PREDATION RISK AND MOLTING DECISIONS IN THE HAWAIIAN CRAB *LEPTODIUS SANGUINEUS*: THE UPS AND DOWNS OF AN ARMORED EXOSKELETON

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predation Risk and Molting Decisions in the Hawaiian Crab *Leptodius* *sanguineus*: The Ups and Downs of an Armored Exoskeleton.

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Abstract

The exoskeleton of crustaceans serves many functions, including defensive armor against predators. Since the exoskeleton is softer just before, during and just after molt, this period of transition increases prey vulnerability to predators. Given this increased vulnerability, behavioral responses to minimize risk are expected. The cost of molting should also influence life-history decisions, especially when to molt and how much to grow; delaying molt when the risk of predation is high would seem an appropriate defensive adaptation. Morphological defences sensitive to risk levels could be induced during the animal's lifetime as well. Relatively few studies have addressed the trade-offs between defences and growth. In this thesis, I quantify some aspects of the vulnerability of the xanthid crab *Leptodius sanguineus* to a portunid crab predator (*Thalamita crenata*) and investigate how behavioral and morphological defences vary throughout the molt cycle.

Prey size had a large impact on vulnerability of intermolt crabs. Below a predator/prey size ratio of 2.8, intermolt crabs resisted the predator from 1 to 105 min. For freshly molted crabs, this size ratio was irrelevant: most crabs were killed in less than one minute. This increased vulnerability may be reduced somewhat by changes in behavior. For example, to avoid predator detection, freshly molted *Leptodius sanguineus* emphasized crypsis by reducing movement in the presence of a predator: they buried into the sand more slowly and made more pauses compared to intermolt crabs. Large male crabs, when exposed to predation risk, were also more likely to molt under cover under.

With increases in predation risk (detected chemically), crabs delayed their molt. Differences in foraging behavior cannot explain these results. In most cases, size increment was unaffected: crabs did not compensate for a delayed molt by a larger size increment at their next molt. Although crabs did not produce relatively larger claws under predation risk, carapace toughness did appear to increase. The carapace toughens during the 20 days following a molt. The anterior region gets harder than the posterior region, but the rate of hardening of the posterior region may be increased under predation risk. Crabs that delayed molt under the highest level of predation risk produced a tougher carapace in the anterior region than did control crabs.

This study provides direct evidence that molting is plastic. Crabs may thus modify their molt cycle and exoskeletal strength in response to variable predation risk.

To Patrick

À Camille, Yvette
Philippe et Simon

Acknowledgments

I landed in Vancouver on the 8th of January 1990. Within that first week at SFU, I was taught what graduate school is all about: hacker hockey. I wish long life to this practice and thank everyone who participated over the years. Hacker hockey symbolizes the spirit of the Behavioral Ecology Research Group, people with many different interests always ready to welcome newcomers. I am glad to have been part of this group, whose members introduced me to the many cultural and scientific events of the far west: concerts, good parties and dance, pot lucks, eagle counting, interesting research, lab meetings, Les Écologistes and a series of great conferences. A hand of applause for all past and present members.

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Chapter 1

General Introduction

In any environment, the optimal life history strategy will be the one that appropriately trades-off defences, growth and reproduction. Although this statement may be broadly accepted, most life history studies have been devoted to the examination of trade-offs related to reproduction. Relatively few studies have addressed the trade-offs associated with defences and growth. In this thesis, I consider such a trade-off in the xanthid crab, *Leptodius sanguineus.*

After planktonic zoeal life stages and metamorphosis to a benthic form (Wenner 1992, McConaugha 1992), crustacean growth proceeds through a series of cycles where stages of tissue growth, maintenance, repair, and size increment alternate (Table 1.1). Growth is therefore discontinuous, abrupt, and episodic (Hopkins 1993). Reproductive bouts are interspersed between episodes of growth, or occur only after growth is completed and sexual maturity is reached at a terminal molt. The timing, intensity and magnitude of these life history events vary among species and are expected to vary among populations and individuals as well, i.e. life history traits often exhibit phenotypic plasticity. (A review of factors influencing crustacean growth is provided in Chapter 3.)

