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The effect of chemical intervention on chicken microbial quality in Khartoum State, Sudan

Sitona Salah Ahmed Yosef and Elniema A. Mustafa*

Department of Food Safety and Veterinary Public Health, College of Veterinary Medicine, University of Bahri, Khartoum, Sudan

KEYWORDS

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ABSTRACT

This descriptive and experimental study was conducted between June and September 2020 in six poultry slaughterhouses in Khartoum State Sudan. The objectives of this study were to evaluate the effect of chemical interventions on reduction of poultry carcasses bacterial load and to assess the management measures adopted in poultry processing plants and their effects on reducing bacterial load. Slaughterhouses were classified into three groups according to the chemical interventions used. Both checklist and swab samples were applied to assess the status of the prerequisite programs (PRPs), activities during operational processes (OP) and total bacterial count (TBC). The samples were taken directly from the processing line from three steps: after final wash, after drying and after freezing. According to the checklist assessment, the study revealed that the slaughterhouses used sodium chloride failed to comply with the acceptable limit and scored 69.1%. The mean of TBC after final wash was found 183±19.6 CFU, after chemical intervention 123±24 CFU, after freezing 133.8±30 CFU with statistical significant difference 0.020 with $p \leq 0.05$. Whereas, slaughterhouses used acetic acid failed to comply with the acceptable limit and scored 73%, the mean of TBC after final wash was found 260.8±18.8 CFU, after chemical intervention 158.4±34 and after freezing 299±1 CFU with statistical significant difference 0.001, with $p \leq 0.05$.Also, slaughterhouses used hydrogen peroxide failed to comply with the acceptable limit and scored 61.9% and the mean of TBC after final wash was found 247.2±29CFU, after chemical intervention 10±10 CFU, after freezing 115.6±21 CFU, with highly statistical significant difference 0.000, with $p \leq 0.05$. This study concluded that the chemical interventions had reduced the bacterial load but the slaughterhouses need more management to minimize contamination in the final product.

**Corresponding Author*
Email Address: elniema.mustafa@hotmail.com

INTRODUCTION

The highest number of cases of food borne pathogens such as Campylobacter and Salmonella reported in European Union were largely associated with fresh poultry meat and eggs and pork (EFSA, 2011). In one hand, the decontamination of poultry carcasses is gaining increased interest, especially because poultry is implicated as a risk factor in human campylobacteriosis (Chen *et al.*, 2012).

On the other hand, operational process steps in modern poultry industry are designed to reduce bacterial contamination and extends shelf life.

In industrial poultry processing, hot water immersion is used to facilitate feather withdraw but it also reduces bacterial load accompanying that process step. Therefore, poultry carcasses should be quickly chilled by reducing their temperature from approximately 40 to 4 °C (James *et al.*, 2006). Other operational process steps such as washing and sanitizing procedures have generally proven effective for reducing overall bacterial populations as well as numbers of specific bacterial pathogens on meat (Sofos, 1994).

Chemical interventions usually applied to inhibit microorganisms because of their ability to disrupt cellular membranes or other cellular constituents and interrupt physiological processes (Loretz *et al.*, 2010).

Chemical interventions primarily comprised organic acids, chlorine-based treatments, or phosphate-based treatments. Acetic acid, acidified sodium chlorite, and trisodium phosphate can reduce bacterial load in the range from 1.0 to 2.2 orders of magnitude (Loretz *et al.*, 2010). Microbial reductions obtained for inoculated bacteria, including aerobic bacteria, non-pathogenic E. coli, E. coli O157:H7 and Salmonella spp., varied between 0.7 log and 4.9 logs (Loretz *et al.*, 2010). The reduction of bacterial load in meat carcasses by applying hydrogen peroxide in poultry chiller water was also reported by Midgley and Small (2006).

The objectives of this study were to evaluate the effect of chemical interventions on reduction of poultry carcasses bacterial load and to assess the management measures adopted in poultry processing plants in the study area and their effect on reducing bacterial load.

MATERIALS AND METHODS

Study area and design

This descriptive and experimental study was conducted between June and September 2020 in six poultry slaughterhouses in Khartoum State, Sudan. The slaughterhouses were categorized into three groups according to the type of chemical intervention currently used. Group (1) used sodium chloride with an addition rate of 0.15 g/l, group (2) used acetic acid with an addition rate of 0.2 ml/l, and group (3) used hydrogen peroxide with an addition rate of 0.05 ml/l. Each chemical intervention was investigated in 2 slaughterhouses. Both checklist and microbiological tests were used in this evaluation.

Data collection

The assessment of the status of some of the prerequisite programs (PRPs) and other related activities during operational processing (OP) that could directly affect meat safety in slaughterhouses were investigated using a checklist.

Checklist design

A structured checklist was designed with three sections. Section one comprised good manufacturing practices (GMPs) and weighed 30%, section two comprised good hygiene practices (GHPs) and weighed 30% and section three comprised operational processing (OPs) and weighed 40%.

