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Assessment of meat quality distributions of breast fillets with woody breast condition in the raw and cooked state

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Abstract The objective of this study was to determine meat quality distributions and assess hardness distributions in raw and cooked breast fillets with the woody breast (WB) condition, in addition to evaluating the relationship between water properties and WB severity. A total of 90 breast fillets were collected and categorized as normal (NORM), mild (MILD) and severe (SEV). Breast weight, drip loss, compression measurements, cook loss, shear and texture profile analysis (TPA) values were measured for each sample by fillet location (cranial to caudal) and sampling depth (cranial-superficial, cranial-internal, middle-superficial, and middle-internal) in the raw and cooked meat state. Low-field NMR relaxation measurements were also collected for both the raw and cooked fillets. Results indicate that severe WB expressed increased hardness, a higher water content (bound water and free water) and reduced meat quality attributes in raw and cooked meat. Breast fillet hardness and meat quality distributions were unevenly distributed between fillets, compression measurements were higher mainly in the cranial region, and progressively decreased toward the caudal region for both

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raw and cooked fillets. Shear force and energy values were higher in the cranial region than in the middle region, and TPA values were higher in superficial regions rather than internal portions. Additionally, low-field NMR could be used to predict WB through variation in water properties as thermal processing reduces water distributing abilities in affected fillets.

Keywords Woody breast \cdot Meat quality \cdot Hardness \cdot Texture analysis \cdot Low-field NMR

Abbreviations

WB	Woody breast
NORM	Normal
MILD	Mild
SEV	Severe
TPA	Texture profile analysis
NMR	Nuclear magnetic resonance
MORS	Meullenet-Owens Razor Shear
BMORS	Blunt Meullenet-Owens Razor Shear
CF	Compression force
CE	Compression energy
Н	Fillet height
BW	Breast weight
DL	Drip loss
CL	Cook loss
CS	Cranial-superficial
CI	Cranial-internal
MS	Middle-superficial
MI	Middle-internal
SF	Shear force
SE	Shear energy
WHC	Water holding capacity

Introduction

Woody breast (WB) is an emerging and challenging meat quality defect affecting the broiler Pectoralis major muscles in the modern global poultry industry (Petracci et al. 2019; Caldas-Cueva and Owens 2020a). Woody breast is generally characterized by distinct hardness throughout the fillet utilizing a subjective palpation technique. In the most severe cases, affected fillets tend to express muscle rigidity throughout the fillet with a prominent ridge-like bulge in the caudal region. Furthermore, the WB condition has been known to exhibit higher pH, increased drip loss, higher fat and collagen contents, and thus lower levels of protein (Soglia et al. 2016a; Tijare et al. 2016). Chemical composition and water property alterations caused by WB result in fillets with poorer meat quality, reduced yields, and reduced sensory acceptance (Cai et al. 2018; Bowker and Zhuang 2019; Pang et al. 2020a, b; Xing et al. 2020b). The WB incidence has progressively increased over the last decade causing over 500 million dollars in economic losses to the industry (Caldas-Cueva and Owens 2020a).

Woody breast fillets have commonly been identified and classified utilizing a subjective tactile evaluation with varying scoring systems proposed by a multitude of researchers (Tijare et al. 2016; Sun et al. 2018, 2021a; Bowker and Zhuang 2019). Due to these subjective differences, objective compression tests have been developed to assess and categorize the hardness of chicken fillets. Previous studies have reported a strong correlation between WB and compression force, regardless of sample size, test settings, storage method (fresh or frozen), and storage time (Mudalal et al. 2015; Sun et al. 2018; Soglia et al. 2017; Pang et al. 2020a; Campo et al. 2020). Several texture analysis methodologies, such as the Meullenet-Owens Razor Shear (MORS), the blunted version of MORS (BMORS), Warner-Bratzler Shear, Allo-Kramer Shear and Texture Profile Analysis (TPA) have been utilized to detect the WB condition. Previous research suggests that meat quality of breast fillets has been affected by storage method, treatment/processing strategies, and differences in fillet region (Tijare et al. 2016; Sun et al. 2018, 2021a; Petracci et al. 2019; Tasoniero et al. 2019; Bowker and Zhuang 2019; Xing et al. 2020b). In fact, recent studies have consistently reported that the WB condition has a negative effect on cooking time, yield, and final quality attributes of WB meat-based products (Caldas-Cueva et al. 2020b, 2021a; b; Sun et al. 2021b, c).

