WHITE STRIPING AND WOODEN BREAST DEFECTS INFLUENCE MEAT QUALITY AND MUSCLE PROTEIN CHARACTERISTICS IN BROILER BREAST MEAT

Brian Bowker*, Hong Zhuang, Elizabeth Barton and Jose Alvarez-Manilla Sanchez

USDA-ARS, U.S. National Poultry Research Center, Quality & Safety Assessment Research Unit, Athens, Georgia, USA

*Corresponding author: brian.bowker@ars.usda.gov

Abstract –The objective was to determine the effects of the wooden breast (WB) and white striping (WS) myopathies on meat quality and protein characteristics of broiler breast meat. Breast fillets (*Pectoralis major*) from a commercial processing plant were segregated into four groups: normal (neither WS nor WB), moderate (moderate for WS and WB), severe (severe for WS and WB), and WS (severe WS, no WB). Despite having greater pH_u, WS and WB fillets exhibited decreased salt-induced water uptake and increased cook loss compared to normal samples. Peak shear force and shear energy values on raw samples were greatest for the severe fillets, least for the normal, and intermediate for moderate and WS fillets. Average sarcomere length was greater in fillets exhibiting the WS and WB conditions. Compared to normal fillets, sarcoplasmic protein solubility was diminished in WS and WB fillets, but myofibrillar protein solubility was not different between the groups. Increased desmin degradation was observed in WS and WB fillets. Data demonstrate that the WB condition has a more detrimental impact on meat quality than the WS condition alone, but that both myopathies cause changes in muscle protein characteristics related to meat quality attributes.

Key Words – chicken, meat quality, myopathy

I. INTRODUCTION

With the fast growth rates of modern broilers and an increase in the proportion of birds raised to heavier weights for the cut-up and further processing markets, myopathies and meat quality abnormalities associated with larger birds are becoming a major concern for the poultry industry. In recent years, reports of meat quality defects known as white striping (WS) and wooden breast (WB) have been reported. WS is characterized by white striations on the surface of the *Pectoralis major* muscle, and has been shown to negatively impact visual consumer appeal [1]. Breast muscles with the WB condition exhibit an abnormally hard or rigid texture and tend to have a hardened ridge along the caudal portion of the muscle [2]. Breast fillets with the WB condition often display WS as well. Proximate analysis and muscle histology data have shown that breast muscles with the WS and WB conditions exhibit extensive myodegeneration/regeneration, lipidosis, and fibrosis [2-4]. Although both WS and WB have been found to negatively impact various meat quality traits, the reports have varied and the underlying effects of these conditions on muscle protein characteristics associated with meat quality are not well understood. Thus, the objective of this study was to determine the effects of the WB and WS myopathies on the quality attributes and underlying muscle protein characteristics of broiler breast fillets.

II. MATERIALS AND METHODS

Boneless, skinless breast fillets (*Pectoralis major*) from mixed sex Ross broilers (3.2 to 4.1 kg liveweight) were collected from the post-chill deboning line of a commercial processing plant at 3 h postmortem. Fillets were assigned WS and WB scores based on published criteria [1, 5]. Representative fillets (n = 12 per group) from 4 distinct categories were selected for evaluation: normal (neither WB nor WS), moderate (moderate for both WS and WB), severe (severe for both WS and WB), and WS (severe WS, no WB). At 24 h postmortem, muscle pH and color measurements (L*a*b*, dorsal surface) were recorded and fillets were packaged and frozen.

Fillets were thawed at 4°C overnight prior to quality and biochemical assessment. Purge moisture was collected from the packaging and evaluated for protein content using a biuret assay. Meat texture was assessed in the raw intact fillets using Meullenet-Owens Razor Shear

(MORS) force measurements (8 shears per fillet). Fillets were then individually homogenized for 20 sec in a food processor. Salt-induced water uptake (%) was measured in triplicate using cold 0.6 M NaCl solution, a 15 min incubation, and centrifugation at 3,000×*g*. The swollen pellet was heated for 20 min in an 80°C water bath to determine cook loss (%).

