

## RESEARCH ARTICLE

# Omega-3 Fatty Acids Enriched Flaxseed Oil Effects on Meat Quality and Fatty Acid Profile of Broiler Chicks

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## ABSTRACT

The study was planned to evaluate effect of adding flaxseed oil on meat quality, sensory attributes and fatty acids profile of broiler chicken meat. Further, optimization of supplemental flaxseed oil duration and level was also tested. A n=300 day-old broiler chicken (Ross-308) were procured from the hatchery and randomly divided into 5 treatments, T0 (control) T1 and T2 (3% and 4% flaxseed oil, 14-35 day), while T3 and T4 having (3% and 4% flaxseed oil, 21-35 day) groups respectively. Each treatment contained 6 replicates with 10 chicks per replicate. The T3 tested diet linearly raised n-3 (PUFA) contents. The n-6/n-3 ratio was also significantly lowered from 7.72 to 1.12%. Regardless of dietary flaxseed oil level, LC-PUFA contents were higher in the T2 group. Meat quality traits were not statistically affected while T1 exhibited a significantly increased meat tenderness value, but interestingly, all other sensory attributes remained statistically unaffected. An equivalent amount of n-3 LC-PUFA may be found in 3% dietary flaxseed oil for the last 21 days of rearing with optimized meat quality and sensory attributes. Healthy broiler meat enriched with n-3 fatty acids may be produced by dietary manipulation.

**Keywords:** Flaxseed oil, n-3 LC-PUFA, Meat quality, Sensory attributes, Broiler meat

## INTRODUCTION

The health benefits of dietary polyunsaturated fatty acids (PUFA) are ubiquitously recognized globally as they play a significant role in the development and maintenance of the human body. The empirical scientific studies indicate that proper intake of Omega-3 and Omega-6 PUFA or with a proper ratio of (n-6/n-3 PUFA) may generally inhibit the progression of various diseases but particularly control diabetes, coronary heart disease, rheumatoid arthritis and nervous system problems <sup>[1]</sup>. The World Health Organization (WHO) has also observed that Omega-3 long-chain PUFA may reduce cardiovascular diseases <sup>[2]</sup>. Omega-3 PUFA consumption also reduces plasma triglycerides, blood pressure, and resting heart rate and might also improve myocardial efficiency, lower inflammation, and improve vascular function. The content of Omega-3 PUFA has a significantly positive impact on public health nutrition, but unfortunately, their intake is still scarce for many reasons.

Marine products are the best source of Omega-3 fatty acids and are consumed predominantly in a few areas of the world <sup>[3]</sup>. However, their dietary inclusion is also rare due to their seasonal availability, consumer preferences and affordability worldwide. This scenario has compelled the scientific community to search for alternative sources for Omega-3 fatty acids, which may successfully serve the purpose. Chicken meat is the most consumed protein source in the world, and efforts are being made to produce broiler chicken meat containing high levels of omega-3 LC PUFA, particularly (C20:5 n-3, EPA) eicosapentaenoic and (C22:6 n-3, DHA) docosahexaenoic acid by the inclusion of natural flaxseed oil <sup>[4]</sup>. Although fish oil is the primary source of these fatty acids, its utilization is restricted in the broiler diet due to meat's high cost and poor sensory attributes. Furthermore, it was reported that dietary fish oil has some adverse effects on the growth performance of chicken and drip loss% of meat, making it less acceptable for processors and consumers <sup>[5]</sup>.



Flaxseed oil is the most valuable common plant-based feed ingredient to enrich n-3 fatty acid poultry products. The flaxseed contains an ample quantity of alpha linolenic acid (ALA) (>50%) and fat (~40-42%) contents along with other nutritional properties (high protein and metabolisable energy content) [6]. However, its high inclusion is also inhibited in poultry feed due to the high cost and presence of anti-nutritional factors such as mucilage, linatinedi-peptide, cyanogenic glycosides, phytic acid and trypsin inhibitor [7]. So, the rational utilization of flaxseed may show promising results on bird growth and may economize bird feed [3,8].

Therefore, the present study was planned firstly to evaluate the two levels of flaxseed oil for different feeding durations as a way to improve the functional value of chicken meat on meat quality, coloring index and fatty acid profile, especially the n-3 LC-PUFA contents and sensory analysis of broiler chicken meat: and Secondly to verify that how long dietary manipulation is suitable to achieve high quality nutritionally enriched chicken meat for human consumption.

