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Microflora of Prepackaged Luncheon Meats

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TABLE OF CONTENTS

Introduction	3
Sources of Experimental Samples	4
Analytical Methods	5
Procurement of Samples	5
Bacteriological Analysis	5
Results and Discussion	7
Quantitative Microbiological Flora and pH	7
Consumer Acceptability of Samples	10
Factors Influencing the Quantitative Microflora	11
Qualitative Microflora	13
Factors Influencing the Qualitative Microflora	18
Summary and Conclusions	19
Literature Cited	20

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R. F. BROOKS AND R. L. HENRICKSON*

INTRODUCTION

New prepackaging methods for fruits, vegetables, meats, and other commodities have resulted in a marked change in the buying habits of housewives. This development also has raised a number of questions regarding the influence of various packaging materials and methods upon characteristics of food products which may affect their acceptance by consumers.

In the prepackaging of meats, for example, variation was noted in the effects of different packaging materials on the retention of desirable color in meats and meat products during storage in the retail display case. The present practice of using different wrapping materials for prepackaged fresh meats and for luncheon meats is largely a result of these practical observations.

Methods and materials used in the fabrication and handling of prepackaged luncheon meats would be expected to influence the qualitative and quantitative microbiological flora. The differences in flora, in turn, should affect the stability of these products during their life in store display cases and home refrigerators.

The purpose of this study was to determine: (1) the "typical" microflora of various prepackaged table-ready meat products in the retail trade; and (2) the influence (if any) of such microflora upon the general stability characteristics of these products.

Luncheon meats were selected for study, since the materials and methods used in their fabrication represent the ultimate in opportunity for bacterial contamination. These products are made largely from packing-house trimmings, which require more processing and handling than the meat cuts. Several of the common luncheon meats contain considerable amounts of spices and other flavoring and seasoning ingredients, which are notor-

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ious sources of bacterial contamination. Finally, there is an unfortunate tendency on the part of many wholesale and retail dealers to regard luncheon meats as the "poor cousins" of the meat industry. They often get less care in handling, refrigeration, storage, fabrication, and distribution. It is perhaps surprising that the general quality of these products is as high as it is, considering some of the conditions to which they are subjected.

Very little investigation has been carried out on the microbiology of prepackaged luncheon meats. Only one reference was found to such a study. Steinke and Foster (10) studied the effects of various storage temperatures on bacterial growth and pH in liver sausages and bologna packaged in Saran. They found that these products retained their normal appearance and showed no appreciable increase in bacterial population and no change in pH when stored at 51°F. (10°C.) or lower for 10 weeks. After 12 and 15 weeks, bologna stored at 51°F. (10°C.) showed a progressive increase in bacterial count, but no pH change. Bacteria in liver sausage after 13 weeks at 51°F. (10°C.) dropped to about one-third of the initial count. At a storage temperature of 61° F (16° C), bacterial growth in both products was delayed for about two weeks, but soon thereafter it rose to several millions per gram. At higher temperatures, bacterial multiplication was very rapid, rising from a few thousand to several millions per gram in three to four days. The predominant organisms in both products, both initially and terminally, were aerobic micrococci. *Bacillus* spp. were also observed in liver sausage held at higher storage temperatures, eventually becoming predominant in the sausage held at 86°F. (30°C.) Their presence in liver sausage while absent in bologna, was ascribed to a difference in the initial flora (contamination) of the two products.

SOURCES OF EXPERIMENTAL SAMPLES

Fifty random samples of each of seven types of prepackaged luncheon meat have been examined bacteriologically, viz.: (1) bologna, (2) pickle-pimiento loaf, (3) spiced luncheon meat, (4) chopped ham loaf, (5) salami, (6) New England brand sausage, and (7) souse. These seven products were selected for study because they represented the largest volume of table-ready meats available in the retail trade.

The 350 samples (Table 1) were obtained at random from seven retail stores within a radius of 50 miles of the laboratory over a period of one year.

By including the indicated variety of products, store sources, and packaging procedures in the study, it was felt that the information obtained would be representative of the average situation in the prepackaged luncheon meat field.

ANALYTICAL METHODS

Procurement of Samples:

Samples were purchased from the retail stores at random (Table 1) and placed in a refrigerated carrying case for return to the laboratory. In the case of local stores (I, II, and III), samples were analyzed on the day of purchase with a maximum holding period of one hour between removal from the store display case and sample preparation in the laboratory.

