SEPTEMBER, 1973
Vol. 36 P. 443-486 No. 9

Journal of Milk and Food Technology

Official Publication

INTERNATIONAL ASSOCIATION MILK, FOOD & ENVIRONMENTAL SANITARIANS

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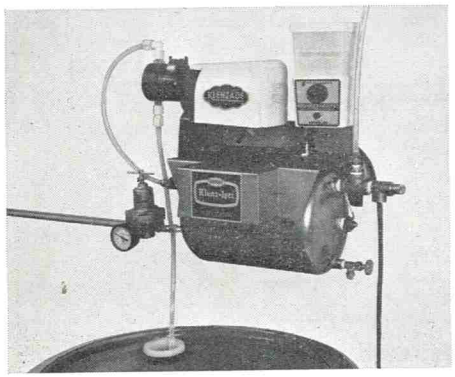

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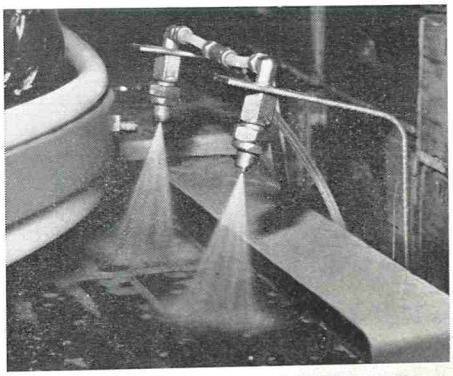

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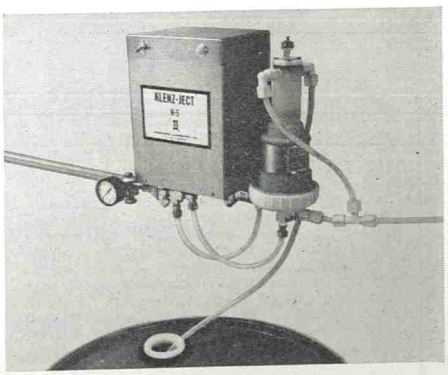

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COMPILED AND EDITED BY

J. C. FLAKE, A. E. PARKER, J. B. SMATHERS, A. K. SAUNDERS AND E. H. MARTH

PUBLISHED BY

INTERNATIONAL ASSOCIATION OF MILK, FOOD, AND ENVIRONMENTAL SANITARIANS, INC.

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International Association of Milk, Food, and Environmental Sanitarians, Inc.
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Prices: Single Copies, $1.00 each: 100 or more copies, 65 cents each.
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The Journal of Milk and Food Technology is issued monthly beginning with the January number. Each volume comprises 12 numbers. Published by the International Association of Milk, Food and Environmental Sanitarians, Inc. with executive offices of the Association Blue Ridge Rd., P. O. Box 437, Shelbyville, Ind.

2nd Class postage paid at Shelbyville, Indiana 46176.

EDITORIAL OFFICES: Dr. Elmer H. Marth Dept. of Food Science, University of Wisconsin, Madison, Wis. 53706. H. L. Thomasson Managing Editor, P. O. Box 437, Shelbyville, Indiana 46176.

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Journal of

MILK and FOOD TECHNOLOGY

INCLUDING MILK AND FOOD SANITATION

Official Publication

International Association of Milk, Food and Environmental Sanitarians, Inc.

Reg. U. S. Pat. Off.

Vol. 36 September, 1973 No. 9

Contributed Research Papers To Be Part Of Program For 1974 Annual Meeting Of IAMFES

Contributed research papers will be included in the program of the 1974 Annual Meeting of IAMFES, according to a recent decision of the Executive Board. The meeting is scheduled for August 12-15, 1974 at the St. Petersburg Hilton Hotel, St. Petersburg, Florida.

Research workers in governmental, industry, university, or other laboratories are invited to submit papers. Membership in IAMFES is NOT necessary for presenting papers at the annual meeting. Abstracts of papers to be presented at the annual meeting must be submitted by February 15, 1974. Forms for Abstracts and detailed instructions will appear in a subsequent issue of the *Journal of Milk and Food Technology.*

Papers should report results of applied research in such fields as:

- **food, dairy, and environmental sanitation and hygiene**
- **foodborne disease hazards (microbiological, chemical, etc.)**
- **food and dairy microbiology, including methodology**
- **food and dairy engineering**
- **food and dairy chemistry, including methods for food control**
- **food additives**
- **food and dairy technology**
- **food service and food administration**
- **food and dairy fermentations**
- **quality control in the food and dairy industries**
- **mastitis, causes and control**
- **environmental health**
- **waste disposal, pollution, water quality**

Three papers per hour will be scheduled. Hence authors will have 15 to 18 minutes for their presentations and several minutes will be allowed for discussion after the paper has been presented. Papers presented at the meeting should not have been published before they are given nor should they have been presented elsewhere.

MICROBIAL FLORA, CHEMICAL CHARACTERISTICS, AND SHELF LIFE OF FOUR SPECIES OF POND-REARED SHRIMP[1]

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(Received for publication April 5, 1973)

ABSTRACT

Fresh pond-reared shrimp stored on sterile ice developed serious off-odors (musty) and melanosis in 14-17 days. Initial microbial counts were 10^4-10^5/g. Counts were lower after 7 days of refrigerated storage, regained their initial level after 21-22 days, and then continued to increase. The initial microbial flora consisted of *Aeromonas, Pseudomonas,* and *Vibrio* species. Coryneform bacteria were predominant after 21 days, and *Acinetobacter* and *Pseudomonas* species after 28 days. Total volatile nitrogen (TVN) levels of freshly harvested pond shrimp were similar to those of fresh Gulf shrimp, amino-nitrogen (AA-N) levels of pond shrimp were about twice as high. During the first 14 days of storage, TVN levels increased and AA-N levels decreased. Values for TVN/AA-N of spoiled pond shrimp were lower than those of spoiled Gulf shrimp and were in the range of fresh white or brown Gulf shrimp. Amino acid analyses showed that during the first 14 days of iced storage increases occurred in lysine-ornithine, histidine, threonine, alanine, valine, methionine, isoleucine, leucine, tyrosine, and phenylalanine, with sharp decreases in proline and glycine.

Production and processing of shrimp is an important segment of United States and Gulf Coast fisheries. Commercial landings of shrimp in the United States in 1971 were 388 million lb. valued at dockside at $167 million *(10)*. Shrimp accounted for 26% of the total value of U. S. commercial fishery landings. Imports during 1971 amounted to 191 million lb. Nearly all shrimp harvested commercially are caught by fishing vessels on near-shore fishing grounds. In recent years, extensive efforts have been made to rear shrimp in natural and artificial ponds *(8)*. Vanderzant et al. *(12)* reported that the microbial flora of pond-reared brown shrimp (*Penaeus aztecus*) differed from Gulf Coast shrimp and consisted predominantly of coryneform bacteria and *Vibrio* species. Data on chemical characteristics of boat shrimp such as total volatile nitrogen, amino nitrogen, and amino acid distribution are available *(4, 5)*. Similar information on pond-reared shrimp is lacking. This paper reports on the microbial flora, chemical characteristics, and shelf life of four species of pond-reared shrimp stored on ice for 4-5 weeks.

[1]Published with the approval of the Director of the Texas Agricultural Experiment Station, College Station.

MATERIALS AND METHODS

Shrimp cultivation and sampling

The 0.5-acre artificial ponds used in this study are located on the West Galveston Bay shore in Brazoria County, Tex. Postlarvae (seed stock, mean length 10 mm) of *P. vanami, P. occidentalis,* and *P. setiferus* were supplied by the Ralston Purina Company hatchery in Crystal River, Florida; those of *P. aztecus* by the Dow Chemical Co. hatchery in Freeport, Tex. At the time of stocking the estimated number of postlarvae per pond were 50,000 for *P. vanami* and *P. occidentalis,* 20,000 for *P. setiferus,* and 15,000 for *P. aztecus.* Each species was raised in a separate pond. The ponds were stocked in June-July and the shrimp harvested in September 1972. Shrimp in the ponds were fed daily at a rate of 5% of their estimated total body weight. Shrimp were fed 25 and 35% protein rations prepared by Ralston Purina Co. Ponds were approximately 3-5 ft in depth. Detailed information on pond management procedures are presented in another report *(8)*.

Chemical and microbiological procedures

Shrimp were obtained from ponds with a seine and were deheaded aseptically using sterile rubber gloves. The tails were placed in a sterile nylon net bag which was placed in sterile crushed ice in a 40 × 30 × 30 cm insulated ice chest. A perforated plastic partition was placed in each chest, 4-5 cm from the bottom. Water was drained from the ice chests and sterile ice was added when needed. Samples for analysis were withdrawn with sterile forceps. Shrimp were examined by a trained three-member panel 2 to 3 times each week for appearance (melanosis, color, slime) and off-odors.

Total volatile nitrogen (TVN) and amino nitrogen (AA-N) were determined in the trichloroacetic acid extracts as described by Cobb et al. *(3)*. A fully automated Spinco Model 120 C amino acid analyzer was employed for amino acid analysis.

Aerobic plate counts of shrimp were determined with the spreadplate method by placing 0.1 ml of appropriate dilutions on Trypticase Soy agar (TSA, BBL) plates with 3% NaCl. Preparation of samples and dilutions was as described by Surkiewicz *(9)*. Duplicate plates were incubated at 25 C for 2 days. To determine microbial types, approximately 40 colonies were picked at random from countable plates. Diagnostic procedures and schemes for identification of the microbial flora were presented previously *(11)*.

TABLE 1. DISTRIBUTION OF MICROBIAL FLORA OF POND-REARED SHRIMP DURING REFRIGERATED STORAGE

Storage time (days)	Sample[a]	Percentage distribution									
		Acineto-bacter	*Aero-monas*	*Bacillus*	Coryne-form	*Entero-bacter*	*Flavo-bacterium*	*Micro-coccus*	*Morax-eila*	*Pseudo-monas*	*Vibrio*
0	A		65							30	5
	B		80							5	15
	C		30							60	10
	D		83							13	4
7	A		50							50	
14	A		40				5		15	35	5
21	A		15		65		5		15		
	B		5		95						
	C		30		45			15	10		
	D			10	60				25	5	
28	A	95			5						
	B	97				3					
	C							3		97	
	D	60					15		15	10	

[a]A = *P. vanami*, B = *P. occidentalis*, C = *P. aztecus*, D = *P. setiferus*

RESULTS AND DISCUSSION

Freshly harvested pond shrimp were judged of excellent quality with no detectable defects with respect to appearance or odor. After 7 days some deterioration in appearance (grey flesh, some reddening of pigment, slight melanosis) had occurred in all species except *P. setiferus*. This condition of the 3 species worsened after 14 days. Only a trace of melanosis was detectable on *P. setiferus*. Musty off-odors were detected in *P. vanami, P. occidentalis*, and *P. setiferus*. *P. aztecus* had a faint putrid off-odor. After 21 days of ice storage all species exhibited strong musty or musty-amine type off-odors, with dark grey flesh and diffuse greening under the shell. Samples were considered unacceptable in appearance and odor at 17-21 days of storage. Strong putrid off-odors were noticeable after 25 days when significant increases in bacterial count had occurred (Fig. 1).

Bacterial counts of freshly harvested pond shrimp ranged from 3×10^4 to 10^5/g (Fig. 1). With storage on ice, bacterial counts of the samples decreased sharply at 7 days, increased to their initial level after 21 to 22 days and continued to increase with counts at 28 days ranging from 2.5×10^6 to 3×10^7 per g.

The microbial flora of freshly harvested pond shrimp consisted of *Aeromonas, Pseudomonas*, and *Vibrio* species (Table 1). *Aeromonas* and *Pseudomonas* species continued to dominate the microbial flora of *P. vanami* after 7 and 14 days of refrigerated storage. A similar distribution of the microbial flora was noted for the other shrimp species at that time. Although minor variations in distribution of microbial types were noted between shrimp species, coryneform bacteria predominated after 21 days and *Acinetobacter* or *Pseudomonas* species after 28 days of refrigerated storage.

The initial bacterial counts of the freshly harvested shrimp in this study were slightly lower than those of *P. aztecus* reported previously *(12)*. *Aeromonas* or *Pseudomonas* species were the predominant isolates of the freshly harvested shrimp, whereas in a previous study *(12)* coryneform bacteria and to a lesser extent *Vibrio* species predominated in *P. aztecus*. Changes in ponds and characteristics of the water and shrimp stock may have caused these differences. In addition, type of feed, feeding rate, and fertilization of the ponds in 1972 differed from those employed in 1970 *(12)*. The reduction in count after 7 days of iced storage probably was caused by the washing effect of the melting ice and the inability of some

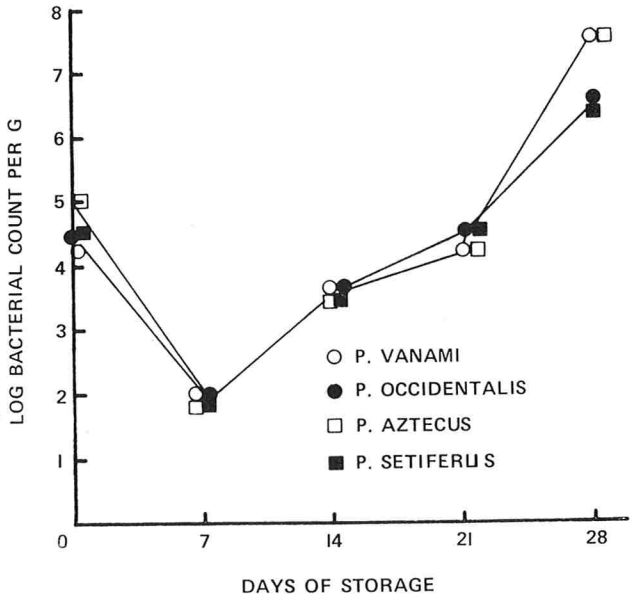

Figure 1. Agar plate counts of pond-reared shrimp stored in ice for 28 days.

microbial species to survive and/or grow at low temperatures. A similar change in microbial population of iced shrimp was reported by Campbell and Williams *(1)* and Carroll et al. *(2)*. Changes in the distribution of microbial flora during refrigerated storage may have been caused by differences in growth rate, symbiotic or antagonistic effects between species, or through reduction of the salinity of shrimp by melting ice. According to Carroll et al. *(2)* some microbial species may be unable to tolerate lowered salinity. Contact of shrimp with melting ice occurs during iced storage on commercial boats and is important in delaying deterioration of quality.

TVN levels of pond shrimp (Table 2) were similar or slightly higher than average values (13.36-15.73 mg N/100 g) for white or brown Gulf shrimp *(5)*. AA-N levels of pond shrimp were approximately twice those of Gulf shrimp (22.45-23.97 mM/100 g). During refrigerated storage, TVN levels increased during the first 14 days, leveled off, and then continued to decrease during the next 7 days. TVN levels of *P. vanami* and *P. occidentalis* increased again after 28-34 days when considerable increases in microbial population occurred. AA-N values for stored samples were lower than the initial values (0 days) particularly for *P. occidentalis*. TVN/AA-N ratios increased sharply during the first 7-14 days of storage, remained about the same for the next 11 days and increased again (*P. vanami* and *P. occidentalis*) after 28 and 34 days of storage. During the first 14 days, TVN/AA-N ratios increased at the rate of 0.017 (*P. vanami*), 0.013 (*P. occidentalis*), 0.017 (*P. aztecus*), and 0.028 (*P. setiferus*) mg N/mM/day. The value for *P. setiferus* is similar to an increase in TVN/

TABLE 2. TOTAL VOLATILE NITROGEN (TVN), AMINO NITROGEN (AA-N) AND TVN/AA-N RATIOS OF FOUR SPECIES OF SHRIMP DURING REFRIGERATED STORAGE

Iced storage (days)	*P. vanami*			*P. occidentalis*			*P. aztecus*			*P. setiferus*		
	TVN[a]	AA-N[b]	TVN/AA-N[c]	TVN	AA-N	TVN/AA-N	TVN	AA-N	TVN/AA-N	TVN	AA-N	TVN/AA-N
0	13.7	50.1	0.27	21.7	55.2	0.39	17.1	46.6	0.37	19.8	47.5	0.42
7	20.2	45.8	0.44	19.7	39.6	0.50	22.1	39.6	0.56	23.8	38.7	0.61
14	25.2	49.3	0.51	25.1	44.0	0.57	26.0	42.3	0.61	37.2	45.8	0.81
17	24.6	43.1	0.57	24.6	37.0	0.66	24.1	40.5	0.60	32.1	37.0	0.87
21	20.6	35.7	0.58	16.9	25.1	0.67	20.7	36.1	0.60	31.0	38.7	0.80
25	21.8	38.7	0.56	20.0	29.1	0.69	24.5	41.4	0.59	28.2	35.2	0.80
28	29.9	39.6	0.76	—	—	—	—	—	—	—	—	—
34	—	—	—	37.6	29.9	1.26	—	—	—	—	—	—

[a]mg N/100 g shrimp
[b]mM/100 g shrimp
[c]mgN/mM

TABLE 3. AMINO ACID ANALYSIS OF EXTRACTS FROM *P. vanami* DURING STORAGE ON STERILE ICE

Amino Acid	mM/100g shrimp						
	0[a]	7	14	17	21	25	28
Lysine-ornithine	0.36	0.58	1.14	1.03	1.09	1.06	1.26
Histidine	0.16	0.19	0.24	0.22	0.17	0.17	0.17
Ammonia	0.75	1.01	2.00	1.86	1.69	1.73	2.77
Arginine	3.74	2.97	3.33	2.97	3.05	2.61	2.14
Taurine	0.47	0.40	0.52	0.46	0.36	0.39	0.33
Aspartic acid	0.01	0.02	0.01	0.02	0.02	0.02	0.04
Threonine	0.16	0.26	0.53	0.46	0.42	0.45	0.42
Serine-glutamine-asparagine	1.36	1.75	1.65	1.44	1.06	1.45	0.66
Glutamic acid	0.36	0.40	0.50	0.43	0.38	0.30	0.42
Proline	8.09	7.20	3.91	5.86	4.00	5.54	5.46
Glycine	26.03	23.20	21.94	13.36	10.18	10.35	16.31
Alanine	4.03	3.60	6.81	5.63	5.36	5.08	4.86
Valine	0.26	0.49	0.87	0.81	0.72	0.83	0.64
Methionine	0.13	0.22	0.31	0.29	0.26	0.29	0.27
Isoleucine	0.21	0.29	0.45	0.43	0.36	0.42	0.36
Leucine	0.33	0.47	0.66	0.62	0.51	0.59	0.50
Tyrosine	0.11	0.17	0.24	0.22	0.22	0.20	0.21
Phenylalanine	0.08	0.18	0.27	0.25	0.03	0.25	0.26
TOTAL	46.64	43.40	45.38	36.36	29.88	31.73	37.08

[a]Days stored on sterile ice

AA-N of 0.032 mg N/mM/day reported for sterile (filtered) shrimp juice *(4)*. This suggests that TVN production during this period was probably caused by tissue enzyme activity. TVN/AA-N ratios for spoiled pond shrimp (after 17-21 days) were within the range of values (<0.9 mg N/mM) reported for freshly harvested Gulf shrimp *(5)* of excellent quality. This difference in TVN/AA-N ratios of spoiled samples probably resulted from high AA-N levels in the pond shrimp. Although all samples were unacceptable after 21 days, only the TVN value of *P. setiferus* exceeded 30 mg N/100g, which in the Australian and Japanese markets is used as indicative of spoilage *(7)*. TVN values of spoiled Gulf shrimp (initially handled aseptically and stored on sterile ice) usually were less than 30 mg N/100g *(5)*. With spoiled boat-shrimp (handled and stored under commercial conditions) TVN values usually exceeded 30 mg N. This difference in TVN level of spoiled boat shrimp and samples handled aseptically and stored on sterile ice may reflect a difference in type of microbial flora and activities.

The amino acid analyses for *P. vanami* are in Table 3. The analyses for the other species were similar. After 14 days increases were noted in the levels of lysine-ornithine, histidine, threonine, alanine, valine, methionine, isoleucine, leucine, tyrosine, and phenylalanine. The lysine-ornithine component increased sharply probably because of arginase activity *(4)*. Urea was present in the 14-day extracts ranging from 0.317 to 0.700 mM/100g shrimp. Increases in histidine, threonine, leucine, and isoleucine indicate that considerable proteolytic activity occurred. Sharp decreases in proline and glycine occurred during this period. This reduction is not caused by the washing action of the melting ice because taurine which is not metabolized *(4)* did not change. Reduction in the level of glycine could affect the desirable "sweet" flavor of freshly harvested shrimp and result in bitterness *(6)*. This defect has been reported for shrimp stored for long periods in sterile ice *(2)*. The significant changes in chemical characteristics examined in this study occurred during the first 14-17 days. After this period arginase activity and probably urease activity also decreased and hence NH_3 (TVN) production. This may have resulted from the activity of proteolytic enzymes. NH_3 production again was detected when microbial activity had increased sharply (at 28-34 days).

The high AA-N levels in pond shrimp may have resulted from an abundance of protein-rich feed. Deterioration of the appearance of pond shrimp, particularly melanosis, was somewhat faster than that of Gulf shrimp stored under similar condition. In a previous study *(4)* it was noticed that this problem occurred more rapidly in shrimp from waters with high organic matter. The cause and mechanism of the more rapid development of melanosis in shrimp from waters with high organic matter is not known.

ACKNOWLEDGMENT

This study was supported in part by the National Sea Grant Program NOAA, U.S. Department of Commerce through institutional grant No. 2-35213 made to Texas A&M University. The authors thank Mr. H. Holcomb for technical assistance in pond management and harvesting of samples.

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REFERENCES

1. Campbell, L. L. Jr., and O. B. Williams. 1952. The bacteriology of Gulf Coast shrimp. IV. Bacteriological, chemical, and organoleptic changes with ice storage. Food Technol. 6:125-126.

2. Carroll, B. J., G. B. Reese, and B. Q. Ward. 1968. Microbiological study of iced shrimp: Excerpts from the 1965 Iced-Shrimp Symposium. Circ. 284, U.S. Department of Interior, Washington, D.C. 17 pp.

3. Cobb, B. F., III, I. Alaniz, and C. A. Thompson, Jr. 1973. Biochemical and microbial studies on shrimp: Volatile nitrogen and amino nitrogen analysis. J. Food Sci. 38:431-436.

4. Cobb, B. F., III, and C. Vanderzant. 1971. Biochemical changes in shrimp inoculated with *Pseudomonas, Bacillus* and a coryneform bacterium. J. Milk Food Technol. 34:533-540.

5. Cobb, B. F., III, C. Vanderzant, C. A. Thompson, Jr., and C. S. Custer. 1973. Chemical characteristics, bacterial counts, and potential shelf life of shrimp from various locations on the northwestern Gulf of Mexico. J. Milk Food Technol. 36: (*In press*).

6. Hashimoto, Y. 1965. Taste-producing substances in marine products. pp. 57-61. *In* R. Kreuzer, (ed.) The technology of fish utilization, Fishing News (Books) Ltd., London, England.

7. Montgomery, W. A., G. S. Sidhu, and G. L. Vale. 1970. The Australian prawn industry. 1. Natural resources and quality aspects of whole cooked fresh prawns and frozen prawn meat. CSIRO Food Preservation Quart. 30:21-27.

8. Parker, J. C., and H. W. Holcomb. 1972. Comparative growth and production of *Penaeus aztecus, Penaeus setiferus,* and *Penaeus occidentalis* from experimental ponds in Brazoria County, Texas. Proc. 4th Annual Workshop, World Mariculture Soc., Jan. 1972.

9. Surkiewicz, B. F. 1966. Microbiological methods for examination of frozen and/or prepared foods. J. Ass. Off. Anal. Chem. 49:276-281.

10. United States Department of Commerce. 1972. Fisheries of the United States. 1971. NOAA, NMFS, CFS-5900, Wash., D.C. p. 30.

11. Vanderzant, C., and R. Nickelson. 1969. A microbiological examination of muscle tissue of beef, pork, and lamb carcasses. J. Milk Food Technol. 32:357-361.

12. Vanderzant, C., R. Nickelson, and P. W. Judkins. 1971. Microbial flora of pond-reared brown shrimp (*Penaeus aztecus*). Appl. Microbiol. 21:916-921.

