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Chapter

White Striping and Wooden Breast Myopathies in the Poultry Industry: An Overview of Changes in the Skin, Bone Tissue and Intestinal Microbiota and Their Economic Impact

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Abstract

Considerable advances in the poultry industry have been observed in the last sixty years. Consequently, new technological and biological processes have accompanied the development of animals and inputs. With these new possibilities and growth in the sector, there was also the emergence of new paradigms, one of which being the different types of myopathies. In the poultry industry, the breast is one of the noble parts and, therefore, a lot has been studied about the occurrences, alterations and/or different myopathies that alter its quality characteristics. Here we will highlight White striping (WS) and Wooden breast (WB), both investigated more recently due to changes in quality characteristics and considerable losses. The objectives of this review will be to discuss the biochemical parameters of these meats affected by myopathies WS and WB and their consequences on the development of skin, bone and intestinal microbiota lesions; correlate with the impacts of these occurrences to economic losses associated with partial and total condemnations of the carcasses. Another approach is that fast-growing animals have a drop in their quality of life, impacting the well-being of birds since the inflammatory process and excess weight have a direct correlation with dermatitis, corns, arthritis and other comorbidities.

Keywords: quality of poultry meat, broilers, animal welfare, carcasses condemnation

1. Introduction

Approximately 60 years ago, there was a considerable advance in the broiler industry. Consequently, new technological and biological processes have

accompanied the development of both animals and inputs. In the last 20 years, there has been an increase in the preference for chicken meat by the general population, the trigger for this event being the affordable price of this protein source, ease and diversity in its preparation, as well as properties associated with the healthiness generated when it comes of white meats. These factors led to an increase in the poultry sector, seeking fast-growing strains, to increase production in a shorter time [1].

With these new possibilities and growth in the sector, there was also the emergence of new paradigms, one of which being the different types of myopathies. In the poultry industry, the breast is one of the parts considered noble and therefore, the occurrences, alterations and/or different myopathies that alter the quality characteristics of the breasts have been studied a lot. The most common are: Pale, Soft and Exudative (PSE), Dry, Firm and Dark (DFD), Deep pectoral myopathy or Green chest, Acid chest, White striping (WS), Wooden breast (WB) and Spaghetti meat (SM). The last three myopathies are the most recent when compared to the others mentioned. Still, there is a need to investigate further, due to changes in quality characteristics and considerable losses for the sector. In addition to permeating hypotheses to better describe what happens, why it occurs and possibly, understand the mechanism to minimize or even suppress them. Several researchers from different countries such as the USA, Italy, Spain, Brazil, Finland, Canada, China and England are looking for these answers. However, until now, little is known about the effects or action of specific proteins and protein groups on these anomalies. The difference between protein content and collagen and protein degradation rate was described in [2]. Although, protease groups are crucial to understanding what can happen with muscle activity.

Although information about the incidence of these myopathies is limited and sometimes contradictory, it is assumed that myopathic chicken breasts appear in all countries where fast-growing hybrids are used, with WS being the most common affecting up to 50% of the breasts of chicken in Italy, France, Spain and Brazil. In the Northeast of Brazil, the proportion of these myopathies is reported between 10% and 20%. In the USA there is an incidence of 98% of birds developing these myopathies, of which 55% were classified as moderate and severe [1].

The trend in the production of Brazilian chicken cuts is to continue increasing, and with that, there is a concern with the quality of this product and with the need to slaughter larger chickens with higher yield and in less time. When all these factors are achieved, a satisfactory result is achieved for the industry and the producer [3]. In a relatively short time (**Table 1**) [4], genetic selection, associated with management, nutrition and other factors, significantly accelerated the development of the muscle tissue of these birds, especially the breast (*Pectoralis major*), which

Year	Carcass weight (kg)	Age (days)	Breast size (%)
2001	2.207	43	15.8
2007 ^a	2.200	36	18.6
2012 ^a	2.200	35	21.1
2014 ^a	2.200	34	21.5
2017 ^a	2.200	34	22.0

^aRoss 308 Broiler Performance Objectives.

