Growth and condition of post-moult male snow crab (*Chionoecetes opilio*) in the laboratory

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Abstract

We monitored soft-shell and hard-shell legal-sized male snow crabs sampled periodically in the field or fed to satiation at 4.5 °C in the laboratory for changes in nutritional condition. In soft-shell crabs, feeding resulted in a significant decrease of water content in all tissues. This decrease was associated with increased lipid (digestive gland) and protein (muscle and haemolymph) contents. The weight of the muscle in the merus and the weight of the digestive gland also increased. Feeding had no impact on the nutritional condition of hard-shell crabs in the laboratory. In the wild, soft-shell crabs required a much longer period of time to recover to the hard-shell condition than was postulated from earlier studies; nutritional condition of soft-shell crabs did not improve markedly over summer and into autumn. The results of the present study are relevant to aquaculture and stock management. Landed legal-sized hard shell adolescent crabs could be held through terminal moult and grown to a marketable condition in a reasonably short period of time. Nutritional condition indices may also provide valuable information on several aspects of production and contribute to a precautionary management of the snow crab stocks. © 2002 Elsevier Science B.V. All rights reserved.

Keywords: Chionoecetes opilio; Snow crab; Condition indices; Proteins; Lipids; Growth

1. Introduction

The global crustacean harvest and aquaculture production is roughly 8 million metric tons, less than 5% of which can be ascribed to aquaculture. This percentage increases for shrimp and prawn species but is much lower for crab species despite high market values (FAO FISHSTAT database). This situation may be explained by the crabs' predatory and

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cannibalistic food habits (Dutil et al., 1997; Lovrich and Sainte-Marie, 1997) and also by the lack of knowledge on rearing methods. Profitability for crab aquaculture, as in other species, depends on a very short production cycle. This is a stringent requirement, particularly for cold-water aquaculture. Snow crabs (*Chionoecetes opilio*) live in water colder than 3–4 °C (Williams, 1984; Tremblay, 1997) and become metabolically deficient above 7.0 °C (Foyle et al., 1989b). Females require about 6 years (Alunno-Bruscia and Sainte-Marie, 1998) and males 10 years (Sainte-Marie et al., 1995) to reach maximum size in the northern Gulf of St. Lawrence. Males reach legal size (95-mm carapace width) in 8 years and one moult cycle may take as much as 1 or 2 years in large-sized individuals (Comeau et al., 1991; Sainte-Marie et al., 1995).

Two avenues have been considered to achieve profitability in snow crab aquaculture, live storage of landed hard-shell legal-sized adult individuals, i.e., male snow crabs larger than 95-mm carapace width in the terminal moult stage (Conan and Comeau, 1986; Sainte-Marie et al., 1995), and rearing of landed legal-sized adolescent individuals, i.e., individuals which may grow to a larger size at terminal moult. Landed inter-moult snow crabs of legal size are able to survive long periods of food deprivation with low rates of mortality and no marked effects on body condition at low temperatures, indicating that snow crabs, like lobsters, could be stored and marketed live year-round (Hardy et al., 1994, 2000). Soft-shell snow crabs in the wild require several months to reach the hard-shell condition (Watson, 1971; O'Halloran and O'Dor, 1988; Taylor et al., 1989). The rearing conditions that may promote faster growth rates and would reduce the duration of the post-moult period in legal-sized snow crabs under controlled conditions are unknown.

Although crustaceans increase in size at moult (e.g. Reilly and Saila, 1978; Hunt and Lyons, 1986), growth of soft tissues takes place during post-moult and inter-moult (El Haj et al., 1984; Mayrand et al., 2000). Intensive physiological and biochemical processes occur during these periods (El Haj and Whiteley, 1997), allowing the animals to recover from the demanding moult period and to proceed with the growth of soft tissues in preparation for the next moult until a terminal moult is achieved, as is the case in the snow crab (Conan and Comeau, 1986). Rearing conditions should promote the rapid growth of individual organs and, in particular, the rapid build up of muscle tissue during the post-moult period. Snow crabs below the legal size (66-mm carapace width) and fed different rations after terminal moult began feeding soon after moulting, but animals fed a high ration consistently ate all the food only after 30 days at 3 °C (Mayrand et al., 2000). Muscle growth was slow in the first month of the experiment, but more energy was directed, in decreasing order of priority, towards gonadal growth, muscle growth and reserve build-up in the digestive gland in the second month (Mayrand et al., 2000). While a number of studies have been conducted describing seasonal changes in the biochemical composition of various crustacean species (Barnes et al., 1963; Fernandes et al., 1994) including crabs (Heath and Barnes, 1970; Du Preez and McLachlan, 1983), very few studies have compared growth rates of soft tissues measured in the laboratory to those observed concurrently in the wild, particularly in decapod crustaceans (Jussila and Mannonen, 1997).

The present study examined growth and nutritional condition indices in fed legal-sized male snow crabs. We collected legal-sized soft-shell individuals from the fishery, fed them

natural food, and monitored changes in the mass and composition of several tissues during the post-moult and into the inter-moult period under controlled conditions. We had two objectives: to determine the length of time required to reach market quality from the soft-shell condition and to develop condition indices to be used in assessing the nutritional status in the wild and in aquaculture facilities. We compared muscle growth as well as the size and composition of the digestive gland in laboratory-fed soft-shell crabs to those of inter-moult hard-shell crabs in the wild.

2. Materials and methods

Recently moulted snow crabs (S_0) were sampled within a few hours to a couple of days after moulting from a group of crabs which had been caught in May and were held in our facility until moulting. Temperature and salinity of the holding tanks were not controlled during the holding and moulting period (18 May–17 June 1993) and averaged 4.9 ± 1.8 °C (3.4–8.6 °C) and 26.3 ± 2.0 (22.6–28.9), respectively.

Legal-sized (carapace width > 95 mm) male snow crabs were also collected from the St. Lawrence estuary in the summer and autumn of 1993 using a beam trawl and baited traps. Crabs with wounds or more than one limb missing were rejected. Soft-shell crabs were collected in early August (S_1, not fed; S_1F_1–S_1F_7, fed and sampled every 2 weeks from late September until late December) and in early October (S_2, not fed; S_2F, fed and sampled in late December) (Table 1). Hard-shell crabs were collected in early October (samples H_2, not fed; samples H_2F, fed and sampled in late December). Reference crabs (S_1, S_2, H_2) were not fed and were sampled after a few days in the holding tanks.

