

Distribution of Coliphages in Various Foods

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ABSTRACT

The distribution of coliphages in various foods and the relationship between the incidences of coliphages and bacterial indicators were investigated. A total of 120 food samples comprising twelve products and including fresh meats, shellfish, vegetables and processed meats, were analyzed for indigenous coliphages using *Escherichia coli* hosts C, C-3000 and B. Bacterial analyses included enumeration of *E. coli*, fecal coliforms and coliforms, as well as aerobic plate counts and *Salmonella* analyses. Coliphages were detected (≥ 10 PFU/100 g) in 56% of samples and eleven of twelve products. Coliphages, *E. coli*, fecal coliforms and coliforms were recovered at a level of at least 30 organisms per 100 g in 43, 43, 68 and 81% of samples, with overall mean recoveries of 13, 19, 93 and 4300 organisms/100 g, respectively. Highest and lowest recoveries of coliphages and *E. coli* were from fresh meats and vacuum-packaged processed meats, respectively. Significant nonparametric correlations between coliphages, *E. coli*, fecal coliforms and coliforms were found among all food samples.

The ecology and distribution of bacteriophages of *Escherichia coli*, i.e., coliphages, as well as their potential value as indicators of fecal contamination or enteric pathogens in water and wastewater have been studied extensively in recent years (12,23,28,41,47-50). Coliphage analyses have been suggested as a more rapid, economical and simple alternative to conventional analysis of water or wastewater for indicator bacteria, enteric pathogens and enteroviruses. A number of investigators also have demonstrated that coliphages are superior to coliform bacteria as indicators of viral inactivation or removal during water and wastewater treatment (11,15,16,28,47). As with *E. coli*, coliphages can be recovered in relatively high densities from the feces of various warm-blooded animals, including man (9,36,45). Likewise, coliphages have been recovered from raw sewage, sewage effluent, natural waters, aquatic sediments

and drinking water in association with coliforms and enteric pathogens (23,28,41,44,47,49,50). Highly significant correlations ($P < 0.001$) and quantitative relationships between coliphages and total coliforms or fecal coliforms (23,49) and between coliphages and enteroviruses in water also have been reported (47).

The acceptability of coliphage indicators has been questioned by some investigators with regard to host specificity (19,43,48) and their possible non-fecal origin or alternate host bacteria (6,37,42,43,48). However, these problems have been addressed by use of appropriate host bacteria (17,18,20,39,43). Furthermore, recent studies have shown that the relative incidence of coliphages in water and wastewater having alternate bacterial hosts of non-fecal origin is low (8,17,39,50). Although male or F plasmid-specific coliphages can infect *E. coli* as well as other *Enterobacteriaceae* carrying an F plasmid, the F-pili required for phage adsorption are not produced by bacteria below 30°C (4,39,43). The fecal origin or ecological specificity of coliphages as a group has also been disputed with regard to the demonstrated ability of some coliphages to proliferate at temperatures less than 30°C in the presence of host *E. coli* (37,42,48). Distinct physiological types of coliphages have recently been recognized with respect to the effect of temperature on their infectivity as follows: high temperature (HT), mid temperature (MT) and low temperature (LT) types having the ability to proliferate between 30 to 42°C, 15 to 42°C and 15 to 30°C, respectively (42). The distribution of these coliphages in water closely reflected the temperature and degree of fecal contamination of the environment from which they were recovered (42).

Very little information is available concerning the distribution, sanitary significance, ecology or types of indigenous coliphages occurring in various foods. The recovery of coliphages and bacteriophages of *Salmonella* from milk was reported as early as 1937 but no quantitative or comparative data were presented (29). Coliphages have since been isolated from fresh oysters (27,48) as well as fresh and processed meats (24). A significant correlation between coliphage and coliform counts in oysters taken

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from water stations at various distances from a sewage outfall was reported by Kott and Gloyna (27). Coliphages were recovered from all fresh chicken and pork sausage samples as well as from 33% of delicatessen meats examined by Kennedy et al. (24).

