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## Beta-galactosidase from *Bacillus licheniformis*

An application to amend the *Australia New Zealand Food Standards Code* with  
a beta-galactosidase preparation produced by a genetically modified strain of  
*Bacillus licheniformis*

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Regulatory Affairs



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## EXECUTIVE SUMMARY

The present application seeks to amend Schedule 18 - Processing Aids of the Australia New Zealand Food Standards Code (the Code) to approve a beta-galactosidase enzyme preparation produced by Novozymes A/S.

### ***Proposed change to Australia New Zealand Food Standards Code – Schedule 18 – Processing aids***

The table in S18—4, Permitted enzymes (section 1.3.3—6)—Enzymes of microbial origin, is proposed to be amended to include a genetically modified strain of *Bacillus licheniformis* as permitted source for beta-galactosidase.

The application is applied for assessment by the general procedure.

### ***Description of enzyme preparation***

The enzyme is a beta-galactosidase (EC 3.2.1.23), commonly known as lactase.

Beta-galactosidases catalyze the hydrolysis of terminal non-reducing beta-D-galactose residues in beta-D-galactosides. The most common and well-known reaction is the hydrolysis of D-lactose into D-glucose and D-galactose.

The enzyme is produced by submerged fermentation of a *Bacillus licheniformis* microorganism expressing a beta-galactosidase from *Bifidobacterium bifidum*.

The commercial enzyme product, Saphera, is available in two product strengths as liquid preparations complying with the JECFA recommended purity specifications for food-grade enzymes.

The producing micro-organism, *Bacillus licheniformis*, is absent from the commercial enzyme product.

### ***Use of the enzyme***

The lactase preparation is used as a processing aid during the manufacture of milk and other lactose containing products. Lactase converts lactose to glucose and galactose helping to produce lactose free or lactose reduced milk/dairy products.

The enzyme is used during milk production in either of two methods to produce low-lactose milk. In the classical, low-lactose process, milk is pasteurized and then cooled. At this point the lactase is added and allowed to react with the milk. The milk is then heat treated again to preserve the milk and to stop the action of the lactase enzyme.

In the low-lactose UHT process, milk is UHT treated and cooled to ambient temperature, at which time sterile filtered lactase is added and the product sterile packed. The lactase is allowed to react with lactose in the milk over several days, and the action of the lactase stops when there is no more lactose present. In the final dairy product the substrate is depleted,



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which means that any remaining low level of food enzyme does not have any action or any function, and is thus, like any other protein, inert.

## **Benefits**

The benefits of the action of the food enzyme in milk/dairy processing are:

- Lactose-reduced products are easier and safer to digest for lactose-intolerant individuals
- Sweeter dairy products due to glucose and galactose formation
- Less sugar addition needed to obtain the wanted product sweetness – thus, reduced caloric value
- Reduced sandiness in ice cream because no lactose crystals are formed
- Softer scoop of ice cream due to lower freezing point

## **Safety evaluation**

The safety of the strain and the enzyme product has been thoroughly assessed:

- The production organism has a long history of safe use as production strain for food grade enzyme preparations and is known not to produce any toxic metabolites.
- The genetic modifications in the production strain are well-characterized and safe and the recombinant DNA is stably integrated into the production organism and unlikely to pose a safety concern.
- The enzyme preparation complies with international specifications ensuring absence of contamination by toxic substances or noxious microorganisms
- Sequence homology assessment to known allergens and toxins shows that oral intake of the beta-galactosidase does not pose food allergenic or toxic concern.
- Two mutagenicity studies *in vitro* showed no evidence of genotoxic potential of the enzyme preparation.
- An oral feeding study in rats for 13-weeks showed that all dose levels were generally well tolerated and no evidence of toxicity.

Furthermore, the safety of the beta-galactosidase preparation was confirmed by external expert groups, as follows:

- Denmark: The enzyme preparation was safety assessed resulting in the authorization of the enzyme product by the Danish authorities.
- USA: A GRAS determination was done and notified to the US FDA in March 2015 (GRN000572). In the reply letter from FDA dated August 28<sup>th</sup>, 2015, the agency has no questions regarding Novozymes' determination that the beta-galactosidase enzyme preparation is GRAS for its intended use.
- Mexico: Based on a dossier submitted by Novozymes, the Mexican food authorities, COFEPRIS, has approved the enzyme in February 2016.

## **Conclusion**

Based on the Novozymes safety evaluation (confirmed by the above-mentioned bodies), we respectfully request the inclusion of this enzyme in the table in S18—4, Permitted enzymes (section 1.3.3—6)—Enzymes of microbial origin.

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# INTRODUCTION

The present dossier describes a beta-galactosidase enzyme preparation produced by submerged fermentation of a *Bacillus licheniformis* microorganism expressing a beta-galactosidase from *Bifidobacterium bifidum*. The Novozymes A/S trade name used for the beta-galactosidase enzyme preparation is Saphera.

The enzyme is a beta-galactosidase (EC 3.2.1.23), commonly known as lactase. The enzyme catalyzes the hydrolysis of D-lactose into D-glucose and D-galactose.

The lactase enzyme preparation is intended to be used as a processing aid in the dairy industry for making lactose reduced/free products e.g. milk, yogurt, cream and ice cream.

The following sections describe in detail the construction of the genetically modified *Bacillus licheniformis* used as the production organism, the production process, the product specification, the application of the enzyme preparation and finally the safety evaluation of the product including the toxicology program, which has been carried out confirming the safety of the product for its intended use.

The documentation has been elaborated according to the Application Handbook from Food Standards Australia New Zealand as of March 1<sup>st</sup> 2016, applied as relevant for an enzyme application, i.e. outlining the following section:

- Section 3.1.1 – General requirements
- Section 3.3.2 – Processing aids, subsections A, C, D, E, F

**NB!** When reading this document it should be noticed that in some reports, the lactase enzyme preparation is described by its commercial name, Saphera or by the internal production batch codes PPL 34537 or OFFR 6-7.

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## CHAPTER 3.1, GENERAL REQUIREMENTS FOR APPLICATIONS

### A. Executive Summary

An Executive Summary is provided as a separate copy together with this application.

### B. Applicant details

(a) ***Applicant's name/s***

[REDACTED]

(b) ***Company/organisation name***

Novozymes Australia Pty Ltd

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(d) ***Telephone number***

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(f) ***Nature of applicant's business***

Biotechnology

(g) ***Details of other individuals, companies or organisations associated with the application.***

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## C. Purpose of the application

This application is submitted to provide for amendment of the Australia New Zealand Food Standards Code – Schedule 18 – Processing aids, Table S18—4 to include a genetically modified strain of *Bacillus licheniformis* as permitted source for beta-galactosidase.

## D. Justification for the application

### ***The need for the proposed change***

Table S18—4 of Schedule 18 contain a list of permitted enzymes of microbial origin. There are a number of approved beta-galactosidases EC 3.2.1.23 from different sources, but not from the source *Bacillus licheniformis*.

*Bacillus licheniformis* is an approved host and production strain for a number of enzymes in the Schedule 18, i.e. alpha-amylase, chymotrypsin, endo-1,4-beta-xylanase, glycerophospholipid cholesterol acyltransferase, maltotetraohydrolase, pullulanase, serine proteinase.

### ***The advantages of the proposed change over the status quo***

The lactase preparation is used as a processing aid during the manufacture of milk and other lactose containing products. Lactase converts lactose to glucose and galactose helping to produce lactose free or lactose reduced milk/dairy product.

The benefits of the action of the food enzyme in milk/dairy processing are:

- Lactose-reduced products are easier and safer to digest for lactose-intolerant individuals
- Sweeter dairy products due to glucose and galactose formation
- Less sugar addition needed to obtain the wanted product sweetness – thus, reduced caloric value
- Reduced sandiness in ice cream because no lactose crystals are formed
- Softer scoop of ice cream due to lower freezing point

The benefits, which are described above, are not exclusively obtainable by means of enzyme treatment but can be achieved without the use of enzymes, or with a reduced use of enzymes, through e.g. modified maybe more expensive or less environmentally friendly production processes or recipe changes.

Some of the alternative production processes (acid hydrolysis, chromatographic separation and membrane techniques such as ultrafiltration (UF) and microfiltration have been described by Harju in 2012<sup>1</sup>.

As a response to international customer interests, registration activities have been done globally. The beta-galactosidase preparation is approved in Denmark under the commercial name, Saphera. Also, the beta-galactosidase is the subject of a GRAS determination and notification to US FDA as well as approval obtained in Mexico.

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## **D.1 Regulatory impact information**

### **D.1.1 Costs and benefits of the application**

The application is not likely to place costs or regulatory restrictions on industry or consumers. Inclusion of the beta-galactosidase enzyme in Table S18—4 of Schedule 18 will provide the food industry with the opportunity to produce “lactose-reduced” or “lactose-free” milk products with the desired functional properties under environmentally friendly and cost efficient production conditions. For government, the burden is limited to necessary activities for a variation of Schedule 18.

### **D.1.1 Costs and benefits of the application**

The application is not likely to cause impact on international trade.

## **E. Information to support the application**

### **E.1 Data requirements**

No public health and safety issues related to the proposed change are foreseen. As outlined in sections 3.3.2 C, D, E, F, the beta-galactosidase is produced by submerged fermentation of a genetically modified *Bacillus licheniformis* strain.

The safety of the strain and the enzyme product has been thoroughly assessed:

- The production organism has a long history of safe use as production strain for food grade enzyme preparations and is known not to produce any toxic metabolites.
- The genetic modifications in the production strain are well-characterized and safe and the recombinant DNA is stably integrated into the production organism and unlikely to pose a safety concern.
- The enzyme preparation complies with international specifications ensuring absence of contamination by toxic substances or noxious microorganisms
- Sequence homology assessment to known allergens and toxins shows that oral intake of the beta-galactosidase does not pose food allergenic or toxic concern.
- Two mutagenicity studies *in vitro* showed no evidence of genotoxic potential of the enzyme preparation.
- An oral feeding study in rats for 13-weeks showed that all dose levels were generally well tolerated and no evidence of toxicity.

## **F. Assessment procedure**

Because the application is for a new source organism for an existing enzyme in the Code, it is considered appropriate that the assessment procedure is characterized as “General Procedure, Level 1”.

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## **G. Confidential commercial information (CCI)**

Detailed information on the construction and characteristics of the genetically modified production strain is provided in Appendix 6. A summary of this information is given in section 3.3.2 E. The formal request for treatment of selected parts of Appendix 6 as confidential commercial information (CCI) is included as Appendix 1.1.

## **H. Other confidential information**

Apart from the selected parts of Appendix 6 identified as confidential commercial information (CCI), no other information is requested to be treated as confidential.

## **I. Exclusive capturable commercial benefit (ECCB)**

This application is not expected to confer an Exclusive Capturable Commercial Benefit.

## **J. International and other national standards**

### ***J.1 International Standards***

Use of enzymes as processing aids for food production is not restricted by any Codex Alimentarius Commission (Codex) Standards.

### ***J.2 Other national standards or regulations***

With few exceptions on national, commodity standards, use of enzymes as processing aids for food production is in general not restricted by standards or regulations in other countries.

## **K. Statutory declaration**

The Statutory Declaration is provided as a separate document together with this submission.

## **L. Checklist**

This application concerns an enzyme product intended to be used as a processing aid. Therefore, the relevant documentation according to the Application Handbook from Food Standards Australia New Zealand as of March 1<sup>st</sup> 2016, are the following sections:

- Section 3.1.1 – General requirements
- Section 3.3.2 – Processing aids, subsections A, C, D, E, F

Accordingly, the checklist for General requirements as well as the Processing aids part of the checklist for applications for substances added to food was used and is included as Appendix 1.2 and 1.3.

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## CHAPTER 3.3, GUIDELINES FOR APPLICATIONS FOR SUBSTANCES ADDED TO FOOD

### 3.3.2 PROCESSING AIDS

The lactase enzyme preparation described in this application is representative of the commercial food enzyme product, Saphera, on which approval is sought.

#### A. Technical information on the processing aid

##### A.1. Information on the type of processing aid

Saphera belongs to the category of processing aids described in Table S18—4 of Schedule 18, Enzymes of microbial origin.

Saphera is to be used in the food industry as a processing aid during the manufacture of milk and other lactose containing products. Lactase converts lactose to glucose and galactose helping to produce lactose free or lactose reduced milk/dairy products.

The enzyme is used during milk production in either of two methods to produce low-lactose milk. In the classical, low-lactose process, milk is pasteurized and then cooled. At this point the lactase is added and allowed to react with the milk. The milk is then heat treated again to preserve the milk and to stop the action of the lactase enzyme.

In the low-lactose UHT process, milk is UHT treated and cooled to ambient temperature, at which time sterile filtered lactase is added and the product sterile packed. The lactase is allowed to react with lactose in the milk over several days, and the action of the lactase stops when there is no more lactose present. In the final dairy product the substrate is depleted, which means that any remaining low level of food enzyme does not have any action or any function, and is thus, like any other protein, inert.

The highest dosages are needed in cold hydrolysis processes (4-12°C), where dosages up to 7500 LAU(B) per kg lactose are used

Examples of benefits when applying Saphera during dairy processing, is provided in the Saphera application information sheet, Appendix 2.1.

##### A.2. Information on the identity of the processing aid

###### A.2.1. Enzyme

Generic name:	Beta-galactosidase (also referred to as Lactase)
IUB nomenclature:	Beta-galactosidase
IUB No.:	EC 3.2.1.23
CAS No.:	9031-11-2

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#### A.2.2. Enzyme preparation

Commercial name: Saphera

The lactase enzyme preparation is available under the commercial name Saphera as a single enzyme formulation in 2 product variants, a high-strength, liquid enzyme preparation, Saphera 2600 L, and a sterile, liquid enzyme formulation, Saphera 900 L S.

The Product Data Sheets for Saphera 2600 L and Saphera 900 L S are enclosed as Appendix 2.2 and 2.3. The typical compositions of Saphera 2600 L and Saphera 900 L S are shown below:

	<u>Saphera 2600 L</u>	<u>Saphera 900 L S</u>
Enzyme solids (TOS <sup>a</sup> )	approx. 2.3 %	approx. 0.8 %
Glycerol	approx. 60 %	approx. 60 %
Water	approx. 37.7 %	approx. 39.2 %

Saphera 2600 L and Saphera 900 L S are standardized in lactase units to an activity of 2600, respectively 900 LAU(B)/g. The Novozymes method used to determine the LAU(B) activity is enclosed in Appendix 3.1.

Beta-galactosidase converts o-NitroPhenyl beta-D-galactopyranoside (ONPG) to o-NitroPhenyl (ONP). ONP has a yellow colour when in an alkaline solution and can be quantified at 405 nm. The colour development is monitored, so the change in absorption per time unit can be calculated. The increase in absorption is proportional to the enzyme activity.

#### A.2.3. Host organism

The host strain is a modified (non-sporulating, protease deficient) *Bacillus licheniformis* strain derived from a natural isolate of *B. licheniformis*, DSM 9552. The taxonomic classification is as follows:

Name: *Bacillus licheniformis*  
Class: Bacilli  
Order: *Bacillales*  
Genus: *Bacillus*  
Species: *licheniformis*

For a more detailed description of the host organism and the genetic modifications, please see section 3.3.2 E.

#### A.2.4. Donor organism

The beta-galactosidase is from *Bifidobacterium bifidum*. The introduced gene is chemically synthesized on sequence data from a public database and is not protein engineered.

For a more detailed description of the donor and the donor gene, please see section 3.3.2 E.

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<sup>a</sup> TOS = Total Organic Solids, defined as: 100% - water - ash - diluents



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### **A.3. Information on the chemical and physical properties of the processing aid**

The enzyme is a beta-galactosidase (EC 3.2.1.23), commonly known as lactase. Beta-galactosidases catalyze the hydrolysis of terminal non-reducing beta-D-galactose residues in beta-D-galactosides. The most common and well-known reaction is the hydrolysis of D-lactose into D-glucose and D-galactose (simply called lactose, glucose and galactose, respectively).

The enzyme preparation is available in two formulations as liquid products stabilized with glycerol.

The food enzyme object of the present dossier is not added to final foodstuffs but used as a processing aid during food manufacturing.

No reaction products, which could not be considered normal constituents of the diet, are formed during the production or storage of the enzyme treated food.

### **A.4. Manufacturing process**

The manufacturing process is composed of a fermentation process, a purification process, a formulation process and finally a quality control of the finished product, as outlined by Aunstrup et al. 1979<sup>2</sup>. This section describes the processes used in manufacturing of the lactase enzyme product.

The enzyme preparation is manufactured in accordance with current Good Manufacturing Practices, Food. The quality management system used in the manufacturing process complies with ISO 9001:2008 (Appendix 4).

The raw materials are Food Grade Quality and have been subjected to appropriate analysis to ensure their conformity with the specifications.

#### **A.4.1. Fermentation**

The lactase is produced by submerged fed-batch pure culture fermentation of the genetically modified strain of *Bacillus licheniformis*, described in section 3.3.2 E.

##### **A.4.1.1. Raw materials for fermentation**

The production strain is grown in a medium consisting of compounds providing an adequate supply of carbon and nitrogen plus minerals and vitamins necessary for growth. The choice of raw materials used in the fermentation process (the feed, the seed fermenter, the main fermenter and dosing) is listed below.

Carbohydrates (e.g. sucrose, glucose, maltose, starch hydrolysates)

Vegetable protein (e.g. potato protein, soy bean meal)

Ammonia

Salts (e.g. KOH, (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, CaCO<sub>3</sub>, MgSO<sub>4</sub>, Na<sub>2</sub>HPO<sub>4</sub>)

Trace metals (e.g. MnSO<sub>4</sub>, FeSO<sub>4</sub>, CuSO<sub>4</sub>, ZnSO<sub>4</sub>)

Alkali and acid for pH adjustments (e.g. Citric acid, H<sub>3</sub>PO<sub>4</sub>, NH<sub>3</sub>, NaOH)

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Antifoaming agents (if necessary, e.g. polypropylene glycol, modified polyalkoxyether)  
Potable water

#### *A.4.1.2. Hygienic precautions*

All equipment is designed and constructed to prevent contamination by foreign micro-organisms.

All valves and connections not in use for the fermentation are sealed by steam at more than 120°C.

After sterilization a positive pressure of more than 0.2 atmosphere is maintained in the fermentation tank.

The air used for aeration is sterilized by passing a sterile filter.

The inside of each fermentation tank is cleaned between fermentations by means of a high-pressure water jet and inspected after the cleaning procedures have been completed.

#### *A.4.1.3. Preparation of the inoculum*

The inoculum flask containing the prepared medium is autoclaved and checked. Only approved flasks are used for inoculation.

The stock culture suspension is injected aseptically into the inoculum flask and spread onto the medium in the flask. Once growth has taken place in the inoculum flask (typically after a few days at 30°C), the following operations are performed:

- Strain identity and traceability: ampoule number is registered
- Microbial purity: a sample from the inoculum flask is controlled microscopically for absence of microbial contaminants.

When sufficient amount of biomass is obtained and when the microbiological analyses are approved, the inoculum flask can be used for inoculating the seed fermentor.

#### *A.4.1.4. The seed fermentation*

The raw materials for the fermentation medium are mixed with water in a mixing tank. The medium is transferred to the seed fermenter and heat sterilized (e.g. 120°C / 60 min).

The seed fermentation tank is inoculated by transferring aseptically a suspension of cells from the inoculum flask.

The seed fermentation is run aerobically (sterile airflow), under agitation. The overpressure is kept above 0.2 atmosphere at all times, to prevent contamination.

Once a sufficient amount of biomass has developed, microbiological analyses are performed to ensure absence of contamination. The seed fermentation can then be transferred to the main fermentation tank.

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#### A.4.1.5. *The main fermentation*

The raw materials for the medium are mixed with water in a mixing tank. The medium is transferred to the main fermenter and heat sterilized (e.g. 120°C / 60 min). If necessary, the pH is adjusted after sterilization, with sterile pH adjustment solutions.

The fermentation in the main tank is run as normal submerged fed-batch fermentation.

The main fermentation is run aerobically (sterile airflow), under vigorous agitation. The overpressure is kept above 0.2 atmosphere at all times, to prevent contamination. The fermentation is run at a well-defined temperature.

Fresh medium is added aseptically when the pH increases above its set point, and the dissolved oxygen concentration rises. The feed rate is adjusted so that there is no accumulation of carbohydrates.

Other parameters are measured at regular intervals

- Refractive index
- Enzyme productivity
- Residual glucose
- Residual ammonia.

Samples are also taken at regular intervals to check absence of microbial contamination.

#### A.4.2. *Recovery*

The recovery process is a multi-step operation designed to separate the enzyme from the microbial biomass and partially purify, concentrate, and stabilize the food enzyme.

The steps of this process involve a series of typical unit operations:

- Pre-treatment
- Primary separation
- Concentration
- Pre and germ filtration
- Preservation and stabilization
- Pre and germ filtration (if needed)

##### A.4.2.1. *Raw materials for recovery*

The raw materials typically used in the recovery process are as follows:

Potable water

Diatomite or Perlite

Acids and bases for pH adjustment (e.g. Acetic acid, NaOH)

Flocculants (e.g. Calcium chloride, polymer of dimethylamin and epichlorhydrin, anionic polyacrylamide and poly(aluminium hydroxy)chloride)

Stabilisation: Glycerol

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#### A.4.2.2. *Pre-treatment*

To facilitate the separation, flocculants are used in a pH-controlled process.

#### A.4.2.3. *Primary separation*

The cell mass and other solids are separated from the broth by well-established techniques such as pre-coat vacuum drum filtration or centrifugation. The precoat used in the filter and the filter aid used in the process is diatomaceous earth (diatomite or perlite).

The primary separation is performed at well-defined pH and temperature range.

#### A.4.2.4. *Pre and germ filtration*

For removal of residual cells of the production strain and as a general precaution against microbial degradation, filtration on dedicated germ filtration media is applied. Pre-filtration is included when needed.

The filtrations are performed at well-defined pH and temperature intervals, and result in an enzyme concentrate solution free of the production strain and insoluble substrate components from the fermentation.

#### A.4.2.5. *Concentration*

Ultrafiltration and/or evaporation are applied for concentration and further purification. The ultrafiltration is applied to fractionate high molecular weight components (enzymes) from low molecular weight components and is used to increase the activity/dry matter ratio. Evaporation is used to increase the activity while maintaining the activity/dry matter ratio.

The pH and temperature are controlled during the concentration step, which is performed until the desired activity and activity/dry matter ratio has been obtained.

#### A.4.2.6. *Preservation and stabilization*

For enzymatic, physical and microbial stabilization glycerol is added to the enzyme concentrate. pH is adjusted by acetic acid or sodium hydroxide.

#### A.4.2.7. *Final filtration*

A polish filtration is performed to remove any precipitations followed by a final germ filtration. The enzyme concentrate is stored at 5-10 °C.

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#### A.4.2.8. *Process control*

Apart from the process controls performed during the various fermentation steps and described above, the following microbial controls are also performed.

Samples are withdrawn from both the seed fermenter and the main fermenter:

- a) before inoculation
- b) at regular interval during cultivation
- c) before transfer/harvest

The samples during all steps are examined by:

- a) microscopy
- b) plating culture broth on a nutrient agar and incubating for 24-48 hours.

Growth characteristics are observed macroscopically and microscopically.

During the microbiological control steps, the number of foreign micro-organisms should be insignificant. The fermentation parameters, i.e. enzyme activity, temperature and oxygen as well as pH are also monitored closely. A deviation from the normal course of the fermentation may signal a contamination.

If a significant contamination develops, the fermentation is terminated. The fermentation is regarded as “significantly contaminated” if two independent samples show presence of contaminating organisms after growth on nutrient agar.

Any contaminated fermentation is rejected for enzyme preparations to be used in a food grade application.

### **A.5. *Specification for identity and purity***

The beta-galactosidase enzyme product complies with the purity criteria recommended for Enzyme Preparations in Food, Food Chemical Codex, 9th edition, 2014.

In addition to this, the beta-galactosidase enzyme product also conforms to the General Specifications for Enzyme Preparations Used in Food Processing as proposed by the Joint FAO/WHO Expert Committee on Food Additives in Compendium of Food Additive Specifications, available online at: <http://www.fao.org/ag/agn/jecfa-additives/search.html?lang=en>.

Analytical data for an unstandardized, representative batch of the beta-galactosidase enzyme product is shown in the table below. These data show compliance with the purity criteria of the specification.

Control parameter	Unit	Specification	Batch OFFR 6-7
Beta-galactosidase activity	LAU(B)/g		8690
Heavy Metals <sup>a</sup>	ppm	Max 30	4.2
Pb	ppm	Max 5	ND <sup>b</sup> (DL < 0.5) <sup>c</sup>
As	ppm	Max 3	ND (DL < 0.1)
Cd	ppm	Max 0.5	ND (DL < 0.05)
Hg	ppm	Max 0.5	ND (DL < 0.03)
Total viable count	/g	Not more than 50000	100
Total coliforms	/g	Not more than 30	<10
Enteropathogenic E. coli	/25g	Not detected	ND
Salmonella	/25g	Not detected	ND
Antibiotic activity		Not detected	ND
Production strain	/g	Not detected	ND

a) Heavy Metals =  $\Sigma$  of Ag, As, Bi, Cd, Cu, Hg, Mo, Ni, Pb, Sb, Sn

b) ND = Not Detected

c) DL = Detection Limit

The methods of analysis used to determine compliance with the specifications are enclosed (Appendix 3).

The lactase enzyme preparation is available under the commercial name Saphera as a single enzyme formulation in 2 product variants, a high-strength, liquid enzyme preparation, Saphera 2600 L, and a sterile, liquid enzyme formulation, Saphera 900 L S.

The Product Data Sheets for Saphera 2600 L and Saphera 900 L S are enclosed as Appendix 2.2 and 2.3. The typical compositions of Saphera 2600 L and Saphera 900 L S are shown below:

	<u>Saphera 2600 L</u>	<u>Saphera 900 L S</u>
Enzyme solids (TOS <sup>b</sup> )	approx. 2.3 %	approx. 0.8 %
Glycerol	approx. 60 %	approx. 60 %
Water	approx. 37.7 %	approx. 39.2 %

Saphera 2600 L and Saphera 900 L S are standardized in lactase units to an activity of 2600, respectively 900 LAU(B)/g. The Novozymes method used to determine the LAU(B) activity is enclosed in Appendix 3.1.

The beta-galactosidase enzyme preparation does not contain known food allergens as detailed in the Product Data Sheets in Appendix 2.2 and 2.3.

The lactase preparation is used in milk/dairy processing for the hydrolysis of lactose during processing raw materials and intermediates typically known to contain high concentrations of lactose. The enzyme is denatured by heat during processing or the substrate is depleted, and therefore it does not exert a function in final food.

<sup>b</sup> TOS = Total Organic Solids, defined as: 100% - water - ash - diluents

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#### **A.6. Analytical method for detection**

The beta-galactosidase enzyme preparation is to be used in the food industry as a processing aid. This information is not required in the case of an enzymatic processing aid.

### **B. Information related to the safety of a chemical processing aid**

Not applicable – this application does not concern a chemical processing aid.

### **C. Information related to the safety of an enzyme processing aid**

#### **C.1. General information on the use of the enzyme as a food processing aid in other countries**

The enzyme is used as processing aid in the dairy industry in a range of countries, where there are no restrictions of the use of enzyme processing aids or where the enzyme is covered by country positive list or specific approval. Additional global registration activities for the beta-galactosidase enzyme object of the present dossier are in progress to further widen the range of countries where this enzyme can be used in regulatory compliance.

The safety of the beta-galactosidase preparation has been evaluated and confirmed by external expert groups, as follows:

- Denmark: The enzyme preparation was safety assessed resulting in the authorization of the enzyme product by the Danish authorities.
- USA: A GRAS determination was done and notified to the US FDA in March 2015 (GRN000572). In the reply letter from FDA dated August 28<sup>th</sup>, 2015, the agency has no questions regarding Novozymes' determination that the beta-galactosidase enzyme preparation is GRAS for its intended use.
- Mexico: Based on a dossier submitted by Novozymes, the Mexican food authorities, COFEPRIS, has approved the enzyme in February 2016.

#### **C.2. Information on the potential toxicity of the enzyme processing aid**

##### **(a) Information on the enzyme's prior history of human consumption and/or its similarity to proteins with a history of safe human consumption**

A wide variety of enzymes are used in food processing. Enzymes including lactase have a long history of use in food (Pariza and Foster, 1983<sup>3</sup> and Pariza and Johnson, 2001<sup>4</sup>).

Lactase has been used extensively for more than 25 years in various industrial food applications such as for reduction of lactose, with their major application in low lactose milk processes. Lactase enzyme preparations from various sources are widely authorized, e.g. in Australia, Brazil, Canada, China, Denmark, France, Mexico, South Korea.

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(b) *Information on any significant similarity between the amino acid sequence of the enzyme and that of known protein toxins*

A sequence homology assessment of the beta-galactosidase enzyme to known toxins and allergens was conducted. No homologies to toxins or allergens were found. The complete search report is enclosed in Appendix 5.1.

Furthermore, safety studies as described below were performed on a representative batch (PPL 34537) that was produced according to the description given in section 3.3.2 A.4, omitting stabilization and standardization. A summary of the safety studies is enclosed in Appendix 5.2.

The following studies were performed:

- Ames Test. Test for mutagenic activity (Appendix 5.3)
- *In vitro* micronucleus test (Appendix 5.4)
- Subchronic (13 week) oral toxicity study in rats (Appendix 5.5)

The main conclusions of the safety studies can be summarized as follows:

- Beta-galactosidase, PPL 34537 did not induce gene mutations in bacteria either in the presence or absence of metabolic activation (S-9) when tested under the conditions employed in this study.
- Beta-galactosidase, PPL 34537 did not induce micronuclei in cultured human peripheral blood lymphocytes following treatment in the presence or absence of an aroclor induced rat liver metabolic activation system (S-9).
- Oral administration of batch PPL 34537 to Sprague-Dawley rats at doses up to 100% of the tox test batch (672 mg TOS/kg bw/day for 13 weeks was well-tolerated and did not cause any adverse change. The NOAEL was considered to be 100% of the tox test batch (equivalent to 672 mg TOS/kg bw/day).

Based on the present toxicity data it can be concluded that the beta-galactosidase enzyme preparation, represented by batch PPL 34537 exhibits no toxicological effects under the experimental conditions described.

### **C.3. *Information on the potential allergenicity of the enzyme processing aid***

(a) *Information of the source of the enzyme processing aid*

The beta-galactosidase enzyme is produced by a *Bacillus licheniformis* microorganism expressing the beta-galactosidase from *Bifidobacterium bifidum*. *Bacillus licheniformis* is a soil and plant living saprophyte, recognized as non-pathogenic species for humans, animals and plants (see Section 3.3.2 D).

(b) *Analysis of similarity between the amino acid sequence of the enzyme and that of known allergens*

Enzymes have a long history of safe use in food, with no indication of adverse effects or reactions. Moreover a wide variety of enzyme classes (and structures) are naturally present in food.



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The allergenicity potential of enzymes was studied by Bindslev-Jensen et al (2006<sup>5</sup>) and reported in the publication: "Investigation on possible allergenicity of 19 different commercial enzymes used in the food industry". The investigation comprised enzymes produced by wild-type and genetically modified strains as well as wild-type enzymes and protein engineered variants and comprised 400 patients with a diagnosed allergy to inhalation allergens, food allergens, bee or wasp. It was concluded from this study that ingestion of food enzymes in general is not likely to be a concern with regard to food allergy.

Additionally, food enzyme are used in small amounts during food processing resulting in very small amounts of the enzyme protein in the final food. A high concentration generally equals a higher risk of sensitization, whereas a low level in the final food equals a lower risk (Goodman et al, 2008<sup>6</sup>).

A sequence homology assessment of the beta-galactosidase enzyme to known toxins and allergens was conducted. No homologies to toxins or allergens were found. The complete search report is enclosed in Appendix 5.1.

Consequently, oral intake of the beta-galactosidase is not anticipated to pose any food allergenic concern.

#### **C.4. Safety assessment reports prepared by international agencies or other national government agencies, if available**

The certificate of approval of the lactase enzyme preparation by the Danish authorities following their safety evaluation in accordance with guidelines from the European Food Safety Authority (EFSA) is enclosed as Appendix 2.4.

In addition, the US FDA did not question the conclusion that the lactase enzyme preparation object of the present dossier is GRAS for its intended use, cf. <http://www.accessdata.fda.gov/scripts/fcn/fcnDetailNavigation.cfm?rpt=grasListing&id=572>).

### **D. Additional information related to the safety of an enzyme processing aid derived from a microorganism**

#### **D.1. Information on the source microorganism**

The beta-galactosidase enzyme is produced by a *Bacillus licheniformis* microorganism expressing the beta-galactosidase from *Bifidobacterium bifidum*. The host strain is a modified (non-sporulating, protease deficient) *Bacillus licheniformis* strain derived from a natural isolate of *B. licheniformis*, DSM 9552.

#### **D.2. Information on the pathogenicity and toxicity of the source microorganism**

*Bacillus licheniformis* is a soil and plant living saprophyte, recognized as non-pathogenic species for humans, animals and plants (Priest FG, 1993<sup>7</sup>, de Boer AS et al, 1994<sup>8</sup>, EPA, 1997<sup>9</sup>). *B. licheniformis* is also common in foods including natural agricultural products such as cereals.

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*B. licheniformis* is classified as a group 1 microorganism according to EU Directive 2000/54/EC of the European Parliament and of the Council of 18 September 2000 on the protection of workers from risks related to exposure to biological agents at work. A group 1 biological agent means one that is unlikely to cause human disease.

The host strain is sporulation deficient.

Industrial strains belonging to the *B. licheniformis* species have a long history of safe use in food enzyme manufacturing. They have been used for decades in the production of enzymes, and in more than a decade as recombinant organisms for the production of a variety of bio-industrial products like food grade enzymes, vitamins, antibiotics, and additives (Schallmey et al, 2004<sup>10</sup>).

The Food and Drug Administration has affirmed that mixed carbohydrase and protease enzyme products derived from *B. licheniformis* are generally recognized as safe (GRAS) in the production of certain foods including nutritive sweeteners, see 21CFR §184.1027. In the supplementary information to the final rule in the Federal Register, FDA emphasized that "Published scientific literature as well as standard books on food microbiology demonstrate that *B. licheniformis* is widely recognized as a common contaminant found in many foods. None of these references report any toxicity or pathogenicity associated with the presence of this organism in food."

In addition, the FDA did not question the conclusion that various other food enzymes obtained from genetically modified *B. licheniformis* strains are GRAS under the intended conditions of use (GRN no. 22, GRN no. 24, GRN no. 72, GRN no. 79, GRN no. 265, GRN no. 277, GRN 472 (GRAS Notice Inventory<sup>11</sup>).

The non-pathogenicity and non-toxicity of *B. licheniformis* is thus strongly supported by the historic record of this organism.

### **D.3. Information on the genetic stability of the source organism**

The inserted recombinant DNA is genetically stable during fermentation, as the inserted DNA is integrated into the chromosome.

The genetic stability of the production strain was tested at large-scale fermentation. The strain stability during fermentation was analyzed by Southern blotting. No instability of the strain was observed.

For a more detailed description of the strain construction and characteristics, please see section 3.3.2 E.

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## **E. Additional information related to the safety of an enzyme processing aid derived from a genetically-modified microorganism**

### ***E.1. Information on the methods used in the genetic modification of the source organism***

This section contains summarized information on the modifications of the host strain, on the content and nature of the introduced DNA and on the construction of the final production strain, as well as the stability of the inserted gene. The detailed information is provided in the confidential Appendix 6.

#### *E.1.1. Host organism*

The parental strain is a natural isolate and the taxonomic classification is as followed:

Name: *Bacillus licheniformis*  
Class: Bacilli  
Order: *Bacillales*  
Genus: *Bacillus*  
Species: *licheniformis*

The classification was confirmed by Deutsche Sammlung von Mikroorganismen und Zellkulturen GmbH and deposited as DSM 9552.

The recipient strain, AEB1763, used in the construction of the *Bacillus licheniformis* production strain, was derived from the parental strain by modification of several chromosomal loci to cause deletion of genes encoding a number of proteases. Also a gene essential for sporulation was deleted, eliminating the ability to sporulate, together with three additional genes encoding unwanted proteins that can be present in the culture supernatant. The lack of these proteins represents improvements in product purity, safety and stability.

#### *E.1.2. Introduced DNA*

The expression plasmid, pPP2771, used to transform the *Bacillus licheniformis* recipient strain AEB1763, is based on the well-known *Bacillus* vector pE194 (Horinouchi and Weisblum, 1982<sup>12</sup>) from *Staphylococcus aureus* and a standard *E. coli* vector. No elements of these vectors are left in the production strain. The plasmid contains the expression cassette consisting of a fragment of a hybrid bacillus promoter with promoter elements from *Bacillus licheniformis*, *Bacillus amyloliquefaciens* and *Bacillus thuringiensis*, the lactase coding sequence and a terminator. This promoter is followed by a chemically synthesized lactase gene based on sequence data from a public database.

The lactase gene, *galT1*, is derived from the *bbgIII* gene from *Bifidobacterium bifidum* (NCIMB 41171). In *galT1* the sequence encoding the wild type, mature lactase is fused to the signal peptide from the *aprH* gene of *Bacillus clausii* to enable efficient secretion (DSM 8716). Finally a *Bacillus licheniformis* terminator sequence is inserted to terminate transcription. Following the terminator, a non-coding DNA sequence inserted to enable targeted integration on the genome of the recipient strain. Only the expression cassette with

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elements between the promoter and the terminator are present in the final production strain. This has been confirmed by Southern blot analysis and PCR analysis followed by DNA sequencing.

#### *E.1.3. Construction of the Recombinant Microorganism*

The production strain, *Bacillus licheniformis* PP3930, was constructed from recipient strain AEB1763 through the following steps:

- 1) Plasmid pPP2771 was integrated into four specific loci in strain AEB1763 by targeted homologous recombination to these loci using a two-step integration approach. After the initial integration of pPP2771 at the four target loci, excision of the bacterial backbone occurs by homologous recombination. Thus, only the expression cassette of the *galT1* gene containing the promoter and the transcriptional terminator is left at each target locus.
- 2) The resulting four-copy *galT1* strain was named PP3930.

Sequence confirmation of the inserted expression cassettes and the flanking regions at both of the integration loci was performed in the production strain.

#### *E.1.4. Antibiotic Resistance Gene*

No functional antibiotic resistance genes were left in the strain as a result of the genetic modifications. The absence of these genes in the production strain was verified by Southern blot analysis using the relevant antibiotic resistance gene probes.

#### *E.1.5. Stability of the Introduced Genetic Sequences*

The presence of the introduced DNA sequences was also determined by Southern hybridization to assess the stability and potential for transfer of genetic material as a component of the safety evaluation of the production microorganism. The transforming DNA is stably integrated into the *Bacillus licheniformis* chromosome and, as such, is poorly mobilized for genetic transfer to other organisms and is mitotically stable.

## **F. Information related to the dietary exposure to the processing aid**

### ***F.1. A list of foods or food groups likely to contain the processing aid or its metabolites***

The lactase enzyme preparation is used as a processing aid during the manufacture of milk and other lactose containing products. Lactase converts lactose to glucose and galactose helping to produce lactose free or lactose reduced milk/dairy products.

The typical use of lactase is to reduce the content of lactose in cow's milk. This lactose-reduced milk product is consumed as is or used as an ingredient in low-lactose food applications.

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## ***F.2. The levels of residues of the processing aid or its metabolites for each food or food group***

The lactase enzyme preparation is used at minimum levels necessary to achieve the desired effect and according to requirements for normal production following cGMP.

The enzyme is used during two methods of milk production.

In the classical, low lactose process milk is pasteurized and then cooled. At this point the lactase is added and allowed to react with the milk. The milk is then heat treated again to preserve the milk and stop the action of the lactase enzyme.

In the low-lactose UHT process, milk is UHT treated and cooled to ambient temperature, at which time sterile filtered lactase is added and the product sterile packed. The lactase is allowed to react with lactose in the milk over several days, and the action of the lactase stops when there is no more lactose present. In the final dairy product the substrate is depleted, which means that any remaining low level of food enzyme does not have any action or any function and is thus, like any other protein, inert.

### *Use level*

The enzyme preparation is used at minimum levels necessary to achieve the desired effect and according to requirements for normal production following cGMP.

The conditions of use of the lactase preparation during food processing do not only depend on the type of application, but also on the food production process of each individual food manufacturer. In order to ensure optimal effectiveness of the enzyme at an acceptable economic cost the dosage, reaction time, process conditions and processing steps are adjusted.

The highest dosages are needed in cold hydrolysis processes (4-12°C), where dosages up to 7500 LAU(B) per kg lactose are used. This corresponds to 2.88 g of Saphera 2600 L per kg lactose equivalent to 66 mg TOS/kg lactose (cf. Section 3.3.2 A 2.2).

### *Enzyme residues in the Final Food*

The lactase preparation is used in milk/dairy processing for the hydrolysis of lactose during processing raw materials and intermediates typically known to contain high concentrations of lactose. The enzyme is denatured by heat during processing or the substrate is depleted, and therefore it does not exert a function in final food.

#### *F.2.1. Estimates of human consumption*

##### *Method used for the dietary exposure assessment*

The dietary exposure assessment is based on food consumption data found in the EFSA Comprehensive European Food Consumption Database<sup>13</sup>.

According to this database the maximum average intake of "Milk and dairy products" over 19 countries and all age groups, except infants, ranges from 1.70 to 38.23 g/kg body weight (bw)/day. Infants (up to and including 11 months – see "Use of the EFSA Comprehensive European Food Consumption Database in Exposure Assessment"<sup>14</sup> have been excluded, since lactose intolerance is a very rare occurrence in infants, and since onset of lactose

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intolerance starts at 2-3 years of age and is first complete at 5-10 years of age (Swallow, 2003<sup>15</sup>).

*Theoretical Maximum Daily Intake (TMDI)*

The highest dosage of 7500 LAU(B) per kg lactose, equivalent to 66 mg TOS/kg lactose, and the highest average intake of "Milk and dairy products", 38.23 g/kg body weight (bw)/day, is used for estimating the TMDI.

It is assumed that cow's milk is the dairy product with the highest consumption and therefore, for the sake of simplifying the intake calculation, that "Milk and dairy product" = "Cow's milk". Cow's milk contains approx. 5% lactose, meaning that 38.23 g milk and dairy product contains approx. 1.9 g lactose.

Based on this 1.9 g lactose/kg bw/day will result in an exposure of:  
 $66 \text{ mg TOS}/1000 \text{ g lactose} \times 1.9 \text{ g lactose/kg bw/day} = 0.127 \text{ mg TOS/kg bw/day}$ .

Thereby, the Theoretical Maximum Daily Intake (TMDI) of the food enzyme by consumers is:  
**0.127 mg TOS/kg bw/day**

*F.2.2. Safety Margin Calculation*

The safety margin is calculated as dose level with no adverse effect (NOAEL) divided by the estimated human consumption (TMDI). The NOAEL dose level in the 13 weeks oral toxicity study in rats was 10 ml/kg/day corresponding to 672 mg TOS/kg/day (cf. Section 3.3.2 C 2).

The estimated human consumption is 0.127 mg TOS/kg/day

The safety margin can thus be calculated to be  $672/0.127$  or approximately **5300**.

***F.3. For foods or food groups not currently listed in the most recent Australian or New Zealand National Nutrition Surveys (NNSs), information on the likely level of consumption***

Not relevant.

***F.4. The percentage of the food group in which the processing aid is likely to be found or the percentage of the market likely to use the processing aid***

In the estimate on human consumption given in F.2.1 above, it is assumed that all milk and dairy products containing lactose are produced using the beta-galactosidase object of this dossier as a processing aid at the highest recommended dosage.

***F.5. Information relating to the levels of residues in foods in other countries***

As described in F.2.1 above, a "worst case" calculation is made assuming that all organic matter originating from the enzyme is retained in the processed food product. The dietary exposure is estimated on EU intake data on milk and dairy products, representing the highest relevant per capita consumption of these products.

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**F.6. For foods where consumption has changed in recent years, information on likely current food consumption**

No significant changes in recent years are observed.

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## List of Appendices

- 1) General Requirements
- 2) Product information
- 3) Methods of analysis used to determine compliance with the specifications
- 4) Documentation regarding the manufacturing process
- 5) Safety documentation
- 6) Documentation regarding the production microorganism

# Appendix 1

## General Requirements

1. Formal request for treatment of confidential commercial information (CCI)
2. Checklist for General requirements
3. Checklist for applications for substances added to food

## Appendix 1.1

### Formal request for treatment of confidential commercial information (CCI)

Novozymes respectfully request that the selected and marked parts of Appendix 6 are treated as confidential commercial information (CCI).

The documents in Appendix 6 contain detailed description of the construction of the genetically modified production strain and the introduced DNA. While individual steps in the DNA construction might be well known or publicly available information, the exact steps and sequence of those constitutes information that represent the state-of-the-art of one of Novozymes' core technologies, which has been obtained as a result of substantial investment in research and development within rDNA technology. Therefore, the selected parts of Appendix 6 are claimed confidential for an unlimited period of time.

August 2016



Senior Science Manager  
Regulatory Affairs  
Novozymes A/S

## Appendix 1.2

### Checklist for General requirements

General requirements (3.1.1)		
Check	Page No.	Mandatory requirements
	4	A Form of application
<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/> <i>Application in English</i>
		<input checked="" type="checkbox"/> <i>Executive Summary (separated from main application electronically)</i>
<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/> <i>Relevant sections of Part 3 clearly identified</i>
		<input checked="" type="checkbox"/> <i>Pages sequentially numbered</i>
		<input checked="" type="checkbox"/> <i>Electronic copy (searchable)</i>
		<input checked="" type="checkbox"/> <i>All references provided</i>
<input checked="" type="checkbox"/>	7	B Applicant details
<input checked="" type="checkbox"/>	8	C Purpose of the application
	8	D Justification for the application
<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/> <i>Regulatory impact information</i>
		<input checked="" type="checkbox"/> <i>Impact on international trade</i>
<input checked="" type="checkbox"/>	9	E Information to support the application
		<input checked="" type="checkbox"/> <i>Data requirements</i>
	9	F Assessment procedure
<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/> <i>General</i>
		<input type="checkbox"/> <i>Major</i>
		<input type="checkbox"/> <i>Minor</i>
		<input type="checkbox"/> <i>High level health claim variation</i>
		G Confidential commercial information
<input checked="" type="checkbox"/>	10	<input checked="" type="checkbox"/> <i>CCI material separated from other application material</i>
	10	<input checked="" type="checkbox"/> <i>Formal request including reasons</i>
	23	<input checked="" type="checkbox"/> <i>Non-confidential summary provided</i>
	10	H Other confidential information
<input checked="" type="checkbox"/>		<input type="checkbox"/> <i>Confidential material separated from other application material</i>
		<input type="checkbox"/> <i>Formal request including reasons</i>
<input checked="" type="checkbox"/>	10	I Exclusive Capturable Commercial Benefit
		<input type="checkbox"/> <i>Justification provided</i>
	10	J International and other national standards
<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/> <i>International standards</i>
		<input checked="" type="checkbox"/> <i>Other national standards</i>
<input checked="" type="checkbox"/>		K Statutory Declaration
	10	L Checklist/s provided with application
<input checked="" type="checkbox"/>		<input checked="" type="checkbox"/> <i>3.1.1 Checklist</i>
		<input checked="" type="checkbox"/> <i>All page number references from application included</i>
		<input checked="" type="checkbox"/> <i>Any other relevant checklists for Chapters 3.2–3.7</i>

## Appendix 1.3

### Checklist for applications for substances added to food

Processing aids (3.3.2)		
Check	Page No.	Mandatory requirements
<input checked="" type="checkbox"/>	11	A.1 Type of processing aid
<input checked="" type="checkbox"/>	11	A.2 Identification information
<input checked="" type="checkbox"/>	13	A.3 Chemical and physical properties
<input checked="" type="checkbox"/>	13	A.4 Manufacturing process
<input checked="" type="checkbox"/>	17	A.5 Specification information
<input checked="" type="checkbox"/>	19	A.6 Analytical method for detection
<input type="checkbox"/>		B.1 Industrial use information (chemical only)
<input type="checkbox"/>		B.2 Information on use in other countries (chemical only)
<input type="checkbox"/>		B.3 Toxicokinetics and metabolism information (chemical only)
<input type="checkbox"/>		B.4 Toxicity information (chemical only)
<input type="checkbox"/>		B.5 Safety assessments from international agencies (chemical only)
<input checked="" type="checkbox"/>	19	C.1 Information on enzyme use on other countries (enzyme only)
<input checked="" type="checkbox"/>	19	C.2 Toxicity information of enzyme (enzyme only)
<input checked="" type="checkbox"/>	20	C.3. Allergenicity information of enzyme (enzyme only)
<input checked="" type="checkbox"/>	21	C.4. Overseas safety Assessment Reports
<input checked="" type="checkbox"/>	21	D.1 Information on source organism (enzyme from microorganism only)
<input checked="" type="checkbox"/>	21	D.2 Pathogenicity and toxicity of source microorganism (enzyme from microorganism only)
<input checked="" type="checkbox"/>	22	D.3 Genetic stability of source organism (enzyme from microorganism only)
<input checked="" type="checkbox"/>	23	E.1 Nature of genetic modification of source organism (enzyme from GM source microorganism)
<input checked="" type="checkbox"/>	24	F.1 List of foods likely to contain the processing aid
<input checked="" type="checkbox"/>	25	F.2 Anticipated residue levels in foods
<input checked="" type="checkbox"/>	26	F.3 Information on likely level of consumption
<input checked="" type="checkbox"/>	26	F.4 Percentage of food group to use processing aid
<input checked="" type="checkbox"/>	26	F.5 Information on residues in foods in other countries (if available)
<input checked="" type="checkbox"/>	27	F.6 Where consumption has changed, information on likely consumption

## Appendix 2

### Product information

1. Saphera application sheet
2. Product Data Sheet for Saphera 2600 L
3. Product Data Sheets for Saphera 900 L S
4. DK approval certificate

Lactose-free dairy products  
**The next generation of lactase**

# Novozymes Saphera®



**Set a new standard**  
for production control and  
for creating new high-quality  
lactose-free dairy products

**Saphera®** is the only lactase on the market that achieves:

- Better control of lactose elimination
- Better sweetness stability during product shelf life
- Improved suitability in fermented dairy products

**Dairy producers gain improved production control and the versatility to create high-quality lactose-free products with this innovative lactase. Originated from *Bifidobacterium bifidum*, Saphera® differs from traditional yeast-based products.**

#### **Better control of lactose elimination**

With Saphera®, the desired lactose level can be more precisely measured and easily reached. This is because much less oligosaccharides are formed during the reaction compared to yeast lactases, particularly when producing 0.01% lactose-free milk.

#### **Sweetness enhancement**

Due to the formation of galactose and glucose, Saphera® enables the dairy producer to reduce the amount of added sugars and still achieve the same degree of sweetness in lactose-free dairy products. This offers the opportunity for cleaner, more attractive product labels.

#### **Sweetness stability in sugared dairy products**

With Saphera®, producers are able to maintain a stable level of sweetness and taste during the shelf life of sugared dairy products. This is due to the absence of invertase side activity.

#### **Improved suitability in fermented dairy products**

Saphera® is uniquely well suited for yogurt production, better than conventional yeast-based lactases. The pH and temperature profile, and the absence of invertase and amylase side activities make Saphera® the optimal solution.

#### **Clean taste**

Saphera® is exceptionally pure. This purity reduces the risk of off-flavors during shelf life, particularly useful in dairy products with long shelf life to keep a pure, clean and consistent natural milk flavor.

**Rethink Tomorrow**

**novozymes®**   
Food & Beverages



### Broad application

Saphera® is applicable for a broad range of dairy products including fresh, ESL and UHT milk, milk drinks, cream, ice cream, fermented dairy products and dulce de leche. Saphera® is available in a variety of formulations to meet varying processing needs. The high product purity also enables improved filterability in UHT processes.

### Saphera® product range

Saphera® is sold as a liquid standard product (Saphera® 2600 L) for use in batch process and in-line dosing systems (e.g. TetraPak Aldose). It is sold as a sterile liquid product (Saphera® 900 LS) for use in aseptic in-line dosing systems (e.g. TetraPak FlexDos).

### Novozymes Saphera®

Product name	Product version	Activity
Saphera® 2600 L	Standard liquid version for use in batch process and in-line dosing systems (e.g. TetraPak Aldose)	2600 LAU(B)/g
Saphera® 900 LS	Sterile liquid version for use in aseptic in-line dosing systems (e.g. TetraPak FlexDos)	900 LAU(B)/g

**Table 1.**

Novozymes Saphera® product range

### Novozymes Technical Service

Our experienced technical service representatives have a proven track record of optimizing processes for lower cost-in-use and better quality. Our technical service teams are located in every region of the world. We can help you optimize your process from within your factory, and we can assist you in developing new lactose-free products.

### Quality you can trust

Novozymes is dedicated to quality. From the time a new idea is born to when it's an integrated part of your process, we continue to monitor our products. We provide safe production strains, and assessed and approved raw materials – under strict, controlled processes that have full traceability. All to ensure safe, stable solutions that add value for you.



### Learn more

For more market insights and innovation inspiration, or to find out more about Saphera®, contact your local Novozymes representative at [foodandbeverages@novozymes.com](mailto:foodandbeverages@novozymes.com)

### About Novozymes

Novozymes is the world leader in biological solutions. Together with customers, partners and the global community, we improve industrial performance while preserving the planet's resources and helping build better lives. As the world's largest provider of enzyme and microbial technologies, our bioinnovation enables higher agricultural yields, low temperature washing, energy efficient production, renewable fuel and many other benefits that we rely on today and in the future. We call it Rethink Tomorrow.

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# Saphera 2600 L

In this product the key enzyme activity is provided by

beta-galactosidase that hydrolyzes terminal non-reducing beta-D-galactosides releasing beta-D-galactose residues

## PRODUCT CHARACTERISTICS/PROPERTIES

Component name	Beta-galactosidase
Activity	2600 LAU-B/g
Side activities	The product contains no significant activity of Protease
Color	Light yellow
Physical form	Liquid
<i>Color can vary from batch to batch. Color intensity is not an indication of enzyme activity.</i>	

## PRODUCT SPECIFICATION

	Lower Limit	Upper Limit	Unit
Lactase unit LAU-B	2600		/g
Total viable count (100)	-	100	/g
Coliform bacteria	-	10	/g
E.coli	Not Detected		/25 g
Salmonella	Not Detected		/25 g
Heavy metals		Max 30	mg/kg
Lead		Max 5	mg/kg
Arsenic		Max 3	mg/kg
Cadmium		Max 0.5	mg/kg
Mercury		Max 0.5	mg/kg

## COMPOSITION

Ingredients	Appr. % (w/w)
Glycerol CAS no. 56-81-5	60
Water CAS no. 7732-18-5	38
Beta-galactosidase CAS no. 9031-11-2*	2

\*Defined as enzyme conc. (dry matter basis)

## ALLERGEN

Allergen	Substance contained <sup>1</sup>	Allergen	Substance contained <sup>1</sup>
Celery	no	Molluscs	no
Cereals containing gluten <sup>2</sup>	no	Mustard	no
Crustaceans	no	Nuts <sup>3</sup>	no
Egg	no	Peanuts	no
Fish	no	Sesame	no
Lupin	no	Soy	no
Milk (including lactose)	no	Sulphur dioxide/sulphites more than 10 mg per kg or l	no

<sup>1</sup>Definition of substances according to EU Regulation 1169/2011 as amended. List covers allergens mentioned in 21 USC 301 (US) and GB 7718-2011 (China).

<sup>2</sup>i.e. wheat rye barley oats spelt kamut

<sup>3</sup>i.e. almond hazelnut walnut cashew pecan nut Brazil nut pistachio nut macadamia nut and Queensland nut

## NUTRITIONAL VALUES

The product has a typical nutritional value of approximately 639 kJ/100 g enzyme product.

• Protein	2 g/100 g
• Polyols	60 g/100 g
• Moisture	38 g/100 g

## GM STATUS

This product is not a GMO.

Production organism

Bacillus licheniformis

The enzyme product is manufactured by fermentation of a microorganism that is not present in the final product. The production organism is improved by means of modern biotechnology.

# Saphera 2600 L

## STORAGE CONDITION

**Recommended storage:** 0-10 °C (32-50 °F)

Packaging must be kept intact, dry, and away from sunlight. Please follow the recommendations and use the product before the best before date to avoid the need for a higher dosage.

**Best before:** You will find the best before date in the certificate of analysis or on the product label.

The product gives optimal performance when stored as recommended and used within 24 months of the production date.

Novozymes guarantees delivery at least 12 months prior to the best-before date.

The product can be transported at ambient temperature. Following delivery the product should be stored as recommended.

## SAFETY AND HANDLING PRECAUTIONS

Enzymes are proteins. Inhalation of dust or aerosols may induce sensitization and may cause allergic reactions in sensitized individuals. Some enzymes may irritate the skin, eyes, and mucous membranes upon prolonged contact. See the MSDS or Safety Manual for further information regarding safe handling of the product and spills.

## PATENT

Saphera® 2600L can only be used for non-fermented dairy products and is sold without a license to patents originating from WO 2009/071539 for production of fermented dairy products. Please contact Novozymes if you want to use a lactase in the Saphera® product family for production of yoghurt or other fermented milk products.

:

## COMPLIANCE

The product complies with the recommended purity specifications for food-grade enzymes given by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) and the Food Chemical Codex (FCC).

Kosher and Halal certificates are available from the Customer Center or sales representative.

## CERTIFICATIONS

Novozymes is a signatory to United Nations Global Compact, United Nations Convention on Biological Diversity and report on our sustainability performance through Global Reporting Initiative (GRI). See all our commitments under sustainability on [www.novozymes.com](http://www.novozymes.com).



## FOOD SAFETY

Novozymes has carried out a hazard analysis and prepared an HACCP plan describing the critical control points (CCPs). The HACCP plan is supported by a comprehensive prerequisite program implemented in Novozymes' GMP practices.

The product is produced according to Novozymes' HACCP plan, GMP practices, and additional requirements controlled by Novozymes' Quality Management System.

The product complies with FAO/WHO JECFA- and FCC-recommended purity requirements regarding mycotoxins.

## PACKAGING

The product is available in different types of packaging. Please contact the sales representative for more information.

# Product Data Sheet



1 of 2

Valid from 2016-04-11

## Saphera 900 L S

In this product the key enzyme activity is provided by

beta-galactosidase that hydrolyzes terminal non-reducing beta-D-galactosides releasing beta-D-galactose residues

### PRODUCT CHARACTERISTICS/PROPERTIES

Component name	Beta-galactosidase
Activity	900 LAU-B/g
Side activities	The product contains no significant activity of Protease
Color	Light yellow
Physical form	Liquid
Approximate density (g/ml)	1.15
<i>Color can vary from batch to batch. Color intensity is not an indication of enzyme activity.</i>	

### PRODUCT SPECIFICATION

	Lower Limit	Upper Limit	Unit
Lactase unit LAU-B	900		/g
Heavy metals		Max 30	mg/kg
Lead		Max 5	mg/kg
Arsenic		Max 3	mg/kg
Cadmium		Max 0.5	mg/kg
Mercury		Max 0.5	mg/kg

### COMPOSITION

Ingredients	Appr. % (w/w)
Glycerol CAS no. 56-81-5	60.20
Water CAS no. 7732-18-5	39
Beta-galactosidase CAS no. 9031-11-2*	0.80

\*Defined as enzyme conc. (dry matter basis)

### ALLERGEN

Allergen	Substance contained <sup>1</sup>	Allergen	Substance contained <sup>1</sup>
Celery	no	Molluscs	no
Cereals containing gluten <sup>2</sup>	no	Mustard	no
Crustaceans	no	Nuts <sup>3</sup>	no
Egg	no	Peanuts	no
Fish	no	Sesame	no
Lupin	no	Soy	no
Milk (including lactose)	no	Sulphur dioxide/sulphites more than 10 mg per kg or l	no

<sup>1</sup>Definition of substances according to EU Regulation 1169/2011 as amended. List covers allergens mentioned in 21 USC 301 (US) and GB 7718-2011 (China).

<sup>2</sup>i.e. wheat rye barley oats spelt kamut

<sup>3</sup>i.e. almond hazelnut walnut cashew pecan nut Brazil nut pistachio nut macadamia nut and Queensland nut

### NUTRITIONAL VALUES

The product has a typical nutritional value of approximately 614 kJ/100 g enzyme product.

• Protein	1 g/100 g
• Polyols	60 g/100 g
• Moisture	39 g/100 g

### GM STATUS

This product is not a GMO.

Production organism

Bacillus licheniformis

The enzyme product is manufactured by fermentation of a microorganism that is not present in the final product. The production organism is characterized as self-cloned according to the EU definition.

# Saphera

## 900 L S

### STORAGE CONDITION

**Recommended storage:** 0-10 °C (32-50 °F)

Packaging must be kept intact, dry, and away from sunlight. Please follow the recommendations and use the product before the best before date to avoid the need for a higher dosage.

**Best before:** You will find the best before date in the certificate of analysis or on the product label.

The product gives optimal performance when stored as recommended and used within 12 months from the production date.

The product can be transported at ambient temperature. Following delivery the product should be stored as recommended.

### SAFETY AND HANDLING PRECAUTIONS

Enzymes are proteins. Inhalation of dust or aerosols may induce sensitization and may cause allergic reactions in sensitized individuals. Some enzymes may irritate the skin, eyes, and mucous membranes upon prolonged contact. See the MSDS or Safety Manual for further information regarding safe handling of the product and spills.

### COMPLIANCE

The product complies with the purity specifications for food-grade enzymes given by the Joint FAO/WHO Expert Committee on Food Additives (JECFA) and the Food Chemical Codex (FCC) and is commercially sterile according to WHO/FAO CAC/RCP 40-1993.

Kosher and Halal certificates are available from the Customer Center or sales representative.

### CERTIFICATIONS

Novozymes is a signatory to United Nations Global Compact, United Nations Convention on Biological Diversity and report on our sustainability performance through Global Reporting Initiative (GRI). See all our commitments under sustainability on [www.novozymes.com](http://www.novozymes.com).



### FOOD SAFETY

Novozymes has carried out a hazard analysis and prepared an HACCP plan describing the critical control points (CCPs). The HACCP plan is supported by a comprehensive prerequisite program implemented in Novozymes' GMP practices.

The product is produced according to Novozymes' HACCP plan, GMP practices, and additional requirements controlled by Novozymes' Quality Management System.

The product complies with FAO/WHO JECFA- and FCC-recommended purity requirements regarding mycotoxins.

### PACKAGING

The product is available in different types of packaging. Please contact the sales representative for more information.

TO WHOM IT MAY CONCERN

11-08-2015

File no.: 2015-29-7101-00025 and 2015-29-7101-00033

### **Lactase enzyme product**

The Danish Veterinary and Food Administration hereby certifies having accepted in 2015 the lactase enzyme product from Novozymes A/S (file no. 2015-29-7101-00025). The product, produced by a *Bacillus licheniformis* strain expressing a lactase from *Bifidobacterium bifidum*, has been accepted to be used for processing of milk and dairy products up to a level of 7500 LAU(B) per kg lactose.

The evaluation of the safety of the lactase product has been made in accordance with the principles laid down in the Guidance of EFSA prepared by the Scientific Panel of Food Contact Material, Enzymes, Flavourings and Processing Aids on the submission of a Dossier on Food Enzymes. The EFSA journal (2009) 1305, 1-26.

Yours sincerely



Danish Veterinary and Food Administration  
Division of Chemistry and Food Quality



## Appendix 3

### Methods of analysis used to determine compliance with the specifications

1. Enzyme activity, LAU(B)/g
2. Total aerobic viable count
3. Total coliforms
4. *Escherichia coli* (E. coli).
5. *Salmonella*
6. Antimicrobial activity
7. Production strain

Analysis of Heavy Metals, Lead, Arsenic, Cadmium, and Mercury are performed at an external laboratory, Technological Institute (TI), Denmark.

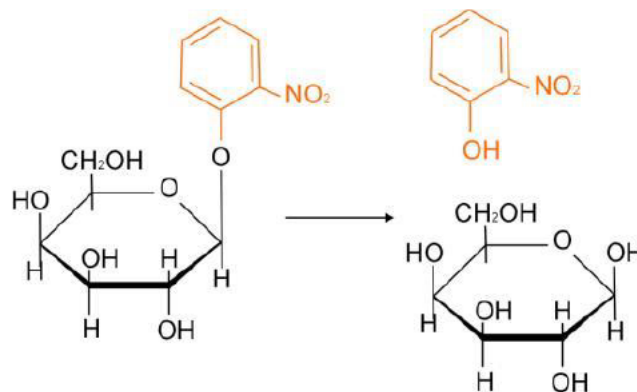
## Analytical methods

### Lactase activity analysis by Konelab (LAU(B))

#### Principle

Lactase, EC.3.2.1.23, hydrolyses ONPG, (o-NitroPhenyl  $\beta$ -D-galactopyranoside), and hereby ONP, (o-NitroPhenyl) is released.

o-NitroPhenyl has a yellow colour when in a alkaline solution and can be quantified at 405 nM.



#### Reaction conditions

Parameter [reaction 1]	Reaction conditions [reaction 1]
Temperature	30°C
pH	6.5
Substrate conc.	1.46 mg/ml
Enzyme conc.	0.0175-0.105 LAU(B)/ml
Reaction time	600 sec.
Concentration of Magnesium is critical for the analysis	1 mM MgSO <sub>4</sub> ·7H <sub>2</sub> O
Parameter [Stop]	Reaction conditions [Stop]
Concentration of Stop reagent	0.2 M Na <sub>2</sub> CO <sub>3</sub>
Parameter [Detection]	Reaction conditions [Detection]
pH	Approx. 12
Reaction time	126 sec.
Wave length	405 nM



## Definition of unit

The activity is determined relative to a Lactase standard.

## Method parameters

### Specificity

The method is specific for Lactases and other agents which are able to hydrolyse the substrate ONPG. Glycerol does not have an interfering effect in the method.

### Range

Normal range: 0.140 - 0.840 LAU(B)/ml

Calibration range: The same as the normal range.

### Limit of determination

The Limit of determination is 3.5 LAU(B)/g for a minimum preparation of 1 g sample dissolved in 25 mL (25 ml/g)

## Equipment

Equipment	
Konelab 30 Analyzer	Thermo Fisher Scientific
Diluter	e.g. Hamilton Microlab
Analytical balance	e.g. Sartorius, Mettler
Balance	e.g. Sartorius
pH meter	e.g. Radiometer, Metrohm
Magnetic stirrer plates	-

## Chemicals

Name	Chemical formula	Brand
ONPG ( <i>o</i> -NitroPhenyl $\beta$ -D-galactopyranoside)	$C_{12}H_{15}NO_8$	Sigma N1127 (store frozen up to 6 month after opening).
Magnesium sulphate heptahydrate	$MgSO_4 \cdot 7H_2O$	i.e Sigma 63140
MES (2-(N-morpholino)ethanesulfonic acid)	$C_6H_{13}NO_4S$	i.e Sigma 8250
Brij 35	$(C_2H_4O)_n C_{14}H_{22}O$	i.e Sigma B4184
Sodium carbonate	$Na_2CO_3$	i.e Sigma-Aldrich 31432
4N NaOH	NaOH	-
UltraPure water	$H_2O$ with resistivity $\geq 18.2$ M $\Omega$ *cm at 25 °C	e.g. MilliPore

Always check out the Material Safety Data Sheet (MSDS) for all the chemicals.

## Reagents

0.1 M  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$

Example: Preparation of 500 ml

Step	Action
1	Weigh out 12.33 g of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ .
2	Transfer to a 500 ml flask.
3	Fill to the mark by Ultrapure water.
4	Stir until all is dissolved.
5	Storability: 1 month at room temperature.

15% (w/v) BRIJ 35 stock solution

Example: Preparation of 2000 mL

Step	Action
1	Transfer 1000 mL 30% (w/v) Brij 35 to a 2000 mL graduated flask.
2	Rinse the BRIJ flask with Ultrapure water and transfer the water to the 2000 mL flask.
3	Fill up to 2000 mL with Ultrapure water.
4	Mix vigorously and pour in a labelled capped bottle.
5	Storability: 2 months at approx. 4° C in a refrigerator.

Sample MES buffer (0.05 M MES, 1 mM  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , 450 mg/l Brij 35, pH 6.5).

Example: Preparation of 10 L

Step	Action
1	Weigh out 97.60 g of MES.
2	Transfer to a 10 L flask and add approximately 9500 ml of Ultrapure water.
3	Add 100ml of 0.1M $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ . (Alternatively weigh out 2.466 g of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ )
4	Add 30 ml of Brij 35 stock solution 15 W/v%.
5	Add 80 ml 4N NaOH.
6	Stir until fully dissolved.
7	Adjust pH to $6.5 \pm 0.05$ by 4N NaOH.
8	Fill to the mark by UltraPure water.
9	Storability: 1 month at room temperature.

Substrate MES buffer (0.05 M MES, 1 mM  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , pH 6.5)

LAU(B)-Buf/S

Example: Preparation of 1 L

Step	Action
1	Weigh out 9.76 g of MES.
2	Transfer to a 1 L flask and add approximately 800 ml of Ultrapure water.
3	Add 10ml of 0.1M $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ . Alternatively weigh out 0.2466 g of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$
4	Add 8 ml 4N NaOH.
5	Stir until fully dissolved.
6	Adjust pH to $6.5 \pm 0.05$ by 4N NaOH.
7	Fill to the mark by UltraPure water.
8	Storability: 1 month at room temperature.

Substrate 1.67 mg/ml ONPG

LAU(B)-Sub/S

Preparation of 100 ml:

Step	Action
1	Weigh out $167 \pm 0.5$ mg of ONPG
2	Transfer to a 100 ml volumetric flask.
3	Fill up to 100 ml with Substrate MES buffer (0.05 M MES, 1 mM $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ , pH 6.5)
4	Stir until the ONPG is dissolved.
5	Storability: 1 month at 2-8 °C.

Stopsolution: 1 M  $\text{Na}_2\text{CO}_3$

LAU Stop/S

Example: Preparation of 1L

Step	Action
1	Weigh out 105.99 g of $\text{Na}_2\text{CO}_3$ .
2	Transfer to a 1L flask and add approximately 900 ml of Ultrapure water. Generates heat.
3	Stir until fully dissolved.
4	Fill to the mark by ultrapure water.
5	Storability: 2 months at room temperature.

## Standard

The standard is available upon request.

The standard is available upon request.

Step	Action																																
1	Weigh out an amount of enzyme standard corresponding to 4200 LAU(B)																																
2	Dissolve the standard in Sample MES buffer in a 500 ml measuring flask.																																
3	Stir on a magnetic stirrer for 15-60 minutes. Storability: 24 hours at room temperature.																																
4	<p>Working solutions: The stock solution is stirred/whirl mixed just before the final dilution is prepared in the Konelab cup. The standard curve is a 5 point curve with a factor 6 between lowest and highest standard point. Recommended total volume of Hamilton dilution is 1500 µL. The standard solutions are prepared by diluting the stock solution with Sample MES buffer on a diluter directly into the sample cups, according to this table:</p> <table><tr><th rowspan="2">Standard no.</th><th colspan="2">Example</th><th rowspan="2">Dilution ratio</th><th rowspan="2">Concentration (LAU(B)/ml)</th></tr><tr><th>Stock solution µl</th><th>Diluent µl</th></tr><tr><td>1</td><td>25</td><td>1475</td><td>60</td><td>0.140</td></tr><tr><td>2</td><td>60</td><td>1440</td><td>25</td><td>0.336</td></tr><tr><td>3</td><td>100</td><td>1400</td><td>15</td><td>0.560</td></tr><tr><td>4</td><td>125</td><td>1375</td><td>12</td><td>0.700</td></tr><tr><td>5</td><td>150</td><td>1350</td><td>10</td><td>0.840</td></tr></table> <p>Storability: Must be prepared directly before use.</p>	Standard no.	Example		Dilution ratio	Concentration (LAU(B)/ml)	Stock solution µl	Diluent µl	1	25	1475	60	0.140	2	60	1440	25	0.336	3	100	1400	15	0.560	4	125	1375	12	0.700	5	150	1350	10	0.840
Standard no.	Example		Dilution ratio	Concentration (LAU(B)/ml)																													
	Stock solution µl	Diluent µl																															
1	25	1475	60	0.140																													
2	60	1440	25	0.336																													
3	100	1400	15	0.560																													
4	125	1375	12	0.700																													
5	150	1350	10	0.840																													

## QC sample

The QC sample is available upon request.

Prepare a QC sample with known enzyme content on the same way as for the samples below.

## Samples

Step	Action
1	Weigh out 0.28 – 1.0 g of the sample precisely and dissolve in a measuring flask with Sample MES buffer. Use the same preparation of buffer that is used for the preparation of the standard and QC sample.
2	Stir for 15-60 minutes on a magnetic stirrer. Storability: 24 hours at room temperature
3	The samples are further diluted with Sample MES buffer to achieve an activity of approx. 0.49 LAU(B)/ml. Storability: Must be prepared directly before use

## Blank

No reagent blank is used in the method.

## Procedure

Step	Action
1	Stat up the Konelab
2	Place the reagents in the Konelab:

	Reagent	Konelab reagent name	Reagent container volume*	Syringe speed	Stability in reagent container
	1.67 mg/ml ONPG	LAU(B)-Sub/S	20 mL	Slow	1 day
	Substrate MES buffer	LAU(B)-Buf/S	20 mL	Slow	1 day
	1 M Na <sub>2</sub> CO <sub>3</sub>	LAU Stop/S	20 mL	Slow	1 day
3	Place standards, QC sample and samples in the Konelab in the mentioned order. <i>NOTE:</i> 30 samples can be analyzed in one analytical run.				
4	Start analysis on Konelab				

### Calculation

Step	Action
1	The activity of the enzyme samples is determined relative to the standard curve.
2	On the basis of the results in Abs for the five standards, a standard curve is drawn with the activities of the standards in LAU(B)/ml as the x-values and the Abs of the standards as the y-values. A linear algorithm is used.
3	<p>The enzyme activity of the diluted samples is read from the standard curve. The activity of the samples is calculated using the formula:</p> $\text{Activity LAU(B)/g} = \frac{S \cdot V \cdot F}{W}$ <p> S = Reading from the standard curve in LAU(B)/ml  V = Volume of the measuring flask used in ml  F = Dilution factor for second dilution  W = Weight of sample in g </p>
4	<p>EXAMPLE: 0.300 g sample is dissolved in a 250 ml measuring flask and further diluted 20 times using a diluter</p> <p>On the Equipment a Signal of 0.89091 is measured.</p> <p>From the standard curve an activity of 0.3276 Unit/ml is calculated.</p> $\text{Activity} = \frac{0.3276 \cdot 250 \cdot 20}{0.300} = 5460 \text{ LAU(B)/g}$

### Approval of analytical run

Approval of standard curve:

Parameter	Requirement
Quality of fit (lower R <sup>2</sup> limit)	R <sup>2</sup> > 0.9945 If R <sup>2</sup> < 0.9945 one standard point may be removed.
Curve appearance	The curve is an increasing linear curve.

Approval of QC sample:

The measured activity of the QC sample must be the declared value ± 2 · SD

Approval of samples

The analytical result (= average of 2 weighings on 2 standard curves) must be ≤ 2.5 %

### Statement of analysis results

The analysis result is stated with three significant digits.

Configurations  
LAU(B) test definition:

---  
Me  
Wa  
Me

**LAU(B)-SUB/s**

---

**LAU(B)-Buf/s**

**LAU(B) Stop/s**

## Handling of enzymes and chemicals

Enzymes and enzyme solutions should be handled in a fume hood or in closed containers.

Avoid inappropriate handling of enzymes and enzyme solutions, which may result in aerosol/dust generation.

Avoid inhalation of dust aerosols and contact with skin and eyes.

Handling of chemicals and disposal of waste must be performed according to valid procedures.

## Validity

Valid from December 2014

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### Novozymes A/S

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*Novozymes is the world leader in bioinnovation. Together with customers across a broad array of industries we create tomorrow's industrial biosolutions, improving our customers' business, and the use of our planet's resources. Read more at [www.novozymes.com](http://www.novozymes.com).*



# Enumeration of Total Viable Count

**Scope** All Novozymes Enzyme Business QC laboratories involved in analysis of samples from Novozymes production and GLP studies.

**Principle** **Total Viable Count (TVC)** is defined as the number of organisms which form colonies on a non-selective agar medium (Tryptic Soy Agar, TSA) after aerobic incubation for 3 days at 30-35°C. TSA is a rich non-selective agar medium.

The method outlined below conforms to the principles of (Ref. □) with the following exceptions:

- The test only covers the enumeration of microorganisms capable of growing on TSA (Total aerobic Microbial Count).
- The dilution water has an addition of 4% Tween 80.
- EP describes the use of duplicates. This method uses single tests.
- The agar plates are incubated for 3 days, not for 3-5 days.
- Growth promotion test of TSA is performed according to in-house procedures and not according to the description in EP.

Routine samples are analysed by the spiral plater (100 µl) or spread plate technique (100 µl or 1 ml) as described below:

Sample type	Requested test (LIMS code)	Technique	Volume spread	Lowest Dilution	No. of plates	Plate size	Detection limit
Enzyme samples and fluid hyaluronic acid	TVC	Spiral plating or spread plating	100 µl	10 <sup>-1</sup>	1 plate	9 cm	100 CFU / g or ml
	TVC(100)	Spread plating	1 ml	10 <sup>-1</sup>	4 plates	14 cm	10 CFU / g or ml
CIP-samples	CIP_TVC	Spiral plating or spread plating	100 µl	Undiluted	1 plate	9 cm	10 CFU / ml
		Petrifilm	1 ml		N/A	N/A	1 CFU / ml
FeF samples	FEF_TVC	Spread plating	1 ml	10 <sup>-1</sup>	4 plates	14 cm	10 CFU / g or ml

Depending on sample type, level of contamination and the detection limit needed for the specific sample, alternative procedures may be used.

**IMPORTANT:** Petrifilm must only be used to analyze CIP samples if pH of the CIP water is within range 6.6-8.5 (Ref. □ and □).

*Continued on next page*

## Enumeration of Total Viable Count, *Continued*

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**Definition of units**

The result is stated as:

- Total Viable Count (TVC) / g or ml
- 

**Samples**

All sample types.

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**Detection limit**

The detection limit of this method is dependent on the sample volume and the dilution in use (See "Principle").

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**Equipment**

- Balance ( $\pm 0.1$  g)
  - Magnetic stirrer
  - Petri dishes (9 cm or 14 cm)
  - Suitable sterile pipettes for transfer of 100  $\mu$ l or 1 ml (4x0.25 ml)
  - Spiral plater (for the spiral plate technique)
  - Sterile Drigalski spreaders (for the spread plate technique)
  - Incubator (30-35°C)
  - Stereo microscope or microscope
  - Plastic spreader (*Petrifilm test*)
- 

**Media and reagents**

- Tween 80 buffer 4%, 90 ml (if necessary with a magnet) prepared acc. to [EB-ME-0052](#)
  - EP buffer, 90 ml buffered sodium chloride-peptone solution pH 7.0, prepared acc. to [EB-ME-0067](#)
  - TSA plates (9 or 14 cm) prepared acc. to [EB-ME-0041](#)
  - 3M™ Petrifilm™ Aerobic Count Plates (*Petrifilm test*)
- 

**Safety**

It is the responsibility of the laboratory leader, that all personnel are aware of the correct handling of enzymes and reagents.

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*Continued on next page*

## Enumeration of Total Viable Count, *Continued*

### Sample preparation

Enzyme samples and other solid samples are prepared as follows:

Sample type	Action
<b>Enzyme samples</b> <b>FeF samples</b> <b>Other solid samples</b>	Transfer 10 g of solid sample or 10 ml of liquid sample to 90 ml Tween 80 buffer 4%.  <i>NOTE:</i> Immediately homogenize the sample by stirring or by shaking. Solid samples are homogenized on a magnetic stirrer for app. 20 minutes though min. 1 hour for Sweetzyme (batch code 1A).
<b>Non-enzyme fluid samples (e.g. CIP samples)</b>	Non-enzyme fluid samples are analyzed undiluted. If needed, 10-fold dilutions may be prepared with Tween 80 buffer 4%.
<b>Fluid hyaluronic acid (HA)</b>	Transfer 10 ml of liquid sample to 90 ml EP buffer. <i>IMPORTANT:</i> Homogenize on a magnetic stirrer for min. 1 hour. It is recommended to shake the sample after approx. 30 min.

*TIP:* All enzyme products must be analyzed from at least a  $10^{-1}$  dilution due to possible inhibition of microorganisms in undiluted enzyme. If an enzyme product is known to contain growth inhibiting components (e.g. rodalone or proxel) consider analyzing further dilutions prepared with Tween 80 buffer 4% (e.g.  $10^{-2}$  and  $10^{-3}$  dilutions). In this case be aware that the quantification limit is lower than the spec. limit of the sample.

*IMPORTANT:* Valid for US laboratories: TVC analysis must also be performed using a  $10^{-2}$  dilution if the spec. limit of the sample is > 30.000 and/or for samples from Recovery 1 and 2.

*Continued on next page*

## Enumeration of Total Viable Count, *Continued*

### Plating

Plating must be done within 15 minutes from end of homogenisation. If this is not possible, the sample can be stored at 2-8°C for up to 4 hours.

Test	Action
<b>TVC</b>	Transfer 100 µl from the 10 <sup>-1</sup> dilution onto the surface of a TSA plate (9 cm). Repeat this for any of the necessary dilutions. <i>Or</i> Perform a spiral plating of 100 µl from the 10 <sup>-1</sup> dilution in accordance with the directions for the specific spiral plater.
<b>TVC(100)</b> <i>or</i> <b>TVC_FeF</b>	Transfer 1 ml from the 10 <sup>-1</sup> dilution onto the surface of 4 TSA plates (14 cm) with app. 0.25 ml onto each plate. Repeat this for any of the necessary dilutions.
<b>TVC_CIP using TSA plates</b>	Transfer 100 µl from the undiluted sample onto the surface of a TSA plate (9 cm). Repeat this for any of the necessary dilutions. <i>Or</i> Perform a spiral plating of 100 µl from the undiluted sample in accordance with the directions for the specific spiral plater.

Leave the plates on the table with lid on until the sample has been soaked into the agar.

Test	Action
<b>TVC_CIP using Petrifilm</b>	1. Transfer 1 ml from the undiluted sample to the center of the film. 2. Place plastic spreader, recessed side down, on center of sample and press down, gently and firmly to distribute inoculum. 3. Wait at least one minute for gel to form

### Incubation

Incubate the TSA agar plates at 30-35°C for 3 days.

Incubate the Petrifilm with clear side up at 35-39°C for 2 days.

*Continued on next page*

## Enumeration of Total Viable Count, *Continued*

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### Reading

#### **TSA agar plates – Spread plate technique:**

Count the number of colonies on the plates.

Size of agar plate	Interval of reading
9 cm	1–300 colonies per plate
14 cm	1–750 colonies per plate

#### **TSA agar plates – Spiral plate technique:**

The number of typical colonies on each plate is counted and the result is calculated in accordance with the directions for the specific spiral plater. Danish sites may refer to (Ref. □).

*IMPORTANT:* Small colonies, e.g. lactobacillus, may erroneously be misread as product crystallizations. If in doubt use stereo microscope for macroscopic observation and/or prepare a slide culture of a colony for light microscopy.

#### **Petrifilm**

Count the number of colonies on the film. Interval of reading is 1-250 colonies (Ref. □).

*IMPORTANT:* Discoloration from enzyme residues may occur. In case this is observed the result must be considered invalid.

*TIP:* Refer to (Ref. □) to get familiarized with reading Petrifilms.

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*Continued on next page*

## Enumeration of Total Viable Count, *Continued*

### Calculation

#### General principles:

The calculation is based on the number of colonies ( $C_x$ ) on the plate, and the sample volume analysed ( $V_x$ ).

The result is stated with two significant figures (e.g.  $2.2 \times 10^1$ ).

When Using results from	Then the result is	Where
One dilution	$\frac{C_x}{V_x}$	$C_x$ = no. of colonies $V_x$ = volume analysed
2 or more dilutions	$\frac{C_1 + C_2}{V_1 + V_2}$	$C_1$ = no. of colonies in lowest dilution $C_2$ = no. of colonies in next dilution $V_1$ = volume analyzed in lowest dilution $V_2$ = volume analyzed in next dilution

**IMPORTANT:** When using more than one dilution, the numbers from each dilution are compared (the likelihood of product inhibitions, contamination of the sample, analytical errors etc. is considered). In general, the highest dilution is used. If the result is stated on the basis of other dilutions, the reason must be given in the raw data.

When the sample volume is 0.1 ml then  $V_x$  and  $C_x$  are:

Dilution	Undiluted	$10^{-1}$	$10^{-2}$
$V_x$	0.1 ml	0.01 ml	0.001 ml
$C_x$	No. of colonies on the plate	No. of colonies on the plate	No. of colonies on the plate

EXAMPLE:

EXAMPLE: Examples of calculating spread plate of 0.1 ml sample:

$C_x$	$V_x$ (g or ml)	Dilution	Result
0	0.01	$10^{-1}$	< 100 / g or ml
123	0.1	$10^0$	$\frac{123}{0.1} = 1.2 \times 10^3$ / g or ml
334	0.01	$10^{-1}$	> $3.0 \times 10^4$ / g or ml
253 24	0.01 0.001	$10^{-1}$ $10^{-2}$	$\frac{253+24}{0.01+0.001} = 2.5 \times 10^4$ / g or ml

*Continued on next page*

## Enumeration of Total Viable Count, *Continued*

### Calculation (*continued*)

When the sample volume is 1 ml (four 14 cm agar plates with 0.25 ml on each plate) then  $V_x$  and  $C_x$  are:

Dilution	Undiluted	$10^{-1}$	$10^{-2}$
$V_x$	1 ml	0.1 ml	0.01 ml
$C_x$	sum of colonies on the 4 plates	sum of colonies on the 4 plates	sum of colonies on the 4 plates

EXAMPLE: Examples of calculating spread plate of 1 ml sample:

$C_x$	$V_x$ (g or ml)	Dilution	Result
0	0.1	$10^{-1}$	$< 10 / \text{g or ml}$
123	1	$10^0$	$\frac{123}{1} = 1.2 \times 10^2 / \text{g or ml}$
426	0.1	$10^{-1}$	$\frac{426}{0.1} = 4.3 \times 10^3 / \text{g or ml}$
3134	0.1	$10^{-1}$	$> 3.0 \times 10^4 / \text{g or ml}$
853 84	0.1 0.01	$10^{-1}$ $10^{-2}$	$\frac{853+84}{0.1+0.01} = 8.5 \times 10^3 / \text{g or ml}$

NOTE: Calculation at Danish laboratories may follow:

- Spread plating of 1 ml: [PSL-MSP-0069](#)
- Spread plating of 100  $\mu\text{l}$ : [PSL-MSP-0082](#)
- Spiral plating of 100  $\mu\text{l}$ : [PSL-MSP-0075](#)

### Accuracy and precision

CV% (surface plating) = 25%

CV% (spiral plating) = 29%

REFERENCE: LUNA no. [2003-34435](#)

### Filing

All documentation should be filed in accordance with the local filing SOP.

*Continued on next page*

## Enumeration of Total Viable Count, *Continued*

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**Contingencies** All deviations from this SOP should be discussed with the Method Responsible Scientist and should be documented.

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**References**

- European Pharmacopoeia, Chapter 2.6.12. Microbiological examination of non-sterile products (Total viable aerobic count).
- [PSL-MSP-0075](#): Beregning ved anvendelse af spiralplater (In Danish).
- [PSL-TE-3001](#): Spiralplater (In Danish).
- LUNA No. [2010-19643-01](#): Validation of pH Range Adjustment for Water Samples Using Petrifilm.
- [3M Petrifilm Interpretation Guide](#)
- [3M Petrifilm™ Aerobic Count Plates](#)

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**Revision** Removed TVC-AIR samples. Analysis of air samples performed according to EB-SM-5001. Specified that Tween 80 is used throughout the document cf. [CISAR-PSL-Mik.Ba-64777](#). In the section "Principle", the amount analyzed and the detection limit when using Petrifilm have been corrected from 100 ul to 1 ml and from 10 to 1 CFU/ml, respectively.

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## Analytical methods

### Enumeration of coliform bacteria using violet red bile agar

#### IMPORTANT

This method is used for the analysis of glucose isomerase and liquid products (with the exception of liquid xylanase).

#### Principle

Coliform bacteria (coliforms) are broadly defined as gram-negative, oxidase-negative, nonsporogenous rods which grow in aerobic or facultative anaerobic conditions. More specifically, coliforms are capable of fermenting lactose (due to the production of galactosidase) in the presence of bile at 37°C. Coliforms are not a taxonomically defined group of bacteria, and consequently there is no common agreement on which microorganisms truly belong to the coliforms. However, Novozymes defines coliforms as organisms belonging to the genera *Escherichia*, *Citrobacter*, *Enterobacter*, *Klebsiella*, *Serratia*, and *Hafnia*. The presence of coliforms, especially *E. coli*, can be used as an indicator of the bacteriological hygiene of an enzyme product.

Violet Red Bile Agar (VRBA) is a selective and indicative agar:

Principle	Description
Selective principle	Crystal violet and bile salts inhibit growth, primarily of the gram-positive accompanying flora. This favors growth of the fast-growing gram-negative enterobacteria.
Indicative principle	Degradation of lactose to acid is indicated by the pH indicator neutral red, which changes its color to red and in some cases also by precipitation of bile acids. Coliforms degrade lactose.

Routine testing is performed in the following way:

Sample type	Requested test (LIMS code)	Technique	Volume spread	Lowest dilution	No. of plates	Plate size	Detection limit
Enzyme samples	COLIFORM	Pour plate with cover layer	2.5 ml	10 <sup>-1</sup>	1 plate	14 cm	4 CFU/g or ml

Depending on the sample type, the level of contamination, and the detection limit needed for the specific sample, alternative procedures may be used.

The method outlined below conforms to ISO 4832 with the following deviations:

- ISO 4832 and ISO 6887-1 describe the use of a peptone salt solution or buffered peptone water as diluent. This Novozymes method uses Tween buffer 4%
- ISO 4832 describes the use of duplicates. This Novozymes method uses single tests

### Definition of units

The result is stated as:

- Coliform bacteria/g or ml

### Detection limit

The detection limit of this method is dependent on the sample volume and the dilution in use (see "Principle").

### Equipment

- Balance ( $\pm 0.1$  g)
- Magnetic stirrer
- Petri dishes (14 cm)
- Suitable sterile pipette for transfer of 1 ml or 10 ml (2.5 ml)
- Incubator (34–38°C)

### Media and reagents

- Tween buffer, 90 ml buffered sodium chloride-peptone solution pH 7.0 (if necessary with a magnet) containing 4% Tween 80, pH 7.0
- Violet Red Bile Agar (VRBA), Merck 1.01406

NOTE: If the agar is freshly prepared in the laboratory, suspend the media with 200 ml of exchanged water and leave for 15 min. Ensure that the media are thoroughly dissolved before the melting procedure by regular shaking. In addition, stir the agar immediately before cooling in the water bath and again before pouring into the Petri dishes.

### Sample preparation

The samples are prepared as follows:

Step	Action
1	Transfer 10 g of solid sample or 10 ml of liquid sample into 90 ml of Tween buffer 4%
2	Immediately homogenize the sample by stirring or shaking. Solid samples are homogenized on a magnetic stirrer for approx. 20 minutes

IMPORTANT: All enzyme products must be analyzed from a 10<sup>-1</sup> dilution due to possible inhibition of microorganisms in undiluted enzyme.

TIP: Further 10-fold dilutions can be prepared with Tween buffer 4%.

## Plating

Plating is performed using the pour plate technique:

Description
1. Transfer 2.5 ml from the $10^{-1}$ dilution into an empty Petri dish (14 cm).
2. Pour approx. 40–45 ml of VRBA ( $47 \pm 2^{\circ}\text{C}$ ) into the Petri dish (= bottom layer) and mix carefully. Leave this to solidify.
3. Pour approx. 10 ml of VRBA ( $47 \pm 2^{\circ}\text{C}$ ) onto the bottom layer (= covering layer). Leave this to solidify.

IMPORTANT: Agar used for BB samples must be cooled to  $45 \pm 2^{\circ}\text{C}$ .

## Incubation

Incubate the plates at  $34\text{--}38^{\circ}\text{C}$  (target =  $36^{\circ}\text{C}$ ) for 1 day under aerobic conditions:

## Reading

Count the number of typical colonies:

Count colonies on plates with	Typical colonies
1–375 colonies per plate	Purplish red with a diameter of $\geq 0.5$ mm, sometimes surrounded by a reddish zone of precipitated bile.

## Calculation

### General principles:

The calculation is based on the number of colonies ( $C_x$ ) on the plate and the sample volume analyzed ( $V_x$ ).

The result is stated with two significant figures (e.g.,  $2.2 \times 10^1$ ).

When using results from	Then the result is	Where
1 dilution	$\frac{C_x}{V_x}$	$C_x$ = no. of colonies $V_x$ = volume analyzed
2 or more dilutions	$\frac{C_1 + C_2}{V_1 + V_2}$	$C_1$ = no. of colonies in lowest dilution $C_2$ = no. of colonies in next dilution $V_1$ = volume analyzed in lowest dilution $V_2$ = volume analyzed in next dilution

IMPORTANT: When using more than one dilution, the numbers from each dilution are compared (the likelihood of product inhibitions, contamination of the sample, analytical errors, etc., is considered). In general, the highest dilution is used. If the result is stated on the basis of other dilutions, the reason must be given in the raw data.

When the sample volume is 2.5 ml, then  $V_x$  and  $C_x$  are:

Dilution	$10^{-1}$	$10^{-2}$
$V_x$	0.25 ml	0.025 ml
$C_x$	No. of colonies on the plate	No. of colonies on the plate

EXAMPLE: Examples of calculating pour plate of 2.5 ml of sample on a 14 cm agar plate:

$C_x$	$V_x$ (g or ml)	Dilution	Result
0	0.25	$10^{-1}$	$\frac{0}{0.25} = < 4/\text{g or ml}$
3	0.25	$10^{-1}$	$\frac{3}{0.25} = 12/\text{g or ml}$
412	0.25	$10^{-1}$	$\frac{375}{0.25} = 1.5 \times 10^3/\text{g or ml}$
53 8	0.25 0.025	$10^{-1}$ $10^{-2}$	$\frac{53+8}{0.25+0.025} = 2.2 \times 10^2/\text{g or ml}$

## Accuracy and precision

CV% = 29%

## References

1. ISO 4832, 2nd ed. (1991) Microbiology – General Guidelines for the enumeration of coliforms – colony count technique.
2. ISO 6887-1, 1st ed. (1999) Microbiology of food and animal feeding stuffs – Preparation of test samples, initial suspensions and decimal dilutions for microbiological examination – Part 1: General rules for the preparation of the initial suspension and decimal dilutions

## Handling of enzymes and chemicals

Enzymes and enzyme solutions should be handled in a fume hood or in closed containers.

Avoid inappropriate handling of enzymes and enzyme solutions, which may result in aerosol/dust generation.

Avoid inhalation of dust aerosols and contact with skin and eyes.

Handling of chemicals and disposal of waste must be performed according to valid procedures.

## Validity

Valid from December 2011.

### Novozymes A/S

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## Analytical methods

### Detection of *Escherichia coli* (*E. coli*) in 25 g

#### Principle

*E. coli* is a Gram-negative, indole-positive, facultative anaerobic rod. It is considered to be a fecal indicator.

Detection of *E. coli* in 25 g is carried out as a qualitative analysis using nonselective enrichment in buffered peptone water (BPW) followed by isolation of  $\beta$ -D-glucuronidase-positive *E. coli* on a selective indicative agar medium (TBX agar).  $\beta$ -glucuronidase-negative *E. coli* strains (3–4%) form colorless colonies on TBX agar (e.g., *E. coli* O157). The detection of *E. coli* O157 is performed as ImmunoMagnetic Separation (IMS) using Dynabeads® anti-O157 and plating onto two selective indicative agar media (CT-SMAC agar and CHROMagar O157). Suspect *E. coli* O157 colonies are verified using the *E. coli* O157 latex test.

Suspect colonies from TBX agar and/or *E. coli* O157 latex-positive isolates from CT-SMAC agar and/or CHROMagar O157 are reported as *E. coli* detected in 25 g.

Important: The media used have the following characteristics:

Media	Characteristics
BPW broth	Nonselective broth.
TBX agar	<b>Selective properties:</b> Growth of accompanying Gram-positive flora is largely inhibited by the use of bile salts. <b>Indicative properties:</b> The presence of the enzyme $\beta$ -D-glucuronidase differentiates most <i>E. coli</i> spp. from other coliforms. <i>E. coli</i> absorbs the chromogenic substrate 5-bromo-4-chloro-3-indolyl- $\beta$ -D-glucuronide (X- $\beta$ -D-glucuronide). The enzyme $\beta$ -glucuronidase splits the bond between the chromophore 5-bromo-4-chloro-3-indolyl and the $\beta$ -D-glucuronide. <i>E. coli</i> colonies are colored blue-green. <b>NOTE:</b> For the recovery of sublethally injured <i>E. coli</i> , plates are incubated at 34–38 °C and not 44 °C as recommended by Merck (inhibits growth of accompanying Gram-positive flora).
CT-SMAC agar (Sorbitol MacConkey agar)	Polypeptone favors the growth of <i>E. coli</i> O157:H7. Sorbitol-negative bacterial (in particular O157:H7) colonies are colorless. Sorbitol-positive bacteria give rise to red colonies due to the change of the color of the pH indicator (neutral red). Contaminating bacteria are inhibited by the association of bile salts, crystal violet, cefixime, and potassium tellurite.
CHROMagar O157 and CT-CHROMagar O157	A typical <i>E. coli</i> O157 will grow as a pink-mauve colony, whereas most other microorganisms are either inhibited or grow as blue or colorless colonies.

## Definition of units

The result is stated as:

- DET (*E. coli* detected in 25 g) or
- ND (*E. coli* not detected in 25 g)

## Standards

A positive reference strain can be used (e.g., *E. coli* ATCC 11229).

If a reference strain of *E. coli* O157 is included, it must be *E. coli* O157 without the genes coding for verotoxins (e.g., ATCC 43888).

## Detection limit

Theoretical detection limit: 1 *E. coli* in 25 g

## Equipment

Balance ( $\pm 0.1$  g)

Magnetic stirrer

Incubator (34–38°C)

Sterile inoculation loops (1- $\mu$ l)

Sterile swabs

Vortex mixer

Pipettes and sterile tips

For ImmunoMagnetic Separation (either mIMS or aIMS):

- For manual ImmunoMagnetic Separation (mIMS):



- MPC-S rack and magnet (Invitrogen cat. no. 120.20) + Eppendorf tubes, 1.5-ml (Eppendorf cat. no. 0030 10.086) + MX-3 mixer (Dynal cat. no. 159.09) – mixer is optional.
- For automatic ImmunoMagnetic Separation (aIMS):
  - BeadRetriever™ (Invitrogen cat. no. 159-50) + tubes and tips (Invitrogen cat. no. 150-51)

## Media and reagents

Buffered peptone water (BPW) (450 ml)

Chromocult® TBX agar plates (9-cm)

Cefixime-tellurite Sorbitol MacConkey agar (CT-SMAC agar plates, 9-cm)

CHROMagar O157 agar plates (9-cm) or CT-CHROMagar O157 (app. 5- or 9-cm)

Tryptone soya agar plates (TSA)

Dynabeads® anti-O157, Dynal cat. no. 710.04

Washing buffer (PBS-Tween 20 buffer), Sigma no. P-3563

*E. coli* O157 Latex Test Kit (for verification), Oxoid no. DR620

## Safety

The *E. coli* O157 Latex Test Kit (Oxoid DR0620) is labeled R22 – Harmful if swallowed due to 0.1% sodium azide.

## Transfer of sample to BPW

25 g of sample is transferred to 450–900 ml of BPW, depending on the sample type

## Enrichment

The nonselective enrichment is performed as follows:

Incubate BPW at 34–38°C for 16–20 hr (minimum 16 hr).

## Detection of $\beta$ -D-glucuronidase-positive *E. coli*

Detection of  $\beta$ -D-glucuronidase-positive *E. coli* is performed as follows:

- Streak the enriched sample onto the surface of a TBX agar plate using a sterile 10- $\mu$ l inoculation loop. If using two BPW bottles, streak on one agar plate from each bottle
- Incubate the plate at 34–38°C for 18–24 hours
- Examine the plate for growth of typical *E. coli* colonies:

Organism	Growth on Chromocult® TBX agar
<i>E. coli</i>	Blue-green or dark-blue-to-violet colonies (Salmon-GAL and X-glucuronide reaction)
Coliforms (not <i>E. coli</i> )	Salmon-to-red colonies (Salmon-GAL reaction but no X-glucuronide reaction)
Other Gram-negatives	Colorless colonies, except for some organisms which possess $\beta$ -D-glucuronidase activity. These colonies appear light-blue to turquoise


## Detection of *E. Coli* O157

ImmunoMagnetic Separation (IMS) is performed either as manual IMS (mIMS) or automated IMS (aIMS):

**Manual IMS (mIMS):**

Step	Action
1	Place one Eppendorf tube per sample in the rack without the magnet inserted. Gently vortex the Dynabeads® anti-O157 and add 20 µl of Dynabeads® anti-O157 to each tube. Use a lid opener to open the lids of the Eppendorf tubes
2	Gently add 1 ml of the pre-enriched sample to the Eppendorf tube. Use a new pipette/tip for each sample. Close the lid. <i>NOTE:</i> If the sample is divided between two BPW bottles, take 500 µl from each bottle
3	Incubate the tubes for approx. 10 min at room temperature. Gently rotate the rack without the magnet on an MX-3 mixer (Dyna) or by hand
4	Insert the magnet into the rack. Tilt the rack frequently for approx. 3 minutes to ensure complete collection of beads. With correct capture, a distinct circular-to-oval brownish pellet is formed at the tube site halfway between the top and bottom of the tube
5	Open the tubes gently using the lid opener. Place a Pasteur pipette at the water surface opposite the pellet. Gently pipette up the supernatant and liquid in the cap of the tube. Slow down pipetting when the surface of the liquid passes the pellet in order to make sure that no beads leave the tube through the pipette. If beads leave the sample, return the supernatant to the tube and repeat step 4. Use a new pipette/tip for each sample
6	Carefully add 1 ml of washing buffer to each sample. Do not touch the tube with the pipette/ tip as this can cross-contaminate the samples as well as the buffer. Close the lids and remove the magnet from the rack. Wash the bead complex by rotating the rack three times. Repeat steps 4 to 6 twice, but the last time only resuspending the pellet in 100 µl of washing buffer

**Automatic ImmunoMagnetic Separation (aIMS):**

Step	Action
1	Load one sample tube for each sample into a sample rack.   <i>NOTE:</i> Each sample tube consists of five tubes called tubes 1 to 5 (tube 1 is to the left (= slip end) and tube 5 is to the right)
2	Gently vortex the Dynabeads® anti-O157 until the pellet in the bottom of the tube disappears, then aseptically add 10 µl of properly mixed Dynabeads® anti-O157 into sample tubes 1 and 2
3	Aseptically add 500 µl of washing buffer to sample tubes 1 and 2. Aseptically add 1000 µl of washing buffer to sample tubes 3 and 4. Aseptically add 100 µl of washing buffer to sample tube 5
4	Add 500 µl of the enriched test sample to sample tubes 1 and 2; be careful not to contaminate other tubes.



