QUALITY ASSESSMENT OF WEATHERVANE SCALLOP (*Patinopecten caurinus*) AND PURPLE-HINGE ROCK SCALLOP (*Crassadoma gigantea*) FROM ALASKA

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Abstract

In Alaska, the Pacific weathervane scallop (*Patinopecten caurinus*) is the only species commercially harvested. In the Eastern Gulf of Alaska, harvesters report occurrences of poor quality and difficult to market scallops from some fishing areas, designating the product 'weak meat' scallops. The purple-hinge rock scallop (PHRS, *Crassadoma gigantea*), has aquaculture potential. A recent grow-out study in Alaska was promising, verifying this potential. Our goal was to assess the quality of Alaskan scallops using physical and chemical analyses to understand the perceived differences in meat quality. 'Weak meat' (WS) and standard quality (SS) whole weathervane scallops from Yakutat, two batches of scallop muscle from Kodiak (KS1, KS2) and a group of whole PHRS from Southeast Alaska were sampled. Compared to KS1, KS2 and SS, muscle condition was lower ($P<0.05$), moisture content was higher ($P<0.05$) and glycogen content was below the detection limit (<0.62%) in WS. There were no differences ($P>0.05$) in proximate composition between PHRS and Kodiak scallops, however, PHRS had higher ($P<0.05$) levels of ω-6 fatty acids. Results confirmed anecdotal information from harvesters that WS was inferior in quality when compared to either SS or KS. Furthermore, we concluded that PHRS meat was of high quality, similar to Kodiak scallops.

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CHAPTER 1

General Introduction

1.1 Justification and Need

Scallops are distributed worldwide and the adductor muscle is a highly valued food source. There is a wide body of literature examining many aspects of scallops and many species (Hennick 1970, Whyte et al. 1990, Cranford 1995, Jeong et al. 1999, Shriver et al. 2002, Shumway and Parsons 2006, Pacheco-Aguilar et al. 2008, Makri 2009). Of the species found in Alaskan waters, the Pacific weathervane scallop (*Patinopecten caurinus* Gould 1850) is the only one commercially harvested. The purple-hinge rock scallop (*Crassadoma gigantea* Gray 1825), formerly classified as *Hinnites multirugosus* Gale 1928, has potential for aquaculture and is highly prized by local communities and harvested for subsistence in coastal Alaska.

The commercial weathervane scallop fishery is prosecuted by catcher-processor vessels that shuck, wash, grade, and freeze scallop muscles on board. The Eastern Gulf of Alaska (GOA) off Yakutat and the Western GOA off Kodiak Island are the main harvest areas in the state. The Kodiak area is known for its high productivity, and fast growing scallops with consistently high quality muscle (Rosenkranz 2011). However, vessel operators have noted that weathervane scallop muscles from some areas of the Eastern GOA are poor quality characterized by stringy texture, off-white to grayish color and a spongy consistency suggesting high moisture content (NPFMC 2008). The Alaska scallop industry calls these scallops 'weak meat' scallops and has difficulty marketing them. The fleet is forced to move to other harvest areas resulting in a negative economic

impact to the fishery and also underutilization of the Guideline Harvest Range (GHR) in the Eastern GOA. This has the potential to negatively impact the GHR for the following year and, consequently, the industry (Scallop Plan Team 2007). The 'weak meat' problem needs investigation into the potential biochemical origin but the biochemical profile of adductor muscle quality of 'weak meat' scallops has not been characterized. The first objective of this project was to compare some physical and biochemical properties of interest for two groups of weathervane scallops harvested off Yakutat. One group was harvested from a commercial fishing area known for its high numbers of 'weak meat' scallops, the other group from an area of standard quality scallops. Quality parameters of Yakutat scallops were also compared to commercially harvested weathervane scallops from Kodiak Island, AK, a region where 'weak meat' scallops have not been observed.

The second objective was to compare quality aspects of purple-hinge rock scallops harvested from Elfin Cove Oysters (Port Althorp, Southeast Alaska) with commercially harvested weathervane scallops from Kodiak. Aquaculture of purple-hinge rock scallop has been investigated in California and while results were promising, there were barriers to implementing commercial culture operations. Renewed interest has resulted in a recent grow-out study to determine the feasibility of culturing this species in Alaska. While purple-hinge rock scallops are locally prized, they would be a new species for the seafood market and meat quality assessment is necessary to promote market acceptance.

1.2 World Shellfish Fisheries and Aquaculture

In 2006, total world fisheries production was 144 million tonnes (mt), of which 92 mt were from capture fisheries and 52 mt from aquaculture production (FAO 2009). Since the early 1950's, when annual aquaculture production was less than 1 mt, there has been an annual growth rate in aquaculture production of almost 7 % while capture harvest has remained static for several years (Fig. 1.1). More than 110 mt (76%) of the total world fisheries production was used for human consumption and resulted in a per capita supply of food fish of 16.7 kg in 2006 (FAO 2009). Utilization of fish for human consumption was predominantly in the live and fresh form, followed by frozen, canned and cured (Fig. 1.2).

Bivalve shellfish i.e. oysters, clams, mussels and scallops contributed 2.3% and approximately $78.9 billion to world fisheries production in 2005. Between 1995 and 2005 growth in shellfish production was roughly 5% per year and increased from 9 to 13 mt (Pawiro 2009). Since 1995, aquaculture contribution to world shellfish production has increased from 78% of total production to 87%, while capture production has fallen from 22% to 13% of total production (Fig. 1.3, FAO 2007). China is the largest producer of shellfish (70%), followed by Japan (5.8%), the United States (US; 5.2%), South Korea (2.8%) and Thailand (2.8%); the remaining 13.4% comes from Canada, Chile, France, Italy and Spain combined (Pawiro 2009). In 2008, oysters and clams (33.8% and 37.1%, respectively) accounted for the bulk of world shellfish production followed by scallops (16.4%) and mussels (12.7%). Table 1.1 shows world shellfish production by species (FAO 2007).

1.2.1 World Scallop Fisheries and Aquaculture

At least 30 scallop species are commercially important. The distribution and significance of the major commercial species are listed in Table 1.2 (FAO 1990). World scallop production increased from 0.1 mt in 1950 to 2.2 mt in 2008, accounting for 1.4% of total world fishery production. In 1950, scallop production was entirely from capture. Scallop culture emerged in 1970 and by 2008 accounted for 65%, or 1.4 mt, of total world scallop production (Fig. 1.4) and almost 1% of world fisheries production. China led scallop culture production in 2008 with 1.2 mt worth $1.6 billion, followed by Japan, Chile and Peru (Table 1.3). Overall, scallops accounted for $\geq$ 45% of the global bivalve export market value in 2005 (Pawiro 2009).

1.2.2 United States Scallop Fisheries and Aquaculture

Atlantic sea scallop (*Placopecten magellanicus* Gmelin 1791), bay scallop (*Aequipecten irradians* Lamarck 1819), calico scallop (*Argopecten gibbus* Linnaeus 1758) and weathervane scallop are currently the most commercially important species in the US and have the greatest economic value (Hackney and Rippon 2000). In 2008, Atlantic sea, bay and weathervane scallops from the US accounted for 26.7% of the world's scallop capture and 9.2% of total scallop production (FAO 2007). Atlantic sea scallops dominate the domestic market with 99% of the total US scallop production, and New Bedford, MA has the distinction of being the main scallop port in the US (Turk 2000). Three other species, pink scallop (*Chlamys rubida* Hinds 1845), spiny scallop (*Chlamys hastata* Sowerby 1842) and purple-hinge rock scallop are important in discrete local areas but have little economic value (Fisher 2000).

Scallop culture is a well-established industry in some parts of the world. Japan began investigating the potential for scallop culture in the 1930's and commenced commercial production in the mid-1960's (Kosaka and Ito 2006). Building on Japan's success, and borrowing from the procedures developed there, China and Chile began their own scallop culture industries in the 1980's and all three countries have well developed economically important scallop culture industries (Guo and Luo 2006, von Brand et al. 2006). Also building on the technology developed in Japan, small but developing culture industries exist in eastern Canada, Russia, Britain, France, Norway, Ireland, Italy and Spain (Ivin et al. 2006, Norman et al. 2006, Parsons and Robinson 2006). In spite of the success of scallop culture elsewhere in the world, efforts in the US fail to meet domestic demand and remain limited to small scale operations of local economic significance (Blake and Shumway 2006).

1.2.3 Alaska Scallop Fisheries and Aquaculture

Weathervane scallops are the only scallop species commercially harvested in Alaska. The fishery began in 1967 in response to the collapse of the sea scallop fishery on the east coast of the US, and the decline of the red king crab (*Paralithodes camtschaticus* Tilesius 1815) fishery in Alaska (Kruse et al. 2005). The fishery started in Kodiak but expanded to Yakutat in 1968, the Alaska Peninsula in 1975, Southeast Alaska in 1980, Cook Inlet in 1983, the Bering Sea in 1987 and Prince William Sound in 1992 (Woodby et al. 2005). The development of this fishery is typical of most emerging fisheries and includes the following stages: discovery, bandwagon growth, fallback and redevelopment (Kruse et al. 2005). The fishery was largely unregulated until 1993 when

Alaska implemented an interim management plan. In 1994, a federal Fisheries Management Plan (FMP) was adopted that delegated management of the fishery to Alaska (Turk 2000). Statewide harvest for the 2007/2008 commercial season was 458,313 pounds of meats with a wholesale value of $2,722,379 (NPFMC 2009).

Alaskan shellfish growers are able to produce high quality shellfish products due to pristine water quality and low human activity in the shellfish farm localities. Oliveira et al. (2006) reported that Alaska maricultured oysters are of high quality, and often command a high market value. However, aquaculture production of scallops currently does not exist in Alaska. Weathervane, purple-hinge rock, pink and spiny scallops have been studied for their aquaculture potential in Alaska, but none have been viable to date (RaLonde 1993). The remoteness of shellfish farms in the state, transportation costs, state regulations, and long winters coupled to low water temperature considerably increase operational costs and contribute to the difficulties of scallop culture. MacDonald et al. (1991) indicate that pink and spiny scallops are too small and grow too slowly to be economically viable for aquaculture operations in Alaska. Collection of wild weathervane scallop spat has been unsuccessful, growth rates are too slow, and except for oyster spat under 20 mm, Alaska regulations prohibit the importation of shellfish spat (RaLonde 1993). Rock scallops have been extensively studied and successfully cultured to market size in as little as two years, however wild spat collection in Alaska has been unsuccessful (Monical 1980, Leighton and Phleger 1981, MacDonald and Bourne 1989, RaLonde 1993, Lauzier and Bourne 2006).

1.3 Scallop General Biology

The scallop family (Pectinidae) is a group of filter feeding bivalves containing over 30 genera and 350 species and occurring in all the worlds' oceans. The biology and anatomy of scallops have been thoroughly reviewed (Shumway and Parsons 2006). Scallops are monomyarian bivalves, having lost the anterior adductor and possessing only a single centrally located adductor muscle. This muscle is separated into two pieces that differ in size and function by connective tissue. The larger portion, called the quick muscle, is the highly prized meat and the dominant feature in the body. The quick muscle is capable of rapid contractions that open and close the valves allowing the animal to swim; a feature that sets scallops apart from other bivalves. The smaller portion, the catch muscle, contracts slowly and with considerable force allowing the valves to remain closed for extended periods with little energy expenditure (Chantler 2006). Scallops are a broadcast spawning species and release eggs and sperm into the water to be fertilized externally.

1.3.1 Pacific Weathervane Scallop

Weathervane scallops are found along the west coast from Point Reyes, California to the Eastern Bering Sea (Foster 1991). Scallop beds are discrete, elongated in shape and oriented north to south in the direction of the prevailing current. They are found at depths of 20-125 fathoms (37-228 meters), but commercial fishing effort is primarily at depths of 21-79 fathoms (38-144 meters) (Rosenkranz and Spafard 2010). Substrate type is typically sand, gravelly sand, or clayey silt (Turk 2000). Distribution is patchy throughout the range and commercial harvest is limited to areas of local abundance; the

Alaskan fishery is by far the largest, but small fisheries occur in Oregon and Washington (Lauzier and Bourne 2006).

The weathervane scallop is one of the largest and oldest lived species of scallop, reaching shell heights of up to 250 mm, meat weights of 340 g and 28 years of age (Kruse et al. 2005). Commercial catches typically do not exceed 180 mm shell height and 40 g meat weight (Hennick 1973). Annuli, or growth rings, are formed annually and scallops are sexually mature after three or four annuli are present. Yakutat scallops appear to grow more slowly and are smaller at maturity than Kodiak scallops (Hennick 1970).

Males and females of the species are separate, although one hermaphroditic specimen has been observed (Hennick 1971). Reproduction occurs at the expense of nutrient reserves stored during non-spawning periods. Scallops build reserves in the form of glycogen, lipid and protein and store them in tissues such as the digestive gland and adductor muscle. These reserves are utilized prior to spawning to build gonadal tissue, consequently lowering the quality of the adductor muscle; after spawning occurs the reserves are restored (Barber and Blake 1981). Timing of bivalve spawning has been linked to water temperature and food availability, with bivalves in more northern latitudes spawning in a single annual event (Malachowski 1988). Hennick (1970) reports that in Alaska, spawning is complete and takes place annually between early June and mid-July, presumably when plankton is abundant enough to sustain scallop larvae and replenish adult energy reserves.

1.3.2 Purple-hinge Rock Scallop

Purple-hinge rock scallops are found along the west coast from Alaska to Baja California from the low tide zone to depths of 80 m (44 fathoms) (Lauzier and Bourne 2006). Populations are locally abundant but distribution is patchy throughout its range. At about 20-30 mm, they abandon the free-swimming lifestyle seen in other scallops and permanently attach themselves to rocky substrate (MacDonald and Bourne 1989). Commercial harvest for purple-hinge rock scallops is not economically feasible due to distribution and difficulties harvesting scallops that have embedded into the substrate, however divers pry scallops out of rocks in recreational and subsistence fisheries (Leighton and Phleger 1981, MacDonald and Bourne 1989, MacDonald et al. 1991).

Like weathervane scallops, purple-hinge rock scallops are a large, long-lived species. A shell height of 250 mm has been reported, though 100 to 175 mm is more common (MacDonald and Bourne 1989, Leighton 1991, Lauzier and Bourne 2006). Shells become massive in comparison to other scallop species, and are often encrusted with plant or animal growth, making typical aging techniques impractical. Nonetheless, specimens of 27 years of age have been reported from British Columbia, Canada (Lauzier and Bourne 2006). The spawning season of purple-hinge rock scallop is poorly defined, and literature information scarce. Lauzier and Bourne (2006) reported that purple-hinge rock scallop males and females are separate, although one hermaphroditic specimen has been observed. Lauren (1982) registered incomplete spawning in June for this species in Puget Sound, Washington with development of unspawned oocytes continuing until September. Furthermore, bimodal spawning has been observed in San Diego, California

and Puget Sound, Washington; however, in Humboldt Bay, California spawning occurred throughout the year (Malachowski 1988). Local environmental factors such as water temperature, food availability or tidal fluctuations may negatively impact larval survival, and the poorly defined spawning cycle seen in purple-hinge rock scallops may be an adaptation to offset larval mortality (Malachowski 1988).

1.4 Scallop Quality Parameters

Shellfish quality refers to aspects of consumer preference such as freshness, nutrition and the sensorial attributes of color, texture and flavor (Boulter 1996). Storage and handling practices during rigor mortis, resolution of rigor mortis, nucleotide degradation (autolysis) and microbial growth affect freshness, flavor and texture. Nucleotide degradation and microbial growth specifically affect the freshness and flavor of fish and shellfish (Ocano-Higuera et al. 2006). Tenderness (texture) is an important aspect of scallop quality and can be measured by both instrumental and sensorial methods (Chung and Merritt 1991). The autolytic, microbial and textural changes in scallops under various storage and handling practices have been widely reported (Hiltz and Dyer 1973, Maxwell-Miller et al. 1982, Chung and Merritt 1991, Ocano-Higuera et al. 2006, De Vido de Mattio et al., 2009 Makri 2009).

