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Full Length Research Paper

The effects of residual blood of carcasses on the microbiological quality of poultry

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To prove the residual blood factors that are responsible for high contamination of carcasses, this study was conducted to compare the microbial level in poultry both perfectly and imperfectly bled. At 3°C storage temperature, the average number of bacteria of perfectly bled poultry was 7.05×10^2 ufc/g which was significantly (P≤ 0.05) lower than the average number of bacteria of imperfectly bled poultry ($1 .78 \times 10^4$ ufc/g). At 7°C storage temperature, the average number of bacteria of perfectly bled poultry was $6 .18 \times 10^3$ ufc/g which was significantly (P≤ 0.05) lower than the average number of bacteria of imperfectly bled poultry (3.60×10^4 ufc/g). The meat samples were taken from the chest and thigh to reflect the heterogeneity of the contamination. The rise in the storage temperature increased the bacteria in the two types of poultry; but the number of bacteria of perfectly bled poultry was still minimal. The residues of blood increased the degree of contamination of carcasses. It was found out that there were effects of season on the results of 80 samples from chicken carcasses (n = 80; 40 were used in the winter and 40 in the summer).

Key words: Residual blood, total aerobic mesophilic flora (TAMF), perfectly bled poultry, imperfectly bled poultry.

INTRODUCTION

Very little attention had been directed towards studying the effects of residual blood content and its relationship with microbial growth in the muscle. Since blood is a perfect medium for microorganisms' growth, if increased levels of residual blood were present or localized in a muscle, a corresponding susceptibility to microbial contamination may, therefore, exist (Gill and Newton, 1978). However, Warriss (1977) showed that there is no evidence that differences in meat composition do not affect the spoilage flora, nor does the failure to bleed animals alter meat qualities in a manner which affects bacterial growth. In contrast, it had been suggested by Thorton (1949) and Hess (1968) that the flesh of a carcass which was imperfectly bled must be consumed immediately because of rapid decomposition of meat qualities.

This was attributed to an increased muscle pH, caused by residual blood which favours the growth of bacteria (Gotze, 1969; Heinz et al., 1972). Furthermore, Lawrie (1966, 1977, and 1979) suggested that the delay in the bleeding of slaughtered animals caused bacteraemia even in healthy animals, and also suggested that increased residual blood content in muscles promoted spoilage, gamey flavours and discoloration. The circulating blood in a healthy animal at slaughter time is virtually free of microorganisms (Gunstone, 1980; Dill, 1976), and precautions against contamination should be taken when blood is collected, for food purposes (Richards, 1970). Gunstone (1980) proposed the aerobic count of no more than 100,000 organisms/ml is set as a standard number for blood plasma. Tybor et al. (1975) and Dill (1976) indicated that no pathogenic organisms were found in freshly collected plasma and reported low aerobic counts for blood plasma.

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Table 1. Experimental general plan.

Parameter	Season							
	Winter				Summer			
Number of experience	1		2		3		4	
C.C. T	3°C		7°C		3°C		7°C	
Type of slaughtering	PB	IB	PB	IB	PB	IB	PB	IB
Number of carcasses samples	n=10	n=10	n=10	n=10	n=10	n=10	n=10	n=10

n, The number of samples; PB, perfectly bled poultry; IB imperfectly bled poultry.

Jay (1978) and Empey and Scott (1939) noted that the contact of knife and meat can cause infection in muscle tissue (bacteraemia) from an organism entering into the bloodstream when touching the carotid and jugular vein. From observations of previous authors, we built our hypothesis which was based on the fact that the presence of residual blood in the carcasses would result in an increase in the concentration of microorganisms in imperfectly bled carcasses compared to perfectly bled ones. Then, an approach must be developed to avoid problems associated between residual blood and meat microbiology.

The discharge of the maximum quantity of blood leads to a suitable control of meat microbiology and eliminates many problems associated with meat quality. Since limited research has been conducted regarding the effects of residual blood of carcasses on meat quality, this study was designed to determine the residual blood on carcasses in the effect on the meat microbiology, to demonstrate the relationship between meat residual blood on carcasses and the degree of bacterial contamination. Also, to compare the decline of total aerobic mesophilic flora (TAMF) in perfectly bled meat, with imperfectly bled meat and to identify the associated residual blood factors that is responsible for high contamination of poultry carcasses.

