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Effect of strain, sex and process parameters on water to protein ratio of chicken cuts

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Abstract

To keep up with chicken meat demand, genetic improvement, field performance, and technological processes have been sought. However, some consequences of this are already being observed, such as constant divergences in physicochemical parameters of in nature chicken cuts, since legislation has not been revised at the same speed. Thus, this work aimed to evaluate water content, protein and water content/protein ratio (W/P) for different conditions (slaughter, lineage, sex, and processing) and compliance with current legislation (Brazilian and European). Breast and half breast, drumstick and thigh cuts of in nature chicken were assessed. A significant difference was found in sex, with lower values in the females. Chickens have water content levels higher than those required by Brazilian legislation. In the different cuts evaluated, discrepancies were observed in relation to legislation. There was no compliance with W/P ratio to European legislation, where incompatibility approached 26.67% for skinless breast, 6.67% for breast and half chest and 64.44% for thigh and drumstick. The results reinforce the need for continuity of studies that support the revision of current legislation, clarifications regarding the calculations used to determine the established standards and clarifications of the collection and analysis methodologies currently employed.

Keywords: poultry; legislation; water absorption; water content/protein ratio.

Practical Application: The results show that variations in the water content/protein ratio are inherent to the processing and the lineage and sex of chickens, which are not foreseen in the current Brazilian legislation, indicating the need to review this one.

1 Introduction

The high demand for meat products will make companies face many challenges, one of which is meeting current legislation. In this sense, companies already face difficulties in meeting parameters of water content, protein, and water to protein ratio of *in nature* chicken cuts provided in Brazilian legislation - Normative Instruction nº 32 of 3 December 2010 (Brazil, 2010). The European commission regulation (EC) nº 543/08 of June 16, 2008 (European Union, 2008) only sets maximum values for water content/protein (W/P) ratio in chicken breasts with and without skin, thighs and drumsticks.

Genetic changes in chicken's lineage are carried out in order to improve animal conversion, increase average weight, and carcass yields in slaughterhouses (Valentim et al., 2019). Thus, there are also physiological changes in birds, such as percentage of water and protein, or in the centesimal composition of carcass musculature (Ferrari et al., 2016). Given these evolutions, physiological studies of muscles should be conducted concomitantly taking into account variable parameters like weight and age; as well as changes in the technological process (air or water immersion cooling, as well as other specific parameters) in order to be updated with current legislation.

The established regulations do not always follow this entire technological advance, and may hinder production in the whole productive chain due to the incompatibility between the established and obtained by the industries.

Poultry meat processing is similar in many parts of the world and consists of basic steps: receiving, hanging, numbing, bleeding, scalding, plucking, eviscerating, pre-cooling, cutting and packing (Sams, 2016). Questions regarding compliance with water content, protein, and W/P ratio parameters begin at the reception of chickens. A European study conducted in 2012 confirmed that younger birds have slightly higher physiological water content and lower protein content (Elahi & Topping, 2012).

Among the operations involved in chicken processing, post mortem cooling is very important to maintain the meat quality. Postmortem temperature has a critical factor of quality, and reduction of carcass temperature must start as soon as possible after slaughter (Mir et al., 2017). With this procedure, post-mortem biochemical reactions are reduced, preventing rapid pH drop and uncontrolled action of natural proteolytic enzymes, besides inhibiting microbiological growth (Mastrogiacomo, 2006). Besides, the temperature reduction in the carcass by the heat exchange with the chiller, where water absorption percentage is related to the residence time, system air injection (bubbling), carcass final temperature, carcass size, and other parameters of low significance since carcasses absorb water mainly in the skin, surrounding fat and subcutaneous tissue (Assis, 2009).

If these parameters are not known and controlled in each process, they may result in absorption percentages higher than

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those established by the legislation. For pale, soft and exudative (PSE) cuts the high value of water content and small protein quantity results in a high W/P ratio, due to the low water retention capacity of PSE meat. Also, low pH, close to the isoelectric point of the proteins, presenting negative and positive charges in equal quantity decreases the space between the thick and thin filaments and do not allow the binding of these molecules with water (Mantese, 2002).

In chicken processing, the product should be cooled in the pre-chiller and chiller by immersing the carcasses in ice water or in refrigerated air. At this stage the muscle tissue incorporates water that should come out of the chicken before frozen, otherwise the incorporated water will freeze along the product, which will have its exceeded weight, and can lead to fraud problems and economically harm the consumer (Kato, 2013).

In this sense, this work aims to evaluate the influence of different fasting times on water content, protein and W/P ratio in different chicken's cuts. Besides evaluating different cuts *in nature*, the process conditions was fixed, having as variations the process steps, lineage (Cobb and Ross) and sex (male and female).