## MATERIAL AND METHODS

### Ethical Approval

All the experimental procedures followed the Animal Care and Use Committee (dr/1376: 06-05-2019) Department of Poultry Production, University of Veterinary and Animal Sciences, Lahore, Pakistan.

### Experimental Plan, Management and Slaughtering

A total of 300-day-old broiler chickens unsexed (Ross-308) were randomly distributed into 5 experimental groups according to a completely randomized design (each group contained 6 replicates and 10 broiler chickens per group) for 35 days. Five *isonitrogenous* and *isocaloric* diets were prepared in mash form (Table 1). The experimental room was properly fumigated and disinfected before the arrival of broiler chickens. All these were raised under standard management conditions. A photoperiod of 23 h of light and 1 h of darkness was provided. Further, a temperature of 35-32°C was provided during the first week of rearing, which was gradually reduced to 5°C until 21°C. According to the schedule, birds were vaccinated for Newcastle disease, infectious bursal disease and infectious bronchitis.

The diets were enriched with the two flaxseed oil levels and fed in two different durations to evaluate effective level of flaxseed. The T<sub>0</sub> was the control diet, but T<sub>1</sub> and T<sub>2</sub> basal diets contained 3% and 4% flaxseed oil provided from 14-35 days of production, while T<sub>3</sub> and T<sub>4</sub> were enriched with previously mentioned concentrations but fed from 21-35 days of production. Further, recipes and chemical composition of all the experimental diets have

| Ingredients%                     | Control <sup>1</sup> | Flaxseed Oil <sup>2</sup> |       |
|----------------------------------|----------------------|---------------------------|-------|
|                                  |                      | 3%                        | 4%    |
| Corn grain                       | 58.49                | 59.50                     | 56.00 |
| Wheat bran                       | 3.00                 | 2.30                      | 4.70  |
| Canola meal                      | 4.77                 | 4.76                      | 4.86  |
| Corn gluten 60%                  | 1.00                 | 1.00                      | 1.00  |
| Soybean meal                     | 24.0                 | 24.0                      | 24.0  |
| Soybean oil                      | 3.30                 | 00                        | 0     |
| Flaxseed oil                     | 00                   | 3.00                      | 4.00  |
| CaCO <sub>3</sub>                | 1.10                 | 1.10                      | 1.10  |
| Dicalcium phosphate              | 1.80                 | 1.80                      | 1.80  |
| Lysine-SO <sub>4</sub>           | 0.40                 | 0.40                      | 0.40  |
| DL-methionine                    | 0.16                 | 0.16                      | 0.16  |
| Threonine                        | 0.98                 | 0.98                      | 0.98  |
| Ajinomot                         | 0.10                 | 0.10                      | 0.10  |
| NaCl                             | 0.30                 | 0.30                      | 0.30  |
| NaHCO <sub>3</sub>               | 0.10                 | 0.10                      | 0.10  |
| Vitamin premix                   | 0.30                 | 0.30                      | 0.30  |
| Minerals premix                  | 0.20                 | 0.20                      | 0.20  |
| Total                            | 100                  | 100                       | 100   |
| <b>Calculated Composition, %</b> |                      |                           |       |
| Protein                          | 19.3                 | 19.3                      | 19.3  |
| Metabolizable energy (MJ/Kg)     | 2988                 | 2979                      | 2985  |
| Ether extract                    | 6.03                 | 5.70                      | 6.70  |
| Ash                              | 6.10                 | 6.08                      | 6.10  |
| Crude fiber                      | 3.90                 | 3.90                      | 4.10  |
| Calcium                          | 0.91                 | 0.91                      | 0.91  |
| Available-phosphorus             | 0.42                 | 0.42                      | 0.42  |

<sup>1</sup> Diet: T<sub>0</sub>: Control group  
<sup>2</sup> Diets containing 3 and 4% flaxseed oil fed either during the last 21 days of production (T<sub>1</sub> and T<sub>2</sub>, respectively) or during the last 14 days of production (T<sub>3</sub> and T<sub>4</sub>, respectively)

been shown in Table 1 and formulated based on the recommendation [9].