MATERIALS AND METHODS

This study aimed to investigate the influence of residual blood on the post mortem evaluation of TAMF. Large numbers of chickens are slaughtered in two different ways: first, slaughter without stunning for a maximum discharge of blood, that is perfectly bled (PB). The second way is to slaughter by light lead ball to keep nearly all the blood in the carcass; that is imperfectly bled (IB). The concept of microbiological meat quality has two notions. The first one is the absence of sensory degradation products due to the presence of spoilage microorganisms, and the second is, there is no risk of intoxication for the consumer because of the presence of pathogenic microorganisms. Since this study is to evaluate the hygienic quality of the perfectly bled poultry with the imperfectly bled one, the method of controlling the hygienic quality was used.

This is based on the enumeration of TAMF, which shows the degree of bacterial contamination of meat overall (Roberts, 1980), and which was used as a method to control the hygienic quality of carcasses (Cartier, 1993). The degree of bacterial contamination of meat allows comprehensive changes of fluorometric microvolume assay technology (FMAT) in two substrates in a selective manner, and therefore selects the substrate which is more favourable to the proliferation of germs. The enumeration of TAMF was used to determine the sanitary quality of poultry products and can indicate its state of freshness or decomposition. The total flora count was used to judge the conditions in which the poultry was produced, transported, and stored. The study requires a large number of animals, so this study uses poultry as biological material In addition.

Conditions of pre-slaughter and slaughter

For the chickens in the same conditions before and after slaughter to prevent the intervention of certain intrinsic and extrinsic factors, and many other physicochemical factors, such as temperature, oxygen availability, storage mode and the type of lighting, the plucking was made by soaking the chicken in hot water (about 50°C). Samples for microbiological analysis were collected immediately after evisceration, by excision, with a sterile scalpel, placed separately in sterile bottles, kept cold in an insulated container and returned to the laboratory for bacteriological analysis. The samples were taken from inside the muscles to avoid contamination of the surface which is exposed to the highest values of contamination (Anderson et al., 1987; Lasta and Fornoug, 1988). Samples were collected from the chest and thigh to reflect the heterogeneity of the contamination of carcasses (Le Touze et al., 1985; Lasta et al., 1988, 1992; Cartier, 1993). For each experiment, a sample of 100 g was used, they were aseptically cut and then carefully milled by using the grinder ultra-turax. Each sample was divided into 10 portions of 10 g. 1 ml of the previous preparation and successive dilutions was cultured in depth in a sterile Petri dish, was added to 15 ml of culture medium plate count agar medium (PCA) and incubated at 30°C for 72 h, according to the standard test (ISO 4833-2008 NM).

The cultivation was performed for 24 h for post slaughter; half of the samples were stored at 3°C, and the other at 7°C .The effects of carcasses residual blood on the microbiological poultry quality by the two experiments was evaluated. In the first experiment before evaluation, the samples were stored at 3°C; the growth of TAMF of the perfectly bled poultry with the imperfectly bled one was evaluated for the comparison between the changes of 2,2,2-trifluoroethyl methacrylate (TFMA) in the two substrates. In the second experiment, the same comparison was realized with samples stored at 7°C. Increases in the number of FMAT in the summer led us to see the effect of season on the results; that is why the study made the same comparison in summer (experiments 3 and 4).

A total of 80 samples from chicken carcasses (n = 80, 40 per treatment) were used to test the effects of residual blood ($P \leq 0.1$), as shown in Table 1. The term imperfectly bled was used by Thorton

Table 2. Number of TAMF (CFU / g) recorded in the carcasses poultry perfectly bled and imperfectly bled after 24 h of storage.

Parameter	Winter				Summer			
	1		2		3		4	
	3°C		7°C		3°C		7°C	
	PB	IB	PB	IB	PB	IB	PB	IB
TAMF (CFU/g)	4×10^2	4.1×10^4	1.47×10^3	$3.5 \times 1\ 0^4$	2.5×10^2	5.7×10^4	5×10^7	1.16×10^8
	2×10^2	5.6×10^4	4.04×10^4	5.3×10^3	5.6×10^2	7.6×10^3	10^5	7.68×10^7
	3.2×10^2	5.2×10^4	4.9×10^3	5.52×10^4	7.6×10^2	1.36×10^4	1.6×10^5	7.25×10^7
	10^3	8.5×10^4	6.15×10^3	5.13×10^4	3×10^2	5.4×10^3	10^4	1.05×10^8
	1.2×10^3	3.6×10^4	5.52×10^3	1.4×10^4	0.3×10^2	3.2×10^3	2×10^4	7.2×10^8
	8.50×10^2	5.8×10^4	8.50×10^2	4.9×10^4	9.97×10^2	3.61×10^4	2×10^5	3.51×10^7
	6.00×10^2	4.2×10^4	6.95×10^2	5.02×10^4	$1\ .20 \times 10^3$	4.69×10^4	2×10^5	96×10^8
	10^3	8.06×10^4	1.5×10^3	4.05×10^4	1.2×10^3	3.61×10^4	10^4	9.8×10^7
	3.5×10^3	10^4	4×10^3	1.65×10^4	5.60×10^2	1.20×10^4	9×10^4	4.69×0^8
	9.00×10^2	3.2×10^4	10^3	4.40×10^4	8×10^2	1.5×10^4	6×10^4	4.92×10^7

PB, Perfectly bled poultry; IB, imperfectly bled poultry; TAMF, total aerobic mesophilic flora.

