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IDENTIFICATION OF *SALMONELLA* LOAD IN BROILER PRIMARY PRODUCTION AND PROCESSING IN BAHRI LOCALITY – SUDAN

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ABSTRACT

This study aimed to evaluate the hygienic biosecurity practices of *salmonella* contamination in primary production and processing. A total of 32 samples from two broiler farms and their slaughterhouses were randomly collected in Bahri locality, Sudan. Of these, 8 samples from broiler farms (4 feacal, 2 composite litter, 2 boots sock) and 24 samples from slaughterhouses (12 pre-chill carcasses and 12 post-chill carcasses). *Salmonella* load was carried out in selective media (XLD) after enrichment in Rapapport vasilidus broth. The percentage of *Salmonella* load from the total number of samples was (34.38%). The result represented 5 (62.5%) *Salmonella* enumerate from farms samples, 1 (20%) from feacal, 2 (40%) from composite litter and 2(40%) from boot sock. Six (25%) *Salmonella* enumerate were obtained from slaughterhouses of which 4 (66.7%) from pre-chill carcass and 2 (33.3%) from post-chill carcasses. With respect to farms samples, boot socks and litter yielded the highest percentage of positive results for *Salmonella* load when compared with feacal samples. In contrast pre-chill carcasses samples were slightly higher in load than post-chill carcasses. There was a significant relationship of *Salmonella* load in farm and slaughterhouse. Also, the results revealed that the load of *Salmonella* decreased during different stages of processing plant.

INDEX: Salmonella, Broiler farm, Poultry slaughterhouses.

I. INTRODUCTION

The incidence of *Salmonellosis* has always been a major concern for broiler consumers in many developing countries including India, Egypt, Brazil and Zimbabwe[1] and is the most seriously perceived food risks in chicken meat, even in the developed countries.[2]

Campylobacter and *Salmonella* infections from poultry ranked first and fourth, respectively from the perspective of illness cost and loss of quality-adjusted life years.[3] Studies reported that poultry meat and eggs are vehicles for *Salmonella* transmission to humans.[4].[5] stated that chickens less than14 days are extremely susceptible to infection by *Salmonella* spp. Infected day-old-chick flocks can rapidly spread *Salmonella* throughout the poultry house and the rest of the birds will ingest the bacteria and become infected in few days of rearing.[6]

One of the main causes for hindering the diagnosis of *Salmonella* at farm level is that poultry are often unapparent carriers.[7] Despite this, the organism can be directly diagnosed at farm level or at the slaughterhouse by isolation of the bacterium.

Sanitary measures to ensure food safety throughout the production, processing and marketing chain is important for consumers' health. *Salmonella* load on the farm was significantly associated with load of the same pathogen at processing. Strict biosecurity measures and sanitation, among other practices, are recommended by U.S. Department of Agriculture's Food Safety Inspection Service for pre-harvest pathogen control in broiler chickens.[8] Implementing stringent biosecurity measures will prevent the adverse effects of pathogenic microorganism on poultry farms.[9] To reduce pathogenic microorganism in poultry carcasses, hazard analysis and critical control point (HACCP) shall be applied.[10]

II. MATERIALS AND METHODS

Areas and type of samples collected

A total of 32 samples were randomly collected from two broiler farms having their own slaughterhouses in Bahri locality, Sudan during the period between May to June 2017**.** These samples comprised 4 feacal samples, 2 composite litter samples and 2 samples of boot sock from the farms, in addition to 12 pre-chilled and 12 post-chilled carcasses rinses from the poultry slaughterhouses (Table 1).

Table 1: Site and type of samples collection.

Source	Feacal	Composite litter	Boot swab	Pre-chill carcasses	Post-chill carcasses
Farm 1	2	1	1	-	-
Farm 2	2	1	1	-	-
Slaughterhouse 1	-	-	-	6	6
Slaughterhouse 2	-	-	-	6	6

Feacal and Litter Samples: *Salmonella* bacteria were enumerated according to ISO 6579:2002. By the pre-enrichment in peptone water and enrichment in Rapapport vasilidus broth after created a 5-fold dilution series, the cultures from all samples were plated onto XLD agar and incubated at 37°C for 24 hour under aerobic conditions.