The crabs used in feeding experiments were weighed (total body weight—TW; ± 0.1 g) and identified individually with a Carlin dangler wire tag (Floy Tag, Seattle, WA, USA) tied to one of the pereopods. Feeding experiments were conducted in two rectangular tanks (1.2×3.6 m; 30-cm deep) each divided into four 1-m² compartments. Each tank was set up with its own temperature control and recirculation system in a semi-open design. Water quality (pH, ammonia, dissolved oxygen) was monitored in the first weeks of the experiment to ensure that the flow rate (8 l/min) was sufficient to maintain adequate water quality in the tanks. Temperature was controlled during the experiment whereas salinity was not. They averaged 4.5 ± 0.2 °C and 26.9 ± 1.4, respectively. Crabs were fed twice a week a ration roughly equivalent to 0.5% of their total mass per day using equal portions of shrimp (*Pandalus borealis*) and capelin (*Mallotus villosus*). The initial density of crabs in the tanks was approximately 25 kg m² but was not constant over the course of the experiment as the crabs were sampled periodically. Not all food was consumed between meals, indicating that crabs were fed ad libitum; however, food ingestion was not determined.

2.1. Dissection and analyses

We measured carapace width (± 0.1 mm), chela height (± 0.1 mm), total body weight (TW; ± 0.1 g); length, height, width and muscle weight (± 0.1 g) of the second

Table 1
Source, treatment, and moult stage of legal-sized male snow crabs used in a growth experiment in tanks at 4.5 °C

Groups	n	Source and treatment	Moult stage
S_0	16	Post-moult crabs from holding tanks at the laboratory. Collected in spring 1993 (6–13 May). Sampled shortly after moulting between 18 May and 17 June 1993 (day 0 = 1 June 1993). No nourishment.	A–B
S_1	10	Soft-shell crabs captured on the north shore of the St. Lawrence estuary between 5 and 13 August. No nourishment; sampled on 16 August (day 60).	Mainly C_1–C_3
S_2	10	Soft-shell crabs captured on the south shore of the St. Lawrence estuary between 1 and 5 October. No nourishment; sampled on 5 October (day 110).	Mainly C_1–C_3
S_2F	7	Soft-shell crabs captured on the south shore of the St. Lawrence estuary between 1 and 5 October. Kept in tanks and nourished from 6 October until sampling on 20 December (day 186).	Mainly C_4
H_2	10	Hard-shell crabs captured on the south shore of the St. Lawrence estuary between 1 and 5 October. No nourishment; sampled on 6 October (day 111).	C_4–D_0
H_2F	10	Hard-shell crabs captured on the south shore of the St. Lawrence estuary between 1 and 5 October. Held in tanks and nourished from 6 October until sampling on 21 December (day 187).	C_4
S_1F_1–S_1F_7	10	Soft-shell crabs captured on the north shore of St. Lawrence estuary between 5 and 13 August. Held in tanks and nourished from 19 August until sampling on 27 September (S_1F_1, day 102), 12 October (S_1F_2, day 117), 26 October (S_1F_3, day 131), 8 November (S_1F_4, day 144), 23 November (S_1F_5, day 159), 7 December (S_1F_6, day 173) and December 20 (S_1F_7, day 186).	C_1–D_0

Moult stages were determined using the scale described in O'Halloran and O'Dor (1988).
All crabs from the north shore were caught using a beam trawl. Those from the south shore were caught using baited traps.

right merus (± 0.1 mm); weight of the digestive gland (DW; ± 0.1 g); and weight of the remaining viscera including testes and vasa deferentia (VW, ± 0.1 g). The volume of the merus was estimated as the product of its length, height and width. The right chela was weighed (± 0.01 g). The left chela and samples of the dorsal shell, merus muscle, and digestive gland were weighed (± 0.01 g) and dried at 96 °C to constant weight (48 h) for the determination of water content.

Moult stage was assessed from setal development observations (O'Halloran and O'Dor, 1988). The hardness of the lower surface of the right chela was measured with a durometer using a 7-lb (31.14 N) gauge (PTC Instruments model 307HS). The durometer scale is in arbitrary units (0–100) and actual forces can be calculated since the force–scale relationship is linear (Foyle et al., 1989a). Shell hardness was not

measured on crabs that had recently moulted (S_0) since their shell was too soft to allow the use of the durometer. The distinction between two maturity status, i.e., adult (large-clawed) and adolescent (small-clawed) crabs, was based on the discriminant function established using the relation between chela height and carapace width for a sample of male crabs from the same area (Sainte-Marie and Hazel, 1992).

The haemolymph was sampled with a disposable ice-cold syringe via the arthrodial membrane at the base of one of the rear pereopod's coxa. The 5-ml sample was immediately transferred to an ice-cold tube and centrifuged for 5 min at 10,000 rpm. The plasma was then split into 0.5-ml aliquots and held in microtubes at -20 °C for assays. Plasma glucose (mg/100 ml) was determined using an enzymatic diagnostic kit (Sigma #315 Glucose Trinder) and plasma proteins (g/l) with a dye binding method using Coomassie Brilliant Blue G-250 (Bio-Rad protein assay). We also weighed and dried a sample of haemolymph (2–12 g) for the determination of water content.

Samples of the digestive gland held at -80 °C were used for the determination of lipid and protein contents. Lipid content was determined gravimetrically (Soxhlet extraction procedure, Soxtec unit 1043). Protein content was calculated by subtracting N from non-protein sources from total nitrogen. Nitrogen was extracted on a Kheltec system using the micro-Kjeldahl method with or without protein precipitation with K_2SO_4 and assessed spectrophotometrically using the Berthelot reaction (Haslemore and Rougham, 1976). We used a factor of 6.25 to convert protein nitrogen into protein contents.

2.2. Statistical analysis

Variables were checked for normality and homogeneity of variance using Wilks–Shapiro and Levene tests. Variables with normal distribution and homogenous variances were tested by one-way ANOVA followed by Tukey's a posteriori multiple comparisons test (Zar, 1996). When these conditions were not met, Kruskal–Wallis tests were conducted followed by Tukey's tests. A posteriori tests were carried out only when the ANOVA or Kruskal–Wallis showed significant differences ($P < 0.05$). Merus muscle weight is expressed as a ratio to merus volume. Digestive gland, viscera and chela weights are expressed as ratios with the crab total body weight and crab total body weight as a ratio with its carapace width.

Relationships between variables were examined using least-square regression analysis. ANCOVA analyses were used to check for the influence of maturity status on these relationships. In the event of a significant interaction between maturity status and the independent variable, we used multiple regression coupled with an ANOVA.

All statistical analyses were carried out using the SAS for Windows software release 6.12. All averages given in the text are arithmetic means with standard deviations.

