ABSTRACT The purpose of this study was to investigate the relationships between muscle water properties, water-holding capacity (WHC), and woody breast (WB) severity in intact raw broiler breast fillets. Broiler pectoralis major deboned at 3 h postmortem was collected from a commercial plant and categorized as normal (NORM), moderate WB, or severe WB (SEV). Meat drip loss was calculated based on weight loss during overnight storage at 4°C. Water properties of the intact fillets were determined with time domain nuclear magnetic resonance and the T2 relaxation times were determined using an inverse Laplace algorithm (CONTIN). Three T2 water components, hydration water (T2b), intra-myo-fibrillar water (T21), and extra-myo-fibrillar water (T22), were identified. With increasing WB severity, the time constant of each water component and the relative content of T22 (P22) increased while the relative areas of T2b and T21 (P2b and P21, respectively) decreased. Spearman correlation analysis showed that there were significant correlations between the WB condition score and either the time constant or normalized area for each T2 component. T22 normalized areas (A22) were most strongly correlated with the WB score (r = 0.75); however, the weakest correlation was found between the WB score and T21 areas (A21). Pearson correlation analysis revealed that the strongest correlation (r = 0.64) was found between A22 and drip loss; however, there was no correlation between A21 and drip loss. Within the NORM group, drip loss was significantly correlated to the time constants for both T2b and T21. Within the SEV group, only A22 was significantly correlated to drip loss. These data indicate that the WB condition has a significant impact on the distribution of water within the intact muscle tissue. The content of extra-myo-fibrillar water in broiler breast fillets may be a key factor responsible for the poor WHC measurements in WB meat.

Key words: drip loss, myofibrillar water, NMR T2 relaxation, pectoralis major, water-holding capacity

INTRODUCTION

The woody breast (WB) condition is an emerging myopathy in broiler chickens. Woody breast meat is characterized by palpable hardness and rigidity throughout the raw pectoralis major (Siervo et al., 2014; Mudalal et al., 2015) and the exact etiology leading to this defect is not well understood (Petracci et al., 2019). The WB condition has been estimated to cost the industry millions of dollars annually in the United States due to its impacts on meat quality (Kuttappan et al. 2016). In a review, Petracci et al. (2019) pointed out that the characterization of WB meat quality profile and the underlying mechanisms of impaired traits are essential to minimize the negative influence of the myopathy on quality of the end-use chicken products and consumer acceptability.

In addition to having unique texture properties in the raw state, WB meat has also been shown to have altered moisture characteristics and water-holding capacity (WHC)/binding capacity when compared to normal (NORM) breast meat. WB fillets exhibit greater
water/moisture content than NORM fillets (Soglia et al., 2016a, 2016b). During refrigerated storage the WB condition resulted in increased moisture losses compared to NORM fillets (Mudalal et al., 2015; Soglia et al., 2016b; Kuttappan et al., 2017; Bowker et al., 2018; Dalgaard et al., 2018; and Sun et al., 2018). In further processing, WB meat showed poor emulsifying and gelling abilities (Xing et al., 2017; Chen et al., 2018; Petracci et al., 2019) and cook loss of intact broiler fillets (to remove skin, connective tissue, bone, and mucoid exudate) and weighed. Fillets were scored with a 3-point scale in which, 1 = NORM, 2 = moderate WB (MOD), and 3 = severe WB (SEV) based on palpable hardness and rigidity according to the criteria used by Chatterjee et al. (2016) and Bowker and Zhuang (2019). Fillets were also scored for white striping (WS) with a 3-point scale based on the prevalence and thickness of white striations on the ventral (skin-side) surface of the muscle (Kuttappan et al., 2012). A total of 144 fillets, 48 for each category, were selected for this study.

Breast fillet weight, pH, and color were measured at approximately 6 h postmortem (Zhuang and Bowker, 2018). Muscle pH was measured in the cranial end of each fillet using a Hach H280 GB pH meter equipped with a spear tip probe (EW-5998-20, Cole-Parmer, Vernon Hills, IL). Raw color values (CIE L*a*b*) were measured on the dorsal surface (bone side) of each fillet using a Minolta spectrophotometer (VTL CM-700, Konica Minolta, Ramsey, NJ). The spectrophotometer was calibrated by following the manufacturer’s instruction for zero (directing the specimen measuring port to midair) and white (directing the port to the white calibration cap) before measurements. Compression force was measured on raw intact breast fillets using a 12-mm diameter probe with a 50 kg loading cell on a texture analyzer (model TA-XT-Plus, Texture Technologies Corp., Hamilton, MA). The trigger force was set at 20 g, and the test speed of the probe was 1 mm/s. For each fillet, 3 compression force measurements were conducted on the middle portion of the ventral side of the muscle. Data for compression force measurements were calculated based on 30% of the fillet height. Similar to the procedure of Sun et al. (2018), the fillets were placed in Ziploc bags and stored at 4°C overnight before fillet weight was measured again (at about 24 h postmortem) for calculating drip loss as a measure of WHC (den Hertog-Meischke et al., 1997). Drip loss was expressed as the percentage of weight change in the breast fillet before and after storage.