Table 3. Means of TAMF (CFU/g) recorded in the carcasses poultry perfectly bled and imperfectly bled after 24 h of storage.

Parameter	Winter				Summer			
	3°C		7°C		3°C		7°C	
	PB	IB	PB	IB	PB	IB	PB	IB
Minimum	6.66×10^2	1.81×10^4	6.65×10^3	3.61×10^4	9.97×10^2	4.92×10^4	5.10^6	2.70×10^8
Mean of TAMF (ufc/g)	7.05×10^2	$1\ .78 \times 10^4$	$6\ .18 \times 10^3$	3.60×10^4	9.24×10^2	4.85×10^4	4.62×10^6	2.93×10^8
Maximum	1.2×10^3	4.69×10^4	4.04×10^4	5.52×10^4	3.5×10^3	8.5×10^4	5×10^4	9.60×10^8
ISO 4833-2008	5.10^5 - 5.10^6							

n, The number of samples; PB , perfectly bled poultry; IB, imperfectly bled poultry; TAMF, total aerobic mesophilic flora.

(1949) and Hess (1968).

Data analysis

Data were statistically analysed using means and standard error of means (Table 3), the effects of residual blood of carcasses on TAMF was determined by student t-test and correlation coefficient matrix of meat quality by Pearson coefficient correlation, all analysis were performed by using SYSTAT 10.2. Version 7600.LSD values at $p< 0.01$ and $p<0.05$ were obtained for comparison of mean values (Steel et al., 1997).

RESULTS AND DISCUSSION

The effect of residual blood of carcasses on the microbiological quality of poultry is reported in Table 2. In winter and at 3°C as in the first experiment, the average number of TAMF of perfectly bled poultry was 7.05×10^2. The average number of TAMF of imperfectly bled poultry was 1.78×10^4. Paired samples t test showed that the residuals bloods differed significantly ($P<0.01$). At 7°C, in the second experiment, the average number of TAMF in perfectly bled poultry was 6.18×10^3. The average number of TAMF in perfectly bled poultry was 6.18×10^3. The average number of TAMF in imperfectly bled poultry was $3.6x10^4$. Paired samples t test showed that the residuals bloods were differed significantly ($P<0.05$). In summer at 3°C, in the third experiment, the average TAMF of perfectly bled poultry was 9.24×10^2 and the average TAMF of imperfectly bled poultry was $4.85x10^4$. Residuals bloods differed significantly ($P<0.01$). At 7°C, in the fourth experiment, the average pHu in perfectly bled poultry was 4.62×10^6, and the TAMF in imperfectly bled poultry was 2.93×10^8. Paired samples t test showed that the residuals bloods differed significantly ($P<0.01$).

According to Goepfert (1975, 1977), Gray and Johnson (1976) and Duitschaever et al. (1973, 1976), the incident of aerobic bacteria was attributed to contamination during collection, processing and post-processing procedures. Samples must have aerobic plate counts between 5.10^5 and 5.10^6 ufc/g (ISO 4833-2008). In the winter at 3°C, in

Table 4. The effect of season on quality of poultry.

Parameter	Season			
	Winter		Summer	
Number of cases	n = 10	n = 10	n = 10	n = 10
Number of experience	1	2	3	4
Minimum	30	6.95×10^2	2×10^2	10^3
Maximum	1.2×10^3	4.04×10^4	3.5×10^3	5×10^4
Mean of TAMF for PB	7.05×10^2	6.18×10^3	9.24×10^2	4.62×10^6
Minimum	3.2×10^3	5.3×10^3	10^4	$35,18 \times 10^6$
Maximum	4.69×10^4	5.52×10^4	8.5×10^4	9.60×10^8
Mean of TAMF for IB	1.78×10^4	3.60×10^4	4.85×10^4	2.93×10^8

n, The number of samples; PB, the perfectly bled poultry; IB, the imperfectly bled poultry; TAMF, total aerobic mesophilic flora.

the first experiment, the comparison between the number of FMAT in the perfectly bled poultry with the number of TAMF in imperfectly bled poultry showed that the average number of TAMF of perfectly bled poultry (7.05 × 102 ufc/g) was lower than the average number of FMAT of imperfectly bled poultry (1 .78 × 104 ufc/g). At 7°C, in the second experiment, the average number of TAMF in perfectly bled poultry (6 .18 × 103 ufc/g) was lower than the average number of TAMF of the national and international standards (5.10^5 and 5.10^6 ufc/g) (ISO 4833-2008).

