Studies on the Contamination of Products Produced by Rendering Plants

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SUMMARY

Studies on the bacterial contamination in rendered product and the environment of five rendering plants were carried out. From a total of 180 samples examined, total bacterial and anaerobic spore counts were conducted on 135.

Plants with melter systems produced a sterile product which was recontaminated before reaching the finished stage. Two plants with continuous rendering systems did not achieve sterilization of the product during the heating process. Spore forming organisms regularly survived heating in the continuous rendering system.

Salmonellae were isolated from samples collected in four of the five plants under study. Pathogenic Clostridia, especially Cl. novyi, Cl. septicum and Cl. perfringens were present in samples from all plants. Other pathogens found were Staphylococci, Streptococci, Corynebacteria and Pasteurella.

RÉSUMÉ

L'auteur a procédé à l'étude de la contamination bactérienne des sous-produits animaux et de l'environnement de cinq usines de ces sous-produits. Des 180 échantillons examinés, il en a soumis 135 à un comptage complet des microbes et des spores anaérobies.

Les usines pourvues d'une système de fonte donnèrent un produit stérile qui fut contaminé ultérieurement, avant d'atteindre son stade final. Deux usines pourvues d'un système de transformation continuelle ne réussirent pas à stériliser le produit au cours de son traitement par la chaleur. Des microbes sporulants survécurent régulièrement dans le système de transformation continuelle.

L'auteur a isolé des salmonellas dans les échantillons prélevés à quatre des cinq usines faisant l'objet de son étude. Les échantillons prélevés dans chacune des usines contenaient des clostridies pathogènes, entre autres Cl. novyi, Cl. septicum et Cl. perfringeus. Des staphylocoques, des streptocoques, des corynébactéries et des pasteurelles représentaient les autres microbes pathogènes qu'il a isolés.

INTRODUCTION

In recent years there has been an increasing awareness of the dissemination of animal pathogens by contaminated feedstuffs, especially meat and bone and fish meal. The attention of most investigators focused on salmonella. The presence of salmonella in meat and bone meal from rendered products is well documented (1,4,5,6, 7,8,9,10,11) and outbreaks of salmonellosis have been associated with feeding of contaminated feedstuffs (1,4). Introduction of exotic serotypes to localities where these were previously not known is also reported (1,5,10). It is conceivable that the incidence of passive carriers can be augmented in this way, which could cause increased salmonella contamination in the food processing industry and consequently greater human exposure.

Detailed studies on the ecology of contamination in rendering plants indicate

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that salmonellae did not survive the heating process and the product became recontaminated with salmonellae-carrying materials in the environment of the rendering plant and storage facilities (6,7,9,11). Other reports however indicated that salmonellae survived the heating process (4, 9). Airborn infection seemed to be the main source of contamination. Poor sanitary practices were incriminated as the cause of contamination and segregation of the heated product, with storage in sealed containers, is recommended to maintain a wholesome product (6,9,11).

Reports of other pathogens other than salmonella are less frequent. *Bacillus anthracis, Clostridium botulinum* and Staphyloccus have been reported (7).

In this study, the effect of different rendering systems on the type and degree of bacterial contamination during processing and in the final product, was investigated.

MATERIALS AND METHODS

PLANTS

Five plants in southern Ontario were selected for study. Plants A, B and C were of the conventional "melter" type (Fig. 1). The material was boiled for two to five hours, then the temperature was raised to 248-285°F for one half-hour to four hours. During the heating process the material was continuously mixed by rotating blades inside the tank. The following differences existed between the melter plants. Plant A was not attached to a packing plant. The "septic" receiving area was completely separated from the "clean" finished product area. Maximum heat applied was 265°F for three hours. The handling of the material was exclusively mechanical. Plant B processed offal from hogs only. This was a small plant with excellent sanitation. Septic and clean areas were segregated. Maxi-

