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Food Chemistry 93 (2005) 337-348

Food Chemistry

www.elsevier.com/locate/foodchem

# Effect of heat treatment on changes in texture, structure and properties of Thai indigenous chicken muscle

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Received 19 April 2004; received in revised form 28 September 2004; accepted 28 September 2004

#### Abstract

Changes in texture, microstructure, colour and protein solubility of Thai indigenous and broiler chicken *Pectoralis* muscle stripes cooked at different temperatures were evaluated. The change in shear value of both chicken muscles was a significant increase from 50 to 80 °C but no change from 80 to 100 °C. A significant decrease in fibre diameter was obtained in samples heated to an internal temperature of 60 °C and the greatest shrinkage of sarcomeres was observed with internal temperatures of 70–100 and 80–100 °C for broiler and indigenous chicken muscles, respectively (P < 0.05). Cooking losses of indigenous chicken muscles increased markedly in the temperature range 80–100 °C and were significantly higher than those of the broiler (P < 0.001). With increasing temperature, from 50 to 70 °C, cooked chicken muscle became lighter and yellower. Relationships between changes in sarcomere length, fibre diameter, shear value, cooking loss and solubility of muscle proteins were evaluated. It was found that the solubility of muscle protein was very highly correlated with the texture of cooked broiler muscle while sarcomere length changes and collagen solubility were important factors influencing the cooking loss and texture of cooked indigenous chicken muscle. © 2004 Elsevier Ltd. All rights reserved.

Keywords: Heat treatment; Texture; Structure; Properties; Chicken muscle

# 1. Introduction

Thai indigenous chicken has a unique taste and texture and is very popular among consumers. Indigenous and broiler chickens are consumed at approximately the same commercial live weight. However, indigenous chickens generally have slower growth rates than the commercial broilers. The indigenous chicken muscles, both *Pectoralis m.* and *Biceps femoris m.*, possess firmer textures, particularly after cooking than those of the commercial broilers (Wattanachant, Benjakul, & Ledward, 2004). This may be related to the difference in total and soluble collagen contents between the muscles of the two chicken breeds (Wattanachant et al., 2004).

Thermal processing, in meat and poultry strongly, influences texture, protein changes, cooking yield, and other important quality factors, such as juiciness, colour, and flavour, which are associated with palatability and consumer acceptance of the final product. (Califano, Bertola, Bevilacqua, & Zaritzky, 1997; Murphy & Marks, 2000). The principal proteins responsible for meat texture include stromal (mostly collagen) and myofibrillar proteins (Califano et al., 1997; Dawson, Sheldon, & Miles, 1991). According to Bailey and Light (1989), during the cooking of meat, there is first an increase in toughness, between 40 and 50 °C, owing to the beginning of denaturation of myofibrillar proteins, a further increase between 60 and 70 °C, because of shrinkage of intramuscular collagen at 65 °C, and a third increase in the range of 70-90 °C when shrinkage and dehydration of the actomyosin occurs. However,

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<sup>0308-8146/</sup>\$ - see front matter © 2004 Elsevier Ltd. All rights reserved. doi:10.1016/j.foodchem.2004.09.032

Christensen, Purslow, and Lersen (2000) studied the effect of heating temperature on changes in whole meat, single muscle and perimysial connective tissue of beef semitendinosus muscle and found that the changes were divided into two phases. The initial rise in meat toughness was due to thermal shrinkage of intramuscular connective tissue at temperatures between 40 and 50 °C. The second rise in meat toughness could be due to heat denaturation of myofibrillar proteins at temperatures above 60 °C. The difference in results from Bailey and Light (1989) might be because Christensen et al. studied the single fibres. Palka and Daun (1999) found that tenderizing of beef semitendinosus muscle occurred when processed at the high temperature of 121 °C. In contrast, unacceptable effects on textural characteristics, including toughening, drying, and loss of particulate shape, were observed after high temperature processing of chicken breast meat (Dawson et al., 1991). The different results obtained from the different specie muscles may be caused by the difference in muscle type, structure and compositions, such as the content and nature of the intramuscular collagen. The increase in collagen content and collagen cross-linking in meat (often associated with older animals and specific muscle types) generally increases the toughness of cooked meat (Dawson et al., 1991; Rochdi, Foucat, & Renou, 2000). Changes in collagen solubility with heating temperature could affect the textural and water-binding properties of the product (Eilert & Mandigo, 1993).

