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Research Article

Effect of Poor Post-Slaughter Handling on Physicochemical and Microbial Quality of Fresh Broiler in Bangladesh

Md. Kamruzzaman, Md. Rasel Islam, Ananya Raiyan, Md. Mohasin Hossain, Md. Ashrafuzzaman Zahid and Rashida Parvin*

Department of Nutrition and Food Technology, Faculty of Applied Science and Technology, Jashore University of Science and Technology, Jashore-7408, Bangladesh

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***Corresponding author:** Rashida Parvin, Department of Nutrition and Food Technology, Faculty of Applied Science and Technology, Jashore University of Science and Technology, Jashore-7408, Bangladesh, E-mail: rakhiparvin@yahoo.com

ORCiD: https://orcid.org/0000-0002-4091-6115

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Abstract

The slaughtering method and post-handling play an important role in meat processing. Countries followed different slaughtering methods, post-handling, and preservation techniques for post-slaughtered meat. Stress-oriented slaughtering, unhygienic handling, processing, and improper preservation are triggering factors of rapid oxidation and microbial proliferation in meat during storage. Bangladesh is a Muslim-oriented country but many vendors do not follow the halal slaughtering, and handling of meat in an unhygienic way due to a lack of food safety knowledge. Thus, the study aims to assess the effect of poor post-slaughtering handling practices on the physiochemical properties which include moisture, ash, protein content, fat content, drip loss, cooking loss, water holding capacity, pH, fat oxidation, color, heme iron, mineral content, and microbial qualities of fresh samples by total plate count during 0 days, 15 days and 30 days frozen storage at -18°C. The live broiler was purchased randomly and slaughtered instantly from different slaughter shops in Jashore City, Bangladesh. The laboratory analysis was performed following the standard method of meat sample analysis. The outcome of this study revealed that heme iron content, minerals like Ca, Fe, Zn, and lightness values decreased significantly ($p < 0.05$) during 30 days of storage at -18 °C. Post-handling had no significant effect ($p > 0.05$) on protein and ash content at 0 and 15 days of storage but slowly decreased with increasing storage time. The continuous increase in pH, cooking loss, and TBARS value indicate rises in lipid oxidation noticed in all samples, especially during 16-30 days of frozen storage ($p < 0.05$). The total viable count significantly raised ($p < 0.05$) and reached the highest value of 7.27 log10 CFU/g and 7.44 log10 CFU/g respectively after 15 days and 30 days of frozen storage compared to fresh condition. Bacteriologically meat samples were of very poor quality during storage compared to fresh ones.

Introduction

Meat is considered a valuable food in terms of nutrients, and it is one of the main components of the daily diet of a significant percentage of humans. Because of its distinct chemical composition, nutritional value, and balanced protein level, meat is a crucial part of the human diet. Moreover, studies have demonstrated that consuming extra protein helps maintain healthy body weight and composition, in part by enhancing satiety and enhancing energy and endurance [1]. Muscle meat proteins have a high bioavailability, with a net protein utilization value of about 0.75, whereas plant protein has a value of 0.5–0.6 [2].

The procedure of killing animals is the first stage in the production of meat. The act of killing an animal to produce meat is known as slaughtering. Maximum blood drainage is guaranteed by excellent slaughtering practices, although the technique chosen depends on how much blood is bled [3]. Traditional slaughtering refers to the process of killing animals for food using methods that have been practiced for generations within a specific cultural or regional context. The methods can vary widely across different cultures and communities. In many traditional slaughtering practices, the emphasis is often placed on ensuring a humane and respectful treatment of the animal. Around the world, a variety of ways of slaughter have been employed. These consist of the Halal technique, the hanging technique, and the stunning (CO_2 and electrical) technique [4]. Improper slaughtering results in the existence of blood in

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carcasses, and hemoglobin of blood enhances lipid oxidation to decrease the shelf life of meat during storage.

Poultry meat can become contaminated by microorganisms throughout the slaughter, processing, and preparation stages. Products made with poultry meat have low sensory ratings and limited storage durations because of the fast start of lipid oxidation and its high microbiological load [5]. Meat contamination may result from suppliers of bird meat using inadequate sanitation and hygiene practices [6]. Infections with *E. Coli*, Salmonella, and Staphylococcus aureus are exceedingly prevalent in developing nations and represent a major risk to public health [7]. In underdeveloped countries like Bangladesh, factors like the level of sanitation, the modes of transportation, and the storage conditions all play a part in the contamination of meat and the spread of numerous harmful bacterial strains [8]. After the slaughtering operation, the next problem processors deal with how to keep the quality of meat from deteriorating when the meat product is not intended for immediate consumption.