High-yielding broilers have continually increased total breast yield but have coincidentally increased the total incidence of WB; thus, making the size and thickness of the *Pectoralis major*, from heavy deboned markets, adverse in nature. Breast fillets are normally portioned/trimmed to present a more uniform size, shape, and thickness for further processing or food markets (Bowker et al. 2018). Therefore, it is important to understand how WB hardness affects meat quality throughout the fillet. Sihvo et al. (2014), Soglia et al. (2016a), and Tasoniero et al. (2019) affirmed that muscle degeneration, which leads to abnormal hardness, is higher on the ventral surface, particularly in the cranial region of WB fillets. Sun et al. (2017) reported that compression force values were higher in the cranial region than in the middle region of WB fillets. Additionally, the negative impact of WB on marination and cooking performance were less severe on the dorsal side when compared to the ventral side of affected fillets (Bowker et al. 2018). Researchers have also asserted that the WB myopathy has a significant impact on the distribution of water within the muscle tissue of WB meat, which exhibits altered moisture properties (more extramyofibrillar water and less intra-myofibrillar and hydration water) compared to normal fillets (Tasoniero et al. 2017; Soglia et al. 2016a, b; Pang et al. 2020b). In this context, although the instrumental quality evaluation of WB meat has been widely investigated, variation in meat quality, particularly the hardness distribution in WB fillets and variation in water/moisture properties, are still not fully understood. This lack of knowledge in meat quality evaluation, by affected fillet region, is important to investigate as it is difficult to reduce economic losses by fillet portioning without knowing the limits of acceptability. Thus, the objective of this study was to determine: (1) the relationship between hardness distribution and WB severity of breast fillets in different locations for both the raw and cooked state; (2) meat quality differences among WB classes by shearing location in cooked meat; and (3) water properties measured by low-field NMR of WB fillets in the raw and cooked state.

Materials and methods

Sample preparation and tactile evaluation

High breast-yielding broilers were processed at 6 weeks of age utilizing a commercial processing system. A total of 90 breast fillets (n = 30/category, deboned at 2 h postmortem) were collected and categorized as normal (NORM), mild (MILD), and severe (SEV) for WB by the degree of palpable hardness according to Tijare et al. (2016). After scoring, all fillets were individually packed in zip-sealed plastic bags and stored in a 4 °C walk-in cooler for further analysis. Seventy-five (n = 25/category) breast fillets were used to conduct meat quality analysis, while the remaining 15 breast fillets (n = 5/category) were separated for NMR relaxation measurements.

Compression and drip loss measurements in raw breast fillet

Compression analysis was completed on day of processing according to the method described by Sun et al. (2018) with minor modifications. Compression analysis was achieved using a 6 mm flat probe affixed to a TA.XT Plus Texture Analyzer (Texture Technologies Corp., Scarsdale, NY). In this study, 5 locations (1 to 5 marked as cranial, cranial-middle, middle, middle-caudal, caudal) were considered in the breast fillet as shown in Fig. 1A. Breast fillets were compressed in duplicate to 30% of their initial height at each location. Following analysis, the average compression force (CF), compression energy (CE), and fillet height (H) at each location were recorded. The trigger force was set at 5 g, probe height set to 45 mm (higher than the thickest fillet sample), pre- and post-probe speeds were both set to 10 mm/s, and the test speed of the probe was 5 mm/s. Additionally, breast weight (BW) was measured on day of processing and drip loss (DL) of each fillet was calculated as the percent loss of refrigerated breast weight versus the weight at 24 h postmortem.

Breast fillet cooking and instrumental shear analysis

Breast weight was recorded prior to cooking, then all fillets were packaged in individual retort pouches separately. Breast fillets were fully immersed in a water bath at 84 °C and cooked until their central temperature reached 76 °C (Brambila et al. 2018; Xing et al. 2020b). After cooking, fillets were cooled to room temperature and reweighed. Cook loss (CL) was calculated by percent loss of fillet weight from pre- and post-cooking weights. Then breast fillets were individually packaged in aluminum foil sheets



Fig. 1 Instrumental Compression and texture analysis of breast fillets at different locations or sampling depth in both raw and cooked meat. (A) Compression tests were conducted in duplicate at each location from 1 to 5 (from cranial to caudal region of breast fillet, respectively) in raw and cooked breast fillets. (B) Texture analysis was conducted in both the cranial and middle region with sampling depth treatment. CS: cranial-superficial, CI: cranial-internal, MS: middle-superficial, MI: middle-internal

and stored overnight at 4 °C for texture analysis the following day.