	If the sample is divided between two BPW bottles, take 500 µl from each bottle
<b>5</b>	Repeat step 4 for the remaining samples
<b>6</b>	Aseptically insert the sterile protective sample tip combs into the instrument
<b>7</b>	Insert the rack of filled tubes into the instrument and lock it in place
<b>8</b>	Check that everything is properly aligned. Close the instrument door
<b>9</b>	Select the EPEC/VTEC program sequence by scrolling with the arrow key, then press the Start button

### Streaking onto selective indicative agar plates:

Each IMS product (from mIMS or aIMS) is tested for the presence of *E. coli* O157 using selective indicative agar plates:

Step	Action
1	Gently vortex the pellet (IMS product)
2	Streak 50 µl of IMS product onto the surface of a CT-SMAC agar plate, then streak another 50 µl of IMS product onto the surface of a CHROMagar O157 plate (or a CT-CHROMagar O157 plate) as follows: Spread the bead-bacteria complex over one half of the plate with a sterile cotton swab. This ensures the break-up of the bead-bacteria complexes. Dilute further by streaking with a loop
3	Incubate the plates at 34–38°C for 18–24 hours

### Reading:

Agar	Description
CT-SMAC agar	On CT-SMAC agar, typical <i>E. coli</i> O157 colonies are transparent and almost colorless with a pale yellowish-brown appearance and a diameter of approx. 1 mm. Sorbitol-positive organisms form bright-red (pink) colonies. In some cases, suspect colonies are so few that they can only be recognized in the bacterial lawn in the primary streaking zone. In this case, subculture suspect colony material onto a new CT-SMAC agar plate. If the growth is too weak after 18–24 hr, the plates can be reincubated for up to 24 hr. In this case, representative sorbitol-negative colonies (transparent) should be verified using the <i>E. coli</i> O157 Latex Test Kit from Oxoid (see below)
CHROMagar O157 and CT-CHROMagar O157	A typical <i>E. coli</i> O157 will grow as a pink-mauve colony, whereas most other microorganisms are either inhibited or grow as blue or colorless colonies

### Verification of *E. coli* O157:

Suspect colonies on CT-SMAC agar and CHROMagar O157 (or CT-CHROMagar O157) are verified as *E. coli* O157 using the *E. coli* O157 Latex Test Kit from Oxoid. The verification is performed according to the manufacturer's description.

### Interpretation of results

*E. coli* detected (DET) in 25 g

- Presence of typical colonies on TBX agar
- Presence of O157 latex-positive colonies from CT-SMAC agar and CHROMagar O157 (or CT-CHROMagar O157), i.e., suspect *E. coli* O157

*E. coli* not detected (ND) in 25 g

- Absence of typical colonies on TBX agar

- Absence of O157 latex-positive colonies from CT-SMAC agar and CHROMagar O157 (or CT-CHROMagar O157), i.e., suspect *E. coli* O157

## Sensitivity and specificity

Sensitivity: 100%

Specificity: 100%

## References

ISO 16649-2, 1st ed. (2001) Microbiology of food and animal feeding stuffs – Horizontal method for the enumeration of presumptive *Escherichia coli* – Part 2: Colony-count technique at 44°C using 5-bromo-4-chloro-3-indolyl- $\beta$ -D-glucuronic acid.

ISO 16654, 1st ed. (2001): Microbiology of food and animal feeding stuffs – Horizontal method for the detection of *Escherichia coli* O157.

The detection of *E. coli* O157 is in accordance with ISO 16654 with the following exception:

- Enrichment is performed using buffered peptone water at 34–38°C for 16–20 hours. ISO 16654 uses a modified TSB + novobiocin at 41.5°C  $\pm$  1°C for 18–24 hours
- ImmunoMagnetic Separation is only performed after 16–20 hours  
ISO 16654 states after 6 hr and again, if necessary, after 12–18 hours (i.e., a total elapsed time of 18–24 hours)
- Verification is performed using the *E. coli* O157 latex test. ISO 16654 states the indole test and serological test

## Handling of enzymes and chemicals

Enzymes and enzyme solutions should be handled in a fume hood or in closed containers.

Avoid inappropriate handling of enzymes and enzyme solutions, which may result in aerosol/dust generation.

Avoid inhalation of dust aerosols and contact with skin and eyes.

Handling of chemicals and disposal of waste must be performed according to valid procedures.

## Validity

Valid from November 2011.

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## Analytical methods

### Detection of *Salmonella* spp.

#### Principle

Detection of *Salmonella* spp. is carried out as a qualitative test.

The test is based on a non-selective enrichment of 25 g of sample in 450 ml of buffered peptone water for 18-24 hours followed by *Salmonella* specific PCR. Optionally, a secondary enrichment in the selective RVs broth may be added after enrichment in BPW. The methods are in-house methods evaluated and validated at Novozymes.

#### Definition of unit

The result is stated as:

DET (*Salmonella* detected in 25 g)

ND (*Salmonella* not detected in 25 g)

#### Standards

A positive reference strain can be included in the test, e.g., *Salmonella adabraka*, *Salmonella havana*, or *Salmonella senftenberg*.

#### Detection limit

Theoretical detection limit: 1 *Salmonella* sp. in 25 g.

#### Equipment

General equipment	
Balance	-
Incubator for BPW and agar plates	(34–38°C)
Incubator or water bath for RVs	(40.0–42.0°C)
Vortex mixer	
Automatic pipettes and sterile tips	10-100 µl, 100-1000 µl, and 1 ml

<b>PCR specific equipment and materials</b>	
AB 7500 Fast Real-Time PCR System	-
Microcentrifuge	E.g. Ole Dich microcentrifuge
Heating block	E.g. Stuart Block Heater SBH200D
Automatic pipette dedicated to PCR	10-100 µl
Automatic pipette dedicated to PCR	100-1000 µl
Pipette tips dedicated to PCR, DNA and DNase free	100 µl and 1000 µl
Sterile pasteur pipettes	-
Powder free gloves (PCR)	-
FastReactionTubes 0,1ml 8/strip	-
Tube Cap Strips 8 Caps/Strip	-
Tubes RNase-Free 1.5mL	-

Mesia and reagents

#### **Enrichment broths**

Buffered Peptone Water (BPW) (450 ml)

Optional: Rappaport Vassiliadis soya peptone broth (RVs broth) (Oxoid CM0866)

#### **PCR specific reagents**

MicroSEQ® *Salmonella* spp. Detection Kit (Life Technologies Cat. No. 4403930).

PrepSEQ™ Rapid Spin Sample Preparation Kit (Life Technologies Cat. No. 4407760)

EP buffer, 90 ml buffered sodium chloride-peptone solution pH 7.0.

Nuclease-Free Water (e.g. Sigma Cat. No. 101210442)

#### **Nonselective enrichment**

The nonselective enrichment is performed in the following way:

<b>Step</b>	<b>Action</b>
1	Transfer 25 g or 25 ml of sample to 450-ml BPW preheated to 40-42°C
2	Incubate BPW at 34-38°C for 18-24 hours

#### **Optional: Selective enrichment in RVs broth**

The selective enrichment in RVs is performed in the following way:

- Transfer 500 µl or 0.5 ml from BPW to 10-ml RVs tubes equilibrated to minimum room temperature
- Incubate the RVs broth at 40.0–42.0°C for 22-26 hours

Note: If a water bath is used to incubate the RVs, there is no need to equilibrate the temperature of the broth.

#### **DNA purification**

Cautions: When performing DNA purification there is a cross contamination risk, therefore, the following precautions should be taken:

- Set up only the materials and reagents needed for the particular work process in the LAF bench
- Always use a new pipette tip for each sample at each step
- Always handle one sample at a time keeping the remaining samples physically separated
- When opening reagent bottles put lids/caps upside down behind the bottle

- Always keep open bottles/reagents separate from the waste bin and not in the path where the pipette is transferred
- Always close (eppendorf) tubes when they are not handled
- Avoid passing your hands and pipettes above open bottles/tubes

IMPORTANT: Work in LAF bench when performing DNA purification and use gloves when performing lysis procedure.

IMPORTANT: Mix any reagents before use.

1. Mix BPW or RVs broth

CAUTION: Be careful that the broth does not touch the lid when mixing.

2. Transfer 2 ml BPW broth (or optionally RVs broth) to a 2 ml eppendorf tube and microcentrifuge (hereafter named centrifuge) for 3 minutes at max speed, e.g. 15.000g

TIP: Store the BPW or RVs broth at a cool temperature (2-8°C) until PCR has been successfully completed. The RVs broth may be stored for max. 2 days at a cool temperature (Ref.**Error! Reference source not found.**).

3. Discard the supernatant without touching the pellet, e.g. using a sterile pasteur pipette

4. Add 650 µl EP buffer and re-suspend the pellet thoroughly

5. Insert a spin column into a labeled tube

6. Load 650 µl of sample onto the spin column and cap the column

7. Load the tube into the centrifuge. Make sure the lid points toward the center of the centrifuge. Then centrifuge for 3 minutes at max speed, e.g. 15.000g

8. Remove the tube from the centrifuge, and then discard the used spin column

CAUTION: Make sure that any liquid on the outside of the spin column is scraped off on the edge of the eppendorf tube.

9. Aspirate, and then discard the supernatant

10. Add 50 µl of Lysis Buffer to the pellet. Re-suspend by pipetting up and down, or vortex until the pellet is re-suspended

11. Cap the tube, and then incubate at 95±3°C for 10 minutes

12. Allow the sample to cool for 2 minutes at room temp, then centrifuge for 1 minute at max. speed, e.g. 15.000g

13. Add 250 µl of Nuclease-Free water, then centrifuge for 1 minute at max. speed, e.g. 15.000g

14. Proceed with PCR, or store the tube at ±18°C. *Remark:* Avoid loading the black pellet when transferring to the lyophilized qPCR strip sample

TIP: Material may be stored at cool (2-8°C) for max. 2 hours after completion of step 3, 8 or 13.

### **PCR preparation**

IMPORTANT: Use gloves or wash your hands thoroughly after the PCR preparation. The negative control contains 0 – 0.01 % Na-azide.

IMPORTANT: Use a Pathogen Detection Negative Control for each PCR run.

1. Open the storage pouch containing the assay beads (MicroSEQ® *Salmonella* spp. Detection Kit)

IMPORTANT: Do not remove the desiccant from the storage pouch.

2. Remove the appropriate number of individual tubes or 8-tube strips

3: *NOTE:* Frozen samples and/or controls only: thaw these completely, vortex, then briefly spin them down using a microcentrifuge

4. Examine the assay beads in the 8-tube strips. Gently tap the tubes as needed to settle all assay beads to the bottom of each tube

5. Gently remove, and then discard the concave caps. Avoid disturbing the beads from the bottom of the tubes

6. For each sample or control, transfer 30 µl into a tube containing the appropriate assay beads. Beads dissolve in 1 to 5 seconds.

IMPORTANT: Dispense all unknown samples first followed by the negative control

7. Add additional tubes as needed so that each strip contains a full set of 8 tubes

8. Cap the tubes, sealing each tube with the flat (transparent) optical strip caps provided in the kit. Cap the tubes firmly with the strip cap tool to avoid collapsing, bending, or misaligning the tubes.

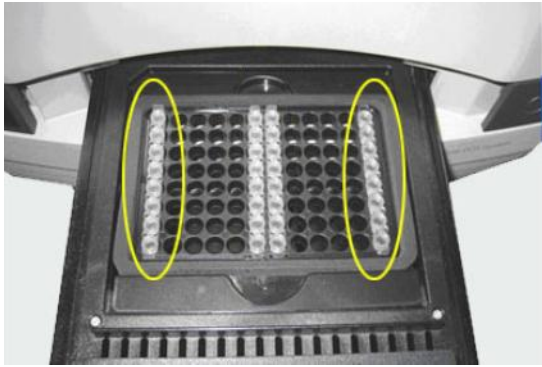
IMPORTANT: Avoid scratching the optical strip caps as this will interfere with the reading of fluorescence

9. Confirm that the strips are straight and that each tube is in line with the adjacent tube

10. Make sure reagents are thoroughly mixed and at the bottom of the tubes

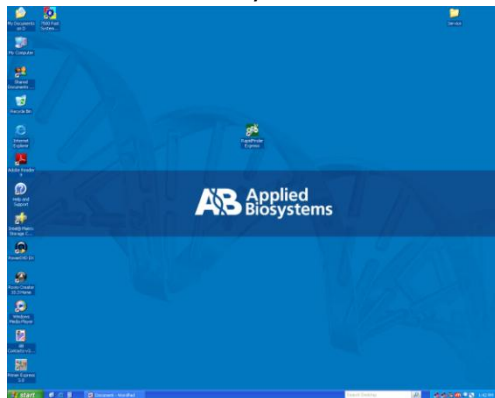
11. Carefully insert two or more 8-tube strips containing samples, starting from the center of the plate holder and moving out. This layout minimizes bending or misaligning the tube strips

12. If column 1 (leftmost) and column 12 (rightmost) of the Plate Holder are not used, insert two fully capped, empty, 8-tube strips into these columns (see below photo)

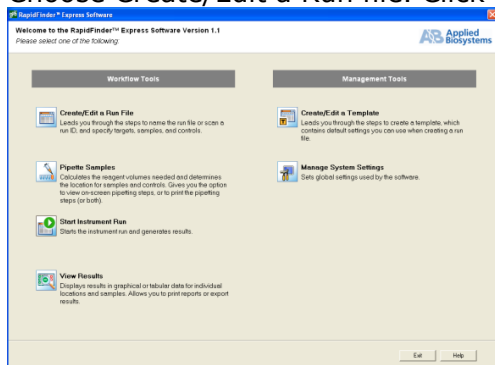


## Run PCR reactions

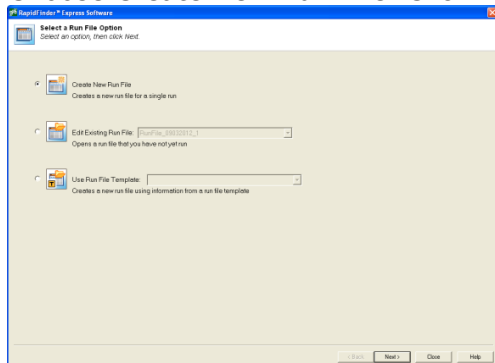
1. Turn on the PCR system first. Then turn on computer and open "RapidFinder Express"



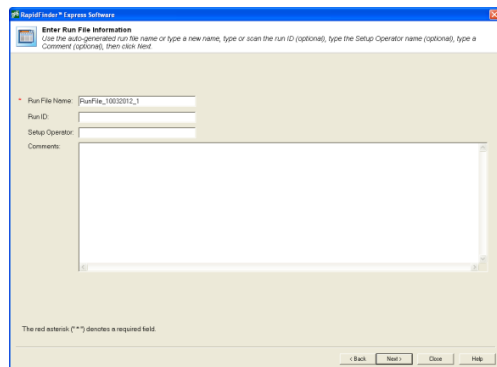
2. Choose Create/Edit a Run file. Click next



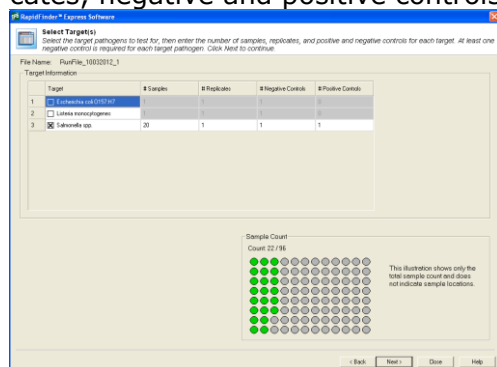
3. Choose Create New Run File. Click next



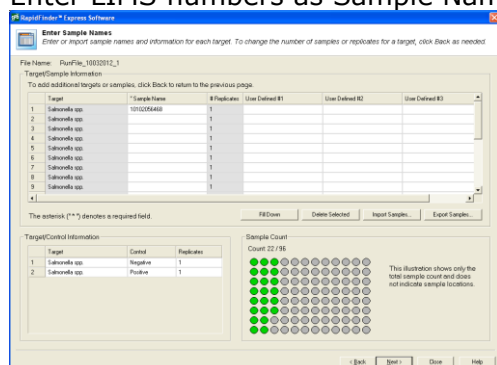
4. Enter Run File Information. Use default Run File Name and enter initials for Setup Operator. Click next



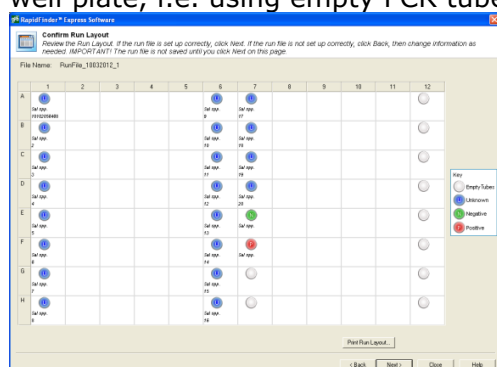
5. Select Targets: Choose *Salmonella* spp. Enter number of samples. Enter '1' for replicates, negative and positive controls, respectively. Click next



6. Enter LIMS numbers as Sample Names. Click next

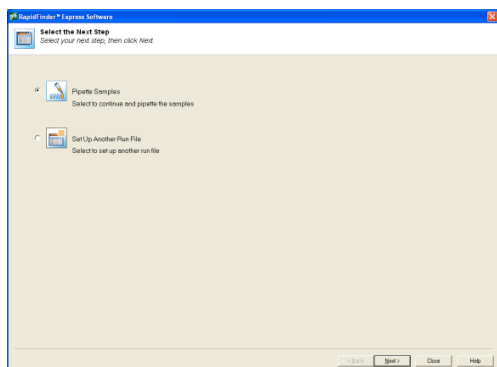


7. Confirm Run Layout: Make a layout of the program securing equilibrium on the 96-well plate, i.e. using empty PCR tubes. Click next

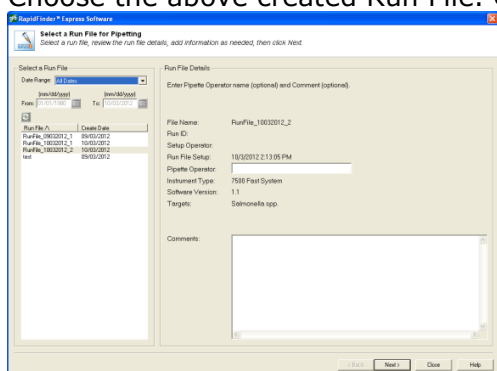


8. Choose Pipette Samples. Click next

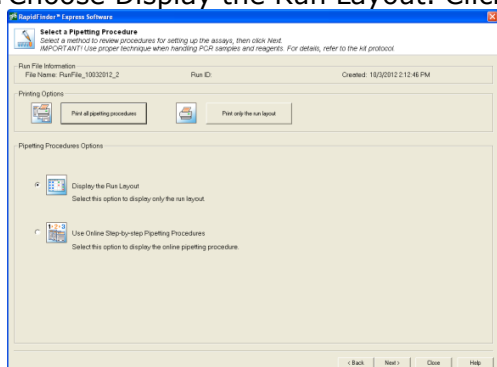




9. Choose the above created Run File. Click next

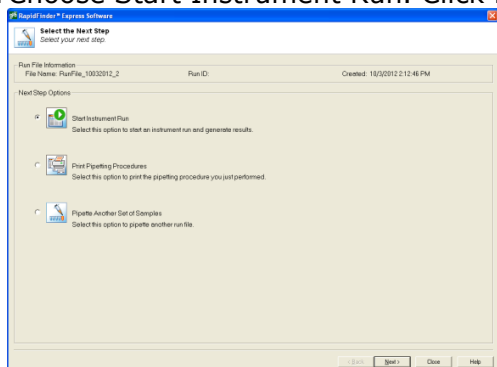


10. Choose Display the Run Layout. Click next



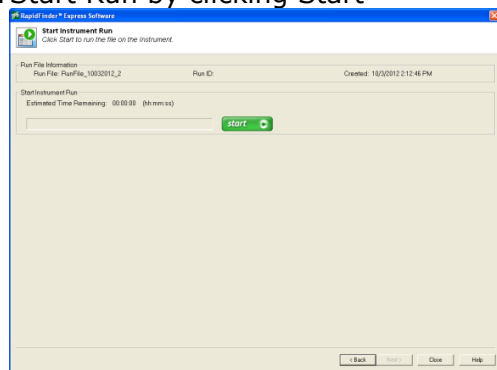
11. Confirm Run Layout. Click next

12. Choose Start Instrument Run. Click next

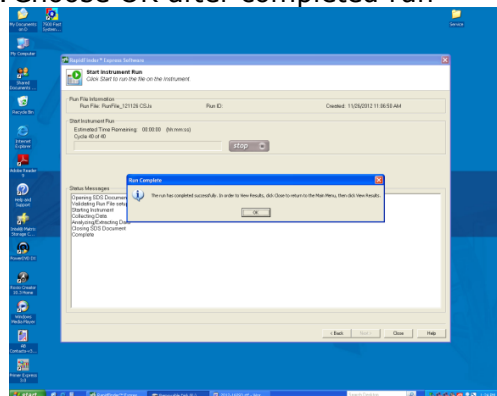


13. Load PCR tubes

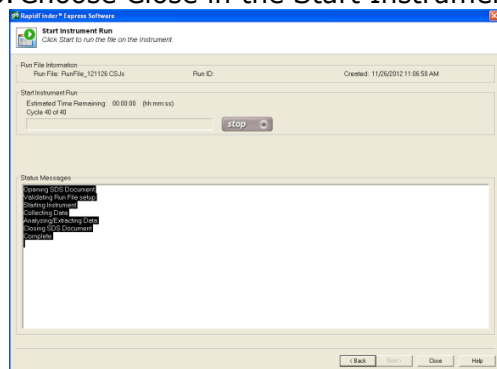
#### 14. Start Run by clicking Start



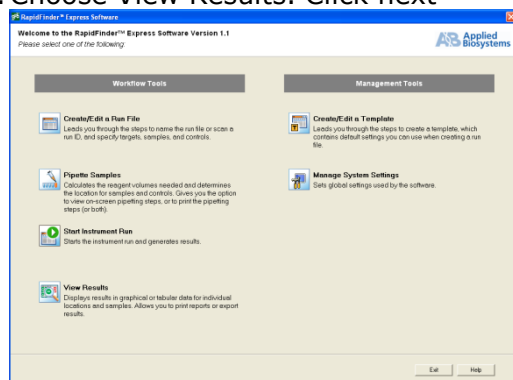
#### 15. Choose OK after completed run



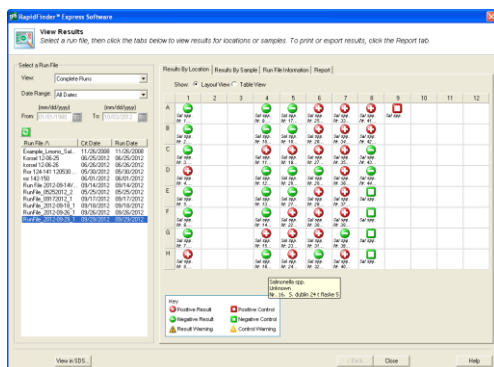
#### 16. Choose Close in the Start Instrument Run window



#### 17. Choose View Results. Click next



#### 18. Choose relevant Run File and the sheet "Results By Location"



## 19. Interpretation of results

If...	Then report result as...
Negative control is marked as "+"	Re-perform PCR procedure with all "+" samples and negative controls
Sample is marked as "-"	<i>Salmonella</i> spp. not detected (ND)
Sample is marked as "+"	<i>Salmonella</i> spp. detected (DET)

TIP: The threshold value for a *Salmonella* positive sample is 35.69 cycles.

Note: If a positive PCR result is obtained, cultivation may be performed from the BPW or RVs broth if verification of the *Salmonella* type is desired. See flow chart or (Ref. **Error! eference source not found.**).

20. Click Close then Exit to close the RapidFinder Express software

21. Choose Shutdown to turn computer off, then turn off PCR system

**IMPORTANT:** AFTER PCR run:

- **NEVER EVER** open tubes
- Throw tubes in the trash in the PCR room. Do not re-use tubes
- Before leaving the room:
  - Remove and throw gloves in the trash
  - Wash hands

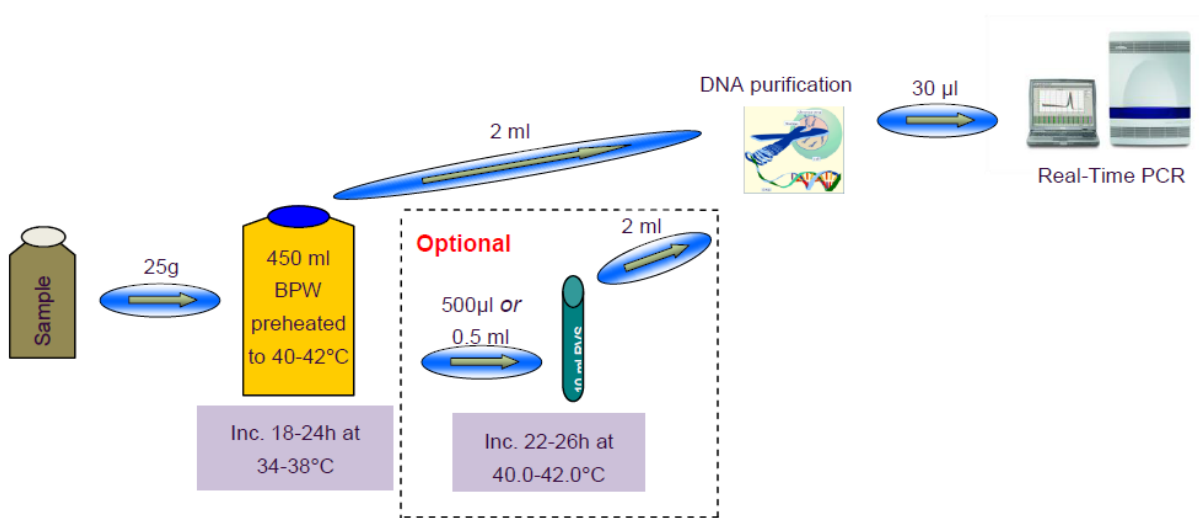
## Accuracy, sensitivity and specificity

Accuracy: 100%

Sensitivity: 100%

Specificity: 100%

## Flow chart:



## Handling of enzymes and chemicals

Enzymes and enzyme solutions should be handled in a fume hood or in closed containers. Avoid inappropriate handling of enzymes and enzyme solutions, which may result in aerosol/dust generation.

Avoid inhalation of dust aerosols and contact with skin and eyes.

Handling of chemicals and disposal of waste must be performed according to valid procedures.

## Validity

Valid from March 2013.

---

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## Detection of Antimicrobial activity

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**Scope** All Novozymes QC laboratories involved in analysis of samples from Novozymes production and GLP studies.

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**Principle** **Detection of Antimicrobial activity** is based on the measurement of inhibition of bacterial growth under specific circumstances.  
The method is in accordance with JECFA (1992)

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**LIMS code** ANTIMIC

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**Definition of units** The result is stated as:

- DET (Antimicrobial activity detected) or
- ND (Antimicrobial activity not detected)

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**Samples** All sample types.

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**Standards** *Staphylococcus aureus*, ATCC 6538  
*Escherichia coli*, ATCC 11229  
*Bacillus cereus*, ATCC 2  
*Bacillus circulans*, ATCC 4516  
*Streptococcus pyogenes*, ATCC 12344  
*Serratia marcescens*, ATCC 14041  
**NOTE:** The test organisms must be traceable.

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**Detection limit** Not known.

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*Continued on next page*

## Detection of Antimicrobial activity, *Continued*

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### Equipment

Balance ( $\pm 0.1$  g)  
Sterile pipettes for transfer of 100  $\mu$ l, 1 ml and 10 ml  
Inoculation loops 1  $\mu$ l  
Paper discs, e.g. S&S Analytical Filter Papers No. 740-E (12.7 mm in diameter), autoclaved  
Bio Safety Cabinet, Class II  
Sterile gloves  
Refrigerator (2-8°C)  
Incubator (34-38°C)  
-80°C freezer  
Ruler or Vernier gauge  
Petri dishes, 9 cm

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### Media and reagents

Tween buffer 4%  
Tryptone Soya agar (TSA), 90 ml in 250 ml Blue cap bottles  
Tryptone Soya agar plates, 9 cm with app. 15 ml agar (TSA)  
CASO broth, 50 ml  
*IMPORTANT:* Preparation in the local laboratory shall be done according to the current valid WW Media direction.  
Ciprofloxacin discs (5  $\mu$ g or 10  $\mu$ g) (bought ready to use).

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### Safety

It is the responsibility of the laboratory leader that all personnel are aware of the correct handling of enzymes and reagents.

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*Continued on next page*

## Detection of Antimicrobial activity, *Continued*

**Day 1:** Handling the test organisms must be performed in a Bio Safety Cabinet, Class II.  
**Preparation of test organisms in CASO**

Step	Action
1	Inoculate each of the 6 test organisms using a 1 µl inoculation loop (the strains are taken directly from a Cryo tube that has been stored in a -80°C freezer) in separate CASO broth, 50 ml.
2	Contemporary, streak out each test organism, using the same inoculation loop as in step 1, on the surface of a TSA plate to look for purity.
3	Incubate the CASO broth and TSA plates overnight at 34-38°C.

**Day 2:** The number of test organisms is tested in each of the CASO broths.  
**Number of test organisms in CASO**

Step	Action
1	Make a 10 <sup>-4</sup> dilutions of the following test organisms: <ul style="list-style-type: none"><li>• Bacillus cereus</li><li>• Bacillus cirkulans</li></ul>
2	Make a 10 <sup>-5</sup> dilutions of the following test organisms: <ul style="list-style-type: none"><li>• Staphylococcus aureus</li><li>• Escherichia coli</li><li>• Streptococcus pyrogenes</li><li>• Serratia marcescens</li></ul>
3	Determine the Total viable count of each dilution by spread plate or spiral plate on TSA plates. Incubate overnight at 34-38°C

*Continued on next page*



## Detection of Antimicrobial activity, *Continued*

### Day 2: Infection control (purity of the Cryo tubes)

Purity of the test organisms from the Cryo tubes are controlled the following way:

Step	Action
1	Control the purity of the 6 test organisms on TSA plates from the day before.
2	Write down the result (+ or – infection).

### Day 2: Preparation of test plates and purity of the test organisms in CASO broth

Preparation of test plates must be done in a Bio Safety Cabinet, Class II and wearing sterile gloves.

Step	Action
1	For each test organism a bottle containing 90 ml of Tryptone Soya agar (TSA) is melted and cooled (to app. 47°C)
2	Transfer 10 ml of CASO broth inoculated with <i>S. aureus</i> to a 250 ml Blue cap bottle with 90 ml melted and cooled Tryptone Soya agar (TSA). Mix carefully.
3	Pour app. 10 ml of the TSA-microorganism mixture onto an already prepared TSA plates (containing app. 15 ml TSA). Distribute the TSA-microorganism mixture evenly on the surface of the TSA plate, and allow solidifying.
4	Make another 9 plates as described in step 3.
5	Control the purity of the CASO broth by streaking out from the last drop of the bottle with a 1 µl inoculation loop onto the surface of one TSA plate.
6	Repeat step 2-5 for the rest of the microorganisms. <i>IMPORTANT:</i> Transfer only 5 ml of the CASO broth containing <i>Streptococcus pyogenes</i> to 90 ml of melted and cooled TSA.
7	Incubate the TSA plates prepared in step 5 overnight at 34-38°C.

*Continued on next page*

## Detection of Antimicrobial activity, *Continued*

### Day 2: Control of test plates with Ciprofloxacin

Control the test plates with Ciprofloxacin to determine whether the test organisms are capable of making an inhibition zone on the test plate.  
The control must be done on one test plate per test organism.

Step	Action
1	Put one disc of Ciprofloxacin onto the middle of a test plate.
2	Place the test plate overnight at 2-8°C.
3	Incubate the test plate overnight at 34-38°C.

### Day 3: Purity in CASO broth

Check the purity of the TSA plates from the day before.  
Write down the result of the purity test for each of the test organisms (+ or – infection).

### Day 3: Number of colonies on TSA plates

Count the number of colonies on the TSA plates from the day before.  
Write down the results.  
*IMPORTANT:* To approve the test plates all readings must be  $>10^6$  CFU.

### Day 3: Reading of in- hibition zone

Reading of the TSA plates with Ciprofloxacin from the day before is done by measuring the diameter of the inhibition zone on each of the test plates using a ruler or a Vernier gauge.  
Write down the results.  
*IMPORTANT:* Each zone must be  $\geq 25$  mm.

### Sample preparation

The samples are prepared as followed:

- Transfer 10 g of solid sample or 10 ml of liquid sample to 90 ml Tween buffer 4%
- Immediately homogenize the sample by stirring or by shaking. Solid samples are homogenized on a magnetic stirrer for app. 20 minutes

*Continued on next page*

## Detection of Antimicrobial activity, *Continued*

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### Test procedure

The test is performed in the following way:

*TIP:* It is recommended to work with dry plates, and to place the disc on the test plate immediately before addition of sample so the filters do not absorb moisture from the plates and thus cannot absorb the sample.

Step	Action
1	Place a sterile paper disc on each of the 6 test plates (one test plate per micro organism). <i>NOTE:</i> Up till 5 sterile paper discs can be placed on one plate (giving the possibility of analysing up to 5 samples per set of 6 test plates).
2	Inoculate each paper disc with 100 µl of the 10 <sup>-1</sup> dilution of the sample prepared above.
3	Place the plates overnight at 2-8°C.
4	Incubate the plates overnight at 34-38°C.
5	Measure the diameter of the inhibition zone on each of the plates using a ruler or a Vernier gauge.
6	Write down the results (inhibition zone in mm.).

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*Continued on next page*

## Detection of Antimicrobial activity, *Continued*

**Interpretation of results** Results are given according to the sheet below:

Is there...	with a zone measuring...	...the result is
0 inhibition zones	0 mm	Not detected (ND)
X inhibition zones	<16 mm	Not detected (ND)
1 inhibition zones	≥16 mm	Not detected (ND)
2 inhibition zones	≥16 mm	Not detected (ND)
3 inhibition zones	≥16 mm	Detected (DET)

If the result is Detected (DET) a remark is given on which of the test organisms that shows obvious antimicrobial activity in the sample and the size of the zone is stated.

**IMPORTANT:** If the result is Detected (DET) the Responsible Scientist is contacted.

**Accuracy and precision** Not validated.

**Archiving** All documentation should be archived in accordance with the local archiving SOP.

**Contingencies** All deviations from this SOP should be discussed with the Method Responsible Scientist and should be documented.

**References** Joint FAO/WHO Expert Committee on Food Additives (JECFA). Compendium of food additive specifications, Volume 1, Rome 1992, appendix A to annex 1.

**Revision** Both 5 µg and 10 µg Ciprofloxacin discs can be used (Luna no. 2008-31511)

## Detection of production strains

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### Scope

All Novozymes QC laboratories involved in analysis of samples from Novozymes production and GLP studies.

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### Principle

**The production strain** is defined as the organism used for fermentation of a given Novozymes product. Agar media and incubation conditions used for detection of a specific production strain is listed in [BD 001-IN-000](#)

**The reference strain** is defined as an isolate of the production strain used in the laboratory as a reference during the analysis.

Strains not listed in [BD 001-IN-000](#) are detected according to specific **Analytical Directions** prepared and approved by the EB Method Responsible Scientist. Analytical Directions are typically used in connection with GLP studies.

When analyzing samples from Novozymes production, the detection is carried out by spread plating of 0.1 g or 0.1 ml of sample.

When analyzing samples from GLP studies, the detection is carried out by spread plating or enrichment of 1 g of sample acc. to the specific Analytical Direction.

Detection of morphologically typical colonies (compared with the reference strain) indicates the presence of the production strain.

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### Definition of units

When analyzing samples from Novozymes production, the result is stated as:

- DET (The productions strain detected in 0.1 g or 0.1 ml) *or*
- ND (The productions strain not detected in 0.1 g or 0.1 ml)

When analyzing samples from GLP studies, the result is stated as:

- DET (The productions strain detected in 1 g) *or*
- ND (The productions strain not detected in 1 g)

**IMPORTANT:** When detected, the app. number of production strain / g or ml is stated.

---

### Samples

Novozymes products

---

*Continued on next page*

## Detection of production strains, *Continued*

---

### Detection limit

The detection limit of this method is dependent on the sample volume and the dilution in use.

Sample volume	Size and number of agar plates	Dilution	Detection limit
1 ml, spread plate	14 cm (4 plates)	10 <sup>-1</sup>	10 colonies / g or ml
10 ml, spread plate	14 cm (40 plates)	10 <sup>-1</sup>	1 colonies / g or ml

---

### Equipment

Balance (± 0.1 g)  
Magnetic stirrer  
Petri dishes (14 cm and 9 cm)  
Suitable sterile pipettes for transfer of 10 ml, 1 ml (4x0.25 ml) and 0.25 ml  
Sterile Drigalski spatula  
Incubator  
(relevant incubation temperatures are listed in [BD 001-IN-000](#))

---

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## Detection of production strains, *Continued*

### Media and reagents for Bacterial strains

Dilution buffer: Tween buffer 4%, 90 ml (If necessary, with a magnet) prepared acc. to [EB-ME-0052](#)

Agar media:

Abbreviation	Full name	Prepared acc. to EB Media direction (link)	Purpose
AT-2	AT-2 agar	<a href="#">EB-ME-0001</a>	Detection & verification
B-TSA	Basic Tryptic Soy Agar	<a href="#">EB-ME-0055</a>	Detection
B-TSA w.CAM	Basic Tryptic Soy Agar with or without Chloramphenicol (CAM) <i>NOTE:</i> The addition of CAM is optional	<a href="#">EB-ME-0056</a>	Detection
Schaeffers	Schaeffers agar	<a href="#">EB-ME-0036</a>	Verification
Sch.starch	Schaeffers agar with 1% starch	<a href="#">EB-ME-0037</a>	Verification
Skim milk	Tryptic Soy Agar with 1 % skim milk	<a href="#">EB-ME-0038</a>	Verification
TBX w.AMP	Chromocult®TBX agar + ampicillin (100 mg/l)	<a href="#">EB-ME-0066</a>	Detection
TSA	Tryptic Soy Agar	<a href="#">EB-ME-0041</a>	Detection
TSA w.CAM	Tryptic Soy Agar with or without Chloramphenicol (CAM) <i>NOTE:</i> The addition of CAM is optional	<a href="#">EB-ME-0057</a>	Detection
TSA w.kana	Tryptic Soy Agar with kanamycin	<a href="#">EB-ME-0058</a>	Detection

*Continued on next page*

## Detection of production strains, *Continued*

**Media and reagents for Fungal strains**

Dilution buffer: Tween buffer 4%, 90 ml (If necessary, with a magnet) prepared acc. to [EB-ME-0052](#)  
Agar media:

Abbreviation	Full name	Prepared acc. to EB Media direction (link)	Purpose
Cove-T-2	Cove-T-2 agar	<a href="#">EB-ME-0013</a>	Detection & verification
DG-18	DG-18 agar	<a href="#">EB-ME-0017</a>	Verification
Phytate	Phytate agar	<a href="#">EB-ME-0028</a>	Verification
Sch.starch	Schaeffers agar with 1% starch	<a href="#">EB-ME-0037</a>	Verification
YPG	YPG agar with or without tetracycline <i>NOTE:</i> The addition of tetracycline is optional	<a href="#">EB-ME-0044</a>	Detection
YPSS	YPSS agar with or without tetracycline <i>NOTE:</i> The addition of tetracycline is optional	<a href="#">EB-ME-0045</a>	Detection
YSG	Yeast/Soy Peptone/Glucose	MSA-SUB-FS-0064	Verification

If verification on Schaeffers agar with starch is performed then Lugol's iodine solution (0.5%) is used. Lugol's solution is prepared acc. to [EB-ME-0021](#).

### Safety

It is the responsibility of the laboratory leader, that all personnel are aware of the correct handling of enzymes and reagents.

*Continued on next page*



## Detection of production strains, *Continued*

### Sample preparation

The samples are prepared as follows:

Step	Action
1	Transfer 10 g of solid sample or 10 ml of liquid sample to 90 ml Tween buffer 4%.
2	Immediately homogenize the sample by stirring or by shaking. Solid samples are homogenized on a magnetic stirrer for app. 20 minutes.

**IMPORTANT:** All enzyme products must be analyzed from a  $10^{-1}$  dilution due to possible inhibition of micro organisms in undiluted enzyme.

### Plating

Plating must be done within 15 minutes from end of homogenization. If this is not possible, the sample can be stored at 2-8°C for up to 4 hours.

**NOTE:** Relevant agar plates and incubation conditions (time and temperature) are listed in [BD 001-IN-000](#)

Step	Action
1	<b>NOTE:</b> Prepare the test plates: <ul style="list-style-type: none"><li>When analyzing samples from Novozymes production: Transfer 1 ml from the <math>10^{-1}</math> dilution onto the surface of 4 relevant agar plates (14 cm) with app. 0.25 ml on each plate.</li><li>When analyzing samples from Tox batches (GLP): Analyse according to the relevant Analytical Direction.</li></ul>
2	Prepare the 2 positive control plates: <ul style="list-style-type: none"><li>Transfer 0.25 ml from the <math>10^{-1}</math> dilution onto the surface of 1 relevant agar plate (14 cm), and streak the bacteria reference strain or point inoculate the fungal production strain onto the inoculated plate.</li><li>Streak the bacteria reference strain or point inoculate the fungal strain onto another agar plate (not inoculated with sample).</li></ul>
3	Leave the plates on the table until the sample has been soaked into the agar.

*Continued on next page*

## Detection of production strains, *Continued*

### Reading

The colonies on the test-plates are compared morphologically with the colonies of the reference strain.

If ...	Then ...
No suspect colonies are observed on the test-plates ...	The test is ended and the result is stated as: <b>ND</b> (the production strain is Not Detected)
Suspect colonies are observed on the test-plates ...	The test is continued as described below (Verification).

**IMPORTANT:** The reference strain must grow on both of the two positive control plates. If not, the test is repeated.

### Verification

Suspect colonies from the test plates and the reference strain are streaked or point inoculated onto one or more of the agar plates (9 cm or 14 cm) listed in [BD 001-IN-000](#) (column "Verification"). Inoculation and reading of these agar media are described below. The plates are incubated as described in the column "Verification". If necessary, these media can be supplemented with other agar media, e.g. the agar medium used for the detection.

### AT-2 agar

Detection of pullulanase activity:

	Description
<b>Principle</b>	Pullulanase-producing strains degrade the amylopectin in the agar. As a result, blue zones (haloes) will surround the colonies of the isolate.
<b>Inoculation</b>	Point inoculation
<b>Reading</b>	Colonies of the isolate are compared morphologically with the colonies of the reference strain. The surface of the plates is carefully flooded with Lugol's solution (0.5%). Blue zones surrounding the colonies in a reddish-brown medium indicate pullulanase activity.  <i>NOTE:</i> If the production strain produces amylase in addition to pullulanase, clear zones will surround the colony. Between the clear zone and the reddish-brown medium a narrow blue zone might be seen.

*Continued on next page*

## Detection of production strains, *Continued*

---

**Cove-T-2 agar** Detection of amdS-transformed fungi:

	Description
<b>Principle</b>	GMO strains transformed with the marker amdS grow well on the agar, while other strains appear with feeble or no growth.
<b>Inoculation</b>	Point inoculation.
<b>Reading</b>	<i>NOTE:</i> Colonies of the isolate are compared morphologically with the colonies of the reference strain. Vigorous growth on Cove-T-2 indicates presence of an amdS-transformed strain.

---

**DG-18 agar** Comparison of morphology of fungi:

	Description
<b>Principle</b>	DG-18 is a general growth medium for Fungi. The agar is used for comparison of morphology of fungal isolates with the reference strain.
<b>Inoculation</b>	Point inoculation
<b>Reading</b>	<i>NOTE:</i> Colonies of the isolate are compared morphologically with the colonies of the reference strain.

---

**Phytate agar** Detection of phytase activity:

	Description
<b>Principle</b>	Phytase-producing strains degrade phytate in the agar. As a result, clear zones (haloes) will surround the colonies of the isolate.
<b>Inoculation</b>	Point inoculation
<b>Reading</b>	<i>NOTE:</i> Colonies of the isolate are compared morphologically with the colonies of the reference strain. Before inoculation the plates are opaque. The presence of phytase activity is indicated by presence of clear zones (haloes) surrounding the colonies.

---

*Continued on next page*

## Detection of production strains, *Continued*

### Schaeffers agar

Sporulation test (*Bacillus* spp.):

	Description
<b>Principle</b>	Schaeffers agar induces sporulation of wild type strains, but the production strains show no sporulation on Schaeffers agar after incubation for 2-3 days.
<b>Inoculation</b>	Point inoculation
<b>Reading</b>	<i>NOTE:</i> Colonies of the isolate are compared morphologically with the colonies of the reference strain. The colonies are examined by microscopy for sporulation. The production strain shows no sporulation after incubation for 2-3 days.

### Schaeffers starch agar

Detection of amylase activity (all isolates) and sporulation test (*Bacillus* spp.):

	Description
<b>Principle</b>	<u><i>Bacillus</i> spp.:</u> Schaeffers agar induces sporulation of wild type <i>Bacillus</i> strains, but the <i>Bacillus</i> production strains show no sporulation on Schaeffers agar after incubation for 2-3 days. <u><i>Bacillus</i> spp. &amp; Fungi:</u> Amylase producing strains degrade the starch in the agar. As a result, in clear zones (haloes) will surround the colonies of the isolate.
<b>Inoculation</b>	Point inoculation
<b>Reading</b>	Colonies of the isolate are compared morphologically with the colonies of the reference strain. <u><i>Bacillus</i> spp.:</u> The colonies are examined by microscopy for sporulation. The production strain shows no sporulation after incubation for 2-3 days. <u><i>Bacillus</i> spp. &amp; fungi:</u> The surface of the plates is carefully flooded with Lugol's solution (0.5%). Clear zones around the colonies in a blue (dark blue) indicates amylase activity.

*Continued on next page*

## Detection of production strains, *Continued*

---

**Skim milk agar** Detection of proteolytic activity:

	Description
<b>Principle</b>	Protease-producing strains degrade the skim milk in the agar. As a result, clear zones (haloes) surround the colonies of the isolate.
<b>Inoculation</b>	Point inoculation
<b>Reading</b>	<i>NOTE:</i> Colonies of the isolate are compared morphologically with the colonies of the reference strain. Before inoculation the plates are opaque. Presence of clear zones (haloes) surrounding the colonies of the isolate after end of incubation indicate the presence of a proteolytic activity.

---

**Calculation** The result is stated on the basis of the number of typical colonies.

- No typical colonies: ND (Production strain not detected in 0.1 g or 0.1 ml)
- Typical colonies: DET (Production strain detected in 0.1 g or 0.1 ml).

If detected, the app. number of production strains / g or ml is stated.

*IMPORTANT:* If any production strain is detected, the Method Responsible Scientist is contacted immediately. In addition, QCC-cor is informed by mail.

---

**Accuracy and precision** The theoretical detection limit is:

- When analysing samples from Novozymes production:  
10 production strains / g or ml
- When analysing samples from GLP studies:  
1 production strains / g

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**Archiving** All documentation should be archived in accordance with the local archiving SOP.

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*Continued on next page*

## Detection of production strains, *Continued*

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**Contingencies** All deviations from this SOP should be discussed with the Method Responsible Scientist and should be documented.

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**References** [BD 001-IN-000](#)

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**Revision** “EB – Productions Strain list” changed to [BD 001-IN-000](#)

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## **Appendix 4**

### **Documentation regarding the manufacturing process**

1. Statement on compliance of Good Manufacturing Practices, Food
2. ISO 9001:2008 certificate

To Whom It May Concern

**Statement on Good Manufacturing Practice - GMP**  
- general description of production, control and hygiene

Novozymes A/S is a manufacturer of enzymes used in the food industry. We hereby certify that: The products are produced according to good manufacturing practices for manufacturing, packing, or holding human food in order to prevent serious food hazards. Furthermore, our documented quality system is ISO 9001<sup>1</sup> certified by Bureau Veritas Certification, accredited by UKAS. The quality system includes:

- Production operations are conducted in accordance with adequate sanitation principles.
- HACCP plan. Critical control points (CCPs) are identified and controlled, and the products are released if in compliance with these requirements.
- Critical measuring equipment is identified and calibrated at regular intervals.
- Instructions on cleaning of equipment, utensils and rooms are established and cleaning is documented.
- The personnel is trained in hygienic practices in order to prevent contamination of products and equipment.
- The personnel is trained in the quality system.
- The buildings and equipment are monitored periodically with special reference to maintenance.
- The production of our food enzymes complies with EC regulation 853/2004/EC, including amendments, on *the hygiene of foodstuffs*.
- The packaging materials used for our food enzyme products comply with EC regulation 1935/2004/EC, and related legislation including amendments on materials and articles intended to come into contact with foodstuffs.
- The production is under control of and inspected by the authorities according to EC regulation 853/2004/EC, including amendments, on *the official control of foodstuffs* as interpreted and implemented in Danish legislation.

<sup>1</sup>The scope of the 9001 certificate is: Development, Production and Sales of Biopolymers and Industrial Enzymes.



**BUREAU VERITAS**  
Certification



**Novozymes A/S**  
Krogshøjvej 36, 2880 Bagsværd, Denmark

This is a multi-site certificate. Additional site details are listed in the appendix to this certificate.

*Bureau Veritas Certification Holding SAS – UK Branch certifies that the Management System of the above organization has been audited and found to be in accordance with the requirements of the Management System standards detailed below.*

*Standard*

**ISO 9001:2008**

*Scope of certification*

**Development , Production and Sales of Industrial Enzymes.**

Certification cycle start date: **26-03-2015**

Subject to the continued satisfactory operation of the organization's Management System, this certificate expires on: **25-03-2018**

Original certification date: **25-03-1996**

**Certificate no.: DK005049**

**Version: 1**

**Revision date: 24-03-2015**



**Signed on behalf of BVCH SAS UK Branch**

Certification body address:  
Local Office:

5<sup>th</sup> Floor, 66 Prescott Street, London, E1 8HG, United Kingdom  
Oldenborggade 1B, 7000 Fredericia, Denmark



Further clarifications regarding the scope of this certificate and the applicability of the Management System requirements may be obtained by consulting the organization.  
To check this certificate validity, please call **(+45) 77 311 000**.



## **Novozymes A/S**

*Standard*

## **ISO 9001:2008**

*Scope of certification*

**Head Office:**  
**Development , Production and Sales of Industrial Enzymes.**

<b>Site:</b>	<b>Address:</b>	<b>Scope:</b>
Novozymes A/S	Krogshøjvej 36, 2880 Bagsværd Denmark	Development , Production and Sales of Industrial Enzymes.
Novozymes A/S	Hillerødgade 31 & 42 2200 København N Denmark	Development , Production and Sales of Industrial Enzymes.
Novozymes A/S	Hallas Allé 1 4400 Kalundborg Denmark	Development , Production and Sales of Industrial Enzymes.
Novozymes (China) Biotechnology Co. Ltd.	150 Nanhai Road TEDA 300457 Tianjin China	Development , Production and Sales of Industrial Enzymes.
Novozymes (China) Investment Co. Ltd.	No. 22 Xinxi Zhong Lu Shangi Zone, Haidian District, Beijing 10085 China	Development , Production and Sales of Industrial Enzymes.

Revision date: **24-03-2015**

**Signed on behalf of BVCH SAS UK Branch**

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## **Novozymes A/S**

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### **ISO 9001:2008**

*Scope of certification*

<b>Site :</b>	<b>Address:</b>	<b>Scope :</b>
Suzhou Hongda Enzyme Co. Ltd.	Sha Xi Town Taicang City Jiang Su Province 215421 China China	Development , Production and Sales of Industrial Enzymes.
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Novozymes Nebraska	600 S. 1st Street NE 68008 Blair USA	Development , Production and Sales of Industrial Enzymes.
Novozymes Brazil	Rua Professor Francisco Ribeiro 683 CEP 83707-660 Bairro Barigüi Araucária – Paraná Brazil	Development , Production and Sales of Industrial Enzymes.
Novozymes Switzerland	Neumatt CH-4243 Dittingen Switzerland	Development , Production and Sales of Industrial Enzymes.

Revision date: **24-03-2015**



*Signed on behalf of BVCH SAS UK Branch*

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Local Office:

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## **Novozymes A/S**

*Standard*

### **ISO 9001:2008**

*Scope of certification*

<b>Site :</b>	<b>Address :</b>	<b>Scope :</b>
Novozymes South Asia Private Limited	Genisys Building Plot No. 32, 47-50 EPIP Area Bangalore 560 066 Karnataka India	Development , Production and Sales of Industrial Enzymes.
Novozymes South Asia Private Limited	Survej No. 193 Hoody Village, Whitefield Road Bangalore, 560 048 India	Development , Production and Sales of Industrial Enzymes.
Novozymes South Asia Private Limited	20th KM Hosur Road Electronics City Bangalore 560 100 India	Development , Production and Sales of Industrial Enzymes.
Sales Region Headquarter (HG)	Krogshøjvej 36 2880 Bagsværd Denmark	Development , Production and Sales of Industrial Enzymes.
Sales Region North America (NA) Novozymes North America Inc.	77 Perry Chapel Church Road Franklinton North Carolana 27525-0576 USA	Development , Production and Sales of Industrial Enzymes.

Revision date: **24-03-2015**



*Signed on behalf of BVCH SAS UK Branch*

Certification body address:  
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## **Novozymes A/S**

*Standard*

### **ISO 9001:2008**

*Scope of certification*

<b>Site :</b>	<b>Address :</b>	<b>Scope :</b>
Sales Region China (CH) Novozymes China Investment Co Ltd.	No. 14, Xinx Road Shangdi Zone Beijing 100085 China	Development , Production and Sales of Industrial Enzymes.
Sales Regions EEMEA (Eastern Europe, Middle East and Africa) and CWE (Central Western Europe) Novozymes Switzerland AG	Neumattweg 16 4243 Dittingen Switzerland	Development , Production and Sales of Industrial Enzymes.
Sales Region South East Asia, India and Australia (SEAIA) Novozymes Malaysia Sdn. Bhd	Lot 5, Technology Park Lebuhraya Puchong- Sg Besi Malaysia Bukit Jalil Kuala Lumpur 57000 Malaysia	Development , Production and Sales of Industrial Enzymes.
Sales Region Japan & Korea (JK) Novozymes Japan Ltd.	Makuhari Techno Garden CB-5/3, Nakase, 1-chome, Mihama-ku Chiba-shi 261-8501 Japan	Development , Production and Sales of Industrial Enzymes.

Revision date: **24-03-2015**



*Signed on behalf of BVCH SAS UK Branch*

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Certification



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## **ISO 9001:2008**

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Sales Region Latin America (LA) Novozymes Latin America Ltda	Rua Professor Francisco Ribeiro, 683 Arucária Paraná (PR) 83707-660 Brazil	Development , Production and Sales of Industrial Enzymes.

Revision date: **24-03-2015**

  
*Signed on behalf of BVCH SAS UK Branch*

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Further clarifications regarding the scope of this certificate and the applicability of the Management System requirements may be obtained by consulting the organization.  
To check this certificate validity, please call **(+45) 77 311 000**.

## Appendix 5

### Safety documentation

1. Lactase produced by PP3930. Assessment of sequence homology to known toxins and allergens. Novozymes Report No.: 2016-03250
2. Summary of toxicity data. Lactase, batch PPL34537 from *Bacillus licheniformis* Novozymes Report No.: 2014-11425
3. Lactase, batch PPL34537: Test for mutagenic activity with strains of *Salmonella typhimurium* and *Escherichia coli*. Novozymes Study No. 20138037. Novozymes Report no.: 2013-17652
4. Lactase, batch PPL34537: Induction of micronuclei in cultured human peripheral blood lymphocytes. Covance Laboratories Study No. 8292559. Novozymes Report No.: 2014-00365
5. Lactase, batch PPL34537. Toxicity Study by Oral Gavage Administration to Sprague-Dawley Rats for 13 Weeks. Huntingdon Life Sciences Study No. LKG0074. Novozymes Report No.: 2014-11424

# lactase produced by PP3930 Assessment of sequence homology to known toxins and allergens

Esben Friis  
LUNA# 2016-03250-01

February 23, 2016

## **Contents**

<b>1</b>	<b>Sequence homology of lactase produced by PP3930 to known toxins</b>	<b>2</b>
<b>2</b>	<b>Allergen analysis of lactase produced by PP3930</b>	<b>2</b>
<b>A</b>	<b>Scripts for toxin homology search</b>	<b>5</b>
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# 1 Sequence homology of lactase produced by PP3930 to known toxins

## Uniprot database

Protein sequences that contain the word *toxin* in the description field were extracted from UNIPROT (Database date: 11-Feb-2016). This database contains entries from SWISSPROT and TREMBL. 50251 entries were found. Each of the sequences was placed in its uniquely named Fasta file. The lactase produced by PP3930 sequence was placed in a separate file "lactase.fasta". The awk script in appendix A was used to invoke the sequence alignment program ClustalW 2.0.10 to align each sequence to lactase produced by PP3930. A summary file containing the length of each sequence and number of identical residues is also created. From this, the identity percentage to the lactase produced by PP3930 sequence or the compared toxin sequence is calculated, whichever is longest. This is chosen because the toxin sequences have many different lengths, both much shorter and much longer than the lactase produced by PP3930 sequence. By always using the longest sequence, artificial high scores from very short or very long toxins are avoided. The largest homology encountered was 15.0%, indicating that the homology to any toxin sequence in this database is indeed random and very low. First 1000 results with largest homology are shown in appendix B.

# 2 Allergen analysis of lactase produced by PP3930

## Allergen Database

- <http://allergenonline.org>. This is the home page of the The Food Allergy Research and Resource Program (FARRP) allergen protein database. The present report use data downloaded 10-Feb-2016 (version 16). Appendix D shows a list.

## Analyses

1. more than 35% identity in the amino acid sequence of the expressed protein (i.e.without the leader sequence, if any), using a window of 80 amino acids and a suitable gap penalty (using Clustal-type alignment programs or equivalent alignment programs). This is one of the recommended test methods of the EFSA scientific opinion [1], and also of the earlier publication from the FAO/WHO Expert group [2]. The queries were done using Fasta 3.4, using the scripts in appendix C
2. same as item 1, but with scaling enabled. In this way, matches with high identity, but over windows shorter than 80 amino acids can be identified. For example a match with 50%

identity over 60 amino acids would still have enough identical amino acids to exceed the 35% threshold over 80 amino acids:  $60 \cdot 0.50/80 = 0.375 = 37.5\%$ .

3. Search for identity over 8 contiguous amino acids. The queries were done using Fasta 3.4, using the scripts in appendix C
4. Alignment of lactase produced by PP3930 to any allergen identified by 35% or larger identity over any 80 amino acid window, or 100% identity over any 8 amino acid window. This is to show the identity over the full length of the protein. These queries were performed using the global alignment "needle", which is an implementation of the Needleman-Wunsch global alignment algorithm [3] in the program package EMBOSS [4].

Items 1 is in compliance with the recommendations from WHO/FAO Expert group [2]. Item 2 is a further analysis to detect high homology over shorter sequences, in compliance with the recommendations in the EFSA scientific opinion [1]. Item 4 adds some more detailed information for hits identified by the methods in item 1 and 3.

## Results

### Database: allergenonline.org

#### 35% identity over 80 amino acids

The following allergens had one or more matches using the method described in item 1 above (see appendix E for a complete list).

(No hits found)

#### 35% identity over 80 amino acids with scaling

The following allergens had one or more matches using the method described in item 2 above (see appendix E for a complete list).

(No hits found)

#### Identity over full length

Identities between lactase produced by PP3930 and allergens identified by 35% or larger identity over any 80 amino acid window, or 100% identity over any 8 amino acid window.

### 100% identity over 8 amino acids

The following allergens had one or more matches using the method described in item 3 above.

(No hits found)

## References

- [1] Scientific opinion on the assessment of allergenicity of GM plants and microorganisms and derived food and feed. EFSA panel on genetically Modified Organisms (GMO panel). European Food Safety Authority (EFSA), Parma 2010. (The document may be downloaded from <http://www.efsa.europa.eu/en/scdocs/scdoc/1700.htm>)
- [2] Evaluation of Allergenicity of Genetically Modified Foods (Report of a Joint FAO/WHO Expert Consultation on Allergenicity of Foods Derived from Biotechnology 22–25 January 2001), Food and Agriculture Organization of the United Nations (FAO), Rome 2001. [http://www.who.int/foodsafety/publications/biotech/ec\\_jan2001/en/](http://www.who.int/foodsafety/publications/biotech/ec_jan2001/en/)
- [3] Needleman, S. B. and Wunsch, C. D. (1970) *J. Mol. Biol.* **48**, p 443-453.
- [4] Rice, P. Longden, I. and Bleasby, A. (2000): "EMBOSS: The European Molecular Biology Open Software Suite" *Trends in Genetics* **16**, No 6. p 276-277

## A Scripts for toxin homology search

Awk scripts for alignment of sequences to the lactase produced by PP3930 sequence and calculation of sequence lengths and identities. First the script used to run the alignments. The script is stored in a file called "runaligns".

```
#!/bin/tcsh
cat lactase.fasta $1 >tmp.txt
clustalw tmp.txt
grep -v ">" $1 | gawk '{printf "%s",$0} END {printf "\n"}' | wc | \
    gawk '{print $3-1}' > $1.len
cat tmp.aln | gawk '{printf "%s",$0} END {printf "\n"}' | \
    sed 's/[~\*]//g' | wc | gawk '{print $3-1}' > $1.idt
echo $1 | gawk '{printf "%s ",$0}' >> summary
cat $1.len $1.idt | gawk '{printf "%s ",$0} END {printf "\n"}' >>summary
mv tmp.aln $1.out
```

Before start, the file "summary" must be deleted. The analysis is automatically done for all .fasta files in the current directory (and subdirectories, if present) by the command:

```
find . -name "*.fasta" -exec runaligns {} \;
```

Afterwards the sequence length and identity information can be found in the file summary. This file is processed through the following Python script, which calculates the percentages as described in the text.

```
#!/usr/bin/python
import string,commands

compare_length = 413
data = []

f = open ("summary","r")

buffer = "XX"
i=0

while buffer != "":
    buffer = f.readline()
    if buffer != "":
        data.append(string.split(buffer))
        data[i][1] = int(data[i][1])
        data[i][2] = int(data[i][2])

        i = i+1
f.close()
```

```

for i in range(len(data)):
    fullname = commands.getoutput("grep "+string.upper(data[i][0][2:-6])+" description.txt")
    percentid = 100.0*float(data[i][2])/float(max(data[i][1],compare_length))
    if (percentid >= 10.0):
        printlist = [data[i][0][2:-6], data[i][1], data[i][2], \
                      percentid, \
                      fullname[18:83] ]
        print '%-13s %4d %4d %5.1f    %-60s' % tuple(printlist)

```

## B Toxin homology results

UNIPROT entries, that contain the word "toxin", but not "fragment" in the description field and their identity to lactase produced by PP3930. The columns are

1. sequence database accession number
2. sequence length
3. number of identical residues after alignment to lactase produced by PP3930
4. percent identity compared to lactase produced by PP3930 or the sequence, whichever is longest.
5. sequence description

1000 sequences with largest homology are shown.

X6H4M2	1523	229	15.0	X6H4M2_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
A3VL73	1512	224	14.8	A3VL73_9RHOB	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:EAQ10992....}
L8JRY0	1295	193	14.8	L8JRY0_9BACT	SubName: Full=RTX toxin {ECO:0000313 EMBL:ELR70224.1};
AOA076K205	1070	192	14.7	AOA076K205_	SubName: Full=Putative RTX toxin and hemolysin-type calcium ...
AOA0E2N9W0	1522	222	14.6	AOA0E2N9W0_	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
A8TUW5	1188	190	14.6	A8TUW5_9PROT	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
V7GAK9	1522	222	14.6	V7GAK9_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
X5YPX3	1525	222	14.6	X5YPX3_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
X6J2E5	1525	222	14.6	X6J2E5_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
X6JX35	1525	219	14.4	X6JX35_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
K9RXK7	1158	187	14.3	K9RXK7_SYNP3	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
X6EJK5	1603	230	14.3	X6EJK5_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
X6F1R8	1603	229	14.3	X6F1R8_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
X6FC97	1603	229	14.3	X6FC97_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
A6FPB9	1274	185	14.2	A6FPB9_9RHOB	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
B9JJP1	1622	231	14.2	B9JJP1_AGRRK	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
B8M939	1045	184	14.1	B8M939_TALSN	SubName: Full=Hansenula MRAKII killer toxin-resistant protei...
C3MGZ7	1256	184	14.1	C3MGZ7_RHISN	SubName: Full=Putative RTX toxin hemolysin-type protein {ECO...
X5ZE95	1523	215	14.1	X5ZE95_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
B6QYA4	1292	182	14.0	B6QYA4_9RHOB	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:EEA969...
K9TLY5	1139	182	14.0	K9TLY5_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
U4E257	1525	214	14.0	U4E257_9VIBR	SubName: Full=Putative RTX toxin and related Ca2+-binding do...
W1J3J6	1016	183	14.0	W1J3J6_9ENTR	SubName: Full=C component of insecticidal toxin complex (Tc)...
X6IT24	1525	214	14.0	X6IT24_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
AOA024B2T5	971	181	13.9	AOA024B2T5_	SubName: Full=Insecticidal toxin complex protein TccC3 {ECO:...
AOA0POMJ37	1107	181	13.9	AOA0POMJ37_	SubName: Full=RTX-I toxin determinant A from serotypes 1/9 {...}
B7K4C8	1394	194	13.9	B7K4C8_CYAP8	SubName: Full=RTX cytolytic toxin protein A {ECO:0000313 EMB...
AOA076K3H6	1177	180	13.8	AOA076K3H6_	SubName: Full=Putative RTX toxin and hemolysin-type calcium ...
Q2VZW5	1352	187	13.8	Q2VZW5_MAGSA	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
AOA034TCR5	1050	178	13.7	AOA034TCR5_	SubName: Full=RTX toxin {ECO:0000313 EMBL:GAJ71685.1};
AOA0F6EZD2	1256	178	13.7	AOA0F6EZD2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AIY11346.1};
AOA0K8ZGA0	1450	198	13.7	AOA0K8ZGA0_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KJE59101.1};
AOA0L6YTE6	1450	198	13.7	AOA0L6YTE6_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KOA13934.1};

AOA0R1BNN9	1450	198	13.7	AOA0R1BNN9_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KRJ74933.1};
A6CZA9	1480	203	13.7	A6CZA9_9VIBR	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
G2JJE2	1450	198	13.7	G2JJE2_ACIBA	SubName: Full=RTX toxin {ECO:0000313 EMBL:AEP06532.1};
O85157	1043	179	13.7	O85157_PHOLU	SubName: Full=Insecticidal toxin complex protein TccC {ECO:0...
Q2KC97	1797	247	13.7	Q2KC97_RHIEC	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
Q2W222	1274	178	13.7	Q2W222_MAGSA	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
X5HXU4	1022	179	13.7	X5HXU4_AERHY	SubName: Full=Structural toxin protein RtxA {ECO:0000313 EMB...
DOHH25	1505	205	13.6	DOHH25_VIBMI	SubName: Full=RTX toxin related protein {ECO:0000313 EMBL:EE...
EO5IS2	1436	195	13.6	RHSB_DICD3	RecName: Full=Probable deoxyribonuclease RhsB; EC=3.1.-.-; A...
I4YZB7	1527	207	13.6	I4YZB7_9RHIZ	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
J3AB06	1214	177	13.6	J3AB06_9CAUL	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA074V6D5	940	176	13.5	AOA074V6D5_	SubName: Full=RTX toxins or related Ca2+-binding protein {EC...
AOA077FLU1	1236	176	13.5	AOA077FLU1_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA0C4XWH7	1384	187	13.5	AOA0C4XWH7_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0Q0W3A9	1518	205	13.5	AOA0Q0W3A9_	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
X5UVA6	1573	213	13.5	X5UVA6_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
X6C6F8	1649	222	13.5	X6C6F8_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
AOA0J9F132	1043	175	13.4	AOA0J9F132_	SubName: Full=Toxin {ECO:0000313 EMBL:KMW72153.1};
V7HL06	1649	221	13.4	V7HL06_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
X6KK79	1573	210	13.4	X6KK79_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
AOA034TCJ9	1085	173	13.3	AOA034TCJ9_	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
AOA080K4U8	940	174	13.3	AOA080K4U8_	SubName: Full=RTX toxin or related Ca2+-binding protein {ECO...
AOA085WUN2	1237	174	13.3	AOA085WUN2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KFE71395.1};
AOA0A0QUL1	1052	173	13.3	AOA0A0QUL1_	SubName: Full=RTX-III toxin determinant A {ECO:0000313 EMBL:...
AOA0A0QVB1	1052	174	13.3	AOA0A0QVB1_	SubName: Full=RTX-III toxin determinant A {ECO:0000313 EMBL:...
AOA0A1V8I7	1534	204	13.3	AOA0A1V8I7_	SubName: Full=Insecticide toxin TcdB-like protein {ECO:00003...
AOA0F7LW2	1043	173	13.3	AOA0F7LW2_	SubName: Full=Toxin {ECO:0000313 EMBL:AKH64265.1};
AOA0G2ZFW1	1236	174	13.3	AOA0G2ZFW1_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AKJ00466.1};
AOA0J5T1N6	959	173	13.3	AOA0J5T1N6_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA0K1PFQ3	1250	174	13.3	AOA0K1PFQ3_	SubName: Full=Insecticidal toxin complex protein TccB1 {ECO:...
AOA0LOM000	908	173	13.3	AOA0LOM000_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
A0Z9Q6	1067	173	13.3	A0Z9Q6_NODSP	SubName: Full=RTX toxins and Ca2+-binding proteins {ECO:0000...
BOBQR7	1052	173	13.3	BOBQR7_ACTPJ	SubName: Full=RTX-III toxin determinant A from serotype 8 {E...
D9PBQ3	1052	173	13.3	D9PBQ3_ACTPL	SubName: Full=RTX-III toxin determinant A from serotype 8 {E...
K9QV50	1183	174	13.3	K9QV50_NOSS7	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
M1P8D8	1287	174	13.3	M1P8D8_DESSD	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
P55131	1052	173	13.3	RTX32_ACTPL	RecName: Full=RTX-III toxin determinant A from serotype 8; A...
X6AGC7	1649	220	13.3	X6AGC7_9RHIZ	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
AOA023RIR6	1456	192	13.2	AOA023RIR6_	SubName: Full=RTX toxin-like protein {ECO:0000313 EMBL:AHX61...
AOA090T468	1712	226	13.2	AOA090T468_	SubName: Full=RTX toxins and related Ca2+-binding proteins {...
AOA093V6K6	1329	175	13.2	AOA093V6K6_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0E4GSA0	1451	191	13.2	AOA0E4GSA0_	SubName: Full=RTX toxins determinant A and related Ca2+-bind...
AOA0J5QCB3	1087	172	13.2	AOA0J5QCB3_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA0R0TPB5	1450	192	13.2	AOA0R0TPB5_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KRI74544.1};
A1SYJ0	1092	172	13.2	A1SYJ0_PSYIN	SubName: Full=RTX toxin-like protein {ECO:0000313 EMBL:ABM04...
D3POF7	1716	226	13.2	D3POF7_AZOS1	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
G0YF58	1045	172	13.2	G0YF58_PHOTE	SubName: Full=Insecticidal toxin complex protein TccC {ECO:0...
G6EIS7	1213	172	13.2	G6EIS7_9SPHN	SubName: Full=Structural toxin protein RtxA {ECO:0000313 EMB...
Q2S9B2	1361	180	13.2	Q2S9B2_HAHCH	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
U7R4B6	1043	172	13.2	U7R4B6_PHOTE	SubName: Full=Toxin {ECO:0000313 EMBL:ERT14620.1};
AOA034TGP3	1133	171	13.1	AOA034TGP3_	SubName: Full=RTX toxin {ECO:0000313 EMBL:GAJ71684.1};
AOA061PXJ9	1251	171	13.1	AOA061PXJ9_	SubName: Full=RTX toxin {ECO:0000313 EMBL:GAK21698.1};
AOA093XDC7	1252	171	13.1	AOA093XDC7_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0M9EAF6	912	171	13.1	AOA0M9EAF6_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KPA17796.1};
H6NFP1	1302	171	13.1	H6NFP1_9BACL	SubName: Full=RTX toxins and-related Ca2+-binding protein {E...
J3CA95	1308	172	13.1	J3CA95_9RHIZ	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
J3F666	1660	218	13.1	J3F666_9PSED	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
O85153	1485	195	13.1	O85153_PHOLU	SubName: Full=Insecticidal toxin complex protein TcaC {ECO:0...

Q7MZV7	1043	171	13.1	Q7MZV7_PHOLL	SubName: Full=Insecticidal toxin complex protein TccC1 {ECO:...
Q93NP0	1052	171	13.1	Q93NP0_ACTPL	SubName: Full=RTX-toxin IIIA {ECO:0000313 EMBL:AAK50053.1};
U5TWR2	1381	181	13.1	U5TWR2_KLEPN	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA031GXC3	1006	170	13.0	AOA031GXC3_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA090P2W8	1316	171	13.0	AOA090P2W8_	SubName: Full=RTX toxins and related Ca2+-binding proteins {...
AOA0E2NVU7	1620	211	13.0	AOA0E2NVU7_	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
AOA0H6SN78	1203	169	13.0	AOA0H6SN78_	SubName: Full=RTX toxin RtxA {ECO:0000313 EMBL:CSC43151.1};
AOA0H6V206	1176	169	13.0	AOA0H6V206_	SubName: Full=RTX toxin RtxA {ECO:0000313 EMBL:CSC65864.1};
AOA0H6WM51	1048	169	13.0	AOA0H6WM51_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:00003...
AOA0M1MWL2	1279	170	13.0	AOA0M1MWL2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KOR74258.1};
AOA0NOE4N9	1181	169	13.0	AOA0NOE4N9_	SubName: Full=Hemagglutinin repeat/Pre-toxin domain with VEN...
AOA0NOX934	1447	188	13.0	AOA0NOX934_	SubName: Full=Putative insecticidal toxin complex protein B ...
AOA0P9REQ9	1018	169	13.0	AOA0P9REQ9_	SubName: Full=Putative Insecticidal toxin complex protein {E...
AOA0ROB4F0	1069	170	13.0	AOA0ROB4F0_	SubName: Full=Toxin {ECO:0000313 EMBL:KRG52160.1};
A6FML4	1028	170	13.0	A6FML4_9RHOB	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
E5AU98	1059	170	13.0	E5AU98_BURRH	SubName: Full=Insecticidal toxin complex protein TccA {ECO:0...
G0JX94	1068	169	13.0	G0JX94_STEMA	SubName: Full=Zeta toxin family protein {ECO:0000313 EMBL:AE...
L8M6B9	1096	169	13.0	L8M6B9_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
Q2K6Y1	1326	172	13.0	Q2K6Y1_RHIEC	SubName: Full=Putative RTX toxin hemolysin-type protein {ECO...
Q66A99	994	170	13.0	Q66A99_YERPS	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
Q7MZ20	936	170	13.0	Q7MZ20_PHOLL	SubName: Full=Insecticidal toxin complex protein TccC7 {ECO:...
U2GRE5	1154	170	13.0	U2GRE5_9PROT	SubName: Full=RTX toxin {ECO:0000313 EMBL:ERJ28533.1};
VOVNK4	1024	170	13.0	VOVNK4_ECOLX	SubName: Full=Hemolysin toxin protein A {ECO:0000313 EMBL:ES...
AOA072CPB3	1256	168	12.9	AOA072CPB3_	SubName: Full=RTX toxin hemolysin-type protein {ECO:0000313 ...
AOA075HCN3	1533	197	12.9	AOA075HCN3_	SubName: Full=RTX toxin, Ca2+-binding protein {ECO:0000313 E...
AOA075MW27	1240	168	12.9	AOA075MW27_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA093USE2	1242	168	12.9	AOA093USE2_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0A0CQR0	1043	168	12.9	AOA0A0CQR0_	SubName: Full=Toxin {ECO:0000313 EMBL:KGM28239.1};
AOA0A1HPQ2	1703	220	12.9	AOA0A1HPQ2_	SubName: Full=RTX toxin, putative {ECO:0000313 EMBL:CDF92448...
AOA0H6JCZ3	1773	228	12.9	AOA0H6JCZ3_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:00003...
A4BAS2	1629	210	12.9	A4BAS2_9GAMM	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:EAR11028...
B9JP23	927	168	12.9	B9JP23_AGRRK	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
F6IPV7	1535	198	12.9	F6IPV7_9SPHN	SubName: Full=Structural toxin protein RtxA {ECO:0000313 EMB...
I2STR4	998	168	12.9	I2STR4_ECOLX	SubName: Full=Hemolysin toxin protein A {ECO:0000313 EMBL:EI...
K0WZS3	1725	223	12.9	K0WZS3_PSEFL	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
M4VGU8	1186	168	12.9	M4VGU8_9PROT	SubName: Full=RTX toxins-related Ca2+-binding protein {ECO:0...
N1NUB5	1016	168	12.9	N1NUB5_XENNE	SubName: Full=C component of insecticidal toxin complex (Tc)...
N1NUG4	1542	199	12.9	N1NUG4_XENNE	SubName: Full=A component of insecticidal toxin complex (Tc)...
Q6YGS9	1377	178	12.9	Q6YGS9_ECOLX	SubName: Full=Vacuolating autotransporter toxin {ECO:0000313...
Q9XDI1	1208	168	12.9	Q9XDI1_LEGPN	SubName: Full=Structural toxin protein RtxA {ECO:0000313 EMB...
S2K911	1446	187	12.9	S2K911_9PSED	SubName: Full=Insecticidal toxin complex protein TcaC {ECO:0...
V6G128	998	168	12.9	V6G128_ECOLX	SubName: Full=Hemolysin toxin protein A {ECO:0000313 EMBL:EI...
AOA009I5V9	1326	170	12.8	AOA009I5V9_	SubName: Full=Pre-toxin domain with VENN motif family protei...
AOA034TDC8	1596	205	12.8	AOA034TDC8_	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
AOA061Q823	1001	167	12.8	AOA061Q823_	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
AOA0FOGDR4	1519	195	12.8	AOA0FOGDR4_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AIY11347.1};
AOA0P9WJ79	1018	167	12.8	AOA0P9WJ79_	SubName: Full=Putative Insecticidal toxin complex protein {E...
B2BCY3	1489	191	12.8	B2BCY3_YERPU	SubName: Full=Toxin complex subunit TcaC {ECO:0000313 EMBL:A...
E0MRE4	1725	220	12.8	E0MRE4_9RHOB	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:EFL88467...
F7RTG8	1273	167	12.8	F7RTG8_9GAMM	SubName: Full=RTX toxin, putative {ECO:0000313 EMBL:EGM68164...
H2FXH0	968	167	12.8	H2FXH0_OCESG	SubName: Full=Structural toxin protein RtxA {ECO:0000313 EMB...
M7CTJ2	974	167	12.8	M7CTJ2_MORMO	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
O85151	1095	167	12.8	O85151_PHOLU	SubName: Full=Insecticidal toxin complex protein TcaA {ECO:0...
Q307Q7	1485	190	12.8	Q307Q7_PHOLU	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
Q7N936	1485	190	12.8	Q7N936_PHOLL	SubName: Full=Insecticidal toxin complex protein TcaC {ECO:0...
AOA034TT43	1854	236	12.7	AOA034TT43_	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
AOA0A8P2X7	1016	166	12.7	AOA0A8P2X7_	SubName: Full=C component of insecticidal toxin complex (Tc)...
AOA0F2HXE6	1005	165	12.7	AOA0F2HXE6_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KJR28404.1};



AOA0J5FTT1	1016	166	12.7	AOA0J5FTT1_	SubName: Full=Insecticidal toxin complex (Tc) protein C {ECO:0000313 EMBL:K117395.1};
AOA0L1LMB0	922	165	12.7	AOA0L1LMB0_	SubName: Full=Toxin {ECO:0000313 EMBL:K117395.1};
AOA0M4LKL7	923	166	12.7	AOA0M4LKL7_	SubName: Full=Toxin {ECO:0000313 EMBL:ALD99946.1};
AOA0NOEMB5	923	166	12.7	AOA0NOEMB5_	SubName: Full=Insecticidal toxin complex protein {ECO:0000313 EMBL:K117395.1};
AOA0P9YDX9	925	165	12.7	AOA0P9YDX9_	SubName: Full=Insecticidal toxin complex protein {ECO:0000313 EMBL:K117395.1};
AOA0Q8AEK4	1154	166	12.7	AOA0Q8AEK4_	SubName: Full=Toxin {ECO:0000313 EMBL:K117395.1};
AOA0R4FN65	1016	166	12.7	AOA0R4FN65_	SubName: Full=C component of insecticidal toxin complex (Tc)...
D3VHIO	1016	166	12.7	D3VHIO_XENNA	SubName: Full=C component of insecticidal toxin complex (Tc)...
D9P6I6	1050	166	12.7	D9P6I6_ACTPL	SubName: Full=RTX-III toxin determinant A from serotype 8 {E...
EORIS1	975	166	12.7	EORIS1_PAEP6	SubName: Full=RTX toxin-like Ca2+-binding protein {ECO:0000313 EMBL:K117395.1};
E4UMN3	1284	165	12.7	E4UMN3_ARTGP	SubName: Full=Killer toxin alpha/beta {ECO:0000313 EMBL:EFQ9...}
E9RGH6	995	166	12.7	E9RGH6_ACTVL	SubName: Full=Toxin {ECO:0000313 EMBL:BAJ78235.1};
F2C8C1	1478	187	12.7	F2C8C1_STRSA	SubName: Full=Diarrheal toxin {ECO:0000313 EMBL:EGF14161.1};...
F3SK83	1478	188	12.7	F3SK83_STRSA	SubName: Full=Diarrheal toxin {ECO:0000313 EMBL:EGG39484.1};...
F4XXN8	1147	166	12.7	F4XXN8_9CYAN	SubName: Full=RTX toxins family Ca2+-binding protein {ECO:0000313 EMBL:K117395.1};
F7UXB7	1287	166	12.7	F7UXB7_EEGSY	SubName: Full=RTX toxin {ECO:0000313 EMBL:BAK43375.1};
H0EYF4	1087	166	12.7	H0EYF4_GLAL7	SubName: Full=Putative Killer toxin subunits alpha/beta {ECO:0000313 EMBL:K117395.1};
H1W6M6	1315	167	12.7	H1W6M6_9CYAN	SubName: Full=Putative Hemolysin-type calcium-binding toxin,...
J7SKP9	1480	188	12.7	J7SKP9_MORMO	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:AGG30...}
K4Z9Z8	1069	166	12.7	K4Z9Z8_PAEAL	SubName: Full=Insecticidal toxin complex protein {ECO:0000313 EMBL:K117395.1};
K8FZJ3	1830	233	12.7	K8FZJ3_9XANT	SubName: Full=RTX toxin {ECO:0000313 EMBL:EKQ61710.1};
K9EZL7	924	165	12.7	K9EZL7_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 EMBL:K117395.1};
L7GWZ7	923	166	12.7	L7GWZ7_PSESX	SubName: Full=Insecticidal toxin complex protein {ECO:0000313 EMBL:K117395.1};
L7U8I5	1024	165	12.7	L7U8I5_MYXSD	SubName: Full=Toxin {ECO:0000313 EMBL:AGC45241.1};
P55130	1049	165	12.7	RTX31_ACTPL	RecName: Full=RTX-III toxin determinant A from serotype 2; A...
Q113P6	1363	173	12.7	Q113P6_TRIEI	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
AOA034TUQ8	937	164	12.6	AOA034TUQ8_	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
AOA061PGR0	832	164	12.6	AOA061PGR0_	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
AOA077PQ26	1039	164	12.6	AOA077PQ26_	SubName: Full=Insecticidal toxin complex protein TccC {ECO:0000313 EMBL:K117395.1};
AOA0H5BBN0	963	164	12.6	AOA0H5BBN0_	SubName: Full=Alkaline phosphatase {ECO:0000313 EMBL:BAR9959...}
AOA0H6PTJ8	1425	179	12.6	AOA0H6PTJ8_	SubName: Full=RTX toxin RtxA {ECO:0000313 EMBL:CSC08031.1};
AOA0K8LB73	1423	179	12.6	AOA0K8LB73_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 EMBL:K117395.1};
A7WK64	1177	164	12.6	A7WK64_LYSSH	SubName: Full=Cry48Aa protein {ECO:0000313 EMBL:CAJ86545.1};...
B7RNR5	952	164	12.6	B7RNR5_9RHOB	SubName: Full=Putative RTX toxin hemolysin-type calcium-bind...
C8C508	1706	215	12.6	C8C508_BORPT	SubName: Full=Adenylate cyclase toxin {ECO:0000313 EMBL:ACU8...}
E3EHS8	1255	164	12.6	E3EHS8_PAEPS	SubName: Full=RTX toxin {ECO:0000313 EMBL:AD056340.2};
H8GKK7	1044	164	12.6	H8GKK7_METAL	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 EMBL:K117395.1};
L8M7Q3	1167	164	12.6	L8M7Q3_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 EMBL:K117395.1};
Q2BFU2	1415	178	12.6	Q2BFU2_9BACI	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
R4QC48	1323	167	12.6	R4QC48_HELXP	SubName: Full=Toxin {ECO:0000313 EMBL:AGL67076.3};
AOA061PZG9	1756	219	12.5	AOA061PZG9_	SubName: Full=RTX toxin {ECO:0000313 EMBL:GAK21699.1};
AOA063BT73	1005	163	12.5	AOA063BT73_	SubName: Full=HC-toxin synthetase {ECO:0000313 EMBL:KDB12477...}
AOA085P7F3	1614	202	12.5	AOA085P7F3_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KFD77649.1};
AOA0D8KUS9	1978	248	12.5	AOA0D8KUS9_	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:KJF726...}
AOA0E4CJ39	949	163	12.5	AOA0E4CJ39_	SubName: Full=RTX toxins determinant A and related Ca2+-bind...
AOA0F2RXE4	983	163	12.5	AOA0F2RXE4_	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:KJS433...}
AOA0H6P3P1	1009	163	12.5	AOA0H6P3P1_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:0000313 EMBL:K117395.1};
AOA0MOUH96	1357	169	12.5	AOA0MOUH96_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 EMBL:K117395.1};
AOA0POQK41	982	163	12.5	AOA0POQK41_	SubName: Full=Toxin {ECO:0000313 EMBL:ALL40423.1};
AOA0P9QWH9	1465	183	12.5	AOA0P9QWH9_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K117395.1};
AOA0P9U630	1411	177	12.5	AOA0P9U630_	SubName: Full=Putative Insecticidal toxin protein {ECO:0000313 EMBL:K117395.1};
AOA0R4FMU3	1541	192	12.5	AOA0R4FMU3_	SubName: Full=A component of insecticidal toxin complex (Tc)...
A4ABP6	1452	181	12.5	A4ABP6_9GAMM	SubName: Full=Anthrax toxin LF subunit {ECO:0000313 EMBL:EAQ...}
A6E282	1327	166	12.5	A6E282_9RHOB	SubName: Full=RTX toxin and related Ca2+-binding protein-lik...
C6X784	1905	239	12.5	C6X784_METGS	SubName: Full=Putative rhizobiocin/RTX toxin and hemolysin-t...
D3V2U5	1506	188	12.5	D3V2U5_XENBS	SubName: Full=B component of insecticidal toxin complex (Tc)...
D3VGF7	1541	192	12.5	D3VGF7_XENNA	SubName: Full=A component of insecticidal toxin complex (Tc)...
EOSAK8	1434	179	12.5	RHSA_DICD3	RecName: Full=Probable deoxyribonuclease RhsA; EC=3.1.-.-; A...

E4V536	1224	163	12.5	E4V536_ARTGP	SubName: Full=Killer toxin alpha/beta {ECO:0000313 EMBL:EFRO...
K4ZKF8	1100	163	12.5	K4ZKF8_PAEAL	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
V0Z831	1024	163	12.5	V0Z831_ECOLX	SubName: Full=Hemolysin toxin protein A {ECO:0000313 EMBL:ES...
W4HRJ2	811	163	12.5	W4HRJ2_9RHOB	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA077PHH9	1481	184	12.4	AOA077PHH9_	SubName: Full=B component of insecticidal toxin complex (Tc)...
AOA080K2N0	1695	210	12.4	AOA080K2N0_	SubName: Full=RTX toxin or related Ca2+-binding protein {ECO...
AOA085WHZ5	973	162	12.4	AOA085WHZ5_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KFE67308.1};
AOA090R3P4	898	162	12.4	AOA090R3P4_	SubName: Full=RTX toxin putative {ECO:0000313 EMBL:GAL09796...
AOA0B1Z5B3	941	162	12.4	AOA0B1Z5B3_	SubName: Full=Insecticidal toxin complex protein TccC2 {ECO:...
AOA0D2T6X1	1018	162	12.4	AOA0D2T6X1_	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
AOA0D9AU22	941	162	12.4	AOA0D9AU22_	SubName: Full=Insecticidal toxin complex protein TccC2 {ECO:...
AOA0E3W1D8	1417	176	12.4	AOA0E3W1D8_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0H5WPM2	1271	162	12.4	AOA0H5WPM2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:CRZ85881.1};
AOA0N9NNY3	1417	176	12.4	AOA0N9NNY3_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0Q7RJD3	1978	245	12.4	AOA0Q7RJD3_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KQY73131.1};
AOA0R2YEJ3	995	162	12.4	AOA0R2YEJ3_	SubName: Full=Toxin {ECO:0000313 EMBL:KRP46695.1};
AOA0R4FMW4	969	162	12.4	AOA0R4FMW4_	SubName: Full=C component of insecticidal toxin complex (Tc)...
A1EAA3	998	162	12.4	A1EAA3_ECOLX	SubName: Full=Hemolysin toxin protein A {ECO:0000313 EMBL:AB...
C2HXU5	1586	196	12.4	C2HXU5_VIBAB	SubName: Full=RTX (Repeat in toxin) cytotoxin {ECO:0000313 E...
C9SKM3	1305	162	12.4	C9SKM3_VERA1	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
D2ZPN4	1559	194	12.4	D2ZPN4_METSM	SubName: Full=Putative toxin-antitoxin system toxin componen...
D3VGF9	969	162	12.4	D3VGF9_XENNA	SubName: Full=C component of insecticidal toxin complex (Tc)...
F2Q8L6	955	162	12.4	F2Q8L6_SALEE	SubName: Full=Insecticidal toxin {ECO:0000313 EMBL:CAX67951...
I3NIC9	1489	185	12.4	I3NIC9_YERPS	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:C...
I4YK51	1039	162	12.4	I4YK51_9RHIZ	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
J3HXI7	1374	170	12.4	J3HXI7_9BURK	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
N1NQ21	969	162	12.4	N1NQ21_XENNE	SubName: Full=C component of insecticidal toxin complex (Tc)...
Q3SAJ8	1706	211	12.4	Q3SAJ8_9BORD	SubName: Full=Adenylate cyclase toxin {ECO:0000313 EMBL:AAZ5...
W6JBM9	951	162	12.4	W6JBM9_9ENTR	SubName: Full=Insecticidal toxin SepC/TccC {ECO:0000313 EMBL...
AOA024BWN0	1288	161	12.3	AOA024BWN0_	SubName: Full=Toxin {ECO:0000313 EMBL:AHZ25568.1};
AOA060H5F9	1035	161	12.3	AOA060H5F9_	SubName: Full=RTX toxin Ca2+-binding protein {ECO:0000313 EM...
AOA061PX55	1224	160	12.3	AOA061PX55_	SubName: Full=RTX toxin {ECO:0000313 EMBL:GAK19300.1};
AOA061Q3Z9	1940	238	12.3	AOA061Q3Z9_	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
AOA093VPW1	1229	161	12.3	AOA093VPW1_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0A0CT86	936	160	12.3	AOA0A0CT86_	SubName: Full=Toxin {ECO:0000313 EMBL:KGM28057.1};
AOA0A0HNW3	1067	160	12.3	AOA0A0HNW3_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KGM88636.1};
AOA0B2ES17	1311	161	12.3	AOA0B2ES17_	SubName: Full=Toxin {ECO:0000313 EMBL:KHL89529.1};
AOA0B4IRG5	999	161	12.3	AOA0B4IRG5_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AIV05674.1};
AOA0D5RK06	1294	160	12.3	AOA0D5RK06_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AJZ40009.1};
AOA0F4TQK6	1294	160	12.3	AOA0F4TQK6_	SubName: Full=Toxin {ECO:0000313 EMBL:KJZ46339.1};
AOA0F6FGK9	998	160	12.3	AOA0F6FGK9_	SubName: Full=Hemolysin toxin protein {ECO:0000313 EMBL:AJA2...
AOA0F7LM29	948	160	12.3	AOA0F7LM29_	SubName: Full=Toxin {ECO:0000313 EMBL:AKH64254.1};
AOA0G1U216	1068	160	12.3	AOA0G1U216_	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
AOA0H6JMW8	986	160	12.3	AOA0H6JMW8_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:00003...
AOA0H6LPS4	1009	161	12.3	AOA0H6LPS4_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:00003...
AOA0K1QOC3	933	160	12.3	AOA0K1QOC3_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AKU98854.1};
AOA0LOLCD1	967	160	12.3	AOA0LOLCD1_	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:KND47671...
AOA0LOMX15	1140	160	12.3	AOA0LOMX15_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0LON221	991	160	12.3	AOA0LON221_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0N8TFN1	1085	161	12.3	AOA0N8TFN1_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KPZ19657.1}; SubNa...
AOA0QOEI63	1085	161	12.3	AOA0QOEI63_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KPY50799.1};
AOA0ROANM7	1061	161	12.3	AOA0ROANM7_	SubName: Full=Toxin {ECO:0000313 EMBL:KRG46297.1};
A3W655	1655	204	12.3	A3W655_9RHOB	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:EAQ24050....}
A4TXL1	897	160	12.3	A4TXL1_9PROT	SubName: Full=RTX toxins and related Ca2+-binding proteins {...}
A6B2T1	985	160	12.3	A6B2T1_VIBPQ	SubName: Full=RTX toxin {ECO:0000313 EMBL:EDM59502.1};
B3EM99	1279	161	12.3	B3EM99_CHLPB	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
C7BMY6	927	160	12.3	C7BMY6_PHOAA	SubName: Full=Insecticidal toxin complex protein tccc2 {ECO:...
I2S547	998	161	12.3	I2S547_ECOLX	SubName: Full=Hemolysin toxin protein A {ECO:0000313 EMBL:EI...

I4KYK6	1493	183	12.3	I4KYK6_PSEFL	SubName: Full=Insecticidal toxin complex protein TcaC1 {ECO:...
I4L6N1	995	160	12.3	I4L6N1_9PSED	SubName: Full=Insecticidal toxin complex protein TccC2 {ECO:...
I4YZR4	953	161	12.3	I4YZR4_9RHIZ	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
P55129	1023	160	12.3	RTX12_ACTPL	RecName: Full=RTX-I toxin determinant A from serotypes 5/10;...
Q3K7R3	928	160	12.3	Q3K7R3_PSEPF	SubName: Full=Putative insecticidal toxin {ECO:0000313 EMBL:...
Q46716	998	160	12.3	Q46716_ECO57	SubName: Full=Exotoxin {ECO:0000313 EMBL:ALH93973.1}; SubNam...
TOF4N5	1296	160	12.3	TOF4N5_HELFX	SubName: Full=Toxin {ECO:0000313 EMBL:EPZ72814.1};
TONXK1	954	161	12.3	TONXK1_PHOTE	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:EQ...
U7QUH2	954	161	12.3	U7QUH2_PHOTE	SubName: Full=Toxin {ECO:0000313 EMBL:ERT10897.1};
W1IQG7	1051	161	12.3	W1IQG7_9ENTR	SubName: Full=Insecticidal toxin complex protein TccC {ECO:0...
AOA010L7B1	1039	159	12.2	AOA010L7B1_	SubName: Full=Pre-toxin domain with VENN motif family protei...
AOA086SUJ9	1286	159	12.2	AOA086SUJ9_	SubName: Full=Killer toxin subunits alpha/beta-like protein ...
AOA090P6A2	1537	188	12.2	AOA090P6A2_	SubName: Full=RTX toxins and related Ca2+-binding proteins {...
AOA090R9U6	1046	159	12.2	AOA090R9U6_	SubName: Full=RTX toxins and related Ca2+-binding proteins {...
AOA0A3FJ62	1170	159	12.2	AOA0A3FJ62_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KGR36076.1};
AOA0G3BWE4	1447	176	12.2	AOA0G3BWE4_	SubName: Full=Structural toxin protein RtxA {ECO:0000313 EMB...
AOA0H4KU17	868	159	12.2	AOA0H4KU17_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AKO97025.1};
AOA0K9N586	1303	159	12.2	AOA0K9N586_	SubName: Full=Toxin {ECO:0000313 EMBL:KMZ51180.1};
AOA0LON4C9	1710	209	12.2	AOA0LON4C9_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0N9WRZO	1495	183	12.2	AOA0N9WRZO_	SubName: Full=Toxin {ECO:0000313 EMBL:ALI00598.1};
AOA0Q6NMK7	1978	241	12.2	AOA0Q6NMK7_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KQU93220.1};
AOA0Q7CQ26	1978	242	12.2	AOA0Q7CQ26_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KQW60697.1};
AOA0Q7DL43	1978	242	12.2	AOA0Q7DL43_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KQW73920.1};
AOA0Q8XD46	1978	242	12.2	AOA0Q8XD46_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KRC79525.1};
AOA0Q8ZOF2	1978	242	12.2	AOA0Q8ZOF2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KRC99918.1};
A6AYI7	1011	159	12.2	A6AYI7_VIBPQ	SubName: Full=RTX toxin {ECO:0000313 EMBL:EDM61098.1};
B3V688	1712	209	12.2	B3V688_9EURY	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:ACF09812....
B3ZYH5	976	159	12.2	B3ZYH5_BACCE	SubName: Full=Clostridial binary toxin B/anthrax toxin PA fa...
C4SEI1	1493	182	12.2	C4SEI1_YERMO	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:E...
C5CZA8	944	159	12.2	C5CZA8_VARPS	SubName: Full=Putative membrane protein involved in toxin up...
C8C4L1	1022	159	12.2	C8C4L1_ACTPL	SubName: Full=RTX-I toxin determinant A {ECO:0000313 EMBL:AC...
D2TDG7	1094	159	12.2	D2TDG7_ERWP6	SubName: Full=Putative insecticidal toxin {ECO:0000313 EMBL:C...
G8QBG5	1434	175	12.2	G8QBG5_PSEFL	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:A...
K9TCW9	1700	208	12.2	K9TCW9_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
M1RRU6	1588	193	12.2	M1RRU6_9PROT	SubName: Full=RTX toxin related protein {ECO:0000313 EMBL:AG...
M2ZTJ2	1626	199	12.2	M2ZTJ2_9PROT	SubName: Full=RTX toxin {ECO:0000313 EMBL:EME70692.1};
N1NR59	1519	186	12.2	N1NR59_XENNE	SubName: Full=B component of insecticidal toxin complex (Tc)...
O85155	965	159	12.2	O85155_PHOLU	SubName: Full=Insecticidal toxin complex protein TccA {ECO:0...
QOK3D5	1552	190	12.2	QOK3D5_CUPNH	SubName: Full=RTX toxin exported protein {ECO:0000313 EMBL:C...
AOA024C932	1313	159	12.1	AOA024C932_	SubName: Full=Toxin {ECO:0000313 EMBL:AHZ26951.1};
AOA0A7S1A5	1834	222	12.1	AOA0A7S1A5_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AJA45248.1};
AOA0D0JLD3	1504	182	12.1	AOA0D0JLD3_	SubName: Full=Toxin {ECO:0000313 EMBL:KIP96133.1};
AOA0F4XQW4	1492	180	12.1	AOA0F4XQW4_	SubName: Full=Toxin {ECO:0000313 EMBL:KKA08299.1};
AOA0J5QA03	1970	238	12.1	AOA0J5QA03_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA0L0LZ72	1149	158	12.1	AOA0L0LZ72_	SubName: Full=Toxin subunit {ECO:0000313 EMBL:KND55378.1};
AOA0L1P552	1280	158	12.1	AOA0L1P552_	SubName: Full=Toxin {ECO:0000313 EMBL:KNH48185.1};
AOA0P0Z828	1111	158	12.1	AOA0P0Z828_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA0P7KQS1	948	158	12.1	AOA0P7KQS1_	SubName: Full=Toxin {ECO:0000313 EMBL:KPN93642.1};
AOA0P9NAX9	1468	177	12.1	AOA0P9NAX9_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9V8N8	1468	177	12.1	AOA0P9V8N8_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0R3A5S3	994	158	12.1	AOA0R3A5S3_	SubName: Full=Toxin {ECO:0000313 EMBL:KRP67548.1};
B8K258	1157	158	12.1	B8K258_BACTU	SubName: Full=Spherical crystal protein {ECO:0000313 EMBL:AC...
D3VOQ1	932	158	12.1	D3VOQ1_XENBS	SubName: Full=C component of insecticidal toxin complex (Tc)...
EOF4H2	1671	202	12.1	EOF4H2_ACTPL	SubName: Full=RTX toxin protein {ECO:0000313 EMBL:EFM96323.1...
E5AUA2	1489	180	12.1	E5AUA2_BURRH	SubName: Full=Insecticidal toxin complex protein TcaC {ECO:0...
F2KBS0	1493	181	12.1	F2KBS0_PSEBN	SubName: Full=Putative toxin {ECO:0000313 EMBL:AEA66852.1};
F9RFQ6	1129	158	12.1	F9RFQ6_VIBSN	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:EGU31431....
I2UHR4	998	158	12.1	I2UHR4_ECOLX	SubName: Full=Hemolysin toxin protein A {ECO:0000313 EMBL:EI...