Morphological changes of muscle tissue during heating have been established (Jones, Carroll, & Cavanaugh, 1977; Palka & Daun, 1999). Most previous reports have focussed on the effects of heat treatment on the structure of muscle fibres in relation to the texture of beef (Bertola, Bevilacqua, & Zaritzky, 1994; Yu & Lee, 1986; Zayas & Naewbanij, 1986). Effects of thermal processing on the texture of chicken (Dawson et al., 1991) and chicken products (Murphy & Marks, 2000) have been reported. However, there is no information regarding the effect of heating on the chicken commonly consumed in Thailand.

Therefore, the objective of this study was to evaluate changes in texture, microstructure and properties of Thai indigenous chicken and broiler *Pectoralis* muscle during heating in the range 50–100 °C.

# 2. Materials and methods

## 2.1. Sample preparation

Twenty Thai indigenous chickens (*Gallus domesticus*) aged 16 weeks and 20 commercial broilers (*CP707*) aged 38 days of similar live weights  $(1.5 \pm 0.2 \text{ kg})$ , obtained from a local farm in Songkhla Thailand, were killed by conventional neck cut, bled for 2 min, scalded at 60

°C for 2 min, plucked in a rotary-drum picker for 30 s and eviscerated. Pectoralis major muscles were dissected from the carcasses after chilling at 4 °C for 24 h. The skin was removed and the muscles were trimmed of obvious fat and connective tissue. Breast muscles (Pectoralis major) from both broiler and indigenous chickens were subjected to 24 h aging at 4 °C prior to further study. The muscles were cut to the size  $2.0 \times 2.0 \times 6.0$ cm. The muscle strips were individually weighed, packed into tightly sealed plastic bags, and stored at 4 °C for 24 h. Samples were heated in boiling water to the following internal temperatures: 50, 60, 70, 80, 90 and 100 °C. Copper-constantan thermocouples were used for temperature measurement of samples and water bath environment. Heat penetration time, to obtain the internal temperature designated, was recorded. After heating, the samples were chilled with cold water to about 10 °C and stored at 4 °C until analyzed.

#### 2.2. Cooking loss

Cooking losses were calculated from differences in the weight of raw and cooked muscle strips. The measurements were conducted in seven replications.

#### 2.3. Shear value

Muscle samples, raw and cooked, were cut to the size of  $1.0 \times 2.0 \times 0.5$  cm for shear analysis using the Texture Analyzer equipped with a Warner-Bratzler shear apparatus (Dawson et al., 1991). The operating parameters consisted of a cross head speed of 2 mm/s and a 25 kg load cell. The shear force, perpendicular to the axis of muscle fibres, was measured in 20 replicates for each treatment on both chicken breeds. The peak of the shear force profile was regarded as the shear force value.

# 2.4. Colour

The colour of muscle samples in the anterior and posterior locations was determined in seven replications of each sample (n = 14) using a Hunterlab colorimeter and reported as the complete International Commission on Illumination (CIE) system colour profile of  $L^*$ , redness  $(a^*)$ , and yellowness  $(b^*)$ .

#### 2.5. Microstructure of muscle

The microstructure of muscle was determined using a scanning electron microscope (SEM) according to the procedure of Palka and Daun (1999) with a slight modification. Pieces  $(1 \times 1 \times 0.5 \text{ cm})$  were excised from raw and cooked muscle samples and fixed in 2.5% glutaral-dehyde in 0.1 M phosphate buffer, pH 7.3, for 2 h at room temperature (26–28 °C). The specimens were then rinsed with distilled water and dehydrated in a serial

solution of 25%, 50%, 70%, 95% and absolute ethanol (twice), for 1 h in each solution. The samples were cut in liquid nitrogen using a razor blade. The fragments of dried specimens were mounted on aluminium stubs and coated with gold. The specimens were examined and photographed in a SEM, using an accelerating voltage of 5 or 10 kV. The micrographs and videoprints were taken at magnification of 500× (5 kV) for transverse sections and  $10,000 \times (10 \text{ kV})$  for longitudinal ones. The areas of muscle fibres and the length of sarcomeres were measured on videoprints, using a special morphometric facility. Four videoprints from each sample were taken for transverse sections and 10 measurements of fibre area on each were made (n = 40). The fibre diameter was calculated from the fibre area. Three videoprints from each sample were taken for longitudinal sections and 10 measurements of sarcomere length on each were made (n = 30).

