# Effects of Rigor Status during High-Pressure Processing on the Physical Qualities of Farm-Raised Abalone (*Haliotis rufescens*)

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**Abstract:** High-pressure processing (HPP) is used to increase meat safety and shelf-life, with conflicting quality effects depending on rigor status during HPP. In the seafood industry, HPP is used to shuck and pasteurize oysters, but its use on abalones has only been minimally evaluated and the effect of rigor status during HPP on abalone quality has not been reported. Farm-raised abalones (*Haliotis nufescens*) were divided into 12 HPP treatments and 1 unprocessed control treatment. Treatments were processed pre-rigor or post-rigor at 2 pressures (100 and 300 MPa) and 3 processing times (1, 3, and 5 min). The control was analyzed post-rigor. Uniform plugs were cut from adductor and foot meat for texture profile analysis, shear force, and color analysis. Subsamples were used for scanning electron microscopy of muscle ultrastructure. Texture profile analysis revealed that post-rigor processed abalone was significantly (P < 0.05) less firm and chewy than pre-rigor processed irrespective of muscle type, processing time, or pressure. L values increased with pressure to 68.9 at 300 MPa for pre-rigor processed foot, 73.8 for post-rigor processed foot, 90.9 for pre-rigor processed adductor, and 89.0 for post-rigor processed adductor. Scanning electron microscopy images showed fraying of collagen fibers in processed adductor, but did not show pressure-induced compaction of the foot myofibrils. Post-rigor processed abalone meat was more tender than pre-rigor processed meat, and post-rigor processed foot meat was lighter in color than pre-rigor processed foot meat, suggesting that waiting for rigor to resolve prior to processing abalones may improve consumer perceptions of quality and market value.

Keywords: abalone, high-pressure processing, rigor, texture

**Practical Application:** This study demonstrated the effects of rigor status during high-pressure processing (HPP). Postrigor processed abalone was more tender than pre-rigor processed abalone, and color lightened with increasing pressure. These results have important implications because abalone is typically processed pre-rigor. Post-rigor HPP may improve processing efficiency and marketability since mechanical tenderization and chemical bleaching agents would not be necessary to enhance product quality.

# Introduction

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Abalones are highly perishable molluscan shellfish known for their distinctive firm and crisp raw texture (Brown and others 2008). Abalone meat may be categorized as foot or adductor muscle (Fallu 1991), with foot meat typically containing higher quantities of collagen and pigment than the naturally tender, white adductor meat (Olaechea and others 1993; Allen and others 2006). Whiter abalone meat is more desirable to consumers but can be hard to achieve in farm-raised abalone fed algal diets, resulting in the need for trimming or chemical bleaching of the meat (Oakes and Ponte 1996; Allen and others 2006; Brown and others 2008). Common processes to extend abalone shelf-life include canning, drying, and freezing, which are economical but can negatively affect physical quality of the meat.

High-pressure processing (HPP), the application of uniform hydrostatic pressure, is used in the shellfish industry to process oysters

MS 20141044 Submitted 6/16/2014, Accepted 10/13/2014. Authors Hughes and Skonberg are with Univ. of Maine, School of Food and Agriculture, 5735 Hitchner Hall, Orono, ME, 04469-5735, U.S.A. Author Greenberg is with Univ. of Maine, Aquaculture Research Inst, 5735 Hitchner Hall, Orono, ME, 04469-5735, U.S.A. Author Yang is with US Army Natick Soldier Research, Development, and Engineering Center, 16 Kansas St, Natick, MA, 01760, U.S.A. Direct inquiries to author Skonberg (e-mail: Denise.Skonberg@umit.maine.edu). and lobsters but has only been minimally investigated for use on abalone. HPP-induced changes at the cellular level may include enzyme inactivation, pathogen cell wall damage, pigment break down (bleaching), and protein deformation (textural changes) (Ronner 1995; Corwin and Shellhammer 2002; Ichinoseki and others 2006; Waite and others 2009; Del Olmo and others 2010; Perera and others 2010). Pressures over 400 MPa have been shown to toughen meat such as chicken and beef (Del Olmo and others 2010; Duranton and others 2012), although there is not abundant research on the effects of HPP on abalone. Red abalone increased in chewiness and cohesiveness, but did not increase in firmness, when pressurized at 500 or 550 MPa (Briones-Labarca and others 2012). However, chewiness and firmness both contribute to perceived toughness, an undesirable attribute for abalone meat. Evaluation of processing pressures of less than 500 MPa to prevent postprocessing chewiness is limited. However, a recent study reported that disk abalone treated at 400 MPa for 3 minutes exhibited decreased water holding capacity and similar shear values compared to unprocessed controls, although chewiness was not evaluated (Jo and others 2014). Further investigation of the effect of different processing times at pressures of less than 500 MPa, particularly on textural attributes of HPP abalone, is warranted.