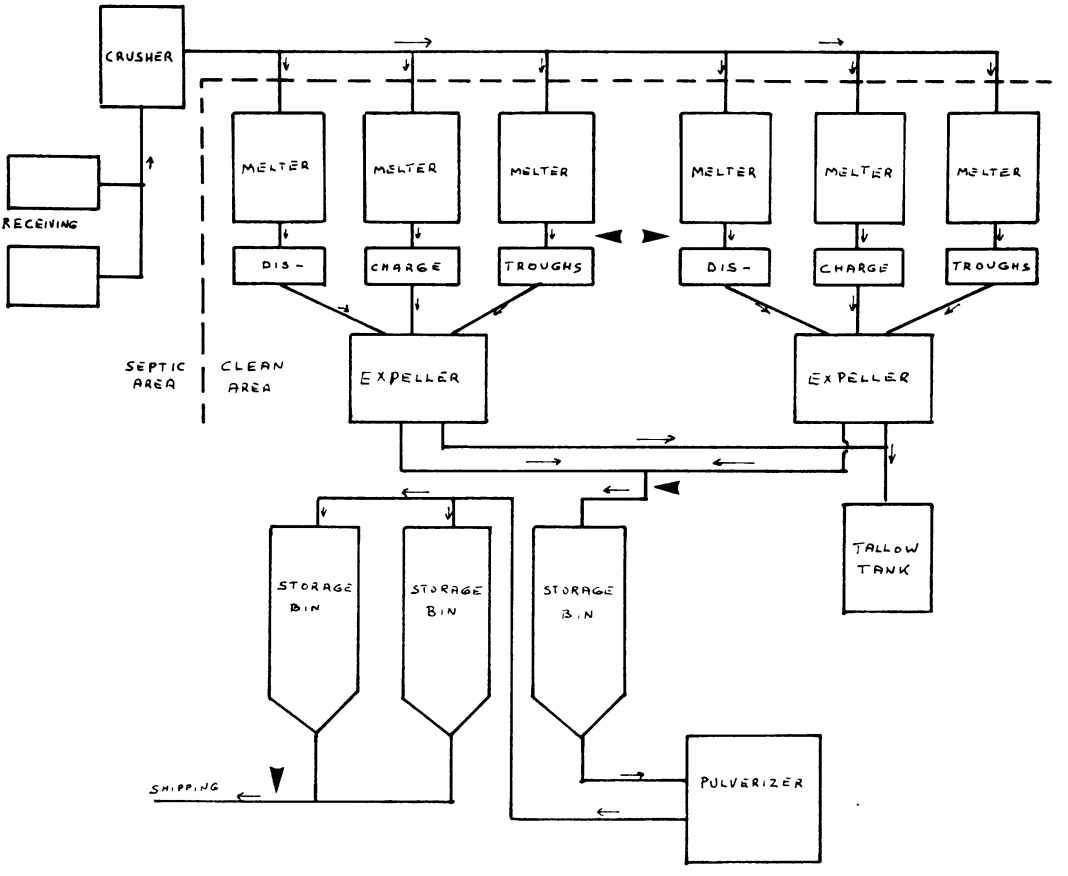

Fig. 1. Schematic outline of a melter plant.
◂Points where samples were taken. — — — Separation of "septic" and "clean" areas.

mum heat applied was 285°F for half an hour. The heated material was shovelled manually into a press which produced cracklings in the form of a pressed cake. Plant C was an older establishment with septic and clean areas in the same room. Maximum heat applied was 248°F for two hours with manual shovelling of the heated material into the expeller.

Plants D and E were continuous rendering systems (Fig. 2). The material was liquefied by adding warm tallow at approximately 130°F. The core of the system was a heating pipe of approximately 50 feet in length in which the material was heated to 235-300°F and which discharged into a vacuum chamber with 27″ of vacuum. The material circulated through the heater-evaporator system with fresh material being continuously added and withdrawn.