**TD-NMR Measurements**

A 1H-NMR analyzer (Bruker LF90 Proton-NMR Minispec, Bruker Optics, The Woodlands, TX) was used for TD-NMR measurements of muscle water properties in raw intact fillets. Transverse relaxation data (T2) were measured using the Carr-Purcell-Meiboom-Gill pulse sequence (Carr and Purcell, 1954; Meiboom and Gill, 1958) with a τ-value (90°–180° pulse separation) of 1 ms. For each fillet sample, whose temperature at

**MATERIALS AND METHODS**

**Sample Preparation and Physical Measurements**

Boneless and skinless breast fillets (more than 200 and prescreened) were collected from the deboning line of a commercial processing plant for the large birds at approximately 3 h postmortem. Broiler birds (Ross Line 308) were slaughtered at 8 to 9 wk and processed following the standard operation procedure for retail broiler meat products in the United States, including electrical stunning, soft scalding, immersion chill, automatic evisceration, and hand deboning. Fillets were placed in plastic bags on ice and transported to the U.S. National Poultry Research Center (45 min) where they were trimmed (to remove skin, connective tissue, bone, and mucoid exudate) and weighed. Fillets were scored with a 3-point scale in which, 1 = NORM, 2 = moderate WB (MOD), and 3 = severe WB (SEV) based on palpable hardness and rigidity according to the criteria used by Chatterjee et al. (2016) and Bowker and Zhuang (2019). Fillets were also scored for white striping (WS) with a 3-point scale based on the prevalence and thickness of white striations on the ventral (skin-side) surface of the muscle (Kuttappan et al., 2012). A total of 144 fillets, 48 for each category, were selected for this study.

Breast fillet weight, pH, and color were measured at approximately 6 h postmortem (Zhuang and Bowker, 2018). Muscle pH was measured in the cranial end of each fillet using a Hach H280 GB pH meter equipped with a spear tip probe (EW-5998-20, Cole-Parmer, Vernon Hills, IL). Raw color values (CIE L*a*b*) were measured on the dorsal surface (bone side) of each fillet using a Minolta spectrophotometer (VTL CM-700, Konica Minolta, Ramsey, NJ). The spectrophotometer was calibrated by following the manufacturer’s instruction for zero (directing the specimen measuring port to midair) and white (directing the port to the white calibration cap) before measurements. Compression force was measured on raw intact breast fillets using a 12-mm diameter probe with a 50 kg loading cell on a texture analyzer (model TA-XT-Plus, Texture Technologies Corp., Hamilton, MA). The trigger force was set at 20 g, and the test speed of the probe was 1 mm/s. For each fillet, 3 compression force measurements were conducted on the middle portion of the ventral side of the muscle. Data for compression force measurements were calculated based on 30% of the fillet height. Similar to the procedure of Sun et al. (2018), the fillets were placed in Ziploc bags and stored at 4°C overnight before fillet weight was measured again (at about 24 h postmortem) for calculating drip loss as a measure of WHC (den Hertog-Meischke et al., 1997). Drip loss was expressed as the percentage of weight change in the breast fillet before and after storage.

**TD-NMR Measurements**

A 1H-NMR analyzer (Bruker LF90 Proton-NMR Minispec, Bruker Optics, The Woodlands, TX) was used for TD-NMR measurements of muscle water properties in raw intact fillets. Transverse relaxation data (T2) were measured using the Carr-Purcell-Meiboom-Gill pulse sequence (Carr and Purcell, 1954; Meiboom and Gill, 1958) with a τ-value (90°–180° pulse separation) of 1 ms. For each fillet sample, whose temperature at
measurements was approximately 4°C, 16 scans were acquired with a total number of 200 echoes. The decay curves were analyzed with the CONTIN regularization algorithm (Provencher, 1982) provided by Minispec (contin-lit_mq_nf, Bruker Biospin GmbH, Rheinstetten, Germany), resulting in the corresponding distributions of the relaxation times and T2 parameters, hydration water (T2b), intra-myoﬁbrillar water (T21), extra-myoﬁbrillar water (T22), and their time constants, relative proportions, and normalized areas per 100 g of raw meat. The normalized area of T2 parameters was calculated by the following equation:

Normalized area of T2x = 100 g × [the area of a T2 peak/ intact ﬁllet weight (g)].

Like the other peak areas in chemical analysis, the normalized area of the T2 parameter here is proportional to amount of water that is present in meat or indicates the amount of water that is present in meat.

Statistical Analysis

Data (including raw meat characteristics and water properties) were subjected to a one-way ANOVA using the PROC MIXED procedure of SAS (version 9.4, SAS Institute Inc., Cary, NC). Significant differences (P < 0.05) between means were identiﬁed using a Tukey’s mean separation method. The relationships between WB score and water properties were analyzed by calculating Spearman correlation coefﬁcients (r) using the PROC CORR procedure of SAS. Similarly, the relationships between drip loss values and water properties were analyzed with Pearson correlation coefﬁcients (r). For the purposes of discussion, in this study the following descriptors were used to describe the relative strength of the correlations: very weak (r < 0.20), weak (r = 0.20–0.39), moderate (r = 0.40–0.59), strong (r = 0.60–0.79), and very strong (r = 0.80–0.99) (Bowker and Zhuang, 2019).