The demonstrated value of coliphages as indicators of fecal contamination in water and wastewater as well as their frequent recovery and apparent association with bacterial indicators in a limited number of foods warranted further investigation of coliphage distribution in various foods. The objective of this study was to investigate the comparative distribution of coliphages infective for three *E. coli* host strains in a wide variety of foods using methods developed in a previous study (25). Additionally, the relationships between the incidence and levels of coliphages and bacterial indicators were investigated.

MATERIALS AND METHODS

Samples

The following samples were obtained at retail markets in Gainesville, FL: fresh chicken (breasts), fresh ground beef, fresh pork sausage, fresh oysters (pints), fresh mushrooms, fresh lettuce (leaf), biscuit dough (refrigerated), frozen chicken pot pie, pickle and pimento loaf (sliced at delicatessen), roast turkey breast (sliced at delicatessen), roast turkey breast (vacuum packaged, 6 oz) and roast chicken breast (vacuum packaged, 6 oz). Ten samples of each product were analyzed for a total of 120 samples. Samples were transported to the laboratory in refrigerated containers and were finely chopped and/or mixed using aseptic techniques. Approximately 250 to 340 g of a relatively homogeneous sample was available for subsequent analyses. Samples were held in sterile Whirlpak bags at 1 to 2°C until time of analysis (6 to 12 h).

Bacteriological analyses

All bacteriological media were obtained from Difco Laboratories (Detroit, MI) unless otherwise indicated. Procedures used for bacteriological analyses of samples conformed to those presented in the *Compendium of Methods for the Microbiological Examination of Foods* (2). A 50-g subsample was diluted 1:10 in Butterfield's phosphate buffer, blended and serially diluted as deemed appropriate. The 3-tube MPN procedure was used for enumeration of coliforms, fecal coliforms and *E. coli*. Aerobic plate counts were done using the surface-spread technique; sets of plates were incubated at 35°C for 48 h (APC 35) and 20°C for 5 d (APC 20).

Coliphage analyses

Subsamples (100 g) were diluted (wt/wt) 1 to 3 with EC medium eluent in sterile blender jars and blended (4000-4500 RPM, 10 min) with a Waring blender attached to a variable autotransformer. Sample suspensions (200 to 250 ml) were clarified by centrifugation (3000 × g, 5 min). Approximately 100 to 150 ml of clarified sample suspension was obtained for subsequent coliphage assays.

Cultures of *E. coli* B (ATCC 11303), *E. coli* C (ATCC 13706) and a male strain, *E. coli* C-3000 (ATCC 15597), were used for recovery of indigenous coliphages from samples.

EC medium was used for preparation of bottom agar plates

and overlay medium as described previously (24,25). For each sample, portions (4 ml) of clarified sample suspension were prepared and plated with 1.5 ml of molten (45°C) overlay agar (EC medium, 1.5% agar) and 0.3 ml of each host strain (6-h culture in EC medium, 35°C); five plates were prepared in this manner for each host strain with each sample. In addition, two 1-ml portions were prepared with each host for samples of fresh chicken, pork and oysters to obtain less crowded and more countable plates in samples having high densities of coliphages. Hence, the detection sensitivity was 10 plaque-forming units (PFU) per 100 g of sample. Assay plates were prepared and incubated at 35°C for 16 to 18 h.

Correlation determinations

Coliphage counts (PFU/100 g), bacterial indicator counts (converted to MPN/100 g) and aerobic plate counts (colony-forming units/g) were paired for samples of each product and all samples combined for correlation determinations using a nonparametric statistical procedure, Kendall's Tau analysis (7). Because coliphages and fecal indicator bacteria were not detected in a large percentage of samples, parametric correlation and/or linear regression analyses could not be realistically applied to much of the data obtained and, therefore, are not reported.