Table 1.
Advance in the production of breast meat (proportional to the carcass size) concerning the commercial Ross 308 line between the years 2001 and 2017 [4].

currently exceeds one-fifth of the total weight of the birds, and certainly represents the most valuable and noble part for the broiler industry.

The results of this project on White striping (WS) and Wooden breast (WB) in poultry will be useful to better understand what happens in these animals, what their impact on meat and carcass as a whole and ensures the slaughtering and consumer industries that will have, even with myopathies, viable products in terms of nutritional, physical-chemical, biochemical and technological aspects.

The birds are staying on the farms for 9 days less and increasing 6.2% in their weight in brisket. This gain is significant for the sector since to keep these animals on the farms, generates costs of food, handling and maintenance of the production system as a whole. In this sense, [5] described that a great ally for the sector's economy is the genetic selection (**Figure 1**), as it brought standardization of broilers concerning body weight, carcass yield and feed conversion, reaching thus, a possibility of slaughtering birds with greater weight in a shorter time. It is known that genetic selection in conjunction with the accelerated growth of these animals does not always achieve positive responses and the incidence of abnormal physiological consequences begins to appear more frequently and is extremely visible and significant in chicken breasts [6].

According to information published by [6], one of the most common changes in the broilers' breasts is deep pectoral myopathy (or green muscle disease) which, according to [7], occurs when oxygenation in the smaller pectoral muscles (commercially known

Figure 1.
Growth, efficiency and productivity of commercial chickens from 1957, 1978 and 2005 [6].

as “chest file” ceases or “sassami”) with degeneration, necrosis and atrophy. In reference [8] reported that this anomaly can also be caused when blood circulation ceases due to intense muscle exercise, with voluntary movement of the wings where the muscle is unable to expand and ischemic necrosis of the chest muscle occurs.

Therefore, during the deboning process, this muscle is condemned, however, when there is production of whole chicken, it is not possible to identify it, because according to Ordinance N°. 210, of November 10, 1998, the supra-coracoid muscle is exposed only when the carcass follows a more detailed evaluation at the Department of Final Inspection (DIF) or in the boning room [9]. However, in a review published by [6] that mentions research by Pereira et al. [10] found that myopathy can be a technopathy caused by changes in technology and, if possible, adapt the pre-slaughter management to reduce the damage caused, and these can be consumed, because it is not an issue food security, but rather a product quality problem. Reference [11] pointed out that the green color probably came from the transformation of myoglobin in anaerobic conditions and not from inflammation process.

Others anomalies in the chest and small chest file have caused damage to the refrigerators. In [12] observed in Finland that there was an increase in chicken breasts with abnormalities that was characterized in the pectoralis major muscle, with pale and hard external areas with white streaks, in which they caused rejection by the consumer and, consequently, there were economic losses in industries. They did not find a relationship between these anomalies and any antemortem symptoms.

Through electron microscopy analysis, this type of anomaly was called WS (**Figure 2**). Still, concerning anomalies, another occurrence in chicken breasts was investigated by [13] and called WB, however, the breast had a yellowish color and a certain hardness accompanied by inflammatory processes and necrosis. Both anomalies had similar histological characteristics. However, there is no information about the implication of these anomalies in the quality of the products, as well as the training mechanisms.

White streaks of chicken breast called WS (**Figure 2**), according to [8] are related to adipose tissue according to histological and chemical analyzes. While the characteristics of chicken breast with WB are related to the connective tissue that was characterized by muscle hardening [12]. Thus, these two myopathies highlighted in the present proposal, present specific differences, being that WS was characterized by the development of white fibers of the connective tissue and that

Figure 2.
Breast fillets displaying different degrees of white striping. Score 0 indicates no white striping and score 3 indicates severe white striping [8].

was developed in parallel to the muscle fibers, with the inclusion of adipose decision [14]. While WB according to [15] presented heterogeneity of color, excessive superficial exudate and loss of muscle elasticity.

Published by [16] illustrates in a very creative way how the muscle affected with WB develops pressure resistance. These authors explain the probable etiologies for the development of myopathy and address interesting issues such as nutritional, environmental and genetic aspects.