MICROBIAL FLORA AND LEVEL OF *VIBRIO PARAHAEMOLYTICUS* OF OYSTERS *(CRASSOSTREA VIRGINICA)*, WATER AND SEDIMENT FROM GALVESTON BAY[a]

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(Received for publication April 25, 1973)

ABSTRACT

Aerobic plate counts at 25 C of freshly harvested oysters ranged from 2.3×10^4 to 3.0×10^7 and those of sediment samples from $<10^2$ to 3.0×10^6/g. Counts of water samples were nearly always $<10^2$/ml. *Vibrio, Aeromonas,* and *Moraxella* species predominated in the fresh oysters. *Vibrio parahaemolyticus* was isolated from 39 of 66 oyster samples and from 9 of 30 sediment and water samples. Isolation was most effective with prior enrichment of samples in trypticase soy broth with 7% NaCl and subsequent plating on thiosulfate citrate bile salts sucrose agar. *V. parahaemolyticus* was detected in only 1 of 8 refrigerated retail oyster samples. Aerobic plate counts at 25 C of refrigerated retail oysters were not much different from those of similar lots shucked under aseptic conditions in the laboratory (before shucking and washing in the plants). *Aeromonas* and *Moraxella* species were predominant in oysters at the retail level.

For many years isolations of *Vibrio parahaemolyticus* seemed to be limited to Japan where it is a major cause of gastroenteritis associated with consumption of seafoods particularly during summer months *(19)*. In recent years this organism has been isolated from shellfish and marine environments in the United States and many other countries *(2-4, 7-10, 16, 22, 32, 33)*. Although *V. parahaemolyticus* was the probable cause of several unconfirmed outbreaks of foodborne illness in the United States in 1969 *(25)*, the first confirmed outbreaks occurred in 1971 which involved consumption of crab *(14, 26)*. Other seafood-associated outbreaks of gastroenteritis caused by *V. parahaemolyticus* have been reported in 1972 *(27)*. Isolations of *V. parahaemolyticus* from oysters have been reported from widely different marine environments in the United States *(3, 4, 10, 22, 30)*. A seasonal incidence of this organism with a peak during the summer months is reported for oysters and/or water from Puget Sound *(3)*, Great Bay and Little Bay areas of New Hampshire *(4)*, and Chesapeake Bay *(8)*. A similar distribution was reported from Japan *(13)* and the Baltic Sea *(16)*.

Foodborne illness caused by *V. parahaemolyticus* in Japan usually is associated with consumption of raw seafoods. In the United States most seafoods are subjected to some heat treatment before consumption. Under these conditions outbreaks were caused by gross mishandling of the food. The oyster is the only mollusc that is generally eaten raw. Landings in the United States in 1971 were 54.6 million pounds valued at $30 million *(24)*. To evaluate the potential of oysters in foodborne illness, information is needed concerning the distribution of *V. parahaemolyticus* in oysters from sub-tropical waters such as the Northern Gulf of Mexico. Information about the effect of commercial handling practices on survival and/or growth of *V. parahaemolyticus* in oysters is scarce. With respect to the microbial flora of oysters, reports by Colwell and Liston *(5)*, Lovelace et al. *(11)*, and Murchelano and Brown *(15)* indicate that *Pseudomonas, Vibrio, Achromobacter, Alcaligenes,* and *Flavobacterium* species were predominant. The range for gram-positive forms was 15 to 20%. This report provides information about the level and type of microbial flora of oysters and the distribution of *V. parahaemolyticus* in oysters, water, and sediment.

MATERIALS AND METHODS

Samples

All samples were taken from various approved areas in Galveston Bay by personnel of the Texas State Department of Health at LaMarque or of the Marine Laboratory, Texas A&M University at Galveston, Tex. Oysters were collected in sterile plastic bags and transported to the laboratory in an insulated container at 5-10 C. Samples usually were examined within 6 to 12 hr after collection. Commercial oyster samples in the market surveys were obtained from two processing plants in the Texas Gulf Coast area. Preparation of oysters for microbiological examination was as described in *Recommended Procedures for the Examination of Sea Water and Shellfish (1)*.

[a]Published with the approval of the Director of the Texas Agricultural Experiment Station, College Station.

Microbiological procedures

The shucked oysters were blended for 2 min in a sterile Waring blendor. A 50-g portion then was blended for 1.5 min with 450 ml of sterile 0.1% trypticase peptone (BBL) with 3% NaCl. Bacterial counts were determined with the spread plate method by placing 0.1 ml of appropriate dilutions on trypticase soy agar (TSA, BBL) plates with 3% NaCl. Plates were incubated at 25 C for 2 days. To determine microbial types, approximately 40 colonies were picked at random from countable plates. Diagnostic procedures and schemes for identification of the microbial flora were presented in a previous report *(29)*.

Isolation of *V. parahaemolyticus* was accomplished by placing appropriate quantities of oyster-trypticase-peptone homogenate (equivalent to 1 to 10^{-4} g of oyster) in trypticase soy broth (TSB, BBL) with 7% NaCl. After 24 hr at 42 C, the tubes were streaked with a wire loop on thiosulfate citrate bile salts sucrose agar (TCBS, BBL) plates and also on a starch medium originally proposed by Twedt et al. *(23)* and later modified by Vanderzant and Nickelson *(30)*. The latter medium, designated MT, consisted of 2% trypticase peptone, 0.2% yeast extract, 1% corn starch, 7% NaCl, and 1.5% agar (pH 8.0). Isolation was also made without enrichment by spreading 0.1 ml of oyster-trypticase homogenate and appropriate dilutions directly on the surface of MT plates. Plates were incubated aerobically at 42 C for 24 to 48 hr. Blue-green colonies from TCBS medium and white to creamy, circular, smooth, amylase-positive colonies from MT medium were picked as suspect *V. parahaemolyticus*. The tests applied to these isolates and typical reactions for *V. parahaemolyticus* are as follows: Gram reaction (neg.), morphology (rods, exhibiting pleomorphism), cytochrome oxidase (+), catalase (+), starch hydrolysis (+), triple sugar iron agar (alkaline/acid, H_2S-, gas-), lysine decarboxylase (+), ornithine decarboxylase (+), lysine deaminase (−), NH_3 from arginine (−), growth in 1% trypticase broth with 0, 3, 7, 8, and 10% NaCl (−+++−), indole production (+), methyl red (+), Voges-Proskauer (−), motility (+), nitrate reduction (+), citrate utilization (+), urease (−), gelatin liquefaction (+), sensitive to pteridine 0/129 (+) and novobiocin (+), acid from sucrose (−), lactose (−), mannitol (+), arabinose (+), and cellobiose (∓). Hemolytic activity against fresh human erythrocytes (Kanagawa test) and serological identification were also employed. Methods are described in detail in the *Bacteriological Analytical Manual for Foods (28)* and by Vanderzant and Nickelson *(30)*.

RESULTS

Bacterial counts of freshly harvested oysters ranged from 2.3×10^4 to 3.0×10^7/g with an average (geometric) count of 4.0×10^5/g (Fig. 1). Bacterial counts of sediment samples ranged from $<10^2$ to 3.0×10^6/g, those of water samples were, except for one, always $<10^2$ per ml. The temperature of the waters from which the oysters were taken ranged from 12 C in November to 30 C in June, July, and September (Fig. 2). The salinity of the water ranged from 7 ppt in May to 26 ppt in April (Fig. 3).

In general, species of *Vibrio, Aeromonas,* and *Moraxella* were predominant in freshly harvested oysters (Table 1). These species constituted 20% or more of the microbial flora in 5 to 8 of the 12 monthly

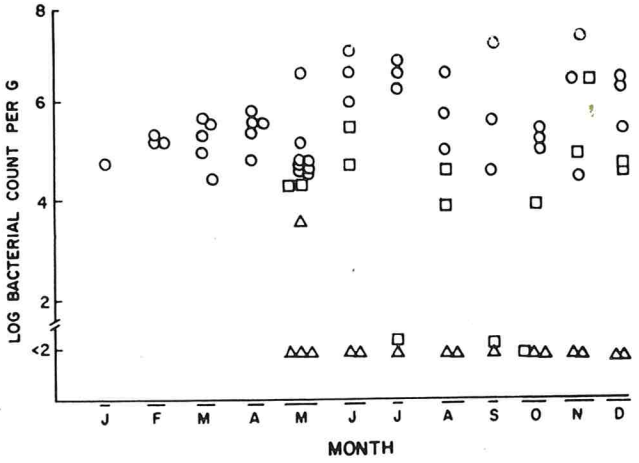

Figure 1. Aerobic plate count of freshly harvested oysters (open circles), water (open triangles), and sediment (open squares) samples.

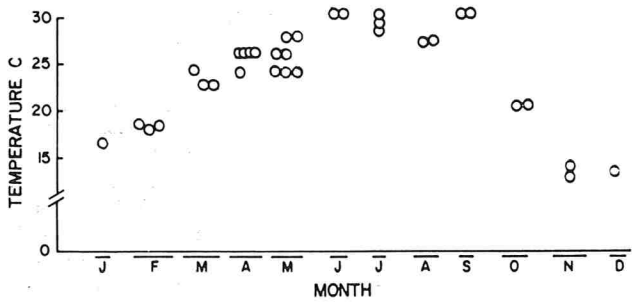

Figure 2. Temperature of waters from which oysters were taken.

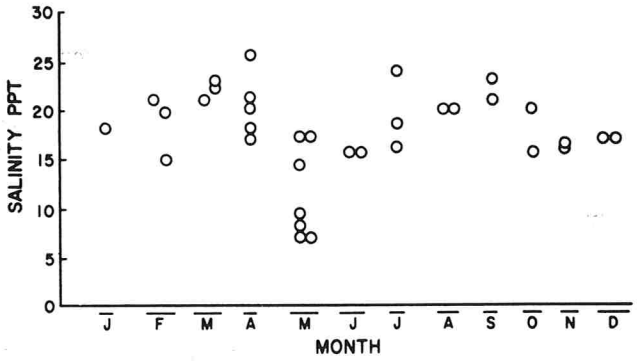

Figure 3. Salinity of waters from which oysters were taken.

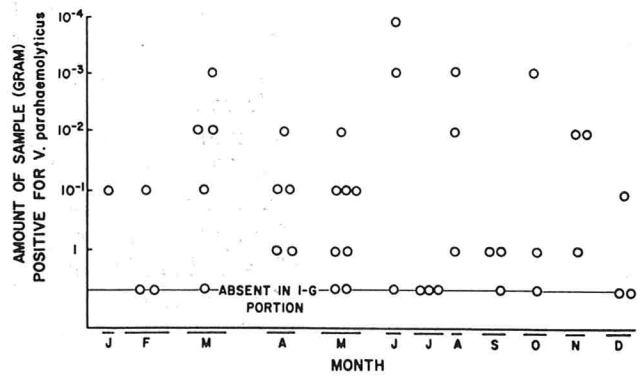

Figure 4. Level of *V. parahaemolyticus* in oysters from January through December 1972.

TABLE 1. DISTRIBUTION OF MICROBIAL FLORA OF 31 OYSTER SAMPLES

Type	% of total flora		Type present in no. of samples
	Range	Average	
Vibrio	0-80.8	22.3	23
Aeromonas	0-71.4	21.2	25
Moraxella	0-65.0	18.9	24
Pseudomonas	0-79.3	14.9	25
Acinetobacter	0-57.5	8.7	20
Coryneforms	0-63.3	7.1	13
Flavobacterium	0-30.0	1.5	5
Achromobacter	0-14.7	1.5	8
Bacillus	0-7.5	<1	3
Micrococcus	0-2.5	<1	1
Staphylococcus	0-2.5	<1	1
Enterobacteriaceae	0-32.5	3.2	8

TABLE 2. NUMBER OF SUSPECT AND CONFIRMED *V. parahaemolyticus* ISOLATES FROM OYSTERS, WATER, AND SEDIMENT SAMPLES[a]

Isolates	Oysters	Water & sediment
Suspect *V. parahaemolyticus*	278	41
No. and % of *V. parahaemolyticus*	80(29)	20(50)
No. and % of Kanagawa-pos. *V. parahaemolyticus*	16(20)	1(5)

[a]Includes isolates from MT medium with direct plating and from MT and TCBS media with enrichment in TSB (7% NaCl).

TABLE 3. NUMBER OF SAMPLES OF OYSTERS, WATER, AND SEDIMENT WITH SUSPECT AND CONFIRMED *V. parahaemolyticus* AS DETERMINED ON MT AND TCBS MEDIA

No. and type of sample	Method used		
	MT[a]	MT(ER)[b]	TCBS(ER)[c]
Oysters, suspect[d] 66	2	52	59
Oysters, confirmed 39	2	16	34
Water and sediment, suspect 16	1	11	13
Water and sediment, confirmed 9	0	5	7
Total suspect, 82	3	63	72
Total confirmed, 48	2	21	41

[a]MT = Sample plated directly on MT medium.

[b]MT(ER) = Sample plated on MT medium following enrichment in TSB (7% NaCl).

[c]TCBS(ER) = Sample plated on TCBS agar following enrichment in TSB (7% NaCl).

[d]Includes 43 samples of freshly harvested oysters shucked in the laboratory and 23 samples of commercially shucked oysters.

averages. Large variations between samples were noted for *Vibrio, Aeromonas, Moraxella, Pseudomonas, Acinetobacter,* coryneform bacteria, and *Flavobacterium. Bacillus, Micrococcus, Staphylococcus,* and *Enterobacteriaceae* were not often detected and only in low numbers.

V. parahaemolyticus was present (in 1 to 10^{-4}-g portions) in 30 of the 43 freshly harvested oysters. In only a few of the samples was *V. parahaemolyticus* present in 0.001-g portion (Fig. 4). *V. parahaemolyticus* was detected in 9 of the 30 water and sediment samples and only in 1- or 0.1-g portions.

A comparison of the frequency of isolation of *V. parahaemolyticus* with three different procedures showed that by direct plating on MT medium *V. parahaemolyticus* was confirmed in only 2 of the 43 oyster samples. With prior enrichment in TSB (7% NaCl) and subsequent plating on MT medium, *V. parahaemolyticus* was detected in 13 samples, with a similar enrichment but plated on TCBS agar in 25 samples. It should be recognized that by direct plating on MT medium, 0.1 ml of a 1:10 dilution of the sample was spread on the plates, which will not detect very low levels of *V. parahaemolyticus.* A comparison of the levels of *V. parahaemolyticus* in oysters as determined by plating enrichments (TSB, 7% NaCl) on MT and TCBS agar media showed higher levels on TCBS agar with 18 samples, higher levels on MT medium with 6 samples, and a similar level with 6 samples.

Many of the typical colonies, both on MT and TCBS agar media, which were picked for further identification were "false-positive." With enrichment in TSB, the percentage of false-positive isolates for TCBS agar (33%) was much smaller than on MT medium (72%). Many of those on MT medium were *V. alginolyticus.* The number of suspect *V. parahaemolyticus* isolates and the percentage confirmed are presented in Table 2. Only 29% of the suspected isolates could be confirmed by further testing. Twenty percent of the confirmed *V. parahaemolyticus* showed beta hemolysis with fresh human erythrocytes (Kanagawa reaction).

The number of samples with suspect and confirmed *V. parahaemolyticus* is given in Table 3. *V. parahaemolyticus* was confirmed in 39 of 66 oyster samples and in 9 of 30 water and sediment samples. Percentage of confirmation was greater (57 versus 33%) when isolates were picked from TCBS plates as compared with MT(ER) plates.

Eight market surveys were conducted between May to December 1972 in which oysters were sampled at various stages during processing and marketing (Table 4). Aerobic plate counts of oysters in the shell (shucked in the laboratory) ranged from 1.5×10^5 to 3.0×10^7/g with an average (geometric) count of 3.9×10^6/g. Bacterial counts of identical lots at the retail level were similar or slightly lower than before processing. *V. parahaemolyticus* was present at low levels (0.01- to 1-g portion) in 5 of the 8 freshly harvested samples but none of these samples contained *V. parahaemolyticus* (in 1-g portion) at

TABLE 4. AEROBIC PLATE COUNT AND LEVEL OF *V. parahaemolyticus* IN OYSTERS AT VARIOUS STAGES DURING PROCESSING AND MARKETING IN 8 MARKET SURVEYS

Sample	APC per g[a]	Portion of sample (g) positive for *V. parahaemolyticus* [TCBS(ER)][b]							
In shell (shucked in lab.)	1.5×10^5-3.0×10^7 (Avg. 3.9×10^6)	1	10^{-2}	–[c]	10^{-2}	–	–	1	10^{-1}
After shucking	3.0×10^5-6.5×10^6 (Avg. 1.3×10^6)	–	–	1	–	–	–	10^{-1}	1
After shucking and washing	6.7×10^4-3.0×10^6 (Avg. 5.0×10^5)	–	1	10^{-3}	1	–	1	–	1
At retail level	8.2×10^4-8.7×10^6 (Avg. 6.0×10^5)	–	–	10^{-3}	NT[d]	–	–	–	–

[a]APC = Aerobic plate count
[b]TCBS(ER) = Level of *V. parahaemolyticus* by plating on TCBS medium following enrichment in TSB with 7% NaCl.
[c]1-g portion negative for *V. parahaemolyticus*.
[d]NT = Not tested.

TABLE 5. DISTRIBUTION OF MICROBIAL FLORA OF OYSTERS BEFORE SHUCKING AND AT RETAIL LEVEL IN 8 MARKET SURVEYS

Microbial Type	Before shucking[a]			At retail level		
	% of total flora		Type present in no. of samples[b]	% of total flora		Type present in no. of samples[c]
	Range	Average		Range	Average	
Vibrio	0-25.0	5.8	3	0-60.0	15.4	3
Aeromonas	0-71.4	37.3	5	0-44.4	18.9	6
Moraxella	3.6-41.4	26.5	6	2.5-37.5	19.8	7
Pseudomonas	0-46.7	11.7	4	0-12.5	6.1	5
Acinetobacter	0-13.3	5.1	3	0-60.0	12.1	4
Coryneforms	0-44.8	7.9	2	0-85.0	13.2	2
Flavobacterium	0-7.5	2.3	3	0-52.9	8.3	2
Achromobacter	0	0	0	0-7.5	1.6	2
Bacillus	0-6.9	1	1	0	0	0
Micrococcus	0-2.5	<1	1	0	0	0
Staphylococcus	0	0	0	0-2.5	<1	1
Enterobacteriaceae	0-5.0	2	2	0-18.5	4.5	3

[a]Shucked under aseptic conditions in laboratory.
[b]Based on 6 samples.
[c]Based on 7 samples.

the retail level. In one oyster sample *V. parahaemolyticus* was present in 0.001-g portion after shucking and washing and at the retail level, and not before shucking and washing. The cause of this is not known. A comparison of the microbial flora of oysters shucked in the laboratory (market survey samples) and shucked, washed oysters in cans at the retail level (Table 5) indicates that species of *Aeromonas* and *Moraxella* predominated.

DISCUSSION

In this study, *V. parahaemolyticus* was present in 70% of the freshly harvested oysters and in 30% of the water and sediment samples. Although the highest level of *V. parahaemolyticus* was recorded in June, no definite seasonal incidence was apparent. This is probably because of the relatively high water temperature in Galveston Bay as compared with Puget Sound, Chesapeake Bay, and estuaries of New Hampshire where numbers are greater in summer months and decrease sharply when water temperatures drop below 15 C *(3, 4, 8)*.

In general, *V. parahaemolyticus* was detected more frequently and at somewhat higher levels by plating on TCBS agar following enrichment of samples in TSB with 7% NaCl. In addition, fewer false-positive isolates were picked from TCBS than from MT medium. The percentage of oyster samples positive for *V. parahaemolyticus* and the level of this organism were somewhat lower than reported by Baross and Liston *(3)*. This probably resulted from a difference in classification of *V. parahaemolyticus*. In their study, hemolytic vibrios meeting the general classification of *V. parahaemolyticus* without regard to sucrose fermentation were reported as *V. parahaemolyticus*. In the present study only isolates with all basic characteristics of *V. parahaemolyticus* including lack of sucrose fermentation, identical to Sakazaki's biotype 1 *(20)*, were included as confirmed *V. parahaemolyticus*. If samples with sucrose-fermenting strains of "*V. parahaemolyticus*" had been included, 94% of the freshly harvested samples would have contained this organism. Japanse workers *(12, 21)* reported that 88-96% of *V. parahaemolyticus* cultures isolated from patients with gastroenteritis are beta-hemolytic against fresh human erythrocytes (Kanagawa-positive), but only 0.5-1% of the isolates from raw fish. In their opinion, human pathogenicity is closely related to this specific hemolytic activity. Administration of non-hemolytic strains to human volunteers confirmed their lack of pathogenicity. In our

study, 20% of fresh isolates of *V. parahaemolyticus* from oysters were Kanagawa-positive. Some loss of hemolytic activity occurred when these isolates were maintained on laboratory media for 6 to 12 months. Pathogenicity of these strains to humans has not yet been established. A majority (79%) of *V. parahaemolyticus* isolates from oysters, water, and sediment were serologically nontypable.

Reduction in the level of *V. parahaemolyticus* after processing at the retail level was probably caused by a dilution effect of water during washing after shucking and the sensivity of this organism to refrigeration *(6, 31)*. In addition, washing may have reduced the salinity of the surface area of the oyster. *V. parahaemolyticus* is unstable without salt and is destroyed readily under these conditions *(6)*. No relation could be established between level of *V. parahaemolyticus* and aerobic plate count.

Oysters harvested from approved waters usually are of acceptable quality at time of harvesting with areobic plate counts (APC) at 35 C on Standard Methods agar (SMA) ranging from a few hundred to a few thousand per gram. High aerobic plate counts of shucked oysters at the wholesale level reflect microbial growth in the interval between harvesting and shucking, and improper handling or inadequate refrigeration during processing and shipping *(17, 18)*. To be acceptable at this point the APC at 35 C must not exceed 5×10^5/g with a fecal coliform density (MPN) of not more than 230/100 g. The relatively high bacterial counts of freshly harvested oysters in this study most likely reflect certain changes in the agar plate method such as *(a)* plate incubation at 25 C, *(b)* use of TSA as compared with SMA plating medium, and *(c)* inclusion of NaCl in the plating medium, which allowed a greater number of bacteria to develop. A comparison of bacterial counts of a limited number of samples on TSA (3% NaCl, at 25 C) and SMA (no added NaCl, at 35 C) showed counts on TSA 1.6 to 3.7 logs higher. The average (geometric) count on TSA was 4×10^5, that on SMA 10^3 per g. Higher counts on TSA also may have resulted from improved recovery of sublethally injured cells. Colwell and Liston *(5)* also reported higher counts with plate incubation at 25 than at 37 C.

Which of the bacterial counts, at 35 C on SMA without salt or at 25 C on TSA with salt provides more useful information is a controversial point. High viable counts at 35-37 C in seafoods usually indicate unsanitary handling. High viable counts at 20-25 C reflect increases in count of psychrotrophic and some mesophilic species initially present on the freshly harvested seafood or acquired by contact with contaminated equipment or surfaces in the plant. These species are frequently responsible for quality deterioration during refrigerated storage. For some of these species a temperature of 35-37 C may constitute the upper limit for growth on laboratory media. In addition, some species of the natural microbial flora of shellfish exhibit a partial or complete salt dependence as demonstrated by a growth stimulating effect of NaCl added to standard media *(5)*.

The bacterial counts of freshly harvested oysters from Galveston Bay were somewhat higher (geometric mean 4×10^5/g) than counts of oyster gill tissue (10^3-10^4/g) or mantle fluid (10^4/ml) from Chesapeake Bay oysters *(5)*. This difference in count may have been caused by differences in marine environment or enumeration techniques. Bacterial counts of oysters in excess of 10^6/g were recorded more often from May to September probably because of higher air and water temperatures. The slightly lower counts of oysters at the retail level as compared with the shellstock probably reflects the effacts of sanitary handling, washing, and adequate refrigeration.

The microbial flora of freshly harvested oysters from Galveston Bay was dominated by gram-negative rods (*Vibrio, Aeromonas, Moraxella, Pseudomonas, Acinetobacter*) and was similar to that of Pacific oysters (*Crassostrea gigas*) from Washington State *(5)* and oysters (*Crassostrea virginica*) from Long Island Sound and Chesapeake Bay *(11, 15)*. In addition to *V. parahaemolyticus, V. alginolyticus* and *V. anguillarum* were isolated from oysters. Among *Vibrio, V. alginolyticus* and *V. parahaemolyticus* usually were predominant throughout the year. *V. alginolyticus* predominated in August and September. Baross and Liston *(3)* reported that *V. alginolyticus* was abundant in the summer only which suggests that the temperature of the water is a critical factor.

REFERENCES

1. American Public Health Association. 1970. Recommended procedures for the examination of sea water and shellfish, 4th ed. American Public Health Association, New York.

2. Baross, J., and J. Liston. 1968. Isolation of *Vibrio parahaemolyticus* from the Northwest Pacific. Nature 217: 1263-1264.

3. Baross, J., and J. Liston. 1970. Occurrence of *Vibrio parahaemolyticus* and related hemolytic vibrios in marine environments of Washington State. Appl. Microbiol. 20:179-186.