Quality can also refer to the physiological condition of an animal and its ability to respond to its environment. Maguire et al. (1999) used a combination of physiological, biochemical and behavioral tests, including condition indices and carbohydrate content, to assess the response of scallops to acute and chronic stress. Condition indices measure the physiological activity of an animal at a given time or environmental condition and can

be computed for specific tissues (i.e., adductor muscle, gonad) in relation to other tissues after taking morphometric measurements (Beltran-Lugo et al. 2005). Biochemical composition is an important quality indicator and is closely associated with condition indices. Storage of nutrient reserves in the adductor muscle contributes to the nutritional value of the meat and also provides the reserves necessary for processes such as gametogenesis (King et al. 1990, Ocano-Higuera et al. 2006)

1.5 Objectives

The goal of this project was to assess Alaskan scallops to better understand the perceived differences in muscle quality. A variety of morphological characteristics, chemical composition and texture analysis were used to measure quality. Specific objectives included:

1. Quantification of the variability in muscle quality of 'weak meat' and standard weathervane scallops from Yakutat, AK and comparison to prime quality weathervane scallops from Kodiak, AK,
2. Characterization of muscle quality of purple-hinge rock scallops and comparison to prime quality weathervane scallops from Kodiak, AK.

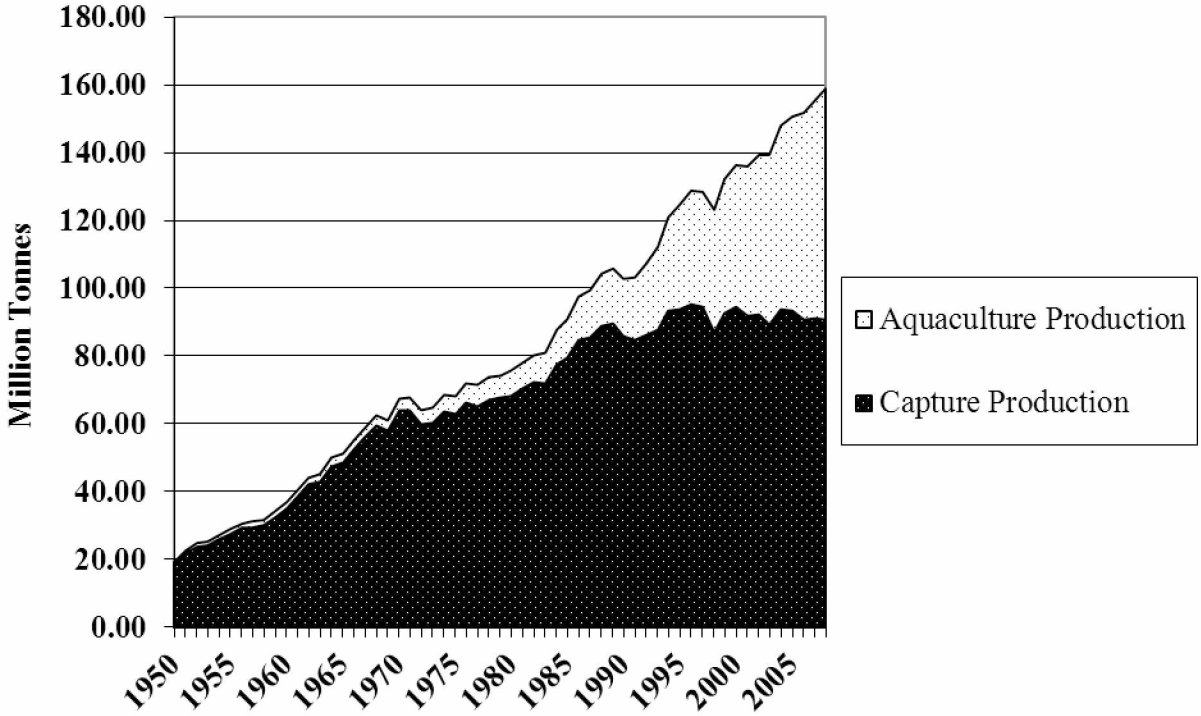

Figure 1.1. World fishery production, 1950-2008 (FAO 2007).

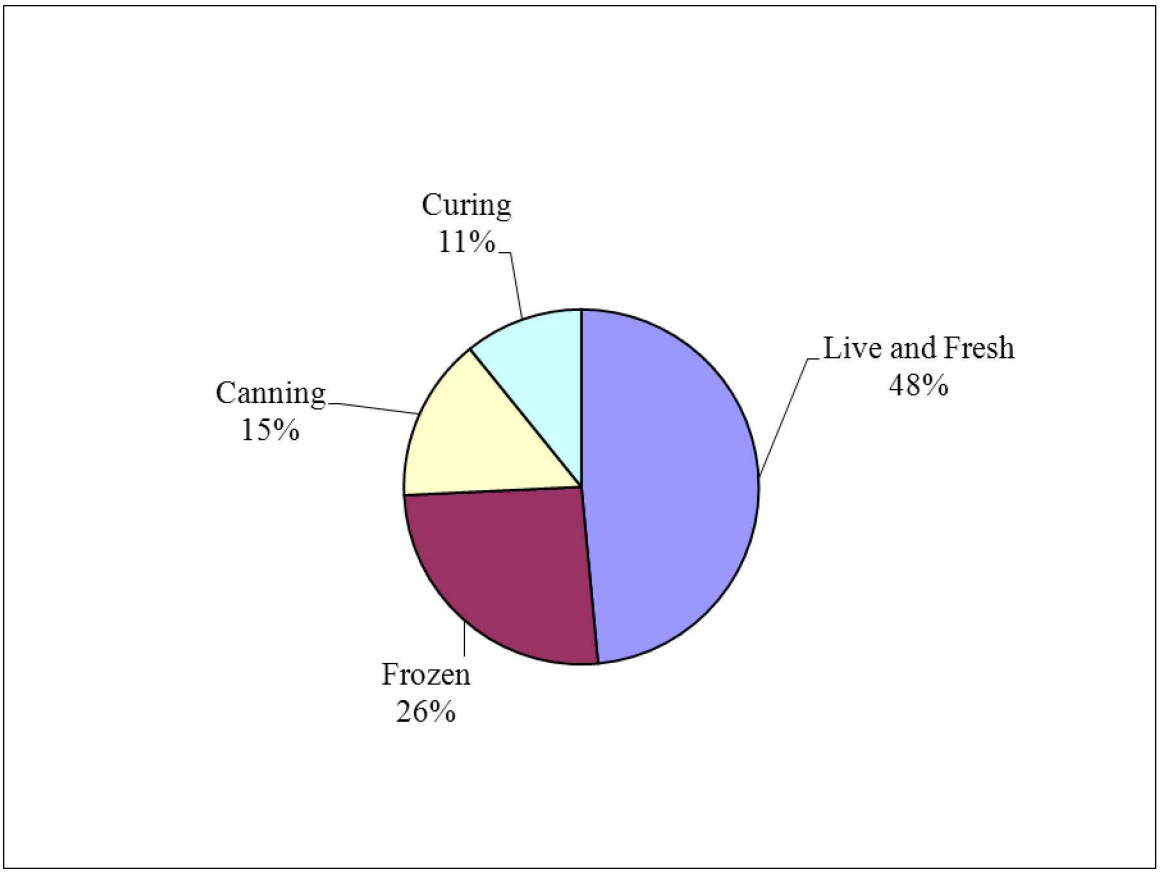

Figure 1.2. Utilization of world fishery production for human consumption in 2006 (FAO 2009).

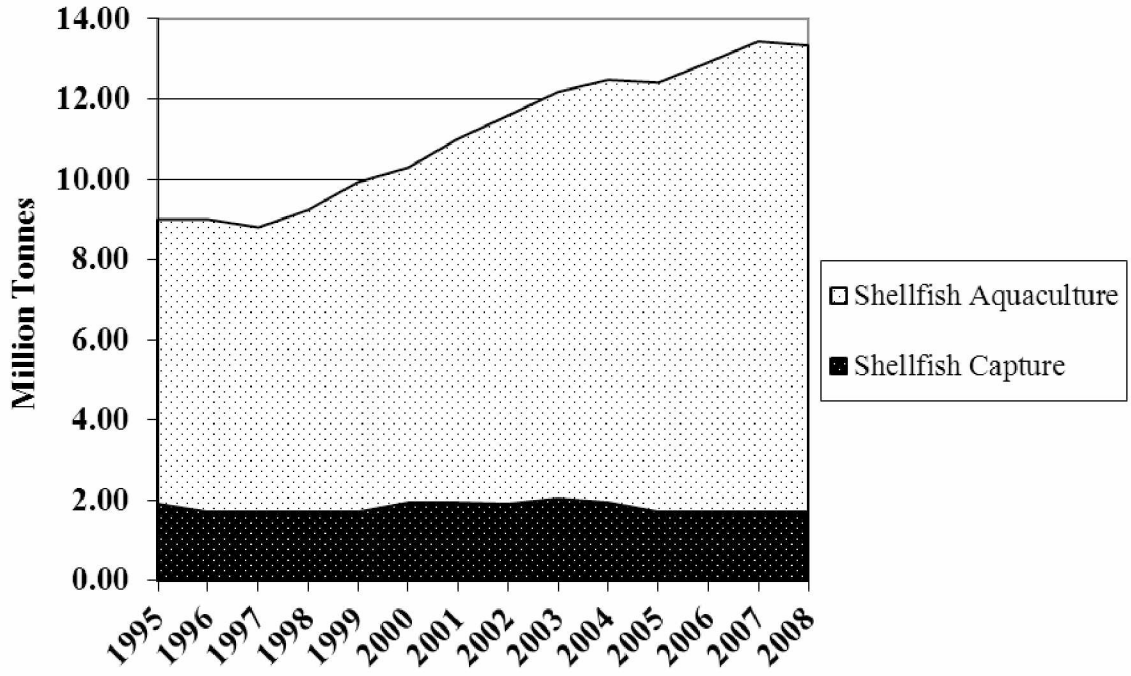

Figure 1.3. World shellfish production, 1995-2008 (FAO 2007).

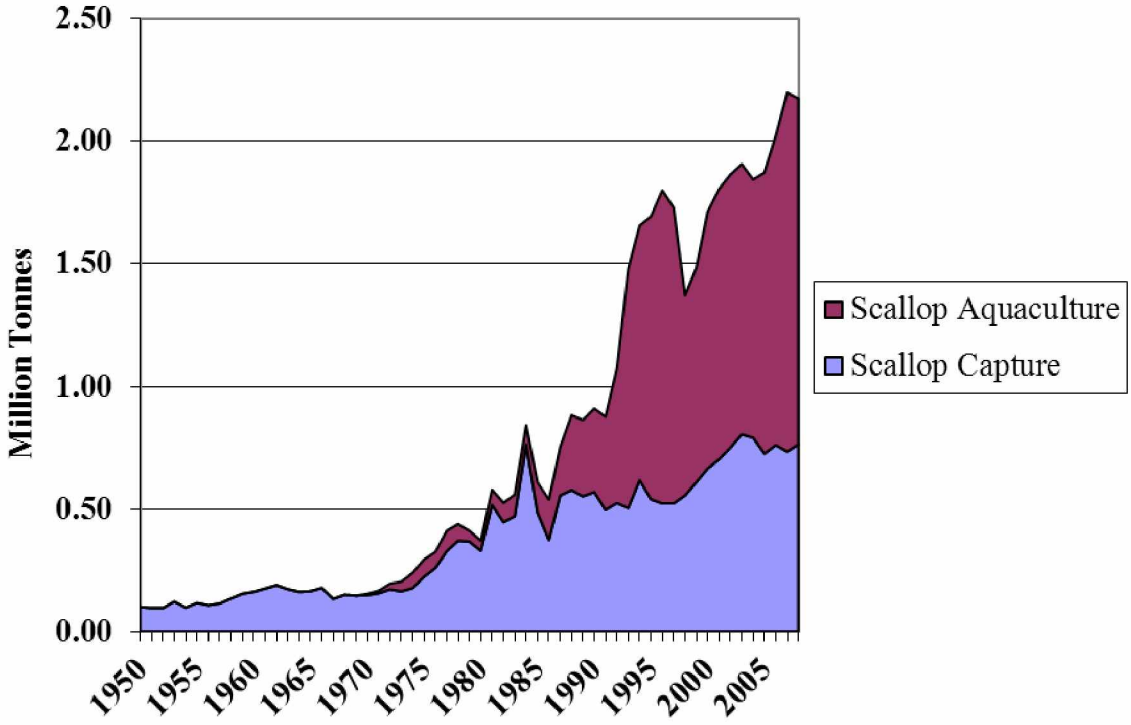

Figure 1.4. World scallop production, 1950-2008 (FAO 2007).

Table 1.1.

World shellfish production (million tonnes) by species, 1995-2005 (FAO 2007).

Year	Oyster	Clams, cockles and arkshells	Scallops (Pectens)	Mussels
1995	3.24	2.73	1.69	1.35
1996	3.22	2.70	1.80	1.29
1997	3.12	2.65	1.73	1.32
1998	3.43	2.91	1.37	1.51
1999	3.55	3.33	1.48	1.59
2000	3.86	3.15	1.71	1.57
2001	3.98	3.63	1.80	1.62
2002	4.06	3.88	1.86	1.78
2003	4.21	4.27	1.91	1.81
2004	4.29	4.49	1.84	1.86
2005	4.32	4.38	1.87	1.85
2006	4.40	4.56	2.02	1.93
2007	4.55	4.98	2.20	1.71
2008	4.29	5.17	2.17	1.71

Table 1.2.

Distribution and economic significance of major scallop species (FAO 1990).

Ocean	Species	Countries	Significance
Atlantic			
N -E Atlantic	*Pecten maximus*	France, U.K, Spain	+++
	Chlamys varia	France, Spain	++
	Chlamys opercularis	Norway	++
N-W Atlantic	*Placopecten magellanicus*	Canada, U.S.A.	+++
	Argopecten irradians	U.S.A.	+++
S-W Atlantic	*Chlamys purpuratus*	Chile, Argentina	+
Mediterranean	*Pecten maximus*	Italy, Greece	++
Black	*Chlamys farreri*	Algeria	+
Pacific			
N-E Pacific	*Patinopecten caurinus*	Canada, U.S.A.	++
N-W Pacific	*Pecten yessoensis*	Japan, USSR	+++
	Chlamys farreri	China	++
S-E Pacific	*Chlamys farreri*	Chile	+
S-W Pacific	*Pecten alba*	Australia	+++
	Amusium balloti	New Zealand	++

Table 1.3.

Scallop culture production by country in 2008 (FAO 2007).

Country	Tonnes	$1000
China	1,148,194	1,621,735
Japan	225,600	338,400
Chile	21,277	270,218
Peru	14,802	139,404
Republic of Korea	421	2,251
Canada	309	1,294
Russian Federation	85	442
Ireland	60	516
United Kingdom	42	291
Norway	28	32
Channel Islands	9	34
Brazil	3	15

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CHAPTER 2

Quality assessment of weathervane scallops (*Patinopecten caurinus*) from Alaska[1]

2.1 Abstract

Weathervane scallops (*Patinopecten caurinus*) have been commercially harvested in Alaska since 1967. From the beginning, vessel operators in the Eastern Gulf of Alaska have reported poor scallop muscle quality due to 'weak meats', characterized by high moisture content and off-white to grayish color. The Alaska scallop industry has difficulty marketing poor quality scallops. The objective of this research was to quantify variability in muscle of weathervane scallop. Two groups of 40 whole scallops each, weak (WS) and standard (SS), from Yakutat and two batches (17 individuals and 30 individuals) of muscle from Kodiak (KS1 and KS2, respectively) were sampled. Physical measurements and chemical composition analyses were conducted to determine quality differences between groups. Moisture content was significantly ($P<0.05$) higher in WS than SS. Glycogen content was similar ($P<0.05$) for SS, KS1 and KS2 but was below the detection limit in WS. Muscle condition indices were significantly ($P<0.05$) lower in WS than in SS. The ω-3 levels were lowest and ω-6 levels were highest in WS, resulting in the lowest ω3/ω6 ratio for WS ($P<0.05$). Results indicate that WS are lower in overall quality than SS and Kodiak scallops. Further investigation into the causes of 'weak meat' scallops is warranted.