The quality of meat may decay due to long periods of storage. Thus, the storage temperature and initial microbial load are the prime causes of raw meat quality deterioration and hamper shelf life [9]. Many studies on meat shelf -life practiced on 0 to 4 °C refrigeration temperature and -18 °C to -20 °C for frozen storage. If the microbial contamination exceeds 7 log CFU g-1 the microbial metabolites generate a bad -odor, loss of WHC, and enhanced chemical reaction [10]. Therefore, the freshness of meat for consumer acceptability depends on the processing and storage condition of meat practiced by local vendors. The fact that physicochemical and microbiological properties change throughout the supply chain, along with knowledge and practices related to sanitary and hygienic slaughter, distribution, and retail marketing, is one of the main obstacles facing meat slaughter in retail establishments [11].

Nonetheless, not much research has been done to evaluate changes in the physicochemical qualities of broilers, and microbiological contamination, related to locally practiced slaughtering and post-handling of chicken meat along the production chain from retail locations in Jashore city, Bangladesh. The objective of the study was to understand the effect of poor post-slaughtering practices on the physicochemical characteristics, and bacteriological quality of fresh meat during frozen storage at 0 days, 15 days, and 30 days in Jashore City, Bangladesh.

Materials and methods

Ethics statement

The present investigation was conducted in the Department of Nutrition and Food Technology, Jashore University of Science and Technology, Bangladesh. The full procedure including vendor consent and slaughtering protocol was performed by following the Animal Slaughter & Meat Quality Control Act, 2011 (Act No.16 of 2011) of Bangladesh, after considering the animal-related ethical factors.

Sample collection and slaughter method

A small sample size of 40 broiler chickens of approximately 1.5kg weight, of the same marketable age, were randomly purchased from different local vendors due to limited resources in terms of time, money, and manpower in the preliminary study at Jashore meat market. The birds were slaughtered by local vendors following a rapid slaughtering method similar to the Islamic ritual or Halal method. No stunning was performed throughout the experiment. Next, the chickens were dipped in hot water for 2-3minutes at 55-60 °C and the feathers were plucked out from the skin using a feather-picking machine which was in unhygienic condition. Subsequently, the body parts are eviscerated, washed with clean water, and packed in plastic bags for storage. The processed meat is divided into three different groups based on storage duration 0 days, 15 days, and 30 days. One group was quickly utilized for physicochemical quality analysis and the rest of the groups were vacuum-packed, frozen, and stored for further analysis at the end of each selected storage time.

Quality evaluation of chicken meat

The standard methods of the Association of Official Agricultural Chemists [12] were followed to determine the proximate composition. The determination of pH, lipid oxidation rate, Water-Holding Capacity (WHC), Drip loss, thawing loss, cooking loss, heme iron content, and instrumental color analysis (Konica Minolta chroma meter CR-400, Konica Minolta Sensing, Inc., Japan) were undertaken in the departmental laboratory. Additionally, microbial quality evaluations were performed alongside the given parameters.

Proximate analysis: The proximate analysis of ash, moisture, protein, and fat content of chicken muscle was performed according to the methods of AOAC [12].

Determination of pH: The pH of the meat sample was determined by the protocol given by Dadgar, et al. [13] which involves measuring the acidity or alkalinity of the meat sample. pH is a measure of the concentration of hydrogen ions in a solution and is an important parameter in assessing the quality and freshness of meat. The pH of meat can influence its color, texture, water-holding capacity, and overall palatability. About 5g of meat was homogenized with 20mL deionized water at 13,600 rpm for 40 sec and a Mettler Toledo pH meter was used to measure the pH.

Determination of drip loss: The drip loss in meat was determined by following the Wang, et al. [14] procedure. Drip loss in meat refers to the loss of moisture or liquid from the meat during storage or processing. The principle of the drip loss test involves measuring the weight of the liquid that is released from the meat sample. Drip loss is an indicator of the water-holding capacity of meat and is influenced by factors such as muscle structure, pH, and storage conditions. For drip loss measurement, the weighed sample (50g) was stored under controlled conditions (typically refrigerated at 4 ºC for 24 hours and calculated using the equation below.