I9XFL8	919	158	12.1	I9XFL8_RHILT	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
K8R5K8	1213	158	12.1	K8R5K8_9BURK	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
K9QRX4	1894	229	12.1	K9QRX4_NOSS7	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
M7CK10	1480	179	12.1	M7CK10_MORMO	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:EMP53...
P55128	1023	158	12.1	RTX11_ACTPL	RecName: Full=RTX-I toxin determinant A from serotypes 1/9; ...
Q7N7Y7	938	158	12.1	Q7N7Y7_PHOLL	SubName: Full=Insecticidal toxin complex protein TccC5 {ECO:...
Q93NP2	1022	158	12.1	Q93NP2_ACTPL	SubName: Full=RTX toxin IA {ECO:0000313 EMBL:AAK50051.1};
S0DVV9	1226	158	12.1	S0DVV9_GIBF5	SubName: Full=AM-toxin synthetase (AMT) {ECO:0000313 EMBL:CC...
S9YK37	1307	158	12.1	S9YK37_9CETA	RecName: Full=Multidrug and toxin extrusion protein {ECO:000...
TOQJY1	1332	161	12.1	TOQJY1_PHOTE	SubName: Full=Insecticidal toxin complex protein TcdB2 {ECO:...
T5A8IO	1393	168	12.1	T5A8IO_OPHSC	SubName: Full=Killer toxin alpha/beta {ECO:0000313 EMBL:EQLO...
V5WSW7	1350	164	12.1	V5WSW7_PAEPO	SubName: Full=RTX toxin {ECO:0000313 EMBL:AH818551.1};
V8R2R5	928	158	12.1	V8R2R5_9PSED	SubName: Full=Insecticidal toxin complex protein TccC2 {ECO:...
AOA014NAGO	975	157	12.0	AOA014NAGO_	SubName: Full=Toxin TccC3 {ECO:0000313 EMBL:EXU76398.1};
AOA031FRGO	1969	237	12.0	AOA031FRGO_	SubName: Full=RTX toxin+-binding protein {ECO:0000313 EMBL:E...
AOA067LGD0	2004	241	12.0	AOA067LGD0_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KDP89547.1};
AOA077QL98	1506	180	12.0	AOA077QL98_	SubName: Full=B component of insecticidal toxin complex (Tc)...
AOA084A6I7	1037	157	12.0	AOA084A6I7_	SubName: Full=RTX-I toxin determinant A from serotypes 1/9 {...
AOA090S861	864	157	12.0	AOA090S861_	SubName: Full=RTX toxins determinant A and related Ca2+-bind...
AOA099LTY2	1006	156	12.0	AOA099LTY2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KGG10752.1};
AOA0A0CVJ5	1365	164	12.0	AOA0A0CVJ5_	SubName: Full=Toxin {ECO:0000313 EMBL:KGM29663.1};
AOA0F6LN77	999	156	12.0	AOA0F6LN77_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KIS77576.1};
AOA0H6D2Y2	1248	156	12.0	AOA0H6D2Y2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:CSC44920.1};
AOA0H6EXW7	1248	157	12.0	AOA0H6EXW7_	SubName: Full=RTX toxin {ECO:0000313 EMBL:CSB19839.1};
AOA0H6PGY8	974	156	12.0	AOA0H6PGY8_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:00003...
AOA0H6QCA9	1254	157	12.0	AOA0H6QCA9_	SubName: Full=RTX toxin {ECO:0000313 EMBL:CSC09699.1}; EC=5....
AOA0K1PG08	1457	175	12.0	AOA0K1PG08_	SubName: Full=Insecticidal toxin complex protein TccB1 {ECO:...
AOA0K9MWR3	1296	156	12.0	AOA0K9MWR3_	SubName: Full=Toxin {ECO:0000313 EMBL:KMZ48250.1};
AOA0LOQB80	1296	157	12.0	AOA0LOQB80_	SubName: Full=Toxin {ECO:0000313 EMBL:KNE15784.1};
AOA0L1MJD9	1361	163	12.0	AOA0L1MJD9_	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
AOA0L1MJF0	1498	180	12.0	AOA0L1MJF0_	SubName: Full=Toxin {ECO:0000313 EMBL:KNH28536.1};
AOA0L8TL15	999	156	12.0	AOA0L8TL15_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KKC74140.1};
AOA0M5IWV0	851	156	12.0	AOA0M5IWV0_	SubName: Full=Structural toxin protein RtxA {ECO:0000313 EMB...
AOA0M8NG51	1304	156	12.0	AOA0M8NG51_	SubName: Full=Toxin {ECO:0000313 EMBL:KOS31806.1};
AOA0M9ESG5	1038	157	12.0	AOA0M9ESG5_	SubName: Full=Hc-toxin efflux carrier toxa {ECO:0000313 EMBL...
AOA0NOBW33	948	157	12.0	AOA0NOBW33_	SubName: Full=Toxin {ECO:0000313 EMBL:KOY03544.1};
AOA0NOC8E7	999	156	12.0	AOA0NOC8E7_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KOY32530.1};
AOA0NOEI46	1158	157	12.0	AOA0NOEI46_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0N1JNF6	1615	193	12.0	AOA0N1JNF6_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9XS59	1017	156	12.0	AOA0P9XS59_	SubName: Full=Toxin RelE {ECO:0000313 EMBL:KPY38469.1};
AOA0Q0SMG2	1271	156	12.0	AOA0Q0SMG2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KQA63789.1};
A3N292	1022	157	12.0	A3N292_ACTP2	SubName: Full=RTX-I toxin determinant A {ECO:0000313 EMBL:AB...
A7K4R4	1011	157	12.0	A7K4R4_VIBAE	SubName: Full=RTX toxin {ECO:0000313 EMBL:EDN56406.1};
B3H1M6	1002	156	12.0	B3H1M6_ACTP7	SubName: Full=RTX toxin protein {ECO:0000313 EMBL:ACE61703.1...
B5YBP5	1039	156	12.0	B5YBP5_DICT6	SubName: Full=Leukocidin/Hemolysin toxin family {ECO:0000313...
D3QWL7	1154	157	12.0	D3QWL7_ECOCB	SubName: Full=Putative macrophage toxin {ECO:0000313 EMBL:AD...
E0F5N9	1022	157	12.0	E0F5N9_ACTPL	SubName: Full=RTX-I toxin determinant A from serotypes 5/10 ...
E3DDQ6	1209	156	12.0	E3DDQ6_ERWSE	SubName: Full=Insecticidal toxin complex protein TcaB (Toxin...
G4TFH1	1111	156	12.0	G4TFH1_PIRID	SubName: Full=Related to KRE33-Killer toxin REsistant protei...
I2RLF1	997	157	12.0	I2RLF1_EC0LX	SubName: Full=Hemolysin toxin protein A {ECO:0000313 EMBL:EI...
K9QTC3	1706	205	12.0	K9QTC3_NOSS7	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
M6UFF0	1383	166	12.0	M6UFF0_9LEPT	SubName: Full=Insecticide toxin TcdB middle/N-terminal domai...
M7CV39	1413	169	12.0	M7CV39_MORMO	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:EMP52...
Q2W244	1139	157	12.0	Q2W244_MAGSA	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
Q693A0	1045	156	12.0	Q693A0_YEREN	SubName: Full=Putative insecticidal toxin complex protein {E...
Q7CUW1	1937	233	12.0	Q7CUW1_AGRFC	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:AAK890...
Q7N7Y4	970	157	12.0	Q7N7Y4_PHOLL	SubName: Full=Insecticidal toxin complex protein TccC3 {ECO:...
Q8X7W9	1144	157	12.0	Q8X7W9_EC057	SubName: Full=Putative macrophage toxin {ECO:0000313 EMBL:AA...

S6GWE9	1267	156	12.0	S6GWE9_9PSED	SubName: Full=Putative insecticidal toxin protein {ECO:00003...
S9QXX6	1167	157	12.0	S9QXX6_9RHOB	SubName: Full=RTX toxins/related Ca2+-binding protein {ECO:0...
TOF3P5	1296	157	12.0	TOF3P5_HELPHX	SubName: Full=Toxin {ECO:0000313 EMBL:EPZ92706.1};
T2T3Q2	1295	157	12.0	T2T3Q2_HELPHX	SubName: Full=Toxin {ECO:0000313 EMBL:EQD99512.1};
V6L708	1304	156	12.0	V6L708_HELPHX	SubName: Full=Toxin {ECO:0000313 EMBL:EST40205.1};
W1J1L4	952	156	12.0	W1J1L4_9ENTR	SubName: Full=C component of insecticidal toxin complex (Tc)...
W6I337	1580	189	12.0	W6I337_9PROT	SubName: Full=Insecticidal toxin complex protein TcaC {ECO:0...
X2HYF9	1303	156	12.0	X2HYF9_HELPHX	SubName: Full=Toxin {ECO:0000313 EMBL:AHN36352.1};
AOA017T4E0	1575	188	11.9	AOA017T4E0_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA062DYP5	1713	204	11.9	AOA062DYP5_	SubName: Full=Pre-toxin domain with VENN motif family protei...
AOA077PBT7	1474	175	11.9	AOA077PBT7_	SubName: Full=B component of insecticidal toxin complex (Tc)...
AOA088X658	806	155	11.9	AOA088X658_	SubName: Full=Cytolethal distending toxin A/C family protein...
AOA098VXV5	1467	175	11.9	AOA098VXV5_	SubName: Full=Killer toxin insensitive protein 3-like protei...
AOA0A0HKN7	1736	207	11.9	AOA0A0HKN7_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KGM87461.1};
AOA0A1E9P6	1269	155	11.9	AOA0A1E9P6_	SubName: Full=Putative autotransporter adhesin/RTX toxin, li...
AOA0A2CL52	974	155	11.9	AOA0A2CL52_	SubName: Full=Toxin secretion ABC transporter ATP-binding pr...
AOA0A8KP40	1285	155	11.9	AOA0A8KP40_	SubName: Full=Autotransporter {ECO:0000313 EMBL:KP065347.1};...
AOA0A8P399	1156	155	11.9	AOA0A8P399_	SubName: Full=A component of insecticidal toxin complex (Tc)...
AOA0B2EJD4	1312	156	11.9	AOA0B2EJD4_	SubName: Full=Toxin {ECO:0000313 EMBL:KHL85470.1};
AOA0E0Y501	1285	155	11.9	AOA0E0Y501_	SubName: Full=Serine protease pet (Plasmid-encoded toxin pet...
AOA0J6H5Q3	1357	161	11.9	AOA0J6H5Q3_	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
AOA0J6H6P4	933	155	11.9	AOA0J6H6P4_	SubName: Full=Toxin {ECO:0000313 EMBL:KMM90133.1};
AOA0J8H4H1	1323	158	11.9	AOA0J8H4H1_	SubName: Full=Toxin {ECO:0000313 EMBL:KMT68339.1};
AOA0J8H4T0	1296	155	11.9	AOA0J8H4T0_	SubName: Full=Toxin {ECO:0000313 EMBL:KMT68088.1};
AOA0J9F897	931	155	11.9	AOA0J9F897_	SubName: Full=Toxin {ECO:0000313 EMBL:KMW74558.1};
AOA0K9NOZ6	1295	155	11.9	AOA0K9NOZ6_	SubName: Full=Toxin {ECO:0000313 EMBL:KMZ50386.1};
AOA0LOPPC9	1296	155	11.9	AOA0LOPPC9_	SubName: Full=Toxin {ECO:0000313 EMBL:KNE08225.1};
AOA0L1MBQ3	1572	187	11.9	AOA0L1MBQ3_	SubName: Full=Toxin {ECO:0000313 EMBL:KNH25674.1};
AOA0L6DOK0	1323	158	11.9	AOA0L6DOK0_	SubName: Full=Toxin {ECO:0000313 EMBL:KNX43652.1};
AOA0L6D7N3	1303	155	11.9	AOA0L6D7N3_	SubName: Full=Toxin {ECO:0000313 EMBL:KNX46152.1};
AOA0L8BIB0	1978	236	11.9	AOA0L8BIB0_	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:KOF142...
AOA0M4H170	1994	237	11.9	AOA0M4H170_	SubName: Full=RTX toxin {ECO:0000313 EMBL:ALD13312.1};
AOA0M9EIV2	883	155	11.9	AOA0M9EIV2_	SubName: Full=RTX-I toxin determinant A from serotypes 1/9 {...
AOA0P9LCV6	1617	193	11.9	AOA0P9LCV6_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0P9U4D9	1491	178	11.9	AOA0P9U4D9_	SubName: Full=Putative insecticidal toxin complex protein B ...
AOA0Q7A1W4	1492	178	11.9	AOA0Q7A1W4_	SubName: Full=Toxin {ECO:0000313 EMBL:KQW30036.1};
AOA0Q8YU97	964	155	11.9	AOA0Q8YU97_	SubName: Full=Toxin {ECO:0000313 EMBL:KRC96803.1};
AOA0R4FMT9	1519	181	11.9	AOA0R4FMT9_	SubName: Full=B component of insecticidal toxin complex (Tc)...
AOQLI5	825	155	11.9	MAV_MYCA1	RecName: Full=Putative NAD(+)--arginine ADP-ribosyltransfera...
A5CT14	2004	238	11.9	A5CT14_CLAM3	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:CAN02239....
B7LBY5	1285	155	11.9	B7LBY5_ECO55	SubName: Full=Serine protease pet (Plasmid-encoded toxin pet...
C4SCQ5	1299	155	11.9	C4SCQ5_YERMO	SubName: Full=RTX toxin and Ca2+-binding protein {ECO:000031...
C7BJB9	926	155	11.9	C7BJB9_PHOAA	SubName: Full=Insecticidal toxin complex TccC {ECO:0000313 E...
D2U4E2	1198	155	11.9	D2U4E2_9ENTR	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
D3V3H9	901	155	11.9	D3V3H9_XENBS	SubName: Full=C component of insecticidal toxin complex {ECO...
D3VGF8	1519	181	11.9	D3VGF8_XENNA	SubName: Full=B component of insecticidal toxin complex (Tc)...
EOE9Q4	1022	155	11.9	EOE9Q4_ACTPL	SubName: Full=RTX-I toxin determinant A from serotypes 5/10 ...
EOEZM2	1022	155	11.9	EOEZM2_ACTPL	SubName: Full=RTX-I toxin determinant A from serotypes 5/10 ...
EOFC15	1022	155	11.9	EOFC15_ACTPL	SubName: Full=RTX-I toxin determinant A from serotypes 5/10 ...
E5AUA0	1181	155	11.9	E5AUA0_BURRH	SubName: Full=Insecticidal toxin complex protein TccB {ECO:0...
KOG960	1022	155	11.9	KOG960_ACTSU	SubName: Full=RTX-I toxin determinant A {ECO:0000313 EMBL:AF...
K9QLH8	1683	200	11.9	K9QLH8_NOSS7	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
K9QT96	932	155	11.9	K9QT96_NOSS7	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
M5BA51	2002	238	11.9	M5BA51_9MICO	SubName: Full=Conserved secreted protein, putative RTX toxin...
M6QKI1	1688	201	11.9	M6QKI1_9LEPT	SubName: Full=Toxin HINT domain protein {ECO:0000313 EMBL:EM...
O95271	1327	158	11.9	TNKS1_HUMAN	RecName: Full=Tankyrase-1; Short=TANK1; EC=2.4.2.30; AltName...
QOFTY6	1769	211	11.9	QOFTY6_PELBH	SubName: Full=RTX toxin, putative {ECO:0000313 EMBL:EAU47613...
Q6J4J3	950	155	11.9	Q6J4J3_PLUXY	SubName: Full=Insecticidal Bt toxin receptor APN2 {ECO:00003...

ROEGR9	1491	177	11.9	ROEGR9_CAUVI	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
R4QAN6	1323	157	11.9	R4QAN6_HELXP	SubName: Full=Toxin {ECO:0000313 EMBL:AGL71178.1};
R8BW59	1311	156	11.9	R8BW59_TOGMI	SubName: Full=Putative killer toxin subunits alpha beta prot...
V4JFQ2	1436	171	11.9	V4JFQ2_9GAMM	SubName: Full=RTX toxin related Ca2+-binding protein {ECO:00...
WOWO43	1335	159	11.9	WOWO43_ZYGBA	SubName: Full=Related to Killer toxin-resistance protein 5 {...
W8PNS3	1370	163	11.9	W8PNS3_9PSED	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
X5JHX6	888	155	11.9	X5JHX6_9NOST	SubName: Full=Toxin secretion ABC transporter ATP-binding pr...
AOA023Z7Z0	951	154	11.8	AOA023Z7Z0_	SubName: Full=Hemolysin toxin protein {ECO:0000313 EMBL:AHY7...
AOA077PD61	1503	178	11.8	AOA077PD61_	SubName: Full=B component of insecticidal toxin complex (Tc)...
AOA077Q4R2	1030	154	11.8	AOA077Q4R2_	SubName: Full=C component of insecticidal toxin complex (Tc)...
AOA086RUE3	1296	154	11.8	AOA086RUE3_	SubName: Full=Toxin {ECO:0000313 EMBL:KFH28475.1};
AOA086T4H6	1375	162	11.8	AOA086T4H6_	SubName: Full=Killer toxin subunits alpha/beta-like protein ...
AOA086ZG83	1490	176	11.8	AOA086ZG83_	SubName: Full=Putative diarrheal toxin {ECO:0000313 EMBL:KFI...
AOA089Q1C3	989	154	11.8	AOA089Q1C3_	SubName: Full=Toxin {ECO:0000313 EMBL:AIRO6093.1};
AOA0A1HYZ9	1372	162	11.8	AOA0A1HYZ9_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0A2B503	972	154	11.8	AOA0A2B503_	SubName: Full=Toxin secretion ABC transporter ATP-binding pr...
AOA0A2VXC7	1443	170	11.8	AOA0A2VXC7_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0B2EAS0	1312	155	11.8	AOA0B2EAS0_	SubName: Full=Toxin {ECO:0000313 EMBL:KHL82051.1};
AOA0C1ZRM5	1042	154	11.8	AOA0C1ZRM5_	SubName: Full=Toxin {ECO:0000313 EMBL:KIF60085.1};
AOA0D6VJV1	1528	180	11.8	AOA0D6VJV1_	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:K...
AOA0E1W3D2	1528	180	11.8	AOA0E1W3D2_	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:E...
AOA0H0Y3P6	1005	154	11.8	AOA0H0Y3P6_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KLI68771.1};
AOA0H3EFD1	1376	163	11.8	AOA0H3EFD1_	SubName: Full=Vacuolating autotransporter toxin {ECO:0000313...
AOA0H5FUX1	1500	177	11.8	AOA0H5FUX1_	SubName: Full=Putative insecticidal toxin complex protein {E...
AOA0H5GFY0	1500	177	11.8	AOA0H5GFY0_	SubName: Full=Putative insecticidal toxin complex protein {E...
AOA0J6GE41	1503	177	11.8	AOA0J6GE41_	SubName: Full=Toxin {ECO:0000313 EMBL:KMM83031.1};
AOA0J9F852	1364	161	11.8	AOA0J9F852_	SubName: Full=Toxin {ECO:0000313 EMBL:KMW74533.1};
AOA0K9MP87	1323	156	11.8	AOA0K9MP87_	SubName: Full=Toxin {ECO:0000313 EMBL:KMZ45605.1};
AOA0L1P546	933	154	11.8	AOA0L1P546_	SubName: Full=Toxin {ECO:0000313 EMBL:KNH48183.1};
AOA0M8NWW9	1313	155	11.8	AOA0M8NWW9_	SubName: Full=Toxin {ECO:0000313 EMBL:KOS35951.1};
AOA0M9B2C8	1369	162	11.8	AOA0M9B2C8_	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
AOA0M9W666	1295	154	11.8	AOA0M9W666_	SubName: Full=Toxin {ECO:0000313 EMBL:KOS32609.1};
AOA0N8VN45	1043	154	11.8	AOA0N8VN45_	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
AOA0Q6DOW5	1616	191	11.8	AOA0Q6DOW5_	SubName: Full=Insecticidal toxin {ECO:0000313 EMBL:KQT63456....}
AOA0Q9EQ07	1979	233	11.8	AOA0Q9EQ07_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KRD66014.1};
AOA0R4FN23	1156	154	11.8	AOA0R4FN23_	SubName: Full=A component of insecticidal toxin complex (Tc)...
A5PDA6	1933	228	11.8	A5PDA6_9SPHN	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
A6E154	983	154	11.8	A6E154_9RHOB	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:EDM315...
A8EM04	1471	173	11.8	A8EM04_BURPE	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:E...
A8EM19	1528	181	11.8	A8EM19_BURPE	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:E...
B7MC95	1376	163	11.8	B7MC95_ECO45	SubName: Full=Vacuolating autotransporter toxin {ECO:0000313...
C9Q790	1924	227	11.8	C9Q790_9VIBR	SubName: Full=RTX toxin putative {ECO:0000313 EMBL:EEX65724....}
C9SGR7	1242	154	11.8	C9SGR7_VERA1	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
D3VHH3	1156	154	11.8	D3VHH3_XENNA	SubName: Full=A component of insecticidal toxin complex (Tc)...
E3DDQ7	1431	169	11.8	E3DDQ7_ERWSE	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
E3EHS9	1529	181	11.8	E3EHS9_PAEPS	SubName: Full=RTX toxin {ECO:0000313 EMBL:AD056341.2};
E4V1K1	1258	154	11.8	E4V1K1_ARTGP	SubName: Full=Killer toxin alpha/beta {ECO:0000313 EMBL:EFR0...
F2BUS8	1478	175	11.8	F2BUS8_STRSA	SubName: Full=Diarrheal toxin {ECO:0000313 EMBL:EGF05514.1};...
F7UEQ7	1978	234	11.8	F7UEQ7_RHIRD	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:EGP559...
H7FNV2	1285	154	11.8	H7FNV2_9FLAO	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
I4K1X3	937	154	11.8	I4K1X3_PSEFL	SubName: Full=Insecticidal toxin complex protein TccC2 {ECO:...
L2FERO	1296	154	11.8	L2FERO_COLGN	SubName: Full=Killer toxin alpha beta {ECO:0000313 EMBL:ELA2...
L8MBA3	1294	154	11.8	L8MBA3_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
N1NRV3	1156	154	11.8	N1NRV3_XENNE	SubName: Full=A component of insecticidal toxin complex (Tc)...
P11103	1013	154	11.8	PARP1_MOUSE	RecName: Full=Poly [ADP-ribose] polymerase 1; Short=PARP-1; ...
P26446	1011	154	11.8	PARP1_CHICK	RecName: Full=Poly [ADP-ribose] polymerase 1; Short=PARP-1; ...
P27008	1014	154	11.8	PARP1_RAT	RecName: Full=Poly [ADP-ribose] polymerase 1; Short=PARP-1; ...
Q2SFE6	2095	248	11.8	Q2SFE6_HAHCH	SubName: Full=RTX toxins and related Ca2+-binding protein {E...

Q7N7Z1	915	154	11.8	Q7N7Z1_PHOLL	SubName: Full=Insecticidal toxin complex protein TccC2 {ECO:...
S3MJM4	937	154	11.8	S3MJM4_PSESY	SubName: Full=Putative insecticidal toxin complex protein C2...
U4RBV3	1323	156	11.8	U4RBV3_HELPX	SubName: Full=Toxin {ECO:0000313 EMBL:EPZ94982.1};
W1IN06	1471	174	11.8	W1IN06_9ENTR	SubName: Full=B component of insecticidal toxin complex (Tc)...
W8PP50	912	154	11.8	W8PP50_9PSED	SubName: Full=Toxin {ECO:0000313 EMBL:AHL35703.1};
AOA031FT93	925	153	11.7	AOA031FT93_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:E...
AOA031G3P8	1505	176	11.7	AOA031G3P8_	SubName: Full=Toxin {ECO:0000313 EMBL:EZP31473.1};
AOA031G3Q8	1507	177	11.7	AOA031G3Q8_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA059L1E8	1109	153	11.7	AOA059L1E8_	SubName: Full=Toxin {ECO:0000313 EMBL:KDD68056.1};
AOA059L1Y7	1357	159	11.7	AOA059L1Y7_	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
AOA061PX21	962	152	11.7	AOA061PX21_	SubName: Full=RTX toxin {ECO:0000313 EMBL:GAK19302.1};
AOA077NG66	1524	178	11.7	AOA077NG66_	SubName: Full=Insecticidal toxin complex protein TccB (Modul...
AOA077P3L7	1030	152	11.7	AOA077P3L7_	SubName: Full=C component of insecticidal toxin complex (Tc)...
AOA090BP84	865	153	11.7	AOA090BP84_	SubName: Full=Hansenula MRKII killer toxin-resistant protei...
AOA090P2X0	873	152	11.7	AOA090P2X0_	SubName: Full=RTX toxins and related Ca2+-binding proteins {...
AOA090QIX4	863	152	11.7	AOA090QIX4_	SubName: Full=RTX toxins and related Ca2+-binding proteins {...
AOA0A2VU88	1664	194	11.7	AOA0A2VU88_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0B2DTA3	1303	153	11.7	AOA0B2DTA3_	SubName: Full=Toxin {ECO:0000313 EMBL:KHL77604.1};
AOA0C1WN19	1490	174	11.7	AOA0C1WN19_	SubName: Full=Toxin {ECO:0000313 EMBL:KIF63853.1};
AOA0H5GMI1	1500	175	11.7	AOA0H5GMI1_	SubName: Full=Putative insecticidal toxin complex protein {E...
AOA0L6D4L2	1296	152	11.7	AOA0L6D4L2_	SubName: Full=Toxin {ECO:0000313 EMBL:KNX44925.1};
AOA0N1CR67	903	153	11.7	AOA0N1CR67_	SubName: Full=Toxin {ECO:0000313 EMBL:KPG79182.1};
AOA0N1GZ45	998	152	11.7	AOA0N1GZ45_	SubName: Full=Putative HC-toxin efflux carrier TOXA {ECO:000...
AOA0N7H2T3	1440	169	11.7	AOA0N7H2T3_	SubName: Full=Toxin {ECO:0000313 EMBL:ALI09620.1};
AOA0N8SXR7	1449	169	11.7	AOA0N8SXR7_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0N9VRS2	933	152	11.7	AOA0N9VRS2_	SubName: Full=Toxin {ECO:0000313 EMBL:ALI00599.1};
AOA0P7MYJ0	1369	160	11.7	AOA0P7MYJ0_	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
AOA0P9W539	1064	152	11.7	AOA0P9W539_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
CONC57	1068	153	11.7	CONC57_AJECG	SubName: Full=Killer toxin resistant protein {ECO:0000313 EM...
C3QP10	964	153	11.7	C3QP10_9BACE	SubName: Full=Diphtheria toxin, C domain protein {ECO:000031...
C7BMQ9	953	152	11.7	C7BMQ9_PHOAA	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:CA...
D2TDG8	1210	152	11.7	D2TDG8_ERWP6	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:CAY72...
E0SGL7	1658	194	11.7	RHSC_DICD3	RecName: Full=Putative deoxyribonuclease RhsC; EC=3.1.-.-; A...
E2XVN8	1506	176	11.7	E2XVN8_PSEFL	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:E...
E9FBL5	908	153	11.7	E9FBL5_METRA	SubName: Full=Diphtheria toxin, translocation domain protein...
F3H2L6	923	152	11.7	F3H2L6_PSESX	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
H8H5S3	815	153	11.7	H8H5S3_HELPX	SubName: Full=Toxin-like outer membrane protein {ECO:0000313...
H8NTY2	1130	152	11.7	H8NTY2_RAHAQ	SubName: Full=Insecticidal toxin complex protein TcaC {ECO:0...
I3AP72	939	153	11.7	I3AP72_SERPL	SubName: Full=Toxin protein {ECO:0000313 EMBL:EIJ20915.1};
I4K0U3	905	152	11.7	I4K0U3_PSEFL	SubName: Full=Insecticidal toxin complex protein TccC2 {ECO:...
I4L5Y3	1527	179	11.7	I4L5Y3_9PSED	SubName: Full=Insecticidal toxin complex protein TcaC2 {ECO:...
I4YYX3	741	152	11.7	I4YYX3_9RHIZ	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
K9TR73	1740	203	11.7	K9TR73_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
L8LWF5	1165	152	11.7	L8LWF5_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
L9KIN4	1103	153	11.7	L9KIN4_TUPCH	RecName: Full=Multidrug and toxin extrusion protein {ECO:000...
M1PTS1	1153	153	11.7	M1PTS1_DESSD	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
M3J5B4	1442	169	11.7	M3J5B4_CANMX	SubName: Full=Killer toxin-resistance protein, putative {ECO...
N1NU07	1021	153	11.7	N1NU07_XENNE	SubName: Full=A component of insecticidal toxin complex (Tc)...
P09805	1146	153	11.7	KTXA_KLULA	RecName: Full=Killer toxin subunits alpha/beta; AltName: Ful...
P26831	1628	191	11.7	NAGH_CLOPE	RecName: Full=Hyaluronoglucosaminidase; Short=Hyaluronidase;...
Q1GFJ9	867	152	11.7	Q1GFJ9_RUEST	SubName: Full=RTX toxins and related Ca2+-binding proteins-l...
Q48JB4	1157	152	11.7	Q48JB4_PSE14	SubName: Full=Insecticidal toxin complex protein, putative {...
Q6EIX3	1177	153	11.7	Q6EIX3_BACTU	SubName: Full=Cry {ECO:0000313 EMBL:AAV88348.1}; SubName: Fu...
Q6PFX9	1320	155	11.7	TNKS1_MOUSE	RecName: Full=Tankyrase-1; Short=TANK1; EC=2.4.2.30; AltName...
Q7N7Z0	1476	173	11.7	Q7N7Z0_PHOLL	SubName: Full=Insecticidal toxin complex protein TcdB1 {ECO:...
Q7N8C3	1471	172	11.7	Q7N8C3_PHOLL	SubName: Full=Insecticidal toxin complex protein TccB3 {ECO:...
Q8YPF5	1003	152	11.7	Q8YPF5_NOSS1	SubName: Full=Toxin secretion ABC transporter ATP-binding pr...
R8BS00	1071	153	11.7	R8BS00_TOGMI	SubName: Full=Putative killer toxin subunits alpha beta prot...

S6JAH5	948	152	11.7	S6JAH5_9PSED	SubName: Full=A component of insecticidal toxin complex (Tc)...
TOEPZ2	935	152	11.7	TOEPZ2_HELPHX	SubName: Full=Toxin {ECO:0000313 EMBL:EPZ67774.1};
TOF8R9	1323	155	11.7	TOF8R9_HELPHX	SubName: Full=Toxin {ECO:0000313 EMBL:EPZ74269.1};
TOP1I3	1407	164	11.7	TOP1I3_PHOTE	SubName: Full=Insecticidal toxin complex protein TcdA1 {ECO:...
U1TBE4	960	153	11.7	U1TBE4_PSEFL	SubName: Full=Toxin {ECO:0000313 EMBL:ERH61510.1};
WOTAK9	866	152	11.7	WOTAK9_KLUMA	SubName: Full=Hansenula MRAKII killer toxin-resistant protei...
W8QHR4	926	152	11.7	W8QHR4_9PSED	SubName: Full=Toxin {ECO:0000313 EMBL:ABL35810.1};
AOA059L161	1513	175	11.6	AOA059L161_	SubName: Full=Toxin {ECO:0000313 EMBL:KDD68058.1};
AOA061PQA0	1642	190	11.6	AOA061PQA0_	SubName: Full=RTX toxin and related Ca2+-binding protein {EC...
AOA087IFS5	1005	151	11.6	AOA087IFS5_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KFK53227.1};
AOA090SQV7	971	151	11.6	AOA090SQV7_	SubName: Full=RTX toxins and related Ca2+-binding proteins {...
AOA098RQC7	1977	229	11.6	AOA098RQC7_	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:KGE823...
AOA099MOR7	1007	151	11.6	AOA099MOR7_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KKG13062.1};
AOA0A6D8N8	1501	174	11.6	AOA0A6D8N8_	SubName: Full=Toxin {ECO:0000313 EMBL:KHA71102.1};
AOA0J6JOD7	1504	175	11.6	AOA0J6JOD7_	SubName: Full=Toxin {ECO:0000313 EMBL:KMN17903.1};
AOA0K1PHT8	1027	151	11.6	AOA0K1PHT8_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0LOPVL4	1323	153	11.6	AOA0LOPVL4_	SubName: Full=Toxin {ECO:0000313 EMBL:KNE10435.1};
AOA0LOPZB1	1296	151	11.6	AOA0LOPZB1_	SubName: Full=Toxin {ECO:0000313 EMBL:KNE11720.1};
AOA0LOQOR0	1291	151	11.6	AOA0LOQOR0_	SubName: Full=Toxin {ECO:0000313 EMBL:KNE12195.1};
AOA0LOQ2A9	1291	151	11.6	AOA0LOQ2A9_	SubName: Full=Toxin {ECO:0000313 EMBL:KNE12518.1};
AOA0MOMSA7	1323	154	11.6	AOA0MOMSA7_	SubName: Full=Toxin {ECO:0000313 EMBL:KOO66235.1};
AOA0MOUL17	1463	169	11.6	AOA0MOUL17_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0M2MAA8	1615	187	11.6	AOA0M2MAA8_	SubName: Full=Toxin {ECO:0000313 EMBL:KKI25163.1};
AOA0NOFVD7	967	151	11.6	AOA0NOFVD7_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0NOL6G0	1511	175	11.6	AOA0NOL6G0_	SubName: Full=Toxin {ECO:0000313 EMBL:KPG94069.1};
AOA0P6RMG2	1494	174	11.6	AOA0P6RMG2_	SubName: Full=Toxin {ECO:0000313 EMBL:KPG93377.1};
AOA0P9J3M1	991	151	11.6	AOA0P9J3M1_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9LH36	1444	168	11.6	AOA0P9LH36_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0Q0F3B6	1615	187	11.6	AOA0Q0F3B6_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0QORVG5	1300	151	11.6	AOA0QORVG5_	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
AOA0R2YEB0	1524	177	11.6	AOA0R2YEB0_	SubName: Full=Toxin {ECO:0000313 EMBL:KRP46694.1};
AOA0R4CY34	1021	151	11.6	AOA0R4CY34_	SubName: Full=A component of insecticidal toxin complex (Tc)...
C7BMQ8	1488	173	11.6	C7BMQ8_PHOAA	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:CA...
C7BN44	1545	179	11.6	C7BN44_PHOAA	SubName: Full=Similar to different toxins like syringomycin ...
D3VGF6	1021	151	11.6	D3VGF6_XENNA	SubName: Full=A component of insecticidal toxin complex (Tc)...
F3LIJ2	1088	151	11.6	F3LIJ2_9GAMM	SubName: Full=RTX toxin {ECO:0000313 EMBL:EGG93293.1};
G0YF57	1567	181	11.6	G0YF57_PHOTE	SubName: Full=Insecticidal toxin complex protein TccB {ECO:0...
G8Q748	1493	173	11.6	G8Q748_PSEFL	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:A...
J3FRY0	1981	230	11.6	J3FRY0_9PSED	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
K9NQT2	1549	180	11.6	K9NQT2_9PSED	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:A...
M7TBM7	1314	152	11.6	M7TBM7_BOTF1	SubName: Full=Putative hc-toxin synthetase protein {ECO:0000...
Q2EMV9	1817	210	11.6	PAR14_MOUSE	RecName: Full=Poly [ADP-ribose] polymerase 14; Short=PARP-14...
Q2SJA2	952	151	11.6	Q2SJA2_HAHCH	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
Q7MZV6	1564	181	11.6	Q7MZV6_PHOLL	SubName: Full=Insecticidal toxin complex protein TccB1 {ECO:...
Q7N7X5	959	151	11.6	Q7N7X5_PHOLL	SubName: Full=Insecticidal toxin complex protein TccC4 {ECO:...
Q87X47	1615	187	11.6	Q87X47_PSESM	SubName: Full=Insecticidal toxin protein, putative {ECO:0000...
R8BG26	1040	151	11.6	R8BG26_TOGMI	SubName: Full=Putative killer toxin subunits alpha beta prot...
TOP881	2009	234	11.6	TOP881_AERSA	SubName: Full=RTX toxin-like protein {ECO:0000313 EMBL:EQC03...
U4RH49	1323	153	11.6	U4RH49_HELPHX	SubName: Full=Toxin {ECO:0000313 EMBL:EPZ97500.1};
W1HM31	1009	151	11.6	W1HM31_KLEPN	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0A2W4F4	1874	215	11.5	AOA0A2W4F4_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0A4GPP1	1483	170	11.5	AOA0A4GPP1_	SubName: Full=Toxin {ECO:0000313 EMBL:KGU85192.1};
AOA0A6D946	1370	158	11.5	AOA0A6D946_	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
AOA0F5KHA2	800	150	11.5	AOA0F5KHA2_	SubName: Full=Cytolethal distending toxin A/C family protein...
AOA0F6HYL8	1038	150	11.5	AOA0F6HYL8_	SubName: Full=Toxin HINT domain protein {ECO:0000313 EMBL:EK...
AOA0F7LPL8	954	150	11.5	AOA0F7LPL8_	SubName: Full=Toxin {ECO:0000313 EMBL:AKH63811.1};
AOA0G3AY52	973	150	11.5	AOA0G3AY52_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0H4XLC2	1245	150	11.5	AOA0H4XLC2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AKQ69052.1};



AOA0H6RW05	1953	225	11.5	AOA0H6RW05_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:00003...
AOA0H6SB44	1656	190	11.5	AOA0H6SB44_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:00003...
AOA0H6TLK7	1217	150	11.5	AOA0H6TLK7_	SubName: Full=RTX toxin RtxA {ECO:0000313 EMBL:CSC55598.1};
AOA0J6HCX1	922	150	11.5	AOA0J6HCX1_	SubName: Full=Toxin {ECO:0000313 EMBL:KMM92323.1};
AOA0K8L7C9	1368	157	11.5	AOA0K8L7C9_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0K9N7V3	1457	167	11.5	AOA0K9N7V3_	SubName: Full=Diarrheal toxin {ECO:0000313 EMBL:KMZ52879.1};...
AOA0LOPIE1	1291	150	11.5	AOA0LOPIE1_	SubName: Full=Toxin {ECO:0000313 EMBL:KNE06147.1};
AOA0L1IUZ5	1463	168	11.5	AOA0L1IUZ5_	SubName: Full=Putative killer toxin sensitivity protein (IKI...
AOA0NOF4H4	1043	150	11.5	AOA0NOF4H4_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0N1ZID9	1339	154	11.5	AOA0N1ZID9_	SubName: Full=Insecticide toxin TcdB middle/N-terminal domai...
AOA0P9MSI1	1446	166	11.5	AOA0P9MSI1_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0P9VDC7	941	150	11.5	AOA0P9VDC7_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0QOE035	1636	188	11.5	AOA0QOE035_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0Q6DOZ9	1536	177	11.5	AOA0Q6DOZ9_	SubName: Full=Toxin {ECO:0000313 EMBL:KQT63455.1};
A3J1R3	1334	154	11.5	A3J1R3_9FLA0	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
D3VOQ0	1481	170	11.5	D3VOQ0_XENBS	SubName: Full=B component of insecticidal toxin complex (Tc)...
G2GXS5	1823	210	11.5	G2GXS5_9ENTR	SubName: Full=RTX toxin {ECO:0000313 EMBL:EGY29457.1};
G5JQ85	999	150	11.5	G5JQ85_STRCG	SubName: Full=Toxin-antitoxin system, toxin component family...
I3AG85	930	150	11.5	I3AG85_SERPL	SubName: Full=Toxin protein {ECO:0000313 EMBL:EIJ18128.1};
I3NI94	1192	150	11.5	I3NI94_YERPS	SubName: Full=Putative (AF046867) insecticidal toxin complex...
JOC9P2	1098	150	11.5	JOC9P2_RHILT	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
J2EXL3	1253	150	11.5	J2EXL3_PSEFL	SubName: Full=Insecticidal toxin complex protein TcaB1 {ECO:...
J3ES81	1981	227	11.5	J3ES81_9PSED	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
M6K5S1	977	150	11.5	M6K5S1_9LEPT	SubName: Full=Toxin HINT domain protein {ECO:0000313 EMBL:EM...
N1NPM3	1530	176	11.5	N1NPM3_XENNE	SubName: Full=B component of insecticidal toxin complex (Tc)...
Q693A4	1481	170	11.5	Q693A4_YEREN	SubName: Full=Putative insecticidal toxin complex protein {E...
Q7MZU2	965	150	11.5	Q7MZU2_PHOLL	SubName: Full=Insecticidal toxin complex protein TccC6 {ECO:...
Q7MZV5	966	150	11.5	Q7MZV5_PHOLL	SubName: Full=Insecticidal toxin complex protein TccA1 {ECO:...
R4TLX6	1293	150	11.5	R4TLX6_9VIRU	SubName: Full=Zeta toxin and ParB nuclease domain protein {E...
S6GUV3	940	150	11.5	S6GUV3_9PSED	SubName: Full=Putative insecticidal toxin complex protein {E...
U5MQ68	1435	165	11.5	U5MQ68_CL0SA	SubName: Full=Toxin A {ECO:0000313 EMBL:AGX41831.1};
U7QVZ8	1178	150	11.5	U7QVZ8_PHOTE	SubName: Full=Toxin {ECO:0000313 EMBL:ERT12199.1};
V522Y6	1854	213	11.5	V522Y6_9ENTR	SubName: Full=Toxin A {ECO:0000313 EMBL:CCG85374.1};
W0VTS3	1335	154	11.5	W0VTS3_ZYGBA	SubName: Full=Related to Killer toxin-resistance protein 5 {...
AOA014MUY8	984	149	11.4	AOA014MUY8_	SubName: Full=Toxin TccC3 {ECO:0000313 EMBL:EXU70978.1};
AOA031HCH7	994	149	11.4	AOA031HCH7_	SubName: Full=Zeta toxin family protein {ECO:0000313 EMBL:EZ...
AOA059XJ93	964	149	11.4	AOA059XJ93_	SubName: Full=Insecticidal toxin SepC/TccC {ECO:0000313 EMBL...
AOA061PHM8	875	149	11.4	AOA061PHM8_	SubName: Full=RTX toxin {ECO:0000313 EMBL:GAK16508.1};
AOA075R1D7	1293	149	11.4	AOA075R1D7_	SubName: Full=Putative toxin 56 {ECO:0000313 EMBL:AIG26387.1...
AOA077NHL6	1481	169	11.4	AOA077NHL6_	SubName: Full=B component of insecticidal toxin complex (Tc)...
AOA077SFQ6	1273	149	11.4	AOA077SFQ6_	SubName: Full=Zeta toxin family protein {ECO:0000313 EMBL:CD...
AOA093VDJ1	1264	149	11.4	AOA093VDJ1_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA099SLD6	1617	185	11.4	AOA099SLD6_	SubName: Full=Toxin {ECO:0000313 EMBL:KGK93697.1};
AOA099SXB9	922	149	11.4	AOA099SXB9_	SubName: Full=Toxin {ECO:0000313 EMBL:KGK96636.1};
AOA0A1HR48	1442	165	11.4	AOA0A1HR48_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:CDF92...
AOA0A4GQT7	1523	174	11.4	AOA0A4GQT7_	SubName: Full=Toxin {ECO:0000313 EMBL:KGU87336.1};
AOA0A4HRW0	939	149	11.4	AOA0A4HRW0_	SubName: Full=Insecticidal toxin complex protein TccC2 {ECO:...
AOA0C2RNL7	1512	173	11.4	AOA0C2RNL7_	SubName: Full=Toxin {ECO:0000313 EMBL:KIK87207.1};
AOA0C6PDA9	1464	167	11.4	AOA0C6PDA9_	SubName: Full=Dermonecrotic toxin {ECO:0000313 EMBL:CCJ56339...
AOA0E1NNU2	984	149	11.4	AOA0E1NNU2_	SubName: Full=Insecticidal toxin {ECO:0000313 EMBL:ABG13692.1...
AOA0E1VV90	1485	170	11.4	AOA0E1VV90_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0E3MQ73	996	149	11.4	AOA0E3MQ73_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0E8XLT2	1505	171	11.4	AOA0E8XLT2_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0F4TT39	949	149	11.4	AOA0F4TT39_	SubName: Full=Toxin {ECO:0000313 EMBL:KJZ46542.1};
AOA0F4V1X1	905	149	11.4	AOA0F4V1X1_	SubName: Full=Toxin {ECO:0000313 EMBL:KJZ62515.1};
AOA0F6W1X6	2041	233	11.4	AOA0F6W1X6_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:AKF05...
AOA0H2YIA8	984	149	11.4	AOA0H2YIA8_	SubName: Full=Insecticidal toxin {ECO:0000313 EMBL:ABG18164.1...
AOA0J6ILE9	1504	172	11.4	AOA0J6ILE9_	SubName: Full=Toxin {ECO:0000313 EMBL:KMN13133.1};

AOA0J8S3S6	1189	149	11.4	AOA0J8S3S6_	SubName: Full=Toxin RTX-I translocation ATP-binding protein ...
AOA0K2CYC7	975	149	11.4	AOA0K2CYC7_	SubName: Full=Toxin {ECO:0000313 EMBL:ALA12432.1};
AOA0K2CYN4	975	149	11.4	AOA0K2CYN4_	SubName: Full=Toxin {ECO:0000313 EMBL:ALA12593.1};
AOA0L8IR97	850	149	11.4	AOA0L8IR97_	SubName: Full=Putative insecticidal toxin complex protein C3...
AOA0M1V5X0	984	149	11.4	AOA0M1V5X0_	SubName: Full=Insecticidal toxin {ECO:0000313 EMBL:EDR32051.1...
AOA0M2M500	886	149	11.4	AOA0M2M500_	SubName: Full=Toxin {ECO:0000313 EMBL:KKI25161.1};
AOA0N1JDY8	1617	185	11.4	AOA0N1JDY8_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0N7H1R8	1522	173	11.4	AOA0N7H1R8_	SubName: Full=Toxin {ECO:0000313 EMBL:ALI06700.1};
AOA0N8S9G3	714	149	11.4	AOA0N8S9G3_	SubName: Full=Zonular occludens toxin {ECO:0000313 EMBL:KPY0...
AOA0P9ITF9	959	149	11.4	AOA0P9ITF9_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0P9J3K2	1619	185	11.4	AOA0P9J3K2_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9NYZ4	1619	185	11.4	AOA0P9NYZ4_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9ZP75	885	149	11.4	AOA0P9ZP75_	SubName: Full=ADP-ribosylating toxin family protein {ECO:000...
AOA0Q0A0P1	1549	177	11.4	AOA0Q0A0P1_	SubName: Full=Insecticidal toxin complex protein TcdA1 {ECO:...
AOA0Q0BMP6	886	149	11.4	AOA0Q0BMP6_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0Q8BRQ8	1506	171	11.4	AOA0Q8BRQ8_	SubName: Full=Toxin {ECO:0000313 EMBL:KRA04595.1};
AOA0Q8YUA8	1476	169	11.4	AOA0Q8YUA8_	SubName: Full=Toxin {ECO:0000313 EMBL:KRC96804.1};
AOA0ROLTE4	1284	149	11.4	AOA0ROLTE4_	SubName: Full=Toxin RTX-I translocation ATP-binding protein ...
AOA0R3CF01	1511	172	11.4	AOA0R3CF01_	SubName: Full=Toxin {ECO:0000313 EMBL:KRP93665.1};
A9CX50	1830	208	11.4	A9CX50_9GAMM	SubName: Full=RTX toxin, putative {ECO:0000313 EMBL:EDQ02608...
B1BSY0	875	149	11.4	B1BSY0_CLOPF	SubName: Full=Iota toxin component Ib {ECO:0000313 EMBL:EDT1...
B5IMP3	2003	228	11.4	B5IMP3_9CHRO	SubName: Full=Structural toxin protein RtxA {ECO:0000313 EMB...
G0YF59	1488	169	11.4	G0YF59_PHOTE	SubName: Full=Insecticidal toxin complex protein TcaC {ECO:0...
G2H2F8	1696	194	11.4	G2H2F8_9ENTR	SubName: Full=RTX toxin {ECO:0000313 EMBL:EGY27821.1};
G3KFK0	1165	149	11.4	G3KFK0_BACTA	SubName: Full=Crystal toxin protein 1D {ECO:0000313 EMBL:AEO...
I4Z2L6	1738	199	11.4	I4Z2L6_9RHIZ	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
L2FZJ6	1384	158	11.4	L2FZJ6_COLGN	SubName: Full=Killer toxin subunits alpha beta {ECO:0000313 ...
M7C078	1032	149	11.4	M7C078_CHEMY	RecName: Full=Multidrug and toxin extrusion protein {ECO:000...
M9V7X5	975	149	11.4	M9V7X5_9BACL	SubName: Full=Toxin 1 {ECO:0000313 EMBL:AGJ74029.1};
P41809	1802	205	11.4	HKR1_YEAST	RecName: Full=Signaling mucin HKR1; AltName: Full=Hansenula ...
Q2BFU3	1218	149	11.4	Q2BFU3_9BACI	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
Q2W7U4	1857	211	11.4	Q2W7U4_MAGSA	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
Q3KHR5	942	149	11.4	Q3KHR5_PSEPF	SubName: Full=Putative insecticidal toxin {ECO:0000313 EMBL:...
Q45336	1451	166	11.4	Q45336_BORPT	SubName: Full=Dermonecrotic toxin {ECO:0000313 EMBL:AAA20995...
Q46221	875	149	11.4	Q46221_CLOPF	SubName: Full=Iota toxin component Ib {ECO:0000313 EMBL:CAA5...
Q74TJ0	984	149	11.4	Q74TJ0_YERPE	SubName: Full=Insecticidal toxin {ECO:0000313 EMBL:CAL21008.1...
Q87X45	886	149	11.4	Q87X45_PSESM	SubName: Full=Insecticidal toxin protein, putative {ECO:0000...
Q9L877	1189	149	11.4	Q9L877_BACTU	SubName: Full=Toxin Cry1Ca6 {ECO:0000313 EMBL:AAF37224.1};
Q9R152	1013	149	11.4	PARP1_CRIGR	RecName: Full=Poly [ADP-ribose] polymerase 1; Short=PARP-1; ...
R9VWV6	975	149	11.4	R9VWV6_9CAUD	SubName: Full=Toxin 1 {ECO:0000313 EMBL:AGN89341.1};
S6JLH5	1337	152	11.4	S6JLH5_9PSED	SubName: Full=Putative insecticidal toxin complex protein {E...
TOP9U7	1488	169	11.4	TOP9U7_PHOTE	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:EQ...
T2SHS6	1296	149	11.4	T2SHS6_HELPH	SubName: Full=Toxin {ECO:0000313 EMBL:EQD92321.1};
U4RAS1	1110	149	11.4	U4RAS1_HELPH	SubName: Full=Toxin {ECO:0000313 EMBL:EPZ94607.1};
V5Z4X4	1029	149	11.4	V5Z4X4_9ENTR	SubName: Full=Dermonecrotic toxin DNT {ECO:0000313 EMBL:CCG8...
V8ROL4	1042	149	11.4	V8ROL4_9PSED	SubName: Full=Toxin {ECO:0000313 EMBL:ETF05656.1};
W6K8J7	1238	149	11.4	W6K8J7_9PROT	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
AOA013RV46	1267	147	11.3	AOA013RV46_	SubName: Full=Pre-toxin domain with VENN motif family protei...
AOA031G4M5	1559	176	11.3	AOA031G4M5_	SubName: Full=Toxin {ECO:0000313 EMBL:EZP31482.1};
AOA072NL58	670	147	11.3	AOA072NL58_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA077P6P2	1184	148	11.3	AOA077P6P2_	SubName: Full=A component of insecticidal toxin complex (Tc)...
AOA090RQS1	1068	147	11.3	AOA090RQS1_	SubName: Full=RTX toxin putative {ECO:0000313 EMBL:GAL09797...
AOA0A0XE42	1464	166	11.3	AOA0A0XE42_	SubName: Full=Dermonecrotic toxin {ECO:0000313 EMBL:AIW94618...
AOA0A3K136	1279	148	11.3	AOA0A3K136_	SubName: Full=Insecticidal toxin, SepC/Tcc class domain prot...
AOA0A5HCQ2	975	147	11.3	AOA0A5HCQ2_	SubName: Full=Toxin {ECO:0000313 EMBL:KXG81482.1};
AOA0A6DCA7	1045	148	11.3	AOA0A6DCA7_	SubName: Full=Toxin {ECO:0000313 EMBL:KHA72805.1};
AOA0A6DCJ7	1614	182	11.3	AOA0A6DCJ7_	SubName: Full=Insecticidal toxin {ECO:0000313 EMBL:KHA72855...
AOA0B2EBT4	1290	148	11.3	AOA0B2EBT4_	SubName: Full=Toxin {ECO:0000313 EMBL:KHL82830.1};

AOAODOHXR6	779	147	11.3	AOAODOHXR6_	SubName: Full=Clostridial binary toxin B/anthrax toxin PA fa...
AOAOD5CMI2	2002	227	11.3	AOAOD5CMI2_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AJW80512.1};
AOA0E3DRZO	800	147	11.3	AOA0E3DRZO_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0E8XLQ5	1197	147	11.3	AOA0E8XLQ5_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0F4XDJ4	1350	153	11.3	AOA0F4XDJ4_	SubName: Full=Killer toxin-resistance protein 5 {ECO:0000313...
AOA0G3A3U8	1936	218	11.3	AOA0G3A3U8_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AKJ06937.1};
AOA0H2YBK8	1197	147	11.3	AOA0H2YBK8_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:ABG15...
AOA0H2YMY3	1197	147	11.3	AOA0H2YMY3_	SubName: Full=Toxin subunit {ECO:0000313 EMBL:ABG19819.1};
AOA0H3LPZ7	1464	165	11.3	AOA0H3LPZ7_	SubName: Full=Dermonecrotic toxin {ECO:0000313 EMBL:CAE34341...
AOA0H3NK74	1175	148	11.3	AOA0H3NK74_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:CBY25...
AOA0H3NRB5	967	147	11.3	AOA0H3NRB5_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0H5FQ03	1175	148	11.3	AOA0H5FQ03_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:CRX73...
AOA0H5GWG8	969	148	11.3	AOA0H5GWG8_	SubName: Full=Putative insecticidal toxin complex protein {E...
AOA0H5IV66	1175	148	11.3	AOA0H5IV66_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:CRY16...
AOA0H6HVM6	1056	147	11.3	AOA0H6HVM6_	SubName: Full=RTX toxin RtxA {ECO:0000313 EMBL:CSB23061.1};
AOA0H6WM18	1161	147	11.3	AOA0H6WM18_	SubName: Full=RTX toxin RtxA {ECO:0000313 EMBL:CSC91335.1};
AOA0I9XJH8	941	147	11.3	AOA0I9XJH8_	SubName: Full=Protoplast regeneration and killer toxin resis...
AOA0J9HQR5	935	147	11.3	AOA0J9HQR5_	SubName: Full=Toxin {ECO:0000313 EMBL:KMW71534.1};
AOA0KOYYE5	1197	147	11.3	AOA0KOYYE5_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0KOZBC3	1197	147	11.3	AOA0KOZBC3_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0K1ALU4	1197	147	11.3	AOA0K1ALU4_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0K1AW77	1197	147	11.3	AOA0K1AW77_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0K1B7V4	1197	147	11.3	AOA0K1B7V4_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0K1BL92	1197	147	11.3	AOA0K1BL92_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0K1C2H4	1197	147	11.3	AOA0K1C2H4_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0K1C952	1197	147	11.3	AOA0K1C952_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0K1CM45	1197	147	11.3	AOA0K1CM45_	SubName: Full=Putative insecticidal toxin complex protein Tc...
AOA0K8L4X2	1053	148	11.3	AOA0K8L4X2_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0LONAP9	1314	149	11.3	AOA0LONAP9_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0LOQXS2	1291	148	11.3	AOA0LOQXS2_	SubName: Full=Toxin {ECO:0000313 EMBL:KNE23374.1};
AOA0M1PR92	1005	148	11.3	AOA0M1PR92_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KOR96893.1};
AOA0N9TWQ0	1464	166	11.3	AOA0N9TWQ0_	SubName: Full=Dermonecrotic toxin {ECO:0000313 EMBL:ALH65512...
AOA0P9IDK1	956	147	11.3	AOA0P9IDK1_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0P9JPX3	1581	178	11.3	AOA0P9JPX3_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0P9U3K2	885	148	11.3	AOA0P9U3K2_	SubName: Full=ADP-ribosylating toxin family protein {ECO:000...
AOA0P9UAH7	1581	178	11.3	AOA0P9UAH7_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0P9X4T6	885	148	11.3	AOA0P9X4T6_	SubName: Full=ADP-ribosylating toxin family protein {ECO:000...
AOA0P9Y8S1	886	147	11.3	AOA0P9Y8S1_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0Q0AMB1	1444	163	11.3	AOA0Q0AMB1_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0Q3MC31	1291	147	11.3	AOA0Q3MC31_	SubName: Full=Toxin {ECO:0000313 EMBL:KQK52891.1};
AOA0Q6DCE0	1042	147	11.3	AOA0Q6DCE0_	SubName: Full=Toxin {ECO:0000313 EMBL:KQT63094.1};
AOA0Q6R2N0	889	147	11.3	AOA0Q6R2N0_	SubName: Full=Toxin {ECO:0000313 EMBL:KQV19401.1};
AOA0R2Y879	1506	170	11.3	AOA0R2Y879_	SubName: Full=Toxin {ECO:0000313 EMBL:KRP44729.1};
A5D094	1078	148	11.3	A5D094_PELTS	SubName: Full=RTX toxins and related Ca2+-binding proteins {...
A6CZNO	737	148	11.3	A6CZNO_9VIBR	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
A7ISL2	927	147	11.3	A7ISL2_9GAMM	SubName: Full=RTX A toxin {ECO:0000313 EMBL:ABA39419.1};
B9WDQ8	1453	164	11.3	B9WDQ8_CANDC	SubName: Full=Killer toxin-resistance protein, putative {ECO...
D7IRS5	991	148	11.3	D7IRS5_9BACE	SubName: Full=Toxin-antitoxin system toxin component {ECO:00...
E2XVN7	1325	150	11.3	E2XVN7_PSEFL	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:E...
E5AU93	1202	147	11.3	E5AU93_BURRH	SubName: Full=Insecticidal toxin complex protein TccB {ECO:0...
E9EJN3	1215	147	11.3	E9EJN3_METRA	SubName: Full=Killer toxin alpha/beta {ECO:0000313 EMBL:EFZ0...
G0YF56	966	148	11.3	G0YF56_PHOTE	SubName: Full=Insecticidal toxin complex protein TccA {ECO:0...
G2XA97	1305	147	11.3	G2XA97_VERDV	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
G8Q750	844	147	11.3	G8Q750_PSEFL	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
IOEKJ4	1353	153	11.3	IOEKJ4_HELCO	SubName: Full=Toxin-like outer membrane protein {ECO:0000313...
I3BP01	1117	147	11.3	I3BP01_9GAMM	SubName: Full=Insecticidal toxin complex/plasmid violence pr...
I4L351	931	148	11.3	I4L351_9PSED	SubName: Full=Insecticidal toxin complex protein TcaA2 {ECO:...
J2N3Z0	1847	209	11.3	J2N3Z0_9PSED	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...

J2Y652	929	147	11.3	J2Y652_PSEFL	SubName: Full=Insecticidal toxin complex protein TccC1 {ECO:...
J2YW56	1847	209	11.3	J2YW56_9PSED	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
K4ZS67	1121	147	11.3	K4ZS67_PAEAL	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
K9ETK2	896	147	11.3	K9ETK2_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
N4TIH4	1713	193	11.3	N4TIH4_FUSC1	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
Q53550	1196	147	11.3	Q53550_CLOB0	SubName: Full=Progenitor toxin L nontoxic-nonhemagglutinin c...
Q7VTS2	1464	166	11.3	Q7VTS2_BORPE	SubName: Full=Dermonecrotic toxin {ECO:0000313 EMBL:CAE43702...
Q8D1P6	1516	171	11.3	Q8D1P6_YERPE	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:AAM83...
Q8D1P7	1197	147	11.3	Q8D1P7_YERPE	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:AAM83...
S0E8X1	941	147	11.3	S0E8X1_GIBF5	SubName: Full=Related to protoplast regeneration and killer ...
S6HQJ6	1766	199	11.3	S6HQJ6_9PSED	SubName: Full=Putative insecticidal toxin protein {ECO:00003...
T5CZ87	1298	148	11.3	T5CZ87_HELPH	SubName: Full=Toxin {ECO:0000313 EMBL:EQ63044.1};
W2TLB6	880	147	11.3	W2TLB6_NECAM	SubName: Full=Zonula occludens toxin {ECO:0000313 EMBL:ETN82...
W6APX8	1410	160	11.3	W6APX8_9GAMM	SubName: Full=Putative insecticidal toxin component B {ECO:0...
W6IOTO	1621	183	11.3	W6IOTO_9PROT	SubName: Full=Insecticidal toxin complex protein TccB {ECO:0...
AOA021XE05	823	146	11.2	AOA021XE05_	SubName: Full=Zeta toxin {ECO:0000313 EMBL:EYR82822.1};
AOA095TTK1	763	146	11.2	AOA095TTK1_	SubName: Full=ABC transporter family protein {ECO:0000313 EM...
AOA0A1HU99	919	146	11.2	AOA0A1HU99_	SubName: Full=Insecticidal toxin protein, putative {ECO:0000...
AOA0A1I4X4	1495	168	11.2	AOA0A1I4X4_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:CDF97...
AOA0A2VJ25	1836	206	11.2	AOA0A2VJ25_	SubName: Full=HC-toxin synthetase {ECO:0000313 EMBL:KGQ07588...
AOA0A8NX28	1030	146	11.2	AOA0A8NX28_	SubName: Full=C component of insecticidal toxin complex (Tc)...
AOA0A8PON5	1391	156	11.2	AOA0A8PON5_	SubName: Full=A component of insecticidal toxin complex (Tc)...
AOA0C2A1C0	943	146	11.2	AOA0C2A1C0_	SubName: Full=Toxin {ECO:0000313 EMBL:KIF63155.1};
AOA0D9AUU6	1352	152	11.2	AOA0D9AUU6_	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
AOA0E1NR85	1496	168	11.2	AOA0E1NR85_	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:A...
AOA0E3MS27	966	146	11.2	AOA0E3MS27_	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
AOA0F7Y4U4	2367	264	11.2	AOA0F7Y4U4_	SubName: Full=RTX toxin {ECO:0000313 EMBL:CRI55927.1};
AOA0H2YLI4	1496	168	11.2	AOA0H2YLI4_	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:A...
AOA0H5E8H4	956	146	11.2	AOA0H5E8H4_	SubName: Full=Putative insecticidal toxin complex protein {E...
AOA0H6VZ57	1057	146	11.2	AOA0H6VZ57_	SubName: Full=RTX toxin RtxA {ECO:0000313 EMBL:CSC76969.1};
AOA0J6GF15	1762	198	11.2	AOA0J6GF15_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0J6HF11	1493	167	11.2	AOA0J6HF11_	SubName: Full=Toxin {ECO:0000313 EMBL:KMM92325.1};
AOA0J9F8F7	1178	146	11.2	AOA0J9F8F7_	SubName: Full=Toxin {ECO:0000313 EMBL:KMW74532.1};
AOA0M1V8W7	1496	168	11.2	AOA0M1V8W7_	SubName: Full=Putative insecticidal toxin complex protein {E...
AOA0N2SK27	1496	168	11.2	AOA0N2SK27_	SubName: Full=Toxin {ECO:0000313 EMBL:KJG86603.1};
AOA0P9H213	939	146	11.2	AOA0P9H213_	SubName: Full=Putative insecticidal toxin complex protein C2...
AOA0P9I177	1026	146	11.2	AOA0P9I177_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0P9LLF1	922	146	11.2	AOA0P9LLF1_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0P9U9Z2	1026	146	11.2	AOA0P9U9Z2_	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
AOA0P9VVA4	922	146	11.2	AOA0P9VVA4_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0Q0C096	937	146	11.2	AOA0Q0C096_	SubName: Full=Putative insecticidal toxin protein {ECO:00003...
AOA0Q0DLS3	1449	163	11.2	AOA0Q0DLS3_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0Q0FP17	939	146	11.2	AOA0Q0FP17_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0R3L4G0	776	146	11.2	AOA0R3L4G0_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KRR02772.1};
AOA0R4CPQ2	1391	156	11.2	AOA0R4CPQ2_	SubName: Full=A component of insecticidal toxin complex (Tc)...
A6E153	844	146	11.2	A6E153_9RHOB	SubName: Full=Rhizobiocin/RTX toxin {ECO:0000313 EMBL:EDM315...
A6FF64	1515	169	11.2	A6FF64_9GAMM	SubName: Full=Insecticidal toxin complex protein TcaC {ECO:0...
B1N6H9	884	146	11.2	B1N6H9_9BACT	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:ABM53525...
B6SD27	1683	189	11.2	B6SD27_9VIRU	SubName: Full=Putative YD-repeat toxin {ECO:0000313 EMBL:ACJ...
C3IAE2	1458	163	11.2	C3IAE2_BACTU	SubName: Full=Insecticidal toxin complex protein TcaC (Toxin...
C7BNH8	1468	164	11.2	C7BNH8_PHOAA	SubName: Full=Insecticidal toxin complex protein tcdB2 {ECO:...
D3VHH4	1391	156	11.2	D3VHH4_XENNA	SubName: Full=A component of insecticidal toxin complex (Tc)...
I4L321	966	146	11.2	I4L321_9PSED	SubName: Full=Insecticidal toxin complex protein TccC1 {ECO:...
I4L5Q0	1477	166	11.2	I4L5Q0_9PSED	SubName: Full=Insecticidal toxin complex protein TcaC1 {ECO:...
J3E9M8	1909	214	11.2	J3E9M8_9PSED	SubName: Full=C-terminal region of Pasteurella multocida tox...
K4ZTB4	936	146	11.2	K4ZTB4_PAEAL	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
K5AF64	1195	146	11.2	K5AF64_PAEAL	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
N1NQQ5	1391	156	11.2	N1NQQ5_XENNE	SubName: Full=A component of insecticidal toxin complex (Tc)...

N1NTR9	1030	146	11.2	N1NTR9_XENNE	SubName: Full=C component of insecticidal toxin complex (Tc)...
Q1IA36	990	146	11.2	Q1IA36_PSEE4	SubName: Full=Insecticidal toxin, SepC/Tcc class {ECO:000031...
Q63UE1	766	146	11.2	Q63UE1_BURPS	SubName: Full=Putative toxin transport-related membrane prot...
Q66A38	982	146	11.2	Q66A38_YERPS	SubName: Full=Insecticidal toxin {ECO:0000313 EMBL:CAH21532.1...
Q74PN2	1496	168	11.2	Q74PN2_YERPE	SubName: Full=Insecticidal toxin complex {ECO:0000313 EMBL:C...
Q9H2K2	1166	146	11.2	TNKS2_HUMAN	RecName: Full=Tankyrase-2; Short=TANK2; EC=2.4.2.30; AltName...
Q9S5D5	1464	164	11.2	Q9S5D5_BORBO	SubName: Full=Dermonecrotizing toxin {ECO:0000313 EMBL:BAA34...
TOEWM5	1269	146	11.2	TOEWM5_HELPH	SubName: Full=Toxin {ECO:0000313 EMBL:EPZ68468.1};
TONVRO	1361	152	11.2	TONVRO_PHOTE	SubName: Full=Insecticidal toxin complex protein TccB2 {ECO:...
TOP8Q9	1476	166	11.2	TOP8Q9_PHOTE	SubName: Full=Insecticidal toxin complex protein TccB1 {ECO:...
T1RTB7	915	146	11.2	T1RTB7_ECOLX	SubName: Full=Pertussis toxin liberation protein C {ECO:0000...
U7YQY9	1361	152	11.2	U7YQY9_PHOTE	SubName: Full=Toxin {ECO:0000313 EMBL:ERT12200.1};
W1J1L0	1400	157	11.2	W1J1L0_9ENTR	SubName: Full=A component of insecticidal toxin complex (Tc)...
W2ECV5	1008	146	11.2	W2ECV5_9BACL	SubName: Full=Toxin-like protein {ECO:0000313 EMBL:ETK29444...
W6UWR4	1017	146	11.2	W6UWR4_9PSED	SubName: Full=Dermonecrotic/RTX toxin {ECO:0000313 EMBL:EUB7...
AOA059L273	1210	145	11.1	AOA059L273_	SubName: Full=Insecticidal toxin complex protein TcaA2 {ECO:...
AOA059L2J0	931	145	11.1	AOA059L2J0_	SubName: Full=Toxin {ECO:0000313 EMBL:KDD68325.1};
AOA061ZS50	1841	205	11.1	AOA061ZS50_	SubName: Full=Pre-toxin domain with VENN motif family protei...
AOA072CAH1	2048	227	11.1	AOA072CAH1_	SubName: Full=Rhizobiocin/RTX toxin and hemolysin-type calci...
AOA093VB53	1340	149	11.1	AOA093VB53_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA099SLB7	934	145	11.1	AOA099SLB7_	SubName: Full=Insecticidal toxin complex protein C1 {ECO:000...
AOA099SPE2	1444	161	11.1	AOA099SPE2_	SubName: Full=Toxin {ECO:0000313 EMBL:KKG93698.1};
AOA0B1Z178	1352	150	11.1	AOA0B1Z178_	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
AOA0B2EGY5	1312	145	11.1	AOA0B2EGY5_	SubName: Full=Toxin {ECO:0000313 EMBL:KHL85924.1};
AOA0B8NYZ2	883	145	11.1	AOA0B8NYZ2_	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:GAM59186...
AOA0B8PYF3	1373	152	11.1	AOA0B8PYF3_	SubName: Full=RTX toxins {ECO:0000313 EMBL:GAM68118.1};
AOA0C2A3P1	902	145	11.1	AOA0C2A3P1_	SubName: Full=Toxin {ECO:0000313 EMBL:KIF63855.1};
AOA0C2MXA5	1573	175	11.1	AOA0C2MXA5_	SubName: Full=Toxin {ECO:0000313 EMBL:KIK85430.1};
AOA0D5A362	986	145	11.1	AOA0D5A362_	SubName: Full=Toxin secretion ABC transporter ATP-binding pr...
AOA0D5ZOM1	975	145	11.1	AOA0D5ZOM1_	SubName: Full=Toxin {ECO:0000313 EMBL:AKA38047.1};
AOA0D9AUZ8	1483	165	11.1	AOA0D9AUZ8_	SubName: Full=Toxin {ECO:0000313 EMBL:KJH84529.1};
AOA0E1NJJ3	1374	152	11.1	AOA0E1NJJ3_	SubName: Full=Putative insecticidal toxin complex protein {E...
AOA0G2GGD1	1546	171	11.1	AOA0G2GGD1_	SubName: Full=Putative hc-toxin synthetase {ECO:0000313 EMBL...
AOA0H3AZ46	1197	145	11.1	AOA0H3AZ46_	SubName: Full=Toxin subunit {ECO:0000313 EMBL:ACA66782.1};
AOA0J6K2A2	1474	164	11.1	AOA0J6K2A2_	SubName: Full=Toxin {ECO:0000313 EMBL:KMM90132.1};
AOA0K6HFJ1	773	145	11.1	AOA0K6HFJ1_	SubName: Full=Ca2+-binding protein, RTX toxin-related {ECO:0...
AOA0LONMR6	870	145	11.1	AOA0LONMR6_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0L6DH18	1291	145	11.1	AOA0L6DH18_	SubName: Full=Toxin {ECO:0000313 EMBL:KNX49189.1};
AOA0M2MIF8	922	145	11.1	AOA0M2MIF8_	SubName: Full=Toxin {ECO:0000313 EMBL:KKI28033.1};
AOA0M8MZ11	1309	145	11.1	AOA0M8MZ11_	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
AOA0NOWHEO	1444	160	11.1	AOA0NOWHEO_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0N1JCT2	922	145	11.1	AOA0N1JCT2_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0N8R7H2	1446	160	11.1	AOA0N8R7H2_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9KOA1	1444	161	11.1	AOA0P9KOA1_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9X548	934	145	11.1	AOA0P9X548_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9XMC2	1444	161	11.1	AOA0P9XMC2_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0Q0ARN6	939	145	11.1	AOA0Q0ARN6_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0Q0BHD3	922	145	11.1	AOA0Q0BHD3_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0Q6YEM5	1475	163	11.1	AOA0Q6YEM5_	SubName: Full=Toxin {ECO:0000313 EMBL:KQW09095.1};
AOA0Q8BVP4	996	145	11.1	AOA0Q8BVP4_	SubName: Full=Toxin {ECO:0000313 EMBL:KRA04593.1};
AOA0R3A5T3	1606	178	11.1	AOA0R3A5T3_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0R4FMG4	1030	145	11.1	AOA0R4FMG4_	SubName: Full=C component of insecticidal toxin complex (Tc)...
A7ISK7	927	145	11.1	A7ISK7_9GAMM	SubName: Full=RTX structural toxin {ECO:0000313 EMBL:ABA4264...
B2ACW9	1872	207	11.1	B2ACW9_PODAN	SubName: Full=Podospora anserina S mat+ genomic DNA chromoso...
B2BCY2	1197	145	11.1	B2BCY2_YERPU	SubName: Full=Toxin complex subunit TcaB {ECO:0000313 EMBL:A...
C3FB27	965	145	11.1	C3FB27_BACTU	SubName: Full=Iota toxin component Ib {ECO:0000313 EMBL:EEM5...
D2TBD5	2090	232	11.1	D2TBD5_ERWP6	SubName: Full=Toxin A {ECO:0000313 EMBL:CAY72286.1};
D3VFD3	1030	145	11.1	D3VFD3_XENNA	SubName: Full=C component of insecticidal toxin complex (Tc)...

D3VHI1	1475	164	11.1	D3VHI1_XENNA	SubName: Full=B component of insecticidal toxin complex (Tc)...
D4HZV6	1051	145	11.1	D4HZV6_ERWAC	SubName: Full=Dermonecrotic toxin (DNT) (PMT) (Mitogenic tox...
F3J4C8	1444	161	11.1	F3J4C8_PSEAP	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:E...
J2Y4U8	1495	166	11.1	J2Y4U8_PSEFL	SubName: Full=Insecticidal toxin complex protein TcaC1 {ECO:...
J3A395	1572	175	11.1	J3A395_9PSED	SubName: Full=C-terminal region of Pasteurella multocida tox...
J3FI30	711	145	11.1	J3FI30_9PSED	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
M3GTD3	1502	167	11.1	M3GTD3_9LIST	SubName: Full=Diarrheal toxin/FtsK/SpoIIIE family protein {E...
O25331	1943	216	11.1	O25331_HELPHY	SubName: Full=Toxin-like outer membrane protein {ECO:0000313...
O85156	1565	174	11.1	O85156_PHOLU	SubName: Full=Insecticidal toxin complex protein TccB {ECO:0...
Q1I6V0	2350	261	11.1	Q1I6V0_PSEE4	SubName: Full=Putative RTX toxin {ECO:0000313 EMBL:CAK16632...
Q1IFC6	1520	168	11.1	Q1IFC6_PSEE4	SubName: Full=Putative insecticidal toxin complex protein {E...
Q2KUX5	1464	162	11.1	Q2KUX5_BORA1	SubName: Full=Dermonecrotic toxin {ECO:0000313 EMBL:CAJ50595...
Q2W246	2065	229	11.1	Q2W246_MAGSA	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
Q2WCV1	1424	158	11.1	Q2WCV1_YEREN	SubName: Full=Putative insecticidal toxin complex protein {E...
Q665G1	952	145	11.1	Q665G1_YERPS	SubName: Full=Putative insecticidal toxin {ECO:0000313 EMBL:...
Q7W4W3	1464	163	11.1	Q7W4W3_BORPA	SubName: Full=Dermonecrotic toxin {ECO:0000313 EMBL:CAE38827...
Q887R0	922	145	11.1	Q887R0_PSESM	SubName: Full=Insecticidal toxin protein, putative {ECO:0000...
Q93GI2	927	145	11.1	Q93GI2_MORBO	SubName: Full=Cytotoxin {ECO:0000313 EMBL:ABR28451.1}; SubNa...
S7T085	1102	145	11.1	S7T085_DESML	SubName: Full=Putative insecticidal toxin protein {ECO:00003...
U7R071	916	145	11.1	U7R071_PHOTE	SubName: Full=Toxin {ECO:0000313 EMBL:ERT13744.1};
V8R571	1215	145	11.1	V8R571_9PSED	SubName: Full=Insecticidal toxin complex protein TcaA2 {ECO:...
W2E4N5	959	145	11.1	W2E4N5_9BACL	SubName: Full=Mosquitocidal toxin Mtx {ECO:0000313 EMBL:ETK2...
AOA031G4M3	898	143	11.0	AOA031G4M3_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:E...
AOA031G8I9	904	143	11.0	AOA031G8I9_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:E...
AOA061PJQ6	2092	230	11.0	AOA061PJQ6_	SubName: Full=RTX toxin {ECO:0000313 EMBL:GAK16509.1};
AOA077MZE2	747	143	11.0	AOA077MZE2_	SubName: Full=Insecticidal toxin complex protein TccC {ECO:0...
AOA077QHL3	1524	167	11.0	AOA077QHL3_	SubName: Full=Insecticidal toxin complex protein TccB (Modul...
AOA077R4R0	1098	143	11.0	AOA077R4R0_	SubName: Full=Related to SKT5-protoplast regeneration and ki...
AOA081FU95	786	143	11.0	AOA081FU95_	SubName: Full=RTX toxins and related Ca2+-binding protein {E...
AOA084BEP9	1239	144	11.0	AOA084BEP9_	SubName: Full=Killer toxin sensitivity protein IKI3 {ECO:000...
AOA095RWM2	805	143	11.0	AOA095RWM2_	SubName: Full=Cytolethal distending toxin A/C family protein...
AOA0A0CV22	960	143	11.0	AOA0A0CV22_	SubName: Full=Toxin {ECO:0000313 EMBL:KGM28687.1};
AOA0A0CXT4	1183	143	11.0	AOA0A0CXT4_	SubName: Full=Toxin {ECO:0000313 EMBL:KGM29662.1};
AOA0A3KG85	806	143	11.0	AOA0A3KG85_	SubName: Full=Cytolethal distending toxin A/C family protein...
AOA0A6FJF5	1540	170	11.0	AOA0A6FJF5_	SubName: Full=Toxin {ECO:0000313 EMBL:KHA72856.1};
AOA0B1Z186	1483	163	11.0	AOA0B1Z186_	SubName: Full=Toxin {ECO:0000313 EMBL:KHK64390.1};
AOA0B2DX43	1295	143	11.0	AOA0B2DX43_	SubName: Full=Toxin {ECO:0000313 EMBL:KHL77447.1};
AOA0B8NY52	733	144	11.0	AOA0B8NY52_	SubName: Full=RTX toxin, putative {ECO:0000313 EMBL:GAM59450...
AOA0C1WTN2	896	143	11.0	AOA0C1WTN2_	SubName: Full=Toxin {ECO:0000313 EMBL:KIF59622.1};
AOA0D0ILMO	1044	143	11.0	AOA0D0ILMO_	SubName: Full=Toxin {ECO:0000313 EMBL:KIP93907.1};
AOA0D6AWN8	1013	143	11.0	AOA0D6AWN8_	SubName: Full=Toxin secretion ABC transporter ATP-binding pr...
AOA0D9ASX4	941	144	11.0	AOA0D9ASX4_	SubName: Full=Toxin {ECO:0000313 EMBL:KJH83842.1};
AOA0E1E6J8	2124	233	11.0	AOA0E1E6J8_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AIC18716.1};
AOA0E1QH35	902	144	11.0	AOA0E1QH35_	SubName: Full=Putative Clostridial binary toxin B superfamil...
AOA0E3HOB4	1508	166	11.0	AOA0E3HOB4_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:AKA85...
AOA0F4XSB5	927	143	11.0	AOA0F4XSB5_	SubName: Full=Toxin {ECO:0000313 EMBL:KKA08298.1};
AOA0F6GGB2	800	143	11.0	AOA0F6GGB2_	SubName: Full=Cytolethal distending toxin A/C family protein...
AOA0F6LGC3	800	143	11.0	AOA0F6LGC3_	SubName: Full=Cytolethal distending toxin A/C family protein...
AOA0F7BNH8	1427	157	11.0	AOA0F7BNH8_	SubName: Full=Toxin {ECO:0000313 EMBL:AKF82362.1};
AOA0G3C2G3	2178	239	11.0	AOA0G3C2G3_	SubName: Full=RTX toxin {ECO:0000313 EMBL:AKJ33327.1};
AOA0G3GGY1	1523	168	11.0	AOA0G3GGY1_	SubName: Full=Toxin {ECO:0000313 EMBL:AKK00379.1};
AOA0G3GHU5	1443	159	11.0	AOA0G3GHU5_	SubName: Full=Toxin {ECO:0000313 EMBL:AKK00811.1};
AOA0H3NQQ2	1165	143	11.0	AOA0H3NQQ2_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:CBY25...
AOA0H5GYD5	956	143	11.0	AOA0H5GYD5_	SubName: Full=Putative insecticidal toxin complex protein {E...
AOA0H5IV57	1165	143	11.0	AOA0H5IV57_	SubName: Full=Putative toxin subunit {ECO:0000313 EMBL:CRY16...
AOA0H6S6D0	768	143	11.0	AOA0H6S6D0_	SubName: Full=RTX toxins and Ca2+-binding protein {ECO:00003...
AOA0H6WJE3	986	143	11.0	AOA0H6WJE3_	SubName: Full=RTX toxin RtxA {ECO:0000313 EMBL:CSC91723.1};
AOA0J6IUN5	1454	160	11.0	AOA0J6IUN5_	SubName: Full=Toxin {ECO:0000313 EMBL:KMN16028.1};

AOA0J6K994	2128	235	11.0	AOA0J6K994_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KMM92472.1};
AOA0K2X3W3	1290	144	11.0	AOA0K2X3W3_	SubName: Full=Toxin-like outer membrane protein {ECO:0000313 ...
AOA0K2Y4E9	1779	195	11.0	AOA0K2Y4E9_	SubName: Full=Monoglycosyltransferase toxin {ECO:0000313 EMB...
AOA0K8QML0	754	143	11.0	AOA0K8QML0_	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
AOA0L1POB8	2128	234	11.0	AOA0L1POB8_	SubName: Full=RTX toxin {ECO:0000313 EMBL:KNH46483.1};
AOA0NOFZX2	812	143	11.0	AOA0NOFZX2_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0N8QP54	1449	160	11.0	AOA0N8QP54_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0N9M7Y6	970	143	11.0	AOA0N9M7Y6_	SubName: Full=Toxin {ECO:0000313 EMBL:ALG77265.1};
AOA0N9W678	932	143	11.0	AOA0N9W678_	SubName: Full=Toxin {ECO:0000313 EMBL:ALI06702.1};
AOA0P9ISC2	948	144	11.0	AOA0P9ISC2_	SubName: Full=Putative insecticidal toxin protein {ECO:00003...
AOA0P9J5M2	906	143	11.0	AOA0P9J5M2_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0P9JBZ8	1449	160	11.0	AOA0P9JBZ8_	SubName: Full=Putative insecticidal toxin complex protein B ...
AOA0P9K309	939	144	11.0	AOA0P9K309_	SubName: Full=Putative Insecticidal toxin protein {ECO:00003...
AOA0P9UFM5	948	144	11.0	AOA0P9UFM5_	SubName: Full=Putative insecticidal toxin protein {ECO:00003...
AOA0P9UTF6	826	144	11.0	AOA0P9UTF6_	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:K...
AOA0Q8C526	1531	168	11.0	AOA0Q8C526_	SubName: Full=Toxin {ECO:0000313 EMBL:KRA04594.1};
AOA0Q8FYB2	823	143	11.0	AOA0Q8FYB2_	SubName: Full=Zeta toxin {ECO:0000313 EMBL:KRA64124.1};
AOA0R3ACU7	1477	162	11.0	AOA0R3ACU7_	SubName: Full=Toxin {ECO:0000313 EMBL:KRP70810.1};
AOA0R3BET1	1476	162	11.0	AOA0R3BET1_	SubName: Full=Toxin {ECO:0000313 EMBL:KRP80931.1};
AOYTD3	801	143	11.0	AOYTD3_LYNBP	SubName: Full=Hemolysin-type calcium-binding toxin {ECO:0000...
C3IAE4	1088	143	11.0	C3IAE4_BACTU	SubName: Full=Insecticidal toxin complex protein TcaA (Toxin...
C4SEH6	939	144	11.0	C4SEH6_YERMO	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
D2U4E6	1581	174	11.0	D2U4E6_9ENTR	SubName: Full=Insecticidal toxin complex protein {ECO:000031...
D3UWD9	1184	144	11.0	D3UWD9_XENBS	SubName: Full=A component of insecticidal toxin complex (Tc)...
D9PD25	1668	183	11.0	D9PD25_ACTPL	SubName: Full=RTX toxin protein {ECO:0000313 EMBL:EFL80249.1...
E4UN84	1287	143	11.0	E4UN84_ARTGP	SubName: Full=Killer toxin alpha/beta {ECO:0000313 EMBL:EFQ9...
E5AU92	1101	144	11.0	E5AU92_BURRH	SubName: Full=Insecticidal toxin complex protein TcaA {ECO:0...
E5B8D4	1049	143	11.0	E5B8D4_ERWAM	SubName: Full=Dermonecrotic toxin (DNT) (PMT) (Mitogenic tox...
F3UU36	1471	162	11.0	F3UU36_STRSA	SubName: Full=Diarrheal toxin {ECO:0000313 EMBL:EGJ41852.1};...
G2WSX8	1330	146	11.0	G2WSX8_VERDV	SubName: Full=Killer toxin subunits alpha/beta {ECO:0000313 ...
I3VZ41	1295	143	11.0	I3VZ41_ECOLD	SubName: Full=Secreted autotransporter toxin {ECO:0000313 EM...
I4L436	1611	178	11.0	I4L436_9PSED	SubName: Full=Insecticidal toxin complex protein TcaB2 {ECO:...
I4YXH0	895	144	11.0	I4YXH0_9RHIZ	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
J2V723	851	144	11.0	J2V723_9RHIZ	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
K2T2J2	1446	159	11.0	K2T2J2_PSESY	SubName: Full=Insecticidal toxin protein {ECO:0000313 EMBL:E...
K2TB53	953	143	11.0	K2TB53_PSESY	SubName: Full=Putative insecticidal toxin complex {ECO:00003...
K9RJQ1	2084	229	11.0	K9RJQ1_9CYAN	SubName: Full=Ca2+-binding protein, RTX toxin {ECO:0000313 E...
L7H6J5	1620	178	11.0	L7H6J5_PSEFL	SubName: Full=Insecticidal toxin complex protein TcaB1 {ECO:...
M3TLH9	887	144	11.0	M3TLH9_ENTHI	SubName: Full=Zonula occludens toxin, putative {ECO:0000313 ...

## C Scripts for allergen analysis

### Script for making the search for identity over a window

```
#!/bin/csh
#
# USAGE: windowmatch <query sequence> <library> <windowlength> <cutoff> <raw fasta output>
# e.g. windowmatch BG025.fasta allergenonline.fasta 80 35.0 ../rawfasta.txt
#
awk -v window=$3 -f ./makewindows.awk $1
find . -name "window$3_*.fasta" -exec /z/linux/fasta/fasta34 -Q -b 100000 -d 100000 -w 100 {} \
    $2 2 \; | tee $5 | grep -A 2 ">>" \
    | awk -v window=$3 -v threshold=$4 '/^>>/ {name= substr($1,3,200); getline; getline; percent=gensub("%","", "g", $4); \
    overlap= int($9); if (1.0*percent >= 1.0*threshold && 1*overlap >= 1*window) \
    {printf "%s\t%3.1f%% identity i %2d aa overlap.\n", \
    name, percent, overlap}}' | sort -r -n --key=10
```

The script is invoked by the following command, where parameter 2 is the length of the window, and parameter 3 is the identity threshold:

```
windowmatch lactase.fasta allergenonline.fasta 80 35.0 >
allergenonline_window80_result.txt
```

and

```
windowmatch lactase.fasta allergen.org.fasta 80 35.0 >
allergenorg_window80_result.txt
```

### Script for making the search for scaled identity over a window

```
#!/bin/csh
#
# USAGE: windowmatch_scale <query sequence> <Library> <windowlength> <cutoff> <raw fasta output>
# e.g. windowmatch BG025.fasta allergenonline.fasta 80 35.0 myfastaoutput.txt
#
awk -v window=$3 -f ./makewindows.awk $1
find . -name "window80_*.fasta" -exec /z/linux/fasta/fasta34 -Q -b 100000 -d 100000 -w 100 {} \
    $2 2 \; | tee $5 | grep -A 2 ">>" \
    | awk -v window=$3 -v threshold=$4 '/^>>/ {name= substr($1,3,200); getline; getline; percent=$4; \
    overlap= int($9); newpercent=(1.0*percent*overlap)/(1.0*window); if (newpercent >= 1.0*threshold && overlap < window) \
    {printf "%s\t%3.1f%% identity i %2d aa overlap, scaled to %3.1f%% identity i %d aa overlap\n", \
    name, percent, overlap, newpercent, window }}' | sort -r -n --key=10
```

The script is invoked by the following command, where parameter 2 is the length of the window,



and parameter 3 is the identity threshold. This script allows identification of matches with higher identity over shorter windows than 80 amino acids. For example a match with 50% identity over 60 amino acids would still have enough identical amino acids to exceed the 35% threshold over 80 amino acids:  $60 \cdot 0.50 / 80 = 0.375 = 37.5\%$ .

```
windowmatch_scale lactase.fasta allergenonline.fasta 80 35.0 >
allergenonline.window80_result_scale.txt
```

and

```
windowmatch_scale lactase.fasta allergen.org.fasta 80 35.0 >
allergenorg.window80_result_scale.txt
```

## Common awk script used by the two previous scripts

The file is named `makewindows.awk`

```
BEGIN { seq=""
        if (window < 1)
            window = 6
    }
    {
        if (substr($0,1,1) != ">")
        {
            gsub("[^A-Za-z]", "")
            seq = sprintf("%s%s", seq, $0)
        }
    }
    END {
        for (i=1; i<length(seq)-window+2; i++)
        {
            filename = sprintf ("window%d_%04d.fasta", window, i)
            printf ">window%d_%04d\n", window, i > filename
            printf "%s\n", substr(seq,i>window) > filename
        }
    }
}
```

## Script for making the Needleman-Wunsch alignment and comparison

```
#!/bin/csh
#
# USAGE: fullmatch <query sequence> <library> <cutoff> <raw needle output>
# e.g. fullmatch BG025.fasta 35.0 ../rawneedle.txt
#
```

```

needle -asequence $1 -bsequence $2 \
-gapopen 10.0 -gapextend 0.5 -outfile /dev/stdout \
| tee $4 | awk -v threshold=$3 '2:/{name = substr($3,1,80) } \
/Identity/{ matches = $3; percent = strtonum(gensub("\\(", "", 1, $NF)); \
if (percent >= threshold) {printf "%-80s %-10s = %5.1f%%\n", \
name, matches, percent } } ' | sort -r -n --key=4

```

The script is invoked by the following command, where parameter 1 is the identity threshold:

```

fullmatch lactase.fasta allergenonline.fasta 10.0 >
allergenonline_fullresult.txt

```

and

```

fullmatch lactase.fasta allergen.org.fasta 10.0 >
allergenorg_fullresult.txt

```

## D List of allergens from allergenonline

List of allergens that have been tested by the EFSA scientific opinion recommended allergen analysis described in section 2. The sequences were downloaded via <http://allergenonline.org>.