TABLE 1 -- SOURCES OF PREPACKAGED LUNCHEON MEATS PURCHASED FROM STORES HAVING EITHER COMPLETE OR PARTIAL SELF-SERVICE.

Store No.	Degree of self-service*	Packaging practice	No. of samples
I	Complete	Store-prepackaged	76
II	Partial	Prepackaged by suppliers	21
III	Complete	Part in store under refrigeration; part by suppliers	74
IV	Complete	Store-prepackaged	30
V	Partial	Store-prepackaged	48
VI	Partial	Prepackaged by suppliers	36
VII	Partial	Mixed-store and suppliers	46
Other**			19
Total			350

*"Complete" self-service = all meats prepackaged for retail sale.

"Partial" self-service = specialty items only prepackaged for retail sale.

**The 19 samples in this category were obtained from a miscellaneous group of stores outside the regular sampling area. Since only three or four samples were obtained from any one of these stores, the group is treated as one "miscellaneous" source.

When samples were purchased outside the local area (Stores IV, V, VI, & VII), two or more towns were visited during a half-day. The samples were returned to the laboratory under refrigeration, held in a standard refrigerated display case at 36°F. (2°C.) overnight, and analyzed the following day, with a maximum holding period of 24 hours.

In selecting the samples from the retail display case, no attempt was made to choose the freshest or best-appearing samples. Complete randomization of sample selection was practiced in order to determine the average or normal microbiological condition of the products. In some instances, coding of the packages was practiced by the store or supplier to indicate packaging date. This information was later used to relate the "age" of samples to their bacterial content and general acceptability.

Bacteriological Analysis:

Approximately 50 grams of the product, estimated by proportion of the total package weight, were taken aseptically by cutting off a section

of the pile of sliced product, and placed in a tared sterile Waring Blendor jar. The exact weight of sample was determined by difference. This method of sampling was preferable to that of weighing out exactly 50 grams, which would involve considerably more exposure and handling of the product and, consequently, much greater opportunity for contamination. The average accuracy of estimation of the 50-gram sample size is indicated by the mean of sample weights—49.8 grams, with a standard deviation from the mean of 6.5 grams. Seventy-two percent of the samples were within the weight range of 43.3 to 56.3 grams (mean minus and plus its standard deviation).

To the weighed sample in the blendor jar was added 100 ml. of sterile distilled water, to give an approximate three-fold dilution; the sample was then blended for three minutes to produce a slurry which could be pipetted for further serial dilution. Preliminary tests showed that a minimum of three minutes blending time was required for adequate disintegration and dispersal of the sample to produce a smooth slurry free from large particles.

Aliquots of the diluted slurry were plated in the usual manner, using tryptone-glucose-beef extract agar (Difco Laboratories, 4). Duplicate sets of plates of each sample were prepared in order to allow incubation at 98° F (37° C) and 59° F (15° C). Immediately after completing the preparation of the bacteriological plates, the pH of the meat slurry was determined by the Beckman Model G pH meter.

After incubation for two days at 98° F (37° C) or five days at 59° F (15° C), appropriate plates were counted and the number of bacteria per gram of original sample computed by the application of the proper dilution factor. The dilution factor was a combination of the original dilution in the slurry and that of the further ten-fold dilutions used in preparing the plates. At the time of counting the plates, the relative abundance of the various colony types appearing on the plates was noted, and representative pure cultures were isolated for further study and characterization.

For the purpose of identification of the pure cultures isolated from the luncheon meat samples, the following characteristics were determined: morphology (shape, size, and grouping of cells); Gram stain reaction; motility; sporulation; type of growth on agar stroke; agar colony form; growth pattern and final pH in nutrient broth; nitrate reduction; chromogenesis; indol production from tryptone broth; hydrogen sulfide formation; digestion of gelatin and casein; action in litmus milk; starch hydrolysis; and acid and/or gas production from glucose, fructose, galactose, lactose, maltose, and sucrose.

These tests were performed routinely on all cultures. While it was

not possible to identify the cultures positively to the species level in all cases, due to normal variation in characters of strains freshly isolated, in the majority of instances an identification of the culture as "similar, if not identical, to" a given species could be made. In a few cases, certain special characteristics were determined, such as catalase production, which have been shown to be of particular value in dealing with special groups of bacteria.

