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Microbial Load of Poultry By-Products Following Rendering Process

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Abstract: Rendering is a practical example of effective heat treatment to destroy microorganisms in raw poultry by-products and its conversion into rendered safe materials almost free from pathogens. The most important and valuable use for these rendered by-products is as feed ingredients for livestock, poultry and aquaculture. So, this study was applied on rendered poultry products before and after rendering on samples obtained from 10 rendering plants associated with poultry processing plants. Total bacterial count (TBC), fungal count (TFC), coliforms count (TCC) and isolation of *Salmonella* and *Campylobacter* spp. were determined. Results showed that there was a reduction of 99.96% in TBC, 99.99% in TCC, 100% in *Campylobacter* spp. count while *Salmonella* spp. percentage was reduced from 70% to 10% and, TFC reduced only by 60%. However, there was an evidence for post processing recontamination, as *Salmonella* spp., *Escherichia coli, Klebsiella* spp., *Enterobacter* spp., *Citrobacter* spp., *Penicillium* spp., *Yeast* spp., *Aspergillus fumigatus* and *pantoea* spp. this recontamination thought to occur from the environment in the processing plant. Conclusively, rendering process was found to be effective in reducing microbial load of raw poultry by-products. Also, presence of pathogens expected to be related to rendering plant environment contamination. So, the hygienic condition of processing plant should be monitored regularly and properly in order to reduce contamination.

Key words: Rendering • Poultry By-Products • Bacteria • Fungi

INTRODUCTION

Burial, incineration, composting and rendering are different methods used for disposal of animal and poultry carcasses and their wastes [1, 2].

Rendering is a classical example of effective heat treatment to destroy microorganisms and separate water, fat and protein contained in animal or poultry tissues under controlled and specific processes. Rendering converts raw inedible animal tissue into stable, value added materials resulting in many useful products like poultry by-product meal. Temperature and length of time of the cooking process can impact the quality of the finished product [3, 4].

National Renderers Association (NRA) [5] found that ground raw parts of slaughtered poultry carcasses as heads, feet, undeveloped eggs and intestine are highly contaminated with microorganisms including bacteria, viruses, virus-like particles, fungi, yeast and associated microbial toxins that constitute a potential risk to animal and human health [6, 7]. During rendering process, raw materials are cooked at a predetermined, continuously monitored temperature and atmospheric pressure in batch steam cookers (115°C to 145°C for 40 to 90 minutes) that inactivate many bacteria, viruses and molds [8].

After attempting to quantify microbial loads in raw poultry rendering materials, Glenn [9] discovered difficulties in enumerating bacteria by traditional aqueous buffer dilution methods due to the high fat content of the rendered material. Also, the high fat content of rendered products complicates traditional bacterial enumeration methodology. So, it was imperative to develop accurate test methods to detect these pathogens in high fat rendered materials to prevent false results [10]. However, the most important and valuable use for these rendered by-products is as feed ingredients for livestock, poultry and aquaculture [3] and this may result in human illness [11] specially *Salmonella* serotypes.

So, the objective of this study was to evaluate the effect of rendering process on the microbial load of poultry by-products.

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MATERIALS AND METHODS

The present study was conducted in some poultry processing plants to evaluate the effect of heat-pressure treatment followed during rendering process on the microbial contents of poultry by-products. Samples were obtained from ten rendering plants containing dry batch cookers in which raw materials are exposed to treatment of 140°C and pressure of 2 bars for 40-90 minutes.

Samples to Be Examined: Samples were collected after screening of poultry processing wastes and before rendering in the cooker. These samples were collected before cooking just near to the cooker before putrefaction using sterile gloves and plastic bags then transported in an ice tank to the laboratory as quick as possible. For sampling after cooking process, poultry meal samples were collected using sterile gloves and plastic bags then transported inside an ice box and stored at 4°C prior to analysis as described by Troutt *et al.* [12].

Microbiological Examination: After samples arrival at the laboratory, they were examined to determine Total Bacterial Counts (TBC), coliform count, mould& yeast count, isolation of coliforms, isolation of *Salmonella* spp. and enumeration, isolation & identification of *Campylobacter* spp.

Determination of TBC, Coliform Count, Fungal Count and Campylobacter Count: All were applied according to Kinley [4]. Samples were serially diluted using 0.1% sterile peptone water. Aliquots of each dilution were spread-plated onto Plate Count Agar for determination of total bacterial counts (TBC), MacConkey agar plates for total coliform count (TCC) and Saboraud's Dextrose Agar containing 0.5 mg of chloramphenicol for total fungal counts (TFC) and CCDA plates for Campylobacter count. Plates were incubated at 37°C for 24 h for total bacterial and coliform count, at room temperature for 3-5 days for fungi and under micro-aerophilic conditions at 42°C for 48-72h for Campylobacter count. After that fungal colonies were purified then undergo microscopical staining and identification using Lactophenol Cotton Blue stain according to Quinn et al. [13]. Bacterial and fungal counts were reported as CFU/g.

Isolation of Coliforms: Ten grams of sample were mixed with 90 mL pre-enrichment broth and incubated at 37°C for 24 h. For coliform testing inoculation of Mac-Conkey

Agar plates was applied and incubated at 37°C for 48 h according to Troutt *et al.* [12] followed by purification and biochemical identification of colonies according to Macfaddin [14].