When or when the animal dies, there is blood circulation failure as a result of bleeding, which causes depletion of oxygen and nutrients. The metabolism then uses the oxygen associated with myoglobin to continue the aerobic process. When O₂ reaches its critical limits, the main metabolic pathway for ATP generation becomes the glycogen reserve. This scenario fully characterized as anaerobiosis, generates lactic acid and reduces the concentration of ATP until it no longer exists in the process. In the sequence, the actin-myosin interactions begin, forcing the muscle to enter a phase of continuous contraction until the muscle enters an irreversible phase contraction known as rigor mortis. Thus, glycogen levels begin to decrease and lactic acid is the product of this metabolism, which accumulates in the muscle fiber, acting as an indicator of the post-mortem glycolysis rate and directly results in a reduction in pH after 24 hours of slaughter. Finally, there is a proteolytic rupture of the muscular structure, which can last up to two weeks, with increased flexibility and tenderness of the meat, this last stage is known as *post rigor mortis* [6, 17].

There are several negative consequences concerning the development of these myopathies directly on the quality of chickens cited by [1], which in turn affects consumer preferences. Therefore, the search for possible solutions to prevent this occurrence is one of the main objectives for food and animal production scientists. Therefore, a prior indication of the pathological pathways of these myopathies is necessary.

2. Economic impact and losses for the poultry industry

A variable portion of chicken breasts affected by the aforementioned myopathies were reported by [1] and may be due to: This author lists in the following sequence, factors that are a consequence of myopathies, being: (a) condemnation/cut (whole breast, carcass); (b) lower yield and value since there will be changes in the water retention capacity [WHC], emulsification and gelation capacity; (c) manual separation in the deboning line to be intensified (addition and training of personnel for classification/sorting - highest cost); and (d) there will be rejection from consumers, since undesirable sensory changes occur in this meat". Consequently, all these factors are responsible for economic loss in the poultry sector.

As there are different forms of myopathies already cataloged, if all of them manifested together, certainly a refrigerator could lose millions in a few hours of slaughter, taking into account that there are systems that work with 350.00 heads of birds slaughtered in a 12-hour period (personal note author). Knowing this, several studies were carried out to try to quantify the percentage and some of the dollar values of the damage that WB and WS can generate.

In a study carried out in the United States, it was reported that more than 50% of the studied squad developed WB [18]. Additionally, study with growing birds on farms under commercial conditions observed that 96.1% developed WB [19]. In 2017, in Italy, [20] observed 474 carcasses and of these, 53.2% developed WB. Regarding values, there are two articles regarding this survey, with Kuttappan et al. [16] indicating annual economic losses for the American market in the range of \$ 200 million. For the Brazilian market, there are estimated values of

\$ 70,632.00 thousand/day, this being the only work that associated WB with WS to calculate economic losses [21].

The data provided above are reported by different authors, however, the group responsible for preparing this review has worked directly with slaughterhouses in the Southern Region of Brazil and some data are being obtained gradually. Data collected during 30 days of slaughter monitoring during the year 2020 raised the following numbers: of the total and partial convictions of the carcasses as a consequence of dermatoses, myopathies, arthritis, contusion/fracture and contamination, a loss of around \$ 162,926.49 occurs a total of 2 million birds slaughtered. This value represents about X% of monthly loss for the sector. If there is an extrapolation that these condemnations can be minimized in the same way as WS my WB myopathies and that they can be associated with the same etiologies, that is, with the rapid growth of these animals, and this equation can be adjusted so that the gain of weight is gradually adaptable to the physiology of these birds, these changes could be minimized and the condemnations, in turn, would gradually decrease, which would generate greater revenue for the slaughterhouses.

Given the great economic loss caused by these myopathies, their characterization and understanding concerning their development, changes in meat quality standards, possible changes in carcasses beside the affected muscles and how these occurrences can affect meat processing, whether or not being associated with a loss of welfare of these animals is necessary information for the sector. The compilation of this information, in a systematic way, could minimize the negative impact of these myopathies, resulting in a higher yield for the sector, as well as an improvement in the quality of the final product that will reach the consumer.

