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STUDIES WITH A BACTERIOPHAGE INHIBITORY MEDIUM

I. INHIBITION OF PHAGE AND GROWTH OF SINGLE STRAIN LACTIC STREPTOCOCCI AND LEUCONOSTOC

D. R. HENNING, W. E. SANDINE, P. R. ELLIKER AND HELEN A. HAYS

Department of Microbiology, Oregon State University, Corvallis

(Received for publication March 29, 1965)

SUMMARY

A phage resistant starter culture medium was examined for its ability to inhibit bacteriophages under several conditions. The medium also was tested for its ability to support growth of single strain starter culture organisms and for its effect on viability of starter bacteria during storage at 2 and -20 C. When prepared according to manufacturer's directions, the medium prevented phage proliferation and eliminated phages from infected cultures within three transfers. Preparation of the medium with water of 500 ppm hardness did not affect its phage-inhibiting properties. Single strains of Streptococcus lactis, Streptococcus cremoris and Streptococcus diacetilactis grew well in the medium though there was variation between strains; Leuconostoc species were not able to grow well. Storage survival of starter organisms in the phage resistant medium also revealed considerable variation between strains; Leuconostoc sp. and S. diacetilactis survived poorly when stored in the medium, especially at 2 C, while S. lactis and S. cremoris survived well when held at -20 C.

Bacteriophage infection of lactic streptococcus starter cultures is one of the most important causes of insufficient acid production during controlled dairy fermentations. The economic and public health significance of this is well known and has been discussed by others (2, 4).

1

Several approaches toward preventing the consequences of bacteriophage infection during dairy fermentations have been studied. Exclusion of bacteriophage from starter propagation rooms and aseptic handling of cultures have led to the development of special culture-handling equipment (9, 19). Removal of the calcium ion, which is essential for phage development (14), either by ion exchange or chelation has been attempted (10, 11, 15). A novel approach that employs specific antiphage antibodies for neutralization of phages present in culture media has been used successfully by Erskine (7). Other methods in commercial use to minimize phage infection are the rotation of cultures and use of the direct seed method of inoculation (17). Any of the above approaches can be used singly or in combination with other methods.

Recently, a new phage inhibitory medium (PIM) has been made available to the dairy industry. This medium, called "Marstar", has been reported to inhibit development of lactic phages through the calcium-sequestering action of phosphate salts. However, no evaluation of this medium has been published and the present work was undertaken with this in mind. Studies were made on the growth of single strain lactic streptococci in the PIM. The ability of the medium to inhibit development of a variety of phages under different conditions also was tested.

EXPERIMENTAL PROCEDURES

Single strain cultures used were taken from the collection maintained in the Department of Microbiology at Oregon State University or isolated from commercial mixed strain lactic starter cultures by serial dilution plating with lactic agar (5). Phages employed were taken from the collection at Oregon State or isolated from cheese whey collected from commercial plants.

The PIM was prepared according to the manufacturer's directions except where otherwise stated; 100 ml were dispensed aseptically into 250-milliliter flasks before inoculation.

Single strain growth.

Single strains of *Leuconostoc* and lactic streptococci were cultured for eleven daily transfers in the PIM. Plate counts were made on lactic agar (5) after each incubation period of 16 to 18 hr at 21 C. Plates containing *Leuconostoc* were incubated at 25 C for 48 hr before counting. Ten per cent nonfat milk in 100-milliliter amounts sterilized at 121 C for 15 min was used as a medium for comparison.

Fhage inhibition.

Milk cultures were incubated for 18 hr at 21 C and then inocuated at the 1% rate into PIM and sterile 10% nonfat milk. The cultures were then infected with known amounts of homologous phage. Cultures were incubated daily at 21 C for 18 hr and 1-milliliter aliquots were then titrated for plaque-forming units (pfu) per milliter by the overlay method (1). Microscopic examinations of PIM cultures were made after the last transfer to establish the presence or absence of the starter organism.

Culture storage.

The PIM and 10% nonfat milk cultures of two *Leuconostoc* strains and three lactic streptococci were frozen or refrigerated after incubation at 21 C for 20 hr. Samples were plated periodically to determine the effect that the PIM had upon survival of the starter organisms. The frozen cultures were stored at -20 C and the refrigerated cultures were stored at 2 C.

¹Supported by a grant-in-aid from Marschall Dairy Laboratory, Inc., Madison, Wisconsin, Technical Paper No. 1974, Oregon Agricultural Experiment Station, Corvallis.

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TABLE 1. CROSS INFECTION PATTERNS BY BACTERIOPHAGES FOR STRAINS OF LACTIC STREPTOCOCCI ISOLATED FROM DIFFERENT BRANDS OF COMMERCIAL MIXED STRAIN STARTER CULTURES

8 T.		Single strains ^a									
Phage	A1 ^b	A2	В1	B2	C1	C2	D1	D2	E1		
al°	$+^{d}$	+	+	_		_		-	+		
a2	+	+	+	-	-	-	-	+	+		
b1	+	+	÷				-		+		
b2		—	-	+	-		-	-			
cl		4-	+		+	-		+	+		
c2	+	-	+	-	-	+	_	-	+		
d1	-	+	_	_	-		- -	+	_		
d2	+	+	+	-	-	-	-+-	+	+		
el	+	+	+	1	+		λ <u></u>	+	,+		

^aAll strains tested were isolated from different commercial mixed strain cultures. For example, strains A1 and A2 are from different mixed strain cultures supplied by supplier A.

^bSpecies identity of cultures is: A1, A2, C1 and C2 are S. *diacetilactis*; B1, B2, D1, D2, and E1 are S. *cremoris*.

[°]Phages are identified by giving them the same numbers as the homologous host used during their primary isolation.

 a_+ means inhibition on lawn of the single strain by 1 drop of sterile phage lysate.

PIM preparation with hard water.

The PIM was prepared with hard water (8) at a level of 500 ppm (29.25 gr per gallon). The control media were 10% nonfat milk and PIM prepared with distilled water. Three lactic streptococci were grown in the above media for one transfer and then transferred at the 1% level into fresh media of the same composition. The cultures were infected with homologous phage and incubated at 21 C for 18 hr and then the phage titer determined. The following day a second transfer was made of all cultures and the plate count and phage titer determined after incubation as before.

RESULTS

Table 1 shows cross reaction patterns of phages isolated using a particular host for strains of lactic streptococci isolated from different commercial brands of mixed strain starter cultures. The table emphasizes one of the reasons why so much repeated difficulty with bacteriophage inhibition of starter cultures occurs in the dairy industry. For example, phage d2 isolated on host D2, obtained by plating a mixed strain culture supplied by company D, lysed strains isolated from another mixed strain culture (D1) from the same company as well as strains isolated from mixed strain cultures from suppliers A, B and E. Similar cross reaction patterns may be seen from the rest of the table, and it is evident that many different brands of starter cultures contain the same

strains where bacteriophage sensitivity is concerned. Thus, the need for a simple, effective phage control medium is emphasized.

Table 2 presents data on growth of single strains of lactic streptococci in PIM and nonfat milk. Inspection of the data indicates that strains may vary in their growth response in the two media, some doing somewhat better in PIM than nonfat milk and others

TA	BLE	2.	POPULATION	Ac	CHIEVED	IN	PIN	M A	ND	No	NF	AT Ì	MILK
BY	LAC	CTIC	STREPTOCO	CCI	INCUBA	TED	18	HR	AT	21	С	FOR	THE
			NUMBER	OF	TRANS	FER	s I	NDIC	CATI	ED			

			Averag	çe plate	count/ml	x 107	2	a	
Org	anism	1st trans NFM	1st transfer NFM PIM		ıd sfer PIM	3ro trans NFM	d sfer PIM	Preferen	
S	lactis			į.	. I				
5.	27	111ь	60	112	44	103	51	-	
	C2	122	168	56	201	17	158	+	
	C10	179	325	223	346	251	450	0	
	7962	57	126	133	123	101	147	0	
	7963	117	32	295	470	226	460	+	
	11454	280	- 240	249	381	205	410	· +	
	11955a	117	63	97	29	72	28		
	a	61	43	90	47	70	45	0	
	b	74	67	95	36	97	45	. —	
S.	cremoris						a		
	144F	75	148	172	65	76	18		
	W	67	10	82	22	54	28	0	
	C13	156	146	128	171	161	434	0	
	9625	94	59	108	33	69	52	0	
	11602a	30	26		50	69	64	- 0	
	18-1	147	12	156	164	51	179	+	
	27-1	93	38	109	48	84	56	. 0	
	Da-1	38	28	52	19	103	17		
	Da-5	131	114	108	78	128	92	0	
	31-9	44	19	29	11.5	16	11	0	
	CC-2	88	.24	84	45	75	35		
	11E	116	99	131	76	119	165	0	
S.	diacetilacti	is		20- 2 5 760					
	RM1	442	463	328	510	360	390	0	
	6B-1	158	67	113	98	121	50		
	63-3	171	66	98	62	119	49		
	Da-20	76	74	84	58	89	57	0	
	23-2	120	45	110	49	103	61	0	
	4R-1	97	87	79	64	94		0°	
	4R-5	135	132	90	130	93	24		
	3D-1	110	94	91	45	70	48	0	
	110-3	143	93	84	66	119	76	0	
	31-2	107	66	161	61	171	57	-	
	31-8	133	64	172	42	175	59		
	CC-1	145	54	88	50	103	58	C	

 $^{*}+$ indicates greater than twice as many cells developing in PIM as compared to NFM after 3 transfers; – indicates less than half as many, and 0 indicates the cell counts were within these limits.

^bPlate count (average of 2 plates) was 111 x 10⁷ per ml.

'Second transfer data.

doing the reverse. For example, if we arbitrarily call the development of twice as many cells in PIM an indication of growth preference by a strain for this medium, comparisons between species and strains can be made. Doing this we find three out of nine *S. lactis*, two out of 12 *S. cremoris* and none of 12 *S. diacetilactis* strains reveal preferences for nonfat milk. Finally, three out of nine *S. cremoris*, seven out of 12 *S. lactis* and seven out of 12 *S. diacetilactis* strains are within the arbitrary limits indicating no preference for either medium. No lactic streptococci were found, however, which did not grow reasonably well in the phage resistant medium, though there was apparent slight reduction of growth of *S. diacetilactis* under these conditions.

Growth of *Leuconostoc* organisms in PIM and nonfat milk after one and 11 transfers may be seen in Table 3. One strain of each of the three species of lactic streptococci was included for control purposes. It may be seen that in all cases the *Leuconostoc* species grew poorly in PIM relative to the milk. *Leuconostoc citrovorum* 91404 was not detectable in the PIM by plating after the eleventh daily culturing.

Table 4 presents data comparing the ability of homologous bacteriophages for *S. cremoris* to replicate in PIM and nonfat milk. The titers of each phage were markedly reduced after only one transfer in the PIM. Furthermore, after three transfers in PIM, all evidence of phage as revealed by the plaquing technique was gone. These data are typical of three-transfer values obtained on over 20 different lactic phages, including types for all three species of lactic streptococci. In all cases, microscopic examination revealed that the single strains were still present in the infected PIM cultures after three

TABLE 3. GROWTH OF SINGLE STRAINS OF LACTIC STREPTOCOCCI AND Leuconostoc in PIM and Nonfat Milk

	Averag	e Plate (Count/mlª x 10 ⁵	after	
	First tr	ansfer	Eleventh transfer		
* es \$	PIM	NFM	PIM	NFM	
LeF_8	170	480	0.42	370	
Da-3	110	550	6.6	370	
CAF_7	22.6	340	0.00015	350	
CAF_{19}	0.6	130	0.0027	122	
91404	24.3	300	0.00000	310	
	30,400	29,100	36,500	26,700	
	10,500	13,700	12,000	13,000	
s 18-16	6,100	14,600	9,600	10,800	
	LcF ₈ Da-3 CAF ₇ CAF ₁₉ 91404 \$ 18-16	$\begin{tabular}{ c c c c c } \hline Averag \\ \hline First tr \\ \hline PIM \\ \hline \\ LcF_8 & 170 \\ Da-3 & 110 \\ CAF_7 & 22.6 \\ CAF_{19} & 0.6 \\ 91404 & 24.3 \\ & 30,400 \\ 10,500 \\ \hline \\ s & 18-16 & 6,100 \\ \hline \end{tabular}$	$\begin{tabular}{ c c c c c } \hline Average Plate 0 \\ \hline \hline First transfer \\ \hline PIM & NFM \\ \hline \\ LcF_8 & 170 & 480 \\ Da-3 & 110 & 550 \\ CAF_7 & 22.6 & 340 \\ CAF_{19} & 0.6 & 130 \\ 91404 & 24.3 & 300 \\ 30,400 & 29,100 \\ 10,500 & 13,700 \\ s 18-16 & 6,100 & 14,600 \\ \hline \end{tabular}$	$\begin{tabular}{ c c c c c c c c c c c c c c c c c c c$	

"Average from duplicate plates; for convenience, values are expressed in terms of the same power of ten (10^5) .