#### 2.6. Solubility of proteins

Solubility of proteins in the raw and heated samples was determined in different solvents according to the procedure of Roussel and Cheftel (1990) with some modifications. Solvents used included: 0.6 M KCl (S1); 20 mM Tris, pH 8.0 (S2); 20 mM Tris, pH 8.0, containing 1% (w/v) SDS (S3); 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea (S4); 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea, and 2% (v/v)  $\beta$ -mercaptoethanol (S5), and 0.5 M NaOH (S6). A minced muscle sample (2 g) was added to 20 ml of the solvent and agitated with a magnetic stirrer for 4 h at room temperature, except that the S5 suspension was heated in the water bath at 100 °C for 2 min before agitation. All samples were centrifuged at 12,100g for 30 min. To the supernatant (4 ml) was added cold 50% (w/v) TCA to a final concentration of 10%. Samples were kept at 4 °C for 18 h and then centrifuged at 2500g for 20 min. The precipitate was solubilised in 0.5 M NaOH. Protein contents were determined by the Biuret method (Robinson & Hodgen, 1940) using BSA as a standard. Solubility was expressed as the percentage of total protein extracted by 0.5 M NaOH.

#### 2.7. SDS–PAGE analysis

Raw and cooked muscle samples (3 g) were homogenised in 5% (w/v) SDS (27 ml) at 11,000 rpm for 60 s with a homogeniser. Five percent (w/v) SDS was solubilised in exudates from cooked muscle samples. All mixtures were incubated at 85 °C for 1 h, the extract was then centrifuged at 6100g for 10 min. The protein content of the supernatant was analysed according to the Biuret method (Robinson & Hodgen, 1940). SDS–PAGE was carried out by the method of Laemmli (1970). The supernatants were mixed at a ratio of 1:1 (v/v) with the SDS–PAGE sample buffer containing 1%  $\beta$ -ME and boiled for 3 min. The samples (20 µg) were loaded onto the gel made of 4% stacking and 10% separating gels and then subjected to electrophoresis using a mini vertical Bio-Rad apparatus (Bio-Rad Laboratoies Pty Ltd, Regents Park, Australia). After electrophoresis, the gels were stained with 0.02% Coomassie Brilliant Blue R-250 in 50% methanol and 7.5% acetic acid and destained with 50% methanol and 7.5% acetic acid for 30 min, followed by destaining with 5% methanol and 7.5% acetic acid for 24 h.

# 2.8. Soluble collagen

Soluble collagen in heated and unheated samples was extracted according to the method of Eilert and Mandigo (1993) with a slight modification. Muscle samples (2 g) were homogenized with 8 ml of 25% Ringer's solution (32.8 mM NaCl, 1.5 mM KCl, and 0.5 mM CaCl<sub>2</sub>). The homogenates were heated for 15 min at 50 °C and centrifuged for 30 min at 2300g. The supernatant was decanted, and the pellet was suspended with the same solution and was recentrifuged. The supernatant solutions were combined. The sediment and supernatants were hydrolyzed with 6 M HCl at 110 °C for 24 h. The hydroxyproline content in the hydrolysate was analyzed by the method of Bergman and Loxley (1963) and converted to collagen content using the factor of 7.25 (Liu, Nishimura, & Takahashi, 1996). Soluble collagen was expressed as percent of the total collagen.

# 2.9. Statistical analyses

Data were evaluated statistically as a one-way ANO-VA using the SPSS 10.0 computer programme. Significant differences between heat treatment means were analyzed by Duncan's multiple range test and a paired sample T test was applied to analyze the significant difference of means between chicken breeds (Steel & Torrie, 1980).

#### 3. Results and discussion

#### 3.1. Heat penetration

The heat penetration times of broiler and indigenous *Pectoralis* muscle stripes were recorded during heating of the samples as shown in Table 1. The indigenous *Pectolaris* muscle stripes took a longer time to reach the end-point temperature than the broiler muscle stripes. This might be due to the thicker perimysium and firmer muscle of the former as reported by Wattanachant, Benjakul, and Ledward (in press).

Table 1	
Heat penetration of broiler and indigenous Pectoralis muscle	

Temperature (°C)	Time to end-point temperature (min)		
	Indigenous	Broiler	
50	$2.27 \pm 0.09$	$2.00 \pm 0.06$	
60	$3.00 \pm 0.16$	$2.83\pm0.02$	
70	$3.85 \pm 0.24$	$3.23 \pm 0.14$	
80	$5.17 \pm 0.07$	$4.83 \pm 0.04$	
90	$8.17 \pm 0.15$	$5.27 \pm 0.11$	
100	$12.00 \pm 0.37$	$8.45\pm0.07$	

Data are presented as means  $\pm$  SD. n = 2.

Heating medium: boiling water bath (100 °C).