Meat quality evaluation of cooked breast fillets was conducted according to the procedures by Maxwell et al. (2018) and Xing et al. (2020b) with mild modification. The main factors of WB category (NORM, MILD and SEV) and sampling location [cranial-superficial (CS), cranialinternal (CI), middle-superficial (MS), and middle-internal (MI)] were investigated in the present study. Breast fillet strips (10 mm \times 10 mm \times 30 mm) were collected from each breast fillet (Fig. 1B) parallel to muscle fibers. Briefly, each sample was sheared perpendicular to muscle fiber direction in duplicate using the TA. XT Plus Texture Analyzer (Texture Technologies Corp., Scarsdale, NY) equipped with a HDPS blade. Results were then reported as average peak shear force (SF, N) and total shear energy (SE, N.mm).

Texture profile analysis

Cooked samples were also subjected to texture profile analysis (TPA) using the same TA. XT Plus Texture Analyzer equipped with a P/36 R probe according to Xing et al. (2020b) with slight modification. Breast fillet cubes (15 mm \times 15 mm \times 15 mm) were collected from the breast fillet (Fig. 1B) and a duplicate compression cycle test was set to compress 50% of the original height, elapsing a total of 5 s. The pre- and post-speeds were set at 5 mm/s, test speed was set to 5 mm/s, and a trigger force of 5 g was utilized. Average values for hardness, springiness, cohesiveness, gumminess, chewiness, and resilience were recorded for TPA results.

Nuclear magnetic resonance (NMR) relaxation measurements

Water distribution properties of woody breast was carried out using a Niumag Pulsed NMR analyzer set with experimental parameter settings published by Tasoniero et al. (2017) and modified to better fit the current experiment. For the test set, cube samples (20 mm \times 20 mm \times 20 mm) were excised from the cranial region of breast fillets in both raw meat and immediately following the cooking process. Both raw and cooked samples were kept at room temperature and thermostatically cooled at 25 °C, respectively. Each sample was placed in the bottom of a nuclear magnetic tube with a diameter of 60 mm and loaded into the NMR probe. NMR measurements (Instrument Model: MesoMR23-060 V-I, NIUMAG Corp.) were calculated using the following settings: NMR spectra was acquired using a Carr-Purcell-Meiboom-Gill pulse sequence with a main frequency and offset frequency of 25 Hz and 411,671.61 MHz, respectively. In the obtained

NMR spectra, relaxation times of (0.1-10 ms), (10-100 ms), and (100-1000 ms) represented T_{2b} , T_{21} , and T_{22} , respectively. Individual water content of bound water (S_{2b}) , immobilized water (S_{21}) , and free water (S_{22}) were calculated as the ratio of peak area for each corresponding relaxation period to the total area of relaxation time (0-1000 ms). Additionally, NMR inversion images were collected from each fillet in both the raw and cooked sample using Multi-Slice Echo.

Statistical analysis

Data from this study were analyzed using the GLM procedure in JMP 15 (SAS, 2020). Compression measurements were analyzed by testing the main effect of WB category, fillet location, and the interaction of WB category and fillet location in both the raw and cooked meat state. Drip loss, cook loss, and water properties were analyzed by testing the main effect of WB severity for raw and cooked meat samples. Instrumental shear and TPA values of cooked meat were analyzed by testing the main effect of WB category, sampling depth, and the interaction of WB category and sampling depth. Means were then separated using a Tukey's HSD test, as appropriate, with a significance level set at $P \leq 0.05$.

Results

Breast weight, water holding capacity and hardness distribution of breast fillets

Meat quality characteristics for various regions in both raw and cooked fillets exhibiting various WB severities are expressed in Table 1. As expected, BW increased as WB severity increased (P < 0.05). DL was higher in MILD and SEV fillets over NORM fillets (P < 0.05). Furthermore, CL increased as WB severity increased (P < 0.05).