Samples

Approximately 100 gm of material was collected into sterile bags using sterile tongue depressors. Hot fluid samples were collected into sterile jars. Samples were collected immediately after the heating process, following fat extraction, from the finished product and from surfaces of the equipment and buildings where dust was likely to accumulate. In addition, samples of dust in the air were collected by exposing a widemouth jar of 2″ diameter with 50 ml of MacConkey or nutrient broth for six to eight hours. Swab samples from surfaces were also taken. No counts were made from air and swab samples. Bacteriological examinations were initiated on each sample on the day of collection.

Bacteriological Methods

Bacterial counts were made by the pour plate technique using tryptic soy agar enriched with yeast extract 3%, glucose 1% and buffered with sodium phosphate to pH 7.0. Ten gm of each sample was mixed in a Waring blendor with 1% peptone water, and further diluted to 1:500 and 1:5000 for inoculation of the plates. For total counts the plates were incubated for 24 hours at 37°C. For anaerobic spore counts a portion of each dilution was heated to 80°C for ten minutes before inoculation. The plates were incubated anaerobically for 48 hours at 37°C.

Three to 5 gm portions of each sample

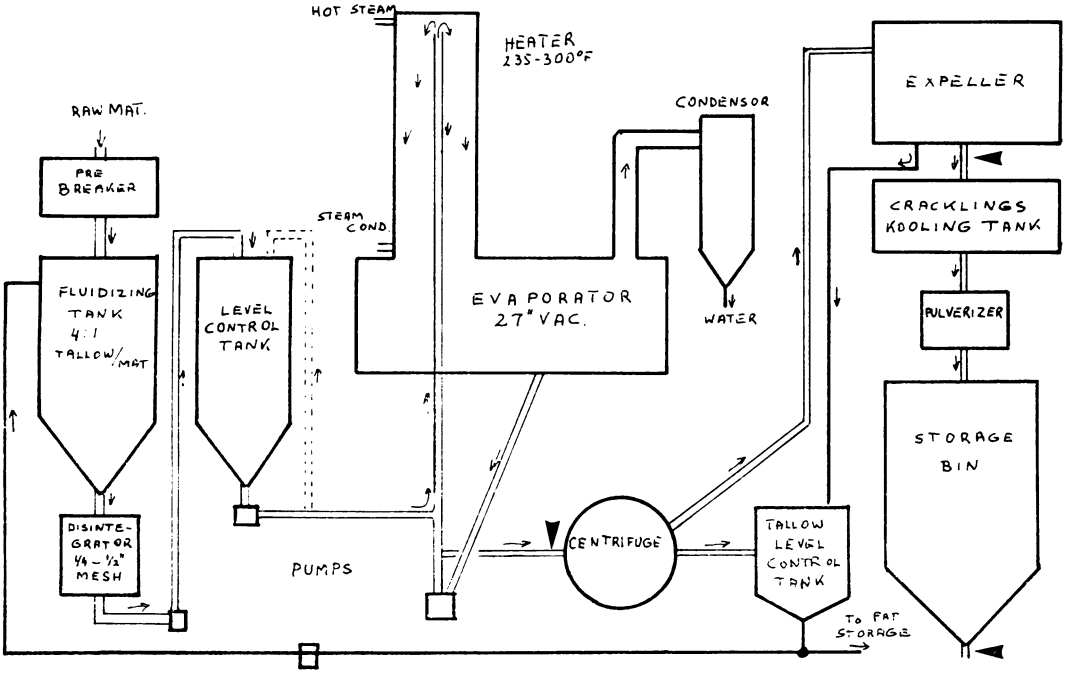

Fig. 2. Schematic outline of a continuous rendering plant using recycling of the material in a pressure-vacuum system.
◀Points where samples were taken.