Throughout the molt cycle, the exoskeleton varies in strength. Morphological defences used during intermolt are progressively less effective from stage D1 to E and regain effectiveness from then until the end of stage C3 (see Table 1.1). This period of transition between armored exoskeletons increases prey vulnerability to predators. These animals might therefore be expected to assess probability of death during the different stages of the molt cycle and adjust their behaviors and investment in defences accordingly. The trade-off between growth and survival becomes of paramount importance in

Table 1.1. Major events of the molt cycle of decapod crustaceans. Days and percentages of the molt cycle are from *Homarus* and *Pachygrapsus crassipes*.

Stage	**Events**	**Authors**
Period: Postmolt		
A1	Lasts 4-12h, 0.3 to 1% of the molt cycle	Warner 1977, Aiken 1980
	Water absorbed for expansion	Lockwood 1967, Warner 1977
	Exoskeleton very soft	Lockwood 1967
	Immobile	Warner 1977
	No feeding	Lockwood 1967
	Tanning continues from premolt	Roer 1980, Mangum 1992
	Active transport of calcium begins	Roer 1980, Mangum 1992
A2	Lasts 1-2 days, 1-2% of the molt cycle	Warner 1977, Aiken 1980
	Mineralization begins, carapace still soft	Lockwood 1967, Mangum 1992
	Slowly regains mobility	Lockwood 1967
	May eat exuvium	Skinner 1985
B1	Lasts 1.5-3 days, 3% of the molt cycle	Warner 1977
	Main period of cuticle calcification	Lockwood 1967
	Exoskeleton still flexible	Lockwood 1967
	No feeding	Lockwood 1967
B2	Lasts 2.5-4 days, 5% of the molt cycle	Warner 1977
	Legs and claws crack if bent	Lockwood 1967, Warner 1977
	Formation of the endocuticle begins	Skinner 1985
	Feeding resumes	Warner 1977
C1	Lasts about 5 days, 6-8% of the molt cycle	Warner 1977, Aiken 1980
	Calcification continues	Lockwood 1967, Skinner 1985
	Main period of tissue growth	Lockwood 1967, Warner 1977
	Main period of formation of the endocuticle	Skinner 1985

Table 1.1. (Cont.)

C2	Lasts 7-10 days, 8-14% of the molt cycle	Warner 1977, Aiken 1980
	Tissue growth continues	Lockwood 1967
	Integument rigid but gives under slight pressure	Lockwood 1967, Warner 1977
C3	Lasts 7.5-11 days, 9-15% of the molt cycle	Warner 1977, Aiken 1980
	May last up to six weeks in *Astacus*	Welinder et al. 1975
	Integument rigid, but calcification incomplete in lateral and frontal parts of the carapace	Lockwood 1967
Period: Intermolt		
C4	Lasts 15-22days, 30-60% of the molt cycle	Warner 1977, Aiken 1980
	Takes 20% in diecdysal species in the tropics in which case proecdysis takes 60%	Lockwood 1967, Spivak 1988
	Calcification complete	Roer & Dillaman 1984
	Membranous layer laid down	O'Brien et al. 1991
	Tissue growth complete	Lockwood 1967
	Period of maintenance	Lockwood 1967, Skinner 1985
	Accumulation of glycogen reserves	Skinner 1985
	Period of tissue differentiation and basal growth of limb buds	Hopkins 1992, Weis et al. 1992
	Maintained through a feedback mechanism control between ecdysteriods and Molt Inhibiting Hormones, cycling every 24h	Hopkins 1992
	Size is fixed, but weight can vary depending on nutritional status; usually weight is at equilibrium	Gurney et al. 1990

Table 1.1. (Cont.)

Period: Premolt

D1	Lasts 3.5-6 days, 8-9% of the molt cycle	Warner 1977, Aiken 1980
	Reduction in ecdysteroids causes decrease in MIH	Hopkins 1992
	Feeding ceases (not for tropical species)	Lockwood 1967
	Apolysis (membranous layer breakdown)	Skinner et al. 1992
D2	Lasts 3.5-6 days, 7-8% of the molt cycle	Warner 1977, Aiken 1980
	MIH production decreases	Hopkins 1992
	Ecdysteroid production increases	Hopkins 1992
	Regeneration of missing limbs	Weis et al. 1992, Skinner 1985
	New epi- and exocuticle secreted	Skinner et al. 1992
	Muscle atrophy in limbs	Skinner 1985
	Calcium is reabsorbed; cuticle cracks in places	Lockwood 1967
	Degradation of inner layers of exoskeleton	Skinner et al. 1992
D3	Lasts 1.5-3.5 days, 2-4% of the molt cycle	Warner 1977, Aiken 1980
	All processes from stage D2 continue	Hopkins 1992, Skinner et al. 1992
D4	12-15h, 1% of the molt cycle	Warner 1977, Aiken 1980
	Remains of the old cuticle are detached	Lockwood 1967, Warner 1977
	Ecdysteroids decrease to allow molting	Hopkins 1992, Chang et al. 1993
	Water uptake begins	Lockwood 1967, Mangum 1992