Carcasses Samples: Method of carcass culturing was done according to ISO 6579:2002. Twenty five grams of sample were cut and weighted, and then 250 ml of the buffered peptone water was added. The samples were put in mechanical shaker for 1 minute to ensure consistent agitation[11] and then put in incubator at 24°C for 18-24 hours. Then one ml from the culture was added to 9 ml Rapapport vasilidus broth and incubated overnight at 41°C. Then 10 micro-liter was streaked onto XLD plates and incubated at 37°C for 24 hours under aerophilic conditions.

The plates with feacal, boot sock, litter and carcass bacterial cultures were then examined for morphologically typical *Salmonella* colonies (pink colonies with black center). Presumptive colonies were confirmed by Gram-stain, motility test, oxidase test, catalase test, Citrate test, Indole test, Methyl red test, Hydrogen sulphide production and Voges - Proskauer test.

III. RESULTS

Salmonella enumerate load in environmental and processing plant samples

In this evaluation a total of 5 *Salmonella* enumerate were obtained from 8 environmental samples and a total of 6 *Salmonella* enumerate were obtained from 24 (pre-and post carcasses rinses) of processing plant samples performed on birds from the same flocks (Table 2).

Table 2: *Salmonella* enumerate from farms and slaughterhouse samples.

Type of sample	Total number	Positive results
Farm samples	8	5 (62.5%)
Slaughterhouse samples	24	6 (25.0%)

The cultural characteristics, bacterial morphology and biochemical reactions results are shown in Table (3).

Table 3: Cultural characteristics, bacterial morphology and biochemical reactions of *Salmonella* spp.

Type of the test	Result
Aerobic growth	+
XLD	Pink with black in center
Nutrient agar	Circular, smooth, opaque
Gram stain	-
Shape	Rod
Motility	+
Catalase	+
Oxidase	-
Indol	-
Methyl red	+
V/P	-
Citrate	+
H2S	+
OF	+

***Salmonella* load in different environmental samples**

With respect to farms samples; litter 2 (100%) with mean (SD) log10MPN 4.06, and boot socks 2 (100%) with mean (SD) log10 MPN 4.7 yielded the highest percentage of positive results for *Salmonella* load than feacal samples 1 (25%) with mean (SD) log10 MPN 2.2. The Mean and Standard Deviation of *Salmonella* load in environmental samples in the two farms is shown in Table (4).

Table 4: The mean and standard deviation of *Salmonella* load in environmental samples.

Type of samples	Farms	N	Mean	Std. Deviation
Feacal sample	Farm 1	2	4.4	3.1
	Farm 2	2	.0000	.00
Litter sample	Farm 1	2	5.1	3.59
	Farm 2	2	4.3	3.07
Boot sock sample	Farm 1	2	4.6	3.26
	Farm 2	2	4.6	3.23

Boot socks, litter, and fecal samples, all had similar sensitivities for detecting *Salmonella* on the farm. There was insignificant difference in the mean of *salmonella* count between feacal, litter and boot samples with $p > 0.05$ (.423, .922 and .995, respectively) as evident in Table (5).

Table 5: Paired sample t test of *salmonella* load in feacal, litter and boot sock samples.

Type of sample	Variances	Sig. (2-tailed)	Mean Difference
Feacal sample	Equal variances assumed	.423	2.19
Litter sample	Equal variances assumed	.922	.36950
Boot sample	Equal variances assumed	.995	.02250

***Salmonella* enumerate in processing plant samples**

In this evaluation, a total of 6 (25%) *salmonella* enumerate were obtained from 24 pre-and post carcasses rinses. Pre-chill carcasses (66.7%) were higher than post-chill carcasses (33.3%) as shown in Figures (1 & 2).

