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Research Article
A Comparison of the Carcass and Meat Quality of ISA (F15) Spent Hens Slaughtered at Two Different Ages

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Abstract

Objective: A total of 16 spent hens (ISA F15) were slaughtered at about 220 and 337 days of age and studied for their muscle characteristics and sensory attributes. Methodology: Tenderness was estimated from breast muscle using penetrometer PNR 10 and sensory trained panel. Water Holding Capacity (WHC), percentage of released water, cooking loss, pH and Myofibril Fragmentation Index (MFI) were studied. Results: Penetration values were found negatively correlated with tenderness and MFI. Tenderness scores were negatively correlated with animal age and positively with pH. Overall acceptability of the meat of spent hens was judged higher irrespective of age. Multiple regressions analysis revealed that penetration depth was mainly explained by juiciness and MFI. Conclusion: In addition, the results revealed that sensory parameters representing textural properties of meat (tenderness, cohesiveness, chewiness and residues) were mainly explained by cooking losses.

Key words: Spent hens, poultry meat, penetrometer, sensory analysis, age

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.
INTRODUCTION

In Algeria, there is an intense expansion of eggs production leading to abundant availability of spent hens. Also, since the beginning of the 1980’s, the Algerian ministry of agricultural oriented consumer’s meat consumption to white meat as an alternative to beef and lamb for numerous economical and health reasons. Poultry is well known to contain low cholesterol and fat with very high omega-3 fatty acids contents\(^2\). In addition, consumers acknowledge the relatively low price, the typically convenient portions and the lack of religious restriction against its consumption\(^3\). In Algeria, spent hens may after the end of their economic laying cycle contribute significantly to the domestic consumption of meat. Nevertheless, much remain to be done to increase the value of spent hens meat in Algeria, which is perceived as very tough by consumers. Moreover, there is little knowledge on muscle characteristics and meat quality of ISA (F15) spent hens, the largest produced poultry in the country.

Technological and sensory traits like pH, water-holding capacity, cooking loss and tenderness of meat are very important and are related directly to the quality of the protein constituents of the meat product\(^4\). Therefore, the aims of this study were first to obtain the information on chemical and physical characteristics of ISA (F15) spent hens meat produced in Algeria. This is for us an open research window to increase our knowledge on spent hen ISA (F15) muscle to propose alternatives for the Algerian poultry industries. Secondly, age effect at time of slaughter on the sensory attributes, mainly tenderness of hens breast muscle has been investigated. In addition the potential use of penetrometer to predict meat tenderness scores of spent hens assessed by trained panellists was investigated. This approach may allow us in the future to choose the final destination of the spent hens carcasses toward direct consumption or for industrial transformation.

MATERIALS AND METHODS

**Animals and sampling:** Two groups of 16 spent aged hens (ISA F15) obtained from henhouse of SARL Nutri Aliment Plus “NAP” Constantine (Algeria) were slaughtered at about 337 (group 1) and 220 (group 2) days of age. They were slaughtered in compliance to Halal slaughter method in accordance with the Islamic legislation, exactly as described by Ibrahim et al\(^5\). Briefly, the Halal slaughter involves subjecting the birds to a transverse neck cut by using a sharp knife severing the esophagus, trachea, jugular veins and carotid arteries and also by starting the process of slaughtering by reciting Allah’s name and Tasmiyah (Bismillah Allahu Akbar). Carcasses were then chilled at 4°C for 14 h, before sampling of the superficial pectoral muscles (breast muscles) for (1) Textural, (2) Sensory analysis and (3) Biochemical and technological traits measurements.

**Texture measurements by penetrometry:** For instrumental texture, the petrotest PNR 10 penetrometer was used according to the protocol developed by our group to assess the texture of meat samples\(^6\). Briefly, 10 parallel epipedic samples (4 × 2 × 1.5 cm) were dissected from m. superficial pectoral of each animal. The instrument device is provided with a discerning body (2.5 g), which penetrates in free fall (perpendicularly to the muscular fibers) the sample under its own weight, during approximately 5 sec. The depth of penetration was measured in mm and expressed as unit of penetration (UP), where 1 UP = 0.1 mm. From each muscle sample, 10 pieces were taken and measured. The measurements were repeated 5 times for each piece of muscle; therefore 50 measures of penetration depth are obtained by sample and expressed as one mean.