Species	Common	IUIS Allergen	Type	Group*	Length	GI#9	First Version
Acarus siro	Mite	Aca s 13	Aero Mite	Acarus Aca s 13	131	118638268	9
Acarus siro	Mite	Unassigned	Aero Insect	Acarus siro Group 4 allergen	1517	118638278	9
Actinidia chinensis	Kivi	Unassigned	Food Plant	Actinidia Act c 1 Act d1380	190358935	1190358935	9
Actinidia deliciosa	Kivi	Act d 1	Food Plant	Actinidia Act c 1 Act d1380	15984	115984	7
Actinidia deliciosa	Kivi	Unassigned	Food Plant	Actinidia Act c 1 Act d1380	166317	1166317	7
Actinidia deliciosa	Kivi	Unassigned	Food Plant	Actinidia Act c 1 Act d1380	193806886	1193806886	12
Actinidia chinensis	Kivi	Act c 1	Food Plant	Actinidia Act c 10 LTP 115	378548410	1378548410	13
Actinidia chinensis	Kivi	Act c 5.0102	Food Plant	Actinidia Act c 5 Kiwellin	1441482354	1441482354	14
Actinidia chinensis	Kivi	Act c 8.0101	Food Plant	Actinidia Act c 8 Act d1159	281552896	1281552896	11
Actinidia deliciosa	Kivi	Act d 8.0101	Food Plant	Actinidia Act c 8 Act d1157	281552898	1281552898	11
Actinidia deliciosa	Kivi	Act d 10.0201	Food Plant	Actinidia Act d 10 LTP 92	378548411	1378548411	13
Actinidia deliciosa	Kivi	Act d 10.0101	Food Plant	Actinidia Act d 10 LTP 92	378405189	1378405189	13
Actinidia deliciosa	Kivi	Act d 11	Food Plant	Actinidia Act d 11 Kirola MLP	332319679	1332319679	12
Actinidia chinensis	Kivi	Unassigned	Food Plant	Actinidia Act d 2 thaumatin like protein	168064399	168064399	7
Actinidia chinensis	Kivi	Unassigned	Food Plant	Actinidia Act d 2 thaumatin like protein	1441482370	1441482370	14
Actinidia deliciosa	Kivi	Act d 2.0101	Food Plant	Actinidia Act d 2 thaumatin like protein	171057064	171057064	7
Actinidia deliciosa	Kivi	Unassigned	Food Plant	Actinidia Act d 2 thaumatin like protein	146737976	146737976	9
Actinidia deliciosa	Kivi	Act d 4.0101	Food Plant	Actinidia Act d 4 Phycocystatin	140807635	140807635	7
Actinidia deliciosa	Kivi	Act d 5.0101	Food Plant	Actinidia Act d 5 Kiwellin	185701136	185701136	7
Actinidia deliciosa	Kivi	Unassigned	Food Plant	Actinidia Act d 5 Kiwellin	1441482346	1441482346	14

Actinidia deliciosa	Kivi	Unassigned	Food Plant	Actinidia Act d 5	1213	441482348	114
Actinidia deliciosa	Kivi	Unassigned	Food Plant	Actinidia Act d 5	1213	441482350	114
Actinidia deliciosa	Kivi	Unassigned	Food Plant	Actinidia Act d 5	1213	441482352	114
Actinidia deliciosa	Kivi	Act d 9.0101	Food Plant	Actinidia Act d 9,	1109	100	116
Actinidia arguta	Hardy Kivi	Unassigned	Food Plant	Actinidia arguta	1213	441482362	114
Actinidia arguta	Hardy Kivi	Unassigned	Food Plant	Actinidia arguta	1213	441482364	114
Actinidia arguta	Hardy Kivi	Unassigned	Food Plant	Actinidia arguta	1213	441482366	114
Actinidia eriantha	(Climber (plant)	Unassigned	Food Plant	Actinidia eriantha	1213	441482356	114
Actinidia eriantha	(Climber (plant)	Unassigned	Food Plant	Actinidia eriantha	1213	441482358	114
Actinidia eriantha	(Climber (plant)	Unassigned	Food Plant	Actinidia eriantha	1213	441482360	114
Aedes aegypti	Yellow fever mosquito	Aed a 1	Venom or Salivary	Aedes Aed a 1 apyrase	1562	156272	17
Aedes aegypti	Yellow fever mosquito	Unassigned	Venom or Salivary	Aedes Aed a 1 apyrase	1562	1193806340	110
Aedes aegypti	Yellow fever mosquito	Aed a 2	Venom or Salivary	Aedes Aed a 2	1321	1205525919	19
Aedes aegypti	Yellow fever mosquito	Aed a 3	Venom or Salivary	Aedes Aed a 3	1253	12114497	17
Aedes aegypti	Yellow fever mosquito	Unassigned	Venom or Salivary	Aedes Aed a 3	1273	194468546	17
Agrostis alba	Bent grass	Unassigned	Aero Plant	Agrostis Agr a 1	126	1320606	17
Agrostis alba	Bent grass	Unassigned	Aero Plant	Agrostis Agr a 1	135	175139987	17
Agrostis alba	Bent grass	Unassigned	Aero Plant	Agrostis Agr a 1	135	175139989	17
Alnus glutinosa	Alder	Aln g 1	Aero Plant	Alnus Aln g 1	1160	1261407	17
Alnus glutinosa	Alder	Aln g 1.0101	Aero Plant	Alnus Aln g 4	185	13319651	17
Alternaria alternata	Fungus	Alt a 1.0101	Aero Fungi	Alternaria Alt a 1	1157	11842045	17
Alternaria alternata	Fungus	Unassigned	Aero Fungi	Alternaria Alt a 1	1115	121913174	17
Alternaria alternata	Fungus	Alt a 1.0102	Aero Fungi	Alternaria Alt a 1	1157	145680856	17
Alternaria alternata	Fungus	Unassigned	Aero Fungi	Alternaria Alt a 1	1133	1390980892	113
Alternaria alternata	Fungus	Unassigned	Aero Fungi	Alternaria Alt a 1	1130	1508123617	115
Alternaria alternata	Fungus	Alt a 10.0101	Aero Fungi	Alternaria Alt a 10 ADH	1497	176666767	17
Alternaria alternata	Fungus	Alt a 12	Aero Fungi	Alternaria Alt a 12	1110	11350779	17
Alternaria alternata	Fungus	Alt a 13.0101	Aero Fungi	Alternaria Alt a 13	1231	174611808	110
Alternaria alternata	Fungus	Alt a 3	Aero Fungi	Alternaria Alt a 3 HSP	1152	114423730	17
Alternaria alternata	Fungus	Alt a 4	Aero Fungi	Alternaria Alt a 4	1436	185701160	17





Ambrosia artemisifolia	Short ragweed	Amb a 8.0102	Aero Plant	Ambrosia Amb a 8 profilin	133	62249512	17
Ambrosia artemisifolia	Short ragweed	Amb a 9.0101	Aero Plant	Ambrosia Amb a 9	183	62249470	17
Ambrosia artemisifolia	Short ragweed	Amb a 9.0102	Aero Plant	Ambrosia Amb a 9	183	62249481	17
Ambrosia trifida	Giant ragweed	Amb t 5	Aero Plant	Ambrosia Amb t 5 Ra5G	173	114091	17
Amphioctopus fangsiao	Octopus	Unassigned	Food Animal	Amphioctopus arginine kinase	348	340742817	112
Anacardium occidentale	Cashew	Ana 0 1.0102	Food Plant	Anacardium Ana 0 1	1536	21666498	17
Anacardium occidentale	Cashew	Ana 0 1.0101	Food Plant	Anacardium Ana 0 1	1538	21914823	17
Anacardium occidentale	Cashew	Ana 0 2	Food Plant	Anacardium Ana 0 2	1457	25991543	17
Anacardium occidentale	Cashew	Ana 0 3	Food Plant	Anacardium Ana 0 3	1138	124473800	17
Ananas comosus	Pineapple	Ana c 2.0101	Aero Plant	Ananas Ana c 2 Bromelain precursor	351	75277440	17
Ananas comosus	Pineapple	Ana c 1.0101	Food Plant	Ananas profilin Ana c 1	1131	75306610	110
Anisakis simplex	Parasitic fish worm	Ani s 1	Food Animal	Anisakis Ani s 1 protease inhibitor	1194	47606452	17
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 1 protease inhibitor	1163	442577863	114
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 1 protease inhibitor	1163	442577865	114
Anisakis simplex	Parasitic fish worm	Ani s 10.0101	Food Animal	Anisakis Ani s 10	1231	272574378	111
Anisakis simplex	Parasitic fish worm	Ani s 11.0101	Food Animal	Anisakis Ani s 11	1307	323575361	112
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 11	1160	323575363	112
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 11	1287	323575365	112
Anisakis pegriffii	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 12	1264	442577845	114
Anisakis pegriffii	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 12	1264	442577847	114
Anisakis pegriffii	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 12	1264	442577849	114
Anisakis simplex	Parasitic fish worm	Ani s 12.0101	Food Animal	Anisakis Ani s 12	1295	323575367	112
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 12	1264	442577851	114
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 12	1264	442577853	114
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 12	1264	442577855	114
Anisakis pegriffii	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 2 paranyosin	1869	442577833	114
Anisakis simplex	Parasitic fish worm	Ani s 2	Food Animal	Anisakis Ani s 2 paranyosin	1473	18453086	17
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 2 paranyosin	1869	42559536	19

Anisakis simplex	Parasitic fish worm	Ani s 3	Food Animal	Anisakis Ani s 3 tropomyosin	1284	14423976	17
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 3 tropomyosin	1284	350285785	13
Anisakis simplex	Parasitic fish worm	Ani s 4	Food Animal	Anisakis Ani s 4	114	47606398	17
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 4	1115	110346534	18
Anisakis simplex	Parasitic fish worm	Ani s 5.0101	Food Animal	Anisakis Ani s 5 SXP/FAI-2 family protein	1152	121308878	18
Anisakis simplex	Parasitic fish worm	Ani s 7.0101	Food Animal	Anisakis Ani s 7 UAS-recognized allergen	1096	119524036	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676636	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676682	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676684	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676686	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676688	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676690	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676692	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676694	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676696	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis Ani s 8 SXP/FAI-2 family protein 2	1150	155676698	19
Anisakis simplex	Parasitic fish worm	Ani s 9.0101	Food Animal	Anisakis Ani s 9	147	157418806	19
Anisakis simplex	Parasitic fish worm	Unassigned	Food Animal	Anisakis simplex troponin-like	1161	6065738	17
Anthoxanthum odoratum	Sweet vernal grass	Unassigned	Aero Plant	Anthoxanthum Ant o 1	126	1320607	17
Anthoxanthum odoratum	Sweet vernal grass	Ant o 1.0101	Aero Plant	Anthoxanthum Ant o 1	132	175139986	17
Anthoxanthum odoratum	Sweet vernal grass	Unassigned	Aero Plant	Anthoxanthum Ant o 1	132	175139990	17
Apis cerana	Indian honeybee	Unassigned	Venom or Salivary	Apis Api m 1 Api d 1 Api c 1	1134	17435005	17



Apis cerana cerana	Indian honeybee	Api c 1.0101	Venom or Salivary	Apis Api m 1 Api d 1	134	12958583	115
Apis dorsata	Giant honeybee	Api d 1.0101	Venom or Salivary	Apis Api m 1 Api d 1	134	47117012	17
Apis mellifera	Honeybee	Api m 1	Venom or Salivary	Apis Api m 1 Api d 1	167	24418862	17
Apis mellifera	Honeybee	Unassigned	Venom or Salivary	Apis Api m 10 icarapin	223	94471622	17
Apis mellifera	Honeybee	Api m 10.0101	Venom or Salivary	Apis Api m 10 icarapin	175	94471624	17
Apis mellifera carnica	Honeybee	Unassigned	Venom or Salivary	Apis Api m 10 icarapin	112	594708629	116
Apis mellifera carnica	Honeybee	Unassigned	Venom or Salivary	Apis Api m 10 icarapin	119	594708627	116
Apis mellifera carnica	Honeybee	Unassigned	Venom or Salivary	Apis Api m 10 icarapin	125	594708625	116
Apis mellifera carnica	Honeybee	Unassigned	Venom or Salivary	Apis Api m 10 icarapin	141	594708623	116
Apis mellifera	Honeybee	Api m 2	Venom or Salivary	Apis Api m 2	382	585279	17
Apis mellifera	Honeybee	Unassigned	Venom or Salivary	Apis Api m 3 acid phosphatase	388	1208342441	110
Apis mellifera	Honeybee	Api m 3.0101	Venom or Salivary	Apis Api m 3 acid phosphatase	388	74835477	112
Apis dorsata	Giant honeybee	Unassigned	Venom or Salivary	Apis Api m 4 Melittin	126	1126955	17
Apis mellifera	Honeybee	Api m 4.0101	Venom or Salivary	Apis Api m 4 Melittin	170	5622	17
Apis mellifera	Honeybee	Unassigned	Venom or Salivary	Apis Api m 4 Melittin	127	69552	17
Apis mellifera	Honeybee	Api m 5.0101	Venom or Salivary	Apis Api m 5 dipeptidylpeptidase	1775	187281543	115
Apis mellifera	Honeybee	Unassigned	Venom or Salivary	Apis Api m 6	192	94400907	17
Apis mellifera	Honeybee	Unassigned	Venom or Salivary	Apis Api m 6	194	88770352	110
Apis mellifera	Honeybee	Api m 11.0101	Venom or Salivary	Apis mellifera Api m 11.416	11.416	58585070	115
Apis mellifera	Honeybee	Api m 11.0201	Venom or Salivary	Apis mellifera Api m 11.423	11.423	62910925	115
Apis mellifera	Honeybee	Api m 12.0101	Venom or Salivary	Apis mellifera Api m 12.1770	12.1770	29329817	115
Apis mellifera	Celery	Api g 1.0101	Food Plant	Apium Api g 1	154	1346568	17
Apis mellifera	Celery	Api g 1.0201	Food Plant	Apium Api g 1	159	14423646	19
Apis mellifera	Celery	Api g 2.0101	Food Plant	Apium Api g 2	118	256600126	112
Apis mellifera	Celery	Api g 4	Food Plant	Apium Api g 4	134	4761578	17
Apis mellifera	Celery	Api g 5.0101	Food Plant	Apium Api g 5	186	33300920	110
Apis mellifera	Celery	Api g 6.0101	Food Plant	Apium graveolens Api g 16 LTP 2	167	550540827	115
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Agglutinin (Lectin)	273	253289	17
Arachis hypogaea	Peanut	Arachis h 1	Food Plant	Arachis Ara h 1	1614	11168390	17
Arachis hypogaea	Peanut	Arachis h 1	Food Plant	Arachis Ara h 1	1626	11168391	17

Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 1	1299	46560474	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 1	1303	46560472	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 1	1428	46560476	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 1	1619	312233063	112
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 1	1418	375332427	113
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 1	1418	347447588	113
Arachis hypogaea	Peanut	Ara h 2.0201	Food Plant	Arachis Ara h 2	1172	26246447	17
Arachis hypogaea	Peanut	Ara h 2.0101	Food Plant	Arachis Ara h 2	1169	31322017	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 2	1166	15418705	110
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 2	1158	1224747150	110
Arachis hypogaea	Peanut	Ara h 3.0101	Food Plant	Arachis Ara h 3 Glycinin	1507	13703107	17
Arachis hypogaea	Peanut	Ara h 3.0201	Food Plant	Arachis Ara h 3 Glycinin	1530	15712199	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 3 Glycinin	1538	121314465	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 3 Glycinin	1219	22136348	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 3 Glycinin	1512	1112380623	18
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 3 Glycinin	1530	119732457	110
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 3 Glycinin	1510	224036293	110
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 3 Glycinin	1512	312233065	112
Arachis hypogaea	Peanut	Ara h 5	Food Plant	Arachis Ara h 5	1131	15902968	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 5	1131	284810529	111
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 5	1131	431812555	114
Arachis hypogaea	Peanut	Ara h 6	Food Plant	Arachis Ara h 6	1129	15923742	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 6	1144	11725991	17
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 6	1127	159163254	19
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 6	1145	175114094	110
Arachis hypogaea	Peanut	Ara h 7.0101	Food Plant	Arachis Ara h 7	1160	15931948	17
Arachis hypogaea	Peanut	Ara h 7.0201	Food Plant	Arachis Ara h 7	1164	1158121995	110
Arachis hypogaea	Peanut	Ara h 8.0101	Food Plant	Arachis Ara h 8	1157	13749626	17
Arachis hypogaea	Peanut	Ara h 8.0201	Food Plant	Arachis Ara h 8	1153	1145904610	19
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 8	1157	1169786740	19
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis Ara h 8	1157	1110676574	112

Arachis hypogaea	Peanut	Ara h 9.0101	Food Plant	Arachis Ara h 9 LTP	116	161087230	110
Arachis hypogaea	Peanut	Ara h 9.0201	Food Plant	Arachis Ara h 9 LTP	92	161610680	110
Arachis hypogaea	Peanut	Ara h 10.0101	Food Plant	Arachis hypogaea Ara h 169	110	113200509	115
Arachis hypogaea	Peanut	Ara h 10.0102	Food Plant	Arachis hypogaea Ara h 150	110	152001239	115
Arachis hypogaea	Peanut	Ara h 11.0101	Food Plant	Arachis hypogaea Ara h 137	111	71040655	115
Arachis hypogaea	Peanut	Unassigned	Food Plant	Arachis hypogaea Ara h 137	111	122218640	116
Argas reflexus	European pigeon tick	Arg r 1	Venom or Salivary	Argas Arg r 1	159	158371884	17
Argas reflexus	European pigeon tick	Unassigned	Venom or Salivary	Argas Arg r 1	144	1322812205	112
Artemisia vulgaris	Mugwort	Art v 1	Aero Plant	Artemisia Art v 1	132	127818335	17
Artemisia vulgaris	Mugwort	Art v 2.0101	Aero Plant	Artemisia Art v 2	162	148887203	19
Artemisia vulgaris	Mugwort	Art v 3.0101	Aero Plant	Artemisia Art v 3	137	173621307	17
Artemisia vulgaris	Mugwort	Art v 3.0201	Aero Plant	Artemisia Art v 3	114	189544578	111
Artemisia vulgaris	Mugwort	Art v 3.0202	Aero Plant	Artemisia Art v 3	116	189544584	111
Artemisia vulgaris	Mugwort	Art v 3.0301	Aero Plant	Artemisia Art v 3	117	189544590	111
Artemisia vulgaris	Mugwort	Unassigned	Aero Plant	Artemisia Art v 3	117	189544595	111
Artemisia vulgaris	Mugwort	Art v 4.0101	Aero Plant	Artemisia Art v 4	133	12955969	115
Artemisia vulgaris	Mugwort	Art v 4.0201	Aero Plant	Artemisia Art v 4	133	12955971	115
Artemisia vulgaris	Mugwort	Art v 6.0101	Aero Plant	Artemisia Art v 6	136	162530283	18
Artemisia vulgaris	Mugwort	Art v 5.0101	Aero Plant	Artemisia mugwort Art v 82	15	162530285	115
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	134	12735096	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	134	12735098	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	133	12735102	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	133	12735106	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	1267	12735108	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	1267	12735110	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	1267	12735112	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	134	12735114	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	134	12735118	17
Ascaris lumbricoides	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	134	12735100	17
Ascaris suum	Parasitic roundworm	Asc s 1	Worm (parasite)	Ascaris Asc s 1	168	1295550	17
Ascaris suum	Parasitic roundworm	Asc s 1	Worm (parasite)	Ascaris Asc s 1	1365	177416849	17
Ascaris suum	Parasitic roundworm	Unassigned	Worm (parasite)	Ascaris Asc s 1	134	1343197079	112

Ascaris suum	Parasitic roundworm	Asc s 13.0101	Worm (parasite)	Ascaris lumbricoidea/suum	1206	1170109	15
Ascaris lumbricoidea	Parasitic roundworm	Asc 1 3.0101	Worm (parasite)	Glutathione S-transferase			
Ascaris lumbricoidea	Parasitic roundworm	Asc 1 3.0101	Worm (parasite)	Ascaris troponosin Asc	287	224016002	10
Aspergillus fumigatus Fungus				1 3			
Aspergillus fumigatus Fungus		Asp f 1	Aero Fungi	Aspergillus Asp f 1	125	13021324	17
Aspergillus fumigatus Fungus		Asp f 1	Aero Fungi	Aspergillus Asp f 1	150	19280360	17
Aspergillus fumigatus Fungus		Unassigned	Aero Fungi	Aspergillus Asp f 1	176	154039254	17
Aspergillus fumigatus Fungus		Asp f 10	Aero Fungi	Aspergillus Asp f 10	395	1963013	17
Aspergillus fumigatus Fungus		Asp f 11	Aero Fungi	Aspergillus Asp f 11	178	15019414	17
Aspergillus fumigatus Fungus		Asp f 18.0101	Aero Fungi	Aspergillus Asp f 18	1495	12143220	17
Aspergillus niger	Fungus	Unassigned	Aero Fungi	Aspergillus Asp f 18 and Asp n 18		1289172	17
Aspergillus fumigatus Fungus		Asp f 22	Aero Fungi	Aspergillus Asp f 22	1438	113925873	17
Aspergillus fumigatus Fungus		Unassigned	Aero Fungi	Aspergillus Asp f 22	1438	18328046	17
Aspergillus fumigatus Fungus		Asp f 23.0101	Aero Fungi	Aspergillus Asp f 23	392	121215170	17
Aspergillus fumigatus Fungus		Unassigned	Aero Fungi	Aspergillus Asp f 23	392	183305621	17
Aspergillus fumigatus Fungus		Asp f 27.0101	Aero Fungi	Aspergillus Asp f 27	163	191680605	17
Aspergillus fumigatus Fungus		Asp f 28.0101	Aero Fungi	Aspergillus Asp f 28	108	191680607	17
Aspergillus fumigatus Fungus		Asp f 29.0101	Aero Fungi	Aspergillus Asp f 29	110	191680609	17
Aspergillus fumigatus Fungus		Asp f 3	Aero Fungi	Aspergillus Asp f 3	168	12769700	17
Aspergillus fumigatus Fungus		Asp f 34.0101	Aero Fungi	Aspergillus Asp f 34	185	1133920236	18
Aspergillus fumigatus Fungus		Asp f 4	Aero Fungi	Aspergillus Asp f 4	286	13005839	17
Aspergillus fumigatus Fungus		Unassigned	Aero Fungi	Aspergillus Asp f 4	322	183300369	17
Aspergillus fumigatus Fungus		Unassigned	Aero Fungi	Aspergillus Asp f 4	322	1666434194	16
Aspergillus fumigatus Fungus		Asp f 5	Aero Fungi	Aspergillus Asp f 5	1634	13776613	17
Aspergillus fumigatus Fungus		Unassigned	Aero Fungi	Aspergillus Asp f 5	1634	185541646	111
Aspergillus fumigatus Fungus		Asp f 6	Aero Fungi	Aspergillus Asp f 6	1221	11648970	17
Aspergillus fumigatus Fungus		Unassigned	Aero Fungi	Aspergillus Asp f 6	1210	183305645	17
Aspergillus fumigatus Fungus		Asp f 7	Aero Fungi	Aspergillus Asp f 7	270	183300389	17
Aspergillus fumigatus Fungus		Unassigned	Aero Fungi	Aspergillus Asp f 7	270	1666431137	16
Aspergillus fumigatus Fungus		Asp f 8	Aero Fungi	Aspergillus Asp f 8	1111	1686524	17

Aspergillus fumigatus	Fungus	Unassigned	Aero Fungi	Aspergillus Asp f 8	111	18306635	17
Aspergillus fumigatus	Fungus	Asp f 9	Aero Fungi	Aspergillus Asp f 9	302	2879890	17
Aspergillus niger	Fungus	Asp n 14	Aero Fungi	Aspergillus Asp n 14	804	2181180	17
Aspergillus niger	Fungus	Asp n 14	Aero Fungi	Aspergillus Asp n 14	804	4235093	17
Aspergillus oryzae	Fungus	Asp o 21	Aero Fungi	Aspergillus Asp o 21	499	194706935	17
Aspergillus oryzae	Fungus	Asp o 21.0101	Aero Fungi	Aspergillus Asp o 21	499	166531	115
Aspergillus fumigatus	Fungus	Unassigned	Aero Fungi	Aspergillus Endo-chitosanase	238	174629604	116
Aspergillus fumigatus	Fungus	Unassigned	Aero Fungi	Aspergillus Endo-chitosanase	242	174666748	116
Aspergillus flavus	Fungus	Unassigned	Aero Fungi	Aspergillus Oryzin Asp o 13, fl 13	403	174665726	17
Aspergillus oryzae	Fungus	Asp o 13	Aero Fungi	Aspergillus Oryzin Asp o 13, fl 13	403	129235	17
Aspergillus versicolor	Fungus	Unassigned	Aero Fungi	Aspergillus versicolor serine protease	403	129441150	116
Bacillus sp.	Bacteria	Unassigned	Bacteria airway	Bacillus lentus	361	1125905	19
Bacillus lentus	Bacteria	Unassigned	Bacteria airway	Bacillus lentus subtilisin	269	267048	19
Bacillus licheniformis	Bacteria	Unassigned	Bacteria airway	Bacillus licheniformis subtilisin	379	1135016	19
Bacillus licheniformis	Bacteria	Unassigned	Bacteria airway	Bacillus licheniformis subtilisin	374	11127680	19
Balanus rostratus	Crustacean	Unassigned	Food Animal	Balanus r tropomyosin	284	125659386	19
Bassia scoparia	summer cypress	Unassigned	Aero Plant	Bassia scoparia Koc s	1167	1914410012	116
Batillus cornutus	Japanese turban shell	Unassigned	Food Animal	Batillus Tur c1	284	219806888	110
Bertholletia excelsa	Brazil nut	Ber e 1	Food Plant	Bertholletia Ber e 1	146	112754	17
Bertholletia excelsa	Brazil nut	Ber e 2	Food Plant	Bertholletia Ber e 2	465	130313867	17
Betula pendula	European white birch	Unassigned	Aero Plant	Betula glutathione S-transferase	237	1573005958	116
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	151	1320545	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	1534898	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	1534900	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	1534910	17
Betula pendula	European white birch	Bet v 1.1601	Aero Plant	Betula Bet v 1	160	1321714	17
Betula pendula	European white birch	Bet v 1.1701	Aero Plant	Betula Bet v 1	160	1321716	17
Betula pendula	European white birch	Bet v 1.1801	Aero Plant	Betula Bet v 1	160	1321718	17
Betula pendula	European white birch	Bet v 1.1502	Aero Plant	Betula Bet v 1	160	1321720	17
Betula pendula	European white birch	Bet v 1.1901	Aero Plant	Betula Bet v 1	160	1321722	17
Betula pendula	European white birch	Bet v 1.2001	Aero Plant	Betula Bet v 1	160	1321724	17

Betula pendula	European white birch	Bet v 1.2101	Aero Plant	Betula Bet v 1	160	1321726	17
Betula pendula	European white birch	Bet v 1.2201	Aero Plant	Betula Bet v 1	160	1321728	17
Betula pendula	European white birch	Bet v 1a/n	Aero Plant	Betula Bet v 1	160	1168710	17
Betula pendula	European white birch	Bet v 1.0108	Aero Plant	Betula Bet v 1	160	1542861	17
Betula pendula	European white birch	Bet v 1.0109	Aero Plant	Betula Bet v 1	160	1542863	17
Betula pendula	European white birch	Bet v 1.0110	Aero Plant	Betula Bet v 1	160	1542865	17
Betula pendula	European white birch	Bet v 1.0111	Aero Plant	Betula Bet v 1	160	1542867	17
Betula pendula	European white birch	Bet v 1.0112	Aero Plant	Betula Bet v 1	160	1542869	17
Betula pendula	European white birch	Bet v 1.0113	Aero Plant	Betula Bet v 1	160	1542871	17
Betula pendula	European white birch	Bet v 1.0114	Aero Plant	Betula Bet v 1	160	1542873	17
Betula pendula	European white birch	Bet v 1.2301	Aero Plant	Betula Bet v 1	160	1241458	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	12564220	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	12564222	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	12564224	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	12564228	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006928	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006945	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006953	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006955	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006957	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006959	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006961	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006965	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	160	14006967	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	14376216	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	14376219	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	14376220	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	14376221	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	14376222	17
Betula pendula	European white birch	Bet v 1 b1	Aero Plant	Betula Bet v 1	160	14590392	17
Betula pendula	European white birch	Bet v 1 b2	Aero Plant	Betula Bet v 1	160	14590394	17
Betula pendula	European white birch	Bet v 1 b3	Aero Plant	Betula Bet v 1	160	14590396	17
Betula pendula	European white birch	Bet v 1.0701	Aero Plant	Betula Bet v 1	160	1168706	17
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	11514622	17
Betula pendula	European white birch	Bet v 1x	Aero Plant	Betula Bet v 1	121	130908931	17

Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	38492423	17
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	143	129734	17
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	120	14006963	17
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	120	14006947	17
Betula pendula	European white birch	Bet v 1.0203	Aero Plant	Betula Bet v 1	160	1452742	18
Betula pendula	European white birch	Bet v 1	Aero Plant	Betula Bet v 1	159	159162097	19
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	159	1560188693	115
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	159	1550544347	115
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	160	1565807648	115
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	159	1560188694	115
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	159	1560188692	115
Betula pendula	European white birch	Bet v 1.0101	Aero Plant	Betula Bet v 1	160	117938	115
Betula pendula	European white birch	Bet v 1.0102	Aero Plant	Betula Bet v 1	160	1452732	115
Betula pendula	European white birch	Bet v 1.0103	Aero Plant	Betula Bet v 1	160	1452734	115
Betula pendula	European white birch	Bet v 1.0104	Aero Plant	Betula Bet v 1	160	1452736	115
Betula pendula	European white birch	Bet v 1.0106	Aero Plant	Betula Bet v 1	160	1452740	115
Betula pendula	European white birch	Bet v 1.0107	Aero Plant	Betula Bet v 1	160	1452744	115
Betula pendula	European white birch	Bet v 1.0201	Aero Plant	Betula Bet v 1	160	1450885	115
Betula pendula	European white birch	Bet v 1.0202	Aero Plant	Betula Bet v 1	160	1452730	115
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1	159	1661918055	116
Betula platyphylla	Japanese white birch	Unassigned	Aero Plant	Betula Bet v 1	160	112583681	17
Betula platyphylla	Japanese white birch	Unassigned	Aero Plant	Betula Bet v 1	160	112583683	17
Betula platyphylla	Japanese white birch	Unassigned	Aero Plant	Betula Bet v 1	160	112583685	17
Betula sp.	Birch	Unassigned	Aero Plant	Betula Bet v 1	151	1298736	17
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 1b	151	1320546	17
Betula sp.	Birch	Unassigned	Aero Plant	Betula Bet v 1b	151	1298737	17
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 2	133	1157830684	19
Betula pendula	European white birch	Bet v 2.0101	Aero Plant	Betula Bet v 2	133	1169563	111
Betula pendula	European white birch	Unassigned	Aero Plant	Betula Bet v 2	133	1576017922	115
Betula pendula	European white birch	Bet v 3.0101	Aero Plant	Betula Bet v 3	1205	1488605	115
Betula pendula	European white birch	Bet v 4.0101	Aero Plant	Betula Bet v 4	185	1809536	115
Betula pendula	European white birch	Bet v 6.0102	Aero Plant	Betula Bet v 6	1308	110764491	17
Betula pendula	European white birch	Bet v 7	Aero Plant	Betula Bet v 7	1173	121886603	17
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella chymotrypsin-like	1252	1757943154	116
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella 36 kDa allergen	120	1544618	17

Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella 36 kDa allergen	125	544619	17
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella arginine kinase	366	221602737	110
Blattella germanica	German cockroach	Bla g 1.0201	Aero Insect	Blattella Bla g 1	1492	4240396	17
Blattella germanica	German cockroach	Bla g 1.0101	Aero Insect	Blattella Bla g 1	1412	4572592	17
Blattella germanica	German cockroach	Bla g 11.0101	Aero Insect	Blattella Bla g 11 alpha Amylase	515	85002763	115
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 2	330	62738637	17
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 2	362	145105726	19
Blattella germanica	German cockroach	Bla g 2.0101	Aero Insect	Blattella Bla g 2	362	1176397	111
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 2	334	315113421	112
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 3	1657	262272875	111
Blattella germanica	German cockroach	Bla g 3.0101	Aero Insect	Blattella Bla g 3	1657	262272877	111
Blattella germanica	German cockroach	Bla g 4	Aero Insect	Blattella Bla g 4	182	1166673	17
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 4	182	144952778	19
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 4	181	212675308	110
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 4	191	194350815	111
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 4	190	194350817	111
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella Bla g 5	1200	144952780	19
Blattella germanica	German cockroach	Bla g 5.0101	Aero Insect	Blattella Bla g 5	1200	2326190	111
Blattella germanica	German cockroach	Bla g 6.0101	Aero Insect	Blattella Bla g 6	151	82704032	18
Blattella germanica	German cockroach	Bla g 6.0201	Aero Insect	Blattella Bla g 6	151	82704034	18
Blattella germanica	German cockroach	Bla g 6.0301	Aero Insect	Blattella Bla g 6	154	82704036	18
Blattella germanica	German cockroach	Bla g 7.0101	Aero Insect	Blattella Bla g 7	284	8101069	17
Blattella germanica	German cockroach	Unassigned	Aero Insect	Blattella delta GST	216	161137518	111
Blomia tropicalis	Mite	Blo t 1	Aero Mite	Blomia Blo t 1.01	221	14276828	17
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Blo t 1.02	333	13667928	18
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Blo t 1.02	333	12	18
Blomia tropicalis	Mite	Blo t 10.0101	Aero Mite	Blomia Blo t 10	284	156938889	19
Blomia tropicalis	Mite	Blo t 11	Aero Mite	Blomia Blo t 11	1875	21954740	17
Blomia tropicalis	Mite	Unassigned	Aero Insect	Blomia Blo t 12	169	1723586656	116
Blomia tropicalis	Mite	Blo t 12	Aero Mite	Blomia Blo t 12	144	1902012	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Blomia Blo t 12	143	13943777	17
Blomia tropicalis	Mite	Blo t 13	Aero Mite	Blomia Blo t 13.01	130	11377869	17
Blomia tropicalis	Mite	Blo t 21.0101	Aero Insect	Blomia Blo t 21	129	160679570	19



Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Elo t 21	129	111120432	8
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Elo t 21	129	111494253	8
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Elo t 21	129	111120424	8
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Elo t 21	129	111120428	8
Blomia tropicalis	Mite	Blo t 3.0101	Aero Mite	Blomia Elo t 3	1266	25989482	17
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Elo t 3	1266	133667930	8
Blomia tropicalis	Mite	Unassigned	Aero Insect	Blomia Elo t 4 alpha amylase	1506	133667932	8
Blomia tropicalis	Mite	Blo t 5	Aero Mite	Blomia Elo t 5	1134	4204917	17
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Elo t 5	1134	111120436	9
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Elo t 5	1134	111120450	9
Blomia tropicalis	Mite	Unassigned	Aero Mite	Blomia Elo t 5	1119	160285626	9
Blomia tropicalis	Mite	Unassigned	Aero Insect	Blomia Elo t 7 non_luis1 allergen	1192	133667936	8
Blomia tropicalis	Mite	Unassigned	Aero Insect	Blomia Elo t 8	1236	137958149	8
Blomia tropicalis	Mite	Blo t 8.0101	Aero Insect	Blomia Elo t 8	1236	256665455	11
Bombus pennsylvanicus	Bumblebee	Bom p 1.0101	Venom or Salivary	Bombus Bom p 1	1136	47117013	112
Bombus pennsylvanicus	Bumblebee	Bom p 4.0101	Venom or Salivary	Bombus Bom p 4 protease	243	75009997	112
Bombus terrestris	Bumblebee	Bom t 1.0101	Venom or Salivary	Bombus Bom t 1	1136	114423832	17
Bombus terrestris	Bumblebee	Unassigned	Venom or Salivary	Bombus Bom t 4 protease	20	1313471465	112
Bombyx mori	Silkworm	Bomb m 1.0101	Aero Insect	Bombyx Bomb m 1	1355	1826558675	115
Bos taurus	Bovine	Unassigned	Food Animal	Bos Alpha-s1 casein	193	162650	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Alpha-s1 casein	1214	162794	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Alpha-s1 casein	176	162927	17
Bos taurus	Bovine	Bos d 9.0101	Food Animal	Bos Alpha-s1 casein	1214	130794348	8
Bos taurus	Bovine	Unassigned	Food Animal	Bos Alpha-s1 casein	1205	159793197	9
Bos taurus	Bovine	Unassigned	Food Animal	Bos Alpha-s1 casein	1172	159793201	9
Bos taurus	Bovine	Unassigned	Food Animal	Bos Alpha-s1 casein	1129	159793217	9
Bos taurus	Bovine	Bos d 10.0101	Food Animal	Bos Bos d 10	222	127806963	115
Bos grunniens mutus	Yak	Unassigned	Food Animal	Bos Bos d 11 beta casein	1259	1942073448	116
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 11 beta casein	1224	162797	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 11 beta casein	1224	162805	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 11 beta casein	1224	1459292	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 12	1190	162811	17
Bos taurus	Bovine	Bos d 12.0101	Food Animal	Bos Bos d 12	1190	127881412	115

Bos taurus	Bovine	Unassigned	Aero Animal	Bos Bos d 2	1172	12497701	19
Bos taurus	Bovine	Bos d 3	Aero Animal	Bos Bos d 3	1101	12493414	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 4	1142	125774	17
Bos taurus	Bovine	Bos d 4.0101	Food Animal	Bos Bos d 4	1142	1163283	115
Bos taurus	Bovine	Bos d 5	Food Animal	Bos Bos d 5	1178	1520	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 5	114	1162750	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 5	1178	1125910	19
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 5	1178	1195957138	110
Bos taurus	Bovine	Bos d 6	Food Animal	Bos Bos d 6	1607	1162648	17
Bos taurus	Bovine	Unassigned	Food Animal	Bos Bos d 6	1607	11336842	17
Bos taurus	Bovine	Unassigned	Vaccine	Bos collagen alpha2	11364	127806257	111
Bos taurus	Bovine	Unassigned	Food Animal	Bos lactotransferrin	1708	130794292	18
Brassica napus	Rape	Bra n 1	Food Plant	Bra n 1	1125	175107016	19
Brassica napus	Rape	Unassigned	Aero Plant	Bra n Bra r 2	183	1212801	17
Brassica napus	Rape	Unassigned	Aero Plant	Bra n Bra r 2	183	1212802	17
Brassica rapa	Turnip	Unassigned	Aero Plant	Bra n Bra r 2	180	1212805	17
Brassica rapa subsp. rapa	Turnip	Unassigned	Aero Plant	Bra n Bra r 2	183	159800146	17
Brassica juncea	Mustard	Bra j 1	Food Plant	Brassica Bra j 1 2S	1129	132363444	19
Brassica oleracea var. oleracea	Wild cabbage	Unassigned	Aero Plant	Brassica Bra o 3 LTP full length	1112	1922434456	116
Brassica rapa	Turnip	Bra r 1.0101	Food Plant	Brassica Bra r 1	1178	117697	19
Brassica rapa subsp. rapa	Turnip	Bra r 2.0101	Contact	Brassica Bra r 2	191	132363456	19
Brassica rapa	Turnip	Bra r 5.0101	Food Plant	Brassica Calcim binding protein Group I	179	11255540	115
Brassica napus	Rape	Unassigned	Food Plant	Brassica napus 2S	1109	126985163	17
Candida albicans	Yeast	Cand a 1.0101	Contact	Candida Cand a 1 Alcohol dehydrogenase	1350	1608690	115
Candida albicans	Yeast	Cand a 3.0101	Contact	Candida Cand a 3 Peroxisomal protein	1236	137548637	17
Candida albicans	Yeast	Unassigned	Contact	Candida Emolase 1	1440	1232054	17
Canis familiaris	Dog	Can f 1.0101	Aero Animal	Canis Can f 1 Lipocalin	1174	12598974	111
Canis familiaris	Dog	Can f 2	Aero Animal	Canis Can f 2 Lipocalin	1177	129292272	17
Canis familiaris	Dog	Can f 2	Aero Animal	Canis Can f 2 Lipocalin	1179	129292274	17
Canis familiaris	Dog	Can f 2.0101	Aero Animal	Canis Can f 2 Lipocalin	1180	12598976	111
Canis familiaris	Dog	Can f 3	Aero Animal	Canis Can f 3 Serum albumin	1265	1633938	17

Canis familiaris	Dog		Can f 3	Aero Animal	Canis Can f 3 Serum	1585	13319897	17
Canis familiaris	Dog				albumin			
Canis familiaris	Dog		Can f 3.0101	Aero Animal	Canis Can f 3 Serum	1608	125631688	115
Canis familiaris	Dog				albumin			
Canis familiaris	Dog		Can f 4.0101	Aero Animal	Canis Can f 4	1174	1262232390	112
Canis familiaris	Dog				epithelial 18 kDa			
Canis familiaris	Dog		Unassigned	Aero Animal	Canis Can f 4	1174	1625295108	116
Canis familiaris	Dog				epithelial 18 kDa			
Canis familiaris	Dog		Can f 5.0101	Aero Animal	Canis Can f 5	1260	1868	115
Canis familiaris	Dog		Can f 6.0101	Aero Animal	Canis Can f 6 Lipocalin	1190	1374092884	113
Cannabis sativa	Hemp		Can s 3.0101	Aero Plant	Cannabis LTP Can s 3	191	1571256597	115
Capsicum annuum	Bell pepper		Cap a 1	Food Plant	Capsicum Cap a 1	1246	116609959	17
Capsicum annuum	Bell pepper		Cap a 2	Food Plant	Capsicum Cap a 2	1131	116555785	17
Carica papaya	Papaya		Unassigned	Food Plant	Carica Car p 1	1345	1129614	19
Carpinus betulus	Hornbeam		Car b 1.0102	Aero Plant	Carpinus Car b 1	1159	1402745	17
Carpinus betulus	Hornbeam		Car b 1.0103	Aero Plant	Carpinus Car b 1	1160	11545875	17
Carpinus betulus	Hornbeam		Car b 1.0104	Aero Plant	Carpinus Car b 1	1160	11545877	17
Carpinus betulus	Hornbeam		Car b 1.0105	Aero Plant	Carpinus Car b 1	1160	11545879	17
Carpinus betulus	Hornbeam		Car b 1.0108	Aero Plant	Carpinus Car b 1	1160	11545893	17
Carpinus betulus	Hornbeam		Car b 1.0301	Aero Plant	Carpinus Car b 1	1161	11545895	17
Carpinus betulus	Hornbeam		Car b 1.0302	Aero Plant	Carpinus Car b 1	1161	11545897	17
Carpinus betulus	Hornbeam		Unassigned	Aero Plant	Carpinus Car b 1	140	1239735	17
Carpinus betulus	Hornbeam		Car b 1.0113	Aero Plant	Carpinus Car b 1	1160	167472845	110
Carpinus betulus	Hornbeam		Car b 1.0109	Aero Plant	Carpinus Car b 1	1160	167472837	110
Carpinus betulus	Hornbeam		Car b 1.0112	Aero Plant	Carpinus Car b 1	1160	167472843	110
Carpinus betulus	Hornbeam		Car b 1.0111	Aero Plant	Carpinus Car b 1	1160	167472841	110
Carpinus betulus	Hornbeam		Car b 1.0110	Aero Plant	Carpinus Car b 1	1160	167472839	110
Carpinus betulus	Hornbeam		Unassigned	Aero Plant	Carpinus Car b 1	180	11008578	112
Carpinus betulus	Hornbeam		Unassigned	Aero Plant	Carpinus Car b 1	180	11008579	112
Carpinus betulus	Hornbeam		Unassigned	Aero Plant	Carpinus Car b 1	180	11008580	112
Carpinus betulus	Hornbeam		Car b 1.0101	Aero Plant	Carpinus Car b 1	1159	1402743	115
Carpinus betulus	Hornbeam		Car b 1.0106	Aero Plant	Carpinus Car b 1	1160	11545881	115
Carpinus betulus	Hornbeam		Car b 1.0107	Aero Plant	Carpinus Car b 1	1160	11545889	115
Carpinus betulus	Hornbeam		Car b 1.0201	Aero Plant	Carpinus Car b 1	1159	1402747	115
Carya illinoensis	Pecan		Car i 1.0101	Food Plant	Carya Car i 1 Seed storage protein	1143	128207731	17
Carya illinoensis	Pecan		Car i 4.0101	Food Plant	Carya Car i 4 11s legumin	1505	1158998780	114

Carya illinoensis	Pecan	Unassigned	Food Plant	Carya Car i 4 11s legumin	1505	15898782	114
Caryota mitis	Fishtail Palm	Unassigned	Aero Plant	Caryota profilin	131	121277849	8
Castanea sativa	European chestnut	Cas s 1	Aero Plant	Castanea Cas s 1	160	16555781	17
Castanea sativa	European chestnut	Unassigned	Aero Plant	Castanea Cas s 1	159	212291466	110
Castanea sativa	European chestnut	Cas s 1.0101	Aero Plant	Castanea Cas s 1	159	212291464	110
Castanea sativa	European chestnut	Unassigned	Aero Plant	Castanea Cas s 1	159	212291468	110
Castanea sativa	European chestnut	Cas s 5	Food Plant	Castanea Cas s 5	316	1359600	17
Castanea sativa	European chestnut	Unassigned	Food Plant	Castanea Cas s 5	298	307159110	112
Catharanthus roseus	Madagascar periwinkle	Unassigned	Aero Plant	Catharanthus cyclophilin	178	659835152	116
Cavia porcellus	Domestic guinea pig	Cav p 1	Aero Animal	Cavia Cav p 1	115	32469617	17
Cavia porcellus	Domestic guinea pig	Cav p 2.0101	Aero Animal	Cavia Cav p 2	170	325910590	112
Cavia porcellus	Domestic guinea pig	Cav p 3.0101	Aero Animal	Cavia Cav p 3 lipocalin	170	325910592	112
Chamaecyparis obtusa	Japanese cypress	Cha o 1.0101	Aero Plant	Chamaecyparis Cha o 1	375	1514943	17
Chamaecyparis obtusa	Japanese cypress	Unassigned	Aero Plant	Chamaecyparis Cha o 2	1514	47606004	17
Chamaecyparis obtusa	Japanese cypress	Unassigned	Aero Plant	Chamaecyparis Cha o 2	1419	114841683	18
Charybdis feriatus	Crab	Cha f 1.0101	Food Animal	Charybdis Cha f 1	264	17024506	17
Chenopodium album	Pigweed	Che a 1	Aero Plant	Chenopodium Che a 1	168	122074346	17
Chenopodium album	Pigweed	Che a 2	Aero Plant	Chenopodium Che a 2	131	29465666	17
Chenopodium album	Pigweed	Unassigned	Aero Plant	Chenopodium Che a 2	133	238886048	111
Chenopodium album	Pigweed	Che a 3	Aero Plant	Chenopodium Che a 3	186	29465668	17
Chionoecetes opilio	Snow Crab	Unassigned	Food Animal	Chionoecetes tropomyosin	284	308191688	112
Chironomus kienensis	Midge	Chi k 10	Aero Insect	Chironomus Chi k 10	285	17321108	17
Chironomus thummi	Midge	Chi t 1.01	Aero Insect	Chironomus Chi t 1	151	121219	17
Chironomus thummi	Midge	Chi t 1.02	Aero Insect	Chironomus Chi t 1	151	121227	17
Chironomus thummi	Midge	Chi t 2.0101	Aero Insect	Chironomus Chi t 2	158	2506460	17
Chironomus thummi	Midge	Chi t 3.0601	Aero Insect	Chironomus Chi t 3	161	156405052	17
Chironomus thummi	Midge	Chi t 3.0901	Aero Insect	Chironomus Chi t 3	151	121237	17
Chironomus thummi	Midge	Chi t 3.0501	Aero Insect	Chironomus Chi t 3	161	121244	17
Chironomus thummi	Midge	Chi t 3.0701	Aero Insect	Chironomus Chi t 3	161	156405054	17
Chironomus thummi	Midge	Chi t 3.0702	Aero Insect	Chironomus Chi t 3	161	121248	17

Chironomus thummi	Midge	Chi t 3.0801	Aero Insect	Chironomus Chi t 3	162	121249	17
Chironomus thummi	Midge	Chi t 3.0301	Aero Insect	Chironomus Chi t 3	161	156406306	17
Chironomus thummi	Midge	Chi t 3.0101	Aero Insect	Chironomus Chi t 3	160	1707908	17
Chironomus thummi	Midge	Chi t 3.0401	Aero Insect	Chironomus Chi t 3	161	1707911	17
Chironomus thummi	Midge	Chi t 3.0201	Aero Insect	Chironomus Chi t 3	162	12506461	17
Chironomus thummi	Midge	Chi t 4	Aero Insect	Chironomus Chi t 4	151	121256	17
Chironomus thummi	Midge	Chi t 9	Aero Insect	Chironomus Chi t 9	151	121259	17
Citrus sinensis	Navel orange	Cit s 1.0101	Food Plant	Citrus Cit s 1	125	152782810	17
Citrus sinensis	Navel orange	Cit s 2.0101	Food Plant	Citrus Cit s 2	131	156000996	17
Citrus limon	Lemon	Cit 1 3.0101	Food Plant	Citrus LTP Cit s 3/Cit 1 3	120	152783176	17
Citrus sinensis	Navel orange	Unassigned	Food Plant	Citrus LTP Cit s 3/Cit 1 3	120	152783177	17
Citrus sinensis	Navel orange	Cit s 3	Food Plant	Citrus LTP Cit s 3/Cit 1 3	191	150199132	17
Davidiella tassiana	Fungus	Cla h 10	Aero Fungi	Cladosporium / Davidiella Cla h 10	1496	176666769	17
Davidiella tassiana	Fungus	Cla h 5.0101	Aero Fungi	Cladosporium / Davidiella Cla h 5	111	15777795	110
Davidiella tassiana	Fungus	Cla h 6	Aero Fungi	Cladosporium / Davidiella Cla h 6	1440	1467660	17
Davidiella tassiana	Fungus	Cla h 6	Aero Fungi	Cladosporium / Davidiella Cla h 6	1440	16015094	17
Davidiella tassiana	Fungus	Cla h 7.0101	Aero Fungi	Cladosporium / Davidiella Cla h 7	1204	1467629	110
Davidiella tassiana	Fungus	Cla h 8.0101	Aero Fungi	Cladosporium / Davidiella Cla h 8	1267	137780015	18
Davidiella tassiana	Fungus	Cla h 9.0101	Aero Fungi	Cladosporium / Davidiella Cla h 9	1518	160116876	110
Davidiella tassiana	Fungus	Unassigned	Aero Fungi	Cladosporium / Davidiella Heat shock 170 kDa protei	1643	1729764	17
Davidiella tassiana	Fungus	Unassigned	Aero Fungi	Cladosporium / Davidiella Hydrophobin	1105	122796153	17
Davidiella tassiana	Fungus	Unassigned	Aero Fungi	Cladosporium / Davidiella putative hydrolase	1274	176446100	110
Davidiella tassiana	Fungus	Unassigned	Aero Fungi	Cladosporium / Davidiella Putative nuclear transpo	1125	121748151	17

Cladosporium cladosporioides	Fungus	Clas c 14.0101	Aero Fungi	Cladosporium Clas c 14	1325	1301015198	115
Cladosporium cladosporioides	Fungus	Clas c 9.0101	Aero Fungi	Cladosporium Clas c 9	1388	148361511	111
Clupea harengus	Atlantic herring	Clupea h 1.0101	Food Animal	Clupea Clupea h 1	1109	242253963	111
Clupea harengus	Atlantic herring	Clupea h 1.0201	Food Animal	Clupea Clupea h 1	1110	242253965	111
Clupea harengus	Atlantic herring	Clupea h 1.0301	Food Animal	Clupea Clupea h 1	1109	242253967	111
Cochliobolus lunatus	Fungus	Cur 1 3.0101	Aero Fungi	Cochliobolus (Curvularia) Cur 1 3	1108	14686755	115
Cochliobolus lunatus	Fungus	Cur 1 2.01	Aero Fungi	Cochliobolus (Curvularia) enolase Cur 1 2.01	1440	14686753	118
Coffea arabica	Coffee	Cof a 3.0101	Food Plant	Coffea Cof a 3	165	494319676	115
Coffea arabica	Coffee	Cof a 1.0101	Food Plant	Coffea Cof a 1	1263	296399179	115
Coffea arabica	Coffee	Cof a 2.0101	Food Plant	Coffea Cof a 2	180	494319674	115
Coprinus comatus	Shaggy mane	Cop c 1	Food Fungi	Coprinus Cop c 1	181	4538529	117
Corylus avellana	European hazelnut	Cor a 1.0103	Aero Plant	Corylus Cor a 1	1160	122684	117
Corylus avellana	European hazelnut	Cor a 1.0104	Aero Plant	Corylus Cor a 1	1160	122686	117
Corylus avellana	European hazelnut	Cor a 1.0102	Aero Plant	Corylus Cor a 1	1160	122690	117
Corylus avellana	European hazelnut	Cor a 1.0201	Aero Plant	Corylus Cor a 1	1160	1321731	117
Corylus avellana	European hazelnut	Cor a 1.0301	Aero Plant	Corylus Cor a 1	1160	1321733	117
Corylus avellana	European hazelnut	Cor a 1.0401	Food Plant	Corylus Cor a 1	1161	15726304	117
Corylus avellana	European hazelnut	Cor a 1.0402	Food Plant	Corylus Cor a 1	1161	111762102	117
Corylus avellana	European hazelnut	Cor a 1.0403	Food Plant	Corylus Cor a 1	1161	111762104	117
Corylus avellana	European hazelnut	Cor a 1.0404	Food Plant	Corylus Cor a 1	1161	111762106	117
Corylus avellana	European hazelnut	Cor a 1.0101	Food Plant	Corylus Cor a 1	1160	122688	115
Corylus avellana	European hazelnut	Cor a 11	Food Plant	Corylus Cor a 11	1448	19338630	117
Corylus avellana	European hazelnut	Cor a 12.0101	Food Plant	Corylus Cor a 12	1159	49617323	115
Corylus avellana	European hazelnut	Cor a 13.0101	Food Plant	Corylus Cor a 13	1140	29170509	117
Corylus avellana	European hazelnut	Cor a 14.0101	Food Plant	Corylus Cor a 14 2S albumin	1147	226437844	111
Corylus avellana	European hazelnut	Cor a 2.0101	Aero Plant	Corylus Cor a 2 profilins	1131	12659206	117
Corylus avellana	European hazelnut	Cor a 2.0102	Aero Plant	Corylus Cor a 2 profilins	1131	12659208	117
Corylus avellana	European hazelnut	Unassigned	Food Plant	Corylus Cor a 2 profilins	1131	1576017879	115
Corylus avellana	European hazelnut	Unassigned	Food Plant	Corylus Cor a 2 profilins	1133	1576017878	115
Corylus avellana	European hazelnut	Unassigned	Food Plant	Corylus Cor a 2 profilins	1133	1576017819	115

Corylus avellana	European hazelnut	Unassigned	Food Plant	Corylus Cor a 2 profilins	131	1576017779	115
Corylus avellana	European hazelnut	Unassigned	Food Plant	Corylus Cor a 2 profilins	133	1576017777	115
Corylus avellana	European hazelnut	Unassigned	Food Plant	Corylus Cor a 2 profilins	133	1576017776	115
Corylus avellana	European hazelnut	Unassigned	Food Plant	Corylus Cor a 2 profilins	133	1576017776	115
Corylus avellana	European hazelnut	Cor a 8	Food Plant	Corylus Cor a 8	1115	13507262	17
Corylus avellana	European hazelnut	Unassigned	Aero Plant	Corylus Cor a 9	1514	1557792009	116
Corylus avellana	European hazelnut	Cor a 9	Food Plant	Corylus Cor a 9	1515	18479082	17
Crangon crangon	Shrimp	Cra c 1.0101	Food Animal	Crangon Cra c 1 tropomyosin	1284	1238477263	112
Crangon crangon	Shrimp	Cra c 2.0101	Food Animal	Crangon Cra c 2 arginine kinase	1366	1238477265	112
Crangon crangon	Shrimp	Cra c 4.0101	Food Animal	Crangon Cra c 4 sarcoplasmic calcium-binding prote	1193	1238477327	112
Crangon crangon	Shrimp	Cra c 5.0101	Food Animal	Crangon Cra c 5 myosin light chain	1153	1238477331	112
Crangon crangon	Shrimp	Cra c 6.0101	Food Animal	Crangon Cra c 6 troponin C	1150	1238477333	112
Crangon crangon	Shrimp	Cra c 8.0101	Food Animal	Crangon Cra c 8 triosephosphate isomerase	1249	1238477329	112
Crassostrea gigas	American oyster	Unassigned	Food Animal	Crassostrea Tropomyosin	1233	15419048	17
Crassostrea gigas	American oyster	Unassigned	Food Animal	Crassostrea Tropomyosin	1284	219806594	110
Crassostrea virginica	Eastern oyster	Unassigned	Food Animal	Crassostrea Tropomyosin	1160	3468408	17
Crocus sativus	Saffron crocus	Cro s 2.0101	Aero Plant	Crocus profilin Cro s 2	1131	158700651	17
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria class IV chitinase	1281	156550550	17
Cryptomeria japonica	Japanese cedar	Cry j 1.0102	Aero Plant	Cryptomeria Cry j 1	1374	493634	18
Cryptomeria japonica	Japanese cedar	Cry j 1.0101	Aero Plant	Cryptomeria Cry j 1	1374	493632	115
Cryptomeria japonica	Japanese cedar	Cry j 1.0103	Aero Plant	Cryptomeria Cry j 1	1374	1516728	115
Cryptomeria japonica	Japanese cedar	Cry j 2	Aero Plant	Cryptomeria Cry j 2	1514	11171004	17
Cryptomeria japonica	Japanese cedar	Cry j 2	Aero Plant	Cryptomeria Cry j 2	1514	124898904	17
Cryptomeria japonica	Japanese cedar	Cry j 2	Aero Plant	Cryptomeria Cry j 2	1514	124898906	17
Cryptomeria japonica	Japanese cedar	Cry j 2	Aero Plant	Cryptomeria Cry j 2	1514	124898908	17
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	1114841607	18
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	1114841617	18
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	1114841629	18
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	1114841635	18
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	1114841641	18

Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	114841653	18
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	114841657	18
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	114841663	18
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	1514	114841665	18
Cryptomeria japonica	Japanese cedar	Cry j 2.0101	Aero Plant	Cryptomeria Cry j 2	1514	1506868	19
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Cry j 2	165	123299282	19
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria Isoflavone	306	19847822	17
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	reductase-like protein			
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria pollen	1165	291621332	112
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	allergen CJP-8			
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria pollen	1472	293329689	112
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	allergen CPA63			
Cryptomeria japonica	Japanese cedar	Unassigned	Aero Plant	Cryptomeria thaumatin	1225	139002766	18
Cucumis melo	Muskmelon	Cuc m 1	Food Plant	Cucumis Cuc m 1	1731	1807698	17
Cucumis melo	Muskmelon	Unassigned	Food Plant	Cucumis Cuc m 2	1131	31559374	17
Cucumis melo	Muskmelon	Cuc m 2	Food Plant	Cucumis Cuc m 2	1131	156263793	17
Cucumis melo var. reticulatus	Netted muskmelon	Unassigned	Food Plant	Cucumis Cuc m 2	1131	157021110	17
Cucumis melo	Muskmelon	Cuc m 3.0101	Food Plant	Cucumis Cuc m 3	141	146396595	19
Cucumis melo var. inodorus	Muskmelon	Unassigned	Food Plant	Cucumis Cuc m 3	1151	1171464770	19
Cupressus arizonica	Arizona Cypress	Cup a 1	Aero Plant	Cupressus Cup a 1/Cup s1346		16562326	17
Cupressus arizonica	Arizona Cypress	Cup a 1	Aero Plant	Cupressus Cup a 1/Cup s1367		19069497	17
Cupressus arizonica	Arizona Cypress	Unassigned	Aero Plant	Cupressus Cup a 1/Cup s1347		118197955	18
Cupressus sempervirens	Mediterranean Cypress	Cup s 1.0101	Aero Plant	Cupressus Cup a 1/Cup s1367		18101711	17
Cupressus sempervirens	Mediterranean Cypress	Cup s 1.0102	Aero Plant	Cupressus Cup a 1/Cup s1367		18101713	17
Cupressus sempervirens	Mediterranean Cypress	Cup s 1.0103	Aero Plant	Cupressus Cup a 1/Cup s1367		18101715	17
Cupressus sempervirens	Mediterranean Cypress	Cup s 1.0104	Aero Plant	Cupressus Cup a 1/Cup s1367		18101717	17
Cupressus sempervirens	Mediterranean Cypress	Cup s 1.0105	Aero Plant	Cupressus Cup a 1/Cup s1367		18101719	17
Cupressus arizonica	Arizona Cypress	Unassigned	Aero Plant	Cupressus Cup a 4	1165	1261865475	111
Cupressus arizonica	Arizona Cypress	Unassigned	Aero Plant	Cupressus Cup s 3	1199	19929163	17
Cupressus sempervirens	Mediterranean Cypress	Cup s 3.0102	Aero Plant	Cupressus Cup s 3	1225	138456228	17
Cupressus sempervirens	Mediterranean Cypress	Cup s 3.0101	Aero Plant	Cupressus Cup s 3	1225	138456226	111



Cochliobolus lunatus	Fungus	Cur 1 4. 0101	Aero Fungi	Curularia Cur 1 4	1506	193507493	115
Cynodon dactylon	Bermuda grass	Cyn d 1	Aero Plant	Cynodon Cyn d 1	125	451274	17
Cynodon dactylon	Bermuda grass	Cyn d 1	Aero Plant	Cynodon Cyn d 1	138	451275	17
Cynodon dactylon	Bermuda grass	Cyn d 1	Aero Plant	Cynodon Cyn d 1	134	691726	17
Cynodon dactylon	Bermuda grass	Cyn d 1.0204	Aero Plant	Cynodon Cyn d 1	1244	10314021	17
Cynodon dactylon	Bermuda grass	Cyn d 1.0201	Aero Plant	Cynodon Cyn d 1	1244	15384338	17
Cynodon dactylon	Bermuda grass	Cyn d 1.0202	Aero Plant	Cynodon Cyn d 1	1262	16076693	17
Cynodon dactylon	Bermuda grass	Cyn d 1	Aero Plant	Cynodon Cyn d 1	1262	16076695	17
Cynodon dactylon	Bermuda grass	Cyn d 1.0203	Aero Plant	Cynodon Cyn d 1	1262	16076697	17
Cynodon dactylon	Bermuda grass	Cyn d 1.0101	Aero Plant	Cynodon Cyn d 1	1246	7687901	110
Cynodon dactylon	Bermuda grass	Cyn d 12	Aero Plant	Cynodon Cyn d 12	1131	12154730	17
Cynodon dactylon	Bermuda grass	Unassigned	Aero Plant	Cynodon Cyn d 7	171	11247373	17
Cynodon dactylon	Bermuda grass	Unassigned	Aero Plant	Cynodon Cyn d 7	173	11247375	17
Cynodon dactylon	Bermuda grass	Cyn d 7	Aero Plant	Cynodon Cyn d 7	182	1871507	17
Cynodon dactylon	Bermuda grass	Unassigned	Aero Plant	Cynodon Group 4 like-allergen FAD-linked oxidoredu	1522	41393750	17
Cyprinus carpio	Carp	Cyp c 1.0101	Food Animal	Cyprinus Cyp c 1	1109	11797825	17
Cyprinus carpio	Carp	Cyp c 1.0201	Food Animal	Cyprinus Cyp c 1	1109	11797827	17
Dactylis glomerata	Orchard grass	Dac g 1	Aero Plant	Dactylis Dac g 1	1264	18093991	17
Dactylis glomerata	Orchard grass	Dac g 1.0101	Aero Plant	Dactylis Dac g 1	1240	33149333	17
Dactylis glomerata	Orchard grass	Dac g 2	Aero Plant	Dactylis Dac g 2	1196	1093120	17
Dactylis glomerata	Orchard grass	Dac g 2	Aero Plant	Dactylis Dac g 2	1122	44007040	17
Dactylis glomerata	Orchard grass	Dac g 3	Aero Plant	Dactylis Dac g 3	196	1825459	17
Dactylis glomerata	Orchard grass	Dac g 4.0101	Aero Plant	Dactylis Dac g 4	155	132363463	19
Dactylis glomerata	Orchard grass	Dac g 5	Aero Plant	Dactylis Dac g 5	1290	114423124	17
Dactylis glomerata	Orchard grass	Dac g 5	Aero Plant	Dactylis Dac g 5	1265	18093971	17
Daucus carota	Carrot	Unassigned	Food Plant	Daucus cyclophilin	1171	1373939374	113
Daucus carota	Carrot	Dau c 1.0101	Food Plant	Daucus Dau c 1	1168	11335877	17
Daucus carota	Carrot	Dau c 1.0102	Food Plant	Daucus Dau c 1	1154	11663522	17
Daucus carota	Carrot	Dau c 1.0103	Food Plant	Daucus Dau c 1	1154	12154732	17
Daucus carota	Carrot	Dau c 1.0104	Food Plant	Daucus Dau c 1	1154	12154734	17
Daucus carota	Carrot	Dau c 1.0105	Food Plant	Daucus Dau c 1	1154	12154736	17
Daucus carota	Carrot	Dau c 1.0201	Food Plant	Daucus Dau c 1	1154	118652047	17
Daucus carota	Carrot	Unassigned	Food Plant	Daucus Dau c 1	1154	119912791	17

Daucus carota	Carrot		Dau c 1.0301	Food Plant	Daucus Dau c 1	154	302379147	12
Daucus carota	Carrot		Unassigned	Food Plant	Daucus Dau c 1	154	302379149	12
Daucus carota	Carrot		Unassigned	Food Plant	Daucus Dau c 1	154	302379151	12
Daucus carota	Carrot		Unassigned	Food Plant	Daucus Dau c 1	154	302379153	12
Daucus carota	Carrot		Unassigned	Food Plant	Daucus Dau c 1	154	302379155	12
Daucus carota	Carrot		Unassigned	Food Plant	Daucus Dau c 1	154	302379157	12
Daucus carota	Carrot		Unassigned	Food Plant	Daucus Dau c 1	154	302379159	12
Daucus carota	Carrot		Dau c 4	Food Plant	Daucus Dau c 4	134	18652049	7
Daucus carota	Carrot		Dau c 5.0101	Food Plant	Daucus Dau c 5	306	373939378	13
Daucus carota	Carrot		Unassigned	Food Plant	Isoflavone reductase			
Dermatophagoides farinae	House dust mite		Der f 13.0101	Aero Mite	Dermatophagoides Der f 131	131	37958167	11
Dermatophagoides farinae	House dust mite		Der f 15	Aero Mite	Dermatophagoides Der f 155	155	5815436	7
Dermatophagoides pteronyssinus	House dust mite		Der p 15.0101	Aero Mite	Dermatophagoides Der f 1532	1532	67975089	7
Dermatophagoides pteronyssinus	House dust mite		Der p 15.0102	Aero Mite	Dermatophagoides Der f 1568	1568	78128018	7
Dermatophagoides farinae	House dust mite		Der f 16	Aero Mite	Dermatophagoides Der f 1480	1480	121591547	7
Dermatophagoides farinae	House dust mite		Unassigned	Aero Mite	Dermatophagoides Der f 356	356	37786884	8
Dermatophagoides pteronyssinus	House dust mite		Der f 20.0201	Aero Mite	Dermatophagoides Der f 356	356	156938897	9
Dermatophagoides pteronyssinus	House dust mite		Der p 20.0101	Aero Mite	Dermatophagoides Der f 356	356	685432792	15
Dermatophagoides farinae	House dust mite		Der f 24.0101	Aero Mite	Dermatophagoides Der f 118	118	477541860	14
Dermatophagoides pteronyssinus	House dust mite		Der p 24.0101	Aero Mite	Dermatophagoides Der f 118	118	1922664427	16
Dermatophagoides farinae	House dust mite		Der f 25.0101	Aero Mite	Dermatophagoides Der f 1247	1247	442565872	14
Dermatophagoides farinae	House dust mite		Der f 25.0201	Aero Mite	Dermatophagoides Der f 1247	1247	685432812	15
Dermatophagoides farinae	House dust mite		Unassigned	Aero Mite	Dermatophagoides Der f 1427	1427	37958175	8
Dermatophagoides farinae	House dust mite		Der f 27.0101	Aero Mite	Dermatophagoides Der f 1427	1427	685432794	15
Dermatophagoides farinae	House dust mite		Der f 28.0101	Aero Mite	Dermatophagoides Der f 1659	1659	442565876	14

Dermatophagoides farinae	House dust mite	Der f 28.0201	Aero Mite	Dermatophagoides Der f 1654	28	685432788	115
Dermatophagoides farinae	House dust mite	Der f 29.0101	Aero Mite	Dermatophagoides Der f 164	29	37958141	8
Dermatophagoides farinae	House dust mite	Der f 30.0101	Aero Mite	Dermatophagoides Der f 171	30	442565878	114
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der f 180	30	15072346	17
Dermatophagoides farinae	House dust mite	Der f 1	Aero Mite	Dermatophagoides Der p 321	1 Der f 1 Der m 1	730035	17
Dermatophagoides farinae	House dust mite	Der f 1.0101	Aero Mite	Dermatophagoides Der p 321	1 Der f 1 Der m 1	27530349	17
Dermatophagoides farinae	House dust mite	Der f 1.0102	Aero Mite	Dermatophagoides Der p 276	1 Der f 1 Der m 1	76097507	17
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 321	1 Der f 1 Der m 1	156106765	9
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 263	1 Der f 1 Der m 1	37958161	112
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 305	1 Der f 1 Der m 1	387178006	113
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 303	1 Der f 1 Der m 1	3063387429	115
Dermatophagoides farinae	House dust mite	Der f 1.0108	Aero Mite	Dermatophagoides Der p 321	1 Der f 1 Der m 1	119633260	115
Dermatophagoides farinae	House dust mite	Der f 1.0109	Aero Mite	Dermatophagoides Der p 321	1 Der f 1 Der m 1	119633262	115
Dermatophagoides farinae	House dust mite	Der f 1.0110	Aero Mite	Dermatophagoides Der p 321	1 Der f 1 Der m 1	119633264	115
Dermatophagoides pteronyssinus	House dust mite	Der m 1.0101	Aero Mite	Dermatophagoides Der p 30	1 Der f 1 Der m 1	127205	17
Dermatophagoides pteronyssinus	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725560	17
Dermatophagoides pteronyssinus	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725562	17
Dermatophagoides pteronyssinus	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725564	17
Dermatophagoides pteronyssinus	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725566	17
Dermatophagoides pteronyssinus	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725568	17
Dermatophagoides pteronyssinus	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725570	17
Dermatophagoides pteronyssinus	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725572	17
Dermatophagoides pteronyssinus	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725574	17
Dermatophagoides	House dust mite	Der p 1	Aero Mite	Dermatophagoides Der p 222	1 Der f 1 Der m 1	21725576	17



Dermatophagoides pteronyssinus	House dust mite	Der p 14.0101	Aero Mite	Dermatophagoides Der p 1662 14 / Der f 14	20385544	17
Dermatophagoides farinae	House dust mite	Der f 2.0102	Aero Mite	Dermatophagoides Der p 146 2 / Der f 2	217306	17
Dermatophagoides farinae	House dust mite	Der f 2.0103	Aero Mite	Dermatophagoides Der p 138 2 / Der f 2	217308	17
Dermatophagoides farinae	House dust mite	Der f 2.0105	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	17978844	17
Dermatophagoides farinae	House dust mite	Der f 2.0108	Aero Mite	Dermatophagoides Der p 146 2 / Der f 2	55859470	17
Dermatophagoides farinae	House dust mite	Der f 2.0107	Aero Mite	Dermatophagoides Der p 146 2 / Der f 2	55859468	17
Dermatophagoides farinae	House dust mite	Der f 2.0106	Aero Mite	Dermatophagoides Der p 146 2 / Der f 2	55859466	17
Dermatophagoides farinae	House dust mite	Der f 2.0109	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	76097511	17
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 146 2 / Der f 2	256631558	11
Dermatophagoides farinae	House dust mite	Der f 2.0112	Aero Mite	Dermatophagoides Der p 140 2 / Der f 2	37958157	12
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	387178018	13
Dermatophagoides farinae	House dust mite	Der f 2.0101	Aero Mite	Dermatophagoides Der p 138 2 / Der f 2	217304	15
Dermatophagoides farinae	House dust mite	Der f 2.0116	Aero Mite	Dermatophagoides Der p 146 2 / Der f 2	124696217	15
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21465915	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725582	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725584	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725586	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725588	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725590	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725592	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725594	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725596	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725600	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	21725602	17

Dermatophagoides pteronyssinus	House dust mite	Der p 2	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	12172604	17
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	76097509	17
Dermatophagoides pteronyssinus	House dust mite	Der p 2.0114	Aero Mite	Dermatophagoides Der p 146 2 / Der f 2	98644635	17
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 130 2 / Der f 2	110560872	19
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	157829757	19
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 145 2 / Der f 2	164415695	19
Dermatophagoides pteronyssinus	House dust mite	Der p 2.0101	Aero Mite	Dermatophagoides Der p 145 2 / Der f 2	9280643	10
Dermatophagoides pteronyssinus	House dust mite	Der p 2.0110	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	256095984	11
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	387178014	13
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 129 2 / Der f 2	387178008	13
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 146 2 / Der f 2	86450747	17
Dermatophagoides pteronyssinus	House dust mite	Der p 21.0101	Aero Mite	Dermatophagoides Der p 140 21	85687540	17
Dermatophagoides pteronyssinus	House dust mite	Der p 23.0101	Aero Mite	Dermatophagoides Der p 190 23 Peritrophin-like protein	1171466145	14
Dermatophagoides farinae	House dust mite	Der f 3	Aero Mite	Dermatophagoides Der p 232 3 / Der f 3	1314736	17
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 259 3 / Der f 3	163638970	19
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 259 3 / Der f 3	218203816	10
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 259 3 / Der f 3	218203818	10
Dermatophagoides farinae	House dust mite	Der f 3.0101	Aero Mite	Dermatophagoides Der p 259 3 / Der f 3	1311457	15
Dermatophagoides pteronyssinus	House dust mite	Der p 3	Aero Mite	Dermatophagoides Der p 261 3 / Der f 3	1511476	17
Dermatophagoides pteronyssinus	House dust mite	Der p 4	Aero Mite	Dermatophagoides Der p 496 4	15059162	17
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 19 4	1351935	17
Dermatophagoides pteronyssinus	House dust mite	Der p 5.0102	Aero Mite	Dermatophagoides Der p 132 5	1913285	17
Dermatophagoides pteronyssinus	House dust mite	Der p 5	Aero Mite	Dermatophagoides Der p 132 5	28798085	17

Dermatophagoides pteronyssinus	House dust mite	Der p 5.0101	Aero Mite	Dermatophagoides Der p 148	5	19072	115
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 120	6 / Der f 6	1404371	17
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 279	6 / Der f 6	218203826	110
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 279	6 / Der f 6	218203828	110
Dermatophagoides farinae	House dust mite	Der f 6.0101	Aero Mite	Dermatophagoides Der p 279	6 / Der f 6	6808630	111
Dermatophagoides pteronyssinus	House dust mite	Der p 6	Aero Mite	Dermatophagoides Der p 120	6 / Der f 6	1352239	17
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 1213	7 / Der f 7	37958165	18
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 1213	7 / Der f 7	218203832	110
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 1213	7 / Der f 7	685432798	116
Dermatophagoides pteronyssinus	House dust mite	Der p 7.0101	Aero Mite	Dermatophagoides Der p 1215	7 / Der f 7	1045602	17
Dermatophagoides pteronyssinus	House dust mite	Der p 7	Aero Mite	Dermatophagoides Der p 1215	7 / Der f 7	10189811	17
Dermatophagoides pteronyssinus	House dust mite	Der f 7.0101	Aero Mite	Dermatophagoides Der p 1213	7 / Der f 7	1311689	110
Dermatophagoides pteronyssinus	House dust mite	Der p 8.0101	Aero Mite	Dermatophagoides Der p 1219	8	807138	17
Dermatophagoides pteronyssinus	House dust mite	Unassigned	Aero Mite	Dermatophagoides Der p 1219	8	60920878	17
Dermatophagoides farinae	House dust mite	Der f 18	Aero Mite	Dermatophagoides farinae Der f 18 Der p 1462	18	27550039	17
Dermatophagoides pteronyssinus	House dust mite	Der p 18.0101	Aero Mite	Dermatophagoides farinae Der f 18 Der p 1462	18	167975085	17
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides farinae Der f 21 Chew	136	140089314	19
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides farinae Der f 21 Chew	136	140089316	19
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides farinae Der f 21 Chew	136	140089320	19
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides farinae Der f 21 Chew	136	140089322	19
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides farinae Der f 21 Chew	136	140089324	19
Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides farinae Der f 21 Chew	136	140089326	19
Dermatophagoides farinae	House dust mite	Der f 21.0101	Aero Mite	Dermatophagoides farinae Der f 21 Chew	136	1567768173	115

Dermatophagoides farinae	House dust mite	Unassigned	Aero Mite	Dermatophagoides Profilin	130	1685432824	116
Dolichovespula maculata	Whiteface hornet	Dol m 1.02	Venom or Salivary	Dolichovespula Dol m 1 Phospholipase A1B	1303	1709642	17
Dolichovespula maculata	Whiteface hornet	Dol m 1.0101	Venom or Salivary	Dolichovespula Dol m 1 Phospholipase A1B	1317	288917	18
Dolichovespula maculata	Whiteface hornet	Dol m 2.0101	Venom or Salivary	Dolichovespula Dol m 2 Hyaluronidase	1331	1511604	111
Dolichovespula arenaria	Yellow jacket	Dol a 5.0101	Venom or Salivary	Dolichovespula Venom allergen 5	1203	156719	111
Dolichovespula maculata	Whiteface hornet	Dol m 5.0101	Venom or Salivary	Dolichovespula Venom allergen 5	1227	156715	111
Dolichovespula maculata	Whiteface hornet	Dol m 5.02	Venom or Salivary	Dolichovespula Venom allergen 5	1212	1552080	111
Epicroccum nigrum	Fungus	Epi p 1.0101	Aero Fungi	Epicroccum Epi p 1	118	124636820	19
Equus caballus	Horse	Equ c 1.0101	Aero Animal	Equus Equ c 1	1187	11575778	111
Equus caballus	Horse	Equ c 2.0101	Aero Animal	Equus Equ c 2	129	13121755	17
Equus caballus	Horse	Equ c 2.0102	Aero Animal	Equus Equ c 2	119	13121756	17
Equus caballus	Horse	Equ c 3.0101	Aero Animal	Equus Equ c 3	1607	1399672	17
Equus caballus	Horse	Equ c 4.0101	Aero Animal	Equus Equ c 4 and Equ c 15	1228	1126514234	18
Erimacrus isenbeckii	Horsehair crab	Unassigned	Food Animal	Erimacrus tropomyosin	1284	1125995169	18
Erimacrus isenbeckii	Horsehair crab	Unassigned	Food Animal	Erimacrus tropomyosin	1284	1125995171	18
Eriocheir sinensis	Chinese mitten crab	Unassigned	Food Animal	Eriocheir tropomyosin	1284	1134305330	18
Euphausia pacifica	North Pacific Krill	Unassigned	Food Animal	Euphausia	1284	1156712754	19
Euphausia superba	Krill	Unassigned	Food Animal	Euphausia	1284	1156712752	19
Euroglyphus maynei	House dust mite	Eur m 1.0101	Aero Mite	Euroglyphus Eur m 1	1321	13941388	17
Euroglyphus maynei	House dust mite	Unassigned	Aero Mite	Euroglyphus Eur m 1	1327	13941390	17
Euroglyphus maynei	House dust mite	Eur m 2.0102	Aero Mite	Euroglyphus Eur m 2	1135	13941386	17
Erynnis japonica	Crimson seabream	Unassigned	Food Animal	Erynnis parvalbumin	1109	1327342663	112
Erynnis japonica	Crimson seabream	Unassigned	Food Animal	Erynnis parvalbumin	1108	1327342661	112
Fagopyrum esculentum	Buckwheat	Unassigned	Food Plant	Fagopyrum BW 8 kDa protein	1133	117907758	17
Fagopyrum tataricum	Buckwheat	Unassigned	Food Plant	Fagopyrum BW 8 kDa protein	1133	1144228127	18
Fagopyrum esculentum	Buckwheat	Unassigned	Food Plant	Fagopyrum esculentum 13S globulins IgE binding	1453	1584592120	115
Fagopyrum esculentum	Buckwheat	Unassigned	Food Plant	Fagopyrum esculentum 13S globulins IgE binding	1453	1584592116	115
Fagopyrum esculentum	Buckwheat	Unassigned	Food Plant	Fagopyrum Fag e 2 Fag t127		161970231	17







Gallus gallus	Chicken	Unassigned	Food Animal	Gallus Gal d 1	1210	209979542	110
Gallus gallus	Chicken	Gal d 2	Food Animal	Gallus Gal d 2	1155	63052	17
Gallus gallus	Chicken	Gal d 2.0101	Food Animal	Gallus Gal d 2	386	129293	17
Gallus gallus	Chicken	Gal d 2	Food Animal	Gallus Gal d 2	386	808969	17
Gallus gallus	Chicken	Gal d 2	Food Animal	Gallus Gal d 2	385	15826578	17
Gallus gallus	Chicken	Unassigned	Food Animal	Gallus Gal d 2	385	34811333	17
Gallus gallus	Chicken	Gal d 3.0101	Food Animal	Gallus Gal d 3	705	757851	17
Gallus gallus	Chicken	Gal d 3	Food Animal	Gallus Gal d 3	705	1351295	17
Gallus gallus	Chicken	Gal d 4	Food Animal	Gallus Gal d 4	147	126608	17
Gallus gallus	Chicken	Gal d 4	Food Animal	Gallus Gal d 4	124	212279	17
Gallus gallus	Chicken	Gal d 4.0101	Food Animal	Gallus Gal d 4	147	63581	115
Gallus gallus	Chicken	Gal d 5	Food Animal	Gallus Gal d 5	1615	63748	17
Gallus gallus	Chicken	Unassigned	Food Animal	Gallus Gal d 6 YGP42	1284	13	114
Gallus gallus	Chicken	Unassigned	Food Animal	Gallus gallus Gal d 7	1192	55584149	116
Gallus gallus	Chicken	Unassigned	Food Animal	Gallus parvalbumin	1110	225877920	110
Glossina morsitans	Tsetse fly	Unassigned	Venom or Salivary	Glossina Glo m 5	258	289740263	111
Glossina morsitans	Tsetse fly	Unassigned	Venom or Salivary	Glossina Glo m 5	259	289742475	111
Glossina morsitans	Tsetse fly	Unassigned	Venom or Salivary	Glossina Glo m 5	222	289742483	111
Glossina morsitans	Tsetse fly	Glo m 5.0101	Venom or Salivary	Glossina Glo m 5	259	8927462	111
Glycine max	Soybean	Gly m 1.0101	Food Plant	Glycine Gly m 1	180	123506	112
Glycine max	Soybean	Gly m 3.0102	Food Plant	Glycine Gly m 3	131	3021373	17
Glycine max	Soybean	Unassigned	Food Plant	Glycine Gly m 3	131	156938901	19
Glycine max	Soybean	Gly m 3.0101	Food Plant	Glycine Gly m 3	131	3021375	115
Glycine max	Soybean	Gly m 4	Food Plant	Glycine Gly m 4	158	18744	17
Glycine max	Soybean	Unassigned	Food Plant	Glycine Gly m 5.0101 alpha subunit beta congl	1605	18536	17
Glycine max	Soybean	Unassigned	Food Plant	Glycine Gly m 5.0101 alpha subunit beta congl	218	169927	17
Glycine max	Soybean	Gly m 5.0101	Food Plant	Glycine Gly m 5.0101 alpha subunit beta congl	1543	19867357	115
Glycine max	Soybean	Unassigned	Food Plant	Glycine Gly m 5.0201 alpha prime beta congl	1639	169929	17
Glycine max	Soybean	Unassigned	Food Plant	Glycine Gly m 5.0201 alpha prime beta congl	1621	15425631	115
Glycine max	Soybean	Gly m 5.0201	Food Plant	Glycine Gly m 5.0201	1569	19867361	115



Glycine max	Soybean	Unassigned	Food Plant	Glycine Trypsin inhibitor	204	256636	7
Glycine max	Soybean	Unassigned	Food Plant	Glycine Trypsin inhibitor	208	510515	7
Glycophagus domesticus	Storage mite	Gly d 2.0101	Aero Mite	Glycophagus Gly d 2	128	6179520	7
Glycophagus domesticus	Storage mite	Gly d 2.0201	Aero Mite	Glycophagus Gly d 2	125	7160811	7
Glycophagus domesticus	Storage mite	Unassigned	Aero Mite	Glycophagus Gly d 2	141	33772588	7
Halictis discus	Disk abalone	Unassigned	Food Animal	Halictis Hal m 1 tropomyosin	284	219806586	10
Halictis diversicolor	Abalone	Unassigned	Food Animal	Halictis Hal m 1 tropomyosin	284	9954249	7
Halictis discus	Disk abalone	Unassigned	Food Animal	Halictis paramyosin	860	318609972	12
Helianthus annuus	Sunflower	Hel a 2	Aero Plant	Helianthus Hel a 2	133	3581965	7
Helianthus annuus	Sunflower	Hel a 3.0101	Food Plant	Helianthus Hel a 3	116	31324341	15
Helianthus annuus	Sunflower	Unassigned	Food Plant	Helianthus Seed 2S albumin	141	112745	9
Helix aspersa	Brown garden snail	Hel as 1.0101	Food Animal	Helix Hel as 1 tropomyosin	284	4468224	7
Hevea brasiliensis	Para rubber tree	Hev b 1.0101	Contact	Heves Hev b 1	138	18839	15
Hevea brasiliensis	Para rubber tree	Hev b 10.0101	Contact	Heves Hev b 10	233	348137	7
Hevea brasiliensis	Para rubber tree	Hev b 10.0102	Contact	Heves Hev b 10	205	5777414	7
Hevea brasiliensis	Para rubber tree	Hev b 10.0103	Contact	Heves Hev b 10	205	10862818	7
Hevea brasiliensis	Para rubber tree	Hev b 11.0101	Contact	Heves Hev b 11	295	14575525	7
Hevea brasiliensis subsp. brasiliensis	Para rubber tree	Hev b 11.0102	Contact	Heves Hev b 11	295	27526732	7
Hevea brasiliensis	Para rubber tree	Hev b 12	Contact	Heves Hev b 12	116	20135538	7
Hevea brasiliensis	Para rubber tree	Hev b 13	Contact	Heves Hev b 13	391	30909057	7
Hevea brasiliensis	Para rubber tree	Hev b 14.0101	Contact	Heves Hev b 14 heveamine	208	313870630	12
Hevea brasiliensis	Para rubber tree	Hev b 15.0101	Contact	Heves Hev b 15	70	571257122	15
Hevea brasiliensis	Para rubber tree	Hev b 2.0101	Contact	Heves Hev b 2	374	1184668	7
Hevea brasiliensis	Para rubber tree	Hev b 2	Contact	Heves Hev b 2	374	32765543	7
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Heves Hev b 2	374	124294783	8
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Heves Hev b 2	374	124294785	8
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Heves Hev b 2	374	124365249	8
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Heves Hev b 2	374	124365251	8
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Heves Hev b 2	374	124365253	8
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Heves Hev b 2	374	268037674	11

Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 2	374	270315180	111
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 2	373	359359690	113
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 2	374	387778882	113
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 2	374	387778880	113
Hevea brasiliensis	Para rubber tree	Hev b 3.0101	Contact	Hevea Hev b 3	1204	3818475	111
Hevea brasiliensis	Para rubber tree	Hev b 4.0101	Contact	Hevea Hev b 4	366	46410859	17
Hevea brasiliensis	Para rubber tree	Hev b 5	Contact	Hevea Hev b 5	151	1480457	17
Hevea brasiliensis	Para rubber tree	Hev b 6	Contact	Hevea Hev b 6	187	2832430	17
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 6	143	73535415	17
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 6	1204	158342650	19
Hevea brasiliensis	Para rubber tree	Hev b 7.01	Contact	Hevea Hev b 7	388	1916805	17
Hevea brasiliensis	Para rubber tree	Hev b 7.02	Contact	Hevea Hev b 7	388	13087805	17
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 7	388	13288200	17
Hevea brasiliensis	Para rubber tree	Hev b 7	Contact	Hevea Hev b 7	388	16707018	17
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 7	387	41581137	17
Hevea brasiliensis	Para rubber tree	Hev b 8.0203	Aero Mite	Hevea Hev b 8	131	6979171	111
Hevea brasiliensis	Para rubber tree	Hev b 8.0101	Contact	Hevea Hev b 8	131	3183706	17
Hevea brasiliensis	Para rubber tree	Hev b 8	Contact	Hevea Hev b 8	131	11513601	17
Hevea brasiliensis	Para rubber tree	Hev b 8.0201	Contact	Hevea Hev b 8	131	6979167	111
Hevea brasiliensis	Para rubber tree	Hev b 8.0202	Contact	Hevea Hev b 8	131	6979169	111
Hevea brasiliensis	Para rubber tree	Hev b 8.0102	Contact	Hevea Hev b 8	131	15689740	115
Hevea brasiliensis	Para rubber tree	Hev b 8.0204	Contact	Hevea Hev b 8	131	8919948	115
Hevea brasiliensis	Para rubber tree	Hev b 9	Contact	Hevea Hev b 9	1445	9981744	17
Hevea brasiliensis	Para rubber tree	Unassigned	Contact	Hevea Hev b 9	1445	11423687	19
Holcus lanatus	Velvet grass	Hol 1.0101	Aero Plant	Holcus Hol 1.1	265	1414703	17
Holcus lanatus	Velvet grass	Hol 1.0102	Aero Plant	Holcus Hol 1.1	248	1167836	17
Holcus lanatus	Velvet grass	Unassigned	Aero Plant	Holcus Hol 1.1	263	13860384	17
Holcus lanatus	Velvet grass	Unassigned	Aero Plant	Holcus Hol 1.5	120	75140046	17
Holcus lanatus	Velvet grass	Hol 1.5.0201	Aero Plant	Holcus Hol 1.5	240	12266623	17
Holcus lanatus	Velvet grass	Hol 1.5.0101	Aero Plant	Holcus Hol 1.5	264	12266625	17
Holcus lanatus	Velvet grass	Unassigned	Aero Plant	Holcus Hol 1.5	296	11991229	17
Homarus americanus	American lobster	Hon a.1.0102	Food Animal	Homarus Hon a.1	284	12660868	17
Homarus americanus	American lobster	Hon a.1.0101	Food Animal	Homarus Hon a.1	284	12660866	115
Hordeum vulgare subsp. vulgare	Barley	Unassigned	Aero Plant	Hordeum Alpha-amylase inhibitor BDAT-1	152	13367714	17
Hordeum vulgare subsp. vulgare	Barley	Unassigned	Aero Plant	Hordeum Alpha-amylase inhibitor component Csa	144	118955	17

Hordeum vulgare subsp. vulgare	Barley	Unassigned	Aero Plant	Hordeum Alpha-amylase inhibitor component Caa	145	439275	17
Hordeum vulgare	Barley	Unassigned	Aero Plant	Hordeum Alpha-amylase inhibitor component Cba	149	585290	17
Hordeum vulgare subsp. vulgare	Barley	Hor v 15.0101	Food Plant	Hordeum Hor v 15	146	19003	115
Hordeum vulgare	Barley	Unassigned	Food Plant	Hordeum Hor v 20	1289	1708280	115
Hordeum vulgare	Barley	Hor v 20.0101	Food Plant	Hordeum Hor v 20	1286	288709	115
Hordeum vulgare	Barley	Unassigned	Aero Plant	Hordeum Lrp 1	1117	167077	17
Hordeum vulgare	Barley	Unassigned	Food Plant	Hordeum Lrp 1	1134	19039	17
Hordeum vulgare	Barley	Unassigned	Aero Plant	Hordeum Typsin inhibitor CHe	144	1405736	17
Hordeum vulgare subsp. vulgare	Barley	Unassigned	Aero Plant	Hordeum Typsin inhibitor CHe	148	19009	17
Humulus japonicus	Japanese hop	Hum j 1	Aero Plant	Humulus Humj1	1155	13313263	17
Humulus scandens	Japanese hop	Unassigned	Aero Plant	Humulus profilin-like protein	131	34851176	17
Humulus scandens	Japanese hop	Unassigned	Aero Plant	Humulus profilin-like protein	131	34851174	17
Juglans nigra	Black walnut	Jug n 1.0101	Food Plant	Juglans Jug r 1 Jug n 1161	13121942	31321942	17
Juglans regia	English walnut	Jug r 1.0101	Food Plant	Juglans Jug r 1 Jug n 1139	11794252	1794252	17
Juglans nigra	Black walnut	Jug n 2.0101	Food Plant	Juglans Jug r 2	1481	31321944	17
Juglans regia	English walnut	Jug r 2.0101	Food Plant	Juglans Jug r 2	1593	6580762	17
Juglans regia	English walnut	Unassigned	Food Plant	Juglans Jug r 3	1119	20948145	111
Juglans regia	English walnut	Jug r 4.0101	Food Plant	Juglans Jug r 4 seed storage protein	1507	156788031	17
Juniperus ashei	Mountain cedar	Jun a 2	Aero Plant	Juniperus Jun a 2	1507	9955725	17
Juniperus ashei	Mountain cedar	Jun a 3.0101	Aero Plant	Juniperus Jun a 3	225	9087177	18
Juniperus rigida	Cedar	Unassigned	Aero Plant	Juniperus Jun a 3	225	38456224	17
Juniperus rigida	Cedar	Unassigned	Aero Plant	Juniperus Jun a 3	225	38456222	17
Juniperus virginiana	Red cedar	Unassigned	Aero Plant	Juniperus Jun a 3	1110	151316532	17
Juniperus ashei	Mountain cedar	Jun a 1.010101	Aero Plant	Juniperus Jun a/v 1	1367	14138877	17
Juniperus oxycedrus	Juniper	Unassigned	Aero Plant	Juniperus Jun a/v 1	1367	115139849	17
Juniperus virginiana	Red cedar	Jun v 1.0102	Aero Plant	Juniperus Jun a/v 1	1367	8843917	17
Juniperus virginiana	Red cedar	Jun v 1.0101	Aero Plant	Juniperus Jun a/v 1	1367	8843921	17
Juniperus oxycedrus	Juniper	Jun o 4	Aero Plant	Juniperus Jun o 4	1165	15391446	17
Lates calcarifer	Asian Seabass	Lat c 1.0101	Food Animal	Lates Lat c 1	1109	15653743	115
Lates calcarifer	Asian Seabass	Lat c 1.0201	Food Animal	Lates Lat c 1	1109	148526356	115
Lens culinaris	Lentil	Len c 3.0101	Food Plant	Lens Len c 3	1118	160735410	115

Lens culinaris	Lentil	Len c 1.0101	Food Plant	Len Len c 1	418	28639109	17
Lens culinaris	Lentil	Len c 1.0102	Food Plant	Len Len c 1	415	28639111	17
Lepidoglyphus destructor	Storage mite	Lep d 10.0101	Aero Mite	Lepidoglyphus Lep d 10	284	6900304	115
Lepidoglyphus destructor	Storage mite	Lep d 13.0101	Aero Mite	Lepidoglyphus Lep d 13	131	6523380	115
Lepidoglyphus destructor	Storage mite	Lep d 2.0102	Aero Mite	Lepidoglyphus Lep d 2	141	21213898	17
Lepidoglyphus destructor	Storage mite	Lep d 2.0202	Aero Mite	Lepidoglyphus Lep d 2	141	21213900	17
Lepidoglyphus destructor	Storage mite	Lep d 2	Aero Mite	Lepidoglyphus Lep d 2	141	1882223	17
Lepidoglyphus destructor	Storage mite	Lep d 2	Aero Mite	Lepidoglyphus Lep d 2	141	1882222	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Lepidoglyphus Lep d 2	141	34495274	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Lepidoglyphus Lep d 2	141	34495278	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Lepidoglyphus Lep d 2	140	34495280	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Lepidoglyphus Lep d 2	141	34495282	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Lepidoglyphus Lep d 2	141	34495284	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Lepidoglyphus Lep d 2	141	34495286	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Lepidoglyphus Lep d 2	141	34495288	17
Lepidoglyphus destructor	Storage mite	Unassigned	Aero Mite	Lepidoglyphus Lep d 2	141	34495290	17
Lepidoglyphus destructor	Storage mite	Lep d 2.0101	Aero Mite	Lepidoglyphus Lep d 2	98	1587450	115
Lepidoglyphus destructor	Storage mite	Lep d 2.0201	Aero Mite	Lepidoglyphus Lep d 2	141	999458	115
Lepidoglyphus destructor	Storage mite	Lep d 5.0102	Aero Mite	Lepidoglyphus Lep d 5	171	34495292	17
Lepidoglyphus destructor	Storage mite	Lep d 5.0103	Aero Mite	Lepidoglyphus Lep d 5	169	34495294	17
Lepidoglyphus destructor	Storage mite	Lep d 5.0101	Aero Mite	Lepidoglyphus Lep d 5	110	6523378	115
Lepidoglyphus destructor	Storage mite	Lep d 7.0101	Aero Mite	Lepidoglyphus Lep d 7	216	16706282	115
Lepidorthobus whiffiaonis	Flat fish	Lep w 1.0101	Food Animal	Lepidorthobus Lep w 1 parvalbumin	109	1208608078	110
Lepima saccharina	Silverfish	Lep s 1	Aero Insect	Lepima Troponyosin	284	120387027	17
Lepima saccharina	Silverfish	Unassigned	Aero Insect	Lepima Troponyosin	243	120387029	17



Ligustrum vulgare	Privet	Lig v 1.0101	Aero Plant	Ligustrum Lig v 1	145	3256210	7
Ligustrum vulgare	Privet	Lig v 1.0102	Aero Plant	Ligustrum Lig v 1	145	3256212	7
Lilium longiflorum	Trumpet lily	Unassigned	Aero Plant	Lilium polygalacturonase	413	73913442	8
Litchi chinensis	Lychee nut	Lit c 1	Food Plant	Litchi Lit c 1	131	15809696	7
Litchi chinensis	Lychee nut	Unassigned	Food Plant	Litchi Lit c 1	131	83317152	7
Litopenaeus vannamei	Whiteleg Shrimp	Lit v 4.0101	Food Animal	Litopenaeus Lit v 4  sarcoplasmic Ca+  binding	193	223403273	11
Litopenaeus vannamei	Whiteleg Shrimp	Lit v 1.0101	Food Animal	Litopenaeus Lit v 1	284	170791252	10
Litopenaeus vannamei	Whiteleg Shrimp	Lit v 2.0101	Food Animal	Litopenaeus Lit v 2	356	115492980	8
Litopenaeus vannamei	Whiteleg Shrimp	Lit v 3.0101	Food Animal	Litopenaeus Lit v 3  myosin	177	184198734	10
Lolium perenne	Perennial ryegrass	Lol p 1	Aero Plant	Lolium Lol p 1	263	126385	7
Lolium perenne	Perennial ryegrass	Lol p 1.0102	Aero Plant	Lolium Lol p 1	252	168314	7
Lolium perenne	Perennial ryegrass	Lol p 1.0101	Aero Plant	Lolium Lol p 1	263	168316	10
Lolium perenne	Perennial ryegrass	Lol p 1.0103	Aero Plant	Lolium Lol p 1	263	6599300	10
Lolium perenne	Perennial ryegrass	Lol p 11	Aero Plant	Lolium Lol p 11	134	47606808	7
Lolium perenne	Perennial ryegrass	Lol p 2.0101	Aero Plant	Lolium Lol p 2	97	126386	7
Lolium perenne	Perennial ryegrass	Lol p 2	Aero Plant	Lolium Lol p 2	88	939932	7
Lolium perenne	Perennial ryegrass	Lol p 3	Aero Plant	Lolium Lol p 3	97	126387	7
Lolium perenne	Perennial ryegrass	Lol p 4.0101	Aero Plant	Lolium Lol p 4	423	55859464	7
Lolium perenne	Perennial ryegrass	Lol p 5	Aero Plant	Lolium Lol p 5	301	4416516	7
Lolium perenne	Perennial ryegrass	Lol p 5	Aero Plant	Lolium Lol p 5	301	663467	7
Lolium perenne	Perennial ryegrass	Lol p 5.0101	Aero Plant	Lolium Lol p 5	339	455288	10
Lolium perenne	Perennial ryegrass	Lol p 5.0102	Aero Plant	Lolium Lol p 5	307	332278195	12
Lupinus albus	white lupine	Unassigned	Food Plant	Lupinus albus congluten   beta	531	89994190	14
Lupinus angustifolius	blue lupin	Unassigned	Food Plant	Lupinus Lup an 1  conglutin beta	521	149208401	9
Lupinus angustifolius	blue lupin	Unassigned	Food Plant	Lupinus Lup an 1  conglutin beta	455	149208403	9
Lupinus angustifolius	blue lupin	Lup an 1.0101	Food Plant	Lupinus Lup an 1  conglutin beta	611	169950662	10
Lycium barbarum	wolfberry	Unassigned	Food Plant	Lycium ltp	151	363805423	13
Macrobrachium rosenbergii	Giant River Prawn	Mac r 1.0101	Food Animal	Macrobrachium rosenbergii shrimp  tropomyosin	284	288819271	11
Macrobrachium rosenbergii	Giant River Prawn	Unassigned	Food Animal	Macrobrachium rosenbergii shrimp  tropomyosin	284	555698675	15

Malassezia furfur	Yeast		Mal a f 2	Contact	Malassezia Mala f 2	177	3445490	17
Malassezia furfur	Yeast		Mal a f 3	Contact	Malassezia Mala f 3	166	3445492	17
Malassezia furfur	Yeast		Mal a f 4	Contact	Malassezia Mala f 4	342	4587985	17
Malassezia symphodialis	Yeast		Mal a s 1	Contact	Malassezia Mala s 1	350	1261972	17
Malassezia symphodialis	Yeast		Mal a s 10.0101	Contact	Malassezia Mala s 10 heat shock protein	773	28564467	14
Malassezia symphodialis ATCC 42132	Yeast		Unassigned	Contact	Malassezia Mala s 10 heat shock protein	773	465797105	14
Malassezia symphodialis	Yeast		Mal a s 11	Contact	Malassezia Mala s 11 first 38 aa signal	1237	28569698	17
Malassezia symphodialis ATCC 42132	Yeast		Unassigned	Contact	Malassezia Mala s 11 first 38 aa signal	1202	465795607	14
Malassezia symphodialis	Yeast		Mal a s 12.0101	Contact	Malassezia Mala s 12	1618	78038796	17
Malassezia symphodialis ATCC 42132	Yeast		Mal a s 13	Contact	Malassezia Mala s 13 Thioredoxin Rev	1107	465793078	14
Malassezia symphodialis	Yeast		Mal a s 5	Contact	Malassezia Mala s 5	1172	4138171	17
Malassezia symphodialis ATCC 42132	Yeast		Unassigned	Contact	Malassezia Mala s 5	1172	465794772	14
Malassezia symphodialis	Yeast		Mal a s 6	Contact	Malassezia Mala s 6	1162	4138173	17
Malassezia symphodialis	Yeast		Mal a s 7	Contact	Malassezia Mala s 7	1187	4138175	17
Malassezia symphodialis	Yeast		Mal a s 8	Contact	Malassezia Mala s 8	1179	7271239	17
Malassezia symphodialis	Yeast		Mal a s 9	Contact	Malassezia Mala s 9	342	19069920	17
Malassezia symphodialis ATCC 42132	Yeast		Unassigned	Contact	Malassezia Mala s 9	342	465794420	14
Malus x domestica	Apple		Mal d 1.0301	Food Plant	Malus Mal d 1	159	1313966	17
Malus x domestica	Apple		Mal d 1.0401	Food Plant	Malus Mal d 1	160	1313968	17
Malus x domestica	Apple		Mal d 1.0402	Food Plant	Malus Mal d 1	160	1313970	17
Malus x domestica	Apple		Mal d 1.0403	Food Plant	Malus Mal d 1	160	1313972	17
Malus x domestica	Apple		Mal d 1.0206	Food Plant	Malus Mal d 1	159	12443824	17
Malus x domestica	Apple		Mal d 1.0103	Food Plant	Malus Mal d 1	159	4590364	17
Malus x domestica	Apple		Mal d 1.0203	Food Plant	Malus Mal d 1	159	4590366	17
Malus x domestica	Apple		Mal d 1.0204	Food Plant	Malus Mal d 1	159	4590368	17
Malus x domestica	Apple		Mal d 1.0104	Food Plant	Malus Mal d 1	159	4590376	17

Malus x domestica	Apple	Mal d 1.0105	Food Plant	Value Mal d 1	159	4590378	17
Malus x domestica	Apple	Mal d 1.0106	Food Plant	Value Mal d 1	159	4590380	17
Malus x domestica	Apple	Mal d 1.0107	Food Plant	Value Mal d 1	159	4590382	17
Malus x domestica	Apple	Mal d 1.0205	Food Plant	Value Mal d 1	159	4590388	17
Malus x domestica	Apple	Mal d 1.0208	Food Plant	Value Mal d 1	158	121685277	17
Malus x domestica	Apple	Mal d 1.0304	Food Plant	Value Mal d 1	159	27922941	17
Malus x domestica	Apple	Mal d 1.0108	Food Plant	Value Mal d 1	159	4768879	11
Malus x domestica	Apple	Mal d 1.0201	Food Plant	Value Mal d 1	159	862307	11
Malus x domestica	Apple	Mal d 1.0102	Food Plant	Value Mal d 1	159	886683	11
Malus x domestica	Apple	Mal d 1.0101	Food Plant	Value Mal d 1	159	1747852	15
Malus x domestica	Apple	Mal d 1.0109	Food Plant	Value Mal d 1	159	115418742	15
Malus x domestica	Apple	Mal d 1.0207	Food Plant	Value Mal d 1	159	115418744	15
Malus x domestica	Apple	Mal d 1.0302	Food Plant	Value Mal d 1	159	115418738	15
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 2	126	11478293	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 2	126	160418842	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 2	126	160418848	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 2	126	130316292	18
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 2	158	1218059718	10
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 2	158	1218059715	10
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 2	193	1392507603	13
Malus x domestica	Apple	Mal d 2.0101	Food Plant	Value Mal d 2	1245	13643249	15
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 3	115	150659891	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 3	115	150659889	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 3	115	150659885	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 3	115	150659879	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 3	115	150659859	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 3	115	138492338	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 3	115	114423814	19
Malus x domestica	Apple	Mal d 4.0302	Food Plant	Value Mal d 4	131	128881453	17
Malus x domestica	Apple	Mal d 4.0102	Food Plant	Value Mal d 4	131	128881457	17
Malus x domestica	Apple	Mal d 4.0202	Food Plant	Value Mal d 4	131	128881455	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 4	131	160418854	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 4	131	160418858	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 4	131	160418862	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 4	131	160418866	17
Malus x domestica	Apple	Unassigned	Food Plant	Value Mal d 4	131	1164510842	19

Malus x domestica	Apple	Unassigned	Food Plant	Malus Mal d 4	131	164510858	9
Malus x domestica	Apple	Unassigned	Food Plant	Malus Mal d 4	131	164510860	9
Malus x domestica	Apple	Unassigned	Food Plant	Malus Mal d 4	177	218059730	10
Malus x domestica	Apple	Unassigned	Food Plant	Malus Mal d 4	115	218059733	10
Malus x domestica	Apple	Unassigned	Food Plant	Malus Mal d 4	131	218059728	10
Malus x domestica	Apple	Mal d 4.0301	Food Plant	Malus Mal d 4	131	4761584	11
Malus x domestica	Apple	Mal d 4.0201	Food Plant	Malus Mal d 4	131	4761586	11
Malus x domestica	Apple	Mal d 4.0101	Food Plant	Malus Mal d 4	131	4761588	11
Manihot esculenta	Cassava	Unassigned	Food Plant	Manihot Man e 5.0101	177	21586695	17
Manihot esculenta	Cassava	Man e 5.0101	Food Plant	Manihot Man e 5.0101	177	332713934	14
Manilkara zapota	Sapodilla plum	Unassigned	Food Plant	Manilkara Thaumatin like protein 1	112	442580988	14
Manilkara zapota	Sapodilla plum	Unassigned	Food Plant	Manilkara Thaumatin like protein 1	9	442570282	14
Manilkara zapota	Sapodilla plum	Unassigned	Food Plant	Manilkara Thaumatin like protein 1	207	663434113	15
Marsipensaeus japonicus	Kuruma Shrimp	Unassigned	Food Animal	Marsipensaeus tropomyosin	1284	1125995159	8
Mercurialis annua	Annual mercury grass	Mer a 1	Aero Plant	Mercurialis Mer a 1	133	2295898	17
Macrurus magellanicus	Patagonian Grenadier	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	98	308191450	12
Macrurus magellanicus	Patagonian Grenadier	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	108	308191461	12
Macrurus magellanicus	Patagonian Grenadier	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	74	308191474	12
Macrurus novaezelandiae	Blue hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	183	308191475	12
Merluccius australis australis	southern hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	108	308191452	12
Merluccius australis polylepsis	Southern hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	108	308191453	12
Merluccius australis polylepsis	Southern hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	108	308191464	12
Merluccius bilinearis	Silver hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	108	308191465	12
Merluccius bilinearis	Silver hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	108	308191478	12
Merluccius bilinearis	Silver hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	194	308191488	12
Merluccius capensis	Shallow-water cape hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	108	308191466	12
Merluccius gnyi	Southern Pacific hake	Unassigned	Food Animal	Merluccius sp. Parvalbumin Hake	108	308191455	12

Merluccius gayi	Southern Pacific hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	191	1308191489	112
Merluccius merluccius	European hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	108	1311116	112
Merluccius merluccius	European hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	108	1308191469	112
Merluccius paradoxus	Deep-water cape hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	108	1308191457	112
Merluccius paradoxus	Deep-water cape hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	108	1308191470	112
Merluccius paradoxus	Deep-water cape hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	108	1308191483	112
Merluccius polli	Benguela hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	108	1308191471	112
Merluccius polli	Benguela hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	169	1308191484	112
Merluccius productus	North Pacific hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	108	1308191459	112
Merluccius productus	North Pacific hake	Unassigned	Food Animal	Merluccius sp.	Parvalbumin Hake	108	1308191472	112
Mesocricetus auratus	Golden hamster	Unassigned	Aero Animal	Mesocricetus auratus	Mes a 1	1172	13124669	116
Metapanaeus ensis	Greasyback shrimp	Met e 1	Food Animal	Metapanaeus Met e 1	Tropomyosin	1274	1607633	17
Mimachlamys nobilis	Noble scallop	Unassigned	Food Animal	Mimachlamys Tropomyosin		284	9954253	17
Morus nigra	Black mulberry	Mor n 3.0101	Food Plant	Morus Mor n 3 mulberry	LTP	191	288561913	111
Morus alba var. atropurpurea	White Mulberry	Unassigned	Food Plant	Morus winter accumulating protein		157	1610664572	115
Morus bombycis	Mulberry	Unassigned	Food Plant	Morus winter accumulating protein		157	154311115	112
Morus bombycis	Mulberry	Unassigned	Food Plant	Morus winter accumulating protein		157	154311119	112
Mus musculus	Mouse	Mus m 1	Aero Animal	Mus Mus m 1		180	120178291	17
Mus musculus	Mouse	Mus m 1.0101	Aero Animal	Mus Mus m 1		180	1295910	115
Mus musculus domesticus	Mouse	Mus m 1.0102	Aero Animal	Mus Mus m 1		180	1199881	115
Musa acuminata	Banana	Unassigned	Food Plant	Musa Allergen Endo-Beta-1,3-Glucanase		312	183754908	17
Musa acuminata AAA Group	Banana	Unassigned	Food Plant	Musa Allergen Endo-Beta-1,3-Glucanase		340	16073860	114
Musa acuminata	Banana	Mus a 4.0101	Food Plant	Musa Mus a 4		1200	188191901	17
Musa acuminata	Banana	Mus a 2.0101	Food Plant	Musa Mus s 2		1318	117932710	115
Musa acuminata	Banana	Mus xp 1	Food Plant	Musa profilin banana		1131	114161635	17
Myrmecia pilosula	Juniper ant	Unassigned	Venom or Salivary	Myrmecia Myr p 1		1112	11911819	17