RESULTS

Fillet Quality Attributes

Table 1 shows that raw intact ﬁllets with the WB (MOD or SEV) condition had greater average weight, drip loss, pH, and WS score compared with NORM ﬁllets (P < 0.05). There were no differences in weight, drip loss, and pH value between MOD and SEV samples (P > 0.05). SEV ﬁllet samples had greater compression force than NORM samples (P < 0.05). For CIE L*a*b* color measurements in this study, greater a* values in SEV ﬁllets and greater b* values in SEV and MOD ﬁllets were observed compared with NORM ﬁllets (P < 0.05), and there were no differences in L* values among the 3 WB conditions (P > 0.05).

TD-NMR Parameters

Figure 1 shows the representative distribution of T2 relaxation time collected from intact ﬁllets with CONTIN regularization analysis, where 3 distinct water components were identiﬁed despite the WB condition. Table 2 shows the average time constants (T2b, T21, and T22), relative areas or proportion (P2b, P21, and P22), and normalized areas (A2b, A21, and A22, normalized to 100 g of meat) of the 3 T2 components. The time constant of T2b in MOD and SEV ﬁllets was similar (0.43 and 0.44 ms, respectively) (P > 0.05), and greater than that in NORM ﬁllets (0.39 ms) (P < 0.05). The time constants of T21 and T22 showed signiﬁcant increases with the severity of the WB conditions. The proportions of T2b or P2b in intact broiler breast meat was less than 1% of the total water. In MOD and SEV ﬁllets, P2b was similar (0.40 and 0.35%, respectively). However, P2b was greater in NORM ﬁllets (0.79%) than WB ﬁllets (P < 0.05). The proportion of T21 or P21 was more than 60% in intact broiler breast meat and was different between the 3 WB conditions (P < 0.05). P21 in NORM ﬁllets (81%) was greater than both MOD (65%) and SEV (60%) ﬁllets (P < 0.05). The effects of WB on P22 were opposite to those on P21. P22 was greater in MOD (34%) and SEV (39%) ﬁllets than that in NORM ﬁllets (18%) (P < 0.05). The normalized area of T2b or A2b in MOD and SEV ﬁllets (2.41 and 2.25 unit/100 g meat) was less than that in NORM ﬁllets (3.65 unit/100 g meat) (P < 0.05), whereas A21 was just the opposite. A21 was greater in MOD and SEV ﬁllets (407.21 unit/100 g meat) than that in NORM ﬁllets (385.21 unit/100 g meat) (P < 0.05). A22 showed signiﬁcant increases with the severity of the WB condition. A22 was greater in MOD (223.40 unit/100 g meat) and SEV

Table 1. Raw meat characteristics of broiler breast ﬁllets with the WB condition (mean ± SD, n = 48).

| Parameter | NORM | MOD | SEV |
|-----------|------|-----|-----|
| Weight (g) | 461 ± 64 | 571 ± 58 | 555 ± 55 |
| Drip loss (%) | 1.14 ± 0.53 | 1.64 ± 0.77 | 1.84 ± 0.71 |
| pH | 5.98 ± 0.13 | 6.14 ± 0.18 | 6.11 ± 0.15 |
| L* (dorsal) | 60.5 ± 1.9 | 62.2 ± 2.6 | 60.7 ± 2.5 |
| a* (dorsal) | -0.8 ± 0.5 | -0.1 ± 0.8 | 0.4 ± 0.9 |
| b* (dorsal) | 11.4 ± 1.2 | 13.8 ± 1.7 | 14.7 ± 1.6 |
| WS score | 1.28 ± 0.34 | 1.85 ± 0.44 | 2.33 ± 0.58 |
| Compression force (N) | 19.7 ± 4.4 | 26.9 ± 6.0 | 36.4 ± 9.8 |

Table 2 shows that the average time constants (T2b, T21, and T22), relative areas or proportion (P2b, P21, and P22), and normalized areas (A2b, A21, and A22, normalized to 100 g of meat) of the 3 T2 components. The time constant of T2b in MOD and SEV ﬁllets was similar (0.43 and 0.44 ms, respectively) (P > 0.05), and greater than that in NORM ﬁllets (0.39 ms) (P < 0.05). The time constants of T21 and T22 showed significant increases with the severity of the WB conditions. The proportion of T2b or P2b in intact broiler breast meat was less than 1% of the total water. In MOD and SEV ﬁllets, P2b was similar (0.40 and 0.35%, respectively). However, P2b was greater in NORM ﬁllets (0.79%) than WB ﬁllets (P < 0.05). The proportion of T21 or P21 was more than 60% in intact broiler breast meat and was different between the 3 WB conditions (P < 0.05). P21 in NORM ﬁllets (81%) was greater than both MOD (65%) and SEV (60%) ﬁllets (P < 0.05). The effects of WB on P22 were opposite to those on P21. P22 was greater in MOD (34%) and SEV (39%) ﬁllets than that in NORM ﬁllets (18%) (P < 0.05). The normalized area of T2b or A2b in MOD and SEV ﬁllets (2.41 and 2.25 unit/100 g meat) was less than that in NORM ﬁllets (3.65 unit/100 g meat) (P < 0.05), whereas A21 was just the opposite. A21 was greater in MOD and SEV ﬁllets (407.21 unit/100 g meat) than that in NORM ﬁllets (385.21 unit/100 g meat) (P < 0.05). A22 showed significant increases with the severity of the WB condition. A22 was greater in MOD (223.40 unit/100 g meat) and SEV

