HERBAL-ORIGIN ADDITIVES - NOT ONLY TO REPLACE CHEMICAL COCCIDIOSTATS BUT ALSO TO IMPROVE THE MEAT QUALITY OF BROILER CHICKENS

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Abstract

Broiler chicken production is widely applicable due to its rapid development, ease of feeding, low conversion ratio, and nutrients required by the human body. Natural products have emerged as a more naturalistic method of preventing coccidiosis, as have herb-based additives used to improve production performance and meat quality. Thus, the aim of this study was to analyse the impact of feed supplemented with an herbal-origin additive on broiler chickens' meat quality characteristics. The feeding trial was carried out with 1-d-old 600 Ross 308-line combination broiler chickens, which were divided into 2 groups with 4 replicate pens: the control group was fed a standard compound feed (with usual coccidiostats), and the experimental group was fed a standard compound diet (without usual coccidiostats) + 500 g/t herbal-origin additive. At the end of the trial, 10 broilers (35-d-old) were randomly selected from each group and euthanized; breast and thigh muscles were collected post-mortem. The morphological composition of carcasses and the physico-chemical composition of muscles were determined. Results showed that the herbal-origin additive in feed significantly lowered total breast fillet weight and leg muscle yield, but increased breast muscle thickness and carcass weight compared to the control group ($P<0.05$). Compared to the control group, the experimental group lowered pH in thigh muscles by 0.22, decreased fat content in the breast muscles, and on the contrary, increased it in the thigh; significantly lowered breast muscle redness and drip loss, but increased water binding capacity ($P<0.05$). An uneven trend was discovered in the cooking loss results: the experimental group decreased the indicator in the breast muscles, while the one in the thighs significantly increased ($P<0.05$). Although the tendentious herbal additive mechanisms were not revealed during our study, this kind of inclusion in poultry feed can positively impact some breast and thigh muscle quality features.

Keywords: *physico-chemical properties, coccidiostats alternatives, natural origin additives, breast muscle, thigh muscle*

1. INTRODUCTION

Meeting the increasing consumer demand for animal-based products that are healthy, natural, and nutrient-rich presents a significant challenge for animal nutritionists. Broiler chickens, due to their soft and tender meat, minimal fat content, and rapid growth, are specifically raised for meat production, which makes them a valuable source of animal protein globally, contributing to food security and employment opportunities (Kryger *et al.,* 2010). Achieving optimal productivity is essential for sustainable broiler production, but it should be balanced with the welfare of the poultry, the people involved in the production process, and the environment. In the manufacturing of broiler meat, consumers are also increasingly concerned about issues such as antimicrobial resistance, the use of additives, nutritional value, flavour, traceability, genetically modified organisms, and other factors that impact the safety and health of both the poultry and the general public (Najeeb *et al.,* 2014; Mehdi *et al.,* 2018). It is essential to address these concerns and promote sustainable practises that ensure the safety and quality of broiler meat while protecting animal welfare and the environment. Despite the increasing demand for antibiotic-free animal products, raising animals without the use of antibiotics remains a challenging task (Sacchetti *et al.,* 2005; Masud *et al.,* 2020).

The intensification of broiler chicken production has presented a challenge in finding alternatives that can improve productivity, carcass yield, animal health, and meat quality (Mehdi *et al.,* 2018). Despite being a popular branch of livestock production, poultry farming is faced with various challenges,

including the occurrence of infectious and non-infectious diseases due to inadequate biosecurity and husbandry procedures. One of the infectious diseases that affect broiler chickens is avian coccidiosis, caused by the protozoan *Eimeria* and affecting the gastrointestinal tract (Abd El-Hack *et al.,* 2021). Coccidiosis can cause stunted growth, subpar performance, increased susceptibility to secondary illnesses, and even death (De Gussem, 2007). Regular use of anticoccidials can lead to residual effects and increased resistance.