In keying out the cultures, the accepted arrangement as given in the 6th edition of Bergey's Manual (Breed et al., 2) was used; in addition, the alternate generic key developed by Skerman (9) was used as a check on the placing of a culture at the genus level. In all cases, the two keys led to the same generic identification.

RESULTS AND DISCUSSION

Quantitative Microbiological Flora and pH

Data obtained from the quantitative study of the microflora, and from the pH measurements, are summarized in Table 2. As would be expected, the psychrophilic counts (15° C incubation) averaged considerably higher than the mesophilic (37° C) counts; this was interpreted as a reflection of the selective action of refrigeration, during fabrication and storage of the product, in perpetuating or stimulating the development and growth of low-temperature flora, while simultaneously inhibiting mesophilic organisms.

There was a direct, statistically significant relationship between the mesophilic (37° C) and psychrophilic (15° C) bacterial populations (Table 3), the latter being quite consistently larger. The values found for the coefficient of correlation, based on actual bacteria counts at the two incubation temperatures, are highly significant for all products except New England brand sausage, and are moderately significant in the case of that product. Rank correlation coefficient values are highly significant for all products. These values bear out the general observation made throughout the study that the bacterial populations reflect the selective influence of refrigeration on the relative levels of mesophilic and psychrophilic microflora.

In the pickle-pimiento loaf, chopped ham loaf, and New England brand sausage, an appreciable part of the high mean counts at both 15° C and 37° C was contributed by relatively few samples having unusually high bacterial populations, compared with the remaining samples.

The same influence was noted in the 37° C counts only on salami.

TABLE 2 -- QUANTITATIVE MICROFLORA AND pH OF 350 SAMPLES OF PREPACKAGED LUNCHEON MEATS

Product	Aerobic Bacteria /g. product						pH		
	15°C. incubation			37°C. incubation			Minimum	Maximum	Mean*
	Minimum	Maximum	Mean	Minimum	Maximum	Mean			
Bologna	290	36,000,000	1,810,000	290	3,980,000	104,000	5.80	6.78	6.32
Pickle-pimiento loaf	300	150,000,000	8,660,000	660	150,000,000	4,740,000	4.91	6.87	5.81
Spiced luncheon meat	285	53,300,000	1,220,000	280	7,000,000	154,000	6.10	6.70	6.31
Chopped ham loaf	550	312,000,000	9,570,000	525	31,000,000	1,566,000	5.68	6.60	6.18
Salami	900	40,000,000	6,940,000	900	85,000,000	6,830,000	5.40	6.60	6.13
New England brand sausage	730	267,000,000	18,800,000	280	28,700,000	4,470,000	5.57	6.65	6.06
Souse	310	8,760,000	704,000	280	3,630,000	197,000	4.20	4.88	4.53

*Mean values were determined after converting pH to H-ion concentration.

TABLE 3 -- SUMMARY OF CORRELATION COEFFICIENT VALUES - 37°C. VS. 15°C. BACTERIA COUNTS.

Product	Correlation coefficient based on:	
	Actual bacteria counts on individual samples $r \pm P.E.r$	Ranks of bacteria counts on individual samples $pr \pm P.E.pr$
Bologna	+0.622±0.045	+0.807±0.033
Pickle-pimiento loaf	+0.912±0.016	+0.849±0.027
Spiced luncheon meat	+0.994±0.001	+0.759±0.040
Chopped ham loaf	+0.685±0.052	+0.803±0.035
Salami	+0.809±0.033	+0.903±0.018
New England brand sausage	+0.316±0.086	+0.818±0.032
Souse	+0.522±0.069	+0.757±0.041

NOTE: All "r" values significant at odds >99:1, except New England brand sausage, for which odds are >19:1 but <49:1. All "pr" values significant at odds >99:1.

Its mean value was the highest of any product analyzed (Table 2), and the maximum count was exceeded in only one other product—pickle-pimiento loaf. A rather large proportion of the samples (13 out of 50) showed mesophilic counts greater than 1,000,000 per gram, and in eight of them it was greater than 10,000,000, which accounted for the high average count. A possible explanation for the frequency of high counts is the variety and amount of species used in this product, since natural spices are highly contaminated in many cases (6).

At 15° C incubation temperature, the average count lay in the middle of the range for all products, indicating that the majority of the assumed contaminants entering the product on the spices were of the mesophilic type. The ratio of mesophilic to psychrophilic counts was the highest in salami of any of the seven products, closely approaching unity, and further supported the previous assumption of a "preferential" contamination by mesophiles.