Muscle strip size  $2.0\times2.0\times6.0$  cm packed individually in tightly sealed plastic bag.



Fig. 1. Effect of heating temperature on shear value of broiler and indigenous *Pectoralis* muscle. Bars indicate the standard deviation from 20 determinations.



Fig. 2. SEM micrographs of transverse sections of raw and cooked broiler and indigenous Pectoralis muscle.



Fig. 2 (continued)

# 3.2. Changes in textural characteristic and muscle structure

The effects of heat treatment on the shear value of broiler and indigenous chicken muscles are shown in Fig. 1. The indigenous chicken muscles were tougher, than those of the broiler at all temperatures tested (P < 0.001). The changes in shear value of the two chicken muscles were similar. A significant increase in shear value was found at 60 and 80 °C for both chicken muscles (P > 0.05), and at temperatures above 80 °C, no change in the shear value with heating up to 80 °C might be due to the combination effect of the denaturation of myofibrillar proteins, the shrinkage of intramuscular collagen, as well as the shrinkage and dehydration of the actomyosin (Bailey & Light, 1989).

The changes in microstructure of raw and cooked *Pectoralis* muscles from both chicken breeds are presented in Figs. 2 and 3. On the transverse sections (Fig. 2); gaps between muscle fibres were visible in raw and samples cooked at 50 °C. With increasing temperature, the structure of chicken meat became denser and with more compact fibre arrangements at 60–70 °C, especially for the indigenous chicken muscle. The more

compact fibre arrangement resulted in increased shear values. Denaturation and disintegrating of perimysial and endomysial collagen, together with the denaturation of myofibrils, were observed in the range 80-100 °C. This result contributed to negligible or no changes in shear value after 80 °C. On the longitudinal sections (Fig. 3), very slight changes were observed in the range 50-60 °C. However, shrinkage of the sarcomere was obvious with increasing heating temperature from 80 to 100 °C. Weakening of myofibrils, with loss in the structure of Z-disks, occurred at temperatures of 80-100 °C for broiler muscles. The sarcomeres of the cooked indigenous chicken muscles were more compact (with Z-disks still visible) with increasing temperature up to 90 °C; the structure of the Z-disks of indigenous muscle was still visible on treatment at 100 °C. These results suggest that the Z-disk structures of the indigenous chicken muscles were more stable than those of the broiler muscles.

Microstructural measurements of raw and cooked Thai indigenous and broiler chicken muscles are presented in Figs. 4 and 5. A statistically significant decrease in fibre diameter was observed in samples heated to the end-point temperature of 60 °C. This shrinkage might be due to the thermal denaturation of



Fig. 3. SEM micrographs of longitudinal sections of raw and cooked broiler and indigenous Pectoralis muscle.

intramuscular collagen, which was found at 60.7 and 61.7 °C for indigenous chicken and broiler Pectoralis muscle, respectively (Wattanachant et al., in press). The fibre diameter of both chicken muscles was expanded when heated to higher end-point temperatures and tended to stabilise in the range 80-100 °C (P > 0.05). The increase in fibre diameter when heating above 60 °C might be due to the denaturation and progressive loss of crimp structure, and shrinkage of collagen fibres in intramuscular connective tissue, leading to swelling of muscle fibres. The change in fibre diameter was related to the change in sarcomere length, as shown in Fig. 5. Sarcomere length decreased with increasing temperature. The sarcomere length decreased non-significantly when heated to temperatures of 50–60 °C for broiler muscle and 50-70 °C for the indigenous chicken muscle (P > 0.05). However, the greatest shrinkage of the sarcomeres was observed in samples heated to endpoint temperatures of 70–100 °C for the broiler muscle and 80–100 °C for the indigenous chicken muscle (P < 0.05). The result indicates that the shrinkage of chicken meat during cooking in the range of 50–100 °C occurred in two phases. At a temperature of about 50–60 °C, the shrinkage was primarily transverse but was primarily parallel to the fibre axis at 70–100 °C. Changes in muscle fibres observed in this study are in agreement with Offer, Restall, and Trinick (1984) and Palka and Daun (1999) who studied the process in cooked bovine muscles.