HPP has been shown to proportionally increase whiteness and opacity with increased pressure in mackerel, cod, bluefish, carp,

trout, salmon, and red mullet (Ohshima and others 1993; Angsupanich and Ledward 1999; Master and others 2000; Sequeria-Munoz and others 2006; Yagiz and others 2007; Yagiz and others 2009; Erkan and others 2010). However, one study (Briones-Labarca and others 2012) reported that abalone L values decreased with high pressure (550 MPa, 8 min) compared to the unprocessed control abalone. Further investigation of the effect of different processing pressures on abalone meat color is warranted, particularly since whitening would be a desirable effect of HPP.

HPP is known to effect different changes on meat quality depending on whether the meat is pre-rigor or post-rigor during HPP. Specifically, beef processed pre-rigor has been shown to be more tender than unprocessed and post-rigor processed beef, unless high temperatures are used in conjunction with HPP (Mac-Farlane 1973; Bouton and others 1977; Kennick and others 1980; Jung and others 2000; Sikes and others 2010). The effect of rigor status during processing on seafood quality has been infrequently discussed except as it pertains to gaping in fish such as cod and salmon (Lauritzsen and others 2004; Esaiassen and others 2008; Larsen and others 2008). Evaluations of rigor status during processing of shellfish such as lobster and scallop are very limited (Gornik and others 2009; Jiménez-Ruiz and others 2013), and have not been reported for abalone, which are usually processed pre-rigor. The objectives of this study were to assess the effects of abalone rigor status during HPP, and the effects of different HPP parameters (pressure and time), on the color, texture, and ultrastructure of abalone adductor and foot meat.

## Materials and Methods

# **Rigor observation**

A guideline of time to rigor and rigor resolution was needed to inform the pre-rigor and post-rigor processing schedule. Whole abalones (The Abalone Farm, Cayucas, Calif., U.S.A.; n = 2) were shucked, eviscerated, and placed on ice at 2 °C to ascertain the time to onset of rigor, and subsequent resolution. The abalone meats were assessed by compression force (Newtons, N), foot side up, immediately after shucking, at 6 h, at 12 h, and every 2 h from then until rigor resolution using a texture analyzer (TA-XT2i, Texture Technologies Corp., Scarsdale, N.Y., U.S.A.). Force was recorded by the texture analysis software (Exponent 32, version 5,0,6,0, 2010, Texture Technologies Corp.). A 10 mm cylinder probe was used at a test speed of 2 mm/s to a depth of 5 mm.

# **Experimental design**

The HPP study had a  $2 \times 2 \times 3$  factorial experimental design, with rigor status (pre- and post-rigor), pressure (100 and 300 MPa), and processing time (1, 3, and 5 min) as the treatment variables (Table 1). Post-rigor unprocessed meats were used as the control because all meats were post-rigor at the time of analyses. Samples were coded according to rigor status (PRE or POST), processing time (1, 3, or 5 min) then pressure (100 or 300 MPa), for example, PRE-5-300 was processed pre-rigor for 5 min at 300 MPa. Shucked abalones were processed whole, but were divided into adductor and foot for all analyses. Each of the 12 processing treatments (plus the unprocessed control treatment) contained 12 abalones (N = 156). Reagents were analytical grade and were purchased from Fisher Scientific (Waltham, Mass., U.S.A.) unless otherwise noted.

Processing

Live, July-harvested, farm-raised abalones (The Abalone Farm, Cayucas, Calif., U.S.A.; Haliotis rufescens; N = 156) were divided into 2 groups, PRE and POST, according to the experimental design. Individual abalones were weighed. The mean weight (n = 40) of a random sampling of in-shell abalones was 88.9 g  $\pm$  11.5. Abalones had not been fed a minimum of 5 days before shucking to promote glycogen depletion. Abalones were shucked, eviscerated, packed in plastic bags, and stored on ice at 2 °C until further processing. POST abalones, including control, were stored on ice for at least 40 h after shucking and PRE were stored on ice between 1 and 6 h after shucking. Samples were sealed in plastic HPP bags (Winpak, Winnepeg, MB, Canada) under 99% vacuum. There were 6 abalones per bag, and treatments were randomly processed using a 1 L HPP unit (Engineering Pressure Systems Inc., Haverhill, Mass., U.S.A.). The temperature of the vessel during pressurization ranged from 23 to 28 °C. Hydraulic fluid (20:1 water:Hydrolubric 120-B [Houghton Intl. Inc., Norristown, Pa., U.S.A.]) was used to achieve hydrostatic pressure. The come-up time ranged from 3 to 4.5 min and depressurization was immediate.