TABLE 4. GROWTH (PFU/ML) OF HOMOLOGOUS S. cremoris Bacteriophages in Phage Resistant Medium (PIM) and Nonfat Milk (NFM) When Tested After the Transfers Indicated

		PFU/ml after 1	transfer
Phage	MoI ^a	NFM	PIM
40	1.5 x 10 ⁻²	40 x 10 ⁷	26 x 10 ³
33	3.5 x 10 ⁻²	60×10^{7}	50 x 10 ¹
6	33.0 x 10 ⁻¹	40×10^5	10 x 10 ³
10	5.0 x 10 ⁻³	10 x 10 ⁷	40 x 10 ³
27	9.6 x 10 ⁻¹	12×10^{8}	62×10^{4}
		PFU/ml after 3	transfers
5	8.0 x 10 ⁻⁶	40 x 10 ⁵	$< 10^{\circ}$
12	3.8 x 10-1	10×10^4	$< 10^{\circ}$
4	4.3 x 10 ⁻¹	25×10^5	$< 10^{b}$
15	2.6 x 10-1	50×10^4	$< 10^{\text{b}}$

^aMoI–Multiplicity of infection, the ratio of number of phage particles to the number of bacteria.

^bNo plaques on plate receiving 1 ml of infected culture diluted 1:10.

transfers. However, this was not the case with all infected milk cultures; some revealed no cells surviving while others showed the presence of phage resistant mutants. These mutants never appeared to be as active as the parent strains.

Results in Table 5 suggest that *Leuconostoc* survive better in nonfat milk than in the phage resistant medium at 2 and -20 C. The same is true for *S. diacetilactis*, especially at 2 C. There appeared to be less difference between the *S. lactis* and *S. cremoris* strains tested under these conditions; however, storage of these organisms in phage resistant medium at -20 C appeared less detrimental than storage at 2 C. In addition, *S. lactis* survived somewhat better than *S. cremoris* at 2 C in both media.

Table 6 gives data showing that when water containing 500 ppm of hardness is used, the phage inhibiting properties of the PIM are not impaired. This table also shows that phage particles can be eliminated from cultures by transferring host cells in PIM for several transfers. Also it can be seen from the plate count data that two of the single strains tested were reduced to extremely low populations in nonfat milk as a result of phage activity.

DISCUSSION

The ability of PIM to minimize phage development in mixed strain lactic starter cultures and to remove phage particles from infected cultures has been confirmed by these studies. Preparation and

Creation and Creat			Average plate count per mla x 107					
			Refrigerated (2 C)			Frozen (20 C)		
Organism	Medium	1 day	7 days	21 days	1 day	7 days	21 days	
L. citrovorum LcF _o	PIM	0.68	0.27	0.65	0.017	3.00	0.10	
8	NFM	3.30	3.10	3.50	2.40	, 2.40	3.30	
L. citrovorum Da—3	PIM	0.63	1.10	1.21	0.042	0.34	0.21	
Da-5	NFM	3.90	5.10	3.90	4.60	3.00	3.02	
S. lactis E	PIM	330.00	450.00	58.00	340.00	100.00	78.00	
	NFM	280.00	250.00	14.00	170.00	140.00	78.00	
S. cremoris 1	PIM	120.00	95.00	0.20	63.00	91.00	71.00	
	NFM	150.00	120.00	0.00189	130.00	55.00	63.00	
S. diacetilactis 18-16	PIM	57.00	0.014	0.00159	56.00	46.00	38.00	
	NFM	120,00	59.00	0.0107	100.00	140.00	51.90	

TABLE 5. STORAGE SURVIVAL OF SINGLE STRAIN LACTIC STREPTOCOCCI IN NONFAT MILK AND PIM

"Average from duplicate plates; for convenience, values are expressed in terms of the same power of ten (10^7) .

TABLE 6. NUMBERS OF PHAGES AND HOST LACTIC STREPTOCOCCI IN NONFAT MILK (NFM), PHAGE RESISTANT MEDIUM (PIM) AND PIM PREPARED WITH HARD WATER (HPIM) AFTER THE TRANSFERS INDICATED

				$pfu/ml x 10^4$		
· ,	Af	ter first transfer		1	After second transfer	
Phage	NFM	PIM	HPIM ^a	NFM	PIM	HPIM
S. cremoris 5	60,000	<1.000 ^b	<1.000 ^b	4,000	<0.010°	<1.000 ^b
S. cremoris 15	75,000	$< 1.000^{b}$	<1.000 ^ь	1,400	<0.010°	$< 1.000^{b}$
S. lactis 24	200,000	$< 1.000^{\text{b}}$	<1.000 ^b	6,000	0.020	$< 1.000^{b}$
			Average	plate count/ml x 10	5	
	-		Aft	er second transfer		
Host	NFM		PIM		HPIM	
S. cremoris 5	430		7,000	-	6,600	
S. cremoris 15	0.001		67,000)	44,000	
S. lactis 24	0.004		5,200		4,700	

^aPIM prepared with water at 500 ppm hardness.

"No plaques on plate receiving culture diluted 10-4

"No plaques on plate receiving culture diluted 10-2.

use of PIM in accordance with the manufacturer's directions would therefore be expected to provide added protection for the dairy industry against bacteriophage problems.

Results also indicate that growth of single strains of lactic streptococci in PIM varies considerably between strains, emphasizing the well-known nutritional complexity of these bacteria. These differences may be minimized in mixed strain starter cultures and studies on this are now in progress and will be reported in the second paper in this series. The somewhat reduced growth of certain strains of S. *diaceti*- lactis in PIM coupled with the inability of any of the strains of this species tested to grow better in PIM indicates the medium may cause reduced citrate fermentation. Further evidence for this comes from the inability of most strains of the *Leuconostoc* to grow well in the PIM. These findings might suggest that the PIM be limited to use for Cheddar cheese cultures where development of *Leuconostoc* sp. is not essential and growth of *S. diacetilactis* undersirable (16, 18). The medium would be useful for Cottage cheese cultures also, especially if the cheese were creamed with dressing to enhance the flavor (6, 12, 13). Use of the PIM for buttermilk cultures would be questionable, and could lead to insufficient or "green" flavor in the final product.

Since the growth of all single strain cultures in PIM does not proceed to the same population level as in nonfat milk, an examination of PIM growth responses of component strains in commercial mixed strain starter cultures is needed if these cultures are to be propagated in PIM. It is reasonable to expect that mixed strain cultures would not maintain the same strain balance in PIM as in nonfat milk. In view of this, it may be worthwhile to prepare new mixtures of strains that have the same relative growth abilities in PIM.

From the storage studies, it would appear that refrigerated or frozen storage of lactic streptococci in either PIM or nonfat milk may result in significant loss of viable cells within 3 weeks. The survival ability in PIM as compared to milk appeared to depend upon the single strain under consideration.

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THE USE OF ANTIMICROBIAL SOAPS AND DETERGENTS FOR HAND WASHING IN FOOD SERVICE ESTABLISHMENTS

FRANCIS D. CRISLEY AND MILTON J. FOTER²

Robert A. Taft Sanitary Engineering Center U. S. Department of Health, Education, and Welfare, Cincinnati, Ohio

Considerable interest has been generated in the use of the newer formulations that contain skin sanitizing agents for hand washing in food processing and service establishments. In some areas of the country, the degerming agents have been suggested so often by public health officials that the use of antimicrobial soaps has approached the status of a recommendation. It is the purpose of this paper to briefly review this situation and some of the problems involved in choosing between some of the most common chemical agents that have been suggested for this purpose.

Sanitization of the skin involves a limiting surface that Lane and Blank (22) have described generally as a continuous, relatively smooth layer of dead, flattened, keratinized cells made somewhat irregular by various ridges and furrows, by orifices of sweat glands, and by hair follicles and outgrowths of hair. The cutaneous glands secrete a film over these cells, and this film constitutes the absolute limiting boundary between man and his environment. In this film are included salt, urea, and other substances left behind by evaporation of sweat; sebum, which covers all areas except the palms and soles; and a uniform layer of fat. The cells contain proteins, lipids, and water. The protein is largely keratin, which is insoluble in weak acids, weak alkalies, and salt solutions. If the outer layers of the cells of the epidermis are brought into equilibrium with a solution that has a pH to either side of pH 3.70, the isoelectric point of keratin, swelling of the cells will occur. The skin reacts more strongly to alkalies than to acids. Numerous measurements have shown the pH of the skin to range from 3.5 to 7.0.

Although the relative amounts and kinds of the substances of the skin may vary among individuals and from time to time in each individual (as a result of changes in physiological conditions), the structure and composition of the skin generally provide a good environment for bacterial growth. Foci for the establishment of bacterial flora exist in hair follicles, the sweat and subaceous glands, and the numerous ridges and furrows. Price (29) has classified the bacteria found on the human skin into two groups, the transient and the resident. The transient types are acquired by contact with other persons or objects in the environment. The resident flora comprise organisms that have established themselves and live in dynamic balance as parasites or saprophytes in the skin.

From the standpoint of sanitation in the food establishment, the ideal situation would be sterilization of the skin on the hands of food handlers; however, it is generally agreed that it is impossible to render skin sterile without destroying it. Transient bacteria are readily removed with ordinary soap and water. The resident organisms, however, are more difficult to remove; and scrubbing in hand-washing procedures is a recognition of this fact. In many persons, staphylococci make up a significant part of the resident flora. Because of the pathogenicity of some staphylococci and their ability to produce enterotoxin, major stress has been placed on the destruction, removal, and control of these organisms by hand-washing procedures. Although there is a paucity of reports on the efficacy of germicidal agents for hand washing in food service establishments, much valuable information can be gleaned from the voluminous literature on preoperative or surgical procedures and, to a lesser degree, from studies of the control of bacteria that produce body odor.

Even if surface bacteria are removed, the bacterial population is easily re-established by the emergence of resident organisms from the deeper structures and the addition of transient types acquired by continual contact with objects in the external environment. Price (29) has shown that under normal conditions the skin flora is fully re-established within a week after degerming of the skin. For this reason, the advantages of the deposition of a germicidal residue on the skin after hand washing to exert a continuous antibacterial action on the emerging organisms has been studied. The reliability of data on the efficacy of chemical agents known to be strongly retained on the skin surface is questionable when one moves from the study of operating-room hand-washing procedures (in which most of the rigorous testing has been done), through the simple

¹The contents of this paper are derived from the available literature and the opinions of the authors and should not be construed to represent the views of the Public Health Service. ²Present address: Division of Environmental Engineering and Food Protection, Public Health Service, U. S. Department of Health, Education, and Welfare, Washington, D. C.

control of body odor, to the hands of the food handler. Optimum control of skin bacteria by antibacterial agents in soaps and hand-washing detergents may depend on continuous use of the antibacterial preparation at work and at home, since washing with ordinary soap might tend to remove residual antibacterial agents quickly. Also, the food handler must usually wash utensils in strong cleansing detergents at least intermittently during the day, which causes swelling of the epidermal layer and disruption of the film of active agent deposited by germicidal soaps. To add to these factors, many of the germicidal agents presently in use are soluble in soap fats and fat solvents, and much of the germicide could be lost from the hands by contact with animal and vegetable oils in foods such as salads, meats, etc. Since optimal reduction of skin bacteria by some antibacterial agents has been reported to be based upon continuous use of the agent for as long as 7 days, these limitations must be considered when such agents are proposed for food-handling operations.

Another factor that has not been adequately considered is the ability of certain transient organisms to change their status and become more or less permanent residents. Price (29) has given the term "colonization" to this mechanism. While many transient types tend to disappear spontaneously from the skin, apparently because the conditions are not suited to their survival or colonization, Price (29) has reported that colon bacilli placed on the hands do not so disappear. He found that prolonged presence of unusual contaminants from wounds, such as Staphylococcus aureus, streptococci, Escherichia coli, Bacillus pyocyaneous (Pseudomonas aeruginosa), resulted in their colonization on the hands as part of the resident flora. Price (29) also noted the appearance and persistence for over a year of a nonpathogenic Trichophyton on the skin of his hands and arms, although this organism had not been encountered previously and was not present in the air of the laboratory. To what extent the colonization mechanism may operate in food handlers exposed to salmonellae in poultry, or to bacteria in abscesses or other pockets of infection in meat carcasses, has not been adequately studied, to the author's knowledge; this perhaps should be considered in the choice of antibacterial hand-washing compounds.