Period: Molt

E	Animal escape the old exoskeleton	Lockwood 1967
	Takes up water rapidly to unfold the new cuticle	Lockwood 1967, Skinner et al. 1992

influencing crustacean reproduction, and consequently, population dynamics, and deserves more attention.

In this thesis, I use an experimental approach to quantify some aspects of the vulnerability of *L. sanguineus* to a portunid crab predator. I determine whether prey size or molt stage influence vulnerability to predation and whether *L. sanguineus* possesses morphological defences and behavioral adaptations, such as molt delay, that could reduce the risk of predation throughout the molt cycle.

The portunid predator was the blue snapping crab, *Thalamita crenata.* Members of the family Portunidae are called swimming crabs due to the characteristic transformation of the 5th leg into a paddle used for swimming (George and Jones 1984). *T. crenata* are marine or estuarine crabs, found in pools, channels, mangrove creeks and back lagoons, on reef platforms or on the seaward side of coral reefs, where they live in the subtidal muddy sand into which they dig for protection (George and Jones 1984, Schreiber and Cases 1984, Sukardjo and Toro 1988). *Thalamita crenata* reach a maximum size of 60 mm carapace width (George and Jones 1984), and even carapace widths of 80 mm have been recorded (Bob Elner, pers. comm.). They feed on other crustaceans (including *L. sanguineus*; pers. obs.), molluscs, and worms (George and Jones 1984), mostly at night and during high tide; at low tide, they remain buried and inactive (Murugesan and Paulpandian 1987).

Little is known of the natural history of *Leptodius sanguineus*. Most of the information here therefore comes from personal observations. *L. sanguineus* live buried in the sand, under or inside pieces of coral rubble. They reach a maximum carapace width of 35 mm, and prey on bivalves, gastropods and, possibly, hermit crabs. They are cryptic, slow moving crabs, in comparison with the agile swimming crabs, and possess a strong exoskeleton characteristic of the xanthid family. They are found either solitary or aggregated in local microhabitats, at densities of about 2 to 6 individuals per piece of coral rubble. They molt and reproduce throughout their life, breeding from at least May to October (and possibly year round). Females carry eggs from

7 mm carapace width. I found two pairs in amplexus, but could not determine if the female must be freshly molted for fertilization to occur.

Both crab species were collected from Gravel Island, a coral rubble and sand bar located in Kaneohe Bay, Oahu, Hawaii. Gravel Island is partly exposed at low tide and completely submerged at high tide. The principal inhabitants are species of hermit crabs, stomatopods, bivalves, gastropods, sponges, and small grapsid crabs; birds and fishes visit occasionally. Live corals and macroalgae are absent. *Leptodius sanguineus* and *Thalamita crenata* are abundant on Gravel Island, although predator numbers and sizes seem to fluctuate through time.

In Chapter 2, I investigate *T. crenata* handling time (from capture to death) of *L. sanguineus* for different predator:prey size ratios during the prey's intermolt stage, and less than a day after molt, and describe the prey's defensive and protective behaviors. I also describe *L. sanguineus* behaviors aimed at avoiding predator detection, such as reduction in movements (for both molt stages), and molting in protected locations (the latter in Chapter 3).

In the course of an animal's life time, the different trade-offs encountered at successive phases can give rise to selection for very different life histories (Ydenberg 1989). The potential cost of molting itself is expected to influence life history decisions, especially when to molt and how much to grow. Because larvae are transported in the plankton, the location of settlement for juvenile and adult life is somewhat unpredictable. Since conditions most likely vary greatly among habitats, the appropriate life history will depend on the local environment, and might be expected to be environmentally induced. Moreover, because of temporal variation within habitats, the optimal molt timing will vary through time. Selection should therefore favor the ability to assess current predation risk and adjust behaviors throughout the life cycle. Adjustments in the timing of the molt, allowing delay when the risk of predation is high, would seem an appropriate defensive adaptation.