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$$\text{Drip loss}(\%) = \frac{w1 - w2}{w1} \times 100$$

Here, W1= weight of the sample before freezing; W2= weight of the sample after 24 h refrigeration

Determination of thawing loss: The thawing loss of meat was measured by the Dadgar, et al. [13] procedure. The determination of thawing loss in meat involves measuring the weight loss that occurs when frozen meat is allowed to thaw. Thawing loss is an important parameter in assessing the quality of frozen meat and is influenced by factors such as the rate of thawing, temperature, and the meat's water-holding capacity. This test provides information about the ability of meat to retain moisture during the thawing process. For thawing loss measurement, weigh the frozen meat sample and record the initial weight (W1) using a balance machine. Place the frozen meat sample in a plastic bowl suitable for thawing. Thaw the meat sample under controlled conditions at room temperature until fully thawed. After thawing, remove any surface moisture using absorbent material and take weight again for recording the final weight (W2).

$$\text{Thawing loss}(\%) = \frac{w1 - w2}{w1} \times 100$$

Here, *W1*= weight of the sample before freezing

W2 = weight of the meat sample after thawing

Determination of cooking loss: The cooking loss was determined by the procedure reported by Dadgar, et al. [13]. It involves measuring the weight loss during the cooking or heating process. Cooking loss is a valuable parameter that reflects the ability of meat to retain water during the cooking process, providing insight into its water-holding capacity and overall quality. About 50g of meat samples were vacuum-packed individually and immersed in hot water in a water bath for cooking at 80±0.5 °C for 30 minutes. Then they cooled at room temperature and took the weight for calculation of weight loss percentage during cooking.

$$\text{Cooking loss}(\%) = \frac{w1 - w2}{w1} \times 100$$

Here, *W1* = weight of the sample before cooking;

W2 = weight of the sample after cooking.

Determination of Water Holding Capacity (WHC): The water-holding capacity of meat was assessed as free water content according to the filter press method given by Honikel and Hamm [15]. The principle is based on the ability of meat to bind and retain water, which is crucial for maintaining juiciness, tenderness, and overall quality. WHC is influenced by factors such as protein content, meat structure, and processing conditions. The WHC should be calculated by

$$\%\text{WHC} = \frac{C - F}{2} \times 100$$

Here, C = Weight of the meat sample

F = Weight of meat flake after pressure treatment

Determination of heme iron: The Heme iron content of meat samples was measured employing the method explained by Ozer & Saricoban [16] with minor modifications. The determination of heme iron in meat involves extracting heme iron from myoglobin and hemoglobin, converting it to a stable pigment, and quantifying the absorbance or color change. The principle is based on the specific reaction of heme iron with reagents that form a complex, allowing for its measurement, first, make a meat sample paste and weigh it with a balance machine. Meat sample paste (1 g) was added to 9 ml of acidified acetone solution (acetone: distilled water: HCl = 90:8:2) in a beaker. The beaker was closed with foil paper and permitted to stand in darkness condition at room temperature for 1 h. The beaker content was filtrated using Whatman GFA as glass filter paper, and the absorbance was evaluated at 640 nm by pouring it into a cuvette.

The calculation for heme iron content was performed by calculating the whole pigment as hematin employing the following formulas:

Heme-iron (mg/100gm of sample) = Total heme pigment (ppm) (A640× 680) ×0.0822

Determination of TBARS (Thiobarbituric Acid Reactive Substances): The TBARS in meat was measured by the Bozkurt and Erkmen [17] method. It indicates the formation of Thiobarbituric acid reactive substances (TBARS) as a result of lipid oxidation. TBARS are formed when malondialdehyde (MDA) reacts with Thiobarbituric acid under acidic conditions. The test is widely used to assess the degree of lipid oxidation, which can affect the quality and shelf life of meat. To determine TBARS, 3 g ground samples were homogenized at high speed with 27 ml 3.86% perchloric acid using a digital homogenizer for 20 seconds and kept for 1 h at low temperature to settle down properly. Next, the mixture was passed through a centrifugation machine at 2000 rpm for 10 min and clarified using Whatman no.1 filter paper to separate the lipid part from other macronutrients. By using a pipette, 2 ml of 20-mm TBA solution and 2 ml filtrated solution were transferred into a test tube, and also made a blank sample by the addition of 2 ml distilled water to 2 ml of 20-mm TBA solution. Then, all solutions were carefully stored at room temperature for 15 h. Finally, the concentration of TBARS was evaluated by using a spectrophotometer (Cary 60 Uv-vis, Agilent Technologies, Seoul, Korea) where the reading of absorbance was taken at 531 nm. All samples were arranged in triplicate for analysis and the value was expressed as mg malonaldehyde per kg wet sample.

$$\text{TBARS}\left(\mu\text{mol MDA / kg sample}\right) = \frac{Absorbance\ at\ 532\ nm - Blank\ Absorbance}{Molar\ Absorptivity\left(\varepsilon\right) \times Sample\ Volume\left(L\right) \times Sample\ Weight\left(kg\right)}$$

Determination of color: The determination of the color of meat involves assessing the visual appearance of the meat,

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which is influenced by factors such as myoglobin content, oxygenation state, and pH. The color of meat is an important quality parameter as it indicates freshness, and doneness, and can influence consumer preferences. The primary pigments responsible for meat color are myoglobin and its derivatives. The meat samples free from color defects, blood spots, and bruises were used for color recording by a Hunter Lab scan Colorimeter (Minolta CR-300, Minolta Corp., Tokiyo, Japan) following the Dadgar, et al. [13] method. Recorded color values obtained from the instrument as lightness (L*), redness (a*), yellowness (b*), hue angle (h), and chroma (c) values from three different locations on the cut surface of individual meat samples.