The maximum counts found in souse were the lowest among the products studied, as was the mean 15° C (psychrophilic) count. Although two products—bologna and spiced luncheon meat—had somewhat lower mean counts at 37° C, the differences were not outstanding, and it was concluded that the over-all bacterial population of souse was the lowest among the products analyzed. A logical explanation of this fact may be indicated by the lower pH values found for this product (Table 2). It had by far the lowest pH level, which undoubtedly exerted an inhibitory effect on the survival and continued growth of many contaminating bacteria. Perhaps the most striking observation in regard to the pH readings on this product was their uniformity, which was unexpected in view of the heterogeneity of constituents in souse.

The pH data (Table 2) indicate that, with the exception of souse, the acidity of the products was insufficient to exert a marked preservative

effect. The general similarity in range and mean of pH values for these products would seem to indicate that the relatively large variations in their bacterial counts could not be attributed to differences in product acidity. Scattergraphs of pH vs. bacterial counts (at both 15° C and 37° C incubation) failed to reveal any relationship between these characteristics.

The bactericidal or bacteriostatic properties of certain spices and flavoring ingredients, some of which are normal constituents of the products tested, were not noticeably reflected by low bacterial counts in these products. In fact, as noted previously, spices may actually have been a major source of bacterial contamination in highly-spiced products such as salami.

Consumer-Acceptability of Samples

Formal taste panel studies of the consumer-acceptability of each sample were not made, but all of the samples were examined organoleptically at the time of plating. Of the 350 packages sampled, 343 were tasted and judged to be palatable, although there was some variation in their degree of palatability.

The remaining seven samples (Table 4) were judged unpalatable due to undesirable ("spoiled") odor, accompanied in one case (chopped ham

TABLE 4 -- BACTERIAL POPULATION LEVELS IN SEVEN UNACCEPTABLE LUNCHEON MEAT SAMPLES

Product	No. of samples	Bacteria count per gram	
		15°C.	37°C.
Pickle-pimiento loaf	2	15,700,000	28,000,000
Chopped ham loaf	1*	312,000,000	150,000,000
Salami	2	9,250,000	13,800,000
		32,000,000	5,700,000
New England brand sausage	2	4,250,000	7,800,000
		267,000,000	10,200,000

*Sample showed visible bacterial growth on product.

loaf) by visible bacterial growth or "slime" on the surface of the meat. These samples were not tasted. Other workers (1, 3) have shown that deterioration in meat products is commonly first noticed by off-odor production, followed by development of "slime" or visible bacterial growth.

Comparing the data of Table 4 with those of Table 2, in three of the unacceptable samples the 15° C count was the maximum recorded for that particular product. For the 37° C count, this was true in only one of the seven samples. The bacterial counts (at one or both incubation temperatures) in other samples, while not the highest found for the product, were well above the mean count, and were believed to have played a major role in rendering the samples unacceptable.

The two unacceptable samples of pickle-pimiento loaf were obtained from the same store on the same day, and were known to have been packaged and placed in the retail case six days previously. These were the only unpalatable samples whose "age" was determinable. The reason for the wide discrepancy in bacterial counts between these two supposedly duplicate samples is not known, but might have been some unusual difference in initial contamination during handling and packaging, or possibly the removal of the higher-count package from the refrigerated case for a period of time and its subsequent return.

Although the number of samples involved was small, and by no means statistically significant, the results support the observation made throughout the study, that the psychrophilic organisms are probably of greater importance than the mesophiles, so far as stability of the product is concerned.

Factors Influencing the Quantitative Microflora

1. *Age of Sample:* Although the "age" of the samples (length of time they had been in the display case since packaging) was not known for a sufficient number of samples to allow statistical analysis, a marked trend was noted for samples more than a week old to show appreciably larger bacterial populations than samples which had been in the case for less than a week. This relationship was particularly evident in the case of the psychrophilic counts. It supports the generally accepted rule-of-thumb used by market managers, that packages not purchased from the display case within a week should be removed and discarded because of potential spoilage or loss of consumer acceptability.
2. *Time of Sampling:* In certain products the quantitative populations seemed to be related to the season of the year during which the samples were analyzed. But this apparent relationship was not consistent for all products and the isolated instances in which it was found could not be explained, other than theoretically. Thus, the total counts on bologna, spiced luncheon meat, chopped ham loaf, and souse tended to show a lower level during the warm months (April—September) than during the remainder of the year. No such trend was noted in the remaining products. A logical explanation for this relation is not readily apparent. Possibly, during the warmer months of the year more care is taken to ensure adequate refrigeration of the product to guard against spoilage, but this would not explain why the lowered counts were not found in all products.
3. *Packaging Material:* For three of the products, it was possible to compare the bacteria counts found in conventionally-wrapped (cellophane)