Protein solubility was measured in triplicate with 0.25 mM potassium phosphate (pH 7.2) buffer (sarcoplasmic protein solubility) and in 0.1 M potassium phosphate/1.1 M KI buffer (total protein solubility). Myofibrillar solubility was calculated from the difference between total and sarcoplasmic protein solubility. Sarcomere length and myofibrillar fragmentation index were also measured in triplicate.

Myofibrillar and sarcoplasmic protein fractions were separated by subcellular fractionation [6] in buffer containing 50 mM KCl, 20 mM Tris, pH 7.0, 2 mM EDTA, 4 mM $MgCl_2$, 5 mM 2-mercaptoethanol, 0.1 mM PMSF, and 1% (v/v) Triton X-100, and in rigor buffer (75 mM KCl, 10 mM KH_2PO_4, 2 mM $MgCl_2$ 2 mM EGTA, pH 7.0). Protein fractions were loaded onto 4-20% Tris-glycine gels for SDS-PAGE analysis and for transfer to PVDF membrane to detect desmin by western blotting.

Data were analyzed as a one-way ANOVA (SAS v. 9.2) and significant differences ($P < 0.05$) between means were identified using the Tukey's means separation method.

III. RESULTS AND DISCUSSION

In this study, the left and right *Pectoralis major* muscles from butterfly breast fillets were separated from each other and independently scored for WS and WB. Although only one fillet per carcass was utilized for subsequent meat quality and biochemical analysis, samples were selected for further evaluation only if the left and right breast muscles exhibited identical scores for both WS and WB. Because the determination of WS and WB scores was based on subjective evaluations, only those samples that unquestionably fit into one of the four distinct categories of interest were utilized.

Table 1. Physical characteristics of broiler breast meat (lsmeans).

Trait	Normal	Moderate	Severe	WS	SEM
Fillet wt. (g)	351[b]	460[a]	459[a]	439[a]	17
L*	57.8	56.7	58.1	55.9	1.1
a*	-0.1	-0.5	-0.4	-0.5	0.3
b*	14.2	14.2	14.8	13.9	0.6
pH_u	5.94[c]	6.14[ab]	6.10[b]	6.21[a]	0.05
Sarcomere Length (µm)	1.65[b]	1.73[a]	1.72[a]	1.74[a]	0.02

[ab] lsmeans with different letters differ ($p<0.05$)

As the WS and WB conditions are associated with larger birds and high breast meat yield [7, 8], it was expected that average fillet weights were greater in samples exhibiting these myopathies. Although the ventral surfaces (skin-side) of the affected fillets exhibited white striations, slightly pale color, and often a layer of viscous substance (severe fillets only), differences in the lean color on the dorsal surfaces (bone-side) were not observed between the categories (Table 1).

Compared to normal fillets, breast meat with the WS and WB conditions exhibited inferior WHC attributes as evidenced by decreased salt-induced water uptake and greater cook loss values (Table 2). In broiler breast meat, poor WHC is typically associated with decreased meat pH. However, the WS, moderate, and severe fillets exhibited increased pH_u compared to normal samples. While this has been observed previously [9], it is unknown if the increase in pH_u is due to changes in the metabolic status of the muscle at slaughter, the postmortem activity of glycolytic enzymes, or the postmortem buffering capacity of the muscle. These data suggest that the decreased WHC in breast meat with the WS and WB conditions is not a function of meat pH.

The ability of breast meat to bind and retain water during processing and cooking is also thought to be influenced by muscle protein functionality. Using solubility measurements as indicators of

Table 2. Water-holding capacity measurements of broiler breast meat (lsmeans).

Trait	Normal	Moderate	Severe	WS	SEM
Thaw loss %	6.08[a]	5.93[ab]	4.75[bc]	3.98[c]	0.61
SIWU[1] %	57.3[a]	41.2[b]	34.5[c]	46.1[b]	3.3
CL[2] %	34.5[b]	40.8[a]	42.8[a]	40.8[a]	0.9

[ab] lsmeans with different letters differ ($p<0.05$)
[1] SIWU = salt-induced water uptake
[2] CL = cook loss of swollen SIWU pellet

Table 3. Protein solubility measurements[1] of broiler breast meat (lsmeans).

