and fruit salads in pleated portion cups of the appropriate sizes, holding them on trays in the refrigerator until needed. Left-overs, too, can be economically saved for later use by refrigerating or freezing them in wax or plastic-coated paper containers.

An excellent sanitation technique that also saves staff time is to use paper cups for water. A small stack of cups (upside-down with a napkin underneath) and a water pitcher can be put on each table so customers can serve themselves.

If this kind of idea appears to be useful in talking to restaurant and other operators, it should not be hard for each sanitarian to build a broad inventory of ideas of all kinds from his own observations once he starts to keep an eye out for them.

This technique was pointed up by the featured speaker at a convention of advertising experts who make their living by getting people to buy things by mail. He was heckled with one piercing question: "What *single thing* can I say to sell more goods?"

Without a moment's hesitation, the speaker flashed back: "Don't tell people how good your goods are. Tell them how good your goods make them." To this man who had sold millions of dollars worth of merchandise this was the strongest action-getting approach that can be devised.

Although sanitarians are not in the direct mail business, there is a lot to be learned from this homely anecdote because it shows how anyone can increase his effectiveness – lengthen his reach – by spending additional time thinking how to help the people he deals with to be as good as they want to be.

This is the finest kind of enforcement – and the strongest in the long run.

# FURTHER STUDIES ON THE SELECTIVITY OF VIOLET RED BILE AGAR[1,2]

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Direct plating on selective media is the method of choice for the routine enumeration of coliform bacteria in most frozen foods (*4, 8*), yet little is known regarding the selectivity of these media under various conditions of use. It is often desirable, and sometimes essential, to know what types of bacteria are being counted when the red colonies on a plate are being tallied. The problem becomes more acute when only a few colonies per plate can raise serious questions regarding the sanitary history of the product.

During the course of a recent project[2], opportunity was afforded to study factors which influenced the coliform count of several different types of frozen foods. Some pertinent data are reported here to serve as a basis for a better understanding of the meaning of the coliform count of frozen foods determined on Violet Red Bile (VRB) Agar.

## MATERIALS AND METHODS

The plating procedures have been described previously (*5*). Pie samples were taken from the contents only, unless specific mention was made that crust was included. Purplish-red colonies 1 mm. or more in diameter were termed "typical" colonies (*3*), while purplish-red colonies of less than 1 mm. diameter were called "small" colonies. The results would not have differed materially had an arbitrary colony size of 0.5 mm. diameter (*11, 12*) been used, since a considerable proportion of the small colonies were less than 0.5 mm. in diameter. Isolated colonies were streaked on Eosin Methylene Blue Agar, then the plates were incubated for 24 hours at 37C. This method had been found to be about 90% effective for presumptive identification of *Escherichia* from VRB Agar when compared with isolation followed by IMVIC tests (*5*).

## RESULTS AND DISCUSSION

Some previously reported results obtained on chicken pies (*5*) are included in Part A of Table 1 for comparative purposes. Approximately 67% of the typical colonies and 35% of the small colonies were identified as *Escherichia* when the sample was devoid of crust. When a representative portion of crust was included in the samples (B, Table 1), only 33% of the typical and 13% of the small colonies from chicken pies were confirmed as *Escherichia*. Lesser proportions of the typical colonies from turkey and beef pies were confirmed as *Escherichia*, but the pro-

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TABLE 1—PRESUMPTIVE IDENTIFICATION OF *Escherichia* FROM COLONIES ON VIOLET RED BILE AGAR PLATES.

| Product | "Typical" | | | "Small" | | |
|---|---|---|---|---|---|---|
| | No. Tested | No. Pos. | % Pos. | No. Tested | No. Pos. | % Pos. |
| A. Chicken pies, no crust[a] | 61[b] | 41 | 67 | 51 | 18 | 35 |
| B. Chicken pies, with crust | 40 | 13 | 33 | 103 | 13 | 13 |
| Turkey pies, with crust | 20 | 5 | 25 | 87 | 11 | 13 |
| Beef pies, with crust | 22 | 4 | 18 | 83 | 8 | 10 |
| Total, pies with crust | 82 | 22 | 27 | 273 | 32 | 12 |
| Crust only | 25 | 0 | 0 | 52 | 7 | 14 |
| C. Dinners — meat, gravy | 30 | 9 | 30 | 13 | 5 | 38 |
| Dinners — vegetables | 23 | 13 | 57 | 27 | 8 | 30 |
| Macaroni-cheese | 23 | 12 | 52 | 19 | 12 | 63 |
| Cream pies | 49 | 9 | 18 | 37 | 8 | 22 |
| Raw milk[a] | 42 | 4 | 10 | 0 | — | — |