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[1] Brenner, K. A., A. C. M. Oliveira, G. Rosenkranz, R. Burt, M. Spafard, P. J. Bechtel, C. A. Crapo & R. RaLonde. 2011. Quality assessment of weathervane scallops (*Patinopecten caurinus*) from Alaska. Prepared for submission to the Journal of Shellfish Research.

2.2 Introduction

The Pacific weathervane scallop (*Patinopecten caurinus*) is distributed along the west coast of North America from Point Reyes, CA to the Bering Sea (Foster 1991). The highest abundances in Alaska occur in the Western Gulf of Alaska (GOA) off Kodiak Island, in the Eastern GOA off Yakutat, and in the Bering Sea (Kruse et al. 2000). Weathervane scallops have been commercially harvested in Alaska since 1967, in response to the collapse of the sea scallop fishery on the east coast of the US. From 1994-2002 the eastern GOA region around Yakutat accounted for 31% of statewide scallop landings. Currently, Kodiak and Yakutat still dominate landings but other harvest areas include the Alaska Peninsula, Southeast Alaska, Cook Inlet and the Bering Sea as seen in Fig. 2.1 (Kruse et al. 2005).

The commercial weathervane scallop fishery in Alaska is prosecuted primarily by catcher-processor vessels. Scallops are shucked then muscles are washed, graded, packaged, and plate frozen onboard. The Kodiak area is known for its high productivity, and fast growing scallops with consistently high quality muscle (Rosenkranz 2011). However, vessel operators harvest weathervane scallops from some areas of the Eastern GOA that have poor quality muscles characterized by stringy texture, off-white to grayish color and a spongy consistency suggestive of high moisture content (NPFMC 2008). The Alaska scallop industry calls these scallops 'weak meat' scallops and has difficulty marketing them. The fleet is forced to move to other harvest areas resulting in underutilization of the Guideline Harvest Range (GHR) in the eastern GOA. This has the

potential to negatively impact the GHR for the following year and has a negative economic impact on the industry (Scallop Plan Team 2007).

The 'weak meat' problem could have a biochemical origin but the biochemical profile of adductor muscle of 'weak meat' scallops has not been characterized. Therefore, the objective of this research was to compare physical and biochemical properties for two groups of weathervane scallops harvested in Yakutat, AK. One group was harvested from a commercial fishing area known for its high numbers of 'weak meat' scallops, the other group from an area of standard quality scallops. Muscle quality parameters of Yakutat scallops were compared to weathervane scallops from Kodiak Island, AK, a region where 'weak meat' scallops have not been observed.

2.3 Materials and Methods

2.3.1 Sample Procurement

One poly bag (1.13 Kg) of weathervane scallop meats were obtained from the F/V Provider in April 2009 (KS1) and ~6.9 Kg (three 2.3 Kg boxes, two poly bags/box) of weathervane scallop meats harvested in August 2009 were purchased at a local seafood market (Island Seafoods, Kodiak, AK) in October 2009 (KS2). In July and August 2009, forty whole 'weak meat' scallops (WS) and forty whole standard scallops (SS) were collected from the F/V Arctic Hunter in the Eastern GOA by Alaska Department of Fish and Game (ADFG) observers. The SS samples were harvested from a depth of 36 fathoms from scallop beds currently fished by scallop fishermen, while WS samples were harvested from a depth of 40 fathoms from beds abandoned by scallop fishermen due to the presence of poor quality scallops. Specimens were frozen whole and shipped to the

Fishery Industrial Technology Center in Kodiak, AK for analysis. All samples were kept frozen at -30°C for one to two months until processed.

2.3.2 Morphology, Age and Condition

Morphometric measurements were recorded for WS and SS. Wet weights of whole scallop, shell, gonadal-visceral (body) tissue, and adductor muscle (meat) were determined using a Sartorius digital scale with manufacturer accuracy of 0.01g (Model LP2200P, Gottingen, Germany). Shell height, width, and depth and meat height and width were measured using electronic digital calipers (200 mm range, VWR, Radnor, PA). Measurements for KS included meat weight for KS1 and meat height and weight for KS2. The WS and SS scallops were shucked after whole weights were recorded and the adductor muscle (meat) and gonad-viscera (body) were separated. Shells were gently cleaned under running tap water with a brush, and transported to the ADFG Kodiak Island facility. Scallop body and meat tissues were individually frozen and stored at -30°C for two months until analysis.

Age was determined from shells of WS and SS in the ADFG lab by counting annuli (growth rings) on the upper valve. This technique has been used extensively to determine mollusk growth and has been used successfully for weathervane scallop (Haynes and Hitz 1971, Ignell and Haynes 2000). Muscle condition index (MCI), was calculated for WS and SS based on that of Beltran-Lugo et al (2005) in order to assess physiological condition of the meat relative to total soft tissue: MCI (%) = weight of adductor muscle/weight of total soft tissues x 100.

2.3.3 Sample Preparation

Individual scallops were too small to be assayed for all chemical analyses so pooling was necessary. A pooling methodology applied by Linehan et al. (1999) was adopted. Linehan et al. (1999) determined that the value of any assay obtained from a sample pool of oysters was not significantly different than the weighted average obtained by analysis of single individuals, demonstrating that this strategy could be used to meet tissue requirements for laboratory analysis. This pooling scheme was successfully applied for biochemical analysis of Alaska oyster samples (Oliveira et al. 2006).

Scallop meats were thawed at 4°C. The KS1 meats were randomly pooled into three samples of three meats and two samples of four meats (n=5), and for the KS2 samples, five meats were randomly pooled (n=6). The WS (n=8) and SS samples (n=8) were pooled into eight samples of five meats each using constrained randomization to maintain consistent weight between samples. Samples were homogenized using a Cuisinart Mini-prep model DLC-1SS (Cuisinart Corp., East Windsor, NJ) until a uniform paste was obtained (± 2 min). Homogenized samples were stored in zip-lock bags and kept at 4°C throughout analysis.

2.3.4 Proximate Composition

Percent moisture and ash were determined according to AOAC methods 952.08 and 938.08, respectively (Helrich 1990). Percent nitrogen was determined using a LECO model TruSpec N nitrogen analyzer (LECO Corp., St. Joseph, MI). Percent protein was calculated by multiplying %N by 6.25 (Helrich 1990). Lipid was extracted using the method described by Folch et al. (1957) with slight modifications described below. A

quantity of 12 g of tissue was combined with 90 mL of a 2:1 mixture of chloroform:methanol with 0.01% butylated hydroxytoluene (BHT), mixed vigorously with a glass stir bar, and refrigerated under nitrogen atmosphere for 24 h. Contents were filtered through a 42 mm Buchner funnel and transferred to a 125 mL separatory funnel. A volume of 18 mL of 0.88% (W:V) KCl was added to the separatory funnel and the contents were thoroughly shaken and allowed to sit until clear. The lower phase was filtered through anhydrous sodium sulfate and the solvent was evaporated using a TurboVap-LV Evaporator (Caliper Life Sciences, Hopkinton, MA) operated at 40°C under a nitrogen atmosphere. Lipid content was determined gravimetrically.

The total carbohydrate content was determined using the anthrone method, a combination of the methods of Strickland and Parsons (1972) and Clegg (1956) as previously described by Oliveira et al. (2006). The anthrone reagent was prepared by dissolving 0.2 g of anthrone (Sigma-Aldrich, St. Louis, MO) in 8 mL of HPLC-grade ethyl alcohol (VWR Inc., Radnor, PA) and 30 mL of distilled water. After complete dissolution of the anthrone in the solvents, 100 mL of concentrated sulfuric acid was added to the mixture. A quantity of 15 g of homogenized tissue was mixed with 10 mL of distilled water and 13 mL of 52% perchloric acid (VWR) in a 100 mL capped flask. The contents were stirred for 30 min at room temperature using a magnetic bar and a standard laboratory stir plate (VWR). The mixture was transferred to a 100 mL volumetric flask, diluted to 100 mL and filtered using Whatman #4 filter paper into a 250 mL volumetric flask. The filtrate was diluted to 250 mL, and a 10 mL portion was transferred to a 100 mL volumetric flask and diluted to 100 mL. A volume of 1 mL was

transferred to a 25 mL screw-top tube, 5 mL of anthrone reagent was added, and the tube was placed in a boiling water bath for 7 min. The tubes were rapidly cooled in an ice water bath and solutions were read at 624 nm on a Cary 50 UV-Visible Spectrophotometer (Varian Instruments, Walnut Creek, CA). In parallel, 1 mL of distilled water was used as the blank, and 1 mL of a 0.1 mg/mL solution of oyster glycogen Type II (Sigma-Aldrich) was used as the standard. Blank and standard solutions were subjected to identical colorimetric reaction as described for scallop meat samples. Percent total carbohydrate was calculated using the following formula:

% Total carbohydrate = (25 x A2)/(A1 x W)

Where:

W = weight of sample (g)

A1 = absorbance of glycogen standard

A2 = absorbance of sample

It is important to point out that in the case of scallop meats, carbohydrates in the tissue are predominantly present as glycogen (Barber and Blake 1981, Brokordt et al. 2000). Therefore, % total carbohydrate quantified was reported and discussed as % glycogen. Glycogen content for the KS1 group was determined by subtraction: % glycogen = 100 - (% moisture + % protein + % ash + % lipid).

2.3.5 Lipid Classes

Lipid class analysis was based on the methods of Parrish (1987) and Ackman et al. (1990), as described by Oliveira and Bechtel (2005) and was carried out on an Iatroscan^{TM} TLC/FID Analyzer (Model MK-6s, Iatron Laboratories, Tokyo, Japan). The

hydrogen flow rate was 160 mL/min, air flow was 1.6 L/min and scan time was 30 s. The six standards used to identify lipid class were tripalmitin (triacylglycerol, TAG), palmitic acid (free fatty acids, FFA), (S)-1,2-dipalmitin (diacylglycerol, DAG), cholesterol (sterol, ST), DL-α-palmitin (monoglyceride, MG) and L-α-phosphatidylcholine (phospholipid, PL) and were obtained from Sigma-Aldrich. Solvent tanks (Shell-USA, Fredericksburg, VA) were lined with Whatman #2 filter paper and 159.3 mL of solvent was added. The closed system was allowed to equilibrate for 20 min. The solvent system was hexane:diethyl ether:formic acid (80:25:1.2). Sample solutions contained 13 ug/μL of oil in chloroform and composite standard solution contained a mixture of 16.6 ug/μL of each standard in chloroform. For each rack of 10 rods (Chromarods-SIII, Iatron Laboratories, Tokyo Japan), a 1 μL portion in five 0.2 μL aliquots was spotted for each sample (rods 2 to 9) and composite standard (rods 1 and 10). Chromarods were suspended in the solvent tank for 10 min before being lowered into the solvent for 30 min. Developed rods were dried at 105°C for 3 min before being scanned. Peaks were integrated using the Peak Simple Program (v. 2.83, SRI Instruments, Torrence, CA).

2.3.6 Fatty Acid Methyl Ester Profiles

Fatty acid methyl esters (FAME) were prepared according to the procedure of Maxwell and Marmer (1983) using 1 mg of methyl tricosanoate (Sigma-Aldrich) as the internal standard. FAMEs were quantified as previously described by Oliveira et al. (2006), with the exception that the sample injection volume was 2 μL.

2.3.7 Texture and pH

Twenty scallop adductor muscles from groups KS2, WS and SS were selected for texture analysis. For WS and SS, the meat weights of all 40 scallops were listed in descending order and the meats with the middle 20 weights were chosen. For KS2, seven random scallop meats were pulled from two of the 2.3 Kg boxes, and 6 meats were randomly pulled from the other 2.3 Kg box. Texture was measured using a TA-HDi Texture Analyzer (Texture Technologies Corp., Scarsdale, NY) equipped with a P/0.5 flat end probe set to travel 40 mm/min with a 5 kg load cell. Total distance travelled was 40 mm. The peak force (g) necessary to puncture the meat was recorded based on surimi gel puncture tests (Park 2005) using Stable Microsystems Texture Expert Exceed software (v. 2.64, Scarsdale, NY).

The pH was determined on the pooled meats for WS, SS and KS2 using the temperature compensated WTW pH3400i handheld pH meter with the WTW model 103702 probe attached (Weilheim, Germany). The probe was two-point calibrated to pH 4 and pH 7 before use and inserted directly into the sample immediately after tissue homogenization.

2.3.8 Statistical Analysis

Data were checked for normality by the Kolmogorov-Smirnoff test prior to statistical analysis. Non-parametric analysis was used because data did not comply with normal distribution assumptions. In cases where three or more groups were compared a Kruskal-Wallis ANOVA was used. Multiple comparisons of mean ranks for all pairs of groups were run post hoc to determine significances ($P<0.05$). In the case of two groups

being compared a Mann-Whitney U t-test was performed. Analysis was carried out using Statistica (v.10, StatSoft, Tulsa, OK).

2.4 Results

2.4.1 Morphology, Age and Condition

Table 2.1 shows that WS were significantly larger in whole weight, meat weight, body weight and shell height than SS, though yields of meat were significantly lower ($P<0.05$) in WS than SS. Morphological data is in agreement with the observation that WS were significantly older (14 annuli) than SS (5 annuli), as depicted in Fig. 2.2. Muscle Condition Index was significantly lower in WS than SS (Fig. 2.3), and this result is in agreement with anecdotal information from scallop harvesters in the Yakutat region.

2.4.2 Proximate Composition

Significant differences were observed between groups of scallop samples ($P<0.05$) (Table 2.2). The WS group had the lowest protein content and KS1 and KS2 the highest, with SS having intermediate protein values. Moisture content was inversely related to protein content, and the WS group had the highest moisture values and KS samples had the lowest. The ash content of WS group was about 1% higher than values determined for scallops from Kodiak, and this difference was statistically significant ($P<0.05$). Interestingly, WS measured no detectable levels of glycogen, whereas SS, KS1 and KS2 samples had glycogen content between 1.6% and 2.2%. Lipid content was generally low (<1%) and not significantly different between groups. The moisture to protein ratios (M:P) were significantly higher in WS samples than in KS1 and KS2 samples. Lower protein content and absence of glycogen in WS samples suggest that

these scallops are of inferior quality to SS scallops, which had comparable gross composition to Kodiak scallops.

2.4.3 Lipid Classes

Scallop adductor muscle is a lean tissue with lipid content ≤ 1%, and as expected most extractable lipids were phospholipids (Table 2.3). Phospholipids constituted a minimum of 81.6% and maximum of 90.8% of the lipid classes investigated, with KS2 having the highest content and WS the lowest. Triacylglycerols were below the detection limit (0.5%), except for KS1 which had only trace amounts. Significant differences ($P<0.05$) were detected in the levels of free fatty acids and WS had the highest content and KS2 the lowest. The contribution of sterols to the total lipid classes of the scallops followed a similar pattern to the one observed for free fatty acids, with WS having the highest proportion of this lipid class and KS2 the lowest.