# 3.3. Changes in physical properties of muscle

The amount of water bound by a tissue system decreases with increasing temperature (Palka & Daun, 1999). The cooking losses of broiler and indigenous chicken muscles are presented in Fig. 6. The cooking





Fig. 4. Effect of heating temperature on fibre diameter of broiler and indigenous *Pectoralis* muscle. Bars indicate the standard deviation from 40 determinations.

losses of broiler and indigenous chicken muscles were not significantly different in the range 50–70 °C. The greatest increases in cooking loss for indigenous chicken muscles were observed in the range 80–100 °C. Significantly higher cooking losses were found in indigenous chicken muscle compared with those of the broiler (P < 0.001). The difference might be related to the difference in content of crosslinked collagen between chicken breeds (Wattanachant et al., 2004). For the older indig-



Fig. 5. Effect of heating temperature on sarcomere length of broiler and indigenous *Pectoralis* muscle. Bars indicate the standard deviation from 30 determinations.

enous birds, the more highly crosslinked collagen remained insoluble and shrank during heat treatment and effectively squeezed the heat-denatured myofibrillar gel. This led to the loss of moisture and a tougher texture.

Changes in colour of broiler and indigenous chicken muscles during heating are shown in Fig. 7. The CIE system values of lightness  $(L^*)$  and yellowness  $(b^*)$ , of both chicken muscles, increased significantly with



Fig. 6. Effect of heating temperature on cooking losses of broiler and indigenous *Pectoralis* muscle. Bars indicate the standard deviation from seven determinations.



Fig. 7. Effect of heating temperature on colour of broiler and indigenous *Pectoralis* muscle. Bars indicate the standard deviation from 14 determinations.

increasing temperature in the range 50–70 °C (P < 0.05) and no changes they were observed when heated to higher temperatures (P > 0.05). The redness ( $a^*$ ) of indigenous chicken muscle increased significantly when it was heated to an end-point temperature of 70 °C and decreased when heated to higher temperatures (P < 0.05). On the other hand, the  $a^*$  value of broiler muscle decreased with heat treatment up to 60 °C, followed by continuous increase up to 90 °C. A sharp decrease was noticeable at 100 °C. The increase in  $L^*$  and  $b^*$  value and decrease in  $a^*$  value of broiler muscle after cooking at 95 °C and 98 °C have been reported (Fletcher, Qiao, & Smith, 2000; Qiao, Fletcher, Smith, & Northcutt, 2002). With increasing heating temperature, meat tended to be lighter and also turned to a brown-grey hue. The lightening is due to an increased reflection of light, arising from light scattering by denatured proteins (Young & West, 2001). The loss of chroma and change in hue resulted from the changes in myoglobin. Myoglobin is one of the more heat-stable of the sarcoplasmic proteins, which is almost completely denatured between 80 and 85 °C (Lawrie, 1994). According to Lawrie, 1994 and Young and West (2001), the compound involved in increasing redness of muscles should be globin hemochrome, in which the iron is in the  $Fe^{2+}$  state. Its colour is typically dull red. Globin hemichrome, with the iron in the Fe<sup>3+</sup> state, is largely responsible for the browngrey hue. The balance between hemochromes and hemichromes is affected by the state of the meat before cooking and other factors, including species, animal maturity and muscle type (Young & West, 2001).

#### 3.4. Changes in solubility of proteins

Changes in solubility of muscle proteins in S1–S5 are shown in Fig. 8. The contents of soluble protein, in both S1 and S2, markedly decreased with increasing temperature (P < 0.05). The decrease in total soluble proteins of chicken breast patties with increasing temperatures has been reported by Murphy and Marks (2000). This result indicated denaturation or other structural changes in the proteins during heating. The indigenous chicken muscle had a lower content of protein solubilised in S1 and S2 than did broiler muscle, especially in raw muscle. This might be attributed to the lower myofibrillar protein in the former chicken muscle (Wattanachant, 2004). Gradual increases in protein solubility in the presence of denaturating agents, especially SDS and urea (S3 and S4), were observed in the muscle of indigenous chickens up to 70 °C (P < 0.05). The higher content of proteins solubilised in S3, S4 and S5 in indigenous chicken muscle suggested that there were more contributions of hydrogen bonds, hydrophobic interactions and disulfide bonds to the structure compared with that of broiler muscle.

SDS–PAGE patterns of muscle and exudates of both breeds during heating are shown in Figs. 9 and 10. The decrease in myosin heavy chains was observed in the indigenous muscle with increasing temperature. However, the changes in SDS–PAGE patterns for broiler muscle were not noticeable. Murphy and Marks (2000) studied the effect of temperature on properties of chicken



Fig. 8. Effect of heating temperature on solubility of broiler and indigenous *Pectoralis* muscle. Bars indicate standard deviation from triplicate determinations. (*Note*: S1 = 0.6 M KCl; S2 = 20 mM Tris, pH 8.0; S3 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS; S4 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS, 8 M urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS W urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS W urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS W urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS W urea; S5 = 20 mM Tris, pH 8.0, containing 1% (w/v) SDS W ur



Fig. 9. SDS-PAGE pattern of raw and cooked broiler and indigenous *Pectoralis* muscles; (H) and (L) high and low molecular weight protein standards; (1) raw; (2–7) muscles cooked to 50, 60, 70, 80, 90, and 100 °C, respectively.