RESULTS

Recovery of coliphages and indicator bacteria

Coliphages (≥ 10 PFU/100 g) were detected in 50, 46 and 18% of foods samples with *E. coli* host strains C, C-3000 and B, respectively (Table 1). Coliphages of any host strain were likewise recovered from 56% of samples and eleven of twelve product types examined. *Escherichia coli*, fecal coliforms and total coliforms were detected at levels ≥ 30 MPN/100 g in 43, 68 and 81% of all samples, respectively. At a detection level of ≥ 30 PFU/100 g, coliphages (any host strain) were detected in 43% of all samples as with *E. coli*. The percentage occurrences of coliphages and *E. coli* in various products generally reflected mean recoveries (Table 2) in that highest incidences ($\geq 90\%$) were generally found with fresh meats. Neither coliphages nor bacterial indicators were detected in vacuum-packaged roast turkey. Coliphages and *E. coli* were detected in only one of ten samples of delicatessen pickle and pimento loaf. *Escherichia coli* was detected in only one of ten samples of fresh mushrooms, whereas coliphages were detected in eight of ten samples.

Coliphages of host strains *E. coli* C, C-3000 and B as well as numbers of bacterial indicators and aerobic plate counts (APC 20) recovered from twelve products are summarized in Table 2. Ranges of coliphage or bacterial indicator counts among samples for most products were generally large, i.e., greater than two orders of magnitude. Highest mean recoveries of both coliphages (≥ 1000 PFU/100 g) and *E. coli* (≥ 1000 MPN/100 g) were noted for fresh chicken and meats. The mean recovery of coliphages (*E. coli* C or *E. coli* C-3000) from oysters was also relatively high although the corresponding recovery of *E. coli* was comparatively low, i.e., 31

TABLE 1. Percentage occurrence of coliphages and bacterial indicators in twelve retail foods^a.

Product ^b	Coliphages				Bacterial Indicators		
	Host C	Host C-3000	Host B	Any host	<i>E. coli</i>	Fecal coliforms	Total coliforms
Fr. chicken	100	100	80	100	100	100	100
Fr. pork	100	100	90	100	90	100	100
Fr. ground beef	100	80	10	100	100	100	100
Fr. oysters	80	70	20	80	70	90	100
Fr. mushrooms	80	40	10	80	10	70	100
Fr. lettuce	20	30	— ^c	40	20	40	100
Chicken pot pie	40	50	—	50	20	80	90
Biscuit dough	—	30	—	30	50	80	90
Deli. loaf	10	10	—	10	20	70	100
Deli. roasted turkey	40	30	—	40	30	90	100
Pkg. roasted turkey	—	—	—	—	—	—	—
Pkg. roasted chicken	30	10	—	40	—	—	40
Overall	50	46	18	56	43	68	81

^aPositive sample = ≥ 10 PFU/100 g for coliphages or ≥ 30 MPN/100 g for bacterial indicators.

^bFr. = fresh; deli. = delicatessen; pkg. = vacuum packaged.

^c—, no recovery.

MPN/100 g. Lowest recoveries of coliphages and indicator bacteria were found with vacuum-packaged roast turkey and chicken. Mean recoveries of coliphages ranged from <10 PFU/100 g for vacuum-packaged turkey to 4200 PFU/100 g in fresh chicken; some samples of fresh chicken, pork sausage and oysters had coliphage counts in excess of 10,000 PFU/100 g. *E. coli* or fecal coliform counts in excess of 10,000 MPN/100 g were noted for some samples of fresh chicken and pork, with counts in excess of 100,000 MPN/100 g in some samples of ground beef. Mean total coliform recoveries were highest ($>100,000$ MPN/100 g) for fresh mushrooms, lettuce, oysters and ground beef. Mean recoveries of coliphages, *E. coli* and fecal coliforms generally did not parallel those of coliforms for fresh products. *Salmonella* was detected in four of ten samples of fresh chicken, two of fresh pork sausage and one sample of refrigerated biscuit dough. Coliphages and all bacterial indicators were recovered from each sample in which *Salmonella* was detected, with the exception of the biscuit sample in which no coliphages were recovered (data not presented).