Isolation of *Salmonella* **spp:** Ten grams of each sample were mixed with 90 mL pre-enrichment broth like buffered peptone water and incubated at 37°C for 18 h. For *Salmonella* detection, the sample enriched on Selenite-F broth and incubated at 37°C for 18 h, followed by plating onto S-S agar as done by Kinley [4]. After that purification of suspected colonies on nutrient agar plates followed by biochemical identification of colonies according to Macfaddin [14].

Isolation of *Campylobacter* **spp:** Ten grams of each sample were mixed with 90 mL of Bolton broth incubated under micro-aerophilic conditions at 42°C for 48-72 h. Followed by plating onto CCDA and Karmali agar under the same conditions followed by confirmation of colonies according to ISO [15].

RESULTS AND DISCUSSION

Raw poultry by-products exposed to rendering temperature of 140°C and pressure of 2 bars for 40-90 minutes (Table 1), which equivalent to requirements of The European Commission for Health and Consumer Protection Directorate [16] resulted in 99.96% reduction in TBC, 99.99% reduction in TCC and 100% reduction in *Campylobacter* spp. count. This agree with results obtained by several scholars [8, 12, 17] and Hess *et al.* [18] found that heat treatment and pressure of rendering equipment can make complete elimination of contaminants and this can be maintained if the product could be well handled and stored to prevent recontamination after processing.

Table (2) illustrated that in the final product low reduction percent in TFC (60%), *Salmonella* spp. that was isolated from 10% of samples, *Escherichia coli* 20% of samples, *Enterobacter* spp. 90% of samples and *Klebsiella* spp. 70% of samples. These results agree with Haapapuro *et al.* [19] who said that rendered animal co-products contain high number of microorganisms, including pathogenic bacterial species such as *Campylobacter, E. coli* and *Salmonella* spp. which may cause enteric affections in birds, animals and their consumers. Kinley *et al.* [20] found that total bacterial counts were in the range of 1.7 to 6.68 log10 CFU/g, with

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Microbial count	Before rendering	After rendering	Reduction%
TBC (cfu/g)	24x10 ⁸	77x10 ⁴	99.96
TCC (cfu/g)	31x10 ⁸	$2x10^{4}$	99.99
TFC (cfu/g)	16x10 ⁶	64x10 ⁵	60
Campylobacter count	34x10 ⁶	nil	100
T.B.C: Total Bacterial Count	T.F.C: Total Fungal Count	T.C.C: Total Coliform Count	

Table 1: The average total bacterial, coliform, fungal and Campylobacter spp. counts of poultry by-products before and after rendering:

Table 2: prevalence of bacterial and fungal isolates recovered from poultry by-products before and after rendering: N=10 samples

	Bacterial isolates										Fungal isolates						
						Campylobacter spp.				Aspergillus spp.							
	Salmonella	Proteus	Enterobacter	Citrobacter	Klebsiella				Pantoea	Hafnia			Morganilla	Penicillium	Mucor	Yeas	
Processing stage	Spp.	spp.	spp.	spp.	spp.	C.coli	C.jejuni	E.coli	spp.	spp.	A. niger	A.fumigatus	morgani	spp.	spp.	spp.	
% of positive	70	40	100	100	100	100	100	100	50	30	10	20	20	0	30	100	
samples before																	
rendering																	
% of positive	10	0	90	10	70	0	0	20	60	0	10	0	10	20	30	20	
samples after																	
rendering																	

the highest in blood meal and the lowest in meat meal, Salmonella was detected in 8.7% of the samples but did not find E. coli in any of the samples and coliforms were detected in only four samples. Additionally, Crump et al. [11] and Loken et al. [21] isolated Salmonella spp. from 14% of samples containing less than one coliform bacterium per gram. Bensink [22] found that 70% of examined meals were contaminated with Salmonella spp. and Hess et al. [18] who isolated Salmonella spp. from processing plant environment and raw material. Results showed that 35.9% of samples were positive for Salmonella spp. but the product samples collected at time of discharge from the extractor cookers were negative for Salmonella. While total bacterial counts were in the range of 107 to 108 CFU/g. of samples. The results were disagreed with Cooke [23] and Lo Fong Wong [24] who examined commercial animal feeds in several European countries and found a low level of Salmonella spp. contamination (less than one percent). Regarding fungal isolates of final products, Aspergillus fumigatus was isolated from 10% and Penicillium spp. isolated from 20%. Both can produce mycotoxins that have harmful effects if consumed by birds or animals and can accumulate and affect consumer health [25, 26].

Presence of all of these pathogens may be related to post-rendering recontamination from the environment of the rendering plant [11, 20, 27-29].

CONCLUSION AND RECOMMENDATIONS

Rendering process resulted in reducing microbial load of raw poultry by-products. Complete reduction of *Campylobacter* spp. but not *Salmonella* spp. was attained. Microbial counts present in the final product may be due to ineffective rendering process conditions or high microbial load of raw material. So, the quality of raw material and hygienic condition of processing plant should be monitored properly.

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