Determination of mineral: The presence of Calcium (Ca), magnesium (Mg), iron (Fe), zinc (Zn), and copper (Cu) in meat samples was measured by a wet method of digestion where 0.5g sample was digested in the ratio of 2:1 of HNO_3/H_2O_2 mixture for 24hr. The digested mixture was placed in a volumetric flask with and volume of up to 100 mL using deionized water. The minerals were identified by using a Spectrophotometer of different wavelengths for different minerals and expressed as mg/kg wet samples.

Microbiological analysis: The total plate count in meat was determined by the procedure of Islam, et al. [18]. The determination of the total plate count in meat involves assessing the total number of viable microorganisms (bacteria) present on the meat surface. The principle is based on the ability of microorganisms to form colonies when provided with suitable growth conditions. This test indicates the overall microbial load and the hygienic quality of the meat. For this analysis, 5g blended samples were aseptically placed in a sterilized beaker to mix with distilled water thoroughly for 2 mins. Then 1mL of diluted mixture was transferred in a previously prepared nutrient agar media in triplicate and the inoculated Petri dishes were in a suitable incubator at the appropriate temperature 37°C and time 24hr for microbial growth. After incubation, count the visible colonies on the agar surface and calculate it as follows

$$\text{Total plate count}\left(\text{CFU / g or CFU / ml}\right) = \frac{\textit{Number of Colonies Counted}}{\textit{Dilution Factor} \times \textit{Volume Plated}}$$

Statistical analysis

The experiments were run in triplicate with three different lots of samples. All data were subjected to analysis of variance (ANOVA) and mean comparison was carried out using Duncan's multiple range test. Analysis was performed using the Statistical Package for Social Science package (SPSS 25.0 for Windows, SPSS Inc., Chicago, IL, USA), and the level of significance was determined at $p < 0.05$.

Results & discussion

Effect of poor post-slaughtering practices on proximate composition of chicken meat at different storage times

Moisture content: Table 1 shows the moisture content of chicken meat samples was 75.96 %, 73.59 %, and 72.91 % found at 0, 15, and 30 days during frozen conditions. There was a significant ($p < 0.05$) decrease in moisture content observed in 30 days compared to 0 days. The moisture content value of the broiler meat samples during frozen storage between day 15 and day 30 did not differ significantly ($p > 0.05$). Freezer burn occurs when moisture in the chicken meat evaporates and forms ice crystals on the surface. Freezer burn results in the loss of moisture, leading to dry and dehydrated areas on the meat [19].

Ash content: From Table 1 we can observe that the ash content of chicken meat samples was found 1.30 %, 1.17 %, and 1.10 % at 0, 15, and 30 days during the frozen condition. It was noted that there was no significant difference in the ash content of chicken meat during storage. The content of ash in chicken meat samples did not change significantly during 1 month of storage in frozen condition, except the content of fat, which reduced significantly ($p > 0.05$), this is in agreement with Hamed, et al. [19] and Bida and Faruruwa [20].

Protein content: The protein contents of meat were 22.38, 21.34, and 18.21 gm/100 gm found in 0, 15, and 30 days during frozen conditions (Table 1). The protein content of the broiler meat samples did not differ significantly ($p > 0.05$) between 0 days and 15 days but there was a significant ($p < 0.05$) decrease in protein content observed between 0 days and 30 days. Again, a significant ($p < 0.05$) difference was observed between 15 and 30 days during frozen storage. This reduction may be due to microbial activity and drip loss during freezing or could be due to the denaturation of myofibril proteins produced by prolonged freezing as well as related to the enzymatic activities of psychotropic bacteria growth, which causes proteolysis [19].

Fat content: Table 1 also reflects that the fat contents of the sample were 2.31, 1.92, and 1.08 gm /100 gm in 0, 15, and 30 days during the frozen conditions. There was a significant ($p < 0.05$) difference in fat content observed between 0 and 30 days during frozen storage. The fat content of the broiler meat samples did not differ significantly ($p > 0.05$) between 0 and 15 days but a significant ($p < 0.05$) difference was observed between 15 and 30 days. It was noted that a decrease in fat content with an increasing frozen storage period was caused by lipid oxidation. Lipid oxidation was more pronounced, and more likely associated with the higher blood retained in the meat [20].

Therefore, poor post-slaughtering practices had no noticeable influence on ash content but the other parameters

Table 1: Effect of poor post-slaughtering practices on proximate composition of chicken meat at different storage times.