The degree to which mechanical cleansing alone is responsible for removal of skin bacteria is important (29) in the consideration of hand-washing procedures and provides a standard by which the degerming efficiency of antibacterial hand-washing agents can be measured. Price (29) concluded that the amount of friction produced at the skin surface by scrubbing appeared to be the most important

variable factor in dislodging the resident flora. More firmly imbedded than the transient bacteria that are only lightly attached to the skin by extraneous grease or oils, the resident organisms cannot be removed by soap and water or simple rinsing without the use of friction. Price (29) showed that rubbing the hands together was more effective than rinsing, but less effective than scrubbing with a soft brush. A soft brush was less efficient than a stiff one. Brushing the skin without soap reduced the resident flora more rapidly than when soap was used, because soap served as a friction-reducing lubricant for brush bristles. Soap, however, increased the efficiency of removal of grease, dirt, and transient bacteria. Price (29) also found brushing with soap in hard water to be more effective than in soft water for removing the basic flora, because hard water precipitated the soap. This finding naturally leads to the possibility that modern detergents, which are not so readily precipitated by hard water, may interfere somewhat with the mechanical cleansing process.

An understanding of these complexities of skin sanitation is needed before an attempt can be made to discuss some of the major types of antibacterial agents that are presently being considered for hand soaps and detergents in food service establishments. Many of the same agents have been proposed for use in lotions or creams. This type of application may be objectionable because of the possibility that the resident bacteria can multiply in the deeper skin layers beneath the preparation (24) and also because of the enhanced possibilities for introduction of the agent into foods.

THE BISPHENOLS

Essentially diphenols, the bisphenols are compounds that contain two hydroxyl (OH) groups, only one of which is neutralized by alkalies in soaps and detergents. The second hydroxyl group on the molecule remains free and is completely active against bacteria. In this characteristic the bisphenols are superior to the older phenolics, in which the single hydroxyl groups are easily inactivated by soaps. Of the many bisphenols that have been synthesized, two are most commonly associated with hand soaps. They are hexachlorophene and bithionol. Both are bacteriostatic agents that act by inhibiting the growth of bacteria rather than by killing the organisms.

Hexachlorophene (also known as G-11, AT-7, Gamophen, Hexosan, Exofine, Phisohex, Surgicen, and Surofene³ is 2, 2'--Dihydroxy-3, 3', 5, 5', 6, 6'-hexachlorodiphenylmethane or bis-(3, 5, 6-tri-chloro-2-hydroxyphenyl) methane (26). It is usually

³Mention of commercial products does not infer endorsement by the Public Health Service.

employed in a concentration of 1 to 3% in liquid or solid soaps, lotions, or emulsions (26). Some skin sensitivity reactions have been known to occur in some individuals (26).

Reduction of skin bacteria has been reported to be considerably greater by hexachlorophene soaps than by ordinary soap (14, 24). No significant reduction occurs immediately after application of the agent (14, 24); and, when used as a single scrub, hexachlorophene soaps are not much more effective than ordinary soaps (5, 19, 31). Routine use for 5 to 10 days, however, has been reported (2, 4) to result in a reduction of bacteria as high as 85 to 95% from the original numbers. Only 4% of the food handlers regularly washing with hexachlorophene formulation for a period of over a year harbored coagulase-positive staphylococci, as opposed to a 16% coagulase-positive rate among workers using hexachlorophene-free soap (7). These findings may indicate that some residual antibacterial effect may be present even on individuals who use regular soap away from work. Shemano and Nickerson (33), using hexachlorophene labelled with C-14, found that some of the agent remained on the skin for a considerable time, although some was lost, especially during the first day, by washing with ordinary soap and water. Whether residual agent that remains adsorbed to the skin is as fully active against bacteria as free hexachlorophene is not clear. Some loss of the agent by washing with ordinary soaps between hexachlorophene ablutions is commonly accepted (16).

Following degerming with hexachlorophene and the attainment of a low bacterial level, the normal count returns about 7 days after the use of the agent is stopped. This delay, however, may not be solely due to a residual adsorbed on the skin, since regeneration times of 1 to 7 days occur after disinfection by other means (29), depending on the thoroughness of treatment.

The activity of hexachlorophene is greatly reduced by organic matter such as body fluids, pus, serum, albumin, milk, etc., and non-ionic detergents and emulsifying agents (31). Although there is no evidence that hexachlorophene-resistant bacteria develop as a result of exposure to the agent (31), Price (28) pointed out that the microbial flora on the hands of different people vary in susceptibility to the agent with some individuals harboring a resistant flora. This finding introduces some uncertainty as to the reliability of hexachlorophene soaps that is probably more serious in the operating room than in food service establishments; nevertheless it must be considered.

In general, the Gram-positive bacteria are most susceptible to hexachlorophene, and Gram-negatives such as *E. coli* and *Salmonella* are not greatly affect-

ed (31). Many Gram-negative types are represented among the transient species encountered in food by the food handler. Post and Balzer (27) have reported that hexachlorophene appeared to have some effect on the transient as well as the resident bacterial flora on the hands of four culinary workers. The effect on the Gram-negative organisms was erratic. The authors admitted that the results were inconclusive and in need of further clarification. Furthermore, the small number of workers studied and the diversity of their culinary duties suggest that additional studies should be carried out on a greater number of subjects and more attention given to the type of culinary operation performed and the extent to which workers' contracts with dishwater and food influence the reduction of bacterial flora by hexachlorophene.

Bithionol (also known as XL-7, Actamer, Lorothiodol, and TBP) is termed 2, 2'-thiobis (4, 6-dichlorophenol) or bis (2-hydroxy-3, 5-dichlorophenyl) (26). It is usually employed in concentrations of 1 to 3% in liquid or solid soap formulations for surgical scrub or other skin disinfection. Lower concentrations may be used, one brand containing only 0.4%.

Soap containing 1 to 2% bithionol is reported to be at least as active as soap with hexachlorophene. It is also more active against the Gram-positive than the Gram-negative bacteria (1) and is reported to be fungistatic (1, 31). After 10 to 12 days of continuous use, the bacterial load on hands is said to be reduced by 89 to 97.4% and levels off with no further reduction (1). It is said to be nonirritating when used in soap.

Bithionol, like hexachlorophene, is claimed to resist removal by soap and water (1). It is said to be strongly absorbed by animal tissues such as skin and hair and works best in the acidic range of pH 5.0 to 6.5 (1). Apparently an active residual is maintained (1) in the presence of alkali in soaps or detergents, but more study is needed on this aspect of both bithionol and hexachlorophene.

THE IODINE COMPOUNDS

Free Iodine

Elemental iodine is one of the most effective antimicrobial agents known (31). It is essentially bactericidal, dilutions possessing bacteriostatic and bactericidal action being practically identical (18). Under a variety of exaggerated test conditions, iodine in the proper concentrations is uniformly active against a broad spectrum of pathogenic organisms, including the tubercle bacillus, pathogenic fungi, viruses, and even bacterial and fungal spores. Although effective for antiseptic washes and for irrigation purposes over a wide pH range (17), the activity of iodine solutions is markedly enhanced under acid conditions.

Reddish (32) points out that the well-recognized efficacy of iodine is partly due to the margin of safety

under which it has been employed. It is used as a skin antiseptic in hospitals in a concentration of 2%, although a level of 0.02% in solution is germicidal within one minute to a variety of pathogenic organisms, including *S. aureus*. Tinctures of iodine have low surface tensions, and the solvent action of the alcohol dissolves skin oils and facilitates penetration into the epidermal layer, thus destroying both the transient and the resident bacterial flora. Aqueous solutions of iodine have also been used successfully and possess certain advantages as preoperative skin disinfectants, and disinfectants for surgical instruments, clinical thermometers, drinking water, and cating and drinking utensils.

The iodophors are chemical complexes of diatomic iodine and solubilizing agents or carriers, usually synthetic nonionic surfactants. A portion of the iodine becomes firmly bound in the complex and is unrecoverable, but the remainder is "available" and is believed to be responsible for the germicidal activity. The iodophors have been reported to be effective sanitizers, good disinfectants *in vitro*, nonallergenic, relatively nontoxic, and noncorrosive.

The activity of the iodophors is directly related to the amount of titratable iodine present in solution. Titratable iodine content is very pertinent in evaluating commercial iodophors (10). Blatt and Maloney (10) compared three commercial iodophors with aqueous or alcoholic solutions of elemental iodine on the basis of equivalent amounts of titratable iodine and found no significant differences in germicidal effectiveness. These results indicate that germicidal activity is contributed solely by titratable iodine, and any enhancement of germicidal activity is at least partially due to the wetting action of the detergent compound. Blatt and Maloney (10) also found that, once all of the titratable iodine was removed from the compound and after the compound had been allowed to stand, no further iodine could be demonstrated by titration. In the analysis of iodine preparations, it is pertinent that the amount of titratable iodine does not always represent the actual amount of active iodine to be expected under actual conditions of use, because the amount dissociating from the complex at any time is dependent upon the dissociation constant, which is influenced by pH and temperature (12). This amount may be only a fraction of the amount recoverable by titration.

Solutions of elemental iodine, phosphoric acid buffered iodine- I_2 and solutions of certain iodophors with a low pH (even after the addition of test bacteria) are believed to be among the best sanitizing agents (31). Many of the marketed liquid iodophors contain phosphoric acid. A cationic iodophor that possesses an alkaline pH and contains 3.2% elemental

iodine has been recommended for disinfection of the skin, for operative procedures, and for disinfection of thermometers and surgical instruments (31). It has been reported (21) to be responsible for a reduction in major postoperative wound infection from 14.8% to 6.8%, although minor infections were not significantly reduced.

Comparisons have been made of the effectiveness of free iodine preparations, iodophors, and hexachlorophene. Goldenberg, et al. (19), by culturing washings of the insides of surgeons gloves, found that iodine-detergent surgical scrub was almost three times as effective after 4 minutes of scrubbing (23.5% positive cultures) as a 3% hexachlorophene detergent was after 10 minutes of washing (66.7% positive culture). Recently, King and Price (20), employing the widely used method of Price (29), compared the degerming activity of solutions of several iodophor formulations with simple alcoholic and aqueous solutions that have approximately the same iodine content, and with a scrub with ordinary face soap. Although the iodophors were less effective in reducing bacteria on the skin than were the iodine solutions, they were more effective than ordinary soap. A 2minute exposure to a tincture of 1% iodine in 70% ethyl alcohol reduced the bacterial flora to less than 20% of the pre-exposure levels (equivalent to 13 minutes of soap and brush scrubbing). The same exposure to the most efficient iodophor reduced the flora to only 54%, equivalent to the efficiency attained in 4 minutes of scrubbing with white soap and brush, or to 35 seconds of exposure to 1% iodine in 70% alcohol. The authors believed that about the only advantage of an iodophor surgical scrub over one incorporating tincture of iodine is that the former is more pleasant and less irritating.

Experimental detergent-iodine cakes containing about 0.7 to 1.0% available iodine have been produced and patented (15), but, to our knowledge, are not yet available commercially (15). The exact details of their composition are not known, and an evaluation should be made when more information is available.

As skin degerming agents for the surgical scrub, the iodophors appear to be generally less efficacious than elemental iodine, but this may be a result of the paucity of information about the amount of titratable and dissociated iodine present in the various compounds under actual conditions of use. The available information does not reveal the presence of true residual activity as occurs with hexachlorophene. The iodophors are attractive to potential users because of the claim that detergency and disinfection can be accomplished simultaneously with the same agent.

THE CHLOROCARBANILIDES

TCC is a 3, 4, 4, trichlorocarbanilide (6). Available information from the manufacturer (6) indicates bacteriostatic activity against staphylococci in dilutions of 1:5 million to 1:10 million, with some fungistatic action against skin fungi. TCC is used in soaps in concentrations ranging from 0.5 to 2.0% and is claimed to be unaffected by either nonionic or anionic detergent. Data for Gram-positive bacteria only are included (6). Handwashing tests with TCC were strictly controlled to eliminate the use of other handwashing agents, so no skin retention data are available.

Bacteriostat CH3479 (Irgasan CF-3) is 3-trifluoromethyl 4, 4' dichlorocarbanilide (3). It is used in concentrations of 1% in deodorant soaps or shampoos and in detergents in 0.2 to 0.4% for residual bacteriostatic effect on cotton fabrics. Although ineffective against Gram-negative bacteria, it is claimed by the manufacturer (3) to be more effective than TCC against Gram-positive bacteria.

Although extensive evaluations of TCC and CH3479 are not yet available, these compounds seem to require their exclusive use by the food handler in order to be fully effective.