### Colorimetric and texture analyses

**Preparation.** Treatments were analyzed in random order. The whole abalones were sliced horizontally below the base of the adductor to separate the foot from the adductor muscle. Square plugs measuring  $20 \times 20 \times 7$  mm by a digital caliper were cut from the center of each muscle. Plugs from the foot were cut from the ventral side, and plugs from the adductor were cut from the dorsal side. Plugs were stored in plastic bags at 4 °C until analyses.

**Color.** Color analyses of each treatment (n = 12) were performed using a colorimeter (LabScan XE, Hunter Labs, Reston, Va., U.S.A.). The Hunter *L*, *a*, *b* values were recorded by the colorimeter software (Universal, version 4.10, 2001, Hunter Labs). The colorimeter was standardized using white and black tiles. Each plug was evaluated 3 times by rotating  $120^{\circ}$  from the previous reading, and the 3 values from each plug were averaged.

Texture analyses. Twelve abalones per treatment were divided into 2 groups to conduct 2 different texture analyses: texture profile analysis (TPA) and shear by an A/CKB craft knife blade. Following color analysis, individual muscle plugs were placed on the calibrated texture analyzer platform (TA-XTi2, Texture Technologies Inc.) noncut surface side up. For TPA of each treatment (n = 6), the texture analyzer was configured with a 10 mm probe, 2 mm/s test speed, 30% compression, and a 5 s gap between the 2 compressions. TPA emulates chewing, and can be useful for comparison to the consumer experience (Bourne 2002). Force (Newtons, N), area (N  $\times$  s), and time (s) were calculated by the texture analysis software (Exponent 32, version 5,0,6,0 2010, Texture Technologies Inc.) from the force-deformation curve to quantify firmness (resistance to compression, N), springiness (how well meat springs back after compression, unitless), resilience (immediate springiness as the probe is withdrawn between "bites," unitless), and chewiness (characterizes elastic resistance of meat, unitless; Bourne 2002). Shear force analysis of each treatment was conducted using plugs (n = 6) placed so the blade cut across the muscle fibers. The texture analyzer blade was configured to a 6 mm depth and a 5 mm/s test speed.

## Scanning electron microscopy

Abalone samples were prepared for microwave enhanced fixation by using a razor blade to slice pieces (n = 3) no thicker

## Table 1-Experimental treatments.

|                                | Pressure                             |                  |  |  |
|--------------------------------|--------------------------------------|------------------|--|--|
| Processing parameters          | 100 MPa                              | 300 MPa          |  |  |
| 1 min                          | 1-100                                | 1-300            |  |  |
| 3 min                          | 3-100                                | 3-300            |  |  |
| 5 min                          | 5-100                                | 5-300            |  |  |
| Muscle analyzed                | Adductor or foot                     | Adductor or foot |  |  |
| Rigor status during processing | Pre-rigor (PRE) or Post-rigor (POST) | PRE or POST      |  |  |

than 1 mm from the center of the muscle. Foot and adductor muscles from control, minimum (1–100) and maximum (5–300) HPP treatments were selected to optimize observation of potential visual treatment effects. Iced specimens were fixed in 2.5% glutaraldehyde (Electron Microscopy Services, Hatfield, Pa., U.S.A.), in phosphate buffer for one 7 s cycle on high, followed by a 20 s rest, and a final 7 s cycle on high (7/20/7). The initial fixation was followed by 2 buffer rinses before 1% osmium tetroxide (Electron Microscopy Services), fixation for one 7/20/7 cycle and a final deionized water rinse. Fixed specimens were dehydrated in serially increasing concentrations of ethanol, and stored in 100% ethanol until critical point drying (Samdri PVT-3, Tousimis Research Corp., Rockville, Md., U.S.A.). Dried samples were transferred to stubs affixed with carbon-coated tape and silver conductive adhesive (type 503, Electron Microscopy Sciences, Hatfield, Pa.,

U.S.A.). The samples were sputter coated (Cressington 108 Auto, Redding, Calif., U.S.A.) at 40 mA and 0.08 mbar for 90 s to generate a 32 nm layer of gold–palladium on the surface. Samples were stored in a dessicator until imaging. The scanning electron microscope (AMRay 1820 Digital SEM, Bedford, Mass., U.S.A.) was degaussed initially and between samples. An accelerating potential of 10 kV and spotsize of 10 were selected, and magnification up to 2000 times ( $2000 \times$ ) was used. Foot myofibril widths (n = 10) were measured and averages were used for statistical analyses.