The first section GMP consisted of building standards, separation between clean and dirty area, water proof flooring, system of ventilation, and compliance of equipment with international standards. The second section was GHP contained application of hygiene policy procedures, cleaning and waste management system, and the third section consisted of activities related to operational processing (OP) that could directly affect the safety of meat such as defeathering temperature and washing after evisceration, changing of immersion water and addition of ice in chiller tank, chemical interventions used and rate of addition, workers' movement in the slaughterhouse and their protective equipment.

Sampling

Ninety samples were collected using sterile swabs from six slaughterhouses with the purpose of evaluating total bacterial count (TBC). For each chemical intervention in each slaughterhouse a total of 15 swab samples were taken directly from the processing line from three process steps: after final washing, after chemical intervention and after freezing of the final product. The collected swabs were then aseptically transferred into sterile containers, and then kept in an icebox and transported at 4 ºC to the laboratory of the College of Veterinary Medicine, University of Bahri.

Procedure for evaluation of bacterial load

Total bacterial count was used as described by Marshall (1992). The TBC was calculated by using original dilution and then added to 9 ml normal saline for the first dilution, then 1 ml was taken from first dilution and added to 9 ml normal saline for the second dilution. Then 1 ml of the fifth dilution that prepared previously was taken and added to labeled petri dish with about 20 ml plate count agar and spread to facilitate absorption and incubated for 24 hours at 37 ºC. Bacterial colonies were counted and documented for analysis.

TBC means were compared to the Sudanese Standards and Metrology Organization (SSMO) where the acceptable total bacterial load in whole poultry carcass is supposed to be 3×105 CFU by Aeropic Plate Count (APC) test.

Statistical analysis

The obtained data were coded and analyzed by using statistical packaging for the social sciences SPSS/PC software program, version 21 for windows. Data were analyzed for descriptive statistical analysis and ANOVA test.

RESULTS

The results of this study showed that the mean TBC in the final product in all investigated broiler meat in the slaughterhouses had exceeded the acceptable limits set by SSMO.

The mean of TBC in the three process steps was found 183± 19.6 CFU/ml after final wash, 123±24.2 CFU/ml after chemical intervention (sodium chloride) and 133.83±0.7 CFU/ml after freezing with significant differences (0.02) between the three process steps with $p \leq 0.05$ (Table 1).

Table 1: Average of total bacterial count in the three process steps in the slaughterhouses using sodium chloride.

Process step	Mean	SD	SE	Sig
1- After final wash	183.667	76.199	19.674	
2- After chemical intervention	123.000	94.101	24.297	0.02
3- After freezing	133.800	119.196	30.776	

SD= Standard deviation; SE= Standard Error, Sig = Significance

Table 2: Average of total bacterial count in the three process steps in the slaughterhouses using acetic acid.

Process step	Mean	SD	SE	Sig
1- After final wash	260.900	59.748	18.894	
2- After chemical intervention	158.400	109.389	34.593	0.001
3- After freezing	299.000	3.162	1.000	

SD= Standard deviation; SE= Standard Error, Sig = Significance

The mean of TBC in the three process steps was 260.9±18.8 CFU/ml after final wash, 158.4±34.5 CFU/ml after chemical intervention (acetic acid) and 299.0±1.00 CFU/ml after freezing of the final product. The results show significant differences (0.001) between the three process steps with $p \leq 0.05$ (Table 2)

The mean of TBC in the three process steps was found 247.2±29.4 CFU/ml after final wash, 10±10.0 CFU/ml after chemical intervention (hydrogen peroxide) and 115.6±21.0 CFU/ml after freezing. There were significant differences (0.00) between the three process steps, with $p \leq 0.05$ (Table 3).

Table 3: Average of total bacterial count in the three process steps in the slaughterhouses using hydrogen peroxide.

Process step	Mean	SD	SE	Sig
1- After final wash	247.200	65.7856	29.420	
2- After chemical intervention	10.000	22.3618	10.000	0.000
3- After freezing	115.600	47.1310	21.077	

SD= Standard deviation; SE= Standard Error, Sig = Significance

Table 4: Assessment of PRPs and OP for the investigation of slaughterhouses

Chemical intervention	Assessment of the checklist sections			
	GMP	**GHP**	**OP**	**Total**
1- Sodium chloride	13.90%	22.00%	33.10%	69.20%
2- Acetic acid	26.30%	15.20%	31.50%	73.00%
3- Hydrogen peroxide	17.80	18.70%	25.00%	61.50%

PRPs= Prerequisite programs, OP= Operational processes

Table 5: Assessment of PRPs and OPs of slaughterhouses and average of TBC (±SE) in the three stages of slaughter processing

Chemical intervention	PRPs and OP assessment		The mean of TBC (CFU/ml) Process steps		
	Acceptable	**Unacceptable**	**Final wash**	**Chemical intervention**	**Freezing**
1-Sodium chloride	39.80%	61.20%	183.6 ±19	123±24	133.8 ±30
2- Acetic acid	27.00%	73.00%	260.9±19	158.4±34	299±1
3- Hydrogen peroxide	38.50%	61.50%	247.2±29	10±10	115.6±21

PRPs= Prerequisite programs, SE= Standard error of mean, OP= Operational processes, TBC= Total bacterial count, CFU= Colony forming unit

The assessment of the status of PRPs and their effects on operational processing in terms of bacterial load is shown in Table (5). The PRPs assessment though found unacceptable with regard to the three chemical interventions, yet their effect on bacterial load showed a decrease in TBC after final wash step and an increase after freezing process step.