Some coliphages are more heat resistant than enteric bacteria (1,5,10,38,40). The comparative recoveries of coliphages and indicator bacteria in the processed meat products is of interest in this regard (Tables 1 and 2). Coliphages were not recovered at levels greater than 10 PFU/100 g in these products in the absence of fecal or total coliforms which are considered indicators of processing inadequacy, post-processing contamination and/or temperature abuse. Processed meats obtained by the slice at delicatessens are subject to substantially more post-processing contamination and temperature abuse than corresponding products obtained in vacuum packages (31,34). The higher recoveries of indicator bacteria, particularly *E. coli* and fecal coliforms, in delicatessen meats as compared to vacuum-packaged processed meats reflect these

conditions (Table 2). Coliphages and each bacterial indicator were recovered from samples of delicatessen roast turkey (single manufacturer), with mean counts of fecal and total coliforms being 210 and 5300 MPN/100 g, respectively. *Escherichia coli* counts of 9300 MPN/100 g were also found in one sample of delicatessen turkey (data not presented). Coliphages or indicator bacteria were not detected in any samples of vacuum-packaged roast turkey produced by the same manufacturer supplying the delicatessen product. Coliphages (90 PFU/100 g) were recovered in one sample of delicatessen pickle and pimento loaf, with the concomitant recovery of 290 and 1500 MPN/100 g of fecal and total coliforms, respectively. *Escherichia coli* and fecal coliforms were not recovered in samples of vacuum-packaged roast chicken, whereas coliphages were recovered from four of ten samples at a level of 10 PFU/100 g, which is the minimum detection level. However, total coliforms were also recovered in four of ten samples of the vacuum-packaged roast chicken, with levels up to 240,000 MPN/100 g.

The overall mean recovery and occurrence of indigenous coliphages infective for *E. coli* C and C-3000 were similar and substantially higher than that with *E. coli* B (Tables 1 and 2). Likewise, recoveries of coliphages with *E. coli* C or C-3000 were consistently higher than those with *E. coli* B for individual samples. The percentage occurrence of coliphages was 50, 46 and 18% for *E. coli* C, C-3000 and B, respectively. Because the relative recovery of coliphages using *E. coli* B was very low for most products, coliphage counts with this host were not used for comparative purposes. Coliphages were detected in 56% of samples using both *E. coli* C and C-3000, whereas coliphages were detected in 50 and 46% of samples using only *E. coli* C and only C-3000, respectively (Table 1). Coliphages infective for *E. coli* C and C-3000 were exclusively detected in twelve and seven samples,

TABLE 2. Summary of coliphage and bacterial analyses of twelve retail foods.

Product ^a	Coliphages ^b				Bacteria		
	Host C	Host C-3000	Host B	<i>E. coli</i> ^c	Fecal coliforms ^c	Total coliforms ^c	APC ^d
Fr. chicken							
Mean	3.62	3.18	1.36	3.32	3.91	4.62	6.23
Range	2.66-4.04	1.78-4.45	ND-2.54	2.54-4.63	3.36-6.63	3.63-6.68	5.04-7.04
Fr. pork							
Mean	2.92	2.76	1.92	3.20	3.42	3.74	5.56
Range	1.00-4.77	1.30-4.38	ND-4.42	ND-4.97	1.56-4.97	1.56-4.97	3.54-8.00
Fr. ground beef							
Mean	2.04	2.34	0.00	3.45	3.53	5.77	7.58
Range	1.00-3.04	ND-2.51	ND-1.30	2.36-5.38	2.36-5.38	3.97-6.38	7.92-8.04
Fr. oysters							
Mean	2.69	2.04	0.60	1.49	2.15	5.23	6.86
Range	ND-4.92	ND-4.90	ND-4.68	ND-2.88	ND-2.88	2.63-6.38	4.30-8.49
Fr. mushrooms							
Mean	1.15	0.78	0.00	0.30	1.36	6.08	8.18
Range	ND-2.26	ND-2.43	ND-1.00	ND-2.36	ND-2.63	5.02-6.38	6.04-8.94
Fr. lettuce							
Mean	0.30	0.70	ND	0.70	1.23	5.18	7.00
Range	ND-2.00	ND-3.76	ND	ND-2.36	ND-4.97	4.63-6.38	6.04-7.45
Chicken pot pie							
Mean	0.60	1.00	ND	0.30	1.66	2.36	3.88
Range	ND-1.78	ND-2.73	ND	ND-1.96	ND-2.88	ND-4.18	3.04-5.30
Biscuit dough							
Mean	ND	0.48	ND	1.18	2.28	2.71	4.57
Range	ND	ND-2.57	ND	ND-3.63	ND-3.63	ND-3.63	3.11-5.04
Deli. loaf							
Mean	0.30	0.00	ND	0.48	1.83	2.74	6.88
Range	ND-1.95	ND-1.48	ND-2.54	ND-3.36	ND-3.36	1.56-3.36	5.46-8.40
Deli. roasted turkey							
Mean	0.60	0.70	ND	0.90	2.32	3.72	5.56
Range	ND-1.60	ND-2.94	ND	ND-3.97	ND-3.97	ND-5.04	4.08-6.45
Pkg. roasted turkey							
Mean	ND	ND	ND	ND	ND	ND	3.18
Range	ND	ND	ND	ND	ND	ND	1.00-7.30
Pkg. roasted chicken							
Mean	0.30	0.00	ND	ND	ND	1.46	6.00
Range	ND-1.00	ND-1.00	ND	ND	ND	ND-4.38	1.00-7.89