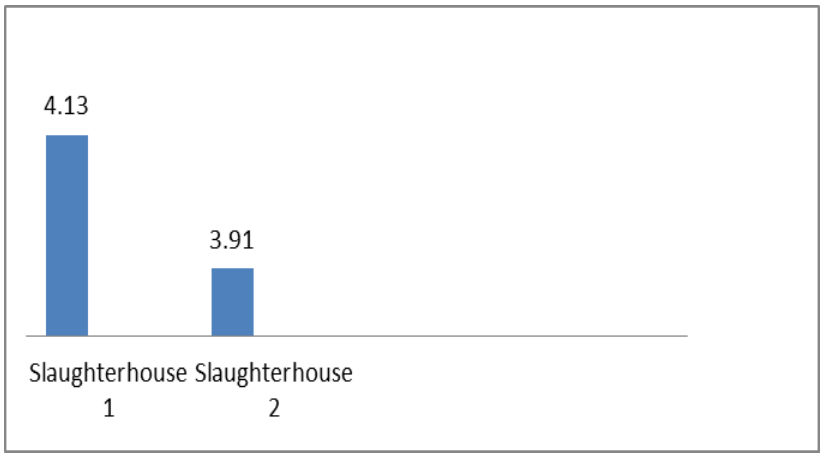

Figure 1: The mean distribution of *Salmonella* load in pre-chill carcasses rinses.

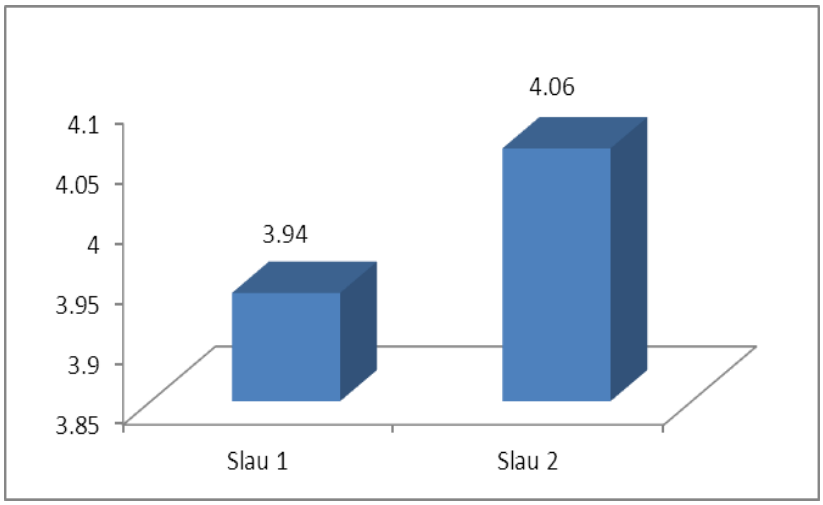

Figure 2: The mean distribution of *Salmonella* load in post-chill carcasses rinses.

According to the cultural characteristics, bacterial morphology and biochemical reactions, the mean and standard deviation are shown in Table (6).

Table 6: The Mean and Standard Deviation of *Salmonella* load in processing plant samples.

Type of sample	Slaughterhouse	N	Mean	Std. Deviation
Pre-chill carcass rinses	No. 1	6	4.13	.17290
	No. 2	6	3.91	.26904
Post-chill carcass rinses	No. 1	6	3.94	.35815
	No. 2	6	4.06	.15377

Paired sample t test revealed that there was insignificant difference in the two samples of pre and post-chilling process steps p >0.05(0.89) (Table 7).

Table 7: Paired sample t test of *Salmonella* load in the pre and post chill carcasses rinses.

Type of samples obtain from pre and post-chill carcasses	Mean	Sig. (2-tailed)
The *Salmonella* mean (pre chill carcass) and *Salmonella* mean (post chill carcass)	.0156	.89

Comparison between *Salmonella* load in broiler farms and slaughterhouses

A greater proportion of variability in carcass rinse loads for *Salmonella* was examined in farm samples. The load of *Salmonella* in broiler farms samples (62.50%) were higher than those found in slaughterhouse samples (25.00%) as appears in Figure (3).