DISCUSSION

The objectives of this study were to evaluate the effect of chemical interventions on reduction of poultry meat bacterial load and to assess the management measures adopted in poultry processing plants and their effects on reducing bacterial load.

Microorganisms are considered good indicators of the hygienic conditions present during food manufacturing process (Hoffmann *et al.*, 2004). For example, high coliform counts indicate post-processing contamination and/or unsuitable sanitization (Kottwitz *et al.*, 2010).

This study investigates the use of different chemical interventions such as sodium chloride, acetic acid and hydrogen peroxide that were added to the pre-chilling and chilling water tanks. It was revealed that all chemical interventions used had significantly reduced the TBC of the product. This will eventually improve the quality and shelf life of product. This is also stated by Guastalli *et al.* (2016) who reported that chemicals were added to the pre-chilling and chilling water tanks with the purpose of minimizing carcass bacterial load.

In the present study chemical intervention using sodium chloride after the final wash reduced TBC, though TBC increased after freezing, with significant differences (0.02) between the three process steps, with $p \leq 0.05$. The increase of TBC observed after freezing may be due to inadequate sanitation in this process step.

This study also shows that chemical intervention using acetic acid after the final wash reduced TBC, despite that the TBC increased after freezing, with significant differences (0.001) between the three process steps, with $p \leq 0.05$. Dickson and Anderson (1992) attributed the reduction of bacterial load when using acetic, lactic and citric acids to the fact that these acids have bactericidal effects.

The increase in TBC after freezing may be attributed to inadequate sanitation in this process step. Moreover, EFSA (2011) stated that acetic acid, which is a natural component of vinegar, is not expected to raise any safety concern (Midgley and Small, 2006).

Research studies indicating the efficacy of acetic acid in reducing bacterial load in meat were also conducted by Loretz *et al.* (2010) who evaluated acetic acid on inoculated beef carcass surfaces under laboratory conditions. They found microbial reductions obtained for inoculated bacteria, (including aerobic bacteria, nonpathogenic *E. coli*, *E. coli* O157:H7, and *Salmonella* spp.) varied between 0.7 log and 4.9 logs.

This study also shows that chemical intervention using hydrogen peroxide after the final wash reduced TBC despite its increase after freezing with significant differences (0.000) between the three process steps, with $p \leq 0.05$. The increase in TBC after freezing may be due to inadequate sanitation and the probable cross contamination that may occur in this process step. This finding was in line with those published by Gorman *et al.* (1995) who stated that decontamination can be accomplished with the use of hydrogen peroxide which inactivates microorganisms by acting as an oxidant. Similar finding was reported by Reagan *et al.* (1996) who studied some procedures that included hydrogen peroxide (5%) as intervention treatment on beef carcasses and revealed that hydrogen peroxide reduced aerobic plate counts by 1.14 log cfu/cm². The reduction of bacterial load in meat carcasses by applying hydrogen peroxide in poultry chiller water was also reported by Midgley and Small (2006**)** who found that hydrogen peroxide as a bactericide reduced aerobic organisms by 95–99.5% with 6,600 ppm or higher and *E. coli* by 97–99.9% with 5,300 ppm or higher.

It worth noting in this study that the high microbial load in the final product after the freezing step may be due to inadequate hygiene activities which applied in slaughterhouses such as cross contamination from the hands of workers. In addition, the researchers observed poor sanitation and cleaning and disinfection of freezers and tools. This complied with that stated by Sheridan *et al.* (1992) who identified personal equipment, such as knives, mesh gloves, and aprons as reservoirs of bacteria in the abattoir.

Assessment of the status of PRPs and their effects on operational processing in terms of bacterial load in the present study revealed low acceptable scores. Despite this nonconforming PRPs, the three process steps showed a decrease in TBC in chemical intervention process steps. These findings proved the significance of chemical intervention in reducing bacterial load in meat carcasses. The studies of Loretz *et al.* (2010) and Dickson and Anderson (1992) also attributed the decrease in TBC by applying chemical intervention to the fact that using sanitizing agents have generally proven effective for reducing overall bacterial populations as well as numbers of specific bacterial pathogens on meat.

CONCULISION AND RECOMUNDITIONS

This study concluded that using chemical intervention after the final wash reduced TBC in broiler carcasses and that there was an increase in TBC after freezing process step. To avoid high bacterial count in the final product, good hygiene measures must be applied.

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