At the end of the trial, birds were slaughtered according to the local standard for Halal slaughtering (PS 3733:2016) in compliance with the Ethical Review Committee's institutional guidelines, UVAS (dr/1380: 13/06/ 2019). The birds were slaughtered on 35 days of production at the Meat Science and Technology department's meat processing plant, UVAS. The carcasses were subjected to an ice-water bath for 30 min, packed in a polystyrene tray, wrapped with cling film, and finally placed in a chiller at 0-4°C for 4 h before deboning.

## pH

After 24 h of slaughtering, the pH was measured from breast muscle (*Pectoralis major*) for all the treatments of each group using a calibrated portable pH meter (WTW, pH 3210 SET 2, Germany) with a meat penetrating probe (WTW, SenTix® Sp, pH electrode, Germany) as described by [10]. The pH meter was calibrated on 4.0, 7.0 and 10.0 buffer solutions at the start of each trial.

## Color

The color values were measured from breast fillets (*Pectoralis major*) with the help of a chroma meter (Konica Minolta® CR-410, Japan) 50 mm port size and D65 illuminant [10]. The chroma meter was calibrated using a standard white tile (L\* 94.93, a\* 0.13, b\* 2.55) and color scale [11]. The color parameters were comprised of lightness (L\*), redness (b\*), yellowness (a\*), chrome (c) and hue angle (h). After deboning, breast fillets were placed in food-grade polystyrene trays, wrapped with a commercially available 250 mm thick cling film and stored in a chiller (Model: S80100VVC, Tecnodom S.P.A., Vigodarzere, PD, Italy) at 0-4°C. Color values were taken from the center of the breast muscle, avoiding the visible fascia.

## Cooking Loss (%)

The samples from each treatment were weighed with the help of an electronic weighing balance (SF-400, capacity 7000±1 g, China) and vacuum-packed in plastic bags using a C300 twin vacuum packer (Multivac®, Ltd., Serial no. 219528, Germany) in plastic bags (SR 150×200, PA/PE 90). The samples were cooked in a water bath (Mettler, WNB45, Germany), operating at 80°C to determine the cooking loss. The samples were cooked until the internal core temperature reached 72°C. The cooked meat temperature was recorded with the help of a thermometer (TP101, CixiSinco, China, -50°C to 300°C) [12]. The samples were cooled down to room temperature and again weighed to calculate the cooking loss by the following formula:

$$\% \text{ Cooking Loss} = (W_1 - W_2 / W_1) \times 100$$

$W_1$  = weight of meat before cooking

$W_2$  = weight of meat after cooking

## Drip Loss (%)

After the deboning, breast fillets were randomly selected from each treatment to calculate the drip loss percentage. The drip loss percentage was calculated using the suspension method as described [13]. A digital compact weighing balance (SF-400, 7000±1 g) was used to weigh the samples. The drip loss percentage was calculated by the formula given below

$$\% \text{ Drip Loss} = (W_1 - W_2 / W_1) \times 100$$

$W_1$  = weight of meat before the suspension

$W_2$  = weight of meat after suspension (24 h)

## Warner-Bratzler Shear Force Value

The texture analysis was performed with the help of a texture meter (TA.XT plus®, Stable Micro System, Ltd., Surrey, UK, Serial no. 41851) fitted and calibrated with a 5 kg load cell. From the cooked breast file samples, stripes of the meat were obtained and the fillets were cut down in a direction parallel to the muscle fiber orientation (2×1×1 cm<sup>3</sup>) with scalpel blades [14]. The breast fillets were placed in polystyrene trays and cooled at 0-4°C in a display chiller (ALVO, Model MD-12, Technosight, 72" × 42" × 48). A minimum of 3 values for the shear force were recorded from each sample and the force required to cut the muscle fiber was N/cm<sup>2</sup>.

## Fatty Acid Analysis

The breast muscle (*Pectoralis major*) samples were randomly selected from each treatment group to extract and measure the lipid contents to determine fatty acids. Total lipid contents were extracted according to the method [15]. A finely minced meat sample of 50 g was added to 400 mL of organic solvent chloroform/methanol (v/v 2:1). The solvent and the minced meat sample were homogenized (6000 rpm, 2 times, 30 sec each time) using a homogenizer (DAIHAN Scientific, HG-15D-Set-A, South Korea). The homogenate was filtered and shaken by adding a 0.2 mL volume of 0.9% NaCl solution. The mixture was centrifuged (Eppendorf, 5810, Germany) at a low speed (3000 rpm, 15 min) to separate the two phases. A rotatory evaporator separated the lower organic phase from the upper layer (Daihan Scientific, WEV-1001L, 25W, South Korea).