Figure 1. Representative distribution of T2 relaxation time for normal breast (NORM), moderate woody breast (MOD), and severe woody breast (SEV) ﬁllets based on CONTIN regularization analysis.
(273.36 unit/100 g meat) fillets than that in NORM fillets (88.80 unit/100 g meat) (P < 0.05).

**Relationships Between the WB Condition and Water Properties**

Table 3 shows the Spearman correlations between the WB score and the water property parameters. There were significant Spearman correlations (P < 0.001) between WB scores and the time constants of T2b, T21, and T22, and their corresponding normalized areas A2b, A21, and A22, which were 0.52, 0.56, 0.58, 0.29, and 0.75, respectively. Table 4 shows the Pearson correlations between drip loss or WHC and water property parameters. With the exception of A21, there were significant Pearson correlations (P < 0.001) between drip loss and the time constants of T2b, T21, and T22, and the normalized areas A2b and A22, which were 0.38, 0.45, 0.38, −0.40, and 0.64, respectively. Table 5 shows the Pearson correlations between drip loss and water properties within each WB condition. For time constants, significant Pearson correlations between drip loss and time constants of T2b and T22 were only observed in NORM fillets. For the normalized areas, there were significant correlations between drip loss and normalized areas for all T2 components in NORM fillets. However, in SEV fillets there was only a significant relationship between drip loss and A22. For MOD fillets, significant relationships were noted between drip loss and A21 or A22. In addition, A21 was positively correlated with purge loss in NORM samples (r = 0.38); however, it was negatively correlated with purge loss in MOD samples (r = −0.29).

**DISCUSSION**

The raw meat characteristics of broiler breast fillets with the WB condition (Table 1) are well in agreement with data published in previous reports. The average weights of WB fillets were more than that of NORM fillets (Mudalal et al., 2015; Chatterjee et al., 2016; Dalle Zotte et al., 2017; Dalgaaard et al., 2018; Sun et al., 2018). The WS scores were positively related to the WB condition (Bowker and Zhuang, 2019). Many studies observed greater drip loss (Mudalal et al., 2015; Soglia et al., 2016b; Kuttappan et al., 2017; Bowker et al., 2018; Sun et al., 2018) and pH values (Mudalal et al., 2015; Brambila et al., 2017; Dalle Zotte et al., 2017; Kuttappan et al., 2017; Bowker et al., 2018; Zhuang and Bowker, 2018; Baldi et al., 2019) in WB meat compared with the NORM ones. So far the CIE L*a*b* color data reported in literature have not shown consistent relationships between the WB condition and CIE L*a*b* color measurements on the dorsal side of fillets. Trocino et al. (2015) and Brambila et al. (2017) found no significant differences between WB fillets and NORM fillets regardless of the CIE L*a*b* parameters. However, Chatterjee et al. (2016) and Zhuang and Bowker (2018) showed the difference in a* value. Mudalal et al. (2015) found the difference in b* value. Baldi et al. (2019) reported differences in L* as well as

**Table 2. Effect of the WB condition on water properties time constants (T), relative areas (P), and normalized areas (A) (mean ± SD, n = 48).**

| Parameter | NORM | MOD | SEV |
|-----------|------|-----|-----|
| T2b       | 0.39 ± 0.03b | 0.43 ± 0.03a | 0.44 ± 0.02b |
| T21       | 49.04 ± 2.911 | 54.74 ± 4.20b | 57.21 ± 4.02a |
| T22       | 204.88 ± 19.24c | 218.20 ± 13.60b | 234.64 ± 22.54a |
| Relative area (%) |      |      |     |
| P2b       | 0.79 ± 0.25a | 0.40 ± 0.18b | 0.35 ± 0.14c |
| P21       | 81.02 ± 3.93c | 65.48 ± 8.63c | 60.42 ± 7.55c |
| P22       | 18.18 ± 4.08c | 34.18 ± 8.74c | 39.23 ± 7.62c |
| Normalized area (unit/100 g meat) |      |      |     |
| A2b       | 3.65 ± 0.95c | 2.41 ± 0.75b | 2.25 ± 0.53d |
| A21       | 385.21 ± 26.44b | 407.63 ± 34.15a | 404.47 ± 51.13a |
| A22       | 88.80 ± 31.06c | 223.40 ± 90.02b | 273.36 ± 95.82a |

* *Means within a row lacking a common superscript differ (P < 0.05).
Abbreviations: MOD, moderate woody breast; NORM, normal breast; SEV, severe woody breast; WB, woody breast.