Myrmecia pilosula	Jumper ant	Myr p 1.0101	Venom or Salivary	Myrmecia Myr p 1	1112	1312284	115
Myrmecia pilosula	Jumper ant	Myr p 2	Venom or Salivary	Myrmecia Myr p 2	175	1587177	17
Myrmecia pilosula	Jumper ant	Myr p 2.0101	Venom or Salivary	Myrmecia Myr p 2	175	1438761	110
Myrmecia banksi	Giant Bull Ant	Myr p 3.0101	Venom or Salivary	Myrmecia Myr p 3	184	151241753	115
Neptunea polycostata	Wrinkled Neptune	Unassigned	Food Animal	Neptunea tropomyosin	1284	1219806590	110
Nicotiana tabacum	Tobacco	Unassigned	Aero Plant	Nicotiana villin	1520	157283139	17
Nicotiana tabacum	Tobacco	Unassigned	Aero Plant	Nicotiana villin	1569	157283137	17
Octopus vulgaris	Octopus	Unassigned	Food Animal	Octopus tropomyosin	1284	183715936	17
Olea europaea	Olive tree	Ole e 1	Aero Plant	Olea Ole e 1	1145	14424429	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1137	11362128	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1136	11362129	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1136	11362130	17
Olea europaea	Olive tree	Ole e 1.0104	Aero Plant	Olea Ole e 1	1145	11362131	17
Olea europaea	Olive tree	Ole e 1	Aero Plant	Olea Ole e 1	1137	11362132	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1136	11362133	17
Olea europaea	Olive tree	Ole e 1.0103	Aero Plant	Olea Ole e 1	1145	11362136	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1136	11362137	17
Olea europaea	Olive tree	Ole e 1.0105	Aero Plant	Olea Ole e 1	1146	12465127	17
Olea europaea	Olive tree	Ole e 1.0106	Aero Plant	Olea Ole e 1	1146	12465129	17
Olea europaea	Olive tree	Ole e 1.0107	Aero Plant	Olea Ole e 1	1146	12465131	17
Olea europaea	Olive tree	Ole e 1.0101	Aero Plant	Olea Ole e 1	1130	113196763	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1134	137724597	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1135	137724593	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1132	137548763	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1131	13329768	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1132	13329766	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1132	13329764	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1131	13329762	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1131	13329760	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1129	13329748	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1131	13329744	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1132	13329738	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1132	13329732	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1132	13325115	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1140	1145313982	19
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 1	1140	1145313984	19

0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 1	140	145313988	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 1	140	145313990	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 1	140	145313992	9
0lea europaea	0live tree	0le e 10	Aero Plant	0lea Ole e 10	1123	25466664	17
0lea europaea	0live tree	0le e 11.0102	Aero Plant	0lea Ole e 11.0101 and 364 0102		168270856	11
0lea europaea	0live tree	0le e 11.0101	Aero Plant	0lea Ole e 11.0101 and 364 0102		269996495	11
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 12	1308	449061782	14
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 13	1226	449061783	14
0lea europaea	0live tree	0le e 2	Aero Plant	0lea Ole e 2	1134	3914427	17
0lea europaea	0live tree	0le e 2	Aero Plant	0lea Ole e 2	1134	3914428	17
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 2	1131	1576017874	115
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 2	1131	1576017774	115
0lea europaea	0live tree	0le e 2.0101	Aero Plant	0lea Ole e 2	1134	12465133	115
0lea europaea	0live tree	0le e 3.0101	Aero Plant	0lea Ole e 3	184	1337403	17
0lea europaea	0live tree	0le e 3	Aero Plant	0lea Ole e 3	152	137725377	17
0lea europaea	0live tree	0le e 5.0101	Aero Plant	0lea Ole e 5	130	1122064581	18
0lea europaea	0live tree	0le e 5	Aero Plant	0lea Ole e 5	1152	39840779	17
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	145313972	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347106	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1144	160347108	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347112	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347120	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347122	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347124	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347126	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347130	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347134	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160347138	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160962543	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160962547	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160962557	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160962569	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160962577	9
0lea europaea	0live tree	Unassigned	Aero Plant	0lea Ole e 5	1152	160962583	9

Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 5	144	160962587	9
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 5	152	160962591	9
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 5	152	160962597	9
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 5	152	160962611	9
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 5	152	160962613	9
Olea europaea	Olive tree	Ole e 6.0101	Aero Plant	Olea Ole e 6	150	12276458	11
Olea europaea	Olive tree	Ole e 7	Aero Plant	Olea Ole e 7	121	122002032	17
Olea europaea	Olive tree	Ole e 8	Aero Plant	Olea Ole e 8	171	16901654	17
Olea europaea	Olive tree	Ole e 8.0101	Aero Plant	Olea Ole e 8	171	16901652	11
Olea europaea	Olive tree	Ole e 9	Aero Plant	Olea Ole e 9	1460	114279169	17
Olea europaea	Olive tree	Unassigned	Aero Plant	Olea Ole e 9	1101	1166235350	9
Omaastrephes bartasii	red squid	Unassigned	Food Animal	Omaastrephes tropomyosin	1284	183715934	17
Onchocerca volvulus	Parasitic nematode	Unassigned	Worm (parasite)	Onchocerca tropomyosin	1284	142559586	112
Oncorhynchus keta	chum salmon	Onc k 5.0101	Food Animal	Oncorhynchus Onc k 5	1193	1296040357	115
Oncorhynchus mykiss	rainbow trout	Onc m 1.0101	Food Animal	Oncorhynchus Rainbow trout parv Onc m 1	1108	1288559139	111
Oncorhynchus mykiss	rainbow trout	Onc m 1.0201	Food Animal	Oncorhynchus Rainbow trout parv Onc m 1	1107	1288559140	111
Oratosquilla oratoria	hantais shrimp	Unassigned	Food Animal	Oratosquilla tropomyosin	1284	162286975	9
Oreochromis mossambicus	Mozambique tilapia	Ore m 4.0101	Food Animal	Oreochromis Ore m 4 tropomyosin	1284	1410060781	114
Oryctolagus cuniculus	European rabbit*	Ory c 3.A.0101	Aero Animal	Oryctolagus Ory c 3	193	11993600	115
Oryctolagus cuniculus	European rabbit*	Ory c 3.B.0101	Aero Animal	Oryctolagus Ory c 3	190	11993592	115
Oryza sativa	Rice	Unassigned	Food Plant	Oryza Glyoxalase I	1291	184029333	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Glyoxalase I	1291	16580747	17
Oryza sativa	Rice	Ory s 1.0101	Aero Plant	Oryza Ory s 1	1263	1173557	18
Oryza sativa	Rice	Unassigned	Aero Plant	Oryza Ory s 1	1267	18118439	17
Oryza sativa (japonica cultivar-group)	Rice	Ory s 1	Aero Plant	Oryza Ory s 1	1267	109913547	18
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Aero Plant	Oryza putative polcalcin Phl p 7	182	145736119	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor	157	123616954	18
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor	165	1218193	17



Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 157	218197	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 111	1304216	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 109	1304217	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 113	1304218	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 166	1398913	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 160	1398915	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 157	1398916	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 160	1398918	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 157	2827316	17
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 166	114152865	18
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 163	114152864	18
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 160	23495787	18
Oryza sativa (japonica cultivar-group)	Rice	Unassigned	Food Plant	Oryza Trypsin alpha-amylase inhibitor 160	23616947	17
Ostrya carpinifolia	European hop hornbeam	Ost c 1.0101	Aero Plant	Ostrya Ost c pollen allergen	300872635	112
Pachycondyla chinensis	Asian needle ant	Unassigned	Venom or Salivary	Pachycondyla Pac c 3 allergen	169822894	110
Pandalus borealis	Caribbean shrimp	Pan b 1.0101	Food Animal	Pandalus Pan b 1	1312831088	112
Penaeus stimpsoni	Lobster	Pan s 1.0101	Food Animal	Penaeus Pan s 1	13080761	111
Paralithodes camtschaticus	Kamchatka crab	Unassigned	Food Animal	Paralithodes tropomyosin	125995163	18
Paralithodes camtschaticus	Kamchatka crab	Unassigned	Food Animal	Paralithodes tropomyosin	125995165	18
Parietaria judaica	Weed	Par j 1	Aero Plant	Parietaria Par j 1	1741844	17
Parietaria judaica	Weed	Par j 1.0102	Aero Plant	Parietaria Par j 1	11532058	17

Parietaria judaica	Weed	Par j 1.0101	Aero Plant	Parietaria Par j 1	133	992612	115
Parietaria judaica	Weed	Par j 1.0103	Aero Plant	Parietaria Par j 1	139	95007033	115
Parietaria judaica	Weed	Par j 1.0201	Aero Plant	Parietaria Par j 1	138	706811	115
Parietaria judaica	Weed	Par j 2.0102	Aero Plant	Parietaria Par j 2	133	1532056	17
Parietaria judaica	Weed	Par j 2.0101	Aero Plant	Parietaria Par j 2	133	12497750	17
Parietaria judaica	Weed	Par j 3.0102	Aero Plant	Parietaria Par j 3 profilin	131	14423869	17
Parietaria judaica	Weed	Par j 3.0101	Aero Plant	Parietaria Par j 3 profilin	132	14423876	17
Parietaria judaica	Weed	Par j 3.0201	Aero Plant	Parietaria Par j 3 profilin	131	444175753	114
Parietaria judaica	Weed	Par j 4.0101	Aero Plant	Parietaria Par j 4	184	1201071363	115
Parietaria officialis	Weed	Par o 1	Aero Plant	Parietaria Par o 1	112	75139847	17
Parietaria officialis	Weed	Par o 1	Aero Plant	Parietaria Par o 1	117	1311509	17
Parietaria officialis	Weed	Par o 1	Aero Plant	Parietaria Par o 1	115	1311510	17
Parietaria officialis	Weed	Par o 1	Aero Plant	Parietaria Par o 1	115	1311511	17
Parietaria officialis	Weed	Par o 1	Aero Plant	Parietaria Par o 1	115	1311512	17
Parietaria officialis	Weed	Par o 1	Aero Plant	Parietaria Par o 1	130	1311513	17
Parietaria officialis	Weed	Par o 1	Aero Plant	Parietaria Par o 1	124	1836011	17
Parietaria officialis	Weed	Unassigned	Aero Plant	Parietaria Par o 1	125	1836010	17
Paspalum notatum	Bahia grass	Unassigned	Aero Plant	Paspalum group 13 pollen allergen	169	1338930686	112
Paspalum notatum	Bahia grass	Unassigned	Aero Plant	Paspalum group 13 pollen allergen	169	1338930684	112
Paspalum notatum	Bahia grass	Unassigned	Aero Plant	Paspalum group 13 pollen allergen	169	1338930682	112
Paspalum notatum	Bahia grass	Unassigned	Aero Plant	Paspalum group 13 pollen allergen	169	1338930680	112
Paspalum notatum	Bahia grass	Unassigned	Aero Plant	Paspalum group 13 pollen allergen	169	1338930678	112
Paspalum notatum	Bahia grass	Unassigned	Aero Plant	Paspalum group 13 pollen allergen	169	1338930676	112
Paspalum notatum	Bahia grass	Unassigned	Aero Plant	Paspalum group 13 pollen allergen	1391	1338930674	112
Paspalum notatum	Bahia grass	Unassigned	Aero Plant	Paspalum group 13 pollen allergen	1395	1338930672	112
Paspalum notatum	Bahia grass	Pas n 1.0101	Aero Plant	Paspalum Pas n 1 beta	1265	1168419914	110

Penaeus monodon	Black tiger shrimp	Pen m 1.0101	Food Animal	expansin	1284	60892782	115
Penaeus monodon	Black tiger shrimp	Pen m 2	Food Animal	Penaeus Pen m 2	366	127463265	17
Penaeus monodon	Black tiger shrimp	Unassigned	Food Animal	Penaeus Pen m 2	366	1308154236	112
Penaeus monodon	Black tiger shrimp	Pen m 3.0101	Food Animal	Penaeus Pen m 3 myosin light chain	1177	1317383196	112
Penaeus monodon	Black tiger shrimp	Pen m 4.0101	Food Animal	Penaeus Pen m 4 sarcoplasmic calcium binding	1193	1317383198	112
Penicillium crustosum	Fungus	Pen cr 26.0101	Aero Fungi	Penicillium crustosum Pen cr 26 60s P1	1107	1371537645	113
Penicillium chrysogenum	Fungus	Pen ch 18.0101	Aero Fungi	Penicillium Pen 18	1494	17963902	17
Penicillium chrysogenum	Fungus	Pen ch 18	Aero Fungi	Penicillium Pen 18	1494	114215732	17
Penicillium citrinum	Fungus	Unassigned	Aero Fungi	Penicillium Pen 18	1457	14588118	17
Penicillium citrinum	Fungus	Unassigned	Aero Fungi	Penicillium Pen 18	1358	112006501	17
Penicillium oxalicum	Fungus	Pen o 18.0101	Aero Fungi	Penicillium Pen 18	1503	112006497	17
Penicillium brevicompactum	Fungus	Pen b 26.0101	Aero Fungi	Penicillium Pen b 26	1107	159894749	17
Penicillium citrinum	Fungus	Pen c 19	Aero Fungi	Penicillium Pen c 19	1503	114423733	17
Penicillium citrinum	Fungus	Pen c 22	Aero Fungi	Penicillium Pen c 22	1438	113991101	17
Penicillium citrinum	Fungus	Pen c 24	Aero Fungi	Penicillium Pen c 24	1228	138326693	17
Penicillium citrinum	Fungus	Pen c 3	Aero Fungi	Penicillium Pen c 3	1167	15326864	17
Penicillium citrinum	Fungus	Pen c 30.0101	Aero Fungi	Penicillium Pen c 30	1733	182754305	17
Penicillium citrinum	Fungus	Pen c 32.0101	Aero Fungi	Penicillium Pen c 32	1290	1121584258	18
Penicillium chrysogenum	Fungus	Pen ch 13.0101	Aero Fungi	Penicillium Pen ch 13	1397	16684758	17
Penicillium chrysogenum	Fungus	Pen ch 13	Aero Fungi	Penicillium Pen ch 13	1398	121069093	17
Penicillium citrinum	Fungus	Unassigned	Aero Fungi	Penicillium Pen ch 13	1397	14587983	17
Penicillium chrysogenum	Fungus	Pen ch 20	Aero Fungi	Penicillium Pen ch 20	1117	1999009	17
Penicillium chrysogenum	Fungus	Pen ch 35.0101	Aero Fungi	Penicillium Pen ch 35	1324	1300679427	115
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta americana Per a 11	1494	1821092692	116
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta americana Per a 12	1407	1821092694	116
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta GST	1216	160678789	17
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta GST	1216	1359326557	115
Periplaneta americana	American cockroach	Per a 7.0102	Aero Insect	Periplaneta Per 7	1284	14378573	17

Periplaneta americana	American cockroach	Per a 7.0101	Aero Insect	Periplaneta Per 7	1284	14468639	17
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per 7	1284	1239740599	111
Periplaneta fuliginosa	Smokybrown cockroach	Unassigned	Aero Insect	Periplaneta Per 7	1284	19310971	17
Periplaneta americana	American cockroach	Per a 1.0201	Aero Insect	Periplaneta Per a 1	1446	12231297	17
Periplaneta americana	American cockroach	Per a 1.0104	Aero Insect	Periplaneta Per a 1	1274	12253610	17
Periplaneta americana	American cockroach	Per a 1.0103	Aero Insect	Periplaneta Per a 1	1395	12580504	17
Periplaneta americana	American cockroach	Per a 1.0102	Aero Insect	Periplaneta Per a 1	1228	12897849	17
Periplaneta americana	American cockroach	Per a 1.0101	Aero Insect	Periplaneta Per a 1	1231	14240399	17
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 1	1124	130144660	17
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 1	1395	1284518361	111
Periplaneta americana	American cockroach	Per a 10.0101	Aero Insect	Periplaneta Per a 10	1266	160678799	17
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 2	1351	160678785	17
Periplaneta americana	American cockroach	Per a 2.0101	Aero Insect	Periplaneta Per a 2	1351	1313870534	112
Periplaneta americana	American cockroach	Per a 3.0201	Aero Insect	Periplaneta Per a 3	1631	11531589	17
Periplaneta americana	American cockroach	Per a 3.0202	Aero Insect	Periplaneta Per a 3	1470	11580794	17
Periplaneta americana	American cockroach	Per a 3.0203	Aero Insect	Periplaneta Per a 3	1393	11580797	17
Periplaneta americana	American cockroach	Per a 3.0101	Aero Insect	Periplaneta Per a 3	1685	12833325	19
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 3	1688	1284518363	111
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 3	1685	1289721058	111
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 4	1183	160678787	17
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 4	1163	1215794707	110
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 4	1167	1212675312	110
Periplaneta americana	American cockroach	Per a 6.0101	Aero Insect	Periplaneta Per a 6	1151	160678791	18
Periplaneta americana	American cockroach	Unassigned	Aero Insect	Periplaneta Per a 9	1356	150428904	18
Periplaneta americana	American cockroach	Per a 9.0101	Aero Insect	Periplaneta Per a 9	1356	1167782135	19
Perna viridis	Asian green mussell	Unassigned	Food Animal	Perna Troponyosin	1284	19954251	17
Persea americana	Avocado	Pers a 1	Food Plant	Persea Pers a 1	1326	13201547	17
Phalaris aquatica	Canary grass	Unassigned	Aero Plant	Phalaris Pha a 1	120	1409328	17
Phalaris aquatica	Canary grass	Pha a 1	Aero Plant	Phalaris Pha a 1	1269	12498576	17
Phalaris aquatica	Canary grass	Pha a 5.0101	Aero Plant	Phalaris Pha a 5	1320	12498577	17
Phalaris aquatica	Canary grass	Unassigned	Aero Plant	Phalaris Pha a 5	1305	12498578	17
Phalaris aquatica	Canary grass	Unassigned	Aero Plant	Phalaris Pha a 5	1294	12498579	17
Phalaris aquatica	Canary grass	Unassigned	Aero Plant	Phalaris Pha a 5	1175	12498580	17
Phaseolus vulgaris	Kidney bean	Pha v 3.0101	Food Plant	Phaseolus Pha v 3	1115	1289064177	111

Phaseolus vulgaris	Kidney bean	Pha v 3.0201	Food Plant	Phaseolus Pha v 3	118	289064179	111
Phleum pratense	Common timothy	Phl p 1.0102	Aero Plant	Phleum Phl p 1	1263	473360	17
Phleum pratense	Common timothy	Phl p 1.0101	Aero Plant	Phleum Phl p 1	1263	3901094	17
Phleum pratense	Common timothy	Phl p 1	Aero Plant	Phleum Phl p 1	1241	28373838	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 1	1240	45823012	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 1	1262	1582250	110
Phleum pratense	Common timothy	Phl p 11	Aero Plant	Phleum Phl p 11	1143	123452313	17
Phleum pratense	Common timothy	Phl p 12.0103	Aero Plant	Phleum Phl p 12	1131	12415700	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 12	1131	110644906	18
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 12	1131	110644908	18
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 12	1131	110644910	18
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 12	1131	110644912	18
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 12	1131	110644914	18
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 12	1131	110644916	18
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 12	1131	110644918	18
Phleum pratense	Common timothy	Phl p 12.0102	Aero Plant	Phleum Phl p 12	1131	12415698	110
Phleum pratense	Common timothy	Phl p 12.0101	Aero Plant	Phleum Phl p 12	1131	1453976	115
Phleum pratense	Common timothy	Phl p 13	Aero Plant	Phleum Phl p 13	1394	14526572	17
Phleum pratense	Common timothy	Phl p 2	Aero Plant	Phleum Phl p 2	1122	1415896	17
Phleum pratense	Common timothy	Phl p 4.0101	Aero Plant	Phleum Phl p 4	1508	154144332	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 4	1500	145108973	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 4	1500	145108967	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 4	1500	189014266	110
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 4	1500	189014268	110
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 4	1500	189014270	110
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 4	1500	189014272	110
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 4	1500	1405944794	114
Phleum pratense	Common timothy	Phl p 4.0201	Aero Plant	Phleum Phl p 4	1508	154144334	115
Phleum pratense	Common timothy	Phl p 5.0101	Aero Plant	Phleum Phl p 5	1312	1398830	17
Phleum pratense	Common timothy	Phl p 5	Aero Plant	Phleum Phl p 5	1257	1422005	17
Phleum pratense	Common timothy	Phl p 5	Aero Plant	Phleum Phl p 5	1280	1481397	17
Phleum pratense	Common timothy	Phl p 5	Aero Plant	Phleum Phl p 5	124	175139900	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1285	11092249	17
Phleum pratense	Common timothy	Phl p 5.0202	Aero Plant	Phleum Phl p 5	1281	11684718	17
Phleum pratense	Common timothy	Phl p 5.0104	Aero Plant	Phleum Phl p 5	1276	11684720	17
Phleum pratense	Common timothy	Phl p 5.0102	Aero Plant	Phleum Phl p 5	1286	12398757	17

Phleum pratense	Common timothy	Phl p 5.0105	Aero Plant	Phleum Phl p 5	1276	13135497	17
Phleum pratense	Common timothy	Phl p 5.0106	Aero Plant	Phleum Phl p 5	1276	13135499	17
Phleum pratense	Common timothy	Phl p 5.0107	Aero Plant	Phleum Phl p 5	1276	13135501	17
Phleum pratense	Common timothy	Phl p 5.0108	Aero Plant	Phleum Phl p 5	1276	13135503	17
Phleum pratense	Common timothy	Phl p 5.0103	Aero Plant	Phleum Phl p 5	1312	13309039	17
Phleum pratense	Common timothy	Phl p 5.0203	Aero Plant	Phleum Phl p 5	1295	13309041	17
Phleum pratense	Common timothy	Phl p 5.0206	Aero Plant	Phleum Phl p 5	1290	13309045	17
Phleum pratense	Common timothy	Phl p 5.0207	Aero Plant	Phleum Phl p 5	1287	13309047	17
Phleum pratense	Common timothy	Phl p 5	Aero Plant	Phleum Phl p 5	1275	113430402	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725606	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725608	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725610	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725612	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725614	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725616	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725618	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725620	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725622	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725624	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725626	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725628	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725630	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1287	121725632	17
Phleum pratense	Common timothy	Phl p 5	Aero Plant	Phleum Phl p 5	1102	128948464	17
Phleum pratense	Common timothy	Phl p 5.0109	Aero Plant	Phleum Phl p 5	1284	129500897	17
Phleum pratense	Common timothy	Phl p 5.0201	Aero Plant	Phleum Phl p 5	1284	12398759	110
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 5	1309	1345108717	113
Phleum pratense	Common timothy	Phl p 6.0102	Aero Plant	Phleum Phl p 6	1138	13004465	17
Phleum pratense	Common timothy	Phl p 6.0101	Aero Plant	Phleum Phl p 6	1138	13004467	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 6	1106	13004469	17
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum Phl p 6	1111	128374072	17
Phleum pratense	Common timothy	Phl p 7.0101	Aero Plant	Phleum Polcalin (Phl p 17)	178	13367732	110
Phleum pratense	Common timothy	Unassigned	Aero Plant	Phleum pollen allergen group 3	1100	1283806867	111
Phodopus sungorus	Siberian hamster	Unassigned	Aero Plant	Phodopus sungorus lipocalin	1151	1530376029	116

Phoenix dactylifera	Date palm	Pho d 2	Aero Plant	Phoenix Pho d 2	131	21322677	17
Pinus pinea	Pine	Unassigned	Aero Plant	Pinus pinea albumin	110	749495809	116
Pistacia vera	*Pistachio	Unassigned	Food Plant	Pistacia 11S globulin	1472	156001070	19
Pistacia vera	*Pistachio	Pis v 2.0101	Food Plant	Pistacia 11S globulin	1496	110349083	110
Pistacia vera	*Pistachio	Pis v 2.0201	Food Plant	Pistacia 11S globulin	1472	110349085	110
Pistacia vera	*Pistachio	Pis v 1.0101	Food Plant	Pistacia Pis v 1 2S albumin	1149	110349081	110
Pistacia vera	*Pistachio	Pis v 3.0101	Food Plant	Pistacia Pis v 3 vicilin	1519	133711974	110
Pistacia vera	*Pistachio	Pis v 4.0101	Food Plant	Pistacia Pis v 4	1230	149786150	19
Pisum sativum	Pea	Pis s 1.0102	Food Plant	Pisum Pis s 1	1415	142414629	17
Pisum sativum	Pea	Pis s 1.0101	Food Plant	Pisum Pis s 1	1415	142414627	17
Pisum sativum	Pea	Pis s 2.0101	Food Plant	Pisum Pis s 2	1613	17339551	115
Plantago lanceolata	Narrow-leaved plantain	Pla 1 1.0101	Aero Plant	Plantago Pla 1 1	1131	114422359	17
Plantago lanceolata	Narrow-leaved plantain	Pla 1 1.0102	Aero Plant	Plantago Pla 1 1	1131	114422361	17
Plantago lanceolata	Narrow-leaved plantain	Pla 1 1.0103	Aero Plant	Plantago Pla 1 1	1131	114422363	17
Plantago lanceolata	Narrow-leaved plantain	Unassigned	Aero Plant	Plantago Pla 1 1	165	129163773	17
Platanus x acerifolia	London plane tree	Unassigned	Aero Plant	Platanus acerifolia Pla 118	1118	110224778	116
Platanus x acerifolia	London plane tree	Unassigned	Aero Plant	Platanus acerifolia Pla 193	193	930156468	116
Platanus x acerifolia	London plane tree	Pla a 1	Aero Plant	Platanus Pla a 1	1179	126190140	17
Platanus x acerifolia	London plane tree	Pla a 2	Aero Plant	Platanus Pla a 2	1377	149523394	17
Platanus orientalis	oriental plane	Pla or 1.0101	Aero Plant	Platanus Pla or 1	1170	162949336	19
Platanus orientalis	oriental plane	Pla or 2.0101	Aero Plant	Platanus Pla or 2	1378	162949338	19
Plodia interpunctella	Indian meal moth	Plo i 1.0101	Aero Insect	Plodia Plo i 1 Arginine kinase	355	15886861	17
Plodia interpunctella	Indian meal moth	Plo i 2.0101	Aero Insect	Plodia Plo i 2 thioresoxin	1106	1308193268	112
Poa pratensis	Kentucky bluegrass	Poa p 1	Aero Plant	Poa p 1	120	1280414	17
Poa pratensis	Kentucky bluegrass	Poa p 1	Aero Plant	Poa p 1	126	1320620	17
Poa pratensis	Kentucky bluegrass	Poa p 1.0101	Aero Plant	Poa p 1	1263	14090265	17
Poa pratensis	Kentucky bluegrass	Poa p 5	Aero Plant	Poa p 5	1303	111991227	17
Poa pratensis	Kentucky bluegrass	Unassigned	Aero Plant	Poa p 9	1373	1113560	17
Poa pratensis	Kentucky bluegrass	Unassigned	Aero Plant	Poa p 9	1307	1113562	17
Poa pratensis	Kentucky bluegrass	Unassigned	Aero Plant	Poa p 9	1131	1539056	17

Poa pratensis	Kentucky bluegrass	Unassigned	Aero Plant	Poa Poa p 9	333	113561	17
Polistes annularis	Paper wasp	Pol a 5.0101	Venom or Salivary	Polistes Pol 5	1209	160780	17
Polistes dominulus	Paper wasp	Pol d 5	Venom or Salivary	Polistes Pol 5	1227	151093377	17
Polistes exclamans	Paper wasp	Pol e 5.0101	Venom or Salivary	Polistes Pol 5	1226	151093375	17
Polistes fuscatus	Paper wasp	Pol f 5	Venom or Salivary	Polistes Pol 5	1205	1549188	17
Polistes gallicus	Paper wasp	Pol g 5	Venom or Salivary	Polistes Pol 5	1206	125091511	17
Polistes annularis	Paper wasp	Pol a 1.0101	Venom or Salivary	Polistes Pol a 1 Pol d 301	1301	15815249	111
Polistes dominulus	Paper wasp	Pol d 1.0104	Venom or Salivary	Polistes Pol a 1 Pol d 316	1316	145510893	17
Polistes dominulus	Paper wasp	Pol d 1.0103	Venom or Salivary	Polistes Pol a 1 Pol d 316	1316	145510891	17
Polistes dominulus	Paper wasp	Pol d 1.0102	Venom or Salivary	Polistes Pol a 1 Pol d 316	1316	145510889	17
Polistes dominulus	Paper wasp	Pol d 1.0101	Venom or Salivary	Polistes Pol a 1 Pol d 337	1337	145510887	17
Polistes gallicus	Paper wasp	Unassigned	Venom or Salivary	Polistes Pol a 1 Pol d 142	142	141017429	17
Polistes annularis	Paper wasp	Pol a 2.0101	Venom or Salivary	Polistes Pol a 2	1367	15815251	111
Polistes dominulus	Paper wasp	Pol d 4.0101	Venom or Salivary	Polistes Venom serine protease	1277	130909091	17
Polybia paulista	wasp	Unassigned	Venom or Salivary	Polybia p hyaluronidase	345	1302201583	112
Polybia paulista	wasp	Unassigned	Venom or Salivary	Polybia p hyaluronidase	288	1302425085	112
Polybia paulista	wasp	Unassigned	Venom or Salivary	Polybia p venom allergen 5	1141	1290792375	111
Polybia paulista	wasp	Unassigned	Venom or Salivary	Polybia p venom allergen 5	1207	1302595972	112
Polybia paulista	wasp	Pol p 1.0101	Venom or Salivary	Polybia Pol p 1.0101 phospholipase	1322	1166216292	9
Polybia paulista	wasp	Unassigned	Venom or Salivary	Polybia Pol p 1.0101 phospholipase	1302	1315190620	112
Pontastacus leptodactylus	Danube crayfish	Pon 1 4.0101	Food Animal	Pontastacus Pon 1 4	1192	1134309	115
Portunus pelagicus	blue swimmer crab	Por p 1.0101	Food Animal	Portunus Por p 1 tropomyosin	1284	1448278534	114
Portunus sanguinolentus	Crab	Unassigned	Food Animal	Portunus Por p 1.0101 tropomyosin	1284	1119674937	8
Portunus trituberculatus	Crab	Unassigned	Food Animal	Portunus Por p 1.0101 tropomyosin	1284	1151505281	9
Procambarus clarkii	red swamp crayfish	Unassigned	Food Animal	Procambarus red crayfish arginine kinase	1357	1375298901	113
Procambarus clarkii	red swamp crayfish	Unassigned	Food Animal	Procambarus tropomyosin	1284	1225348412	110
Protopis juliflora	mesquite	Pro j 2.0101	Aero Plant	Protopis Pro j 2	1133	1625293889	115



Protortonia cacti	Arthropod	Unassigned	Food Animal	Protortonia	335	1237769615	11
Prunus dulcis	Almond	Unassigned	Food Plant	Prunus persica Pru p 2  241		190613941	10
Prunus dulcis x Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  160		190613871	10
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  246		190613905	10
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  246		190613909	10
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  246		190613907	10
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  246		190613911	10
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  242		190613903	10
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  246		25091405	12
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  242		25091406	12
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus persica Pru p 2  246		359744030	13
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		2677826	17
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		1513216	17
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		44409496	17
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		44409474	17
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		44409451	17
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		159162378	19
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		82492285	17
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		748758672	16
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus PRP (Bet v 1  family)		748758670	16
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus Pru 3	119	1313575730	12
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus Pru 3	117	1313575732	12
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus Pru 3	117	1313575734	12
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus Pru 3	117	1313575736	12
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus Pru 3	117	16715520	17
Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus Pru 3	117	1313575726	12

Prunus avium	Cherry	Unassigned	Food Plant	Prunus Pru 3	117	313575728	112
Prunus domestica	Plum	Pru d 3	Food Plant	Prunus Pru 3	191	19297015	17
Prunus persica	Peach	Pru p 3.0101	Food Plant	Prunus Pru 3	191	3287877	17
Prunus persica	Peach	Unassigned	Food Plant	Prunus Pru 3	117	154793477	17
Prunus persica	Peach	Unassigned	Food Plant	Prunus Pru 3	117	313575718	112
Prunus persica	Peach	Unassigned	Food Plant	Prunus Pru 3	117	1544369592	115
Prunus avium	Cherry	Pru av 4	Food Plant	Prunus Pru 4 Profilin   peach cherry almond	131	4761682	17
Prunus dulcis	Almond	Pru du 4.0101	Food Plant	Prunus Pru 4 Profilin   peach cherry almond	131	24473794	17
Prunus dulcis x Prunus persica	Plant hybrid	Unassigned	Food Plant	Prunus Pru 4 Profilin   peach cherry almond	131	190613937	110
Prunus persica	Peach	Pru p 4.01	Food Plant	Prunus Pru 4 Profilin   peach cherry almond	131	27528310	17
Prunus persica	Peach	Pru p 4.02	Food Plant	Prunus Pru 4 Profilin   peach cherry almond	131	27528312	17
Prunus avium	Cherry	Pru av 2	Food Plant	Prunus Pru av 2	1245	1144346	17
Prunus dulcis	Almond	Unassigned	Food Plant	Prunus Pru du 6 Amandin   531		285588247	111
Prunus dulcis	Almond	Unassigned	Food Plant	Prunus Pru du 6 Amandin   178		1523916668	115
Prunus dulcis	Almond	Pru du 6.0101	Food Plant	Prunus Pru du 6 Amandin   551		1307159112	115
Prunus dulcis	Almond	Pru du 6.0201	Food Plant	Prunus Pru du 6 Amandin   504		1307159114	115
Prunus persica	Peach	Pru p 7.0101	Food Plant	Prunus Pru p 7   Peamaclein	163	408407790	114
Prunus dulcis	Almond	Unassigned	Food Plant	Prunus Seed allergenic protein 2 (Conglutin gamma)	125	76107131	18
Pseudocardium sachalinensis	Mollusc	Unassigned	Food Animal	Pseudocardium tropomyosin	1284	219806598	110
Punica granatum	Pomegranate	Pun g 1.0101	Food Plant	Punica Pun g 1	1120	155979767	115
Punica granatum	Pomegranate	Pun g 1.0201	Food Plant	Punica Pun g 1	1120	155979765	115
Punica granatum	Pomegranate	Pun g 1.0301	Food Plant	Punica Pun g 1	1120	155979763	115
Pyrus communis	Pear	Pyr c 3.0101	Food Plant	Pyrus LTP Pyr c 3 IUIS   115		16715524	111
Pyrus communis	Pear	Unassigned	Food Plant	Pyrus LTP Pyr c 3 IUIS   194		355525862	113
Pyrus communis	Pear	Unassigned	Food Plant	Pyrus LTP Pyr c 3 IUIS   194		355525860	113
Pyrus communis	Pear	Unassigned	Food Plant	Pyrus LTP Pyr c 3 IUIS   194		355525856	113
Pyrus communis	Pear	Pyr c 1.0101	Food Plant	Pyrus Pyr c 1	1159	114423877	19
Pyrus communis	Pear	Pyr c 4	Food Plant	Pyrus Pyr c 4	1131	4761680	17
Pyrus communis	Pear	Pyr c 5	Food Plant	Pyrus Pyr c 5	1308	13243234	17
Quercus alba	Oak	Que a 1.0201	Aero Plant	Quercus Que a 1	1159	1167472847	110
Quercus alba	Oak	Que a 1.0401	Aero Plant	Quercus Que a 1	1160	1167472851	110

Quercus alba	Oak	Que a 1.0301	Aero Plant	Quercus Que a 1	160	167472849	110
Rana esculenta	Frog	Ran e 1	Food Animal	Rana Ran e 1	110	20796729	17
Rana sp. CH-2001	Frog	Unassigned	Food Animal	Rana Ran e 1	110	20796733	17
Rana esculenta	Frog	Ran e 2	Food Animal	Rana Ran e 2	109	20797081	17
Rana sp. CH-2001	Frog	Unassigned	Food Animal	Rana Ran e 2	109	20797085	17
Rattus norvegicus	Rat	Rat n 1	Aero Animal	Rattus Rat n 1	181	127533	17
Rattus norvegicus	Rat	Rat n 1	Aero Animal	Rattus Rat n 1	181	81890324	17
Rattus norvegicus	Rat	Rat n 1.0101	Aero Animal	Rattus Rat n 1	177	204261	15
Rhizopus oryzae	Fungus	Unassigned	Aero Fungi	Rhizopus Rhi o 1.0101	1401	695094784	16
Rhodotorula	Fungus	Rho m 1.0101	Aero Fungi	Rhodotorula Rho m 1	1439	130314940	11
Rhodotorula	Fungus	Rho m 2.0101	Aero Fungi	Rhodotorula Rho m 2	1342	154654335	17
Ricinus communis	Castor bean	Ric c 1.0101	Food Plant	Ricinus Ric c 1	1258	121068	15
Rubus idaeus	Raspberry	Rub i 1.0101	Food Plant	Rubus Rub i 1	137	110180625	8
Rubus idaeus	Raspberry	Rub i 3.0101	Food Plant	Rubus Rub i 3	117	110180623	8
Salmo salar	Salmon	Sal s 1	Food Animal	Salmo Sal s 1	108	18281421	17
Salmo salar	Salmon	Unassigned	Food Animal	Salmo Sal s 1	109	209734468	10
Salmo salar	Salmon	Sal s 1.0101	Food Animal	Salmo Sal s 1	109	1322183	15
Salmo salar	Salmon	Unassigned	Food Animal	Salmo Sal s 2 enolase	1432	385145180	13
Salmo salar	Salmon	Sal s 2.0101	Food Animal	Salmo Sal s 2 enolase	1434	197632415	15
Salmo salar	Salmon	Unassigned	Food Animal	Salmo Sal s 3 aldolase	363	385145176	13
Salmo salar	Salmon	Sal s 3.0101	Food Animal	Salmo Sal s 3 aldolase	363	213511774	15
Salsola kali	Thistle	Sal k 1.0201	Aero Plant	Salsola pectin methylesterase Sal k 1.01 & 1.02	362	151242679	8
Salsola kali	Thistle	Sal k 1.0302	Aero Plant	Salsola pectin methylesterase Sal k 1.01 & 1.02	339	159895728	8
Salsola kali	Thistle	Sal k 1.0301	Aero Plant	Salsola pectin methylesterase Sal k 1.01 & 1.02	339	159895730	8
Salsola kali	Thistle	Unassigned	Aero Plant	Salsola pectin methylesterase Sal k 1.01 & 1.02	339	225810697	10
Salsola kali	Thistle	Sal k 1.0101	Aero Plant	Salsola Sal k 1	142	25090947	10
Salsola kali	Thistle	Unassigned	Aero Plant	Salsola Sal k 3 pollen allergen	757	225810699	10
Salsola kali	Thistle	Sal k 4.0101	Aero Plant	Salsola Sal k 4 profilin	133	239916566	11
Salsola kali	Thistle	Unassigned	Aero Plant	Salsola Sal k 4 profilin	133	159912885	15

Salsola kali	Thistle	Sal k 4.0201	Aero Plant	Salsola Sal k 4 profilin	133	300490499	15
Salsola kali	Thistle	Sal k 5.0101	Aero Plant	Salsola Sal k 5	151	300490501	15
Salvelinus fontinalis	Brook trout	Unassigned	Food Animal	Salvelinus parvalbumin	109	28857438	11
Salvelinus fontinalis	Brook trout	Unassigned	Food Animal	Salvelinus parvalbumin	108	28857440	11
Sarcoptes scabiei type hominis	Scabies mite	Unassigned	Venom or Salivary	Sarcoptes Apolipoprotein Ssagl.2	330	27462848	17
Sarcoptes scabiei	Scabies mite	Unassigned	Aero Mite	Sarcoptes	310	1507480520	15
Sarcoptes scabiei	Scabies mite	Unassigned	Venom or Salivary	Sarcoptes cysteine protease C08	340	46406002	17
Sarcoptes scabiei	Scabies mite	Unassigned	Venom or Salivary	Sarcoptes cysteine proteases F04	338	46406012	17
Sarcoptes scabiei	Scabies mite	Unassigned	Venom or Salivary	Sarcoptes cysteine proteases F04	339	46406014	17
Sarcoptes scabiei	Scabies mite	Unassigned	Venom or Salivary	Sarcoptes cysteine proteases F04	273	46406016	17
Sarcoptes scabiei	Scabies mite	Unassigned	Venom or Salivary	Sarcoptes Glutathione S-transferase Mu	1219	27462836	17
Sarcoptes scabiei	Scabies mite	Unassigned	Venom or Salivary	Sarcoptes Glutathione S-transferase Mu	1219	60920770	17
Sardinops sagax	South American pilchard	Sar sa 1.0101	Food Animal	Sardinops Sar sa 1 parvalbumin	1109	193247972	10
Scapharca broughtonii	Clam	Unassigned	Food Animal	Scapharca tropomyosin	1284	219806592	10
Schistosoma japonicum	Schistosoma	Unassigned	Protozoan	Schistosoma profilin	1129	29841461	17
Schistosoma japonicum	Schistosoma	Unassigned	Protozoan	Schistosoma tegumental antigen	191	2739154	17
Schizophyllum commune	Mushroom	Sch c 1.0101	Food Fungi	Schizophyllum Sch c 1	1576	302681819	15
HA-8							
Scomber japonicus	Chub mackerel	Unassigned	Food Animal	Scomber Parvalbumin	1109	29420793	17
Scomber scombrus	Atlantic mackerel	Unassigned	Food Animal	Scomber Parvalbumin	1109	28857436	11
Scylla paramamosain	green mud crab	Unassigned	Food Animal	Scylla arginine kinase	357	375298903	13
Scylla serrata	giant mud crab	Unassigned	Food Animal	Scylla sp. (mud crab) tropomyosin	1284	151505279	9
Sebastes marinus	ocean perch (red fish)	Seb m 1.0101	Food Animal	Sebastes Seb m 1	1109	242253959	11
Sebastes marinus	ocean perch (red fish)	Seb m 1.0201	Food Animal	Sebastes Seb m 1	1110	242253961	11
Secale cereale	Rye	Sec c 20.0101	Food Plant	Secale Sec c 20	123	1699225	15
Secale cereale	Rye	Sec c 20.0201	Food Plant	Secale Sec c 20	129	1699228	15
Secale cereale	Rye	Sec c 38.0101	Food Plant	Secale Sec c 38.01	126	1944865	10
Secale cereale	Rye	Unassigned	Aero Plant	Secale Sec c 4	1520	15859456	17
Secale cereale	Rye	Unassigned	Aero Plant	Secale Sec c 4	1518	15859454	17

Scale cereale	Rye	Unassigned	Aero Plant	Scale Sec c 5	16	75140047	17
Scale cereale	Rye	Sec c 5.0101	Food Plant	Scale Sec c 5	1292	332205751	112
Sepia esculenta	cuttlefish	Unassigned	Food Animal	Sepia tropomyosin	1284	83715928	17
Sepioteuthis lessoniana	*Bigfin reef squid	Unassigned	Food Animal	Sepioteuthis tropomyosin	1284	83715930	17
Sesamum indicum	Sesame	Unassigned	Food Plant	Sesamum seed maturation-like protein	1345	171863012	116
Sesamum indicum	Sesame	Ses i 1	Food Plant	Sesamum Ses i 1	1153	13183175	17
Sesamum indicum	Sesame	Unassigned	Food Plant	Sesamum Ses i 1	1153	1209165427	110
Sesamum indicum	Sesame	Ses i 2	Food Plant	Sesamum Ses i 2	1148	5381323	17
Sesamum indicum	Sesame	Ses i 3	Food Plant	Sesamum Ses i 3	1585	13183177	17
Sesamum indicum	Sesame	Ses i 4.0101	Food Plant	Sesamum Ses i 4 oleosin	166	10834827	113
Sesamum indicum	Sesame	Unassigned	Food Plant	Sesamum Ses i 5 oleosin	145	198250943	110
Sesamum indicum	Sesame	Ses i 5.0101	Food Plant	Sesamum Ses i 5 oleosin	145	5381321	115
Sesamum indicum	Sesame	Ses i 6.0101	Food Plant	Sesamum Ses i 6	1459	5381325	115
Sesamum indicum	Sesame	Ses i 7.0101	Food Plant	Sesamum Ses i 7	1497	13183173	115
Sinapis alba	White mustard	Sin a 1	Food Plant	Sinapis Sin a 1.01	1145	1009434	17
Sinapis alba	White mustard	Sin a 1	Food Plant	Sinapis Sin a 1.01	1145	1009436	17
Sinapis alba	White mustard	Sin a 1	Food Plant	Sinapis Sin a 1.01	1145	1009438	17
Sinapis alba	White mustard	Sin a 1	Food Plant	Sinapis Sin a 1.01	1145	1009440	17
Sinapis alba	White mustard	Sin a 1	Food Plant	Sinapis Sin a 1.01	1145	1009442	17
Sinapis alba	White mustard	Sin a 1.0101	Food Plant	Sinapis Sin a 1.01	1145	51338768	17
Sinapis alba	White mustard	Sin a 2.0101	Food Plant	Sinapis Sin a 2.01 11S	1510	62240390	17
Sinapis alba	White mustard	Unassigned	Food Plant	Sinapis Sin a 2.01 11S	1523	62240392	17
Sinapis alba	White mustard	Sin a 3.0101	Food Plant	Sinapis Sin a 3.01 LTP	192	156778059	112
Sinapis alba	White mustard	Sin a 4.0101	Food Plant	Sinapis Sin a 4.01 profilin	1131	156778061	112
Sinonovacula constricta	*Chinese razor clam	Unassigned	Food Animal	Sinonovacula tropomyosin [Song paper]	1284	156145810	115
Solanum lycopersicum (Lycopersicon esculentum)	Tomato	Unassigned	Food Plant	Solanum lycopersicum Sola l 6	196	1460373045	116
Solanum tuberosum	Potato	Unassigned	Food Plant	Solanum profilin-like	1131	177416979	17
Solanum tuberosum	Potato	Unassigned	Food Plant	Solanum profilin-like	1131	177999277	17
Solanum lycopersicum (Lycopersicon esculentum)	Tomato	Sola l 1.0101	Food Plant	Solanum Sola l 1 profilin (Lyc e l)	1131	16555787	17
Solanum lycopersicum (Lycopersicon	Tomato	Lyc e 1	Food Plant	Solanum Sola l 1 profilin (Lyc e l)	1131	11724229	17



Solenopsis invicta	Red fire ant	Unassigned	Venom or Salivary	Solenopsis Sol i 1	126	1336813	17
Solenopsis invicta	Red fire ant	Sol i 1.0101	Venom or Salivary	Solenopsis Sol i 1	1346	51093373	17
Solenopsis invicta	Red fire ant	Sol i 2.0101	Venom or Salivary	Solenopsis Sol i and Sol r Venom allergen II	138	549179	17
Solenopsis richteri	Black fire ant	Sol r 2.0101	Venom or Salivary	Solenopsis Sol i and Sol r Venom allergen II	119	6136162	17
Solenopsis invicta	Red fire ant	Sol i 3.0101	Venom or Salivary	Solenopsis Venom allergen III	1234	2293571	111
Solenopsis richteri	Black fire ant	Sol r 3.0101	Venom or Salivary	Solenopsis Venom allergen III	1211	6136163	17
Sorghum halepense	Johnson grass	Sor h 1.0101	Aero Plant	Sorghum Sor h 1	1266	674275729	115
Sorghum halepense	Johnson grass	Sor h 1.0201	Aero Plant	Sorghum Sor h 1	1266	674275731	115
Sorghum halepense	Johnson grass	Sor h 13.0101	Aero Plant	Sorghum Sor h 13	1422	674275737	115
Sorghum halepense	Johnson grass	Sor h 13.0201	Aero Plant	Sorghum Sor h 13	1410	674275739	115
Stachybotrys chartarum	Fungus	Sta 3.0101	Aero Fungi	Stachybotrys Sta c 3	1144	253970748	114
Staphylococcus aureus	Bacteria	Unassigned	Bacteria skin	Staphylococcus enterotoxin SEA	1233	1633233	9
Staphylococcus aureus	Bacteria	Unassigned	Bacteria skin	Staphylococcus enterotoxin SEB	1254	83308249	9
Staphylococcus aureus	Bacteria	Unassigned	Bacteria skin	Staphylococcus enterotoxin SEC	1266	1462026	9
Staphylococcus aureus	Bacteria	Unassigned	Bacteria skin	Staphylococcus enterotoxin SED	1258	1119654	9
Staphylococcus aureus	Bacteria	Unassigned	Bacteria skin	Staphylococcus enterotoxin TSST 1	1234	1136457	9
Stemphylium callistephi	Fungus	Unassigned	Aero Fungi	Stemphylium major allergen alt al-like	1137	49476467	17
Stemphylium sp. CDI012	Fungus	Unassigned	Aero Fungi	Stemphylium major allergen alt al-like	1137	152060760	9
Stemphylium vesicarium	Fungus	Unassigned	Aero Fungi	Stemphylium major allergen alt al-like	1137	49476465	17
Strongyloides stercoralis	Parasitic nematode	Unassigned	Worm (parasite)	Strongyloides L3/10Ag. 01	229	5669875	17
Suidasia medanensis	Mite	Unassigned	Aero Mite	Suidasia putative Sui m141 12	1141	145738062	17
Sus scrofa	Pig	Unassigned	Aero Animal	Sus Porcine Pepsin	1385	1118572885	111
Syringia vulgaris	Lilac	Syr v 3.0101	Aero Plant	Syringae Syr v 3	181	11423847	17
Syringia vulgaris	Lilac	Syr v 1.0101	Aero Plant	Syringae Syr v I	1145	631911	17
Syringia vulgaris	Lilac	Syr v 1.0102	Aero Plant	Syringae Syr v I	1145	631912	17
Syringia vulgaris	Lilac	Syr v 1.0103	Aero Plant	Syringae Syr v I	1145	631913	17
Tabanus yao	Horse Fly	Tab y 1.0101	Venom or Salivary	Tabanus Tab y 1 Aprase	554	1323473390	112
Tabanus yao	Horse Fly	Tab y 2.0101	Venom or Salivary	Tabanus Tab y 2 Hyaluronidase	1349	1304273371	112

Tabanus yao	Horse Fly	Tab y 5.0101	Venom or Salivary	Tabanus Tab y 5	1266	304273369	112
Thaumetopoea pityocampa	Pine moth	Tha p 1.0101	Contact	Thaumetopoea Tha p 1	1126	301030229	112
Thaumetopoea pityocampa	Pine moth	Tha p 2.0101	Contact	Thaumetopoea Tha p 2	1115	408387552	114
Theragra chalcogramma	Alaska pollock	Unassigned	Food Animal	Theragra parvalbumin	1109	14531020	17
Theragra chalcogramma	Alaska pollock	Unassigned	Food Animal	Theragra parvalbumin	1109	14531018	17
Thunnus albacares	Yellowfin tuna	Thu a 2.0101	Food Animal	Thunnus Thu a 2 enolase	432	385145178	113
Thunnus albacares	Yellowfin tuna	Unassigned	Food Animal	Thunnus Thu a 2 enolase	12	576011132	115
Thunnus albacares	Yellowfin tuna	Unassigned	Food Animal	Thunnus Thu a 3 aldolase	364	291195949	112
Thunnus albacares	Yellowfin tuna	Thu a 3.0101	Food Animal	Thunnus Thu a 3 aldolase	37	576011088	115
Todarodes pacificus	Japanese flying squid	Unassigned	Food Animal	Todarodes Tod p 1	1284	83715932	17
Trachurus japonicus	Japanese horse mackerel	Unassigned	Food Animal	Trachurus parvalbumin	1107	77799800	17
Tresus keenae	clam	Unassigned	Food Animal	Tresus tropomyosin	1284	219806600	110
Triatoma protracta	Western conenose	Tri a p 1	Venom or Salivary	Triatoma Tri a p 1	1169	15426413	17
Arthroderma benhamiae	Fungus	Unassigned	Contact	Trichophyton (Arthroderma) Tri m 4	1726	23894232	17
Arthroderma vanbreuseghemii	Fungus	Unassigned	Contact	Trichophyton (Arthroderma) Tri m 4	1726	219687753	110
Arthroderma benhamiae	Fungus	Unassigned	Contact	Trichophyton (Arthroderma) Tri r 2	1292	23894240	17
Arthroderma benhamiae	Fungus	Unassigned	Contact	Trichophyton (Arthroderma) Tri r 2	1404	23894244	17
Trichophyton rubrum	Fungus	Tri r 2	Contact	Trichophyton (Arthroderma) Tri r 2	1412	5813790	17
Trichophyton schoenleinii	Fungus	Unassigned	Contact	Trichophyton (Arthroderma) Tri r 2	1405	174663809	112
Trichophyton rubrum	Fungus	Tri r 4	Contact	Trichophyton tri 4 allergen (Arthroderma)	1726	5813788	17
Trichophyton schoenleinii	Fungus	Unassigned	Contact	Trichophyton tri 4 allergen (Arthroderma)	1726	23894227	17
Triticum aestivum	Wheat	Unassigned	Aero Plant	Triticum Tri a 14 LTP-amylase inhibitor	1113	4417370	111
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum 5a2 protein	194	16840998	17
Triticum aestivum	Wheat	Unassigned	Aero Plant	Triticum sAI QM16.17	1143	195957140	110
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum sAI QM16.17	1143	121711	17
Triticum turgidum subsp. durum	Wheat	Unassigned	Food Plant	Triticum sAI QM16.17	1143	121916	17
Triticum aestivum	Wheat	Unassigned	Aero Plant	Triticum aestivum Tri a160	141	1827354945	116





Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum omega-5 gliadin Tri a 19	1346	208605348	110
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum omega-5 gliadin Tri a 19	1366	1508732623	115
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum putative leucine-rich repeat protein	1137	166840986	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum serine carboxypeptidase II	1260	166840984	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum serine carboxypeptidase II	1444	125987805	110
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Thaumatin-like	173	135917	112
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 12	1131	190684061	111
Triticum aestivum	Wheat	Tri a 12.0103	Food Plant	Triticum Tri a 12	1131	1548948852	114
Triticum aestivum	Wheat	Tri a 12.0101	Gladiin	Triticum Tri a 12	1131	1548948848	115
Triticum aestivum	Wheat	Tri a 12.0102	Gladiin	Triticum Tri a 12	1131	1548948850	115
Triticum aestivum	Wheat	Tri a 12.0104	Gladiin	Triticum Tri a 12	1131	1207366248	115
Triticum aestivum	Wheat	Tri a 15.0101	Gladiin	Triticum Tri a 15	1121	1283465829	111
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 20	1302	1170702	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 20	1291	1170708	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 20	1251	1170736	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 20	1327	1170738	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 20	1279	11063270	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 20	1285	162484809	17
Triticum aestivum	Wheat	Tri a 20.0101	Gladiin	Triticum Tri a 20	1279	1508732621	115
Triticum aestivum	Wheat	Tri a 21.0101	Gladiin	Triticum Tri a 21 alpha, beta-gliadin	1281	1283476402	111
Triticum aestivum	Wheat	Tri a 25.0101	Gladiin	Triticum Tri a 25	1125	18980491	115
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 26	1830	121743	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 26	1648	121751	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 26	1660	121779	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 26	139	121793	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 26	1705	122090	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 26	1815	1170743	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 26	1838	1736319	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 26	1101	1897811	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 26	1794	1508732625	115
Triticum aestivum	Wheat	Tri a 26.0101	Gladiin	Triticum Tri a 26	1848	1288860106	115
Triticum aestivum	Wheat	Tri a 26.0201	Gladiin	Triticum Tri a 26	1795	171084277	115

Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 27.0101	1203	30793446	17
Triticum aestivum	Wheat	Tri a 28.0101	Gladiin	Triticum Tri a 28	1119	66841026	17
Triticum aestivum	Wheat	Tri a 29.0101	Aero Plant	Triticum Tri a 29	1120	2537783731	111
Triticum aestivum	Wheat	Tri a 29.0201	Aero Plant	Triticum Tri a 29	1120	283465827	111
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 29	1145	121701	17
Triticum turgidum subsp. durum	Wheat	Unassigned	Food Plant	Triticum Tri a 29	1145	121920	17
Triticum aestivum	Wheat	Tri a 30.0101	Food Plant	Triticum Tri a 30	1168	121713	17
Triticum aestivum	Wheat	Tri a 31.0101	Food Plant	Triticum Tri a 31	1253	11124572	17
Triticum aestivum	Wheat	Unassigned	Aero Plant	Triticum Tri a 32	1218	190684059	111
Triticum aestivum	Wheat	Tri a 32.0101	Aero Plant	Triticum Tri a 32	1218	75324900	114
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 33	1399	1885350	17
Triticum aestivum	Wheat	Tri a 33.0101	Gladiin	Triticum Tri a 33	1398	15734506	115
Triticum aestivum	Wheat	Tri a 34.0101	Gladiin	Triticum Tri a 34 GAPDH	1337	2537783729	111
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1307	121773	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1356	121783	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1373	75317968	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1229	886963	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1261	886965	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1276	886967	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1285	175219081	17
Triticum aestivum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1326	162550933	17
Triticum aestivum	Wheat	Tri a 36.0101	Food Plant	Triticum Tri a 36	1369	133531566	112
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 36	1304	1170730	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 36	1323	1170732	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 36	1244	1170734	17
Triticum aestivum	Wheat	Unassigned	Gladiin	Triticum Tri a 36	1283	1508732827	115
Triticum turgidum subsp. durum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1295	121926	17
Triticum turgidum subsp. durum	Wheat	Unassigned	Food Plant	Triticum Tri a 36	1285	121930	17
Triticum aestivum	Wheat	Tri a 37.0101	Food Plant	Triticum Tri a 37 alpha	137	14007850	114
Triticum aestivum	Wheat			Triticum Tri a 39			

Triticum aestivum	Wheat	Unassigned	Aero Plant	serine proteinase inhibitor-lik	184	154101366	110
Triticum aestivum	Wheat	Unassigned	Aero Plant	serine proteinase inhibitor-lik	184	122065237	111
Triticum aestivum	Wheat	Tri a 39.0101	Aero Plant	serine proteinase inhibitor-lik	184	403213259	114
Tyrophagus putrescentiae	Dust mite	Unassigned	Aero Mite	Tyrophagus Tyr p 10	1284	148615631	9
Tyrophagus putrescentiae	Dust mite	Unassigned	Aero Mite	Tyrophagus Tyr p 10	1201	156938915	9
Tyrophagus putrescentiae	Dust mite	Tyr p 10.0101	Aero Mite	Tyrophagus Tyr p 10	1284	48249227	9
Tyrophagus putrescentiae	Dust mite	Tyr p 13	Aero Mite	Tyrophagus Tyr p 13	1131	151860756	17
Tyrophagus putrescentiae	Dust mite	Unassigned	Aero Mite	Tyrophagus Tyr p 13	1130	121296500	9
Tyrophagus putrescentiae	Dust mite	Unassigned	Aero Mite	Tyrophagus Tyr p 13	1131	156938917	9
Tyrophagus putrescentiae	Dust mite	Tyr p 2	Aero Mite	Tyrophagus Tyr p 2	1141	2182106	17
Tyrophagus putrescentiae	Dust mite	Tyr p 24.0101	Aero Mite	Tyrophagus Tyr p 24	1153	219815476	111
Tyrophagus putrescentiae	Dust mite	Tyr p 3.0101	Aero Mite	Tyrophagus Tyr p 3	1285	167540622	111
Tyrophagus putrescentiae	Dust mite	Unassigned	Aero Mite	Tyrophagus Tyr p 8	1218	452215228	114
Venerupis philippinarum	Clam	Unassigned	Food Animal	Venerupis tropomyosin	1284	219806573	110
Vespa affinis	Lesser banded hornet	Unassigned	Food Animal	Vespa affinis Phospholipase A1	1334	157601175	115
Vespa affinis	Lesser banded hornet	Unassigned	Food Animal	Vespa affinis Phospholipase A1	1334	157601171	115
Vespa magnifica	Hornet	Unassigned	Venom or Salivary	Vespa magnifica Vesp m1	357	1315133295	112
Vespa magnifica	Hornet	Unassigned	Venom or Salivary	Vespa magnifica Vesp m1	225	1319801357	112
Vespa crabro	European hornet	Unassigned	Venom or Salivary	Vespa Vesp c 1 phospholipase	1301	1313471397	112
Vespa crabro	European hornet	Vesp c 5.0101	Venom or Salivary	Vespa Vesp c 5	1202	1549184	17
Vespa crabro	European hornet	Vesp c 5.0102	Venom or Salivary	Vespa Vesp c 5	1202	1549185	17
Vespa mandarinia	Wasp	Vesp m 5.0101	Venom or Salivary	Vespa Vesp c 5	1202	16136165	17
Vespula germanica	Wasp	Unassigned	Venom or Salivary	Vespula Phospholipase A1- Vesp m/v 1	1300	174035843	17
Vespula saculifrons	Wasp	Ves m 1.0101	Venom or Salivary	Vespula Phospholipase A1- Vesp m/v 1	1300	11709545	18

Vespula vulgaris	Wasp	Ives v 1.0101	Venom or Salivary	Vespula Phospholipase A1- Ves m/v 1	336	897647	17
Vespula flavopilosa	Wasp	Ives f 5.0101	Venom or Salivary	Vespula Ves f 5	1204	549189	17
Vespula germanica	Wasp	Ives g 5.0101	Venom or Salivary	Vespula Ves f 5	1204	549190	17
Vespula germanica	Wasp	Unassigned	Venom or Salivary	Vespula Ves f 5	1204	74035841	17
Vespula maculifrons	Wasp	Ives m 5.0101	Venom or Salivary	Vespula Ves f 5	1204	549191	17
Vespula maculifrons	Wasp	Unassigned	Venom or Salivary	Vespula Ves f 5	1227	85681830	17
Vespula pennsylvanica	Wasp	Ives p 5.0101	Venom or Salivary	Vespula Ves f 5	1204	549192	17
Vespula squamosa	Wasp	Ives s 5.0101	Venom or Salivary	Vespula Ves f 5	1205	549193	17
Vespula vidua	Wasp	Ives vi 5.0101	Venom or Salivary	Vespula Ves f 5	1206	549194	17
Vespula vulgaris	Wasp	Ives v 5.0101	Venom or Salivary	Vespula Ves f 5	1227	162551	17
Vespula vulgaris	Wasp	Ives v 5	Venom or Salivary	Vespula Ves f 5	1204	4826574	17
Vespula vulgaris	Wasp	Ives v 5	Venom or Salivary	Vespula Ves f 5	1209	11514279	17
Vespula maculifrons	Wasp	Unassigned	Venom or Salivary	Vespula Ves m 2 Hyaluronidase	31	313118253	112
Vespula squamosa	Wasp	Unassigned	Venom or Salivary	Vespula Ves s 1 phospholipase	1298	313471398	112
Vespula germanica	Wasp	Unassigned	Venom or Salivary	Vespula Ves v 2	331	116174180	8
Vespula germanica	Wasp	Unassigned	Venom or Salivary	Vespula Ves v 2	323	116174182	8
Vespula vulgaris	Wasp	Ives v 2	Venom or Salivary	Vespula Ves v 2	331	11346323	17
Vespula vulgaris	Wasp	Ives v 2.0101	Venom or Salivary	Vespula Ves v 2	340	62147665	17
Vespula vulgaris	Wasp	Unassigned	Venom or Salivary	Vespula Ves v 2	331	109157163	8
Vespula vulgaris	Wasp	Ives v 3.0101	Venom or Salivary	Vespula Ves v 3 dipeptidylpeptidase IV	776	167782086	9
Vigna radiata	mung bean	Vig r 1.0101	Food Plant	Vigna Vig r 1 PR 10	155	60418924	17
Vigna radiata	mung bean	Vig r 2.0101	Food Plant	Vigna Vig r 2	453	108743976	115
Vigna radiata	mung bean	Vig r 2.0201	Food Plant	Vigna Vig r 2	454	158251953	115
Vigna radiata var. radiata	mung bean	Vig r 4.0101	Food Plant	Vigna Vig r 4	272	1000708	115
Vigna radiata	mung bean	Vig r 6.0101	Food Plant	Vigna Vig r 6 Cytokinin-specific binding protein	155	4190976	114
Vitis sp.	Grape	Unassigned	Food Plant	Vitis Lipid transfer protein P3	191	145559502	8
Vitis sp.	Grape	Vit v 1	Food Plant	Vitis Vit v 1 LTP	37	462719	17
Vitis sp.	Grape	Unassigned	Food Plant	Vitis Vit v 1 LTP	38	462717	17
Xiphias gladius	Swordfish	Xip g 1.0101	Food Animal	Xiphias Xip g 1 beta-parvalbumin	109	222352960	110
Zea mays	Corn	Unassigned	Aero Plant	Zea group 13 pollen allergen	1410	89892725	17
Zea mays	Corn	Unassigned	Aero Plant	Zea group 13 pollen	1404	89892727	17

Zea mays	Corn	Unassigned	Aero Plant	Zea group 13 pollen allergen	1411	89892729	17
Zea mays	Corn	Unassigned	Aero Plant	Zea pollen specific protein	1170	1588669	17
Zea mays	Corn	Zea m 1.0101	Aero Plant	Zea Zea m 1 beta-expansin	1269	28630919	17
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 beta-expansin	1269	28630923	17
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 beta-expansin	1269	14193761	18
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 beta-expansin	1245	114794319	18
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 isoform	1263	89892721	17
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 isoform	1252	89892723	17
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 isoform	199	105969643	18
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 isoform	1269	105969645	18
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 isoform	1270	115502167	19
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 1 isoform	1269	115502168	19
Zea mays	Corn	Zea m 12.0104	Food Plant	Zea Zea m 12 profilin	131	2842324	17
Zea mays	Corn	Unassigned	Food Plant	Zea Zea m 12 profilin	131	110644952	18
Zea mays	Corn	Unassigned	Food Plant	Zea Zea m 12 profilin	131	110644954	18
Zea mays	Corn	Unassigned	Food Plant	Zea Zea m 12 profilin	131	110644956	18
Zea mays	Corn	Unassigned	Food Plant	Zea Zea m 12 profilin	131	110644958	18
Zea mays	Corn	Unassigned	Food Plant	Zea Zea m 12 profilin	131	110644960	18
Zea mays	Corn	Unassigned	Food Plant	Zea Zea m 12 profilin	131	110644962	18
Zea mays	Corn	Unassigned	Food Plant	Zea Zea m 12 profilin	130	110644964	18
Zea mays	Corn	Zea m 12.0101	Food Plant	Zea Zea m 12 profilin	131	1313138	15
Zea mays	Corn	Zea m 12.0102	Food Plant	Zea Zea m 12 profilin	137	1313140	15
Zea mays	Corn	Zea m 12.0103	Food Plant	Zea Zea m 12 profilin	131	1313142	15
Zea mays	Corn	Zea m 12.0105	Food Plant	Zea Zea m 12 profilin	131	11493677	15
Zea mays	Corn	Zea m 14.0101	Food Plant	Zea Zea m 14	120	168576	15
Zea mays	Corn	Zea m 14.0102	Food Plant	Zea Zea m 14	199	168578	15
Zea mays	Corn	Unassigned	Aero Plant	Zea Zea m 25 thioredoxin	128	16841002	17
Ziziphus mauritiana	Chinese-date	Ziz m 1.0101	Food Plant	Ziziphus Ziz m 1	330	16125281	17

## **E Results from the EFSA scientific opinion recommended allergen analysis of lactase produced by PP3930 using allergenonline database**

### **E.1 35% or larger identity over any 80 amino acid window**

(blank=No matches found) Count of significant hits described in text based on identity > 35%.

### **E.2 35% or larger identity over any 80 amino acid window (with scaling)**

(blank=No matches found) Count of significant hits described in text based on identity > 35%.

—

Esben Peter Friis <epf@novozymes.com>

February 23, 2016

**Toxicology & Product Safety**

Date: July 30, 2014  
File: 2014-11425-01  
Ref.: LiNi

**SUMMARY OF TOXICITY DATA**

**Lactase, batch PPL34537 from *Bacillus licheniformis***

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## 1. ABSTRACT

The below series of toxicological studies were undertaken to evaluate the safety of Lactase, batch PPL34537.

All studies were carried out in accordance with current OECD guidelines and in compliance with the OECD principles of Good Laboratory Practice (GLP). The studies were performed at Novozymes A/S (Denmark), Huntingdon Life Sciences (UK) and Covance Laboratories Ltd. (UK) during the period November 2013 to August 2014.

The main conclusions of these studies can be summarized as follows:

- Lactase, batch PPL34537 did not induce gene mutations in the Ames test, neither in the presence nor absence of S-9 mix.
- Lactase, batch PPL34537 did not cause an increase in the induction of micronuclei in cultured human lymphocytes in this *in vitro* micronucleus test using human lymphocytes, either in the presence nor in the absence of S-9 mix.
- Daily oral administration (by gavage) of Lactase, batch PPL34537 to rats at dosages of up to 10.0 mL/kg bw/day for thirteen weeks resulted in no adverse treatment-related effects. Consequently, the No Observed Adverse Effect level (NOAEL) was considered to be 10 mL/kg bw/day (equivalent to 0.672 g TOS/kg bw/day or 70665 LAU(B)/kg bw/day).

Based on the present toxicity data it can be concluded that Lactase, represented by batch PPL34537, exhibits no toxicological effects under the experimental conditions described.

## 2. TEST SUBSTANCE

Lactase is a liquid enzyme concentrate containing a lactase (E.C. number 3.2.1.23), which catalyses the hydrolysis of lactose into galactose and glucose.

### 2.1 Production organism

The production organism is a genetically modified strain of *Bacillus licheniformis*.

*B. licheniformis* has a long history of safe use for the production of food enzymes and is recommended for QPS (Qualified Presumption of Safety) by EFSA under the condition that absence of toxigenic potential is verified. Absence of toxigenic potential has been verified for the parental strain. The recipient *B. licheniformis* strain was developed from the parental strain through a series of modification steps including classical mutagenesis and genetic modification. The classical mutagenesis steps resulted in a strain with reduced spore-forming ability.

## 2.2 Characterization

The toxbatch Lactase, batch PPL34537 was used for the conduct of all the toxicological studies. The characterization of the toxbatch is presented in Table 1.

Table 1. Characterization data of Lactase, batch PPL34537

Batch number	PPL34537
Activity	3260 LAU(A)/g
	6730 LAU(B)/g
Water (KF) (% w/w)	90.7
Dry matter (% w/w)	9.3
Ash (% w/w)	2.9
Total Organic Solids (TOS <sup>1</sup> ) (% w/w)	6.4
Specific gravity (g/mL)	1.050

<sup>1</sup> % TOS is calculated as 100% - % water - % ash - % diluents.

## 3. MUTAGENICITY

### 3.1 Bacterial Reverse Mutation assay (Ames test)

Lactase, batch PPL34537 was examined for mutagenic activity in the bacterial reverse mutation assay using *Salmonella typhimurium* strain TA1535, TA100, TA1537, and TA98 and *Escherichia coli* WP2uvrApKM101. The study was carried out according to the OECD test guideline 471 (adopted in 1997) and in compliance with GLP.

Crude enzyme preparations, like the present batch of Lactase, contain the free amino acids histidine and tryptophan, most often in an amount, which exceeds the critical concentration for incorporation in the direct standard assay. To overcome this problem all strains were exposed to Lactase in liquid culture known as “treat and plate assay”.

Two independent experiments were performed, with and without the inclusion of metabolic activation (S-9 mix). In each experiment cultures of bacteria were exposed to six doses of the test substance (5000, 2500, 1250, 625, 313, and 156 µg dry matter/mL) in a phosphate buffered nutrient broth for 3 hours. After incubation, the test substance was removed by centrifugation prior to plating.

Lactase contains an abundance of various nutrients, and composes a rich growth medium to the test bacteria. These circumstances are reflected in the present study, as the viability was somewhat reduced at the highest dose levels in several test series with *Salmonella* strains. On the contrary growth stimulation was observed in the test series with the *E. coli* strain as demonstrated by increases in the viable count of exposed cultures compared to the solvent control. However, these conditions had no significant influence on the revertant colony count.

No treatments of any of the *Salmonella* and *E. coli* strains with Lactase resulted in any increases in revertant numbers that meet the criteria for a positive or equivocal response.

It was concluded that Lactase, batch PPL34537 did not induce gene mutations in bacteria either in the presence or absence of metabolic activation when tested under the conditions employed in this study.

### 3.2 *In vitro* Micronucleus assay

In order to assess the clastogenic and the aneugenic activity of Lactase, batch PPL34537 its effects on the frequency of micronuclei was investigated in cultured human peripheral blood lymphocytes applying the cytokinesis-block methodology.

The study was conducted according to GLP and in compliance with the OECD guideline: Guideline 487: In vitro micronucleus test (Adopted in 2010).

Heparinized whole blood cultures, pooled from two male donors, were established, and division of the lymphocytes was stimulated by adding phytohaemagglutinin (PHA) to the cultures.

Treatments covering a broad range of concentrations, separated by narrow intervals, were performed both in the absence and presence of metabolic activation (S-9) from Aroclor 1254-induced rats. The test article was formulated in water for irrigation (purified water) and the highest concentration tested in the Micronucleus Experiment, 5000 µg test substance (weighed out as received)/mL, was determined following a preliminary cytotoxicity Range-Finder Experiment.

Treatments were conducted for 48 hours following mitogen stimulation by phytohaemagglutinin (PHA). The test article concentrations for micronucleus analysis were selected by evaluating the effect of Lactase, batch PPL34537 on the replication index (RI).

Appropriate negative (vehicle) control cultures were included in the test system under each treatment condition. The proportion of micronucleated binucleate (MNBN) cells in the vehicle cultures fell within current 95-percentile of the observed historical vehicle control (normal) ranges. Mitomycin C (MMC) and Vinblastine (VIN) were employed as clastogenic and aneugenic positive control chemicals respectively in the absence of rat liver S-9.

Cyclophosphamide (CPA) was employed as a clastogenic positive control chemical in the presence of rat liver S-9. Cells receiving these were sampled in the Micronucleus Experiment at 24 hours (CPA, MMC) or 48 hours (VIN) after the start of treatment. All positive control compounds induced statistically significant increases in the proportion of cells with micronuclei. All acceptance criteria were considered met and the study was therefore accepted as valid.

Treatment of cells with Lactase, batch PPL34537 in the absence and presence of S-9 resulted in frequencies of MNBN cells which were similar to and not significantly ( $p \leq 0.05$ ) higher than those observed in concurrent vehicle controls for the majority of all concentrations analysed (all treatments). A single exception was observed at the lowest concentration analysed (3000 µg/mL) following 3+21 hour exposure in the absence of S-9 treatment. However, this increase was not large with both replicate cultures exhibiting MNBN cell frequencies within the historical vehicle control (normal) range with no instances of MNBN cell values exceeding normal values for any other Lactase, batch PPL34537 treated culture (all concentrations, all treatments). As such, this isolated statistical increase was not considered of biological importance.

It was concluded that Lactase, batch PPL34537 did not induce micronuclei in cultured human peripheral blood lymphocytes following treatment in the absence and presence of an aroclor induced rat liver metabolic activation system (S-9).

## 4. GENERAL TOXICITY

### 4.1 13 Week Oral Toxicity Study in Rats

The objective of this study was to assess the systemic toxic potential of Lactase, batch PPL34537, when administered orally by gavage to Sprague-Dawley (CrI:CD(SD)) rats for 13 weeks. Three groups, each comprising 10 males and 10 females, received doses of 10, 33 or 100% of the Lactase batch (equivalent to 0.067, 0.222 or 0.672 g TOS/bw kg/day, or 7067, 23319 or 70665 LAU(B)/kg bw/day). A similarly constituted control group received the vehicle (reverse osmosis water) at the same volume-dose (10 mL/kg bw).

During the study, clinical condition, detailed physical and arena observations, sensory reactivity, grip strength, motor activity, body weight, food consumption, water consumption (by visual assessment), ophthalmic examination, haematology (peripheral blood), blood chemistry, organ weight, macropathology and histopathology investigations were undertaken.

General appearance and behaviour, sensory reactivity responses, grip strength and motor activity were not affected by treatment, there were no deaths during the treatment period and there was no effect of treatment on bodyweight gain or on food and water consumption. There were no treatment-related ophthalmic findings.

There were no treatment-related haematological findings. Organ weights were unaffected and there were no treatment-related macroscopic or microscopic findings.

Analysis of the blood plasma during Week 13 revealed high alkaline phosphatase activity in males receiving 33% of the Lactase batch and in both sexes receiving 100% of the Lactase batch and low urea and creatinine and high potassium concentrations in males receiving 100% of the Lactase batch. However, as there was no change in organ weight or any macroscopic or microscopic findings and no other biochemical markers for organ damage were observed, these findings were considered most likely to represent adaptive changes associated with administration of high doses of a xenobiotic and were therefore considered of no toxicological significance.

It was thus concluded that oral administration of Lactase, batch PPL34537, to Sprague-Dawley rats at doses up to 100% of the Lactase batch (0.672 g TOS/kg bw/day or 70665 LAU(B)/kg bw/day) for 13 weeks was well-tolerated and did not cause any adverse change. The no-observed-adverse-effect level (NOAEL) was considered to be 100% of the toxbatch (equivalent to 0.672 g TOS/kg bw/day or 70665 LAU(B)/kg bw/day).

## 5. REFERENCES

### 5.1 Study reports

Novozymes A/S: Study No.: 20138037. Lactase, batch PPL34537: Test for mutagenic activity with strains of *Salmonella typhimurium* and *Escherichia coli*. (December 2013). LUNA file: 2013-17652.

Covance Laboratories: Study No.: 8292559. Novozymes Reference No.: 20136071: Lactase, batch PPL34537: Induction of micronuclei in cultured human peripheral blood lymphocytes. (January 2014). LUNA file: 2014-00365.

Huntingdon Life Sciences: Study No.: LKG0074. Novozymes Reference No.: 20136077: Lactase, batch PPL34537: Toxicity Study by Oral Gavage Administration to Sprague-Dawley Rats for 13 Weeks. LUNA file: 2014-11424.

**Toxicology & Product Safety**

Date: 27 September 2013  
Project: DEV00917  
File: 2013-17652-01  
Ref.: PBjP/PSCK

## **R E P O R T**

**Lactase, batch PPL34537:  
Test for Mutagenic Activity with Strains of  
*Salmonella typhimurium* and *Escherichia coli*.**

**Study No. 20138037**

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## GLP Compliance Statement and Study Director Authentication

**REPORT:** Lactase, batch PPL34537: Test for Mutagenic Activity with Strains of *Salmonella typhimurium* and *Escherichia coli*

**STUDY NO.:** 20138037

The study described in this report was conducted at the department of Toxicology & Product Safety, Novozymes A/S in compliance with the OECD principles of Good Laboratory Practice OECD, ENV/MC/CHEM(98)17 (as revised 1997 and issued January 1998).

This report is a complete and accurate account of the methods employed and the results obtained.

9 Dec 2013

Date

Study Director  
Novozymes A/S



## QUALITY ASSURANCE STATEMENT

REPORT: Lactase, batch PPL34537:  
Test for Mutagenic activity with strains of Salmonella  
typhimurium and Escherichia coli.

STUDY NUMBER 20138037

The conduct of this study has been subject to appropriate inspections and the report has been reviewed according to the relevant Standard Operation Procedures of Novozymes A/S Quality Assurance.

Inspection/Audit	Dates of inspection	Inspection results reported to Study Director and Study Management
Protocol	10 JUL 2013	10 JUL 2013
Analysis	16 JUL 2013	17 JUL 2013
Report	21 NOV 2013	21 NOV 2013

I hereby confirm that the report reflects the raw data.

2 dec. 2013

Date

  
Quality Assurance

## 1. General Information

**STUDY** Lactase, batch PPL34537: Test for Mutagenic Activity with Strains of *Salmonella typhimurium* and *Escherichia coli*.  
Study No. 20138037

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**DATES OF STUDY**  
Study initiation date: 9 July 2013  
Experimental start date: 15 July 2013  
Experimental completion: 12 August 2013

The study completion date is the date the final report is signed by the Study Director.

## 2. Summary

Lactase (Batch Number: PPL34537) was examined for mutagenic activity in the bacterial reverse mutation assay using *Salmonella typhimurium* strain TA1535, TA100, TA1537, TA98 and *Escherichia coli* WP2uvrApKM101.

Crude enzyme preparations, like the present batch of Lactase contain the free amino acids histidine and tryptophan, most often in an amount, which exceeds the critical concentration for incorporation in the direct standard assay.

To overcome this problem all strains were exposed Lactase in liquid culture ("treat and plate assay"). Bacteria were exposed to 6 doses of the test substance in a phosphate buffered nutrient broth for 3 hours with 5 mg (dry matter) per mL as highest concentration. After incubation the test substance was removed by centrifugation prior to plating.

The study was conducted with and without the metabolic activation system S9 - a liver preparation from male rats, pre-treated with Aroclor 1254, and the co-factors required for mixed function oxidase activity (S9 mix). All results were confirmed by conducting two complete and independent experiments.

Lactase contains an abundance of various nutrients, and composes a rich growth medium to the test bacteria. These circumstances are reflected in the present study.

In the present study the viability was somewhat reduced at the highest dose levels in several test series with *Salmonella* strains. On the contrary growth stimulation was observed in the test series with the *E.coli* strain as demonstrated by increases in the viable count of exposed cultures compared to the solvent control. However, these conditions had no significant influence on the revertant colony count.

No treatments of any of the *Salmonella* and *E.coli* strains with Lactase resulted in any increases in revertant numbers that meet the criteria for a positive or equivocal response.

It is concluded that the results of the bacterial mutagenicity tests described in this report give no indication of the presence of mutagenic components in this preparation of Lactase (Batch No. PPL34537), when tested under the conditions applied in this study.



### 3. Introduction

Bacterial reverse mutation assays have been recognized and used for more than three decades as a rapid, sensitive and reliable method of evaluating the mutagenic potential of chemicals. Bacterial systems offer several advantages to other test systems. They can be grown in large numbers in a short time, enabling the detection of very rare mutational events. Further, extensive knowledge of bacterial genetics has allowed the construction of special strains, which are more sensitive than wild-type strains are to a variety of agents. The reversion of bacteria from growth-dependence on a particular amino acid to growth in the absence of that amino acid is the most widely used marker in reverse-mutation assays. The genetic target is small, specific and selective, and the phenotypic effect of the reverse mutation is easily detected.

A wide range of strains within the species *Salmonella typhimurium* (Ames strains) and *Escherichia coli* have been constructed in order to make the test system more sensitive and selective to different classes of chemical mutagens.

By incorporation of the post-mitochondrial supernatant (S9) from the livers of rats pre-treated with an enzyme inducer Aroclor 1254, the metabolising systems present in mammalian cells are mimicked to facilitate the detection of a wide range of pro-mutagens.

This report describes experiments performed to assess the activity of Lactase (Batch Number: PPL34537) in amino acid dependent strains of *Salmonella typhimurium* and *Escherichia coli* capable of detecting both induced frame-shift (TA1537 and TA98) and base-pair substitution mutations (TA1535, TA100, and *E. coli* WP2uvrApKM101).

Lactase (Batch Number: PPL34537) is a microbial enzyme preparation derived from submerged pure culture fermentation of a non-pathogenic and non-toxicogenic strain. It contains a variety of unspent medium residues, including low concentrations of free amino acids like histidine and tryptophan.

This complexity poses several problems during mutagenicity testing in vitro. In the Ames test it composes a rich growth medium to the test bacteria, resulting in completely different and poorly defined environments of exposed cultures compared to control cultures. The main problem is the content of utilizable histidine and tryptophan in the test material, since the principle of the Ames test is the histidine auxotrophy of the *Salmonella* strains and tryptophan auxotrophy of the *E. coli* strains.

As a result, the density of the bacterial background lawn increase with increasing doses ("feeding effect") followed by dose related increases in the number of spontaneous revertant. These increases are obviously artificial.

To overcome this problem all strains applied in the present study were treated with Lactase in liquid culture ("treat and plate assay").

Two complete and independent experiments were conducted with all five bacterial strain. Additionally, a third supplementary test series was carried out with TA1535 and TA1537 with incorporation of metabolic activation.

The study was conducted in accordance with the general recommendations in OECD Guideline for testing of chemicals, No. 471: Bacterial Reverse Mutation Assay" (July 1997 concerning the general specifications of the test. However the exposure of test bacteria in liquid culture ("treat and plate") is not specifically described in any guidelines.

### 4. Materials

#### 4.1 Test substance

A sample of Lactase (Batch Number: PPL34537, TKS 12114) was received from Recovery Pilot Plant; Novozymes A/S 3 december 2012, and immediately stored in a

freezer. The substance was a brown liquid with a declared content of 9.3 % (w/w) dry matter.

A standard solution of 5% (w/v) dry matter was prepared in deionized water and sterilized by filtration. The sterility was confirmed by plate counting. Solution was stored refrigerated and used as test substance.

#### 4.2 Positive control substances

Chemical	Source	Lot.No.
2-Nitrofluorene (2-NF)	Sigma-Aldrich	S 43858 V
Acridine mutagen (ICR-191)	Sigma	110M1173W
1-Methyl-3-Nitro-N-NitrosoGuanidine (MNNG)	Aldrich-Chemie	15427 LO
2-Aminoanthracene (2-AA)	Sigma-Aldrich	STBB1901

All positive control substances were dissolved in dimethyl sulphoxide (spectrophotometric grade).

#### 4.3 Liver homogenate – S9

A commercial preparation of S9 from Aroclor 1254 induced Sprague Dawley rats was obtained from Cappel/MP Biomedicals, LLC, 29525 Fountain Parkway, Solon, Ohio 44139. Specifications of the preparation, the enzymatic properties and metabolic activation from the supplier are archived as raw data.

The tubes with S9 were received frozen in dry ice and were immediately stored in a  $-80^{\circ}\text{C}$  ultra low freezer at Toxicology & Product Safety, Novozymes.

#### 4.4 Plates

As selective substrate for reverted bacteria Vogel-Bonner medium E agar plates with 2% glucose were prepared in-house as described in Appendix 3.

All plates were stored refrigerated in closed plastic bags and examined for contamination and dryness before use.

#### 4.5 Bacteria

##### *Salmonella typhimurium*

Four strains of *Salmonella typhimurium* were used:

*S. typhimurium* TA1535

*S. typhimurium* TA100

*S. typhimurium* TA1537

*S. typhimurium* TA98

All these strains contain mutations in the histidine operon, thereby imposing a requirement for histidine in the growth medium. They all contain GC base-pairs at the site of the histidine mutation, and are therefore selective for agents which react predominantly with these bases. Three mutations in the histidine operon are involved:

The *hisG46* mutation (TA1535 and TA100) is a missense mutation which is reverted to prototrophy by a variety of mutagens that cause base-pair substitutions.

The *hisC3076* mutation (TA1537) contains a frame-shift which appears to have added a GC base-pair. This mutation is reverted for example by ICR-191 and epoxides of polycyclic hydrocarbon. The *hisD3052* mutation (TA98) also contains a frame-shift mutation with a sequence of repeated GC, which is reverted with the deletion of two of these base-pairs. It is readily reverted by aromatic amines and derivatives.

All four strains contain the deep rough (*rfa*) mutation, which deletes the polysaccharide side chain of the polysaccharide coat of the bacterial cell surface. This deletion increases cell permeability to more hydrophobic substances and, furthermore, greatly decreases the pathogenicity of these organisms.



The *uvrB* deletion renders the strains incapable of excision repair, making them more sensitive both to the mutagenic and lethal effects of a wide variety of mutagens (e.g. poly-aromatic hydrocarbons), since the strains cannot excise DNA adducts. These two deletions include the nitrate reductase (*chl*) and biotin (*bio*) genes also.

Strain TA98 and TA100 are derived from strain TA1538 and TA1535 respectively by the addition of a plasmid, pKM101, which confers resistance to ampicillin. This plasmid also carries a gene (*muc<sup>+</sup>*), which in some strains (*recA<sup>+</sup>/lexA<sup>+</sup>*) have proven to participate in "SOS" DNA-repair. This repair pathway is induced by DNA damage and confers resistance to the lethal effects of many mutagens at the expense of increased mutability. Bacteria carrying pKM101 have therefore a higher spontaneous mutation rate.

### **Escherichia coli**

One strain was used:

*Escherichia coli* WP2uvrApKM101

This strain contains an ochre mutation in the *trpE* locus and can be mutated to tryptophan independence either by a base-pair reversion of an A-T base-pair in the *trpE* locus, or more likely, by a base-pair substitution within a number of transfer RNA loci elsewhere in the chromosome. The latter causes the original defect to be suppressed (*ochre* suppression) and involves only base-pair substitution transitions at G-C base-pairs.

Like the *uvrB* mutation in the *Salmonella* strains, the *uvrA* mutation causes the bacteria to be deficient in the excision of bulky lesions from the DNA, so, it is more readily mutated by certain agents (ultraviolet radiation, polycyclic hydrocarbons). Further the strain contains the pKM101 plasmid as described above for the *Salmonella* strains.

## **4.6 Bacterial cultures**

The test strains of *Salmonella typhimurium* LT2 were obtained from Prof. B.N. Ames, Biochemistry Department, University of California, Berkeley, CA 94720, U.S.A.

*Escherichia coli* WP2uvrApKM101 was obtained from Covance Laboratories Ltd, Otley road, Harrogate, North Yorkshire HG3 1PY, England.

New batches of culture stocks frozen in 8% dimethyl sulphoxide are prepared at intervals from a central stock held in a  $\pm 150^{\circ}\text{C}$  freezer. They are regularly checked for appropriate amino acid requirement, spontaneous reversion rate, genetic characters and response to diagnostic mutagens.

Samples of each strain were grown up overnight in Nutrient broth in a  $37 \pm 1^{\circ}\text{C}$  water bath with shaking. Fresh cultures were prepared before each test.

## **4.7 S9 mix**

Composition of a 10% V/V S9 mix (final concentrations):

Co-factors:

-phosphate buffer (0.2M, pH 7.4) .....	100 mM
-salts (1.65M KCl, 0.4 M MgCl <sub>2</sub> ) .....	33 and 8 mM
-glucose-6-phosphate, mono-Na salt (0.2M) .....	5 mM
-NADP, di-Na salt (0.1M) .....	4 mM
S9 preparation .....	10% V/V

A freshly prepared solution of the co-factors mixed 9:1 (v/v) with freshly thawed still cold S9 preparation. This S9 mix was prepared freshly each day, and immediately used. Unused reagent was discarded.

#### 4.8 Dilution of test substance

Serial dilutions of a sterile standard solution of 5% dry matter (w/v) Lactase batch PL34537 were prepared in sterile deionised water corresponding to the final dose levels:

5000 µg - 2500 µg - 1250 µg - 625 µg - 313 µg - 156 µg dry matter per mL.

The dilutions were prepared freshly immediately prior to use and applied in experiments in the presence and absence of S9 mix.

#### 4.9 Top agar

0.6 % soft agar was sterilised by autoclaving.

Bottles with 200 mL melted soft agar were kept at about 55°C and added 20 mL 0.5 mM L-histidine/biotin solution for strains of *Salmonella* or 20 mL 0.5 mM tryptophan solution for *Escherichia coli*. This molten agar was divided into 2 mL aliquots in sterile glass tubes and placed in a heat-block at  $45 \pm 1^\circ\text{C}$ .

### 5. Methods

#### 5.1. Treat and plate assay

This procedure was applied with all strains.

For each assay sterile tubes were added:

- 4 mL Nutrient broth
- 4 mL S9 mix or 0.2M phosphate buffer (pH 7.4)
- 1 mL bacterial culture
- 1 mL test substance solution (6 doses) or diagnostic mutagen solution (positive control) or sterile deionised water (solvent control).

These incubation mixtures were incubated with shaking at  $37 \pm 1^\circ\text{C}$  for 3 hours.

After incubation all bacterial suspensions were washed two times by centrifugation for 10 minutes at 1272 X g. After the first washing the bacterial pellets were resuspended in 5 mL phosphate buffer (pH 7.4, 0.2M) and finally they were re-suspended in 1 mL phosphate buffer.

Tubes with 2 mL of top agar were added 0.1 mL of all washed bacterial suspensions.

#### 5.2 Selective incubation and scoring

For each dose of the test substance and the standard mutagens three similar tubes with top agar were prepared and five tubes were prepared for the solvent control.

These tubes were poured on to minimal glucose agar plates. When the soft agar set, the plates were inverted and incubated at  $37 \pm 2^\circ\text{C}$  for about 72 hours.

After incubation the numbers of revertant colonies were counted automatically (Perceptive Instruments). Plates with less than about 20 colonies were counted manually.

#### 5.3 Viable cell count

0.1 mL aliquots of a  $10^{-6}$  dilution of each bacterial suspension were poured on to minimal glucose agar plates (added the required amino acids in excess) in duplicates. When the soft agar set, the plates were inverted and incubated at  $37 \pm 2^\circ\text{C}$  for about 72 hours after which they were scored as described above.

#### 5.4 Controls

The following controls were run with each experiment:

##### Genotype checking:

- Sensitivity for crystal violet: *rfa* (all *Salmonella* strains)
- Sensitivity for Mitomycin C: *uvrB* (all *Salmonella* strains) and *uvrA* (*E. coli* WP2uvrApKM101)
- Resistance to ampicillin: *Salmonella* TA98 and TA100 and *E.coli* uvrApKM101



0.1 mL bacterial culture was spread on to nutrient agar medium. To the surface of the dried plate was added a disc of ampicillin/(Rosco Neo-Sensitabs) and two 9 mm  $\phi$  sterile filter discs, one with 10 $\mu$ l 0.1% crystal violet and the other with 10 $\mu$ l 0.01% Mitomycin C. The plate was incubated for about 72 hours at 37  $\pm$  2°C.

#### **Sterility of the test substance and S9 mix**

0.1 mL of the 5% (w/v) standard solution of the test substance and 0.1 mL of the S9 mix was plated on to complete medium and incubated for 48-72 hours at 37  $\pm$  2°C.

**Diagnostic mutagens** were used for each strain with and without S9 mix, as follows:

<b>Mutagen</b>	<b>S9</b>	<b>Strain</b>	<b><math>\mu</math>g/mL</b>
MNNG	-	TA 1535	1.0
MNNG	-	TA 100	1.0
2-NF	-	TA 98	20.0
ICR-191	-	TA 1537	0.01
MNNG	-	WP2uvrApKM101	7.5
2-AA	+	TA 98	5.0
2-AA	+	TA 1537	5.0
2-AA	+	TA 1535	5.0
2-AA	+	TA 100	5.0
2-AA	+	WP2uvrApKM101	20.0

## **6. Results and discussion**

### **Genetic characters**

All *Salmonella* strains used in these experiments were sensitive to crystal violet and Mitomycin C. TA98 and TA100 were both resistant to ampicillin. *E.coli* WP2uvrApKM101 was sensitive to Mitomycin C and resistant to ampicillin. These results are as expected.

### **Solvent and positive controls**

In general the solvent control values presented in this report are within the normal ranges experienced in our laboratory (Appendix 1) and/or the ranges reported in the literature with the *Salmonella* strains. The solvent controls values for the *E.coli* cultures applied in this study were in general rather high and just exceeded our historical range in the experiments without S9. However, these experiments were accepted since the solvent control levels were set against rather high viable cell counts and the cultures responded very well to the positive controls.

With one exception all positive control chemicals induced significant increases in revertant colony numbers which fulfilled our criteria for an acceptable response and thereby demonstrated the sensitivity of the test system and the efficiency of the metabolic activation system (S9 mix). The exception to this was both main experiments with the strain TA1537 with the addition of S9-mix (Table 4 and 9), where the effect of 2-aminoanthracene was in the low end of our historical control data (Appendix 2). Therefore this test was repeated, and in this experiment the culture responded very well to the diagnostic mutagene.



**Sterility of test substance and S9 mix**

All standard solution of the test substance applied in this study were sterile. The same apply to the S9-mix used with one exception. 5 colonies were demonstrated on the sterile plate with S9-mix applied in the first experiment with TA98 and TA1537. From the revertant colony plates in these test series this contamination could not be verified and it was considered as an accidental contamination during preparation of the sterile control.

**Lactase (Batch Number: PPL34537)**

The results are represented in Table 1-12.

We consider a test substance as positive when it has induced at least a doubling in the mean number of revertant colonies per plate compared to the appertaining solvent control in one or more of the strains, in the presence or absence of S9 mix, if this response is dose related (at least 3 doses) and reproducible.

In case of a dose related and reproducible numerical increase, which is below a doubling but at least 50% higher than the solvent control, the result is considered as equivocal and needs further clarification.

Lactase is a fluid enzyme preparation. It contains an abundance of various nutrients, and composes a rich growth medium to the test bacteria. This means, that comparison of viable counts between exposed cultures and control culture in a "treat and plate" assay reflects growth stimulation/inhibition as well as cell killing. Variation in the viable counts may cause some variation in the number of spontaneous revertant colonies.

In the present study the viability was somewhat reduced at the highest dose levels in several test series with *Salmonella* strains. On the contrary growth stimulation was observed in the test series with the *E.coli* strain as demonstrated by increases in the viable count of exposed cultures compared to the solvent control. However, these conditions had no significant influence on the revertant colony count.

No treatments of any of the *Salmonella* and *E.coli* strains with Lactase resulted in any increases in revertant numbers that meet the criteria for a positive or equivocal response.

In the second experiment with TA1535 with S9 the revertant levels were relatively high for cultures exposed to the test article in general and without any obvious dose relation. Concurrently, low viabilities at the highest two doses were evident in this test series. Therefore, a similar 3.Experiment was performed, demonstrating no reduction in viabilities and revertants levels equal to the solvent control of all doses.

**7. Conclusion**

The results of the bacterial mutagenicity tests described in this report give no indication of the presence of mutagenic components in this preparation of Lactase (Batch No. PPL34537), when tested under the conditions applied in this study.

**Table 1-10****Table 1.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium TA100* following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 1.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Base-pair substitution type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	102 83 77	87	23 13	18	74 90 96	87	225 252	239
2500	100 85 96	94	30 31	31	109 103 115	109	115 163	139
1250	109 100 95	101	75 70	73	105 127 112	115	112 99	106
625	120 116 121	119	161 154	158	131 103 88	107	190 156	173
313	116 110 91	106	127 159	143	120 90 104	105	216 214	215
156	116 95 125	112	100 107	104	104 94 105	101	171 178	175
Solvent control	103 112 117 135 124	118	112 110	111	88 86 101 99 96	94	167 169	168
2AA 5.0	-	-	-	-	1979 1963 1961	1968	114 122	118
MNNG 1.0	3861 3839 3710	3803	112 99	106	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

MNNG - 1-Methyl-3-Nitro-N-NitrosoGuanidine

**Table 2.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium TA1535* following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the treat and plate assay.

**20138037 EXP 1.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Base-pair substitution type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	8 8 8	8	44 43	44	19 9 5	11	259 215	237
2500	16 12 8	12	47 54	51	5 5 10	7	178 195	187
1250	9 10 8	9	80 93	87	9 8 8	8	248 251	250
625	6 9 8	8	140 154	147	14 8 12	11	217 194	206
313	11 8 8	9	126 112	119	10 11 9	10	255 224	240
156	9 5 12	9	125 137	131	6 13 16	12	318 309	314
Solvent control	9 12 10 6 13	10	142 162	152	11 8 6 7 11	9	268 287	278
2AA 5.0	-	-	-	-	164 188 153	168	103 111	107
MNNG 1.0	4452 4366 4206	4341	201 183	192	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

MNNG - 1-Methyl-3-Nitro-N-NitrosoGuanidine



**Table 3.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium* TA98 following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 1.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Frame-shift mutation type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	42 43 44	43	82 46	64	23 23 16	21	88 73	81
2500	27 36 42	35	106 101	104	21 26 26	24	130 124	127
1250	58 43 41	47	124 121	123	16 19 20	18	96 99	98
625	48 35 36	40	98 95	97	21 30 19	23	91 103	97
313	25 52 47	41	88 70	79	31 24 23	26	85 59	72
156	54 48 43	48	137 111	124	21 36 20	26	77 78	78
Solvent control	44 52 54 46 52	50	96 78	87	26 28 30 24 27	27	75 82	79
2AA 5.0	-	-	-	-	3483 3798 3677	3653	25 23	24
2-NF 20.0	1463 1368 1426	1419	117 114	116	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

2-NF - 2-Nitrofluorene

**Table 4.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium TA1537* following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 1.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Frame-shift mutation type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	3 6 6	5	67 77	72	7 8 5	7	72 64	68
2500	15 7 5	9	119 84	102	9 9 6	8	41 57	49
1250	12 12 10	11	93 114	104	6 7 5	6	74 80	77
625	3 8 14	8	120 116	118	5 2 4	4	62 79	71
313	16 10 11	12	106 104	105	3 3 7	4	107 95	101
156	11 13 12	12	133 140	137	6 6 5	6	91 112	102
Solvent control	14 7 10 13 10	11	232 203	218	4 4 6 9 10	7	95 104	100
2AA 5.0	-	-	-	-	77 62 77	72	67 40	54
ICR-191 0.01	1177 1254 1212	1214	149 93	121	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

ICR-191 - Acridine Mutagen

**Table 5.**

Number of revertant colonies per plate obtained with *E.coli WP2uvrApKM101* following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 1.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Base-pair substitution type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	236 273 253	254	515 478	497	341 346 367	351	541 478	510
2500	256 290 261	269	444 414	429	326 261 308	298	436 421	429
1250	341 280 298	306	308 356	332	394 300 341	345	489 430	460
625	313 295 263	290	269 288	279	311 357 311	326	311 309	310
313	263 298 303	288	326 294	310	352 327 363	347	383 364	374
156	385 300 337	341	378 301	340	405 355 308	356	436 414	425
Solvent control	287 282 276 282 278	281	276 332	304	319 336 305 268 306	307	374 363	369
2AA 20.0	-	-	-	-	2433 2115 2291	2280	238 237	238
MNNG 7.5	3474 3472 3236	3394	226 225	226	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

MNNG - 1-Methyl-3-Nitro-N-NitrosoGuanidine



**Table 6.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium TA100* following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 2.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Base-pair substitution type							
	Without S9				With S9			
	Revertants		Viable cells *		Revertants		Viable cells *	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	91 101 77	90	93 78	86	146 146 128	140	90 88	89
2500	86 106 106	99	73 75	74	120 128 122	123	135 163	149
1250	146 124 136	135	122 111	117	130 112 112	118	167 132	150
625	117 101 143	120	162 152	157	112 136 121	123	159 152	156
313	124 128 149	134	192 208	200	143 143 143	143	115 116	116
156	128 131 115	125	163 130	147	117 131 111	120	136 124	130
Solvent control	125 127 110 142 130	127	154 172	163	124 146 126 145 125	133	126 141	134
2AA 5.0	-	-	-	-	1898 1756 1976	1877	90 80	85
MNNG 1.0	5552 5384 5484	5473	132 131	132	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

MNNG - 1-Methyl-3-Nitro-N-NitrosoGuanidine

**Table 7.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium* TA1535 following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the treat and plate assay.