4. Bartley, C. H., and L. W. Slanetz. 1971. Occurrence of *Vibrio parahaemolyticus* in estuarine waters and oysters of New Hampshire. Appl. Microbiol. 21:965-966.

5. Colwell, R. R., and J. Liston. 1960. Microbiology of shellfish. Bacteriological study of the natural flora of Pacific oysters (*Crassostrea gigas*). Appl. Microbiol. 8:104-109.

6. Covert, D., and M. Woodburn. 1972. Relationships of temperature and sodium chloride concentration to the survival of *Vibrio parahaemolyticus* in broth and fish homogenate. Appl. Microbiol. 23:321-325.

7. Fishbein, M., I. J. Mehlman, and J. Pitcher. 1970. Isolation of *Vibrio parahaemolyticus* from the processed meat of Chesapeake Bay blue crabs. Appl. Microbiol. 20:176-178.

8. Kaneko, T., and R. R. Colwell. 1973. Ecology of *Vibrio parahaemolyticus* in Chesapeake Bay. J. Bacteriol. 113: 24-32.

9. Krantz, G. E., R. R. Colwell, and T. E. Lovelace. 1969. *Vibrio parahaemolyticus* from the blue crab *Callinectus sapidus* in Chesapeake Bay. Science 164:1286-1287.

10. Landry, W. L., B. Wentz, and M. Fishbein. 1970. Recovery of *Vibrio parahaemolyticus* during a market survey of Gulf Coast seafoods in the Dallas area. Paper presented at the Fall Meeting of the Texas Branch ASM. Dallas.

11. Lovelace, T. E., H. Tubiash, and R. R. Colwell. 1968. Quantitative and qualitative commensal bacterial flora of *Crassostrea virginica* in Chesapeake Bay. Proc. Natl. Shellfish Assoc. 58:82-87.

12. Miyamoto, Y., T. Kato, Y. Obara, S. Akiyama, K. Takizawa, and S. Yamai. 1969. In vitro hemolytic characteristic of *Vibrio parahaemolyticus*: Its close correlation with human pathogenicity. J. Bacteriol. 100:1147-1149.

13. Miyamoto, Y., K. Nakamura, and K. Takizawa. 1962. Seasonal distribution of *Oceanomonas* spp., halophilic bacteria, in the coastal sea. Its significance in epidemiology and marine industry. Jap. J. Microbiol. 6:141-158.

14. Molenda, J. R., W. G. Johnson, M. Fishbein, B. Wentz, I. J. Mehlman, and T. A. Dadisman, Jr. 1972. *Vibrio parahaemolyticus* gastroenteritis in Maryland: Laboratory aspects. Appl. Microbiol. 24:444-448.

15. Murchelano, R. A., and C. Brown. 1968. Bacteriological study of the natural flora of the Eastern oyster, *Crassostrea virginica*. J. Invertebrate Pathol. 11:519-520.

16. Nakanishi, H., L. Leistner, H. Hechelmann, and J. Baumgart. 1968. Weitere Untersuchungen über das Vorkommen von *Vibrio parahaemolyticus* und *Vibrio alginolyticus* bei Seefischen in Deutschland. Archiv. Lebensmittelhygiene 19:49-53.

17. Presnell, M. W. 1970. Cooperative bacteriological study of commercial practices of oyster harvesting and processing in Alabama. U. S. Dept. of Health, Education, and Welfare, Special report, GCWHL 71-1.

18. Presnell, M. W., and C. B. Kelly. 1961. Bacteriological studies of commercial shellfish operations on the Gulf Coast. U. S. Dept. of Health, Education and Welfare, Techn. Report F 61-9.

19. Sakazaki, R. 1969. Halophilic *Vibrio* infections. In H. Riemann (ed.) Foodborne infections and intoxications. Academic Press, New York.

20. Sakazaki, R., S. Iwanami, and H. Fukumi. 1963. Studies on the enteropathogenic, facultatively halophilic bacteria, *Vibrio parahaemolyticus*. I. Morphological, cultural, and biochemical properties and its taxonomical position. Jap. J. Med. Sci. Biol. 16:161-188.

21. Sakazaki, R., K. Tamura, T. Kato, Y. Obara, S. Yamai, and K. Hobo. 1968. Studies on the enteropathogenic, facultatively halophilic, bacteria, *Vibrio parahaemolyticus*. III. Enteropathogenicity. Jap. J. Med. Sci. Biol. 21:325-331.

22. Thomson, W. K., and D. A. Trenholm. 1971. The isolation of *Vibrio parahaemolyticus* and related halophilic bacteria from Canadian Atlantic shellfish. Can. J. Microbiol. 17:545-549.

23. Twedt, R. M., and R. M. Novelli. 1971. Modified selective and differential isolation medium for *Vibrio parahaemolyticus*. Appl. Microbiol. 22:593-599.

24. U. S. Department of Commerce. 1972. Fisheries of the United States 1971. NOAA, NMFS, CFS-5900, Washington, D. C.

25. U. S. Department Health, Education, and Welfare. 1969. Foodborne outbreaks. Annual Summary. Center for Disease Control. Atlanta, Georgia.

26. U. S. Department of Health, Education and Welfare. 1971. *Vibrio parahaemolyticus*-Gastroenteritis-Maryland. Morbidity and Mortality 20:356.

27. U. S. Department of Health, Education, and Welfare. 1972. *Vibrio parahaemolyticus*. Morbidity and Mortality 21: 245, 341, 430.

28. U. S. Department of Health, Education, and Welfare. 1972. Bacteriological analytical manual for foods. Washington, D. C.

29. Vanderzant, C., and R. Nickelson. 1969. A microbiological examination of muscle tissue of beef, pork, and lamb carcasses. J. Milk Food Technol. 32:357-361.

30. Vanderzant, C., and R. Nickelson. 1972. Procedure for isolation and enumeration of *Vibrio parahaemolyticus*. Appl. Microbiol. 23:26-33.

31. Vanderzant, C., and R. Nickelson. 1972. Survival of *Vibrio parahaemolyticus* in shrimp tissue under various environmental conditions. Appl. Microbiol. 23:34-37.

32. Vanderzant, C., R. Nickelson, and J. C. Parker. 1970. Isolation of *Vibrio parahaemolyticus* from Gulf Coast shrimp. J. Milk Food Technol. 33:161-162.

33. Ward, B. Q. 1968. Isolations of organisms related to *Vibrio parahaemolyticus* from American estuarine sediments. Appl. Microbiol. 16:543-546.

J. Milk Food Technol., Vol. 36, No. 9 (1973)

NEW WASTE WATER PRACTICES IN DAIRY PLANTS

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(Received for publication June 18, 1973)

ABSTRACT

Water pollution legislation is now having a national impact on pollution control activities in the Dairy Industry. The legislative programs that are being enforced are establishing uniform standards throughout the 50 states to abate pollution. It is apparent that individual plants will be forced to evaluate their present operation in terms of reducing waste water. The waste water from dairy plants presents certain characteristics which make treatment difficult. The wastes are produced over a short period during the day and this presents peak loads to a treatment facility. The strength of waste water will also vary. Rather than the dairy plant constructing its own treatment facility, the predominant means of disposal is to discharge to a municipal sewer system. For this method of treatment, certain charges are made against the dairy plant to pay for operation of the facility. These charges are becoming substantial, dollarwise, because of the high strengths and volumes of waste water being treated daily. By collecting product/water rinsings from the HTST and CIP system waste water loads can be reduced. Further equipment augmentation to existing CIP systems results in additional reduction in daily water usage.

We must now acknowledge that the recent emphasis on ecology has made its impact on the Dairy Industry. New regulations and controls for fluid and solids wastes, noise pollution, and air pollution are the principal areas of concern to the processor. This article discusses the fluid waste water segment of the problem.

Water has always played an important part in operation of a milk plant. Before the days of automation and central cleaning systems, daily take-down of gasketed pipelines and fittings, hand washing of storage tank and processing vats, and manually cleaning of 10-gal milk cans were the accepted practices of good housekeeping sanitation. From this cleaning procedure, dairy plants would send large volumes of water that contained milk fat, solids, washing compounds, and sanitizers to the drain as waste water. Continued reassembly of pipelines and fittings often resulted in leaks in the system. Not only was this a source of contamination, but product leaking from manual valves, gaskets, or pumps, eventually ended up being flushed to the drain.

CLEANING-IN-PLACE

Some 15 years ago cleaning-in-place (CIP) was introduced to the dairy industry. Along with this new technique for cleaning were related items that would help make this new procedure a success. Air-operated valves replaced out-dated manual valves, all welded pipeline systems replaced the gasketed take-down lines, and permanent spray devices were installed in tanks, thus eliminating the need for manual cleaning.

With a CIP system, a tank or line circuit could be pre-rinsed, washed, post-rinsed, and sanitized as part of a daily cleaning procedure. The important time, temperature, and pH relationships were maintained by installing a CIP "Programmer." The plant was cleaned on a daily basis by a CIP unit that consisted of a 50-gal tank. The small size of this unit allowed it to be installed at a low capital investment and minimal floor space. With this system, a loop was developed between the equipment being cleaned and the CIP unit. The end result was that all rinses and chemicals were sent to the drain and ended up as part of the total plant waste water. This system was then termed a "Throw-a-way" type (Fig. 1).

As CIP operation became more prevalent throughout the industry, so did the degree of automation in new plant design. During the 1960's new developments on the West Coast for milk processing plants emerged which found plants installing minimal equipment which was of large volume capacity in design. These plants were limited product, efficient dairy processing centers, and were termed "high-efficiency" plants. Along with their production and efficiency came a concern for the CIP operation and water usage. For these plants a two-tank CIP system was installed that included one tank for rinse and one for wash. Like the early CIP units, the tanks were supplied with proper valves, steam controls, level controls, and chemical pumps. The only items sent to the drain in this system were the pre-rinse and post-rinse waters. Because the wash solution was returned to a tank and its strength monitored, this procedure was termed a "re--use" system (Fig. 2). Both systems were used throughout the nation as the accepted methods for cleaning-in-place circulation systems.

ENVIRONMENTAL CONCERNS

During the early 1970's the subject of national environmental control became an intense target for

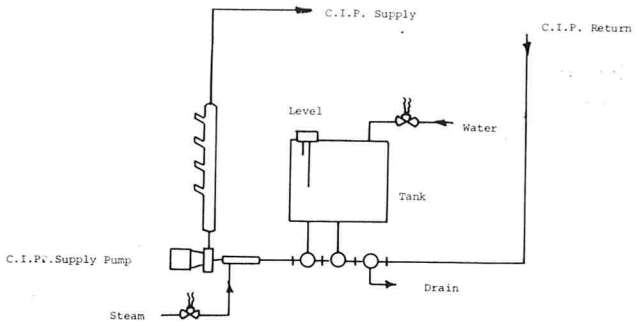

Figure 1. "Throw-away" type of CIP system.

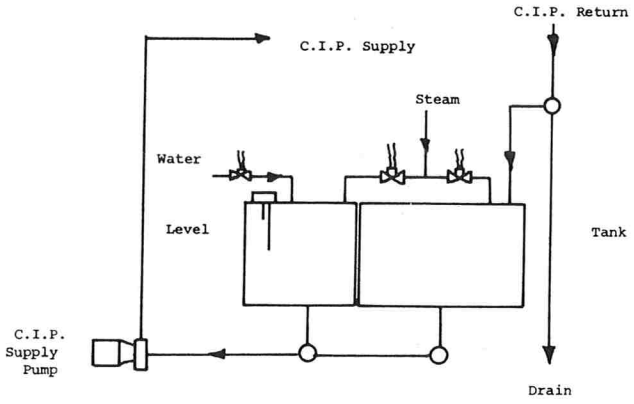

Figure 2. "Re-use" type of CIP system.

public criticism. One outgrowth of this criticism was a renewal in enforcement of old existing laws such as the *Rivers and Harbors Act of 1899* that required a plant to apply for permission to discharge water into navigable streams and tributaries. Thus, plants had to either acquire a permit or find new areas for dumping their waste water. For many plants, the municipal sewage system seemed to be the logical answer. Since most plants have locations in urban areas, about 87% are now dumping waster into municipal sewage systems. Unfortunately, municipal sewage systems have been an expensive answer to this problem. Depending on plant locality and local conditions, the dairy plant is, in many instances, the largest contributor of waste water to the community sewage system. Revision of monthly surcharges to cope with the increased BOD content and waste water volume emitted by these plants, required them to tighten their belts and review all areas that contributed to the waste water problem.

Plant Operations That Yield Waste Water

High-temperature-short-time operation

The first area of concern for reducing waste water loads is operation of the HTST system. This system in today's modern plant could include a plate heat exchanger, homogenizer, timing pump, automatic desludging separator/clarifier, vacuum treatment unit, and constant level tank. The capacity of these components can range from 100 to 150 gal/min. It is generally recognized that the system is started up and shut down using water in lieu of product. Water can also be used when changing products as

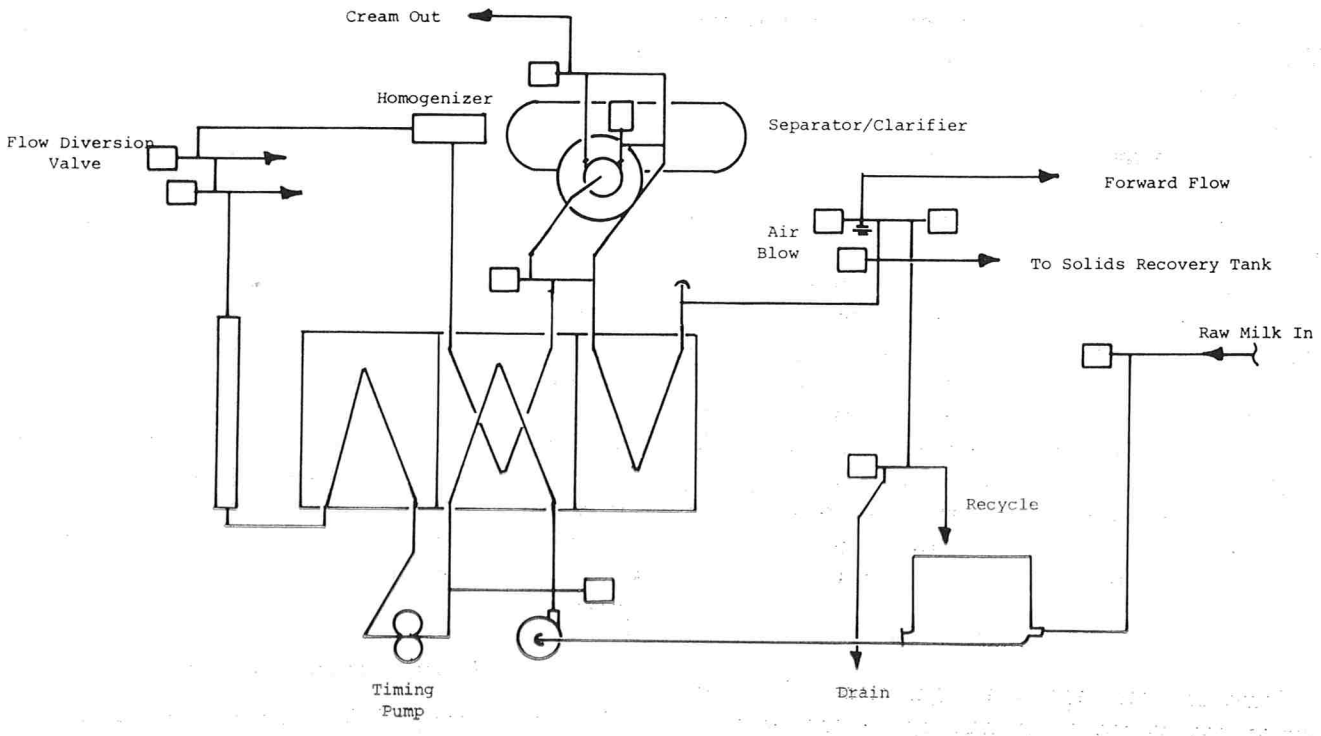

Figure 3. High-temperature-short-time solids recovery system.

often occurs during the day's operation. Any change in mode of operating the HTST will result in an intermixing of product and water. The intermixed product must be disposed of because it is not a legal milk product. To keep the product from reaching the drain and raising the BOD level, many plants are installing a waste collecting tank (Fig. 3). Through the use of air operated valves, level controls, and a timed flow through the system, an accurate reliable method of BOD reduction may be obtained during operation of the HTST. The collected product, depending on the locality, may be used elsewhere in the plant or sold as animal feed. This type of waste collection equipment may also be considered for CIP cleaning of the HTST system to optimize savings of water and cleaning solution used for CIP purposes. The equipment can be controlled by a CIP programmer to insure day-to-day dependability.

Central cleaning system area

In the previous discussion concerning CIP systems, both systems were sending rinse water to the drain. In CIP cleaning, the first rinse is critical as this will remove any milk fat that adheres to the side of a vessel or remains in a pipeline. It is for this reason that plants are installing a recovery tank for this rinsing. As is similar in the HTST system, a tank is provided to store this product that is a combination of water and milk fat. Further efficiency in the system serves to recover the wash water and maintain it at the proper temperature and concentration. The final rinse water is also recovered and stored in the pre-rinse tank where it is used on the next piece of equipment before being discharged to the drain.

Thus, through proper system engineering, the plant CIP system can be a very efficient operation. Whereas the automated circulation cleaning unit has been a large contributor to waste water loads, its water, steam, and chemical consumption can be materially reduced.

Filling area

Another area of the plant that contributes to waste water loads is the high speed filling and packaging area. Drainage in this area contains product from damaged cartons, broken cases, rinsings from machines, and lubricants from casers, stackers, and conveyor chains. A plant recovery program should be established to dispose of damaged or broken cartons and the product in the cartons. A review should be made of the system that supplies the conveyor with lubricants. It is important to use conveyor lubricants at low concentration, particularly since they do contain about 25% hexane solubles.

Separator clarifier area

Many plants have installed either automatic separators or clarifiers in the operation. In either event the machines are set to desludge automatically every 15 to 30 min depending on the operation. The resulting sludge should be collected and not allowed to reach the plant drains.

Hose stations

All plants should have positive shut-offs installed on hoses to prevent excess amounts of water from going to the drain when the hose is not in use.

Conclusion

In conclusion, the waste water problem is an individual plant problem as well as an industry problem. Finalized enforcement of legislation will leave plants with no choice but to conserve water. By improving on the foundations that are already established, solutions to waste problems for many plants may already exist. For others, a plant waste treatment facility may be the answer. Such a facility is not inexpensive in design or operation. The Dairy Industry traditionally has been generally good in water conservation practices but increasing costs are forcing the industry to re-evaluate old traditional methods of operating.

ADAPTABILITY OF POTATO DRYING TO YAM PROCESSING[1]

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(Received for publication March 2, 1973)

Abstract

One of the problems related to the food supply in Nigeria and other West African countries at present is that of efficient preservation and processing of raw products. In this brief review of potato processing techniques, properties of West African yams are compared with those of potato with a view to explore possibilities of adapting potato processing techniques to yams. Yams referred to in this article are of the family *Dioscoreaceae,* and they are different from the yams known in the U.S.A. The American yams are of the family *Convovulaceae* and are a variety of sweet potatoes. Possible new foods from West African yams, similar to convenience foods made from potato are also proposed. Of the varieties of yam grown in Nigeria, *Dioscorea alata* (water yam) is compositionally closest to potato. Properties of other varieties also are comparable to those of potato. Because there are similarities between the two tuberous crops, various methods for dehydrating potato could be adapted to drying of the West African yam.

Introduction and Background Information

The objective of this review of potato dehydration is to provide a basis for developing techniques to dry varieties of West African yams which have characteristics similar to the potato.

Yams are tuberous crops indigenous to the humid, tropical, southern parts of West Africa where they are used as an important source of carbohydrate. They are the most important root crop produced in Nigeria. West African yams are different from the yams known in the United States of America. American "yams" are a variety of sweet potatoes. American varieties of sweet potatoes are classified into two main groups: *(a)* varieties grown primarily for feed and industrial uses and *(b)* varieties grown primarily for food. The food types are further divided into two subgroups: varieties with soft or moist flesh when cooked, and varieties with firm or dry flesh when cooked *(5, 7).* In the United States, growers, shippers, and buyers of the moist-fleshed types frequently use the term "yam" *(5).* In fact the term "yam" as it is used in the United States is a misnomer for a variety of sweet potato. The group of plants which has been classified by plant scientists as yams are quite distinct from and not even closely related to the sweet potato. In general, the sweet potato is a member of the family *Convolvulaceae* — the morning glory family *(5).* It is a dicotyledon with net-veined leaves. On the other hand, the yam is a member of the family *Dioscoreaceae.* It is a monocotyledon and in general has arrow shaped leaves, and flower parts occur in groups of three or multiples of three. At present yams are not grown commercially in the United States except for one kind, *Dioscorea batatis,* the chinese yam which is being cultivated on a small commercial scale in California *(1, 5).*

There are many varieties of yam grown in Nigeria, and early work of the Agricultural Department of Nigeria recognized six groups or species of the genus *Dioscorea* from which the cultivated yam of Nigeria is derived *(11).* The cultivated species are: *Dioscorea rotundata* (white yam), *D. cavanensis* (yellow yam), *D. alata* (water yam), and *D. dumentorum* (trifoliate yam, "esuru").

D. rotundata (white yam) is the most popular and the many varieties within this group are widely grown in different parts of Nigeria. The subvarieties are classified as to the nature of carbohydrate content—whether mealy or hard or suitable for use as pounded yam "iyan" or to be eaten as such when boiled in water. The other varieties are also important and are used widely as food, although most of the food products, "iyan" (pounded yam), "amala" (yam flour reconstituted in boiling water), and sometimes cakes and fried yams similar to "French fries", are all derived from *D. rotundata.*

One of the species of *Dioscorea* that requires special attention, although used less as food, is *D. alata* (water yam). It differs from other varieties in character and use of the tuber. This variety normally affords greater yields and does better than other varieties in soils of very low fertility. It has a thick dry coat next to the skin enclosing the flesh which has more water than the flesh of other varieties of yams. This is readily noticeable when the tuber is cut. The cut tuber undergoes browning more readily when exposed to air than the other varieties. The tuber is not suitable for pounding, but may be grated before boiling and made into porridge with vegetable oil,

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TABLE 1. PROXIMATE ANALYSIS OF YAM TUBERS,[1] POTATOES, AND SWEET POTATOES

Species	Moisture content (%)	CHO (%)	Fat (%)	Crude Protein (%)	Fiber & ash (%)
D. alata	65-73	22-29	0.03-0.21	1.12-2.78	1.3-3.4
D. cavanensis	83	15	0.05	1.02	0.93
D. rotundata	58-73	23	0.12	1.09-1.99	1.03-3.1
D. dumentorum	79	17	0.28	2.78	1.02
Potatoes	68-82	14-27	0.02-0.18	1.14-2.98	1.06
Sweet potatoes	58-81	17-43	0.18-1.66	0.45-4.37	1.20

[1]Potato and sweet potato are included in this table for comparison (19). Adapted from D. G. Coursey *(3)*.

green vegetables, shrimp, flavorings such as "egunsi" (powdered melon seeds after the shells of the seeds have been removed), and other condiments. It has been shown that *D. alata* contains relatively more protein than the other varieties *(10)*, and its protein content (Table 1) is comparable to that of the potato *(3)*.

The greatest problems with yams, and which need immediate solutions, are the seasonal variability in supplies and the inadequacy of yam food products for providing essential nutrients. There are times of the year between the rainy months (June to late October) when there are large supplies of the different varieties of yam. However, because efficient preservation and processing are lacking, yams and yam products are very scarce and expensive during the dry months.

The respiration rate of yams is much higher than that of other plant products. Perhaps this causes the very rapid conversion of polysaccharides in yams to hexose sugars during traditional storage. The high respiration rate accounts for the early "death" of the yam tissue. No successful storage method has been developed for yam tubers. Hence, the only and surest way to effectively distribute yams and food products derived from yams throughout the year would be through use of a dehydration process to prepare various food products similar to dehydrated potato products.

Canning is usually a convenient method to preserve and distribute food products. American "yams" (sweet potatoes) are usually canned in sucrose or common table syrup. Usually, the potatoes are firmed by soaking in a 0.5% citric acid solution for 20-24 hr before canning. Use of a 40% syrup is then necessary to mask the acid flavor resulting from soaking in the acid solution. Canning of West African yams may not be economically feasible at the present time. Aside from the fact that such syruped canned yams similar to canned potatoes may not meet consumer acceptability, the cost of canning in Nigeria and other West African countries at the moment is exorbitant. It would, therefore, be of greater advantage to explore the possibilities of improving the existing techniques of yam processing.