3. Results

Maturity status and crab size differed among samples. While crabs in most samples were adults, S_1F_6 and S_1F_7 crabs were mainly adolescents (Table 2). Differences in

Table 2
Numbers of adult and adolescent male snow crabs and corresponding carapace width (mm ± SD) in each sample of a growth experiment at 4.5 °C

Sample	Adolescent (percentage)	Adults (percentage)	Average carapace width (mm)
S_1F_1	0 (0%)	10 (100%)	121.16 (± 6.18)
S_1F_2	1 (10%)	9 (90%)	119.77 (± 9.44)
S_1F_3	3 (30%)	7 (70%)	116.46 (± 5.84)
S_1F_4	3 (30%)	7 (70%)	114.66 (± 10.76)
S_1F_5	2 (20%)	8 (80%)	110.17 (± 9.81)
S_1F_6	8 (80%)	2 (20%)	99.03 (± 5.80)
S_1F_7	9 (90%)	1 (10%)	97.34 (± 5.94)
H_2	0 (0%)	10 (100%)	114.09 (± 8.13)
H_2F	1 (10%)	9 (90%)	103.18 (± 4.68)
S_0	3 (18.75%)	13 (81.25%)	117.35 (± 11.99)
S_1	0 (0%)	10 (100%)	119.94 (± 8.62)
S_2	0 (0%)	10 (100%)	122.83 (± 9.33)
S_2F	1 (14.29%)	6 (85.71%)	117.66 (± 15.63)

Soft-shell crabs were sampled periodically during the experiment (S_1F_1–S_1F_7). Reference wild soft-shell (S_1, S_2) and hard-shell (H_2) crabs were sampled after a few days in holding tanks and were not fed. Additional soft-shell (S_2F) and hard-shell (H_2F) crabs were fed and sampled at the end of the experiment. S_0 were crabs sampled within a few days following ecdysis.

carapace widths were significant ($F = 9.52$, $P < 0.001$; Table 2). S_1F_6 (99.0 ± 5.8 mm), S_1F_7 (97.3 ± 5.9 mm) and H_2F (103.2 ± 4.7 mm) crabs were significantly smaller than S_1F_1 (121.2 ± 6.2 mm), S_1F_2 (119.8 ± 9.4 mm), S_0 (117.3 ± 12.0 mm), S_1 (119.9 ± 8.6 mm), and S_2 (122.8 ± 9.3 mm) crabs. While crabs sampled in the S_1F_1–S_1F_7 series had progressively smaller carapace widths, only S_1F_6 and S_1F_7 crabs differed significantly. Total body weights showed similar variations as observed for carapace widths ($F = 11.81$, $P < 0.001$). The regressions between TW and carapace width for adults and adolescents differed significantly ($F = 3.986$, $P = 0.048$), with adults showing a slightly smaller increase in weight for each unit increase in width. During the feeding experiments, TW did not increase ($P > 0.05$) except for S_1F_3 crabs ($t = 2.8$, $P = 0.02$).

Shell hardness did not increase in fed soft-shell crabs in the laboratory experiments (S_1F_1 through S_1F_7). While it did increase between early August and early October in the field samples, shell hardness of crabs that moulted in 1993 never reached the level measured in hard-shell crabs in early October (Fig. 1). Shell hardness varied considerably among samples ($F = 16.69$, $P < 0.001$). We could not detect significant groupings of samples using Tukey's a posteriori test. However, we were able to observe certain tendencies: S_0 crabs had a much softer shell than crabs in all other groups, but shell hardness could not be measured with the gauge selected. S_1 crabs had softer shells than any other crabs from the remaining samples (hardness of ∼ 45). Crabs in groups H_2, H_2F, and S_2F were all similar and had the hardest shells (∼ 83). S_1F_1–S_1F_7 and S_2 crabs formed a third group with intermediate shell hardness (∼ 67). S_2F crabs included an outlier with a shell hardness of 55 compared to an average 84 in the rest of the sample. When this outlier was excluded from the analysis, the trend described above became significant. Similarly, relative chela weight varied among samples ($F = 21.96$,
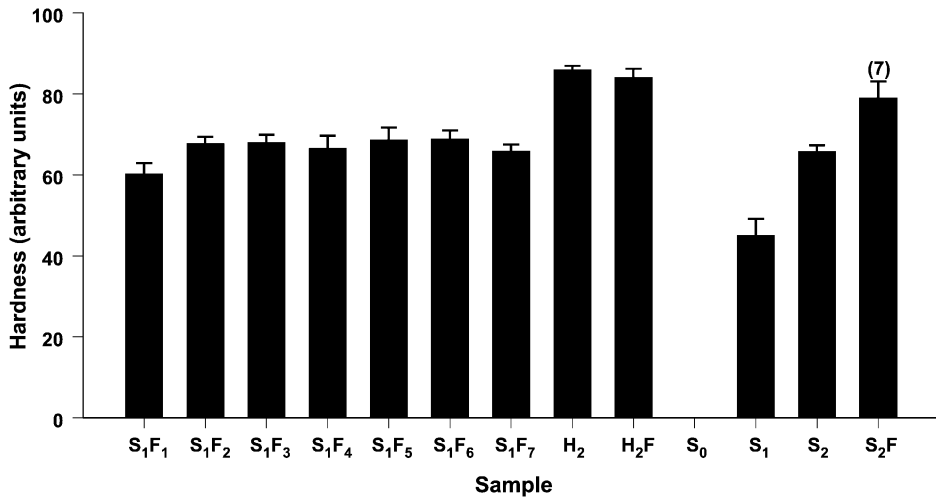
Fig. 1. Shell hardness of male snow crabs in a growth experiment at 4.5 °C. Soft-shell crabs were sampled periodically during the experiment ($S_1F_1-S_1F_7$). Reference wild soft-shell (S_1, S_2) and hard-shell (H_2) crabs were sampled after a few days in holding tanks and were not fed. Additional soft-shell (S_2F) and hard-shell (H_2F) crabs were fed and sampled at the end of the experiment. S_0 were crabs sampled within a few days following ecdysis. Error bars represent standard error; (n) indicates the sample size when different from 10. There is no shell hardness data for S_0 since the shell was too soft to measure with the durometer.

$P < 0.001$). The Tukey test detected two groups of data: S_0 vs. all the other samples. The mean ratio was much smaller in the S_0 sample (0.014 ± 0.003 g chela weight/g TW) than in the other samples. There were no significant differences among the $S_1F_1-S_1F_7$ and S_2 samples (~ 0.025 g chela weight/g TW) or among the H_2, H_2F, and S_2F samples (~ 0.040 g chela weight/g TW). However, S_1 (0.021 ± 0.004 g chela weight/g TW) and S_2F (0.034 ± 0.008 g chela weight/g TW) crabs were significantly different.