**20138037 EXP 2.**

Test Substance Concentration µg per mL	Number of revertants (number of colonies/plate) Base-pair substitution type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	4 8 15	9	67 61	64	14 16 16	15	15 16	16
2500	10 8 10	9	132 131	132	12 21 16	16	15 19	17
1250	10 13 17	13	141 184	163	12 8 11	10	166 188	177
625	7 8 12	9	192 196	194	14 9 12	12	- -	-
313	16 13 14	14	159 143	151	13 11 17	14	211 242	227
156	9 6 13	9	204 193	199	11 13 14	13	245 243	244
Solvent control	11 17 11 14 11	13	166 167	167	12 9 9 8 9	9	154 161	158
2AA 5.0	-	-	-	-	130 142 131	134	3 7	5
MNNG 1.0	6142 6040 5928	6037	196 183	190	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

MNNG - 1-Methyl-3-Nitro-N-NitrosoGuanidine



**Table 8.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium* TA98 following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 2.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Frame-shift mutation type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	31 35 31	32	251 238	245	33 33 39	35	135 184	160
2500	38 26 37	34	198 168	183	45 28 23	32	141 125	133
1250	28 28 30	29	178 159	169	26 26 28	27	236 185	211
625	32 33 37	34	132 138	135	30 36 29	32	179 169	174
313	32 32 37	34	131 132	132	48 33 26	36	145 94	120
156	33 23 43	33	231 229	230	25 19 34	26	219 232	226
Solvent control	36 30 36 32 33	33	274 177	226	35 33 30 42 22	32	305 352	329
2AA 5.0	-	-	-	-	2477 2501 2423	2467	93 107	100
2-NF 20.0	1580 1489 1518	1529	145 272	209	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

2-NF - 2-Nitrofluorene

**Table 9.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium* TA1537 following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 2.**

Test Substance Concentration µg per mL	Number of revertants (number of colonies/plate) Frame-shift mutation type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	7 13 9	10	59 47	53	6 5 6	6	26 23	25
2500	8 8 11	9	101 80	91	7 6 7	7	30 26	28
1250	7 7 8	7	115 143	129	4 9 4	6	68 52	60
625	15 13 7	12	156 184	170	4 6 7	6	68 70	69
313	11 11 11	11	190 166	178	12 7 8	9	84 51	68
156	12 14 11	12	208 215	212	5 9 9	8	51 49	50
Solvent control	14 13 12 16 12	13	156 148	152	5 6 3 9 5	6	70 63	67
2AA 5.0	-	-	-	-	61 75 72	69	65 49	57
ICR-191 0.01	1564 1564 1554	1561	152 163	158	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

ICR-191 - Acridine Mutagen

**Table 10.**

Number of revertant colonies per plate obtained with *E.coli* WP2uvrApKM101 following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 2.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Base-pair substitution type							
	Without S9				With S9			
	Revertants		Viable cells*		Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean	Single plates	Mean	Single plates	Mean
5000	221 255 243	240	313 318	316	322 304 379	335	460 455	458
2500	292 293 259	281	253 231	242	340 332 334	335	295 293	294
1250	279 229 304	271	337 342	340	310 300 336	315	441 448	445
625	287 290 273	283	224 251	238	326 332 314	324	348 353	351
313	271 268 266	268	259 282	271	277 331 301	303	329 325	327
156	247 263 255	255	224 241	233	297 310 313	307	292 285	289
Solvent control	287 288 279 276 247	275	236 214	225	329 322 319 329 301	320	313 348	331
2AA 20.0	-	-	-	-	1417 1467 1507	1464	242 238	240
MNNG 7.5	1016 1087 1048	1050	158 171	165	-	-	-	-

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

MNNG - 1-Methyl-3-Nitro-N-NitrosoGuanidine



**Table 11.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium TA1535* following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the treat and plate assay.

**20138037 EXP 3.**

Test Substance Concentration µg per mL	Number of revertants (number of colonies/plate) Base-pair substitution type			
	With S9			
	Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean
5000	13 9 14	12	162 142	152
2500	9 6 11	9	131 140	136
1250	9 18 8	12	132 131	132
625	12 14 8	11	59 65	62
313	7 18 11	12	115 105	110
156	8 7 15	10	105 84	95
Solvent control	6 14 15 9 8	10	149 146	148
2AA 5.0	282 241 203	242	19 17	18

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

**Table 12.**

Number of revertant colonies per plate obtained with *Salmonella typhimurium TA1537* following exposure to Lactase batch PPL34537 in the absence and presence of metabolic activation in the Treat and plate assay.

**20138037 EXP 3.**

Test Substance Concentration (µg dry matter/per mL)	Number of revertants (number of colonies/plate) Frame-shift mutation type			
	With S9			
	Revertants		Viable cells*	
	Single plates	Mean	Single plates	Mean
5000	11 17 22	17	128 112	120
2500	16 18 12	15	53 42	48
1250	18 18 18	18	40 48	44
625	16 11 9	12	128 126	127
313	14 19 10	14	111 104	108
156	19 20 12	17	168 132	150
Solvent control	15 16 17 12 16	15	130 104	117
2AA 5.0	289 335 329	318	91 95	93

Abbreviations:

\*Corresponding to no. of viable cells x 10<sup>6</sup> on revertant plates

2AA - 2-Aminoanthracene

## Appendix 1 Historical control data for solvent controls

**Solvent control ranges** (purified water) for *S. typhimurium* strains and *E.coli* WP2uvrApKM101 in the treat and plate assay. (SOP: TOX-SM-1006 and TOX-SM-1007)

Strain	S9	Number of determinations	Mean number of revertants per plate	SD	Range *)	
					lower	upper
TA1535	÷	20	10	3	6	19
	+	20	11	3	7	22
TA100	÷	21	109	16	76	142
	+	20	117	16	94	151
TA1537	÷	20	10	4	5	17
	+	22	13	5	6	24
TA98	÷	20	22	7	11	33
	+	20	31	7	22	44
WP2 uvrA pKM101	÷	21	214	26	178	270
	+	21	257	32	213	331

The above are pooled data from a number of independent determinations selected from studies throughout the year 2012. Only determinations, which were obviously vitiated by errors, have been omitted.

\*) Ranges stated are the maximum and minimum mean revertant colony counts from the data sets sampled.

## Appendix 2 Historical control data for positive controls

**Positive control ranges** for *S. typhimurium* strains and *E.coli* WP2uvrApKM101 in the treat and plate assay. (SOP: TOX-SM-1006 and TOX-SM-1007).

Strain	S9	Number of determinations	Chemical	Mean number of revertants per plate	SD	Range *)	
						lower	upper
TA1535	÷	20	MNNG 1 µg/ml	4176	739	2541	5623
	+	20	2-AA 5 µg/ml	189	41	114	261
TA100	÷	21	MNNG 1 µg/ml	3819	687	2113	5121
	+	20	2-AA 5 µg/ml	2144	454	1072	2964
TA1537	÷	20	ICR-191 0.01µg/ml	1934	764	1189	4611
	+	20	2-AA 5 µg/ml	179	71	84	335
TA98	÷	20	2-NF 20 µg/ml	1221	340	693	2022
	+	20	2-AA 5 µg/ml	2083	548	1046	3002
WP2 uvrA pKM101	÷	21	MNNG 7.5 µg/ml	1424	335	989	2442
	+	21	2-AA 20 µg/ml	1516	288	807	2086

The above are pooled data from a number of independent determinations selected from studies throughout the year 2012. Only determinations, which were obviously vitiated by errors, have been omitted.

\*) Ranges stated are the maximum and minimum mean revertant colony counts from the data sets sampled.



## Appendix 3 Preparation of media

### 1. Top-agar - histidine-deficient soft agar

Agar, Merck	0.6 g
NaCl	0.5 g
Distilled water to	100 mL

The medium was autoclaved for 15 minutes at 121°C. After cooling to approximately 55°C, 10 mL of a sterile aqueous solution of 0.5 mM biotin - 0.5 mM histidine was added aseptically.

### 2. Nutrient broth - histidine-rich broth

Difco nutrient broth	8 g
NaCl	5 g
Distilled water to	1 litre

The medium was autoclaved for 15 minutes at 121°C.

### 3. Nutrient agar - histidine-rich agar medium

Agar, Merck	15 g
Oxoid nutrient broth No. 2	25 g
Distilled water to	1 litre

The medium was autoclaved for 15 minutes at 121°C.

### 4. Minimal medium

This was Vogel-Bonner minimal "E" medium with 2% glucose, prepared as follows:

#### Solution A (Vogel-Bonner medium E, 20X)

MgSO <sub>4</sub> 7H <sub>2</sub> O	4 g
Citric acid, monohydrate	40 g
K <sub>2</sub> HPO <sub>4</sub>	200 g
NaH <sub>2</sub> NH <sub>4</sub> 4H <sub>2</sub> O	70 g
Distilled water to	1000 mL

The solution was sterilized by filtration.

#### Solution B (40% glucose)

Glucose	40 g
Distilled water to	100 mL

This solution was sterilized by filtration.


#### Solution C (Agar base)

Agar, Merck	16.7 g
Distilled water to	1000 mL

Solution C was autoclaved for 15 minutes at 121°C. After cooling to approximately 55°C, 450 mL of solution C was aseptically added 25 mL of solution A and 25 mL of solution B.



# Final Report

Study Title	Lactase, PPL34537: Induction of micronuclei in cultured human peripheral blood lymphocytes
Study Director	
Test Facility	Covance Laboratories Ltd. Otley Road, Harrogate North Yorkshire, HG3 1PY England
Covance Study Number	8292559
Sponsor	Novozymes A/S Toxicology Krogshoejvej 36 DK-2880 Bagsvaerd Denmark
Novozymes Reference Number	NZ 20136071
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Page Number	1 of 42

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### COMPLIANCE STATEMENT AND SIGNATURE PAGE


I, the undersigned, hereby declare that the work was performed under my supervision and that the findings provide a true and accurate record of the results obtained.

The study was performed in accordance with the agreed protocol and with Covance Laboratories Ltd. Standard Operating Procedures, unless otherwise stated, and the study objectives were achieved.

This study was conducted in accordance with the following:

- The United Kingdom (GLP Monitoring Authority, Medicines and Healthcare products Regulatory Agency [MHRA]) Good Laboratory Practice Regulations 1999, Statutory Instrument 1999 No.3106 as amended by the Good Laboratory Practice (Codification Amendments Etc.) Regulations, 2004
- The OECD Principles on Good Laboratory Practice ENV/MC/CHEM (98) 17 (Revised in 1997, Issued January 1998).

With the exception that analyses for achieved concentration of test article formulations were not conducted as part of this study due to the nature of the test article preparation. Although test article stability in the vehicle was not determined in this study, all test article formulations were freshly prepared prior to treatment on the day of use.

  
Study Director  
Covance Laboratories Ltd.

08 January 2014  
Date

## QUALITY ASSURANCE STATEMENT


This study has been reviewed by the GLP Quality Assurance Unit of Covance and the report accurately reflects the raw data. The following inspections were conducted and findings reported to the study director (SD) and associated management.

Critical procedures, which are performed routinely in an operational area, may be audited as part of a "process" inspection programme. This can be in addition to phases scheduled on an individual study basis. Selected process inspections conducted and considered applicable to this study are included below.

In addition to the inspection programmes detailed below, a facility inspection programme is also operated. Details of this programme, which covers all areas of the facility annually (at a minimum), are set out in standard operating procedures.

Inspection Dates		Phase	Date Reported to SD and SD Management
From	To		
01 Oct 2013	01 Oct 2013	Protocol Review	01 Oct 2013
09 Dec 2013	11 Dec 2013	Draft Report and Data Review	11 Dec 2013
30 Dec 2013	31 Dec 2013	Sign off Inspection Record	31 Dec 2013
08 Jan 2014	08 Jan 2014	Final Report Review	08 Jan 2014

Inspection Dates		Phase	Date Reported to SD and SD Management
From	To		
08 Oct 2013	08 Oct 2013	Slide Analysis	08 Oct 2013
09 Oct 2013	09 Oct 2013	Wash Off	10 Oct 2013
10 Oct 2013	10 Oct 2013	Slide Staining	10 Oct 2013
10 Oct 2013	10 Oct 2013	S9 Quality Control Checks	10 Oct 2013
15 Oct 2013	15 Oct 2013	Dose Preparation	15 Oct 2013
18 Oct 2013	18 Oct 2013	Archiving	18 Oct 2013
24 Oct 2013	24 Oct 2013	Archiving	25 Oct 2013

 \_\_\_\_\_ 8 January 2014  
Date  
Quality Assurance Unit

**RESPONSIBLE PERSONNEL**

Study Monitor	[REDACTED]
Study Director	[REDACTED]
Genetic Toxicology Operations	[REDACTED]
Quality Assurance Contact	[REDACTED]

## 1. SUMMARY

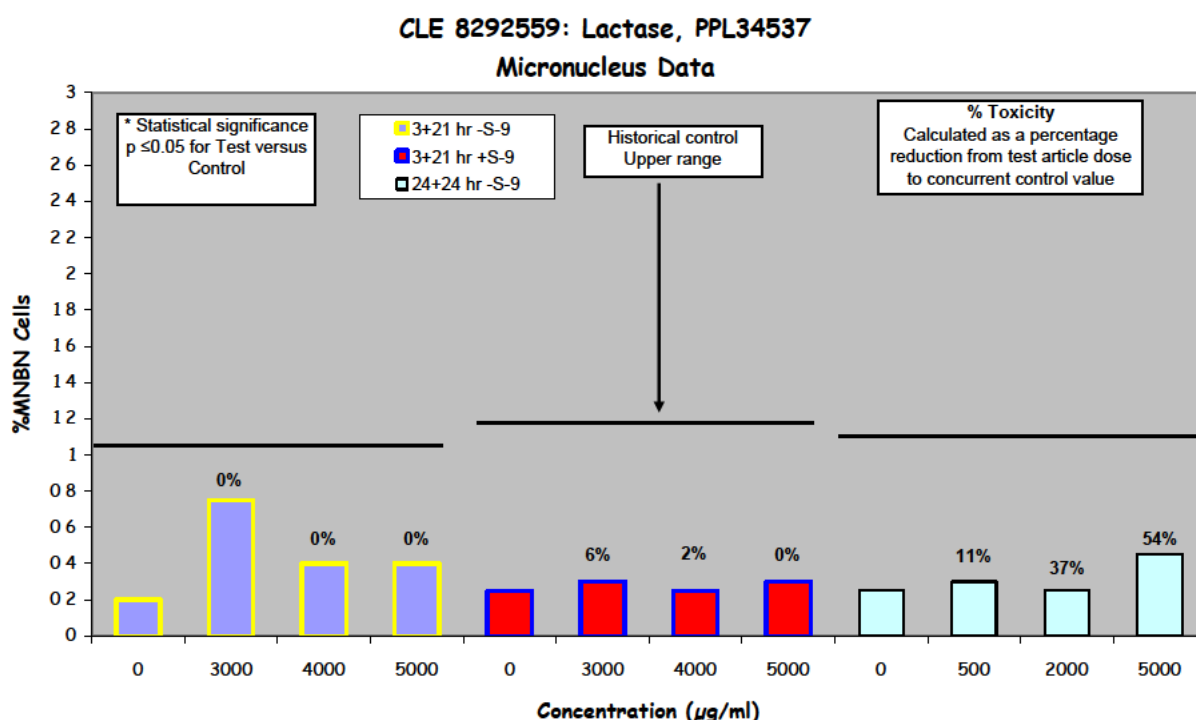
Lactase, PPL34537 was tested in an *in vitro* micronucleus assay using duplicate human lymphocyte cultures prepared from the pooled blood of two female donors in a single experiment. Treatments covering a broad range of concentrations, separated by narrow intervals, were performed both in the absence and presence of metabolic activation (S-9) from Aroclor 1254-induced rats. The test article was formulated in water for irrigation (purified water) and the highest concentrations used in the Micronucleus Experiment, 5000 µg/mL (an acceptable maximum concentration for *in vitro* micronucleus studies according to current regulatory guidelines), was determined following a preliminary cytotoxicity Range-Finder Experiment.

Treatments were conducted (as detailed in the following summary table) 48 hours following mitogen stimulation by phytohaemagglutinin (PHA). The test article concentrations for micronucleus analysis were selected by evaluating the effect of Lactase, PPL34537 on the replication index (RI). Micronuclei were analysed at three concentrations and a summary of the data is presented in the following table:

Treatment	Concentration (µg/mL)	Cytotoxicity (%) <sup>s</sup>	Mean MNBN cell frequency (%)	Historical Control Range (%) <sup>#</sup>	Statistical significance
3+21 hour -S-9	Vehicle <sup>a</sup>	-	0.20	0.10 – 1.10	-
	3000	0	0.75		p≤0.01
	4000	0	0.40		NS
	5000	0	0.40		NS
	*MMC, 0.80	ND	9.90		p≤0.001
3+21 hour +S-9	Vehicle <sup>a</sup>	-	0.25	0.20 – 1.20	-
	3000	6	0.30		NS
	4000	2	0.25		NS
	5000	0	0.30		NS
	*CPA, 6.25	ND	3.45		p≤0.001
24+24 hour -S-9	Vehicle <sup>a</sup>	-	0.25	0.05 – 1.15	-
	500.0	11	0.30		NS
	2000	37	0.25		NS
	5000	50	0.45		NS
	*VIN, 0.10	ND	3.16		p≤0.001

<sup>a</sup> Vehicle control was purified water  
<sup>\*</sup> Positive control  
<sup>#</sup> 95<sup>th</sup> percentile of the observed range  
<sup>s</sup> Based on replication index  
 NS Not significant  
 ND Not determined

This data is graphically presented as follows:



Appropriate negative (vehicle) control cultures were included in the test system under each treatment condition. The proportion of micronucleated binucleate (MNBN) cells in the vehicle cultures fell within the current 95<sup>th</sup> percentile of the observed historical vehicle control (normal) ranges. Mitomycin C (MMC) and Vinblastine (VIN) were employed as clastogenic and aneugenic positive control chemicals respectively in the absence of rat liver S-9. Cyclophosphamide (CPA) was employed as a clastogenic positive control chemical in the presence of rat liver S-9. Cells receiving these were sampled in the Micronucleus Experiment at 24 hours after the start of treatment; all compounds induced statistically significant increases in the proportion of cells with micronuclei.

All acceptance criteria were considered met and the study was therefore accepted as valid.

Treatment of cells with Lactase, PPL34537 in the absence and presence of S-9 resulted in frequencies of MNBN cells which were similar to and not significantly ( $p \leq 0.05$ ) higher than those observed in concurrent vehicle controls for the majority of all concentrations analysed (all treatments). A single exception was observed at the lowest concentration analysed (3000 µg/mL) following 3+21 hour in the absence of S-9 treatment. However, this increase was not large with both replicate cultures exhibiting MNBN cell frequencies within the historical vehicle control (normal) range with no instances of MNBN cell values exceeding normal values for any other Lactase, PPL34537 treated culture (all concentrations, all treatments). As such, this isolated statistical increase was not considered of biological importance.



It is concluded that Lactase, PPL34537 did not induce micronuclei in cultured human peripheral blood lymphocytes following treatment in the absence and presence of an Aroclor 1254-induced rat liver metabolic activation system (S-9). Concentrations were tested up to 5000 µg/mL, a recommended regulatory maximum concentration for *in vitro* micronucleus assays.

## 2. GENERAL STUDY INFORMATION

### 2.1 Objective

The objective of this study was to evaluate the clastogenic and aneugenic potential of Lactase, PPL34537 by examining its effects on the frequency of micronuclei in cultured human peripheral blood lymphocytes treated in the absence and presence of a rat liver metabolic activation system (S-9).

### 2.2 Introduction

Chromosome defects are recognised as the basis of a number of human genetic diseases (Mitelman, 1991). Assays for the detection of chromosome damage in mammalian cells *in vitro* are recommended in regulatory guidelines as a complement to Ames tests in a genotoxicity test battery. There is a large database on the use of chromosomal assays for screening purposes (Preston *et al.*, 1981; Fenech, 1998; Fenech *et al.*, 2003). The use of human peripheral blood lymphocytes is recommended because the cells are only used in short-term culture and maintain a stable karyotype (Evans & O’Riordan, 1975). Experiments with these cells can also be performed in conjunction with a rat liver metabolising system (S-9) since, for short incubation periods, no toxicity is induced by the liver homogenate itself.

An alternative to measuring structural aberrations in mitotic cells is to measure micronuclei. These are produced from whole chromosomes or acentric fragments that are unable to attach to the spindle at mitosis and appear during the next interphase as small darkly staining bodies adjacent to the main daughter nucleus. Cytochalasin B (Cyto-B), if added to cultures, inhibits cytokinesis (cell division) but not karyokinesis (nuclear division) resulting in the formation of binucleate cells (Fenech & Morley, 1985). If micronuclei are counted in binucleate cells, then a measurement of micronucleus induction resulting from cell division can be obtained.

Theoretical considerations, together with published data (Lorge *et al.*, 2006), indicate that most aneugens and clastogens will be detected by a short term treatment period of 3-6 hours in the presence and absence of S-9 followed by removal of the test article and a growth period of 1.5-2.0 cell cycles (Fenech & Morley, 1986).

The most efficient approach is to test lymphocytes 44-48 hours post-mitogen stimulation by PHA, when cycle synchronisation will have dissipated (Fenech, 2007).

The test article was added at 48 hours following culture initiation (stimulation by PHA). Cells were exposed to the test article for 3 hours in the absence and presence of S-9 (from rats induced with Aroclor 1254). These cultures were sampled 24 hours after the beginning of treatment (i.e. 72 hours after culture initiation). In addition, a continuous 24 hour treatment (equivalent to approximately 1.5 to 2 times the average generation time of cultured lymphocytes from the panel of donors used in this laboratory) in the absence of S-9 was included. These cultures were sampled 48 hours after the beginning of treatment (i.e. 96 hours after culture initiation).

### 2.3 Study Timetable

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Study Initiation Date:	30 September 2013
Experimental Start Date:	02 October 2013
Experimental Completion Date:	08 November 2013
Study Completion Date:	Is the date the final report is signed by the Study Director

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### 2.4 Regulatory Test Guidelines

OECD Guideline 487 (OECD, 2010) and accepted scientific/regulatory principles described in current guidelines for clastogenicity testing *in vitro* (Fenech, 1998; Fenech *et al.*, 2003; Rosefort *et al.*, 2004; Elhajouji *et al.*, 1998; Migliore & Nieri, 1991; Galloway *et al.*, 1994; Aardmea *et al.*, 1998; Miller *et al.*, 1998; Fenech *et al.*, 1999; Thybaud *et al.*, 2007).

### 2.5 Protocol Adherence

This study was conducted according to the Protocol, with the exception of the protocol deviation (Section 7.4). This deviation did not affect the integrity or interpretation of the results of the study.

### 2.6 Major Computer Systems

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Application Name	Application Function
CMS (Covance Management Systems)	Scheduling
Pristima	Formulations
eNotes	Electronic communication system
Vitroabs, CBPI, Vitronuc	Slide coding, data generation and collation
Microsoft Office / Adobe Acrobat	Report generation

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Version numbers of the applications are maintained on file at Covance.

### 2.7 Archive Statement

The raw data, including documentation, study protocol, final report and study correspondence resulting from this study will be retained in the test facility archives for five years from the date of report finalisation. After completion of this period, the Sponsor will be contacted in order to determine their requirements for further retention or disposition of the archived materials (excluding facility records, non-transferable electronic data and facility copies of protocol/final report, which will be retained by Covance in accordance with test facility SOPs). Where continued retention is requested, the archived materials may subsequently be transferred to alternative Covance Archive locations. In this event, the Sponsor will be informed, and documented chain of custody records will be maintained.

### 3. MATERIALS

#### 3.1 Test Article

Lactase, PPL34537, batch number PPL34537, was received as a frozen brown liquid (nominal volume as 100 mL) on 25 September 2013. Activity was stated as 3260 LUA(A)/g but for the purpose of this study purity was considered to be 100%. The expiry date was given as 20 November 2022. Lactase, PPL34537 is a high molecular weight protein with a molecular weight confirmed as approximately 130 kDa such that 10 mM would be far greater than 5000 µg/mL. The test article information and [Certificate of Analysis](#) provided by the Sponsor are considered an adequate description of the characterisation, purity and stability of the test article. Determinations of stability and characteristics of the test article were the responsibility of the Sponsor.

Following receipt, the test article was thawed under refrigerated conditions, stirred and sub-divided into three aliquots (1 x 15 mL for use on Range-Finder experiments and 2 x 42 mL aliquots for use on the Micronucleus Experiment). These aliquots were then re-frozen and stored at <-10°C, protected from light in order that the test article could be thawed and re-frozen on no more than one occasion.

Lactase, PPL34537 is a high molecular weight protein, which was formulated under subdued lighting in water for irrigation (purified water) at a concentration of 50 mg/mL (weighed out as received), equivalent to 5000 µg/mL final culture concentration. For each experimental occasion, the test article was de-frosted overnight under refrigerated conditions prior to the day of formulation/treatment. Once de-frosted, the test article was stirred gently prior to weighing and during formulation (where possible) under magnetic stirring conditions. Following removal from the refrigerator the test compound was used within approximately 2.5 hours.

No preliminary solubility trials were carried out as part of this study.

The stock solutions were membrane filter-sterilised (Pall Acrodisc 32 mm, 0.2 µm pore size) and subsequent dilutions made (under stirring conditions) using purified water. The test article solutions were protected from light prior to treatment.

The treatment scheme used is illustrated as follows:

Experiment	Treatment	Concentration Range (mg/mL)			Final Concentration Range (µg/mL)		
Range-Finder	3+21, -S-9	0.1814	to	50.00	18.14	to	5000
	3+21, +S-9	0.1814	to	50.00	18.14	to	5000
	24+24, -S-9	0.1814	to	50.00	18.14	to	5000
Micronucleus Experiment	3+21, -S-9	10.00	to	50.00	1000	to	5000
	3+21, +S-9	10.00	to	50.00	1000	to	5000
	24+24, -S-9	2.500	to	50.00	250.0	to	5000

### 3.2 Controls

Sterile purified water was added to cultures designated as vehicle controls as described in the methods section of this report. The positive control chemicals were supplied and used according to the following table:

Chemical	Stock Concentration (mg/mL) <sup>c</sup>	Final Concentration (µg/mL)	S-9
Mitomycin C (MMC) <sup>a</sup>	0.060	0.60	-
	0.080	0.80	-
Cyclophosphamide (CPA) <sup>b</sup>	0.625	6.25	+
	1.250	12.50	+
Vinblastine (VIN) <sup>a</sup>	0.008	0.08	-
	0.01	0.100	-
	0.012	0.120	-

<sup>a</sup> Obtained from Sigma-Aldrich.

<sup>b</sup> Obtained from Acros Organics, Loughborough, Leicestershire, UK.

<sup>c</sup> In the Micronucleus Experiment, CPA was dissolved in anhydrous analytical grade dimethyl sulphoxide (DMSO), frozen (<-50°C) and thawed immediately prior to use. VIN and MMC were dissolved in purified water immediately prior to use.

For the 3+21 hour treatment in the absence of S-9, MMC was used as the positive control. For the 24+24 hour –S-9 treatment, VIN was used as the positive control.

### 3.3 Metabolic Activation System

The mammalian liver post-mitochondrial fraction (S-9) used for metabolic activation was obtained from Molecular Toxicology Incorporated, USA where it is prepared from male Sprague Dawley rats induced with Aroclor 1254. The batches of MolTox™ S-9 were stored frozen in aliquots at <-50°C prior to use. Each batch was checked by the manufacturer for sterility, protein content, ability to convert known promutagens to bacterial mutagens and cytochrome P-450-catalyzed enzyme activities (alkoxyresorufin-O-dealkylase activities). See [Quality Control Statement for S-9](#).

The S-9 mix was prepared in the following way:

Glucose-6-phosphate (G6P: 180 mg/mL), β-Nicotinamide adenine dinucleotide phosphate (NADP: 25 mg/mL), Potassium chloride (KCl: 150 mM) and rat liver S-9 were mixed in the ratio 1:1:1:2. For all cultures treated in the presence of S-9, an aliquot of the mix was added to each cell culture to achieve the required final concentration of test article in a total of 10 mL. The final concentration of the liver homogenate in the test system was 2%.

Cultures treated in the absence of S-9 received an equivalent volume of 150 mM KCl.

### 3.4 Blood Cultures

Blood from two healthy, non-smoking female volunteers from a panel of donors at Covance was used for each experiment as follows:

Experiment	Donor Sex	Donor Age (years)	Donor Identity
Range-Finder	Female	26, 31	8513, 9959
Micronucleus Experiment	Female	24, 29	8669, 8578

No donor was suspected of any virus infection or exposed to high levels of radiation or hazardous chemicals. All donors are non-smokers and are not heavy drinkers of alcohol. Donors were not taking any form of medication (contraceptive pill excluded). The measured cell cycle time of the donors used at Covance, Harrogate falls within the range  $13 \pm 2$  hours. For each experiment, an appropriate volume of whole blood was drawn from the peripheral circulation into heparinised tubes within two days of culture initiation. Blood was stored refrigerated and pooled using equal volumes from each donor prior to use.

Whole blood cultures were established in sterile disposable centrifuge tubes by placing 0.4 mL of pooled heparinised blood into 8.1 mL pre-warmed (in an incubator set to  $37 \pm 1^\circ\text{C}$ ) HEPES-buffered RPMI medium containing 10% (v/v) heat inactivated foetal calf serum and 0.52% penicillin / streptomycin, so that the final volume following addition of S-9 mix/KCl and the test article in its chosen vehicle was 10 mL. The mitogen Phytohaemagglutinin (PHA, reagent grade) was included in the culture medium at a concentration of approximately 2% of culture to stimulate the lymphocytes to divide. Blood cultures were incubated at  $37 \pm 1^\circ\text{C}$  for approximately 48 hours and rocked continuously (see Protocol deviation Section 7.4).

## 4. METHODS

### 4.1 Test System

The test system was suitably labelled (using a colour-coded procedure) to clearly identify the study number, experiment number, treatment time, donor sex, test article concentration (if applicable), positive and vehicle controls.

### 4.2 Cytotoxicity Range-Finder

S-9 mix or KCl (0.5 mL per culture) was added appropriately. Cultures were treated with the test article and vehicle (1.0 mL per culture). Positive control treatments were not included.

The final culture volume was 10 mL. Cultures were incubated at  $37\pm 1^{\circ}\text{C}$  for the designated exposure time.

### 4.3 Micronucleus Experiment

Immediately prior to treatment, all positive control cultures had 0.9 mL culture medium added to give a final pre-treatment volume of 9.4 mL.

S-9 mix or KCl (0.5 mL per culture) was added appropriately. Cultures were treated with the test article, vehicle, or positive controls (1.0 mL per culture or 0.1 mL per culture for positive control cultures).

The final culture volume was 10 mL. Cultures were incubated at  $37\pm 1^{\circ}\text{C}$  for the designated exposure time.

This scheme is illustrated as follows:

Treatment	S-9	Number of Cultures			
		Cytotoxicity Range-Finder		Micronucleus Experiment	
		3+21*	24+24*	3+21*	24+24*
Vehicle control	-	2	2	4	4
	+	2		4	
Test article	-	1	1	2	2
	+	1		2	
Positive controls	-			2	2
	+			2	

\* Hours treatment + hours recovery

For removal of the test article, cells were pelleted (approximately 300 g, 10 minutes), washed twice with sterile saline (pre-warmed in an incubator set to  $37\pm 1^{\circ}\text{C}$ ), and resuspended in fresh pre-warmed medium containing foetal calf serum and penicillin / streptomycin. Cyto-B (formulated in DMSO) was added to post wash-off culture medium to give a final concentration of 6  $\mu\text{g/mL}$  per culture.

Duration of Treatment (hours)	S-9	Hours after Culture Initiation			Harvest Time
		Addition of Test Chemical	Removal of Test Chemical	Addition of Cyto-B	
3	-	48	51	51*	72
24	-	48	72	72*	96
3	+	48	51	51*	72

\* Approximate times

Changes in osmolality of more than 50 mOsm/kg and fluctuations in pH of more than one unit may be responsible for an increase in chromosome aberrations (Scott *et al.*, 1991; Brusick, 1986). Osmolality and pH measurements on post-treatment incubation medium were taken in the cytotoxicity Range-Finder Experiment.

#### 4.4 Harvesting

At the defined sampling time, cultures were centrifuged at approximately 300 g for 10 minutes, the supernatant removed and discarded and cells resuspended in 4 mL (hypotonic) 0.075 M KCl at 37±1°C for 4 minutes to allow cell swelling to occur. Cells were then fixed by dropping the KCl suspension into fresh, cold methanol/glacial acetic acid (7:1, v/v). The fixative was changed by centrifugation (approximately 300 g, 10 minutes) and resuspension. This procedure was repeated as necessary (centrifuging at approximately 1250 g, 2-3 minutes) until the cell pellets were clean.

#### 4.5 Slide Preparation

Lymphocytes were kept in fixative at 2-8°C prior to slide preparation for a minimum of 3 hours to ensure that cells were adequately fixed. Cells were centrifuged (approximately 1250 g, two to three minutes) and resuspended in a minimal amount of fresh fixative (if required) to give a milky suspension. Several drops of cell suspension were gently spread onto multiple clean, dry microscope slides. Slides were air-dried then stored protected from light at room temperature prior to staining. Slides were stained by immersion in 125 µg/mL Acridine Orange in phosphate buffered saline (PBS), pH 6.8 for approximately 10 seconds, washed with PBS (with agitation) for a few seconds before transfer and immersion in a second container of PBS for approximately 10 minutes. Slides were air-dried and stored protected from light at room temperature prior to analysis.

#### 4.6 Selection of Concentrations for the Micronucleus Experiment

Slides from the cytotoxicity Range-Finder Experiment were examined, uncoded, for proportions of mono-, bi- and multinucleate cells, to a minimum of 200 cells per concentration. From these data the replication index (RI) was determined.

RI, which indicates the relative number of nuclei compared to vehicle controls was determined using the formulae as follows:

$$RI = \frac{\text{number binucleate cells} + 2 (\text{number multinucleate cells})}{\text{total number of cells in treated cultures}}$$



Relative RI (expressed in terms of percentage) for each treated culture was calculated as follows:

$$\text{Relative RI (\%)} = \frac{\text{RI of treated cultures}}{\text{RI of vehicle controls}} \times 100$$

Cytotoxicity (%) is expressed as (100 – Relative RI).

A selection of random fields was observed from enough treatments to determine whether chemically induced cell cycle delay or cytotoxicity had occurred.

A suitable range of concentrations was selected for the Micronucleus Experiment based on these toxicity data.

#### **4.7 Selection of Concentrations for Micronucleus Analysis (Micronucleus Experiment Only)**

Slides were examined, uncoded, for RI to a minimum of 500 cells per culture to determine whether chemically induced cell cycle delay or toxicity had occurred.

The highest concentration selected for micronucleus analysis following all treatment conditions was the highest concentration tested (5000 µg/mL), a recommended maximum concentration for *in vitro* micronucleus studies (OECD, 2010). Two lower concentrations that spanned the toxicity curve (where applicable) were also analysed.

#### **4.8 Slide Analysis**

For each treatment regime, two vehicle control cultures were analysed for micronuclei.

Slides from the CPA, MMC and VIN positive control treatments were checked to ensure that the system was operating satisfactorily. One concentration from each positive control, which gave satisfactory responses in terms of quality and quantity of binucleated cells and numbers of micronuclei, was analysed.

All slides for analysis were coded, using randomly generated letters, by an individual not connected with the scoring of the slides. Labels with only the study number, assay type, experiment number, the sex of the donor and the code were used to cover treatment details on the slides.

Immediately prior to analysis 1-2 drops of PBS were added to the slides before mounting with glass coverslips. Where possible, one thousand binucleate cells from each culture (2000 per concentration) were analysed for micronuclei. The number of cells containing micronuclei and the number of micronuclei per cell on each slide was noted. Observations were recorded on raw data sheets. The microscope stage co-ordinates of the first six micronucleated cells were recorded.

Binucleate cells were only included in the analysis if all of the following criteria were met:

1. The cytoplasm remained essentially intact, and

2. The daughter nuclei were of approximately equal size.

A micronucleus was only recorded if it met the following criteria:

1. The micronucleus had the same staining characteristics and a similar morphology to the main nuclei, and
2. Any micronucleus present was separate in the cytoplasm or only just touching a main nucleus, and
3. Micronuclei were smooth edged and smaller than approximately one third the diameter of the main nuclei.

Micronucleus analysis was not conducted on slides generated from the Range-Finder treatments.

Slide analysis was performed by competent analysts trained in the applicable Covance Laboratories standard operating procedures. The analysts were physically located remote from the Covance facility, but were subject to Covance management and GLP control systems (including QA inspection). All slides and raw data generated by the remote analysts were returned to Covance Laboratories for archiving on completion of analysis.

## **4.9 Analysis of Results**

### **4.9.1 Treatment of Data**

After completion of scoring and decoding of slides, the numbers of binucleate cells with micronuclei (MNBN cells) in each culture were obtained.

The proportions of MNBN cells in each replicate were used to establish acceptable heterogeneity between replicates by means of a binomial dispersion test ([Richardson et al., 1989](#)).

The proportion of MNBN cells for each treatment condition were compared with the proportion in vehicle controls by using Fisher's exact test ([Richardson et al., 1989](#)). Probability values of  $p \leq 0.05$  were accepted as significant. Additionally, the number of micronuclei per binucleate cell were obtained and recorded.

### **4.9.2 Acceptance Criteria**

The assay was to be considered valid if the following criteria were met:

1. The binomial dispersion test demonstrated acceptable heterogeneity (in terms of MNBN cell frequency) between replicate cultures, particularly where no positive responses were seen
2. The frequency of MNBN cells in vehicle controls fell within the historical vehicle control (normal) ranges
3. The positive control chemicals induced statistically significant increases in the proportion of cells with micronuclei. Both replicate cultures at the positive control

concentration analysed under each treatment condition demonstrated MNBN cell frequencies that clearly exceeded the normal range

4. A minimum of 50% of cells had gone through at least one cell division (as measured by binucleate + multinucleate cell counts) in vehicle control cultures at the time of harvest.

#### **4.9.3 Evaluation Criteria**

For valid data, the test article was considered to induce clastogenic and/or aneugenic events if:

1. A statistically significant increase in the frequency of MNBN cells at one or more concentrations was observed
2. An incidence of MNBN cells at such a concentration that exceeded the normal range in both replicates was observed
3. A concentration-related increase in the proportion of MNBN cells was observed.

The test article was considered positive in this assay if all of the above criteria were met.

The test article was considered negative in this assay if none of the above criteria were met.

Results which only partially satisfied the above criteria were dealt with on a case-by-case basis. Evidence of a concentration-related effect was considered useful but not essential in the evaluation of a positive result ([Scott \*et al.\*, 1990](#)).

## 5. RESULTS

### 5.1 Selection of Concentrations for Micronucleus Analysis

The results of the RI determinations from the cytotoxicity Range-Finder Experiment were as follows:

**Text Table 1**  
**Data for 3+21 Hour Treatments -S-9, Range-Finder - Female Donors**

Treatment (µg/mL)	Replicate	Mono	Bi	Multi	Total Number of Cells	RI	Cytotoxicity (%)
Vehicle	A	35	159	6	200	0.86	-
	B	27	164	9	200	0.91	
18.14	A	41	158	1	200	0.80	9
30.23	A	33	164	3	200	0.85	4
50.39	A	32	164	4	200	0.86	3
83.98	A	27	168	5	200	0.89	0
140.0	A	30	167	3	200	0.87	2
233.3	A	37	158	5	200	0.84	5
388.8	A	26	172	2	200	0.88	0
648.0	A	29	169	2	200	0.87	2
1080	A	24	176	0	200	0.88	0
1800	A	30	170	0	200	0.85	4
3000	A	27	173	0	200	0.87	2
5000	A	21	179	0	200	0.90	0

See Section 7.2 for abbreviations.

**Text Table 2**  
**Data for 3+21 Hour Treatments +S-9, Range-Finder - Female Donors**

Treatment (µg/mL)	Replicate	Mono	Bi	Multi	Total Number of Cells	RI	Cytotoxicity (%)
Vehicle	A	42	158	0	200	0.79	-
	B	55	145	0	200	0.73	
18.14	A	45	155	0	200	0.78	0
30.23	A	32	168	0	200	0.84	0
50.39	A	24	176	0	200	0.88	0
83.98	A	22	178	0	200	0.89	0
140.0	A	25	175	0	200	0.88	0
233.3	A	36	164	0	200	0.82	0
388.8	A	37	163	0	200	0.82	0
648.0	A	40	160	0	200	0.80	0
1080	A	28	172	0	200	0.86	0
1800	A	37	163	0	200	0.82	0
3000	A	38	162	0	200	0.81	0
5000	A	34	166	0	200	0.83	0

See Section 7.2 for abbreviations.

**Text Table 3**  
**Data for 24+24 Hour Treatments -S-9, Range-Finder - Female Donors**

Treatment (µg/mL)	Replicate	Mono	Bi	Multi	Total Number of Cells	RI	Cytotoxicity (%)
Vehicle	A	7	178	15	200	1.04	-
	B	21	150	29	200	1.04	
18.14	A	7	165	28	200	1.11	0
30.23	A	10	175	15	200	1.03	1
50.39	A	16	162	22	200	1.03	1
83.98	A	54	106	40	200	0.93	11
140.0	A	30	153	17	200	0.94	10
233.3	A	20	162	18	200	0.99	5
388.8	A	9	159	32	200	1.12	0
648.0	A	14	186	0	200	0.93	11
1080	A	35	165	0	200	0.83	21
1800	A	69	131	0	200	0.66	37
3000	A	50	150	0	200	0.75	28
5000	A	75	125	0	200	0.63	40

See Section 7.2 for abbreviations.

No marked changes in osmolality (shifts of greater than 50 mOsm/kg) or pH (shifts of greater than 1 pH unit) were observed at the highest concentration tested (5000 µg/mL) as compared to the concurrent vehicle controls (individual data not reported).

The results of the cytotoxicity Range-Finder Experiment were used to select suitable maximum concentrations for the Micronucleus Experiment.

The results of the RI determinations from the Micronucleus Experiment were as follows:

**Text Table 4**  
**Data for 3+21 Hour Treatments -S-9, Micronucleus Experiment - Female Donors**

<b>Treatment (µg/mL)</b>	<b>Replicate</b>	<b>Mono</b>	<b>Bi</b>	<b>Multi</b>	<b>Total Number of Cells</b>	<b>RI</b>	<b>Cytotoxicity (%)</b>
Vehicle	A	173	368	13	554	0.71	-
	B	188	344	4	536	0.66	
	C	200	328	6	534	0.64	
	D	193	341	3	537	0.65	
1000	A	163	355	1	519	0.69	0
	B	160	340	14	514	0.72	
2000	A	237	280	13	530	0.58	0
	B	142	387	36	565	0.81	
<b>3000</b>	<b>A</b>	<b>177</b>	<b>393</b>	<b>18</b>	<b>588</b>	<b>0.73</b>	<b>0 #</b>
	<b>B</b>	<b>133</b>	<b>354</b>	<b>36</b>	<b>523</b>	<b>0.81</b>	
<b>4000</b>	<b>A</b>	<b>155</b>	<b>336</b>	<b>27</b>	<b>518</b>	<b>0.75</b>	<b>0 #</b>
	<b>B</b>	<b>176</b>	<b>357</b>	<b>11</b>	<b>544</b>	<b>0.70</b>	
<b>5000</b>	<b>A</b>	<b>197</b>	<b>345</b>	<b>5</b>	<b>547</b>	<b>0.65</b>	<b>0 #</b>
	<b>B</b>	<b>154</b>	<b>355</b>	<b>9</b>	<b>518</b>	<b>0.72</b>	

See Section 7.2 for abbreviations.

# Highlighted concentrations were selected for analysis.

**Text Table 5**  
**Data for 3+21 Hour Treatments +S-9, Micronucleus Experiment - Female Donors**

<b>Treatment (µg/mL)</b>	<b>Replicate</b>	<b>Mono</b>	<b>Bi</b>	<b>Multi</b>	<b>Total Number of Cells</b>	<b>RI</b>	<b>Cytotoxicity (%)</b>
Vehicle	A	145	351	14	510	0.74	-
	B	133	374	25	532	0.80	
	C	141	401	7	549	0.76	
	D	133	495	37	665	0.86	
1000	A	152	410	18	580	0.77	2
	B	127	372	17	516	0.79	
2000	A	113	381	19	513	0.82	5
	B	183	376	11	570	0.70	
<b>3000</b>	<b>A</b>	<b>185</b>	<b>360</b>	<b>25</b>	<b>570</b>	<b>0.72</b>	<b>6 #</b>
	<b>B</b>	<b>141</b>	<b>390</b>	<b>18</b>	<b>549</b>	<b>0.78</b>	
<b>4000</b>	<b>A</b>	<b>120</b>	<b>380</b>	<b>10</b>	<b>510</b>	<b>0.78</b>	<b>2 #</b>
	<b>B</b>	<b>141</b>	<b>362</b>	<b>17</b>	<b>520</b>	<b>0.76</b>	
<b>5000</b>	<b>A</b>	<b>124</b>	<b>441</b>	<b>20</b>	<b>585</b>	<b>0.82</b>	<b>0 #</b>
	<b>B</b>	<b>150</b>	<b>365</b>	<b>30</b>	<b>545</b>	<b>0.78</b>	

See Section 7.2 for abbreviations.

# Highlighted concentrations were selected for analysis.

**Text Table 6**  
**Data for 24+24 Hour Treatments -S-9, Micronucleus Experiment - Female Donors**

Treatment (µg/mL)	Replicate	Mono	Bi	Multi	Total Number of Cells	RI	Cytotoxicity (%)
Vehicle	A	57	395	81	533	1.05	-
	B	87	364	62	513	0.95	
	C	76	380	61	517	0.97	
	D	53	380	70	503	1.03	
250.0	A	81	445	48	574	0.94	7
	B	83	395	37	515	0.91	
<b>500.0</b>	<b>A</b>	<b>91</b>	<b>430</b>	<b>31</b>	<b>552</b>	<b>0.89</b>	<b>11 #</b>
	<b>B</b>	<b>100</b>	<b>405</b>	<b>37</b>	<b>542</b>	<b>0.88</b>	
1000	A	183	340	5	528	0.66	28
	B	133	400	8	541	0.77	
<b>2000</b>	<b>A</b>	<b>200</b>	<b>305</b>	<b>3</b>	<b>508</b>	<b>0.61</b>	<b>37 #</b>
	<b>B</b>	<b>185</b>	<b>350</b>	<b>1</b>	<b>536</b>	<b>0.66</b>	
3000	A	233	285	2	520	0.56	43
	B	225	320	2	547	0.59	
4000	A	284	233	1	518	0.45	54
	B	285	244	1	530	0.46	
<b>5000</b>	<b>A</b>	<b>275</b>	<b>275</b>	<b>1</b>	<b>551</b>	<b>0.50</b>	<b>50 #</b>
	<b>B</b>	<b>273</b>	<b>266</b>	<b>2</b>	<b>541</b>	<b>0.50</b>	

See Section 7.2 for abbreviations.

# Highlighted concentrations were selected for analysis.

## 5.2 Micronucleus Analysis

### 5.2.1 Raw Data

The raw data for the observations on the test article plus positive and vehicle controls are retained by Covance Laboratories Ltd. A summary of the number of cells containing micronuclei is given in Table 9.1 to Table 9.3.

### 5.2.2 Validity of Study

The data in Table 9.1 to Table 9.6, Section 7.3 and Text Table 4 to Text Table 6 confirm that:

1. The binomial dispersion test demonstrated acceptable heterogeneity (in terms of MNBN cell frequency) between replicate cultures (Table 9.4 to Table 9.6)
2. The frequency of MNBN cells in vehicle controls fell within the historical vehicle control (normal) range (Section 7.3)
3. The positive control chemicals induced statistically significant increases in the proportion of MNBN cells. Both replicate cultures at the positive control concentration analysed under each treatment condition demonstrated MNBN cell frequencies that clearly exceeded the normal range (Table 9.1 to Table 9.3)

4. A minimum of 50% of cells had gone through at least one cell division (as measured by binucleate + multinucleate cell counts) in vehicle control cultures at the time of harvest ([Text Table 4](#) to [Text Table 6](#)).

### **5.2.3 Analysis of Data**

Treatment of cells with Lactase, PPL34537 in the absence and presence of S-9 resulted in frequencies of MNBN cells which were similar to and not significantly ( $p \leq 0.05$ ) higher than those observed in concurrent vehicle controls for the majority of all concentrations analysed (all treatments) ([Table 9.1](#) to [Table 9.6](#)). A single exception was observed at the lowest concentration analysed (3000  $\mu\text{g/mL}$ ) following 3+21 hour treatment in the absence of S-9. However, this increase was not large with both replicate cultures exhibiting MNBN cell frequencies within the normal range with no instances of MNBN cell values exceeding normal values for any other Lactase, PPL34537 treated culture (all concentrations, all treatments) ([Section 7.3](#)). As such, this isolated statistical increase was not considered of biological importance.



## **6. CONCLUSION**

It is concluded that Lactase, PPL34537 did not induce micronuclei in cultured human peripheral blood lymphocytes following treatment in the absence and presence of an Aroclor 1254-induced rat liver metabolic activation system (S-9). Concentrations were tested up to 5000 µg/mL, a recommended regulatory maximum concentration for *in vitro* micronucleus assays.

## **7. ASSOCIATED STUDY INFORMATION**

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## 7.2 Abbreviations

Abbreviation	Description
CMS	Covance Management System
CPA	Cyclophosphamide
Cyto-B	Cytochalasin B
DMSO	Dimethyl sulphoxide
G6P	Glucose-6-phosphate
GLP	Good laboratory practice
HEPES	Hydroxyethyl piperazineethane sulphonic acid
KCl	Potassium chloride
MMC	Mitomycin C
MNBN	Micronucleated binucleate cells
NADP	$\beta$ -Nicotinamide adenine dinucleotide phosphate
OECD	Organization for Economic Cooperation and Development
PHA	Phytohaemagglutinin
PBS	Phosphate buffered saline
QA	Quality Assurance
RI	Replication index
RPMI	Roswell Park Memorial Institute
S-9	Rat liver metabolic activation system
SD	Study Director
sd	Standard deviation
SOP	Standard operating procedure
VIN	Vinblastine

### Units of Measure

$\mu\text{g}$	Microgram
$^{\circ}\text{C}$	Degrees Celsius
mOsm/kg	Milliosmole per kilogram
mg	Milligram
mL	Millilitre
mM	Millimolar

### Footnotes to tables

Bi	Binucleate
DF	Degrees of freedom
Mono	Mononucleate
Multi	Multinucleate
NS	Not significant
NSc	Not scored

### 7.3 Historical Vehicle Control Ranges for the Human Peripheral Blood Lymphocyte Micronucleus Assay

		Frequency of MNBN cells/cells scored (%)
		Female donors
3+21 hour-S-9	Number of studies	48
	Number of cultures	128
	Median	0.60
	Mean	0.59
	sd	0.273
	<b>Observed range</b>	<b>0.10 to 1.30</b>
	<b>95% reference range</b>	<b>0.10 to 1.10</b>
3+21 hour +S-9	Number of studies	50
	Number of cultures	129
	Median	0.60
	Mean	0.58
	sd	0.295
	<b>Observed range</b>	<b>0.00 to 1.90</b>
	<b>95% reference range</b>	<b>0.20 to 1.20</b>
24+24 hour –S-9	Number of studies	17
	Number of cultures	120
	Median	0.40
	Mean	0.42
	sd	0.278
	<b>Observed range</b>	<b>0.00 to 1.70</b>
	<b>95% reference range</b>	<b>0.05 to 1.15</b>

Reference ranges are calculated from percentiles of the observed distributions.

Calculated in November 2012 by Covance Laboratories Statistics, for studies started between March 2011 and August 2012.

---

#### 7.4 Protocol Deviation

---

Procedure	Protocol Deviation
Blood culture preparation	The rocking platform used to gently rock the blood cultures for 48 hours after culture set up for the main Micronucleus Experiment was found to be malfunctioning on the day of treatment. As such it cannot be ascertained as to how long the cultures had not been rocked continuously prior to treatment. However, as the vehicle control cultures demonstrated acceptable Replication index values (indicating suitable cell proliferation), this observation was not considered to have any adverse impact on the integrity of the study.

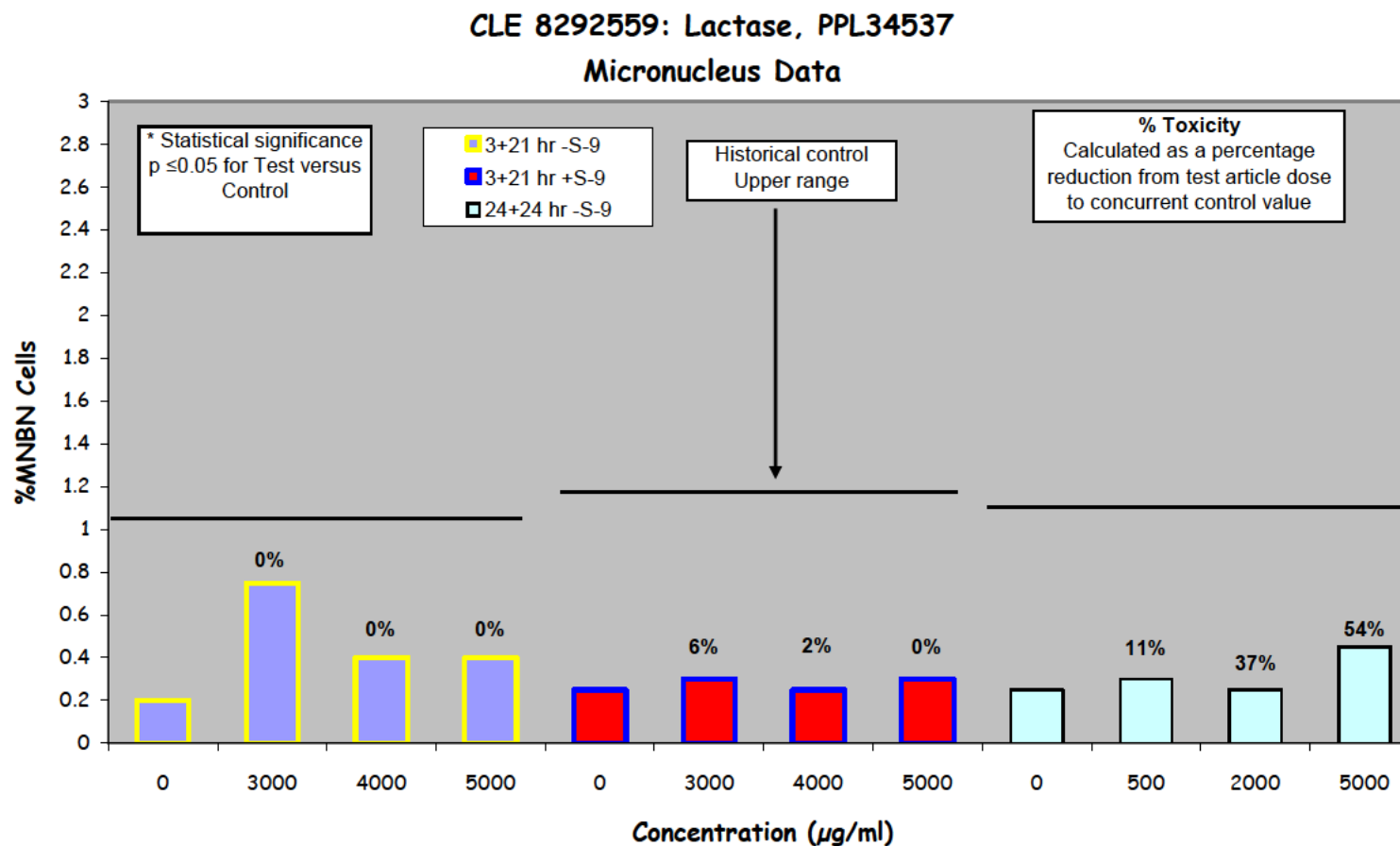
---

This study deviation neither affected the overall interpretation of study findings nor compromised the integrity of the study.

**8. FIGURE**



Figure 8.1: Graphical Plot



## **9. TABLES**

**Table 9.1: Binucleate Cells with Micronuclei: Lactase, PPL34537, 3+21 Hour Treatments in the Absence of S-9 Micronucleus Experiment - Female Donors**

Treatment (µg/mL)	Replicate	Total BN Cells Scored	Total MNBN Cells Scored	Frequency of MNBN Cells/ Cells Scored (%)	Significance § (% Toxicity)
Vehicle	A	1000	2	0.20	-
	B	1000	2	0.20	
	Total	2000	4	0.20	
3000	A	1000	5	0.50	p≤0.01 (0)
	B	1000	10	1.00	
	Total	2000	15	0.75	
4000	A	1000	6	0.60	NS (0)
	B	1000	2	0.20	
	Total	2000	8	0.40	
5000	A	1000	5	0.50	NS (0)
	B	1000	3	0.30	
	Total	2000	8	0.40	
MMC, 0.80	A	1000	85	<b>8.50 #</b>	p≤0.001
	B	1000	113	<b>11.30 #</b>	
	Total	2000	198	9.90	

MNBN = Micronucleated Binucleate

§ Statistical significance (Table 9.4)

NS = Not significant

# = Numbers highlighted exceed historical negative control range (Section 7.3)

**Table 9.2: Binucleate Cells with Micronuclei: Lactase, PPL34537, 3+21 Hour Treatments in the Presence of S-9 Micronucleus Experiment - Female Donors**

Treatment (µg/mL)	Replicate	Total BN Cells Scored	Total MNBN Cells Scored	Frequency of MNBN Cells/ Cells Scored (%)	Significance § (% Toxicity)
Vehicle	A	1000	2	0.20	-
	B	1000	3	0.30	
	Total	2000	5	0.25	
3000	A	1000	3	0.30	NS (6)
	B	1000	3	0.30	
	Total	2000	6	0.30	
4000	A	1000	2	0.20	NS (2)
	B	1000	3	0.30	
	Total	2000	5	0.25	
5000	A	1000	3	0.30	NS (0)
	B	1000	3	0.30	
	Total	2000	6	0.30	
CPA, 6.25	A	1000	35	<b>3.50 #</b>	p<0.001
	B	1000	34	<b>3.40 #</b>	
	Total	2000	69	3.45	

MNBN = Micronucleated Binucleate

§ Statistical significance (Table 9.5)

NS = Not significant

# = Numbers highlighted exceed historical negative control range (Section 7.3)

**Table 9.3: Binucleate Cells with Micronuclei: Lactase, PPL34537, 24+24 Hour Treatments in the Absence of S-9  
Micronucleus Experiment - Female Donors**

Treatment (µg/mL)	Replicate	Total BN Cells Scored	Total MNBN Cells Scored	Frequency of MNBN Cells/ Cells Scored (%)	Significance § (% Toxicity)
Vehicle	A	1000	2	0.20	-
	B	1000	3	0.30	
	Total	2000	5	0.25	
500.0	A	1000	4	0.40	NS (11)
	B	1000	2	0.20	
	Total	2000	6	0.30	
2000	A	1000	4	0.40	NS (37)
	B	1000	1	0.10	
	Total	2000	5	0.25	
5000	A	1000	2	0.20	NS (54)
	B	1000	7	0.70	
	Total	2000	9	0.45	
VIN, 0.10	A	554	15	<b>2.71 #</b>	p≤0.001
	B	521	19	<b>3.65 #</b>	
	Total	1075	34	3.16	

MNBN = Micronucleated Binucleate

§ Statistical significance (Table 9.6)

NS = Not significant

# = Numbers highlighted exceed historical negative control range (Section 7.3)

**Table 9.4: Statistical Analysis of Test Article Data: Lactase, PPL34537, 3+21 Hour Treatments in the Absence of S-9 Micronucleus Experiment - Female Donors**

Binomial Dispersion Test $\chi^2 = 4.19$	DF: 4
Significance: NS	

Treatment ( $\mu\text{g/mL}$ )	Total BN Cells	BN Cells with micronuclei	Proportion	Fisher's exact test	Significance
Vehicle	2000	4	0.002	-	-
3000	2000	15	0.008	0.006	$p \leq 0.01$
4000	2000	8	0.004	0.133	NS
5000	2000	8	0.004	0.133	NS
MMC 0.80	2000	198	0.099	0.000	$p \leq 0.001$

**Table 9.5: Statistical Analysis of Test Article Data: Lactase, PPL34537, 3+21 Hour Treatments in the Presence of S-9 Micronucleus Experiment - Female Donors**

Binomial Dispersion Test $\chi^2 = 0.40$	DF: 4
Significance: NS	

Treatment ( $\mu\text{g/mL}$ )	Total BN Cells	BN Cells with micronuclei	Proportion	Fisher's exact test	Significance
Solvent	2000	5	0.003	-	-
3000	2000	6	0.003	0.387	NS
4000	2000	5	0.003	0.500	NS
5000	2000	6	0.003	0.387	NS
CPA 6.25	2000	69	0.035	0.000	$p \leq 0.001$

NS = Not significant

DF = Degrees of freedom

BN = Binucleate

**Table 9.6: Statistical Analysis of Test Article Data: Lactase, PPL34537,  
24+24 Hour Treatment in the Absence of S-9  
Micronucleus Experiment - Female Donors**

Binomial Dispersion Test $\chi^2 = 5.46$ Significance: NS	DF: 4
--------------------------------------------------------------	-------

Treatment ( $\mu\text{g/mL}$ )	Total BN Cells	BN Cells with micronuclei	Proportion	Fisher's exact test	Significance
Vehicle	2000	5	0.003	-	-
500.0	2000	6	0.003	0.387	NS
2000	2000	5	0.003	0.500	NS
5000	2000	9	0.005	0.150	NS
VIN 0.10	1075	34	0.032	0.000	$p \leq 0.001$

NS = Not significant

DF = Degrees of freedom

BN = Binucleate

## **ATTACHMENTS**



## Certificate of Analysis




### Toxicology

Date: 12. March, 2013  
Project no.: DEV00917  
Luna: 2012-18502-01  
Ref.: SuH

## Certificate of Analysis

Product:	TOX BATCH
Batch:	PPL34537
Type of enzyme:	Lactase
Host organism:	<i>Bacillus licheniformis</i>
Physical form / Colour:	Brownish liquid at room temperature
E.C.:	3.2.1.23

Activity:	3260 LAU(A)/g
Water (KF):	90.7 % w/w
Dry matter:	9.3 % w/w
Ash (600°C):	2.9 % w/w
Total Organic Solids (TOS):	6.4 % w/w
Specific gravity (g/ml):	1.050 g/ml
pH:	7.5
Total viable counts/g:	6800

  
Study Director

## Quality Control Statement for S-9

# MOLTOX<sup>®</sup>

Molecular Toxicology, Inc.

### POST MITOCHONDRIAL SUPERNATANT (S9) QUALITY CONTROL & PRODUCTION CERTIFICATE

Animal Information	Part Number Information	PREP: <u>May 08, 2013</u>
SPECIES: <u>Rat</u>	LOT NO.: <u>3091</u>	EXPIRY: <u>May 08, 2015</u>
STRAIN: <u>Sprague Dawley</u>	PART NO.: <u>11-101</u>	INDUCING AGENT: <u>Aroclor</u>
SEX: <u>Male</u>	VOLUME: <u>5 ml</u>	<u>1254, (Monsanto KL615), 500</u>
AGE: <u>5 - 6 weeks</u>	BUFFER: <u>0.15 M KCl</u>	<u>mg/kg i.p.</u>
WEIGHT: <u>175 - 199 g</u>	STORAGE: <u>At or below -70°C</u>	
TISSUE: <u>Liver</u>		

REFERENCE: Maron, D & Ames, B., *Mutat Res*, 113: 173, 1983.

For Research Purposes Only

BIOCHEMISTRY: Assayed according to the method of Lowry et al., *JBC* 193:265, 1951  
using bovine serum albumin as the standard.

- PROTEIN: 36.2 mg/ml

#### - ALKOXYRESORUFIN-0-DEALKYLASE ACTIVITIES

Activity	P450	Fold - Induction	
BROD	2B1, 2B2	59.4	Assays for ethoxyresorufin-0-deethylase (EROD), pentoxy-, benzyl- and methoxyresorufin-0-dealkylases (PROD, BROD, & MROD) were conducted using a modification of the methods of Burke, et al., <i>Biochem Pharm</i> 34:3337, 1985. Fold-inductions were calculated as the ratio of the sample vs. uninduced specific activities (SA's). Control SA's (pmoles/min/ mg protein) were 72.3, 48.3, 13.3, & 22.3 for BROD, EROD, MROD and PROD, respectively.
EROD	1A1, 1A2	79.8	
MROD	1A1, 1A2	102.7	
PROD	2B1, 2B2	46.2	

#### BIOASSAY:

##### - TEST FOR THE PRESENCE OF ADVENTITIOUS AGENTS

Samples of S-9 were assayed for the presence of contaminating microflora by plating 1.0 ml volumes on Nutrient Agar and Minimal Glucose (Vogel-Bonner E, supplemented with 0.05 mM L-histidine and D-biotin) media. Duplicate plates were read after 40 - 48 h incubation at 35 ± 2°C. The tested samples met acceptance criteria.

##### - PROMUTAGEN ACTIVATION

No. His+ Revertants	
TA98	TA1535
472.4	1590

The ability of the sample to activate ethidium (EtBr) and cyclophosphamide (CPA) to intermediates mutagenic to TA98 and TA1535, respectively, was determined according to Lesca, et al., *Mutation Res* 129: 299, 1984. Data were expressed as revertants per µg EtBr or per mg CPA.

Dilutions of the sample S9, ranging from 0.2 - 10% in S9 mix, were tested for their ability to activate benzo(a)pyrene (BP) and 2-aminoanthracene (2-AA) to intermediates mutagenic to TA100. Assays were conducted as described by Maron & Ames, (*Mutat Res* 113: 173, 1983).

#### µl S9 per plate/number his<sup>+</sup> revertants per plate

Promutagen	0	1	5	10	20	50
BP (5 µg)	91	229	488	672	736	1462
2-AA (2.5 µg)	90	479	1535	1954	2735	2049

Approved: [REDACTED] 05/10/1

MOLECULAR TOXICOLOGY, INC.

## **REPORT**

### **Lactase, batch PPL34537: Toxicity Study by Oral Gavage Administration to Sprague-Dawley Rats for 13 Weeks**

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<b>HLS study number:</b>	LKG0074
<b>Sponsor reference number:</b>	20136077
<b>Version ID:</b>	Final Report
<b>Issue date:</b>	04 August 2014

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## Compliance with Good Laboratory Practice

### Lactase, batch PPL34537: Toxicity Study by Oral Gavage Administration to Sprague-Dawley Rats for 13 Weeks

The study described in this report was conducted in compliance with the following Good Laboratory Practice standards and I consider the data generated to be valid.

The UK Good Laboratory Practice Regulations (Statutory Instrument 1999 No. 3106, as amended by Statutory Instrument 2004 No. 994).

OECD Principles of Good Laboratory Practice (as revised in 1997), ENV/MC/CHEM(98)17.

EC Commission Directive 2004/10/EC of 11 February 2004 (Official Journal No. L 50/44).

The analysis of the test formulation was performed by the Principal Investigator in compliance with the following Good Laboratory Practice standard.

OECD Principles of Good Laboratory Practice (as revised in 1997), ENV/MC/CHEM(98)17.

These principles of Good Laboratory Practice are accepted by the regulatory authorities of the United States of America and Japan on the basis of intergovernmental agreements.



Study Director  
Huntingdon Life Sciences

CBiol MSB

4 August 2014  
Date

## Quality Assurance Statement

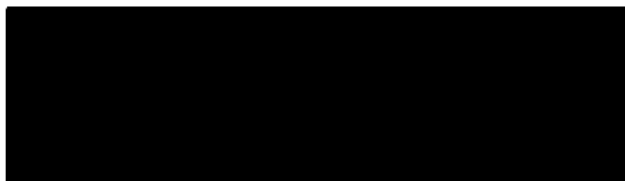
### Lactase, batch PPL34537: Toxicity Study by Oral Gavage Administration to Sprague-Dawley Rats for 13 Weeks

The following inspections and audits have been carried out in relation to this study:

Study Phase	Date(s) of Inspection	Date of Reporting to Study Director and Management
Protocol Audit	22 Oct 2013	22 Oct 2013
Study set up	20 Nov 2013-21 Nov 2013	21 Nov 2013
Protocol Amendment No.1	02 Dec 2013	02 Dec 2013
Study management & conduct	09 Dec 2014-13 Dec 2013	13 Dec 2013
Pre-terminal	19 Feb 2014	19 Feb 2014
Protocol Amendment Nos. 2 and 3	03 Apr 2014	03 Apr 2014
Report Audit	02 Apr 2014-09 Apr 2014	09 Apr 2014
	28 Apr 2014-29 Apr 2014	29 Apr 2014
	08 Jul 2014	08 Jul 2014

In addition, process based inspections were conducted of other routine and repetitive procedures employed on this type of study at or about the time this study was in progress. Similarly an inspection of the facility where this study was conducted was carried out on an annual basis. These inspections were reported to Company Management.

Details of Quality Assurance inspections and audits relating to the formulation analysis phase are indicated in the Test Site QA statement within the Principal Investigator's report included as Annex 2.



Unit Head  
Department of Quality Assurance  
Huntingdon Life Sciences Ltd

30 July 2014  
Date



## Details of Sponsor, Test Facilities and Test Site

<b>Sponsor</b>	Novozymes A/S Krogshøjvej 36 DK-2880 Bagsværd Denmark
<b>Test facility</b> Primary location	Huntingdon Life Sciences Eye Research Centre Eye Suffolk IP23 7PX UK
<b>Test facility</b> Clinical pathology	Huntingdon Life Sciences Huntingdon Research Centre Woolley Road Alconbury Huntingdon Cambridgeshire PE28 4HS UK
<b>Test site</b> Formulation analysis	Novozymes A/S Enzyme Analytical Laboratory 6E1.18 Krogshøjvej 36 Building 6E 2880 Bagsværd Denmark

## Summary

The objective of this study was to assess the systemic toxic potential of Lactase, batch PPL34537 (an enzyme used in the food industry), when administered orally by gavage to Sprague-Dawley (CrI:CD(SD)) rats for 13 weeks. Three groups, each comprising 10 males and 10 females, received doses of 10, 33 or 100% of the Lactase batch (equivalent to 0.067, 0.222 or 0.672 g TOS/kg/day, or 7067, 23319 or 70665 LAU(B)/kg/day). A similarly constituted control group received the vehicle (reverse osmosis water) at the same volume-dose (10 mL/kg body weight).

During the study, clinical condition, detailed physical and arena observations, sensory reactivity, grip strength, motor activity, body weight, food consumption, water consumption (by visual assessment), ophthalmic examination, haematology (peripheral blood), blood chemistry, organ weight, macropathology and histopathology investigations were undertaken.

## Results

General appearance and behaviour, sensory reactivity responses, grip strength and motor activity were not affected by treatment, there were no deaths during the treatment period and there was no effect of treatment on bodyweight gain or on food and water consumption.

There were no treatment-related ophthalmic findings.

There were no treatment-related haematological findings.

Organ weights were unaffected and there were no treatment-related macroscopic or microscopic findings.

Analysis of the blood plasma during Week 13 revealed high alkaline phosphatase activity in males receiving 33% of the Lactase batch and in both sexes receiving 100% of the Lactase batch and low urea and creatinine and high potassium concentrations in males receiving 100% of the Lactase batch. However, as there was no change in organ weight or any macroscopic or microscopic findings and no other biochemical markers for organ damage were observed, these findings were considered most likely to represent adaptive changes associated with administration of high doses of a xenobiotic and were therefore considered of no toxicological significance.

## Conclusion

It is concluded that oral administration of Lactase, batch PPL34537, to Sprague-Dawley rats at doses up to 100% of the Lactase batch (0.672 gTOS/kg/day or 70665 LAU(B)/kg/day) for 13 weeks was well-tolerated and did not cause any adverse change. The no-observed-adverse-effect level (NOAEL) was considered to be 100% of the Toxbatch (equivalent to 0.672 gTOS/kg/day or 70665 LAU(B)/kg/day).

## **1. Introduction**

### **1.1 Objective**

The objective of this study was to assess the systemic toxic potential of Lactase, batch PPL34537 (an enzyme used in the food industry), when administered orally, by gavage, to Sprague-Dawley rats for 13 weeks.

### **1.2 Regulatory compliance**

The study was designed to meet the requirements of the following guideline:

Organisation for Economic Co-operation and Development, Testing of Chemicals  
Guideline No. 408 (revised 1998).

The study was conducted in accordance with the requirements of current, internationally recognised Good Laboratory Practice Standards and the applicable sections of the United Kingdom Animals (Scientific Procedures) Act 1986 Amendment Regulations 2012.

### **1.3 Test system**

The rat was chosen as the test species because it is accepted as a predictor of toxic change in man and the requirement for a rodent species by regulatory agencies. The Sprague-Dawley (CrI:CD(SD)) strain was used because of the historical control data available at this laboratory.

### **1.4 Route of administration**

The oral route of administration was chosen to simulate the conditions of potential human exposure.

### **1.5 Treatment groups and doses**

The doses used in this study (10, 33 or 100% of batch PPL34537; equivalent to 0.067, 0.222 or 0.672 g TOS/kg/day or 7067, 23319 or 70665 LAU(B)/kg/day) were selected in conjunction with the Sponsor, based on findings from a two-week preliminary study performed at this laboratory (Huntingdon Life Sciences Study No. [LKG0071](#)) in which the no-observed-adverse-effect level (NOAEL) was considered to be 100% of Lactase, batch PPL34537 (equivalent to 0.672 g TOS/kg/day or 34230 LAU(A)/kg/day).

The highest dose (100% of Lactase, batch PPL34537) was the maximum practical dose and represents administration of the enzyme, as received, at a volume dosage of 10 mL/kg body weight. The lower doses were selected using an approximate ratio of 3.3 between doses.

## 2. Experimental Procedure

### 2.1 Study schedule and structure

#### 2.1.1 Duration of treatment

Minimum period 13 weeks

Treatment continued throughout the necropsy period and serial observations were recorded at appropriate intervals.

#### 2.1.2 Time schedule

Study initiation: 24 October 2013  
(Protocol signed by Study Director)

Experimental start date: 6 November 2013  
(Animal arrival)

Treatment commenced: 21 November 2013

Necropsy: 20 to 21 February 2014

Experimental completion date: 11 April 2014  
(pathology completed)

Study completion: When the Study Director signs the final report

#### 2.1.3 Identity of treatment groups

The study consisted of one control and three treated groups identified as follows:

Group	Treatment	Dose (% of Toxbatch)	Dose (g TOS/kg/day)	Dose (LAU(B)/kg/day)
1	Control	0	0	0
2	Lactase, batch PPL34537	10	0.067	7067
3	Lactase, batch PPL34537	33	0.222	23319
4	Lactase, batch PPL34537	100	0.672	70665

Group	Number of animals		Cage numbers		Animal numbers	
	Male	Female	Male	Female	Male	Female
1	10	10	1-2	15-16	1-10	131-140
2	10	10	5-6	13-14	21-30	121-130
3	10	10	3-4	9-10	11-14, 16-20, 42	101-103, 105-110, 141
4	10	10	7-8	11-12	31-40	111-120

Some serial observations needed to be performed without the knowledge of the treatment group and, consequently, the animal numbering system was such that it was not easy to identify a treatment group from the animal number.

## 2.2 Test substance, vehicle and formulation

### 2.2.1 Test substance

Information supplied by the Sponsor regarding the test substance is contained in the test substance data sheet, which is retained in study records, and the Certificate of Analysis, which is presented in [Annex 1](#).

The following information is given in summary:

Test substance:	Lactase, batch PPL34537
Intended use:	An enzyme used in the food industry
Description:	Brownish liquid
Storage conditions:	Deep frozen (nominally -20°C). Thawed and refrozen on no more than one occasion.
Supplier:	Sponsor
Batch number:	PPL34537
Water (KF)	90.7% w/w
Dry matter	9.3% w/w
Ash content (600°C)	2.9% w/w
Total organic solids (TOS)	6.4% w/w
Specific gravity (g/mL)	1.050 g/mL
pH	7.5
Total viable counts/g	6800
Activity	6730 LAU(B)/g
Expiry date:	20 November 2022

### 2.2.2 Vehicle

The vehicle was reverse osmosis water.

### 2.2.3 Formulation

Group	Treatment	Dose (% of Toxbatch)	Dose (LAU(A)/kg/day)	Volume dose (mL/kg)
1	Control	0	0	10
2	Lactase, batch PPL34537	10	7067	10
3	Lactase, batch PPL34537	33	23319	10
4	Lactase, batch PPL34537	100	70665	10

Vehicle Reverse osmosis water.

Method of preparation The test enzyme was provided (deep frozen) in a number of containers that were divided further into aliquots for each day of treatment in the Huntingdon Life Sciences Pharmacy Department. At appropriate intervals during the study, containers of test material were thawed overnight in a refrigerator (at approximately 4°C) and divided to provide aliquots of sufficient size for the required daily formulation, which were then re-frozen pending use. The containers were inverted 10 times and gentle magnetic stirring was performed during the aliquoting process.

The day before formulation, the frozen aliquots for that day were removed from the freezer and allowed to thaw overnight in a refrigerator (approximately 4°C).

The test enzyme was then prepared for administration as a series of graded concentrations in the vehicle to provide the required concentrations. Each formulation was prepared by gentle magnetic stirring, in order not to damage the enzyme, of the total required quantity of Lactase, batch PPL34537 and vehicle.

Frequency Daily (administered within 24 hours of preparation).

Test substance accounting Detailed records of compound usage were maintained. The amount of test substance necessary to prepare the formulations and the amount actually used were determined on each occasion. The difference between these amounts was checked before the formulations were dispensed.

#### 2.2.4 Formulation analysis

Liquid formulation	Assessment of homogeneity was not relevant as the liquid enzyme was completely miscible with water and formed a solution.
Achieved concentration	Samples of each formulation prepared for administration in Weeks 1, 6 and 13 of treatment were analysed for achieved concentration of the test substance. A total of 6 x 5 mL samples were taken from the middle of each formulation (Groups 1 to 4) that was prepared for administration in Weeks 1, 6 and 13, and all samples were frozen at approximately -20°C upon completion of sampling. Three samples from each formulation/occasion were subsequently dispatched (deep frozen on dry ice) to the Principal Investigator responsible for formulation analysis. All remaining samples were retained (at approximately -20°C) at Huntingdon Life Sciences as a contingency but will be discarded after finalisation of the formulation analysis report since no further analysis was required.
Analysis	The method of analysis and results are presented in <a href="#">Annex 2</a> .

#### 2.3 Administration

Route	Oral, by gavage, using a suitably graduated syringe and a rubber catheter inserted via the mouth.
Treated at	Constant doses in terms of % of the supplied batch.
Volume dose	10 mL/kg body weight.
Individual dose volume	Calculated from the most recently recorded scheduled body weight.
Control (Group 1)	Vehicle at the same volume dose as the treated groups.
Frequency	Once daily at approximately the same time each day.
Storage	Formulations were administered on the same day as preparation, and stored at ambient temperature prior to dosing.
Formulation	<p>Solutions were stirred gently (in order not to damage the enzyme protein and ensure that a homogenous solution was obtained) using a magnetic stirrer before and throughout the dosing procedure.</p> <p>A daily record of the usage of formulation was maintained based on weights. This balance was compared with the expected usage as a check of correct administration. No significant discrepancy was found.</p>

## 2.4 Animal management

### 2.4.1 Animals, acclimatisation and allocation

#### Animals

Strain/Species	Sprague-Dawley; CrI:CD(SD) rat.
Supplier	Charles River (UK) Ltd.
Number of animals	44 males and 44 females.
	Spare animals were removed from the study room after treatment commenced.
Duration of acclimatisation	At least one week.
Age of the animals at start of treatment	6 to 7 weeks old (approximately).
Weight range of the animals at the start of treatment	Males: 231 g to 292 g Females: 178 g to 222 g

The body weight range of the animals that were supplied for this study was slightly different to that which had been specified within the protocol but this had no impact on the integrity of the study (See Section [2.10](#)).

#### Allocation and identification

Allocation	Randomly allocated on arrival.
	Using the sequence of cages in the battery, one animal at a time was placed in each cage with the procedure being repeated until each cage held the appropriate number of animals. Each sex was allocated separately.
Identification of animals	Each animal was assigned a number and identified uniquely within the study by a microchip inserted shortly after arrival.
Identification of cages	Each cage label was colour-coded according to group and was numbered uniquely with cage and study number, as well as the identity of the occupants.



## Animal replacement

Individuals rejected during the acclimatisation period were replaced with spare animals of suitable weight from the same batch.

Replacement before treatment commenced	Clinical condition - animal had a wet skin abrasion in the area of the microchip insertion	One male
	Body weight range extremes	One female

## 2.4.2 Animal housing, diet and water supply

### Environmental control

Rodent facility	Limited access - to minimise entry of external biological and chemical agents and to minimise the transference of such agents between rooms.
Air supply	Filtered fresh air which was passed to atmosphere and not recirculated.
Temperature and relative humidity	Monitored and maintained within the range of 19-23°C and 40-70%.  There were no deviations from these ranges.
Lighting	Artificial lighting, 12 hours light: 12 hours dark.
Electricity supply	Public supply with automatic stand-by generators.

### Animal accommodation

Cages	Polycarbonate body with a stainless steel mesh lid, changed at appropriate intervals.
Cage distribution	Males and females were blocked by sex and the cages constituting each group were dispersed in batteries so that possible environmental influences arising from their spatial distribution were equilibrated, as far as was practicable. The positions of the cage batteries in the room were changed weekly, following a rotation plan, to further minimise possible effects of spatial variations.
Number of animals per cage	Five of the same sex.
Bedding	Wood based bedding which was changed at appropriate intervals each week.

## **Environmental enrichment**

Aspen chew block	Provided to each cage throughout the study and replaced when necessary.
Plastic shelter	Provided to each cage throughout the study and replaced when necessary.

## **Diet supply**

Diet	Rat and Mouse No. 1 Maintenance Diet.
Availability	Non-restricted (removed overnight before blood sampling for haematology or blood chemistry).

## **Water supply**

Supply	Potable water from the public supply via polycarbonate bottles with sipper tubes. Bottles were changed at appropriate intervals.
Availability	Non-restricted.

## **Supplier certificates of analysis**

Certificates of analysis for the diet were scrutinised and approved before any batch of diet was released for use. Certificates of analysis are routinely provided by the water supplier.

Certificates of analysis were also received from the suppliers of the wood based bedding and Aspen chew blocks.

No specific contaminants were known that may have interfered with or prejudiced the outcome of the study and therefore no special assays were performed.

## **2.5 Serial observations**

### **2.5.1 Clinical and behavioural observations**

Animals were inspected visually at least twice daily for evidence of ill-health or reaction to treatment. Cages were inspected daily for evidence of animal ill-health amongst the occupants. Any deviation from normal was recorded at the time in respect of nature and severity, date and time of onset, duration and progress of the observed condition, as appropriate.

During the acclimatisation period, observations of the animals and their cages were recorded at least once per day.

## **Signs associated with dosing**

Daily during the first week of treatment and twice weekly thereafter, detailed observations were recorded at the following times in relation to dose administration:

Pre-dose observation

One to two hours after completion of dosing all groups

As late as possible in the working day (recorded during Week 1 only)

## **Detailed physical examination and arena observations**

Before treatment commenced and during each week of treatment, detailed physical examination and arena observations were performed on each animal. On each occasion, the examinations were performed at approximately the same time of day (before dosing during the treatment period), by an observer unaware of the experimental group identities.

After removal from the home cage, animals were assessed for physical condition and behaviour during handling and after being placed in a standard arena. Any deviation from normal was recorded with respect to the nature and, where appropriate, degree of severity. Particular attention was paid to possible signs of neurotoxicity, such as convulsions, tremor and abnormalities of gait or behaviour.

Findings were either reported as "present" or assigned a severity grade - slight, moderate or marked.

## **Sensory reactivity and grip strength**

Sensory reactivity and grip strength assessments were performed (before dosing) on all animals during Week 12 of treatment. Animals were tested by an observer who was unaware of the treatment group to which each animal belonged. Before the start of observations, cage labels showing the treatment group were replaced by labels stating only the study, animal and cage numbers. Animals were not necessarily all tested on the same day, but the numbers of animals and the times of testing were balanced across the groups on each day of testing.

The following measurements, reflexes and responses were recorded:

### **Approach response**

A blunt probe was brought towards the animal's head until it was close to the animal's nose (but not touching the whiskers). The animal's reaction was recorded as:

- 1 No reaction or ignores probe/walks past probe
- 2 Normal awareness and reaction e.g. approaches and/or sniffs probe
- 3 Abnormally fearful or aggressive reaction

### **Pinna reflex**

The inside of one ear was touched lightly with a nylon filament and the reaction recorded as:

- 1 No response
- 2 Normal response e.g. ear twitches/flattens or animal shakes its head
- 3 Abnormally fearful or aggressive response

### **Auditory startle reflex**

The animal's response to a sudden sharp noise was assessed and scored as:

- 1 No response
- 2 Weak response e.g. ear twitch only
- 3 Normal response e.g. obvious flinch or startle
- 4 Exaggerated response e.g. all feet off floor

### **Tail pinch response**

The animal's tail was pinched sharply with forceps approximately one third from the tip and the response graded as:

- 1 No response
- 2 Weak response e.g. turns around slowly or weak vocalization without moving away
- 3 Normal response e.g. jumps forward or turns around sharply, usually with vocalization
- 4 Exaggerated response e.g. excessive vocalization, body movement or aggression

### **Grip strength**

Forelimb and hindlimb grip strength was measured using Mecmesin Force Indicators. Three trials were performed.

At any point during the observations, additional comments were made as free text where considered appropriate.

### **Motor activity**

During Week 12 of treatment (before dosing), the motor activity of each animal was measured using a Rodent Activity Monitoring System (Version 2.0.5), with hardware supplied by Pearson Technical Services and software developed and maintained by Huntingdon Life Sciences.

Animals were tested individually in clear polycarbonate cages and motor activity was measured by counting infra-red beam breaks over ten six-minute intervals (one hour total). Ten beams were set at two height levels (five low and five high) to detect cage floor and rearing activity respectively. Animals were not necessarily all tested on the same day, but the numbers of animals and the times of testing were balanced across the groups on each day of testing.

### **2.5.2 Body weight**

The weight of each animal was recorded one week before treatment commenced (Week -1), on the day that treatment commenced (Week 0), weekly throughout the treatment period and before necropsy.

### **2.5.3 Food consumption**

The weight of food supplied to each cage, that remaining and an estimate of any spilled was recorded for the week before treatment started (Week -1) and for each week throughout the treatment period.

### **2.5.4 Water consumption**

Fluid intake was assessed by daily visual observation. No treatment-related effect was observed and consequently quantitative measurements were not performed.

### **2.5.5 Ophthalmic examination**

The eyes of the animals were examined by means of a binocular indirect ophthalmoscope as follows:

<b>Occasion</b>	<b>Animals</b>
Pretreatment	All animals (including spares)
Week 12	All animals of Groups 1 and 4

Prior to each examination, the pupils of each animal were dilated using tropicamide ophthalmic solution (Mydriacyl). The adnexae, conjunctiva, cornea, sclera, anterior chamber, iris (pupil dilated), lens, vitreous and fundus were examined.

### **2.5.6 Haematology, peripheral blood**

During Week 13 of treatment, blood samples were collected after overnight withdrawal of food and prior to dosing. Animals were held under light general anaesthesia induced by isoflurane. Blood samples (nominally 0.5 mL) were withdrawn from the sublingual vein, collected into tubes containing EDTA anticoagulant and examined for the following characteristics using a Bayer Advia 120 analyser:

- Haematocrit (Hct)
- Haemoglobin concentration (Hb)
- Erythrocyte count (RBC)
- Absolute reticulocyte count (Retic)
- Mean cell haemoglobin (MCH)
- Mean cell haemoglobin concentration (MCHC)
- Mean cell volume (MCV)
- Red cell distribution width (RDW)
- Total leucocyte count (WBC)
- Differential leucocyte count:
  - Neutrophils (N)
  - Lymphocytes (L)

Eosinophils (E)  
Basophils (B)  
Monocytes (M)  
Large unstained cells (LUC)  
Platelet count (Plt)  
Morphology:  
Anisocytosis  
Microcytosis  
Macrocytosis  
Hypochromasia  
Hyperchromasia

Blood film (prepared for all samples) - Romanowsky stain, examined for abnormalities by light microscopy, in the case of flags from the Advia 120 analyser. Confirmation or a written description from the blood film was made where appropriate.

Additional blood samples (nominally 0.5 mL) were taken into tubes containing citrate anticoagulant and examined using an ACL series analyser and appropriate reagent in respect of:

Prothrombin time (PT) - using IL PT-Fibrinogen reagent.

Activated partial thromboplastin time (APTT) - using IL APTT reagent.

## **2.5.7 Haematology, bone marrow**

Bone marrow smears were prepared immediately following death on completion of the scheduled treatment period.

Fixation	Smears were air dried and subsequently fixed in methanol.
Retention	Slides were retained by the Department of Biomarkers, Bioanalysis and Clinical Sciences.
Analysis	At the discretion of the pathologist, examination of smears to further evaluate other findings was not considered necessary; the smears are retained in the archives.

## **2.5.8 Blood chemistry**

During Week 13 of treatment, blood samples were collected after overnight withdrawal of food and prior to dosing.

Animals were held under light general anaesthesia induced by isoflurane. Blood samples (nominally 0.7 mL) were withdrawn from the sublingual vein and collected into tubes containing lithium heparin as anticoagulant. After separation, the plasma was examined using a Roche P Modular Analyser in respect of:

Alkaline phosphatase (ALP)  
Alanine aminotransferase (ALT)  
Aspartate aminotransferase (AST)  
Urea  
Creatinine (Creat)  
Glucose (Gluc)  
Total cholesterol (Chol)  
Sodium (Na)  
Potassium (K)  
Total protein (Prot)  
Albumin (Alb)

Albumin/globulin ratio (A/G Ratio) was calculated from total protein concentration and analysed albumin concentration.

## 2.6 Terminal Procedures

All animals were subject to a detailed necropsy. After a review of the history of each animal, a full macroscopic examination of the tissues was performed. All external features and orifices were examined visually. Any abnormality in the appearance or size of any organ and tissue (external and cut surface) was recorded and the required tissue samples preserved in appropriate fixative.

The retained tissues were checked before disposal of the carcass.

Schedule                                      Animals were killed following 13 weeks of treatment.

Method of kill                                Carbon dioxide asphyxiation with subsequent exsanguination.

Sequence                                      To allow satisfactory inter-group comparison.

The organs weighed, tissue samples fixed and sections examined microscopically are detailed as follows:

	Necropsy		Histology	Pathology
Tissue and regions examined	Weigh	Fix		Light microscopy
Abnormalities		*	*	*
Adrenals	*	*	*	*
Aorta		*	*	*
Bone marrow smears		*		
Brain (cerebellum, cerebrum, midbrain)	*	*	*	*
Caecum		*	*	*
Colon		*	*	*
Duodenum		*	*	*
Epididymides	*	*	*	*
Eyes		*	#	#
Femur (femorotibial joint)		b)	*	*
Head		a)	#	#
Heart (including auricular and ventricular regions)	*	*	*	*
Ileum		*	*	*
Jejunum		*	*	*
Kidneys	*	*	*	*
Liver (section from two lobes)	*	*	*	*
Lungs (section from two major lobes including bronchi)		*	*	*

Tissue and regions examined	Necropsy		Histology	Pathology
	Weigh	Fix		Light microscopy
Lymph nodes - mandibular		*	*	*
- mesenteric		*	*	*
- left axillary		*	*	*
Oesophagus		*	*	*
Ovaries	*	*	*	*
Pancreas		*	*	*
Peyer's patches		*	*	*
Pituitary		*	*	*
Prostate		*	*	*
Rectum		*	*	*
Salivary glands - submandibular		*	†	†
- sublingual		*	†	†
- parotid		*	†	†
Sciatic nerves		*	†	†
Seminal vesicles		*	*	*
Skin with mammary glands (inguinal area)		*	*	*
Spinal cord (transverse and longitudinal sections at the cervical, thoracic and lumbar levels)		*	*	*
Spleen	*	*	*	*
Sternum		*	*	*
Stomach		*	*	*
Testes	*	*	*	*
Thymus	*	*	*	*
Thyroid with parathyroids		*	*	*
Trachea		*	*	*
Urinary bladder		*	*	*
Uterus with cervix	*	*	*	*
Vagina		*	*	*

- a) Including nasal cavity, paranasal sinuses and nasopharynx.  
b) Both hindlimbs retained, one sectioned where appropriate.  
\* Organs weighed, samples fixed or sections examined microscopically.  
# Not processed or examined.  
† Only one examined.

## 2.6.1 Organ weights

Requisite organs were weighed for all animals killed at scheduled termination.

## 2.6.2 Fixation

Tissues were routinely preserved in 10% Neutral Buffered Formalin with the exception of those detailed below:

Testes In modified Davidson's fluid.

Eyes In Davidson's fluid.

Bone marrow smears: See Section [2.5.7](#).



### 2.6.3 Histology

Processing	Tissue samples were dehydrated, embedded in paraffin wax and sectioned at a nominal four to five micron thickness. For bilateral organs, sections of both organs were prepared. A single section was prepared from each of the remaining tissues required.
Full List	All animals of Groups 1 and 4.
Abnormalities only	All animals of Groups 2 and 3.
Routine staining	Sections were stained with haematoxylin and eosin.

### 2.6.4 Light microscopy

Tissues preserved for examination were examined as follows:

Category	Animals	Tissues
Scheduled kill	All animals of Groups 1 and 4. All animals of Groups 2 and 3.	All specified in Section 2.6. Abnormalities only.

Findings were either reported as "present" or assigned a severity grade. In the latter case one of the following five grades was used - minimal, slight, moderate, marked or severe. A reviewing pathologist undertook a peer review of the microscopic findings.

## 2.7 Computer systems

The computer systems that were used on this study to acquire and quantify data include:

System name*	System function
ClinAxys II	Used for haematology and blood chemistry.
Liberate Global	In-house system used for reporting in-life, necropsy, pathology and statistics.
Quasar and SAS	In-house statistical analysis packages.
RAMS Rodent Activity Monitoring System	Used for activity monitoring.
StarTox	In-house statistical analysis package.
Xybion Pristima	Used for in-life, necropsy and pathology data collection and pharmacy test substance management.

\* All version numbers of the systems are maintained by Huntingdon Life Sciences

The computer systems utilised at the Test Site that were used in this study are detailed in [Annex 2](#).

## **2.8 Data treatment**

This report contains serial observations pertaining to all weeks of study completed, together with signs data collected during the necropsy period. In the case of detailed physical examination and arena observations, body weight and food consumption data, only information from the final week of the acclimatisation period are presented.

Summary statistics (e.g. means and standard deviations) presented in this report were calculated from computer-stored individual raw data. Group mean values and standard deviations were frequently calculated using a greater number of decimal places than presented in the appendices. It is, therefore, not always possible to derive exact group values from the data presented in the appendices.

Throughout the report the following abbreviations are used:

M	Male
F	Female
SD	Standard deviation
N	Number of animals/cages examined

Weeks of pre-treatment relate to study weeks as follows:

P1	Week -1 of study
----	------------------

### **2.8.1 Serial observations**

#### **Detailed physical examination and arena observations**

Detailed physical examination and arena observations are presented for each animal providing detail of type of sign, day or week of occurrence and information on the duration of the sign applicable.

#### **Signs associated with dosing**

There were no signs associated with dosing and therefore these data are not presented.

#### **Body weight**

Group mean weight changes were calculated from the weight changes of individual animals.

#### **Food consumption**

Weekly group mean food consumptions and standard deviations were derived from unrounded cage values. Overall mean food consumption values were calculated from the weekly group mean values.

#### **Ophthalmic examination**

For each animal that showed findings, observations are presented in the final report. Observations were bilateral unless otherwise indicated.

## Haematology, peripheral blood

The abbreviations used have the following meanings:

CTD	Clotted sample
NVR	No valid result
INS	Instrument error during initial analysis; insufficient volume of blood remaining for analysis

NVR for platelet results is often reported for samples where it is evident platelet ‘clumping’ has occurred. If this is flagged by the instrument it strongly suggests that the platelet count is not accurate due to the presence of these clumps. In these cases the blood film is examined and if the presence of clumps is confirmed then the test is reported as NVR.

The most common morphological changes, anisocytosis, micro/macrocytosis and hypo/hyperchromasia were recorded according to the following convention:

-	No abnormalities detected
+	Slight
++	Moderate
+++	Marked

## Blood chemistry

The abbreviation used have the following meanings:

NVR	No valid result
-----	-----------------

Albumin to globulin ratio (A/G Ratio) was calculated as:

$$\text{A/G Ratio} = \frac{\text{Albumin concentration}}{\text{Total protein} - \text{albumin concentration}}$$

### 2.8.2 Terminal procedures

#### Organ weights

Organ weights were presented both as absolute/unadjusted and adjusted for terminal body weight, using the weight recorded on the day of necropsy.

### 2.8.3 Statistical analysis

All statistical analyses were carried out separately for males and females using the individual animal as the basic experimental unit.

The following data types were analysed:

- Grip strength and motor activity
- Body weight, using gains over appropriate study periods
- Haematology
- Blood chemistry
- Organ weights, absolute and adjusted for terminal body weight

The following sequence of statistical tests was used for grip strength, motor activity, body weight, organ weight and clinical pathology data:

A parametric analysis was performed if Bartlett's test for variance homogeneity (Bartlett 1937) was not significant at the 1% level. The  $F_1$  approximate test was applied. This test is designed to detect significant departure from monotonicity of means when the main test for the comparison of the means is a parametric monotonic trend test, such as Williams' test (Williams 1971, 1972). The test statistic compares the mean square, NMS, for the deviations of the observed means from the maximum likelihood means, calculated under a constraint of monotonicity with the usual error mean square, EMS. The null hypothesis is that the true means are monotonically ordered. The test statistic is  $F_1 = \text{NMS}/\text{EMS}$  which can be compared with standard tables of the  $F$  distribution with 1 and error degrees of freedom. If the  $F_1$  approximate test for monotonicity of dose-response was not significant at the 1% level, Williams' test for a monotonic trend was applied. If the  $F_1$  approximate test was significant, suggesting that the dose response was not monotone, Dunnett's test (Dunnett 1955, 1964) was performed instead.

A non-parametric analysis was performed if Bartlett's test was still significant at the 1% level following both logarithmic and square-root transformations. The  $H_1$  approximate test, the non-parametric equivalent of the  $F_1$  test described above, was applied. This test is designed to be used when the main test for comparison of the means is a non-parametric monotonic trend test, such as Shirley's test (Shirley 1977). The test statistic compares the non-monotonicity sums of squares, NRSS, for the deviations of the observed mean ranks from the maximum likelihood mean ranks with the non-parametric equivalent of the error sums of squares, ERSS =  $N(N+1)/12$ . The test statistic is  $H_1 = \text{NRSS}/\text{ERSS}$  which can be compared to standard tables of the  $\chi^2$ -distribution with 1 degree of freedom. If the  $H_1$  approximate test for monotonicity of dose-response was not significant at the 1% level, Shirley's test for a monotonic trend was applied. If the  $H_1$  approximate test was significant, suggesting that the dose-response was not monotone, Steel's test (Steel 1959) was performed instead.

For grip strength, motor activity and clinical pathology data, if 75% of the data (across all groups) were the same value, for example c, Fisher's Exact tests (Fisher 1973) were performed. Treatment groups were compared using pairwise comparisons of each dose group against the control both for i) values  $<c$  versus values  $\geq c$ , and for ii) values  $\leq c$  versus values  $>c$ , as applicable.

For organ weight data, analysis of covariance was performed using terminal body weight as covariate (Angervall and Carlstrom, 1963), unless non-parametric methods were applied. The treatment comparisons were made on adjusted group means in order to allow for differences in body weight which might influence the organ weights.

Significant differences between Control and treated groups were expressed at the 5% ( $p<0.05$ ) or 1% ( $p<0.01$ ) level. The key to the annotation used on the tables that contain statistical results is given below:

l	Data were log transformed for the statistical analysis
Du	Treated groups compared with Control using Dunnett's test.
Sh	Treated groups compared with Control using Shirley's test.
Wi	Treated groups compared with Control using Williams' test.
*	p<0.05
**	p<0.01

Codes placed above the adjusted means indicate that the comparisons were based on adjusted means.

## 2.9 Quality assurance and archiving procedures

### 2.9.1 Quality assurance

Details of the Quality Assurance inspections and audits undertaken at Huntingdon Life Sciences are presented on the Quality Assurance Statement.

Details of the Quality Assurance inspections and audits undertaken at the Test Site are presented in [Annex 2](#).