3. Proteins and connective tissue

Protein material is a determinant when it comes to meat. In reference [6] comments that the muscular protein organization and nature is important in the way they are metabolically reversed. The skeletal muscle has three proteic classes based on its solubility; the myofibrillar class is the main one and, as myofibrillar proteins are built in a myofibrillar structure in the striated muscle, presenting a challenge for protein turnover. Analyzing these recent advances in the understanding of this protein system, there are indications that myofibrillar proteins are first hydrolyzed before being degraded and reused. It is still not entirely clear how this dissociation occurs, it is suggested that there may be the release of a group of easily hydrolyzable myofilaments, or it may involve the exchange of myofibrillar proteins in the cell's cytoplasm, or both mechanisms may occur at the same time [22].

In addition to the information mentioned above, on quality parameters for chicken meat, it is necessary to target other lines of research that can provide differentiated responses, or that the responses add up to have a system of information that takes the responses that the industry is looking for. Therefore, research on protein turnover in broilers has been carried out for a considerable time, however much is still not known about the subject. Still referring review of [6], the authors report that the protein turnover in laying hens is associated with the differences between sexual maturity, age, stage and feeding posture.

Genetic-based research with genes related to protein degradation (IGF-1, ampk, anthrogin-1, MURF1 and Cathepsin B) is being developed, however, its expression changes due to changes in protein turnover. These genes have already been sequenced for chickens [23–26]. In an article published in 2017, the authors [27] make interesting comments, some of which are briefly as follows: protein synthesis rates are not different for normal meats or WS, however, protein degradation rates

are different between both; increased expression of the MuRF1 and Anthrogin 1 genes are responsible for the highest rate of protein degradation. Despite these sequences, there is still no understanding for these protein changes in birds. This can theoretically lead to greater protein degradation and consequently high catabolic rates, leading to an imbalance in the body of these birds, resulting in the production of proteins that may be deficient, as in the case of collagen for supporting the skin.

In addition to the possibilities involving protein turnover, and as this can influence muscle development, there are still other hypotheses that are being raised to better define the process of meat formation with WS and WB anomalies. One of them is associated with the number of mitochondria present in the tissue, indicating the possibility of a low concentration of these organelles in these muscles [28]. This could explain the need for healing processes in these muscles, which are characterized by muscle striation, indicating the need for deposition of connective tissue.

In addition to this information, studies are addressing the welfare of these birds, since compromising the structure of the carcass as a whole, there may be a direct impairment of their mobility. Reference [29] observed a degree of structural abnormality in all samples of chickens with rapid weight gain. However, there are symptoms in birds that do not affect all individuals affected by WB, however, there may be possible links with the environment [16]. It is not known numerically whether WB or WS affects the behavior, the ability to walk or the welfare of these birds. The hypothesis has been raised that these myopathies would harm mobility and, therefore, have welfare implications for animals.

If there is bibliographic support concerning the mobility of these animals, consequently, this information can be extrapolated to a probable deficiency in the production of connective tissue, since there must be a deviation of nutrients in the metabolism of these animals, to supply the inflammatory processes. Muscle and joint. According to [30], the main histological lesions of the WB muscle, which consist of chronic myodegeneration with regeneration and interstitial edema, accumulation of loose connective tissue or fibrosis and replacement of severely degenerated muscle fibers with connective tissue with excessive fibrosis.

As the connective tissue is crucial for the development of the entire system, as well as muscles, skin and bones, there is a need for a balanced distribution of nutrients throughout the animal's body. In a study published by [31] there is a report on the difference in the organization of collagen, which may be due to the expression of proteoglycan decorin in the extracellular matrix. Decorin is a regulator of collagen crosslinking and is expressed in levels significantly in strains affected by WB, which would lead to tightly compressed collagen fibers due to the high levels of crosslinking of this protein. Besides, the expression of muscle-specific transcriptional regulatory factors for proliferation and differentiation of muscle cells that lead to muscle regeneration in response to muscle damage was significantly elevated for these myopathies. The lack of decorin has been mapped as a destabilizer of the collagen structure due to abnormal collagen crosslinking, leading to fragility of the skin, caused by an abnormal fibrillar network [32].