^aTen samples analyzed for each product; Fr. = fresh; deli. = delicatessen; pkg. = vacuum packaged.

^bCounts expressed as log₁₀ plaque-forming units (PFU) per 100 g; mean = log₁₀ mean.

^cCounts expressed as log₁₀ most probable number (MPN) per 100 g; mean = log₁₀ mean.

^dAPC, aerobic plate count (20°C); counts expressed as log₁₀ colony-forming units (CFU) per g.

^eND, none detected (<10 PFU/100 g or <30 MPN/100 g).

respectively. Hence, the use of both *E. coli* C and C-3000 was necessary to detect coliphages in 19 (16%) of 120 samples.

Relationship between coliphage and bacterial indicators

The ratios of coliphage:*E. coli*:fecal coliform:total coliform counts based upon geometric mean counts for each product can be derived from Table 2. Wide variation in the ratios of coliphage counts to counts of each bacterial indicator were noted as a function of the food product. Likewise, these ratios were variable among individual samples for most products. Ratios of mean coliphage (*E.*

coli C host) to *E. coli* counts ranged from 1:0.1 in fresh oysters to 1:25 in fresh ground beef, whereas ratios with host C-3000 ranged from 1:0.2 in chicken pot pie to 1:13 in ground beef. Lowest ratios of coliphage to fecal coliform counts were observed with processed delicatessen meats and biscuit dough, whereas highest ratios were noted with fresh oysters.

Coliphage (*E. coli* C) and bacterial indicator counts for all 120 samples were cross-tabulated in order to present an overview of the numerical relationships between these organisms (Table 3). Similar results were noted for coliphages of *E. coli* C-3000. At a cut-off level of 100 col-

TABLE 3. Cross-tabulation of coliphage (*E. coli* C) and bacterial indicator counts for all samples (n = 120).

Bacterial indicator	Bacterial range ^a (MPN/100 g)	Coliphage range ^a (PFU/100 g)			
		<10 ²	10 ² -10 ³	10 ³ -10 ⁴	≥10 ⁴
No. of samples					
<i>E. coli</i>	<10 ²	75	5	0	1
	10 ² -10 ³	9	5	3	0
	10 ³ -10 ⁴	6	1	7	1
	≥10 ⁴	0	4	0	3
Fecal coliforms	<10 ²	57	3	0	1
	10 ² -10 ³	22	5	0	0
	10 ³ -10 ⁴	10	3	10	0
	≥10 ⁴	1	4	0	4
Total coliforms	<10 ³	40	1	0	0
	10 ³ -10 ⁴	22	2	3	0
	10 ⁴ -10 ⁵	4	2	6	3
	≥10 ⁵	24	10	1	2