The average number of TAMF in imperfectly bled poultry (3.60×10^4 ufc/g) was also consistent with the national and international standards, but with a higher level compared with the carcasses of perfectly bled. At 7°C, in winter, the hygienic quality of perfectly bled meat was much better compared to that of the imperfectly bled samples. In summer, the results were comparable with national and international standards. At 3°C, in the third experiment, (Table 1) the average number of TAMF in perfectly bled poultry ($9.24x10^2$ ufc/g) was lower than the average number of TAMF of the national and international standards (5.10^5 and 5.10^6 ufc/g) (ISO 4833-2008). The average number of TAMF in imperfectly bled poultry (4.85×10^4 ufc/g) was also consistent with the national and international standards, but with a higher level compared with the carcasses of perfectly bled. At 3°C in summer, the hygienic quality of perfectly bled meat was much better compared to that of the imperfectly bled one.

Despite the hot summer temperature conditions, the hygienic quality of the perfectly bled meat was much better compared to that of the imperfectly bled one. Also, the storage at 3°C allowed the results to remain standard despite the increase in figures in comparison with winter. At 7°C, in the fourth experiment, the average number of TAMF in perfectly bled poultry (4.62×10^6 ufc/g) was adequate than the average number of TAMF of the national and international standards (5.10^5 and 5.10^6 ufc/g) (ISO 4833-2008). The average number of TAMF in imperfectly bled poultry (2.93×10^8 ufc/g) was not consistent with the national and international standards, having a higher level compared to the carcasses perfectly bled. At 7°C in summer, the perfectly bled meat figures were lower than the IB one although both of them remained higher than standard-figures.

Effect of season on the microbiological quality of poultry

The values for TAMF were significantly (P<0.01) higher in summer than winter in the two types of slaughter (PB and IB) (Table 4). The sample size for each season within the type of slaughter was 20 individual; 10 in experience 1 and 10 in experiment 2 in the winter; 10 in experiment 3 and 10 in experiemnt 4 in the summer. Each mean represents the mean of 10 replications. Means were significantly different (p< 0.01).

Effect of conservation carcass temperature on the microbiological quality of poultry

The values for TAMF were significantly (P<0.01) higher in 7 than 3°C in two types of slaughter (PB and IB) (Table 4). The sample size for each conservation carcass temperature within the type of slaughter was 20 individual, 10 in the experience 1 and 10 in the experience 2 for 3°C; 10 in the experience 3 and 10 in the experience 4 for 7°C. Each means represent of 10 replications. Means are significantly different (p< 0.01). These experimental results confirm the observations and assumptions made by Hess (1968) and Lawrie (1966, 1977 and 1979). A rise in the storage temperature increased the TAMF in the two

types of meat but the number of total aerobic mesophilic flora of the perfectly bled meat was still minimal. The comparison of these results (Table 4) show not only the strong relationship between carcass residual blood and the growth of the degree of bacterial contamination, but also indicates the season effect. The lowest level of contamination with the TAMF was recorded during the winter. After this period the values increased to reach the highest rate in the summer, especially at 7°C storage. Results of this study were similar to those reported by Lasta et al. (1992) for TAMF. The residual blood in meat carcasses was responsible for high values of total aerobic mesophilic flora, and hence the deterioration of the quality of meat. Carcasses with less residual blood had lower values of total aerobic mesophilic flora, and therefore better quality.

Conclusion

At the end of the study, it was concluded that carcasses containing residues of blood were not of a good microbiological quality. Residual blood in meat carcasses is responsible for the high values of TAMF. Blood residues in carcasses after slaughter form a serious risk to the health of consumers and consequently, such carcasses should be removed from the market. Carcasses with less residual blood have lower values of TAMF and therefore better quality. The level of contamination of carcasses depends not only on hygiene during slaughter or the state of the live animal as reported by several authors, but also on the ambient temperature as well as the blood remaining in the carcass. Excessive bleeding caused by different techniques of stunning and slaughter, mentioned by some authors, increases the bacterial level of carcasses especially in the case of increasing the storage temperature and in the hot season. Reducing the microbial level of carcasses would absolutely be possible by applying good slaughter practices without stunning and with storage below 3°C.

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