TABLE 5 -- QUANTITATIVE MICROFLORA IN CONVENTIONAL VS. VACUUM-PACKAGED LUNCHEON MEAT SAMPLES

Product	Wrap*	No. of samples	Aerobic bacteria per gram of meat					
			15°C. incubation			37°C. incubation		
			Minimum	Maximum	Mean	Minimum	Maximum	Mean
Pickle-	C	42	300	150,000,000	8,910,000	660	150,000,000	4,960,000
pimiento loaf	V	8	100,000	26,800,000	7,330,000	10,600	26,000,000	3,570,000
Salami	C	43	900	40,000,000	7,440,000	900	85,000,000	7,370,000
	V	7	1,640	15,500,000	3,890,000	2,130	21,000,000	3,550,000
New England brand sausage	C	23	730	267,000,000	15,100,000	280	11,800,000	1,350,000
	V	27	156,000	34,000,000	22,000,000	19,400	28,700,000	7,130,000

*C - conventional cellophane wrap

V - vacuum-packaged

TABLE 6 -- INFLUENCE OF SAMPLE SOURCE ON THE TOTAL AEROBIC MICROFLORA OF PREPACKAGED LUNCHEON MEATS.

Store	No. of samples	Bacteria per gram of product					
		15°C. incubation			37°C. incubation		
		Minimum	Maximum	Mean	Minimum	Maximum	Mean
I	76	310	53,300,000	2,066,000	310	31,000,000	679,000
II	21	590	29,000,000	1,450,000	280	29,000,000	1,400,000
III	74	330	312,000,000	12,555,000	280	28,700,000	3,800,000
IV	30	285	60,700	11,300	285	55,300	9,500
V	48	1,700	40,000,000	5,710,000	280	85,000,000	4,265,000
VI	36	1,640	150,000,000	9,000,000	1,500	150,000,000	5,440,000
VII	46	300	267,000,000	11,000,000	315	36,000,000	2,300,000
All stores (I - VII)	331	285	312,000,000	6,742,000	280	150,000,000	2,625,000

packages vs. vacuum-packaged samples. While the number of samples packaged by the two methods was roughly equal in only one case, the results of this comparison (Table 5) indicate that within the wide ranges of bacteria counts encountered there was no obvious consistent relationship between the packaging method and the quantitative microflora.

4. *Store:* Throughout the survey, it was noted that the lowest bacterial populations were found in those samples which came from stores in which the personnel in charge of meat merchandising were highly conscientious about maintaining good sanitary precautions in the packaging process, adequate refrigeration of the product in the retail case, and proper control of the length of time packages were displayed in the case. The small, neighborhood-type meat outlet (Stores I and IV, Table 6) was outstanding for the consistently low bacteria counts found in its products, reflecting the care that had been taken in packaging and retailing them. Similarly, the larger, super-market meat departments (Stores III, V, and VI), in which the manager was either too busy or uninformed to take extra precautions in handling these products, were regular sources of high-count samples. The need for education of meat handlers in the necessity for sanitary packaging conditions and good refrigeration was obvious.

Qualitative Microflora

A large variety of microbial types were isolated from the samples analyzed. By far the most common genus encountered was *Micrococcus*, which represented nearly two-thirds of the cultures identified (Table 7).