2.4.4 Fatty Acid Methyl Ester Profiles

Fatty acid methyl ester (FAME) classes and the most abundant FAMEs in each class are reported in Table 2.4. The most abundant FAMEs in all scallop meat groups were the saturated FAMEs (SAT) 16:0 and 18:0, the monounsaturated FAMEs (MUFA) 18:1ω9 *cis* and 18:1ω7, and the long chain polyunsaturated FAMEs (PUFA) 20:5ω3 (EPA) and 22:6ω3 (DHA). Palmitic acid (16:0), EPA and DHA had the highest levels and comprised 69.2% of the total extracted lipids (15.5%, 19.3% and 34.4%, respectively). WS had the lowest levels of palmitic acid and EPA while KS2 had the lowest DHA levels. PUFAs constituted about 62.1% of total extracted lipid for all groups, followed by SAT at 24.0% and MUFA at 12.3%; however, PUFA levels were

significantly higher ($P<0.05$) in WS and SS. WS had significantly lower levels of total ω-3, higher levels of total ω-6, and consequently, the lowest ω-3:ω-6.

2.4.5 Texture and pH

Peak force measurements showed that the WS were considerably tougher than SS and KS2 (Fig. 2.4), requiring significantly more force ($P<0.05$) to puncture the muscle. All groups were below pH 7 and WS had significantly higher pH than SS and KS2 (Fig. 2.5).

2.5 Discussion

2.5.1 Morphology, Age and Condition

Age and size data (shell height) of both WS and SS are consistent with catch data from ADFG. The size of retained scallops from the Yakutat area from 1998/99-2006/07 ranged from 100-165 mm, with the majority being between 110-135 mm (NPFMC 2009). From 2005/06-2008/09 the age of commercially retained scallops was 4-21 years and for the 2008/09 season, the majority of the retained scallops were between 7 and 16 years (Rosenkranz unpubl. data). The older age and larger overall size of WS compared to the SS is likely due to harvesters abandoning areas of known WS and they are left to grow undisturbed.

Lucas and Beninger (1985) state that condition indices (CIs) are used as a measure of the physiological state of an animal at a given time. The CIs are influenced by many factors including food quality and quantity (Shriver et al. 2002) and spawning (Li et al. 2008). Meat size is influenced by food supply, water flow velocity, water depth and spawning cycle (Sarro and Stokesbury 2009). The lower MCI and the lower % yield

of meat in WS is an indication that energy is not being partitioned to the meat, or is being utilized by tissues rather than stored. Water depth and spawning cycle influences are unlikely to be factors due to the similar depth and season of harvest of WS and SS, however cumulative stress may trigger different responses for scallops within each of these groups and this possibility cannot be dismissed.

2.5.2 Proximate Composition

The proximate composition of the scallop meats analyzed in this study is within the ranges previously reported for scallops of both the same and other species (Webb et al. 1969, Krzeczkowski et al. 1972, Naidu and Botta 1978, Maxwell-Miller et al. 1982, Anthony et al. 1983, King et al. 1990, Silva and Chamul 2000). Overall, there is scant data available specific to weathervane scallops so exact comparisons of our results to literature values are difficult. However, the general biology and energy utilization of scallops enables an understanding of the differences observed.

The meat and digestive gland are the primary storage sites of nutrient reserves in scallops during periods of somatic growth when food is abundant. Carbohydrate, in the form of glycogen, and protein are stored in the meat, while lipid is stored in the digestive gland (Barber and Blake 1981). These reserves are utilized to support maintenance metabolism during periods of food scarcity and during gametogenesis and in oysters, glycogen may be used for gamete formation during times of stress (Barber and Blake 2006, Liu et al. 2010). All scallop meats in this study had low lipid levels, consistent with the observation that the meat is not a primary storage site for lipid. Protein and glycogen levels of the meats are similarly consistent with regard to how scallop store

energy. The lower protein content and lack of glycogen in WS is an indication that they are under the influence of some stressor.

Li et al. (2008) found that post-spawning Pacific oysters (*Crassostrea gigas*) have a reduction in glycogen as an adaptation to food deprivation. Howgate (2009) states that the glycogen content at death of animals within a species depends upon factors such as nutritional status at the time. Bayne (1973) found that post-spawning oysters postpone or prolong the recovery process including rebuilding glycogen storage. Brokordt et al. (2000) concluded that mobilization of glycogen and protein from adductor muscle to form gametes reduced muscle metabolic capacity and the ability of scallops to recover from exhausting exercise such as predator evasion. Starvation studies conducted on Pacific oyster (Liu et al. 2010) show that over a 90 day period, adductor muscle of starved oysters had a rapid decline in glycogen content followed by declines in protein and lipid content with concomitant increases in ash and moisture levels. Likewise, Phleger et al. (1978) observed markedly reduced glycogen levels in starved aquarium-held purple-hinge rock scallops. A high proportion of water in tissue is an indicator of depleted energy reserves (Lucas and Beninger 1985), which is seen in both the high moisture content and high M:P ratio of WS.

While SS appear to be in better physiological condition based on composition than WS, their condition was not as good as that observed in Kodiak scallops. Quality, from the standpoint of marketability and consumer acceptance is largely based on highly subjective sensory attributes such as color, raw and cooked odor, flavor, and texture and food safety issues such as spoilage (Boulter 1996). In spite of natural and seasonal

variation, many government agencies have tried to establish upper limits of moisture content due to the post-harvest handling practices used in some scallop fisheries. For a time in the 1990's, scallops with moisture content >80% (USA) and >81% (Canada) were required to be labeled as water added (DuPaul et al. 1996). France has established a maximum M:P ratio of 5:1 (Boulter 1996, DuPaul et al. 1996). Based on these guidelines, Kodiak and SS scallops were well under the upper limits, while WS were above the limits.

2.5.3 Lipid Classes

The high phospholipid and sterol contents relative to triacylglycerol content observed in meats in this study are consistent with results from other scallop studies (Napolitano and Ackman 1992, Napolitano et al. 1992, Linder and Ackman 2002, Copeman and Parrish 2004, Palacios et al. 2007). Due to its low lipid content, scallop adductor muscle is considered a 'lean organ' along with gills and mantle. Napolitano and Ackman (1992) observed for sea scallops (*Placopecten magellanicus*) that sterols and phospholipids are the predominant lipid classes in lean organs, whereas triacylglycerols levels are very low. Copeman and Parrish (2004) had similar results with Icelandic scallop (*Chlamys islandica*) adductor muscle as did Palacios et al. (2007) with Pacific lion's paw scallop (*Nodipecten subnodosus*). This is consistent with the fact that phospholipids and sterols are structural lipids comprising cell membrane, while triacylglycerols are energy storage lipids. As observed in prior studies, phospholipids were inversely proportional to TAG content (Bechtel and Oliveira 2006). Ackman and Gunnlaugsdottir (1992) also observed this when measuring neutral and polar lipids in

Japanese sardines with variable lipid content. They reported that sardines with a total lipid content of 0.89% (w/w) had polar and neutral lipids fractions that were 0.7 and 0.19% of the total lipid content, respectively; however, when sardines had 8.66% (w/w) lipids, the polar and neutral lipids fractions were 0.76 and 7.9% of the total lipid content, respectively. While triacylglycerols were detected in one group of samples (KS1) and not in the other three groups, it still comprised only 1% of the total lipid and its presence was likely attributable to seasonal differences in diet composition and/or reproductive stage at time of harvest. Pernet et al. (2003b) found higher triacylglycerol levels in sea scallop (*P. magellanicus*) larvae fed diets rich in this lipid class, and in general levels determined were highly variable between diet and time (Pernet et al. 2003a).

Free fatty acids are usually present in trace amounts in marine organisms and scallops are no exception. Napolitano and Ackman (1992) found ~1% FFA in sea scallop meat in winter and trace levels in summer. Copeman and Parrish (2004) found <1% FFA in the meat of Icelandic scallop. With the exception of KS2, the free fatty acid levels in meats from this study were higher than observed for other scallop species. The presence of large quantities of free fatty acids in tissue is an indication of lipid degradation and breakdown of the structural lipids (Jeong et al. 1999, Pernet et al. 2003a). Lipid degradation can occur during long term storage, or when tissues are improperly stored. WS and SS were frozen whole and high enzymatic activity, typical of tissues such as digestive glands, may have caused hydrolysis of lipids and formation of free fatty acids in the adductor muscle tissue during frozen storage. The slight differences in the free fatty

acid values observed between KS1 and KS2 may be due to the fact that KS1 scallops were frozen for a longer period of time.

2.5.4 Fatty Acid Methyl Ester Profiles

Fatty acid analysis of many species of scallop, including weathervane scallop, have shown that polyunsaturated fatty acids account for about 60% of the total lipids, while saturated and monounsaturated fatty acids account for about 25-30% and 10-15%, respectively (Gruger Jr. et al. 1964, Krzeczkowski et al. 1972, Napolitano et al. 1992, Linder and Ackman 2002, Copeman and Parrish 2004) and the results from the present study were similar. The predominant FA 22:6ω3, 20:5ω3, 18:1ω9 and 16:0 are typical components of phospholipids (Richoux et al. 2005).

The higher levels of PUFA detected in WS and SS could be attributed to temperature, depth and/or diet. Krzeczkowski et al. (1972) found that scallops originating from cold deep environments (40-60 fathoms) have higher PUFA content and the level of unsaturation of polar lipid fatty acids is inversely related to the ambient temperature (Bell et al. 1986, Napolitano et al. 1992). In addition, marine fish and bivalves lack the elongation and desaturation enzymes necessary to synthesize long chain PUFAs and both EPA and DHA are considered nutritionally essential (Bell et al. 1986, Whyte et al. 1990).

The ratio of ω3 to ω6 is used to compare the relative nutritional value of foods because in order to gain the health benefits of ω-3 FA, an increase in ω-3 FA consumption must occur with a concomitant decrease in ω-6 FA consumption (Oliveira et al. 2006). Although there are some significant differences found in the fatty acid

composition, the overall impact of these differences is greatly reduced when the extremely low lipid content of the meat is considered.

2.5.5 Texture and pH

Texture and pH of scallops have been well studied, but have largely been focused on determining changes over time during storage and/or freezing (Maxwell-Miller et al. 1982, Ocano-Higuera 2006, Pacheco-Aguilar et al. 2008, De Vido de Mattio et al. 2009, Makri 2009). Both softening (tenderness) and hardening (toughness or firmness) of texture have been linked to the freezing process (Dunajski 1979, Makri 2009), softening has been linked to increased moisture content in lean fish species (Dunajski 1979), and an inverse relationship between toughness and pH has been observed in fish and mammals (Cowie and Little 1966, Bouton et al. 1971, Purchas and Aungsupakorn 1993, Guerrero et al. 1999). Based on harvester descriptions of WS as 'mushy' and 'soft', the higher moisture content and higher pH, we expected texture results to show that they were less firm than SS, KS1 and KS2. Interestingly, results were opposite of our expectations. Maxwell-Miller (1982) found that toughness increased with size and age in purple hinge rock scallops (*Crassadoma gigantea* Gray 1825), and Dunajski (1979) reports similar findings for fish muscle. A similar relationship might be the factor in the tougher texture observed in the WS meats.

The pH of fresh fish, or fish flesh immediately after harvest, is at or just above neutral (Dunajski 1979, Howgate 2009) and decreases rapidly within several hours post-mortem. De Vido de Mattio et al. (2009) reports that the scallop (*Chlamys tehuelchus*) decreased from pH 6.8 immediately after death to pH 6.0-6.2 after 96 h in cold storage,

similar to our results for SS and KS2. Howgate (2009) states that the post-mortem pH for most fish species is between 6.0-6.8, but for species with high initial glycogen, the pH will be lower. The glycogen in post-mortem muscle is converted to lactate through anaerobic glycolysis, which induces a decrease in muscle pH (Hiltz and Dyer 1971, Dunajski 1979, El Rammouz et al 2004). De Vido de Mattio et al. (2009) reported glycogen content in scallop decreases 40% within the first 12 h of post-mortem storage. Although glycogen was not detected in WS, it is possible that sufficient glycogen could be present in the tissue at the time of death for glycolysis to have occurred. Soon after harvest, a drop in pH of WS adductor muscle to slightly below neutral occurred but insufficient glycogen was available to reduce the normal post-mortem range of pH 6.0-6.8. Another possibility is that post-mortem pH increases can be attributed to the accumulation of alkaline metabolites from bacterial accumulation during chilled storage (Ocano-Higuera et al. 2006); however, since the scallops used in this study were all frozen immediately onboard the vessel, it is unlikely this is the cause of the observed differences in pH.

2.6 Conclusion

Biochemical analysis has quantified the factors that contribute to the poor quality meats observed in WS and determined that SS meats were of similar quality to those from Kodiak scallops. High moisture content in WS could contribute to reports of stringiness upon cooking. The tougher texture explains why WS tear during shucking. The literature suggests that the patterns observed in WS composition may be indicative of nutritional stress. Yakutat is heavily glaciated and run-off from these glaciers could have

a negative impact on feed quality and quantity. Boring worms (*Polychaete* sp.) can also cause small and discolored meats during heavy infestation due to energy redistribution to repair damaged shells (Hennick 1973). While it is beyond the scope of this project to determine the causes of WS, further exploration into environmental impact or parasites and diseases would be worth considering. Results from this project have the potential to identify and quantify the quality parameters that differentiate 'weak meat' scallop muscle from prime quality scallop muscle. This will enable future researchers to focus on the causes and potential solutions to the problem.

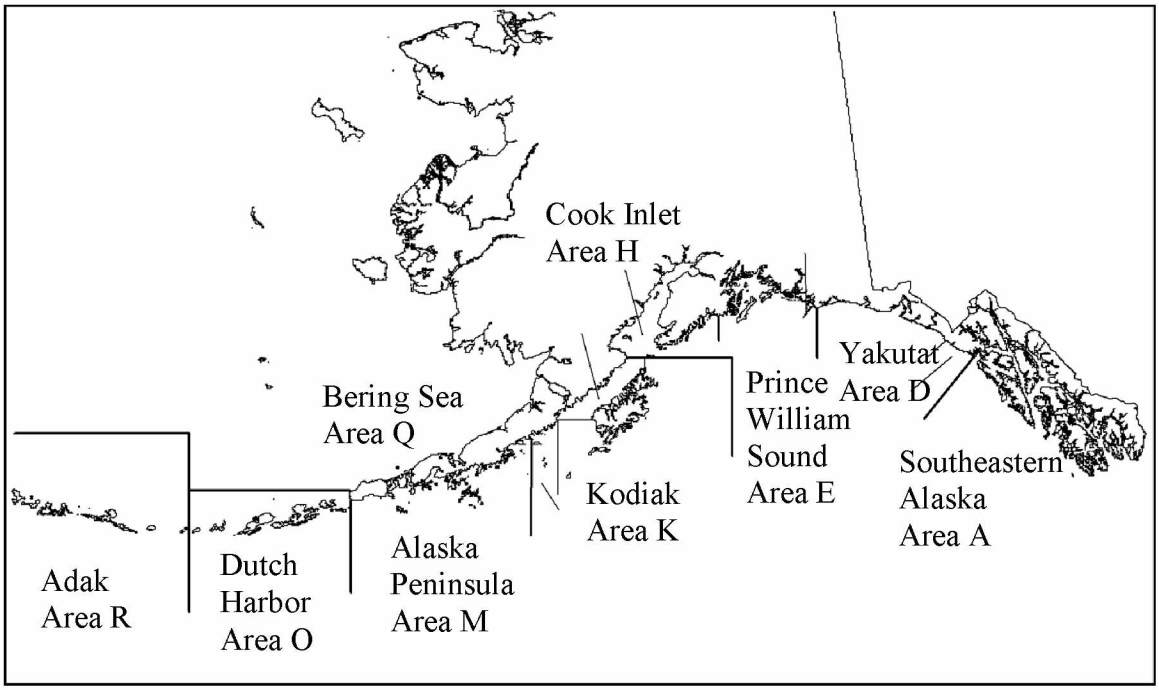

Figure 2.1. Alaska weathervane scallop fishing registration areas (NPFMC 2009).