Relative viscera weight showed marked differences ($F = 20.67$, $P < 0.001$) among samples, with hard-shell crabs having larger VW than soft-shell crabs (Fig. 2). Samples H_2 and H_2F (0.0097 and 0.0111 g VW/g TW) had higher relative viscera weights than crabs in any other sample. There was no variation within the $S_1F_1-S_1F_7$ sample series except for S_1F_6 (0.0061 g VW/g TW), which was significantly greater than S_1F_1 and S_1F_2 (0.0036 and 0.0039 g VW/g TW, respectively). While there were no significant differences among the S_0, S_1, S_2, and S_2F samples, relative viscera weights in S_0 and S_1 were significantly smaller than in the $S_1F_5-S_1F_7$ samples.

Relative digestive gland weight increased significantly with feeding duration and also showed significant variability ($F = 14.29$, $P < 0.001$) from one sample to the next (Fig. 2). The Tukey test did not detect any specific grouping of samples, but it did identify S_1F_1 and S_1F_2 (0.0418 and 0.0425 g DW/g TW) as being significantly smaller than S_1F_6 and S_1F_7 (0.0804 and 0.0796 g DW/g TW). In fact, S_1F_6 and S_1F_7 had the largest digestive glands. The relative digestive gland weight in S_0 (0.0625 g DW/g TW) was significantly larger than in S_1 and S_2 (0.0371 and 0.0365 g DW/g TW, respectively) but
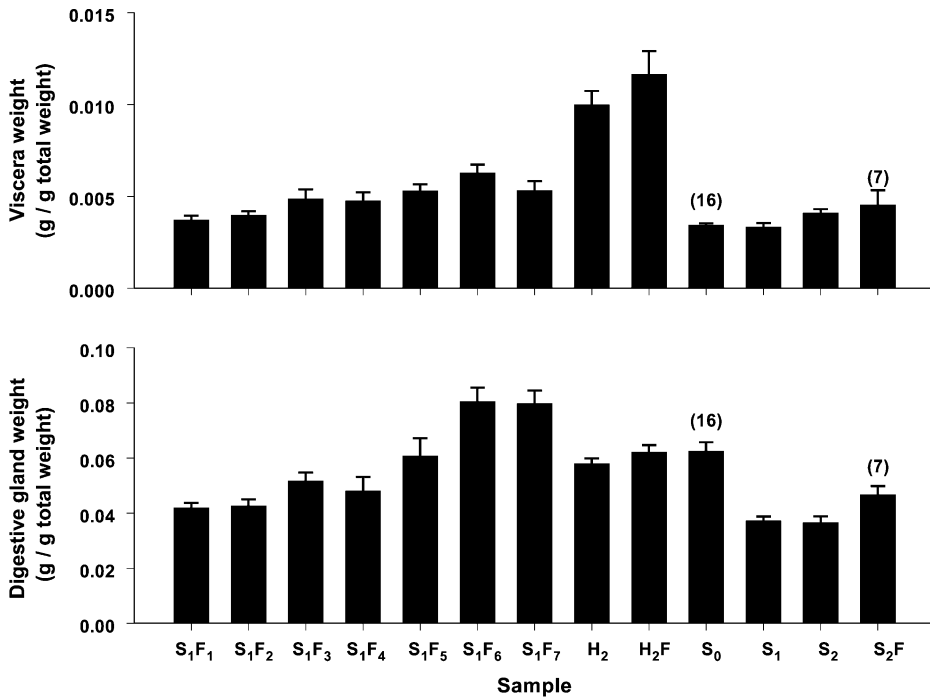
Fig. 2. Weight of the viscera (including gonads but not digestive gland) and digestive gland relative to total weight of male snow crabs in a growth experiment at 4.5 °C. Soft-shell crabs were sampled periodically during the experiment (S_1F_1–S_1F_7). Reference wild soft-shell (S_1, S_2) and hard-shell (H_2) crabs were sampled after a few days in holding tanks and were not fed. Additional soft-shell (S_2F) and hard-shell (H_2F) crabs were fed and sampled at the end of the experiment. S_0 were crabs sampled within a few days following ecdysis. Error bars represent standard error; (n) indicates the sample size when different from 10.

not S_2F (0.0466 g DW/g TW). S_2 was also different from the S_1F_5–S_1F_7 samples. The H_2 and H_2F samples did not differ and were similar to the S_1F_3–S_1F_5, S_0, and S_2F samples.

There were significant differences among samples in the lipid ($F = 4.669$, $P < 0.001$) and water ($F = 4.758$, $P < 0.001$) contents of the digestive glands (Fig. 3). S_2 had the highest (0.722 g water/g DW) and S_1F_7 the lowest (0.594 g water/g DW) water contents. S_2 differed significantly from H_2 (0.625 g water/g DW) and H_2F (0.599 g water/g DW) as well as from S_1F_6 (0.622 g water/g DW) and S_1F_7 (0.594 g water/g DW) samples. S_2 also had a higher water content than the S_1 (0.631 g water/g DW) samples. The decreasing trend of water content observed in the S_1F_1–S_1F_7 series was not significant. These variations were roughly a mirror image of the lipid content (Fig. 3), resulting in a highly significant negative relationship between lipid and water contents (Fig. 4). An analysis of covariance indicated that maturity status had no significant effect on this relationship ($F = 0.746$, $P = 0.39$).

Protein contents in the digestive gland showed significant differences ($F = 7.426$, $P < 0.001$) even though they were lower and had a smaller range than the lipid contents.
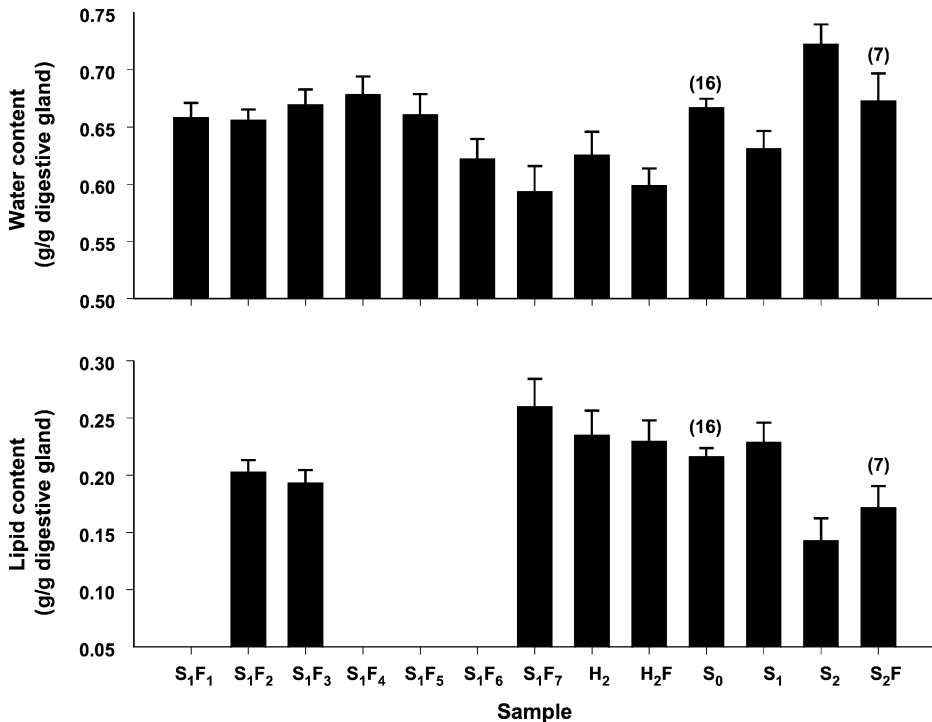
Fig. 3. Water and lipid contents of the digestive gland of male snow crabs in a growth experiment at 4.5 °C. Soft-shell crabs were sampled periodically during the experiment (S_1F_1–S_1F_7). Reference wild soft-shell (S_1, S_2) and hard-shell (H_2) crabs were sampled after a few days in holding tanks and were not fed. Additional soft-shell (S_2F) and hard-shell (H_2F) crabs were fed and sampled at the end of the experiment. S_0 were crabs sampled within a few days following ecdysis. Error bars represent standard error; (n) indicates the sample size when different from 10.

