STUDY ON WATERFOWL MEAT TEXTURE - A COMPARISON BETWEEN THE HISTOLOGICAL AND RHEOLOGICAL METHODS

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Abstract

Among the most used methods in investigating meat structure and texture and its estimated incidence on the sensorial ultimate quality felt by the consumer there is the shear force instrumental measurement using a device with adjustable force and a measurement chamber endorsed with a V gap blade, also known as Warner Bratzler tool. This instrument picks data during sample shearing, in terms of forces necessary to cut the myofibrilar (M force) compound and also the connective compound (C force) of the meat, reported to the elapsed time or distance till total shearing of the sample by the blade. We found out it would be great to compare this instrumental method of muscle toughness estimation with a method belonging to fundamental sciences, using histological and histometrical techniques on the same muscles. The biological material consisted in 20 pieces of breast meat, commercially available as refrigerated duck and goose meat in supermarket (10 samples of duck breast and 10 pieces of goose breast). Out of these material, there were shaped 10 rectangular samples from each breast (10 x 10 = 100 samples per waterfowl species, sized 4 cm length x 1 cm width x 1 cm depth) which were submitted to shear force testing using an Instron type texture analyzer. From the same breast pieces, there were cut 0.5 cm x 0.5 cm x 1 cm samples (5 samples per breast), using the same pattern of muscle fibers orientation like in the previously harvested samples, in order to submit them to paraffin inclusion, microtome cross-section and HEMB coloring, in order to prepare and examine smears using a Motic M230 microscope. Myocytes diameters, cross-section areas were measured and calculated, then reporting to the amount of muscle cell within a 1st order muscle fascicle and to its outer circumference and overall surface, the proportion of pure muscle and connective tissues were derived. The proportions found were compared to those derived from shear forces values readings, knowing the C force instrumentally measured is approx. 0.75-0.9 positively correlated with the connective tissue in muscle. The differences between the instrumental shear force detection of connective tissue and the histological method were quite low and not significantly different. Therefore, it could be stated that the quick instrumental rheological method depicts with accuracy the tissual structure of the muscles, at least at the experimental level we approached.

Key words: duck, goose, meat, shear force, texture, histometry, connective, muscular tissue

INTRODUCTION

Among the sensory traits well appreciated by consumers when eating meat, the tenderness and juiciness are the most relevant [12]. Poultry meat texture is influenced by species [12], age and muscle growth [7], farming system [2], peri-slaughter conditions [5], stunning type and parameters [10], carcass maturation conditions [4], cooking manner [8] and, of course, the method used in its analysis (pure morphological – histological, shear force instrumental or sensorial) [1, 17, 20, 21].

It could be believed that the histological method is more accurate, however, some biases of cross-section and cell morphology could occur during paraffin including technique of microscopic slides preparation. On the other hand, the rheological method is much quicker but the Instron type equipment should be available and be well calibrated to read the actual force values necessary to shear the meat

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sample. Within this context, our goal was to comparatively investigate the connective and myofibrilar compounds participations in waterfowl breast muscles, as components of texture profile analysis and to find out whether there are or not significant differences between the results achieved via morpho-histological analysis and the shear force instrumental analysis.

MATERIAL AND METHOD

Biological material:

- refrigerated breast pieces commercially available on the market, packaged in modified atmosphere (10 pieces duck breast, 10 pieces goose breast);
- 10 samples from each breast (100 samples), cut in 4x1x1 cm rectangles for shear force testing [18];
- 5 samples from each breast (50 samples), cut in 1x0.5x0.5 cm rectangles for paraffin inclusion, microtome cross section and HEMB staining [13].

Methods of examination:

- Shear force assessment, using a Perten Instruments TVT 7600 analyser (100 tests/species) [21];
- Histometry (50 samples x double reading at magnifications of 100 x and 400 x = 100 results, followed by the calculation of tissual proportion in meat, using a Motic M230 and imagistic software (100 tests/species) [15];

Data analysis: Graphpad Prism 8.0 for Windows software was used, in accordance with the appropriate methodology for animal science experiments [16], to calculate descriptive statistics, percentage deviations, ANOVA significance (P values) for outputs.

Starting from peak 2 shear force readings, it was assumed that the numerical value of connective tissue proportion represents 0.8 of these force values achieved for the connective fibrilar compound, after the comparison of many findings related to these traits correlations (0.75-0.92) in comparable studies [3, 6, 9, 11, 14].