**Table 3. Spearman correlations between WB score (1 = NORM, 2 = MOD, 3 = SEV) and water properties time constants (T) or normalized areas (A).**

| Parameter | T2b  | T21  | T22  | A2b  | A21  | A22  |
|-----------|------|------|------|------|------|------|
| WB score  | 0.52*** | 0.68*** | 0.56*** | −0.58*** | 0.29*** | 0.75*** |

***P < 0.001.
Abbreviations: MOD, moderate woody breast; NORM, normal breast; SEV, severe woody breast; WB, woody breast.
component with a T2 relaxation time constant around 0 to 15 ms is the fastest relaxing component in T2. It has been named T2b, and is hypothesized to reflect hydration water, or water tightly associated to the macromolecules in meat. The water component with a T2 relaxation time constant around 40 to 80 ms is named T21 and is hypothesized to reflect the intra-myofibrillar water located within the highly organized myofibrillar lattice space. The slowest relaxing component in T2 is named T22 and presumably corresponds to loosely bound extra-myofibrillar water (Bertram et al., 2002b; Hullberg and Bertram, 2005; Tasoniero et al., 2017; Chen et al., 2018). The TD-NMR results collected from intact fillets in this study (Figure 1) reveal a relaxation pattern very similar to that reported in previously published studies collected with raw chicken meat. Three distinct water components (T2b, T21, and T22), from left to right in the T2 relaxation time distribution, were identified in raw intact broiler breast fillets. Both their time constants and amplitudes appeared to be influenced by the severity of the WB condition.

The average time constant of T2b, ranged from 0.39 to 0.44 ms, the average time constant of T21 from 49.04 to 57.21 ms, and the average time constant of T22 from 204.88 to 234.64 ms (Table 2). These results are similar to those reported earlier with fresh pork meat (Bertram et al., 2002a, 2002b) and chicken breast meat (Petracci et al., 2012; Soglia et al., 2016a; Xing et al., 2017), where the time constants of T2b, T21, and T22 were found centered around 0 to 15, 40 to 80, and 100 to 400 ms, respectively. The differences in T2 constants between our results and those published by Tasoniero et al. (2017) for T21 and T22 might have resulted from the differences in instrumentation, data processing methods, and/or use of intact vs. excised samples. Bertram et al. (2001) also noted differences in T2 time constants and areas among the published literature and attributed them to magnetic equipment, NMR parameters employed, such as repetition time between succeeding scans and the τ-value (time between 90° pulse and 180° pulse), sample temperature, and postmortem time when meat samples were measured.

Our data further showed that with increases in the severity of the WB condition, the means for all T2 time constants in WB fillets were significantly greater (P < 0.05) than those in NORM fillets (Table 2), indicating that the mobility of water in WB fillets is greater than that in NORM fillets. Increased T2 time constants were also observed in the WB muscle by Soglia et al. (2016a) and Tasoniero et al. (2017). Differences (P < 0.05) between NORM and WB samples were also found in both the relative (P) and normalized areas (A) of the 3 water components (Table 2). It should be noted that when data were expressed as the normalized area per 100 g of raw meat, the effects of WB on A2b and A22 were similar to when data were expressed as the relative area P2b and P22. For A21, however, expressing the data as normalized area showed that WB meat contained more intra-myofibrillar water (>400 unit/100 g of meat) than NORM (<390 unit/100 g of meat).

### Table 4. Pearson correlations between drip loss and water properties time constants (T) or normalized areas (A).

| Parameter | T2b | T21 | T22 | A2b | A21 | A22 |
|-----------|-----|-----|-----|-----|-----|-----|
| Drip loss | 0.38*** | 0.45*** | 0.38*** | −0.40*** | −0.04 | 0.64*** |

***P ≤ 0.001.

### Table 5. Pearson correlations between drip loss and water properties time constants (T) or normalized areas (A) within the WB condition.

| Parameter | NORM (n = 48) | MOD (n = 48) | SEV (n = 48) |
|-----------|---------------|--------------|--------------|
| Time constant (ms) | Drip loss (%) | Drip loss (%) | Drip loss (%) |
| T2b | 0.40** | 0.20 | −0.07 |
| T21 | 0.49*** | 0.21 | 0.18 |
| T22 | 0.19 | 0.19 | 0.28 |
| Normalized area (unit/100 g meat) | Drip loss (%) | Drip loss (%) | Drip loss (%) |
| A2b | −0.32* | −0.16 | −0.19 |
| A21 | 0.38*** | −0.29* | −0.24 |
| A22 | 0.60*** | 0.61*** | 0.50*** |

*P ≤ 0.05, **P ≤ 0.01, ***P ≤ 0.001.