2 Materials and methods

2.1 Samples

Samples were obtained from a poultry slaughterhouse located in southern Brazil, under the Federal Inspection Service. The fixed parameters were chicken age of 43 days and 2.8 kg of weight (± 100 g). The samples collections for the treatments occurred in one farm, where the chickens have the same management and feeding.

After the chicken's slaughter, cuts (breast and half breast, drumstick, and thigh) were obtained, then they were frozen in a continuous tunnel (Mod. Madef) at -19 °C for 5 h until reaching -12 °C, and then the samples were sent to the analysis laboratory. This laboratory is recognized by INMETRO, compliant with NBR ISO/IEC 17025 norms. The methodologies used followed Normative Instruction nº 08/2009 (Brazil, 2009): The parameters analyzed in the chicken's cuts were water content, protein and calculated W/P ratio.

2.2 Fasting time evaluation

The influence of food and water fasting time in water content, protein, and W/P ratio was assessed. Four different treatments of fasting from the same group were evaluated: T1 = 0 h, T2 = 6 h, T3 = 12 h and T4 = 18 h. T1 represents the condition without any dietary restrictions. T2 is the minimum period that should be practiced, according to the Brazilian legislation, Ordinance nº 210/1998 (Brazil, 1998) and also most usual for slaughterhouses. T3 is the threshold practice and T4 is an extreme condition.

For each treatment 50 samples were analyzed. Two *in nature* cuts were used as reference. Half breast without skin and bone, and drumstick and thigh with bone and skin. These cuts were selected according to data from slaughterhouse that present great divergence from the parameters studied in the last 3 years.

2.3 Process mapping

In the process mapping, steps with high use of water, with the possibility of aggregation of water content in the carcass and/or cuts (reception/hanging, pre-chiller, chiller 1, chiller 2, post-chiller and after freezing) were assessed. The fixed variables evaluated were lineage (Cobb), sex (male), weight (2.8 kg ± 100g), age (43 days) and fasting time (6 h). The set process parameters used were total cooling time (1:20 h), pre-chiller water temperature (13 °C), chiller 1 water temperature (2 °C), chiller 2 water temperature (2 °C), counter flow bubble (air injection in the pre-cooling system), and freezing temperature (-18 °C).

Sampling was performed at three different times, with n = 10 at each stage, 30 samples at each point (180 samples in total). As a reference for the study, the boneless and skinless breast cut was used. The carcasses were separated, evaluated in relation to the absence of tears or injuries in the skin, since these damages can interfere in the water absorption (Lorenzetti et al., 2018).

2.4 Comparison between the cuts

After the process mapping and identification of the steps that showed significant difference, the different cuts (leg, drumstick and thigh, breast and half breast) of *in nature* chicken were evaluated. The samples were obtained on the platform, pre-chiller and freezing tunnel. For each step, 40 samples were collected and analyzed.

2.5 Evaluation of lineage and sex

The two lineages, Cobb and Ross, most used by the company were evaluated considering the same sex, male. In the male and female sex evaluation, the Cobb lineage was used. Forty samples of each lineage and each sex were analyzed using the skinless breast cut. The samples were obtained in the stages of hanging, pre-chiller and frozen process. All samples followed the same preparation flow.

2.6 Determination of water content

The water analysis was performed weighing 5 g of the sample (in a capsule containing around 15 g of sand and glass rod oven-dried at 103 °C (± 2 °C) for 30 min. The sample and the sand were homogeneously mixed with the aid of the glass rod and oven drying (Biopar, S336AD) at 103 °C (± 2 °C) for 2 h. It was removed from the oven, transferred to a desiccator at room temperature and weighed. The drying, cooling and weighing operations were repeated from 1 h to 1 h until obtaining constant weight (Brazil, 2009). Results were expressed in % (g water content /100 g sample on wet base) and samples were analyzed in triplicate.

2.7 Protein determination

The nitrogen content was determined by the Kjeldahl method, according to methodology described in Normative Instruction nº 20/1999 (Brazil, 1999) and methodology nº 920.123 (Association of Official Analytical Chemists, 2005). The protein content in a sample was obtained by multiplying the nitrogen content by a nitrogen-to-protein conversion factor of 6.25. Results were expressed as % (g protein/100 g sample) and the samples were analyzed in triplicate.