The use of herbal plants as beneficial ingredients in the feed and food production industries has gained interest due to the rising demand for bioactive components of natural origin among consumers (Sacchetti *et al.,* 2005). Herbal plants are rich in bioactive elements, such as phenolic acids, alkaloids, terpenes, tannins, and flavonoids, making them effective natural remedies for various diseases, including avian coccidiosis (Brisibe *et al.,* 2008; Abbas *et al.,* 2012; Idris *et al.,* 2017). Coccidiosis treatment with herbal items is becoming more popular, as they are a safer alternative to synthetic additives that may be hazardous to animal and human health (Muthamilselvan *et al.,* 2016; Abbas *et al.,* 2017). In addition to their medicinal properties, natural plant supplements can also improve the antioxidant defence mechanisms of broiler chickens and reduce the intensity of oxidation processes that have detrimental effects on the quality of poultry products (Mohiti-Asli and Ghanaatparast-Rashti, 2015; Ognik *et al.,* 2016). Lipid oxidation, in particular, directly impacts meat quality, causing changes in colour, flavour, nutrient loss, and reduced shelf life (Contini *et al.,* 2014; Delles *et al.,* 2014). Consumers prefer high-quality products with extended shelf lives and retained sensory and gustatory qualities, thus emphasising the need for effective preservation techniques in the poultry industry. Although there is sufficient research on the impact of herbs on the health and productivity of poultry, few studies have been conducted on the effects of herbs on meat quality. Therefore, the purpose of this study was to analyse the impact of feed supplemented with an herbal-origin additive on broiler chickens' meat quality characteristics.

2. MATERIALS AND METHODS

2.1. Animals and diets

The Law of the Republic of Lithuania on the Care, Storage, and Use of Animals has been followed when conducting research. Guided by Directive 2007/43/EC (European Union, 2007), which describes rules for the protection of chickens kept for meat production, and Directive 2010/63/EU (European Union, 2010) of the European Parliament and of the Council of September 22, 2010 on the protection of animals used for scientific purposes.

The feeding trial was carried out with 1-d-old 600 *Ross 308*-line combination male broiler chickens, which were divided into 2 groups with 4 replicate pens: the control group was fed a standard compound feed (with usual coccidiostats), and the experimental group was fed a standard compound diet (without usual coccidiostats) + 500 g/t herbal-origin additive (powder of *Holarrhena antidysenterica*, *Ficus religiosa*, *Berberis aristata*, *Alium sativum*, *Embelia ribes*, *Azadirachta indica*).

According to Council Directive 2007/43/EC (European Union, 2007), birds were kept in separate pens with wood shaving litter on the floors and stocking densities that did not exceed 33 kg/m^2 for any group. The temperature and lighting schedule adhered to Aviagen Incorporation's recommendations (Aviagen, 2018). The birds had unlimited access to food for five weeks while being fed. The diets were created to satisfy the *Ross 308* broiler chickens needs for energy and nutrients (Aviagen, 2018). Each pen allowed the bird free access to drinkers and hanging feeders for food and water. Table 1 lists the qualitative indicators of compound feeds.

Indicator	Composition	
	Starter feed 1–24 d.	**Starter feed 25–35 d.**
Metabolizable energy, MJ/kg	12.66	13.17
Crude protein*, %	23.00	21.50
Crude fibre*, %	2.31	2.55
Crude ash*, %	6.54	6.04
Crude fat*, %	5.74	7.08
Methionine, %	0.78	0.74
Lysine, %	1.44	1.34
Methionine/Cysteine, %	1.12	1.06
Threonine, %	0.97	0.91
Calcium*, %	0.96	0.88
Phosphorus*, %	0.67	0.61
Sodium, %	0.17	0.16

Premix composition (1 kg of feed): IU: vit. A 1200, vit. D3 500; Fe 54.00, Zn 65.00, Cu 9.0, Mn 58.5, J 0.90, Se 0.14, Co 0.18, vit. E 36, vit. K3 3.50, vit. B1 2.50, vit. B2 7.20, vit. B4 vit. 0.08, B6 5.0, vit. B12 0.03, biotin 0.15, folic acid 1.8, nicotinic acid 40.5, calcium-D pantothenate 12.6.