Limitations: few amount of carcass parts as biological material and its provenience. It is recommended to use at least 100 carcass parts from each species to sample meat and to obtain the parts from fowl raised in an own experimental facility and slaughterhouse, in order to eliminate any biases due to uncontrolled environmental and technological factors; also, the assumption that connective tissue proportion represents 0.8 of the force value given necessary to share the connective fibrilar compound could vary within larger limits, in relation with many factors (meat maturation, freshness status, cooking status etc.).

RESULTS AND DISCUSSIONS

Data on the histological measurements of duck breast muscles are presented in Table 1. The average thickness was calculated at 32.69±0.11µ with variation limits of 30.50-35.15 µ, while the variation coefficient indicated good homogeneity (v=3.81%). These values lead to cross section areas of 816.96 μ^2 (calculated value), and to 818.95 μ^2 (measured value), respectively. The values for the sum of all myocytes comprised within a 1st order muscle fascicle and the 1st order muscle fascicles cross-section areas were used to assess the proportion of pure muscular tissue versus connective tissue, starting from myocytes values. The data are presented in Table 2 and Fig. 1. The myofibrilar compound (muscular tissue) varied between 75.19-85.87%, with an average value of 78.96±0.19% (based on myocytes calculated size) and between 70.44-87.98%, with an average of 78.77±0.24% (based on myocytes measured size). The calculated values were more homogenous than the measured ones, indicating a larger morphological variety of muscle cells shapes within the histological smear. However, the difference between measurement and calculation is quite not relevant (0.2%). In terms of connective tissue, the data varied reversely proportional: 21.04±0.19% (calculated) and 21.23±0.24% (based on measured values).

In terms of comparisons with the rheological method, we decide to keep the tissue structure values issued from measurement as referential. We recorded both peaks of forces needed to shear the rectangular meat samples using the texturometer. Thus, peak 1 (myofibrilar compound) varied between 18.20-19.50 N/cm^2, while peak 2 (connective fibrilar compound) varied between 25.10-28.20 N/cm^2, resulting an average of 26.92±0.10 N/cm^2.

Table 1 – Duck breast – myocytes width and cross-section area

Descriptive stat.	Large diameter (μ)	Small diameter (μ)	Average diameter (μ)	Cross section area (calculated) (μ^2)	Cross section area (measured) (μ^2)
Mean	37.98	27.39	32.69	816.96	818.95
±Std. error	±0.14	±0.13	±0.11	±0.79	±0.56
±Std. deviation	±2.00	±1.59	±1.26	±62.40	±30.84
Min.	35.80	25.20	30.50	708.56	765.55
Max.	42.40	29.80	35.15	929.10	875.24
CV %	5.26	5.81	3.86	7.64	3.77

Table 2 – Duck breast – proportion of main tissue categories in meat, assessed through histological and rheological methods

Descriptive stat.	HISTOLOGICAL				RHEOLOGICAL		
	Myofibrilar compound (%)		Connective compound (%)		Peak 1 & Peak 2 forces (Myofibrilar vs. Connective (N/cm^2)		Connective tissue estimated on Shear Force (%)
	calc.	meas.	calc.	meas.	myofibr.	connect.	
Mean	78.96	78.77	21.04	21.23	18.92	26.92	21.54
±Std. error	0.19	0.24	0.19	0.24	0.06	0.10	0.09
±Std. deviation	3.68	5.79	3.68	5.79	0.39	1.01	0.81
Min.	75.19	70.44	14.13	12.02	18.20	25.10	20.08
Max.	85.87	87.98	24.81	29.56	19.50	28.20	22.56
CV %	4.66	7.35	17.48	27.27	2.04	3.74	3.74

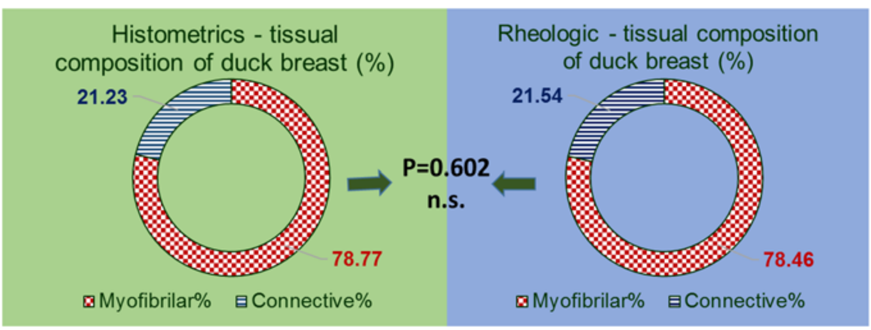

Fig. 1 – Textural compounds of duck breast meat
(a comparison between the histological and rheological methods)

The connective tissue proportion, estimated on the basis of Shear Force value was 21.54±0.09%. By difference from 100% (whole sample), it resulted a proportion of muscular tissue of 78.46% (Fig. 1). The differences between the means obtained through histological measurements and rheological measurements (21.23% vs. 21.54% for connective tissue; 78.77% vs. 78.46% for myocytes proportion) were not significant (P=0.602 > 0.05), thus it could be stated that both used methods provide comparable results.