The objectives of this review, therefore, are to *(a)* compare the apparent properties of yams with those of potato, *(b)* reiterate some of the latest techniques that have been used to dehydrate potato and discuss possibilities of their applicability to yam drying, and *(c)* discuss possible new food products, similar to convenience foods made from potatoes and the possibility of their being acceptable to consumers.

COMPOSITION OF VARIETIES OF YAMS AS COMPARED WITH POTATOES

There have been attempts at providing a detailed chemical composition of yam tubers rather than regarding them only as masses of starch as many nutritionists have done. It has been shown that a small proportion of the total carbohydrate consists of mono- and disaccharides, probably intermediates in the metabolic breakdown of starch. Observations using a paper chromatographic technique have indicated that freshly harvested *D. rotundata* contains two principal sugars; sucrose is present in larger amounts than glucose *(3)*. A trace amount of fructose was also detected. Accumulation of sucrose instead of glucose is evident from the very sweet taste developed by certain subvarieties of *D. rotundata* after some storage.

Yam tubers also contain small but significant amounts of protein, usually between 1-2% and occasionally as high as 3%. The protein contents of some yam species are lower than that of potatoes, but other varieties such as *D. alata* and *D. dumentorum* have comparable levels of protein (Table 1). The amino acid composition of protein in the yam tuber is not known with certainty. A partial analysis of the Japanese varieties, *D. opposita* and *D. japonica*, indicated low contents of sulfur bearing amino acids and lysine but large amounts of tryptophan and arginine *(16)*.

From data in Table 1, it appears that *D. alata* is closest to the potato in composition. The carbohy-

TABLE 2. VITAMIN C CONTENT OF VARIETIES OF YAM[1]

Variety	Vitamin C (mg/100g)
D. rotundata	6.5 - 11.6
D. cavanensis	4.5 - 8.2
D. alata	5.8 - 8.2

[1]Adapted from Coursey *(3)*. The average vitamin C content for the edible portion of potatoes is 17 mg/100 g *(18)*.

TABLE 3. VITAMIN C RETENTION DURING YAM PROCESSING[1]

Process	Percent of Vitamin C retained
Boiling (without peeling)	95
Frying (in Palm oil)	93
Roasting or baking	85
Boiling after peeling	65

[1]Adapted from D. G. Coursey *(3)*

drate, protein, and moisture contents are similar.

Some yams contain some toxic and pharmacologically active substances. It has been noted *(3b)* that the tubers of many wild species of yam are so toxic as to be dangerous to human life, and some have been used deliberately as sources of poison for hunting and fishing. The majority of the toxic materials belong to three classes of compounds: alkaloids, tannins, and saponins.

Alkaloids have been detected in at least seven of the species of yam examined but the largest amounts are found in *D. dumentorum* and *D. hispida* with smaller quantities in *D. alata*. The alkaloid, dioscorine, occurs in the tubers of *D. hispida* and its derivative, dihydrodioscorine, occurs in some varieties of *D. dumentorum* which is closely related to *D. hispida*. Doscorine is a potent neurotoxin, causing a general paralysis of the central nervous system. Dihydrodioscorine is also toxic, acting as a convulsant poison, but the toxicity appears to be milder than that exhibited by the parent compound. It is the dihydrodioscorine that has been found in the West African species of *D. dumentorum (1a)*. Fortunately, the extremely poisonous dioscorine is soluble in water, and hence in the traditional cooking of yams tubers are detoxified by washing employed. The toxin is also soluble in chloroform and methanol.

Little is known of the exact nature of the tannins in yams although occurrence of these compounds have been reported in *D. cirrhosa* and the dark red-fleshed form of *D. alata*.

The third main class of compounds of pharmacological importance occurring in yams are the saponins which are glycosides consisting of a sugar residue (one or more units of glucose or galactose) linked through oxygen to a triterpenoid. Development of foam when pieces of yam tuber are shaken with water is an indication of the presence of these soapy substances. Saponins have very powerful hemolytic actions, and for this reason, they are highly toxic if injected directly into the blood stream. However, they are generally inactive when ingested through the mouth, because they are perhaps hydrolyzed in the digestive system or possibly the ingested substances are not absorbed from the intestine. Saponins, however, have a bitter taste, and yams containing substantial quantities are generally not eaten.

There has been no explanation for *(a)* the various colors associated with varieties of raw yam tubers, *(b)* certain types of discoloration that occurs when the raw tuber is cut, or *(c)* darkening of the reconstituted flour derived from *D. rotundata* "amala." It is not unlikely that yam tubers contain definite (but perhaps small) amounts of phenolic and related substances such as flavones, anthocyanins, and monohydric and polyhydric phenols similar to those found in the potato tuber *(15)*. The yellow coloration of *D. cavanensis* which is not leached into the cooking water apparently results from the presence of fat-soluble pigments possibly carotenoid compounds.

Perhaps the most important of the minor constituents of yam tubers are vitamins, especially vitamin C. These occur in quantities large enough to contribute substantially to human nutrition in yam consuming areas. A more detailed study *(4)* made in Ghana of varieties of *Dioscorea* revealed the vitamin C contents shown in Table 2. In view of the close similarities between potato and yam tubers, it is most likely that techniques and precautions taken in processing potatoes into various forms with a long shelf life might be applicable to yam processing.

POTATO DEHYDRATION TECHNIQUES

No attempt is made in this short discussion to review all techniques used in potato processing. Rather, a few of the recent developments in technology which might lend themselves to yam processing are reviewed. The feasibility of applying these processes to making new products from yams are briefly explored.

Potato flakes by drum drying

Drum drying is the most commonly used method to dry potatoes. In this process, raw potatoes are sliced after peeling and the sliced potatoes are precooked and cooled. Potatoes are then mashed or riced, after which additives to improve product flavor, stability, and texture are added to the riced potato before it is dried. Sodium sulfite and sodium bisulfite are used to prevent oxidative changes during processing and to improve the shelf-life. Usually, an antioxidant "Tenox V,"[3] a mixture of butylated hydro-

xytoluene (BHT), butylated hydroxyanisole (BHA), propyl gallate, citric acid, propylene glycol, and vegetable oil, is used as a preservative against oxidative deterioration.

The mashed potato is dried by feeding it to the top surface of a single-drum drier. A two-way ribbon screw, rotating oppositely to the drum distributes the wet mash uniformly on the surface of the drier. Small-diameter unheated rolls progressively apply fresh mash to that already partially dried, thereby filling the interstices and building up a dense sheet. The sheet is dried by steam in the drum at 75-80 psig and a drum speed of 2 rpm. Under these conditions moisture in the mash is reduced from 80% to 4.5 to 5% in approximately 2 min. The dried sheet is peeled away from the drum surface by a "doctor" knife, but if a good dense sheet is made, it usually peels away from the drum just ahead of the knife and leaves the drum surface quite clean. A slitting roll and a transverse cutting roll convert the sheet into half-inch square flakes.

A process of flakelet manufacture developed by Pader *(12)* uses a high level of monoester of a polyhydric alcohol containing at least three hydroxy groups and a saturated higher fatty acid. The compound is thoroughly mixed with the cooked potatoes before drying. Inclusion of the monoester results in a dehydrated potato product which, on reconstitution has textural and other characteristics of freshly prepared mashed potato. In many instances, the quality of the reconstituted dehydrated product surpasses the quality of freshly prepared mashed potatoes of the same type. In addition, the quality of the reconstituted product is further improved by addition of edible protein such as calcium or potassium caseinate, whole milk solids, skim milk solids, or soybean protein isolates to the cooked potato before drying. The edible protein improves drum drying characteristics of the mashed potato containing the monoester *(12)*.

Development of a food product from yams similar to mashed potatoes is desirable since reconstituted mashed potatoes are quite similar in color and texture to the pounded yam "iyan" which is very popular with people in the yam-growing areas of Nigeria.

Yams are low in protein content and the small amount of protein in the tuber is largely lost when the yam is peeled before processing it into "iyan." Potato flakelets manufactured when protein is incorporated into mashed potatoes suggests that a protein-rich food product from yams is a possibility and would be desirable for development. Meals prepared from yams are usually low in protein although attempts are made to compensate for this deficiency by supplementing these meals with meat and vegetable stew. Incorporation of protein into the "mashed yam" envisaged would increase its nutritional quality as well as improving its drying characteristics, as has been done with potatoes.

Rather than using calcium caseinate or milk proteins which are not readily available in the yam growing areas, plant proteins may be used as additives. Soybean, cotton seed, or peanuts (ground nut) which are commonly cultivated in Nigeria are good and inexpensive sources of protein. Soybeans are grown widely in the Northern states of Nigeria where they are used mainly as cattle feed. Soybean protein is slightly inferior to animal proteins but it is the most desirable of all plant proteins. It contains substantial amounts of the essential amino acids and the glutamic acid content is particularly high. Obstacles to the general use of soybeans for human food products have been the bitter and objectionable odor of such products and the presence of anti-nutritional factors such as a trypsin inhibitor, a hemagglutin, saponin, a goitrogenic factor, anticoagulant factors, and some others *(9)*.

Removal or changing these unpalatable constituents is now possible through heating, soaking, or fermentation with yeast. There is, therefore, no obstacle to using soybean protein isolates in yam processing except, of course, transportation of the soybeans from the Northern States to the yam growing areas in the Southern States.

Cotton seed is another inexpensive source of the essential amino acids. Cotton is grown on the same plots as yams at the time when the yam plant sheds its leaves. Cotton seed is only slightly inferior to the soybean as a protein-containing product, being somewhat deficient in methionine and lysine. The slight lysine deficiency is aggravated by traditional heat treating techniques, but properly processed cotton seed offers an excellent source of protein material. Ironically, the protein-rich cotton seed is fed to goats or discarded as waste in these yam growing areas. On a very limited scale, cotton seed is fermented and made into "ogiri." The greatest problem with defatted cotton seed in areas of the world where it is recognized as a good source of protein is the presence of a toxic polyphenolic pigment, gossypol, which must be removed before it is consumed by monogastric animals including man. Gossypol is innocuous to ruminants such as goats, sheep, and cattle because the pre-digestion process in the rumen converts the gossypol into a nontoxic material before entering the true stomach.

However, edible-grade cotton seed flour can now be

[3]Eastman Chemical Co.

prepared by removing gossypol by several methods of which a fermentation process seems to be most effective. Microorganisms used in the process produce, as metabolic by-products, vitamins such as riboflavin, B_6, nicotinic acid, biotin, folic acid, and vitamin K. Hence, the fermentation increases the nutritional content of cotton seed. Cotton seed flour instead of milk proteins could be widely applicable for production of protein-rich "mashed" yam products.

A problem that needs attention in fortification of yam flour or "mashed yams" with protein is the nonenzymatic browning reaction (Maillard reaction). This could occur between the reducing sugars of yam and the added protein supplements during drum-drying processes. In the spray-drying process, heat contact with the product is short during drying and removal. Quality is more likely to be preserved as overheating is less likely. Proper study of the optimum temperature and conditions of drying is necessary so that availability of added protein is not reduced during drum-drying.

Potato flour by spray drying

Various ways to prepare powdered potatoes have been used in the past but the recently developed spray drying technique, in which the highest possible degree of natural potato flavor in the rehydrated product is retained, is worthy of note. Most of the flavor components of a potato are concentrated in the skin and in those zones of the potato adjacent to the skin. As a result, there have been attempts to remove only the outer peel of the potato before cooking and drying or even to process whole unpeeled potatoes. Disadvantages of this process are *(a)* off-color products because the browning precursors are also concentrated in or near the skin of the potato, *(b)* failure to separate the peel and other undesirable portions of the potato in a product which has an uneven texture and little consumer acceptability, and *(c)* such dehydrated products have poor shelf-life because of the fat in the dehydrated product.

In processes developed by Hollis and Borders *(8)* and Sienkiewicz and Hollis *(14)*, potato material containing a substantial portion of the peel is cooked under carefully controlled conditions. In this process, whole, raw potatoes are washed to remove field dirt, and subjected to carefully controlled precooking conditions, after which the potato cells are conditioned for the rigorous treatment to which they are subjected during slurrying. After slurrying, imperfections are removed mechanically by screening.

Following mechanical separation of undesirable portions, the slurry is dewatered to remove large quantities of free and solubilized starch, soluble reducing sugars, and some of the proteins which have been solubilized. The dewatering process may be effected by centrifugation or horizontal vacuum filter. The resulting filter cake is then reslurried in an aqueous medium and additives such as methyl cellulose, an aerating agent to impart fluffiness, may be added. Emulsifiers (monoglycerides) which offset any pasty, gummy texture that might otherwise be produced in a reconstituted potato product are added. The re-slurried potatoes with additives are spray-dried using spray drying equipment with drying gas or ordinary heated air flowing co-current or counter-current to the slurry being dried. The temperature of spray-drying is critical. It must be such that scorching of potatoes does not occur. Too high a temperature might cause potato solids to "toast," i.e. develop a tan color and sometimes an off-odor and -flavor. Of course, too low a temperature will not remove sufficient moisture from the atomized reslurry.

Processing of yams into flour and reconstituting this in boiling water to give "amala" has been a long practiced process in the home in yam-consuming areas of West Africa and it is still the chief means of processing the yam to spread its consumption over the year. However, it is a very inefficient, laborious, time-consuming, and uneconomical process, in that most of the raw yam is wasted during processing.

In this process, yam tubers, usually subvarieties of *D. rotundata* (white yam), are peeled, washed, and sliced and then sun-dried for several days. The sun-dried slices are crushed in a wooden mortar with a pestle and sieved. Coarse particles are returned to the mortar and again crushed and sieved. This recycling continues until the coarse particles are reduced to a minimum. The flour is then reconstituted in boiling water to give a brown, rather plastic dough which is consumed with vegetable stew, meat, and other condiments.

The mechanical processing of yam into a product which, on reconstitution, will give a meal similar to the traditional "amala" is very desirable and urgently needed. The spray-drying method described above and used for potatoes would lend itself to producing such a product. It is desirable to incorporate those portions of the yam tuber next to the skin in the powdered yam because, as in the potato, flavor components of the yam tuber are likely to be concentrated in the skin. Moreover, the small amount of protein contained in yam tubers is largely in the skin portions which are usually discarded as waste or sometimes fed to goats and sheep. An additional advantage of incorporating the skin portions of the yam tuber into yam flour is the high probability that vitamin C is retained in the product. It has been

observed *(4)* that retention of ascorbic acid during cooking of yams is highest when the yam is unpeeled before cooking (Table 3).

However, no explanation is given for the high retention of ascorbic acid when the yam is fried in palm oil or baked. It is probable that the ascorbic acid of the red palm oil is incorporated into the fried or baked yams. Roasting is normally done without peeling and, therefore, high retention of vitamin C is to be expected.

A processing method which allows retention of the peel of the yam as has been done with potatoes should be explored in developing a nutritious new product from yams. The traditional method of producing yam flour results in a reconstituted product that varies in color from beige or cream-colored to dark brown (almost black). Too dark a brown color of the reconstituted flour (amala) is objectionable but a lighter brown color, close to that of scorched powdered milk is much more acceptable. It is likely that a spray-dried yam flour would yield a rehydrated product which is lighter in color than the "amala" which consumers are used to, but there is no doubt that such a rehydrated product would be acceptable.

Freeze-drying method

Freeze-drying of the whole fresh tuber would be an ideal and superior method for dehydrating yams. Freeze-drying of biological materials is gaining popularity as a means of food preservation and many workers are trying to explore its true potentialities for large scale use.

Drying of a substance from the frozen state called "freeze drying," sublimation drying," or "lyophilization" depends on creation and maintenance of a difference in water vapor pressure between the very dry immediate surroundings of a substance and the ice in the frozen interior of the substance *(17)*. In this process, water vapor is continuously transported away from the substance but ice in the substance never melts. As a result, surfaces are unable to shrink as drying proceeds. When the product is frozen, water is withdrawn from the highly hydrated colloids of the food substance, first by crystallization of pure ice and then, if the temperature is low enough, crystallization of the remaining more concentrated solution. Sublimation of water, the solid constituents being completely immobilized, leaves behind a light, microporous structure of substantially the same dimensions as the original piece. Reabsorption of water into this spongy material is not only very rapid, but is usually quite complete as well because little denaturation of colloid constituents takes place.

Dehydration of the whole yam tuber by freeze-drying so that the characteristics of the yam are retained after rehydration is worth trying on a laboratory scale. The freezing point of yam tissue, like that of most biological materials is somewhat below zero. Investigations *(3)* yielded values between −0.9 C to −1.4 C. The freezing points of the tissues of *D. alata* appear to lie in the upper part of the range. This is as expected since the water content is much higher than the water content of other species. The freezing points of the varieties of *D. rotundata* lie in the lower part of the range.

However, the high cost of freeze-drying would be a limiting factor in its application to large-scale dehydration of yams. It is estimated that freeze-drying costs about five to ten times that of conventional drying of foods. The cost is estimated to be 10 cents (slightly less than Nigerian 10 kobo) per pound of water removed. This estimate is based on 1965 figures *(6)*. With advancement in technology, the cost has, perhaps, been cut from the 1965 estimate. Although freeze-drying is a superior process of preservation for the future, at present it is unappealing economically.

OTHER POSSIBLE NEW FOODS FROM YAM

New yam products similar to potato products are possible. For instance yam "nuts," similar to potato nuts, the production of which is now discontinued because some pieces contain hard, compact areas which are difficult to chew *(15)*, might be tried. A potato snack has been made from freshly cooked potatoes, potato starch, potato flour, vegetable oil, salt, and shortening *(13)*. This product has an appealing taste, crisp texture and light golden brown color. A similar yam product in which flavorings and seasoning materials such as onion, hot pepper, and perhaps "egunsi" from melon seed (*Citrullis vulgaris*) or "origi" obtained from fermented cotton seed and red palm oil would, no doubt, be appealing and well accepted.

CONCLUSIONS

There are many possible products that could be developed from yams and the various methods for dehydrating potatoes could be adapted for drying yams, because there are similarities between the two tuberous crops. However, a well balanced program of fundamental and applied research is urgently called for to study the carbohydrate content of the various species of yams, changes in sugar content during storage, and the ratio of reducing to non-reducing sugars at various storage temperatures. It is also pertinent to investigate the cause of browning and

blackening and factors contributing to the pigmentations of yams.

REFERENCES

1. Anonymous. 1951. Yams, Chinese kind, seen as an important U.S. crop. Sci. News Letter, 60, (12) 185. Cited by Edmond, J. B. and G. R. Ammerman. 1971. Sweet potatoes: Production, processing, marketing. The Avi Publishing Company Inc. Westport, Conn., p. 28.

1a. Bevan, C. W. L., and J. Hirst. 1958. A convulsant alkaloid of *Dioscorea dumentorum*. Chem. Ind. 4:103.

2. Boswell, V. R. 1950. Commercial growing and harvesting of sweet potatoes. U. S. Dept. Agr. Farmer's Bull. 2020. Cited by J. B. Edmond, and G. R. Ammerman. 1971. Sweet potatoes: Production, processing, marketing. The Avi Publishing Company Inc., Westport, Conn., p. 28.

3. Coursey, D. G. 1967. Yams, an account of the nature, origins, cultivation and utilisation of the useful members of the *Dioscoreaceae*. Longmans, London and New York. p. 153-171.

3a. Coursey, D. G. 1967. Yams, an account of nature, origins and utilisation of the useful members of the *Dioscoreacea*. Longmans, London and New York. p. 206-221.

4. Coursey, D. G., and A. Aidoo. 1966. Ascorbic levels in Ghanian yams. J. Sci. Food Agr. 17:446-449.

5. Edmond, J. B., and G. R. Ammerman. 1971. Sweet potatoes: Production, processing, marketing. The Avi Publishing Company Inc. Westport, Conn. p. 28.

6. Hall, C. W., and T. I. Hedrick. 1966. Drying of milk and milk products. The Avi Publishing Company, Inc., Westport, Conn. p. 58.

7. Hayward, H. E. 1938. The structure of economic plants. Macmillan Co., New York. Cited by J. B. Edmond, and G. R. Ammerman. 1971. Sweet potatoes: Production, processing, marketing. The Avi Publishing Company, Inc., Westport, Conn., p. 28.

8. Hollis, F., Jr., and B. Borders. 1965. Process for preparing dehydrated potatoes. U. S. Pat. 3,220,857. Nov. 30.

9. Leiner, I. E. 1958. Effect of heat on plant proteins. p. 83-91. *In* A. M. Altschul (ed) Processed plant protein foodstuffs. Academic Press, New York.

10. Oyenuga, V. A. 1959. Nigeria's feeding stuffs: Their composition and nutritional value, 2nd ed. ref., Ibadan, Ibadan Univ. Press.

11. Oyenuga, V. A. 1967. Agriculture in Nigeria, an introduction. FAO publication, Rome. p. 160-163.

12. Pader, M. 1964. Process for making dehydrated potatoes. U. S. Pat. 3,163,546. Dec. 29.

13. Potter, A. L., and M. L. Belote. 1963. New potato snack item. Bakers Weekly 197 (8):42-43.

14. Sienkjewicz, B., and F. Hollis, Jr. 1966. Process for preparing dehydrated foods. U. S. Pat. 3,261,695. July 19.

15. Talburt, W. F., and O. Smith. 1967. Potato processing. The Avi Publishing Company, Westport, Conn., p. 25.

16. Takahashi, T. 1928. On the protein of the tuber of the yam: Bull. Agr. Chem. Soc. Japan 4:53-55.

17. Van Arsdel, W. B., and M. J. Copley. 1963. Principles of food dehydration, Vol. 1. The Avi Publishing Company, Westport, Conn., p. 157.

18. Watt, B. K., and A. L. Merril. 1963. Composition of foods: raw, processed, prepared. U. S. Dept. Agric. Handbook (8) p. 50.

19. Winton, A. L., and K. B. Winton. 1935. The structure and composition of foods. Wiley, New York. Vol. 11, p. 106.

CHEMICAL CHARACTERISTICS, BACTERIAL COUNTS, AND POTENTIAL SHELF-LIFE OF SHRIMP FROM VARIOUS LOCATIONS ON THE NORTHWESTERN GULF OF MEXICO[1]

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(Received for publication April 5, 1973)

Abstract

Freshly harvested white shrimp (*Penaeus setiferus*) were taken from 13 locations on the northwestern coastline of the Gulf of Mexico. Freshly harvested brown shrimp (*Penaeus aztecus*) were taken from 3 different water depths near Port Aransas, Tex. Brown shrimp taken from commercial fishing boats at time of landing also were examined. Samples were analyzed for amino nitrogen (AA-N), NH_3, total volatile nitrogen (TVN), trimethylamine nitrogen (TMN), bacterial content, and pH. A portion of each sample was placed on sterile ice and allowed to spoil. Spoilage odors appeared in white sea-shrimp after storage for 11-50 days, for brown sea-shrimp in 20-30 days, and in brown boat-shrimp after 0-15 days. Both TVN and AA-N varied considerably from sample to sample and did not show a consistent pattern of change during iced storage. TVN/AA-N ratios increased as samples spoiled. TVN/AA-N ratios greater than 1.3 mg N/mM indicated a short shelf-life of boat shrimp. TMN production was evident in boat-shrimp samples (4 out of 9 samples) with high TVN levels. Bacterial counts of fresh shrimp did not exceed 10^4/g. Nine of the 10 boat-shrimp samples had counts in excess of 10^6/g. Counts of samples spoiled on sterile ice ranged from 2×10^6 to 10^{10}/g. The estimated reduction of the maximum potential shelf-life of boat-shrimp by handling and storage was 0-15 days.

Shrimp are caught in waters varying in many characteristics such as level and type of microbial population, salinity, temperature, organic and inorganic matter, and plankton. Little is known about variations in chemical characteristics, bacterial count, and potential shelf-life of Gulf Coast shrimp harvested from different areas. The microbial flora of shrimp will reflect to some extent the microbiological characteristics of the water. Cobb and Vanderzant *(4)* have shown that different microbial species isolated from shrimp produce different spoilage patterns in refrigerated shrimp. Increased levels of trimethylamine (TMA) in shrimp are often associated with quality deterioration. TMA is generally considered to be the result of bacterial action on trimethylamine oxide (TMAO). Velankar and Govindan *(9)* reported that the TMAO content of shrimp taken from brackish water was lower than in shrimp taken from salt water. Hence, differences in chemical and/or microbiological characteristics of the environment and of the shrimp harvested from these areas could influence the shelf-life during iced storage.