**Sensory analysis:** After textural measurements, the same samples previously used for penetration test were cooked in a water bath until reaching an internal end-point temperature of 75°C\(^7\). Immediately after cooking, the samples were served to the trained panellists (10 persons) to be evaluated on an unstructured scale as reported by Gagaoua et al\(^8\). The panelists scored tenderness (how much force is required to break the sample as you press down with your back molars?), juiciness (how much juice is expressed from the sample over the entire chewing process?), cohesiveness (how does the sample break on the third chew?), chewiness (how many chews does it take to prepare the sample for swallowing?), presence of residues (quantity of residues remaining in the mouth) and overall liking on a 0-10 unstructured scale in which a score of 0, on ascending scale of quality of each attribute was equivalent to tough, not juicy, not cohesive (falls apart), not chewy, nothing and don’t like and score of 10 equivalent to tender, extremely juicy, very cohesive (stays together), very chewy, extreme and like too much. The sessions were carried out in a sensory analysis room equipped with individual boxes under artificial non-colored lighting. Panelists were seated in individual booths with a drinking cup containing water (90%) and apple juice (10%) to cleanse the palate between tastes\(^9\). Each panelist tasted each of 16 samples of meat in 4 sessions (4 samples per session) conducted mid-morning.
**pH measurement:** About 1 g of m. superficial pectoral from each animal was removed in triplicate and placed in glass jar containing 10 mL of a solution with 5 mM iodoacetate and 150 mM potassium chloride adjusted to pH 7.0. Samples were homogenized for two 15 sec bursts, with a 5 sec interval, using a Polytron homogenizer\(^{10,11}\). For pH measurements, an insertion electrode (PHS-3CW microprocessor pH/mV meter, BANTE instrument) was used to measure the pH of the solution in triplicate.

**Water holding capacity and drip losses determinations:** Water holding capacity was evaluated according to the Grau-Hamm method\(^{12}\) with modifications concerning the areas determination by image analysis. Briefly, a sample of 300±5 mg of meat from the m. superficial pectoral was weighed (G) and deposited on a previously dessicated and weighed (P) 11 cm filter paper disk (Whatman No. 1). After that, the sample on the paper was placed between two Plexiglass plates and a weight of 2.25 kg was applied. After 5 min, the areas of meat spot (M) and released juice (T) were drawn on clear plastic and the damp paper filter was weighed (D) after removing the compressed meat sample. The mean of three replicates was used. Water Holding Capacity (WHC) was calculated as \(M \times 100/T\) of the areas. The percentage of released water (PRW) was calculated as \((D-P) \times 100/G\). Traditionally, the areas of meat spot (M) and released juice (T) were measured using a planimeter\(^{12}\), but the method is not very precise and instrument dependent. As an alternative, in this study a simple technique was developed using image analysis. Hence, the areas drawn in clear plastic were first scanned and measured using the open source ImageJ 1.48 software developed by ou groups and as described recently by Hafid et al\(^{13}\) and for their quantification the free hand selection option was used.

**Cooking losses:** For cooking loss, 10 g of each weighed \((P_i)\) muscle (in triplicates) were vacuum packed and frozen at \(-20^\circ C\) until use. When required, the samples were thawed at \(4^\circ C\) for 24 h and cooked in plastic bags at \(80^\circ C\) for 1 h by immersion in a water bath. After cooling, samples were removed from the bags and weighed \((P_j)\). Cooking losses were calculated as \((P_i-P_j) \times 100/P_i\) according to the protocol of Pascual and Pla\(^{14}\).