Storage Day/ Parameters	0 day	15 days	30 days
Moisture (%)	75.96 ± 1.13^a	73.59 ± 0.78^b	72.91±1.15^b
Ash (%)	1.3 ± 0.61^a	1.17 ± 0.28^a	1.1 ±0.19^a
Protein content (g/100g)	22.38 ± 1.19^a	21.34 ±1.15^a	18.21±0.81^b
Fat content (g/100g)	2.31 ± 0.66^a	1.92 ± 0.56^a	1.08 ±0.21^b

Results are expressed as mean± standard deviation ($n = 6$). (a, b, c) means in the same row but superscripts in different columns differ significantly ($p < 0.05$) at different storage days.

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showed major changes with increasing storage time due to frozen storage.

Effect of poor post slaughtering practices on quality parameters of chicken meat at different storage times

pH: pH is one of the vital physical traits of meat quality and safety. The normal pH range for fresh chicken meat is between 5.7 and 6.1. Below 5.7, the meat is referred to as acid, and over 6.1 as DFD (dark, firm, and dry) [21]. The pH of samples was found 5.38, 5.7, and 6.16 in 0, 15, and 30 days of frozen condition which is shown in Table 2. The rate of decline in the pH value depends on the activity of glycolytic enzymes immediately after death, and the ultimate pH was determined by the initial glycogen reserves of the muscle. Different muscles within an animal can have different pH levels. Stress before slaughter can affect the pH [22]. Muscles that are more exercised or used frequently may have a lower pH. The rate of glycolysis is temperature-dependent. Higher temperatures can accelerate postmortem changes and lead to a decrease in pH. Spoilage microorganisms can produce acids, lowering the pH of meat during spoilage. This is a concern for meat that has not been handled or stored properly [23]. A significantly ($p < 0.05$) higher pH value was observed in fresh conditions than 15 days and 30 days of storage. It was noted that meat pH value slowly increases with increasing frozen storage duration due to the formation of nitrogenic basic compounds from spoiled meat. However, during frozen storage, enzymes and bacteria may continue to affect the meat, leading to a subsequent increase in pH [24].

Drip loss, thawing loss, and Water Holding Capacity (WHC): Assessment of drip loss and thawing loss indicate the water binding capacity of meat which is defined as the ability to retain inherent water by the application of external pressure [25]. Table 2 shows the result of drip loss and thawing loss changes during frozen conditions. Poor post-slaughtering practices had a significant impact ($p < 0.05$) on the drip loss of meat during storage. The values of drip loss significantly declined with increasing storage time. pH and WHC were closely related to drip loss and higher pH and WHC indicate less drip loss as in our study. Thawing loss was a critical quality parameter of frozen meat which depends on the degree of histological muscle tissue structure damage and freezing denaturation of protein. The water binding capacity of meat decreases during freezing resulting in a higher loss of meat juice as thawing loss. Thus, this study showed the effect of poor post-slaughtering handling on meat quality by significantly rising ($p < 0.05$) thawing loss of meat during storage. In our study, the water water-holding capacity of fresh raw chicken meat was 81.27 % which was significantly decreased with increasing storage time. The decline in the pH value near the isoelectric point of the myofibrillar protein results in a low WHC due to a reduction in the net charge of the myofibrillar protein, which decreases both the water affinity and space thereafter [26].

Cooking loss: Table 2 shows the cooking loss in chicken meat samples 26.06 %, 29.7 %, and 30.88 % were found at 0 days, 15 days, and 30 days during frozen conditions. There was a significant ($p < 0.05$) difference in cooking loss being observed at 0 days with 15 days and 30 days but it insignificantly ($p > 0.05$) differs between 15 days and 30 days. With increasing storage time, the cooking loss of all samples increased ($p < 0.05$) due to the reduction of water binding capacity. During the extended storage period, Lipid oxidation products have been known to induce protein cross-linking in the muscle. As a result, the denaturation of proteins was enhanced with a concomitant decrease in water-holding capacity after cooking [27].

Heme iron content: Heme iron contents of 2.57, 2.13, and 1.31 mg/100 gm samples were found for 0 days, 15 days, and 30 days during frozen conditions (Table 2). The changes in heme iron content in chicken meat during frozen storage were significantly ($p < 0.05$) reduced between 0 days (2.57 mg/100g) to 30 days (1.31 mg/100mg meat). The heme iron content of fresh chicken meat in 0 days did not differ significantly ($p > 0.05$) compared to 15 days during frozen storage. But there is significant difference was observed in the heme iron content of the sample stored for up to 30 days ($p < 0.05$). Decreases in heme iron content with increasing storage time were probably due to heme breakdown, resulting in the release of non-heme iron. The released iron can stimulate lipid oxidation of muscle during extended storage [28].