The extracted lipid contents were methylated as fatty acid methyl esters (FAME) by gas chromatography (Agilent Technologies, GC System 7890B, USA) to determine the fatty acid quantity. Each fatty acid was identified as a methyl ester by comparing the retention time with a standard.

## Sensory Analysis

The Sensory analysis of poultry breast fillets was performed at the sensory analysis lab, Central Laboratory Complex (CLC), UVAS, by a trained panel of 20 judges [16]. Before starting the sensory trial, all the panelists were aware of the experiment. After opening the tray, samples from each treatment were cooked without salt and spices on a hot plate until they attained the core temperature of 72°C. The core temperature was recorded using a digital food-grade thermometer (TP101, CixiSinco, China, -50°C to 300°C). Each specimen was sub divided into uniform parts to

serve panelists in warm conditions. All the samples were coded and served to the panel and in between subsequent samples, the panelists had a facility to rinse their mouths to remove any carry-over effect. The panelists evaluated the samples for odor, tenderness, juiciness, oiliness, flavor, overall acceptability and shallowness on a 9-point hedonic scale.

**Statistical Analysis**

The collected data were subjected to the One-way analysis of variance (ANOVA) technique. The means were compared using Duncan’s multiple range test with the help of SAS 9.1. The means were considered significantly different at  $P \leq 0.05$  [17].

**RESULTS**

**Meat Quality Parameters**

Table 2 shows the results of meat quality parameters, including colors (redness, yellowness, lightness, chroma and hue). The pH, cooking loss, tenderness and drip loss have been shown in Table 3. The diets enriched with flaxseed oil exhibited non-significant change ( $P > 0.05$ ) in meat pH values among the treatments during both dietary durations. The meat quality parameters, including color, cooking and drip loss percentage, and shear force, were closely linked to the pH of meat. Consequently, no potential statistically significant difference was observed among them. Moreover, flaxseed oil’s supplemental level and duration exhibited no negative impact on broiler meat quality parameters.

**Results of Fatty Acids Analysis**

The fatty acid composition of broiler breast meat is shown in Table 4 and Table 5. The crude fat (CF) percentage among all the dietary treatments was statistically non-significant. The supplementation of flaxseed oil showed

**Table 2. Effect of varying dietary flax seed oil levels on the color values of breast fillets**

| Treatment <sup>1</sup> | a* (Redness) | b* (Yellowness) | L* (Lightness) | C (Chroma) | h (Hue) |
|------------------------|--------------|-----------------|----------------|------------|---------|
| T <sub>0</sub>         | 14.6         | 19.5            | 56.4           | 24.4       | 53.0    |
| T <sub>1</sub>         | 16.3         | 19.7            | 54.9           | 25.7       | 51.8    |
| T <sub>2</sub>         | 13.9         | 19.5            | 55.7           | 25.3       | 50.8    |
| T <sub>3</sub>         | 15.1         | 19.9            | 55.4           | 25.2       | 52.4    |
| T <sub>4</sub>         | 14.9         | 18.5            | 56.7           | 23.9       | 50.9    |
| SEM                    | 0.553        | 0.211           | 0.337          | 0.229      | 0.500   |
| P-Value                | 0.63         | 0.62            | 0.70           | 0.34       | 0.76    |

<sup>1</sup> T<sub>0</sub> = Control group, T<sub>1</sub> and T<sub>2</sub> = 3 and 4% flaxseed oil, respectively, fed during the last 21 days of production, while T<sub>3</sub> and T<sub>4</sub> contained 3 and 4% flaxseed oil, respectively, fed during the last 14 days of production  
Different alphabets on means showing significant differences ( $P \leq 0.05$ ) among treatments

**Table 3. Effect of varying dietary flax seed oil levels on the meat quality**

| Treatment <sup>1</sup> | pH    | Drip loss % | Cooking loss % | Tenderness |
|------------------------|-------|-------------|----------------|------------|
| T <sub>0</sub>         | 5.93  | 4.85        | 11.9           | 19.4       |
| T <sub>1</sub>         | 6.04  | 4.87        | 12.1           | 19.4       |
| T <sub>2</sub>         | 5.99  | 5.15        | 11.1           | 19.7       |
| T <sub>3</sub>         | 6.01  | 4.55        | 11.4           | 19.6       |
| T <sub>4</sub>         | 5.94  | 5.11        | 11.6           | 20.0       |
| SEM                    | 0.013 | 0.205       | 0.142          | 0.218      |
| P-Value                | 0.15  | 0.88        | 0.46           | 0.97       |