^aRanges are inclusive at low but not high limit of indicated ranges.

iphages and *E. coli* per 100 g, coliphage and *E. coli* counts were in agreement for 83% of samples. At a cut-off level of 1000 organisms/100 g, coliphage and *E. coli* counts were in agreement for 88% of samples. *E. coli* was detected in 83 and 100% of samples in which coliphage recoveries (either host strain) were ≥100 and 1000 PFU/100 g, respectively (data not presented). Coliphages (either host) were recovered in 91 and 100% of samples in which *E. coli* recoveries exceeded 10³ and 10⁴ MPN/100 g, respectively (data not presented). Coliphages were not detected in two samples, i.e., delicatessen turkey and pickle loaf, in which *E. coli* counts exceeded 10³ MPN/100 g.

At a cut-off level of 100 coliphages and 1000 fecal coliforms per 100 g, coliphage and fecal coliform counts were in agreement for 83% of samples (Table 3). Fecal coliforms were recovered from 95 and 100% of samples having coliphage recoveries (either host) ≥100 and 1000 PFU/100 g, respectively (data not presented). Fecal coliforms were not detected in two samples, i.e., fresh lettuce and delicatessen turkey, in which coliphage counts exceeded 100 PFU/100 g. Coliphages were detected in 84 and 89% of samples having fecal coliform recoveries in excess of 10³ and 10⁴ MPN/100 g (data not presented). Coliphages were not detected in one sample, i.e., fresh lettuce, having a fecal coliform count in excess of 10⁴ MPN/100 g.

Low levels of coliforms (<1000 MPN/100 g) were generally associated with low levels of coliphages (<100 PFU/100 g) (Table 3). However, there was little association between high levels of coliforms (≥10⁵ MPN/100 g) and high levels of coliphages (≥10³ PFU/100 g). Highest levels of coliforms were found in fresh vegetables which had relatively low recoveries of coliphages, *E. coli* and fecal coliforms. Coliforms were detected in 100% of samples in which coliphage recoveries (either host) were >100 PFU/100 g, respectively. Coliphages (either host) were detected in only two of eighteen sam-

ples in which coliforms (≥30 MPN/g) were not detected; coliphage recovery was 10 PFU/100 g in two samples of vacuum-packaged roast chicken, in which no coliforms were recovered (data not presented).

Correlation determinations between coliphage and bacterial counts for samples of each product and all 120 samples were performed. Correlation coefficients (Kendall's Tau measurements) for all 120 samples and products in which significant (P<0.05) correlations were noted are presented in Table 4. For all samples, highly significant (P<0.00001) correlations between coliphages (either host) and *E. coli*, fecal coliform or coliform counts were found. Coliphage counts of *E. coli* C were less significantly correlated (P<0.01) with APC than fecal indicator bacteria, whereas coliphage counts of *E. coli* C-3000 were not significantly (P>0.05) correlated with APC. Overall, coliphage counts (either host) were more strongly correlated with *E. coli* or fecal coliform counts than coliform counts. The overall correlations reflect the concomitant low or high recoveries of coliphages and bacterial indicators in most processed products as compared to fresh products. A highly significant correlation (P<0.00001) between coliphage counts with host strain C and C-3000 also was found for all 120 samples (data not presented).

There were few significant (P<0.01) correlations between coliphage and bacterial indicator counts noted among individual products (Table 4). Among fresh meats, significant correlations (P<0.01) between coliphage and bacterial indicator counts were found only for samples of fresh pork sausage. In samples of fresh oysters, coliphage counts (either host) were significantly correlated (P<0.01) with coliform counts and aerobic plate counts. In samples of chicken pot pie, some correlation was found between coliphage counts (either host) and coliform counts or aerobic plate counts. Some correlation (P<0.05) was noted between coliphage (host C-3000) and *E. coli* counts in refrigerated biscuits.