Compression measurements for raw breast fillets were different between WB severity (P < 0.0001), fillet location (P < 0.0001), and the interaction between the two (P < 0.01). CF, CE, and H were higher in SEV fillets than in MILD fillets, which were both higher than NORM ones (NORM < MILD < SEV, P < 0.05). CF and CE mainly decreased from locations 1 to 3 (location 1 > 2 > 3, P < 0.05). CF expressed no differences from locations 3 to 5, while CE significantly decreased from location 3 to 5 as location 4 was independent of the two (P < 0.05). H was similar for locations 1 and 2 which were higher than from each of the other locations 3 to 5 (location 1, 2 > 3 > 4 > 5, P < 0.05). A WB category by fillet location interaction was observed and can be seen in Fig. 2A–C with similar tendencies found in CF, CE, and H (P < 0.01). In NORM fillets,

CF was higher in location 1 than in locations 2 through 5 which expressed no differences. CE was higher in location 1 than in locations 3 to 5, with location 2 being intermediate. H in locations 1 and 2 were similar but decreased from locations 3 to 5. In MILD fillets, CF and CE decreased significantly from locations 1 to 3 and were similar from locations 3 to 5. H was similar in locations 1 and 2 but decreased significantly from locations 2 to 5 (P < 0.05). In SEV fillets, CF and CE were similar between locations 1 and 2 but decreased to locations 3 to 5 which expressed no differences. H was similar in locations 1, 2, and 3, intermediate for location 4, and higher than location 5.

For cooked fillets, compression measurements were different between WB category (P < 0.0001) and fillet location (P < 0.0001). CF, CE, and H increased as WB severity increased. CF and CE for locations 1 and 2 were higher than in locations 3 and 5, while location 4 was intermediate (Table 1). H was higher in locations 1 and 2 than location 3, which were higher than locations 4 and 5 (Table 1). CF was significantly different for the interaction of WB category and fillet location interaction (Fig. 2D, P < 0.01), whereas H and CE were not different for this interaction. In NORM fillets, location 2 exhibited the greatest CF over location 3, followed by location 1; however, location 1 expressed higher CF than location 5, with location 4 being intermediate. In MILD fillets, CF in locations 1 and 2 were higher than in locations 3 through 5. In SEV fillets, CF was highest in location 2 than locations 3 through 5, while location 1 was intermediate.

Instrumental texture evaluation of cooked breast fillets

Shear values and instrumental TPA parameters of cooked fillets with various degrees of WB and sampling depth are expressed in Table 2. SF and SE were different between WB category (P < 0.0001), fillet region (P < 0.0001), and their subsequent interaction (P < 0.001, Fig. 2E–F). SF and SE were higher in SEV fillets than in MILD and NORM which expressed no differences. Additionally, both SF and SE increased for the superficial side over the internal layer, regardless of fillet region (CS, CI > MS, MI, P < 0.05). For the cranial region, both SF and SE in CS increased as WB severity increased, and SF, SE in CI were higher in SEV groups than both the MILD and NORM fillets. However, within the middle region, SF and SE values were similar between WB categories regardless of location on the breast (MS, MI, Fig. 2E-F) except for NORM, MS and SEV, MI.

TPA parameters were different between WB category (P < 0.0001) and fillet region (P < 0.0001), with no interaction between the two (P > 0.05). Within WB categories, hardness increased as WB severity increased.

Table 1 Effect of woody breast category and fillet location on compression and water holding capacity (drip loss and cook loss) measurements for raw and cooked breast fillets

Parameter	WB Category ¹			SEM		Locatio	n ²		SEM	<i>P</i> -value			
	NORM	MILD	SEV		1	2	3	4	5		WB	Location	Interaction
Raw													
$CF(N)^3$	1.81 ^c	3.28 ^b	5.68 ^a	0.13	6.32 ^a	5.21 ^b	2.39 ^c	1.95 ^c	2.08°	0.17	< 0.0001	< 0.0001	< 0.0001
CE (N.mm) ⁴	3.15 ^c	6.34 ^b	11.74 ^a	0.29	12.30 ^a	10.51 ^b	5.13 ^c	3.89 ^{cd}	3.55 ^d	0.38	< 0.0001	< 0.0001	< 0.0001
H (mm) ⁵	23.55 ^c	28.77 ^b	33.3 ^a	0.22	33.15 ^a	33.16 ^a	29.53 ^b	25.04 ^c	21.95 ^d	0.29	< 0.0001	< 0.0001	0.0054
BW (g) ⁶	194.24 ^c	252.47 ^b	297.90 ^a	5.38	_	_	_	-	_	_	< 0.0001	-	_
DL (%) ⁷	4.96 ^b	6.16 ^a	6.70 ^a	0.26	_	_	_	-	_	_	< 0.0001	-	_
Cooked													
CF (N)	11.12 ^c	15.11 ^b	16.44 ^a	0.26	16.24 ^a	17.46 ^a	13.29 ^b	12.57 ^{bc}	11.55 ^c	0.34	< 0.0001	< 0.0001	0.0052
CE (N.mm)	28.69 ^c	43.49 ^b	47.63 ^a	0.95	50.16 ^a	51.03 ^a	35.57 ^b	32.90 ^{bc}	30.02 ^c	1.23	< 0.0001	< 0.0001	0.0676
H (mm)	30.50 ^c	34.70 ^b	35.79 ^a	0.26	38.08^{a}	37.01 ^a	32.80 ^b	30.70 ^c	29.71 ^c	0.33	< 0.0001	< 0.0001	0.4490
CL (%) ⁸	21.86 ^c	24.95 ^b	28.63 ^a	0.6	-	-	-	-	-	-	< 0.0001	-	-