Protein contents of S_0 (0.042 g protein/g DW) and S_1 (0.046 g protein/g DW) were significantly smaller than S_2 (0.064 g protein/g DW). Protein content of the S_0 sample was also significantly smaller than S_2F (0.059 g protein/g DW). H_2 (0.066 g protein/g DW) had a higher protein content than S_1F_7 (0.045 g protein/g DW), S_0 (0.042 g protein/g DW) and S_1 (0.046 g protein/g DW), but H_2F (0.053 g protein/g DW) did not differ significantly from the other samples. Protein content was larger in S_1F_2 crabs (0.056 g protein/g DW) than in the S_0 samples.

Feeding promoted a significant increase in the merus muscle weight to merus volume ratio ($F = 34.69$, $P < 0.001$) (Fig. 5). The results of the Tukey analysis indicated that feeding resulted in muscle growth as indicated by an increased filling of the shell; later samples in a series had a greater merus muscle weight to merus volume ratio than earlier ones (S_1F_6–$S_1F_7 > S_1F_1$–S_1F_5, $S_2F > S_1$ and S_2). Furthermore, hard shell crabs had the highest ratios (0.518 and 0.535 g/cm^{-3} for H_2 and H_2F, respectively), but S_1F_6 (0.452 g/cm^{-3}) and S_1F_7 (0.461 g/cm^{-3}) crabs did not differ significantly. S_1 (0.272 g/cm^{-3})
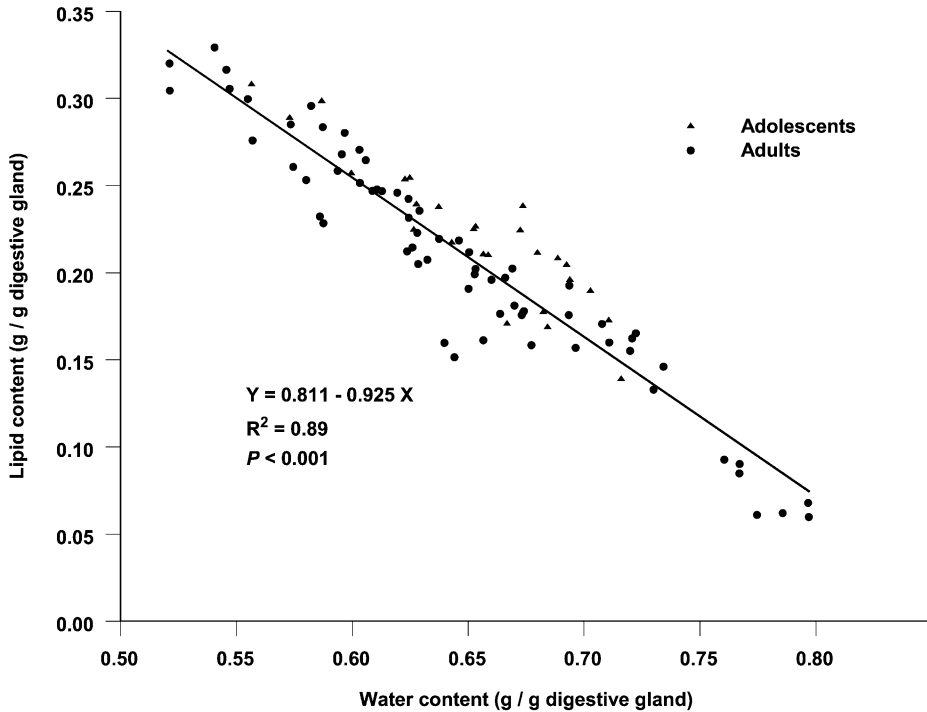
Fig. 4. A least-square linear regression between lipid and water contents in the digestive gland of adult and adolescent male snow crabs in a growth experiment at 4.5 °C.

and S_2 (0.296 g/cm^{-3}) had ratios similar to the S_1F_1 (0.279 g/cm^{-3}) through S_1F_4 (0.326 g/cm^{-3}) samples. S_1F_5 (0.373 g/cm^3) was different from S_1F_1, S_1F_6 and S_1F_7. These variations were inversely related to changes in the muscle water content (Fig. 5), which were also significant ($F = 30.63$, $P < 0.001$). In this case, the Tukey test detected two distinct groups of samples: H_2–H_2F and S_1F_6–S_1F_7 vs. S_1F_1–S_1F_5 and S_0, S_1, S_2 and S_2F. We observed a steady decrease of water content from S_1F_3 through S_1F_6.

Water content of the chela followed a different pattern than water content of the merus muscle. Significant variations were observed ($F = 48.51$, $P < 0.001$) due to the significantly higher values for S_0 (0.842 g water/g chela) and to a lesser extent S_1 (0.727 g water/g chela) than in crabs from other samples ($\sim$ 0.662 g water/g chela). There was a no significant decrease in the water content from S_1F_1 (0.688 g water/g chela) to S_1F_7 (0.643 g water/g chela). Similarly, two distinct groups were observed for water content in the shell ($F = 19.02$, $P < 0.001$): S_0 (0.604 g water/g shell) vs. the rest ($\sim$ 0.444 g water/g shell). The H_2 (0.479 g water/g shell) and H_2F (0.475 g water/g shell) samples had significantly higher water contents than S_1F_6 (0.423 g water/g shell), S_1F_7 (0.427 g water/g shell), S_1 (0.427 g water/g shell), and S_2 (0.411 g water/g shell).