TABLE 7 -- SUMMARY OF GENERA OF MICROORGANISMS FOUND IN SEVEN PREPACKAGED TABLE-READY MEATS

Genus	Frequency of isolation *			Rank		
	37°C.	15°C.	Total	37°C.	15°C.	Total
<i>Micrococcus</i>	213	127	340	I	I	I
<i>Neisseria</i>	10	37	47	IV	II	II
<i>Bacterium</i>	8	29	37	VI	III	III
<i>Bacillus</i>	19	0	19	II	----	IV
<i>Leuconostoc</i>	0	19	19	---	IV	IV
<i>Achromobacter</i>	7	11	18	VII	VI	VI
Yeasts (all)	0	18	18	---	V	VI
<i>Sarcina</i>	11	2	13	III	XI	VIII
<i>Flavobacterium</i>	10	0	10	IV	----	IX
<i>Listeria</i>	7	3	10	VII	X	IX
<i>Alcaligenes</i>	0	8	8	---	VII	XI
<i>Lactobacillus</i>	0	6	6	---	VIII	XII
<i>Corynebacterium</i>	3	2	5	IX	XI	XIII
<i>Microbacterium</i>	0	4	4	---	IX	XIV
<i>Acetobacter</i>	0	2	2	---	XI	XV
<i>Nocardia</i>	1	0	1	X	----	XVI

*Number of samples from which the genus was isolated, from plates incubated at the temperatures noted.

Other genera were found less frequently, some so seldom that they were not considered as common inhabitants of this group of products, but rather as chance, single-batch contaminants. The 557 cultures isolated and identified were distributed among 30 species of psychrophilic types and 28 species of mesophiles (Table 8). Eight species were isolated from plates incubated at both temperatures, indicating an ability on the part of these species to grow over a rather wide temperature range.

TABLE 8 -- DISTRIBUTION OF BACTERIAL SPECIES IN SEVEN LUNCHEON MEAT ITEMS

Species	Frequency of isolation from product:							Total
	A*	L	C	D	G	M	B	
<u>15°C. cultures</u>								
<i>Acetobacter oxydans</i>				2				2
<i>Achromobacter eurydice</i>	14							14
<i>liquefaciens</i>		2						2
<i>Alcaligenes metalcaligenes</i>					8			8
<i>Bacterium ammoniagenes</i>				5				5
<i>insectiphilium</i>					2			2
<i>minutaferula</i>	2	5						7
<i>tegumenticola</i>			2	8	2	2		14
<i>Corynebacterium pseudo-</i> <i>diphtheriticum</i>	1			1				2
<i>Lactobacillus brevis</i>	5							5
<i>Leuconostoc citrovorum</i>						1		1
<i>dextranicum</i>				5		13		18
<i>Listeria</i> sp.							3	3
<i>Microbacterium thermosphactum</i>			4					4
<i>Micrococcus candidus</i>							2	2
<i>citreus</i>	1							1
<i>conglomeratus</i>		10	1		1			12
<i>epidermidis</i>	3						2	5
<i>flavus</i>	1				4			5
<i>luteus</i>	15	12	3		6		6	42
<i>pyogenes</i> var. <i>albus</i>			5		8	10		23
<i>pyogenes</i> var. <i>aureus</i>				4				4
sp.						6		6
<i>ureae</i>	1		2		2	1		6
<i>varians</i>	1	6	1	9	2			19
<i>Neisseria catarrhalis</i>		2	6	2	3			13
<i>flava</i>			2		1			3
<i>perflava</i>		2					1	3
<i>sicca</i>	19	3				1		23
<i>Sarcina lutea</i>	1		1					2
Yeasts (all types)		7	7	1	3			18
Unidentifiable (1 type)							2	2
<u>37°C. cultures</u>								
<i>Achromobacter cycloclastes</i>					3			3
<i>delicatulum</i>		3					1	4
<i>Bacillus alvei</i>	3							3
<i>brevis</i>	4							4
<i>lentus</i>							1	1
<i>sphaericus</i>	2							2
<i>subtilis</i>	1	3	4				1	9

TABLE 8 -- CONTINUED

Species	Frequency of isolation from product:							
	A*	L	C	D	G	M	B	Total
<i>Bacterium castigatum</i>							5	5
<i>minutaferula</i>		2			2			4
<i>Corynebacterium helvolum</i>					3			3
<i>Flavobacterium breve</i>						1	1	2
<i>harrisonii</i>							5	5
<i>lactis</i>					1			1
<i>rhenanus</i>		1						1
<i>Listeria</i> sp.					8		2	10
<i>Micrococcus candidus</i>				2			7	9
<i>conglomeratus</i>			15	27		18		60
<i>flavus</i>	2		2	3	3			10
<i>freudenreichii</i>	10	3						13
sp. I	26		3				4	33
sp. II					7		2	9
<i>ureae</i>	5	6	17	7	5	1	15	56
<i>varians</i>	7	13	2			6		28
<i>Neisseria catarrhalis</i>			1		1			2
<i>flavescens</i>	2						1	3
<i>sicca</i>					5			5
<i>Nocardia polychromogenes</i>	1							1
<i>Sarcina flava</i>			3	1	2		5	11