 1 NORM = normal, fillets were soft and flexible throughout; MILD = mild, fillets were hard primarily in the cranial region with some flexibility in the middle to caudal region; SEV = moderate and severe, fillets were extremely hard and rigid throughout with limited or no flexibility from cranial to caudal region

²1-5 means different locations from cranial to caudal region of breast fillet

 ${}^{3}CF$ = compression force

 4 CE = compression energy

 ${}^{5}\text{H}$ = height of breast fillet on each location

 $^{6}BW = breast weight$

 $^{7}DL = drip loss$

 ${}^{8}CL = cook loss$

 $^{a-d}$ Means within the same row followed by different superscript letters differ significantly (P < 0.05)



Fig. 2 Interaction of woody category and fillet location or sampling depth on compression measurements and shear analysis of raw (A-C) and cooked (D-F) breast fillets. Means with different superscript

letters (a-i) differ significantly (P < 0.05). CF: compression force; *CE* compression energy; *H* fillet height of each location; *SF* shear force; *SE* shear energy

Parameter	WB categ	ory ¹		SEM	Sampling o	lepth ²			SEM	<i>P</i> -value		
	NORM	MILD	SEV		CS	CI	MS	IM		WB	Location	Interaction
Instrumental blade shea	r											
Shear force (N)	26.07 ^b	33.54 ^b	53.38^{a}	2.35	51.33^{a}	49.50^{a}	21.51 ^b	28.32 ^b	2.72	< 0.0001	< 0.0001	0.0005
Shear energy (N.mm)	223.85 ^b	262.18 ^b	382.79 ^a	15.15	398.48^{a}	366.67^{a}	181.81 ^b	211.46 ^b	17.50	< 0.0001	< 0.0001	0.0003
TPA												
Hardness (N)	743.60 ^c	1011.77 ^b	1193.95 ^a	46.56	1151.04^{a}	936.67 ^{bc}	1054.49^{ab}	790.20°	53.77	< 0.0001	< 0.0001	0.7930
Springiness	0.69^{a}	0.65^{b}	0.69^{a}	0.008	0.70^{a}	0.66^{b}	0.71^{a}	0.64^{b}	0.01	0.0001	< 0.0001	0.5924
Cohesiveness	0.66^{b}	0.65^{b}	0.69^{a}	0.005	0.70^{a}	0.66^{b}	0.67^{b}	0.63°	0.005	< 0.0001	< 0.0001	0.2974
Gumminess	498.10 ^b	706.97^{a}	788.67 ^a	33.12	806.15 ^a	621.22 ^b	752.68 ^a	508.26°	38.26	< 0.0001	< 0.0001	0.8073
Chewiness	356.59 ^b	513.06^{a}	525.58 ^a	26.19	582.28 ^a	423.23 ^b	537.06 ^a	337.74°	30.24	< 0.0001	< 0.0001	0.7301
Resilience	$0.307^{\rm b}$	0.301^{b}	0.346^{a}	0.004	0.344^{a}	0.304^{b}	0.323^{a}	0.291°	0.005	< 0.0001	< 0.0001	0.2251
¹ NORM = normal, fillet severe, fillets were extre	s were soft a mely hard a	nd flexible thr nd rigid throu	oughout; MILE ghout with limi) = mild, fill ited or flexib	ets were hard ility from cra	primarily on e	cranial region v region	vith some flex	ibility in mi	ddle to caudal 1	egion; SEV =	moderate and
² Texture analysis was (MI = middle-internal	conducted in	both the crai	iial and middle	e region wit	h superficial	and internal t	reatments, CS	= cranial-sup	erficial, CI =	= cranial-intern	al, MS = midd	e-superficial,

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Springiness was similar in NORM and SEV fillets over MILD fillets (P < 0.05). Cohesiveness and resilience were higher in SEV fillets than in MILD and NORM fillets, which expressed no differences. Gumminess and chewiness were higher in SEV and MILD fillets than in NORM fillets. For sampling depth, hardness was similar between CS and MS which were higher than MI, and intermediate for CI. Springiness was similar between CS and MS which were higher than CI and MI (P < 0.05). Cohesiveness was higher in CS than both CI and MS which were higher than MI. Gumminess, chewiness, and resilience were higher in both CS and MS than in CI, while MI was intermediate.