Reduction of the shelf-life of shrimp through tissue enzymes and microbial activity begins during handling on the boat. These activities can be kept to a minimum if shrimp is handled on board under sanitary conditions and is iced promptly and properly. At present, quality of stored iced shrimp is determined mainly by appearance and odor, tests which frequently lack uniformity and provide little information about its potential shelf-life. This study reports on the potential shelf-life and changes in chemical characteristics and bacterial counts of shrimp from different locations in the Northwestern Gulf of Mexico during iced storage. A chemical analysis (TVN/AA-N ratio) is discussed for use in conjunction with appearance and odor evaluation to determine the suitability of shrimp for processing.

Experimental

Shrimp samples

Freshly harvested white shrimp (*Penaeus setiferus*) were taken from different locations (bays and harbors) on the northwestern coast of the Gulf of Mexico between Galveston and Port Isabel, Tex. Freshly harvested brown shrimp (*Penaeus aztecus*) were taken from different locations in the Gulf of Mexico adjacent to Port Aransas, Tex. Freshly harvested shrimp were taken during June to August 1972. Brown boat-shrimp were obtained from shrimp boats on arrival at different locations on the northwestern coast of the Gulf of Mexico during July and August 1972. Freshly harvested samples were placed on a sheet of sterile plastic and were deheaded using sterile rubber gloves. While grasping the carapace with sterile forceps, tails were removed by cutting with sterile scissors. They were placed in sterile containers, rinsed with sea water from which they had just been removed and placed on sterile ice in an ice chest. Sterile ice was prepared by freezing heat-sterilized water in sterile stainless steel pans. Shrimp tails obtained from fishing boats at time of landing were immediately placed on sterile ice. Sanitized (chlorinated) ice chests ($40 \times 30 \times 30$ cm, Sears, vacucel insulated) were lined with a sterile plastic bag with holes in the bottom. The ice chests contained a partition which suspended the bag 4 or 5 cm above the bottom. Before inserting the shrimp, a layer (15 cm) of sterile ice was placed in the bag. After insertion of shrimp, it was covered with a 10 cm layer of sterile ice. Water was drained from the chambers and ice was added when needed. Samples

[1]Published with the approval of the Director of the Texas Agricultural Experiment Station, College Station.

TABLE 1. CHEMICAL ANALYSES AND AEROBIC PLATE COUNT (APC) OF FRESHLY HARVESTED WHITE SHRIMP TAKEN FROM DIFFERENT LOCATIONS ON THE NORTHWESTERN COASTLINE OF THE GULF OF MEXICO

Location of catch	AA-N[a] mM/100 g	TVN[b] mg N/100 g	NH_3/AA-N mg N/mM	TVN/AA-N mg N/mM	Log APC/g
Galveston					
(West Bay)	22.30	10.34	0.62	0.46	3.8
(Harbor)	24.33	18.65	0.91	0.77	3.8
Bacliff	21.39	9.53	0.78	0.45	4.0
Matagorda	26.83	17.02	0.70	0.63	3.4
Palacios	26.54	15.39	0.58	0.58	2.8
Port Lavaca	23.88[c]	14.68	0.60	0.61	3.5
	19.60[d]	7.33	0.28	0.37	3.9
Indianola	24.65	9.78	0.48	0.40	2.5
Aransas Pass	26.74	11.92	0.60	0.45	2.5
Brownsville (ship channel near entrance)	31.46	15.69	0.85	0.50	3.0
(Mid-channel)	20.26	16.51	0.90	0.82	2.4
Port Isabel	23.32	11.01	0.63	0.47	2.8
Seabrook	25.46	14.98	0.58	0.59	2.8
	18.97	14.27	–	0.75	2.9

[a]α-Amino, proline and ammonia nitrogen
[b]Total volatile nitrogen
[c]Harvested in June
[d]Harvested in August

were withdrawn daily with sterile forceps and examined for appearance and off-odors by a trained three-member panel. Shelf-life was defined as the number of days of refrigerated storage until off-odors developed. At that time refrigerated storage was terminated.

Microbiological and chemical analyses

Aerobic plate counts were determined on Standard Methods Agar (SMA, BBL) as described in a previous report *(8)*. Plates were incubated for 3 days at 25 C.

Shrimp extracts for chemical analyses were prepared by placing five shrimp in a Waring blendor and blending with 7% trichloroacetic acid solution (2 ml per gram of shrimp) until relatively homogeneous. The mixture was centrifuged to remove insoluble protein. Both total volatile nitrogen (TVN) and trimethylamine (TMN) analyses employed the microdiffusion procedure of Conway *(5)* with saturated Na_3PO_4 as releasing agent to prevent production of extraneous NH_3 during analysis. Values were multiplied by 1.3 to correct for incomplete distillation *(3)*. Amino nitrogen (AA-N) and NH_3 were determined by use of a Beckman Model 121C fully Automated Amino Acid Analyzer. Before analysis, shrimp extracts were frozen and recentrifuged to remove residual protein. Measurement of pH was made with a combination electrode on five or more shrimp blended with water (2 ml/g shrimp).

RESULTS AND DISCUSSION

Characteristics of freshly harvested white shrimp

Marked differences in AA-N levels and TVN levels existed between samples of freshly harvested white shrimp from different locations (Table 1). TVN levels were below 19 mg N/100 g and AA-N levels were either at or above 19 mM/100 g. In 50% of the samples the values for TVN/AA-N and NH_3/AA-N were similar indicating that most of the TVN was NH_3. In the other samples, NH_3 levels exceeded TVN levels. This occurred when samples had to be held for several days before amino acid analysis suggesting hydrolysis of compound(s) which yield NH_3. Preliminary experiments indicate that hydrolysis of glutamine is involved. The value of TVN/AA-N of freshly harvested white shrimp was always less than 0.90 mg N/mM. Trimethylamine (TMN) was not detected. Bacterial counts (2.5×10^2 to 10^4/g) were in a range normal for freshly harvested shrimp *(2, 8)*.

The white shrimp taken from different locations were placed on sterile ice until spoilage odors were detectable. The time at which serious off-odors appeared varied considerably (Table 2) but for 11 out of 14 (79%) samples ranged from 25 to 37 days when bacterial counts reached levels of 2×10^6 to 3×10^9/g with a geometric mean of 5×10^7/g. No relationship existed between potential shelf-life on sterile ice (Table 2) and initial bacterial count, AA-N, TVN, NH_3/AA-N or TVN/AA-N. Factors which may have caused differences in shelf-life include type of microbial population, variations in tissue enzyme activity, and differences in chemical characteristics of shrimp of unknown nature. Most samples maintained a commercially acceptable appearance during the test period. Little development of melanosis occurred. The main spoilage odor was "musty".

A comparison of data on chemical analyses of freshly harvested (Table 1) and spoiled white shrimp (Table 2) shows that on spoilage *(a)* AA-N decreased sharply, *(b)* TVN increased in most samples, and *(c)* values for TVN/AA-N and NH_3/AA-N increased. Decreases in AA-N most likely were the result of the

TABLE 2. SHELF LIFE, CHEMICAL ANALYSES AND AEROBIC PLATE COUNT (APC) OF FRESHLY HARVESTED WHITE SHRIMP KEPT IN STERILE ICE UNTIL SPOILAGE OCCURRED

Location of catch	Shelf life days	pH	AA-N[a] mM/100 g	TVN[b] mg N/100 g	NH_3/AA-N mg N/mM	TVN/AA-N mg N/mM	Log APC/g
Galveston							
(West Bay)	35	–	1.87	15.79	5.78	8.44	8.2
(Harbor)	50	8.4	<1	26.81	>8	>8	10.1
Bacliff	50	7.8	<1	NM[c]	>8	NM	LA
Matagorda	32	–	2.91	19.77	4.90	6.79	6.7
Palacios	25	–	2.95	16.82	3.94	5.70	9.5
Port Lavaca	34[d]	–	3.15	20.17	4.65	6.40	6.3
	11[e]	8.0	11.86	–	1.53	–	6.8
Indianola	35	–	5.00	19.27	3.05	3.85	6.5
Aransas Pass	28	–	11.86	30.17	2.01	2.54	7.5
Brownsville (ship channel near entrance,	30	–	8.05	60.70	4.84	7.54	6.5
(Mid-channel)	37	7.7	<1	7.33	>8	>8	8.7
Port Isabel	35	8.1	4.52	20.79	3.91	4.59	8.5
Seabrook	27	7.8	5.66	9.27	1.20	1.64	8.6
	27	8.3	7.62	–	3.18	–	8.0

[a]α-Amino, proline and ammonia nitrogen
[b]Total volatile nitrogen
[c]Level too low for accurate measurement
[d]Harvested in June
[e]Harvested in August

TABLE 3. CHEMICAL ANALYSES AND AEROBIC PLATE COUNT (APC) OF FRESHLY HARVESTED BROWN SEA-SHRIMP TAKEN FROM DIFFERENT DEPTHS IN THE GULF OF MEXICO ADJACENT TO PORT ARANSAS, TEXAS

Location of catch	AA-N[a] mM/100 g	TVN[b] mg N/100 g	NH_3/AA-N mg N/mM	TVN/AA-N mg N/mM	Log APC/g
13 Fathoms[c]	23.27	9.84	0.55	0.42	2.5
18 Fathoms[c]	21.78	19.67	0.87	0.90	2.0
13 Fathoms[d]	21.41	15.18	0.74	0.71	3.2
13 Fathoms[e]	23.62	16.54	0.70	0.70	2.5
18 Fathoms[d]	19.53	16.61	0.69	0.85	3.2
28 Fathoms[d]	25.10	16.51	0.55	0.66	3.4

[a]α-Amino, proline and ammonia nitrogen
[b]Total volatile nitrogen
[c]Harvested June, 1971
[d]Harvested July, 1971
[e]Harvested August, 1971

washing action of melted ice. Only two samples had a TVN content greater than 30 mg N/100 g shrimp, the limit of acceptability used in some sections of the Australian and Japanese markets *(7)*. In the spoiled samples the TVN/AA-N values exceeded those of NH_3/AA-N suggesting the presence of other amines. TMN, however, was not detected. Portions of the shrimp samples from Aransas Pass, Brownsville, and Port Isabel were removed after 14 days of storage on ice and analyzed. The NH_3/AA-N ratios for shrimp from Aransas Pass, Brownsville (ship channel entrance), and Port Isabel were at that time 1.14, 1.33, and 1.13 mg N/mM respectively. These figures correspond to NH_3/AA-N increases of 0.038, 0.034, and 0.040 mg N/mM/day respectively. These increases are in excellent agreement with increases in NH_3/AA-N of 0.032 mg N/mM/day calculated from analyses on stored sterile shrimp juices *(4)* and suggest that these resulted primarily from tissue enzyme activity. In the final phase of the storage of shrimp from Aransas Pass, Brownsville, and Port Isabel, the average daily increases in NH_3/AA-N ratio were 0.062, 0.219, and 0.130 mg N/mM. These increases probably resulted from tissue enzyme and microbial activities *(4)*. The pH of spoiled shrimp ranged from 7.7-8.4, values close to 8.0 which some investigators *(1)* consider indicative of spoilage.

Characteristics of freshly harvested brown shrimp

The results of chemical analyses of freshly harvested brown sea-shrimp (Table 3) were similar to those of freshly harvested white shrimp. AA-N levels were greater than 19 mM/100 g (range 19.53-25.10) and TVN levels were < 20 mg N/100 g (range 9.84-19.67). TVN/AA-N ratios did not exceed 0.90 mg

N/mM. Values for TVN/AA-N ratios were similar to those of NH_3/AA-N. Bacterial counts of freshly harvested samples did not exceed 2.5×10^3/g.

Although the number of samples was limited, the shelf-life of brown sea-shrimp on sterile ice (Table 4) was shorter (range 20-30 days, average 24 days) than that of white shrimp (range 11-50 days, average 33 days). The pattern of changes in AA-N, TVN, NH_3/AA-N, and TVN/AA-N on spoilage of brown shrimp was similar to that when white shrimp spoiled. TMN was not detected in either freshly caught or spoiled brown sea-shrimp. Bacterial counts per gram of spoiled brown sea-shrimp ranged from 2.5×10^6 to $>3 \times 10^9$. The predominant spoilage odor was "musty". The pH of the spoiled samples was relatively high (7.8-8.0).

Characteristics of brown shrimp from commercial boats at time of landing.

The results of analyses on brown boat-shrimp (Table 5) differed considerably from those of freshly harvested shrimp (Tables 1, 3). In general, AA-N levels were low (Avg. 18.43) and TVN levels high (Avg. 26.76) compared with freshly harvested white or brown shrimp (Avg. AA-N 22.45-23.97; Avg. TVN 13.36-15.73). TVN levels in all but one sample exceeded those of freshly harvested shrimp (20 mg N/100 g). The values for TVN/AA-N and NH_3/AA-N exceeded those of freshly caught white or brown shrimp. Bacterial counts of just landed brown boat shrimp ranged from 5×10^5 to 10^8/g with a geometric mean of 1.2×10^7/g. A comparison of the values for the chemical and microbial indices of just landed boat shrimp (Table 5) and those of fresh and spoiled sea-shrimp (Tables 1-4) indicates that some quality deterioration had occurred in the boat shrimp. This is also apparent from the limited additional shelf-life of brown boat shrimp on sterile ice which ranged from 0 (Sample T) to 15 days (Table 6). AA-N levels of the spoiled brown boat shrimp were higher (range 11.63-16.58 mM/100 g, average 14.2) than those of spoiled brown sea-shrimp (range 2.75-14.61, average 7.8) or spoiled white sea-shrimp (<1-11.86). In samples with high TVN levels, TMN was evident. These samples were taken from boats where sanitary handling of the shrimp was poor. Values for TVN/AA-N and NH_3/AA-N of spoiled brown boat shrimp were usually high, TVN/AA-N values were larger than those for NH_3/AA-N. Bacterial counts of spoiled

TABLE 4. SHELF LIFE, CHEMICAL ANALYSES AND AEROBIC PLATE COUNT (APC) OF FRESHLY HARVESTED BROWN SEA-SHRIMP FROM DIFFERENT DEPTHS IN THE GULF OF MEXICO ADJACENT TO PORT ARANSAS, AND KEPT IN STERILE ICE UNTIL SPOILAGE OCCURRED

Location of catch	Shelf life days	pH	AA-N[a] mM/100 g	TVN[b] mg N/100 g	NH_3/AA-N mg N/mM	TVN/AA-N mg N/mM	Log APC/g
13 Fathoms, 18 Fathoms [c,d]	30	—	2.75	10.19	2.07	3.70	6.4
13 Fathoms	21	7.9	12.93	70.2	4.16	5.42	8.8
13 Fathoms	25	—	3.71	—	4.75	—	9.0
18 Fathoms	20	8.0	14.61	53.69	2.84	3.67	>9.5
28 Fathoms	23	7.8	4.99	22.11	3.39	4.43	8.3

[a]α-Amino, proline and ammonia nitrogen
[b]Total volatile nitrogen
[c]For date of catch see Table 3
[d]Samples taken in June pooled because of insufficient amounts of material.

TABLE 5. CHEMICAL ANALYSES AND AEROBIC PLATE COUNT (APC) OF BROWN BOAT-SHRIMP TAKEN FROM COMMERCIAL FISHING BOATS AT LANDING[a]

Sample	pH	AA-N[b] mM/100 g	TVN[c] mg N/100 g	NH_3/AA-N mg N/mM	TVN/AA-N mg N/mM	Log APC/g
R	7.9	20.78	22.62	0.97	1.09	6.3
S	8.1	18.48	24.66	1.11	1.33	7.2
T[d]	—	13.44	45.15	2.36	3.35	>7.5
U	8.2	14.18	24.76	1.35	1.75	6.3
V	7.4	22.21	25.74	0.91	1.15	7.0
W	8.1	18.26	38.43	1.28	2.10	7.9
X	7.3	20.53	19.16	0.91	0.93	7.7
CC	7.5	21.36	22.53	0.67	1.05	5.7
EE	7.1	16.53	20.49	0.86	1.24	7.3
FF	7.2	18.51	24.05	0.99	1.30	8.0

[a]Samples had been on board for a maximum of 6-8 days except for sample CC which had been on board 12 days.
[b]α-Amino, proline and ammonia nitrogen
[c]Total volatile nitrogen
[d]Spoiled at arrival in laboratory

TABLE 6. SHELF-LIFE, CHEMICAL ANALYSES AND AEROBIC PLATE COUNT (APC) OF BROWN BOAT-SHRIMP TAKEN FROM COMMERCIAL FISHING BOATS AT LANDING AND ALLOWED TO SPOIL ON STERILE ICE[a]

Sample	Shelf life days	Estimated total shelf life-days (ETSL)	pH	AA-N[b] mM/100 g	TVN[c] mg N/100 g	TMN mg N/100 g	NH_3/AA-N mg N/mM	TVN/AA-N mg N/mM	Log APC/g
R	11	18	7.9	15.06	97.84	14.98	3.70	6.50	9.3
S	8	15	7.8	11.63	97.89	14.82	4.58	8.41	9.2
U	6	13	8.5	12.41	93.99	16.77	4.64	7.57	9.0
V	5	12	7.5	—	23.44	0	—	—	8.4
W	4	11	7.3	13.39	30.47	0	1.77	2.28	9.0
X	15	22	7.7	15.08	75.82	6.32	3.10	5.03	9.0
CC	11	23	7.5	15.74	32.53	0	1.38	2.06	>8.1
EE	11	18	8.0	16.58	—	—	3.93	—	9.1
FF	3	10	7.7	13.32	31.49	0	1.32	2.36	8.6

[a]Samples had been on board for a maximum of 6-8 days except for sample CC which had been on board 12 days.
[b]α-Amino, proline and ammonia nitrogen
[c]Total volatile nitrogen

samples were greater than 10^8/g. The predominant spoilage odor was putrid. With freshly caught shrimp stored on sterile ice the predominant off-odor was "musty".

Through aseptic handling and storage on sterile ice, brown or white shrimp could be stored for long periods (average values 24 and 33 days) before spoilage odors developed. Even under such conditions the shelf-life is limited because of the activities of the natural microbial flora and tissue enzyme system. The shelf-life of white or brown sea-shrimp on sterile ice was called maximum potential shelf life (MPSL). With the aid of this figure some estimation can be made of the reduction in shelf-life of boat shrimp through commercial handling and storage practices. The average MPSL of brown sea-shrimp caught in July and August was 22 days (Table 4). The brown boat shrimp were also caught in July and August and except for sample CC had been on board an average of 7 days (range 6-8 days). Figures for the estimated total shelf life (ETSL), consisting of the number of days on ice on the boat plus the additional storage on sterile ice before spoilage occurred, are in Table 6. Differences between the average MPSL of brown sea-shrimp and ETSL of commercial brown boat shrimp were −1 (Sample CC) to 15 days (average 7.1 days, Sample T included). Figures for ETSL usually were high for shrimp from boats with excellent sanitary handling practices and storage.

In certain areas of the world high TVN (30 mg/100 g) or TMN (5 mg/100 g) levels are used as an indication of shrimp spoilage *(7)*. In this study 4 of 19 (21%) spoiled sea shrimp samples stored on sterile ice reached this level of TVN and none showed TMN. For spoiled brown boat shrimp the TVN level of 8 out of 9 samples exceeded 30 mg/100 g and 4 samples contained >5 mg TMN/100 g. This discrepancy in TVN and TMN levels of spoiled boat and spoiled sea-shrimp (stored on sterile ice) probably was caused by a difference in microbial activities. *Pseudomonas* species frequently constitute a significant part of the microbial flora of boat shrimp. These species produced large increases in TVN and TMN in shrimp juice *(4)*. On the other hand, microbial counts at time of catch usually are low and typical spoilage bacteria are not numerous at that time. Coryneform bacteria and *Achromobacter* species predominated in sea-shrimp, handled aseptically and stored on sterile ice. This difference in distribution of microbial flora probably is also responsible for the difference in predominant off-odors between spoiled sea-shrimp (musty) and spoiled boat shrimp (putrid).

Some investigators claim that high pH values of shrimp (>7.95) are indicative of spoilage *(1)*. This could not be substantiated in this study.

In general, as spoilage of sea- or boat-shrimp occurred, the value of AA-N decreased and that of TVN increased. TVN and AA-N are produced through tissue enzyme and microbial activities. Losses in TVN and AA-N occur during iced storage when shrimp are continually subjected to washing by water from melting ice. In some samples, increases in TVN after storage were small probably because NH_3 and amines were removed more rapidly than they were produced *(6)*. The value of TVN/AA-N increased, however, because of decreases in AA-N.

The value for TVN/AA-N of freshly harvested brown or white shrimp was always <0.9, for spoiled sea-shrimp >1.64, and for spoiled boat shrimp >2 mg N/mM. The value for TVN/AA-N may be useful in conjuction with an evaluation of appearance and odor as a screening test to determine shrimp quality. In this relation, the ratio TVN/AA-N is to be preferred over NH_3/AA-N or over TVN and AA-N values alone because *(a)* TVN and AA-N can be

measured by simple reliable tests in commercial seafood processing plants *(3), (b)* in spoiled samples TVN consists of NH_3 and various amines, and *(c)* considerable variation existed in AA-N (or TVN) values among freshly harvested samples as well as among spoiled samples. It is recognized that samples of acceptable appearance and odor could have shown certain defects after cooking. Carroll et al. *(2)* reported that some samples of pink and white shrimp rated Grade A with respect to appearance and odor were bitter after cooking.

In a limited field trial with 40 commercial boat samples, samples with TVN/AA-N values >1.3 mg N/mM were usually evaluated as poor (based on appearance and odor) by plant quality control personnel.

ACKNOWLEDGMENT

The authors thank the crew of the *Western Gulf*, experimental fishing boat of Texas Parks and Wildlife Department, for its assistance in obtaining the brown sea-shrimp. This study was supported in part by the National Sea Grant Program, NOAA, U. S. Department of Commerce, through institutional grant 2-35213 made to Texas A & M University.

REFERENCES

1. Bailey, M. E., E. A. Fieger, and A. F. Novak. 1956. Objective tests applicable to quality studies of ice-stored shrimp. Food Res. 21:611-620.

2. Carroll, B. J., G. B. Reese, and B. Q. Ward. 1968. Microbiological study of iced shrimp: Excerpts from the 1965 Iced-Shrimp Symposium. Circ. 284, U. S. Department of Interior, Washington, D. C. 17 pp.

3. Cobb, B. F., III, I. Alaniz, and C. A. Thompson. 1973. Biochemical and microbial studies on shrimp: Volatile nitrogen and amino nitrogen analysis. J. Food Sci. 38:431-436.

4. Cobb, B. F., III, and C. Vanderzant. 1971. Biochemical changes in shrimp inoculated with *Pseudomonas, Bacillus* and a coryneform bacterium. J. Milk Food Technol. 34:533-540.

5. Conway, E. J. 1958. Microdiffusion analysis and volumetric error. The MacMillian Company, New York. p. 199.

6. Iyengar, J. R., K. Visweswariah, M. N. Moorjani, and D. S. Bhatia. 1960. Assessment of the progressive spoilage of ice-stored shrimp. J. Fish. Res. Bd. Canada 17:475-485.

7. Montgomery, W. A., G. S. Sidhu, and G. L. Vale. 1970. The Australian prawn industry. 1. Natural resources and quality aspects of whole cooked fresh prawns and frozen prawn meat. CSIRO Food Preserv. Quart. 30(2):21-27.

8. Vanderzant, C., E. Mroz, and R. Nickelson. 1970. Microbial flora of Gulf of Mexico and pond shrimp. J. Milk Food Technol. 33:346-350.

9. Velankar, N. K., and T. K. Govindan. 1960. Trimethylamine oxide content of marine prawns occurring in the back waters and in the sea off Cochin. Proc. Indian Acad. Sci. 52B:111-115.

A PROCEDURE FOR PROCESSING SMOKED CHANNEL CATFISH (*ICTALURUS PUNCTATUS*)

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(Received for publication April 9, 1973)

Abstract

A study was made to determine the most acceptable procedure to prepare smoked channel catfish Effects of storage time before brining, fish weight, brining times and formulae, smoking times and temperatures, and storage time after processing on final product quality were evaluated. Use of freshly dressed catfish yielded a smoked product indistinguishable from that prepared from frozen catfish. Catfish averaging 10.5 oz (dressed) and brined for 24 hr at 40 F in a mixture prepared by dissolving 9 lb sugar (sucrose), 9 lb sodium chloride, 0.23 oz potassium nitrate, and 0.23 oz sodium nitrite in 72 lb water were highly acceptable. A gradual increase of internal flesh temperature from 90 to 140 F over a 22-hr smoking period, followed by an increase to 180 F over 4 hr with an additional 30 min at 180 F was judged as optimal for producing smoked catfish. Sensory panel evaluations on the finished product showed that smoked catfish were equally acceptable when compared with smoked haddock and chubs.

Production of farm-grown channel catfish in southeastern United States exceeds 50 million lb per year. Approximately half is sold as a frozen product while the remainder is sold as fresh processed catfish. To the authors' knowledge, none has been commercially sold as a smoked product.