Trait	Normal	Moderate	Severe	WS	SEM
Myofibrillar	153.6	154.7	143.8	155.0	6.4
Sarcoplasmic	92.2[a]	74.0[b]	74.4[b]	78.0[b]	2.6

[ab] lsmeans with different letters differ (p<0.05)
[1] Solubility measurements expressed as mg protein/g tissue

Table 4. Meullenet-Owens razor shear force (MORS) measurements of raw broiler breast meat (lsmeans).

Trait	Normal	Moderate	Severe	WS	SEM
Peak (N)	4.39[c]	4.97[b]	6.36[a]	5.17[b]	0.28
Energy (N·mm)	40.2[c]	50.2[b]	67.1[a]	54.1[b]	2.4

[ab] lsmeans with different letters differ (p<0.05)

protein denaturation, this study found that the WS and WB myopathies did not influence the degree of myofibrillar protein denaturation (Table 3). However, sarcoplasmic protein solubility was less in WS and WB fillets than normal samples. These findings are consistent with previous data showing that broiler breast meat WHC attributes are more closely related to sarcoplasmic protein denaturation than myofibrillar protein denaturation [10]. Decreases in sarcoplasmic protein content may also be due to sarcolemma damage causing leakage of fluid from fibers of myopathic fillets.

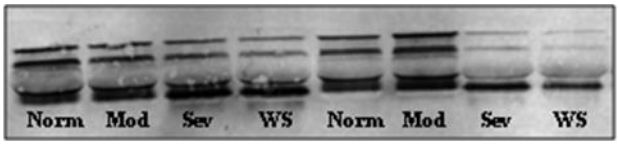
The decreased WHC of WS and WB fillets may be in part caused by differences in breast meat composition. It is well-established that fillets exhibiting these myopathies have greater fat and connective tissue contents and a lower total protein content as a proportion of muscle weight [3, 4]. As a large portion of water within muscle is held within the myofibrils, decreases in overall myofibrillar protein content may decrease the water binding potential of the meat. The increased fat content of fillets exhibiting the WB and WS conditions may also contribute to their greater cook loss.

Surprisingly, thaw loss was slightly greater in normal fillets compared to both WS and severe fillets (Table 2). It is possible that WS and WB fillets lost more muscle exudate than normal samples prior to freezing, thus causing lower subsequent moisture losses. It is also possible that size and shape differences between fillets may have played a role. The composition of the moisture lost from the tissue upon freezing and thawing also seemed to be influenced by the WS and WB conditions. The average protein content of the moisture collected from the packaging after thawing was greater in the normal fillets (136 mg/ml protein) compared to the moderate, severe, and WS samples (105, 96, and 109 mg/ml protein, respectively). As exudate from muscle contains predominantly sarcoplasmic proteins, it is possible that the decreased solubility of the sarcoplasmic protein fraction in WS and WB fillets may have played a role. Additional research is needed, however, to explain the observations related to the exudate expressed from these fillets following freezing and thawing.

The WB and WS conditions had a strong impact on raw meat texture (Table 4). Both peak shear force and total shear energy were greatest in severe fillets and least in normal fillets with moderate and WS fillets intermediate. The overall low shear values observed in this study were likely due to the fact that fillets were aged until 24 h postmortem and then subjected to a freeze-thaw cycle prior to the texture being measured on the raw samples. Similar texture differences between normal and WB fillets have been observed in cooked breast fillets both before and after a freeze-thaw cycle [11]. Sarcomere lengths were greater in WS and WB fillets compared to normal samples (Table 1). This observation is consistent with other reports [5] and suggests that the meat toughness associated with these myopathies is not due to excessive shortening during rigor development. It is likely that meat texture differences in these fillets are a function of connective tissue.

Minor differences in the composition of the myofibrillar and sarcoplasmic protein fractions were detected using SDS-PAGE analysis (data not shown). Within the myofibrillar protein fractions a 115 kDa protein band was more abundant in fillets exhibiting the myopathic conditions. Within the sarcoplasmic protein fractions, the myopathic fillets exhibited a lower relative abundance of the 47 kDa band and a greater relative abundance of bands corresponding to 26 kDa, 70 kDa, and high molecular weight proteins (>150 kDa). Variations in the proportions of the different proteins within the electrophoretic profiles may be due to differences in muscle protein catabolism in the living tissue or

Figure 1. Representative desmin western blot of broiler breast meat.



postmortem changes in muscle protein degradation and denaturation. Further research is needed to determine the mechanistic link between SDS-PAGE results and meat quality traits.