## Statistical analysis

Data were analyzed using SYSTAT 12 (Systat Software, Chicago, Ill., U.S.A.) for one-way analysis of variance (ANOVA) for all one-level (treatment) analyses. Multiway ANOVA was used to assess interactions between rigor status, pressures, and



#### Table 2-Main effects of experimental variables and their interactions.

|   | L  | а  | Ь  | Shear | Firmness | Chewiness | Springiness | Resilience |
|---|----|----|----|-------|----------|-----------|-------------|------------|
| Muscle  | ** | ** | ** | **    | **       | **        | ns          | **         |
| Rigor status                                      | ** | ** | ** | *     | **       | **        | **          | **         |
| Pressure  | ** | ** | ** | ns    | ns       | ns        | **          | ns         |
| Processing time                                   | ** | ns | ns | *     | ns       | ns        | ns          | ns         |
| Pressure × time                                   | ns | ns | *  | ns    | ns       | *         | ns          | ns         |
| Pressure $\times$ muscle                          | ** | ns | ns | ns    | ns       | ns        | ns          | ns         |
| Pressure × rigor                                  | *  | ns | ns | **    | ns       | ns        | ns          | ns         |
| Time × muscle                                     | ns | ns | ns | ns    | ns       | ns        | ns          | *          |
| Time $\times$ rigor                               | ns | ns | ns | *     | ns       | ns        | ns          | ns         |
| Muscle × rigor                                    | ** | ns | ** | ns    | **       | **        | **          | ns         |
| Pressure $\times$ time $\times$ muscle            | *  | ns | ns | ns    | ns       | ns        | ns          | *          |
| Pressure $\times$ time $\times$ rigor             | *  | ns | *  | ns    | ns       | *         | ns          | ns         |
| Pressure $\times$ muscle $\times$ rigor           | ns | ns | ns | ns    | ns       | ns        | ns          | ns         |
| Time $\times$ muscle $\times$ rigor               | ns | ns | *  | ns    | **       | *         | ns          | ns         |
| $Pressure \times time \times muscle \times rigor$ | ns | ns | ns | **    | ns       | ns        | ns          | ns         |

 ${}^{a}P < 0.05$ ;  ${}^{**}P < 0.01$ ; ns, not significant.

processing times using a  $2 \times 2 \times 3$  factorial. Separation of treatment means was accomplished using Tukey's honest significant difference (HSD) *post hoc* test. The Shapiro–Wilk test was used to assess normality and Levene's equality of variances test was used to assess homogeneity. In cases where data did not satisfy either normality or homogeneity, they were evaluated nonparametrically using Kruskal–Wallis. To separate nonparametric treatment means, the Mann–Whitney *post hoc* test was used. For all statistics, a significance level of P < 0.05 was selected.

# **Results and Discussion**

## **Rigor observation**

The time from rigor onset to resolution is dependent on the species, pre-mortem stress, feeding status at slaughter, and temperature (Mørkøre and others 2008; Gornik and others 2009). An evaluation of approximate time of rigor onset through resolution was necessary before the HPP study to determine hold times for shucked abalones prior to pre- or post-rigor processing. The results of the rigor evaluation demonstrated that these abalones were in a pre-rigor state until 12 h postshucking, at which point an increase in force was required to compress the abalone muscle. The time to rigor onset for the farm-raised red abalone meat was very similar to values reported for the lion's paw scallop, which went into rigor between 8 h at 0 °C and 16 h at 5 °C (Jiménez-Ruiz and others 2013). Compression force was highest between 16 and 20 h postshucking, with an average compression force of 36.2 N compared to the average pre-rigor compression force of 10.6 N. Rigor resolution was achieved by 26 h, with an average final compression force of 8.7 N. The onset of rigor occurs as adenosine triphosphate (ATP) levels become depleted, with correlations between fast onset and low initial glycogen levels due to starvation or preslaughter stress having been reported for salmon muscle (Mørkøre and others 2008). No comparable rigor studies have been published for abalone muscle, however because the abalones used for this study had not been starved before shucking, it was expected that they would exhibit a longer rigor to resolution curve than would be expected of glycogen-depleted abalones.