**Myofibril fragmentation index measurement:** Myofibril Fragmentation Index (MFI) was determined using slight modification of the procedure of Li et al\(^{15}\). Briefly, 2 g of m. superficial pectoral of each animal were homogenized, using a polytron homogenizer, for 30 sec in 10 mL of rigor buffer (RB) (pH 7.0) containing 75 mM KCl, 10 mM KH\(_2\)PO\(_4\), 2 mM MgCl\(_2\), 2 mM EGTA and 1 mM NaCN. After centrifugation at 2000 rpm for 15 min at \(4^\circ C\), the supernatant was discarded. The pellet was re-suspended in 10 mL of the RB with stirring, centrifuged again and the supernatant was discarded. The sediment was re-suspended in 10 mL RB and filtered through a filter paper to remove connective tissue and fat. An additional 10 mL of the RB was used to facilitate the passage of myofibrils through the filter paper. The filtrate has been kept in adequate tubes at \(-20^\circ C\). The protein concentration of the suspension was determined using the Bradford protocol\(^{16}\). Then, the suspension was diluted with the same buffer to 0.5±0.05 mg mL\(^{-1}\) protein concentration. Finally, the MFI is the value of absorbance of the myofibrillar suspension, measured at 540 nm multiplied by 200.

**Statistical analysis:** Data were analyzed using XIStat software (Version 2009.1.01, Addinsoft\(^*\)). Effect of age (group of hens) on studied variables was assessed through analysis of variance and Tukey test was used to compare L5 means (p<0.05). Correlation analyses were conducted to study the relationships between sensory attributes, instrumental measurements and biochemical and technological traits. For that purpose, Z-scores were calculated and used to perform correlation analyses\(^{17}\). In addition, Principal Component Analysis (PCA) was conduct to visualize the distribution of the studied traits according to the group effect. For predictive purposes, multiple regression models were performed to explain sensory or instrumental attributes. The maximal number of explanatory variables was fixed at 3. Variables that were significant but contributed less than 2% in terms of explanatory power \(R^2\) were excluded from the model. The standardised coefficients are reported in the model equations and refer to how many standard deviations the explained variable will change, per standard deviation change in the explanatory variable.

**RESULTS**

Means and standard deviations of the mechanical, biochemical and palatability traits of m. superficial pectoral of the two hens groups are given in Table 1. The two groups were highly different in their age but without difference in their body weight. They were very different in terms of pH, percentage of released water (PRW), Cooking Loss (CL), tenderness and residues scores. The group 1 (old spent hens) was characterized by significant lower pH, PRW%, CL% and tenderness scores. Residues scores were significantly different on the...
and are as expected the highest (1.4 fold) within the aged hens (group 1). It can be speculated that the oldest the spent hens are, the lowest are pH, PRW, CL and tenderness and the highest are residuals scores. Otherwise, there is no significant difference in penetration depth between the two ages. The other textural sensory attributes, likely juiciness, cohesiveness, chewiness were not different (p>0.05).

The correlation coefficients between the studied traits are shown in Table 2. Results revealed that penetration depth was negatively correlated with tenderness scores and myofibril fragmentation index (r = -0.54 and -0.59, respectively, p<0.05) and positively correlated with juiciness (r = 0.58, p<0.05). Tenderness was negatively (r = -0.51, p<0.05) and positively (r = 0.50, p<0.05) correlated with cohesiveness and overall liking, respectively. Besides, cohesiveness was negatively (r = -0.58, p<0.05) related to overall liking.

To visualize relationships between the studied variables, Principal Component (PC) analysis was used and the results are given in Fig. 1. The two first PCs explained 47% of the variability (Fig. 1a). Two distinctive groups according to slaughter age are obtained and presented in the bi-plot of Fig. 1b. The young spent hens are grouped in the right side and the aged ones are in the left side. The first PC explaining 32% of variability was mainly characterized by PRW, tenderness, overall liking and pH on the positive side and textural attributes (cohesiveness, chewiness and residues) representing the undesirable properties and age on the negative side. The second PC explaining 15% of variability was mainly characterized by penetration depth on the positive side and Cooking Loss (CL), residues scores and IMF on the negative side. It is noted that ability of PCA to discriminate the two groups of hens according to their age, muscular, technological and sensory properties.