With regards to protein degradation, average MFI values between normal and myopathic fillets were not different (data not shown). Desmin degradation, however, seemed to be influenced by the WS and WB conditions. Using western blot analysis, it was observed that fillets expressing these myopathies had less intact desmin and a greater abundance of desmin degradation products than normal fillets (Fig. 1). These observations suggest that more postmortem proteolysis occurs in fillets with the WB and WS conditions and further indicates that the toughness of these fillets is likely due to connective tissue properties. Further research including multiple postmortem aging periods and enzymatic data is needed, however, to fully determine the influence of these myopathies on postmortem proteolysis.

IV. CONCLUSION

This study demonstrates that broiler breast meat with the WB and WS conditions exhibit inferior WHC and meat texture attributes. The presence of the WB condition, however, negatively influences meat quality to a greater degree than WS alone. Even moderate degrees of WB and WS conditions are adequate to influence muscle protein characteristics and meat quality attributes. Protein characteristics indicate that both the WS and WB conditions decrease sarcoplasmic protein functionality and may increase myofibrillar protein degradation. Further research is needed to elucidate both the etiology of these myopathies and the mechanisms by which alterations in the muscle proteins influence meat quality.

REFERENCES

1. Kuttappan, V.A., Lee, Y.S., Erf, G.F., Meullenet, J.F.C., McKee, S.R., & Owens, C.M. (2012). Consumer acceptance of visual appearance of broiler breast meat with varying degrees of white striping. Poultry Science 91:1240-1247.
2. Sihvo, H.K., Immonen, K., & Puolanne, E. (2014). Myodegeneration with fibrosis and regeneration in the *Pectoralis major* muscle of broilers. Veterinary Pathology 51:619-623.
3. Kuttappan, V.A., Shivaprasad, H.L., Shaw, D.P., Valentine, B.A., Hargis, B.M., Clark, F.D., McKee, S.R., & Owens, C.M. (2013). Pathological changes associated with white striping in broiler breast muscles. Poultry Science 92:331-338.
4. Soglia, F., Mudalal, S., Babini, E., Di Nunzio, M., Mazzoni, M., Sirri, F., Cavani, C., & Petracci, M. (2016). Histology, composition, and quality traits of chicken *Pectoralis major* muscle affected by wooden breast abnormality. Poultry Science 95:651-659.
5. Tijare, V.V., Yang, F.L., Kuttappan, V.A., Alvarado, C.Z., Coon, C.N., & Owens, C.M. (2016). Meat quality of broiler breast fillets with white striping and woody breast muscle myopathies. Poultry Science http://dx.doi.ort/10.3382/ps/pew129
6. Pietrzak, M., Greaser, M.L., & Sosnicki, A.A. (1997). Effect of rapid rigor mortis processes on protein functionality in *Pectoralis major* muscle of domestic turkeys. Journal of Animal Science 75:2106-2116.
7. Kuttappan, V.A., Brewer, V.B., Apple, J.K., Waldroup, P.W., and Owens, C.M. (2012). Influence of growth rate on the occurrence of white striping in broiler breast fillets. Poultry Science 91:2677-2685.
8. Lorenzi, M., Mudalal, S., Cavani, C., & Petracci, M. (2014). Incidence of white striping under commercial conditions in medium and heavy broiler chickens in Italy. Journal of Applied Poultry Research 23:754-758.
9. Mudalal, S., Lorenzi, M., Soglia, F., Cavani, C., & Petracci, M. (2015). Implications of white striping and wooden breast abnormalities on quality traits of raw and marinated chicken meat. Animal 9:728-734.
10. Bowker, B., & Zhuang, H. (2015). Relationship between water-holding capacity and protein denaturation in broiler breast meat. Poultry Science 94:1657-1664.
11. Chatterjee, D., Zhuang, H., Bowker, B., Rincon, A.M., & Sanchez-Brambila, G. (2016). Instrumental texture characteristics of broiler *Pectoralis major* with the wooden breast condition. Poultry Science (In Press).