[a]Data from reference 5.
[b]Includes 31 colonies from VRB Agar plates and 30 colonies from deoxycholate agar plates.

portion of small colonies which proved to be *Escherichia* did not vary with the type of pie. When crust alone was plated, none of 25 typical colonies examined were *Escherichia*, but 14% of the small colonies gave characteristic *Escherichia* reactions on EMB agar. Ingredients used in formulating the crust were examined in an effort to determine whether the small colonies arose from bacteria which were incorporated into the dough or from bacterial contamination of the exterior of the pie. Samples of flour obtained from a processor contained bacteria, including *Escherichia*, which formed small colonies on VRB agar.

The influence of crusts on the coliform counts obtained was determined on 6 brands of pies, some of which were purchased through retail channels. Total counts (Trypticase Soy Agar, 2 days at 32 C.) and enterococcus counts (Thallous Acetate Agar, 2 days at 37 C.) were included in Table 2 in order to demonstrate relative effects on these bacterial populations. Each pie was subjected to a dual sampling procedure whereby a representative portion of crust was included in one sample, while a companion sample was essentially free of crust. The data are arranged so that parallel counts occupy adjacent horizontal locations in Table 2.

Inclusion of the crust material in the sample resulted in a substantial (5 to 10 fold) increase in the apparent coliform count, while total and enterococcus counts were not materially affected (Table 2). In another brand of chicken pie (not shown in Table 2), only 1 colony was found when 24 samples were analyzed without crust. When crust was included as part of the sample, coliforms were recovered from 12 pies in quantities which ranged from 5 to 325 per gm. A third brand of pie yielded only 8 of 24 positive samples, all of which had coliform counts of less than 10 per gm when analyzed without crust. When crust was included as part of the samples, coliforms were recovered from 20 of the samples; 10 of the samples contained 10 or more coliforms per gram. These results indicate that quality control must include examination of the constituents used in the formulation of crusts for pot pies and similar frozen foods. Apparently some processors are not cognizant of this fact.

As shown in Table 2, the "crust count" obscured the coliform content of the interior of the products in many brands and lots of pies. Thus, if the coliform count is used as an indication of the sanitary history of the food, and therefore, the possible presence of enteric pathogens which might endanger the consumer, then the intended objective has not been attained in many laboratories. Gram negative organisms which remained after cooking (2), or after even gross undercooking, would most likely be present in only the central portions of the product. It would be wise to exclude crust material from the sample taken, therefore, in order to gain more knowledge of the coliform content of the interior of the pie, or to plate crust and contents separately.

Reference to the data in Table 1, and especially to Section C of the table, brings to light several points which are worthy of discussion. The vegetable portion of frozen dinners yielded a higher percentage of confirmed typical colonies and about the same percentage of confirmed small colonies than the meat portion of the same dinners. In macaroni-cheese

plates, a considerable portion of both typical and small colonies were *Escherichia*. There was a low confirmation of *Escherichia* from cream pies: about 20%, whether or not the crust was included in the sample. (It is of interest to note that two types of the cream pies examined were of the thaw and serve variety.) In contrast, a low percentage of typical colonies from samples of raw milk were confirmed as *Escherichia* (5). These data clearly demonstrate that the proportions of *Escherichia* included in the coliform count can differ greatly, depending upon the food examined and the conditions of estimation.

As stated previously (5), "it would appear that the term 'coliform count' when applied to frozen pot pies and related products falls within the definition of 'coliform count' as accepted by the dairy bacteriologists," however, one must not lose sight of the product being examined. Recently, Kereluk and Gunderson (8) stated, "Whether or not it is necessary to determine if foods are contaminated with fecal or nonfecal strains of coliform bacteria, the presence of coliform bacteria in frozen foods might indicate whether the foods had been cooked insufficiently or that they were contaminated after cooking or during processing prior to freezing." Until nonfecal coliforms are shown to indicate contamination from a "dangerous source" there is no reason to believe that these types of coliform bacteria, just because they form a colony on VRB Agar, should be considered any less desirable in pot pies than other innocuous microorganisms. Huber *et al.* (7) noted that accompanying a higher total count, greater quantities of coliforms, enterococci and coagulase positive staphylococci could be found in chicken pies. Hartman (unpublished data) found correlations between coliform and total counts and, especially, between enterococcus and total counts in commercial samples which were plated within a short period of production. The coliform-total correlation no longer existed in samples purchased through retail channels (*see also refs. 1, 4*). The coliform count is obviously of little value when performed on samples of unknown history of storage. Litsky *et al.* (9) have commented on some of the other short-comings of coliforms as indicators of pollution. A question then arises. Does the coliform count, or any other