Table 3 and 4 show the different regression models explaining overall liking, tenderness and penetration depth by the other sensory attributes and sensory attributes by studied technological parameters, respectively.
The models showed that chewiness was retained in the models as the main sensory attribute explaining and affecting the variability of tenderness of the studied hens followed by residues attribute. Moreover the models explaining penetration by the sensory attributes retained juiciness as explanatory variable, explaining 29% for all hens or 51% for the aged hens. However, no model was found for the group of aged hens. In addition, the results revealed that the sensory parameters reflecting the texture: tenderness (negatively), cohesiveness, chewiness and residues (all positively) were driven by cooking losses. The explaining powers of the prediction models are variables and vary from 20-39%.

Overall liking was predominantly explained by tenderness. Regression model of aged spent hens explained 71% of its variability by only one variable, tenderness. The group of aged hens judged the tenderest in this study didn’t seem to be appreciated by their high tenderness but for other parameters non-investigated in this experiment.

Muscle characteristics were found to be able to explain more than 52% of the variability of the penetration depth for both groups and using Z-scores for the two groups grouped in one data set. For example, myofibril fragmentation index was among these muscle characteristics. High prediction powers have been recorded. For example, residues of group 1 (aged hens) were explained to 96% by three variables: Cooking loss and weight at slaughter (both positively) and WHC (negatively). These three variables were already retained in the explanatory models of other sensory traits. On another hand and in the second group, 83% of the variability of the

Fig. 1(a-b): Principal component (PC) analysis, (a) Projection of the studied variables in the two first components and (b) Bi-plot of the animal observations corresponding to group 1 (old spent hens in white) and 2 (young spent hens in black). Coefficients in the eigenvectors (loadings) for the first two PC are given.

Table 3: Regression model statistics for predicting penetration depth and sensory attributes from sensory attributes

<table>
<thead>
<tr>
<th>Attributes</th>
<th>Significance</th>
<th>R²</th>
<th>Equation</th>
<th>Significance</th>
<th>R²</th>
<th>Equation</th>
<th>Significance</th>
<th>R²</th>
<th>Equation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Penetration</td>
<td>0.04</td>
<td>0.51</td>
<td>5.66+0.29 Juiciness</td>
<td>No model</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tenderness</td>
<td>No model</td>
<td></td>
<td></td>
<td>0.005</td>
<td>0.58</td>
<td>8.01–0.40 Chewiness</td>
<td>0.013</td>
<td>0.36</td>
<td>-0.60 Chewiness</td>
</tr>
<tr>
<td>Overall liking</td>
<td>0.009</td>
<td>0.71</td>
<td>0.87+0.78 Tenderness</td>
<td>No model</td>
<td></td>
<td></td>
<td>0.004</td>
<td>0.46</td>
<td>0.68 Tenderness</td>
</tr>
</tbody>
</table>

Weight -0.33 0.31
Age -0.76 0.07
pH 0.51 -0.24
WHC -0.51 0.03
PRW 0.77 -0.16
CL -0.53 -0.64
Penetration -0.47 0.68
Tenderness 0.85 -0.17
Juiciness -0.40 0.41
Cohesiveness -0.62 -0.34
Chewiness -0.50 -0.14
Residues -0.62 -0.50
Overall liking 0.51 0.46
IFM 0.25 -0.49

Table 4: Regression model statistics for predicting penetration depth and sensory attributes from physicochemical measurements

<table>
<thead>
<tr>
<th>Attributes</th>
<th>Old spent hens</th>
<th>Young spent hens</th>
<th>Both groups (Z-scores)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Significance</td>
<td>R^2</td>
<td>Equation</td>
</tr>
<tr>
<td>Penetration</td>
<td>0.042</td>
<td>0.52</td>
<td>10.42–0.11 WHC</td>
</tr>
<tr>
<td>Tenderness</td>
<td>0.004</td>
<td>0.76</td>
<td>28.51–3.88 pH</td>
</tr>
<tr>
<td>Juiciness</td>
<td>0.08</td>
<td>0.41</td>
<td>12.19–4.56 weight</td>
</tr>
<tr>
<td>Cohesiveness</td>
<td>no model</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chewiness</td>
<td>0.08</td>
<td>0.42</td>
<td>−16.18+0.64 CL</td>
</tr>
<tr>
<td>Residues</td>
<td>0.002</td>
<td>0.53</td>
<td>−15.78+0.59 CL+3.35 weight−0.17 WHC</td>
</tr>
<tr>
<td></td>
<td>0.007</td>
<td>0.15</td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.008</td>
<td>0.28</td>
<td></td>
</tr>
</tbody>
</table>