*Product A - Bologna
 L - Pickle-pimiento loaf
 C - Spiced luncheon meat
 D - Chopped ham loaf
 G - Salami
 M - New England brand sausage
 B - Souse

Twelve of the 28 species of mesophilic organisms were limited in their occurrence to one product only, and the majority of these were in replicate samples of the product; they were considered to be chance airborne contaminants. Similarly, 15 of the 30 psychrophilic species were single-product isolates. Five of these were either single-sample or replicate-sample isolations, and the product-specificity of the remainder may have been due to pre-fabrication contamination of special ingredients peculiar to the product. An analysis of the bacterial flora of the various ingredients of the products should be helpful in clearing up this question.

The most ubiquitous species isolated was *Micrococcus ureae*, which was found on 37°C. plates from all products and on 15°C. plates from four of the seven types of luncheon meats (Table 8). This species, which is typically found in stale urine and in soil containing urine (2), appears to be more widely distributed in nature than is generally thought, and is by no means limited to its "natural" habitat and source.

Micrococcus varians was the second most common species, being isolated at one or both incubation temperatures from all products except souse.

Micrococcus conglomeratus was isolated from a varying number of samples of all products except bologna and souse, and actually was found in a larger number of samples than any other single species. Of the five products in which this species was found, two of them (pickle-pimiento loaf and salami) yielded the organism as a psychrophilic type. It was isolated from two others (chopped ham loaf and New England brand sausage) as a mesophile and as both a psychrophile and mesophile from one product (spiced luncheon meat). In the latter, however, 15 of the total 16 isolations were from 37°C. plates. The single isolation from salami, as a psychrophile, is not considered significant, but rather a chance contamination.

Several species typically associated with insects (2) were also found in the samples (Table 8), viz.: *Achromobacter eurydice*, *Bacillus alvei*, *Bacterium insectiphilium*, *Bact. minutaferula*, and *Bact. tegumenticola*. The first three species listed were found in each case in only one product and appeared at only one incubation temperature. *Bact. tegumenticola* was isolated from several products, but only at 15°C. incubation, and from relatively few samples of those products in which it occurred. *Bact. minutaferula*, on the other hand, was isolated from only a few samples of three products, but appeared as both a psychrophilic and mesophilic isolate (Table 8). These species did not constitute the most common flora of any product, although in two instances (*A. eurydice* in bologna, *Bact. tegumenticola* in chopped ham loaf) they were isolated an appreciable number of times in comparison to other species.

The significance of the presence of these insect-borne species is obscure. While no direct information is at hand to support this conclusion, the most logical interpretation of their presence in meat products would seem to be that they entered through spices or other ingredients which had been contaminated by insects prior to fabrication of the meat products. An alternate source might be from insect contamination of the product before or during packaging.

The most common mesophilic type found in the bologna samples (and also isolated from a few samples each of spiced luncheon meat and souse) was a white, Gram-variable micrococcus. Its characteristics did not agree sufficiently well with those of any recognized species to merit classification at the species level. It was felt best to designate it simply as "*Micrococcus* sp. I", and not to attempt to identify it, even tentatively, as a variant of a recognized species.

The second most common type of mesophilic organism isolated from salami was a white, Gram-negative micrococcus, which was distinctly different from the type described above. This organism was also isolated twice from souse, and was referred to as "*Micrococcus* sp. II".

One psychrophilic micrococcus type, which was found in 6 samples

of New England brand sausage obtained from a variety of store sources, could not be identified as any of the recognized species of *Micrococcus*. It was designated simply as "*Micrococcus* sp." This organism was distinct from both of the unclassifiable mesophilic micrococci described above.

The cultures identified as *Microbacterium thermosphactum* came from two pairs of samples purchased from different stores. They agreed in all essential respects with the description of this species as given by McLean and Sulzbacher (7, 8). Although these workers originally isolated and characterized the species from a fresh pork sausage habitat, finding it in spiced luncheon meat in this study would indicate that it might be encountered in a variety of products containing pork as a major ingredient.