Note, *- Calculated according to the NRC (1994).

Table 1. Qualitative indicators of compound feeds

2.2. Slaughter performance and physico-chemical analysis

At the end of the feeding trial (35 days of age), 10 broiler chickens were selected from each group (total n = 20) and euthanized in accordance with the recommendations for euthanizing experimental animals (Close *et al.,* 1997). The slaughter was carried out at a commercial slaughterhouse in accordance with established procedures that complied with the laws of the Republic of Lithuania (Order No. B1-866 of October 31, 2012, of the Director of the State Food and Veterinary Service on the approval of requirements for the keeping, care, and use of animals for scientific and educational purposes).

Immediately after slaughter, the eviscerated carcass, breast, thigh, and abdominal fat were weighed and calculated as a proportion of live body weight. The physico-chemical characteristics of 20 breast muscles and 20 thigh muscles (n = 10 breasts/thigh/group) were assessed.

Using a Minolta colorimeter CR-300® (Minolta Camera Co., Osaka, Japan) with an illuminant D65 and a 0° observer, a freshly cut broiler chicken's breast and thigh slices were subjected to colour measurements using the lab system (L*: lightness; a*: redness; and b*: yellowness) after being oxygenated at 4°C for 1 hour after slaughter.

The muscle pH values of broiler chickens were assessed using the Inolab 730 analyzer (WTW GmbH, Weilheim, Germany) 24 hours after the animals were slaughtered.

The procedures outlined by Honikel (2009) were used to determine the drip loss. To prevent evaporation, the weighted, 3 cm-thick transverse slices of the investigated muscles (25–30 g) were put in mesh bags and hung on hooks at 4°C. After 24 hours, the samples were reweighed to assess the weight change. Percentages (%) were used to express drip loss.

The pressing method was used to calculate the water holding capacity (Zamorano and Gambaruto, 1997). On ash-free filter paper, a 300-mg piece of beef was inserted and weighed using an analytical balance. A plastic plate was positioned on top, and a 1 kilogramme weight was used to press it down. After being left for 10 minutes, the weight was removed, and a graphite pencil was used to outline the compressed meat's borders on the filter paper. The inner and outer spot moisture areas are estimated using the planimeter DIGIPLAN Digital Planimeter 300 (Gebrüder Haff GmbH, Pfronten, Germany) to determine the spot area. A percentage (%) was used to express the water retention capacity.

The cooking loss of the breast and thigh muscles of broiler chickens was calculated using Honikel's methods (1998). Approximately 30 g of meat samples were weighed, put in polyethylene bags, and cooked for 30 minutes at 90°C in a circulating water bath. The samples were boiled, allowed to cool at room temperature, then taken out of the bags, drained, and weighed. The cooking loss was determined using the difference in mass between the raw and cooked samples, and the result was expressed as a percentage (%).

Standard techniques were used to conduct a chemical analysis of the broiler chicken muscles, including: dry matter: heating the samples to 105°C until they reached a constant weight; extracting the fat using a Soxtec System and petrol ether at 40–60°C; calculating the protein content using the Kjeldahl method (King-Brink and Sebranek, 1993); and incinerating the samples at 600°C to determine the ash content (AOAC, 2006).

2.3. Statistical Analysis

The data obtained during the study were analysed using SPSS for Windows, version 25.0 (IBM Corp., released in 2017; Armonk, NY, USA). Prior to data analysis, the normality of the data was determined using the Kolmogorov-Smirnov test. For normally distributed data, a parametric independent T-test was performed on all data obtained during each trial period to identify any differences among the different groups. Statistical significance was defined as a *P* value less than 0.05 ($P<0.05$).