Salmon, chub, herring, haddock, sablefish, mullet, and sturgeon are fish species generally smoked in the U. S. and are most common in the Pacific Northwest and Great Lakes area. Federal regulations assuring quality control have been established for processing and distribution of these smoked fishes (*5*). If smoked and handled properly, the product is considered a delicacy and, it might be added, as a luxury when considering its retail price.

With these points in mind, experiments were designed to develop a suitable processing procedure for preparing smoked channel catfish.

Experimental Procedure

Brining and curing procedures

Several preliminary experiments were done before arriving at the most acceptable brining formula, temperature and time of brining, and catfish size for producing the smoked channel catfish (*Ictalurus punctatus*). In addition, methods of killing and storage of the catfish before brining and smoking were evaluated. Those experiments contributing valuable data which were utilized in formulating the final recommended procedure are listed:

Experiment 1. Skinned and unskinned frozen catfish were thawed at room temperature, weighed, and submerged 17.5 hr at 50 F in a brine suggested by Tiemeier (*8*) which consisted of 2.0 lb light brown sugar and 1.5 lb salt (NaCl) in 16.5 lb water. The ratio of brine to fish was 3:2 (w/w).

Experiment 2. Frozen skinned and unskinned catfish were thawed at room temperature and weighed. Fish were then rubbed at a rate of 3/4 oz per pound of fish with a salt mix prepared by combining 1 lb salt, 4 oz sugar (sucrose), 0.12 oz potassium nitrate (KNO_3), and 0.12 oz sodium nitrite ($NaNO_2$). After 24 hr at 50 F the procedure was repeated, thus resulting in a total salt application of 1.5 oz/lb of catfish; 24 hr after the second rub fish were turned. Excess salt was rinsed from the fish with warm tap water after an additional 24 hr (3 days total salting). The fish were then weighed and smoked.

Experiment 3. Catfish examined in this experiment had been subjected to various killing procedures and to various conditions between death and brining. Treatments and conditions were: *(a)* death by electrical stunning, dressed, and brined (control); *(b)* death by suffocation, held in the round 12 hr at 32 F, dressed, and brined; *(c)* death by suffocation, dressed, held 3 months at 0 F, thawed at room temperature, and brined; *(d)* death by suffocation, held in the round 12 hr at 32 F, dressed, held 3 mo at 0 F, thawed at room temperature, and brined; and *(e)* death by electrical stunning, dressed, held 30 months at −40 F, thawed at room temperature, and brined. All catfish in these experiments were skinned. Brine was prepared by dissolving 3.0 lb sugar, 4.5 lb salt, 0.56 oz KNO_3, 0.56 oz $NaNO_2$ and 0.21 oz sodium ascorbate in 24.9 lb water. All catfish were weighed and submerged (with occasional agitation) in an equal weight of brine for 17.5 hr at 50 F before smoking.

Experiment 4. An investigation was made of the effect of brining time and fish size on quality of the finished smoked product was made. The brine formula, consisting of 2 lb sugar, 2 lb salt, and 0.12 oz potassium nitrite (KNO_2) in 16 lb water, was employed at a 1:1 (w/w) ratio with the catfish. Skinned fish had been frozen 3 months and thawed at room temperature. Brining times at 40 F were 4, 8, 16, and 24 hr. Two lots of catfish averaging 8.75 and 12.25 oz each were evaluated at each brining time.

Experiment 5. The effect of a wider range of catfish weight on finished product quality was further studied. Three lots of frozen, skinned catfish averaging approximately 7, 10.5, and 14 oz were thawed at room temperature. Fish were submerged at 40 F for 24 hr in an equal weight of brine formulated by dissolving 9 lb sugar, 9 lb salt, 0.23 oz KNO_3 and 0.23 oz $NaNO_2$ in 72 lb water. Fish were agitated occasionally during brining. Catfish were weighed before and after brining.

Smoking procedures

Temperatures and times of smoke-curing were varied to optimize smoking conditions with those of brining, thus re-

sulting in the most desirable product. Smoke was generated by using moist hickory wood chips in a thermostatically controlled smokehouse manufactured by Griffith Laboratories (Chicago, Illinois). Catfish temperatures referred to below are internal loin temperatures and were measured by inserting thermometers into the thickest portion of the loin muscle. Experiment numbers listed below for smoking correspond to brining experiments.

Experiments 1 and 2. Catfish were suspended by the tail from metal rods and placed in the smoking chamber at 120 F for 20 hr after which half of the skinned and unskinned fish were removed. The temperature of the chamber was then gradually increased to 205 F over a 2.5-hr period, bringing the temperature of the remaining catfish to 175 F.

Experiment 3. Catfish were placed in the smoking chamber at 90 F for 5 hr during which their internal temperature increased to 75 F. Chamber temperature was then maintained at 120 F for 7 hr resulting in a catfish temperature of 110 F. Finally the chamber temperature was brought to 195 F over a 2-hr period to bring the fish temperature to 175 F. Fish were removed after an additional 30 min.

Experiment 4. Catfish from various brining times were hung in the smoking chamber adjusted to 100 F. The chamber temperature was gradually increased to 200 F over a 7-hr period, resulting in a fish temperature of 175 F. Fish were removed after 30 min at 175 F.

Experiment 5. Three lots of catfish representing different weight groups were placed in the smoking chamber at 115 F. The internal flesh temperature gradually increased to 90 F during the first 3 hr; a gradual increase in chamber temperature to 170 F over the next 19 hr resulted in a flesh temperature of 140 F. The chamber was then increased to 192 F during the next 3 hr of smoking and maintained at this temperature for an additional 1.5 hr. Internal fish temperature increased from 140 F to 180 F in 4 hr during the time the chamber tempereture was increased to 192 F and remained at 180 F for 30 min before removal.

After smoking as outlined in Experiments 1 through 4, catfish were removed from the smoking chamber, packaged in polyethylene bags under atmospheric conditions, and held at 37 F until further evaluation. Smoked catfish from Experiment 5 were air-cooled at 37 F before packaging.

Analytical methods

Five-gram portions of homogenized fish fillets were dried at 160 F in a vacuum oven for 24 hr and the moisture content was determined by difference.

Fat determination was according to a method by Young *(11)*. The method consisted of drying 5-g samples of homogenized fish for 5 hr at 230 F, suspending the dried flesh in 10 ml diethyl ether overnight, removing 5 ml of clarified ether containing dissolved fat, evaporating the ether in a tared vessel, weighing, and calculating the percent fat originally contained in the flesh.

Methods outlined in the USDA *Chemistry Laboratory Guidebook (9)* were followed to determine the nitrite and NaCl contents of smoked fish.

Total aerobic plate counts were made from swabs of 10-cm^2 areas of the anterio-dorsal region of the fish. Appropriate dilutions of each swab were made in sterile 0.1% peptone and the organisms were recovered on Standard Methods agar (BBL) using the pour-plate technique. Incubation was at 21 C (69 F) and counts were made after 4 days.

Sensory evaluation

During preliminary tests to determine the most satisfactory brining and smoking conditions (Experiments 1 through 4), a six-member trained panel was requested to assign scores for appearance, color, aroma, texture, and flavor to the finished product. A 20-member untrained panel was later used to evaluate catfish judged to exhibit the most acceptable quality (Experiment 5). Samples of fish loin muscles were scored for aroma, texture, and flavor in a blind test against smoked chubs and haddock obtained from a local delicatessen. In all instances a nine-point hedonic scale (1 = extremely poor; 9 = excellent) was used. Data were subjected to Duncan's multiple range test *(4)*.

RESULTS AND DISCUSSION

Experiments 1 through 4 were exploratory in nature. Information derived from these experiments served to establish a basis for the recommended brining and smoking procedures included in Experiment 5. Rather than presenting details from Experiments 1 through 4, only those data having significant influence on the final recommended procedure for preparing smoked catfish are discussed.

In *Experiments 1 and 2,* a trained six-member panel judged skinned catfish to be more desirable than unskined catfish for smoking, regardless of whether the fish had been brined or dry-rubbed. Skinned catfish consistently exhibited a more attractive color and rated higher in desirable aroma and flavor characteristics. Skin undoubtedly acted as a barrier to the penetration of brine and curing salts and to smoke, thus resulting in a product with lower aroma and flavor. Upon refrigeration, fat deposits were noted between the skin and loin muscle. Such undersirable deposits apparently accumulated during the smoking process when the skin was separated from the loin muscle due to heat. The 2.5-hr extension in smoking time at higher 175 F resulted in a more desirable flavor and, more importantly, was judged as necessary for adequate pasteurization. Salt-rubbed catfish were generally as acceptable as the brined fish of *Experiment 1,* however, the method was not investigated further because of the increased time required in preparing the fish when compared to brining. Brown sugar was judged to have no advantage over sucrose in flavor development.

Experiment 3 yielded at least three important findings. First regardless of slaughtering conditions and time of storage up to 30 months in the frozen state before brining and smoking, finished products were virtually indistinguishable with respect to appearance, color, aroma, texture, and flavor. Secondly, slight to moderate muddy odor and flavor present in some catfish were observed to disappear or become masked during smoking. Fish judged slightly undesirable because of these qualities might therefore be utilized after smoking. Iredale and Rigby *(6)* have also noted this phenomenon in rainbow trout. A third observation was that both internal and external color were improved through the use of KNO_3 and $NaNO_2$ in

Table 1. Mean subjective sensory scores for smoked catfish brined 4, 8, 16 and 24 hr

Brining time (hr)	Appearance	Color	Aroma	Texture	Flavor
4	6.97[a]	7.40[a]	7.42[a]	7.37[a]	6.40[a]
8	7.38[b]	7.50[a]	7.72[a]	7.43[a]	6.80[a]
16	7.37[b]	7.50[a]	7.80[a]	7.90[b]	7.43[b]
24	7.40[b]	7.77[a]	7.62[a]	7.90[b]	7.90[b]

[a, b]Values in the same vertical column bearing the same letter are not significantly different ($P < 0.05$).

Table 2. Mean subjective sensory scores for smoked fishes

Fish	Aroma	Texture	Flavor	Average
Catfish	6.80	7.10	6.30	6.73
Haddock	6.25	5.95	7.10	6.43
Chub	6.70	6.20	5.65	6.18

the brine. Internal pink coloration was initially most intense in muscle nearest the back bone but diffused somewhat during storage. The external appearance was bright and was described as mahogany-gold in color.

Statistical analyses of data from *Experiment 4* showed no differences ($P < 0.05$) in appearance, aroma, color, texture, and flavor between the 8.75- and 12.25-oz average groups of smoked catfish. No significant differences were shown in appearance, aroma, color, or texture of catfish representing various size-brine time combinations when panels were conducted 1, 14, and 31 days after processing. There was no significant difference in flavor between catfish stored 14 and 31 days at 37 F, however both extended storage times showed improvement at a statistically significant level over freshly processed smoked catfish.

Results showing the effect of brining time (*Experiment 4*) on the finished product are in Table 1. The fish were evaluated by a trained panel after 14 days storage at 37 F. Appearance did not differ among 8-, 16- ad 24-hr brines but all were statistically different from the 4-hr brined fish. The 4-hr catfish were somewhat dull and less acceptable than those brined longer. Color and aroma were not shown to be different by the statistical analyses employed. Analysis of ratings for texture showed that there were no significant differences between 4- and 8-hr brines nor were there any differences between 16- and 24-hr brines, although the 4- and 8-hr brines differed and were inferior to the 16- and 24-hr brines. The same pattern was established for flavor. Therefore, based on the analyses of appearance, texture, and flavor it was concluded that 16- and 24-hr brines were superior in producing smoked catfish. Since appearance and flavor scores were numerically greater for catfish brined 24 hr, the longer brining time was judged more desirable.

The brine formula and smoking conditions employed in *Experiment 5* were judged as optimal, based on all previous experiments, for producing smoked catfish. Tested in this experiment was the effect of a wider range of fish sizes (weight) on final quality. Catfish in the 7-oz average group were judged as too salty and showed a 37.5% loss in weight caused by processing. The 10.5- and 14-oz groups were rated as having superior flavor and averaged approximately 28% weight loss during processing. The 10.5-oz size is recommended because it is easily handled during smoking. Heavier fish tended to detach from their tails during smoking and fall from the racks and could therefore result in significant economic loss on a commercial basis. Larger catfish should be attached to the rod in the smoking chamber by a hook through the belly flap rather than by the tail.

Smoked catfish, having been produced in accordance with the brining and smoking procedures described in *Experiment 5* were then evaluated by a 20-member untrained panel, 16 of whom had never sampled smoked fish. The panel evaluation was performed after 1 week of storage at 37 F. Portions of loin muscle from haddock and chubs were also

Table 3. Proximate analyses of smoked and raw fishes

Fish	Time of brine (hr)	Wt before brine (oz)	Wt loss[a] (%)	Moisture[b] (%)	Fat[b] (%)	NaCl[b] (%)	$NaNO_2$[b] (ppm)	Edible wt[a] (%)
Catfish	0	12.3	—[c]	69.5	12.0	TR[d]	0	—
	24	11.5	23.0	58.2	13.1	2.89	39	77.5
		10.2	27.9	58.8	12.3	3.26	44	79.9
Chub	—	—	—	71.0	9.7	2.85	162	59.0
Haddock	—	—	—	59.2	20.9	2.34	TR	—

[a]Calculations are on a wet weight basis, entire smoked carcass.

[b]Calculations are on a wet weight basis, edible flesh; all analyses were on smoked fish with the exception of catfish with 0 hr brine (raw fish).

[c]No test performed.

[d]Trace

included in the blind panel, the results of which are presented in Table 2. Mean scores are shown for aroma, texture, and flavor while average values reflect overall sensory ratings for each fish. Statistical analyses showed no significant differences at the 95% confidence level for any of the sensory characteristics. This was, in part, due to the wide range in ratings obtained from the 20-member panel. Although each of the fish species tested by the panel have natural differences, they also exhibit similarities. There was no attempt to produce a smoked catfish indistinguishable from other smoked fishes. However, we assumed the haddock and chub to be of excellent quality and are therefore categorizing the catfish as having like quality based on the data obtained. In other words, although scores for all smoked fishes shown in Table 2 are acceptable, they undoubtedly would have been higher had the panel been composed entirely of trained members.

Proximate analyses for 24-hr brine catfish from *Experiment 5*, in addition to data for raw catfish, chubs, and haddock are shown in Table 3. Catfish have several competitive advantages over chubs when smoked. The process yield for catfish is approximately 75% (calculated by difference from weight loss data); a similar figure was given by Bratzler and Robinson *(2)* for processed Great Lakes chubs. An even higher process yield for catfish might be achieved through the use of elevated relative humidities in the smoking chamber. Because the head and skin (in addition to viscera) are removed from catfish before brining and smoking, 78% of the resulting product is edible compared to 58% for chubs (Table 3). In addition to supplying 34% more edible flesh than chubs on a purchase-weight basis, the catfish flesh is extremely easy to remove from the bone structure. Fine bones radiating from the backbone of chubs and sometimes inhibiting complete removal of the loin muscle, present no problem in catfish.

Qualities exhibited by the finished product were dependent on the brining formula, temperature and time, ratio of brine to fish, size of fish, smoking temperatures, time of smoking, and storage time after processing. Many of these factors have been also noted to affect quality of smoked chubs *(10)*. Altering any of the recommended procedures may therefore result in smoked catfish exhibiting different characteristics. A processor may be unable to duplicate the product described here, especially if his smokehouse is designed differently *(1)*. It should be noted that, although smoking chamber temperatures and times are given in various experiments above, the time of smoking at specific internal loin muscle temperatures is most important. Necessary modifications in the process should not be extensive.

Procedures outlined in *Experiment 5* to produce smoked catfish are judged as adequate with regard to quality control. No aerobic bacteria were recovered from the smoked product after 31 days storage at 37 F. Although good manufacturing practices and current FDA regulations stipulate levels of $NaNO_2$ only for smoked chubs *(5)*, the sodium chloride content in the water phase of catfish processed in accordance with procedures described in *Experiment 5* approximates 4.8% when calculated by the following formula:

$$\% \text{ NaCl} = \frac{\text{g NaCl in sample} \times 100}{\text{g NaCl} + \text{g } H_2O \text{ in sample}}$$

This satisfies the minimum water phase NaCl for hot-processed smoked fish which has been set at 3.5%.

Catfish is a popular menu item in Southeastern U.S., but it is usually prepared by deepfat or pan frying or by broiling or baking. Smoked country-cured pork products are also highly acceptable. It should be stressed to those unfamiliar with smoked fish, however, that handling and storage of smoked catfish is considerably different from, say, country-cured ham. It is suggested that regulations set forth in the City of Milwaukee Ordinance for smoked fish and smoked fish products *(3)* established in 1964 and current FDA regulations *(5)* should be applied to smoked catfish processing and distribution. Pace and Krumbiegel *(7)* recently summarized the Milwaukee ordinance: *(a)* smoking shall consist of heating every portion of every fish to a minimum temperature of 180 F (82.2 C) for a minimum of 30 min, *(b)* fish shall be removed from the smoking chamber to a separate room for prompt cooling, and must be packaged within 2 hr after completion of smoking, *(c)* the packages shall bear the words "Perishable–Keep Refrigerated," *(d)* the package shall not be sealed so that exchange of air is prevented, *(e)* the package shall bear the processing date and expiration date, *(f)* every smoked fish shall be maintained at a refrigeration temperature of no more than 40 F (4.5 C) from the time of packaging, during transportation, and during display, and *(g)* the expiration date for smoked fish shall be not more than 7 calendar days following the date of smoking. We recommend that the maximum refrigeration temperature under point *(f)* be reduced to 37 F (2.8 C). Adherance to these regulations should preclude the possibility of growth and toxin production by *Clostridium botulinum* in addition to preserving the organoleptic quality of smoked catfish.

ACKNOWLEDGMENTS

We are grateful to Skidaway Institute of Oceanography, Savannah, Ga. for supplying some of the channel catfish used in this study. Technical assistance from L. Bledsoe, N. Perry,

and B. Vaughn and assistance with statistical analyses from D. Parvin and J. C. Elrod, Agricultural Economics Department, is appreciated.

References

1. Berg, I. I. 1971. Smokehouses and the smoke curing of fish. Washington State Dept. of Fisheries, Olympia.

2. Bratzler, L. J. and M. O. Robinson. 1967. Processing Great Lakes chub (*Leucichthys hoyi*). Quart. Bull. Mich. Agr. Expt. Sta. 49:440-444.

3. City of Milwaukee, an Ordinance No. 735, pt. 1, sec. 70-55 through 70-71 of the Milwaukee Code: Smoked fish and smoked fish products, March 17, 1964. City Hall, Milwaukee, Wisconsin 53201.

4. Duncan, D. B. 1955. New multiple range and multiple F tests. Biometrics 11:1-42.

5. Federal Register. 1970. Part 128a. Fish and seafood products. Subpart A. Smoked and smoke-flavored fish. 35: 17401-17402.

6. Iredale, D. G., and D. Rigby. 1972. Effect of smoke-processing on muddy odor and taste in rainbow trout (*Salmo gairdneri*). J. Fish. Res. Bd. Can. 29:1365-1366.

7. Pace, P. J., and E. R. Krumbiegel. 1973. *Clostridium botulinum* and smoked fish production: 1963-1972. J. Milk Food Technol. 36:42-49.

8. Tiemeier, O. W. 1969. Personal communication. Kansas State University, Manhattan.

9. U.S.D.A. 1971. Chemistry laboratory guidebook. CMS, Laboratory Services Div., Washington, D. C.

10. Weckel, K. G., and D. Wosje. 1966. Brine salting of Great Lakes chub (*Leucichthys hoyi*) for smoking. Res. Rept. 24, Exp. Station, Coll. Agr., University of Wisconsin, Madison.

11. Young, C. T. 1973. Unpublished data. University of Georgia, Experiment.

FOOD PRESERVATIVES APPEAR SAFE

Two food preservatives that have been under suspicion have come through tests looking beneficial in several respects involving cholesterol and fat digestion. In fact, one preservative may have medical use for lowering high blood cholesterol levels in humans.

The preservatives are BHA and BHT (butylated hydroxyanisole and butylated hydroxytoluene). They are used in many food products to prevent oxidation of fats which can cause stale flavors to develop during storage. Rat studies had suggested that these materials increase cholesterol and change lipid phosphrus levels of the blood. Their continued use as food preservatives was questioned.

University of Wisconsin food scientist A. L. Branen recently conducted research with these materials fed to young monkeys. Monkeys' digestive systems are more comparable to man's than are the digestive systems of rats. He fed both materials in a diet high in corn oil. Corn oil is a fat reputed to give fewer cholesterol problems than other fats. He traced the levels of various lipids (products of fat digestion and production) and cholesterol in the monkeys' blood and livers. The formation of fatty livers is a health problem associated with fat metablism.

Lipid levels changed in all the treatments, Branen found. But the potentially harmful changes seemed due to the corn oil—not to the BHA or BHT. In fact, high levels of BHA (500 mg. per kg.) seem to prevent many of the changes produced by corn oil. BHT prevented some change, but generally not as well as BHA . This agrees with previous findings by other researchers, Branen points out. It may be that the preservatives prevent oxidation of lipids or interfere with lipid production in some way.

Since BHT at a relatively low level (50 mg. per kg. daily) reduced serum cholesterol, it may have medical use for treating patients with high cholesterol levels, Branen states.

Branen also is studying the role of BHA in preventing mold poisoning in foods. Various Aspergillus organisms can produce aflatoxins. This is related to lipid synthesis by the molds. BHA interferes with lipid synthesis. It appears that 250 parts per million of BHA inhibits both growth and toxin production by such molds, so it acts as a mold preventer as well as an antioxidant.

HOLDERS OF 3-A SYMBOL COUNCIL AUTHORIZATIONS ON AUGUST 20, 1973

"Questions or statements concerning any of the holders of authorizations listed below, or the equipment fabricated, should be addressed to Earl O. Wright, Sec'y-Treas., Dept. of Food Technology, Dairy Industry Bldg., Iowa State University, Ames, Iowa 50010."