Fig. 10. SDS–PAGE pattern of exudates from cooked broiler and indigenous *Pectoralis* muscles; (H) and (L) high and low molecular weight protein standards; (1–6) exudates from muscle cooked to 50, 60, 70, 80, 90, and 100 °C, respectively.

breast patties and reported that the protein subunits with a molecular weight greater than 40 kDa decreased with increasing temperature. As shown in Fig. 10, the low molecular weight proteins were observed in the exudates of cooked muscle at lower heating temperature and decreased with increased temperatures. In contrast, solubilized collagen ( $\beta$  and  $\alpha$  band) was observed with increasing temperature in the exudates from the cooked muscle of both breeds. The  $\beta$  and  $\alpha$  bands of solubilized collagen were confirmed by the SDS-PAGE pattern of the soluble fraction in intramuscular connective tissue from both chicken muscles compared with standard collagen (data not shown). The results also indicated that the muscle protein of indigenous muscle was less heatstable, since a lower temperature caused the disappearance of protein bands at low molecular weight in this muscle compared with that of the broiler.

Changes in solubility of collagen during heating of both chicken muscles are shown in Table 2. The soluble collagen content of both chicken muscles increased gradually (P < 0.05) with increasing temperature from 50 to 100 °C. Larick and Turner (1992) stated that collagen began to shrink at 60-70 °C and was converted to gelatin at 80 °C and that these changes weakened the connective tissue. Therefore, the reduction in shear value above 80 °C (Fig. 1) could also be due to the increase in collagen solubility. The indigenous chicken muscles contained less soluble collagen than those of the broiler muscles, at all temperatures tested except at 100 °C. This resulted in higher shear value in the indigenous muscle than in the broiler. The difference in the amount of heat-soluble collagen between broiler and indigenous chicken might be due to differences in amounts of crosslinked collagen, which are related to the age of the bird. The heat-solubility of collagen decreases with increased collagen crosslinking and crosslinking increases with animal age (Foegeding & Lanier, 1996; Pearson & Young, 1989).

Table 2

Effect of heating temperature on soluble collagen content of broiler and indigenous *Pectoralis* muscle

Temperature	Soluble collager	Significance	
(°C)	Broiler	Indigenous	between breeds
Raw	$3.59 \pm 0.85^{a}$	$2.34 \pm 0.63^{\rm a}$	ns
50	$6.09 \pm 0.64^{b}$	$3.60 \pm 1.18^{ab}$	*
60	$6.54 \pm 1.33^{bc}$	$5.12 \pm 0.92^{bc}$	ns
70	$7.87 \pm 1.11^{bcd}$	$5.17 \pm 0.75^{\rm bc}$	*
80	$7.66 \pm 0.91^{bcd}$	$5.93 \pm 1.00^{cd}$	ns
90	$8.13 \pm 1.40^{cd}$	$7.48 \pm 0.92^{d}$	ns
100	$9.41 \pm 0.34^{d}$	$10.69 \pm 1.15^{\rm e}$	ns

Data are presented as means  $\pm$  SD. n = 3.

Significant differences between breeds were determined by *t* test:

\* P < 0.05; \*\*P < 0.01; \*\*\*P < 0.001; ns = no significant difference. <sup>a-d</sup> Means within a column with differing superscripts are significantly different (P < 0.05).

#### Table 3

Linear correlation for changes in structure, texture, cooking loss and solubility of chicken *Pectolaris* muscle

Variable	Indigenous	Broiler
Sarcomere length: cooking loss	0.92	0.83
Cooking loss: shear value	0.80	0.67
Sarcomere length: shear value	0.77	0.60
Fibre diameter: shear value	0.79	0.10
Cooking loss: fibre diameter	0.57	0.02
Protein solubility: shear value	0.77	0.95
Collagen solubility: shear value	0.88	0.86