Water properties in raw and cooked breast fillets

Water property distributions for NORM, MILD, and SEV fillets are shown in Fig. 3A. T_{2b} relaxation (0–10 ms) corresponds to bound water, T_{21} relaxation (10–100 ms) corresponds to immobilized water, and T_{22} relaxation (100–1000 ms) corresponds to free water. The T_{21} peak decreased as WB severity increased for both raw and cooked fillets. The T_{2b} peak was higher in NORM fillets than SEV fillets, which were both higher than MILD fillets. In addition, the T_{21} peak in raw samples shifted right and the T_{21} peak width of cooked samples increased alongside severity. On the other hand, both T_{2b} and T_{21} peaks shifted left from the raw to cooked meat state.

Peak area proportions were the contents of bound water (S_{2b}) , immobilized water (S_{21}) , and free water (S_{22}) for each sample (Table 3). In the raw state, S_{2b} , S_{21} , and S_{22} were different among WB categories (P < 0.05). Higher S_{2b} peak area ratios were consistently observed in MILD and SEV fillets when compared to NORM fillets (P < 0.05). Free water (S_{22}) was higher in SEV fillets in comparison to NORM fillets (P < 0.05), whereas the S_{22} for MILD fillets did not differ compared to those from the other WB groups. Contrarily, S_{21} was higher in NORM fillets than SEV fillets (P < 0.05); however, no differences were observed in S_{21} for MILD fillets when compared to the other WB categories. In cooked breast fillets, S_{2b} , S_{21} , and S_{22} did not differ among WB categories (P > 0.05).

^{a-c}Means within the same row followed by different superscript letters differ significantly (P < 0.05)

The proton-weighted image of all WB categories in both raw and cooked fillets are presented in Fig. 3B. As expected, brighter yellow color was observed in raw breast meat than in cooked fillets. The brightness of each sample within the images increased visually as the WB severity increased, regardless of meat state (raw or cooked). The raw SEV fillets expressed the highest level of brightness in which more water content was expressed, and the cooked fillets expressed relatively lower levels of brightness.



Fig. 3 NMR relaxation distributions and proton-weighted image of breast fillets with WB condition in raw and cooked meat

Table 3 Water properties ofbreast fillets with different WBconditions in raw and cookedmeat

Parameter	WB Catego	ry ¹		SEM	P-value	
	Norm	Mild	SEV			
Raw						
Bound Water (S _{2b})	0.0059^{b}	0.0184 ^a	0.0207^{a}	0.0029	0.0123	
Immobilized Water (S21)	0.9631 ^a	0.9179 ^{ab}	0.8504 ^b	0.0270	0.0419	
Free Water (S ₂₂)	0.0264 ^b	0.0822^{ab}	0.1614 ^a	0.0290	0.0301	
Cooked						
Bound Water (S2b)	0.0011	0.0015	0.0019	0.0006	0.6944	
Immobilized Water (S21)	0.9838	0.9864	0.9849	0.0040	0.8783	
Free Water (S ₂₂)	0.0151	0.0118	0.0136	0.0030	0.7891	

¹NORM normal, fillets were soft and flexible throughout; *MILD* mild, fillets were hard primarily on cranial region with some flexibility in middle to caudal region; *SEV* moderate and severe, fillets were extremely hard and rigid throughout with limited or flexibility from cranial to caudal region

^{a-b}Means within the same row followed by different superscript letters differ significantly (P < 0.05)

Discussion

In raw breast fillets, BW, DL, CF, H, and CL measurements were all in agreement with previous studies. Breast weight of SEV fillets was higher than NORM fillets (Chatterjee et al. 2016; Dalle Zotte et al. 2017; Sun et al. 2018; Xing et al. 2020a, b), and WB fillets exhibited thicker fillets than NORM fillets, regardless of fillet location (Zambonelli et al. 2016). A potential explanation could be that the increased weight observed in WB fillets can result in dimensional changes, such as an increase in fillet thickness (Mudalal et al. 2015; Zambonelli et al. 2016). A decreased water holding capacity (WHC) has been observed in WB fillets with higher DL in raw meat (Cai et al. 2018; Sun et al. 2018; Bowker et al. 2018) and higher CL in cooked meat (Tijare et al. 2016; Soglia et al. 2016a; Sun et al. 2021a). Several authors have also reported that CF values increased as WB severity increased in breast fillets (Soglia et al. 2017; Sun et al. 2018, 2021a; Pang et al. 2020a). In the current study, CE was used as a new

feature to distinguish differences among WB severities and could potentially better classify WB fillets due the larger range of values as compared to CF. The higher compression measurements observed in WB fillets in raw meat could be explained by histological and compositional alterations caused by this myopathy. Soglia et al. (2016a) reported that fibrosis in WB results in higher connective tissue content than NORM fillets which could lead to higher CF and CE.