<sup>1</sup> T<sub>0</sub> = Control group, T<sub>1</sub> and T<sub>2</sub> = 3 and 4% flaxseed oil, respectively, fed during the last 21 days of production, while T<sub>3</sub> and T<sub>4</sub> contained 3 and 4% flaxseed oil, respectively, fed during the last 14 days of production  
Different alphabets on means showing significant differences ( $P \leq 0.05$ ) among treatments

**Table 4. Effect of varying dietary flax seed oil levels on fatty acid<sup>1</sup> profiles of breast fillets**

| Treatment <sup>2</sup> | Crude fat | SFA               | MUFA              | PUFA               | n-6 PUFA          | n-3 PUFA          | n-6/n-3           |
|------------------------|-----------|-------------------|-------------------|--------------------|-------------------|-------------------|-------------------|
| T <sub>0</sub>         | 1.77      | 35.4 <sup>a</sup> | 42.9 <sup>a</sup> | 21.8 <sup>c</sup>  | 19.3 <sup>a</sup> | 2.50 <sup>c</sup> | 7.72 <sup>a</sup> |
| T <sub>1</sub>         | 1.83      | 32.8 <sup>b</sup> | 39.8 <sup>d</sup> | 27.4 <sup>b</sup>  | 15.4 <sup>c</sup> | 11.9 <sup>b</sup> | 1.29 <sup>b</sup> |
| T <sub>2</sub>         | 1.83      | 31.9 <sup>c</sup> | 39.5 <sup>d</sup> | 28.7 <sup>b</sup>  | 15.2 <sup>d</sup> | 13.5 <sup>a</sup> | 1.12 <sup>b</sup> |
| T <sub>3</sub>         | 1.80      | 32.7 <sup>b</sup> | 41.6 <sup>b</sup> | 25.7 <sup>a</sup>  | 15.8 <sup>b</sup> | 9.87 <sup>d</sup> | 1.60 <sup>b</sup> |
| T <sub>4</sub>         | 1.87      | 32.8 <sup>b</sup> | 40.6 <sup>c</sup> | 26.6 <sup>d</sup>  | 15.3 <sup>d</sup> | 11.3 <sup>c</sup> | 1.36 <sup>b</sup> |
| SEM                    | 0.016     | 0.047             | 0.076             | 0.045 <sup>c</sup> | 0.024             | 0.045             | 0.092             |
| P-Value                | 0.48      | <0.001            | <0.001            | <0.001             | <0.001            | <0.001            | <0.001            |

<sup>1</sup> SFA Saturated fatty acids; MUFA Monounsaturated fatty acids; PUFA Polyunsaturated fatty acids; n-6 PUFA polyunsaturated fatty acid; n-3 PUFA n-3 polyunsaturated fatty acid; n-6/n-3 Ratio of polyunsaturated fatty acid  
<sup>2</sup> T<sub>0</sub> = Control group, T<sub>1</sub> and T<sub>2</sub> = 3 and 4% flaxseed oil, respectively, fed during the last 21 days of production, while T<sub>3</sub> and T<sub>4</sub> contained 3 and 4% flaxseed oil, respectively, fed during the last 14 days of production  
Different alphabets on means showing significant differences ( $P \leq 0.05$ ) among treatments