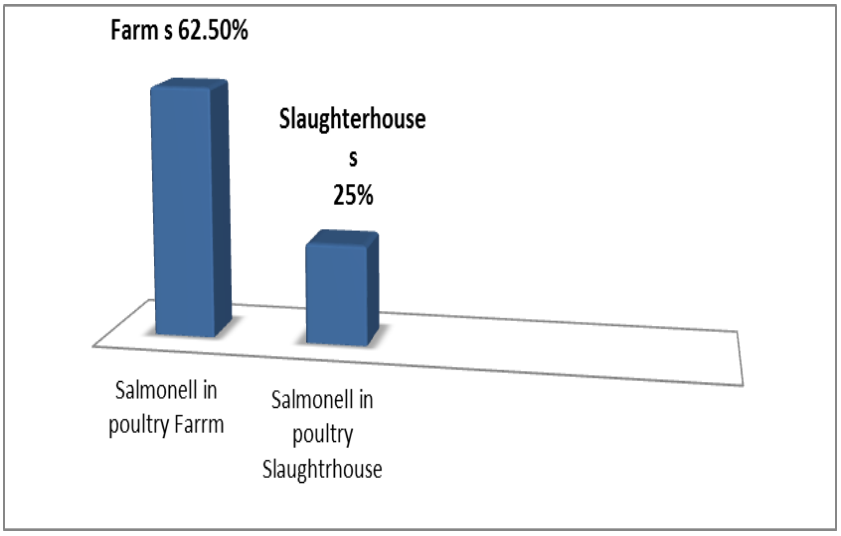

Figure 3: Comparison between *Salmonella* load in the farm and slaughterhouse samples.

IV. DISCUSSION

In this study farms samples, boot socks and litter yielded the highest percentage of positive results for *Salmonella* load when compared to feacal samples. This may be attributed to improper cleaning and disinfection in the rest period. Lower findings were reported by[12] who recorded 68% for boot sock, 22.7% for litter and 13.6% for feacal samples.

In the present study the *Salmonella* load in feacal samples was considered higher. The reason may be attributed to high excretion of *Salmonella* usually occurs in 2 weeks of rearing, coinciding with an immature immune system of chicks.[13]

The result of this study revealed a relationship between *Salmonella* load in farms that was reflected in the final product in the slaughterhouses. The findings of this study revealed that *Salmonella* load in pre-chill carcasses and post-chill carcasses were highest in slaughterhouse (1) than that of slaughterhouse (2). The reason may be due to the highest percentage of *Salmonella* load recovered in feacal samples in farm (1). These findings were similar to those reported by[14-15] who revealed significant relationships between farm and processing plant pathogen prevalences in broiler chicken flocks. Such relationship was also studied by[16] on *Salmonella* in bird and environmental samples and carcass rinses from the same flocks at processing.

[12]reported that *Salmonella* loads decreased as birds progressed through the processing plant. This is because the organism is sensitive to the effect of freezing which can cause a 1-2 log reduction in the level of contamination on poultry meat.[17] The results of the current study indicated that *Salmonella* load in post-chill carcasses was lower than pre-chill carcasses. Much lower loads were obtained by[18] who found 18.2% for per-chill carcasses and 2.4% for post-chill carcasses. Reductions of 52% in *Salmonella* prevalences between re-hang and post-chill carcass rinses were also observed by.[19-20].[21] also reported overall reductions of 0.5 log10 for *Salmonella* in carcass rinses before and after chilling.

The present study showed that the *Salmonella* load in broiler carcasses was higher than that mentioned by[22] who reported 1.6%,[23] in Morocco (2.08%),[24] (1.1%) in Italy. The high numbers of *Salmonella* in this study may be attributed to the poor hygienic measures and improper temperature control in the investigated slaughterhouses. Nevertheless, the results of the present study showed that the *Salmonella* load in broiler carcasses was lower than that reported by[25] in Morocco (57%),[26] in Vietnam (62.79%),[27] in Nepal (46.2%),[28] in Thailand (67.5%) and[29] in India (100%).

V. CONCLUSION AND RECOMMENDATIONS

In conclusion, *Salmonella* load on the farms was found to be associated with load of the same pathogen at processing. Consequently, management practices that reduce pathogens on the farm would be expected to reduce contamination at processing. It is recommended that strict biosecurity measures must be implemented in the farms during rearing time and between successive flocks. Application of HACCP program in the broiler slaughterhouses to improve the safety of broilers production is of paramount importance.

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