#### 3.5. Relationship evaluation

Relationships between changes in sarcomere length, fibre diameter, shear value, cooking loss and solubility of protein (in S2) and collagen were evaluated using linear regression as shown in Table 3. A strong linear correlation was obtained between sarcomere length and cooking loss of indigenous ( $R^2 = 0.92$ ) and broiler muscle ( $R^2 = 0.83$ ), solubility of protein and shear value of broiler muscle ( $R^2 = 0.95$ ), soluble collagen and shear value of indigenous ( $R^2 = 0.88$ ) and broiler muscle  $(R^2 = 0.86)$ , and shear value and cooking loss of indigenous muscle ( $R^2 = 0.80$ ). The solubility of muscle protein showed the highest correlation with the shear value of broiler muscle but did not for indigenous chicken muscle. Lopez-Bote, Warriss, and Brown (1989) found that total soluble protein was a good potential predictor of pig lean meat quality. However, the solubility was not significantly related to shear values or sensory tenderness ratings in young beef (Seideman, Koohmaraie, & Crouse, 1987). Therefore, the correlation appears to depend on the species studied. For indigenous chicken, the result indicated that sarcomere length and collagen solubility were very important factors influencing cooking loss and texture of cooked indigenous chicken muscle, and cooking loss directly affected the shear value. The high correlation between sarcomere length and cooking loss was in agreement with Laakkonen, Wellington, and Sherbon (1970) and Bouton, Harris, and Shorthose (1975) who reported that the change in water content contributes to changes in sarcomere length with temperature. The results suggested that changes in muscle structure during heating might influence the texture and cooking loss of cooked chicken meat, especially for indigenous chicken muscle.

# 4. Conclusions

The indigenous chicken muscles were tougher than the broiler muscle at all heating temperatures. The change in shear value of both chicken muscles occurred with the same pattern: significant increase from 50 to 80 °C and no change during 80–100 °C. A significant decrease in fi-

bre diameter was observed in samples heated to an endpoint temperature of 60 °C, while the greatest shrinkage of the sarcomeres was observed in samples heated to the end-point temperatures in the range 70-100 °C for the broiler and 80-100 °C for the indigenous chicken muscles. The shrinkage of chicken meat during cooking in the range 50-100 °C occurs in two phases. At a temperature of about 50-60 °C the shrinkage was primarily transverse and, at 70-100 °C, primarily parallel to the fibre axis. With increasing heating temperature, cooked chicken muscles became lighter and yellower. Cooking loss and collagen solubility increased with increasing internal temperature. In contrast, the solubility of muscle protein decreased with increasing temperatures and had a very high correlation with the texture of broiler muscle. During heating, sarcomere length changes and collagen solubility were very important factors influencing the cooking loss and texture of cooked indigenous chicken muscle, and the cooking loss directly affected the shear value.

# Acknowledgements

This study was supported in part by the grant provided by Prince of Songkla University and by the Ministry of University Affairs of the Thai Government allocated to the Ph.D. consortium programmes.

# References

- Bailey, A. J., & Light, N. D. (1989). Connective tissue in meat and meat products. London: Elsevier Applied Science.
- Bergman, I., & Loxley, R. (1963). Two improved and simplified methods for the spectrophotometric determination of hydroxyproline. *Analytical Chemistry*, 35, 1961–1965.
- Bertola, N. C., Bevilacqua, A. E., & Zaritzky, N. E. (1994). Heat treatment effect on texture changes and thermal denaturation of proteins in beef muscle. *Journal of Food Processing and Preservation*, 18, 31–46.
- Bouton, P. E., Harris, P. V., & Shorthose, W. R. (1975). Changes in shear parameters of meat associated with structural changes produced by aging, cooking, and myofibrillar contraction. *Journal* of Food Science, 40, 1122–1126.
- Califano, A. N., Bertola, N. C., Bevilacqua, A. E., & Zaritzky, N. E. (1997). Effect of processing conditions on the hardness of cooked beef. *Journal of Food Engineering*, 34, 41–54.
- Christensen, M., Purslow, P. P., & Lersen, L. M. (2000). The effect of cooking temperature on mechanical properties of whole meat, single muscle fibres and perimysial connective tissue. *Meat Science*, 55, 301–307.
- Dawson, P. L., Sheldon, B. W., & Miles, J. J. (1991). Effect of aseptic processing on the texture of chicken meat. *Poultry Science*, 70, 2359–2367.
- Eilert, S. J., & Mandigo, R. W. (1993). Procedure for soluble collagen in thermally processed meat products. *Journal of Food Science*, 58, 948–949.
- Fletcher, D. L., Qiao, M., & Smith, D. P. (2000). The relationship of raw broiler breast meat color and pH to cooked meat color and pH. *Poultry Science*, 79, 784–788.