Table 2: Effect of poor post-slaughtering practices on quality parameters of chicken meat at different storage times

Storage Day/ Parameters	0 day	15 days	30 days
pH	5.38 ± 0.39^{c}	5.7±0.24^{b}	6.16 ±0.04^{a}
Drip loss (%)	4.25±0.15^{a}	3.18±0.26^{b}	1.17±0.08^{c}
Thawing loss (%)	3.10 ± 0.48^{c}	6.63 ± 0.34^{a}	6.04 ± 0.24^{b}
Cooking loss (%)	26.06 ± 3.03^{b}	29.7±2.63^{a}	30.88±1.77^{a}
Water Holding Capacity (%)	81.11±1.46^{a}	76.4 ±1.24^{b}	73.17 ±1.26^{c}
Heam iron (mg/kg)	2.57 ±1.74^{a}	2.13±0.21^{a}	1.31 ±0.27^{b}

Results are expressed as mean± standard deviation ($n = 6$). (a, b, c) means in the same row but superscripts in different columns differ significantly ($p < 0.05$) at different storage days.

Effect of poor post-slaughtering practices on the lipid oxidation rate of chicken meat at different storage times

The effect of Poor post slaughtering practices on the lipid oxidation rate of chicken meat during storage time was monitored by measuring the TBARS value (Figure 1). The continuous increase in TBARS value was noticeable in all samples with increasing storage time ($p < 0.05$). TBARS values of chicken meat slowly increased from 0 days to 15 days, the loss of low molecular weight oxidation products during the advancement of oxidation might lead to the constant TBARS values, and then the rate of fat oxidation significantly increased during 15-30 days of storage ($p < 0.05$). The result indicated that lipid oxidation took place continuously in the chicken meat during frozen storage. The damage to muscle structure due to ice crystallization causes the release of pro-oxidants and oxidative enzymes that enhance the susceptibility to oxidation during frozen conditions [29]. Additionally, Hemoglobin in muscle was reported to accelerate lipid oxidation [30].

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Effect of poor post-slaughtering practices on changes of color attributes of fresh and frozen poultry meat during storage

Meat color is an important assessment criterion. The color profile as lightness (L*), redness (a*), yellowness (b*), Chroma (C*), and hue (h*) of chicken meat samples at different storage periods was represented in Figure 2. The impact of poor post-slaughtering practices had a noticeable effect on different color attributes of fresh meat. Variation in fresh raw chicken meat color caused by stress, and rough handling, during the slaughtering process can affect the meat quality, including its color. Incomplete bleeding during slaughter can leave blood residues in the meat, affecting its color. Poor hygiene practices can lead to bacterial contamination, which may cause discoloration or off-putting odors in the meat. Exposure to air can cause oxidation of myoglobin, the protein responsible for meat color. This can lead to discoloration, and the meat may appear brown or discolored on the surface [26].

The extent of color change of frozen meat is determined, mainly by prevailing conditions in freezing storage and access to oxygen. The current study demonstrated the existence of a direct relationship between the duration of freezing storage and color change (reduced brightness L* but increased yellowness b*) of breast meat soon after thawing. The continuous increase in L* -value was noticeable in all samples during the frozen storage ($p < 0.05$). The L* -value of the meat samples during frozen storage between 15 days and 30 days did not differ significantly ($p > 0.05$). There were no differences in redness (a*) value between 0 days and 15 days ($p > 0.05$). However, there was a significant ($p < 0.05$) difference of a* value at day 0 in fresh condition compared to day 30 during frozen condition. After 15 days of frozen storage, the lightness (L*) and redness (a*) of broiler chicken meat decreased ($p < 0.05$). Decreases in L* value could be caused by declining water holding capacity, which leads to a lower surface light reflectivity. The oxidation of hemoglobin and myoglobin to form methemoglobin or methemoglobin resulted in discoloration of chicken meat during frozen storage [27]. After 15 days of freezing storage, however, the meat was characterized by more intense yellow color (b*) saturation than 0 days in fresh condition. The increasing lipid oxidation and the formation of Met Mb are the main factors contributing to changes in b* value [31].