2.8 W/P ratio

The W/P ratio was calculated by Equation 1 (Brazil, 2010), taking in to account the results obtained in water content and protein analysis.

$$W\ /\ P\ ratio\ =\ Water\ content\ (\%)/\ Protein\ (\%) \quad (1)$$

2.9 Statistical analysis

The results obtained were statistically treated by analysis of variance (ANOVA), followed by comparison of the means differences by the Tukey test or student test, with a 95% confidence level, with Past 4.02 software.

3 Results and discussion

3.1 Fasting time assessment

The influence of pre-slaughter fasting time on W/P ratio evaluated in each treatment is shown in Figure 1. It is possible to observe the variation of parameters with the increase of the fasting period (food and water) for the chicken breast cut (Figure 1a). Larger extrapolations were observed in the upper limit established by the legislation (Brazil, 2010), and the tendency of reduction of this parameter with the increase of fasting time.

In the results for the drumstick and thigh cut (Figure 1b) extrapolations were observed for the limits of 4% without fasting T1 (upper limit) and 2% for 18 h of fasting T4 (lower limit). It should be noted that the 18 h fasting treatment hurts the animal welfare, so it would be an inappropriate practice (Ludtke et al., 2010).

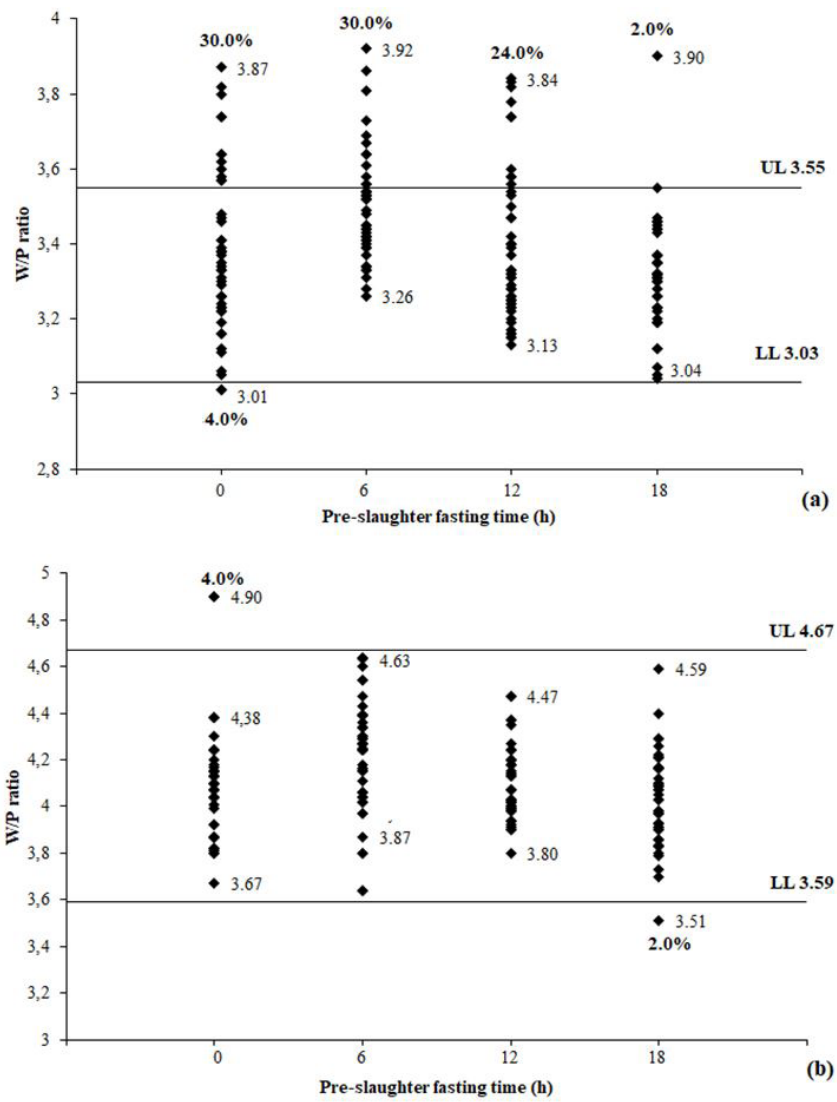

Figure 1. Results of W/P ratio for breast (a), drumstick and thigh (b) cuts in the fasting time. UL = upper limit; LL = Lower Limit (Brazil, 2010). Bold percentages refer to extrapolation of the predicted limits.

By statistical analysis of water content, protein and W/P ratio (Table 1), only the protein parameter in breast cut for treatment without fasting (T1) presented differene ($p<0.05$) in relation to the other fasting times. This fact reflects the physiological condition of the bird without interference from fasting.