3. RESULTS

Table 2 shows the effect of the herbal origin additive on the morphological composition of the carcasses of broiler chickens. The results show that in the experimental group where herbal origin additives were used in broiler chicken feed, the thickness of breast muscles and weight of carcasses of broiler chickens increased statistically reliably ($P<0.05$). Also, under the influence of the additives used, a statistically lower total weight of breast fillet and leg muscle yield of broiler chickens was determined compared to the control group ($P<0.05$). No statistically significant differences were found for other indicators of the morphological composition of the carcasses of broiler chickens.

Indicator	Control	Experimental
Weight of live bird before slaughter, g	3182.95±40.02	3070.32±34.94
Carcass weight without feathers, head, and legs, with viscera, g	2554.16±30.97	2437.92±26.72
Mass of fully eviscerated carcass, without viscera, g	2159.43±31.61	2094.18±30.67
Weight of wings, g	165.30±2.08	165.11±3.90
Weight of thigh muscle (with bone), g	386.38±3.58	342.50±34.97
Weight of drumstick (with bone), g	263.85±10.66	257.28±25.07
Weight of thigh muscle (without bone), g	333.74±3.49	290.70±32.70
Weight of drumstick (without bone), g	198.75±6.60	202.98±25.30
Total weight of breast fillet, g	787.15±13.41[a]	745.28±7.51[b]

Weight of outer breast fillet, g	655.46±19.37	624.26±6.78
Weight of inner breast fillet, g	132.36±14.05	120.72±2.67
Length of breast muscles, cm	20.18±0.66	18.94±0.31
Width of breast muscles, cm	11.80±0.51	12.66±0.21
Thickness of breast muscles, cm	3.36±0.14[a]	3.90±0.15[b]
Length of keel bone, cm	10.90±0.33	11.28±0.43
Weight of abdominal fat, g	25.76±4.81	22.12±3.55
Length of femur bone, cm	8.90±0.19	8.62±0.32
Length of tibia bone, cm	10.74±0.23	11.36±0.31
Weight of carcass, g	489.60±12.01[a]	506.58±18.13[b]
Carcass yield, %	84.55±0.70	85.89±0.66
Breast muscle yield, %	36.45±0.32	35.61±0.55
Leg muscle yield, %	30.10±0.33[a]	28.63±0.37[b]

Note, [a,b] Means in a line with different superscripts differ (P<0.05)

Table 2. Effects of herbal origin additives on broiler chickens' slaughter performance

The results of the chemical breast and thigh muscle properties of broiler chickens are presented in Table 3. Analysing the pH values of the muscles of broiler chickens, it was found that after adding herbal origin additives to broiler chicken feed, the pH value in the thigh muscles of broiler chickens decreased statistically significantly compared to the thigh muscle samples of the control group (P<0.05). Meanwhile, the amount of fat in the breast muscles of the broiler chickens in the experimental group tended to decrease and that of the thigh muscles increased compared to the control group (P<0.05).

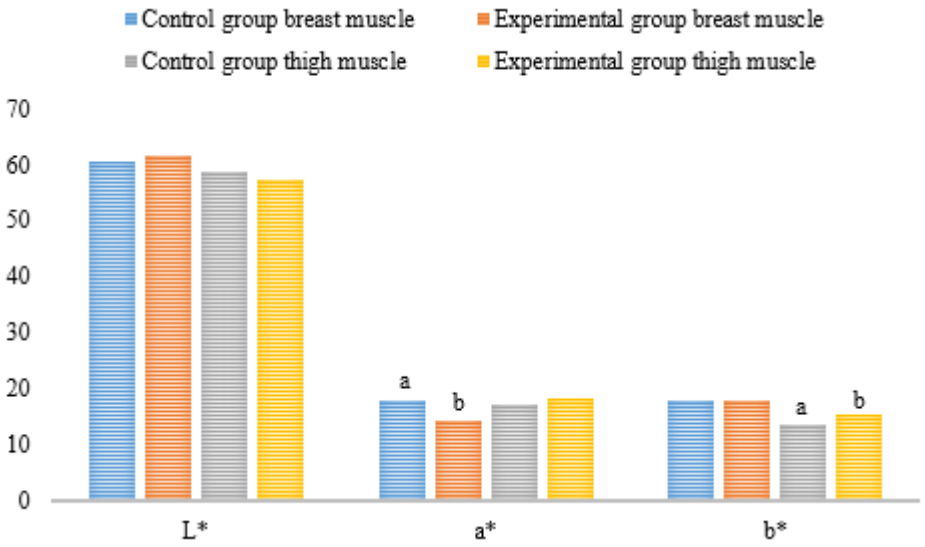
Indicator	Breast muscle		Thigh muscle	
	Control	Experimental	Control	Experimental
pH	5.72±0.05	5.64±0.05	5.98±0.07[a]	5.76±0.05[b]
DM, %	25.09±0.62	24.70±0.66	28.28±0.32	28.16±0.93
Fat, %	3.26±0.30[a]	2.60±0.21[b]	8.68±0.26[a]	9.20±0.28[b]
Ash, %	1.40±0.03	1.37±0.02	1.29±0.02	1.30±0.02
Protein, %	20.43±0.78	20.74±0.62	18.31±0.40	17.66±0.72