3.1 Collagen, bone tissue and fractures

The information mentioned above in this brief literature review shows an overview of the problem, however, through reports from slaughterhouses, in the day-to-day work of the Research Group that is developing this proposal, there is evidence that the animals that develop WS or WB are more likely to have problems associated with the quality of their skin. It has been observed in the field and in

the slaughter plants that these animals have the most fragile skin, with lesions that occur both within the farms and during the plucking process, there may be a greater correlation of bone fractures with these anomalies and consequently a considerable economic loss for the sector, since the carcasses may have partial or total condemnation.

Research developed by [33] related WB myopathy with problems associated with the formation of bone tissue. These authors evolve an interesting line of reasoning based on the development of bone marrow-forming cells and how the increase in adipocytes can influence this process. As a result, they obtained positive correlations, and the levels of calcium and phosphorus in the bone matrix of animals that develop WB are lower when compared to those not affected by myopathy.

A study by [34] that analyzed the efficiency of magnesium in the control of oxidative processes in birds, as well as its correlation with the decreased incidence of development of WS and WB myopathies, concluded that magnesia supplementation protected the tissue against protein oxidation and that it reduced the incidence of WS and WB myopathies to almost half the occurrence in fed animals supplemented with this mineral. In the same study, these authors report that even Calcio and Magnesium use unusual mechanisms of divalent ions for their absorption, one does not harm the absorption of the other, and the same occurred for Phosphorus. The results were innovative since magnesium did not interfere with the action of other minerals, a positive factor, since this benefits the bone matrix, so supplementation with magnesium in the feed of broilers can be a promising alternative as a supplement to mitigate the development of WS and WB myopathies.

4. Microbiota

Another parallel approach to be considered is the intestinal microbiota of birds, as this is an innovative aspect of the proposal since recent studies have verified the importance of the intestinal microbiota in animal performance, health and well-being [35–37]. There is still a close relationship between the diet, microbiota and bioactive compounds that may be present or that are used commercially in poultry feed. Studies that address the interaction of the microbiota are still limited and are at the frontier of knowledge under the paradigm of sustainable poultry production, prioritizing animal welfare.

The microbiota is recognized as the “fifth organ” and the literature suggests that the microbiome plays a crucial role in signal processing and interaction with the environment [38]. The composition of the bacterial microbiota is affected by the bacteria present in the intestine and by the natural microorganisms in the environment [39]. Chicks born in natural conditions receive the microbiota from adults, mainly from the mother. Industrial poultry farming has altered this condition, preventing the chick from coming into contact with the mother, which leads to a delay in the development of the protective intestinal microbiota [40, 41]. The balance of the microbiota can be affected by several factors, both endogenous and exogenous. Poor hygienic-sanitary conditions, stress, food, intoxication and illness, can trigger the increase in bacterial proliferation that can compete for nutrients. They can also determine inflammatory processes, which leads to thickening of the intestinal wall, which will reduce absorption, increase the excretion of metabolites and toxins that trigger enteritis and decrease the transit time of the digesta. Besides, it can increase the turnover of epithelial cells, which allow bacterial and endotoxin translocation to other organs, leading to septicemia [42]. According to [43] it is important to understand and have control over the possible changes in the intestinal microbiota to adapt the management and to include in a rational way additives that

can alter and regulate the microbial ecology, to improve zootechnical performance and reduce some effects of stress or the harm of diseases.

In adult birds, when the microbiota is established, it may contain 400 to 500 microbial species [44]. With variations in the amount and types of microorganisms that may be attached to the epithelium or free in the lumen. When free, they may have an accelerated multiplicative capacity, minimizing loss through peristalsis, and may be associated with other bacteria that are linked to the mucosa. These variations mean that, in general, the small intestine is colonized by facultative microaerophilic bacteria, with their respective representation (in percentage) in the microbiota, which is: *Lactobacillus* (70%), *Clostridiaceae* (11%), *Streptococcus* (6.5%), *Enterococcus* (6.5%). The cecum, on the other hand, has mandatory anaerobic bacteria such as *Clostridiaceae* (65%), *Fusobacterium* (14%), *Bacteroides* (5%), and is also permeated by facultative microaerophilic bacteria such as *Lactobacillus* (8%) and *Streptococcus* and *Enterococcus* [45, 46].