## Color

The abalone foot muscle is known to be darker in color than the adductor due to the presence of pigments such as melanin, carotenoids, and bilichromes such as haliotisrubin (Voltzow 1994; Portela and others 2012). Consumers typically prefer lighter meat,

| Table | 3-Hunt     | er a  | values | of  | abalone  | foot   | and   | adductor     | meat. |
|-------|------------|-------|--------|-----|----------|--------|-------|--------------|-------|
| Each  | value is t | the r | nean ± | sta | ndard de | viatio | on (n | ≥ <b>8).</b> |       |

|         | Fo               | ot               | Adductor                  |                           |  |  |
|---------|------------------|------------------|---------------------------|---------------------------|--|--|
|         | Pre-rigor        | Post-rigor       | Pre-rigor                 | Post-rigor                |  |  |
| Control | $12.0 \pm 2.6$ a | $12.0 \pm 2.6$ a | $2.7 \pm 1.2 \text{ c}$   | $2.7 \pm 1.2$ bc          |  |  |
| 1–100   | $13.6 \pm 2.3$ a | $11.7 \pm 1.9$ a | $4.4 \pm 1.7 \text{ abc}$ | $2.7 \pm 1.0$ bc          |  |  |
| 3–100   | $14.2 \pm 1.9$ a | $11.6 \pm 1.5$ a | $4.2 \pm 1.1 \text{ abc}$ | $2.8 \pm 1.0$ bc          |  |  |
| 5–100   | $14.9 \pm 2.7$ a | $12.2 \pm 2.2$ a | $3.9 \pm 1.2 \text{ bc}$  | $2.5 \pm 1.6 \text{ c}$   |  |  |
| 1–300   | $15.1 \pm 3.2$ a | $14.2 \pm 1.3$ a | $5.9 \pm 1.7 \text{ a}$   | $3.7 \pm 0.9 \text{ abc}$ |  |  |
| 3–300   | $14.9 \pm 2.6$ a | $14.1 \pm 2.4$ a | $5.3 \pm 1.1 \text{ ab}$  | $4.1 \pm 0.9 \text{ ab}$  |  |  |
| 5–300   | $15.0 \pm 2.4$ a | $14.1 \pm 2.7$ a | $5.3 \pm 1.9 \text{ ab}$  | $4.4 \pm 1.3 \text{ a}$   |  |  |

Control is represented as both PRE and POST for simplicity of comparisons across treatments. Values within columns not sharing a lowercase letter are significantly (P < 0.05) different. All treatments were analyzed by ANOVA (Tukey's HSD *post hoc* test).

and abalones with dark pigmentation may be heavily trimmed or bleached (Brown and others 2008). HPP has been reported to increase L and b values and decrease a values in trout, mahi mahi, red mullet, and salmon muscle (Yagiz and others 2007; Yagiz and others 2009; Erkan and others 2010; Ojagh and others 2011). These color changes are presumably due to denaturation of proteins such as myoglobin as well as oxidation of carotenoids and ferrous myoglobin to ferric metmyoglobin. Similarly, HPP oyster meats have been shown to have increased L values, decreased avalues, and unchanged b values compared to unprocessed controls (Lai and others 2010).

Foot and adductor muscle color increased in L value with increasing pressure (Figure 1). The HPP-induced increase in L value (lightening) was most notable in the darker pigmented foot muscle. Foot L values increased from 62.5 (control) to 68.9 (PRE-5-300) and 73.8 (POST-5-300), although the increase was only significant for POST-5-300. Post-rigor processed foot had significantly higher values than pre-rigor processed foot, and L values increased with increasing pressure (Table 2). Adductor L values significantly increased from 84.1 (control) to 90.9 (PRE-5-300) and 88.97 (POST-5-300).

It is known that HPP can oxidize carotenoid pigments in salmon muscle, evidenced by significantly lower a values compared to a raw control (Ojagh and others 2011). However, the results of this study demonstrate increases in both a and b values with pressure (Table 2 and 3). While increasing a values have not been reported in HPP fish or oysters, they have been reported in HPP chicken processed at 400 MPa for a single cycle (Del Olmo and others 2010). Abalone meat processed at 550 MPa for 8 min was reported Table 4-Hunter *b* values of abalone foot and adductor meat. Each value is the mean  $\pm$  standard deviation ( $n \ge 8$ ). Control is represented as both PRE and POST for simplicity of comparisons across treatments. Values within columns not sharing a lowercase letter are significantly (P < 0.05) different, analyzed by ANOVA (Tukey's HSD *post hoc*).