Abbreviations: MOD, moderate woody breast; NORM, normal breast; SEV, severe woody breast; WB, woody breast.
100 g of meat). Expressing T22 data as normalized area (A22) also suggested that the SEV WB fillets contained approximately 3 times more extra-myoﬁbrillar water than NORM fillets. Using much smaller meat samples (<5 g), Sogila et al. (2016a) and Tasoniero et al. (2017) found that P2b (hydration) and P21 (intra-myoﬁbrillar) in WB meat were signiﬁcantly lower than those in NORM meat; however, P21 (extra-myoﬁbrillar) in breast meat with the WB condition was more than 2 times greater than that in NORM. Data in the current study are consistent with published data (Tasoniero et al., 2017; Chen et al., 2018) and further demonstrate that WB meat contains a greater proportion of extra-myoﬁbrillar water and lower proportion of intra-myoﬁbrillar and hydration water than NORM meat. Furthermore, by expressing data as a function of ﬁlet weight (unit/100 g meat), data in this study suggest that WB meat has a greater abundance of both intra- and extra-myoﬁbrillar water compared to NORM meat. This is likely due to both the overall larger size of the WB ﬁllets compared to the NORM ﬁllets as well as the fact that WB meat has a higher moisture content than NORM meat (Petracci et al., 2019).

There were signiﬁcant Spearman correlations (P < 0.001) between WB scores and water properties regardless of the T2 component (T2b, T21, or T22) or expression of T2 components (time constant or normalized area) (Table 3). These results indicate that water properties such as water mobility and the normalized amount in muscle are associated with the severity of the WB condition in broiler breast muscle. Among the time constants, the strongest correlation was found for T21 (r = 0.68), followed by T22 (r = 0.56), and they were all positive. These data suggest that water mobility or water activity may have inﬂuences on physical and technological properties of broiler breast meat with the WB myopathy. There were also signiﬁcant positive relationships (P < 0.001) between WB scores and either A21 or A22 (Table 3). However, the strength of the correlation between A22 and WB scores was approximately 2.5 times higher than between A21 and WB scores and explained more than 50% of the experimental variation. There was also a moderate and negative correlation between WB scores and A2b. These data demonstrate that the abundance of extra-myoﬁbrillar water is more closely associated with the severity of WB meat. These results also suggest that the greater overall water content and higher amount of extra-myoﬁbrillar water in WB ﬁllets might be at least partially responsible for the physical properties, such as hardness and rigidity, used for scoring WB severity in raw broiler breast meat. Using NMR, Tasoniero et al. (2017) also suggested a connection between the increased muscle hardness observed in WB meat and the longer relaxation times of water trapped within the myoﬁbrillar matrix.

The relationships between the measured water properties and drip loss are shown in Table 4. With the exception of A21, the water property parameters were signiﬁcantly correlated to drip loss. The strongest correlation was found between drip loss and A22 (r = 0.64). Moderate correlations were found between drip loss and A2b (r = −0.40, P < 0.001) or T21 (r = 0.45, P < 0.001), whereas, weak correlations were found between drip loss and either T2b or T22 (r = 0.38, P < 0.001). These results indicate that changes in WHC, measured with drip loss, of broiler breast meat with the WB condition may be more likely determined by extra-myoﬁbrillar water content.

The correlations between TD-NMR measurements and drip loss were also analyzed within each WB condition to determine if these relationships were inﬂuenced by the WB modulation. The results suggested that the relationship between drip loss and the various water component measurements varies with the WB condition (Table 5). Similarly, Bertram et al. (2001) found that there were highly signiﬁcant correlations (r > |0.70|) between WHC and either T21 or T22 regardless of the method used for WHC analysis in pork. Bertram et al. (2002b) further demonstrated a strong correlation between the T22 water component and drip loss in pork and concluded that meat WHC was mainly determined by the amount of loosely bound extra-myoﬁbrillar water in the muscle and that the formation of drip loss in NORM pork meat was an ongoing process involving the transfer of water from within myoﬁbrils to the extracellular space. Data in the current study suggest that the mobility of extra-myoﬁbrillar water does not have a signiﬁcant impact on drip loss in broiler breast meat regardless of the WB condition (Table 5). In NORM broiler breast meat, however, drip loss was signiﬁcantly and positively correlated to A21 and A22, which suggests that drip loss is an ongoing process involving the transfer of water from intra-myoﬁbrillar water components to the extracellular space similar to NORM pork. However, drip loss in SEV WB meat is more likely determined by only the amount of loosely bound extra-myoﬁbrillar water. In meat with the MOD WB condition, the abundance of intra-myoﬁbrillar water, in addition to extra-myoﬁbrillar water, may also be involved in its drip loss over refrigerated storage.