Note, [a,b] Means in a line with different superscripts differ (P<0.05)

Table 3. Effects of herbal-origin additives on the chemical muscle properties of broiler chickens

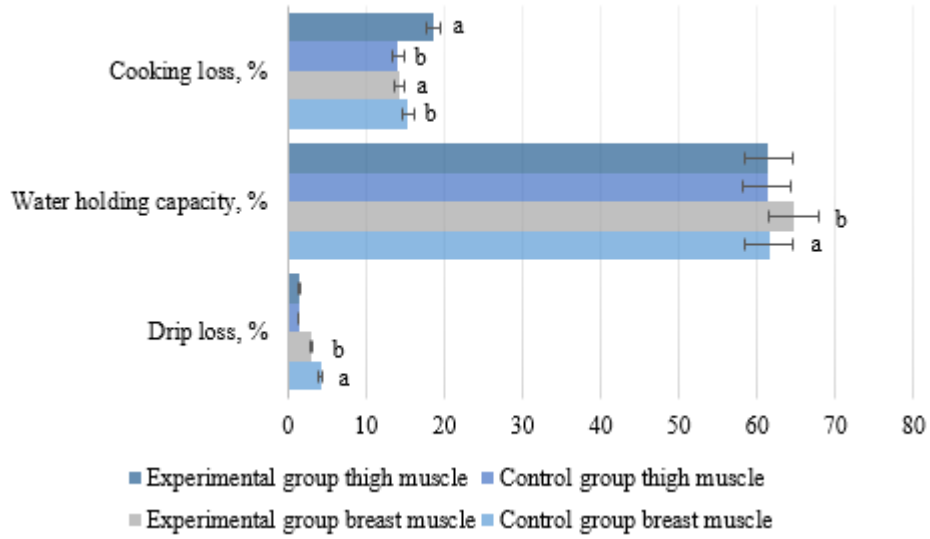
The colour characteristics of the breast and thigh muscles of broiler chickens are presented in Fig. 1. Statistically reliable, no differences in colour lightness (L*) were found in breast and thigh samples of broiler chickens, but differences in pinkness and yellowness were visible. It was found that when origin herbal additives were added to the experimental group's broiler chicken feed, the redness (a*) in the breast muscles of broiler chickens decreased statistically significantly, and the yellowness (b*) in the thigh muscle samples increased, compared to the control group (P<0.05).

Fig. 2 shows the results of the physical properties of the muscles of broiler chickens using herbal-origin additives in broiler chicken feed. After adding herbal origin additives to the feed of broiler chickens (experimental group), it was found that the added additives had a statistically significant effect on the breast samples of broiler chickens, in which drip loss and cooking loss decreased by 1.15% and water holding capacity increased by 3.19%, as well as cooking loss, which increased by 4.41% in the thigh muscles of broiler chickens compared to the control group (P<0.05).