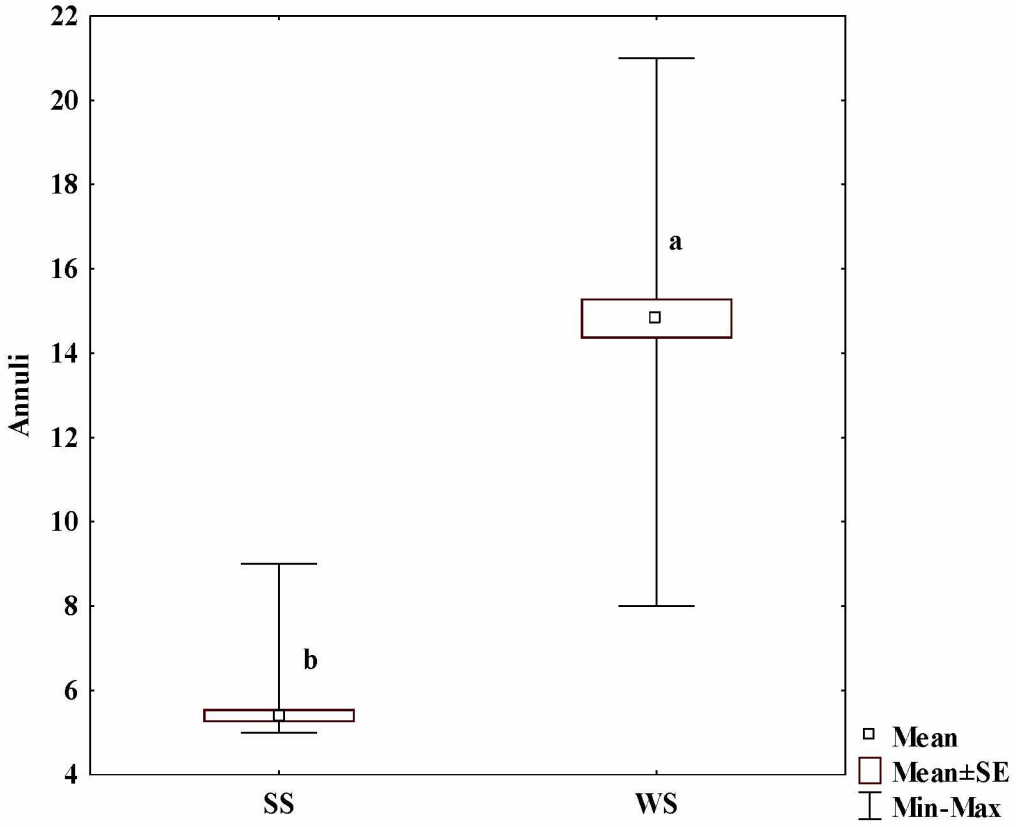

Figure 2.2. Mean annuli indicating age of weathervane scallops from the Eastern Gulf of Alaska. Different letters above boxes indicate significant differences. SS Standard quality scallop, WS Weak meat scallop.

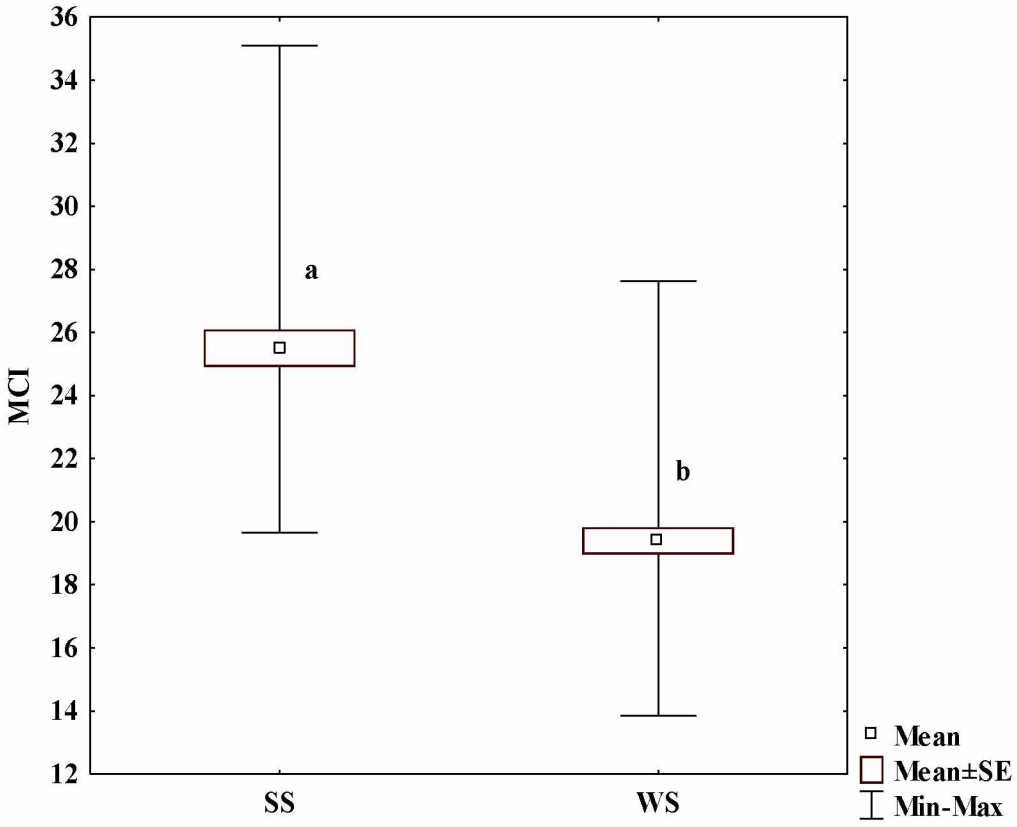

Figure 2.3. Mean Muscle Condition Index (MCI) of weathervane scallops from the Eastern Gulf of Alaska. Different letters above boxes indicate significant differences. SS Standard quality scallop, WS Weak meat scallop.

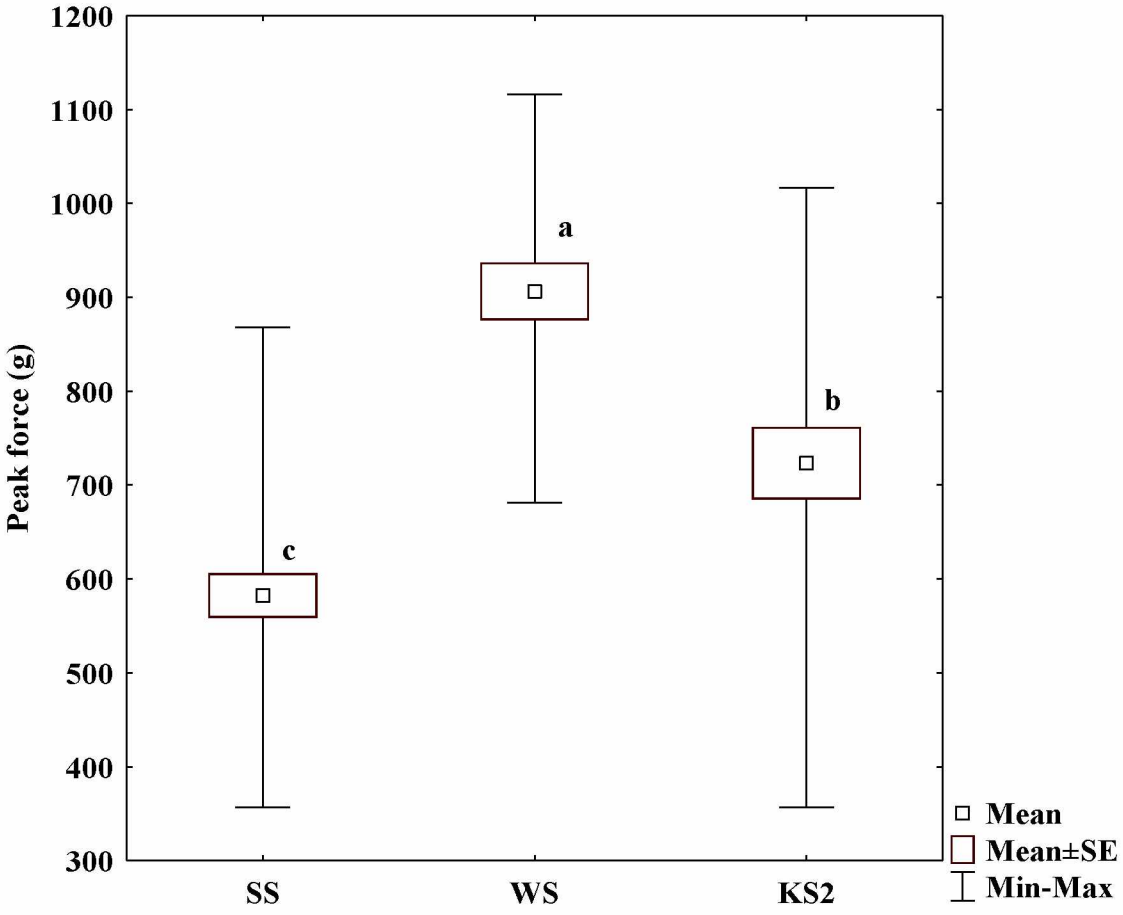

Figure 2.4. Mean peak force needed to penetrate meat of weathervane scallops from the Gulf of Alaska. Different letters above boxes indicate significant differences. SS Standard quality scallop, WS Weak meat scallop, KS2 Kodiak scallop 2.

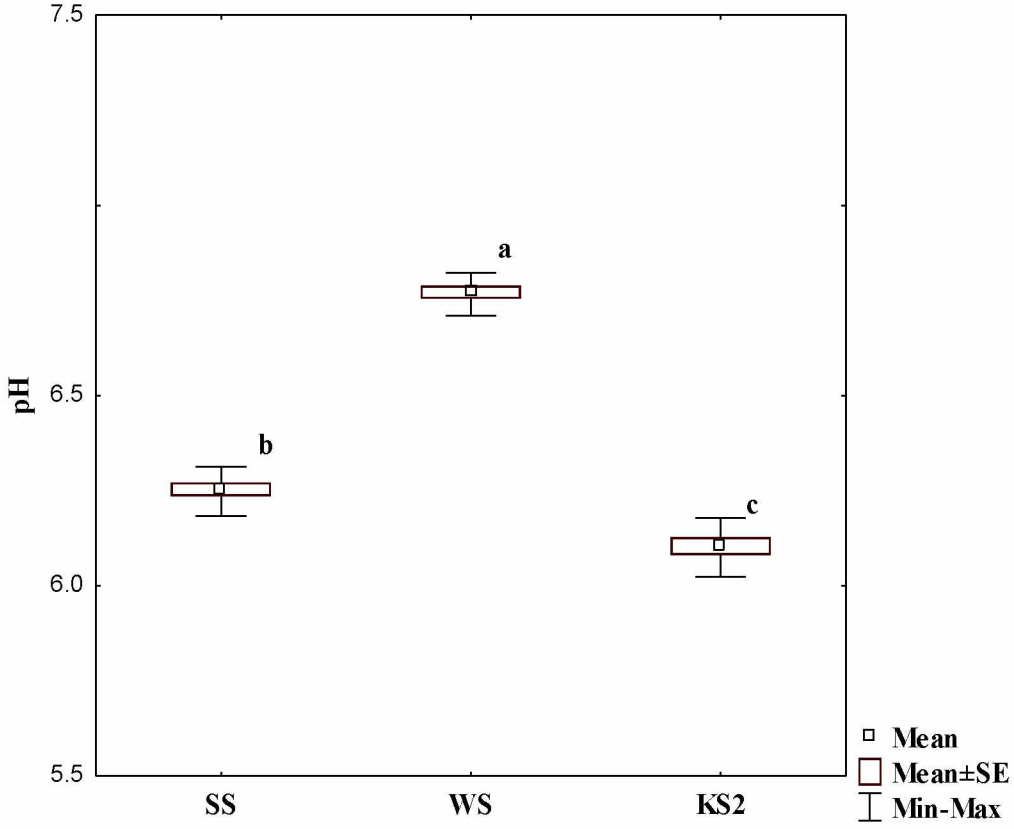

Figure 2.5. Mean pH of weathervane scallops from the Gulf of Alaska. Different letters above boxes indicate significant differences. SS Standard quality scallop, WS Weak meat scallop, KS2 Kodiak scallop 2.

Table 2.1.

Morphological measurements (mean ± SD) taken from weathervane scallops from the Eastern Gulf of Alaska.

	WS (n = 40)	SS (n = 40)
Whole weight (g)	$146.99^{a} \pm 27.74$	$115.34^{b} \pm 21.90$
Body weight (g)	$89.66^{a} \pm 20.68$	$53.49^{b} \pm 10.35$
Muscle (meat) weight (g)	$21.17^{a} \pm 3.70$	$18.04^{b} \pm 3.19$
Meat yield (%)	$14.49^{b} \pm 1.54$	$15.74^{a} \pm 1.49$
Shell height (mm)	$131.68^{a} \pm 8.48$	$116.05^{b} \pm 5.08$

Different superscript letters within a row indicate significant differences ($P<0.05$). SD Standard deviation, SS Standard quality scallop, WS Weak meat scallop.

Table 2.2.

Proximate composition (% wet weight ± SD) of weathervane scallops from the Gulf of Alaska.

Analysis	WS (n = 8)	SS (n = 8)	KS1 (n = 5)	KS2 (n = 6)
Moisture	$81.62^{a} \pm 0.61$	$78.12^{ab} \pm 0.52$	$76.43^{b} \pm 0.48$	$76.77^{b} \pm 0.48$
Protein	$15.35^{b} \pm 0.89$	$17.53^{ab} \pm 0.31$	$19.25^{a} \pm 0.12$	$19.15^{a} \pm 0.48$
Glycogen	BDL	$1.61^{a} \pm 0.49$	$2.19^{a} \pm 0.48$	$1.75^{a} \pm 0.69$
Ash	$2.43^{a} \pm 0.24$	$2.15^{ab} \pm 0.16$	$1.48^{c} \pm 0.08$	$1.55^{bc} \pm 0.03$
Lipid	$0.59^{a} \pm 0.19$	$0.60^{a} \pm 0.08$	$0.65^{a} \pm 0.21$	$0.77^{a} \pm 0.19$
M:P	$5.33^{a} \pm 0.35$	$4.46^{ab} \pm 0.09$	$3.97^{b} \pm 0.05$	$4.01^{b} \pm 0.09$

Different superscript letters within a row indicate significant differences ($P<0.05$). SD Standard deviation, BDL Below detection limit of 0.62%, M:P Moisture:protein ratio SS Standard quality scallop, WS Weak meat scallop, KS1 Kodiak scallop 1, KS2 Kodiak scallop 2.

Table 2.3.

Lipid class composition (mean % total lipids ± SD) of weathervane scallops from the Gulf of Alaska.

Lipid	WS (n = 8)	SS (n = 8)	KS1 (n = 5)	KS2 (n = 6)
Phospholipids	$81.59^{b} \pm 4.33$	$86.19^{ab} \pm 1.97$	$85.21^{ab} \pm 2.04$	$90.84^{a} \pm 0.98$
Sterols	$12.92^{a} \pm 2.84$	$10.41^{ab} \pm 1.54$	$10.32^{ab} \pm 1.41$	$8.13^{b} \pm 0.76$
Free fatty acids	$5.49^{a} \pm 1.57$	$3.40^{ab} \pm 0.55$	$3.45^{ab} \pm 0.52$	$1.03^{b} \pm 0.25$
Triacylglycerols	BDL	BDL	$1.06^{a} \pm 0.69$	BDL

Different superscript letters within a row indicate significant differences ($P<0.05$). SD Standard deviation, BDL Below detection limit (<0.5%), SS Standard quality scallop, WS Weak meat scallop, KS1 Kodiak scallop 1 KS2, Kodiak scallop 2.