Among the cuts, lower levels of water content and protein, and higher W/P ratio were observed in the breast when compared to drumstick and thigh. All results obtained differ from the standards established by Brazilian legislation, except for the breast cut, for water content parameter.

Kotula & Wang (1994) studied different fasting times (0, 3, 6, 12, 18, 24 and 36 h) and observed a decrease in water content in the chest and increased in the thigh with the increase in the fasting time. Even though, when the fasting time exceeded 6 h, there was a significant decrease in breast muscle tenderness. Similarly, in the present study, this behavior was identified in the breast samples, but not reproduced in the thigh.

Feed restriction causes physiological and immunity changes in chickens that should not be ignored. After the food and water withdrawal, dehydration process in the carcass occurs, resulting in live weight loss (Duke et al., 1997). This loss increases linearly with the fasting time. This dehydration influences the quality of chicken meat, as water retention is an important feature related to the appearance of the meat before cooking, its behavior during the cooking and the palatability of the product (Mendes, 2001).

In the evaluation of Ross chickens with 46 days of age, under different fasting times (3, 6, 9, 12, 15, and 18 h), Castro et al. (2008) observed statistical effect ($p<0.05$) on the weight loss in live chickens. However, no statistical differences ($p>0.05$) were observed for sensory characteristics of breast meat, also corroborating with the results of Pereira et al. (2013).

3.2 Process mapping

Figure 2 presents the results obtained in different process steps for water content, protein, and W/P ratio for boneless and skinless breast. After the pre-cooling stage, values of water absorption (8%) in the carcasses complied with the Brazilian legislation (Brazil, 1998).

In the protein content, extrapolations were verified at the low limit, with high percentage of samples below the limit established by the Brazilian legislation in the hanging and frozen stages.

In the water content extrapolations were observed in the upper limit in all process steps. The same occurred for the water content/protein (W/P) ratio with high extrapolation percentages in the hanging and frozen product. In chiller 1 low oscillations and percentage of extrapolations in the upper limit were verified against the others process steps.

Comparison across averages (water content, protein, and W/P ratio) in the different steps of the process are presented in Table 2. In immersion cooling, the European commission regulation (EC) nº 543/08 of June 16, 2008 sets maximum values of 3.40, 3.60 and 4.30 for water content/protein (W/P) ratio in chicken breasts with and without skin, thighs and drumsticks, respectively. Comparing the results with the European standards 93.3% of incompatibility with the combined limits was observed. Only steps 3 and 6 (pre-chiller and frozen product) showed significant difference ($p<0.05$) in the parameters evaluated. In Chiller 1 the differentiation can be due to several reasons, the main is the complexity of the cooling system. The raw material (chicken carcasses) does not have defined geometry, weight and size variability, has specific characteristics for males and females, variable chemical composition, among others (Carciofi, 2005). Carciofi (2005) and Lorenzetti et al. (2018) mention that water temperature and agitation are boundaries for the water cross and become part of the body (water absorbed by carcasses). Olivo & Olivo (2006) report that the carcass residence time in chillers causes great hydration of protein, resulting in high water absorption. According to the legislation, the permanence time of chicken carcasses in the pre-cooling phase should not exceed 30 min (Brazil, 1998).

Table 1. Results of water content, protein and W/P ratio in different fasting times (T1, T2, T3 and T4).

Cut of meat	Treatments (fasting times)			
	T1 (0 h)	T2 (6 h)	T3 (12 h)	T4 (18 h)
	Water content (%)			
Drumstick and thigh (62.82-70.70)*	75.12aA ± 1.18	75.04aA ± 1.34	74.88aA ± 1.28	74.25aA ± 0.77
Breast (67.16-75.40)*	69.86aB ± 1.38	69.89aB ± 1.74	69.59aB ± 1.79	69.49aB ± 2.05
	Protein (%)			
Drumstick and thigh (14.36-18.08)*	22.05aA ± 1.53	21.28aA ± 1.14	21.94aA ± 1.47	22.55aA ± 0.76
Breast (17.81-22.05)*	14.04bB ± 0.66	16.5aB ± 0.83	17.02aB ± 0.63	17.30aB ±0.93
	W/P ratio			
Drumstick and thigh (3.59-4.67)*	3.43aA ± 0.30	3.55aB ± 0.26	3.42aB ± 0.28	3.32aB ± 0.14
Breast (3.28-3.92)*	4.09aA ± 0.24	4.22aA ± 0.23	4.09aA ± 0.15	4.02aA ± 0.19

Means ± standard deviation followed by the same lowercase letter in the line and uppercase in columns, did not differ by Tukey's and Student test ($p<0.05$). *Current legislative limits of the water content and protein in chicken cuts, established by Technical Regulation nº 32 (Brazil, 2010).