TABLE I. Viable Vegetative and Anaerobic Spore Counts of Rendering Products Collected at Various Sites

Plant	Melter-Evaporator		Expeller		Finished Products		Dust from Surfaces	
	Viable Count	Spore Count	Viable Count	Spore Count	Viable Count	Spore Count	Viable Count	Spore Count
A[a]	—	—	170	400	150	15	2,000	2.500
Melter type	—	—	60	1	150	15		
					60	4	640	42
	—	—	2	1	8	1	50,000	100
	—	—	42	S[b]	35	S	S[b]	S
	—	—	3	S	10	S	590	53
B[a]	—	—	50	30	12	4	60	S
Melter type					8			
	—	—	5	2	2	S	6	S
			3	3	3	4	12	8
	—	—	3	2	S	4	350	190
C[a]	—	—	5	7				
Melter type			2	30	S	S	200	50
	—	—	S	S	15	—	200	2
	—	—	85	25	200	65	70	30
			0[c]	0	30	28		
	—	—	76	1	180	2	600	S
	—	—	10	3	1		20,000	0
							20,000	
					280	3	150	30
D[a]	10	48	30	19	6	9	200	3
Continuous rendering	40	40	29	20	40	17		
ystem	40	16			45	19	850	20
	15	53	170	40	100	10	12,000	35
	2	1	35	S	500	S	340	3
			32	2	32	15		
E[a]	2	2	3	2	300	130	40	3
Continuous rendering	3	3	3	1	6	1		240
system	60	6	4	4	310	52	S	
	6	7	8	10	72	56	350	210
	3	2	70	60	127	200	870	750
	12	40	77	85	112			
				9	S		S	
	60	S		2	70	30	0	0
	6	4	190	30	350	50	650	37
			40	4				

Counts are thousands of viable organisms per gram
[a]Samples in one line collected on same day, blanks indicate no sample collected
[b]S = spreader
[c]0 = overgrown

were also inoculated into the following media: 1.) Flask with 50 ml of tetrathionate broth incubated at 42°C for 24 hours, then plated onto SS agar. 2.) Flask with 50 ml of selenite F broth with cysteine (Difco) incubated at 37°C for 24 hours, then plated on brilliant green agar. Pure cultures of suspected salmonellae were tested biochemically and those which conformed to the characteristics of the genus were forwarded for typing to the Public Health Laboratory, Ontario Department of Health, Toronto. 3.) Robertson's cooked meat medium (3) which had been heated to 80°C for ten minutes and sealed with a layer of sterile paraffin oil. After incubation for 72 hours at 37°C a loopful was transferred to lactose-egg-yolk-milk half antitoxin agar plates (12) and to blood agar plates (5% calf blood) and incubated anaerobically for 72 hours at 37°C. 4.) One tube of Brewers thioglycollate medium (Difco) was inoculated and the same procedure followed as with cooked meat medium. A second tube of thioglycollate was incubated without previous heating at 50°C for 8–12 hours, and then sub-cultured to plates as above. Pure cultures were checked for aerobic growth,

grown in cooked meat medium and inoculated to media listed on the "Table for Differentiation of Clostridia" by Cowan and Steel (3). Pathogenicity was tested by intramuscular inoculation of 0.5 ml of fresh broth cultures with added calcium chloride 1.5% to guinea pigs. Final identification of *Cl. novyi* and *Cl. septicum* was obtained by the fluorescent antibody technique through the Diagnostic Laboratory of the Ontario Veterinary College. 5.) A flask containing 125 ml nutrient broth (Difco) which was incubated at 37°C and a loopful sub-cultured after 24 and 48 hours to the following media; a.) blood agar (5% calf blood), b.) MacConkey agar (Difco), c.) Azide agar (Difco), d.) Baird-Parker agar (Oxoid), e.) Sabouraud slope (Difco). After the isolation of a pure culture, differentiation was carried out, following the classification schemes of Cowan and Steel (3) and Bergey's manual (2).