Table 2.4.

Fatty acid methyl ester profiles (mean % w/w ± SD) of weathervane scallops from the Gulf of Alaska.

	WS (n = 8)	SS (n = 8)	KS1 (n = 4)	KS2 (n = 6)
16:0	$14.71^{b} \pm 0.59$	$15.85^{a} \pm 0.27$	$16.21^{a} \pm 0.58$	$15.24^{ab} \pm 0.19$
18:0	$5.69^{a} \pm 0.24$	$5.13^{ab} \pm 0.23$	$5.13^{ab} \pm 0.20$	$4.60^{b} \pm 0.23$
18:1ω9 *cis*	$2.25^{ab} \pm 0.56$	$1.96^{b} \pm 0.08$	$2.99^{a} \pm 0.23$	$3.12^{a} \pm 0.17$
18:1ω7	$3.10^{a} \pm 0.14$	$3.01^{a} \pm 0.11$	$3.06^{a} \pm 0.13$	$3.17^{a} \pm 0.09$
20:5ω3 (EPA)	$17.72^{b} \pm 1.30$	$18.57^{ab} \pm 0.34$	$18.03^{b} \pm 0.49$	$22.63^{a} \pm 0.41$
22:6ω3 (DHA)	$33.27^{b} \pm 2.37$	$37.16^{a} \pm 0.77$	$36.37^{ab} \pm 1.12$	$30.68^{b} \pm 0.67$
EPA + DHA	$51.00^{b} \pm 3.38$	$55.73^{a} \pm 0.69$	$54.40^{ab} \pm 1.08$	$53.31^{b} \pm 1.05$
Σ SAT	$23.81^{ab} \pm 0.93$	$24.04^{ab} \pm 0.38$	$24.93^{a} \pm 0.55$	$22.90^{b} \pm 0.39$
Σ MUFA	$11.14^{ab} \pm 1.74$	$10.43^{b} \pm 0.40$	$13.73^{ab} \pm 0.69$	$13.83^{a} \pm 0.39$
Σ PUFA	$63.46^{a} \pm 2.50$	$63.73^{a} \pm 0.91$	$59.87^{b} \pm 0.61$	$61.23^{ab} \pm 0.61$
PUFA/SAT	$2.67^{a} \pm 0.20$	$2.65^{a} \pm 0.06$	$2.40^{a} \pm 0.07$	$2.67^{a} \pm 0.07$
ω3	$56.07^{b} \pm 1.94$	$58.98^{a} \pm 0.99$	$56.43^{ab} \pm 0.80$	$57.04^{ab} \pm 0.51$
ω6	$7.39^{a} \pm 2.03$	$4.65^{ab} \pm 0.17$	$3.44^{b} \pm 1.00$	$4.06^{b} \pm 0.51$
ω3/ω6	$8.16^{b} \pm 2.43$	$12.69^{ab} \pm 0.58$	$17.30^{a} \pm 4.22$	$14.21^{a} \pm 1.59$

Different superscript letters within a row indicate significant differences ($P<0.05$). SD standard deviation, EPA Eicosapentaenoic acid, DHA Docosahexaenoic acid, SAT Saturated fatty acids, MUFA Monounsaturated fatty acids, PUFA Polyunsaturated fatty acids, SS Standard quality scallop, WS Weak meat scallop, KS1 Kodiak scallop 1, KS2 Kodiak scallop 2.

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CHAPTER 3

Quality assessment of purple hinge rock scallops (*Crassadoma gigantea*) from Alaska

3.1 Abstract

The purple-hinge rock scallop (PHRS, *Crassadoma gigantea* Gray 1825) is highly prized by local communities and harvested for subsistence in Alaska. Scallop culture is a well-established industry in some parts of the world, however efforts in the US remain limited to small scale operations. Aquaculture of PHRS has been investigated in California and while results were promising, there were barriers to implementing commercial culture operations. Renewed interest has resulted in a recent grow-out study to determine the feasibility of culturing this species in Alaska. Assessment of PHRS meat quality is necessary to promote market acceptance. The objective of this study was to characterize meat quality of PHRS and compare them with prime quality weathervane scallops from Kodiak. Ten PHRS from Southeast Alaska and two batches (17 individuals, 30 individuals) of scallop muscle from Kodiak (KS1 and KS2) were sampled. Physical measurements and chemical composition analyses were conducted. PHRS whole weight ranged from 561.0 to 1,202.2 g. Muscle condition indices were between 36.0 and 50.9. Proximate composition and lipid class results were not significantly different ($P>0.05$) between PHRS and weathervane scallops. Overall, PHRS are quite similar to weathervane scallops from a biochemical standpoint and share the same quality attributes.

3.2 Introduction

Scallops are distributed worldwide and the adductor muscle is a highly valued food source. There is a wide body of literature examining many aspects and many species (Hennick 1970, Whyte et al. 1990, Cranford 1995, Jeong et al. 1999, Shriver et al. 2002, Shumway and Parsons 2006, Pacheco-Aguilar et al. 2008, Makri 2009). In Alaskan waters, the Pacific weathervane scallop (*Patinopecten caurinus* Gould 1850) is the only species commercially harvested. The purple-hinge rock scallop (PHRS, *Crassadoma gigantea* Gray 1825), formerly classified as *Hinnites multirugosus* Gale 1928, has potential for aquaculture, and is highly prized by local communities and harvested for subsistence in coastal Alaska.

Scallop culture is a well-established industry in some parts of the world. Japan began investigating the potential for scallop culture in the 1930's and began commercial production in the mid-1960's (Kosaka and Ito 2006). Building on Japan's success, and borrowing from the procedures developed there, China and Chile began their own scallop culture industries in the 1980's and all three countries have well developed economically important scallop culture industries (Guo and Luo 2006, von Brand et al. 2006). Also building on the technology developed in Japan, small but developing culture industries exist in eastern Canada, Russia, Britain, France, Norway, Ireland, Italy and Spain (Norman et al. 2006, Ivin et al. 2006, Parsons and Robinson 2006). In spite of the success of scallop culture elsewhere in the world, efforts in the US fail to meet domestic demand and remain limited to small scale operations (Blake and Shumway 2006).

The PHRS is distributed along the Pacific coast from Baja, California to Alaska, though distribution is patchy throughout the range (MacDonald and Bourne 1989). They have abandoned the free swimming nature of their pectinid relatives and have developed an affinity for permanent attachment and imbed into rocky substrates. While this presents challenges for commercial harvest, it is a potential advantage for mariculture.

Aquaculture of PHRS has been investigated in California and while results were promising, there were barriers to implementing commercial culture operations (Leighton and Phleger 1981). Renewed interest has resulted in a recent grow-out study to determine the feasibility of culturing this species in Alaska. While PHRS are locally prized, they would be a new species for the seafood market and meat quality assessment is necessary to promote market acceptance. The best means to accomplish this effort is to compare the qualities of the PHRS with a known commercially harvested scallop species, the weathervane scallop.

3.3 Materials and Methods

3.3.1 Sample Procurement

In mid-August 2009, ten adult PHRS were harvested from pens at Elfin Cove Oysters in Port Althorp on the north end of Chichagof Island in SE Alaska and shipped live to the Fishery Industrial Technology Center (FITC) in Kodiak, AK. Scallops were held at 4°C and processing was carried out within 2 days of arrival at FITC.

Kodiak weathervane scallop samples and results are the same as those used in Chapter 2. One poly bag (1.13 Kg) of weathervane scallop meats was obtained from the F/V Provider in April 2009 (KS1) and 6.9 Kg (three 2.3 Kg boxes, two poly bags/box) of

weathervane scallop meats harvested in August 2009 were purchased at a local seafood market (Island Seafoods, Kodiak, AK) in October 2009 (KS2). These were brought to FITC and kept frozen at -30 °C for one month until analysis.

3.3.2 Morphology and Condition

Wet weights of whole scallop, shell, gonadal-visceral (body) tissue, and adductor muscle (meat) were measured using a Sartorius model LP2200P electronic digital scale with manufacturer accuracy given at 0.01g (Gottingen, Germany). Shell height, width, and depth and meat height and width were determined using a cloth tape measure and electronic digital calipers (200 mm range, VWR, Radnor, PA). Measurements for KS included meat weight for KS1 and meat height and weight for KS2. The PHRS scallops were shucked after whole weights were recorded and the meat and body were separated. Scallop body and meat tissues were individually vacuum packed and stored at -30°C for three months until analysis. A muscle condition index (MCI), was calculated for PHRS based on that of Beltran-Lugo et al. (2005) in order to assess physiological condition of the meat relative to total soft tissue: MCI (%) = weight of adductor muscle/weight of total soft tissues x 100.

3.3.3 Sample Preparation

Individual scallops were too small to be assayed for all chemical analyses so pooling was necessary. A pooling scheme applied by Linehan et al. (1999) was adopted. Linehan et al. (1999) determined that the value of any assay obtained from a sample pool of oysters was not significantly different than the weighted average obtained by analysis of single individuals, demonstrating that this strategy could be used to meet tissue

requirements for laboratory analysis. This pooling scheme has also been successfully applied for biochemical analysis of Alaska oyster samples (Oliveira et al. 2006).

Scallop meats were thawed at 4°C and the meats from the one KS1 poly bag were randomly pooled into three samples of three meats and two samples of four meats (n=5); for the KS2 samples, five meats were randomly pooled from each poly bag (n=6). PHRS were pooled into five samples of two meats per sample using constrained randomization to maintain consistent weight between samples (n=5). Samples were homogenized using a Cuisinart Mini-prep model DLC-1SS (Cuisinart Corp., East Windsor, NJ) until a uniform paste was obtained (± 2 min). Homogenized samples were stored in zip-lock bags and kept at 4°C throughout analysis.

3.3.4 Proximate Composition

Percent moisture and ash were determined according to AOAC methods 952.08 and 938.08, respectively (Helrich 1990). Percent protein was measured using a Leco model TruSpec N nitrogen analyzer (LECO Corp., St. Joseph, MI). Percent protein was calculated by multiplying %N by 6.25 (Helrich 1990). Lipid was extracted using the method described by Folch et al. (1957) with slight modifications described below. A quantity of 12 g of tissue was combined with 90 mL of a mixture of 2:1 chloroform:methanol with 0.01% butylated hydroxytoluene (BHT), thoroughly mixed with a glass stir bar to break up clumps, and refrigerated under nitrogen atmosphere for 24 h. Contents were filtered through a 42 mm Buchner funnel and transferred to a 125 mL separatory funnel. A volume of 18 mL of 0.88% (W:V) KCl was added to the separatory funnel and the contents were thoroughly shaken and allowed to sit until clear.

The lower phase was filtered through anhydrous sodium sulfate and the solvent was evaporated using a TurboVap-LV Evaporator (Caliper Life Sciences, Hopkinton, MA) operated at 40°C under a nitrogen atmosphere. Lipid was determined gravimetrically.

The total carbohydrate content was determined using the anthrone method, a combination of the methods of Strickland and Parsons (1972) and Clegg (1956) as previously described by Oliveira et al. (2006). The anthrone reagent was prepared by dissolving 0.2 g of anthrone (Sigma-Aldrich, St. Louis, MO) in 8 mL of HPLC-grade ethyl alcohol (VWR) and 30 mL of distilled water. After complete dissolution of the anthrone in the solvents, 100 mL of concentrated sulfuric acid was added to the mixture. A quantity of 15 g of homogenized tissue was mixed with 10 mL of distilled water and 13 mL of 52% perchloric acid (VWR) in a 100 mL capped flask. The contents were stirred for 30 min at room temperature using a magnetic bar and a standard laboratory stir plate (VWR). The mixture was transferred to a 100 mL volumetric flask, diluted to 100 mL and filtered using Whatman #4 filter paper into a 250 mL volumetric flask. The filtrate was diluted to 250 mL, and a 10 mL portion was transferred to a 100 mL volumetric flask and further diluted to 100 mL. A volume of 1 mL was transferred to a 25 mL screw-top tube and 5 mL of anthrone reagent added, and the tube was placed in a boiling water bath for 7 min. The tubes were rapidly cooled in an ice water bath and solutions were read at 624 nm on a Cary 50 UV-Visible Spectrophotometer (Varian Instruments, Walnut Creek, CA). In parallel, 1 mL of distilled water was used as the blank, and a 1 mL of a 0.1 mg/mL solution of oyster glycogen Type II (Sigma-Aldrich) was used as the standard. Blank and standard solutions were subjected to identical

colorimetric reaction as described for scallop meat samples. Percent total carbohydrate was calculated using the following formula:

% Total carbohydrate = (25 x A2)/(A1 x W)

Where:

W = weight of sample (g)

A1 = absorbance of glycogen standard

A2 = absorbance of sample

It is important to point out that for scallop meats, carbohydrates in the tissue are predominantly present as glycogen (Barber and Blake 1981, Brokordt et al. 2000). Therefore, % total carbohydrate quantified is reported and discussed as % glycogen. Glycogen content for KS1 samples was determined by subtraction: % Glycogen = 100 - (% moisture + % protein + % ash + % lipid).

3.3.5 Lipid Classes

Lipid class analysis was based on the methods of Parrish (1987) and Ackman et al. (1990), as described by Oliveira and Bechtel (2005) and was carried out on an Iatroscan^{TM} TLC/FID Analyzer (Model MK-6s, Iatron Laboratories, Tokyo, Japan). The hydrogen flow rate was 160 mL/min, air flow was 1.6 L/min and scan time was 30 s. The six standards used to identify lipid class were tripalmitin (triacylglycerol, TAG), palmitic acid (free fatty acid, FFA), (S)-1, 2-dipalmitin (diacylglycerol, DAG), cholesterol (sterol, ST), DL-α-palmitin (monoglyceride, MG) and L-α-phosphatidylcholine (phospholipid, PL) and were obtained from Sigma-Aldrich. Solvent tanks (Shell-USA, Fredericksburg, VA) were lined with Whatman #2 filter paper and

159.3 mL of solvent was added. The closed system was allowed to equilibrate for 20 min. The solvent system was hexane:diethyl ether:formic acid (80:25:1.2). Sample solutions contained 13 μg/μL of oil in chloroform and composite standard solution contained a mixture of 16.6 μg/μL of each standard in chloroform. For each rack of 10 rods (Chromarods-SIII, Iatron Laboratories, Tokyo Japan), a 1 μL portion in five 0.2 μL aliquots was spotted for each sample (rods 2-9) and composite standard (rods 1 and 10). Chromarods were suspended in the solvent tank for 10 min before being lowered into the solvent for 30 min. Developed rods were dried at 105°C for 3 min before being scanned. Peaks were integrated using the Peak Simple Program (v. 2.83, SRI Instruments, Torrence, CA).

3.3.6 Fatty Acid Methyl Ester Profiles

Fatty acid methyl esters (FAME) were prepared according to the procedure of Maxwell and Marmer (1983) using 1 mg of methyl tricosanoate (Sigma-Aldrich) as the internal standard. FAMEs were quantified as previously described by Oliveira et al. (2006), with the exception that the sample injection volume was 2 μL.

3.3.7 pH

The pH was determined on the pooled meats for PHRS and KS2 using the temperature compensated WTW pH3400i handheld pH meter with the WTW model 103702 probe attached (Weilheim, Germany). The probe was two-point calibrated at pH 7 and pH 10 before use and inserted directly into the sample immediately after tissue homogenization.

3.3.8 Statistical Analysis

Data were checked for normality by the Kolmogorov-Smirnoff test prior to statistical analysis. Non-parametric analysis was used because data did not comply with normal distribution assumptions. In cases where three or more groups were compared a Kruskal-Wallis ANOVA was used. Multiple comparisons of mean ranks for all pairs of groups were run post-hoc to determine significances at ($P<0.05$). In the case of two groups being compared, a Mann-Whitney U t-test was performed. Analysis was carried out using Statistica (v.10, StatSoft, Tulsa, OK).