- Foegeding, E. A., & Lanier, T. C. (1996). Characteristic of edible muscle tissues. In O. R. Fennema (Ed.), *Food chemistry* (3rd ed., pp. 879–942). New York: Marcel Dekker.
- Jones, S. B., Carroll, R. J., & Cavanaugh, J. R. (1977). Structural changes in heated bovine muscle: a scanning electron microscope study. *Journal of Food Science*, 42, 125–131.
- Laakkonen, E., Wellington, G. H., & Sherbon, J. W. (1970). Low temperature, long time heating of bovine muscle. 1. Changes in tenderness, water-binding capacity, pH and amount of water soluble components. *Journal of Food Science*, 35, 175–177.
- Laemmli, U. K. (1970). Cleavage of structural proteins during assembly of head of bacteriophage T4. *Nature*, 227, 680–685.
- Larick, D. K., & Turner, B. E. (1992). Aseptic processing of bovine particulates: flavor development/stability and texture. *Journal of Food Science*, 57, 1046–1050.
- Lawrie, R. A. (1994). Meat science. Oxford England: Pergamon Press.
- Liu, A., Nishimura, T., & Takahashi, K. (1996). Relationship between structural properties of intramuscular connective tissue and toughness of various chicken skeletal. *Meat Science*, 43, 43–49.
- Lopez-Bote, C., Warriss, P. D., & Brown, S. N. (1989). The use of muscle protein solubility measurements to assess pig lean meat quality. *Meat Science*, 26, 167–175.
- Murphy, R. Y., & Marks, B. P. (2000). Effect of meat temperature on properties, texture, and cook loss for ground chicken breast patties. *Poultry Science*, 79, 99–104.
- Offer, G., Restall, D., & Trinick, J. (1984). Water-holding in meat. In A. J. Bailey (Ed.), *Recent advances in chemistry of meat* (pp. 71–86). London: The Royal Society of Chemistry.
- Palka, K., & Daun, H. (1999). Changes in texture, cooking losses, and myofibrillar structure of bovine *m. semitendinosus* during heating. *Meat Science*, 51, 237–243.
- Pearson, A. M., & Young, R. B. (1989). Muscle and meat biochemistry. California: Academic Press.
- Qiao, M., Fletcher, D. L., Smith, D. P., & Northcutt, J. K. (2002). Effects of raw broiler breast meat color variation on marination and cooked meat quality. *Poultry Science*, 81, 276–280.
- Robinson, H. W., & Hodgen, C. G. (1940). The biuret reaction in the determination of serum protein I. A study of the condition necessary for the production of the stable color which bears a quantitative relationship to the protein concentration. *Journal of Biological Chemistry*, 135, 707–725.
- Rochdi, A., Foucat, L., & Renou, J. (2000). NMR and DSC studies during thermal denaturation of collagen. *Food Chemistry*, 69, 295–299.
- Roussel, H., & Cheftel, J. C. (1990). Mechanism of gelation of sardine protein: influence of thermal processing and various additives on the texture and protein solubility of kamaboko gel. *International Journal of Food Science and Technology*, 25, 260–280.
- Seideman, S. C., Koohmaraie, M., & Crouse, J. D. (1987). Factors associated with tenderness in young beef. *Meat Science*, 20, 281–291.
- Steel, R. G. J., & Torrie, J. H. (1980). Principles and procedures of statistics; a biometrical approach (2nd ed.). New York: McGraw-Hill.
- Wattanachant, S. (2004). Chemical compositions, properties and structure of muscle affecting textural characteristics of meat from Thai indigenous chicken and broiler. Ph.D. Thesis, Prince of Songkla University.
- Wattanachant, S., Benjakul, S., & Ledward, D. A. (2004). Compositions, color and texture of Thai indigenous and broiler chicken muscles. *Poultry Science*, 83, 123–128.
- Wattanachant, S., Benjakul, S., & Ledward, D. A. (in press). Microstructure and thermal characteristic of Thai indigenous and broiler chicken muscles. *Poultry Science* (in review).
- Young, O. A., & West, J. (2001). Meat color. In Y. H. Hui, Wai-Kit Nip, R. W. Rogers, & O. A. Young (Eds.), *Meat science and applications* (pp. 39–69). New York: Marcel Dekker Inc..

- Yu, L. P., & Lee, Y. B. (1986). Effects of postmortem pH and temperature on bovine muscle structure and meat tenderness. *Journal of Food Science*, 51, 774–780.
- Zayas, J. F., & Naewbanij, J. O. (1986). The effect of microwave heating on the texture properties of meat and collagen solubilization. *Journal of Food Processing and Preservation*, 10, 203–214.