|         | Fo                        | ot                       | Adductor                  |                           |  |
|---------|---------------------------|--------------------------|---------------------------|---------------------------|--|
|         | Pre-rigor                 | Post-rigor               | Pre-rigor                 | Post-rigor                |  |
| Control | $23.0 \pm 1.8 \text{ bc}$ | $23.0 \pm 1.8$ b         | $10.8 \pm 1.4 \text{ d}$  | $10.8 \pm 1.4 \text{ d}$  |  |
| 1-100   | $22.6\pm1.8~{ m c}$       | $23.7 \pm 1.6 \text{ b}$ | $14.5 \pm 2.7 \text{ b}$  | 11.1 ± 1.5 d              |  |
| 3-100   | $22.7\pm2.8~{\rm c}$      | $23.3 \pm 1.0 \text{ b}$ | $15.2 \pm 1.1 \text{ b}$  | $11.9 \pm 1.8$ bcd        |  |
| 5-100   | $25.2 \pm 1.5$ b          | $23.9 \pm 1.8$ b         | $15.3 \pm 2.3 \text{ b}$  | $11.8 \pm 2.1 \text{ cd}$ |  |
| 1-300   | $26.3 \pm 1.8$ a          | $26.8 \pm 1.3$ a         | $18.8 \pm 1.5$ a          | $13.4 \pm 1.4$ abc        |  |
| 3-300   | $26.1 \pm 2.1$ a          | $26.9 \pm 1.4$ a         | $16.5 \pm 1.8 \text{ ab}$ | $14.7 \pm 1.8$ a          |  |
| 5-300   | $26.4\pm1.7~\mathrm{a}$   | $26.3\pm2.0~\text{a}$    | $16.6\pm1.7~\mathrm{ab}$  | $14.0 \pm 1.9 \text{ ab}$ |  |

Table 5-Shear values (N) of abalone foot and adductor meat. Each value is the mean  $\pm$  standard deviation (n = 6).

|         | F                         | oot                        | Adductor         |                  |  |  |
|---------|---------------------------|----------------------------|------------------|------------------|--|--|
|         | Pre-rigor                 | Post-rigor                 | Pre-rigor        | Post-rigor       |  |  |
| Control | $39.8 \pm 5.6$ a          | 39.8 ± 5.6 a               | $24.3 \pm 3.8$ a | $24.3 \pm 3.8$ a |  |  |
| -100    | $35.0 \pm 1.9$ ab         | 28.6 ± 3.9 c               | $21.2 \pm 5.9$ a | $28.9 \pm 5.2$ a |  |  |
| 6-100   | 33.7 ± 2.7 b              | $38.8 \pm 7.4$ ab          | $26.5 \pm 3.7$ a | $26.7 \pm 4.4$ a |  |  |
| 5-100   | $36.1 \pm 3.2 \text{ ab}$ | $29.5 \pm 6.9 \text{ bc}$  | $29.4 \pm 4.4$ a | $26.9 \pm 7.3$ a |  |  |
| -300    | $32.5 \pm 1.4 \text{ b}$  | $37.0 \pm 2.5 \text{ abc}$ | $26.6 \pm 5.9$ a | $27.1 \pm 4.5$ a |  |  |
| 3-300   | $35.4 \pm 2.3$ ab         | $39.7 \pm 2.7$ a           | $23.1\pm5.8$ a   | $32.3 \pm 4.6 a$ |  |  |
| 5-300   | $33.8\pm3.1~\text{b}$     | $34.6 \pm 1.8 \text{ abc}$ | $23.8\pm4.9\;a$  | $28.6\pm4.2$ a   |  |  |
|         |                           |                            |                  |                  |  |  |

Control is represented as both PRE and POST for simplicity of comparisons across treatments. Values within columns not sharing a lowercase letter are significantly (P < 0.05) different. All treatments were analyzed by ANOVA (Tukey's HSD *post hoc*).

to have L, a, and b values that did not significantly differ from unprocessed controls (Briones-Labarca and others 2012), which is in contrast to the values reported in this study, potentially due to both the longer time and higher pressures. Post-rigor processed

abalone had significantly increasing *a* values with pressure, from 11.98 (control) to 14.14 (POST-5-300) for foot samples, and 2.69 (control) to 4.39 (POST-5-300) for adductor samples. Despite increasing with pressure, *a* values for post-rigor processed samples



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for both foot and adductor were significantly lower than pre-rigor processed samples (Table 2). The *b* values (Table 3) significantly increased with pressure irrespective of muscle type or rigor status. There were no significant differences in *b* values between prerigor processed and post-rigor processed foot samples, however pre-rigor processed adductor samples had significantly higher *b* values than post-rigor processed adductor regardless of treatment (Table 2).

There was no significant effect of processing time on the Hunter L, a, or b values within muscle type or rigor status. The adductor had significantly greater L values and lower a and b values than the foot as expected (Table 2). Finally, post-rigor processed foot was significantly lighter, with greater L and lower a and b values, than pre-rigor processed foot irrespective of treatment. Considering the importance of light coloration to abalone consumers, post-rigor HPP of abalones may be a way to increase acceptance of highly pigmented abalone foot.