Histometrical results issued from goose breast sample analysis are presented in Table 3. The average diameter of the myocytes was 40.46±0.12μ, which led to a cross-section area calculated value of 1249.56±0.92μ^2, close to that found through histological measuring (1254.38±0.85μ^2), in both situations, the results were quite homogenous (v=5.73-6.71%).

Table 3 – Goose breast – myocytes width and cross-section area

Descriptive stat.	Large diameter (μ)	Small diameter (μ)	Average diameter (μ)	Cross section area (calculated) (μ^2)	Cross section area (measured) (μ^2)
Mean	47.25	33.66	40.46	1249.56	1254.38
±Std. error	0.15	0.12	0.12	0.92	0.85
±Std. deviation	2.20	1.32	1.39	83.87	71.85
Min.	44.20	31.90	38.55	1136.51	1156.50
Max.	51.40	35.10	43.15	1408.90	1393.75
CV %	4.66	3.93	3.44	6.71	5.73

Table 4 – Goose breast – proportion of main tissue categories in meat, assessed through histological and rheological methods

Descriptive stat.	HISTOLOGICAL				RHEOLOGICAL		
	Myofibrilar compound (%)		Connective compound (%)		Peak 1 & Peak 2 forces (Myofibrilar vs. Connective (N/cm^2)		Connective tissue estimated on Shear Force (%)
	calc.	meas.	calc.	meas.	myofibr.	connect.	
Mean	75.74	75.41	24.26	24.59	24.67	31.23	24.98
±Std. error	0.14	0.14	0.14	0.14	0.11	0.10	0.09
±Std. deviation	2.10	2.07	2.10	2.07	1.13	1.02	0.82
Min.	72.00	71.32	21.65	23.18	23.10	29.80	23.84
Max.	78.35	76.82	28.00	28.68	27.30	33.50	26.80
CV %	2.77	2.75	8.66	8.42	4.57	3.28	3.28

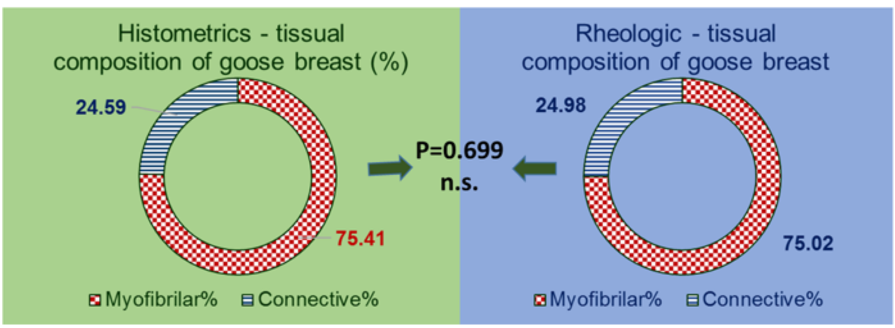

Fig. 2– Textural compounds of goose breast meat
(a comparison between the histological and rheological methods)

In goose meat, the calculus revealed proportion of 75.74±0.14% myofibrilar compounds and 24.26±0.14% connective compounds, while the proportions resulted from measurements reached 75.41% muscular tissue and 24.59% connective tissue, the latter one being considered, as well, of reference when the comparison was made with the rheological findings. As issued from texturometer analysis, peak 1 (myofibrilar compound) varied between 23.10-27.30 N/cm^2, while peak 2 (connective fibrilar compound) varied between 29.80-33.50 N/cm^2, resulting an average of 31.23±0.10 N/cm^2. Therefore, the connective tissue proportion, also estimated on the basis of Shear Force value, reached 24.98±0.09%. The calculated proportion of muscular tissue in goose breast meat, calculated on the basis of shear force reached 75.02% (Fig. 1). ANOVA testing revealed that the differences between the means obtained through histological and rheological measurements (24.59% vs. 24.98% for connective tissue; 75.41% vs. 75.02% for myocytes proportion) were not significant ($P=0.699 > 0.05$), thus it could also be stated that both used methods provide comparable results.

CONCLUSIONS

It was surprisingly interesting to find out that the differences between the instrumental shear force detection of connective tissue and the histological method were quite low, less than 1% and not significantly different.