THE QUATERNARY AMMONIUM COMPOUNDS

The quaternary ammonium germicides ("quats") are another class of synthetic chemical disinfectants which are synthesized to form amines in which the nitrogen in the molecule has a covalence of 5. With the quats, however, the hydrogen atoms are replaced by one or more alkyl groups (CH₃, C₂, H₅, etc.), or a phenyl radical, and one or more alkyl groups containing C_8 to C_{18} carbon chain lengths (31). The quats are characterized further by their ability to depress greatly the surface tension of water. This property places them in a class of chemicals frequently described as wetting or surface active agents, detergents, or dispersing agents. Surface active agents are grouped further between "anionic" detergent (natural soaps and many synthetic soap substatutes); "non-ionic" detergents (sudsing agents); and the quats which are "cationic" detergents (substituted ammonia compounds). Only the quats or cationics are discussed in this review.

The antimicrobial properties of quats are attributed to their chemical reactivity and the ease with which they are adsorbed. Likewise, these properties account for their occasional failure. The quats are inactivated by soap, hard waters, lecithin, and other phospholipids, and are adsorbed by charcoal, bentonite, and agar. They combine readily with proteins and thus are less efficient in the presence of serum, milk, and other food soils (30). By 1954, the quats had been demonstrated to be incompatible with thirty-six chemical agents, among which were iodine, lanolin, pine oil, silicates, polyphosphates, and anionic detergents (23). The number of chemical agents with which the quaternaries are incompatible is about equal to the number of agents with which they are compatible. Thus it is critical that the ingredients of a formulation be compatible with the quaternary used.

The literature is replete with studies on the antimicrobial properties of quats alone and in preparations formulated for a variety of uses. In vitro they appear to be equally effective against many Grampositive and Gram-negative bacteria, according to one source (31). Other workers found that quaternary ammonium compounds were slightly less effective against the Gram-negative organisms tested (8). Mallman (25) summarized these discrepancies, stating, "By selecting the proper laboratory technic, we can show that the cationics are either poor or unusually good disinfectants."

The most widely used quat is benzalkonium chloride, which is a mixture of alkyldimethylbenzyl ammonium chlorides (26) and marketed under a variety of trade names. It has been widely used in hospitals for disinfection in surgery, and in sanitizing utensils, floors, walls, soiled linen, and in other applications. Since benzalkonium chloride (26) is incompatible with anionic detergents, such as soap, and the mineral content of hard waters interferes with the bactericidal action of the quats (11), it would not be suitable in hand-washing procedures for food handlers whose hands are in constant contact with soaps, detergents, waters of varying degrees of hardness, and food.

DISCUSSION AND CONCLUSIONS

Because of the paucity of studies on food handlers per se, it is difficult to make an absolute judgment of the type of antibacterial agent that ought to be incorporated into hand-washing agents for use by food handlers, or whether one should be used at all. Most of the studies on the efficacy of hand washing compounds have been directed to the evaluation of cosmetic applications or hospital procedures. The standards of efficacy in the former are not sufficiently critical for adoption in the food-service environment, and those for the latter may well be too stringent. At present, the choice of an antibacterial agent for the food handler must be made without full knowledge of the extent to which food pathogens may colonize on the skin of the worker, although allowance for such an eventuality should probably be made.

In the existing literature, the development of an antibacterial residue on the skin, such as occurs with the bithionols, is stressed. In itself, the maintenance of a continuing low level of bacteria on the hands may not be sufficient evidence for the presence of an active residue with all agents, since Price (29) has demonstrated that full establishment of the normal skin flora after the skin has been thoroughly degermed may require as much as 1 week, regardless of the method used. Nevertheless, the apparent residual antibacterial activity of the bithionols requires consideration of this aspect in the study of hand sanitizing agents.

The aim of hand sanitation is to prevent the transmission of possible pathogenic organisms from the hands through food or from food to food via the hands of the food handler. Since it is obviously not practicable to depend on the continual and exclusive use of antibacterial hand soaps outside of working hours, optimal control of skin bacteria during the hours in which food is being handled is the best that can be achieved. The maintenance of an efficient antibacterial residual on the skin can then probably be subordinated to other considerations. The antibacterial agent should not be chosen on the basis of activity against staphylococci and the Grampositive bacteria alone. Food handlers may also harbor many Gram-negative bacteria of significance, such as Salmonellae and pathogenic E. coli, as well as Entamoeba histolytica or other pathogens, either as transient flora or possibly as established residents acquired through contact. The control of transient organisms by a more positive method than simple removal by soap and water may be desirable, since transfer from one food to another should be minimized. Also, the consistent use of a broadspectrum antibacterial agent during the working day would help to prevent possible colonization on the skin of bacteria acquired from foods.

If all of these points are considered together, a good hand-washing agent for food service establishments probably should: (a) kill a wide variety of possible pathogens (inhibition is not sufficient because a single viable one transferred to a food may, under optimal conditions, multiply once the agent is diluted by the food); (b) be present, if possible, in sufficient residual concentration from one ablution to another to effect control during the day; and (c) be non-irritating to skin.

Iodine is the only agent reviewed in this report that appears to satisfy the above criteria in most respects. It is bactericidal and active against a wide variety of both Gram-positive and Gram-negative bacteria and other organisms. Although Blatt and Maloney (10) state that skin flora recovers more rapidly after iodophor treatment than after the application of hexachlorophene treatment, this point requires further study, in view of Price's findings (29) on regeneration of the bacterial population of the

skin. Any iodine preparation selected for study should have a high free-iodine content, whether it be a solid soap or a detergent-iodine complex. It should meet all of the general criteria accepted as necessary for performance of germicidal agents whereever sanitation is important (13). Its penetration into skin and its residual effect during the working day should be determined, as well as the degree to which it is removed by contact with food substances. Emollient additives must be incorporated, and their effect on the germicidal efficiency of the preparation must be definitely established.

The need continues for simple and more reliable and reproducible techniques for measuring changes in the skin flora. Particularly, a method should be devised for the study of frequent changes in levels of skin bacteria that occur during the working day as a result of exposure of the food handler to different foods. Such a method could conceivably shed light on the extent to which hazardous bacteria may be transmitted between ablutions as well as the degree to which a sudden contact with heavily contaminated food staples may overwhelm the capacity of a residual bactericide on the skin to control the spread of these organisms.

From the present consideration of the status and efficacy of the available hand sanitizing agents, one may conclude that frequent and thorough use of ordinary hand soaps, with the aid of a good brush during the working day, is, for practical purposes, about as efficient in controlling skin bacteria as the commonly available germicidal soaps. Practical experience has shown that the frequency of handwashing by food service personnel may be greatly increased by installing hand washing facilities in the working area, because of the tendency for personnel to correct one another's lapses in hand sanitation as they occur. This simple expedient tends to greatly enhance the efficacy of the simple soap and brush procedure.

A recent study (9) was reported on the efficiency of bar soaps, without antibacterial additives. The results show that bar soaps do not transfer bacteria among individuals in normal use, nor do they support bacterial growth.

Undoubtedly, some additional benefit will accrue from the use of residual germicidal soaps if they are considered as a supplement to, rather than a replacement for, thorough and frequent scrubbing. While deposition of residual germicide on the skin may be real, it may be nullified by the narrow microbial spectrum affected, possible neutralization by kitchen detergents and food constituents, and the tendency of some germicides to produce sensitivity reactions in some persons.

The use of formulations containing agents with a

broad antibacterial spectrum, such as iodine, presents attractive possibilities which, however, require much further testing in the food service environment.

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BACTERIOLOGICAL COMPARISONS OF HOT PROCESSED AND NORMALLY PROCESSED HAMS

JAMES D. PULLIAM AND DONALD C. KELLEY

College of Veterinary Medicine, Kansas State University, Manhattan

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SUMMARY

A preliminary study of the bacteriological comparisons of hot processed and normally processed hams was made in cured unsmoked and cured smoked hams. Higher bacterial counts were found in the hot processed hams. Staphylococci were isolated from both hot processed and normally processed hams. Coagulase-positive staphylococci were isolated from one hot processed ham.

Hot processing of pork, which could reduce overhead costs in the packing industry (6) was bacteriologically compared with normal pork processing methods. In hot processing, carcasses are cut and hams are pumped with curing pickle immediately after slaughter. The pumped hams are then placed in iced pickle to cool and cure.

MATERIALS AND METHODS

Processing.

The hams from 15 hogs were used. The left side of each carcass was processed in the normal manner, and the right side was cut prior to cooling. Hot processed hams were cut and pumped with pickle within one hour after slaughter. Normally processed carcasses were chilled for 24 to 48 hours before processing. All hams were artery and stitch pumped up to 10% increase in weight with the same type 65° pickle solution. After being pumped, the hot processed hams were placed in iced pickle and cooled at 3 C. All of the hams were then cured for 14 days at 3 C. Half of each ham group was held in the cured, unsmoked state for bacteriological sampling. All hams, unsmoked and smoked, were frozen and held at -20 C for 18 months prior to bacteriological studies.

Sampling.

Composite 25-gram subsurface and deep samples were aseptically cut from each ham after the surface area had been sterilized with a hot spatula. The 25-gram samples were mechanically ground in sterile blenders with 225 ml of sterile water.

Bacteriological comparisons.

Four bacteriological comparisons were used to compare the two processing methods:,

1. Total plate counts were made following standard methods (1) except the plates were incubated at 37 C for 48 hours. Appropriate water blanks were used to make dilutions of 1-10, 1-100, 1-1000, and 1-100,000. Duplicate pour plates, using plate count agar², were made from each dilution.

2. Coliform counts were made in Violet Red Bile $Agar^2$ plates using 1 ml of each of the first three dilutions. All of the counts were run in duplicate at each dilution.

3. To detect the presence of significant staphylococci, 3 to 5 ml of the 1-10 dilution was initially incubated in nutrient broth as an enrichment at 37 C for 72 hours. Isolation of staphylococci was on Mannitol Salt Agar². Tubes of Bacto-Brain Heart Infusion² were inoculated from colonies isolated on Mannitol Salt Agar. Two drops of each 18-hour Bacto-Brain Heart Infusion culture were incubated with 0.5 ml of Bacto-Coagulase Plasma². Tests were run in duplicate using a 1-3 dilution of fresh rabbit plasma the second time. Morphologically typical gram positive cocci that coagulated plasma were presumed to be significant staphylococci.

4. To detect organisms of the family *Enterobacteriaceae* that normally cause food-borne infections, portions of the nutrient broth enrichment cultures were streaked on SS Agar², Desoxycholate Citrate Agar² and E. M. B. Agar². Further identification of the *Enterobacteriaceae* was done using Kligler Iron Agar², Russell Double Sugar Agar², Urea Broth², and Phenol Red Tartrate Agar². Identifications were based on culture characteristics and gram stain characteristics (4, 8). Salmonella isolates were to have been identified by serotyping.

RESULTS

Average number of bacteria per g is shown in Table 1. When total counts were compared statistically by "t" test (9), they were significantly higher (.05 level) in hot processed hams than in normally processed hams before the hams were smoked. With smoked hams, counts were low regardless of processing method.

The coliform counts were less than 1 per g by both processing methods when examined in cured unsmoked hams and in cured smoked hams.

None of the organisms of the family *Enterobacteriaceae* that normally cause food-borne infections were isolated from either group of hams.

Staphylococci were isolated from 67% of the hot processed hams and from 47% of the normally processed hams. Coagulase-positive staphylococci were isolated from one hot processed ham.

DISCUSSION

Even though the freezing process kills about half the bacteria, and frozen storage causes the number of bacteria to decrease slowly (5), the relative numbers and kinds of bacteria found are significantly different in the two processing methods. Average

¹Contribution No. 225, Veterinary Medicine, Kansas Agriculture Experiment Station, Kansas State University, Manhattan, Kansas.

²Difco Laboratories Inc., Detroit, Michigan.