**CONCLUSIONS**

TD-NMR has been used to analyze differences in the water properties between NORM and WB meat using whole intact fillets. Experimental data demonstrate that, in agreement with earlier NMR data collected on small muscle subsamples or ground meat, 3 distinct water components (hydration water, intra-myoﬁbrillar water, and extra-myoﬁbrillar water) were identiﬁed in raw intact broiler breast ﬁllets. In WB meat, water mobility is greater regardless of the water component as evidenced by a shift of T2x values to longer times, and there is more relative extra-myoﬁbrillar water and less relative intra-myoﬁbrillar water and hydration water. When expressed as normalized area/100 g of tissue, the abundance of both intra- and extra-myoﬁbrillar water was greater in WB meat compared to NORM breast meat. Data also suggest that greater extra-myoﬁbrillar water...
content and intra-myofibrillar water mobility may be responsible for the physical and technological properties of raw WB meat, although this remains to be demonstrated with further studies on their relationships. Among the 3 T2 components in WB meat, the T22 normalized area A22 exhibited the greatest correlation with drip loss indicating that extra-myofibrillar water likely plays a key role in the poor WHC of WB meat. Furthermore, this study reveals that, in addition to the amount of extra-myofibrillar water, both water mobility and the amounts of both hydration water and intra-myofibrillar water affect drip loss in NORM broiler breast meat. However, drip loss may be determined primarily by the amount of extra-myofibrillar water in WB meat.

Conflict of Interest Statement: The authors did not provide a conflict of interest statement.

REFERENCES

Baldi, G., F. Soglia, L. Laghi, S. Tappi, P. Rocculi, S. Tavaniello, D. Prioriello, R. Mucci, G. Maiorano, and M. Petracci. 2019. Comparison of quality traits among breast meat affected by current muscle abnormalities. Food Res. Int. 115:369–376.

Bertram, H. C., H. J. Andersen, and A. H. Karlsson. 2001. Comparative study of low-field NMR relaxation measurements and two traditional methods in the determination of water holding capacity of pork. Meat Sci. 57:125–132.

Bertram, H. C., S. Deenstrup, A. H. Karlsson, and H. J. Andersen. 2002a. Continuous distribution analysis of T2 relaxation in meat—an approach in the determination of water holding capacity. Meat Sci. 60:279–285.

Bertram, H. C., P. P. Purslow, and H. J. Andersen. 2002b. Relationship between meat structure, water mobility, and distribution: a low-field nuclear magnetic resonance study. J. Agric. Food Chem. 50:824–829.

Bianchi, M., F. Capozzi, M. A. Cremonini, L. Laghi, M. Petracci, G. Placucci, and C. Cavani. 2004. Influence of the season on the relationships between NMR transverse relaxation data and water-holding capacity of Turkey. J. Sci. Food Agric. 84:1535–1540.

Bowker, B. C., A. D. Maxwell, H. Zhuang, and K. Adhikari. 2018. Marination and cooking performance of portioned broiler breast fillets with the wooden breast condition. Poult. Sci. 97:2966–2970.

Bowker, B., and H. Zhuang. 2019. Detection of razor shear force differences in broiler breast meat due to the woody breast condition depends on measurement technique and meat state1. Poult. Sci. 98:6170–6176.

Brambilla, G. S., D. Chatterjee, B. Bowker, and H. Zhuang. 2017. Descriptive texture analyses of cooked patties made of chicken breast with the woody breast condition. Poult. Sci. 96:3489–3494.

Brambilla, G. S., B. C. Bowker, D. Chatterjee, and H. Zhuang. 2018. Descriptive texture analyses of broiler breast fillets with the wooden breast condition stored at 4°C and −20°C. Poult. Sci. 97:1762–1767.

Brown, R. J. S., F. Capozzi, C. Cavani, M. A. Cremonini, M. Petracci, and G. Placucci. 2000. Relationships between 1H NMR relaxation data and some technological parameters of meat: a Chemometric approach. J. Magn. Reson. 147:89–94.

Cai, K., W. Shao, X. Chen, Y. L. Campbell, M. N. Nair, S. P. Suman, C. M. Beach, M. C. Guyton, and M. W. Schilling. 2018. Meat quality traits and proteome profile of woody broiler breast (pectoralis major) meat. Poult. Sci. 97:337–346.

Carr, H. Y., and E. M. Purcell. 1954. Effects of diffusion on free precession in nuclear magnetic resonance experiments. Phys. Rev. 96:630.

Chatterjee, D., H. Zhuang, B. C. Bowker, A. M. Rincon, and G. Sanchez-Brambilla. 2016. Instrumental texture characteristics of broiler pectoralis major with the wooden breast condition. Poult. Sci. 95:2449–2454.

Chen, H., H. Wang, J. Qi, M. Wang, X. Xu, and G. Zhou. 2018. Chicken breed quality - normal, pale, soft and exudative (PSE) and woody - influences the functional properties of meat batters. Int. J. Food Sci. Technol. 53:654–664.

Dalgaard, L. B., M. K. Rasmussen, H. C. Bertram, J. A. Jensen, H. S. Møller, M. D. Auslyng, E. K. Højbjerg, J. R. Pedersen, D. Elsler Gravesen, and J. F. Young. 2018. Classification of wooden breast myopathy in chicken pectoralis major by a standardised method and association with conventional quality assessments. Int. J. Food Sci. Technol. 53:1744–1752.