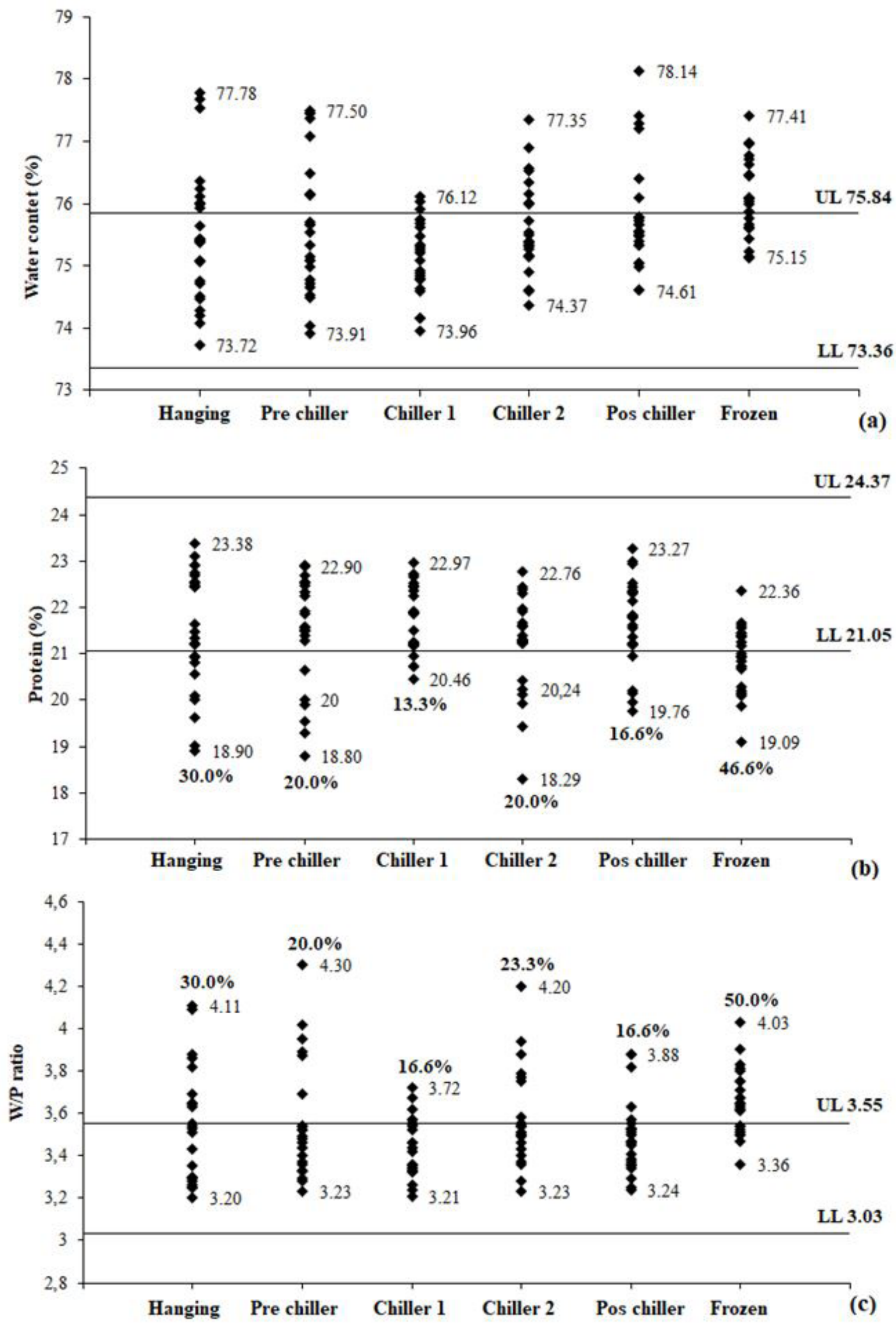

Figure 2. Results of water content (a), protein (b), and W/P ratio (c) for the boneless and skinless breast in different steps of the process. UL = upper limit; LL = Lower Limit (Brazil, 2010). Bold percentages refer to extrapolation of the predicted limits.

Table 2. Values of water content, protein, and W/P ratio in the different steps of the process.