**Table 5. Effect of varying dietary flax seed oil levels on fatty acid profiles of breast fillets**

| Treatment <sup>1</sup> | ALA               | n-3 LCPUFA        | EPA               | DPA               | DHA               |
|------------------------|-------------------|-------------------|-------------------|-------------------|-------------------|
| T <sub>0</sub>         | 1.77 <sup>d</sup> | 0.73 <sup>c</sup> | 0.27 <sup>c</sup> | 0.47 <sup>c</sup> | Trace             |
| T <sub>1</sub>         | 8.60 <sup>b</sup> | 3.37 <sup>a</sup> | 1.13 <sup>a</sup> | 1.47 <sup>a</sup> | 0.77 <sup>a</sup> |
| T <sub>2</sub>         | 10.1 <sup>a</sup> | 3.37 <sup>a</sup> | 1.10 <sup>a</sup> | 1.50 <sup>a</sup> | 0.77 <sup>a</sup> |
| T <sub>3</sub>         | 7.60 <sup>c</sup> | 2.27 <sup>b</sup> | 0.73 <sup>b</sup> | 1.10 <sup>b</sup> | 0.43 <sup>b</sup> |
| T <sub>4</sub>         | 8.90 <sup>b</sup> | 2.37 <sup>b</sup> | 0.77 <sup>b</sup> | 1.13 <sup>b</sup> | 0.47 <sup>b</sup> |
| SEM                    | 0.032             | 0.024             | 0.016             | 0.008             | 0.018             |
| P-Value                | <0.001            | <0.001            | <0.001            | <0.001            | <0.001            |

<sup>1</sup> T<sub>0</sub> = Control group, T<sub>1</sub> and T<sub>2</sub> = 3 and 4% flaxseed oil, respectively, fed during the last 21 days of production, while T<sub>3</sub> and T<sub>4</sub> contained 3 and 4% flaxseed oil, respectively, fed during the last 14 days of production  
Different alphabets on means showing significant differences ( $P \leq 0.05$ ) among treatments

**Table 6.** Effect of varying dietary flax seed oil levels on sensory analysis of breast fillets

| Treatment <sup>1</sup> | Odor  | Tenderness         | Juiciness | Oiliness | Mouth Feel | Swallowness | Overall Acceptability |
|------------------------|-------|--------------------|-----------|----------|------------|-------------|-----------------------|
| T <sub>0</sub>         | 7.80  | 8.00 <sup>ab</sup> | 7.50      | 7.10     | 7.60       | 8.10        | 7.80                  |
| T <sub>1</sub>         | 7.80  | 8.20 <sup>a</sup>  | 7.30      | 7.50     | 7.50       | 7.50        | 8.10                  |
| T <sub>2</sub>         | 7.70  | 7.40 <sup>ab</sup> | 7.80      | 7.10     | 7.20       | 7.30        | 7.20                  |
| T <sub>3</sub>         | 7.70  | 7.70 <sup>ab</sup> | 7.60      | 6.90     | 8.20       | 7.70        | 7.90                  |
| T <sub>4</sub>         | 7.90  | 6.70 <sup>b</sup>  | 7.20      | 7.20     | 7.60       | 7.00        | 7.00                  |
| SEM                    | 0.087 | 0.124              | 0.118     | 0.121    | 0.147      | 0.158       | 0.103                 |
| P-value                | 0.98  | 0.03               | 0.68      | 0.82     | 0.46       | 0.52        | 0.07                  |

<sup>1</sup> T<sub>0</sub> = Control group, T<sub>1</sub> and T<sub>2</sub> = 3 and 4% flaxseed oil, respectively, fed during the last 21 days of production, while T<sub>3</sub> and T<sub>4</sub> contained 3 and 4% flaxseed oil, respectively, fed during the last 14 days of production  
Different alphabets on means showing significant differences (P≤0.05) among treatments

no effect on the fat contents but only improved the fatty acids contents. The maximum level of saturated fatty acids (SFA) was observed in the control group. However, the SFA content was reduced with the increased dietary flaxseed oil. A linear relationship was found in the amount of PUFA contents in the chicken meat as the supplemental level and duration increased. It was found that adding flaxseed oil as a source of ALA linearly increased the Omega-3 PUFA contents and reduced Omega-6 PUFA contents, ultimately decreasing the Omega-6/3 PUFA ratios to 7.72 and 1.12 for the control and T<sub>2</sub> groups, respectively. The Omega-3 LC-PUFA contents were significantly different from each other.

The ALA contents in broiler breast meat were associated with the supplemental level of dietary flaxseed oil, as it deposits directly into the muscles. The highest level of Omega-3 LC-PUFA was observed in the T<sub>1</sub> and T<sub>2</sub> treatment groups as they were supplemented for 3 weeks before slaughtering (Table 5).

### Sensory Analysis Findings

The sensory interpretation showed a non-significant difference among all the treatments (Table 6). However, tenderness was the only attribute having the least acceptance with a 4% dietary flaxseed oil treatment supplemented for 14 days. While observing odor, juiciness, oiliness, flavor, swallow and overall acceptability, the study showed no notable difference between the treatments.