0104 Storage Tanks for Milk and Milk Products As Amended

116 Jacob Brenner Company, Inc. (10/ 8/59)
450 Arlington, Fond du Lac, Wisconsin 54935

28 Cherry-Burrell Corporation (10/ 3/56)
575 E. Mill St., Little Falls, N. Y. 13365

102 Chester-Jensen Company, Inc. (6/ 6/58)
5th & Tilgham Streets, Chester, Pennsylvania 19013

1 Chicago Stainless Equipment (5/ 1/56)
555 Valley Way, Northbrook, Illinois 60062

2 CREPACO, Inc. (5/ 1/56)
100 C. P. Ave., Lake Mills, Wisconsin 53551

117 Dairy Craft, Inc. (10/28/59)
St. Cloud Industrial Park
St. Cloud, Minn. 56301

76 Damrow Company (10/31/57)
196 Western Avenue, Fond du Lac, Wisconsin 54935

115 DeLaval Company, Ltd. (9/28/59)
113 Park Street, So., Peterborough, Ont., Canada

109 Girton Manufacturing Company (9/30/58)
Millville, Pennsylvania 17846

114 C. E. Howard Corporation (9/21/59)
9001 Rayo Avenue, South Gate, California 90280

127 Paul Mueller Company (6/29/60)
P. O. Box 828, Springfield, Missouri 65801

197 Paul Mueller (Canada), Ltd. (9/ 9/67)
84 Wellington St., South, St. Marys, Ont., Canada

233 Stainless Steel Craft Corporation (4/13/72)
4503 Alger St., Los Angeles, Calif. 90039

21 Technova, Inc. Gosselin Division (9/20/56)
1450 Hebert c. p. 758
Drummondville, Quebec, Canada

31 Walker Stainless Equipment Co. (10/ 4/56)
Elroy, Wisconsin 53929

0203 Pumps for Milk and Milk Products as Amended

214R Ben H. Anderson Manufacturers (5/20/70)
Morrisonville, Wis. 53571

212R Babson Bros. Co. (2/20/70)
2100 S. York Rd., Oak Brook, Ill. 60621

29R Cherry-Burrell Corporation (10/ 3/56)
2400 Sixth St., S. W., Cedar Rapids, Iowa 52406

63R CREPACO, Inc. (4/29/57)
100 C. P. Ave., Lake Mills, Wisconsin 53551

205R Dairy Equipment Co. (5/22/69)
1919 So. Stoughton Road, Madison, Wis. 53716

180R The DeLaval Separator Co. (5/ 5/66)
Duchess Turnpike, Poughkeepsie, N. Y. 12602

65R G & H Products, Inc. (5/22/57)
5718 52nd Street, Kenosha, Wisconsin 53140

145R ITT Jabsco, Incorporated (11/20/63)
1485 Dale Way, Costa Mesa, Calif. 92626

26R Ladish Co., Tri-Clover Division (9/29/56)
9201 Wilmot Road, Kenosha, Wisconsin 53140

236 Megator Corporation (5/ 2/72)
125 Gamma Drive, Pittsburgh, Pa. 15238

241 Purity S.A. (9/12/72)
Alfredo Noble #38, Industrial fte. deVigas
Tlalnepantla, Mexico

148R Robbins & Myers, Inc. (4/22/64)
Moyno Pump Division
1345 Lagonda Ave., Springfield, Ohio 45501

163R Sta-Rite Industries, Inc. (5/ 5/65)
P. O. Box 622, Delavan, Wisconsin 53115

72R L. C. Thomsen & Sons, Inc. (8/15/57)
1303 53rd Street, Kenosha, Wisconsin 53140

219 Tri-Canada Cherry-Burrell Ltd. (2/15/71)
6500 Northwest Drive, Mississauga, Ont., Canada L4V 1K4

175R Universal Milking Machine Div. (10/26/65)
National Cooperatives, Inc.
First Avenue at College, Albert Lea, Minn. 56007

52R Viking Pump Div.
Houdaille Industries, Inc. (12/31/56)
406 State Street, Cedar Falls, Iowa 50613

5R Waukesha Foundry Company (7/ 6/56)
Waukesha, Wisconsin 53186

0403 Homogenizers and High Pressure Pumps of the Plunger Type, As Amended

247 Bran and Lubbe, Inc. (4/14/73)
2508 Gross Point Road, Evanston, Illinois 60201

87 Cherry-Burrell Corporation (12/20/57)
2400 Sixth Street, S. W., Cedar Rapids, Iowa 52404

37 CREPACO, Inc. (10/19/56)
100 C.P. Ave, Lake Mills, Wis. 53538

75 Gaulin, Inc. (9 /26/57)
44 Garden Street, Everett, Massachusetts 02149

237 Graco Inc. (6/ 3/72)
60-Eleventh Ave., N.E., Minneapolis, Minn. 55413

0511 Stainless Steel Automotive Milk Transportation Tanks for Bulk Delivery and/or Farm Pick-up Service, As Amended

131R Almont Welding Works, Inc. (9/ 3/60)
4091 Van Dyke Road, Almont, Michigan 48003

98R Beseler Steel Products, Inc. (3/24/58)
417 East 29th, Marshfield, Wisconsin 54449

70R Jacob Brenner Company (8/ 5/57)
450 Arlington, Fond du Lac, Wisconsin 54935

40 Butler Manufacturing Co. (10/20/56)
900 Sixth Ave., S. E., Minneapolis, Minn. 55114

118 Dairy Craft, Inc. (10/28/59)
St. Cloud Industrial Park
St. Cloud, Minn. 56301

66 Dairy Equipment Company (5/29/57)
1818 So. Stoughton Road, Madison, Wisconsin 53716

123 DeLaval Company, Ltd. (12/31/59)
113 Park Street, South Peterborough, Ont., Canada

45 The Heil Company (10/26/56)
3000 W. Montana Street, Milwaukee, Wisconsin 53235

201 Paul Krohnert Mfg., Ltd. (4/ 1/68)
811 Steeles Ave., West Hill, Ontario, Canada L9T 2Y3

232 Litewate Transport Eqiupment (4/ 4/72)
Division of the Stanray Corp.
4220 South 13th Street, Milwaukee, Wis. 53221

80 Paul Mueller (Canada), Ltd. (11/24/57)
84 Wellington Street, So., St. Marys, Ont., Canada
85 Polar Manufacturing Company (12/20/57)
Holdingford, Minn. 56340
144 Portersville Equipment Company (5/16/63)
Portersville, Pennsylvania 16051
71 Progress Industries, Inc. (8/ 8/57)
400 E. Progress Street, Arthur, Illinois 61911
121 Technova Inc. Gosselin Division (12/ 9/59)
1450 Hebert c. p. 758
Drummondville, Quebec, Canada
47 Trailmobile, Div. of Pullman, Inc. (11/ 2/56)
701 East 16th Ave., North Kansas City, Mo. 64116
189 A. & L. Tougas, Ltée (10/ 3/66)
1 Tougas St., Iberville, Quebec, Canada
25 Walker Stainless Equipment Co. (9/28/56)
New Lisbon, Wisconsin 53950

0809 Fittings Used on Milk and Milk Products Equipment, and Used on Sanitary Lines Conducting Milk and Milk Products

79R Alloy Products Corporation (11/23/57)
1045 Perkins Avenue, Waukesha, Wisconsin 53186
138R A.P.V. (Canada) Equipment, Ltd. (12/17/62)
103 Rivalda Rd., Weston, Ont., Canada
245 Babson Brothers Company (2/12/73)
2100 S. York Road, Oak Brook, Illinois 60521
82R Cherry-Burrell Corporation (12/11/57)
2400 Sixth Street, S.W. Cedar Rapids, Iowa 52406
124R DeLaval Company, Ltd. (2/18/60)
113 Park Street, South Peterborough, Ont., Canada
184R The DeLaval Separator Co. (8/ 9/66)
Duchess Turnpike, Poughkeepsie, N. Y. 12602
67R G & H Products, Inc. (6/10/57)
5718 52nd Street, Kenosha, Wisconsin 53140
199R Graco, Inc. (12/ 8/67)
60 Eleventh Ave., N.E., Minneapolis, Minn. 55413
203R Grinnell Company (11/ 7/68)
260 W. Exchange St., Providence, R. I. 02901
218 Highland Equipment Corporation (2/12/71)
74-10 88th St., Glendale, N.Y. 11227
204R Hills McCanna Company (2/10/69)
400 Maple Ave., Carpentersville, Ill. 60110
34R Ladish Co., Tri-Clover Division (10/15/56)
2809 60th St., Kenosha, Wisconsin 53140
239 LUMACO (6/30/72)
Box 688, Teaneck, N. J. 07666
230 ITT Moreland Products, Inc. (3/27/72)
P.O. Box #34, Wrightsville, Pa. 17638
200R Paul Mueller Co. (3/ 5/68)
P. O. Box 828, Springfield, Mo. 65801
244 Pittsburgh Brass Manufacturing Co. (2/10/73)
P.O. Box 387-A, Rd. #6, Irwin, Pa. 15642
149R Q Controls (5/18/64)
Occidental, California 95465
227 Stainless Steel Craft Corporation (1/11/72)
4503 Alger Street, Los Angeles, California
89R Sta-Rite Industries, Inc. (12/23/68)
P. O. Box 622, Delavan, Wis. 53115
73R L. C. Thomsen & Sons, Inc. (8/31/57)
1303 43rd Street, Kenosha, Wisconsin 53140
191R Tri-Canada Cherry-Burrell, Ltd. (11/23/66)
6500 Northwest Drive, Mississauga, Ontario, Canada
L4V 1K4
250 Universal Milking Machine Division (6/11/73)
Universal Coopratives, Inc.
408 First Ave. So.
Albert Lea, Mn. 56007
86R Waukesha Specialty Company, Inc. (12/20/57)
Darien, Wisconsin 53114

0900 Thermometer Fittings and Connections Used on Milk and Milk Products Equipment and Supplement 1, As Amended

32 Taylor Instrument Process Control,
Div. Sybron Corp. (10/ 4/56)
95 Ames Street, Rochester, New York 14601
206 The Foxboro Company (8/11/69)
Neponset Ave., Foxboro, Mass. 02035
246 United Electric Controls (3/24/73)
85 School Street, Watertown, Massachusetts 02172

1000 Milk and Milk Products Filters Using Disposable Filter Media, As Amended

35 Ladish Co., Tri-Clover Division (10/15/56)
2809 60th Street, Kenosha, Wisconsin 53140

1103 Plate-Type Heat Exchangers for Milk and Milk Products, As Amended

20 A.P.V. Company, Inc. (9/ 4/56)
137 Arthur Street, Buffalo, New York 14207
30 Cherry-Burrell Corporation (10/ 1/56)
2400 Sixth Street, S.W., Cedar Rapids, Iowa 52404
14 Chester-Jensen Co., Inc. (8/15/56)
5th & Tilgham Streets, Chester, Pennsylvania 19013
38 CREPACO, Inc. (10/19/56)
100 CP Avenue, Lake Mills, Wisconsin 53551
120 DeLaval Company, Ltd. (12/ 3/59)
113 Park Street, South Peterborough, Ont., Canada
17 The DeLaval Separator Company (8/30/56)
Duchess Turnpike, Poughkeepsie, N. Y. 12602
15 Kusel Dairy Equipment Company (8/15/56)
100 W. Milwaukee Street, Watertown, Wisconsin 53094

1204 Internal Return Tubular Heat Exchangers, for Milk and Milk Products, As Amended

248 Allegheny Bradford Corporation (4/16/73)
P. O. Box 264, Bradford, Pa. 16701
243 Babson Brothers Company (10/31/72)
2100 S. York Road, Oak Brook, Illinois 60521
103 Chester-Jensen Company, Inc. (6/ 6/58)
5th & Tilgham Street, Chester, Pennsylvania 19013
152 The DeLaval Separator Co. (11/18/69)
350 Duchess Turnpike, Poughkeepsie, N. Y. 12602
217 Girton Manufacturing Co. (1/23/71)
Millville, Pa. 17846
238 Paul Mueller Company (6/28/72)
P. O. Box 828, Springfield, Missouri 65801
96 C. E. Rogers Company (3/31/64)
P. O. Box 118, Mora, Minnesota 55051
225 Sanitary Processing Equipment Corporation (11/24/71)
Butternut Drive, East Syracuse, N.Y.

1301 Farm Milk Cooling and Holding Tanks, As Amended

240 Babson Brothers Company (9/ 5/72)
2100 S. York Road, Oak Brook, Illinois 60521

11R CREPACO, Inc. (7/25/56)
100 C. P. Ave.
Lake Mills, Wisconsin 53551

119R Dairy Craft, Inc. (10/28/59)
St. Cloud Industrial Park, St. Cloud, Minn. 56301

4R Dairy Equipment Company (6/15/56)
1919 S. Stoughton Road, Madison, Wisconsin 53716

92R DeLaval Company, Ltd. (12/27/57)
113 Park Street, South Peterborough, Ontario, Canada

49R The DeLaval Separator Company (12/ 5/58)
Duchess Turnpike, Poughkeepsie, N. Y. 12602

10R Girton Manufacturing Company (7/25/56)
Millville, Pennsylvania 17846

95R Globe Fabricators, Inc. (3/14/58)
3350 North Gilman Rd., El Monte, California 91732

179R Heavy Duty Products (Preston), Ltd. (3/ 8/66)
1261 Industrial Road, Preston, Ont., Canada

12R Paul Mueller Company (7/31/56)
P. O. Box 828, Springfield, Missouri 65801

58R Schweitzer's Metal Fabricators, Inc. (2/25/57)
806 No. Todd Avenue, Azusa, California 91702

235 Stainless Steel Craft Corporation (4/13/72)
4503 Alger St., Los Angeles, California 90039

249 Sunset Equipment Co. (4/16/73)
3765 North Dunlap Street
St. Paul, Minnesota 55112

216R Valco Manufacturing Company (10/22/70)
3470 Randolph St., Huntington Pk., Calif. 90256

42R VanVetter, Inc. (10/22/56)
2130 Harbor Avenue S.W., Seattle, Washington 98126

170R The W. C. Wood Co., Ltd. (8/ 9/65)
5 Arthur Street, South, Box 750, Guelph, Ont., Canada

16R Zero Manufacturing Company (8/27/56)
Washington, Missouri 63090

1400 Inlet and Outlet Leak Protector Plug Valves for Batch Pasteurizers, As Amended

122R Cherry-Burrell Corporation (12/11/59)
2400 Sixth St., S. W., Cedar Rapids, Iowa 52406

69 G & H Products Corporation (6/10/57)
5718 52nd Street, Kenosha, Wisconsin 53140

27 Ladish Co. - Tri-Clover Division (9/29/56)
2809 60th Street, Kenosha, Wisconsin 53140

78 L. C. Thomsen & Sons, Inc. (11/20/57)
1303 43rd Street, Kenosha, Wisconsin 53140

1604 Evaporators and Vacuum Pans for Milk and Milk Products

132R A.P.V. Company, Inc. (10/26/60)
137 Arthur Street, Buffalo, New York 14207

111R Blaw-Knox Food & Chemical Equip., Inc. (2/12/59)
P. O. Box 1041
Buffalo, N. Y. 14240

164R Anderson IBEC (4/25/65)
19609 Progress Drive
Strongsville, Ohio 44136

107R C. E. Rogers Company (8/ 1/58)
P. O. Box 118, Mora, Minnesota 55051

186R Marriott Walker Corporation (9/ 6/66)
925 East Maple Road, Birmingham, Mich. 48010

1700 Fillers and Sealers of Single Service Containers, For Milk and Milk Products, As Amended

192 Cherry-Burrell Corporation (1/ 3/67)
2400 Sixth St., S. W., Cedar Rapids, Iowa 52404

137 Ex-Cell-O Corporation (10/17/62)
P. O. Box 386, Detroit, Michigan 48232

220 Haskon, Inc., Package Equipment Division (4/24/71)
2285 University Ave., St. Paul, Minnesota 55114

211 Twinpak, Inc. (2/ 4/70)
2225 Hymus Blvd., Dorval 740, P.Q.

1901 Batch and Continuous Freezers, For Ice Cream, Ices and Similarly Frozen Dairy Foods, As Amended

141 CREPACO, Inc. (4/15/63)
100 C. P. Avenue, Lake Mills, Wisconsin 53551

146 Cherry-Burrell Corporation (12/10/63)
2400 Sixth Street, S. W., Cedar Rapids, Iowa 52404

2201 Silo-Type Storage Tanks for Milk and Milk Products

168 Cherry-Burrell Corporation (6/16/65)
575 E. Mill St., Little Falls, N. Y. 13365

154 CREPACO, Inc. (2/10/65)
100 C. P. Ave., Lake Mills, Wisconsin 53551

160 Dairy Craft, Inc. (4/ 5/65)
St. Cloud Industrial Park
St. Cloud, Minn. 56301

181 Damrow Company, Division of DEC International, Inc. (5/18/66)
196 Western Ave., Fond du Lac, Wisconsin 54935

156 C. E. Howard Corporation (3/ 9/65)
9001 Rayo Avenue, South Gate, California 90280

155 Paul Mueller Co. (2/10/65)
P. O. Box 828, Springfield, Missouri 65801

195 Paul Mueller (Canada), Ltd. (7/ 6/67)
84 Wellington St., So., St. Marys, Ont., Canada

234 Stainless Steel Craft Corporation (4/13/72)
4503 Alger St., Los Angeles, California 90039

165 Walker Stainless Equipment Co. (4/26/65)
Elroy, Wisconsin 53929

2300 Equipment for Packaging Frozen Desserts, Cottage Cheese and Milk Products Similar to Cottage Cheese in Single Service Containers

174 Anderson Bros. Mfg. Co. (9/28/35)
1303 Samuelson Road, Rockford, Illinois 61109

209 Doboy Packaging Machinery (7/23/69)
Domain Industries, Inc.
869 S. Knowles Ave., New Richmond, Wis. 54017

222 Maryland Cup Corporation (11/15/71)
Owings Mills, Maryland 21117

193 Triangle Package Machinery Co. (1/31/67)
6655 West Diversey Ave., Chicago, Illinois 60635

2400 Non-Coil Type Batch Pasteurizers

161 Cherry-Burrell Corporation (4/ 5/65)
575 E. Mill St., Little Falls, N. Y. 13365

158 CREPACO, Inc. (3/24/65)
100 C. P. Avenue, Lake Mills, Wisconsin 53551

187 Dairy Craft, Inc. (9/26/66)
St. Cloud Industrial Park
St. Cloud, Minn. 56301

177 Girton Manufacturing Co. (2/18/66)
Millville, Pennsylvania 17846

166 Paul Mueller Co. (4/26/65)
P. O. Box 828, Springfield, Mo. 65601

2500 Non-Coil Type Batch Processors for Milk and Milk Products

162 Cherry-Burrell Corporation (4/ 5/65)
575 E. Mill St., Little Falls, N. Y. 13365

159 CREPACO, Inc. (3/24/65)
100 C. P. Avenue, Lake Mills, Wisconsin 53551

188 Dairy Craft, Inc. (9/26/66)
St. Cloud Industrial Park
St. Cloud, Minn. 56301

167 Paul Mueller Co. (4/26/65)
Box 828, Springfield, Mo. 65801

196 Paul Mueller (Canada), Ltd. (7/ 6/67)
84 Wellington St., So., St. Marys, Ont., Canada

202 Walker Stainless Equipment Co. (9/24/68)
New Lisbon, Wis. 53950

2600 Sifters for Dry Milk and Dry Milk Products

228 J. H. Day Co. (2/28/72)
4932 Beech Street, Cincinnati, Ohio 45202

171 Entoleter, Inc. (9/ 1/65)
Subsidiary of American Mfg. Co.
P. O. Box 1919, New Haven, Connecticut 06510

229 Russell Finex Inc. (3/15/72)
156 W. Sandford Boulevard, Mt. Vernon, N. Y. 10550

173 B. F. Gump Division (9/20/65)
Blaw-Knox Food & Chem. Equip. Inc.
750 E. Ferry St., P. O. Box 1041
Buffalo, New York 14240

185 The Orville-Simpson Co. (8/10/66)
1230 Knowlton St., Cincinnati, Ohio 45223

176 Sprout, Waldron & Co., Inc. (1/ 4/66)
Munsy, Pennsylvania 17756

172 SWECO, Inc. (9/ 1/65)
6111 E. Bandini Blvd., Los Angeles, California 90022

2800 Flow Meters for Milk and Liquid Milk Products

223 C-E IN-VAL-CO, a division of Combustion Engineering, Inc. (11/15/71)
P. O. Box 556, 3102 Charles Page Blvd., Tulsa, Oklahoma 74101

231 The DeLaval Separator Company (3/27/72)
350 Dutchess Turnpike
Poughkeepsie, New York 12603

226 Fischer & Porter Company (12/ 9/71)
County Line Road, Warminster, Pa. 18974

224 The Foxboro Company (11/16/71)
Foxboro, Massachusetts 02035

NEW LIQUID WASTE TREATMENT PROCESS PURIFIES HIGH PROTEIN WASTEWATER

A patented new chemical liquid waste treatment process, designed specifically to meet the needs of processing industries with wastewater effluents high in protein content, has been developed, it was revealed here today. The new process, called ENVITROL(R), is especially suited to treating waste streams used by plants which process fish, chicken, cattle and hogs.

The Envitrol Corporation, developers of the process, and a subsibiary of Benham-Blair & Affiliates, Inc., Architects-Engineers, reports on-site pilot-plant tests at meat processing plants have shown ENVITROL(R) capable of: (1) reducing BOD content by 80-90 percent; (2) reducing COD content by 80 percent; (3) recovering 90 percent or more of greases and oils, and (4) recovering almost 100 percent of solids, thus producing an essentially clear water effluent. These combined levels could, in some instances, eliminate sewer surcharges and, at the same time, benefit overloaded municipal sewage treatment plants which treat industrial discharges.

William J. Conner, Jr., marketing director for ENVITROL(R), pointed out that the process effects other major economic, as well as environmental, benefits.

"ENVITROL(R)," he said, "greatly increases the the amount of waste material flocculated while accelerating the flocculation rate. This increase in the amount and speed of flocculation results in a higher-than-average degree of suspended solids separation, enabling more efficient solids removal by flotation.

"In turn," Conner added, "this permits a higher recovery rate of saleable protein-rich volatile solids and greases and oils, which can be converted into industrial fats, oils, fertilizers or animal feed to produce additional income for the processing plants."

For more information, write Envitrol Corporation, 6323 N. Grand Boulevard, Oklahoma City, Oklahoma 73118.

A TRUNCATED SEQUENTIAL PROCEDURE FOR DETERMINING SOMATIC CELL COUNT OF MILK BY THE STRIP METHOD[1]

ROY E. GINN, V. S. PACKARD, JR.,[2] AND R. D. COOK
Dairy Quality Control Institute, Inc., St. Paul, Minnesota 55113 and Department of Food Science and Nutrition and Department of Applied Statistics, University of Minnesota, St Paul. Minnesota 55101

(Received for publication April 18, 1973)

ABSTRACT

The Direct Microscopic Somatic Cell Count (DMSCC) "strip" method requires the counting of four strips on two separate milk films. An evaluation of five technicians and 231 milk samples, using a truncated sequential procedure for making cell counts, indicated the potential for significantly reducing the number of strips counted, with little loss in validity of results. For conditions observed in this market, counting of cells could be terminated after the first strip in 85% of the instances with a somatic cell standard set at 1.5 million or higher. Four strips would have to be counted in only 4% of the samples. With the standard set at 1 million or higher, these market conditions would permit one-strip counting 68% of the time, and would necessitate four-strip counting in only 20% of the samples.

The Direct Microscopic Somatic Cell Count (DMSCC) is used as a confirmatory procedure in evaluating milk supplies under the Abnormal Milk Control Program (AMCP) of the U. S. Public Health Service. The method requires that two milk films be prepared and two strips, one horizontal and one vertical, be counted on each film. Because labor costs in the laboratory account for much of the cost of AMCP, procedures for reducing such costs should clearly receive attention.

Since the work of Prescott and Breed *(4)*, considerable research has been done in development and evaluation of screening and confirmatory methods for abnormal milk. The single stain modification of Levowitz and Weber *(2)* was an important step toward reducing the time and complexity of milk film staining. Other innovations, including specially designed glass slides and eyepiece reticles have also been introduced *(3, 5)*. Smith *(8)* found a within-film variance of 130% and a film component variance of 15% of the mean. Schultze et al. *(7)* in a utility-cost study of a number of screening tests assigned the highest rating to the Milk Gel Index, followed by the Wisconsin Mastitis Test when either 1.5 or 1.0 million somatic cell count levels were used as the standard. Also, Schultze et al. *(6)* have developed a statistical procedure, when using the DMSCC as a screening test, for assigning samples to certain categories of test completion depending upon results of a single strip count. Going one step further, and using a slightly different statistical approach, the work reported herein is designed to show the feasibility of count discontinuance after each strip counted when the DMSCC is used as a confirmatory method. As stated previously, current requirements necessitate the counting of four strips, two each on duplicate films.

MATERIALS AND METHODS

During regular testing at the Dairy Quality Control Institute, Inc. laboratories, 10% of those samples showing highest Wisconsin Mastitis Test (WMT) readings (down to but not including a value of 21 mm) are confirmed for somatic cell count by the DMSCC as allowed under the AMCP. For this study, 231 samples were subjected to confirmatory testing. The work was divided among five technicians, each of whom was responsible for preparing and counting her own milk films. No attempt was made to divide the number of samples equally among technicians. Two technicians did 78 tests each, the others, 25, 29, and 19, respectively.

Film preparation and staining were done as described in *Standard Methods for the Examination of Dairy Products (9)*. As recommended by the Subcommittee on Screening Tests, National Mastitis Council *(3)*, glass slides with a clear, circular area of 1 cm^2 permanently outlined on the surface were used. Two films were prepared, and two strips, one horizontal and one vertical, were counted on each film. All technicians used the same microscope and eyepiece. The strip factor was 15,400.

RESULTS AND DISCUSSION

In the discussion that follows, a procedure is presented which provides for discontinuing the cell count, depending upon the results obtained, after the counting of each individual strip. Technically, the procedure can be described as a truncated sequential test, and two assumptions are necessary for its validation: (*a*) that the counts (x_i) per strip within a film follow a common Poisson distribution with mean S,

$$P(x / S) = \frac{e^{-S} S^x}{x!}, \quad x = 0, 1, 2 \ldots \qquad (1)$$

[1]Paper No. 8291 Scientific Journal Series, Minnesota Agricultural Experiment Station, St. Paul.

[2]To whom correspondence should be sent at the Department of Food Science and Nutrition.