Dalle Zotte, A., G. Tasoniero, E. Puolanne, H. Remignon, M. Cecchinato, E. Catelli, and M. Collere. 2017. Effect of “wooden breast” appearance on poultry meat quality, histological traits, and lesions characterization. Czech J. Anim. Sci. 62:51–57.

Den Hertog-Meischke, M. J. A., R. J. L. M. van Laack, and F. J. M. Smulders. 1997. The water-holding capacity of fresh meat. Vet. Q. 19:175–181.

Hullberg, A., and H. C. Bertram. 2005. Relationships between sensory perception and water distribution determined by low-field NMR T2 relaxation in processed pork-impact of tumbling and RN-allele. Meat Sci. 69:709–720.

Kuttappan, V. A., B. M. Hargis, and C. M. Owens. 2016. White striping and wooden breast myopathies in the modern poultry industry: a review. Poult. Sci. 95:2724–2733.

Kuttappan, V. A., C. M. Owens, C. Coon, B. M. Hargis, and M. Vazquez-Anon. 2017. Incidence of broiler breast myopathies at 2 different ages and its impact on selected raw meat quality parameters. Poult. Sci. 96:3095–3099.

Kuttappan, V. A., Y. Lee, G. F. Ehr, J. F. Meullenet, and C. M. Owens. 2012. Consumer acceptance of visual appearance of broiler breast meat with varying degrees of white striping. Poult. Sci. 91:1240–1247.

Meiboom, S., and D. Gill. 1958. Modified spin-echo method for measuring nuclear relaxation times. Rev. Sci. Instrum. 29:688–691.

Mudalal, S., M. Lorenzi, F. Soglia, C. Cavani, and M. Petracci. 2015. Implications of white striping and wooden breast abnormalities on quality traits of raw and marinated chicken meat. Animal 9:728–734.

Pang, B., B. Bowker, Y. Yang, J. Zhang, and H. Zhuang. 2020. Relationships between instrumental texture measurements and subjective woody breast condition scores in raw broiler breast fillets. Poult. Sci. 99:3292–3298.

Petracci, M., L. Laghi, P. Rocculi, S. Rimini, V. Panarese, M. A. Cremonini, and C. Cavani. 2012. The use of sodium bicarbonate for marination of broiler breast meat. Poult. Sci. 91:526–534.

Petracci, M., F. Soglia, M. Madruga, L. Carvalho, Elza Ida, and M. Estévez. 2019. Wooden-breast, white striping, and spaghetti meat: causes, consequences and consumer perception of emerging broiler meat abnormalities. Compr. Rev. Food Sci. F. 18:565–583.

Provencher, S. W. 1982. CONTIN: a general purpose constrained regularization program for inverting noisy linear algebraic and integral equations. Comput. Phys. Commun. 27:229–242.

Silvo, H., K. Immonen, and E. Puolanne. 2014. Myodegeneration with fibrosis and regeneration in the pectoralis major muscle of broilers. Vet. Pathol. 51:619–629.

Soglia, F., L. Laghi, L. Canonicoo, C. Cavani, and M. Petracci. 2016a. Functional property issues in broiler breast meat related to emerging muscle abnormalities. Food Res. Int. 89:1071–1076.

Soglia, F., S. Mudalal, E. Babini, M. Di Nunzio, M. Mazzoni, F. Sirri, C. Cavani, and M. Petracci. 2016b. Histology, composition, and quality traits of chicken Pectoralis major muscle affected by wooden breast. Poult. Sci. 95:651–659.

Sun, X., D. A. Koites, C. N. Coon, K. Chen, and C. M. Owens. 2018. Instrumental compression force and meat attribute changes in woody broiler breast fillets during short-term storage. Poult. Sci. 97:2600–2606.

Tasoniero, G., H. C. Bertram, J. F. Young, A. Dalle Zotte, and E. Puolanne. 2017. Relationship between hardness and myowater properties in Wooden Breast affected chicken meat: a nuclear magnetic resonance study. LWT 86:20–24.
Trocino, A., A. Piccirillo, M. Birolo, G. Radaelli, D. Bertotto, E. Filiou, M. Petracci, and G. Xiccato. 2015. Effect of genotype, gender and feed restriction on growth, meat quality and the occurrence of white striping and wooden breast in broiler chickens. Poult. Sci. 94:2996–3004.

Xing, T., X. Zhao, L. Cai, Z. Guanghong, and X. Xu. 2017. Effect of salt content on gelation of normal and wooden breast myopathy chicken pectoralis major meat batters. Int. J. Food Sci. Technol. 52:2068–2077.

Zambonelli, P., M. Zappaterra, F. Soglia, M. Petracci, F. Sirri, C. Cavani, and R. Davoli. 2016. Detection of differentially expressed genes in broiler pectoralis major muscle affected by White Striping - wooden Breast myopathies. Poult. Sci. 95:2771–2785.

Zhuang, H., and B. Bowker. 2018. The wooden breast condition results in surface discoloration of cooked broiler pectoralis major. Poult. Sci. 97:4458–4461.