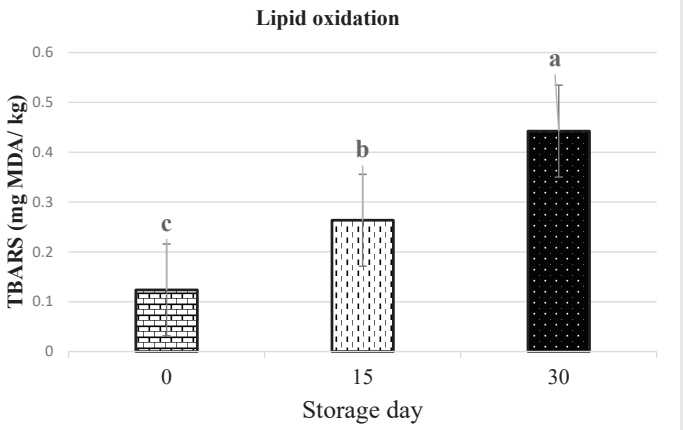

Figure 1: Effect of poor post-slaughtering practices on the lipid oxidation rate of chicken meat at different storage times. Results are expressed as mean ± standard deviation ($n = 6$). (a, b, c) means superscripts in different columns differ significantly ($p < 0.05$) at different storage days.

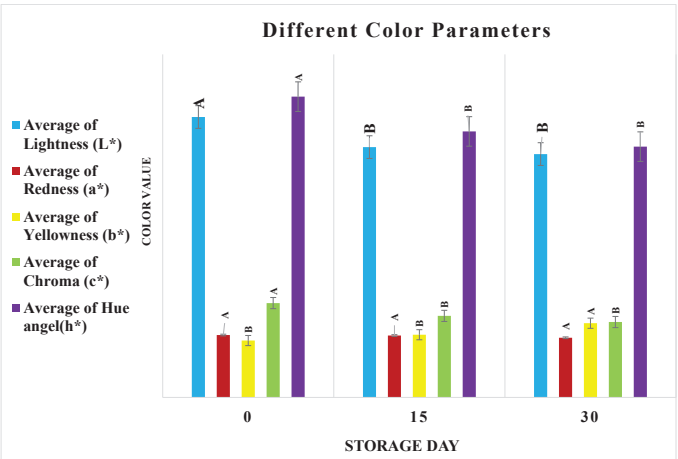

Figure 2: Effect of poor post-slaughtering practices on different color parameters of chicken meat at different storage time. Results are expressed as mean± standard deviation ($n = 6$). (A,B) means superscripts in different columns differ significantly ($p < 0.05$) at different storage days.

Furthermore, the results in Figure 2 indicated that the chroma (C*) values were significantly ($p < 0.05$) decreased with increasing frozen storage periods between 0 days to 30 days. To conclude, the hue angle (h) values revealed significant ($p < 0.05$) decreases with increasing frozen storage periods between 0 and 30 days during frozen storage as a result of poor handling after slaughter.

Effect of poor post-slaughtering practices on the mineral content of fresh and frozen poultry Meat during Storage

Different slaughtering methods from different regions of the world and different storage methods had a significant impact on the mineral composition of chicken meat. The effect of poor post-slaughtering practices and 1 month of frozen storage on the content of Ca, Mg, Fe, Zn, and Cu are shown in Table 3. Here Ca and Mg are the most abundant minerals in freshly slaughtered meat, whereas Fe, Zn, and Cu are available in very tiny amounts. This outcome is nearly similar to the data reported by Hafiz, et al. [32]. The Ca and Mg content of freshly slaughtered chicken meat was higher, similar to the results obtained by Addeen, et al. [33]. Besides this, all mineral content slowly decreased in quantity with increasing storage time. The content of minerals in muscle depends on water-soluble substances like protein, free amino acid, and creatine content of meat, and thawing boosts the loss of minerals from muscle through free water [34]. The presence of a low amount of iron in meat designates maximum removal of blood from veins as a result of traditional slaughtering. There were insignificant changes ($p > 0.05$) were found in Mg and Cu content during the frozen storage time. The values of Ca and Zn were significantly ($p < 0.05$) reduced at the end of 15 days to 30 days, whereas Fe content significantly ($p < 0.05$) decreased during storage from 0 days to 30 days. Above all transition metals, Fe and Cu work as catalysts for oxidation during storage triggering

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the development of TBARS and peroxide values. Here, Fe is a transition metal that acts as a pro-oxidant.

Effect of poor post-slaughtering practices on microbial content of fresh and frozen poultry meat during storage

The total plate counts (TPC) of fresh chicken meat 0, 15, and 30 days during frozen storage are shown in Figure 3. There was a significant ($p < 0.05$) difference in the TPC of chicken meat from 0 days to 30 days. Nurmasytha, et al. [35] reported that the TPC of a market sample of raw chicken breast meat ranged from 5.39 log cfu/ g to 6.27 log cfu/g. In our study, the TPC chicken meat was (6.99 log cfu / g) at 0 days. Fresh chicken meat may be contaminated with different food pathogens due to personal unhygienic culpabilities that occur during various slaughter, storage, transportation, and handling processes, including contaminated water, gastrointestinal contamination, air, dust, sewage, and environmental surfaces [35].