TABLE 1. NUMBER OF BACTERIA IN CURED HAMS AS DETERMINED BY TOTAL PLATE COUNT METHODS

(Cured unsmoked hams)

Normal pro Left side of	cessing carcass	Hot processing Right side of carcass			
Sample no.	Avg./g	Sample no.	Avg./g		
1 L	190	1 R	1430		
3 L	440	3 R	1020		
5 L	90	5 R	340		
7 L	160	7 R	120		
9 L	170	9 R	80		
11 L	20	11 R	0		
13 L	20	13 R	60		
15 L	° 0	15 R	7900		
Arithmetic A	vg./g 136	Arithmetic Av	/g./g 1494		
Range 0	- 440	Range 0 -	- 7900		
Log avg.	/g 54	Log avg./	/g 196		

(Cured smoked hams)

Normal pro Left side of	cessing carcass	Hot process Right side of	sing carcass
Sample no.	Avg./g	Sample no.	Avg./g
2 L	10	2 R	0
4 L	0	4 R	5
6 L	20	6 R	0
8 L	0	8 R	0
10 L	0	10 R	0
12 L	5	12 R	0
14 L	5	14 R	5
Arithmetic A	.vg./g 5.7	Arithmetic Av	g./g 1.4
Range 0	- 20	Range 0 -	- 5
Log avg.	/g 3.3	Log avg./g	g 1.5

number of bacteria found in cured, unsmoked hams was greater than that found by Deibel (3). He conducted counts immediately after curing and before smoking and found the interior of hams to possess very low or undetectable numbers of bacteria, usually fewer than 100 per g (3). Bacteria, particularly staphylococci and lactobacilli, are believed to play an important role in ham curing in Europe where an inoculated cover brine and longer curing times are used (7) and bacterial action is believed to improve the flavor and color. Even though curing brines have been found to average one million bacteria per ml, it is not probable that microorganisms play any role in the manufacture of modern American ham (3).

Low counts in smoked hams of both groups result from the smoking process, which is partial sterilization by heat, drying and chemicals from the smoking process (5). The low numbers of *Enterobacteriaceae* were thought to be due to the nitrite in the curing mixture. Nitrite, particularly in its unstable form, is bactericidal for *Enterobacteriaceae* (2).

The high incidence of staphylococci is expected because of the high salt concentration. Staphylococci are active in many pickling solutions (5) and have been found in 80 to 100% of the brines used in Germany (7).

Cutting and injecting curing pickle into hot hams has greater possibilities for contamination and growth of microorganisms. Although bacterial counts were low in both groups after smoking, hot processed hams had higher counts before smoking, indicating that hot processing may allow greater growth of potential pathogens, such as the coagulase-positive staphylococci which were found. Further studies of hot processed pork are justified.

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BACTERIOLOGICAL SURVEY OF FILLETING PROCESSES IN THE PACIFIC NORTHWEST

IV. BACTERIAL COUNTS OF FISH FILLETS AND EQUIPMENT

WAYNE I. TRETSVEN

Bureau of Commercial Fisheries, Technological Laboratory Seattle, Washington

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SUMMARY

Bacterial counts of the incoming fish, of the fillets produced from them, and of the surfaces of the processing equipment used were determined in 11 filleting plants in Oregon and Washington. The range in counts was wide. Counts among incoming fish in summer and winter and among fish of different quality, however, did not differ significantly. Washing the fish reduced the counts about 80%. During filleting, flushing the boards on which the filleting was done reduced the contamination on the boards by 85% and reduced the contamination of the fillets by 90%. Because the bacterial load on the fillets is due primarily to contamination, comparing the count on the incoming fish with that on the fillets produced from them was found to be a good index of sanitation during processing.

A bacteriological study was made of the filleting operations in the commercial plants in the Pacific Northwest. As in similar studies in other food-processing plants (11, 12), methods of sampling were first studied; the swab method was found to be most applicable (18). Its limitations were then determined, and innovations were developed that enhanced its use (19). A third part of the study involved the bacteriological and physical effects of improper pughing (forking) of the fish (20). The presently reported fourth part was concerned with in-line sampling of the products and facilities from the time the fish are landed until the fillets are packaged in the filleting plants. The specific objectives of the study were:

1. To determine the bacterial counts of the incoming fish, the equipment, and the fillets produced in the filleting plants in the Pacific Northwest.

2. To evaluate the fillet-processing procedures from a bacteriological viewpoint.

3. To assist the industry to raise the quality of fillets by improving filleting conditions based on the data accumulated.

Bacteria commonly are found in great numbers in the slime on the surface of fish (5, 8, 9, 14) with counts usually between 10^3 and $10^5/\text{cm}^2$ (16), and in the digestive tract (7, 8, 14). "The surface count may increase to 10^8 to $10^9/\text{cm}^2$ during storage in ice" (16). Although the flesh of sound fish is considered to be sterile (5, 8, 13, 14), it becomes contaminated either directly or indirectly during the filleting process (5, 8). "In plants where the fish were not washed prior to filleting, there was a direct relation between the bacterial count and the number of days the fish had been out of water" (3). As was found in other food-processing plants (11, 12), Castell (3) showed that washing the incoming products and the processing equipment is an effective means of reducing contamination. Also, without washing, there is a very rapid buildup of contamination. By washing the fish, "It is possible to reduce the surface load of bacteria by about 90% or more" (8). By washing the fish before filleting, the bacterial load on the fillets is reduced from 80 to 99%, which adds from one to six days to the keeping time of fillets stored at 32 to 33 F (6). Ninety percent of the bacteria on the fillets was picked up from the fillet cutting board after the fillet was skinned (7). Mechanical skinning resulted in lower counts on the fillets.

Water sprays were found to be superior to immersion of fish in water for washing (2); also, water sprays were found to be more effective than were sprays of dilute acid solutions or alkaline phosphate solutions (17). No advantage was found in using chlorine-containing solutions for washing fish (8).

Solutions (dips) used for treating fillets were often found to be an important source of contamination (18).

In 1947 (3) the average fillets left the cutting boards with 1,000,000 bacteria per gram. In 1957 a viable count standard of 250,000/g was applied to filleted fish in Canada as a measure of sanitary quality on the assumption that the flesh of the fish entering the plant was essentially sterile.

PROCEDURE

This study involved 46 intermittent and unscheduled in-line bacteriological surveys of 11 filleting plants in Oregon and Washington during 1960 and 1961, 20 during the winter and 26 in the summer. Samples were collected in each plant at least twice, and 20 to 30 samples were collected each time. Surface sampling of the incoming fish, processing equipment, and final fillets were done by the swab technique of Tretsven (18, 19), and the results are reported as bacterial count per 2 cm². Samples (1 ml) of ice and dip solutions were also collected aseptically in clean, sterile, screw-capped glass vials. All of the samples were kept and transported to a laboratory in an insulated sampling kit that maintained the temperature between 0 and 5 C. About 30 to 90 minutes elapsed from the time a sample was collected until it was plated for standard aerobic plate counts by the technique given (18). The logarithmic average of the plate counts was used for comparison of the bacterial counts.

Initially, in an attempt to limit variables, cod (Gadus macrocephalus) was selected as the fish to study. However, owing to the limited amounts of cod caught, the following incoming fish were also sampled in-the-round and during their processing: lingcod (Ophiodon elongatus), rockfishes (Sebastodes sp.), and Dover sole (Microstomus pacificus).

The general quality of the incoming fish was classified as



Figure 1. Range and average bacterial counts on fish, processing equipment and fillets found in-line sampling of eleven filleting plants in Pacific northwest. Broken line, from fish in ocean (theoretically, no count) to fish in plant.

excellent, fair, or poor. Each classification was based on the judgment of the technician who collected all of the samples, and the opinions of the plant personnel doing the filleting. The appearance, feel, odor, and in some cases, knowledge of the fishing and storage conditions aboard the vessel were criteria used in classifying the fish comprising the lot of incoming fish. Poor represents a low level, approaching the borderline of marketable fish for human consumption. Fourteen of the lots were judged to be excellent; 23 were fair; 8 represent mixed lot of fish of two qualities, fair and poor; and one lot was poor.

Results

The results of the 46 in-line samplings, including the range of bacterial counts and the average counts of each item sampled, are shown in Figure 1.

Bacterial counts on 98 samples of the poorer quality (fair and poor) incoming unwashed fish varied from 180,000 to 9,500,000/2 cm² and averaged 640,000/2 cm², whereas those on 45 samples of excellent quality fish varied from 12,000 to 3,000,000/2 cm², and averaged 470,000/2 cm². Counts of the equipment and of the fillets of the poorer quality fish were quite similar to those of the excellent quality fish.

Counts of the incoming fish ranged from 39,000 to 9,500,000/2 cm² and averaged $590,000/^2$ during the summer and 180,000 to 9,500,000/2 cm² during the winter, with an average of 510,000/2 cm². Similarly,

average counts of the equipment and fillets during the summer were essentially the same as those during the winter.

Washing Fish

On 34 direct comparisons involving 142 samples of unwashed fish, the average bacterial count was $570,000/2 \text{ cm}^2$; after commercial washing of the fish, their average count was $120,000/2 \text{ cm}^2$, a reduction of 79% (see Figure 2).

Flushing During the Filleting Process

Eight direct comparisons were made to determine the value of flushing the filleting boards with water. The same commercial filleters and similar fish were used. Before the filleting boards were flushed, the average bacterial count of the filleting boards was 610,000/2 cm², and the average of the fillets was 100,000/2 cm². After about 5 min of continuous flushing of the filleting boards during filleting, the bacterial counts of the boards averaged 96,000/2 cm² and that of the fillets averaged 11,000/2 cm², an 84 and 89% reduction in counts, respectively, due to flushing with water (see Figure 3).

Use of Chlorine

In an attempt to further reduce the bacterial counts of filleting boards and fillets, one plant experimented with water containing about 5 ppm available chlorine to flush the filleting boards continuously while they



Figure 2. Bacterial counts on fish and fillets in 34 in-line samplings.



* ml. (LIQUID, MELTED ICE)

Figure 3. Bacterial counts of filleting boards and fillets, before flushing and after five minutes continuous flushing of the filleting boards with water (8 comparisons).

were in use. In this trial, the bacterial counts of the boards averaged 66,000/2 cm² and of the fillets 60,000/2 cm² when flushed with water; when flushed with the chlorinated water, the boards had a count of 20,000/2 cm², and the fillets a count of 40,000/2 cm².

As a result of these findings, this plant installed in-plant chlorination of the water for their fresh fish and filleting operations. The water containing 5 ppm available chlorine was used in washing the fish and in spraying the filleting operation. Before the change-over, the counts on the fillets averaged 770,000/2 cm², about 20 min after the change-over to the chlorinated water, the bacterial count of the fillets averaged 48,000/2 cm².

DISCUSSION

The operations in the various filleting plants differed in that some were highly mechanized, whereas others were largely manual. In general, however, the same type of facilities was used to process the various species. The fish were filleted by hand in every plant. Most of the species used were kept and handled separately; however, in a few cases, two or more species of fish were on the same filleting line at the same time. In such cases, the filleters were responsible for keeping the fillets of the various species separate. There seemed to be no particular pattern in which the species were filleted. When possible, the oldest or most perishable fish were processed first. Ordinarily there was no attempt to clean up the equipment between the handling of the various species; the bacterial counts obtained therefore cannot be attributed to any one species.

In most of the plants, the incoming fish were washed by spraying them with water while they were on a conveyor or while they were in a revolving drum that was also used as a descaler. In one plant, the fish were washed by being conveyed to the filleters by means of a flume of water. In two of the plants, the fish were not washed.

The results shown in Figure 2 are similar to findings of other investigators in that there is a wide range in bacterial counts for each of the items sampled.

The incoming fish tend to affect the counts at the subsequent filleting stations: for example, higher counts were obtained at the filleting stations and on the final fillet when the count on the incoming fish was high than when it was low.

While counts on the incoming fish varied considerably, the average counts of the fish and items sampled during the summer were quite similar to those of the winter. The average counts of the fish judged to be excellent were also similar to those of poorer quality.

At the beginning of this study, bacterial counts on some of the fish conveyors were higher than were the counts on the incoming fish. During this study, two of the plants installed continuous brushing and washing devices on the conveyors and thereby reduced this source of contamination.

Counts on the gloves of the filleter were higher than those on the unwashed fish. Undoubtedly this is due to the fact that the slime in and about the head adheres to the glove, inasmuch as the filleter grasps the head at the eyes and gills.

We assumed that there was a progressive buildup in bacterial load during the day. We found, however, that the bacterial counts on the filleting boards increased or decreased within a few minutes after the filleting of higher- or lower-count fish. Such changes were noted in the samples regardless of whether the fillet boards were flushed or were not. The bacteria on the fillets are primarily due to contact with slime, blood, viscera, etc., on the filleting board. Owing to the abrasive, scraping, and wiping actions in processing the fish on the board, the contamination of the fillet with bacteria actually growing on or within the boards did not significantly affect the bacterial count.

At each of the inside and skinside (flesh) cuts, the bacterial counts were usually nil, except when the flesh has been broken (5, 20). Not all the fillets were skinned, and a skinning machine was used occasionally in one plant.

Ice surrounding the incoming fish was found to vary considerably in appearance and bacterial count. We attributed this variability to contamination from the fish. High counts were observed in some instances regardless of whether the ice was treated with antibiotics or untreated.