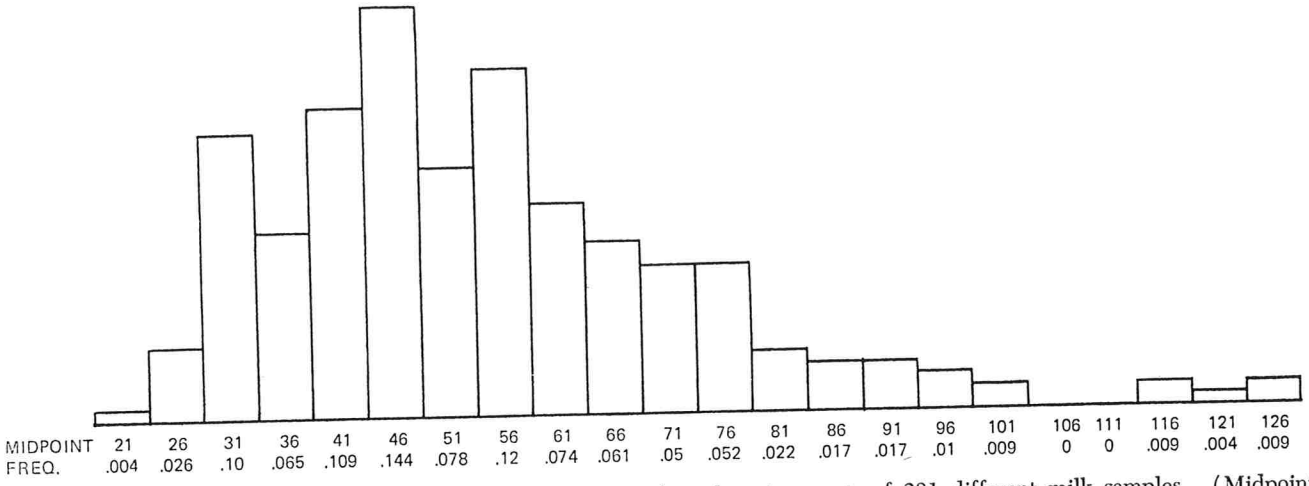

Figure 1. Histogram showing midpoint and frequency of single strip counts of 231 different milk samples. (Midpoint figures are based upon a microscope having a strip factor of 15,400.)

and *(b)* that variation between films is small. For a consideration of these assumptions, the reader is referred to the work of Smith *(8)*. In this investigation and for the five technicians who did the tests, the between film variation averaged 3.0% overall, certainly a very reasonable variation.

Once the above two assumptions are satisfied, it may be further assumed that the strip counts over both films follow a common Poisson distribution, and this is the essential requirement of the method that follows.

Proposed procedure

Depending on the width of the strips and size of film, the maximum allowable concentration of somatic cells per milliliter of milk will be equivalent to a maximum allowable concentration (L) per strip. The proposed procedure tests sequentially the hypothesis that H: S = L vs. HA: S = L − C (where C >0 is a constant with a maximum number (4) of strip counts). This choice of constant will be discussed later, but for now it suffices to indicate that the above hypothesis format was selected so that the probability of error could be controlled at S = L.

In a fully sequential test (1), that is, one in which maximum sample size is not specified in advance, the decision to stop or continue sampling is usually based on the selection of two numbers (A and B) related to type 1 and type 2 errors. Sampling is continued as long as B $<Z_m <$A; the first time $Z_m \geq$ A, accept HA and stop sampling; the first time $Z_m \leq$B, accept H and stop sampling. Here:

$$Z_m = \frac{\prod_{i=1}^{m} P(x_i|\ L\text{-}C)}{\prod_{i=1}^{m} P(x_i|\ L)} \qquad (2)$$

x_i is the ith observation and m is the number of observations taken to that point. For the Poisson distribution and hypothesis in question the criterion B $<Z_m <$A simplifies to

$$G(m) = \frac{\log \frac{A\text{-}mC}{\left(\frac{L\text{-}C}{L}\right)}}{} < x_1+x_2+\ldots+x_m < \frac{\log \frac{B\text{-}mC}{\left(\frac{L\text{-}C}{L}\right)}}{} = F(m) \qquad (3)$$

In the proposed scheme, the above criterion for m=1,2,3 is used, and if sampling has not terminated by the third strip count, then on the fourth count accept H [HA] if $x_1+x_2+x_3+x_4 >$ [≤] 2(2L-C). The constants A and B are chosen to give the desired type 1 and type 2 error values. In general, these constants —and therefore G(m) and F(m)—will be very difficult to determine precisely without the aid of a computer.

In summary, the sampling procedure is implemented as follows: *(a)* Continue counting as long as G(m)

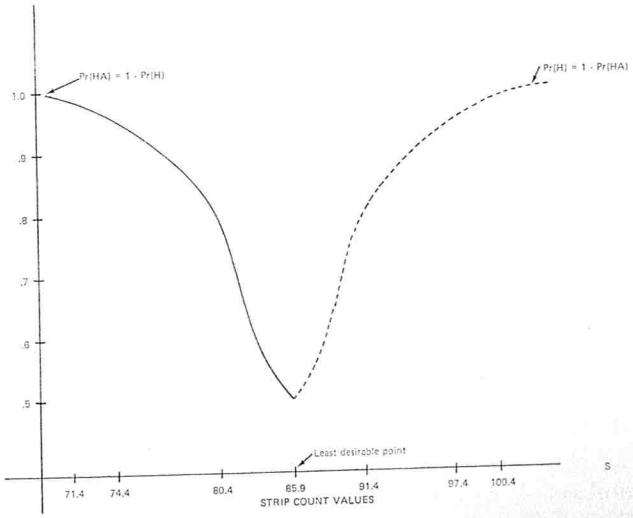

Figure 2. Probability of a correct classification of somatic cell count using a truncated sequential test procedure.

$<x_1+x_2+\ldots+x_m<F(m)$, m = 1,2,3. *(b)* The first time $x_1+\ldots+x_m \leq G(m)$, stop counting and accept HA. *(c)* The first time $x_1+\ldots+x_m \geq F(m)$, stop counting and accept H. *(d)* If counting has not terminated by the third strip, then count a fourth strip and accept H [HA] if $x_1+x_2+x_3+x_4 \leq [>]$ 2(2L-C).

Choice of C

The choice of a particular value for C is to some extent arbitary. However, reasonable recommendations can be given.

It is desirable to choose C so that a large percentage, P, (e.g. .70 <P <.90) of all the samples having true S values less than L fall less than L-C. Choosing C in this manner tends to reduce the percentage of samples with true S value between L-C and L. The proposed procedure would be least effective when S is between L-C and L. That is, around the least desirable point, which is S = (2L-C)/2, the probability of accepting either hypothesis is approximately .5. At the same time, it must be pointed out that the value selected for C should not be too small —as C decreases the expected number of strip counts increases. Thus, if C is chosen small, four strip counts will usually have to be taken and the sequential procedure will lose its advantage.

Clearly, an adequate choice for C necessitates knowledge about the distribution of true concentration (S). If this distribution is centered about S = (2L-C)/2 with a moderately small variance, the procedure will have little value. Not only will it be necessary to count four strips much of the time, but the chosen type 1 and type 2 errors will be meaningless; most of the time the true S value will be between L-C and L. The essential first step, then, is to note the kind of distribution that actually exists. From single strip count data acquired during the testing of 231 samples, the histogram in Fig. 1 was developed. This provides a reasonable picture of the distribution of true S values. Based upon this distribution, consider two examples of the use of this procedure, one in which the somatic cell count standard is placed at 1.5 million, the other at 1.0 million.

Example 1

With the maximum allowable concentration of somatic cells established at 1.5 million/ml, using a strip factor of 15,400, the value for L becomes 97.4. The hypothesis in question is H: S = 97.4 vs. HA: S = 97.4 – C.

From the data which served to develop Fig. 1, C was chosen to be 23, which gives P = 0.88. The numbers A and B needed to determine G(m) and F(m) were selected, with the aid of a computer, by adjusting the fully sequential approximations (1) to yield type 1 and type 2 errors of approximately 0.05. This resulted in the following values for G and F:

m	G(m)	F(m)
1	80	91
2	165	176
3	250	261

Terminal point 2(2L-C) = 343

Figure 2 shows how the exact probability of accepting each hypothesis changes as the true concentration, S, changes. Computations for the power curves in Fig. 2 were carried out on the University of Minnesota CDC 6400.

Thus, it would appear that the sequential procedure would be of real value for data of the type illustrated in Fig. 1. In about 85% of the samples, counting would be terminated after the first strip count. Moreover, it could be expected that a small percentage of the samples would have true S values between 74 and 97, such that, in the least desirable of situations, where S = (2L-C)/2 = 85.5, the counting of four strips would be required in only 4% of the samples. This percentage would become smaller as S deviates from (2L-C)/2.

Example 2

Using a standard of 1.0 million somatic cells/ml and the same strip factor (15,400), the hypothesis becomes H: S=65 vs. HA: S=65-C. Now, from the data of Figure 1, C was chosen to be 15, which give P =.65.

For type 1 and type 2 errors of .05, the values for G(m) and F(m) were determined to be:

m	G(m)	F(m)
1	48	66
2	105	123
3	163	180

Terminal point 2(2L-C) = 230

In this situation, counting could be terminated after a single strip in about 68% of the samples. In the least desirable case (S=57.5), four strip counts would be needed for 29% of the samples.

A comparison of this and the previous example indicates that, for the data derived in this laboratory, as expressed in Fig. 1, the procedure becomes less effective as L decreases.

It must be noted that the power of the truncated sequential test can be significantly less than that of the usual technique on which four strips are always counted. For the power levels to be comparable, a maximum sample size greater than four is necessary. However, preliminary investigations indicate that substantial reduction in labor can still be realized with a maximum sample size chosen to yield power levels comparable to the fixed sample size procedure. This point will be considered in greater detail in a future paper which will include a much broader data base.

References

1. Kendall, M. G., and A. Stuart. 1967. The advanced theory of statistics. Vol. 2. Hafner Publishing Co., New York.

2. Levowitz, D., and M. Weber. 1956. An effective "single solution" stain. J. Milk Food Technol. 19:121.

3. National Mastitis Council, Subcommittee on Screening Tests. 1968. Direct microscopic somatic cell count in milk. J. Milk Food Technol. 31:335-376.

4. Prescott, S. C., and R. S. Breed. 1910. The determination of the number of body cells in milk by a direct method. J Infect. Dis. 7:632-640.

5. Schultze, W. D. 1968. Design of eyepiece reticles for use in the direct microscopic somatic cell count method. J. Milk Food Technol. 31:344-349.

6. Schultze, W. D., J. W. Smith, D. E. Jasper, O. Klastrup, F. H. S. Newbould, D. A. Postle, and W. W. Ullmann. 1971. The direct microscopic somatic cell count as a screening test for control of abnormal milk. J. Milk Food Technol. 34: 76-77.

7. Schultze, W. D., J. W. Smith, D. E. Jasper, O. Klastrup, F. H. S. Newbould, D. S. Postle, and W. W. Ullman. 1972. Collaborative study of some screening tests for detection of abnormal milk. J. Milk Food Technol. 35:639-645.

8. Smith, J. W. 1969. Development and evaluation of the direct microscopic somatic cell count in milk. J. Milk Food Technol. 32:434-441.

9. Standard Methods for the Examination of Dairy Products. 13th Edition. American Public Health Association, New York.

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SEPTEMBER 1973

WASHINGTON, 11TH

Holiday Inn, Everett

WISCONSIN, 20, 21

Holiday Inn, Stevens Point

CONNECTICUT

OCTOBER 1973

INDIANA, 2, 3, 4

Terre Haute

FEBRUARY 1974

KENTUCKY

MARCH 1974

ONTARIO

APRIL 1974

MISSOURI

JUNE 1974

PENNSYLVANIA

(All affiliate secretaries are requested to send annual meeting dates as far in advance as possible so we can keep up-to-date)

NOTICE TO MEMBERSHIP

In accordance with IAMFES Constitution and By-laws, which requires our Second Vice-President and Secretary-Treasurer to be elected by mail ballot, you are hereby notified that President Earl O. Wright, at the annual meeting in Rochester, New York, August 1973, appointed the following members to the nominating committee for 1974: William LaGrange, John Bruhn, Melvin Jefferson, George Hazelwood, Roy Ginn, P. N. Travis and Ray Belknap.

Nominations for the office of Second Vice-President and Secretary-Treasurer are now open and any member wishing to make a nomination should send a picture and biographical sketch of his nominee to the Nomintaing Committee not later than October 15, 1973. To maintain proper balance on Executive Board nominees should be selected from industry, preferably food processing area.

R. A. Belknap, Chairman
Nominating Committee
79 Locust Avenue
Ft. Mitchell, Kentucky 41017

AN ANNOUNCEMENT TO THE MEMBERSHIP: CHANGES IN PROCEDURES APPROVED BY THE EXECUTIVE BOARD

The Executive Board of IAMFES recently met in Rochester, New York and approved several changes in procedures to be followed by the Association. These changes should be of interest to the entire membership and hence are detailed below.

Election of officers

The following procedure will be followed for the next election of officers: (a) a call for nominations will appear in the September issue of the *Journal of Milk and Food Technology,* (b) biographical sketches of nominees will be published in the November issue of the *Journal,* and (c) ballots will be sent by first class mail on January 15, 1974 to all members of record on December 31, 1973. It is hoped that this revised procedure will simplify the entire election process.

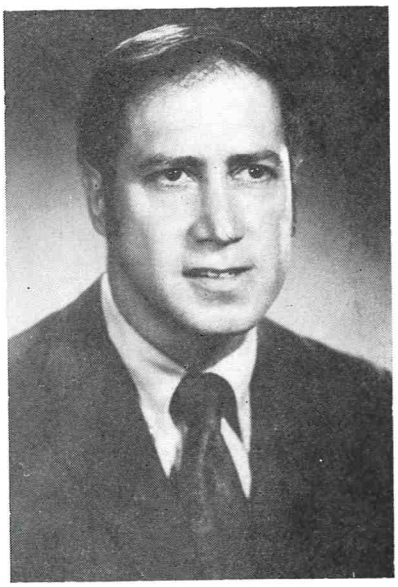
Nominations for recipients of awards

IAMFES presents five awards at the Annual Meeting. They are: (a) Citation Award (plaque), (b) Honorary Life Membership (plaque), (c) C. B. Shogren Award to an affiliate (plaque), (d) Sanitarians Award (plaque and $1000), and (e) University/Industry Award (plaque and $1000).

All members may nominate persons to receive these awards. To facilitate making the nominations, an appropriate form will be sent to all members when they receive the ballot for the election. This form is to be completed and returned to the Chairman of the Recognition and Awards Committee.

Contributed research papers at 1974 Annual Meeting

Research workers are invited to contribute research papers for presentation at the 1974 Annual Meeting of IAMFES. The meeting is scheduled for August 12-15, 1974 at the St. Petersburg Hilton Hotel, St. Petersburg, Florida. Abstracts of papers to be presented at the meeting must be submitted by February 15, 1974. Details about contributed research papers at the 1974 Annual Meeting appears elsewhere in this issue of the *Journal.* Abstract forms will be included in a subsequent issue of the *Journal.*



FOOD SCIENTIST TO HEAD STATE ORGANIZATION

David K. Bandler, assistant professor of Food Science at the N.Y. State College of Agriculture and Life Sciences, Cornell University, has been elected president of the New York State Association of Milk and Food Sanitarians. He will serve as head of the organization for one year.

Founded in 1923, the 800-member association was organized to aid in the improvement of sanitary and nutritional qualities of dairy and other food products. its members include milk and food sanitarians, regulatory officials, educators and representatives of the food and dairy industry throughout New York State.

A member of the Cornell faculty since 1965, Bandler has taught courses in both the Department of Food Science and the N.Y. State Veterinary College. He is a member of the dairy extension faculty with responsibility for the milk flavor and quality control programs in New York State.

A native of New York City, he earned his MPS-CA in 1971 and B.S. in 1955 from Cornell.

Prior to returning to Cornell in 1965, he served as director of research for the N.Y. State Joint Legislative Committee on Imitation Food Products and Problems frm 1956 to 1965. He also served as organizational chairman and secretary-treasurer of the American Cultured Dairy Products Institute, a national association of cultured dairy products manufacturers.

In addition to his teaching responsibilities at Cornell, he has authored numerous articles on nutrition and quality control of milk and other dairy products. He also wrote and produced a weekly radio program, "Food for Thought," heard on 12 stations throughout New York State in 1971-72.

More recently, his work has focused on the development of educational programs aimed at food service personnel and members of the dairy industry involved with the production and handling of school milk supplies.

He is a member of the American Dairy Science Association, International Association of Milk, Food and Environmental Sanitarians, and he is secretary of the New York State Cheese Manufacturers Association.

He is also a member of the Board of Education of the Newfield Central School District, and has served as president since 1970.

Bandler and his wife, the former Lenore M. Ahalt, and their two children, Steven and Robert, live on and operate an 80-acre farm in Newfield.

NEWS AND EVENTS

GREINER NAMED DFISA EXECUTIVE VICE PRESIDENT

Fred J. Greiner, chief executive officer of the Evaporated Milk Association for the past 11 years, has been named executive vice president of the Dairy and Food Industries Supply Association, effective in mid September, announced Roy E. Cairns, president.

During his tenure at EMA from 1962 to the present, Greiner has been responsible for public relation, marketing services and education, production and sanitary standards, research and development, government liason, meetings, and publications. When the organization moved its headquarters from Chicago to Washington, D. C., in 1966, Greiner assumed the additional duties as director of public affairs for the International Association of Ice Cream Manufacturers and the Milk Industry Foundation. His primary responsibilities have included congressional and state legislative relations and environmental concerns.

Prior to joining EMA, Greiner was executive assistant with the Ohio State Council of Retail Merchants from 1960-62, preceded by an eight-year term as chief executive officer of the Ohio Dairy Products Association, 1952-60. At ODPA he directed all administrative functions, its legislative affairs, and state governmental relations.

Other previous positions include secretary of the Columbus Milk Distributors Association, 1950-52, and extension specialist in dairy technology on the faculty of the dairy department (now department of food science and nutrition) at Ohio State University, 1947-50.

Greiner is a native of Zanesville, Ohio. He was awarded his bachelor's degree in dairy technology at Ohio State University in 1947 after having served as a captain in the U. S. Army as an assistant battalion surgeon during World War II.

His activities in the dairy industry and trade association field are numerous. He is chairman of the Dairy Industry Committee, immediate past chairman of the Food Group, past chairman of the Dairy Association Executives Conference, member of the American Dairy Science Association and American Society of Association Executives, and past chairman of the National Institute for Organization Management at Michigan State University. In 1961 Greiner was one of the first thirty-four trade association executives in the U. S. to be awarded the coveted status of Chartered Association Executive by the American Society of Association Executives.

KEYS TO MEMPHIS AND MILK MARKETING

"It is imperative that the dairy industry invest more monies in advertising and nutrition education programs," stated Dr. C. Bronson Lane, Executive Director of the Dairy and Food Nutrition Council of Florida (DAFNC). Addressing the delegates at the 41st Memphis Dairy Council Key Leader Meeting, Lane cited unjustified and unscientific criticism of dairy products, consumer cost concerns, stepped-up activity by competitors, and chaotic regulatory controls as factors responsible for the over-all decline in milk consumption during the last ten years.

"Effective communications with health and education leaders, coupled with sound nutrition education programs, especially for youth, can help reverse this ominous milk consumption trend," Lane continued, "and clear up the misconceptions about dairy products held by confused consumers."

Lane commended the Memphis Dairy Council for its nutrition education endeavors on behalf of the Tennessee dairy industry, and stated that the newly formed DAFNC was adopting many of the successful techniques and programs used by the Memphis organization.

Dr. Lane was presented a key to the city of Memphis by the Mayor's Assistant prior to his presentation.

34TH ANNUAL MEETING OF THE INSTITUTE OF FOOD TECHNOLOGISTS

The 34th Annual Meeting of the Institute of Food Technologists will be held May 12-16, 1974, in New Orleans.

More than 5,000 professional food scientists and technologists will attend, technical and other functions including FOOD EXPO, the nation's largest technical-education exposition for food professionals affiliated with industry, education, research and government.

The New Orleans Rivergate convention and exposition center will be the scene of the various technical sessions and meetings as well as the near-300 booth exposition.

The Marriott and Fairmount Roosevelt Hotels will be co-headquarters for the meeting.

The Institute of Food Technologists, organized in 1939 is an international professional society of more than 12,000 members and is headquarted in Chicago.

DEVELOPING AN AREAWIDE PLAN FOR SOLID WASTE MANAGEMENT

A booklet on solid waste produced by Joseph W. McIntosh, Public Health Consultant, Institute and Area Development, University of Georgia.

Written in an understandable vocabulary. Copies may be obtained for $1.00 from Institute of Community and Area Development, Old College, University of Georgia, Athens, Ga. 30602.

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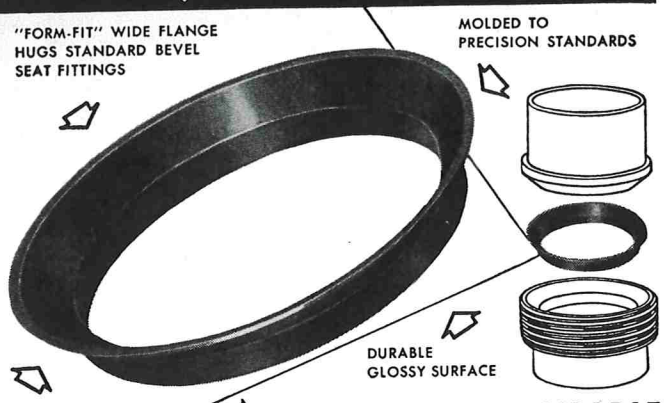

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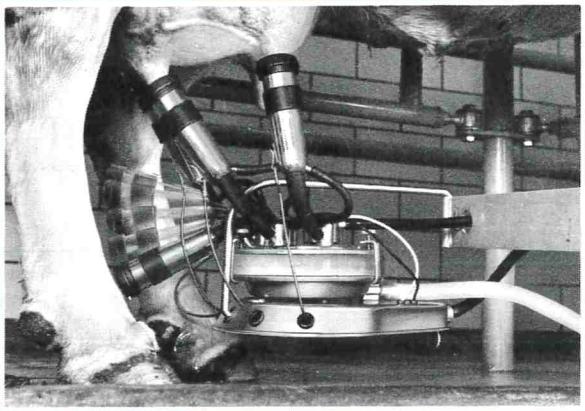
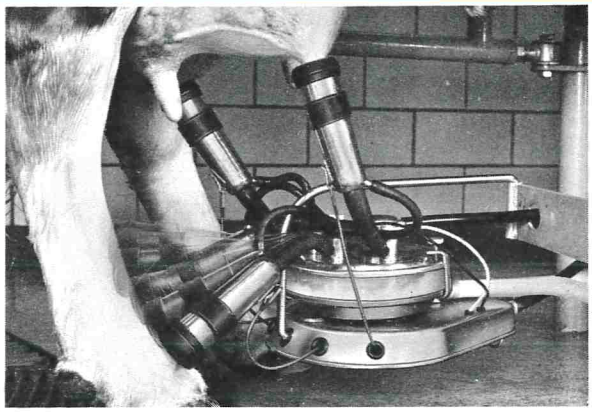
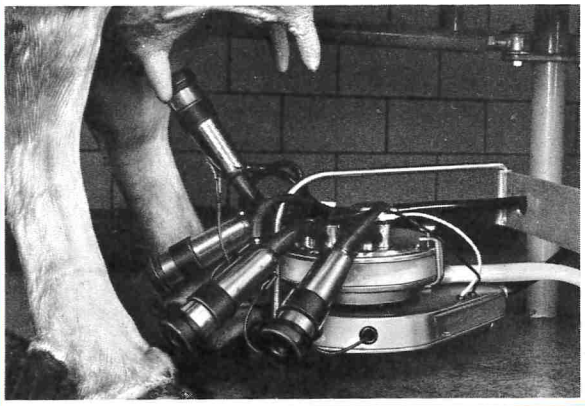


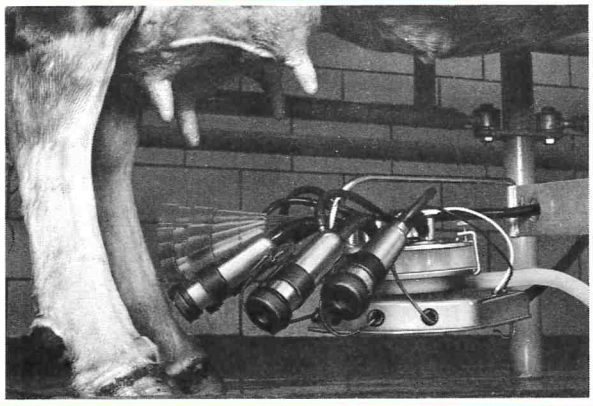
:0220 Two minutes and twenty seconds after unit is attached, left front quarter milks out. QTO teat cup is automatically removed. All other quarters continue milking.



:0254 Thirty-four seconds later, right rear quarter has finished. Milk flow stops and second teat cup is taken off.



:0329 Right front quarter milks out thirty-five seconds later and teat cup is removed automatically.



:0417 Last quarter milks out four minutes, seventeen seconds after unit is attached. Time difference between first and last quarter: one minute and fifty-seven seconds.*

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*Average based on 160,000 individual quarter timings.