Wt: Weight, WHC: Water holding capacity, PRW: Percentage of released water, CL: Cooking loss and MFI: Myofibril fragmentation index

Table 5: A summary of results from selected studies reporting correlations between sensory and instrumental measures of breast meat tenderness

<table>
<thead>
<tr>
<th>Simpson and Goodwin 60</th>
<th>Lyon and Lyon 60</th>
<th>Cavitt et al 23</th>
<th>Cavitt et al 23</th>
<th>Xiong et al 23</th>
<th>Present study</th>
</tr>
</thead>
<tbody>
<tr>
<td>Muscle source and number</td>
<td>Superficial muscles from 48 chickens at 8 weeks of age</td>
<td>Superficial muscles from 288 chickens at 49 days of age</td>
<td>Superficial muscles from 270 chickens at 7 weeks of age</td>
<td>Superficial muscles from 75 male and 75 female at 7 weeks of age</td>
<td>Superficial muscles from 16 hens at 337 and 220 days</td>
</tr>
<tr>
<td>Treatment</td>
<td>Autoclave at 101°C for 20 min</td>
<td>Microwave at 180°C for 4 min</td>
<td>Air convection oven to an internal end-point temperature of 76°C</td>
<td>Air convection oven to an internal end-point temperature of 76°C</td>
<td>Water bath at 85°C for 25 min</td>
</tr>
<tr>
<td>Size of panel used</td>
<td>5 untrained panelists</td>
<td>24 trained panelists</td>
<td>6 trained panelists</td>
<td>6 trained panelists</td>
<td>7 trained panelists</td>
</tr>
<tr>
<td>Size of samples</td>
<td>−</td>
<td>1.9×1.9×1.9 cm</td>
<td>4×2×7 cm</td>
<td>1.9×1.9×1.9 cm</td>
<td>1.9×1.9×1.9 cm</td>
</tr>
<tr>
<td>Correlation coefficients: sensory toughness versus</td>
<td>0.71</td>
<td>0.82 (WB)</td>
<td>0.68 (AK)</td>
<td>0.82</td>
<td>0.82 (WB)</td>
</tr>
</tbody>
</table>

cohesiveness were explained in order by pH (positively), percentage of released water (PRW) and WHC (both negatively).

**DISCUSSION**

The data showed significant effect of age at slaughter on tenderness and also some other quality traits. Similar findings have previously been reported in earlier studies 18,20. Our findings show a linear increase in meat toughness with age at slaughter. The age-related decline in meat tenderness may be due to several factors. Recent studies suggested that probably because of the structural changes of collagen and its cross-linking and differences in the thickness of fibers 18,21. Other factors may also be at the origin of those differences and may occur during post-mortem storage, such as aging, type of rigor, sarcomere length and proteolytic activity 22.

The instrumental measurement of texture by penetrometer was not found to correlate with the age of the studied hens. This finding is in agreement with findings from Schneider et al. 23 who used Allo-Kramer shear force to measure toughness of the hens at different ages.

In spite of the significant differences in tenderness scores between the two groups according to their age, the panelists judged the meat of the two poultry groups very high for the hedonic attribute, overall liking. From these findings, it seems that meat of spent hens is judged acceptable by the panelists irrespective of age. Comparable findings have previously been reported by Horsted et al. 24. According to the results from the regression models, this finding was confirmed. Hence, it can be suggest that tenderness is not the principal factor impacting overall liking of spent hens meat.
Ultimate pH was negatively correlated with animal age in agreement with earlier findings\textsuperscript{25}. The authors explained this decrease of pH with age by the increase of the glycolytic metabolism of the muscles and may be to the reserves in glycogen already found in chicken.