TABLE 2—BACTERIAL COUNTS ON BRAND B WHEN SAMPLED WITH AND WITHOUT CRUSTS.

| Count Per Gram of Sample | | | | | |
|---|---|---|---|---|---|
| With Crusts | | | Without Crusts | | |
| Coliforms | Enterococci | Total Count | Coliforms | Enterococci | Total Count |
| 5 | 140 | 4,200 | 0 | 140 | 8,400 |
| 65 | 260 | 8,800 | 9 | 75 | 2,400 |
| 30 | 110 | 2,300 | 0 | 130 | 9,700 |
| 140 | 270 | 3,800 | 0 | 630 | 7,600 |
| 40 | 610 | 5,800 | 10 | 290 | 4,500 |
| 30 | 290 | 2,200 | 0 | 370 | 7,200 |
| 80 | 140 | 3,700 | 0 | 300 | 1,800 |
| 35 | 400 | 3,500 | 0 | 220 | 3,600 |
| 80 | 3,000 | 11,000 | 0 | 390 | 3,800 |
| 130 | 58 | 2,300 | 8 | 280 | 3,800 |
| 30 | 55 | 3,600 | 0 | 260 | 4,500 |
| 65 | 340 | 3,300 | 0 | 40 | 1,900 |
| 88 | 1,200 | 12,000 | 0 | — | 9,800 |
| 62 | 270 | 5,500 | 0 | 120 | 3,100 |
| 30 | 350 | 8,000 | 14 | 2,200 | 5,800 |
| 17 | 2,000 | 7,800 | 0 | 770 | 6,400 |
| 60 | 2,700 | 10,000 | 15 | 290 | 7,300 |
| 15 | 220 | 2,000 | 0 | 280 | 2,100 |
| 150 | 120 | 5,100 | 8 | 250 | 6,000 |
| 25 | 310 | 2,500 | 0 | 120 | 1,300 |
| 65 | 210 | 3,600 | 13 | 410 | 10,000 |
| 8 | 170 | 28,000 | 0 | 130 | 4,600 |
| 72 | 20 | 1,900 | 13 | 120 | 1,800 |
| 32 | 110 | 2,000 | 0[a] | 160 | 1,400 |
| 57 | 560 | 6,000 | 4 | 350 | 5,000 |

[a]less than 3.

selective count, yield additional information of value for routine purposes than that already obtained by the total count? An increasing body of evidence which is accumulating indicates that once the total count is reduced to a satisfactory level, the other counts seem to fall in line. When simplicity of analysis is sacrificed, as it must be in confirmation of coliform type or determination of coagulase reaction, then the import of the information obtained should justify the effort expended. Sample to sample variations in all counts are such that, in general, effort might better be expended in analyzing many samples for total counts, rather than fewer samples for selected groups of microorganisms.

More information also should be known about the influence of various factors affecting the test (*13*) and modifications in formula on the efficacy of the medium. For example, Thomas *et al.* (*13*) reported that VRB Agar can be stored in prepared form (steamed) for up to 3 weeks before use, but no reports have appeared on the effect of storage at room temperature on the efficacy of autoclave sterilized medium. Furthermore, the specificity of the medium, thus the import of the coliform content of various non-dairy foods, might be increased by utilizing a medium similar to that described by Mossel (*10*), yet modified further (*6*), in order to retard growth of a major portion of the less significant gram negative bacteria.

## SUMMARY

The colony count of frozen pot pies on Violet Red Bile (VRB) Agar was influenced greatly by inclusion of crust in the sample or omission therefrom. The proportions of *Escherichia spp.* included in the VRB agar counts differed greatly, depending upon the food tested and upon the conditions of use of the medium. The significance of these results was discussed in the light of the product being examined.

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