3.4 Results

3.4.1 Morphology and Condition

The PHRS is a large species with massive shells and large meats (Table 3.1). The range of specimen whole weights was 561 g to 1,202 g. Shell heights ranged from of 138 to 189 mm. Yields of meat were <10% however, MCI values were between 36.0 and 50.9.

3.4.2 Proximate Composition

Significant differences were not detected in the proximate composition between PHRS, KS1 and KS2. Table 3.2 shows that PHRS meats are a high protein, low lipid tissue. Glycogen contents were slightly, but not significantly ($P>0.05$), lower in PHRS than KS1 and KS2. Gross composition of PHRS suggested that this group of scallops was of comparable quality to Kodiak scallops.

3.4.3 Lipid Classes

Overall, scallops were very high in phospholipids and triacylglycerols were not detected (<0.5%) except for KS1 which had trace amounts. The two Kodiak groups showed the most variation, and except for having significantly lower abundance of sterols, PHRS were not significantly different (Table 3.3). The predominance of phospholipids in the lipid extracts is expected for tissues with low lipid content, and this result is in agreement with the lipid content < 1% determined for scallops in our study. As observed in prior studies, phospholipids were inversely proportional to triacylglycerol content (Bechtel and Oliveira 2006). Ackman and Gunnlaugsdottir (1992) also observed this when measuring neutral and polar lipids in Japanese sardines with variable lipid content. They reported that sardines with a total lipid content of 0.89% (w/w) had polar and neutral lipids fractions that were 0.7 and 0.19% of the total lipid content, respectively; however, when sardines had 8.66% (w/w) lipids, the polar and neutral lipids fractions were 0.76 and 7.9% of the total lipid content, respectively.

3.4.4 Fatty Acid Methyl Ester Profiles

Fatty acid methyl ester (FAME) classes and the most abundant FAMEs in each class are reported in Table 3.4. The most abundant FAMEs in all scallop meat groups were the saturated FAMEs (SAT) 16:0 and 18:0, the monounsaturated FAMEs (MUFA) 18:1ω9 *cis* and 18:1ω7, and the long chain polyunsaturated FAMEs (PUFA) 20:5ω3 (EPA) and 22:6ω3 (DHA). Palmitic acid (16:0), EPA and DHA were the three most abundant FAMEs and comprised 69.6% of the total extracted lipids (15.8%, 22.8% and 31.0%, respectively). PHRS had the highest EPA and lowest DHA levels of all groups,

while KS1 were highest in palmitic acid. PUFAs constituted about 61.7% of total extracted lipid for all groups, followed by SAT at 24.6%, and MUFA at 12.5%. Total ω-3 FAME was not significantly different between groups but PHRS had significantly higher ω-6 FAME, and consequently the lowest ω3:ω6 ratio.

3.4.5 pH

The pH of PHRS was slightly but significantly lower than that of KS2 (Fig. 3.1) and was well below 7.0 for both groups.

3.5 Discussion

3.5.1 Morphology and condition

Our results reflect observations from other researchers about the size and condition of PHRS. Individuals can reach shell heights of 175-250 mm and the shells become massive and heavy (MacDonald and Bourne 1989, Lauzier and Bourne 2006). Leighton (1991) reported that PHRS meats were 30-55% of soft body weight, reported as MCI in this study, with weights of 50-80 g. Leighton (1991) reported yields of 9-10%. In comparison, weathervane scallop, which can also reach shell heights of 250 mm, typically have meats in the 27-48 g range and have yields of 10-12% (Hennick 1973).

3.5.2 Proximate composition

The proximate composition of the scallop meats analyzed in this study is within the ranges previously reported for scallops of both the same and other species (Webb et al. 1969, Krzeczkowski et al. 1972, Naidu and Botta 1978, Maxwell-Miller et al. 1982, Anthony et al. 1983, King et al. 1990, Silva and Chamul 2000). It must be noted that slight differences do occur between species, and, as in CI, environmental factors can

affect proximate composition (Krzeczkowski et al. 1972, Green and Korhonen 1993). There is scant data available specific to PHRS so exact comparisons are difficult. However, the general biology and energy utilization of scallops enables an understanding of the differences observed in this study.

The meat and digestive gland are the primary storage sites of nutrient reserves in scallops during periods of somatic growth when food is abundant. Carbohydrate, in the form of glycogen, and protein are stored in the meat, while lipid is stored in the digestive gland (Barber and Blake 1981). All scallop meats in this study had low lipid levels, consistent with the observation that the meat is not a primary storage site for lipid. Protein and glycogen levels of the meats are similarly reflective of how scallops store energy.

3.5.3 Lipid Classes

Due to its low lipid content, scallop adductor muscle is considered a 'lean organ' along with gills and mantle. Napolitano and Ackman (1992) observed for sea scallops (*Placopecten magellanicus*) that sterols and phospholipids are the predominant lipid classes in the lean organs, whereas triacylglycerols are very low. The high phospholipid and sterol levels relative to the abundance of triacylglycerols observed in meats in this study are consistent with results from other scallop studies (Napolitano and Ackman 1992, Napolitano et al. 1992, Linder and Ackman 2002, Copeman and Parrish 2004, Palacios et al. 2007). Phospholipids and sterols are structural lipids comprising cell membrane while triacylglycerols are an energy storage lipid. While triacylglycerol was detected in one group of samples (KS1) and not in the other groups, it still comprised

only 1% of the total lipid classes recorded, and its presence was likely attributable to seasonal differences in diet composition and/or reproductive stage at time of harvest. Pernet et al. (2003b) found higher triacylglycerol levels in sea scallop (*Placopecten magellanicus*) larvae fed diets rich in this lipid class, and levels were highly variable between diet and time (Pernet et al. 2003a).

Free fatty acids are usually present in trace amounts in marine organisms and scallops are no exception. Napolitano and Ackman (1992) found ~1% in sea scallop meat in winter and trace levels in summer. Copeman and Parrish (2004) found <1% FFA in the meat of Icelandic scallop. Free fatty acid levels in meats from this study were similar except in KS1 scallops which had the highest FFA and lowest PL levels. The presence of large quantities of FFA in tissue is an indication of lipid degradation and breakdown of the structural lipids (Jeong et al. 1999, Pernet et al. 2003a). Lipid degradation can occur during long term storage, or when tissues are improperly stored. The slight differences in the free fatty acid values observed between PHRS, KS1 and KS2 are likely due to the fact that KS1 scallops were frozen for a longer period of time.

3.5.4 Fatty Acid Methyl Ester Profiles

Fatty acid analysis of many species of scallop have shown that polyunsaturated fatty acids account for about 60% of the total lipids, while saturated and monounsaturated fatty acids account for about 25-30% and 10-15%, respectively (Gruger Jr. et al. 1964, Krzeczkowski et al. 1972, Napolitano et al. 1992, Linder and Ackman 2002, Copeman and Parrish 2004) and the results from the present study were similar. The predominant

FA 22:6ω3, 20:5ω3, 18:1ω9 and 16:0 are typical components of phospholipids (Richoux et al. 2005).

The higher levels of PUFA detected in PHRS could be attributed to temperature, depth and/or diet. Krzeczkowski et al. (1972) found that scallops originating from cold deep environments (40-60 fathoms) have higher PUFA content and the polyunsaturation level of polar lipid fatty acids is inversely related to the ambient temperature (Bell et al. 1986, Napolitano et al. 1992). In addition, marine fish and bivalves lack the elongation and desaturation enzymes necessary to synthesize long chain PUFAs and both EPA and DHA are considered nutritionally essential (Bell et al. 1986, Whyte et al. 1990).

The ratio of ω3 to ω6 is used to compare the relative nutritional value of foods because in order to gain the health benefits of ω-3 FA, an increase in ω-3 FA consumption must occur with a concomitant decrease in ω-6 FA consumption (Oliveira et al. 2006). Although there are some significant differences found in the fatty acid composition, the overall impact of these differences is greatly reduced when the extremely low lipid content of the meat is considered.

3.5.5 pH

In a storage stability study, Maxwell-Miller et al. (1982) reported pH 6.4-6.7 for shucked PHRS meats during two weeks at 5°C. This is much higher than what was observed in this study. However, the pH of fresh fish, or fish flesh immediately after harvest, is at or just above neutral (Dunajski 1979, Howgate 2009) and falls rapidly within several hours post-mortem. De Vido de Mattio et al. (2009) report that the pH of

scallop (*Chlamys tehuelchus*) decreases from pH 6.8 immediately post-mortem to pH 6.0-6.2 after 96 h in cold storage, and their observation closely reflects our results.

3.6 Conclusion

Analysis shows that PHRS is quite similar to weathervane scallop from a biochemical standpoint and shares the same quality attributes. Continuing development to establish this species for culture in Alaska is warranted based on the results of this meat quality study. However, this study was limited in scope and was unable to identify the qualities that make the PHRS a unique and highly valued resource.

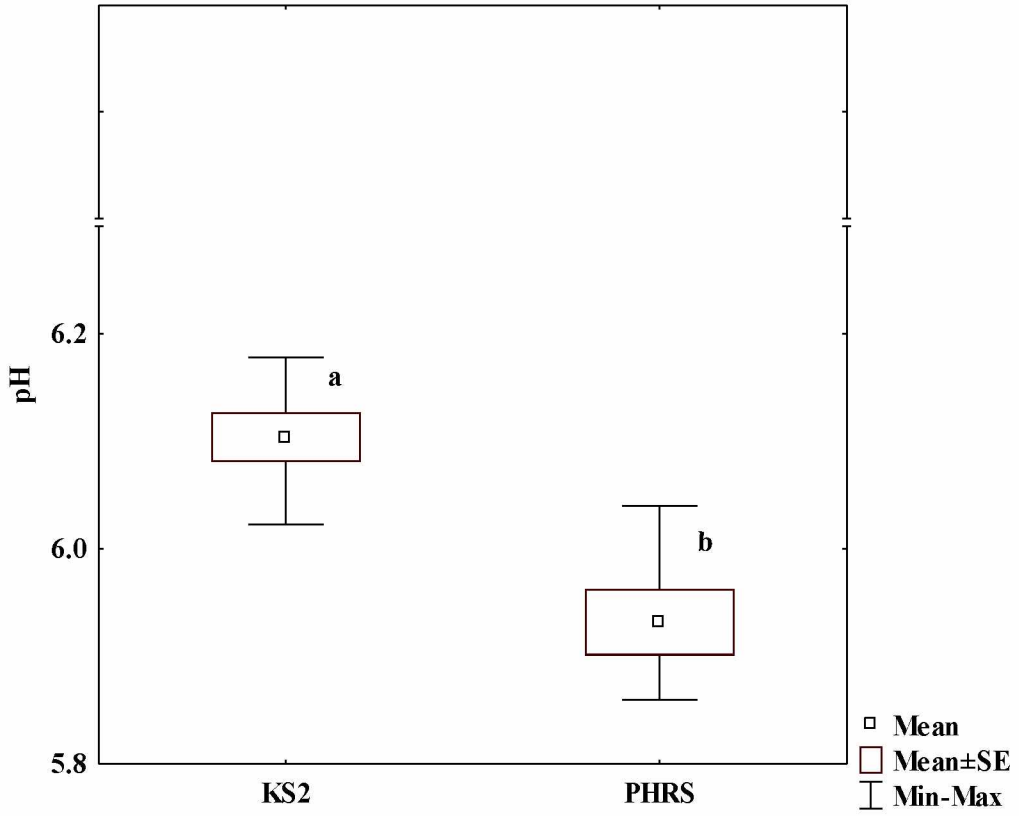

Figure 3.1. Mean pH comparing purple-hinge rock scallops and weathervane scallops from the Gulf of Alaska. Different letters above boxes indicate significant differences. KS2 Kodiak scallop group 2, PHRS Purple-hinge rock scallop.

Table 3.1.

Morphological measurements taken from purple-hinge rock scallops from the Eastern Gulf of Alaska.

	Whole weight (g)	Body weight (g)	Meat weight (g)	Meat yield (%)	Shell height (mm)	MCI
PHRS 1	757.9	48.72	35.58	4.69	167	42.2
PHRS 2	649.7	53.60	30.12	4.64	178	36.0
PHRS 3	1072	90.90	94.35	8.80	169	50.9
PHRS 4	682.2	57.35	43.89	6.43	156	43.4
PHRS 5	561	45.69	36.23	6.46	138	44.2
PHRS 6	938	51.58	39.52	4.21	168	43.4
PHRS 7	714.8	43.62	41.52	5.81	166	48.8
PHRS 8	868.7	63.00	55.57	6.40	175	46.9
PHRS 9	753.5	78.24	67.92	9.01	166	46.5
PHRS 10	1202	114.8	75.92	6.32	189	39.8
Mean	820	64.75	52.06	6.28	167.2	44.2
SD	200.1	23.08	20.98	1.62	13.5	4.36

MCI Muscle condition index, PHRS Purple-hinge rock scallop, SD Standard deviation.

Table 3.2.

Gross proximate composition (% wet weight ± SD) of purple-hinge rock scallops and weathervane scallops from the Gulf of Alaska.

	KS1 (n = 5)	KS2 (n = 6)	PHRS (n = 5)
Moisture	76.43[a] ± 0.48	76.77[a] ± 0.48	76.54[a] ± 0.73
Protein	19.25[a] ± 0.12	19.15[a] ± 0.48	19.67[a] ± 1.60
Glycogen	2.19[a] ± 0.48	1.75[a] ± 0.69	1.39[a] ± 0.41
Ash	1.48[a] ± 0.08	1.55[a] ± 0.03	1.60[a] ± 0.81
Lipid	0.65[a] ± 0.21	0.77[a] ± 0.19	0.81[a] ± 0.21
M:P	3.97[a] ± 0.05	4.01[a] ± 0.09	3.91[a] ± 0.35

Different letters within a row indicate significant difference ($P<0.05$). SD Standard deviation, M:P Moisture:protein ratio, KS1 Kodiak scallop 1, KS2 Kodiak scallop 2; PHRS Purple-hinge rock scallop.

Table 3.3.

Lipid class composition (mean % total lipids ± SD) of purple-hinge rock scallops and weathervane scallops from the Gulf of Alaska.

	KS1 (n = 5)	KS2 (n = 6)	PHRS (n = 5)
Phospholipids	$85.21^{b} \pm 2.04$	$90.84^{a} \pm 0.98$	$90.39^{ab} \pm 1.07$
Sterols	$10.32^{a} \pm 1.41$	$8.13^{ab} \pm 0.76$	$7.81^{b} \pm 0.99$
Free fatty acids	3.45a ± 0.52	$1.03^{b} \pm 0.25$	$1.80^{ab} \pm 0.13$
Triacylglycerols	$1.06^{a} \pm 0.69$	BDL	BDL

Different superscript letters within a column indicate significant differences ($P<0.05$). SD Standard deviation. BDL Below detection limit (0.5%), KS1 Kodiak scallop 1, KS2 Kodiak scallop 2, PHRS Purple-hinge rock scallop.

Table 3.4.

Fatty acid methyl ester profiles (mean % w/w ± SD) of purple-hinge rock scallops and weathervane scallops from the Gulf of Alaska.