## Texture

**Texture profile analysis.** Pre-rigor processed meat significantly toughened due to processing compared to both the control and the post-rigor processed treatments, which were not different in firmness from the control, irrespective of time or muscle type (Table 2). The post-rigor results are similar to those reported for

abalones processed at 550 MPa for 5 min, which were not significantly firmer than unprocessed controls (Briones-Labarca and others 2012). In this study, foot firmness was not different between the control (7.7 N) and the POST-5-300 treatment (8.8 N), however firmness significantly increased for pre-rigor processed treatments (41.6 N for PRE-5-300; Figure 2). Results for adductor were similar, with firmness not being different between the control (1.8 N) and POST-5-300 (1.7 N), but significantly increasing when processed pre-rigor (8.1 N for PRE-5-300). Overall, adductor meat was less firm (approximately 5 times) and less chewy (approximately 5 to 12 times) than foot meat, which was expected given the lower collagen contents of adductor compared to foot (Olaechea and others 1993). In beef pressurized pre-rigor, a massive muscle contraction occurred during pressurization, which caused major internal rupturing and irreversible changes to the collagen fibers (Kennick and others 1980). These pressure-induced changes resulted in tenderization of pressurized pre-rigor meat compared to control meat. As tenderization of prerigor processed abalone was not observed in this study, it can be presumed that a massive contraction similar to the one reported by Kennick and others (1980) did not occur, possibly due to the greater collagen content of abalone compared to beef.

The crisp texture of raw abalone meat is highly desired, so preservation of the raw textural qualities of unprocessed meat is

> Figure 3–TPA values for foot springiness (A), adductor springiness (B), foot resilience (C), and adductor resilience (D). Control is represented as both PRE and POST for simplicity of comparisons across treatments. PRE treatments not sharing a lowercase letter are significantly (P < 0.05) different. POST treatments not sharing an uppercase letter are significantly different. All treatments were analyzed by ANOVA (Tukey's HSD *post hoc*) except PRE-foot springiness, PRE-foot resilience, and PRE-adductor resilience which did not satisfy normality or homogeneity requirements and were analyzed by Kruskal–Wallis (Mann–Whitney *post hoc*).



important (Olaechea and others 1993). Processing of pre-rigor meat caused substantial increases in firmness compared to the control and post-rigor processed meat, however, because firmness values for the highest processing parameters (5300) of post-rigor meat were not significantly different from the control, pressures up to 300 MPa for 5 min could be used to process post-rigor abalones without toughening foot or adductor meat. There were no significant differences between the post-rigor processed meat and the control, although the foot was 3 times as chewy as the adductor. Pre-rigor processed foot and adductor were significantly more chewy than post-rigor processed meat (Figure 3), increasing from 2.9 (POST-5-300) to 21.0 (PRE-5-300) for the foot, and increasing from 0.7 (POST-5-300) to 4.3 (PRE-5-300) for adductor. These results align with those for firmness, and suggest that processing abalones post-rigor will not cause an increase in chewiness of raw abalones.

Springiness, or elasticity, in the foot significantly decreased with HPP, regardless of rigor status, decreasing from 0.893 (control) to 0.759 (PRE-5-300) and 0.668 (POST-5-300; Figure 4). Springiness also decreased with HPP for pre-rigor adductor from 0.909 (control) to 0.725 (PRE-5-300), but was not different for post-rigor adductor, ranging from 0.909 (control) to 0.762 (POST-5-300). The decrease in springiness at the highest pressure and time combination for foot meat was not seen in the post-rigor processed adductor treatments or in similar studies. A trained panel evaluating canned abalone did not detect a difference in springiness between foot and adductor for canned or control

abalone (Sanchez-Brambila and others 2002). Similarly, HPP abalone springiness was not different from controls when evaluated immediately after processing (Briones-Labarca and others 2012).

Pre-rigor processed foot samples were nearly twice as resilient as post-rigor processed samples, at 0.449 for PRE-5-300 compared to the significantly lower 0.254 for POST-5-300 (Figure 4). Adductor meat followed the same trend with values ranging from 0.433 for PRE-5-300 to 0.189 for POST-5-300. Conversely, post-rigor processed foot (0.254) and adductor (0.189) were not different from controls (0.273 foot, 0.256 adductor), but pre-rigor processed foot (0.449) and adductor (0.433) had significantly greater resilience than controls (0.273 foot, 0.256 adductor). These results suggest that HPP up to 300 MPa for 5 min could be utilized to process post-rigor abalone without affecting the firmness, chewiness, or resilience compared to unprocessed raw abalone, despite springiness being negatively affected.