TABLE 4. Correlation determinations between coliphage and bacterial counts for various foods^{a,b,c}.

Product	Coliphage counts		Bacterial counts		
	Host strain	<i>E. coli</i>	Fecal coliforms	Coliforms	APC (20°C)
Fresh pork	C	0.622**	0.578**	0.622**	0.067
	C-3000	0.689**	0.689**	0.733***	-0.044
Fresh oysters	C	0.200	0.044	0.556**	0.600**
	C-3000	0.200	0.178	0.689**	0.644**
Biscuit dough	C	— ^d	—	—	—
	C-3000	0.444*	0.111	0.200	-0.067
Chicken pot pie	C	-0.178	0.156	0.467*	0.556**
	C-3000	-0.222	0.289	0.489*	0.511*
All products	C	0.381*****	0.412*****	0.335*****	0.158**
	C-3000	0.338*****	0.329*****	0.273*****	0.089

^aValues represent measurement of rank correlation (Kendall's Tau values).

^bCorrelation analyses were performed on products having at least three positive samples for coliphages of either *E. coli* host strain; ten samples were analyzed for each product and 120 samples overall.

^c*, **, ***, **** and ***** indicates significance at the 0.05, 0.01, 0.001, 0.0001 and 0.00001 levels, respectively.

^d—, no recovery.

DISCUSSION

The overall incidence and recovery level of coliphages and *E. coli* were similar in 120 samples examined. Likewise, the highest recoveries of coliphages and *E. coli* were found in fresh meats and oysters, whereas the lowest recoveries were noted in vacuum-packaged processed meats. Similar trends were noted for fecal coliforms. Levels of coliphages (*E. coli* C host) recovered in fresh chicken breasts and pork sausage in the present study were similar to or slightly higher than those reported in a previous study (24). Recoveries of coliphages with all host strains of *E. coli*, including *E. coli* B, from retail fresh oysters, ranged from <10 to 8.4×10^4 PFU/100 g in this study as compared to <2 to 17 PFU/100 g with *E. coli* B in a study by Kott and Gloyna (27).

The overall recoveries of coliphages with *E. coli* C and *E. coli* C-3000, a male strain, were strongly correlated ($P < 0.0001$) and very similar with regard to numbers of PFU detected and incidence of recovery (Tables 1 and 2). Recovery of coliphages with either *E. coli* C or C-3000 was consistently higher than those with *E. coli* B. *E. coli* C and C-3000 also have been reported to recover higher numbers of PFU from water and wastewater than *E. coli* B and various other host strains (14,18,20,47). Although recoveries of coliphage with host strain C and C-3000 were analogous overall and for products with high coliphage levels, there was discordance between host strain C and C-3000 in 19 of 120 samples with regard to coliphage detection. Additionally, the distribution of high, mid and low temperature coliphages recovered with each of these host strains generally differed (26). These differences may be attributed to the F plasmid in host strain C-3000 as well as the presence of receptors for different types of coliphages on each host strain (17,18,43). These results indicate that use of both *E. coli* C and C-3000 host strains would provide more represen-

tative recoveries of coliphages than either host alone for sanitary quality evaluations of foods as well as for ecological or taxonomic studies of coliphages in foods.

Based upon linear regression analysis, highly significant quantitative correlations and predictive models between coliphage and total or fecal coliform counts in large numbers of water samples have been reported (23,49). Regression analysis between coliphages and bacterial indicators could not be applied to most of the present data due to the large number of samples in which either coliphages, *E. coli* or fecal coliforms were not recovered. Some numerical relationships between coliphages and *E. coli* or fecal coliforms were noted. For example, at a cut-off level of 100 coliphages and *E. coli* per 100 g, coliphages of host strain C and *E. coli* counts were in agreement for 83% of 120 samples. Although linear or predictive relationships between coliphage and bacterial indicator counts could not be established, a strong overall association between coliphage and bacterial indicators was indicated by nonparametric correlation analyses (Table 4). Coliphage counts with either host were strongly correlated ($P < 0.00001$) with *E. coli*, fecal coliform and total coliform counts and more strongly correlated with *E. coli* and fecal coliform than with total coliform counts. However, few significant correlations ($P < 0.05$) between coliphage and bacterial indicator counts were noted among samples of individual products. Kott and Gloyna (27) reported significant correlations between coliphage and coliform counts in oyster samples obtained from estuarine waters at different distances from a sewage outfall but did not examine oysters for fecal coliforms or *E. coli*. Coliphage and total coliform counts in oysters were strongly correlated in the present study.