### 2.9.2 Archiving procedures

Following completion of this study all raw data, specimens and samples, except those generated or used during any Sponsor's or supplier's analysis, were stored in the archives of Huntingdon Life Sciences. Types of sample and specimen which are unsuitable, by reason of instability, for long term retention and archiving may be disposed of after the periods stated in Huntingdon Life Sciences Standard Operating Procedures.

The final report will be retained indefinitely and all Quality Assurance inspection records for a period of 20 years. All other appropriate specimens and records will be retained for a minimum period of five years from the date of issue of the final report. At the end of the five year retention period the Sponsor will be contacted and advice sought on the above requirements. Under no circumstances will any item be discarded without the Sponsor's knowledge.

All documentation, samples and data pertaining to the aspects of the study undertaken by the Principal Investigator will be transferred to the archive of the Test Site for storage.

## 2.10 Deviations from protocol

The following deviation from protocol occurred:

The body weight range of the animals that were supplied for use in this study was greater than that which had been documented within the protocol. This deviation had no impact on the integrity of the study since all animals were of the correct age at the commencement of treatment.

### **3. Results**

#### **3.1 Formulation analysis**

##### [Annex 2](#)

The levels of Lactase activity measured in Week 1, 6 and 13 were close to nominal, demonstrating acceptable formulation. The absence of Lactase activity in the control samples was confirmed, demonstrating no inadvertent cross contamination had occurred.

The result of the statistical test showed that there was a significant difference between the dose formulations (group) given in Week 1, 6 and 13, comparing preparations of corresponding levels of activity but the difference between the weeks was consistent across groups and in absolute figures, the observed difference was of no significance.

There was a statistically significant difference between the lactase activity (LAU(B)/g) of the high dose group (Group 4) and the Lactase batch, but in absolute figures the observed difference is evaluated to be practically insignificant.

#### **3.2 Clinical observations and mortality**

##### [Table 1, Appendix 1](#)

The general appearance and behaviour of the animals during the detailed physical examination and the arena observations were not affected by treatment and no animals died during the treatment period.

#### **3.3 Sensory reactivity and grip strength**

##### [Table 2, Appendix 2](#)

Sensory reactivity responses and grip strength were unaffected by treatment.

The group mean hind limb grip strength for females receiving 33 or 100% of the Lactase batch was statistically significantly lower than that of the controls. All scores were, however, within the historical control data range (range 0.37-0.53; mean 0.44) and these inter-group differences were therefore considered of no biological significance.

#### **3.4 Motor activity**

##### [Figure 1, Table 3, Appendix 3](#)

Motor activity in Week 12 was unaffected by treatment.

A difference from controls for the 30-minute period in males receiving 100% of the Lactase batch attained statistical significance but since this was an isolated occurrence and, overall, activities were similar for treated and control animals, it was considered to represent normal biological variation.

### **3.5 Body weight**

[Figure 2](#), [Table 4](#), [Appendix 4](#)

Body weight gain was considered to have been unaffected by treatment.

There was some inter-group variability in overall body weight gain. For example, the body weight gain for all treated male groups and for females receiving 10% of the Lactase batch was slightly lower than that of the controls, but the differences did not attain statistical significance, were not dose-related and were, therefore, considered to have arisen by chance.

### **3.6 Food consumption**

[Table 5](#), [Appendix 5](#)

Food consumption was considered to have been unaffected by treatment.

The amount of food consumed by treated animals tended to be slightly lower than that of the controls. The differences were, however, small (<10%) and were not dose-related (the difference between the mean food intake of females receiving 33% of the Lactase batch and the controls was minimal) and were, therefore, considered to have arisen by chance.

### **3.7 Water consumption**

The visual assessment of water intake did not reveal any clear treatment-related effect and, consequently, quantitative measurements were not performed.

A visual assessment of water intake for Day 4 to 5 revealed that males receiving 33 or 100% of the Lactase batch appeared to consume a greater volume of water than the controls but no further differences were seen throughout the study and this isolated finding was therefore considered a chance occurrence.

### **3.8 Ophthalmic examination**

[Appendix 6](#)

There were no treatment-related ophthalmoscopic findings.

### **3.9 Haematology, peripheral blood**

[Table 6](#), [Appendix 7](#)

The haematological examination performed in Week 13 did not identify any finding that was clearly attributable to treatment.

A number of differences from controls occurred, some of which attained statistical significance, but these were minor or lacked dose-relationship and were therefore attributed to normal biological variation. There was a small increase of mean cell haemoglobin and mean cell haemoglobin concentration at all doses in males but there was no dose-response, no similar finding in females and all individual values were within the background range (90-percentile range 16.4 to 18.9 pg for mean cell haemoglobin and 32.4 to 35.9 g/dL for

mean cell haemoglobin concentration (n=303)). There were also small reductions of reticulocyte count and red cell distribution width in males receiving 100% of the Lactase batch. For reticulocyte count the individual values for 6/10 high dose males were below the lower end of the background range (90-percentile range  $0.190 \times 10^{12}$  to  $0.370 \times 10^{12}/L$  (n=15)) but there was no effect on erythrocyte count, or in females, and this difference from controls was therefore considered of no toxicological significance. The small increase of large unstained cell count in females receiving 10% of the Lactase batch was considered incidental in the absence of a similar trend at the higher doses.

### 3.10 Blood chemistry

#### Table 7, Appendix 8

The biochemical analysis of blood plasma during Week 13 revealed, when compared with controls, high alkaline phosphatase activity in males receiving 33% of the Lactase batch and in both sexes receiving 100% of the Lactase batch. Control values tended, however, to be at the low end of the background range with one control male and two control females below the 90-percentile range and, with the exception of one male given 33% of the Lactase batch, the individual values for treated animals were within the background range (90-percentile range 59 to 124 U/L for males (n=299) and 29 to 78 U/L for females (n=309)).

There was a reduction of plasma urea and creatinine concentrations in males receiving 100% of the Lactase batch. The majority of the individual values for urea were below the concurrent control range but all were within the background range (90-percentile range 4.00 to 7.36 mmol/L (n=299)). For creatinine, with the exception of two males, the individual values were within both the concurrent control and background range (90-percentile range 27 to 46  $\mu\text{mol}/L$  (n=299)).

Plasma potassium concentrations were high, when compared with the control, for males receiving 100% of the Lactase batch, with 6/10 animals having individual values that were above the background range (90-percentile range 4.0 to 6.1 mmol/L (n=289)). The control values also tended to be at the upper limit of the background range, with two animals above the 90-percentile range.

All other differences from controls were minor, were only present in one of the sexes, or lacked dose-relationship and were therefore attributed to normal biological variation. Such differences included the minimal reduction of sodium concentration in males receiving 100% of the Lactase batch.

### 3.11 Organ weights

#### Table 8, Appendix 9

The analysis of organ weights for animals killed after 13 weeks of treatment did not reveal any differences from controls that were attributable to treatment.

The adjusted brain weight was slightly low, when compared with the controls, in males given 33 or 100% of the Lactase batch but the differences from controls were minimal and lacked dose-relationship was therefore attributed to normal biological variation.



### **3.12 Macropathology**

[Table 9](#), [Appendix 10](#), [Annex 3](#)

The macroscopic examination of animals killed after 13 weeks of treatment did not reveal any findings that were attributable to treatment.

The nature and incidence of all findings were consistent with the commonly seen background of macroscopic changes in Sprague-Dawley rats at these laboratories.

### **3.13 Histopathology**

[Table 10](#), [Appendix 10](#), [Annex 3](#)

There were no test substance-related findings.

The incidence and distribution of all histopathological findings were consistent with the common background findings in Sprague-Dawley rats at these laboratories.

## 4. Discussion

The oral administration of Lactase, batch PPL34537 to Sprague-Dawley rats at doses up to 100% of the Lactase batch (equivalent to 0.672 gTOS/kg/day or 70665 LAU(B)/kg/day when administered at a dose volume of 10 mL/kg bodyweight) was well-tolerated, with no evidence of adverse change at any of the administered doses.

An effect of treatment upon the liver was indicated by increased plasma alkaline phosphatase activity in both sexes receiving 100% of the Lactase batch and in males receiving 33% of the Lactase batch. There was, however, no change in liver weight, no macroscopic or microscopic findings in the liver and other biochemical markers for liver damage (alanine amino-transferase and aspartate amino-transferase activities) were similar for treated and control animals. These findings were considered most likely to represent adaptive changes associated with administration of high doses of a xenobiotic and were therefore considered of no toxicological significance.

The biochemical analysis of blood plasma during Week 13 also revealed, when compared with controls, a reduction of plasma urea and creatinine concentrations and high plasma potassium concentrations in males receiving 100% of the Lactase batch, which were indicative of an effect upon the kidneys. The majority of individual values were within historical background ranges and there was no histopathological change in the kidneys. Consequently, these findings were considered not adverse.

The findings in this study therefore indicated that the no-observed-adverse-effect level (NOAEL) was 100% of the Lactase batch (equivalent to 0.672 gTOS/kg/day or 70665 LAU(B)/kg/day).

## **5. Conclusion**

It is concluded that oral administration of Lactase, batch PPL34537, to Sprague-Dawley rats at doses up to 100% of the Lactase batch (0.672 gTOS/kg/day or 70665 LAU(B)/kg/day) for 13 weeks was well-tolerated and did not cause any adverse change. The no-observed-adverse-effect level (NOAEL) was considered to be 100% of the Toxbatch (equivalent to 0.672 gTOS/kg/day or 70665 LAU(B)/kg/day).

## 6. References

### Referenced reports

Allen RL (2003).  
Huntingdon Life Sciences Study Number: LKG0071  
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FIGURE 1 Motor activity - group mean scores (beam breaks) for females during Week 12 of treatment

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

— 1M    - - - 2M    - - - 3M    - · - 4M

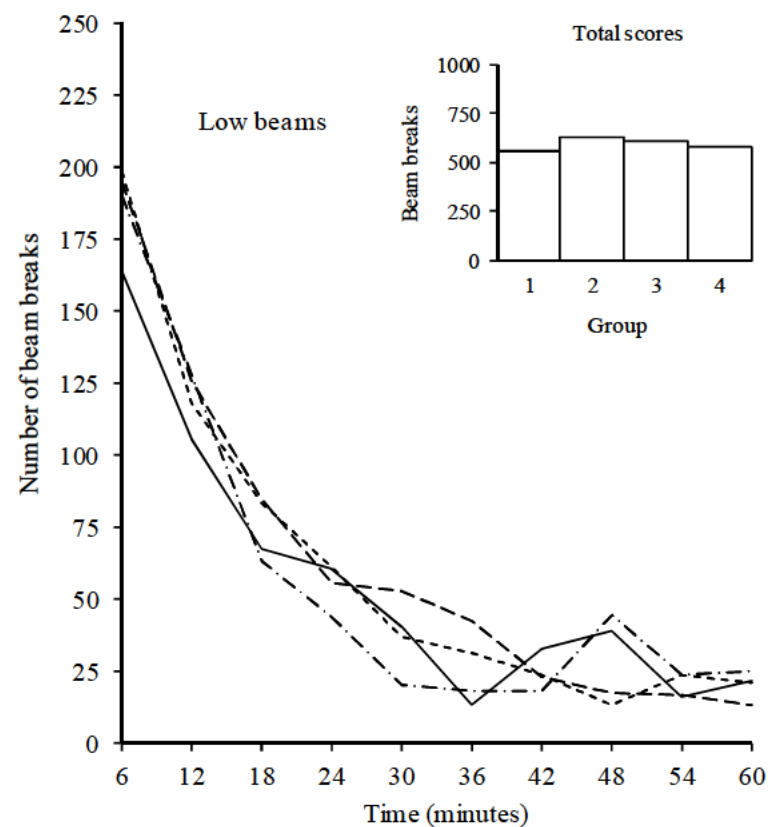
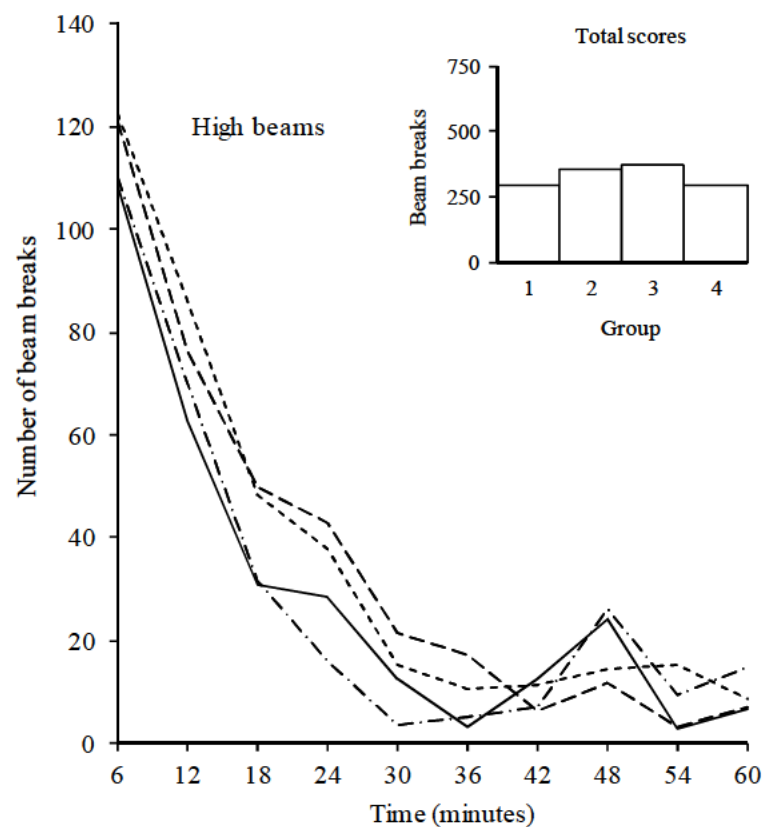


FIGURE 1 (cont) Motor activity - group mean scores (beam breaks) for females during Week 12 of treatment

Dose Group	Control	Lactase, batch PPL34537			
Dose (%)	1	2	3	4	
	0	10	33	100	

— 1F    - - - 2F    - - - 3F    - - - 4F

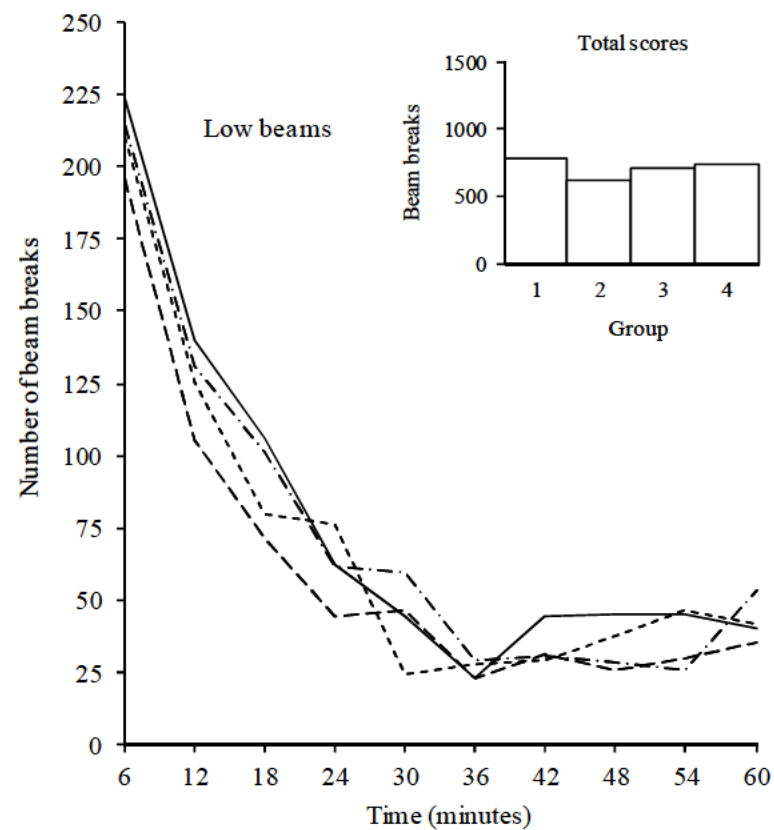
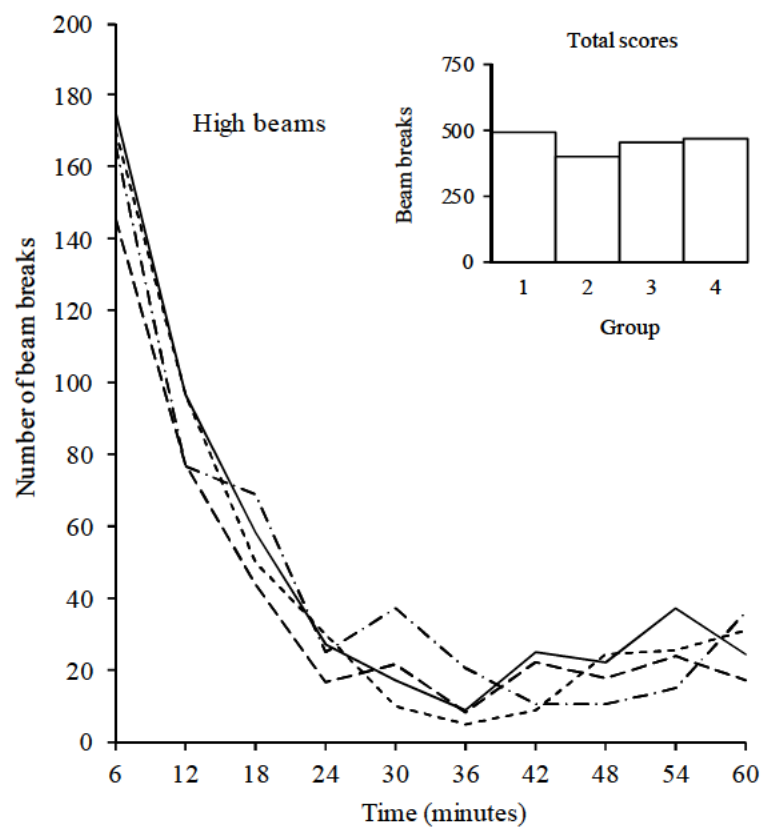


FIGURE 2 Body weight - group mean values (g) for males

Dose Group	Control	Lactase, batch PPL34537			
Dose (%)	1	2	3	4	
	0	10	33	100	

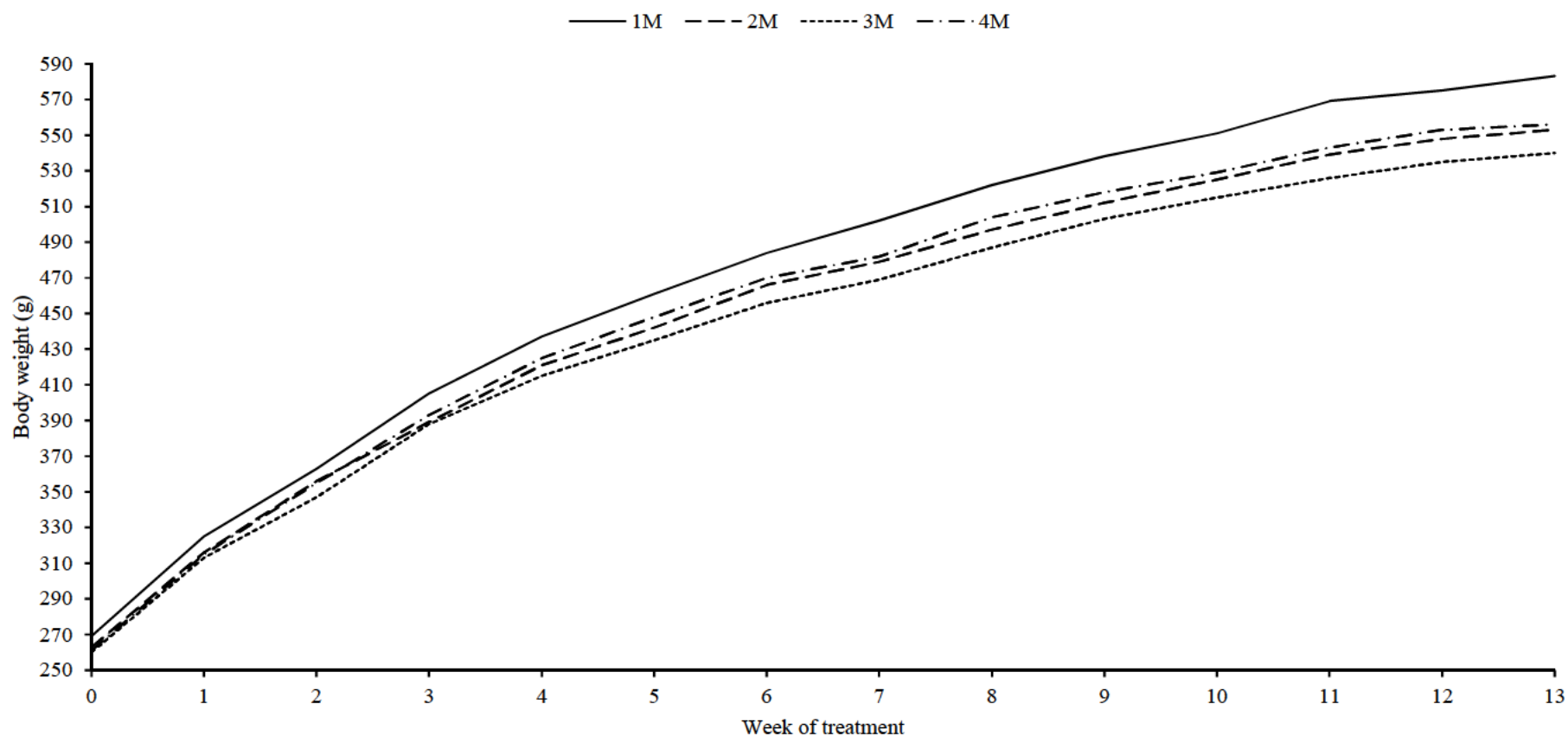


FIGURE 2 (cont) Body weight - group mean values (g) for females

Dose Group	Control	Lactase, batch PPL34537			
Dose (%)	1	2	3	4	
	0	10	33	100	

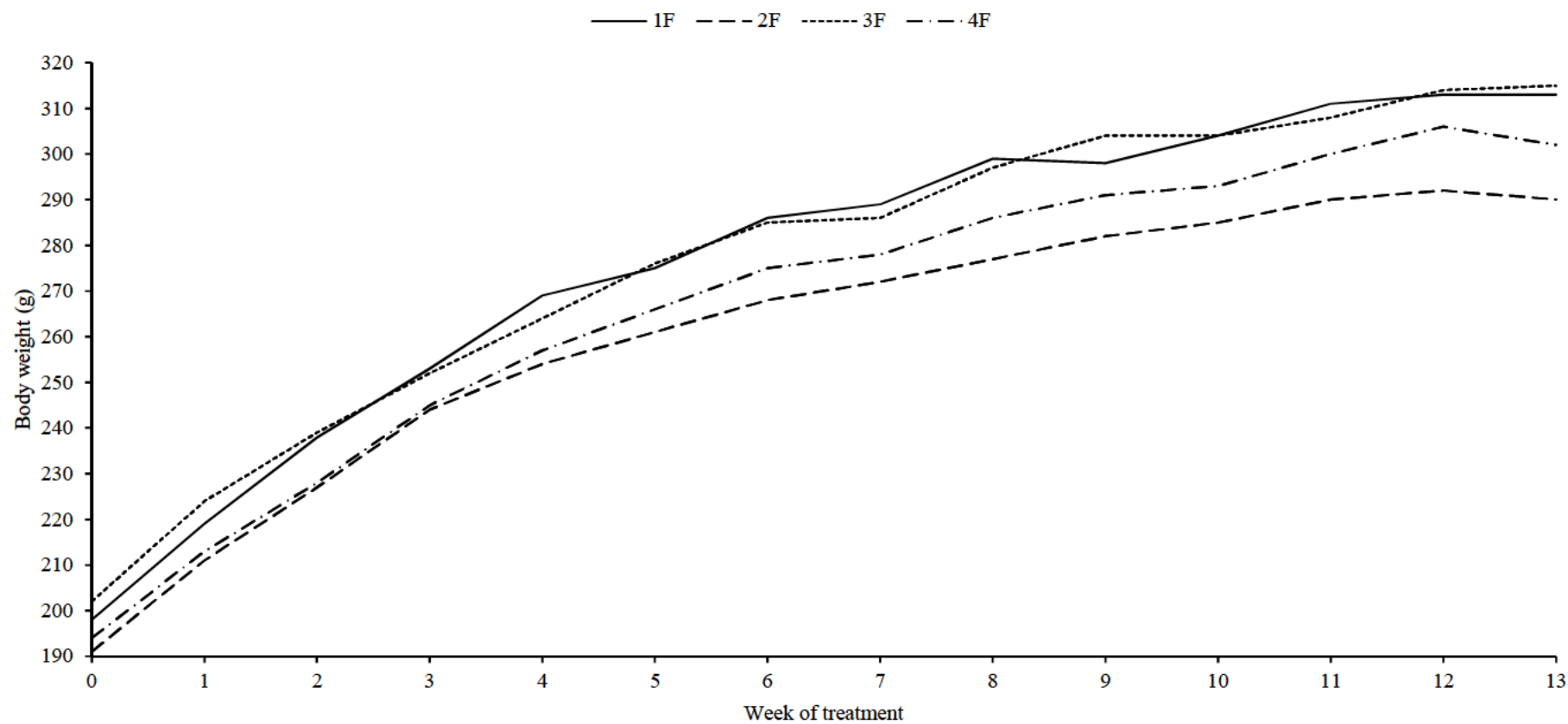




TABLE 1 Detailed physical examination and arena observations - group distribution of observations

Request ID: 5032111

	Control	Lactase, batch PPL34537									
Dose Group	1	2	3	4							
Dose (%)	0	10	33	100							
Category	Observation	Week	Group/Sex: Initial no:	Number of animals affected							
				1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Behavior	Irritable	9		0	0	0	0	0	0	1	0
		12		0	0	0	0	0	0	0	1
		13		0	0	0	0	0	0	0	1
		14		0	0	0	0	0	0	0	1
	Vocalization	2		0	0	0	0	1	2	1	3
		3		0	0	0	0	2	2	1	3
		4		0	0	0	0	2	2	1	3
		5		0	0	0	0	2	3	1	3
		6		0	0	0	0	2	5	3	3
		7		0	0	0	0	2	5	3	3
		8		0	0	0	0	2	4	2	2
		9		0	0	0	0	2	3	1	0
		10		0	0	0	0	2	2	1	0
		11		0	0	0	0	2	1	1	0
		12		0	0	0	0	2	1	1	1
		13		0	0	0	0	0	1	2	1
		14		0	0	0	0	0	1	1	1

TABLE 1 (cont) Detailed physical examination and arena observations - group distribution of observations

Request ID: 5032111

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Category	Observation	Week	Group/Sex: Initial no:	Number of animals affected							
				1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Build (Deformity)	Partially absent appendage, Ear - left	7		0	0	0	0	0	0	0	1
		8		0	0	0	0	0	0	0	1
		9		0	0	0	0	0	0	0	1
		10		0	0	0	0	0	0	0	1
		11		0	0	0	0	0	0	0	1
		12		0	0	0	0	0	0	0	1
		13		0	0	0	0	0	0	0	1
		14		0	0	0	0	0	0	0	1
	Swollen area, Ventral surface	6		0	0	0	0	0	0	1	0
		7		0	0	0	0	0	0	1	0
		8		0	0	0	0	0	0	1	0
		9		0	0	0	0	0	0	2	0
		10		0	0	0	0	0	0	2	0
		11		0	0	0	0	0	0	2	0
		12		0	0	0	0	0	0	2	0
		13		0	0	0	0	0	0	2	0
		14		0	0	0	0	0	0	2	0

TABLE 1 (cont) Detailed physical examination and arena observations - group distribution of observations

Request ID: 5032111

	Control	Lactase, batch PPL34537									
Dose Group	1	2	3	4							
Dose (%)	0	10	33	100							
Category	Observation	Week	Group/Sex: Initial no:	Number of animals affected							
				1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Coat	Hair loss, Dorsal surface	P1		1	0	0	0	1	0	1	0
		1		1	1	0	1	1	2	3	0
		2		2	1	0	1	1	0	1	0
		3		2	2	0	1	1	0	1	2
		4		0	1	0	1	0	0	0	0
		5		0	1	0	0	0	0	0	0
	Hair loss, Forelimbs	P1		0	0	0	0	1	0	0	0
		1		0	0	0	0	1	0	0	0
		2		0	0	0	0	1	0	0	0
		3		0	0	0	0	1	0	0	0
		7		0	0	0	0	0	1	0	2
		8		0	0	0	0	0	1	0	2
		9		0	0	0	0	0	1	0	2
		10		0	0	0	0	2	1	0	2
		11		0	0	0	0	3	1	1	2
		12		0	0	0	0	3	1	1	2
		13		0	0	0	0	3	1	1	2
		14		0	0	0	1	3	1	1	2

TABLE 1 (cont) Detailed physical examination and arena observations - group distribution of observations

Request ID: 5032111

	Control	Lactase, batch PPL34537									
Dose Group	1	2	3	4							
Dose (%)	0	10	33	100							
Category	Observation	Week	Group/Sex: Initial no:	Number of animals affected							
				1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Coat	Hair loss, Head	5		0	0	0	1	0	0	0	0
		6		0	0	0	1	0	2	0	0
		7		1	0	0	1	0	5	1	0
		8		1	0	0	1	1	5	1	0
		9		1	0	1	2	4	5	1	0
		10		1	0	2	2	7	7	3	1
		11		1	1	2	2	7	7	3	1
		12		1	2	2	2	7	7	3	2
		13		1	2	2	2	7	7	3	2
		14		1	2	2	3	7	7	3	2
Eyes	Prominent, Bilateral	9		0	0	0	0	1	0	0	0
		10		0	0	0	0	1	0	0	0
		11		0	0	0	0	1	0	0	0
		12		0	0	0	0	1	0	0	0
		13		0	0	0	0	1	0	0	0
		14		0	0	0	0	1	0	0	0
Skin	Encrustation, Dorsal surface	P1		1	0	0	1	1	0	1	0
		1		1	1	0	0	1	0	1	0
		2		1	1	0	0	1	0	0	0
		3		0	1	0	0	0	0	0	0

TABLE 1 (cont) Detailed physical examination and arena observations - group distribution of observations

Request ID: 5032111

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Category	Observation	Week	Group/Sex: Initial no:	Number of animals affected							
				1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Skin	Encrustation, Head	6		0	0	0	0	0	1	0	0
		7		0	0	0	1	0	2	1	0
		8		1	0	0	1	0	3	1	0
		9		1	0	0	1	3	3	1	0
		10		1	0	0	2	5	4	1	0
		11		1	1	0	2	5	5	1	0
		12		1	2	0	2	5	6	1	0
		13		1	2	0	2	5	6	1	0
		14		1	2	0	2	5	6	1	0
	Encrustation, Lower dorsal surface	3		0	1	0	0	0	0	0	0
		4		0	1	0	0	0	0	0	0
	Encrustation, Upper dorsal surface	6		0	0	0	0	1	0	0	0
Skin abrasion	Wet, Lower dorsal surface	P1		1	0	0	0	0	0	0	0

TABLE 1 (cont) Detailed physical examination and arena observations - group distribution of observations

Request ID: 5032111

	Control	Lactase, batch PPL34537									
Dose Group	1	2	3	4							
Dose (%)	0	10	33	100							
Category	Observation	Week	Group/Sex: Initial no:	Number of animals affected							
				1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Staining	Abnormal color, Brown, Head	5		0	0	0	0	0	1	1	0
		6		0	0	0	0	0	1	1	0
		7		0	0	0	0	0	1	1	0
		8		0	0	0	0	0	1	1	0
		9		0	0	0	0	0	0	0	1
		10		0	0	0	0	0	1	1	1
		11		0	0	0	0	0	1	1	2
		12		0	0	0	0	0	1	1	2
		13		0	0	0	0	0	1	1	2
		14		0	0	0	0	0	1	1	2
	Abnormal color, Brown, Muzzle	P1		0	0	0	0	1	0	0	0
	Abnormal color, Brown, Upper dorsal surface	11		0	0	0	0	0	0	1	0
		12		0	0	0	0	0	0	1	0
		13		0	0	0	0	0	0	1	0
		14		0	0	0	0	0	0	1	0

TABLE 2 Sensory reactivity observations and grip strength - summary of findings during Week 12 of treatment

	Control	Lactase, batch PPL34537										
Dose Group	1	2	3	4								
Dose (%)	0	10	33	100								
Group/sex:			Statistics	1M	2M	3M	4M	Statistics	1F	2F	3F	4F
Number of animals:			test (M)	10	10	10	10	test (F)	10	10	10	10
<u>Parameter</u>		<u>Grade</u>										
Approach response (1-3)		2		10	10	10	10		10	10	10	10
Pinna reflex (1-3)		2		10	10	10	10		10	10	10	10
Auditory startle reflex (1-4)		2		0	0	0	0		0	0	1	1
		3		10	10	10	10		10	10	9	9
Tail pinch response (1-4)		2		0	0	0	0		0	0	0	0
		3		10	10	10	9		10	9	10	9
		4		0	0	0	1		0	1	0	1
Forelimb grip strength (kg)		Mean	Wi	1.06	1.16	1.05	1.11	Wi	0.97	0.85	0.93	0.91
		SD		0.16	0.13	0.11	0.15		0.17	0.15	0.10	0.09
Hindlimb grip strength (kg)		Mean	Wi	0.61	0.58	0.62	0.55	Sh	0.51	0.47	0.46*	0.43**
		SD		0.08	0.06	0.08	0.06		0.06	0.06	0.01	0.03
<u>Additional observations^</u>												
Excessive vocalisation during grip strength procedure - moderate				0	0	0	0		0	1	1	1

^ Each animal may have more than one observation

TABLE 3 Motor activity - group mean scores (beam breaks) during Week 12 of treatment

Dose Group	Control	Lactase, batch PPL34537											
	1	2	3	4									
Dose (%)	0	10	33	100									
Group /Sex	Number of animals	Beam level	6	12	18	24	Time (minutes)		42	48	54	60	Total
Statistics test			Wi	Wi	Wi	Wi	Sh	Wi	Wi	Wi	Sh	Wi	Wi
1M	10	High	108.6	62.5	30.7	28.4	12.5	3.1	12.3	24.1	2.6	6.7	291.5
		SD	20.0	13.7	20.8	18.2	9.6	9.5	16.8	30.0	6.5	13.3	63.4
2M	10	High	121.2	76.4	49.8	43.0	21.3	17.2	6.2	11.5	3.2	7.1	356.9
		SD	30.3	29.0	26.9	33.7	31.8	14.8	7.8	23.4	8.0	20.1	162.0
3M	10	High	122.6	86.1	48.1	37.8	15.0	10.4	11.3	14.6	15.0	8.7	369.6
		SD	9.6	24.0	17.0	26.1	19.7	19.9	19.0	28.5	23.8	14.1	132.1
4M	10	High	110.2	70.0	31.7	15.9	3.7*	5.1	6.9	25.9	9.2	14.9	293.5
		SD	29.9	24.9	32.5	17.5	8.2	13.6	15.8	30.6	11.0	20.1	97.7
Statistics test			Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi
1M	10	Low	163.8	105.9	67.4	60.7	40.6	13.2	32.6	39.3	16.2	21.5	561.2
		SD	17.8	37.3	37.3	42.3	28.7	17.6	39.1	35.5	22.5	21.8	150.8
2M	10	Low	195.1	126.0	84.9	55.8	53.0	42.8	22.9	17.8	17.0	13.1	628.4
		SD	36.0	36.8	33.7	32.8	49.7	33.3	18.1	24.4	21.5	15.6	170.4
3M	10	Low	198.5	118.4	83.8	61.5	37.0	31.6	23.6	13.3	23.5	21.0	612.2
		SD	41.8	35.0	33.3	24.1	28.8	43.1	33.5	14.8	29.2	29.6	82.8
4M	10	Low	190.4	128.1	63.5	44.0	20.1	18.1	18.5	44.4	24.0	25.0	576.1
		SD	52.1	35.8	33.7	31.3	25.0	26.7	24.3	39.2	25.8	28.4	180.4



TABLE 3 (cont) Motor activity - group mean scores (beam breaks) during Week 12 of treatment

	Control	Lactase, batch PPL34537												
Dose Group	1	2	3	4										
Dose (%)	0	10	33	100										
Group /Sex	Number of animals	Beam level	6	12	18	24	Time (minutes)							
							30	36	42	48	54	60	Total	
Statistics test			Wi	Wi	Wi	Wi	Wi	Sh	Wi	Wi	Wi	Wi	Wi	
1F	10	High	175.0	96.9	58.2	27.0	17.2	8.8	24.7	22.2	37.0	24.3	491.3	
		SD	41.9	51.8	30.2	34.9	28.4	14.4	31.6	24.6	38.0	30.8	215.0	
2F	10	High	145.6	77.2	44.1	16.7	21.4	8.4	22.0	17.9	24.0	17.2	394.5	
		SD	25.9	20.9	33.5	19.6	27.4	14.1	30.9	27.4	34.4	20.4	94.9	
3F	10	High	170.1	96.6	50.0	29.7	9.9	4.7	8.8	24.4	25.7	31.1	451.0	
		SD	27.4	32.2	24.8	21.4	15.6	8.2	13.6	25.0	32.3	38.6	128.6	
4F	10	High	166.2	76.5	69.0	24.7	37.1	20.5	10.6	10.3	15.0	36.0	465.9	
		SD	50.8	42.5	36.1	27.6	33.4	34.2	16.8	20.9	22.9	32.8	208.4	
Statistics test			Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	
1F	10	Low	223.6	140.1	105.8	62.1	44.4	23.1	44.7	45.1	45.3	40.3	774.5	
		SD	51.3	54.2	34.6	60.4	46.1	34.3	42.9	35.0	32.5	43.1	233.0	
2F	10	Low	195.9	105.2	71.3	44.3	46.7	23.1	31.3	26.1	29.8	35.8	609.5	
		SD	28.0	37.0	43.0	54.8	35.6	24.9	32.5	29.5	37.7	40.7	155.1	
3F	10	Low	210.5	125.5	79.9	76.6	24.7	28.1	29.5	37.7	46.6	41.9	701.0	
		SD	38.9	35.6	30.0	36.2	23.9	47.4	33.8	33.8	41.4	40.1	115.7	
4F	10	Low	214.6	130.9	101.1	61.8	59.8	29.4	30.4	28.7	25.7	53.4	735.8	
		SD	33.1	41.6	32.5	46.4	42.5	24.0	32.6	32.6	22.6	50.9	166.3	

TABLE 4 Body weight - group mean values (g)

Request ID: 5032123

Dose Group Dose (%)	Control	Lactase, batch PPL34537															Change 0-13
	1 0	2 10	3 33	4 100													
Group /Sex	Week P1	0	1	2	3	4	5	6	7	8	9	10	11	12	13		
Statistics test																Wi	
1M	Mean	207	269	325	363	405	437	461	484	502	522	538	551	569	575	583	314
	SD	13.8	17.4	26.3	29.8	39.9	45.6	51.2	55.5	56.5	61.6	65.1	66.7	69.9	71.0	71.9	55.9
	N	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
2M	Mean	203	263	316	356	389	421	442	466	479	497	512	525	539	548	553	290
	SD	11.5	12.6	17.1	20.8	24.6	27.5	29.9	31.4	33.2	36.9	40.1	42.2	43.5	43.7	43.2	33.9
	N	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
% of 1M																92	
3M	Mean	199	260	313	347	388	415	435	456	469	487	503	515	526	535	540	280
	SD	12.3	15.2	21.3	31.5	37.1	42.8	49.0	54.8	59.3	62.5	66.6	66.1	67.6	66.7	69.0	60.1
	N	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
% of 1M																89	
4M	Mean	201	261	315	355	393	425	448	470	482	504	518	529	543	553	556	295
	SD	9.7	14.3	19.2	22.6	27.4	28.6	31.1	31.5	34.9	38.2	41.0	42.9	45.9	46.9	46.1	37.4
	N	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
% of 1M																94	

TABLE 4 (cont) Body weight - group mean values (g)

Request ID: 5032123

Dose Group Dose (%)	Control	Lactase, batch PPL34537															Change 0-13
	1 0	2 10	3 33	4 100													
Group /Sex	Week P1	0	1	2	3	4	5	6	7	8	9	10	11	12	13		
Statistics test																Wi	
1F	Mean	170	198	219	238	253	269	275	286	289	299	298	304	311	313	313	114
	SD	11.0	13.6	13.5	17.2	20.3	22.3	23.2	24.2	26.5	26.5	25.2	26.7	28.0	28.2	27.8	17.0
	N	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
2F	Mean	166	191	211	227	244	254	261	268	272	277	282	285	290	292	290	99
	SD	5.7	6.6	9.2	8.7	9.2	9.3	10.0	9.0	10.1	9.5	10.1	10.3	13.1	12.8	10.4	9.8
	N	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
% of 1F																87	
3F	Mean	174	202	224	239	252	264	276	285	286	297	304	304	308	314	315	113
	SD	8.3	8.2	8.1	11.2	11.4	11.1	12.8	13.9	16.5	13.8	13.8	17.3	18.6	17.5	17.8	14.7
	N	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
% of 1F																99	
4F	Mean	170	194	213	228	245	257	266	275	278	286	291	293	300	306	302	108
	SD	5.2	5.9	5.8	8.6	11.1	11.8	13.9	17.1	16.0	17.7	18.3	20.5	20.2	19.7	21.3	20.9
	N	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10	10
% of 1F																95	

TABLE 5 Food consumption - group mean values (g/animal/week)

Request ID: 5032113

Dose Group Dose (%)	Control	Lactase, batch PPL34537														
	1 0	2 10	3 33	4 100												
Group /Sex	Week P1	1	2	3	4	5	6	7	8	9	10	11	12	13	Mean 1-13	
1M	Mean	196	205	204	205	208	205	206	210	203	203	209	203	203	191	204
	SD	12.2	6.9	13.3	13.6	9.8	10.7	8.9	8.3	11.5	15.4	13.2	14.4	11.2	10.6	11.4
	N	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
2M	Mean	190	198	198	192	198	199	193	200	187	193	193	196	190	183	194
	SD	1.7	4.0	4.8	4.0	8.8	4.7	2.0	0.7	1.7	6.6	6.5	0.3	5.8	3.0	3.9
	N	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
% of 1M																95
3M	Mean	188	191	190	192	195	191	194	195	191	192	196	191	192	176	191
	SD	3.0	6.4	16.1	14.6	6.1	6.8	6.4	8.5	6.6	9.1	4.5	4.4	1.7	3.4	7.3
	N	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
% of 1M																94
4M	Mean	187	197	202	200	198	200	197	200	193	199	196	196	189	180	196
	SD	6.5	13.0	10.0	10.6	7.9	9.1	7.1	11.2	8.6	12.0	8.3	11.3	10.7	6.5	9.7
	N	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
% of 1M																96

TABLE 5 (cont) Food consumption - group mean values (g/animal/week)

Request ID: 5032113

Dose Group Dose (%)	Control	Lactase, batch PPL34537														
	1	2	3	4												
	0	10	33	100												
Group /Sex	Week P1	1	2	3	4	5	6	7	8	9	10	11	12	13	Mean 1-13	
1F	Mean	129	134	147	142	147	145	149	145	140	138	143	140	136	134	141
	SD	0.8	0.8	1.3	3.7	4.4	0.6	0.4	4.4	2.1	3.4	1.3	4.0	3.8	0.4	2.0
	N	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
2F	Mean	120	128	133	132	133	139	131	133	130	133	130	128	125	123	131
	SD	4.0	3.7	1.3	0.3	1.4	2.0	8.2	3.8	3.5	4.8	8.3	2.4	2.7	3.4	3.0
	N	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
% of 1F																92
3F	Mean	129	136	144	143	142	142	142	142	141	137	135	138	132	126	138
	SD	4.9	1.3	2.3	8.8	0.7	4.9	3.0	5.4	1.7	2.4	3.0	4.2	2.3	1.4	2.6
	N	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
% of 1F																98
4F	Mean	126	129	136	136	139	138	135	133	128	132	130	133	126	121	132
	SD	1.8	0.1	2.4	1.6	0.3	2.0	1.7	0.7	2.7	0.3	1.8	3.8	5.7	4.1	0.5
	N	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
% of 1F																93

TABLE 6 Haematology - group mean values during Week 13 of treatment

Dose Group Dose (%)	Control	Lactase, batch PPL34537							
	1	2	3	4					
	0	10	33	100					
Group /Sex		Hct L/L	Hb g/dL	RBC $\times 10^{12}/L$	Retic $\times 10^{12}/L$	MCH pg	MCHC g/dL	MCV fL	RDW %
Statistics test		Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi
1M	Mean	0.458	15.0	8.72	0.222	17.3	32.8	52.5	14.2
	SD	0.0107	0.47	0.203	0.0281	0.52	0.47	1.19	0.93
	N	10	10	10	10	10	10	10	10
2M	Mean	0.459	15.3	8.73	0.202	17.5	33.4**	52.5	13.9
	SD	0.0209	0.80	0.313	0.0459	0.71	0.45	1.64	1.42
	N	10	10	10	10	10	10	10	10
3M	Mean	0.455	15.2	8.49	0.199	17.9*	33.3**	53.7	13.5
	SD	0.0149	0.58	0.228	0.0369	0.38	0.33	0.86	0.80
	N	10	10	10	10	10	10	10	10
4M	Mean	0.457	15.3	8.62	0.180*	17.7*	33.4**	53.0	13.1*
	SD	0.0164	0.56	0.407	0.0257	0.43	0.27	1.18	0.75
	N	10	10	10	10	10	10	10	10

TABLE 6 (cont) Haematology - group mean values during Week 13 of treatment

Dose Group Dose (%)	Control	Lactase, batch PPL34537									
	1	2	3	4							
	0	10	33	100							
Group /Sex		WBC x10 <sup>9</sup> /L	N x10 <sup>9</sup> /L	L x10 <sup>9</sup> /L	E x10 <sup>9</sup> /L	B x10 <sup>9</sup> /L	M x10 <sup>9</sup> /L	LUC x10 <sup>9</sup> /L	Plt x10 <sup>9</sup> /L	PT sec	APTT sec
Statistics test		Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi
1M	Mean	14.26	2.34	11.08	0.23	0.09	0.40	0.12	1105	21.6	16.5
	SD	2.651	0.680	2.386	0.114	0.036	0.130	0.046	258.8	1.40	0.71
	N	10	10	10	10	10	10	10	10	9	9
2M	Mean	12.25	1.62	9.92	0.17	0.10	0.35	0.09	966	22.0	16.8
	SD	2.134	0.729	1.505	0.065	0.035	0.092	0.036	113.6	0.98	0.91
	N	10	10	10	10	10	10	10	10	10	10
3M	Mean	15.56	2.43	12.19	0.22	0.12	0.49	0.11	1081	21.9	16.4
	SD	4.103	1.251	3.335	0.067	0.040	0.203	0.046	170.7	0.94	1.07
	N	10	10	10	10	10	10	10	9	10	10
4M	Mean	12.37	2.36	9.21	0.20	0.09	0.40	0.11	961	22.3	16.3
	SD	1.766	0.820	1.346	0.111	0.033	0.136	0.072	93.4	0.79	1.13
	N	10	10	10	10	10	10	10	9	10	10

TABLE 6 (cont) Haematology - group mean values during Week 13 of treatment

Dose Group Dose (%)	Control	Lactase, batch PPL34537							
	1 0	2 10	3 33	4 100					
Group /Sex		Hct L/L	Hb g/dL	RBC $\times 10^{12}/L$	Retic $\times 10^{12}/L$	MCH pg	MCHC g/dL	MCV fL	RDW %
Statistics test		Wi	Wi	Wi	Wi	Wi	Wi	Wi	Wi
1F	Mean	0.439	14.9	7.94	0.161	18.7	33.9	55.3	11.7
	SD	0.0196	0.58	0.452	0.0309	0.64	0.28	1.82	0.54
	N	10	10	10	10	10	10	10	10
2F	Mean	0.450	15.3	8.26	0.165	18.6	34.0	54.6	11.6
	SD	0.0144	0.40	0.315	0.0420	0.52	0.45	1.25	0.60
	N	10	10	10	10	10	10	10	10
3F	Mean	0.441	14.9	7.96	0.178	18.7	33.8	55.4	11.6
	SD	0.0124	0.30	0.206	0.0263	0.49	0.37	1.60	0.48
	N	10	10	10	10	10	10	10	10
4F	Mean	0.441	14.9	8.03	0.177	18.5	33.7	54.9	11.8
	SD	0.0127	0.56	0.211	0.0242	0.44	0.46	0.91	0.45
	N	10	10	10	10	10	10	10	10



TABLE 6 (cont) Haematology - group mean values during Week 13 of treatment

Dose Group	Control	Lactase, batch PPL34537									
	1	2	3	4							
Dose (%)	0	10	33	100							
Group /Sex		WBC x10 <sup>9</sup> /L	N x10 <sup>9</sup> /L	L x10 <sup>9</sup> /L	E x10 <sup>9</sup> /L	B x10 <sup>9</sup> /L	M x10 <sup>9</sup> /L	LUC x10 <sup>9</sup> /L	Plt x10 <sup>9</sup> /L	PT sec	APTT sec
Statistics test		Wi	Wi	Du	Wi	Wi	Wi	Du	Wi	Wi	Wi
1F	Mean	7.80	1.02	6.34	0.13	0.03	0.21	0.07	935	23.6	15.3
	SD	2.234	0.611	2.059	0.062	0.018	0.092	0.033	163.4	1.61	1.84
	N	10	10	10	10	10	10	10	10	9	10
2F	Mean	9.14	0.76	7.88	0.12	0.06	0.22	0.12*	1069	23.4	16.2
	SD	1.827	0.315	1.646	0.058	0.022	0.076	0.043	78.8	0.88	0.88
	N	10	10	10	10	10	10	10	10	10	10
3F	Mean	6.98	0.82	5.71	0.11	0.04	0.24	0.06	941	23.0	14.5
	SD	1.721	0.633	1.327	0.044	0.016	0.064	0.036	105.8	1.00	1.67
	N	10	10	10	10	10	10	10	10	10	10
4F	Mean	8.11	1.20	6.45	0.15	0.05	0.20	0.07	1006	24.1	13.9
	SD	2.687	1.023	1.898	0.067	0.032	0.092	0.028	137.6	0.72	1.43
	N	10	10	10	10	10	10	10	9	10	10

TABLE 7 Blood chemistry - group mean values during Week 13 of treatment

		Lactase, batch PPL34537							
Dose Group	Control	1	2	3	4				
Dose (%)		0	10	33	100				
Group /Sex		ALP U/L	ALT U/L	AST U/L	Urea mmol/L	Creat μmol/L	Gluc mmol/L	Chol mmol/L	
Statistics test		Wi	Sh	Wi	Wi	Wi	Wi	Wi	
1M	Mean	74	38	81	5.32	36	8.50	1.56	
	SD	12.8	6.0	14.5	0.716	6.1	1.180	0.409	
	N	10	10	10	10	10	10	10	
2M	Mean	82	43	85	5.20	33	7.72	1.60	
	SD	16.4	16.8	34.0	0.685	4.0	1.361	0.157	
	N	10	10	10	10	10	10	10	
3M	Mean	93*	42	89	4.91	32	7.89	1.85	
	SD	21.6	11.9	17.7	0.595	4.1	1.409	0.376	
	N	10	10	10	10	10	10	10	
4M	Mean	92*	39	83	4.68*	30**	7.98	1.56	
	SD	18.7	4.8	13.8	0.666	4.1	1.568	0.298	
	N	10	10	10	10	10	10	10	

TABLE 7 (cont) Blood chemistry - group mean values during Week 13 of treatment

Dose Group Dose (%)	Control	Lactase, batch PPL34537				
	1	2	3	4		
	0	10	33	100		
Group /Sex		Na mmol/L	K mmol/L	Total Prot g/L	Alb g/L	A/G Ratio
Statistics test		Wi	lWi	Wi	Wi	Wi
1M	Mean	140	5.8	64	35	1.21
	SD	1.1	0.49	2.8	2.0	0.118
	N	10	10	10	10	10
2M	Mean	140	6.2	62	35	1.27
	SD	1.8	0.59	3.1	1.4	0.123
	N	10	10	10	10	10
3M	Mean	139	5.9	64	35	1.20
	SD	1.0	0.45	1.8	1.2	0.111
	N	9	9	10	10	10
4M	Mean	139*	6.7*	65	35	1.14
	SD	1.3	1.33	2.5	1.2	0.084
	N	10	10	10	10	10

TABLE 7 (cont) Blood chemistry - group mean values during Week 13 of treatment

Dose Group Dose (%)	Control	Lactase, batch PPL34537						
	1 0	2 10	3 33	4 100				
Group /Sex		ALP U/L	ALT U/L	AST U/L	Urea mmol/L	Creat μmol/L	Gluc mmol/L	Chol mmol/L
Statistics test		Wi	IWi	Sh	Wi	Wi	Wi	Wi
1F	Mean	35	37	93	5.80	39	7.08	2.18
	SD	9.0	15.5	72.5	0.775	4.0	1.301	0.496
	N	10	10	10	10	10	10	10
2F	Mean	40	30	66	5.63	37	6.71	2.28
	SD	8.4	4.2	11.2	0.628	1.8	0.807	0.358
	N	10	10	10	10	10	10	10
3F	Mean	40	29	69	5.71	41	7.06	2.21
	SD	11.4	5.3	10.9	0.769	2.7	0.577	0.448
	N	10	10	10	10	10	10	10
4F	Mean	52**	30	73	5.92	38	6.69	2.04
	SD	9.6	6.3	14.0	0.711	4.5	0.631	0.294
	N	10	10	10	10	10	10	10

TABLE 7 (cont) Blood chemistry - group mean values during Week 13 of treatment

Dose Group Dose (%)	Control	Lactase, batch PPL34537				
	1 0	2 10	3 33	4 100		

Group /Sex		Na mmol/L	K mmol/L	Total Prot g/L	Alb g/L	A/G Ratio
Statistics test		Wi	Wi	Wi	Wi	Wi
1F	Mean	141	4.0	70	40	1.36
	SD	1.3	0.38	3.6	2.5	0.144
	N	10	10	10	10	10
2F	Mean	140	4.4	69	41	1.44
	SD	1.3	0.25	4.2	2.3	0.183
	N	10	10	10	10	10
3F	Mean	140	4.4	69	41	1.45
	SD	1.2	0.66	3.2	2.3	0.114
	N	10	10	10	10	10
4F	Mean	140	4.3	69	40	1.45
	SD	1.4	0.31	3.0	3.3	0.217
	N	10	10	10	10	10

TABLE 8 Organ weights - group mean unadjusted and adjusted values (g) for animals killed after 13 weeks of treatment

Request ID: 5032114

	Control		Lactase, batch PPL34537								
Dose Group	1		2	3	4						
Dose (%)	0		10	33	100						
Group/ Sex	Terminal Body weight		Adrenals	Brain	Epididymides	Heart	Kidneys	Liver	Spleen	Testes	Thymus
Statistics test		Wi							Sh		
1M	Mean	583	0.057	2.285	1.389	1.651	3.445	20.362	0.922	3.850	0.252
	SD	73	0.012	0.118	0.197	0.209	0.391	4.400	0.080	0.451	0.025
	N	10	10	10	10	10	10	10	10	10	10
2M	Mean	553	0.057	2.227	1.384	1.517	3.343	17.831	0.847	3.802	0.235
	SD	43	0.010	0.076	0.080	0.147	0.289	2.104	0.088	0.296	0.060
	N	10	10	10	10	10	10	10	10	10	10
3M	Mean	542	0.059	2.159	1.397	1.560	3.356	18.357	0.956	3.840	0.266
	SD	68	0.010	0.053	0.111	0.201	0.374	2.298	0.249	0.226	0.059
	N	10	10	10	10	10	10	10	10	10	10
4M	Mean	556	0.063	2.206	1.403	1.511	3.400	18.810	0.899	3.711	0.223
	SD	46	0.008	0.072	0.112	0.120	0.305	2.503	0.201	0.383	0.080
	N	10	10	10	10	10	10	10	10	10	10
Statistics test			Wi	Wi	Wi	Wi	Wi	Wi		Wi	Wi
1M	Adjusted	Mean	0.055	2.273	1.365	1.591	3.322	19.422		3.838	0.246
2M	Adjusted	Mean	0.057	2.230	1.389	1.530	3.369	18.034		3.804	0.237
3M	Adjusted	Mean	0.060	2.167*	1.412	1.600	3.438	18.989		3.848	0.270
4M	Adjusted	Mean	0.063	2.208*	1.405	1.518	3.414	18.915		3.712	0.224

TABLE 8 (cont) Organ weights - group mean unadjusted and adjusted values (g) for animals killed after 13 weeks of treatment

Request ID: 5032114

		Control	Lactase, batch PPL34537								
Dose Group		1	2	3	4						
Dose (%)		0	10	33	100						
Group/ Sex		Terminal Body weight	Adrenals	Brain	Heart	Kidneys	Liver	Ovaries	Spleen	Thymus	Uterus and Cervix
Statistics test		Wi			Sh						
1F	Mean	312	0.063	2.001	1.002	2.053	10.794	0.090	0.572	0.194	0.804
	SD	29	0.007	0.102	0.135	0.226	1.289	0.024	0.060	0.049	0.322
	N	10	10	10	10	10	10	10	10	10	10
2F	Mean	291	0.062	1.990	0.951	1.807	9.812	0.084	0.600	0.230	0.778
	SD	10	0.008	0.073	0.037	0.325	0.760	0.018	0.074	0.062	0.280
	N	10	10	10	10	10	10	10	10	10	10
3F	Mean	315	0.064	2.023	1.012	1.892	10.023	0.087	0.610	0.235	0.881
	SD	17	0.005	0.079	0.059	0.161	0.985	0.010	0.106	0.041	0.346
	N	10	10	10	10	10	10	10	10	10	10
4F	Mean	305	0.061	2.037	0.976	2.037	10.735	0.084	0.594	0.229	0.653
	SD	20	0.008	0.079	0.081	0.165	1.289	0.013	0.080	0.058	0.209
	N	10	10	10	10	10	10	10	10	10	10
Statistics test			Wi	Wi		Wi	Wi	Wi	Wi	Wi	Wi
1F	Adjusted Mean		0.062	1.990		2.017	10.611	0.089	0.565	0.190	0.795
2F	Adjusted Mean		0.064	2.017		1.898	10.269	0.087	0.617	0.239	0.802
3F	Adjusted Mean		0.062	2.007		1.835	9.737	0.086	0.598	0.229	0.867
4F	Adjusted Mean		0.061	2.038		2.039	10.746	0.084	0.595	0.229	0.654

TABLE 9 Macropathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035205

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Tissue/Organ and Findings	Group/Sex No. of animals	Number of animals affected							
		1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Number of animals within normal limits		7	8	8	7	5	6	8	8
Kidneys									
Depression(s)		0	0	0	0	0	1	2	1
Dilated Pelvis		0	0	0	1	0	0	0	0
Liver									
Small		0	0	0	0	0	1	0	0
Strangulated		1	0	0	0	0	0	0	0
Lungs and Bronchi									
Dark area(s)		0	1	1	0	0	0	0	0
Pale area(s)		0	1	1	2	1	0	1	0
Lymph Node, Mandibular									
Enlarged		1	0	0	1	0	0	0	0
Ovaries									
Abnormal color		-	-	-	-	1	0	0	0
Skin and Subcutis									
Hair Loss		0	0	0	0	4	3	0	1



TABLE 9 (cont) Macropathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035205

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Tissue/Organ and Findings	Group/Sex No. of animals	Number of animals affected							
		1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Skin and Subcutis									
Scab(s)		0	0	0	0	4	3	0	0
Stomach									
Depression(s)		0	0	0	0	1	0	0	0
Mass(es)		1	0	0	0	0	0	0	0
Thoracic Cavity									
Adhesion(s)		0	0	1	0	0	0	0	0

TABLE 10 Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Tissue/Organ and Findings	Group/Sex No. of animals	Number of animals affected							
		1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Adrenals	No. examined	10	0	0	10	10	0	0	10
Vacuolation, Cortical	Minimal	0	0	0	1	0	0	0	0
	Slight	2	0	0	1	0	0	0	0
	Total	2	0	0	2	0	0	0	0
Aorta	No. examined	10	0	0	10	10	0	0	10
Bone, Femur Including Joint	No. examined	10	0	0	10	10	0	0	10
Bone, Sternum	No. examined	10	0	0	10	10	0	0	10
Brain	No. examined	10	0	0	10	10	0	0	10
Cecum	No. examined	10	0	0	10	10	0	0	10
Colon	No. examined	10	0	0	10	10	0	0	10

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537							
Dose Group	1	2	3	4					
Dose (%)	0	10	33	100					
					Number of animals affected				
	Group/Sex	1M	2M	3M	4M	1F	2F	3F	4F
Tissue/Organ and Findings	No. of animals	10	10	10	10	10	10	10	10
Duodenum	No. examined	10	0	0	10	10	0	0	10
Epididymides	No. examined	10	0	0	10	-	-	-	-
Esophagus	No. examined	10	0	0	10	10	0	0	10
Heart	No. examined	10	0	0	10	10	0	0	10
Infiltration, Inflammatory Cells, Myocardial	Minimal	0	0	0	1	0	0	0	0
	Slight	1	0	0	0	0	0	0	1
	Total	1	0	0	1	0	0	0	1
Cardiomyopathy, Chronic	Minimal	0	0	0	0	1	0	0	0
	Total	0	0	0	0	1	0	0	0
Ileum	No. examined	10	0	0	10	10	0	0	10
Jejunum	No. examined	10	0	0	10	10	0	0	10

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Tissue/Organ and Findings	Group/Sex No. of animals	Number of animals affected							
		1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Kidneys	No. examined	10	0	0	10	10	1	2	10
Basophilia, Tubular	Minimal	4	0	0	2	0	0	0	2
	Slight	1	0	0	0	0	0	0	0
	Total	5	0	0	2	0	0	0	2
Infiltration, Inflammatory Cells, Interstitial	Minimal	0	0	0	2	0	0	0	2
	Slight	1	0	0	0	0	0	0	0
	Total	1	0	0	2	0	0	0	2
Cast(s), Intratubular	Minimal	0	0	0	0	1	0	0	0
	Total	0	0	0	0	1	0	0	0
Dilatation, Pelvic	Slight	0	0	0	1	0	0	0	1
	Total	0	0	0	1	0	0	0	1
Mineralization, Cortico-Medullary Junction	Minimal	0	0	0	0	0	1	1	2
	Total	0	0	0	0	0	1	1	2

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537											
Dose Group	1	2	3	4									
Dose (%)	0	10	33	100									
					Number of animals affected								
Group/Sex					1M	2M	3M	4M	1F	2F	3F	4F	
Tissue/Organ and Findings					No. of animals	10	10	10	10	10	10	10	10
Kidneys					No. examined	10	0	0	10	10	1	2	10
Scars, Cortical					Minimal	0	0	0	0	0	0	1	1
					Slight	0	0	0	1	0	0	0	0
					Total	0	0	0	1	0	0	1	1
Mineralization, Papilla					Minimal	1	0	0	0	0	0	0	0
					Total	1	0	0	0	0	0	0	0
Lymphocytic Infiltration, Peri-pelvic					Slight	0	0	0	1	0	0	0	0
					Total	0	0	0	1	0	0	0	0
Liver					No. examined	10	0	0	10	10	1	0	10
Aggregates, Lymphoid/Macrophage					Minimal	2	0	0	4	4	0	0	5
					Slight	2	0	0	0	0	1	0	1
					Total	4	0	0	4	4	1	0	6
Necrosis, Hepatocellular, Focal					Minimal	0	0	0	1	0	1	0	1
					Total	0	0	0	1	0	1	0	1

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Tissue/Organ and Findings	Group/Sex No. of animals	Number of animals affected							
		1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Liver	No. examined	10	0	0	10	10	1	0	10
Foci, Cellular Alteration, Clear Cell	Minimal	0	0	0	0	0	0	0	1
	Total	0	0	0	0	0	0	0	1
Glycogen, Decreased	Moderate	0	0	0	0	0	1	0	0
	Total	0	0	0	0	0	1	0	0
Necrosis, Torsion, Lobar	Total	1	0	0	0	0	0	0	0
Fibrosis	Slight	0	0	0	1	0	0	0	0
	Total	0	0	0	1	0	0	0	0
Lungs and Bronchi	No. examined	10	2	2	10	10	0	1	10
Alveolar Macrophages, Foamy	Minimal	0	0	0	1	1	0	1	0
	Slight	0	1	0	0	0	0	0	0
	Total	0	1	0	1	1	0	1	0

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537							
Dose Group	1	2	3	4					
Dose (%)	0	10	33	100					
					Number of animals affected				
	Group/Sex	1M	2M	3M	4M	1F	2F	3F	4F
Tissue/Organ and Findings	No. of animals	10	10	10	10	10	10	10	10
Lungs and Bronchi	No. examined	10	2	2	10	10	0	1	10
Alveolar Macrophages, Increased, Focal	Minimal	1	0	0	0	1	0	0	0
	Slight	0	0	1	0	0	0	0	0
	Total	1	0	1	0	1	0	0	0
Infiltration, Inflammatory Cells, Perivascular	Minimal	0	0	1	0	0	0	0	0
	Slight	2	0	0	0	0	0	0	1
	Total	2	0	1	0	0	0	0	1
Alveolar Macrophages, Pigment	Minimal	1	1	0	0	0	0	0	0
	Total	1	1	0	0	0	0	0	0
Mineralization, Alveoli	Minimal	1	0	1	2	0	0	0	0
	Total	1	0	1	2	0	0	0	0
Inflammation, Alveoli	Minimal	2	0	1	2	0	0	0	0
	Total	2	0	1	2	0	0	0	0

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537								
Dose Group	1	2	3	4						
Dose (%)	0	10	33	100						
					Number of animals affected					
	Group/Sex	1M	2M	3M	4M	1F	2F	3F	4F	
Tissue/Organ and Findings	No. of animals	10	10	10	10	10	10	10	10	
Lungs and Bronchi	No. examined	10	2	2	10	10	0	1	10	
Inflammation, Pleura	Slight	0	0	1	0	0	0	0	0	
	Total	0	0	1	0	0	0	0	0	
Lymph Node, Axillary Lt	No. examined	10	0	0	10	10	0	0	9	
Cellularity Increased, Paracortex	Slight	1	0	0	1	0	0	0	0	
	Total	1	0	0	1	0	0	0	0	
Lymph Node, Mandibular	No. examined	10	0	0	10	10	0	0	10	
Plasmacytosis	Slight	1	0	0	3	2	0	0	0	
	Moderate	2	0	0	2	3	0	0	1	
	Marked	0	0	0	1	0	0	0	0	
	Total	3	0	0	6	5	0	0	1	
Cellularity Increased, Paracortex	Slight	2	0	0	5	3	0	0	1	
	Moderate	1	0	0	0	0	0	0	0	
	Total	3	0	0	5	3	0	0	1	



TABLE 10 (cont)      Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537							
Dose Group	1	2	3	4					
Dose (%)	0	10	33	100					
		Number of animals affected							
	Group/Sex	1M	2M	3M	4M	1F	2F	3F	4F
Tissue/Organ and Findings	No. of animals	10	10	10	10	10	10	10	10
Lymph Node, Mandibular	No. examined	10	0	0	10	10	0	0	10
Erythrocytosis/Erythrophagocytosis, Sinuses	Minimal	2	0	0	0	0	0	0	0
	Total	2	0	0	0	0	0	0	0
Cellularity Increased, Follicles	Slight	3	0	0	4	0	0	0	0
	Moderate	2	0	0	0	0	0	0	0
	Total	5	0	0	4	0	0	0	0
Lymph Node, Mesenteric	No. examined	10	0	0	10	10	0	0	10
Mammary	No. examined	10	0	0	10	10	0	0	10
Nerve, Sciatic	No. examined	10	0	0	10	10	0	0	10
Ovaries	No. examined	-	-	-	-	10	0	0	10
Corpora Lutea, Absent	Total	-	-	-	-	2	0	0	0

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

Dose Group	Control	Lactase, batch PPL34537													
	1	2	3	4											
Dose (%)	0	10	33	100											
						Number of animals affected									
		Group/Sex			No. of animals	1M	2M	3M	4M	1F	2F	3F	4F		
Tissue/Organ and Findings						10	10	10	10	10	10	10	10		
Pancreas Fibrosis	No. examined					10	0	0	10	10	0	0	10		
	Slight					1	0	0	0	0	0	0	0		
	Total					1	0	0	0	0	0	0	0		
Foci, Inflammatory Cells	Minimal					1	0	0	0	0	0	0	0		
	Total					1	0	0	0	0	0	0	0		
Parathyroids	No. examined					9	0	0	10	10	0	0	9		
Peyer's Patches/GALT Cellularity, Increased	No. examined					10	0	0	10	10	0	0	10		
	Moderate					0	0	0	0	1	0	0	0		
	Total					0	0	0	0	1	0	0	0		
Pituitary Cyst(s)	No. examined					10	0	0	10	9	0	0	10		
	Minimal					0	0	0	0	0	0	0	1		
	Total					0	0	0	0	0	0	0	1		

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

Dose Group	Control	Lactase, batch PPL34537											
	1	2	3	4									
Dose (%)	0	10	33	100									

Tissue/Organ and Findings	Group/Sex No. of animals	Number of animals affected							
		1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Prostate	No. examined	10	0	0	10	-	-	-	-
Aggregates, Lymphoid	Minimal	1	0	0	0	-	-	-	-
	Slight	1	0	0	0	-	-	-	-
	Total	2	0	0	0	-	-	-	-
Rectum	No. examined	10	0	0	10	10	0	0	10
Parasites	Total	0	0	0	1	0	0	0	0
Salivary Gland, Mandibular	No. examined	10	0	0	10	10	0	0	10
Salivary Gland, Parotid	No. examined	10	0	0	10	10	0	0	10
Salivary Gland, Sublingual	No. examined	10	0	0	10	10	0	0	10
Seminal Vesicles	No. examined	10	0	0	10	-	-	-	-
Skin and Subcutis	No. examined	10	0	0	10	10	3	0	10

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537							
Dose Group	1	2	3	4					
Dose (%)	0	10	33	100					
					Number of animals affected				
	Group/Sex	1M	2M	3M	4M	1F	2F	3F	4F
Tissue/Organ and Findings	No. of animals	10	10	10	10	10	10	10	10
Skin and Subcutis Scab(s)	No. examined	10	0	0	10	10	3	0	10
	Slight	0	0	0	0	4	2	0	0
	Moderate	0	0	0	0	0	1	0	0
	Total	0	0	0	0	4	3	0	0
Ulceration, Epidermal	Slight	0	0	0	0	1	0	0	0
	Total	0	0	0	0	1	0	0	0
Hyperplasia, Epidermal	Minimal	0	0	0	0	3	2	0	0
	Slight	0	0	0	0	1	1	0	0
	Total	0	0	0	0	4	3	0	0
Spinal Cord Cervical	No. examined	10	0	0	10	10	0	0	10
Spinal Cord Lumbar	No. examined	10	0	0	10	10	0	0	10
Spinal Cord Thoracic	No. examined	10	0	0	10	10	0	0	10

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537								
Dose Group	1	2	3	4						
Dose (%)	0	10	33	100						
				Number of animals affected						
		Group/Sex	1M	2M	3M	4M	1F	2F	3F	4F
Tissue/Organ and Findings		No. of animals	10	10	10	10	10	10	10	10
Spleen		No. examined	10	0	0	10	10	0	0	10
Cellularity Increased, White Pulp		Slight	0	0	0	0	1	0	0	0
		Total	0	0	0	0	1	0	0	0
Hemosiderosis		Minimal	0	0	0	0	1	0	0	0
		Total	0	0	0	0	1	0	0	0
Extramedullary Hemopoiesis, Increased		Minimal	2	0	0	0	1	0	0	3
		Slight	0	0	0	0	0	0	0	1
		Total	2	0	0	0	1	0	0	4
Stomach		No. examined	10	0	0	10	10	0	0	10
Hyperplasia, Epithelial, Nonglandular Region		Slight	0	0	0	0	1	0	0	0
		Total	0	0	0	0	1	0	0	0
Infiltration, Inflammatory Cells, Mucosal/Submucosal, Nonglandular Region		Slight	0	0	0	1	0	0	0	0
		Total	0	0	0	1	0	0	0	0

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

Dose Group Dose (%)	Control	Lactase, batch PPL34537											
	1	2	3	4									
	0	10	33	100									
						Number of animals affected							
		Group/Sex No. of animals	1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10			
Stomach		No. examined	10	0	0	10	10	0	0	10			
Cyst(s), Squamous, Limiting Ridge		Moderate	1	0	0	0	0	0	0	0			
		Total	1	0	0	0	0	0	0	0			
Dilatation, Glands		Minimal	0	0	0	1	0	0	0	0			
		Total	0	0	0	1	0	0	0	0			
Testes		No. examined	10	0	0	10	-	-	-	-			
Thoracic Cavity		No. examined	0	0	1	0	0	0	0	0			
Adhesions		Total	0	0	1	0	0	0	0	0			
Thymus		No. examined	10	0	0	10	10	0	0	10			
Thyroids		No. examined	10	0	0	10	10	0	0	10			
Trachea		No. examined	10	0	0	10	10	0	0	10			
Foci, Inflammatory Cells		Slight	2	0	0	1	0	0	0	1			
		Total	2	0	0	1	0	0	0	1			

TABLE 10 (cont) Histopathology - group distribution of findings for animals killed after 13 weeks of treatment

Request ID: 5035206

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Tissue/Organ and Findings	Group/Sex No. of animals	Number of animals affected							
		1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Urinary Bladder	No. examined	10	0	0	10	10	0	0	10
Uterine Cervix	No. examined	-	-	-	-	10	0	0	10
Uterus	No. examined	-	-	-	-	10	0	0	10
Vagina	No. examined	-	-	-	-	10	0	0	10
Mucification	Total	-	-	-	-	2	0	0	0
Diestrus	Total	-	-	-	-	1	0	0	0
Metestrus	Total	-	-	-	-	3	0	0	7
Proestrus	Total	-	-	-	-	1	0	0	3
Estrus	Total	-	-	-	-	3	0	0	0

APPENDIX 1 Detailed physical examination and arena observations - individual observations

Request ID: 5031943

Dose Group	Control	Lactase, batch PPL34537			
	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
1M	0001	14	Normal Skin abrasion	Within normal limits Wet, Lower dorsal surface	P1, 1-14 P1
	0002	14	Normal	Within normal limits	P1, 1-14
	0003	14	Normal	Within normal limits	P1, 1-14
	0004	14	Normal	Within normal limits	P1, 1-14
	0005	14	Normal	Within normal limits	P1, 1-14
	0006	14	Normal Coat Skin	Within normal limits Hair loss, Head Encrustation, Head	P1, 1-6 7-14 8-14



APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
1M	0007	14	Normal	Within normal limits	P1, 1-14
	0008	14	Normal	Within normal limits	P1, 1-14
	0009	14	Normal	Within normal limits	P1, 1, 4-14
			Coat	Hair loss, Dorsal surface	2-3
			Skin	Encrustation, Dorsal surface	2
	0010	14	Normal	Within normal limits	4-14
			Coat	Hair loss, Dorsal surface	P1, 1-3
			Skin	Encrustation, Dorsal surface	P1, 1

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
2M	0021	14	Normal	Within normal limits	P1, 4-14
			Coat	Hair loss, Dorsal surface	1-3
			Skin	Encrustation, Dorsal surface	1-3
	0022	14	Normal	Within normal limits	P1, 1-14
	0023	14	Normal	Within normal limits	P1, 1-14
	0024	14	Normal	Within normal limits	P1, 1-14
	0025	14	Normal	Within normal limits	P1, 1-14
	0026	14	Normal	Within normal limits	P1, 1-2, 6-14
			Coat	Hair loss, Dorsal surface	3-5
			Skin	Encrustation, Lower dorsal surface	3-4

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

Dose Group	Control	Lactase, batch PPL34537			
	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
2M	0027	14	Normal	Within normal limits	P1, 1-14
	0028	14	Normal	Within normal limits	P1, 1-14
	0029	14	Normal Coat Skin	Within normal limits	P1, 1-11
				Hair loss, Head	12-14
				Encrustation, Head	12-14
	0030	14	Normal Coat Skin	Within normal limits	P1, 1-10
				Hair loss, Head	11-14
				Encrustation, Head	11-14

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

Dose Group	Control	Lactase, batch PPL34537			
	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
3M	0011	14	Normal	Within normal limits	P1, 1-14
	0012	14	Normal	Within normal limits	P1, 1-14
	0013	14	Normal Coat	Within normal limits Hair loss, Head	P1, 1-8 9-14
	0014	14	Normal	Within normal limits	P1, 1-14
	0016	14	Normal	Within normal limits	P1, 1-14
	0017	14	Normal	Within normal limits	P1, 1-14
	0018	14	Normal	Within normal limits	P1, 1-14
	0019	14	Normal Coat	Within normal limits Hair loss, Head	P1, 1-9 10-14
	0020	14	Normal	Within normal limits	P1, 1-14
	0042	14	Normal	Within normal limits	P1, 1-14

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
4M	0031	14	Normal	Within normal limits	P1, 1-14
	0032	14	Normal	Within normal limits	P1, 1-4
			Coat	Hair loss, Head	5-14
			Skin	Encrustation, Head	7-14
	0033	14	Normal	Within normal limits	P1, 1-8
			Coat	Hair loss, Head	9-14
			Skin	Encrustation, Head	10-14
	0034	14	Normal	Within normal limits	P1, 1-13
			Coat	Hair loss, Forelimbs	14
				Hair loss, Head	14
	0035	14	Normal	Within normal limits	P1, 1-14
	0036	14	Normal	Within normal limits	P1, 1-14

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
4M	0037	14	Normal	Within normal limits	P1, 1-14
	0038	14	Normal Coat Skin	Within normal limits	5-14
				Hair loss, Dorsal surface	1-4
				Encrustation, Dorsal surface	P1
	0039	14	Normal	Within normal limits	P1, 1-14
	0040	14	Normal	Within normal limits	P1, 1-14

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

Dose Group	Control	Lactase, batch PPL34537			
	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
1F	0131	14	Normal Coat	Within normal limits Hair loss, Forelimbs	P1, 1-9 10-14
	0132	14	Normal Coat	Within normal limits Hair loss, Head	P1, 1-9 10-14
	0133	14	Normal Coat	Within normal limits Hair loss, Forelimbs Hair loss, Head	1-8 11-14 10-14
			Eyes	Prominent, Bilateral	9-14
			Skin	Encrustation, Head	9-14
			Staining	Abnormal color, Brown, Muzzle	P1
	0134	14	Normal Coat	Within normal limits Hair loss, Forelimbs	P1, 1-9 10-14
	0135	14	Normal Behavior	Within normal limits Vocalization	13-14 2-12
			Coat	Hair loss, Forelimbs	P1, 1-3

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (% of Toxbatch)	0	10	33	100

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
1F	0136	14	Normal	Within normal limits	P1, 1-9
			Coat	Hair loss, Head	10-14
			Skin	Encrustation, Head	10-14
	0137	14	Normal	Within normal limits	4-7
			Coat	Hair loss, Dorsal surface	P1, 1-3
				Hair loss, Head	8-14
			Skin	Encrustation, Dorsal surface	P1, 1-2
	0138	14	Normal	Within normal limits	P1, 1-5, 7-8
			Coat	Hair loss, Head	9-14
			Skin	Encrustation, Head	9-14
				Encrustation, Upper dorsal surface	6
	0139	14	Normal	Within normal limits	P1, 1-8
			Coat	Hair loss, Head	9-14
			Skin	Encrustation, Head	9-14
	0140	14	Normal	Within normal limits	P1, 1-2
			Behavior	Vocalization	3-12
			Coat	Hair loss, Head	9-14
			Skin	Encrustation, Head	10-14



APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (% of Toxbatch)	0	10	33	100

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
2F	0121	14	Normal Coat Skin	Within normal limits Hair loss, Head Encrustation, Head	P1, 1-9 10-14 10-14
	0122	14	Normal Behavior Coat  Skin	Within normal limits Vocalization Hair loss, Forelimbs Hair loss, Head Encrustation, Head	P1, 1-5 6-7, 9 7-14 6-14 6-14
	0123	14	Normal Coat	Within normal limits Hair loss, Head	P1, 1-6 7-14
	0124	14	Normal Behavior Coat Skin	Within normal limits Vocalization Hair loss, Head Encrustation, Head	P1, 1-4 5-10 6-14 8-14
	0125	14	Normal Behavior	Within normal limits Vocalization	P1, 1-5, 9-14 6-8

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
2F	0126	14	Normal Coat	Within normal limits Hair loss, Dorsal surface	P1, 2-14 1
	0127	14	Normal Behavior Coat Skin Staining	Within normal limits Vocalization Hair loss, Head Encrustation, Head Abnormal color, Brown, Head	P1, 1-2 3-8 7-14 7-14 5-8
	0128	14	Normal Behavior Staining	Within normal limits Vocalization Abnormal color, Brown, Head	P1, 1 2-14 10-14
	0129	14	Normal Behavior Coat Skin	Within normal limits Vocalization Hair loss, Dorsal surface Hair loss, Head Encrustation, Head	P1, 3-9 2 1 10-14 12-14
	0130	14	Normal Coat Skin	Within normal limits Hair loss, Head Encrustation, Head	P1, 1-6 7-14 11-14

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (% of Toxbatch)	0	10	33	100

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
3F	0101	14	Normal Behavior	Within normal limits Vocalization	P1, 1-7, 9-14 8
	0102	14	Normal Coat Skin	Within normal limits Hair loss, Head Encrustation, Head	P1, 1-6 7-14 7-14
	0103	14	Normal Behavior Coat	Within normal limits Irritable Vocalization Hair loss, Dorsal surface	14 9 2-13 P1, 1
	0105	14	Normal Behavior Coat Skin	Within normal limits Vocalization Hair loss, Dorsal surface Hair loss, Forelimbs Encrustation, Dorsal surface	4-10 13-14 1-3 11-14 P1, 1
	0106	14	Normal Staining	Within normal limits Abnormal color, Brown, Head Abnormal color, Brown, Upper dorsal surface	P1, 1-4, 9-10 5-8 11-14

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (% of Toxbatch)	0	10	33	100

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
3F	0107	14	Normal Behavior	Within normal limits Vocalization	P1, 1-5, 8-14 6-7
	0108	14	Normal Build (Deformity)	Within normal limits Swollen area, Ventral surface, Sternum appears prominent	P1, 1-5 6-14
			Coat	Hair loss, Head	10-14
	0109	14	Normal Behavior	Within normal limits Vocalization	P1, 2-5, 8-9 6-7
			Coat Staining	Hair loss, Dorsal surface Abnormal color, Brown, Head	1 10-14
	0110	14	Normal Build (Deformity)	Within normal limits Swollen area, Ventral surface, Sternum appears prominent	P1, 1-8 9-14
	0141	14	Normal Coat	Within normal limits Hair loss, Head	P1, 1-9 10-14

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (% of Toxbatch)	0	10	33	100

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
4F	0111	14	Normal Behavior Coat Staining	Within normal limits Vocalization Hair loss, Dorsal surface Abnormal color, Brown, Head	P1, 1, 8-10 2-7 3 11-14
	0112	14	Normal Behavior	Within normal limits Vocalization	P1, 1, 9-14 2-8
	0113	14	Normal	Within normal limits	P1, 1-14
	0114	14	Normal Coat	Within normal limits Hair loss, Forelimbs	P1, 1-6 7-14
	0115	14	Normal Coat Staining	Within normal limits Hair loss, Forelimbs Abnormal color, Brown, Head	P1, 1-6 7-14 9-14

APPENDIX 1 (cont) Detailed physical examination and arena observations - individual observations

Request ID: 5031943

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (% of Toxbatch)	0	10	33	100

Group /Sex	Animal Number	Week of Death	Category	Observation	Week(s) observed
4F	0116	14	Normal Build (Deformity) Coat	Within normal limits Partially absent appendage, Ear - left Hair loss, Head	P1, 1-6 7-14 10-14
	0117	14	Normal Coat	Within normal limits Hair loss, Dorsal surface Hair loss, Head	P1, 1-2, 4-11 3 12-14
	0118	14	Normal Behavior	Within normal limits Irritable Vocalization	P1, 1, 9-11 12-14 2-8, 12-14
	0119	14	Normal	Within normal limits	P1, 1-14
	0120	14	Normal	Within normal limits	P1, 1-14

APPENDIX 2 Sensory reactivity observations and grip strength - individual findings during Week 12 of treatment

	Control	Lactase, batch PPL34537																			
Dose Group	1	2	3	4																	
Dose (% of Toxbatch)	0	10	33	100																	
Group/sex:	1M										2M										
Animal number:	1	2	3	4	5	6	7	8	9	10	21	22	23	24	25	26	27	28	29	30	
<u>Parameter (Grade range)</u>																					
Approach response (1-3)	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
Pinna reflex (1-3)	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
Auditory startle reflex (1-4)	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	
Tail pinch response (1-4)	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	
Forelimb grip strength (kg)	Trial 1	1.10	0.82	1.30	1.03	0.93	0.94	1.28	1.16	1.24	1.03	0.93	0.97	1.28	0.98	1.43	1.23	1.39	0.92	1.18	1.34
	Trial 2	1.00	0.91	1.46	1.08	0.88	0.98	1.06	1.09	1.43	0.97	1.05	1.24	1.26	0.91	1.14	1.34	1.15	0.94	1.16	1.22
	Trial 3	0.98	0.89	1.23	1.08	0.85	1.04	0.92	1.02	1.23	0.82	1.19	1.13	1.28	1.07	1.13	1.26	1.26	0.99	1.12	1.28
	Mean	1.03	0.87	1.33	1.06	0.89	0.99	1.09	1.09	1.30	0.94	1.06	1.11	1.27	0.99	1.23	1.28	1.27	0.95	1.15	1.28
Hindlimb grip strength (kg)	Trial 1	0.57	0.57	0.58	0.64	0.68	0.54	0.66	0.62	0.79	0.67	0.50	0.58	0.60	0.50	0.65	0.64	0.66	0.58	0.62	0.59
	Trial 2	0.65	0.50	0.47	0.55	0.65	0.62	0.55	0.61	0.85	0.48	0.50	0.49	0.65	0.42	0.70	0.65	0.58	0.63	0.56	0.54
	Trial 3	0.56	0.53	0.58	0.58	0.74	0.60	0.62	0.61	0.78	0.54	0.55	0.50	0.65	0.50	0.66	0.62	0.58	0.53	0.58	0.49
	Mean	0.59	0.53	0.54	0.59	0.69	0.59	0.61	0.61	0.81	0.56	0.52	0.52	0.63	0.47	0.67	0.64	0.61	0.58	0.59	0.54
Additional observations	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	

APPENDIX 2 (cont) Sensory reactivity observations and grip strength - individual findings during Week 12 of treatment

	Control	Lactase, batch PPL34537																			
Dose Group	1	2	3	4																	
Dose (% of Toxbatch)	0	10	33	100																	
Group/sex:	3M										4M										
Animal number:	11	12	13	14	42	16	17	18	19	20	31	32	33	34	35	36	37	38	39	40	
<u>Parameter (Grade range)</u>																					
Approach response (1-3)	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
Pinna reflex (1-3)	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
Auditory startle reflex (1-4)	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	
Tail pinch response (1-4)	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	4	3	
Forelimb grip strength (kg)	Trial 1	0.92	1.02	1.22	1.16	1.07	1.22	1.20	1.10	0.78	1.10	1.13	0.86	1.05	1.44	1.26	0.85	1.16	1.15	0.97	1.26
	Trial 2	1.12	0.96	0.95	1.00	1.02	1.32	0.92	1.19	0.89	0.89	1.22	0.98	0.94	1.33	1.19	0.93	1.01	1.36	1.16	1.13
	Trial 3	1.15	1.00	0.96	0.96	1.07	1.23	1.07	1.24	0.89	0.98	1.16	1.04	1.29	1.26	1.41	0.79	0.97	1.02	0.97	1.10
	Mean	1.06	0.99	1.04	1.04	1.05	1.26	1.06	1.18	0.85	0.99	1.17	0.96	1.09	1.34	1.29	0.86	1.05	1.18	1.03	1.16
Hindlimb grip strength (kg)	Trial 1	0.55	0.58	0.49	0.70	0.55	0.71	0.68	0.78	0.52	0.58	0.54	0.49	0.49	0.63	0.57	0.38	0.53	0.56	0.58	0.56
	Trial 2	0.61	0.54	0.51	0.63	0.55	0.74	0.66	0.78	0.54	0.55	0.66	0.56	0.57	0.61	0.52	0.48	0.50	0.48	0.58	0.62
	Trial 3	0.72	0.73	0.52	0.65	0.58	0.65	0.63	0.70	0.49	0.52	0.68	0.50	0.54	0.73	0.65	0.50	0.45	0.54	0.56	0.45
	Mean	0.63	0.62	0.51	0.66	0.56	0.70	0.66	0.75	0.52	0.55	0.63	0.52	0.53	0.66	0.58	0.45	0.49	0.53	0.57	0.54
Additional observations	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	



APPENDIX 2 (cont) Sensory reactivity observations and grip strength - individual findings during Week 12 of treatment

	Control	Lactase, batch PPL34537																			
Dose Group	1	2	3	4																	
Dose (% of Toxbatch)	0	10	33	100																	
Group/sex:	1F										2F										
Animal number:	131	132	133	134	135	136	137	138	139	140	121	122	123	124	125	126	127	128	129	130	
<u>Parameter (Grade range)</u>																					
Approach response (1-3)	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
Pinna reflex (1-3)	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
Auditory startle reflex (1-4)	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	
Tail pinch response (1-4)	3	3	3	3	3	3	3	3	3	3	3	4	3	3	3	3	3	3	3	3	
Forelimb grip strength (kg)	Trial 1	0.82	1.06	1.14	0.92	0.97	0.69	0.80	0.91	1.09	0.88	0.69	0.86	0.62	0.73	0.76	1.07	1.02	0.97	1.08	0.86
	Trial 2	0.82	1.09	1.32	1.14	1.00	0.71	0.78	1.03	0.86	0.96	0.68	1.03	0.56	0.84	0.77	0.75	1.02	1.06	1.02	0.84
	Trial 3	0.82	1.25	1.32	1.18	1.06	0.70	0.83	0.98	0.86	1.03	0.72	0.92	0.56	0.83	0.73	0.76	0.85	1.09	0.90	1.02
	Mean	0.82	1.13	1.26	1.08	1.01	0.70	0.80	0.97	0.94	0.96	0.70	0.94	0.58	0.80	0.75	0.86	0.96	1.04	1.00	0.91
Hindlimb grip strength (kg)	Trial 1	0.38	0.50	0.57	0.49	0.58	0.36	0.50	0.51	0.50	0.44	0.40	0.50	0.42	0.38	0.46	0.57	0.52	0.56	0.64	0.44
	Trial 2	0.46	0.59	0.51	0.47	0.50	0.43	0.59	0.49	0.55	0.56	0.36	0.47	0.38	0.38	0.44	0.50	0.48	0.49	0.50	0.47
	Trial 3	0.40	0.54	0.65	0.60	0.53	0.40	0.51	0.48	0.50	0.61	0.35	0.48	0.47	0.42	0.42	0.48	0.58	0.55	0.44	0.56
	Mean	0.41	0.54	0.58	0.52	0.54	0.40	0.53	0.49	0.52	0.54	0.37	0.48	0.42	0.39	0.44	0.52	0.53	0.53	0.53	0.49
Additional observations	-	-	-	-	-	-	-	-	-	-	-	-	a	-	-	-	-	-	-	-	

a Excessive vocalisation during grip strength procedure - moderate

APPENDIX 2 (cont) Sensory reactivity observations and grip strength - individual findings during Week 12 of treatment

	Control	Lactase, batch PPL34537																			
Dose Group	1	2	3	4																	
Dose (% of Toxbatch)	0	10	33	100																	
Group/sex:	3F										4F										
Animal number:	101	102	103	141	105	106	107	108	109	110	111	112	113	114	115	116	117	118	119	120	
<u>Parameter (Grade range)</u>																					
Approach response (1-3)	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
Pinna reflex (1-3)	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	
Auditory startle reflex (1-4)	3	3	3	2	3	3	3	3	3	3	3	3	3	3	3	3	2	3	3	3	
Tail pinch response (1-4)	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	3	4	
Forelimb grip strength (kg)	Trial 1	0.91	1.37	0.79	0.84	0.91	0.90	1.09	1.05	0.87	0.97	0.78	0.84	1.02	1.03	0.83	0.94	0.90	0.90	0.89	1.04
	Trial 2	0.96	1.10	0.93	0.83	0.86	0.89	0.97	0.84	0.94	0.86	0.79	0.84	0.88	1.01	0.78	0.92	0.98	0.75	0.96	1.01
	Trial 3	0.82	1.09	1.02	0.94	0.92	0.85	1.01	0.71	0.87	0.84	0.90	0.86	1.03	1.12	0.82	0.84	0.98	0.72	0.95	0.95
	Mean	0.90	1.19	0.91	0.87	0.90	0.88	1.02	0.87	0.89	0.89	0.82	0.85	0.98	1.05	0.81	0.90	0.95	0.79	0.93	1.00
Hindlimb grip strength (kg)	Trial 1	0.42	0.51	0.51	0.42	0.46	0.47	0.50	0.42	0.46	0.45	0.42	0.50	0.46	0.45	0.38	0.43	0.42	0.44	0.45	0.45
	Trial 2	0.45	0.48	0.43	0.41	0.48	0.43	0.46	0.49	0.42	0.42	0.41	0.46	0.44	0.42	0.40	0.40	0.48	0.36	0.36	0.48
	Trial 3	0.45	0.42	0.41	0.48	0.46	0.45	0.49	0.40	0.46	0.52	0.43	0.43	0.50	0.48	0.41	0.40	0.40	0.41	0.39	0.50
	Mean	0.44	0.47	0.45	0.44	0.47	0.45	0.48	0.44	0.45	0.46	0.42	0.46	0.47	0.45	0.40	0.41	0.43	0.40	0.40	0.48
Additional observations	a	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	a	-	-	

a Excessive vocalisation during grip strength procedure - moderate

APPENDIX 3 Motor activity - individual scores (beam breaks) during Week 12 of treatment

		Control	Lactase, batch PPL34537										
Dose Group		1	2	3	4								
Dose (% of Toxbatch)		0	10	33	100								
Group /Sex	Animal number	Beam level	6	12	18	24	Time (minutes)						Total
							30	36	42	48	54	60	
1M	1	High	115	94	57	15	23	0	25	22	0	0	351
	2		110	53	42	21	6	0	0	0	0	7	239
	3		102	60	11	25	15	1	0	0	0	0	214
	4		111	60	8	29	6	0	0	0	3	0	217
	5		86	48	49	19	26	0	0	43	1	0	272
	6		127	65	21	37	9	30	31	70	0	20	410
	7		132	61	35	23	18	0	23	30	0	0	322
	8		118	77	1	0	0	0	44	76	21	0	337
	9		120	56	24	62	0	0	0	0	0	0	262
	10		65	51	59	53	22	0	0	0	1	40	291
	1	Low	166	123	89	93	62	15	117	59	14	19	757
	2		142	68	65	20	38	9	8	3	1	13	367
	3		183	81	88	71	22	3	1	0	0	24	473
	4		159	78	3	56	57	9	16	49	26	0	453
	5		141	72	77	33	66	14	35	62	4	10	514
	6		176	137	50	112	90	61	47	81	24	46	824
	7		145	88	67	8	29	10	20	43	2	0	412
	8		190	160	22	10	0	0	81	93	74	40	670
	9		179	166	138	126	2	0	0	0	2	0	613
	10		157	86	75	78	40	11	1	3	15	63	529

APPENDIX 3 (cont) Motor activity - individual scores (beam breaks) during Week 12 of treatment

Dose Group		Control	Lactase, batch PPL34537										
Dose (% of Toxbatch)		1	2	3	4								
		0	10	33	100								
Group /Sex	Animal number	Beam level	6	12	18	24	Time (minutes)						Total
							30	36	42	48	54	60	
2M	21	High	160	76	36	20	9	7	8	0	0	0	316
	22		116	61	53	42	0	29	12	6	0	0	319
	23		127	64	16	54	15	22	0	0	0	7	305
	24		105	70	47	0	0	35	0	0	0	0	257
	25		124	31	35	13	0	1	0	0	0	0	204
	26		96	90	39	60	7	12	23	0	0	0	327
	27		71	63	80	67	27	0	7	66	7	0	388
	28		157	130	72	106	56	35	0	0	0	0	556
	29		159	117	100	64	97	31	12	43	25	64	712
	30		97	62	20	4	2	0	0	0	0	0	185
	21	Low	240	94	86	80	48	33	35	0	0	0	616
	22		200	111	77	77	17	37	48	21	5	3	596
	23		214	112	61	44	32	29	41	22	15	39	609
	24		166	96	22	2	1	70	3	11	5	10	386
	25		224	99	48	45	3	22	0	3	0	0	444
	26		147	79	101	94	100	106	37	0	9	13	686
	27		144	170	106	61	40	3	23	75	44	17	683
	28		186	169	137	95	145	54	11	3	30	7	837
	29		245	172	102	54	115	74	31	43	62	42	940
	30		185	158	109	6	29	0	0	0	0	0	487

APPENDIX 3 (cont) Motor activity - individual scores (beam breaks) during Week 12 of treatment

		Control	Lactase, batch PPL34537										
Dose Group		1	2	3	4								
Dose (% of Toxbatch)		0	10	33	100								
Group /Sex	Animal number	Beam level	6	12	18	24	Time (minutes)						Total
							30	36	42	48	54	60	
3M	11	High	113	68	72	82	64	26	49	69	45	28	616
	12		130	110	61	45	15	0	0	4	5	4	374
	13		110	82	44	8	0	0	0	0	0	0	244
	14		126	69	59	25	19	6	0	0	0	0	304
	16		120	94	57	27	30	62	23	68	62	4	547
	17		123	73	33	31	6	0	0	0	0	0	266
	18		111	45	24	9	0	0	0	5	0	0	194
	19		122	86	22	83	6	10	40	0	0	11	380
	20		140	108	61	30	10	0	1	0	0	0	350
	42		131	126	48	38	0	0	0	0	38	40	421
	11	Low	201	78	81	41	98	33	70	25	35	27	689
	12		195	98	114	47	21	0	0	3	39	50	567
	13		186	165	91	37	14	6	0	0	0	3	502
	14		296	184	130	47	71	15	0	19	0	0	762
	16		171	90	43	111	52	42	37	44	66	29	685
	17		180	118	83	92	40	4	33	20	2	0	572
	18		200	111	53	65	26	145	1	20	12	0	633
	19		152	127	53	69	20	28	91	2	4	11	557
	20		239	132	134	63	21	42	3	0	0	0	634
	42		165	81	56	43	7	1	1	0	77	90	521

APPENDIX 3 (cont) Motor activity - individual scores (beam breaks) during Week 12 of treatment

Dose Group		Control	Lactase, batch PPL34537										
Dose (% of Toxbatch)		1	2	3	4								
		0	10	33	100								
Group /Sex	Animal number	Beam level	Time (minutes)										
			6	12	18	24	30	36	42	48	54	60	Total
4M	31	High	59	44	1	16	0	0	0	50	23	0	193
	32		112	73	1	0	0	0	0	48	25	46	305
	33		146	75	5	0	0	0	0	0	21	14	261
	34		109	31	11	4	0	0	0	0	0	0	155
	35		80	67	43	18	8	0	0	9	18	47	290
	36		99	77	34	14	0	0	49	91	0	36	400
	37		101	46	46	20	0	0	0	38	0	0	251
	38		121	84	52	31	26	8	18	0	0	6	346
	39		111	118	18	0	0	0	0	0	0	0	247
	40		164	85	106	56	3	43	2	23	5	0	487
	31	Low	129	111	38	49	0	0	38	97	21	16	499
	32		154	120	40	0	10	4	0	58	75	73	534
	33		157	111	12	21	6	0	0	0	44	19	370
	34		160	64	38	59	11	0	0	16	0	0	348
	35		133	112	56	38	48	21	7	31	42	31	519
	36		264	129	127	45	23	13	61	76	14	70	822
	37		239	123	74	36	25	8	0	84	0	0	589
	38		223	158	96	71	77	70	54	0	2	0	751
	39		183	198	70	12	0	1	0	0	0	0	464
	40		262	155	84	109	1	64	25	82	42	41	865

APPENDIX 3 (cont) Motor activity - individual scores (beam breaks) during Week 12 of treatment

		Control	Lactase, batch PPL34537												
Dose Group		1	2	3	4										
Dose (% of Toxbatch)		0	10	33	100										
Group /Sex	Animal number	Beam level	6	12	18	24	Time (minutes)		30	36	42	48	54	60	Total
1F	131	High	131	69	25	0	6	1	0	0	5	0	237		
	132		244	179	72	57	75	3	19	35	52	19	755		
	133		192	187	39	0	0	0	35	26	0	0	479		
	134		167	62	90	8	0	0	0	61	58	51	497		
	135		150	43	40	12	0	0	62	11	72	0	390		
	136		234	124	92	47	61	37	95	66	93	23	872		
	137		119	45	62	0	29	15	0	0	3	4	277		
	138		193	85	72	42	1	32	18	13	5	0	461		
	139		141	69	3	0	0	0	0	0	0	61	274		
	140		179	106	87	104	0	0	18	10	82	85	671		
	131	Low	314	204	135	5	73	0	0	1	49	0	781		
	132		213	135	99	66	105	33	42	84	54	58	889		
	133		261	218	80	2	25	1	103	28	4	5	727		
	134		191	86	111	11	0	0	0	79	70	42	590		
	135		153	62	60	58	0	0	101	55	20	3	512		
	136		183	139	135	143	113	71	72	70	72	49	1047		
	137		235	118	109	80	86	31	0	0	10	15	684		
	138		232	183	158	84	41	94	80	81	40	12	1005		
	139		172	80	49	0	0	0	0	0	25	87	413		
	140		282	176	122	172	1	1	49	53	109	132	1097		

APPENDIX 3 (cont) Motor activity - individual scores (beam breaks) during Week 12 of treatment