	KS1 (n = 4)	KS2 (n = 6)	PHRS (n = 5)
16:0	$16.21^{a} \pm 0.58$	$15.24^{b} \pm 0.19$	$16.00^{ab} \pm 0.63$
18:0	$5.13^{ab} \pm 0.20$	$4.60^{b} \pm 0.23$	$7.37^{a} \pm 0.85$
18:1ω9 *cis*	$2.99^{ab} \pm 0.23$	$3.12^{a} \pm 0.17$	$0.83^{b} \pm 0.06$
18:1ω7	$3.06^{b} \pm 0.13$	$3.17^{ab} \pm 0.09$	$4.31^{a} \pm 0.14$
20:5ω3 (EPA)	$18.03^{b} \pm 0.49$	$22.63^{ab} \pm 0.41$	$27.68^{a} \pm 1.44$
22:6ω3 (DHA)	$36.37^{a} \pm 1.12$	$30.68^{ab} \pm 0.67$	$25.87^{b} \pm 1.20$
EPA + DHA	$54.40^{a} \pm 1.08$	$53.31^{a} \pm 1.05$	$53.56^{a} \pm 0.47$
Σ SAT	$24.93^{ab} \pm 0.55$	$22.90^{b} \pm 0.39$	$26.16^{a} \pm 0.78$
Σ MUFA	$13.73^{ab} \pm 0.69$	$13.83^{a} \pm 0.39$	$9.83^{b} \pm 0.32$
Σ PUFA	$59.87^{b} \pm 0.61$	$61.23^{ab} \pm 0.61$	$64.01^{a} \pm 0.82$
P/S	$2.40^{b} \pm 0.07$	$2.67^{a} \pm 0.07$	$2.45^{ab} \pm 0.10$
ω3	$56.43^{a} \pm 0.80$	$57.04^{a} \pm 0.51$	$57.37^{a} \pm 1.00$
ω6	$3.44^{b} \pm 1.00$	$4.06^{ab} \pm 0.51$	$6.64^{a} \pm 0.44$
ω3/ω6	$17.30^{a} \pm 4.22$	$14.21^{ab} \pm 1.59$	$8.68^{b} \pm 0.66$

Different superscript letters within a row indicate significant differences ($P<0.05$). SD Standard deviation, EPA Eicosapentaenoic acid, DHA Docosahexaenoic acid, SAT Saturated fatty acids, MUFA Monounsaturated fatty acids, PUFA Polyunsaturated fatty acids, KS1 Kodiak scallop 1, KS2 Kodiak scallop 2, PHRS Purple-hinge rock scallop.

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CHAPTER 4

General Conclusions

Based on the results obtained during this study, several conclusions can be drawn regarding the quality of Alaskan scallop. The observations of scallop harvesters that 'weak meat' weathervane scallops from Yakutat are of inferior quality have been verified. The high moisture content and tough texture point to difficulties in shucking and toughness after cooking. The lack of glycogen, high pH and low muscle condition are indicative that these scallops are under the influence of some environmental or nutritional stressor. The fact that standard weathervane scallops from the Yakutat area are similar in quality to Kodiak scallops indicates that the factors leading to this stress response in 'weak meat' scallops are localized within the Yakutat area. Future oceanographic research on water quality, currents, tidal influence in areas around scallop beds might allow the causes of 'weak meat' scallops to be determined.

Continuing investigation of purple-hinge rock scallop as an aquaculture species in Alaska is warranted based on the results of this study. The meats of these scallops were large and in good condition. The chemical composition was similar to high quality weathervane scallops, which are highly regarded by consumers. The characteristics that contribute to the high desirability of purple-hinge rock scallops in local subsistence fisheries could be quantified by further research in sensory analysis.

Overall, scallop quality in Alaska seems to vary little between species. The differences that were observed were a result of seasonal and environmental factors.

Appendices

Appendix A

Amino acid analysis

One composite sample of 40 g representing equal weights of each of the six KS2 samples was prepared for amino acids (AA) analysis. Two composite samples of 40 g each, representing equal weights of the eight SS samples and of the eight WS samples were also prepared for (AA) analysis. Three of the five KS1 samples were also used for determination of (AA) composition. Samples were hydrolyzed with 6N HCl and 2% phenol at 110^{o}C for 22 h. Amino acids were quantified using a Beckman 6300 analyzer (GMI Inc., Ramsey, MN) with post-column ninhydrin derivatization. Amino acid profiles were determined by AAA Service Laboratory, (Boring, OR). Table A.1 provides results for this analysis.

Scallop samples for the different groups had AA profiles that appeared comparable, with individual AA showing values in the same order of magnitude. Although to assess differences, analysis of a larger sample size for each group would be required.

Table A.1.

Amino acid composition (% w/w ± SD) of purple-hinge rock scallops and weathervane scallops from the Gulf of Alaska.

	WS (n = 1)	SS (n = 1)	KS1 (n = 3)	KS2 (n = 1)	PHRS (n = 1)
Alanine (ALA)	5.30	5.54	5.25 ± 0.10	5.23	5.77
Arginine (ARG)	8.88	7.53	7.15 ± 0.39	6.90	8.17
Aspartic Acid (ASP)	10.27	10.17	10.15 ± 0.04	9.98	10.75
Glutamic Acid (GLU)	16.13	16.20	15.32 ± 0.16	15.97	17.42
Glycine (GLY)	10.35	12.16	12.73 ± 0.82	13.05	8.45
Histidine (HIS) *	2.21	2.23	2.11 ± 0.02	2.22	2.13
Isoleucine (ILE) *	4.41	4.28	4.45 ± 0.02	4.35	4.42
Leucine (LEU) *	7.96	7.85	7.99 ± 0.05	7.88	8.04
Lysine (LYS) *	8.13	8.08	8.36 ± 0.06	8.16	8.22
Methionine (MET) *	3.18	3.06	3.24 ± 0.05	3.11	3.11
Phenylalanine (PHE) *	4.24	4.19	4.15 ± 0.04	4.16	4.09
Proline (PRO)	2.95	2.92	3.29 ± 0.10	3.19	2.75
Serine (SER)	3.90	3.91	3.71 ± 0.02	3.79	4.32
Threonine (THR) *	4.25	4.18	4.30 ± 0.07	4.23	4.36
Tyrosine (TYR)	3.59	3.57	3.42 ± 0.02	3.38	3.72
Valine (VAL) *	4.24	4.14	4.35 ± 0.08	4.40	4.26
Σ Essential AA *	38.62	38.01	38.97 ± 0.37	38.50	38.63
Σ Non-essential AA	61.38	61.99	61.03 ± 0.37	61.50	61.37

SD Standard deviation, WS Weak meat scallops, SS Standard scallops, KS1 Kodiak scallop 1, KS2 Kodiak scallop 2, PHRS Purple-hinge rock scallop.

Appendix B

Total carbohydrate analysis: Oyster glycogen (type II) calibration curve, detection limits and derivation of equation

A calibration curve plotting absorbance vs. concentration of oyster glycogen type II (Sigma) was determined to establish the optimum absorbance range for the anthrone method (Oliveira et al. 2006). Based on Beer's Law, the optimum range is the linear portion of the curve (Penner 2003). The complete curve is shown in Fig. B.1 (A) and the linear portion of the curve is depicted in Fig. B.1 (B). The lower limit of absorbance is 0.18 and the upper limit is 1.5. This range corresponds exactly to the absorbance range of 0.2 to 1.5 suggested by Penner (2003) as the optimum linear range for spectrophotometric determination of a variety of analytes. Table B.1 gives the absorbance of each of the scallop samples investigated, and it shows that values for PHRS, KS2 and SS were well within the linear limits as shown in Fig. B.1 (B) while WS fell far below the lower limit of detection. Applying Beer's Law and the optimum absorbance range suggested by Penner (2003) to the results shown in Fig. B.1, the lower absorbance limit of 0.18 was adopted. This corresponds to a total carbohydrate content of 0.62%, and this value corresponds to the lower limit of detection of the method.

The equation defined by Oliveira et al. (2006) where % total carbohydrates = 25 x A2/A1 x W (g) is derived in the following way based on the equation of Strickland and Parsons (1972) where mg glucose/m^3 = corrected extinction x F/V. Because the equation was derived for tissue instead of seawater the assumption is made that m^3 = 1 L = 1 Kg. Therefore mg glucose/Kg = corrected extinction factor x F/V where:

F = 1000/Es-Eb

Corrected extinction = extinction value of sample - Eb

V is volume of sample in L

Es is the extinction values of the glucose standard (0.1 mg /mL)

Eb is the extinction value of the blank

In Strickland and Parsons (1972) the sample volume for seawater is given as L; and we used this unit as Kg for tissue weight. So, there is a factor of 1,000 for sample weight (W) because in the Oliveira et al (2006) formula the sample weight is given in grams (or 0.001 Kg = 1 g). This 1,000 value is seen in Strickland and Parsons (1972) formula embedded into the variable 'F'. In addition, Oliveira et al. (2006) report their results in % w/w and therefore:

mg glucose/Kg = 0.1 mg glucose/100g

0.1 mg glucose/100 g = corrected extinction x F/V

1 mg glucose/100 g = % w/w = 10 x corrected extinction x F/V

In Oliveira et al. (2006) these factors are:

F = 0.001/Es-Eb

Es-Eb = A1

Corrected extinction = A2

V (L) = Sample W (g)

After reading the blank, the Varian Cary50 spectrophotometer automatically subtracts the blank value from the values measured for A1 and A2 so there is no need for the blank value to be incorporated into the formula.

The dilution of the tissue in the method described by Oliveira et al. (2006) needs to be accounted for in the equation and the factor used in this case is: 2,500 (25 x 100) because we account for 1 ml taken from 100 mL (1 part in 100), but all carbohydrates present in that 100 mL came from the 10 mL removed from the 250 mL flask, so the 1:25 parts factor needs to be multiplied by 100 only. Using the factors defined by Oliveira et al. (2006) the equation would now be:

$$\% \text{ w/w} = 10 \times 2500 \times A2 \times (0.001/A1)/W = 25 \times A2/A1 \times W \text{ (g)}$$

Total carbohydrate content was calculated for PHRS, SS and KS2 scallops using the formula of Oliveira et al. (2006) (TC1) and the linear regression equation shown in Fig. B 1 (B) (TC2). Both equations incorporate the measured absorbance of the samples. The range of TC1 content for the samples was from 0.77% to 2.68% the range of TC2 content was 0.63% to 2.44. The range of total carbohydrate calculated by these equations is similar and is also within the range of total carbohydrate reported for scallop muscle in the literature.

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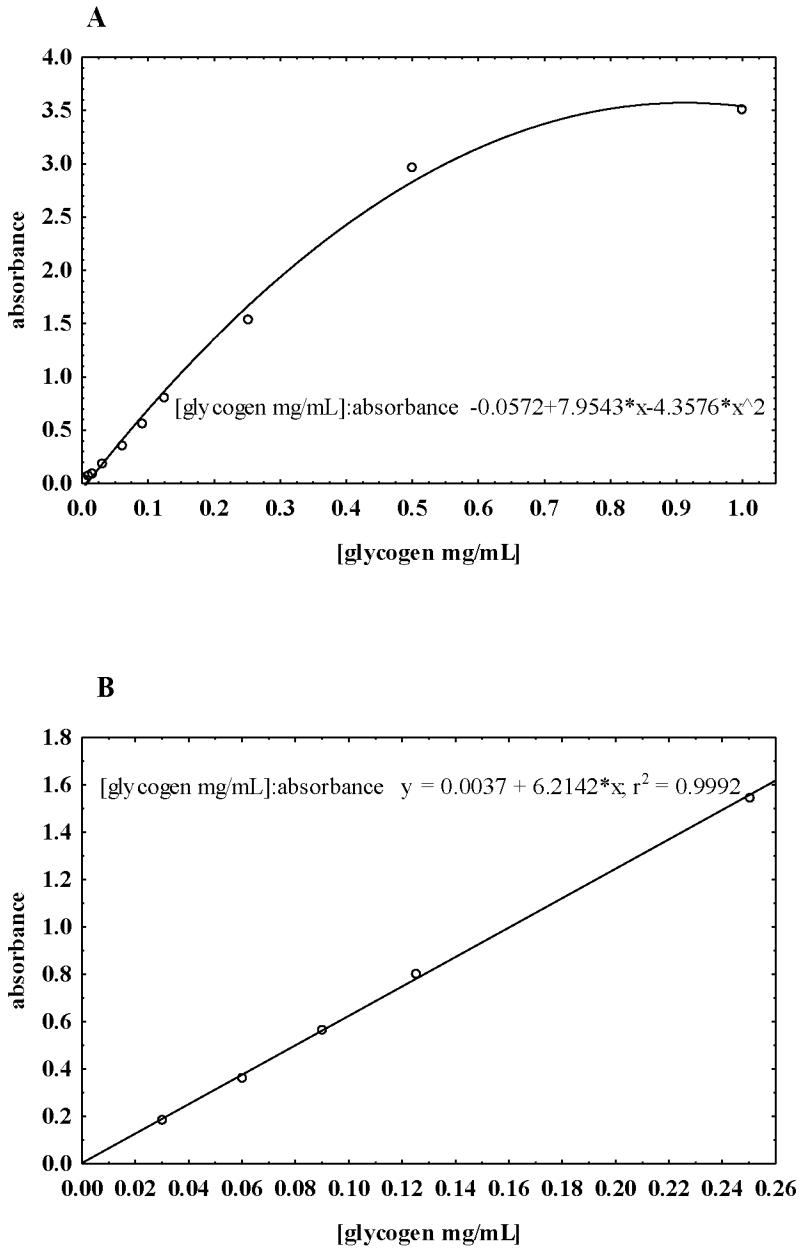

Figure B.1. Glycogen concentration calibration curve showing complete nonlinear curve (A) and the linear portion (B).

Table B.1.

Total carbohydrate content of purple-hinge rock scallops and weathervane scallops from the Gulf of Alaska calculated by two equations.

	Sample weight (g)	A1	A2	% TC1	% TC2
PHRS A	14.8	0.558	0.540	1.62	1.46
PHRS B	14.9	0.558	0.391	1.18	1.05
PHRS C	14.9	0.447	0.322	1.20	0.86
PHRS D	15.4	0.558	0.684	1.99	1.78
PHRS E	14.8	0.449	0.251	0.95	0.67
Avg ± SD	--	--	--	1.39 ± 0.41	1.16 ± 0.45
KS2 1A	15.5	0.563	0.945	2.68	2.44
KS2 1B	15.5	0.563	0.265	0.77	0.68
KS2 2A	15.3	0.563	0.732	2.12	1.91
KS2 2B	15.6	0.563	0.686	1.95	1.76
KS2 3A	15.2	0.449	0.311	1.15	0.81
KS2 3B	15.2	0.449	0.500	1.86	1.31
Avg ± SD	--	--	--	1.75 ± 0.69	1.48 ± 0.68
SS A	15.5	0.420	0.360	1.38	0.93
SS B	15.2	0.420	0.644	2.47	1.69
SS C	15.5	0.420	0.470	1.78	1.21
SS D	15.8	0.420	0.552	2.08	1.42
SS E	15.5	0.420	0.343	1.31	0.88
SS F	15.1	0.420	0.358	1.40	0.94
SS G	15.6	0.420	0.390	1.47	0.99
SS H	15.3	0.420	0.241	0.93	0.63
Avg ± SD	--	--	----	1.61 ± 0.49	1.09 ± 0.34
WS A	14.8	NR	<0.02	BDL	0.04
WS B	14.9	NR	<0.02	BDL	0.04
WS C	15.6	NR	<0.02	BDL	0.04
WS D	14.8	NR	<0.02	BDL	0.04
WS E	14.8	NR	<0.02	BDL	0.04
WS F	15.2	NR	<0.02	BDL	0.04
WS G	15.3	NR	<0.02	BDL	0.04
WS H	15.0	NR	<0.02	BDL	0.04
Avg ± SD	--	--	--		0.04 ± 0.00

PHRS Purple-hinge rock scallop, KS2 Kodiak weathervane scallop 2, SS Standard weathervane scallop, WS 'Weak meat' weathervane scallop, NR Not recorded, BDL Below detection limit, A1 Absorbance of glycogen standard (0.1mg/mL), A2 Absorbance of sample, TC1 % Total carbohydrate by Oliveira et al. (2006) equation, TC2 % Total carbohydrate by Fig. B 1 (B) regression equation.