It was noted that increase in TPC count with increasing frozen storage period ($p < 0.05$). There was a significant ($p < 0.05$) increase of TPC of chicken meat from 0 days to 30 days during frozen storage. However, blood could be retained in the bled sample to some extent and this would serve as the nutrient for microbial growth. The blood enriched with nutrients for microbial growth could induce the enumeration of bacteria, which were contaminated from the skin, viscera, or environment during handling, slaughtering, and dressing. *Salmonella spp., Listeria monocytogenes, Staphylococcus aureus, Enterobacteriaceae, Escherichia coli, Campylobacter spp., and C. perfringens* were found as the dominant bacteria in chicken meat [36]. Also, meat's ultimate pH (> 6.0) significantly affects the growth of spoilage bacteria during frozen storage [37]. Therefore, in this study, the high pH (6.17) of meat and unhygienic handling before storage may be the major reasons for the high total viable count which can reduce the shelf life of meat and make it unfit for human consumption.

Table 3: Effect of poor post-slaughtering practices on the mineral content of chicken meat at different storage times.

Storage day/ Mineral content (mg/ kg)	0 day	15 days	30 days
Ca	115.20 ± 0.31^a	111.71 ± 0.18^a	86.43 ±1.12^b
Mg	305.33 ± 2.23	298.18 ± 1.77	291.22 ± 0.67
Fe	8.05 ± 1.06^a	5.74 ± 2.33^b	4.47 ± 0.14^b
Zn	6.88 ± 0.4^a	5.04 ± 1.17^a	3.86 ± 2.1^b
Cu	1.13 ± 1.32	1.04 ± 0.29	0.72 ± 1.10

Results are expressed as mean ± standard deviation ($n = 6$). (a, b, c) means in the same row but superscripts in different columns differ significantly ($p < 0.05$) at different storage days.

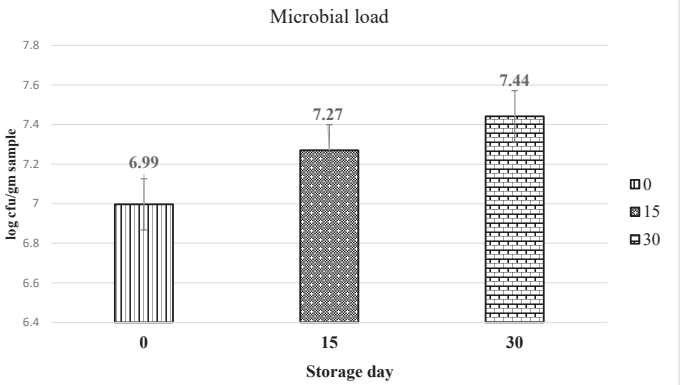

Figure 3: Effect of poor post-slaughtering practices on the microbiological quality of chicken meat at different storage time. Results are expressed as mean ± standard deviation ($n = 6$). (a, b) means superscripts in different columns differ significantly ($p < 0.05$) at different storage days.

To reduce microbial growth in broiler meat, several preventive measures can be implemented. Firstly, ensure immediate chilling of the meat to below -18°C after slaughter to inhibit bacterial growth. Maintain strict hygiene practices during handling, including the use of sanitized equipment and clean surfaces. Employ vacuum packaging to minimize oxygen exposure and slow down microbial proliferation. Additionally, monitor storage temperatures regularly to ensure they remain consistent and within the recommended range. Implementing these measures will help control microbial contamination and improve the overall quality and safety of the meat during storage.

Conclusion

These results indicate that poor post-slaughtering practices affect the chemical and microbiological composition of chicken meat during frozen storage. This study concluded that the meat obtained by local slaughtering results in higher microbial counts and lower physicochemical parameters. The high drip and cook loss observed in this study may be attributed to stressful slaughtering resulting in the depletion of the glycogen reserve in the muscle. Hence, the high ultimate pH observed in poor post-slaughtered handling and preserved meat after 30 days of frozen storage can be attributed to stressful slaughtering and imperfect bleeding. Besides poor post-slaughtering practices, our study faced limitations due to a lack of advanced equipment and time constraints. Future efforts should focus on bridging the gap between laboratory and real-time retail conditions by improving post-slaughter handling, bleeding techniques, and hygiene protocols. Enhancing training for local slaughterhouses and implementing stricter regulations and monitoring systems will be essential to elevate meat quality and safety in retail settings.

Author contributions

Kamruzzaman, M and Rasel, I.: data collection and preparation; Kamruzzaman. M., and Mohasin, H.: creation and writing the initial draft; Kamruzzaman, M., and Rasel, I., writing the original draft: Rashida, P., and Ashrafuzzaman, Z. M., methodology; Ananya, R., and Kamruzzaman, M.: application of statistics. All authors have read and agreed to the published version of the manuscript.

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