The quick instrumental rheological method depicts with accuracy the tissual structure of the muscles, at least at the experimental level we approached.

Further tests are required to establish the real correlation between the methods, for the most accurate confidence interval. Introduction in the protocol of a quick method to assess the collagen content in meat (such as NIRS) could provide data useful to correlate with histometry and shear force readings.

REFERENCES

[1] Aguirre M.E., Owens C.M., Miller R.K., Alvarado C.Z. Descriptive sensory and instrumental texture profile analysis of woody breast in marinated chicken. *Poultry science*, 2018, *97*(4):1456-1461.

[2] Albrecht A., Hebel M., Mittler M., Hurck C., Kustwan K., Heitkönig B., Kreyenschmidt J. Influence of Different Production Systems on the Quality and Shelf Life of Poultry Meat: A Case Study in the German Sector. *Journal of Food Quality*, *2019*.

[3] An J.Y., Zheng J.X., Li J.Y., Zeng D., Qu L.J., Xu G.Y., Yang N. Effect of myofiber characteristics and thickness of perimysium and endomysium on meat tenderness of chickens. *Poultry Science*, 2010, *89*(8):1750-1754.

[4] Benedini R., Parolari G., Toscani T., Virgili R. Sensory and texture properties of Italian typical dry-cured hams as related to maturation time and salt content. *Meat Science*, 2012, *90*(2):431-437.

[5] Hasan S. The Effect of Poultry Preslaughter Fasting and Condition on the Quality of Meat and Luncheon Processed in Syria. *Bioscience Methods*, 2012, *3*.

[6] Hopkins D.L., Allingham P.G., Colgrave M., Van de Ven R.J. Interrelationship between measures of collagen, compression, shear force and tenderness. *Meat Science*, 2013, *95*(2):219-223.

[7] Ismail I., Joo S. T. Poultry meat quality in relation to muscle growth and muscle fiber characteristics. *Korean journal for food science of animal resources*, 2017, *37*(6):873.

[8] Ježek F., Kameník J., Macharáčková B., Bogdanovičová K., Bednář J. Cooking of meat: effect on texture, cooking loss and microbiological quality–a review. *Acta Veterinaria Brno*, 2020, *88*(4):487-496.

[9] Liu A., Nishimura T., Takahashi K. Relationship between structural properties of intramuscular connective tissue and toughness of various chicken skeletal muscles. *Meat science*, 1996, *43*(1):43-49.

[10] Morey A., Owens C.M. Methods for measuring meat texture. In *Poultry quality evaluation* (pp. 115-132). Woodhead Publishing, 2017.

[11] Nishimura T. The role of intramuscular connective tissue in meat texture. *Animal science journal*, 2010, 81.1: 21-27.

[12] Owens C.M., Meullenet J.F.C. Poultry meat tenderness. *Handbook of Poultry Science and Technology*, 2010, *1*:491-514.

[13] Pappas G.S. Laboratory manual of histology. USA, McGraw-Hill Higher Education, 1994.

[14] Purslow P.P. Intramuscular connective tissue and its role in meat quality. *Meat science*, 2005, *70*(3):435-447.

[15] Radu-Rusu R.M., Teuşan V., Teuşan A. Comparative researches concerning some histometric features of the miocytes in somatic musculature of the domestic chicken and waterfowl (I). Pectoral muscles. *Lucrări Ştiinţifice, Seria Zootehnie, 2007,* 50:115-120.

[16] Rossi F., Mirtchev, V. Statistics for Food Scientists. Making Sense of the Numbers. USA, Elsevier, Academic Press, 2016.

[17] Soglia F., Mudalal S., Babini E., Di Nunzio M., Mazzoni M., Sirri F., Petracci M. Histology, composition, and quality traits of chicken Pectoralis major muscle affected by wooden breast abnormality. *Poultry Science*, 2016, *95*(3):651-659.

[18] Steffe J. F. Rheological Methods in Food Process Engineering, USA, Freeman Press, 1996.

[19] Wójcik W., Łukasiewicz M. Nutritional value variability of different poultry species meat in the organic production system. *Animal Science*, 2017, *56*:323-336.

[20] Zhuang H., Savage E. M. Comparisons of sensory descriptive flavor and texture profiles of cooked broiler breast fillets categorized by raw meat color lightness values1. *Poultry science*, 2010, *89*(5):1049-1055.

[21] Zhuang H., Savage E.M. Variation and Pearson correlation coefficients of Warner-Bratzler shear force measurements within broiler breast fillets. *Poultry science*, 2009, 88(1):214-220.