It is well known that protein denaturation and aggregation, moisture loss, collagen solubility, and muscle fiber shrinkage occur in meat during thermal processing, which lead to an increased meat toughness, and thus higher shear values. Chatterjee et al. (2016) and Bowker and Zhuang (2019) reported that shear values (force and energy) were higher in cooked fillets when compared to raw fillets. Tasoniero et al. (2019) suggests that cooked fillets require higher CF than raw fillets, regardless of myopathy severity (normal, moderate, severe) or location (cranial, cranialmedial, medial). As expected, the compression values of cooked fillets were higher than those for raw fillets. The increased hardness observed in cooked fillets could be explained by biochemical changes that occur during thermal processing. The process of thermal manipulation leads to protein denaturation and collagen/muscle fiber shrinkage, while insoluble collagen stabilizes to produce a "stiffer" product. The combination of unfolding proteins and stabilizing collagen then produces a final produce that is harder than before.

In an attempt to better understand hardness distribution of the WB myopathy throughout fillets, 5 locations (see Fig. 1A) were analyzed in this study. In raw breast fillets, CF and CE significantly decreased from the cranial region to the cranial-middle region, with further reduction to the middle region, regardless of WB condition. This data is consistent with the findings of Tasoniero et al. (2019), and our data further indicates no significant changes occur in hardness from the middle to caudal regions. This implies that fillet hardness varies from the cranial to middle region, even though fillet thickness significantly decreases from the cranial to middle region of all fillets, regardless of being affected by WB. Dalle Zotte et al. (2017) reported that WB or breast lesions are mainly focalized in the cranial and/or cranialmiddle region of fillets. Additionally, localized lesions express higher amounts of collagen and pathological fibrosis (Clark and Velleman 2016). The combination of more distinct hardness from the cranial region to the middle region of fillets and these focalized lesions could potentially explain the variability of WB hardness.

For the current study, CF, CE, and H of cooked fillets remained higher in all fillet locations compared to raw fillets. CF in cooked fillets was higher in locations 1 and 2 (cranial/middle) than all other locations which is in agreeance with Tasoniero et al. (2019) who suggest that WB severity/hardness varies in distribution throughout the length of the breast fillet. The results in this study indicate that hardness distribution is uneven for cooked fillets exhibiting the WB condition, which could provide basic information for detecting WB associated with physical measurements.

It has been well established that WB reduces WHC, meat quality attributes, and consumer acceptance (Tijare et al. 2016; Maxwell et al. 2018; Caldas-Cueva and Owens 2020a; Xing et al. 2020b). Instrumental texture measurements have been widely conducted in cooked meat quality analysis. However, shear force values for cooked breast fillets with the WB condition are inconsistent. Tasoniero et al. (2016), Jarvis et al. (2020), and Hasegawa et al. (2020) observed higher Warner–Bratzler shear force and Allo-Kramer shear force in WB fillets compared to NORM fillets. In contrast, Mudalal et al. (2015) and Cai et al. (2018) observed no differences in shear force values for breast fillets. Furthermore, Byron et al. (2020) reported that NORM fillets had higher shear force than SEV fillets in the upper, middle, and lower portions of breast fillets when cooked on the day of processing. For the MORS methodology, in cooked fillets, Chatterjee et al. (2016) and Bowker and Zhuang (2019) indicate that WB requires higher shear force and/or shear energy than NORM fillets. However, Tijare et al. (2016) reported no differences in shear energy between NORM and SEV fillets, regardless of bird age (6-week vs. 9-week), when utilizing the MORS method. In this study, increased shear values (SF and SE) in cooked WB were consistence to the increased hardness in both raw and cooked WB meat which indicates impaired product quality. Variation between shear values throughout the fillet could then be a potential issue as processors aim to provide a more uniform product to consumers. In addition, CS and CI had higher shear values than MS and MI suggesting that shear attributes were more extreme in the cranial region than in the middle region. However, inconsistent shear values have been observed in different studies suggesting that shear values for cooked fillets, affected by WB, are complicated in nature. Variation in results may be due to differences in methodology, bird age, sample portions (whole breast fillet vs portioned tissue), fillet region/location, or cooking method. Therefore, there is an urgent need to develop a nondestructive, standard methodology for assessing texture attributes of fillets affected by the WB condition.