Step	Water content (%) (73.36-75.84)*	Protein (%) (21.05-24.37)*	W/P ratio (3.03-3.55)*
1 - Reception / Hanging	75.49ab ± 0.23	21.48ab ± 0.27	3.53ab ± 0.56
2 - Pre-chiller	75.62ab ± 0.23	21.43ab ± 0.30	3.55ab ± 0.27
3 - Chiller 1	75.15b ± 0.12	21.76b ± 0.15	3.46b ± 0.28
4 - Chiller 2	75.61ab ± 015	21.19ab ± 0.21	3.57ab ± 0.45
5 - Pos Chiller	75.81ab ± 0.18	21.63ab ± 0.96	3.49ab ± 0.35
6 - Frozen Product	75.63a ± 0.12	20.87a ± 0.72	3.65a ± 0.36

Means ± standard deviation followed by the same lowercase letter in the columns, did not differ by Tukey's test ($p < 0.05$). *Current legislative limits of the water content and protein in chicken cuts, established by Technical Regulation nº 32 (Brazil, 2010).

Lorenzetti et al. (2018) identified most pronounced structural changes in the pre-chiller using histological analyzes in chicken breast. However, in the present study, in chiller 1, structural changes continue to occur, although with less intensity.

At the beginning of the cooling process, the carcass presents temperature around 25°C, the pectoral muscle, composed of fibers and inter-fiber space, contains low amount of liquid, which provides high rates of water sorption. This rate strongly depends on the hydrostatic pressure acting on the carcasses, which suggests that the water inlet is controlled by a hydrodynamic mechanism. The filling of the inter-fiber voids of this muscle tissue (pores) and the saturation of the region near the carcass surface causes a decrease in water absorption, which depends mainly on the internal migration of the absorbed water (Carciofi, 2005).

Ferrari et al. (2016), while evaluating the same parameters in carcasses before and after the cooling system, found that the water content obtained before cooling (18.5%) increased after cooling (31.0%), and W/P ratio also increased from 20.4% (before cooling) to 24.8% (after cooling).

Offer & Cousins (1992) point to the existence of two types of intracellular spaces in the post mortem muscle. The spaces between the fibers and perimysium, and fibers and endomysium can be considered as open capillaries at both ends. These capillaries present one of the preferred entrances to the liquids absorption in the muscle during the cooling process in the immersed form.

Another step that showed significant difference was in the frozen product. Slow freezing or storage at improper temperatures can cause ice crystals, favoring the appearance of concentrated salt solutions and other sarcoplasm components (Abreu & Abreu, 2002). Thus, protein denaturation with loss of water retention can be experienced. Defrosting the meat causes considerable loss of fluid and nutrient substances, along with a compaction of protein fibers that will produce a stiffer and drier product.

3.3 Comparison of different cuts

In the cuts, was assessment was made only in the hanging, pre-chiller and frozen product stages that presented significant differentiation. Thus, the continuity of the investigation was performed in the thigh, drumstick, leg, breast and half breast. After the pre-cooling stage, the values of water absorption in the carcasses were followed, with maximum results of 8%, respecting the Brazilian legislation limits (Brazil, 1998). The results of the analysis of the different cuts between the process steps are presented in Table 3. A significant difference ($p < 0.05$) was observed in the thigh and drumstick for water content in the pre-chiller and frozen stages. These results go beyond the limits of the Brazilian legislation. Comparing the data with European legislation, the W/P ratio with standard of 4.30 for the leg group and 3.60 for chicken breast with skin group (thigh, drumstick and leg) there is only 35.56% of compliance, being 20% thigh, 50% drumstick and 36.67% leg. The breast in average met the standard, but the incompatibility was 26.67%.

The breast and thigh have different metabolism, resulting in differences in final pH values, color, meat texture and total lipid content inside the muscles (Cruz et al., 2017). In general, chicken breast meat has low-fat content, mainly due to the natural characteristic of the anatomical region, since it does not require a large amount of energy reserve storage. However, the abdominal cavity and thighs are characterized by being energy reserve regions, were fat levels are much more pronounced, serving as thermal insulator and allowing long-term physical activities.

Some authors reinforce this approach; Dias et al. (2016) evaluated chickens with average weight between 2.4 and 3.2 kg and age of 42 and 50 days, finding for breast and half breast 21.10% protein,72.33% water content, and 3 W/P ratio. For skinless breast 73.90% water content, 22.6% protein and 3.28 W/P ratio, and for thigh cuts 67.76% water content, 16.96% protein and 4.00 W/P ratio. Elahi & Topping (2012) performed

Table 3. Results of water content, protein and W/P ratio for different cuts in the hanging, pre-chiller and frozen product steps.