Haemolymph glucose content was highly variable as shown by the large coefficients of variation calculated from Table 3. There were significant differences among groups
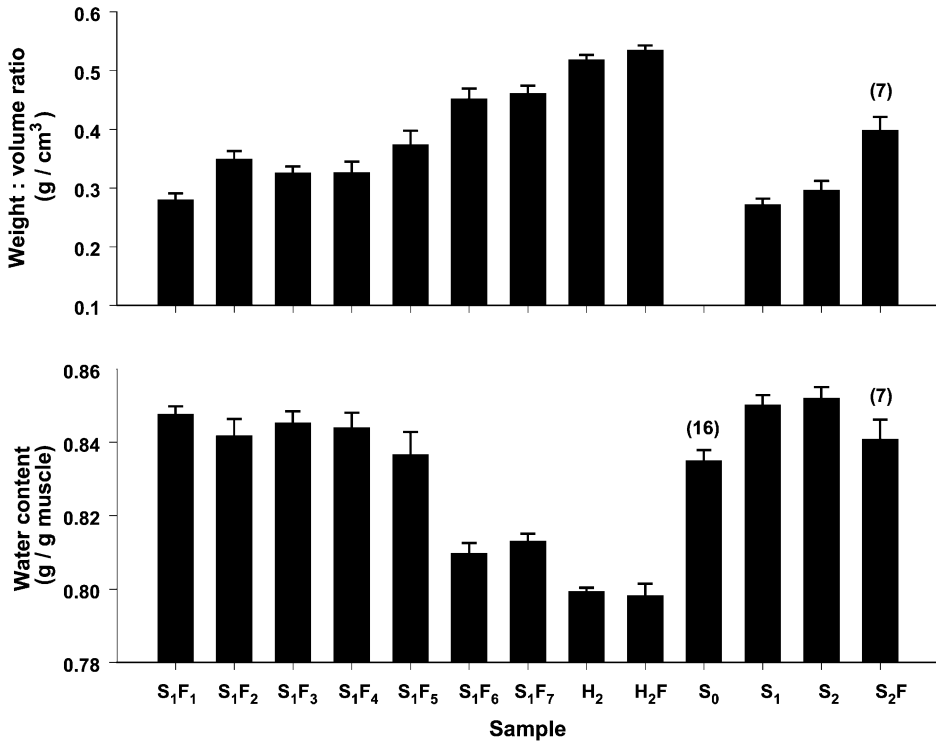
Fig. 5. Muscle weight–volume ratio and muscle water content of the merus of male snow crabs in a growth experiment at 4.5 °C. Soft-shell crabs were sampled periodically during the experiment (S_1F_1–S_1F_7). Reference wild soft-shell (S_1, S_2) and hard-shell (H_2) crabs were sampled after a few days in holding tanks and were not fed. Additional soft-shell (S_2F) and hard-shell (H_2F) crabs were fed and sampled at the end of the experiment. S_0 were crabs sampled within a few days following ecdysis. Error bars represent standard error; (n) indicates the sample size when different from 10.

($\chi^2 = 43.37$, $P < 0.001$), but Tukey tests were unable to detect distinct groupings. S_1F_2 (3.92 mg glucose/100 ml haemolymph) differed from S_1F_7 (8.36 mg glucose/100 ml haemolymph) and S_1F_5 (7.75 mg glucose/100 ml haemolymph) did not differ from either S_1F_2 or S_1F_7. H_2 (6.13 mg glucose/100 ml haemolymph) and H_2F (7.97 mg glucose/100 ml haemolymph) did not differ significantly from the other samples. No significant variations were detected among S_0, S_1, S_2 and S_2F samples although S_2 (3.44 ± 1.63 mg glucose/100 ml haemolymph) was significantly smaller than S_1F_7.

Haemolymph protein contents were greater and haemolymph water contents lower in hard-shell than in soft-shell crabs (Table 3). Protein and water contents showed significant variations ($F = 49.50$, $P < 0.001$ and $\chi^2 = 104.20$ $P < 0.001$, respectively). H_2 and H_2F were detected by the Tukey test as having significantly higher protein (77.38 and 75.40 g protein/l haemolymph, respectively) and lower water (0.913 and 0.919 g water/g haemolymph) contents than any other samples (~ 20 g protein/l haemolymph and ~ 0.954 g water/g haemolymph).

Table 3
Haemolymph composition of male snow crabs in a growth experiment at 4.5 °C

Sample	Glucose (mg/100 ml)	Protein (g/l)	Water (g/g)
S_1F_1	N/A	N/A	0.963 ($\pm$ 0.007)
S_1F_2	3.92 ($\pm$ 3.07)	16.81 ($\pm$ 11.49)	0.954 ($\pm$ 0.007)
S_1F_3	N/A	N/A	0.952 ($\pm$ 0.005)
S_1F_4	N/A	N/A	0.956 ($\pm$ 0.007)
S_1F_5	7.75 ($\pm$ 1.84)	25.87 ($\pm$ 12.32)	0.949 ($\pm$ 0.011)
S_1F_6	N/A	N/A	0.938 ($\pm$ 0.008)
S_1F_7	8.36 ($\pm$ 2.61)	35.39 ($\pm$ 7.75)	0.937 ($\pm$ 0.008)
H_2	6.13 ($\pm$ 1.39)	77.38 ($\pm$ 17.24)	0.913 ($\pm$ 0.014)
H_2F	7.97 ($\pm$ 1.42)	75.40 ($\pm$ 17.16)	0.919 ($\pm$ 0.014)
S_0	6.73 ($\pm$ 4.53)	17.64 ($\pm$ 6.47)	0.959 ($\pm$ 0.003)
S_1	5.07 ($\pm$ 1.64)	5.80 ($\pm$ 4.04)	0.968 ($\pm$ 0.003)
S_2	3.44 ($\pm$ 1.63)	13.16 ($\pm$ 5.86)	0.964 ($\pm$ 0.003)
S_2F	6.56 ($\pm$ 3.69)	28.04 ($\pm$ 9.04)	0.953 ($\pm$ 0.009)

Soft-shell crabs were sampled periodically during the experiment ($S_1F_1 - S_1F_7$). Reference wild soft-shell (S_1, S_2) and hard-shell (H_2) crabs were sampled after a few days in holding tanks and were not fed. Additional soft-shell (S_2F) and hard-shell (H_2F) crabs were fed and sampled at the end of the experiment. S_0 were crabs sampled within a few days following ecdysis. All data presented are means $\pm$ SD.