Water holding capacity is one of the most important traits for meat quality both in fresh meat and in processed products due to economic and sensory reasons. The cooking losses of the meat of the studied aged spent hens of group 1 were significantly (p<0.05) higher (33.4±1.40%) than those of the group 2 (31.14±2.24%). These findings are in agreement with results from\textsuperscript{20,25}. The differences found between the two groups may be due to the collagen content and its solubility during cooking. We can suppose that contraction of the connective tissue may play a major role in the migration of juice out of the muscle cells, as a function of age. In addition, earlier studies from Bertram et al\textsuperscript{26} indicate that the thermal denaturation of structural components of meat is age-dependent. It has been shown that changes in intramuscular connective tissue during growth affect toughness and water distribution.

The other parameter studied is drip loss. Exudate loss, through reduced water holding capacity, presents a diversity of negative effects throughout the poultry chain, impacting on meat quality and processability, consumer satisfaction and nutritional value of the presented meat. In this study, drip loss values were significantly different between the two spent hens ages. It has been shown in several earlier studies\textsuperscript{21,27} to play a significant role in poultry meat. Janisch et al\textsuperscript{21} suggested, that drip loss is related to extracellular protein concentrations and proteomic studies revealed numerous proteins to be involved\textsuperscript{28}.

Correlation analysis between the studied traits (Table 2) revealed that sensory attributes have close and significant correlations with each other (p<0.05) and most are above 0.5. Accordingly, numerous studies reported close relationships between sensory traits\textsuperscript{29}. For example, Lyon and Lyon\textsuperscript{39} reported a positive correlation between tenderness and overall liking of broiler breast meat. Otherwise, as expected, tenderness was negatively correlated with penetration depth values. These findings are in line to those reported in earlier studies as shown in Table S, which summarize results of some selected studies reporting some interrelationships between the sensory and instrumental measures of tenderness of broiler breast muscle. In addition, in their study, Lyon and Lyon\textsuperscript{39} reported that sensory scores were correlated to Warner-Bratzler and Allo-Kramer shear forces values of broiler breast meat. Penetration was found in this study to be significantly correlated with juiciness (r = 0.58, p<0.05). This finding is in agreement to the earlier study by Lyon and Lyon\textsuperscript{10} who found similar results between WBSF values and juiciness scores. In contrast, Caine et al\textsuperscript{31} found no significant correlation between juiciness and Texture Profile Analysis (TPA) or WBSF. Moreover, penetration values in this study were negatively related to myofibrillar fragmentation index (MFI) values. This result is in agreement to those reported by Kriese et al\textsuperscript{22} suggesting that higher the proteases activity are, more tender became the breast meat. Indeed, more investigations are needed to clarify this point.

The predicting models revealed that the sensory parameters representing texture (tenderness, cohesiveness, chewiness and residues) retained cooking loss as an explanatory variable. Despite of the weak powers of prediction, these preliminary results are in agreement with the previous studies reporting the negative impact of cooking losses on tenderness scores of chicken meat\textsuperscript{21,22}. Our findings show that muscle characteristics are not pertinent to explain a great part of the variability of poultry meat quality. Several studies found comparable findings in beef\textsuperscript{13,15}. In addition, the weak prediction powers especially when the dataset of the two groups were grouped are in agreement with the large literature in the field and for other species\textsuperscript{8,33}. Other studies using novel techniques such proteomics will be more efficient in the future for a whole characterization of the involved biological mechanisms.

CONCLUSION

This study revealed that age at slaughter had a significant effect on tenderness with no effect on meat acceptability by the panellists. Numerous relationships were observed between penetration measurements and tenderness scores and MFI. Penetrometry seems to have a great potential of being a promising predicting tool of meat tenderness. From our preliminary findings we can speculate that the old spent hens can be used for direct human consumption if required but we should provide the effect of ageing time on its structural properties to improve its textural properties.

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