Cut of meat	Hanging	Pre-chiller	Frozen product
	Water content (%)		
Thigh (65.33-72.69)*	74.37a ± 0.98	73.59a ± 1.08	73.99a ± 2.67
Drumstick (61.09-70.97)*	74.37a ± 1.05	70.20b ± 1.30	70.82b ± 2.25
Leg (62.82-70.70)*	74.20a ± 1.13	70.96b ± 1.13	72.86b ± 1.77
Breast and half breast (67.16-75.40)*	75.38a ± 1.08	75.15a ± 1.07	75.67a ± 0.90
	Protein (%)		
Thigh (14.40-17.96)*	17.29a ± 1.93	16.97a ± 1.54	16.60a ± 1.14
Drumstick (13.50-18.18)*	17.27a ± 0.72	16.99a ± 058	16.47a ± 0.84
Leg (14.36-18.08)*	17.08a ± 0.75	17.35a ± 0.75	16.55a ± 0.86
Breast and half breast (17.81-22.05)*	21.56a ± 1.27	21.57a ± 1.13	21.37a ± 1.07
	W/P ratio		
Thigh (3.83-4.71)*	4.32a ± 0.16	4.29a ± 0.38	4.49a ± 0.32
Drumstick (3.64-4.72)*	4.31a ± 0.16	4.14a ± 0.16	4.26a ± 0.16
Leg (3.59-4.67)*	4.34a ± 0.18	4.09a ± 0.18	4.35a ± 0.26
Breast and half breast (3.28-3.92)*	3.43a ± 0.26	3.45a ± 0.26	3.52a ± 0.21

Means ± standard deviation followed by the same lowercase letter in the line and uppercase in columns, did not differ by Tukey's and Student test ($p < 0.05$). *Parameters for the evaluation of the total water and protein contained in chicken cuts, established by Technical Regulation nº 32 (Brazil, 2010).

Table 4. Results of water content, protein and W/P ratio in relation to the sex (male and female) and the lineage (Cobb and Ross) in the hanging, pre-chiller and frozen product steps.

Sex	Hanging	Pre-chiller	Frozen product
	Water content (%) (73.36-75.84)*		
Male	75.38^{aA} ± 1.08	75.15^{aA} ± 1.07	75.67^{aA} ± 0.90
Female	73.89^{aB} ± 0.65	73.65^{aB} ± 0.69	74.37^{aB} ± 0.68
	Protein (%) (21.05-24.37)*		
Male	21.56^{aB} ± 1.27	21.57^{aB} ± 1.13	21.37^{aB} ± 1.07
Female	23.30^{aA} ± 0.88	23.25^{aA} ± 0.83	22.62^{aA} ± 0.60
	W/P ratio (3.03-3.55)*		
Male	3.43^{aA} ± 0.26	3.45^{aA} ± 0.26	3.52^{aA} ± 0.21
Female	3.17^{aB} ± 0.13	3.17^{aB} ± 0.11	3.31^{aB} ± 0.10
Lineage	Water content (%) (73.36-75.84)*		
Cobb	75.38^{aA} ± 1.08	75.15^{aA} ± 1.07	75.67^{aA} ± 0.90
Ross	75.51^{aA} ± 0.87	74.51^{aB} ± 1.09	75.68^{aA} ± 1.34
	Protein (%) (21.05-24.37)*		
Cobb	21.56^{aA} ± 1.27	21.57^{aA} ± 1.13	21.37^{aA} ± 1.07
Ross	22.04^{aA} ± 1.02	21.63^{aA} ± 1.21	20.93^{aA} ± 1.06
	W/P ratio (3.03-3.55)*		
Cobb	3.43^{aA} ± 0.26	3.45^{aA} ± 0.26	3.52^{aA} ± 0.21
Ross	3.44^{aA} ± 0.22	3.42^{aA} ± 0.24	3.60^{aA} ± 0.21

Means ± standard deviation followed by the same lowercase letter in the line and uppercase in columns, did not differ by Tukey's and Student test ($p < 0.05$). *Parameters for the evaluation of the total water and protein contained in chicken cuts, established by Technical Regulation nº 32 (Brazil, 2010).

sampling in seven countries of the European Community and obtained 74.99% water content and 22.97% protein in skinless boneless chicken breasts with W/P ratio of 3.26 in these cuts. This study recommends new W/P ratio limits for the different pre-cooling systems as 3.55 in air chiller; 3.65 for air spray chilling and 3.75 for water immersion chiller. These suggested limits were above the maximum limits allowed by the European commission regulation (3.40) and Brazilian legislation (between 3.03 and 3.55) for the different pre-cooling systems. Considering this value, compared to the data obtained in the present study (Table 3), all samples meet the recommendations.