RESULTS

The main difference between the product from the melter-type plants and continuous rendering systems was the sterility of the melter samples in contrast to the evaporator samples which showed consistent growth of spore formers in the range of 10–60,000 organisms per gm (Table I). The range of the counts was broad and no sequence of increased contamination during processing could be shown. Even the counts of dust from surfaces were comparable to those of the finished product. The surface samples with extremely high counts were from moist areas where condensed water or leakage apparently favoured growth. Although none of the plate counts from the melter samples had any growth, occasional colonies of *Bacillus, Staphylococcus* and *Streptococcus* spp. grew on plates inoculated from the enrichment media. In contrast the evaporator samples from the continuous rendering plants showed spore-forming organisms only, but in all samples.

All samples from subsequent stages including the expeller, finished product and surfaces had a variety of bacterial contaminants and pathogens (Table II and III).

Types of Bacteria Present

Salmonella were isolated from four plants (A,C,D,E) in the product as well as from various surfaces. The following serotypes were identified according to frequency. *S.orion, S.derby, S.binza, S.montevideo, S.cerro, S.tennessee.* Of the pathogenic clostrida, *Cl. perfringens* was most frequently found in all plants and in most types of samples. Other pathogens found were *Cl.novyi, Cl. septicum* and *Cl.sordelli. Cl.perfringens* isolates killed guinea pigs within 24 hours. *Cl.novyi, Cl.septicum* and *Cl.sordelli* caused an illness of one to three days characterized by inappetence, lethargy and lameness, but not death. Necrosis of the claw of the injected leg was frequently seen in recovered animals. Non-pathogenic clostridia found in order of frequency were *Cl.sporogenes, Cl.bifermentans, Cl. histolyticum, Cl.tertium, Cl.putrefaciens* and a

TABLE II. Bacteria Isolated from Melter Type Rendering Plants[a]

Evaporator	Expeller	Finished Product	Surfaces
	S. derby		
Cl. perfringens	*Cl. perfringens*	*S. derby*	*S. binza*
Cl. septicum	*Cl. novyi*	*S. cerro*	*S. montevideo*
Clostridia spp.	*Cl. septicum*	*S. binza*	*S. tennessee*
Bacillus spp.	*P. multocida*	*Cl. perfringens*	*Cl. perfringens*
	β-haemolytic streptococci	*Clostridia* spp.	*Cl. septicum*
	Staph. aureus	*Staph. aureus*	*Cl. sordelli*
	Clostridia spp.	*Bacillus* spp.	*Cl bovis*
	Bacillus spp.	*Streptococcus* spp.	*Staph. aureus*
	coliforms	*Staph. aureus*	*Staph.* spp.
			β-haemolytic streptococci
			Clostridia spp.
	Streptococcus spp.	*Staph.* spp.	*Streptococcus* spp.
	micrococci	*Pseudomonas* spp.	*Bacillus* spp.

[a]Pathogens are reported with the name of the species or serotype, the generic name only indicates non-pathogenic species

TABLE III. Bacteria Isolated from Continuous System Rendering Plants[a]

Expeller	Finished Product	Surfaces
S. binza	*S. binza*	*S. binza*
S. montevideo	*S. orion*	*S. montevideo*
S. orion	*Cl. perfringens*	*S. orion*
Cl. perfringens	*Cl. septicum*	*Cl. perfringens*
Cl. novyi	*Clostridia* spp.	*Cl. septicum*
Clostridia spp.	*C. pyogenes*	*Cl. novyi*
coliforms	*C. renalis*	*P. multocida*
Streptococcus spp.	coliforms	*Clostridia* spp.
micrococci	micrococci	coliforms
Bacillus spp.	*Bacillus* spp.	*Streptococcus* spp.
	Pseudomonas spp.	micrococci
		Bacillus spp.
		Pseuedomonas spp.

[a]Pathogens are reported with the name of the species or serotype, the generic name only indicates non-pathogenic species

number of isolates which were not classified as they showed atypical biochemical reactions.