The lack of significant correlations between coliphage and bacterial indicator counts in most products was primarily attributed to the relatively low numbers of samples examined for each product, i.e., ten samples, and the low incidence of coliphages and *E. coli* or fecal col-

iforms in many products. If coliphage indicator systems are to be applied to specific foods, further studies involving more representative numbers of samples are indicated. Other factors which could contribute to a lack of correlation between coliphage and certain bacterial indicator counts in some products include: (a) failure to recover all coliphages or *E. coli* with the method used, (b) the inherent lack of precision associated with the MPN technique, (c) ability of many coliform bacteria to proliferate at refrigeration temperature (33,35), (d) the inability of coliphages to proliferate at temperatures <15°C or in the absence of suitable *E. coli* host bacteria (42), (e) the greater environmental resistance of most coliphages as compared to enteric indicator bacteria (3,12,20,21), and (f) the presence of saprophytic coliforms as well as certain fecal coliforms associated with plants and the environment (13,32). The correlation observed between coliphages and coliform or APC as well as that between *E. coli* and APC in fresh oyster samples suggests that conditions suitable for proliferation of bacterial indicators and coliphages, i.e., presence of suitable *E. coli* hosts and temperatures $\geq 15^\circ\text{C}$, existed for these samples or the waters from which they were obtained.

The public health significance of bacterial indicators, including *E. coli*, in various foods has generally been difficult to establish or define, particularly with regard to their use as indices of food safety, i.e., presence of enteric pathogens (13,30,32,46). The sanitary importance of a particular indicator bacterium in a food product is generally unique to that product with regard to its origin or raw materials, the degree and type of processing or handling to which it is exposed, and its storage or packaging conditions. The sanitary significance of coliphages in a particular food must be considered in the same manner. When coliphages isolated from samples examined in this study were characterized with regard to temperature of infectivity, a relatively high proportion of phages, i.e., HT types, of apparent fecal origin were found in all products examined (26). Although other types of coliphages, i.e., MT or LT types, are capable of proliferation at temperatures as low as 15°C (42), the possible proliferation of these coliphages in most food products imply insanitary conditions with regard to the presence of suitable *E. coli* hosts and improper holding temperatures.

The indicator value of coliphages in thermally processed foods has been questioned (8) because coliphages as a group are very heterogeneous with regard to heat stability and some coliphages are more heat resistant than enteric bacteria or animal viruses (1,10,38,40). However, comparative recoveries of coliphages and bacterial indicators in processed meat samples examined in this study do not indicate survival of heat-resistant phages. Further studies directed toward survival of indigenous coliphages in various foods during processing are indicated to substantiate these observations. However, coliphage indicator systems do not appear as sensitive as some bacterial indicators, e.g., aerobic plate counts or total coliform counts, for monitoring process or handling safety of many ther-

mally processed products.

The strong overall correlation between coliphage and *E. coli* or fecal coliform counts in this investigation as well as the ecological specificity of a large percentage of coliphages isolated from all food types in further studies (26) suggest that many coliphages found in foods are associated with fecal contamination. These results also indicate that coliphages are widely distributed among various food products. Although coliphage analyses used in the present study were completed in ca. 16 h, much shorter assay times (4 to 6 h) have been reported effective for enumeration of coliphages in water and wastewater (49). Similar rapid coliphage assays may provide an alternative to conventional fecal indicator analyses in many food products. Because these results are based on a limited number of samples for a single product type, further studies are indicated to realistically apply a rapid coliphage indicator system to sanitary assessments for any specific food product.

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