## DISCUSSION

The current study highlights the effects of feeding varying flaxseed oil (Omega-3 PUFA) on, meat quality, coloring index, sensory attributes and fatty acids profile, especially the Omega-3 LC-PUFA contents of broiler meat. The broiler breast fillets' pH values showed a non-significant difference among all the treatments. Our results aligned

with Lee et al.<sup>[18]</sup>, who found statistically unchanged effect of feeding flaxseed oils on the egg albumen pH. Moreover, the quail's breast meat pH remained statistically unaffected when the birds were subjected to flaxseed oil containing diet and this finding correlates with our results<sup>[19]</sup>. The findings related to the meat color, including lightness value (L\*), correspond with the muscle biochemistry, as described by<sup>[11]</sup>. As all the dietary treatments pH values remained non-significant, the color values also showed non-significant results, including redness (a\*) and yellowness (b\*) values. The linear correlation in pH and redness (a\*) values were present as a higher pH value shows higher redness (a\*) value, as reported by El Rammouz<sup>[20]</sup>. Further, Qiao et al.<sup>[21]</sup> and Ribeiro et al.<sup>[22]</sup> described the relationship between the pH and yellowness (b\*) values of meat. They stated that with the decline in the meat pH, the yellowness (b\*) values increase, as observed in the current trial.

The non-significant results were recorded in cooking and drip loss percentage among all the treatments as these parameters are also associated with the pH. The effect of the pH value on the drip and cooking losses percentage was described by Ribeiro et al.<sup>[22]</sup>, indicating that lower pH values may lead to more drip and cooking loss and vice versa.

Factually, meat tenderness is perceived as the most important quality factor determining the consumer's ultimate satisfaction. The data regarding the tenderness values illustrated a non-significant difference among various treatments and these findings justified the relationship between the tenderness and meat pH. Similar results were reported by Wang et al.<sup>[23]</sup> in which birds were supplemented with lipid and exhibited no adverse effect on the cooking loss % and meat tenderness. Moreover, our results correlate with the observation of Reddy et al.<sup>[24]</sup>, who recorded significantly higher tenderness values

of all the treatments and remained non-significant with different dietary regimes.

Significant work has been accomplished to include dietary fish meals and oil to enrich broiler meat with omega-3 fatty acids. Though, the inclusion of these sources in the ration had adversely affected the sensory attributes of meat [25]. Sensory attributes of a product are vital as they directly affect the consumer's liking. Flax seed and flax seed oil has a high dietary ALA level and may be used as a replacement for similar marine products. In addition, replacing fish oil with vegetable sources rich in Omega-3 PUFA may significantly improve the meat's sensory attributes [3].

In this study, a high level of flaxseed oil rich in ALA did not affect broiler meat's overall sensory attributes. These results agree with Fletcher [26] also concluded the perpetual effects of dietary Omega-3 PUFA on broiler meat's sensory attributes. Similarly, López-Ferrer et al. [27] used 10 and 17% flaxseed as a dietary ALA source for 8 various durations in broilers. The results showed that high Omega-3 PUFA levels up to 20 days posed no unfavorable effect on chicken meat's sensory attributes, texture, flavor, after taste, liking and overall opinion. Further, meat tenderness was the only conflicting attribute affected by the increased long-duration supplementation of Omega-3 PUFA.

Furthermore, Qiao et al. [21] and Zelenka et al. [28] evaluated the relationship between dietary Omega-3 PUFA and sensory attributes of broiler meat and found variable results. They observed that a higher level of n-3 PUFA did not affect the breast fillet's tenderness, texture and juiciness. However, the odor, taste, oily taste, and after-taste were the attributes that the higher dietary Omega-3 PUFA levels compromised. Moreover, linoleic acid (LA) should also be incorporated into the diet to produce finely fibrous, juicy and enhanced flavored meat. Gonzalez-Esquerra and Leeson [29] reported that the high dietary ALA compromised sensory attributes like aroma and after taste; the flavor was also influenced due to the production of volatile compounds that mediates the off-flavor production. Further, this might be associated with the level of Omega-3 PUFA contents, thermal degradation of lipids and the level of volatile compounds in the meat.

The crude fat (CF) percentage remained unchanged by the duration of the dietary addition of flaxseed oil which shows that the diet containing a higher level of ALA does not affect the fat contents of meat; nonetheless, it only modifies the fatty acids profile. Diets containing high ALA levels increased the n-3 PUFA contents of broiler breast meat and some of its fractions were also converted into biologically more active fatty acids, n-3 LC-PUFA [28].