A clear increase was observed in the haemolymph protein contents as S_1F_2 (16.81 g protein/l haemolymph) was significantly smaller than S_1F_7 (35.39 g protein/l haemolymph) while S_1F_5 (25.87 g protein/l haemolymph) did not differ from either S_1F_2 or S_1F_7. Also, crabs of the S_2F sample had significantly higher protein contents (28.04 g protein/l haemolymph) than S_1 and S_2 crabs (5.80 and 13.16 g protein/l haemolymph, respectively). There were no significant variations in water content in the $S_1F_1 - S_1F_7$ series samples until S_1F_4. All had means of about 0.95 g water/g haemolymph, which is higher than S_1F_6 (0.938 g water/g haemolymph) and S_1F_7 (0.937 g water/g haemolymph). S_1F_5 (0.949 g water/g haemolymph) acted as a transition sample, being significantly different from both S_1F_1 and S_1F_7. The S_0, S_1 and S_2 samples had water contents similar to the S_1F_1 through S_1F_4 samples. However, the water content of S_2F (0.953 g water/g haemolymph) was lower than S_1 (0.968 g water/g haemolymph). Protein content in the haemolymph differed between adults and adolescents ($F = 7.77$, $P < 0.01$) and correlated significantly with water content in the haemolymph ($F = 237.5$, $P < 0.001$), but the interaction term was significant ($F = 7.70$, $P < 0.01$) indicating that slopes differed with a lesser increase in protein content per unit decrease in water content in adolescents (Fig. 6):

$$\text{Adults} \qquad \text{PROTEIN}^{0.7} = 299.3 - 305.1 \times \text{WATER} \qquad (1)$$

$$\text{Adolescents} \qquad \text{PROTEIN}^{0.7} = 211.08 - 212.0 \times \text{WATER} \qquad (2)$$

3.1. Discussion

Condition indices have been used in Atlantic cod (*Gadus morhua*) to provide key information on various population parameters including natural mortality (Dutil and
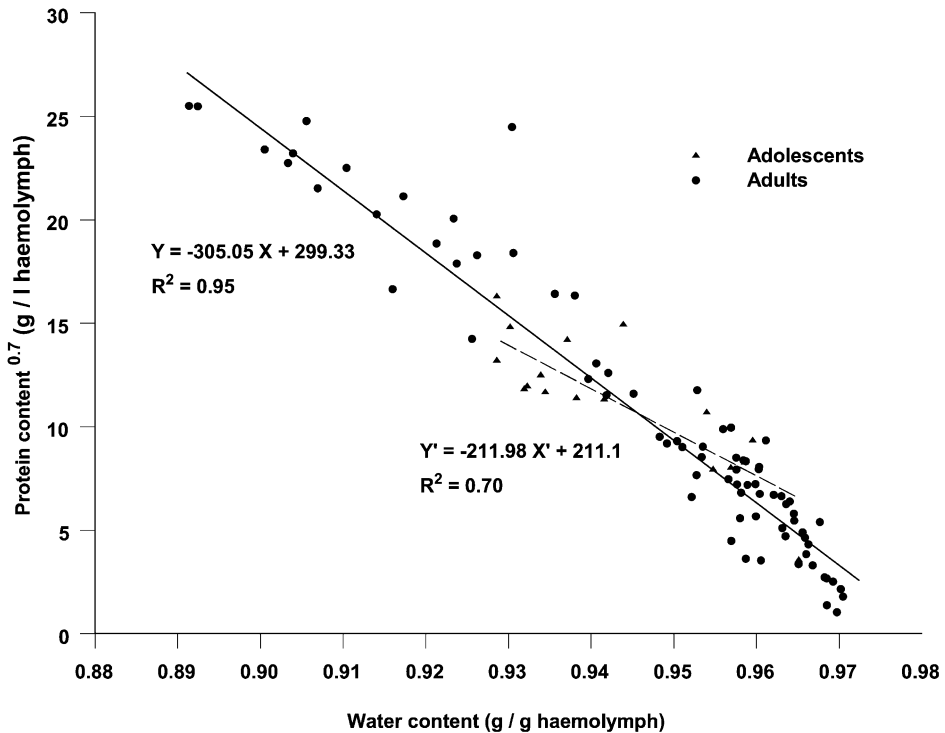
Fig. 6. Least-square linear regressions of haemolymph protein and water contents of adult (solid line) and adolescent (dashed line) male snow crabs in a growth experiment at 4.5 °C. One outlier from the H$_2$F group (fed hard-shell crabs) was eliminated from the calculations of the adult regression. The protein content data were transformed to achieve homogeneity of variance using an exponential factor of 0.7 that was determined by an optimal power (Box–Cox) analysis.

Lambert, 2000), reproductive investment (Lambert and Dutil, 2000), and spawning stock biomass (Marshall et al., 1999). Fulton's condition factor, a ratio of somatic weight to fish length, and the liver–somatic index, a ratio of liver weight to somatic weight, have been correlated with energy content (Lambert and Dutil, 1997). Furthermore, muscle proteins and liver lipids were closely related to water content, Fulton's condition factor, and liver–somatic index (Lambert and Dutil, 1997).

Similar condition indices have been examined in crustaceans. Digestive gland size and composition generally change with starvation (Vogt et al., 1985; Niles et al., 1993) and food availability (Dall, 1974). The composition of muscle and other tissues also changes with food quality (Tsvetnenko et al., 1995) and food availability (Dall, 1974; Moore et al., 2000). The size and lipid content of the digestive gland and the protein content of the merus muscle changed in response to long-term starvation at three different temperatures in hard-shell legal-sized male adult snow crabs (Hardy et al., 2000). The metabolism and growth of muscle in the merus and dry weight of the digestive gland reflected ration level in early post-moult adult male snow crabs below legal size (Mayrand et al., 2000). Our

results concur with those of Hardy et al. (2000) and Mayrand et al. (2000). Because the size of the digestive gland and the ratio of merus muscle weight to merus volume reflect changes in food availability in controlled experiments, we suggest these parameters could potentially be used as indices of the nutritional condition of snow crabs in the wild. Moisture content of the digestive gland has also been proposed as an index of nutritional condition in snow crabs (Hardy et al., 2000). We have compared the lipid–water contents relationship presented in Hardy et al. (2000) to the one obtained in our study and found no significant difference (ANCOVA, $F = 2.87$, $P = 0.09$). The lack of a significant difference in this relationship between fed soft-shell crabs (our study) and starved hard-shell crabs (Hardy et al., 2000) suggests that the relative proportions of lipids and water in the digestive gland are robust indicators of male snow crab nutritional condition.