Poor texture profile traits have been consistently reported for WB fillets. In the current experiment, higher values of hardness, chewiness, cohesiveness, gumminess, and resilience were observed in SEV fillets when compared to NORM fillets, which are in partial agreement with previous studies (Xing et al. 2020b; Chatterjee et al. 2016; Aguirre et al. 2018). On the other hand, Jarvis et al. (2020) reported that TPA values for the cranial portion of breast fillets were higher than the caudal portion of breast fillets. Furthermore, the current study indicates higher values in the CS, MS regions (combined as superficial/ventral portion) over those seen in the CI, MI regions (combined as internal/dorsal portion). These results are consistent with Bowker et al. (2018) who suggest that quality traits throughout fillets with WB are not uniform. These authors also suggest that characteristics are more severe on the ventral side than on the dorsal side of breast fillets. A potential explanation for this could be that myopathic lesions in WB are more evident on the ventral surface of breast fillets, with microscopic confirmation of an increase in connective tissue and collagen (Clark and Velleman 2016). Understanding the hardness distribution of cooked WB fillets could provide strategic information for fillet portioning or creating a trimming strategy for further processing in small particle products to reduce entire loss of affected fillets.

Within the raw state, SEV fillets had higher compositions of bound water and free water, and a decreased amount of immobilized water, when compared to NORM breast meat cubes. Soglia et al. (2016b) reported that WB fillets had an increased proportion and mobility of extramyofibrillar water (free water) compared to NORM fillets. Tasoniero et al. (2017) and Dalgaard et al. (2018) found similar results, as a higher content of extra-myofibrillar water and a lower proportion of intra-myofibrillar water were present in the cranial and middle region of SEV fillets when compared to NORM fillets. Both findings are in agreeance with the data presented in the current study. Furthermore, Pang et al. (2020b) reported that the WB condition has a significant impact on the distribution of water in intact muscle tissue with more extra-myofibrillar water being present in WB than in NORM breast fillets. The current data further shows that a right shifting T_{21} peak increases with WB severity for raw meat. This shift suggests that immobilized water has higher fluidity/mobility in affected raw meat than in NORM meat, and that water distribution in raw meat is more rapidly converted to free water (T_{22}) with increasing severity. The left shifting T_{21} peaks for all muscle conditions after cooking indicates that the degree of freedom in muscle moisture decreases, and that the binding ability of bound water and protein are enhanced. MILD fillets had a lower T_{2b} peak than SEV, which may explain why bound water in MILD fillets is more easily converted to immobilized water. Furthermore, in cooked meat samples, increasing peak width further reveals that the degree of freedom in muscle moisture was higher for WB fillets than in NORM fillets. This indicates that cooked WB has the possibility of losing more water after cooking and/or during storage than unaffected fillets. The results of this study indicate that WB severity could potentially be identified by water composition (bound water, immobilized water, and free water) in the raw meat state. A higher water content in WB meat may explain the higher drip loss generally exhibited, as well as the higher cook loss generally expressed during thermal processing.

Conclusion

In summary, the results from this experiment expressed that the WB condition impaired meat quality attributes such as an abnormal increase in compression force and energy values, as well as cook loss levels in affected fillets. The drip loss levels along with the bound water, and free water were higher in WB fillets compared to NORM fillets. The findings in the present study provide a deeper understanding of the abnormal hardness distribution in breast fillets affected by the WB condition. Higher compression measurements were observed in the cranial region of breast fillets, which then decreased from the cranial region to middle and/or caudal region for both the raw and cooked meat state. Affected WB fillets produced a reduction in meat quality with impaired shear values and TPA measurements. The cranial region produced higher shear values than all other regions, and TPA values were substantially higher in the superficial region of breast fillets. The outcomes of this study provide important information for processors to have a better understanding of meat quality traits and water properties throughout affected WB fillet, which could be used to strategically portion fillets in further processing operations in an attempt to mitigate significant economic losses caused by WB. Future research should focus on developing portioning methodologies to appropriately utilize fillets affected by WB.

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Declarations

Conflict of interest The authors declare no conflict of interest.

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