**Shear.** Shear represents cutting force, and together with TPA, gives a more comprehensive understanding of abalone textural attributes. Unlike the TPA results, the shear values did not demonstrate significant differences that followed specific trends related to pressure or rigor status (Table 4). Most notably, there was no effect of rigor status on foot or adductor shear force, although when evaluated by TPA pre-rigor processed meat was much firmer than the control or post-rigor processed meat (Table 2). Although all shear values for both pre- and post-rigor processed foot meat were lower than the control, differences were not significant. TPA describes many attributes related to compression of meat, while shear



Figure 4–Scanning electron micrographs of abalone adductor meat control (A), PRE-1-100 (B), PRE-5-300 (C), and POST-5-300 (D) taken at 2000 x. Scale bars represent 10  $\mu$ m.



Figure 5–Scanning electron micrographs of abalone foot meat control (A), PRE-1–100 (B), PRE-5–300 (C), and POST-5–300 (D) taken at 1000 x. Scale bars represent 20  $\mu$ m.

specifically records the force required to cut across a muscle, which includes shearing of collagen fibers. Collagen has been shown to be only minimally affected by HPP (Ichinoseki and others 2006) so the lack of difference in shear values among treatments is not unexpected.

Foot meat required significantly greater shear force than adductor meat (Table 4), which was expected based on its higher collagen content and on the higher TPA values recorded for foot meat compared to adductor meat. Pre- and post-adductor shear values (23.8 N PRE-5-300, 28.6 N POST-5-300) were not significantly different from the control (24.3 N). TPA firmness and shear results shared similar values for pre-rigor foot samples, 41.6 N for firmness (PRE-5-300) and 33.8 N for shear (PRE-5-300). Post-rigor foot samples, however, were significantly firmer when evaluated by shear (34.6 N POST-5-300) than by TPA (8.8 N POST-5-300). While HPP up to 500 MPa was shown to reduce shear force values in beef from 5.9 N (unprocessed control) to 4.3 N (100 MPa) and 4.9 N (500 MPa; Ichinoseki and others 2006), the higher shear force values observed in this study were likely due to the much higher collagen content in abalone (3.5%) compared to beef (0.29% to 0.57%); Nakamura and others 2010).

## Scanning electron microscopy

HPP caused unraveling of collagen fibers, but did not cause visible changes in myofibril widths. At higher magnification  $(2000 \times)$ , small fibrils (<1  $\mu$ m) were observed in all adductor images except rigor abalone meat as evidenced by increased firmness, chewiness, the control (Figure 4). The presence of these fibrils in the absence

of shear force differences between control and processed adductor samples suggests pressure-induced unraveling of collagen fibers. It has been shown in beef that HPP can cause stretching of the connective tissue, though changes in texture are minimized due to the continued presence of collagen in the tissue (Ichinoseki and others 2006).

In contrast, the SEM images of myofibrils (1000×) showed little difference among processing treatments (Figure 5). While PRE-1-100 had narrower myofibrils (2.0  $\mu$ m) than the control (2.7  $\mu$ m), there was no difference between myofibril widths of PRE-5-300 (3.0  $\mu$ m), POST-5-300 (2.5  $\mu$ m), and the control refuting that the decrease in fiber widths for the PRE-1-100 was due to processing or rigor status. The lack of difference in both collagen and myofibril images among the HPP treatments was unexpected since firmness values for PRE-5-300 were 6 times higher than for POST-5-300. The SEM images do, however, corroborate shear force values, which were not significantly different among treatments. Overall, the lack of observable differences in collagen and myofibrils suggests that the high collagen content of abalone meat significantly contributes to texture and may inhibit changes in myofibril widths despite pressure induced unraveling of the collagen.

## Conclusions

HPP at 300 MPa for 5 min caused significant toughening of preand resilience values. Compared to pre-rigor processed abalones, post-rigor processed abalones were lighter in color and more tender, suggesting that waiting for rigor to resolve before processing abalones would be beneficial for product quality attributes. Notably, the texture of post-rigor processed abalone was not significantly affected by HPP at 300 MPa for 5 min suggesting that holding abalone meat through rigor resolution will produce raw meat of equal texture attributes to unprocessed meat. In addition, the increased lightness of post-rigor HPP abalone processed at 300 MPa for 5 min produced a whiter meat, which is reportedly desirable to abalone consumers and could increase market value of highly pigmented abalones. Further investigation into the role of collagen in maintaining the texture of abalone meat during HPP is warranted, as well as exploration of the shelf-life of abalone processed at pressures below 500 MPa at different times.

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