Dip solutions were used in some of the plants. When used, the solutions often were found to be quite contaminated, having an average count of 350,000/ml; the fillets before being dipped averaged $140,000/2 \text{ cm}^2$, and after being dipped, $170,000/2 \text{ cm}^2$ (Figure 2).

Processing Index

Early in this study, we noted that the bacterial counts on the final fillets tended to approach, and even to exceed, those of the incoming fish. Thus, the ratio of the count of the incoming fish to the



* ml. (LIQUID, MELTED ICE)

Figure 4. Pattern of good operation showing relatively low count on incoming fish and lower count on final fillet.

count of final fillet is an index of the sanitation during the processing as is also the slope of the line connecting the count of the incoming fish with that of the fillets (Figure 4).

Industry Improvements

Most of the plant managers were interested in our results and asked how they could improve their processing. During the study, a number of changes (some quite costly) were made to improve the quality:

1. A number of the plants improved their fishwashing operations.

2. Two plants installed devices to clean the fish and fillet conveyors continuously.

3. Two plants provided means for flushing the filleting operation with water; and later, one of them installed in-plant chlorination.

4. Reuse of ice in the plants was discontinued.

Since 1960 and 1961, when the results of our study were first made available to the 11 plants, more improvements have been made by them and also by other West Coast plants in the United States and Canada. Practically all of the plants now wash the incoming fish. Most of them flush the filleting boards with plain water or chlorinated water while filleting. About half of the plants have installed in-plant chlorination. Based on the findings of the related study concerning the effects of pughing fish improperly (20), the manager of one plant refused to buy any fish that have been pughed and has eliminated the practice of pughing fish within that plant.

Conclusions

Bacterial counts of the surfaces of the fish, equipment, and fillets furnished a good basis for determining sanitation in 11 filleting plants in Oregon and Washington.

The average bacterial count of incoming summercaught fish was not significantly higher than that of incoming winter-caught fish. The difference in average bacterial counts of the fish classified as fair or poor and those classified as excellent was not significant.

Washing the incoming fish reduced the bacterial count on the fish 79%.

Flushing (washing off) the fish, knife, hands, and filleting boards as the fish were being filleted reduced the bacterial counts of the filleting boards 84% and that of the fillets 89%, compared with counts obtained before flushing.

Acquainting industry with our findings resulted in substantial improvements in equipment, processing methods, and quality of fillets produced.

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UTILIZATION OF DEFOAMERS IN THE DETERGENT INDUSTRY

S. B. CRECELIUS

Economics Laboratory, Inc., St. Paul, Minnesota

In general, in the chemical field defoamers have had a wide application for many years. In chemical processing, the control of foaming has always been a problem, both in aqueous and non-aqueous systems. Through the years a number of different types of defoamers have been developed, one of the most common and well-known being the silicone type, such as Dow Antifoam A and others. In addition to this type, use has been made of various other chemicals, such as long chain hydrocarbons, long chain alcohols, fatty acids and phenolic compounds. Most of these defoamers have found application in aqueous and non-aqueous systems having to do with other chemical applications than the detergent industry.

However, in the detergent and cleaning field we run into a special situation in the field of defoamers. For many years, the existence of foam and detergency were considered synonymous. It was felt that good cleaning action was always accompanied by foam and was a necessary part of the total operation. It has now been found that foam, in a number of mechanical cleaning operations, is actually a decided detriment to good cleaning practice in that it clogs lines, reduces the pressure of cleaning, slows down the proper agitation and mechanical operation of the cleaning devices, and in general presents a problem in the total cleaning operation.

In early attempts to solve these foaming problems by conventional means, it was soon found that defoamers, such as the silicone type, were inadequate for detergent additives. Although they were adequate as defoamers, they were found to plate out or absorb on the surfaces to be cleaned, causing those surfaces to be much more difficult to wet and actually deterring the detergent action. Products such as long chain hydrocarbons often used as defoamers in some applications and also had the effect of introducing gross soil which interfered with rather than helped the total cleaning operation. It was obvious then, that some other type of defoaming mechanism had to be developed to meet this special situation found in detergency. It was through this need that

¹Presented at Klenzade's 25th Educational Seminar, Yosemite National Park, California, April 6-7, 1964.

the present detergent defoamers were actually developed.

Types Foam Encountered

Foam in the cleaning operation can come from a number of sources-the first and most obvious source, of course, is the detergent itself. Detergents, such as ABS or the commonly known heavy duty anionic detergents used most widely in the laundry and hand liquid cleaning operations and known chemically as alkyl benzene sulfonate, give considerable foam when used in almost any cleaning operation. Likewise do the sulfated alcohols, such as sodium lauryl sulfate, the alkylolamids, and most of the less wellknown anionic detergents. Consequently, it was soon learned that these high-foaming anionic detergents could not be used to any great advantage in mechanical cleaning. They do not find their way into these types formulations where mechanical cleaning is used, except in the laundry industry. Soap also gives considerable foam when used in moderately soft water. When used in hard water, soap loses its foaming action; but on the other hand, it also loses considerably part of its detergency in that it is inactivated and precipitated out as a hard water or calcium soap scum.

The nonionic type of detergents may be divided into three main classes. The first class is the alkyl aryl poly ether type, such as the well-known Rohn & Haas Triton X-100, or Igepal CO-730. These are considered to be high to medium foamers. The second class, introduced largely by Wyandotte as the Pluronics, are block polymers of ethylene and propylene oxide. These products can be considered to be low foamers or non-foaming type of detergents, but do not defoam. The third class of nonionic materials is not only non-foaming but actually are defoamers. These are also block polymer-type materials similar to the Pluronics, but have a further modification which gives them their defoaming properties. It is these materials which we intend to discuss today.

To date, we have found no way of controlling or defoaming the foam caused by the highly-active anionic-type detergents, or even the high foaming nonionic-type detergents. Hence, the only course of action here is to not use this type, when formulating a detergent for a non-foaming of operation.

FOAM FROM SOILS

One source of foam in detergent cleaning comes from the soil itself. The greatest source of soil which causes foam is the protein content of the soil. This foam is agitated and increased as it is broken down through hydrolysis and oxidation of this soil in the cleaning process itself. Another source of foam is from the hydrolysis of fats, or rather the saponification of fats, in caustic cleaning. It forms soap, which in turn foams if the water is soft or has been conditioned by polyphosphate builders. This, however, is not nearly as great a source of foam as it was originally thought and is much less a problem than that of the protein. A third source of foam is the carbohydrates, which do contribute somewhat to the foam properties of a cleaning situation. However, they are also much less of a problem than the protein materials.

It can readily be seen then that in any kind of food cleaning operation, such as in the dairy and food processing industry, we are readily confronted with foam problems. This is due primarily to: (a) we do have highly developed mechanical cleaning in these fields with our automated and in-place cleaning systems; and (b) we have in existence the proteins, carbohydrates and fats which cause our greatest foam conditions.

Hence, the control of foam in this industry is quite important. It is equally and likewise important in the control of foam in the food service or dishwashing industry, where all the necessary components also exist. It is important in an animal cage washing industry. It is also important in the beverage and brewery industry, where protein, fats, and carbohydrates are a large component of the food soil which we are necessarily trying to clean.

The defoamer is usually from 1% to 3% of the total solids of the detergent formulation. Not only does this defoamer act as a detergent by the usual addition of wetting, emulsifying, and free rinsing properties, but in addition controls the foam produced by the food soils present. This, in turn, contributes to the increase in wash pressure and prevents the blocking of lines and pumps by foam and allows the cleaning operation to become much more efficient.

DESCRIPTION OF DEFOAMERS

Defoamers of the type discussed here are of the nonionic-type synthetic detergent. They vary in their chemical structure, but may be characterized in general as being of quite high molecular weight. These defoamers are rather new—they are presently in the stage of being processed for patent applications, so that their definite chemical structure cannot be divulged at this time. However, we may discuss some of their physical structure and behavior under different conditions of use.

One characteristic of these defoamers is that their

defoaming ability is definitely tied to the wash solution temperature in which they are used. In general, the defoaming action will not take place in cold water. As a matter of fact, these defoamers, when used in cold water or even in water around or slightly above room temperature, actually become foamers and foam somewhat in the same manner as the higher foam nonionic detergents discussed above. Hence, the use of defoamers in general must take place at temperatures ranging from around 110 to 180 F. Temperatures below 110 F result, in general, in much less defoaming action and at lower degrees you get no defoaming at all. Defoaming action increases up to about 140 F where it begins to level off, and there is very little increase in defoaming action between 140 F to 180 F, beyond which there is no increase at all.



Figure 1. Force loss (ΔF) vs. Temperature. 4% concentration of detergent.

There is some difference in the defoaming action of certain detergents with different structures. This is shown in Figure 1. We studied three different types of non-ionic defoamers at different temperatures, and plotted the wash pressure achieved versus the temperature of the wash solution in which it was accomplished. It will be noted that all of the defoamers perform in a similar manner, at higher temperatures, while down in lower temperatures, between 90 F and 110 F, one of these defoamers gives considerably more action than the other two. This is a typical means which we use to evaluate the performance efficiency of a defoamer in a cleaning operation. The other requirement, of course, is that the defoamer also has good surface active properties.

FUTURE IMPROVEMENTS WHICH WOULD BE DESIRABLE

What, then, are some of the ways in which we would like to improve these detergent defoamers which we now have.

1. We would like, if possible, to be able to develop a defoamer which we could add to an anionic or high foaming type of detergent and get good results of the low surface tension extremely high activity per concentration of this anionic and still reduce the foam of that anionic to a point where it could be used in mechanical cleaning. So far, we have been unsuccessful in finding or developing a defoamer of this type.

2. We would like to be able to find a defoamer which would work at low temperatures (room temperature and below) and still give us a maximum of defoaming capacity. This, however, is not of as great importance as the first point, since most cleaning is done at fairly high temperatures.

3. We would like to develop an efficient defoamer which would be stable on highly caustic type of cleaners, and retain its defoaming efficiency. We have been successful to some degree in this extent and have some experimental defoamers in use, which appear to have much more caustic stability than the original ones which we have used.

4. We would like to develop a defoamer which could be used in the presence of chlorine and would be stable under these conditions. So far, we have been unable to do this, in liquid, since chlorine attacks the surfactant in liquid media about as readily as it will attack the food soil or micro-organisms in the wash solution. Hence, no liquid that we have ever put together with a defoamer has ever been successful in the presence of chlorine. We have, however, devised a method of putting these surfactants in solid materials, in the presence of available chlorine by a process which we call encapsulation. Here we encapsulate the surfactant in a separate preparation so that it does not come directly in contact with the chlorine bearing material and hence, it is quite stable for a long shelf life in a cleaning powder.

So far, we do not have the answer to these problems, although we hope to find it in the future.

THE GREAT AMERICAN DREAM

EARL L. BUTZ

Dean of Agriculture Purdue University West Lafayette, Indiana

The American economic and political system has brought living standards for all our people unparalleled in any other country on the face of the earth. One need only go abroad for a short time to any part of the globe to see convincing proof that this is true.

With only 7 percent of the world's population, we have three-fifths of the world's automobiles, half of the world's electric power production, over half of the world's telephones, two-fifths of the world's steel production capacity, a proportion of our young people in high schools and colleges that far out-strips any other nation, 98 percent of our farms electrified, radios, TV's, refrigerators, deep freezes commonplace in nearly all of our homes, new home construction passing the one million mark again this year for the 16th straight year, every community busy with construction of new schools, new churches, and new business buildings, a food production and processing industry so efficient that our growing population is one of the best fed on earth, and yet we have sufficient food surpluses for substantial distribution overseas.

Truly this is convincing evidence that "The Great American Dream" of yesterday is constantly translated into better living today.

The economic growth of our nation over the past couple of decades has been truly phenomenal. Our economy has doubled in size in less than 20 years. Reliable predictions are that it will double again in the next 20 years. Real income per person in America has increased by about 50 percent in the last 20 years. Indications are that this likewise will increase more than 50 percent in the next 20 years. This is a phenomenal rate of increase in the economic wellbeing of our nation.

If you think for a moment that our people aren't 50 percent better off, on the average, than they were 20 years ago, just drive 100 miles down any road in your home state and look at the new homes, new factories, new schools, new highways, families with two automobiles, TV antennas on the roof, larger share of youngsters in college, new churches going up all over the place, vast amounts of money spent for vacations and recreation, and a score of other evidences of economic well-being. Economic growth of this magnitude can be accomplished only on the solid foundation of research and innovation.