The use of condition indices based on haemolymph protein and water contents could provide us with a method for monitoring individual crabs over long periods of time. Dall (1974) concluded that quantitative measurements of blood constituents are meaningless unless related to blood volume: blood volume may decrease as condition improves, resulting in a decrease of total protein contents in the haemolymph in the absence of any change in protein contents. However, we found that protein contents increased as a result of feeding in our experiment.

Soft-shell crabs harvested in mid-summer (S_1) required a long period of feeding at 4.5 °C under laboratory conditions to regain the hard-shell condition; differences between soft-shell and hard-shell crabs were still present at the end of the experiment. Shell hardening may take between 2 and 3 months post-moult for crabs with a 120-mm carapace (Taylor et al., 1989); this period is even longer for smaller crabs (O'Halloran and O'Dor, 1988). Thus, we expected an increase in hardness over the 5-month period that separated the S_0 and S_2 samples. There was a dramatic increase in shell hardness between recently moulted crabs in June (S_0, moult as in our facility) and crabs sampled in the wild in mid-August (S_1) and a less dramatic increase from mid-August to early October (S_2). However, shell hardness in S_2 crabs was still significantly lower than in H_2 crabs in October (Fig. 1), indicating that shell hardening processes were not completed in the wild even after 150 days. Taylor et al. (1989) used evaluation criteria that allowed only for shells to be classified as soft ("Type 1"), hard and new ("Type 2"), or hard and old ("Type 3"), with no degree of variation within the categories. Foyle et al. (1989a), using a 7-lb gauge durometer, had set the lower limit to 72 to eliminate soft-shell crabs. Their measurements for Type 2 crabs were consistently lower than those for Type 3 crabs. Therefore, Type 2 crabs would be considered hard-shell but still be softer than crabs that made up the H_2 sample. Interestingly, the feeding of crabs that were captured in early October significantly improved the shell hardness in crabs categorized as soft-shell (S_2F vs. S_2) but not in those categorized as hard-shell (H_2F vs. H_2) (Fig. 1). Shell hardening processes in C. opilio may be limited by food availability. However, no significant variation was observed in shell hardness in crabs fed to satiation twice a week in our growth experiment ($S_1F_1 - S_1F_7$) although the shell water content decreased significantly. Shell hardening may also be a question of food quality, as postulated by O'Halloran and O'Dor (1988).

Several results further suggest that large male snow crabs need a very long period of time to recover from moulting. As size increases in crustaceans, moult interval increases and growth rate decreases (Hunt and Lyons, 1986; Westman et al., 1993). Longer moult

intervals with increasing size may result from an increasing demand in energy for moult and growth at moult. The relative weight of the digestive gland in soft-shell male snow crabs in our study was as large in the early post-moult period as it was in hard-shell crabs in early October. However, when samples were obtained from the estuary in early October (S_2), the relative weight of the digestive gland was lower, lipid content was lower and water content was higher than in earlier samples (S_0 and S_1). The water content was even higher in early October than at 60 days post-moult. The small relative weight of the digestive gland in mid-August (S_1) through early October (S_2) suggests a slow post-moult recovery for large soft-shell crabs in the wild. Soft-shell crabs collected in August and fed to satiation at 4.5 °C in the laboratory had recovered by late November, with digestive glands having similar relative weights and similar lipid and water contents as those in hard-shell crabs. Feeding at 4.5 °C through November and December promoted over-compensatory increases in energy reserves as reflected by the high lipid and low water contents in samples S_1F_6 and S_1F_7. Merus muscle growth was also slow in soft-shell crabs, but weight–volume ratios of merus muscle and water contents, which reflect protein contents (Hardy et al., 2000), were similar in fed soft-shell and hard-shell crabs by late November. In contrast, merus muscle weight–volume ratios did not vary significantly during the 60-day period between the S_1 and S_2 field samples, but it did increase in fed soft-shell crabs at 4.5 °C (S_2 vs. S_2F). Snow crabs do not feed in the pre-moult and early post-moult period (O'Halloran and O'Dor, 1988); feeding resumes progressively over a period of up to 4 weeks after ecdysis (Mayrand et al., 2000). Nevertheless, the muscle mass of snow crabs is expected to increase with time after ecdysis under suitable growth conditions (Mayrand et al., 2000), as was the case for fed soft-shell crabs in our study (S_2F and $S_1F_1 - S_1F_7$). Time seems to be a factor, as S_2F crabs generally did not attain levels similar to those of hard-shell crabs while S_1F_7 crabs did (Figs. 3 and 5). Snow crabs that received no food or were fed a low ration mobilized energy reserves from the digestive gland and exhibited low to negative merus muscle growth rates (Mayrand et al., 2000). Thus, we conclude that snow crabs were not under optimal growth conditions in the wild but were able to grow faster at a higher temperature under unlimited food availability. The lack of improvement in the nutritional condition of soft-shell crabs between mid-August (S_1) and early October (S_2) suggests that post-moult recovery is a long process in large-sized individuals that may last much longer than had been estimated in earlier studies (Watson, 1971; O'Halloran and O'Dor, 1988; Taylor et al., 1989). Although gonad size increased in unfed soft-shell crabs after moult to maturity, muscle growth was initially negative in crabs fed a low ration, which suggests that reproduction is a higher priority than muscle growth in adults (Mayrand et al., 2000). Whether the larger relative viscera weights of hard-shell crabs in the present study can be ascribed to a larger gonad size is unknown because gonads were not weighed separately.

The results of our study are relevant to snow crab aquaculture and stock management. Hardy et al. (2000) have shown the feasibility of the live storage of landed legal-sized hard-shell crabs at low temperatures. Fishery yield could potentially be increased by selecting adolescents, i.e., individuals which may grow to a larger size at terminal moult, from among landed legal-sized hard-shell crabs, hold them through terminal moult and grow them to a marketable condition in a reasonably short period of time. Feeding at 4.5 °C in our study resulted in an overall improvement of nutritional condition over time.

Water content of several tissues decreased and meat yield increased markedly. Wild animals did not perform nearly as well over the same period. Seasonal variations in body constituents of crustaceans are not always clearly defined (Du Preez and McLachlan, 1983). Periodic changes in condition have been associated with moult cycle, but breeding (Barnes et al., 1963; Du Preez and McLachlan, 1983; Fernandes et al., 1994) and food availability (Clarke and Holmes, 1986) may also have an influence. Concurrent observations in aquaculture facilities and rivers have shown that wild crayfish had lower energy reserves than farmed crayfish (Jussila and Mannonen, 1997). In our study, wild snow crabs had much lower energy reserves than crabs fed several weeks in the laboratory. This observation warrants further examination of how the snow crab's nutritional condition varies with time and location with the aim of determining whether condition can potentially affect parameters like recruitment and natural mortality. This could provide valuable information on several aspects of production and contribute to a precautionary management of snow crab stocks.

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