Note, [a,b] Means in the parameter column with different superscripts differ (P<0.05)

Fig. 1. Effect of herbal-origin additives on muscle colour coordinates of broiler chickens



Note, [a,b] Means in the parameter column with different superscripts differ (P<0.05)

Fig. 2. Effect of herbal-origin additives on the muscle physical properties of broiler chickens

4. DISCUSSION

Global meat production has increased gradually over the past few decades as a result of the continuous growth in the human population, and this trend is expected to continue steadily in the near future (FAO, 2018). The poultry industry and the scientific community are under enormous pressure to find effective substitutes as potential anti-coccidiosis agents on a daily basis because of the current trend worldwide to reduce the use of antibiotic growth promoters in poultry rations by following the example of Europe (Tewari and Maharana, 2011). There is a lot of information in the literature about the influence of natural herbal supplements on the health, growth performance, and physiological state of broiler chickens (Peric *et al.,* 2010; Mountzouris *et al.,* 2011; Saleh, 2014; Oloruntola *et al.,* 2016; Gerardo *et al.,* 2017; Abd El-Hady *et al.,* 2020). But for consumers, perhaps the most important issue is the quality of the meat, so that it is as healthy as possible and free of extraneous chemical residues.

The present study has demonstrated the significant impact of herbal origin additives on the carcass morphology and weight of broiler chickens. The thickness of breast muscles and weight of carcasses were significantly higher in the experimental group compared to the control group. These results suggest that herbal origin additives may play a role in stimulating muscle growth in broiler chickens. However, it was observed that the total weight of breast fillet and leg muscle yield were lower in the experimental group compared to the control group, indicating a selective effect of the herbal origin additives on certain muscle groups. The underlying mechanisms for this selective effect may be due to differences in the composition or metabolic activity of these muscle groups. Herbal-derived additives may also promote muscle growth and development through the presence of bioactive compounds. These compounds include antioxidants and anti-inflammatory agents that can enhance muscle cell proliferation and differentiation. Our results are in agreement with Vlaicu *et al.* (2021) research results, where the breast mass of chickens was lower in the alfalfa meal and basil group compared to the control group. In addition, Fascina *et al.* (2012) found that adding a combination of plant extracts to the diet of broiler chickens resulted in higher carcass yields. These extracts included turmeric extract, citrus and grape seed extract, Chinese cinnamon essential oil, boldo do chile leaves, fenugreek seeds, and a combination of organic acids. Isabel and Santos (2009) revealed that the breast yield was much higher in hens that received 100 ppm of a blend of cinnamon and cloves compared to the usage of organic acids, despite not having noticed variations in carcass production. When comparing the carcass yields of broilers treated with plant extracts with positive controls treated with flavomycin, Lara *et al.* (2010) observed no discernible difference.

Meat quality may improve with the addition of natural supplements to the diet. In these conditions, supplementation with natural herbal or plant additives high in chemicals with bioactive potential could reduce the performance-harming effects and mitigate some of the impacts of oxidative stress, ultimately enhancing the quality of meat (Leskovec *et al.,* 2019). The study found that the use of herbal-origin additives in broiler chicken feed can significantly impact the pH values and fat content of the muscle tissues. Specifically, the pH value in the thigh muscles of broiler chickens decreased significantly in the experimental group compared to the control group, indicating a potential increase in meat tenderness. Additionally, the amount of fat in the breast muscles tended to decrease, while the amount of fat in the thigh muscles tended to increase in the experimental group compared to the control group. However, no significant differences were found for other indicators of the chemical composition of the meat. These results may have been influenced by the polyphenols and other antioxidants in the herb, which have been shown to have a positive impact on meat quality by reducing lipid oxidation and improving tenderness (Jachimowicz *et al.,* 2022). Additionally, the herbal additives may influence the metabolism and utilisation of fats in the chickens, leading to changes in the distribution of fat within the muscle tissues. On the other hand, just like in a recent study, another author Cristo *et al*. (2022), found no effect on the pH value of the meat of broiler chickens in their studies using vegetable extracts for the prevention of growth stimulants. Meanwhile, in Vlaicu *et al.* (2021) studies using a combination diet of 5% alfalfa meal and 1% basil in broiler chicken feed, a diet containing 5% alfalfa and 1% thyme, diet containing 5% alfalfa and 1% sage, higher amounts of fat were obtained in the breast meat of broiler chickens.