Surely there is nothing second-rate about a nation that can produce such a record, while at the same time it has also tooled up the world's most powerful defense machine. There is no need for us to alibi among the community of nations for anything we do or don't do. We so far outdistance our adversaries in terms of living standards and opportunity for everyone that any honest comparison is ridiculous. The same is true of our total national strength. It's high time we begin to act like the strong, responsible, and proud nation we always have been. We must stop rushing for the aspirin bottle every time some half-pint dictator chooses to sneeze. The highway to national destruction is strewn with appeasers and apologists. No one really respects a nation that pays off to the blackmailer. The international bandits that rule one-third of today's world understand only the language of firmness and strength. And that is the very language the United States is equipped to talk, if we only have the national courage to speak up. It's high time we do so.

A GREAT FUTURE LIES AHEAD

America is on the threshold of its most challenging and most prosperous decade. There is real opportunity ahead for anybody who has imagination enough to participate in the Great American Dream.

This is the age of science and technology, based on research. The frontiers of the mind have replaced the frontiers of geography. A thrilling experience awaits every citizen in this great land who has the capacity and the imagination to "dream constructively."

The scientific and technological advances we will experience in the next decade will be unparalleled in American agriculture and industry.

The geographic frontier in America is gone. No longer can a young man "Go West" and stake out his claim. But the scientific frontier in America is barely scratched. And the scientific frontier has no effective limit. It is limited only by the mind and the imagination of man. Its horizons are vertical, not horizontal.

Organized and imaginative research is the vehicle

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which will push the scientific frontier beyond limits we scarcely dare dream of today. It follows logically, therefore, that if we can keep our economy free and preserve an environment in which individual producers and scientists are free to dream a little about new techniques and new ideas, and to enjoy the fruits of their dreams, we shall experience phenomenal progress in the next generation. It is to this noble purpose that you who sit before me dedicate your energies.

This is a thrilling time to be permitted to live in this great land we call America. Yours is a wonderful opportunity to occupy a front-row seat for the scientific advances occurring in our colleges and universities, in our research foundations and laboratories, and in our business and financial institutions.

LET'S LOOK AT AGRICULTURE

Consider with me for a moment our total agricultural plant in this country, which affords an excellent illustration of an industry in the very middle of a far-reaching scientific and technological revolution.

Changes which have occurred in production and marketing during the past decade have been phenomenal. Still greater changes lie ahead.

American agriculture is now feeding our growing population on science and technology. Today the average farm worker in the United States produces enough food and fiber for himself and 30 others. A generation ago, in 1930, he produced enough for himself and only 9 others. A century previously, in 1830, one farm worker produced enough for himself and 3 others.

Indeed, the productivity of the American farm worker is so high that our problem in America is to learn how to live with plenty, rather than to struggle along with abortive attempts to restrict farm output the way we've been doing.

It is not necessary to travel very far among the world's food deficit nations to realize what a blessing we have in our American food surpluses. If one must choose between the problem of too much food and the problem of not enough food, I'll always choose the problem of too much. It's a blessed problem.

Our modern scientific agriculture, based on research and technology, is so efficient that we now feed and clothe our entire population with only 7 percent of our labor force on farms. The first claim of any society upon its total production resources is to get enough food to keep the population alive. This is true in primitive societies, in semi-developed societies, and in highly developed societies. We do this so efficiently in this country that over nine-tenths of our population is available to produce the wide variety of goods and services that make up the American standard of living.

By way of contrasts, Russian agriculture is so inefficient and so primitive that some 45 percent of their workers are required to produce enough food to keep the population going at a subsistence level. As a consequence, there just isn't enough manpower (or womanpower) left to produce things to make life very pleasant in that country. So long as our output per worker in agriculture remains five times above theirs, there can be little doubt concerning the ultimate outcome of the struggle between our two systems.

It would be possible to fill these pages with specific illustrations, from dozens of vocational areas in this country, of research that has returned its cost an hundredfold. But our interest is in the future. Let's dream a little about that.

Research—Gateway to the Future

It is sometimes said that this is the age of science and technology, based on research. From another point of view, it can be effectively argued that we are not yet in the age of science and technology. Rather, we merely stand on the threshold of this new age.

The important scientific discoveries which we take for granted today are really so recent, viewed against the long backdrop of the history of man, that they occurred only yesterday.

A simple illustration will prove the point. This is the age of speed, ranging from high-powered motorboats on our lakes to 18,000-mile-per-hour satellites soaring around the earth. Man has always tried to travel fast, but through nearly all of the 6,000 years of his recorded history on this earth he could travel no faster than a horse could carry him. This was about 30 miles an hour. This was top speed in Samson's day, in Caesar's day, as well as on the night Paul Revere rode from the North Church to Concord. It was not until the 1830's, with the invention of the locomotive, that man freed himself from the speed limitations of the horse. On that date he pierced the "oat barrier." How long ago was that?

Let's condense the 6,000 years of the written history of man into the 40-year productive span of an ordinary lifetime. Just ten months ago the locomotive was born.

About 1890 the first automobile appeared on the scene. This increased our rate of travel up to perhaps 40 miles per hour. This was six months ago.

In 1903 the Wright Brothers had their first airplane flight. They achieved a speed of 60 miles per hour. This was four and one-half months ago. In 1948 the DC-6 was a pretty fast means of transportation, going up to as high as 280 miles per hour. This was four weeks ago.

In 1956 Air Force fliers pierced the sonic barrier. This was really travelling. This was two weeks ago.

In 1958 Sputnik was launched, and Explorers I and II began circling around the earth at the phenomenal speed of 18,000 miles per hour. This was one week ago.

The most recent shot into space a couple of months ago occurred since we entered this room.

If you were to plot these rates of speed on a graph, the line would continue near the bottom for many centuries, rising slowly near the right edge, and in the last little while rising very rapidly. The line would be going up as a geometric progression. It would resemble the rate of bacterial growth under favorable circumstances. Or as somebody has said, the latter stages of that curve would look like the national debt.

In any event, a curve of this character would be convincing evidence that we are just entering the age of science. We are just beginning to scratch the scientific frontier.

OUR GROWTH POTENTIAL IS GREAT

In the short span of ten years, our population has increased by about 25 million to the current figure of 191 million persons. By 1980, it is estimated that the number will be over 230 million. And equally significant, it is estimated that by 1980 the national economy will have about doubled in size, and that per capita real income will be around 50 percent higher than at present. Such a rise will bring about a substantial increase in the demand for all kinds of goods and services, including many kinds that we don't even dream of today. That branch of American industry and commerce that keeps up with the march of progress through research will capture more than its pro rata share of this growing market. Our greatest error-during the past 20 years-has been in so frequently underestimating our growth potential.

There is a growing area for service and expansion ahead for an industry like yours, which accepts the responsibility for its own welfare and its own future, rather than sitting back and waiting for somebody else to do the job for it. You will play a central role during the next decade in this unfolding drama we call *The American Dream*.

OUR FREE ENTERPRISE SYSTEM WORKS

Our economic system of free enterprise is under constant attack, from within as well as from without. There are many well-meaning people among us who would turn increasingly to government for

security guarantees of one kind or another and junk the economic system which has made us great.

We must cultivate within our society an environment in which individual producers and scientists are free to experiment with new techniques and new ⁶ ideas and to enjoy the fruits of their labors. We shall experience phenomenal progress in the generation ahead if we can preserve our system of free prices and free enterprise.

There are too many people in America today who do not really believe in free enterprise but who still believe they can look to Washington for price supports, price regulations, price ceilings, and so on. We must get the point across to every citizen of our country that the incentives under free prices make our economy great and make it strong, and make it productive.

America's greatest danger today is our general indifference to the organized attempts to limit output per worker, per factory, and per farm. Indeed, most of us belong to groups which subscribe in one way or another to the strangling philosophy of limited output. One of the remarkable phenomena of our age is that America continues to grow in total production and standard of living, in spite of widespread organized attempts to prevent it. Most of us remain indifferent to those abominable practices round about us.

What we all seem to be doing in America today is somehow trying to get more golden eggs out of the goose that lays the golden eggs. Different groups approach the goose differently. Some groups say, "I'll squeeze the goose, and more eggs will come out somehow or other." Some people in government say, "I'll eat the goose and lay the eggs myself." But there are some of us in America—and I would like to count you among that group—who believe that the way to get more golden eggs over a period of years is to feed the goose, put a little fat on its back, pour a little profit back into it; and put a little investment back into it; let it eat an egg itself once in a while, let it enjoy some of the fruits of its own production efforts.

That's the system that made America the great, strong, prosperous nation it is. That's the only system that will keep it that way. Good government must help promote that kind of system.

Our potential production capacity in America is so great that we can never be destroyed from without, unless we first disintegrate and deteriorate from within.

FREE MEN WITH VISION WILL DO THE JOB BEST

We must be careful not to lose at home the very freedoms we struggle abroad to preserve. The whole American system and the whole American way of life are predicated upon the recognition of the dignity and the freedom of the individual.

In recent years, a great many sectors of American agriculture and industry have fallen under governmental controls and restrictions. In the case of agriculture, these controls have followed in the wake of price supports at uneconomic levels.

When a commodity gets itself into the fix of producing for the government rather than producing for a growing market, it almost inevitably finds a ceiling placed on opportunity.

Government production and marketing controls are essentially backward looking—not forward looking. They are based on history—not opportunity. Under these circumstances, producers with above average managerial capacity and ambition are severely limited in what they can do. They suffer, consumers suffer, and all America suffers.

The future of practically all of our business, industry and commerce is closely tied in with increased efficiency of production, processing and distribution, through which we can reach an evergrowing circle of consumers in the market place, here and abroad. Private enterprise must exercise the initiative in getting this job done.

We must strive ever to preserve a proper relationship between industry and government. We must always keep private enterprise the *senior* partner, and government the *junior* partner. It would be easy to reverse this relationship. There are many who would change it. We must be ever vigilant that industry assumes the responsibilities put upon it by our private enterprise system. Otherwise, government *will* become the senior partner. This is inherent in the very nature of government.

We must constantly remind ourselves that the advances of science can be applied most effectively by individual managers in a free industry, unhampered by a excessive governmental regulation and restriction.

We must keep before us always the concept that the fruits of our toil are produced to be consumed in useful outlets and not diverted into purposeless storage or uneconomic uses through politically-inspired governmental price and income support programs.

We must all work together to preserve an atmosphere in which freedom of choice remains one of the basic pillars of our economic system. Under such a system, individual producers and individual processors can grow and prosper as far as their ambition and their ability will take them. The right to succeed will be open to everyone. Conversely, the right to fail will also be present.

In our free society, the right to succeed carries with it the right to fail. If, through legislative action of one kind or another, we remove the right to fail, we ultimately will also remove the right to succeed beyond mediocrity. Men of vision and ambition do not want that. They know their future is most secure in an environment which guarantee freedom to choose, freedom to experiment, freedom to become more efficient, freedom to seek and develop new markets, freedom to dream and freedom to enjoy economic rewards if their dreams are successful.

One of the great challenges facing all of us is to see that our economy is not dominated by government—that government helps us rather than displaces private enterprise. We can do this only if we are willing to throw our influence on the side of keeping government the servant of all of us—not our master. We must keep government at our sides—not on our backs.

Our economic system of individual initiative, private property, competitive pricing, and of profits works for us. It has enabled us in profit-seeking America, most nearly of all the nations of the world, to achieve the communist goal—*Plenty for Everyone in a Classless Society*.

The system is worth preserving.

ASSOCIATION AFFAIRS

DEHART AND SHEURING HONORED AT GEORGIA MEETING

The Georgia Society of Registered Professional Sanitarians enjoyed a very informative and interesting program at its Annual Meeting at Jekyll Island on August 12 and 13. Outstanding speakers presented papers on such subjects as the professional status of sanitarians, public relations and the technique of making environmental health surveys. There were also interesting discussion panels on outdoor recreation development and on inter-professional relationships and communications.

At the Awards Banquet, featured by an address by Governor Carl Sanders of Georgia, two awards of merit were made. Garnett H. DeHart, Chief of the Food Control Section in the Georgia Department of Health was nominated Sanitarian of the Year. Garnett, who began his public career in Georgia 1944, has been successively county sanitarian and regional sanitarian at Macon and food technologist and assistant chief on the state staff before assuming his present position as section chief in 1965. Throughout his career he has manifested enthusiastic devotion to public health and constantly has worked for high levels in sanitation in the state. He has also been active in securing professional recognition for sanitarians and was highly instrumental in the passage and implementation of the Georgia registration law.



Garnett H. DeHart, (left) receives Sanitarian of the Year award consisting of a certificate and a \$100.00 check from Phillip Campbell, Georgia Commissioner of Agriculture.

Garnett is considered to be an outstanding recipient of the Sanitarian of the Year Award.