		Control	Lactase, batch PPL34537												
Dose Group		1	2	3	4										
Dose (% of Toxbatch)		0	10	33	100										
Group /Sex	Animal number	Beam level	6	12	18	24	Time (minutes)		30	36	42	48	54	60	Total
2F	121	High	107	88	33	42	16	12	1	0	0	0	0	299	
	122		128	35	3	0	13	0	0	0	50	0	229		
	123		145	63	99	38	53	1	0	0	0	42	441		
	124		202	87	75	14	0	0	0	0	8	57	443		
	125		134	85	68	27	77	4	2	0	75	24	496		
	126		147	87	0	0	0	0	87	56	0	2	379		
	127		134	62	74	0	0	0	0	5	89	23	387		
	128		139	112	29	0	9	45	54	4	0	0	392		
	129		173	85	42	46	46	17	32	69	18	24	552		
	130		147	68	18	0	0	5	44	45	0	0	327		
	121	Low	204	148	117	85	75	61	13	3	0	0	706		
	122		143	126	20	13	87	5	0	0	84	25	503		
	123		214	92	116	74	39	0	0	0	0	57	592		
	124		231	122	104	169	66	6	28	6	36	122	890		
	125		188	56	77	19	82	64	14	26	31	21	578		
	126		162	79	3	3	13	3	56	65	6	4	394		
	127		204	68	69	0	0	0	0	9	105	54	509		
	128		184	80	61	7	23	42	85	17	0	0	499		
	129		198	109	118	69	82	24	37	78	36	75	826		
	130		231	172	28	4	0	26	80	57	0	0	598		



APPENDIX 3 (cont) Motor activity - individual scores (beam breaks) during Week 12 of treatment

		Control	Lactase, batch PPL34537										
Dose Group		1	2	3	4								
Dose (% of Toxbatch)		0	10	33	100								
Group /Sex	Animal number	Beam level	6	12	18	24	Time (minutes)		42	48	54	60	Total
3F	101	High	164	91	24	58	7	0	0	0	5	0	349
	102		213	77	22	49	15	6	5	32	0	0	419
	103		138	117	57	31	0	26	0	21	100	41	531
	105		155	103	27	15	0	0	0	0	47	77	424
	106		163	88	28	21	1	0	5	28	0	0	334
	107		168	111	81	25	36	9	11	34	27	20	522
	108		216	169	67	64	0	0	30	76	41	94	757
	109		137	50	61	28	40	6	0	0	0	0	322
	110		187	70	90	0	0	0	37	49	0	0	433
	141		160	90	43	6	0	0	0	4	37	79	419
	101	Low	231	82	41	129	31	0	0	0	17	0	531
	102		269	123	65	43	30	144	60	37	0	11	782
	103		156	154	106	121	15	80	23	55	68	76	854
	105		216	157	64	48	0	0	0	0	99	110	694
	106		184	137	57	97	5	0	18	98	19	8	623
	107		192	114	107	77	54	24	27	55	93	67	810
	108		272	167	70	83	0	12	88	73	40	8	813
	109		185	68	143	94	50	15	1	0	24	31	611
	110		177	92	72	12	61	0	78	44	0	18	554
	141		223	161	74	62	1	6	0	15	106	90	738

APPENDIX 3 (cont) Motor activity - individual scores (beam breaks) during Week 12 of treatment

		Control	Lactase, batch PPL34537										
Dose Group		1	2	3	4								
Dose (% of Toxbatch)		0	10	33	100								
Group /Sex	Animal number	Beam level	6	12	18	24	Time (minutes)		42	48	54	60	Total
4F	111	High	91	30	56	0	0	0	14	49	11	44	295
	112		151	89	66	27	74	45	0	0	0	0	452
	113		133	57	90	25	24	27	40	0	0	69	465
	114		149	118	95	8	30	3	0	0	0	69	472
	115		176	71	47	4	0	0	0	51	62	51	462
	116		117	13	18	32	0	0	0	0	0	0	180
	117		192	66	35	47	30	0	0	1	12	0	383
	118		264	153	147	90	94	108	10	0	13	80	959
	119		223	113	61	14	57	5	42	0	52	47	614
	120		166	55	75	0	62	17	0	2	0	0	377
	111	Low	234	187	122	22	0	0	104	92	62	153	976
	112		217	157	127	27	94	43	26	0	20	8	719
	113		181	112	74	131	54	54	29	0	13	71	719
	114		194	166	108	68	94	33	19	38	27	122	869
	115		245	149	87	132	1	0	0	39	57	71	781
	116		183	59	40	85	13	26	23	6	4	17	456
	117		284	164	97	87	90	6	14	46	26	17	831
	118		220	118	77	12	49	28	18	0	2	41	565
	119		182	124	131	46	111	75	71	65	46	34	885
	120		206	73	148	8	92	29	0	1	0	0	557

APPENDIX 4 Body weight - individual values (g)

Request ID: 5031939

Dose Group	Control	Lactase, batch PPL34537			
Dose (% of Toxbatch)	1	2	3	4	
	0	10	33	100	

Group /Sex	Animal Number	Week P1	0	1	2	3	4	5	6	7	8	9	10	11	12	13
1M	0001	210	273	335	357	410	454	484	512	536	561	580	595	618	623	639
	0002	185	238	280	319	346	372	390	401	418	430	436	446	459	466	472
	0003	206	271	336	385	426	462	484	506	526	540	553	567	582	591	594
	0004	183	248	289	321	343	363	380	401	422	434	444	457	465	468	474
	0005	209	269	329	376	422	456	485	511	528	555	567	577	595	601	600
	0006	224	291	353	398	448	487	517	543	558	583	592	606	620	622	629
	0007	225	292	353	398	454	496	523	556	574	600	625	646	663	678	686
	0008	207	261	319	347	380	406	418	439	449	461	477	492	504	514	526
	0009	214	282	350	386	434	458	493	509	533	556	578	591	615	622	636
	0010	206	261	304	346	383	419	439	460	473	500	525	539	564	570	572
2M	0021	189	244	284	317	343	366	382	401	408	421	426	435	442	457	462
	0022	196	261	315	356	385	415	440	474	491	522	534	557	565	576	582
	0023	196	253	304	335	364	392	407	431	445	456	474	480	497	498	506
	0024	194	255	307	351	385	417	445	479	493	511	526	547	558	571	572
	0025	211	276	340	388	430	462	477	504	519	536	550	559	581	590	591
	0026	219	273	329	366	404	442	462	486	503	523	547	559	575	585	592
	0027	203	259	315	355	394	431	458	473	478	481	494	511	526	534	539
	0028	195	256	307	348	383	416	438	460	476	499	517	523	539	554	548
	0029	224	286	339	382	414	447	476	498	510	534	555	566	579	587	595
	0030	206	266	321	358	393	419	430	451	463	486	500	512	529	534	539

APPENDIX 4 (cont) Body weight - individual values (g)

Request ID: 5031939

Dose Group	Control	Lactase, batch PPL34537			
Dose (% of Toxbatch)	1	2	3	4	
	0	10	33	100	

Group /Sex	Animal Number	Week P1	0	1	2	3	4	5	6	7	8	9	10	11	12	13
3M	0011	186	246	294	328	362	388	399	418	426	443	453	467	451	477	486
	0012	187	251	309	312	361	396	428	451	469	486	510	531	549	564	563
	0013	210	275	334	380	419	454	480	509	521	546	570	577	585	596	607
	0014	181	231	270	293	322	340	351	361	367	382	394	405	419	429	430
	0016	212	267	308	335	371	391	404	420	428	447	457	477	496	498	501
	0017	212	270	323	357	400	425	446	461	469	490	507	515	526	531	539
	0018	211	280	345	396	453	492	521	551	576	602	622	635	645	656	667
	0019	205	271	327	369	398	420	428	451	464	476	487	500	518	527	528
	0020	188	250	314	361	416	450	481	508	524	540	559	570	583	585	592
	0042	201	262	305	337	375	397	412	430	444	458	468	474	485	486	492
4M	0031	200	257	300	338	372	401	427	446	462	481	492	498	508	517	525
	0032	215	281	341	378	423	456	469	486	501	522	533	543	550	560	558
	0033	192	253	311	360	405	441	466	491	507	530	546	554	573	585	584
	0034	216	283	344	389	430	456	487	505	525	555	577	592	619	626	636
	0035	194	251	315	348	386	417	439	458	474	494	511	520	535	547	557
	0036	202	262	317	364	402	441	467	496	504	531	549	566	578	590	591
	0037	205	263	312	352	386	415	436	457	464	482	493	502	514	530	528
	0038	185	238	284	320	350	379	392	417	424	444	453	463	478	479	485
	0039	201	273	330	373	417	453	483	506	524	545	560	572	583	595	590
	0040	197	251	296	324	357	389	414	433	440	455	469	479	487	498	503

APPENDIX 4 (cont) Body weight - individual values (g)

Request ID: 5031939

Dose Group	Control	Lactase, batch PPL34537			
Dose (% of Toxbatch)	1	2	3	4	
	0	10	33	100	

Group /Sex	Animal Number	Week P1	0	1	2	3	4	5	6	7	8	9	10	11	12	13
1F	0131	155	178	203	219	229	236	249	263	259	260	268	274	279	275	280
	0132	166	191	205	231	247	260	251	272	280	285	276	292	301	302	297
	0133	174	204	235	249	257	283	300	306	307	326	322	333	340	339	345
	0134	182	204	228	249	264	269	282	290	290	294	298	304	305	302	311
	0135	186	222	233	260	284	299	296	314	325	328	324	331	332	339	343
	0136	152	186	208	222	227	252	262	266	256	271	277	280	275	287	290
	0137	169	196	207	215	231	247	253	253	262	281	276	270	284	287	288
	0138	167	188	206	231	248	259	260	279	279	291	284	295	305	308	295
	0139	177	215	233	264	280	305	314	328	330	340	340	346	360	363	359
	0140	177	201	227	241	262	278	282	289	301	311	311	316	325	329	322
2F	0121	169	193	222	241	256	261	275	282	282	285	298	299	301	300	301
	0122	170	201	225	241	258	269	273	283	288	295	296	302	309	311	304
	0123	161	182	203	215	235	249	253	256	265	277	281	275	284	296	292
	0124	160	188	206	225	244	252	255	266	271	274	276	279	285	289	277
	0125	166	190	207	219	231	242	256	259	254	264	274	276	276	280	284
	0126	158	188	197	220	234	242	246	264	263	269	265	272	265	269	272
	0127	165	182	210	228	241	244	257	265	268	268	275	284	293	288	290
	0128	168	193	212	231	249	258	264	274	281	286	287	294	302	305	302
	0129	178	202	223	227	250	263	274	263	274	274	287	289	294	283	289
	0130	162	192	210	225	240	257	261	267	275	278	284	283	294	299	289

APPENDIX 4 (cont) Body weight - individual values (g)

Request ID: 5031939

Dose Group	Control	Lactase, batch PPL34537			
	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Group /Sex	Animal Number	Week P1	0	1	2	3	4	5	6	7	8	9	10	11	12	13
3F	0101	173	202	225	251	268	275	287	300	305	304	315	319	325	326	332
	0102	182	202	226	235	247	261	269	275	275	288	295	291	294	304	302
	0103	171	201	222	232	241	254	262	266	274	291	292	283	294	298	295
	0105	173	208	224	241	252	264	277	286	282	293	300	308	306	310	314
	0106	186	204	231	250	268	273	289	303	306	307	318	331	339	338	341
	0107	182	212	239	257	259	281	290	293	308	320	324	316	325	332	330
	0108	169	188	211	221	232	245	253	266	259	272	282	277	279	283	288
	0109	171	200	220	239	253	266	284	298	289	303	313	317	316	327	330
	0110	176	211	227	241	254	270	284	286	293	305	309	305	312	322	316
	0141	157	188	214	228	243	253	268	273	274	284	293	295	293	303	302
4F	0111	167	193	207	214	232	247	250	250	259	270	273	271	279	287	285
	0112	168	199	223	241	254	269	281	292	293	301	305	312	313	320	324
	0113	181	202	219	237	265	273	276	291	294	301	300	300	315	316	311
	0114	168	195	212	220	239	246	245	259	264	267	265	268	280	283	275
	0115	173	202	221	237	251	266	282	291	290	298	312	312	316	324	329
	0116	166	190	210	230	249	266	280	289	296	310	314	317	328	335	327
	0117	168	189	210	227	251	265	269	282	288	302	304	307	316	323	312
	0118	165	190	212	229	234	244	264	266	259	271	280	284	281	290	292
	0119	171	184	207	232	248	247	262	276	279	275	288	297	300	300	297
	0120	177	197	212	219	232	244	250	250	259	265	269	261	272	281	272

APPENDIX 5 Food consumption - individual cage mean values (g/animal/week)

Request ID: 5031940

		Control	Lactase, batch PPL34537												
Dose Group		1	2	3	4										
Dose (% of Toxbatch)		0	10	33	100										
Group /Sex	Cage Number	Week P1	1	2	3	4	5	6	7	8	9	10	11	12	13
1M	001	187	200	195	196	201	197	200	204	195	192	200	192	195	183
	002	204	210	213	215	215	212	212	216	211	214	218	213	210	198
2M	005	189	196	195	189	191	196	191	201	186	188	188	196	185	181
	006	191	201	201	195	204	203	194	200	189	198	197	196	194	185
3M	003	186	187	178	181	191	187	189	189	187	186	193	187	191	173
	004	190	196	201	202	199	196	198	201	196	199	200	194	194	178
4M	007	191	206	209	207	203	206	202	208	199	207	202	204	196	185
	008	182	187	195	192	192	194	192	192	187	190	190	188	181	176

APPENDIX 5 (cont) Food consumption - individual cage mean values (g/animal/week)

Request ID: 5031940

Dose Group Dose (%)	Control	Lactase, batch PPL34537													
	1	2	3	4											
	0	10	33	100											
Group /Sex	Cage Number	Week P1	1	2	3	4	5	6	7	8	9	10	11	12	13
1F	015	130	135	146	139	144	144	149	142	138	136	144	137	133	133
	016	129	133	148	144	150	145	149	148	141	141	142	142	139	134
2F	013	117	125	132	131	132	138	125	130	133	130	124	127	123	121
	014	123	131	134	132	134	141	136	135	128	136	136	130	127	126
3F	009	132	137	145	137	142	138	140	138	140	136	132	135	130	125
	010	125	135	142	149	143	145	144	145	143	139	137	141	133	127
4F	011	127	129	138	137	139	136	136	133	126	132	128	130	122	124
	012	125	128	134	135	138	139	134	134	130	132	131	136	130	118



APPENDIX 6 Ophthalmic examination - individual observations before commencement of treatment

Request ID: 2014933

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Structure	Observation	Animal number(s)								
		Group/Sex: No. Examined:	1M 10	2M 10	3M 10	4M 10	1F 10	2F 10	3F 10	4F 10
Cornea	Opacities, Superficial, Faint						0133 0135	0123	0101	0119
Iris	Persistent Pupillary Membrane									0120Lt
Lens	Persistent Hyaloid Artery		0003 0004 0010	0023 0025 0026 0029Lt	0011	0035 0037	0139	0124 0128	0106	0118 0119
Fundus	Hyperreflection, Dorsomedial, Band, Slight					0033Rt				

Lt - Left, Rt - Right  
Only animals with observations are presented

APPENDIX 6 (cont) Ophthalmic examination - individual observations during Week 12 of treatment

Request ID: 2014934

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (% of Toxbatch)	0	10	33	100	

Structure	Observation	Animal number(s)				
		Group/Sex:	1M	4M	1F	4F
		No. Examined:	10	10	10	10
Cornea	Opacities, Superficial, Faint				0133	0119
Iris	Persistent Pupillary Membrane					0120Lt
Lens	Persistent Hyaloid Artery		0003 0004 0010	0035 0037	0139	0118 0119
Fundus	Hyperreflecion, Dorsomedial, Band, Slight			0033Rt		

Lt - Left, Rt - Right  
Only animals with observations are presented

APPENDIX 7 Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2 10    3 33    4 100						
Group /Sex	Animal Number	Hct L/L	Hb g/dL	RBC x10 <sup>12</sup> /L	Retic x10 <sup>12</sup> /L	MCH pg	MCHC g/dL	MCV fL	RDW %
1M	1	0.460	15.0	9.02	0.239	16.7	32.7	51.0	14.9
	2	0.456	15.4	8.49	0.209	18.1	33.8	53.7	15.4
	3	0.474	15.6	8.75	0.219	17.8	32.8	54.3	14.0
	4	0.446	14.6	8.49	0.228	17.2	32.8	52.6	14.1
	5	0.445	14.3	8.37	0.256	17.1	32.1	53.1	13.6
	6	0.469	15.6	8.79	0.223	17.8	33.4	53.3	12.8
	7	0.462	15.1	8.89	0.199	17.0	32.7	52.0	14.0
	8	0.467	15.3	8.83	0.164	17.4	32.8	52.9	13.5
	9	0.444	14.5	8.80	0.217	16.5	32.7	50.5	13.9
	10	0.452	14.7	8.72	0.261	16.9	32.5	51.9	15.9
2M	21	0.492	16.5	8.98	0.215	18.4	33.7	54.8	12.4
	22	0.447	15.0	8.35	0.173	18.0	33.5	53.6	13.5
	23	0.443	14.8	8.52	0.218	17.4	33.5	52.0	13.7
	24	0.436	14.4	8.65	0.229	16.6	33.0	50.4	15.9
	25	0.439	14.4	8.61	0.187	16.7	32.7	51.0	13.7
	26	0.478	16.3	9.13	0.155	17.9	34.2	52.3	12.8
	27	0.460	15.2	8.88	0.269	17.1	33.1	51.8	15.5
	28	0.489	16.4	8.94	0.131	18.3	33.5	54.7	12.1
	29	0.460	15.2	9.07	0.268	16.7	33.0	50.7	16.0
	30	0.443	15.0	8.21	0.176	18.2	33.8	54.0	13.5

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2      3      4 10    33    100					
Group /Sex	Animal Number	WBC x10 <sup>9</sup> /L	N x10 <sup>9</sup> /L	L x10 <sup>9</sup> /L	E x10 <sup>9</sup> /L	B x10 <sup>9</sup> /L	M x10 <sup>9</sup> /L	LUC x10 <sup>9</sup> /L
1M	1	14.12	1.35	12.10	0.18	0.08	0.29	0.13
	2	12.12	1.87	9.60	0.17	0.05	0.23	0.19
	3	11.51	2.91	7.61	0.41	0.06	0.43	0.08
	4	16.47	3.42	12.04	0.16	0.11	0.62	0.11
	5	17.69	3.18	13.40	0.41	0.13	0.38	0.19
	6	16.35	2.62	12.97	0.14	0.11	0.38	0.12
	7	16.34	1.85	13.29	0.35	0.10	0.60	0.16
	8	14.98	2.27	11.92	0.17	0.09	0.43	0.09
	9	9.26	2.18	6.56	0.12	0.02	0.33	0.05
	10	13.76	1.70	11.35	0.18	0.13	0.28	0.12
2M	21	9.81	2.03	7.17	0.19	0.06	0.33	0.03
	22	10.33	0.91	8.86	0.11	0.09	0.24	0.12
	23	16.27	2.94	12.42	0.24	0.13	0.49	0.06
	24	12.52	1.88	9.94	0.15	0.10	0.36	0.08
	25	15.38	2.62	11.67	0.30	0.17	0.50	0.13
	26	11.00	1.27	9.04	0.15	0.13	0.32	0.09
	27	13.27	1.57	11.08	0.08	0.09	0.40	0.06
	28	11.11	0.98	9.45	0.16	0.12	0.28	0.12
	29	11.33	1.06	9.70	0.15	0.08	0.24	0.11
	30	11.50	0.95	9.90	0.12	0.06	0.31	0.14

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2      3      4 10    33    100							
Group /Sex	Animal Number	Plt x10 <sup>9</sup> /L	PT sec	APTT sec	Aniso- cytosis	Micro- cytosis	Macro- cytosis	Hypo- chromasia	Hyper- chromasia	
1M	1	1034	20.2	15.7	-	-	-	-	-	
	2	1014	CTD	CTD	-	-	-	-	-	
	3	1023	20.3	16.5	-	-	-	-	-	
	4	1053	20.9	16.9	-	-	-	-	-	
	5	1096	22.2	15.6	-	-	-	-	-	
	6	1070	22.4	16.1	-	-	-	-	-	
	7	1052	24.7	17.2	-	-	-	-	-	
	8	1824	21.6	17.3	-	-	-	-	-	
	9	887	21.7	15.7	-	-	-	-	-	
	10	997	20.6	17.2	-	-	-	-	-	
2M	21	810	20.6	16.7	-	-	-	-	-	
	22	1003	21.6	17.5	-	-	-	-	-	
	23	1031	23.1	14.6	-	-	-	-	-	
	24	1057	20.8	17.2	-	-	-	-	-	
	25	1104	21.8	16.5	-	-	-	-	-	
	26	859	21.8	16.7	-	-	-	-	-	
	27	1071	22.0	16.5	-	-	-	-	-	
	28	827	23.1	17.5	-	-	-	-	-	
	29	1038	23.6	17.3	-	-	-	-	-	
	30	857	22.0	17.8	-	-	-	-	-	

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2 10    3 33    4 100						
Group /Sex	Animal Number	Hct L/L	Hb g/dL	RBC x10 <sup>12</sup> /L	Retic x10 <sup>12</sup> /L	MCH pg	MCHC g/dL	MCV fL	RDW %
3M	11	0.456	15.4	8.55	0.183	18.0	33.8	53.4	14.3
	12	0.479	16.1	8.70	0.159	18.5	33.5	55.1	12.7
	13	0.448	14.9	8.51	0.211	17.5	33.3	52.6	14.5
	14	0.476	15.9	8.69	0.175	18.3	33.4	54.7	13.0
	16	0.461	15.2	8.55	0.273	17.8	33.0	54.0	13.7
	17	0.445	15.1	8.22	0.186	18.3	33.8	54.1	12.3
	18	0.438	14.5	8.15	0.214	17.8	33.0	53.8	14.2
	19	0.466	15.6	8.78	0.150	17.7	33.4	53.1	12.5
	20	0.436	14.3	8.18	0.236	17.5	32.8	53.3	13.9
	42	0.449	14.9	8.57	0.204	17.4	33.3	52.4	13.9
4M	31	0.479	15.9	9.18	0.175	17.3	33.2	52.2	12.1
	32	0.437	14.6	8.11	0.188	18.0	33.5	53.8	13.5
	33	0.449	14.9	8.60	0.165	17.3	33.1	52.2	13.6
	34	0.435	14.5	7.85	0.202	18.5	33.3	55.4	13.3
	35	0.449	15.0	8.52	0.169	17.6	33.4	52.7	12.4
	36	0.448	14.9	8.62	0.192	17.3	33.3	51.9	14.4
	37	0.467	15.8	8.95	0.197	17.6	33.8	52.1	13.2
	38	0.483	16.0	9.05	0.164	17.7	33.1	53.4	12.1
	39	0.454	15.2	8.76	0.219	17.4	33.5	51.8	13.5
	40	0.464	15.7	8.57	0.127	18.3	33.9	54.1	12.6

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2      3      4 10    33    100					
Group /Sex	Animal Number	WBC x10 <sup>9</sup> /L	N x10 <sup>9</sup> /L	L x10 <sup>9</sup> /L	E x10 <sup>9</sup> /L	B x10 <sup>9</sup> /L	M x10 <sup>9</sup> /L	LUC x10 <sup>9</sup> /L
3M	11	9.53	0.70	8.31	0.14	0.09	0.22	0.07
	12	11.70	1.04	10.05	0.18	0.08	0.26	0.10
	13	16.93	2.29	13.62	0.20	0.15	0.59	0.09
	14	11.03	1.68	8.64	0.22	0.09	0.28	0.11
	16	18.26	3.88	13.16	0.22	0.15	0.70	0.14
	17	18.56	4.30	12.84	0.39	0.13	0.82	0.08
	18	13.71	3.81	8.84	0.25	0.07	0.67	0.08
	19	14.51	1.44	12.30	0.18	0.07	0.43	0.08
	20	19.25	2.75	15.38	0.21	0.18	0.50	0.22
	42	22.15	2.42	18.75	0.23	0.14	0.47	0.14
4M	31	12.26	2.15	9.20	0.18	0.17	0.48	0.07
	32	11.20	2.68	8.01	0.12	0.05	0.29	0.06
	33	15.25	1.96	12.13	0.21	0.09	0.63	0.23
	34	12.44	2.75	8.59	0.16	0.07	0.60	0.26
	35	10.63	0.83	9.20	0.15	0.09	0.28	0.09
	36	13.44	2.79	10.03	0.13	0.06	0.34	0.09
	37	10.15	1.65	7.84	0.11	0.08	0.35	0.12
	38	11.68	2.79	7.96	0.49	0.10	0.27	0.08
	39	15.20	3.89	10.47	0.25	0.10	0.45	0.05
	40	11.48	2.15	8.70	0.17	0.08	0.28	0.09

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2      3      4 10     33     100							
Group /Sex	Animal Number	Plt x10 <sup>9</sup> /L	PT sec	APTT sec	Aniso- cytosis	Micro- cytosis	Macro- cytosis	Hypo- chromasia	Hyper- chromasia	
3M	11	1246	21.9	18.0	-	-	-	-	-	
	12	779	23.2	16.2	-	-	-	-	-	
	13	NVR	20.2	17.2	-	-	-	-	-	
	14	1068	21.2	16.1	-	-	-	-	-	
	16	1055	21.4	16.4	-	-	-	-	-	
	17	1010	23.2	17.4	-	-	-	-	-	
	18	930	21.5	15.4	-	-	-	-	-	
	19	1359	21.8	14.2	-	-	-	-	-	
	20	1113	22.5	16.8	-	-	-	-	-	
	42	1169	22.5	16.3	-	-	-	-	-	
4M	31	1009	22.3	17.3	-	-	-	-	-	
	32	902	22.8	16.0	-	-	-	-	-	
	33	1126	21.5	16.2	-	-	-	-	-	
	34	876	20.8	15.2	-	-	-	-	-	
	35	957	21.8	15.2	-	-	-	-	-	
	36	NVR	22.1	17.8	-	-	-	-	-	
	37	1080	22.9	16.4	-	-	-	-	-	
	38	850	22.6	14.7	-	-	-	-	-	
	39	915	23.6	16.2	-	-	-	-	-	
	40	930	22.5	18.1	-	-	-	-	-	



APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control	Lactase, batch PPL34537						
		1 0	2 10	3 33	4 100				
Group /Sex	Animal Number	Hct L/L	Hb g/dL	RBC x10 <sup>12</sup> /L	Retic x10 <sup>12</sup> /L	MCH pg	MCHC g/dL	MCV fL	RDW %
1F	131	0.465	15.5	8.92	0.133	17.4	33.4	52.1	11.1
	132	0.429	14.5	7.61	0.159	19.1	33.9	56.4	12.9
	133	0.393	13.5	7.19	0.159	18.7	34.3	54.6	11.5
	134	0.436	14.7	7.95	0.195	18.5	33.8	54.8	11.7
	135	0.431	14.6	7.82	0.138	18.7	33.8	55.1	11.2
	136	0.445	15.1	7.99	0.217	18.9	33.8	55.7	11.7
	137	0.443	15.1	8.29	0.163	18.2	34.0	53.5	11.3
	138	0.447	15.2	8.08	0.168	18.8	34.0	55.3	12.0
	139	0.458	15.4	7.79	0.167	19.7	33.6	58.8	11.3
	140	0.441	15.1	7.77	0.107	19.4	34.3	56.7	12.0
2F	121	0.456	15.2	8.37	0.173	18.2	33.4	54.5	10.6
	122	0.442	15.1	8.09	0.140	18.6	34.1	54.6	11.8
	123	0.458	15.5	8.47	0.205	18.3	33.8	54.1	12.1
	124	0.457	15.8	8.47	0.147	18.7	34.7	53.9	11.4
	125	0.438	15.0	7.88	0.208	19.1	34.3	55.6	12.7
	126	0.450	15.3	7.96	0.229	19.3	34.1	56.5	11.6
	127	0.437	15.1	7.81	0.141	19.3	34.6	56.0	11.9
	128	0.476	15.9	8.70	0.176	18.3	33.4	54.7	11.9
	129	0.427	14.6	8.19	0.088	17.8	34.2	52.2	11.5
	130	0.462	15.6	8.62	0.139	18.1	33.8	53.6	10.9

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2 10    3 33    4 100					
Group /Sex	Animal Number	WBC x10 <sup>9</sup> /L	N x10 <sup>9</sup> /L	L x10 <sup>9</sup> /L	E x10 <sup>9</sup> /L	B x10 <sup>9</sup> /L	M x10 <sup>9</sup> /L	LUC x10 <sup>9</sup> /L
1F	131	11.41	2.00	8.72	0.26	0.03	0.29	0.10
	132	6.67	0.43	5.99	0.05	0.02	0.11	0.07
	133	4.28	0.90	3.12	0.08	0.02	0.13	0.03
	134	8.45	1.76	6.27	0.18	0.02	0.16	0.06
	135	5.88	0.62	4.69	0.10	0.02	0.35	0.11
	136	7.74	1.09	6.27	0.15	0.03	0.16	0.03
	137	7.30	0.64	6.32	0.13	0.02	0.15	0.05
	138	6.94	1.78	4.63	0.06	0.07	0.36	0.04
	139	11.42	0.68	10.32	0.13	0.06	0.16	0.08
	140	7.86	0.32	7.05	0.11	0.04	0.23	0.12
2F	121	11.85	1.30	9.86	0.21	0.07	0.24	0.18
	122	8.32	0.73	6.98	0.18	0.03	0.27	0.13
	123	6.27	0.44	5.49	0.09	0.05	0.14	0.06
	124	8.64	0.53	7.60	0.22	0.10	0.11	0.08
	125	11.02	0.73	9.82	0.09	0.04	0.17	0.17
	126	8.94	0.76	7.70	0.11	0.04	0.24	0.09
	127	7.07	0.66	5.89	0.09	0.05	0.22	0.16
	128	10.31	1.33	8.34	0.10	0.08	0.33	0.13
	129	10.93	0.46	10.12	0.08	0.05	0.13	0.10
	130	8.09	0.62	6.95	0.06	0.08	0.31	0.07

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2      3      4 10     33     100						
Group /Sex	Animal Number	Plt x10 <sup>9</sup> /L	PT sec	APTT sec	Aniso- cytosis	Micro- cytosis	Macro- cytosis	Hypo- chromasia	Hyper- chromasia
1F	131	1168	INS	17.2	-	-	-	-	-
	132	541	23.8	15.6	-	-	-	-	-
	133	907	20.9	13.0	-	-	-	-	-
	134	1010	24.3	15.1	-	-	-	-	-
	135	865	21.5	11.6	-	-	-	-	-
	136	961	24.5	16.9	-	-	-	-	-
	137	959	23.0	14.1	-	-	-	-	-
	138	962	23.6	16.6	-	-	-	-	-
	139	909	24.3	16.0	-	-	-	-	-
	140	1065	26.2	16.6	-	-	-	-	-
2F	121	1090	22.1	16.8	-	-	-	-	-
	122	1054	23.2	15.7	-	-	-	-	-
	123	1046	24.4	17.2	-	-	-	-	-
	124	1248	23.1	14.3	-	-	-	-	-
	125	943	23.5	17.4	-	-	-	-	-
	126	1131	22.7	16.1	-	-	-	-	-
	127	1058	23.1	16.3	-	-	-	-	-
	128	1052	23.1	16.1	-	-	-	-	-
	129	1031	25.2	16.4	-	-	-	-	-
	130	1037	23.9	15.7	-	-	-	-	-

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2      3      4 10    33    100							
Group /Sex	Animal Number	Hct L/L	Hb g/dL	RBC x10 <sup>12</sup> /L	Retic x10 <sup>12</sup> /L	MCH pg	MCHC g/dL	MCV fL	RDW %	
3F	101	0.443	15.1	7.62	0.178	19.8	34.0	58.2	11.1	
	102	0.434	14.7	8.10	0.168	18.2	33.9	53.6	12.7	
	103	0.458	15.2	8.00	0.174	19.0	33.3	57.2	11.5	
	105	0.458	15.2	8.13	0.239	18.7	33.2	56.3	11.7	
	106	0.448	15.1	7.99	0.155	18.9	33.7	56.1	11.4	
	107	0.433	14.6	7.92	0.169	18.4	33.7	54.7	11.1	
	108	0.418	14.3	7.60	0.160	18.8	34.2	55.0	11.6	
	109	0.432	14.9	7.98	0.209	18.7	34.4	54.2	11.7	
	110	0.438	14.9	8.24	0.176	18.0	34.0	53.1	11.1	
	141	0.446	15.1	8.04	0.156	18.8	33.9	55.6	11.8	
4F	111	0.436	14.6	8.13	0.153	18.0	33.6	53.6	12.4	
	112	0.449	15.4	8.04	0.192	19.1	34.2	55.9	11.4	
	113	0.449	15.2	8.19	0.179	18.6	33.9	54.9	12.7	
	114	0.452	15.2	8.37	0.154	18.1	33.6	54.0	11.3	
	115	0.429	14.0	7.69	0.215	18.2	32.6	55.8	11.9	
	116	0.443	15.0	8.04	0.202	18.7	33.9	55.1	11.6	
	117	0.453	15.3	8.14	0.141	18.8	33.8	55.7	11.4	
	118	0.434	14.8	7.87	0.159	18.8	34.0	55.2	11.8	
	119	0.449	15.3	8.11	0.182	18.9	34.1	55.3	11.6	
	120	0.413	13.8	7.73	0.192	17.8	33.4	53.4	11.7	

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2 10    3 33    4 100					
Group /Sex	Animal Number	WBC x10 <sup>9</sup> /L	N x10 <sup>9</sup> /L	L x10 <sup>9</sup> /L	E x10 <sup>9</sup> /L	B x10 <sup>9</sup> /L	M x10 <sup>9</sup> /L	LUC x10 <sup>9</sup> /L
3F	101	9.83	2.59	6.72	0.07	0.07	0.33	0.06
	102	6.28	0.57	5.34	0.14	0.04	0.15	0.04
	103	7.05	0.62	5.83	0.12	0.04	0.31	0.13
	105	7.77	0.87	6.38	0.10	0.04	0.30	0.08
	106	8.12	0.65	6.90	0.21	0.06	0.24	0.06
	107	5.66	0.71	4.58	0.13	0.02	0.19	0.03
	108	4.91	0.46	4.13	0.08	0.03	0.17	0.03
	109	4.21	0.52	3.35	0.10	0.03	0.19	0.02
	110	8.40	0.50	7.43	0.06	0.06	0.28	0.07
	141	7.61	0.73	6.42	0.08	0.04	0.22	0.11
4F	111	6.32	0.64	5.38	0.10	0.02	0.13	0.04
	112	10.17	2.50	7.01	0.28	0.05	0.26	0.07
	113	3.89	0.46	3.14	0.09	0.02	0.15	0.04
	114	7.11	0.42	6.37	0.14	0.06	0.08	0.06
	115	6.95	0.65	5.76	0.12	0.03	0.32	0.07
	116	11.14	1.35	9.13	0.24	0.09	0.21	0.11
	117	7.04	1.06	5.62	0.15	0.01	0.15	0.04
	118	5.80	0.50	5.04	0.07	0.03	0.11	0.04
	119	10.72	0.89	9.26	0.16	0.11	0.21	0.11
	120	11.97	3.51	7.78	0.19	0.05	0.36	0.08

APPENDIX 7 (cont) Haematology - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2      3      4 10    33    100							
Group /Sex	Animal Number	Plt x10 <sup>9</sup> /L	PT sec	APTT sec	Aniso- cytosis	Micro- cytosis	Macro- cytosis	Hypo- chromasia	Hyper- chromasia	
3F	101	768	23.0	14.3	-	-	-	-	-	
	102	885	24.4	14.6	-	-	-	-	-	
	103	1003	23.2	15.8	-	-	-	-	-	
	105	945	22.9	14.6	-	-	-	-	-	
	106	859	23.6	15.2	-	-	-	-	-	
	107	1064	23.2	14.2	-	-	-	-	-	
	108	806	22.7	15.6	-	-	-	-	-	
	109	1059	23.8	13.9	-	-	-	-	-	
	110	988	20.7	10.3	-	-	-	-	-	
	141	1028	22.2	16.4	-	-	-	-	-	
4F	111	948	24.6	14.8	-	-	-	-	-	
	112	NVR	24.4	15.4	-	-	-	-	-	
	113	967	25.1	14.3	-	-	-	-	-	
	114	1150	24.6	12.3	-	-	-	-	-	
	115	1231	23.0	12.0	-	-	-	-	-	
	116	1058	23.1	13.2	-	-	-	-	-	
	117	785	23.6	13.3	-	-	-	-	-	
	118	946	24.3	14.7	-	-	-	-	-	
	119	888	24.7	16.3	-	-	-	-	-	
	120	1084	23.6	12.6	-	-	-	-	-	

APPENDIX 8 Blood chemistry - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2 10    3 33    4 100					
Group /Sex	Animal Number	ALP U/L	ALT U/L	AST U/L	Urea mmol/L	Creat μmol/L	Gluc mmol/L	Chol mmol/L
1M	1	92	33	77	5.86	47	7.54	1.75
	2	79	48	83	5.85	40	7.45	0.78
	3	62	35	71	5.04	40	9.49	1.74
	4	66	30	63	4.72	30	7.43	1.64
	5	54	37	88	6.75	29	8.20	1.71
	6	79	34	79	4.62	34	7.03	1.28
	7	90	40	82	4.34	33	10.63	2.28
	8	85	39	76	5.55	41	8.49	1.28
	9	72	38	73	5.38	40	9.48	1.82
	10	63	48	117	5.09	29	9.29	1.32
2M	21	82	50	117	6.07	40	5.64	1.73
	22	87	31	69	4.86	34	7.16	1.70
	23	82	42	78	5.06	31	6.63	1.69
	24	69	42	86	5.54	35	7.02	1.28
	25	73	82	169	6.11	33	9.46	1.68
	26	95	29	58	4.55	28	6.66	1.43
	27	90	41	77	4.37	30	7.87	1.61
	28	53	30	71	5.10	30	9.52	1.54
	29	113	56	74	5.98	39	9.43	1.52
	30	72	26	55	4.39	30	7.80	1.80

APPENDIX 8 (cont) Blood chemistry - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control	Lactase, batch PPL34537			
		1	2	3	4	
Group /Sex	Animal Number	0	10	33	100	
		Na mmol/L	K mmol/L	Total Prot g/L	Alb g/L	A/G Ratio
1M	1	141	5.8	66	36	1.20
	2	142	5.6	65	36	1.24
	3	139	6.1	66	35	1.13
	4	140	5.2	62	31	1.00
	5	139	5.8	65	34	1.10
	6	142	5.1	65	37	1.32
	7	140	6.4	68	38	1.27
	8	140	5.5	60	35	1.40
	9	139	5.5	64	34	1.13
	10	140	6.6	59	33	1.27
2M	21	142	5.9	61	36	1.44
	22	139	6.6	59	34	1.36
	23	138	5.8	64	34	1.13
	24	141	6.0	62	34	1.21
	25	136	6.9	63	33	1.10
	26	139	7.0	67	36	1.16
	27	141	5.0	61	36	1.44
	28	139	6.2	58	33	1.32
	29	139	5.9	67	37	1.23
	30	141	6.2	60	34	1.31



APPENDIX 8 (cont) Blood chemistry - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control 1 0	Lactase, batch PPL34537 2 10    3 33    4 100					
Group /Sex	Animal Number	ALP U/L	ALT U/L	AST U/L	Urea mmol/L	Creat μmol/L	Gluc mmol/L	Chol mmol/L
3M	11	101	31	79	5.82	35	6.31	1.56
	12	113	40	100	5.07	26	6.78	1.23
	13	91	67	134	5.48	33	8.85	1.92
	14	101	56	81	4.90	38	10.34	1.49
	16	91	37	82	4.36	30	6.98	1.83
	17	62	42	79	4.87	34	6.50	2.14
	18	67	38	85	4.41	36	7.67	2.47
	19	126	43	90	5.69	34	7.13	1.98
	20	66	25	71	4.29	26	9.72	2.22
	42	108	45	85	4.23	29	8.59	1.63
4M	31	86	47	92	4.35	23	5.33	1.34
	32	79	32	77	4.26	28	6.25	1.80
	33	115	40	79	4.26	31	7.42	1.19
	34	70	34	101	4.22	28	8.42	1.50
	35	121	39	110	4.09	25	9.92	2.14
	36	81	39	71	4.92	33	6.74	1.73
	37	79	41	72	5.92	30	8.14	1.65
	38	86	34	76	4.72	31	8.82	1.70
	39	119	45	82	5.79	36	8.50	1.38
	40	87	38	68	4.31	35	10.28	1.20

APPENDIX 8 (cont) Blood chemistry - individual values during Week 13 of treatment

Dose Group		Control	Lactase, batch PPL34537			
Dose (% of Toxbatch)		1	2	3	4	
		0	10	33	100	
Group /Sex	Animal Number	Na mmol/L	K mmol/L	Total Prot g/L	Alb g/L	A/G Ratio
3M	11	141	5.3	64	36	1.29
	12	140	5.5	63	34	1.17
	13	NVR	NVR	66	36	1.20
	14	140	5.8	63	36	1.33
	16	138	5.7	67	36	1.16
	17	139	5.8	66	34	1.06
	18	140	6.1	65	33	1.03
	19	139	6.2	61	35	1.35
	20	138	6.8	64	36	1.29
	42	139	5.6	64	34	1.13
4M	31	136	9.4	64	35	1.21
	32	141	5.8	68	35	1.06
	33	138	5.6	65	34	1.10
	34	138	6.8	64	33	1.06
	35	139	6.6	63	35	1.25
	36	138	8.6	71	37	1.09
	37	139	6.3	65	35	1.17
	38	140	5.7	64	35	1.21
	39	139	5.4	65	33	1.03
	40	139	6.4	63	35	1.25

APPENDIX 8 (cont) Blood chemistry - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control	Lactase, batch PPL34537					
		1 0	2 10	3 33	4 100			
Group /Sex	Animal Number	ALP U/L	ALT U/L	AST U/L	Urea mmol/L	Creat μmol/L	Gluc mmol/L	Chol mmol/L
1F	131	31	31	71	5.19	38	6.10	2.44
	132	31	37	70	6.88	41	6.69	2.22
	133	28	58	293	4.43	33	10.31	2.48
	134	47	30	51	6.70	35	6.79	2.54
	135	28	72	116	5.51	45	8.17	2.74
	136	55	22	63	5.15	43	6.46	1.60
	137	33	30	64	5.54	34	6.65	2.51
	138	29	28	80	6.39	40	5.98	2.37
	139	31	31	56	5.96	36	7.36	1.49
	140	35	32	67	6.28	41	6.33	1.37
2F	121	36	25	57	5.21	35	5.84	2.85
	122	35	27	61	5.12	38	6.55	2.23
	123	36	34	59	5.83	38	6.31	2.13
	124	50	39	95	6.18	37	8.07	2.76
	125	44	32	70	4.79	35	7.08	2.38
	126	28	29	68	6.99	40	6.14	1.94
	127	43	32	65	5.28	37	6.72	2.36
	128	42	27	58	5.60	36	5.92	2.18
	129	28	27	70	5.43	36	6.42	1.63
	130	53	31	61	5.85	40	8.09	2.36

APPENDIX 8 (cont) Blood chemistry - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control	Lactase, batch PPL34537			
		1 0	2 10	3 33	4 100	
Group /Sex	Animal Number	Na mmol/L	K mmol/L	Total Prot g/L	Alb g/L	A/G Ratio
1F	131	142	3.3	74	40	1.18
	132	140	4.5	72	41	1.32
	133	139	3.8	72	43	1.48
	134	139	3.8	72	38	1.12
	135	140	3.8	75	45	1.50
	136	141	3.9	68	40	1.43
	137	142	4.5	67	41	1.58
	138	143	4.2	70	40	1.33
	139	140	4.2	66	37	1.28
	140	140	4.3	64	37	1.37
2F	121	140	4.6	74	39	1.11
	122	142	4.4	75	45	1.50
	123	141	4.1	71	42	1.45
	124	137	4.8	72	41	1.32
	125	140	4.0	67	39	1.39
	126	141	4.6	69	43	1.65
	127	140	4.2	68	38	1.27
	128	141	4.2	68	40	1.43
	129	140	4.3	60	38	1.73
	130	140	4.5	69	42	1.56

APPENDIX 8 (cont) Blood chemistry - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control	Lactase, batch PPL34537					
		1 0	2 10	3 33	4 100			
Group /Sex	Animal Number	ALP U/L	ALT U/L	AST U/L	Urea mmol/L	Creat μmol/L	Gluc mmol/L	Chol mmol/L
3F	101	28	29	74	5.06	37	6.66	1.91
	102	43	25	75	7.11	41	6.17	1.63
	103	35	23	60	5.80	38	7.25	2.00
	105	48	24	60	6.02	39	6.56	3.00
	106	38	26	48	6.31	46	6.40	2.58
	107	37	30	82	5.56	42	7.90	1.70
	108	23	41	81	4.81	43	7.53	1.96
	109	41	32	77	5.24	43	7.38	2.66
	110	40	32	64	4.77	41	7.59	2.27
	141	65	26	66	6.44	40	7.15	2.43
4F	111	60	26	69	6.38	37	6.15	1.62
	112	39	24	67	5.32	37	5.84	2.43
	113	59	25	71	5.90	36	5.93	2.15
	114	39	25	59	6.14	38	6.73	1.70
	115	41	26	51	5.26	36	7.81	2.26
	116	63	38	80	5.10	33	7.23	2.00
	117	51	35	73	6.90	43	6.28	2.14
	118	56	29	87	6.87	44	6.95	1.65
	119	53	29	71	6.33	45	7.13	2.37
	120	63	42	101	5.03	32	6.83	2.09

APPENDIX 8 (cont) Blood chemistry - individual values during Week 13 of treatment

Dose Group Dose (% of Toxbatch)		Control	Lactase, batch PPL34537			
		1 0	2 10	3 33	4 100	
Group /Sex	Animal Number	Na mmol/L	K mmol/L	Total Prot g/L	Alb g/L	A/G Ratio
3F	101	138	4.8	69	38	1.23
	102	139	5.6	63	38	1.52
	103	140	4.0	68	40	1.43
	105	140	3.6	70	41	1.41
	106	139	5.4	74	43	1.39
	107	142	4.0	70	43	1.59
	108	140	4.2	74	45	1.55
	109	141	4.1	69	41	1.46
	110	140	4.2	67	41	1.58
	141	141	3.9	68	39	1.34
4F	111	142	4.1	65	41	1.71
	112	142	3.9	71	42	1.45
	113	139	4.6	67	37	1.23
	114	139	4.7	63	38	1.52
	115	141	4.0	73	45	1.61
	116	141	4.2	71	43	1.54
	117	140	4.3	70	40	1.33
	118	138	4.3	70	43	1.59
	119	139	4.8	68	41	1.52
	120	139	4.1	69	34	0.97

APPENDIX 9 Organ weights - individual absolute values (g) for animals killed after 13 weeks of treatment

Request ID: 5031941

Dose Group	Control	Lactase, batch PPL34537			
Dose (% of Toxbatch)	1	2	3	4	
	0	10	33	100	

Group /Sex	Animal Number	Terminal Body weight	Adrenals	Brain	Epididymides	Heart	Kidneys	Liver	Spleen	Testes	Thymus
1M	0001	639	0.066	2.216	1.330	1.727	3.659	22.895	0.954	3.361	0.254
	0002	469	0.044	2.171	1.003	1.287	2.764	16.941	0.850	2.931	0.237
	0003	594	0.079	2.407	1.384	1.897	3.514	17.753	0.949	3.995	0.253
	0004	474	0.057	2.166	1.136	1.465	2.831	13.633	0.872	3.741	0.240
	0005	600	0.051	2.221	1.545	1.863	3.511	21.098	0.930	3.662	0.246
	0006	632	0.048	2.334	1.620	1.652	3.962	24.587	1.085	4.532	0.247
	0007	688	0.053	2.208	1.456	1.823	3.823	28.421	0.901	4.127	0.296
	0008	524	0.049	2.272	1.396	1.408	3.246	18.490	0.853	4.093	0.224
	0009	635	0.072	2.540	1.406	1.796	3.594	22.798	1.001	3.948	0.295
	0010	573	0.050	2.318	1.610	1.588	3.541	17.008	0.822	4.107	0.231
2M	0021	462	0.050	2.189	1.528	1.263	2.708	14.439	0.723	3.417	0.150
	0022	582	0.055	2.235	1.455	1.610	3.494	18.855	0.844	3.710	0.218
	0023	507	0.045	2.189	1.338	1.318	3.170	17.993	0.883	4.066	0.306
	0024	572	0.078	2.173	1.304	1.651	3.484	20.919	0.757	3.374	0.206
	0025	590	0.054	2.382	1.363	1.655	3.637	20.155	1.008	3.967	0.250
	0026	593	0.066	2.255	1.451	1.677	3.760	17.654	0.813	4.068	0.176
	0027	541	0.057	2.207	1.364	1.560	3.319	16.819	0.959	4.021	0.320
	0028	550	0.048	2.198	1.258	1.554	3.229	15.114	0.781	3.679	0.278
	0029	593	0.052	2.321	1.419	1.493	3.326	19.527	0.870	4.184	0.277
	0030	542	0.064	2.124	1.362	1.389	3.298	16.835	0.827	3.532	0.172

APPENDIX 9 (cont) Organ weights - individual absolute values (g) for animals killed after 13 weeks of treatment

Request ID: 5031941

Dose Group	Control	Lactase, batch PPL34537			
Dose (% of Toxbatch)	1	2	3	4	
	0	10	33	100	

Group /Sex	Animal Number	Terminal Body weight	Adrenals	Brain	Epididymides	Heart	Kidneys	Liver	Spleen	Testes	Thymus
3M	0011	486	0.055	2.158	1.373	1.483	3.006	16.078	0.704	3.844	0.277
	0012	566	0.062	2.202	1.431	1.630	3.450	17.962	1.068	3.743	0.199
	0013	604	0.081	2.219	1.395	1.719	3.562	21.496	0.946	3.441	0.284
	0014	433	0.054	2.097	1.265	1.167	2.633	15.533	0.679	4.229	0.179
	0016	502	0.061	2.108	1.172	1.549	3.465	17.600	0.961	3.756	0.284
	0017	540	0.056	2.162	1.563	1.495	3.092	17.617	0.936	3.757	0.393
	0018	670	0.065	2.142	1.491	1.832	3.911	19.811	0.924	3.882	0.287
	0019	530	0.040	2.257	1.393	1.360	3.212	16.701	0.701	3.913	0.270
	0020	593	0.059	2.146	1.415	1.792	3.706	22.636	1.514	3.685	0.220
	0042	495	0.054	2.100	1.467	1.570	3.522	18.139	1.124	4.147	0.262
4M	0031	525	0.074	2.115	1.208	1.417	3.262	17.539	0.745	3.216	0.287
	0032	560	0.064	2.298	1.274	1.653	3.688	18.975	0.849	3.415	0.257
	0033	586	0.069	2.263	1.492	1.700	3.770	18.837	0.785	3.330	0.234
	0034	635	0.072	2.134	1.393	1.542	3.620	20.967	1.053	3.770	0.321
	0035	557	0.050	2.100	1.387	1.536	3.591	23.955	0.935	3.787	0.147
	0036	592	0.060	2.244	1.596	1.576	3.325	19.459	0.910	4.264	0.080
	0037	527	0.059	2.234	1.400	1.448	3.202	16.399	0.821	3.734	0.195
	0038	485	0.060	2.202	1.487	1.366	3.060	17.753	0.855	4.137	0.192
	0039	587	0.066	2.292	1.343	1.548	3.627	19.395	1.381	3.313	0.336
	0040	502	0.052	2.182	1.447	1.328	2.853	14.818	0.658	4.141	0.182



APPENDIX 9 (cont) Organ weights - individual absolute values (g) for animals killed after 13 weeks of treatment

Request ID: 5031941

Dose Group	Control	Lactase, batch PPL34537			
Dose (% of Toxbatch)	1	2	3	4	
	0	10	33	100	

Group /Sex	Animal Number	Terminal Body weight	Adrenals	Brain	Heart	Kidneys	Liver	Ovaries	Spleen	Thymus	Uterus and Cervix
1F	0131	273	0.056	1.968	0.839	1.752	10.371	0.087	0.537	0.234	0.498
	0132	299	0.067	1.995	0.914	1.827	11.827	0.069	0.533	0.128	1.515
	0133	343	0.068	1.920	1.109	2.240	10.877	0.058	0.619	0.188	1.112
	0134	303	0.070	2.040	0.912	1.918	10.213	0.091	0.554	0.204	0.583
	0135	346	0.069	2.185	1.325	2.425	13.153	0.066	0.570	0.209	0.771
	0136	289	0.049	1.833	1.001	1.970	9.442	0.087	0.553	0.155	0.966
	0137	290	0.061	1.938	0.948	2.152	10.066	0.100	0.535	0.240	0.518
	0138	295	0.054	1.976	0.954	1.894	10.062	0.090	0.690	0.168	0.781
	0139	361	0.066	2.130	1.032	2.342	12.521	0.145	0.640	0.281	0.770
	0140	318	0.067	2.025	0.982	2.012	9.405	0.104	0.489	0.134	0.527
2F	0121	296	0.053	2.081	0.965	1.868	10.424	0.122	0.676	0.289	0.664
	0122	306	0.063	1.861	0.960	2.025	10.498	0.085	0.620	0.214	0.585
	0123	297	0.065	1.989	0.931	1.938	10.500	0.086	0.596	0.213	0.591
	0124	285	0.058	1.958	0.945	0.911	10.877	0.100	0.649	0.298	0.440
	0125	283	0.050	2.018	0.903	1.768	9.757	0.091	0.617	0.222	0.959
	0126	275	0.059	1.993	0.923	1.849	8.742	0.068	0.671	0.303	0.678
	0127	289	0.079	1.890	0.944	1.869	9.231	0.063	0.449	0.104	1.403
	0128	305	0.065	2.091	1.039	2.030	9.881	0.089	0.646	0.205	0.633
	0129	288	0.066	1.991	0.930	1.930	8.750	0.073	0.501	0.181	0.996
	0130	290	0.062	2.024	0.974	1.879	9.462	0.067	0.573	0.270	0.831

APPENDIX 9 (cont) Organ weights - individual absolute values (g) for animals killed after 13 weeks of treatment

Request ID: 5031941

Dose Group	Control	Lactase, batch PPL34537			
Dose (% of Toxbatch)	1	2	3	4	
	0	10	33	100	

Group /Sex	Animal Number	Terminal Body weight	Adrenals	Brain	Heart	Kidneys	Liver	Ovaries	Spleen	Thymus	Uterus and Cervix
3F	0101	323	0.062	2.031	1.090	1.979	11.333	0.098	0.640	0.246	0.535
	0102	304	0.069	1.877	0.929	1.885	9.379	0.095	0.541	0.275	0.593
	0103	298	0.057	1.987	0.947	1.594	9.149	0.089	0.462	0.185	0.623
	0105	313	0.059	2.035	1.012	1.986	11.441	0.094	0.817	0.265	1.382
	0106	335	0.074	2.060	1.070	1.982	10.098	0.097	0.657	0.254	0.628
	0107	333	0.063	2.076	1.073	2.114	10.139	0.091	0.653	0.216	0.703
	0108	287	0.058	1.977	0.971	1.794	8.540	0.070	0.615	0.210	0.744
	0109	330	0.067	2.057	1.013	1.841	9.253	0.075	0.582	0.273	1.454
	0110	324	0.065	2.171	1.055	2.042	11.021	0.084	0.670	0.265	0.931
	0141	301	0.061	1.963	0.955	1.701	9.872	0.082	0.458	0.157	1.220
4F	0111	294	0.055	2.124	0.867	1.838	10.395	0.078	0.510	0.182	0.600
	0112	322	0.055	2.065	0.961	2.001	12.586	0.071	0.725	0.223	0.909
	0113	315	0.078	2.060	1.057	2.310	11.283	0.098	0.522	0.253	0.579
	0114	284	0.059	2.029	0.932	2.091	11.137	0.093	0.570	0.203	0.468
	0115	331	0.069	2.028	1.045	2.140	12.850	0.087	0.630	0.211	0.854
	0116	334	0.060	2.124	0.977	2.251	10.523	0.102	0.662	0.363	0.652
	0117	315	0.053	1.961	0.927	1.872	10.117	0.079	0.588	0.148	0.450
	0118	290	0.061	1.867	1.003	1.875	8.841	0.065	0.487	0.230	1.023
	0119	294	0.062	2.014	1.119	1.916	9.198	0.072	0.561	0.269	0.596
	0120	276	0.054	2.100	0.873	2.077	10.417	0.096	0.687	0.204	0.402

APPENDIX 10 Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0001	92 (14)	Treatment	Adrenals: No macropathology findings	Adrenals: Vacuolation, Cortical , slight, bilateral
				Kidneys: No macropathology findings	Kidneys: Mineralization, Papilla , minimal, unilateral
				Liver: Strangulated, right posterior lobe	= Liver: Necrosis, Torsion, Lobar , present Liver: Aggregates, Lymphoid/Macrophage, slight
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Erythrocytosis/Erythrophagocytosis, Sinuses, minimal Lymph Node, Mandibular: Cellularity Increased, Follicles, moderate

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0002	92 (14)	Treatment	Kidneys: No macropathology findings Lymph Node, Mandibular: No macropathology findings	Kidneys: Basophilia, Tubular , minimal, focal Lymph Node, Mandibular: Cellularity Increased, Follicles, slight

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0003	92 (14)	Treatment	Heart: No macropathology findings	Heart: Infiltration, Inflammatory Cells, Myocardial , slight
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, slight
				Lungs and Bronchi: No macropathology findings	Lungs and Bronchi: Infiltration, Inflammatory Cells, Perivascular, slight
				Prostate: No macropathology findings	Prostate: Aggregates, Lymphoid, slight

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Ileum; Jejunum; Kidneys; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0004	92 (14)	Treatment	Kidneys: No macropathology findings	Kidneys: Basophilia, Tubular , slight, focal Kidneys: Infiltration, Inflammatory Cells, Interstitial , slight
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal
				Lymph Node, Mandibular: Enlarged, 10-19mm	= Lymph Node, Mandibular: Plasmacytosis, moderate Lymph Node, Mandibular: Cellularity Increased, Paracortex, moderate Lymph Node, Mandibular: Cellularity Increased, Follicles, slight
				Parathyroids: No macropathology findings	Parathyroids: both, tissue missing

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0005	92 (14)	Treatment	Kidneys: No macropathology findings	Kidneys: Basophilia, Tubular , minimal, focal
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal
				Lungs and Bronchi: No macropathology findings	Lungs and Bronchi: Alveolar Macrophages, Increased, Focal , minimal Lungs and Bronchi: Infiltration, Inflammatory Cells, Perivascular, slight Lungs and Bronchi: Inflammation, Alveoli , minimal
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, moderate Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight Lymph Node, Mandibular: Cellularity Increased, Follicles, slight
				Spleen: No macropathology findings	Spleen: Extramedullary Hemopoiesis, Increased , minimal

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0005	92 (14)	Treatment	Trachea: No macropathology findings	Trachea: Foci, Inflammatory Cells, slight

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Stomach; Testes; Thymus; Thyroids; Urinary Bladder



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0006	92 (14)	Treatment	All tissues: No macropathology findings	All examined tissues: No histopathology findings

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0007	92 (14)	Treatment	Lungs and Bronchi: No macropathology findings	Lungs and Bronchi: Alveolar Macrophages, Pigment , minimal Lungs and Bronchi: Inflammation, Alveoli , minimal
				Pancreas: No macropathology findings	Pancreas: Fibrosis, slight, focal Pancreas: Foci, Inflammatory Cells, minimal, focal
				Trachea: No macropathology findings	Trachea: Foci, Inflammatory Cells, slight

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0008	92 (14)	Treatment	Kidneys: No macropathology findings Lymph Node, Mandibular: No macropathology findings	Kidneys: Basophilia, Tubular , minimal, focal Lymph Node, Mandibular: Erythrocytosis/Erythrophagocytosis, Sinuses, minimal

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0009	92 (14)	Treatment	Adrenals: No macropathology findings	Adrenals: Vacuolation, Cortical , slight, bilateral
				Kidneys: No macropathology findings	Kidneys: Basophilia, Tubular , minimal, focal
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, slight Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight Lymph Node, Mandibular: Cellularity Increased, Follicles, moderate
				Prostate: No macropathology findings	Prostate: Aggregates, Lymphoid, minimal, focal

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1M	0010	92 (14)	Treatment	Lungs and Bronchi: No macropathology findings  Spleen: No macropathology findings  Stomach: Mass(es), limiting ridge, firm, pale, 2-9mm	Lungs and Bronchi: Mineralization, Alveoli , minimal  Lymph Node, Axillary Lt: Cellularity Increased, Paracortex, slight  Spleen: Extramedullary Hemopoiesis, Increased , minimal  = Stomach: Cyst(s), Squamous, Limiting Ridge , moderate

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0021	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0022	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0023	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0024	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0025	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0026	92 (14)	Treatment	Lungs and Bronchi: Dark area(s), multilobular, 2-9mm, 5+ (many)	= Lungs and Bronchi: Alveolar Macrophages, Pigment , minimal

Tissues without comments under macropathology findings were within normal limits at necropsy.

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0027	92 (14)	Treatment	Lungs and Bronchi: Pale area(s), left, $\leq 1$ mm, 2-5 (few) =	Lungs and Bronchi: Alveolar Macrophages, Foamy , slight

Tissues without comments under macropathology findings were within normal limits at necropsy.

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0028	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0029	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2M	0030	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0011	92 (14)	Treatment	Lungs and Bronchi: Pale area(s), multilobular, <=1mm, 5+ (many)	Examined, not correlated

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Lungs and Bronchi



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0012	92 (14)	Treatment	<p>Lungs and Bronchi: Dark area(s), multilobular, 2-9mm, 5+ (many)</p> <p>Lungs and Bronchi: Dark area(s), multilobular, 2-9mm, 5+ (many)</p> <p>Lungs and Bronchi: Dark area(s), multilobular, 2-9mm, 5+ (many)</p> <p>Thoracic Cavity: Adhesion(s), fibrous, pale, 5+ (many), (between right middle and cranial lung lobes and rib-cage.)</p>	<p>= Lungs and Bronchi: Alveolar Macrophages, Increased, Focal , slight</p> <p>= Lungs and Bronchi: Inflammation, Pleura , slight</p> <p>= Lungs and Bronchi: Mineralization, Alveoli , minimal</p> <p>Lungs and Bronchi: Infiltration, Inflammatory Cells, Perivascular, minimal</p> <p>Lungs and Bronchi: Inflammation, Alveoli , minimal</p> <p>= Thoracic Cavity: Adhesions, present</p>

Tissues without comments under macropathology findings were within normal limits at necropsy.

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0013	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0014	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0016	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0017	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0018	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0019	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0020	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3M	0042	92 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0031	92 (14)	Treatment	Adrenals: No macropathology findings	Adrenals: Vacuolation, Cortical , slight, bilateral
				Liver: No macropathology findings	Liver: Fibrosis, slight, focal
				Lungs and Bronchi: No macropathology findings	Lungs and Bronchi: Inflammation, Alveoli , minimal
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Cellularity Increased, Follicles, slight

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0032	92 (14)	Treatment	Kidneys: No macropathology findings	Kidneys: Scars, Cortical , slight, focal
				Lungs and Bronchi: No macropathology findings	Lungs and Bronchi: Mineralization, Alveoli , minimal
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, moderate Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Liver; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0033	92 (14)	Treatment	Heart: No macropathology findings	Heart: Infiltration, Inflammatory Cells, Myocardial , minimal
				Kidneys: No macropathology findings	Kidneys: Basophilia, Tubular , minimal, focal Kidneys: Infiltration, Inflammatory Cells, Interstitial , minimal
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal  Lymph Node, Axillary Lt: Cellularity Increased, Paracortex, slight
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, slight Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight Lymph Node, Mandibular: Cellularity Increased, Follicles, slight
				Rectum: No macropathology findings	Rectum: Parasites , present

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0033	92 (14)	Treatment	Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments: Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Ileum; Jejunum; Lungs and Bronchi; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder	

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0034	92 (14)	Treatment	Adrenals: No macropathology findings	Adrenals: Vacuolation, Cortical , minimal, bilateral
				Lungs and Bronchi: No macropathology findings	Lungs and Bronchi: Inflammation, Alveoli , minimal
				Lymph Node, Mandibular: Enlarged, 10-19mm, 2-5 (few)	= Lymph Node, Mandibular: Plasmacytosis, marked
					Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight
				Trachea: No macropathology findings	Trachea: Foci, Inflammatory Cells, slight

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Urinary Bladder

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0035	92 (14)	Treatment	Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, slight

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0036	92 (14)	Treatment	Kidneys: No macropathology findings	Kidneys: Basophilia, Tubular , minimal, focal
				Lungs and Bronchi: Pale area(s), all lobes, $\leq 1$ mm, 5+ (many)	Examined, not correlated
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Cellularity Increased, Follicles, slight
				Stomach: No macropathology findings	Stomach: Dilatation, Glands , minimal

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Testes; Thymus; Thyroids; Trachea; Urinary Bladder



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0037	92 (14)	Treatment	Liver: No macropathology findings  Lungs and Bronchi: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal  Lungs and Bronchi: Alveolar Macrophages, Foamy , minimal

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0038	92 (14)	Treatment	Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal Liver: Necrosis, Hepatocellular, Focal , minimal
				Lungs and Bronchi: No macropathology findings	Lungs and Bronchi: Mineralization, Alveoli , minimal
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, slight Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight Lymph Node, Mandibular: Cellularity Increased, Follicles, slight
				Stomach: No macropathology findings	Stomach: Infiltration, Inflammatory Cells, Mucosal/Submucosal, Nonglandular Region , slight

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0038	92 (14)	Treatment	Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments: Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Testes; Thymus; Thyroids; Trachea; Urinary Bladder	

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0039	92 (14)	Treatment	Kidneys: Dilated Pelvis, right	= Kidneys: Dilatation, Pelvic , slight, unilateral Kidneys: Infiltration, Inflammatory Cells, Interstitial , minimal Kidneys: Lymphocytic Infiltration, Peri-pelvic, slight, unilateral
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal
				Lungs and Bronchi: Pale area(s), multilobular, <=1mm, 5+ (many)	Examined, not correlated
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, moderate Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0039	92 (14)	Treatment	Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments: Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder	

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4M	0040	92 (14)	Treatment	All tissues: No macropathology findings	All examined tissues: No histopathology findings

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Epididymides; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Prostate; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Seminal Vesicles; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Testes; Thymus; Thyroids; Trachea; Urinary Bladder

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0131	93 (14)	Treatment	Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal
				Lungs and Bronchi: Pale area(s), all lobes, $\leq 1$ mm, 5+ (many)	Examined, not correlated
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, moderate Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight
				Vagina: No macropathology findings	Vagina: Estrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0132	93 (14)	Treatment	Lungs and Bronchi: No macropathology findings  Spleen: No macropathology findings  Vagina: No macropathology findings	Lungs and Bronchi: Alveolar Macrophages, Foamy , minimal Lungs and Bronchi: Alveolar Macrophages, Increased, Focal , minimal  Spleen: Cellularity Increased, White Pulp , slight Spleen: Hemosiderosis , minimal  Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0133	93 (14)	Treatment	Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, slight Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight
				Ovaries: Abnormal color, bilateral, pale	Examined, not correlated Ovaries: Corpora Lutea, Absent , present, bilateral
				Pituitary: No macropathology findings	Pituitary: tissue missing
				Skin and Subcutis: Hair Loss, side(s) of face, 2-5 (few)	Examined, not correlated
				Skin and Subcutis: Scab(s), side(s) of face, dark, <=1mm, 5+ (many)	= Skin and Subcutis: Scab(s), slight  Skin and Subcutis: Ulceration, Epidermal, slight, focal Skin and Subcutis: Hyperplasia, Epidermal, slight
				Stomach: Depression(s), non glandular mucosa, <=1mm, 1 (one)	Examined, not correlated

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0133	93 (14)	Treatment		Stomach: Hyperplasia, Epithelial, Nonglandular Region , slight
				Vagina: No macropathology findings	Vagina: Mucification, present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Peyer's Patches/GALT; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0134	93 (14)	Treatment	Lymph Node, Mandibular: No macropathology findings Vagina: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, moderate Vagina: Estrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0135	93 (14)	Treatment	Kidneys: No macropathology findings	Kidneys: Cast(s), Intratubular , minimal, focal
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, slight
				Ovaries: No macropathology findings	Ovaries: Corpora Lutea, Absent , present, bilateral
				Peyer's Patches/GALT: No macropathology findings	Peyer's Patches/GALT: Cellularity, Increased, moderate
				Vagina: No macropathology findings	Vagina: Diestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Pancreas; Parathyroids; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0136	93 (14)	Treatment	Liver: No macropathology findings  Spleen: No macropathology findings  Vagina: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal  Spleen: Extramedullary Hemopoiesis, Increased , minimal  Vagina: Proestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0137	93 (14)	Treatment	Skin and Subcutis: Hair Loss, side(s) of face, left, 1 (one) Skin and Subcutis: Scab(s), side(s) of face, left, dark, <=1mm, 2-5 (few)  Vagina: No macropathology findings	Examined, not correlated = Skin and Subcutis: Scab(s), slight  Skin and Subcutis: Hyperplasia, Epidermal, minimal  Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0138	93 (14)	Treatment	Heart: No macropathology findings	Heart: Cardiomyopathy, Chronic , minimal
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, moderate Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight
				Skin and Subcutis: Hair Loss, side(s) of face, 2-5 (few)	Examined, not correlated
				Skin and Subcutis: Scab(s), side(s) of face, dark, <=1mm, 5+ (many)	= Skin and Subcutis: Scab(s), slight
					Skin and Subcutis: Hyperplasia, Epidermal, minimal
				Vagina: No macropathology findings	Vagina: Metestrus , present

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0138	93 (14)	Treatment	Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments: Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Ileum; Jejunum; Kidneys; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus	



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0139	93 (14)	Treatment	Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal
				Skin and Subcutis: Hair Loss, side(s) of face, right, 1 (one)	Examined, not correlated
				Skin and Subcutis: Scab(s), side(s) of face, right, dark, <=1mm, 2-5 (few)	= Skin and Subcutis: Scab(s), slight
					Skin and Subcutis: Hyperplasia, Epidermal, minimal
				Vagina: No macropathology findings	Vagina: Mucification, present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
1F	0140	93 (14)	Treatment	Vagina: No macropathology findings	Vagina: Estrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0121	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0122	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0123	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0124	93 (14)	Treatment	Skin and Subcutis: Hair Loss, side(s) of face, 2-5 (few) Skin and Subcutis: Scab(s), side(s) of face, dark, <=1mm, 2-5 (few)	Examined, not correlated = Skin and Subcutis: Scab(s), moderate  Skin and Subcutis: Hyperplasia, Epidermal, slight

Tissues without comments under macropathology findings were within normal limits at necropsy.

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0125	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0126	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0127	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0128	93 (14)	Treatment	Kidneys: Depression(s), bilateral, <=1mm, 5+ (many)  Liver: Small, posterior caudate lobe	Examined, not correlated Kidneys: Mineralization, Cortico-Medullary Junction , minimal, unilateral  = Liver: Glycogen, Decreased , moderate Liver: Aggregates, Lymphoid/Macrophage, slight Liver: Necrosis, Hepatocellular, Focal , minimal

Tissues without comments under macropathology findings were within normal limits at necropsy.

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0129	93 (14)	Treatment	Skin and Subcutis: Hair Loss, side(s) of face, left, 1 (one) Skin and Subcutis: Scab(s), side(s) of face, left, dark, <=1mm, 2-5 (few)	Examined, not correlated = Skin and Subcutis: Scab(s), slight Skin and Subcutis: Hyperplasia, Epidermal, minimal

Tissues without comments under macropathology findings were within normal limits at necropsy.

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
2F	0130	93 (14)	Treatment	Skin and Subcutis: Hair Loss, side(s) of face, 2-5 (few) Skin and Subcutis: Scab(s), side(s) of face, dark, <=1mm, 2-5 (few)	Examined, not correlated = Skin and Subcutis: Scab(s), slight  Skin and Subcutis: Hyperplasia, Epidermal, minimal

Tissues without comments under macropathology findings were within normal limits at necropsy.

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0101	93 (14)	Treatment	Kidneys: Depression(s), bilateral, <=1mm, 2-5 (few)  Lungs and Bronchi: Pale area(s), multilobular, <=1mm, 2-5 (few)	= Kidneys: Scars, Cortical , minimal, focal  = Lungs and Bronchi: Alveolar Macrophages, Foamy , minimal

Tissues without comments under macropathology findings were within normal limits at necropsy.

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0102	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0103	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0105	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0106	93 (14)	Treatment	Kidneys: Depression(s), cortex, right, <=1mm, 1 (one)	Examined, not correlated Kidneys: Mineralization, Cortico-Medullary Junction , minimal, unilateral

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0107	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0108	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0109	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0110	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537			
Dose Group	1	2	3	4	
Dose (%)	0	10	33	100	

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
3F	0141	93 (14)	Treatment	All tissues: No macropathology findings	No histopathology examination

Tissues without comments under macropathology findings were within normal limits at necropsy.

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0111	93 (14)	Treatment	Kidneys: No macropathology findings Parathyroids: No macropathology findings Vagina: No macropathology findings	Kidneys: Dilatation, Pelvic , slight, unilateral Parathyroids: both, tissue missing Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0112	93 (14)	Treatment	Kidneys: No macropathology findings	Kidneys: Infiltration, Inflammatory Cells, Interstitial , minimal
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal
				Spleen: No macropathology findings	Spleen: Extramedullary Hemopoiesis, Increased , minimal
				Vagina: No macropathology findings	Vagina: Proestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus



APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0113	93 (14)	Treatment	Vagina: No macropathology findings	Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0114	93 (14)	Treatment	Kidneys: No macropathology findings  Vagina: No macropathology findings	Kidneys: Basophilia, Tubular , minimal, focal Kidneys: Infiltration, Inflammatory Cells, Interstitial , minimal  Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0115	93 (14)	Treatment	Skin and Subcutis: Hair Loss, forelimb(s), 2-5 (few)  Spleen: No macropathology findings  Vagina: No macropathology findings	Not Applicable  Spleen: Extramedullary Hemopoiesis, Increased , slight  Vagina: Proestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Liver; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0116	93 (14)	Treatment	Heart: No macropathology findings	Heart: Infiltration, Inflammatory Cells, Myocardial , slight
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal Liver: Necrosis, Hepatocellular, Focal , minimal
				Pituitary: No macropathology findings	Pituitary: Cyst(s), minimal
				Spleen: No macropathology findings	Spleen: Extramedullary Hemopoiesis, Increased , minimal
				Vagina: No macropathology findings	Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Ileum; Jejunum; Kidneys; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0117	93 (14)	Treatment	Kidneys: Depression(s), bilateral, <=1mm, 5+ (many)	= Kidneys: Scars, Cortical , minimal, focal Kidneys: Mineralization, Cortico-Medullary Junction , minimal, unilateral
				Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, slight
					Lymph Node, Axillary Lt: tissue missing
				Trachea: No macropathology findings	Trachea: Foci, Inflammatory Cells, slight
				Vagina: No macropathology findings	Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Lungs and Bronchi; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Urinary Bladder; Uterine Cervix; Uterus

= - correlation of macropathology to histopathology finding

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0118	93 (14)	Treatment	Kidneys: No macropathology findings  Liver: No macropathology findings  Vagina: No macropathology findings	Kidneys: Mineralization, Cortico-Medullary Junction , minimal, unilateral  Liver: Aggregates, Lymphoid/Macrophage, minimal Liver: Foci, Cellular Alteration, Clear Cell , minimal  Vagina: Proestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0119	93 (14)	Treatment	Kidneys: No macropathology findings Liver: No macropathology findings Vagina: No macropathology findings	Kidneys: Basophilia, Tubular , minimal, focal Liver: Aggregates, Lymphoid/Macrophage, minimal Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Lungs and Bronchi; Lymph Node, Axillary Lt; Lymph Node, Mandibular; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Spleen; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus

APPENDIX 10 (cont) Macropathology and histopathology - individual findings for animals killed after 13 weeks of treatment

Request ID: 5035207

	Control	Lactase, batch PPL34537		
Dose Group	1	2	3	4
Dose (%)	0	10	33	100

Group /Sex	Animal Number	Day (Week) of Death	Phase	Macropathology findings	Histopathology findings
4F	0120	93 (14)	Treatment	Liver: No macropathology findings	Liver: Aggregates, Lymphoid/Macrophage, minimal
				Lungs and Bronchi: No macropathology findings	Lungs and Bronchi: Infiltration, Inflammatory Cells, Perivascular, slight
				Lymph Node, Mandibular: No macropathology findings	Lymph Node, Mandibular: Plasmacytosis, moderate Lymph Node, Mandibular: Cellularity Increased, Paracortex, slight
				Spleen: No macropathology findings	Spleen: Extramedullary Hemopoiesis, Increased , minimal
				Vagina: No macropathology findings	Vagina: Metestrus , present

Tissues without comments under macropathology findings were within normal limits at necropsy. The following examined tissues had no histopathology findings, notes, or comments:

Adrenals; Aorta; Bone, Femur Including Joint; Bone, Sternum; Brain; Cecum; Colon; Duodenum; Esophagus; Heart; Ileum; Jejunum; Kidneys; Lymph Node, Axillary Lt; Lymph Node, Mesenteric; Mammary; Nerve, Sciatic; Ovaries; Pancreas; Parathyroids; Peyer's Patches/GALT; Pituitary; Rectum; Salivary Gland, Mandibular; Salivary Gland, Parotid; Salivary Gland, Sublingual; Skin and Subcutis; Spinal Cord Cervical; Spinal Cord Lumbar; Spinal Cord Thoracic; Stomach; Thymus; Thyroids; Trachea; Urinary Bladder; Uterine Cervix; Uterus



**Annex 1      Certificate of analysis**



## Toxicology & Product Safety

Date: 20. December, 2013  
Project no.: DEV00917  
Luna: 2012-18502-02  
Ref.: SuH

# Certificate of Analysis

Product:	TOX BATCH
Batch:	PPL34537
Type of enzyme:	Lactase
Host organism:	<i>Bacillus licheniformis</i>
Physical form / Colour:	Brownish liquid at room temperature
E.C.:	3.2.1.23

Activity:	6730	LAU(B)/g
Water (KF):	90.7	% w/w
Dry matter:	9.3	% w/w
Ash (600°C):	2.9	% w/w
Total Organic Solids (TOS):	6.4	% w/w
Specific gravity (g/ml):	1.050	g/ml
pH:	7.5	
Total viable counts/g:	6800	



Study Director

## **Annex 2      Formulation analysis report**



**Novozymes A/S**  
**Enzyme Laboratories**  
**Enzyme Analytical Laboratory**

**CASU**  
**2014APRIL25**  
**Luna no. 2014-07053-01**

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**Huntingdon Life Sciences Study Number: LKG0074**  
**Novozymes Reference No.: 20136077**

## **Investigation Report**

**Lactase, batch PPL34537: Toxicity Study by Oral Gavage**  
**Administration to Sprague-Dawley Rats for 13 Weeks**

**Analysis of Dose Formulation Samples Returned from Huntingdon Life Sciences**

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Investigation Report

NZ Reference No. 20136077

## 1. GLP Compliance

This investigation was conducted at the Enzyme Laboratories, Enzyme Analytical Laboratory, Novozymes A/S, in compliance with the OECD's principles of Good Laboratory Practice, ENV/MC/CHEM(98)17.

## 2. Approval

Approved by:

Date: 18 Jun 2014

Signature: \_\_\_\_\_

Principal Investigator

Investigation Report

NZ Reference No. 20136077

### 3. Quality Assurance Statement

REPORT: Lactase, batch PPL34537: Toxicity Study by Oral Gavage  
Administration to Rats for 13 Weeks  
Analysis of Dose Formulation Samples Returned from HLS

STUDY NUMBER LKG0074

REFERENCE  
NUMBER 20136077

The conduct of this study has been subject to appropriate inspections and the report has been reviewed according to the relevant Standard Operation Procedures of Novozymes A/S Quality Assurance.

Inspection/Audit	Dates of inspection	Inspection results reported to Principal Investigator and Management
Analysis	6 MAR 2014	6 MAR 2014
Report	21 MAY 2014	21 MAY 2014

I hereby confirm that the report reflects the raw data.

18 Jun 2014  
Date



Quality Assurance

Investigation Report

NZ Reference No. 20136077

#### 4. General Information

**Principal Investigator:**

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Krogshoejvej 36, building 6E1.18  
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CASU@novozymes.com

**Laboratory:**

Enzyme Analytical Laboratory (EAL)  
Enzyme Laboratories  
Novozymes A/S  
Krogshoejvej 36, building 6E  
DK-2880 Bagsvaerd, Denmark

**Sponsor/Monitor:**

[REDACTED]  
Toxicology and Product Safety  
Novozymes A/S  
Krogshoejvej 36, building 1A1.13  
DK-2880 Bagsvaerd, Denmark  
Lini@novozymes.com

**Personnel:**

Laboratory employees from EAL  
[REDACTED]

## 5. Purpose

The purpose of this phase of the study was to determine whether the enzyme activity LAU(B)/g in the dose solutions prepared for use in week 1, 6 and 13 were approximately equal comparing preparations of corresponding levels of activity. It was also checked if the activity of the 100% dose solution complied with the enzyme activity of the Tox-batch. Furthermore, the control formulations from week 1, 6 and 13 were analysed for proof of absence of enzyme activity.

Content check analysis is required as part of the OECD guideline for oral toxicity studies (OECD: Repeated dose 90-day oral toxicity study in rodents, Testing of Chemicals guideline No. 408. 21 September 1998).

The samples of the present investigation are dose solutions of Lactase, PPL34537.

## 6. Sample Handling

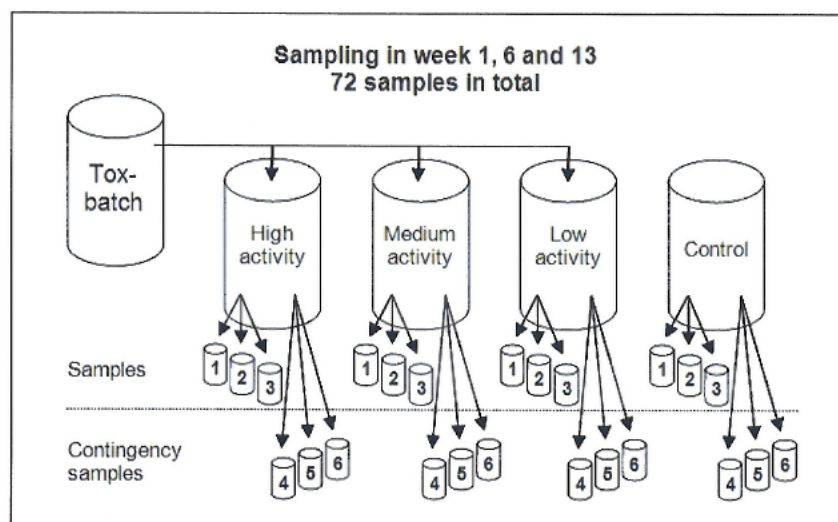
### Sample sampling

During the study, 36 samples were prepared for analysis of enzyme activity and a further 36 samples were prepared as contingency samples (72 samples in total). These were categorised into four groups:

- High activity (100% of Tox-batch activity)
- Medium activity (33% of Tox-batch activity)
- Low activity (10% of Tox-batch activity)
- Control group (0% of Tox-batch activity)

In week 1, 6 and 13, 6 samples of 5 mL were taken from each of the groups and labelled, as illustrated below.





More details about the schedule for the analytical phase are found in the current version 12.0 of PSL-SP-0107.01-D.

#### Sample transportation and registration

Samples (numbered "1", "2" and "3"; 36 samples in total) from Huntingdon Life Sciences were sent (on dry ice) directly to the department Enzyme Analytical Laboratory (EAL) on 2014FEB18. The samples were received frozen in excess dry ice (2014FEB20) and registered on 2014FEB20.

#### Storage of Samples for Analysis

The 36 samples were all intact and following registration in EAL the samples were stored frozen (nominally -18°C) until analysis.

#### Sample Defrost and Date of Analysis

All samples labelled "1", including controls, were defrosted at room temperature before analysis on 2014MAR06.

### 7. Method

The analysis was performed according to the current version 7.0 of PSL-SM-5123.01-D.

Lactase, EC.3.2.1.23, hydrolyzes ONPG (*o*-NitroPhenyl  $\beta$ -Dgalatopyranoside) and hereby ONP (*o*-NitroPhenyl) is released. *o*-NitroPhenyl has a yellow colour when in an alkaline solution and can be quantified at 405 nm.

LACTASE activity is measured in LAU(B)/g. The activity is determined relative to a Bifido Lactase standard.

The samples are analysed as 2 weighings at 1 standard curve as specified for GLP samples in the current version 4.0 of PSL-SP-0598.01-D.

The control samples were analysed as 1 weighing at 1 standard curve as specified for control samples in current version 12.0 of PSL-SP-0107.01-D

## 8. Computer systems

Computer systems used during the formulation analysis phase of this study include:

- JMP® v. 10.0.2, SAS Institute Inc.
- LIMS LabVantage Sapphire R4, LabVantage Solutions Inc.
- MS-Excel 2010, Microsoft Inc.
- Konelab Arena 30 v. 7.2ARIN, Thermo Fisher Scientific

## 9. Deviations

Regarding statistical calculations:

The result output is different from that described in the current valid version 12.0 of PSL-SP-0107.01-D. According to the procedure described in PSL-SP-0107.01-D, sample No. 2 and 3 must be run when there is significant difference between the Week No. or Week No.\*Group as well as significant difference between High and Tox level. However, the observed difference – although statistically significant – is evaluated to be practically insignificant. In additions to the statistical evaluation, calculations have been made to elucidate the deviation observed from sample 1, see table 4. As a consequence of this assumption the mean for group medium is calculated, see table 6.

The deviation to the statistical evaluation confers no impairment to the validity of the result.

## 10. Results and Discussion

The analytical results in LAU(B)/g were evaluated according to valid version 12.0 of PSL-SP-0107.01-D. Results are represented with three significant digits. Results below the methods determination limit are reported as <3.53 LAU(B)/g according to the current version 7.0 of PSL-SM-5123.01-D.

All statistical calculations were carried out using SAS JMP script according valid version 3.0 of PSL-AS-0123. The current SAS JMP ver. 10.0.2 was used for the calculations.

Investigation Report

NZ Reference No. 20136077

**Results from control samples:**

*No activity above the detection limit (<3.53 LAU(B)/g) was found for the control group.*

**Results from samples "1":**

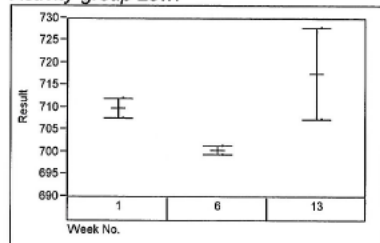
Table 1. Mean results of each sample for the dose groups "High", "Medium" and "Low" given in LAU(B)/g:

Sample No.	Group	Week 1	Week 6	Week 13
1	Low	710	701	718
	Medium	2330	2280	2350
	High	6820	6750	6830

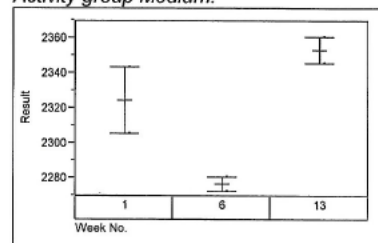
The observed CV is 0.87 %, which is acceptable, i.e. below the LAU(B) method CV approval of 2.5 %.

Below is a visual presentation of the results.

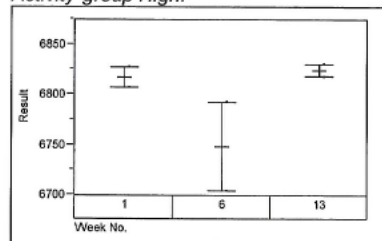
*Activity group Low:*



*Activity group Medium:*



*Activity group High:*



**Investigation of whether dose solutions prepared for use in week 1, 6 and 13 are approximately equal comparing preparations of corresponding levels of activity:**

*The result of the statistical test shows that there was significant difference between the dose formulations (group) given in week 1, 6 and 13, comparing preparations of corresponding levels of activity. The difference between the weeks (Week No.) is consistent across groups. In absolute figures the observed difference is evaluated to be practically insignificant.*

**Table 2:** Results from statistical analysis of difference between weeks.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob > F	Is there sign. diff. ('Prob > F' < 0.05)
Group	2	2	15.332669	101150.4	< 0.0001*	YES*
Week No.	2	2	0.001603	10.5747	0.0043^	YES^
Week No.*Group	4	4	0.000250	0.8233	0.5421	NO

\* The significant difference between groups high, medium and low is as expected.

^ There is significant difference between the weeks (Prob > F is 0.0043). This difference is consistent across Groups (since Week No.\*Group is insignificant).

(Nparm: the number of parameters associated with the effect, and DF: shows the degrees of freedom for the effect test)

**Table 3.** The difference between dose formulations is evaluated by a pairwise comparison (Week No.) using Student's t.

Week	Level	Group	Least Sq Mean
1	Medium	A	2241
6	Medium	B	2208
13	Medium	A	2259

As can be seen from the comparison using Student's t test, the week 6 result for the medium dose is *significantly different when the medium dose group are compared* ( $\alpha=0.05$ ,  $Q=2.26216$ ).

According to the procedure described in PSL-SP-0107.01-D, Sample No. 2 and 3 must be run when there is significant difference in dose within weeks. However, the observed difference – although statistically significant – is evaluated to be practically insignificant. The difference in percentage is seen below (compared to week 13). In most studies this difference in value would not be found significant, but as the observed CV is very low (0.87 %), it is observed here (see table 4).



**Table 4.** Relative recovery deviation in activity compared to week 13

Week No.	Least Sq Mean activity LAU(B)/g			Relative deviation from mean		
	1	6	13	1	6	13
Medium	2241	2208	2259	-0.8%	-2.3%	-

Due to this a deviation from the statistical calculation described in PSL-SP-0107.01-D was made, and the test of difference between Tox and High was done as if no significant difference between weeks was seen for Group medium.

**Table 5.** The difference between dose formulations is evaluated by a pairwise comparison (Week No.\*Group) using Tukey.

Week	Level	Group		Least Sq Mean
1	High	A		6818
6	High	A		6749
13	High	A		6825
1	Medium		B	2325
6	Medium		B	2277
13	Medium		B	2353
1	Low		C	710
6	Low		C	701
13	Low		C	718

Levels not connected by same letter are significantly different.

The pairwise comparison using Tukey show *no significantly different when dose groups are compared amongst Weeks No.\*Group ( $\alpha=0.05$ ,  $Q=3.95606$ )*.

**Table 6.** Mean results for each dose groups "High", "Medium" and "Low" given in LAU(B)/g:

High	Medium	Low
6797	2319	709

**Investigation of whether the activity is approximately equal for group High and the Tox-batch:**

*The result of the statistical test shows that there is significant difference between the dose formulation for group high and the Tox-batch.*

**Table 7:** Results from statistical analysis of difference between group High and Tox-batch

Analyses result for Tox-batch LAU(B)/g	Mean of group High LAU(B)/g	Prob > F	Is there sign. diff. (*Prob > F' < 0.05)
6730	6800	0.0186*	Yes

**Table 8.** Relative deviation in activity comparing Tox-batch level

Level	Group	Least Sq. Mean activity LAU(B)/g	Relative deviation from mean
High	A	6800	1.0%
Tox	B	6730	-

The relative deviation was 1.0% in absolute figures. The average activity of group High is 6797 LAU(B)/g, which is above the Tox average of 6729 LAU(B)/g. The activity of the intended dose is adequate, compared in absolute figures, and the deviation is evaluated to be practically insignificant.

## 11. Conclusion

The result of the statistical test shows that there was significant difference between the dose formulations (group) given in week 1, 6 and 13, comparing preparations of corresponding levels of activity. The difference between the weeks (Week No.) is consistent across groups. The activity values in Week 1, 6 and 13 were close to equal, demonstrating acceptable formulation. In absolute figures the observed difference is evaluated to be practically insignificant.

There is significant difference between the lactase activity (LAU(B)/g) of group High and the Tox-batch, but in absolute figures the observed difference is evaluated to be practically insignificant.

Absence of activity in the control samples was shown.

## **12. Archiving**

The Investigation Plan, all raw data and Investigation Report are archived in Novozymes QM Central Archive by the Principal Investigator.

## **Annex 3      Pathology report**



## **PATHOLOGY REPORT**

### **Lactase, batch PPL34537: Toxicity Study by Oral Gavage Administration to Sprague-Dawley Rats for 13 Weeks**

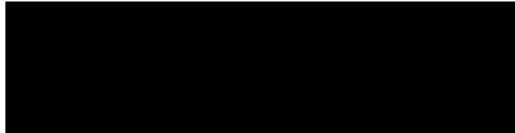
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<b>HLS study number:</b>	LKG0074
<b>Sponsor reference number:</b>	20136077
<b>Version ID:</b>	Final Report
<b>Issue date:</b>	10 July 2014

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## Signature Page

**Lactase, batch PPL34537: Toxicity Study by Oral Gavage Administration  
to Sprague-Dawley Rats for 13 Weeks**



\_\_\_\_\_  
PhD

\_\_\_\_\_  
Date 10 July 2014

Principal Pathologist  
Huntingdon Life Sciences

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# 1. Introduction

## 1.1 Objective

The objective of this study was to assess the systemic toxic potential of Lactase, batch PPL34537 (an enzyme used in the food industry), when administered orally by gavage to Sprague-Dawley rats for 13 weeks.

Group	Treatment	Dose (% of lactase, batch PPL34537)*	Number of animals Main study	
			Male	Female
1	Control	0	10	10
2	Lactase, batch PPL34537	10	10	10
3	Lactase, batch PPL34537	33	10	10
4	Lactase, batch PPL34537	100	10	10

\* Using a dose-volume of 10 mL/kg bodyweight.

# 2. Results

## 2.1 Decedents

There were no decedents in this study.

## 2.2 Macroscopic Pathology

### 2.2.1 Animals killed after 13 weeks of treatment

The macroscopic examination performed after 13 weeks of treatment revealed no test substance related lesions.

The incidence and distribution of all findings were consistent with the common background of macroscopic findings in rats at these laboratories.

## 2.3 Microscopic Pathology

### 2.3.1 Animals killed after 13 weeks of treatment

#### Treatment related findings

There were no test substance-related findings.

The incidence and distribution of all histopathological findings were consistent with the common background findings at these laboratories.

### **3. Discussion**

There were no corresponding microscopic findings to account for the reduced brain weight recorded at necropsy.

### **4. Conclusion**

No test substance-related findings were seen following oral gavage administration of Lactase, batch PPL34537 to Sprague-Dawley rats for 13 weeks.

## **Annex 4      Historical control data**

Historical control data - grip strength for Crl:CD(SD) rats during Week 12/13 of treatment

Study type : 13-Week Toxicity  
Route of administration : Various  
Caging : Gang/Individual  
Approximate age at testing : 16-18 weeks

Date	Number of animals	Males		Number of animals	Females	
		Forelimb	Hindlimb		Forelimb	Hindlimb
Mar-10	10	1.21	0.54	10	1.07	0.43
Apr-10	10	1.27	0.59	10	1.03	0.48
Jun-10	10	1.16	0.58	10	0.98	0.47
Aug-10	10	1.50	0.60	10	1.12	0.45
Sep-10	10	1.11	0.53	10	0.92	0.45
Sep-10	10	1.25	0.45	10	1.07	0.43
Nov-10	10	1.17	0.59	10	0.94	0.44
Dec-10	13	1.09	0.57	15	0.87	0.49
Jan-11	15	1.18	0.59	15	1.10	0.50
Apr-11	10	1.20	0.65	10	1.12	0.53
Jun-11	10	1.06	0.53	10	0.84	0.39

Historical control data - grip strength for Crl:CD(SD) rats during Week 12/13 of treatment

Study type : 13-Week Toxicity  
Route of administration : Various  
Caging : Gang/Individual  
Approximate age at testing : 16-18 weeks

Date	Number of animals	Males		Number of animals	Females	
		Forelimb	Hindlimb		Forelimb	Hindlimb
Sep-11	10	1.17	0.48	10	0.86	0.39
Jan-12	10	1.16	0.47	10	0.86	0.40
Jul-12	10	1.23	0.44	10	1.05	0.37
Sep-12	20	1.00	0.61	19	0.88	0.44
Jan-13	10	1.06	0.58	10	0.98	0.51
Apr-13	10	1.11	0.59	10	1.05	0.47
Apr-13	10	1.21	0.44	10	1.06	0.38
Apr-13	10	1.03	0.51	10	0.85	0.44
Jan-14	10	1.10	0.54	10	0.98	0.39
Total of 20 studies						
Mean		1.16	0.54		0.98	0.44
Minimum		1.00	0.44		0.84	0.37
Maximum		1.50	0.65		1.12	0.53



## **Annex 5      GLP Compliance statements**

**EYE RESEARCH CENTRE GLP COMPLIANCE STATEMENT 2012**



**THE DEPARTMENT OF HEALTH OF THE GOVERNMENT  
OF THE UNITED KINGDOM**

**GOOD LABORATORY PRACTICE**

**STATEMENT OF COMPLIANCE  
IN ACCORDANCE WITH DIRECTIVE 2004/9/EC**

TEST FACILITY	TEST TYPE(S)
Huntingdon Life Sciences Eye Research Centre Occold Eye Suffolk IP23 7PX	Analytical/Clinical Chemistry Environmental Fate Environmental Toxicity Ecosystems Phys.Chem. Testing Residue studies Mutagenicity Toxicology

**DATE OF INSPECTION**  
**18<sup>th</sup> – 20<sup>th</sup> June 2012**

An inspection for compliance with the Principles of Good Laboratory Practice was carried out at the above test facility as part of the UK Good Laboratory Practice Compliance Monitoring Programme.

This statement confirms that, on the date of issue, the UK Good Laboratory Practice Monitoring Authority were satisfied that the above test facility was operating in compliance with the OECD Principles of Good Laboratory Practice.

This statement constitutes a Good Laboratory Practice Instrument (as defined in the UK Good Laboratory Practice Regulations 1999).

[Redacted Signature]

19/9/12

[Redacted Name]

Head, UK GLP Monitoring Authority

**MHRA**

## EYE RESEARCH CENTRE GLP COMPLIANCE STATEMENT 2014



### THE DEPARTMENT OF HEALTH OF THE GOVERNMENT OF THE UNITED KINGDOM

#### GOOD LABORATORY PRACTICE

#### STATEMENT OF COMPLIANCE IN ACCORDANCE WITH DIRECTIVE 2004/9/EC

##### TEST FACILITY

Huntingdon Life Sciences (Eye)  
Eye Research Centre  
Occold  
Eye  
Suffolk  
IP23 7PX

##### TEST TYPE(S)

Analytical/Clinical Chemistry  
Environmental Fate  
Environmental Toxicity  
Ecosystems  
Phys.Chem. Testing  
Residue studies  
Mutagenicity  
Toxicology

##### DATE OF INSPECTION

05 to 07 February 2014

An inspection for compliance with the Principles of Good Laboratory Practice was carried out at the above test facility as part of the UK Good Laboratory Practice Compliance Monitoring Programme.

This statement confirms that, on the date of issue, the UK Good Laboratory Practice Monitoring Authority were satisfied that the above test facility was operating in compliance with the OECD Principles of Good Laboratory Practice.

This statement constitutes a Good Laboratory Practice Instrument (as defined in the UK Good Laboratory Practice Regulations 1999).

[Redacted signature area]

*22/12/14*

[Redacted name]

Head, UK GLP Monitoring Authority



## HUNTINGDON RESEARCH CENTRE GLP COMPLIANCE STATEMENT 2012



### THE DEPARTMENT OF HEALTH OF THE GOVERNMENT OF THE UNITED KINGDOM

#### GOOD LABORATORY PRACTICE

#### STATEMENT OF COMPLIANCE IN ACCORDANCE WITH DIRECTIVE 2004/9/EC

##### TEST FACILITY

Huntingdon Life Sciences  
Woolley Road  
Alconbury  
Huntingdon  
Cambridgeshire  
PE28 4HS

##### TEST TYPE(S)

Analytical/Clinical Chemistry  
Environmental Fate  
Environmental Toxicity  
Ecosystems  
Toxicology  
Residue Studies

*Reissued February 2013 to include Residue  
Studies*

##### DATE OF INSPECTION

20 to 22 November 2012

An inspection for compliance with the Principles of Good Laboratory Practice was carried out at the above test facility as part of the UK Good Laboratory Practice Compliance Monitoring Programme.

This statement confirms that, on the date of issue, the UK Good Laboratory Practice Monitoring Authority were satisfied that the above test facility was operating in compliance with the OECD Principles of Good Laboratory Practice.

This statement constitutes a Good Laboratory Practice Instrument (as defined in the UK Good Laboratory Practice Regulations 1999).



22/2/13



Head, UK GLP Monitoring Authority



## HUNTINGDON RESEARCH CENTRE GLP COMPLIANCE STATEMENT 2012



### THE DEPARTMENT OF HEALTH OF THE GOVERNMENT OF THE UNITED KINGDOM

#### GOOD LABORATORY PRACTICE

#### STATEMENT OF COMPLIANCE IN ACCORDANCE WITH DIRECTIVE 2004/9/EC

##### TEST FACILITY

Huntingdon Life Sciences  
Woolley Road  
Alconbury  
Huntingdon  
Cambridgeshire  
PE28 4HS

*Reissued April 2014 to include Mutagenicity*

##### TEST TYPE(S)

Analytical/Clinical Chemistry  
Environmental Fate  
Environmental Toxicity  
Ecosystems  
Residue Studies  
Mutagenicity  
Toxicology

##### DATE OF INSPECTION

20 to 22 November 2012

An inspection for compliance with the Principles of Good Laboratory Practice was carried out at the above test facility as part of the UK Good Laboratory Practice Compliance Monitoring Programme.

This statement confirms that, on the date of issue, the UK Good Laboratory Practice Monitoring Authority were satisfied that the above test facility was operating in compliance with the OECD Principles of Good Laboratory Practice.

This statement constitutes a Good Laboratory Practice Instrument (as defined in the UK Good Laboratory Practice Regulations 1999).



24/4/14



Head, UK GLP Monitoring Authority