The maximum level of Omega-3 LC-PUFA was determined in the birds fed with 3 and 4% dietary flaxseed oil for 21 days

before slaughtering and processing; however, no difference was found in converting dietary ALA into Omega-3 LC-PUFA between T<sub>1</sub> and T<sub>2</sub> treatment. During the last two weeks of dietary flaxseed oil supplementation, i.e, 3 and 4% T<sub>1</sub> and T<sub>2</sub>, the level has shown a similar trend, as the conversion rate was found to be equal between T<sub>3</sub> and T<sub>4</sub>. This indicates that supplementation duration is more important to convert the dietary ALA into biologically beneficial Omega n-3 LC-PUFA rather than the level of dietary flaxseed oil.

Arnesen et al. [3] reported that 4-week feeding of dietary flaxseed oil yielded 4.7% n-3 LC-PUFA in broilers. The author further added that the duration of supplementation of flaxseed oil for a 2 to 4 wk period effectively enriches broiler meat with Omega n-3 LC-PUFA compared to 6 wk duration. This might be because elongation enzymes like elongase and desaturase for the synthesis and incorporation of long-chain fatty acids into breast fillet's phospholipid fractions. The flaxseed oil supplementation of T<sub>3</sub> and T<sub>4</sub> (3 and 4%) during the last 2 wk before slaughtering was inadequate for the optimal conversion of ALA into n-3 LC-PUFA.

Broiler diets were enriched with 2.5 and 5% flaxseed oil during the starter and finisher stage of production and it was observed that 5% dietary flaxseed oil for 2.5 wk feeding optimized the Omega n-3 LCPUFA contents of broiler meat [30]. However, in the present study, the n-6 to n-3 PUFA ratio of broiler breast fillet was reduced with 2 wk supplementation, i.e. 1.60 and 7.72% of total fatty acids. Moreover, Fletcher [26] and Gonzalez-Esquerra and Leeson [29] also reported similar findings by concluding that 10% flaxseed oil may increase ALA and n-3 PUFA contents but seems unable to increase the Omega n-3 LC-PUFA content of broiler chicken meat.

Dietary manipulations showed that Omega3 PUFA/LC-PUFA enriched broiler meat might be produced. It is viable to effectively reduce the supplement level and duration of flaxseed oil in the broiler ration. Dietary flaxseed oil did not affect the sensory and quality parameters of chicken meat. Replacement of conventional fat sources with flaxseed oil may reduce the Omega-6 to Omega-3 PUFA ratio and increase the overall Omega-3 LC-PUFA contents in meat. Dietary flaxseed oil linearly increased Omega-3 PUFA content and significantly lowered the n-6/n-3 ratio in broiler meat. Furthermore, regardless of the dietary flaxseed oil level inclusion, Omega-3 LC-PUFA contents were observed to be higher in groups with a supplementation duration of 21 days before processing without significant difference in meat quality and sensory analysis parameters except for the tenderness value. The producer and consumer will accept the economical production of omega-3 enriched broiler meat with optimal sensory attributes and meat quality.

The study revealed that meat quality traits were not statistically affected while  $T_1$  exhibited a significantly increased meat tenderness value. The sensory attributes remained statically unchanged. Similarly, amount of n-3 LC-PUFA may be found with 3% dietary flaxseed oil for the last 21 days of rearing with optimized meat quality and sensory attributes. Therefore, it is recommended that healthy broiler meat enriched with n-3 fatty acids may be produced by dietary manipulation.

#### Data Availability Statement

The data that support the findings of this study are available on request from the corresponding author (S. Ali) at the University of Veterinary and Animal Sciences, Lahore, Pakistan.

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#### Competing Interest

The authors have no conflicts of interest to declare. All co-authors have seen and agree with the contents of the manuscript and there is no financial interest to report. We certify that the submission is original work and is not under review at any other publication

#### Authors Contribution

S. A. and M. H. designed, planned and drafted the experiment and converted into the manuscript. N. A., conducted and collected the data. B. A., J. N. analysed the data. R. M. B., K. N. and I. B. performed interpretation of data, conception and reviewed the manuscript. All authors critically revised the manuscript for important intellectual contents and approved the final version.

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