In turn Garnett had the honor to present a special award to Dr. John J. Sheuring, Dairy Science Department, University of Georgia. In nominating John Sheuring for the award it was pointed out that he was one of the earliest advocates for establishment of a professional rank for persons engaged in the field of sanitation. Through his connection with the university programs in better milk and food quality, John recognized the need for professional training



Left to right, C. M. Graham, Jr., President, Georgia Society of Registered Professional Sanitarians, Governor Carl Sanders of Georgia, Eucle George, incoming President, Phillip Campbell, Georgia Commissioner of Agriculture.



Dr. John J. Sheuring receives award from Garnett H. DeHart.

and a status of professional recognition for all sanitarians and worked untiringly to that end.

One of his first steps was participation in the founding of a state affiliate of the International, Milk, Food and Environmental Sanitarians Association in 1952. John has donated a great deal of his time and energy not only to state affiliate activities but also to the national organization which he very capably served as president in 1961.

Largely as a result of the efforts of the state group and of John's activities as secretary, the Sanitarian Registration Act became a fact in 1957. John has continued to serve as secretary of the Georgia Society of Registered Professional Sanitarians since its inception. The Special Award given him at the 1965 annual meeting is considered a fitting recognition of his long-time interest in sanitation and his dedicated efforts on behalf of the professional sanitarian.

*Photos courtesy of Mr. Marion Kelley.

NEWS AND EVENTS

J. C. FLAKE NEW SECRETARY OF NATIONAL MASTITIS COUNCIL

The National Mastitis Council announces the appointment of Dr. J. C. Flake of the Evaporated Milk Association as secretary of the Council. Dr. Flake will assume the administrative duties of Executive Secretary, a position formerly held by George W. Willits.

In addition to the appointment of Dr. Flake, the Executive Committee of the National Mastitis Council at its last meeting decided to maintain the headquarters office of the Council at 118 West First Street, Hinsdale, Illinois.

Plans for the next annual meeting of the National Mastitis Council were discussed and it was decided that the theme of the meeting will be, "Mastitis, What Can We Do About It?" A program is being developed that will explore the role of all agricultural workers in helping farmers control or prevent the ravages of this most costly disease of dairy cattle.

The annual meeting will be held February 3 and 4, 1966 at the O'Hare-Sahara Inn, Chicago, Illinois.

SHORT COURSE IN CHEESE MAKING AT WISCONSIN UNIVERSITY

A special four-week Winter Dairy Course in Cheese Making will be offered by the University of Wisconsin department of dairy and food industries. The course will run from Oct. 25 through Nov. 19. Subjects to be featured in the lecture portion of the course will include principles of cheese making, milk composition and testing, dairy arithmetic, dairy mechanics, dairy bacteriology, starter management, dairy sanitation, and laws and regulations covering cheese. Special laboratory sessions will cover starter making and testing, analysis of milk and cheese, and grading of cheese. Students will be able to gain experience in the manufacture of many varieties of domestic and foreign type cheese using the University's dairy processing facilities. Cottage and other soft type cheese will also be made.

Further details on the short course may be obtained from H. E. Calbert, Chairman, Department of Dairy and Food Industries, University of Wisconsin, Madison, Wis., 53706.

DATES CHANGED FOR RESEARCH AND DEVELOPMENT ASSOCIATES MEETING

The Board of Directors of the Research and Development Associates, Inc. have changed the dates of the fall Military-Industry Conference to be held at the U. S. Army Natick Laboratories, Natick, Mass., to October 26-27, 1965, with a general theme of "Food Preservation and Packaging Techniques."

Registration information may be obtained by contacting Harlan J. Wills, Lt. Col., AUS-Ret'd., Executive Secretary, Research and Development Associates, Inc., U. S. Army Natick Laboratories, Natick, Mass. 01762.



Sanitarians from intermountain states at Colorado State University Food Conference.

FOOD TRAINING CONFERENCE AT COLORADO STATE UNIVERSITY

The first annual Food and Health Conference was held at Colorado State University at Fort Collins on July 20-22, 1965. Sponsored by the USPHS Division of Environmental Engineering and Food Protection, the conference was presented by the CSU Department of Microbiology.

The primary objective of the conference was to provide training for public health personnel whose work requires inspection of various aspects of vending machine operation, ice cream manufacture and vending, and frozen food plants. Participants represented departments from the intermountain states.

Subjects covered and the discussion leaders were: Psychrophilic Growth of Bacteria, N. A. Sinclair; Bacterial Growth Responses to Temperature, Carl A. Frey; The Ice Industry, Dispensing and Bacteria, S. M. Morrison; Food Service and Application of Automation, Burritt Tomlinson; Time-Temperature Effects on the Behavior of Microorganisms in Foods, Robert Angelotti; Problems in Food Vending, Dudley Leonard; Sandwich Vending, C. A. Frey; Frozen Precooked Foods, M. T. Bartram; Vending of Food and Beverages, W. F. Bower; and a panel on Vending Machine Operation and Sanitation lead by D. E. Hartley, S. H. Hopper and Frank Peabody.

The program was prepared under the direction of Dr. S. M. Morrison, Professor of Microbiology at C.S.U.

REPORT ON PESTICIDE RESIDUES IN FOODS¹

The report of a study conducted by the National Academy of Sciences-National Research Council on the question of pesticide residues in foods has been submitted to the Secretary of Agriculture and the Secretary of Health, Education, and Welfare.

The study dealt with the "no residue" and "zero tolerance" concepts as they relate to pesticide registration, the setting of residue tolerances, the enforcement provisions of the Food, Drug, and Cosmetic Act relating to residues in food, and the recommendations of the Federal and State agancies concerning pesticide uses. The study was conducted by the National Academy under contract with the two Departments. The two departments are currently reviewing recommendations of the report.

Eleven recommendations were made in the report. They are:

1. The concepts of "no residue" and "zero tolerance" as employed in the registration and regulation of pesticides are scientifically and administratively untenable and should be abandoned.

2. A pesticide should be registered on the basis of either "negligible residue" or "permissible residue", depending on whether its use results in the intake of a negligible or permissible fraction of the maximum acceptable daily intake as determined by appropriate safety studies.

3. Where the use of a pesticide may reasonably be expected to result in a residue in or on food, registration by the U. S. Department of Agriculture should not be granted unless (a) it is established that the residue is a negligible residue or (b) such residue is not more than a permissible residue established by FDA.

4. When a pesticide is registered on a negligibleresidue basis, the negligible-residue figure should be published, as well as an analytical method for determining whether or not a food contains a residue in excess of the negligible residue. Both the amount and the analytical method should have the concurrence of FDA and be controlling for its enforcement purposes.

5. FDA's regulations on permissible residues should include a published description of the analytical methods used for enforcement purposes and should not be changed without notice and opportunity for comment by interested parties.

6. If a pesticide is known to be too hazardous for a particular use, registration for such use should be refused.

7. Because of the importance that pesticides play in the production of our food supply and the many nonfood uses necessary for protecting the health and economy of the nation, it would seem appropriate that the registration of pesticides should continue to be the responsibility of the U. S. Department of Agriculture.

8. The publication of a reasonable schedule for an orderly transition from the present procedure is necessary, and its duration should be decided by mutual agreement between the Department of Agriculture and the Department of Health, Education, and Welfare.

9. Programs should be developed for continuing centralized leadership, free and prompt exchange of information, training activities, and interlaboratory evaluation. A manual of operating instructions for residue methods should be produced by the U. S. Department of Agriculture and the Department of Health, Education, and Welfare and continuously revised according to changing usage, food habits, and new pesticides and mixtures.

10. A formal program for education in residue analysis is urgently needed and the Departments of Agriculture and Health, Education, and Welfare, and any other agencies concerned should cooperatively sponsor this program with suitable training centers.

11. There should be an expanded research program on the persistence of pesticides in the total environment, and on the toxicology, pharmacology, and biochemistry of pesticides that would improve the reliability and precision of animal studies and their relevance to man.

Single copies of the report are available from the Office of Information, U. S. Department of Agriculture, Washington, D.C. 20250, and from the Press Office of the U. S. Department of Health, Education, and Welfare, Washington, D. C. 20201.

¹From FDA Report on Enforcement and Compliance, July, 1965.

MARYLAND UNIVERSITY SETS WINTER CONFERENCE SCHEDULE

Dates for the 1965-66 Dairy Technology Short Courses and Conferences at the University of Maryland at College Park have been announced. The meetings scheduled are as follows: 21st Annual Dairy Technology Conference, November 10, 1965; Ice Cream Short Course, January 24-February 3, 1966; Ice Cream Conference, February 3, 1966.

Details of the programs and other information will be made available later by Professor W. S. Arbuckle, Department of Dairy Science, University of Maryland.

PUBLIC HEALTH SERVICE ANNOUNCES TWO SHORT COURSES

The Public Health Service will present a 2-week course, Laboratory Analysis of Milk and Milk Products, November 8-19, 1965, at the Robert A. Taft Sanitary Engineering Center, Cincinnati, for professional personnel in responsible positions in State, municipal and other laboratories engaged in milk analysis and dairy products examination. In lectures, demonstrations, and laboratory sessions, the trainee learns to select and perform the laboratory tests used in measuring the quality of milk supplies and to interpret their results.

A one-week course, Institutional Sanitary Food Service, will be presented November 29-December 3, 1965, also at the Robert A. Taft Sanitary Engineering Center for supervisory sanitarians, dietitians and key administrators responsible for food service operations in institutions, with special emphasis on schools and hospitals. Lectures and workshop sessions will enable the trainee to understand and apply the fundamental principles of conducting an efficient institutional food sanitation program.

These courses will be conducted by personnel of the Division of Environmental Engineering and Food Protection. Detailed information about the course is given in the new Training Program Bulletin of Courses which is available on request. Applications for enrollment or requests for information should be addressed to the Director, Training Program, Robert A. Taft Sanitary Engineering Center, 4676 Columbia Parkway, Cincinnati, Ohio 45226. No tuition or registration fee is required.

DIVERSEY NAMES NEW DAIRY HEAD

The Diversey Corporation has announced the appointment of Maynard E. David as the firm's market manager for the dairy industry. David stepped into his new post here July 15, coming from Oklahoma City, Okla., where he had served as Diversey's territory representative since 1961.

David attended Centenary College, Shreveport, La., and has taken courses in dairy marketing and business management at Kansas State and Kansas Universities, respectively. Before joining Diversey four and a half years ago, he was co-owner and manager of Boyle's Dairy in Topeka, Kan.

The Diversey Corporation is a manufacturer of proprietary chemical detergents, including sanitation products and equipment for the food processing and food service industries, as well as cleaning and surface treatment compounds for the metal finishing industry.

TRAINING COURSE IN MILK TECHNOLOGY FOR SANITARIANS

The University of Massachusetts recently received a training grant from the United States Department of Health, Education, and Welfare to conduct a five day in-service training course in milk technology November 15 to 19, 1965. The University will conduct the course cooperatively with Region I of the Federal Agency in which the new Pasteurized Milk Ordinance will be presented and discussed together Dear IAMFES Member:

5 Minutes, Please

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For further information write to Dr. D. J. Hankinson, Department of Food Science and Technology, University of Massachusetts, Amherst, Mass. 01003.

CATALOG DESCRIBES KLEEN-KING PLANT WASH EQUIPMENT

A sixteen-page catalog available from Britt-Tech Corp. of Britt, Iowa, shows a complete line of highpressure wash equipment for food processing and bottling plant cleaning and maintenance.

Seven basic models of the Kleen-King line of "Liquid Brush" wash machines are described. Specific job applications such as cleaning plant interiors and cleaning and sanitizing equipment are outlined in the new catalog.

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NATIONAL SYMPOSIUM ON ANIMAL WASTE MANAGEMENT

A National Symposium on Animal Waste Management is scheduled for May 5-6, 1966, at the Kellogg Center for Continuing Education, Michigan State University, East Lansing, Michigan. The program will be interdisciplinary and the Committee with representatives from each co operating Society is now soliciting technical papers. Readers of Journal of Milk and Food Technology who have material or who know of material which should be presented at this Symposium are invited to forward by November 15, 1965, an abstract, in six copies and in 300 words or less to Dr. E. Paul Taiganides, Program Committee Chairman, Agricultural Engineering Department, The Ohio State University, Columbus, Ohio 43210, USA.

The subject matter of the papers to be submitted may relate to the engineering, agronomic, social, biological, economic, health, production, or any other aspects of the problem of farm waste handling, treatment and disposal. The Symposium is sponsored by the American Society of Agricultural Engineers, the Cooperative State Research Service, the North Central Region Technical Committee 69 on Farm Animal Waste Disposal, the Continuing Education Service of Michigan State University and in cooperation with the U.S. Public Health Service, American Society of Animal Science, Poultry Science Association, the American Society of Civil Engineers and the American Society for Microbiology.

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