The visual appearance of chicken meat, particularly its colour, significantly impacts consumer behaviour and purchasing decisions. Consequently, colour variability poses a significant challenge in the

marketing of chicken meat, as consumers tend to reject meat products that display noticeable colour differences. The results of the analysis of fresh broiler muscle colour revealed no significant differences in lightness (L*) between the experimental and control groups. However, there were significant differences in the redness (a*) and yellowness (b*) of the meat. The addition of herbal-derived additives to the feed of broiler chickens resulted in a statistically significant decrease in redness (a*) in the breast muscles and an increase in yellowness (b*) in the thigh muscles of broiler chickens compared to the control group. The changes in meat colour could be related to the antioxidant properties of the herbal-origin additives. These compounds could have protected the meat from oxidation, which could have resulted in a decrease in redness (a*) in breast muscles and an increase in yellowness (b*) in thigh muscles. It is also possible that the changes in meat colour were due to the alteration of pigments and enzymes in the meat caused by the herbal-origin additives. Following supplementation with plant extracts, Chouliara *et al.* (2007) discovered no alteration in the colour of fresh broiler chicken breast flesh. Sage and rosemary essential oils had little effect on broiler chicken meat colour characteristics, according to Chen *et al.* (2008). In a different investigation, the redness (a*) and yellowness (b*) characteristics of meat were elevated by green tea extract. Myoglobin concentration is likely what causes the changes in broiler chicken meat, but it was discovered that only the thigh meat is affected because it contains a significant number of red muscle fibres, while the breast is often fully made up of white fibres (Qamar *et al.,* 2019).

The addition of herbal-origin additives to broiler chicken feed had a positive effect on the meat quality of broiler chickens. The additives reduced drip and cooking losses in breast muscles, indicating an improvement in water holding capacity. Water-holding capacity is a crucial quality attribute of meat, as it directly affects its processing suitability. The amount of water held within the meat determines its texture, juiciness, and overall sensory properties. Therefore, understanding and optimising water-holding capacity is of great importance for the meat industry, as it can significantly impact the final quality of processed meat products. Additionally, the water-holding capacity in the breast muscles of broiler chickens increased by 3.19%. According to Reis *et al.* (2018), broiler chickens receiving an essential oil-based feed enhancer had meat that could store more water. In a study by Puvača *et al.* (2014), garlic powder was found to significantly boost the ability of broiler diets to hold water. In contrast, cooking loss in thigh muscles increased by 4.41%, which may be attributed to the higher fat content found in the thigh muscles. These results may be due to the presence of active compounds in the herbal-origin additives that affect the water-holding capacity and fat content of the meat.

5. CONCLUSIONS

In summary, the incorporation of herbal-origin additives in broiler chicken feed exerts a significant impact on several morphological and physico-chemical characteristics of chicken carcasses. Notably, the herbal supplement induces a marked increase in breast muscle thickness and carcass weight, alongside a reduction in pH value in the thigh muscle. Conversely, a decrease in the total weight of breast fillet and leg muscle yield was observed with herbal supplementation. Furthermore, a tendency towards decreased fat content in breast muscles and increased fat content in thigh muscles was observed. No significant differences in sample brightness were detected, whereas reductions in redness and increases in yellowness were noted. Additionally, the herbal additive positively affected water holding capacity and drip loss in breast samples but negatively impacted cooking loss in the thigh muscles. Overall, these findings suggest that the use of herbal-derived additives may represent a viable intervention for enhancing the quality and yield of broiler chicken carcasses.

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