

**Amendment 1 to REPORT No. 2500301
Regulatory Document**

DSM 

Date:	17-June-2008
Author:	Beck M
Report Title:	In vitro Intestinal Absorption of 3H-Lignosulfonate using the Caco-2 Monolayer Model
Amendment No	1
Project No.	6309
Compound	Ca-Lignosulfonate, Food Grade

Justification for Amendment

To specify the use of the word 'radiolysis' in the document.


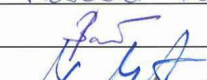
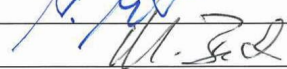

Clarification

In several passages within the Regulatory Document Report 2500301, the expression *radiolysis* was used, which might be unclear or misleading. '*Radiolysis*' is mentioned in the introduction (page 2), in the chapters of the preparation of the tritium-labeled stock solution of the test item (page 4), and in the results and discussion chapter (pages 9, 10, and 12)
In the following paragraph it is explained and commented what was meant.

Distribution
Karin Feltes, NBD/RH
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Approved

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The predominant risk associated with using ^3H -labeled compounds in biological systems is the potential chemical and metabolic instability of the tritium atom itself at its specific site within the test molecule. In aqueous physiological environments of varying pH, tritiated water (HTO) is readily and unavoidably formed by hydrogen exchange processes. This is a well known phenomenon when using tritiated compounds in ADME studies.

Radiolysis is the dissociation of molecules by radiation. This is a process known to occur with many radioisotopically labelled compounds over time (also ^{14}C). Tritium-hydrogen exchange could be seen as one form of radiolysis. For our ADME studies with ^3H -lignosulfonate, we cannot exclude formation of other small MW components by radiolysis.

However, we have shown that the by far major part of the radioactivity permeating through the Caco-2 monolayer can be attributed to tritiated water.