3.4 Evaluation of sex and lineage

Sex (females and males) was assessed in the Cobb lineage in different stages of the process using the skinless breast. The results of the process steps (hanging, pre-chiller and frozen) showed a significant difference ($p < 0.05$) between females and males in all evaluated parameters (Table 4). Males presented higher water content and W/P ratio, and lower protein compared to females. No significant difference ($p > 0.05$) was observed between the evaluated process steps.

A high percentage of male samples presented water content and W/P above the official limits before the pre-refrigeration step. Differences between sexes are related to superiority of males in body weight, weight gain and feed conversion (Api et al., 2017). Males consume more water than females, thus indicating that this difference occurs from day one (Marks & Washburn, 1983). Females reach sexual puberty earlier than males, so the growth adipose tissue occurs earlier, resulting in decrease of weight gain due to the great use of ingested nutrients being intended for adipose tissue growth, and not for muscle growth (Api et al., 2017), interfering with feed conversion. With larger amount of adipose tissue, the centesimal composition will present more lipids and consequently low percentage of water content.

In the Cobb and Ross lineage results obtained for males in the breast cut similar results were observed (Table 4). Water content showed significant difference ($p < 0.05$) in the pre-chiller stage. For protein and W/P ratio no significant differences were observed ($p > 0.05$) across process steps for both lineages. In the mean evaluation, only an extrapolation in relation to the legislation was observed for Ross lineage in the frozen product for the W/P ratio.

In the evaluation of four lineages, Ferrari et al. (2016) did not obtain significant differences across the samples collected before and after the pre-cooling system. Already, Marcato et al. (2010) observed in Cobb lineage with 7.33 days an early intestinal development compared to the Ross lineage, thus justifying the great weight gain, allowing this strain to be slaughtered at a younger age.

Petracci et al. (2013) studied the breast characteristics in two lineages (commercial –SBY, and high breast yield -HBY), considering 4.2 kg and 53-55 days of age. The chemical composition showed significant differences ($p \leq 0.05$) in protein (22.8 and 23.5%) and lipids (1.65 and 1.82%), being small in the HBY, while water content was low in the SBY. The author attributes the difference to the fact that HBY has a high incidence of striated breast fillets, which reduces the water retention capacity. It has shown that muscle microstructure degeneration is capable of determining a quantitative macroscopic reduction of proteins with corresponding increase in water.

Scheuermann et al. (2016) evaluated water content and protein parameters in breast samples of different lineages (Cobb, Ross and Hubbard), concluding that both exceed the limits of Brazilian legislation in 13.92% (1.38% down and 12.54% up) for water content and 10.05% (6.32% down and 3.73% up) for W/P ratio.

Developments in chicken genetics, improvements in field performance and changes in the natural environment can lead to changes in the chemical composition of chicken carcass. These changes make difficult to comply the legislation limits, even if strict quality control is followed in the slaughter process. This fact shows the need of revision of the legal limits, as possible genetic influences in the chicken muscle composition are not considered. It is noteworthy that griller cuts are also commercialized. According to data from the literature (Perreault & Leeson, 1992), there is an effect of age on the composition of some cuts. Therefore, younger chickens should be considered in future studies for inclusion in the legislation. This is also the case of turkey cuts, whose water content and protein levels differ from chicken cuts (Scheuermann et al., 2016), which is not covered by the Normative.

In addition, the W/P ratio may be even more important than the individual assessment of water content and protein, a fact that is already applied in the European Union, and could be replicated to Brazilian legislation. As to the form of sample collection, the probability of extrapolations is high, indicating compliance of 76.69% (Brazil, 2018). Scheuermann et al. (2016) suggests support to the revision of the legislation in order to provide details of the sampling, as it directly affects the outcome. The data obtained in this study strengthen this signaling. While assessing the results, a high percentage of incompatibility for the established parameters was identified.

4 Conclusion

According to the results, the chickens arrive on the slaughter with water content higher than that established by the Brazilian legislation. In the fasting time and lineages evaluated (Cobb and Ross) no significant differences were found. In the process steps, significant differences were found in the pre-chiller and frozen product. The cuts (thigh and drumstick) presented significant differences for water content and W/P ratio in the pre-chiller and frozen product. In addition, significant differences were found between males and females, where females have a high tendency to meet the determined parameters. In accordance with European legislation there were also incompatibilities for W/P ratio, being 26.67% for skinless breast, 6.67% for breast and half chest, and 64.44% for leg group, considering only male Cobb. These results have shown that the parameters provided by legislation do not follow the genetic of chickens. Consequently, more studies are required to update the current legislation values, so that the external interference, whether from field improvements or methodology analysis do not result in mistakenly signal violations by companies.

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