A study by [47] who investigated the microbiota of WB and normal birds to understand the differential expression of plasma metabolites, obtained different results between groups, with non-myopathic broilers produced more heat, with higher body protein content, validated by the higher protein: fat ratio. Lower protein content in myopathic birds was verified, due to the probable high myodegeneration, as observed by the high expression of 3-methylhistidine in plasma. In this work, the authors also reported that there was a predominance of unclassified *Lactobacillus* in birds with myopathy; while the species, *L. acidipiscis* was the predominant bacterium for non-myopathic broilers. The differentially significant metabolites identified in the plasma metabolome between the two groups were homocysteine, cyclic GMP, trimethylamine N-oxide, tyramine, carnitine and acetylcarnitine, all associated with the cardiovascular system. The results of this work suggest that more research on broilers should be carried out with a focus on tissue vascularization.

As WS and WB are proven to be inflammatory processes that permeate the entire carcass, it is possible to raise the hypothesis that alteration in the microbiota may be determinant for birds to be predisposed to develop these anomalies, consequently, there may be changes in the absorption of limiting amino acids or even essential for the synthesis of important proteins such as collagen and other proteins that can provide resistance to blood vessels, bones and skin.

5. Animal welfare: possible changes due to the occurrence of wooden breast and white striping

Brazilian poultry stands out in the international meat market, and according to data from the Brazilian Association of Animal Protein (ABPA 2018), Brazil was in 2nd place in the world production of chicken meat (2017) with 13.05 million tons, second only to the United States, which produced 18.6 million tons. As for exports, Brazil occupies 1st place, exporting approximately 4.3 million tons [48].

Poultry intended for meat production must be assisted to comply with specific conformities for farms, such as maximum animal density, minimum lighting intensity, air quality, water and food availability, among others. Also, meat mortality and inspection data are considered to establish maximum values of stocking density. Dermatitis, parasitic infections and systemic diseases should also be measured to identify signs of poor well-being [49].

Some factors that have directly influenced this growth system are: improvement of lines and inputs, automation of process systems, sanitary conditions for the creation of controlled birds, integrated production system, among others [50].

There is a projection of a 46.4% increase in chicken meat production by 2023. Simultaneously, there is a growing demand for information on ethical aspects of animal production. In a study conducted between the states of Paraná, Santa Catarina and Rio Grande do Sul, evaluating the condemnation of birds, it was observed that injuries were the main cause of condemnation in 2015, followed by dermatoses. These same authors report that the third highest incidence of condemnation is associated with dorsal skull myopathy [49]. In the State of Paraná, which slaughters more than the others in absolute numbers, the percentage of convictions increases. The progressive increase in rates of injury conviction, inadequate bleeding, arthritis and aerosaculitis can indicate important aspects of well-being.

The increase in convictions caused by arthritis may indicate that the industry has undergone significant changes in recent years, which result in a negative impact on the welfare of birds [51]. This information goes from other information obtained with the weight gain of these animals since birds are gaining weight very quickly. These same authors indicate that genetics may be the main cause of skeletal disorders in fast-growing breeds, causing a lack of activity in the birds, which because they are very heavy, aggravates the problem [52, 53]. The high stocking density is also related to the reduction of air quality, increased thermal stress and increased transmission of infectious diseases.

A study by [30] observed that for birds with WB there were changes in the birds' behavior since they associated this difference with the change in the way of walking of these animals. Which may suggest possible effects associated with loss in the welfare of these birds. Another study by [16] have already reported a possible change in the growth conditions of these birds, directly affecting their welfare.

Approach to the welfare of these birds is essential, as it compromises the structure of the carcass as a whole and can directly compromise its mobility. In reference [29] it was observed a degree of structural abnormality in all samples of chickens with rapid weight gain. However, in another study by [16] reported that there are symptoms in birds that did not affect all individuals affected by WB, however, there may be possible links with the environment. However, there is still no quantitative information on whether WS or WB affects the behavior, ability to walk or the welfare of these birds. There is a chance that these myopathies would harm mobility and, therefore, have welfare implications for animals. With this information, it would be interesting to study a variable that can correlate the stress level of birds with the myopathies associated with this proposal, with cortisol being an excellent marker for this purpose.

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Conflict of interest

The authors declare that there is not any conflict of interest.

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