Other nonsporulating pathogens found in order of frequency were coagulase-positive staphylococci, beta-haemolytic streptococci, *Pasteurella multocida, P.haemolytica, P. pseudotubercolosis, Corynebacterium bovis, C.renalis* and *C.pyogenes*. Commonly occurring contaminants were coliform bacteria, *Pseudomonas* spp., coagulase-negative staphylococci, fecal streptococci and micrococci (Table II and III). *Bacillus* spp., fungi and yeasts were present in nearly all samples. Except for a check for *B.anthracis* colonies these organisms were not classified further.

Twenty six samples of dust from the air and 18 swab samples were taken in an attempt to detect the mode of contamination towards the end of the sampling period. As they were not collected simultaneously with the other groups of samples they are not included in the tables, but the bacterial flora of these samples was virtually the same as that of the surface samples.

DISCUSSION

The extent of the bacterial contamination was found to be directly related to the ease with which the plants could be and, in fact, were cleaned. No salmonella was found in plant B and this plant also showed the least bacterial contamination in the product. The sanitation was excellent. The loading area, one floor above the melter, was a spacious room with smooth surfaces, facilitating easy cleaning. The product, a pressed cake, created less dust and thus probably reduced surface contamination. In contrast the construction of plant C made recontamination of the product almost a certainty. Raw material was transported on open conveyor belts in the same room beside and directly above the discharge of the heated material. Pipes, cables and rough surfaces on the ceilings and walls prevented cleaning. The common floor of raw and processing area was usually wet and traffic unrestricted. A storage room of rough concrete with condensed moisture in some places provided a seed bed for microorganisms. The largest variety of bacteria was isolated in this plant. The degree of contamination in plants A, D and E were similar. Enclosed screw type conveyors and storage tanks were used in all three plants. As the inside of this equipment could not be cleaned, pockets of material tended to accumulate, which, with condensed moisture which was occasionally present could serve as a nidus for recontamination of fresh material. While these conveyors are designed as a closed system there were a number of open places through which airborne contamination might have occurred.

The continuous rendering system has many advantages where large amounts of material have to be processed. The flow of the material is rapid and the entire system could be hermetically closed. The failure to sterilize spores in the continuous system studied appears to be due to the short time the material is exposed to heat. In order to produce a sterile product and prevent recontamination the following alterations were suggested: 1.) The temperature in the fluidizing and level control tanks (Fig. 2) should be raised from the usual 130°–140°F to 190°F. 2.) The system should be hermetically sealed and where openings are necessary for cooling purposes air filters installed. 3.) The construction of all machinery where the operating temperature is below 150°F and all surroundings should allow for easy periodic cleaning and disinfection inside and out. 4.) The finished product should either be bagged or loaded in closed cars to prevent contamination.

The feed stuff supplier has a responsi-

bility to eliminate contamination of the product with pathogens. The results of this study are in agreement with those of other investigators (6,7,8,9,10,11) and confirm that salmonellae are destroyed during the heating process, but the final product is recontaminated before leaving the plant. This occurs in both types of plant. Complete sterilization of the rendering material at one point is essential to prevent dissemination of sporeforming pathogens such as pathogenic clostridial species and *B.anthracis*.

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Book Review

OUTLINE OF VETERINARY BACTERIOLOGY AND MYCOLOGY. *G. R. Carter. Published by Lucas Brothers, Columbia, Missouri. 1970. 143 pages. 25 tables. Price $4.95.*

In outline form, this book discusses all the pathogenic bacteria and fungi important in veterinary medicine. Each genus or family of organisms is discussed under the following headings: (1) general characteristics; (2) disease or diseases produced; (3) laboratory diagnosis; and (4) treatment, prevention and control. Although brief, the material is comprehensive and up to date, reflecting the author's experience in teaching, research and diagnostic microbiology.

The book was written as a "core of essential facts" for veterinary students studying bacteriology and mycology and it must be supplemented with lectures, discussions and visual aids. Used in this way, students will find this book extremely helpful. Since laboratory diagnosis is stressed, diagnostic laboratories will want this book as an invaluable reference. Practitioners, unless they do extensive diagnostic work, will probably find one of the standard textbooks of veterinary microbiology more useful. — *J. R. Long.*