Four cultures of a yellow to orange micrococcus were classified as *Micrococcus pyogenes* var. *aureus*, largely on the basis of hydrogen sulfide production; otherwise, the organism could be placed in either *Microc. conglomeratus* or *Microc. citreus*, neither of which is described as a sulfide producer. This type also failed to agree in a few other respects with the recognized description of the species (2), e.g. growth temperature, action in litmus milk, and sugar fermentations. Its characterization as *Microc. pyogenes* var. *aureus* is thus somewhat tentative.

The 10 mesophilic cultures listed as "*Listeria* sp." were identical, but they did not conform well enough to the accepted description (2) of *Listeria monocytogenes*, the only recognized species in this genus, to merit this species designation. It was therefore thought best to refer to them as an unclassifiable type within the genus.

It is of interest to note that *Alcal. metalcaligenes*, which is believed to have its normal habitat in the intestinal canal (2), had previously been isolated by one of the present authors (5) as the most common bacterial type in frozen pork sausage. In the present study, the only product in which this species was found was salami.

Two psychrophilic cultures, isolated from duplicate samples of souse and identical in all of the morphological and physiological characteristics determined, were impossible to identify, even at the genus level. The cells were rod-shaped, averaging about $1.0 \times 8 - 10$ microns in size, motile, non-sporulating, and occurred singly or in irregular artificial clumps. Reaction to the Gram stain varied in the same smear from negative through weak positive to strong positive, with a slight tendency for isolated cells to be negative. Nitrates were reduced to nitrites, gelatin was liquefied, and litmus milk was peptonized but otherwise unchanged. None of the carbohydrates tested was fermented, nor was starch hydrolyzed. On the basis of the characters studied, it was not felt that this organism could be reliably placed in *Achromobacter*, *Alcaligenes*, *Bacterium*, *Brucella*, *Flavobacterium*, *Proteus*, *Pseudomonas*, *Salmonella*, *Shigella*, *Spirillum*, *Vibrio*, or in

any other recognized genus (2).

Perhaps the most unexpected result of the identification of the cultures was the complete absence of members of the Enterobacteriaceae and the virtual absence of Pseudomonadaceae. Only one species of the latter family (*Acetobacter oxydans*) was identified. Both of these groups have been reported by other workers (1, 6) as major components of the microbiological flora of a variety of meats and meat products. No explanation for their absence from the luncheon-meat samples can be given.

Factors Influencing the Qualitative Microflora

In contrast to the situation with respect to the quantitative microflora of the samples, no apparent relationship could be found between the qualitative flora and the age of sample, sampling time, wrapping material used, or store source, based upon the data obtained from this study.

SUMMARY AND CONCLUSIONS

1. The microbial populations of 50 samples each of bologna, pickle-pimiento loaf, spiced luncheon meat, chopped ham loaf, salami, New England brand sausage, and souse varied from a few hundred to several hundred million organisms per gram of product. The samples analyzed were purchased at random from a representative group of stores which re-tailed some or all meat products in the prepackaged form.

2. Among the 350 samples analyzed, there were seven samples (2%) which were not acceptable from a consumer standpoint, because of a noticeable "spoiled" odor. The bacteria counts for these samples, while not consistently the highest found among the samples of the products concerned, were excessively large, usually being well above the product mean. In one case, there was visible bacterial growth or "slime" on the product (chopped ham loaf).

3. Prepackaged, table-ready meat products purchased on the open market may be expected to show a considerable variation in quantitative microbiological flora. Unless the packages are handled in such a way as to be highly contaminated in packaging, or are either so poorly refrigerated or held so long before sale that appreciable growth of microorganisms takes place after packaging, the level of the microbial population does not appear to have a noticeable effect on the consumer acceptability of the product. Only in those cases where the bacteria counts were extremely high, and not always under these conditions, was the product found to be unpalatable. The borderline between total bacteria counts in acceptable and unacceptable samples does not seem to be clear-cut.

4. A similar wide variation exists in the qualitative microflora of these products. Of the many bacterial types found, nearly all were common aerial or insect-borne species, or normal inhabitants of the human upper respiratory tract or skin. No recognized pathogenic species were identified.

5. The responsibility of the meat department manager for the sanitary handling and adequate refrigeration of meat products of this general group is emphasized. Proper attention to these details will result in lower bacterial populations in the products, with an accompanying decrease in likelihood of spoilage or loss of consumer acceptability during the time between packaging and sale of the product.

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