In Chapter 3, I test the hypothesis that crabs respond to variation in predation risk (and cannibalism) by delaying their molt when risk is increased, and compensate for the resulting growth loss by a larger size increment at their next molt. A crab that hides in the presence of a predator will lose some opportunities to feed, but will resume feeding after a certain time has elapsed. However, information on predator visitation rate is expected to influence molting decision. Molt timing and size increment should therefore respond directly to predation risk rather than to reduced energy intake due to suppressed foraging activity. I therefore test the effect of predation risk on foraging activity.

Even when the animal takes the risk to molt, differential investment in other defences can be made. In Chapter 4, I test the hypothesis that high levels of predation risk induce the formation of structural defences. Claws often used in defence could be allocated more energy and made larger, probably at the expense of investment in carapace size (and reproduction). Hardening of the exoskeleton could be hastened under predation risk, or perhaps extended in order to produce a tougher carapace than normal.

In Chapter 5, I summarize the results of this study and discuss them in the context of life history theory and evolution. I also suggest further research which could follow from my work

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Chapter 2

Predation Risk and Periods of Transition : How Does Crab Molt Stage Influence Detection by, Resistance to and Escape from Predators?

Introduction

During an encounter with a predator, the probability of prey survival depends upon the characteristics of both prey and predator as well as the characteristics of the environment in which the interaction takes place. An encounter begins with detection of the prey by a predator and may lead to the prey's death. Selection will act on prey morphology and behavior in such a way as to minimize the probability of mortality. To achieve this goal (given any constraints and trade-offs), the prey may reduce the probability of being detected (Edmunds 1974), attacked or captured by predators (Howland 1974, Weihs and Webb 1984)), or may increase the probability of resistance and escape if captured (Endler 1986). On the other hand, predators have evolved abilities to exploit particular prey, in a manner that increases profitability, by balancing the benefits (usually measured in terms of energy) with the costs in terms of handling time, lost opportunities for other activities, risk of injury and risk of predation (Schoener 1971, Werner and Hall 1974, Krebs et al. 1977, Krebs 1978, Arnold 1984, Krebs and McCleery 1984, Lawton 1989, Lima and Dill 1990).

Prey can interrupt the predation sequence and reduce the probability of being consumed in many ways. To avoid detection, prey may hide (physically, behind structures), or achieve crypsis through some combination of color, shape, odor and reduction or absence of movement (Soane and Clarke 1973, Edmunds 1974, Stein and Magnuson 1976, Sih 1982). If detected, a prey may avoid being attacked by signalling to the predator that it has been seen (Harvey and Greenwood 1978), displaying weapons and threatening to cause harm, walking away (Lawton 1989), or feigning death (Edmunds 1974). Upon attack, prey may prevent capture by startling the predator (Bedford and Chinnick

1966, Endler 1986), deflecting the attack (Edmunds 1974), fleeing and finding a refuge (Webb 1986), or fighting back (Edmunds 1974). If captured, prey may rely on structural defences such as shells, spines and chemicals, or again, fight to escape the predator (Stein 1977, Endler 1986, Hudon 1987, Lawton 1989). At any point in the sequence, the predator may choose to give-up or be forced to stop.

Not all encounters ending in consumption result in prey death. In cases of partial predation, analogous to grazing of plants by herbivores, prey can lose (mutilation) or give away (autotomy) and regenerate body parts without losing their lives. Examples range across taxa: molluscs' foot tips and siphons are nipped away (De Vlas 1979,1985, Peterson and Quammen 1982, Zwarts 1986), tube worms lose parts of their fans (De Vlas 1979), crustaceans lose limbs and claws (Lawton 1989, Smith 1990), while amphibians (Wilbur 1984) and reptiles (Dial and Fitzpatrick 1983) lose parts or all of their tail. Both mutilation and autotomy may help redirect the predator onto another target, facilitating escape. Autotomy is thought to be used as an escape tactic in crustaceans (McVean 1982). For example, decapod crustacean prey were seen to successfully avoid death from the crab *Cancer pagurus* by autotomizing one or two chelipeds (Lawton 1989).

Autotomy may be thought of as a more specialized defence against predators than crypsis, because of the stage in the predation sequence at which it occurs. As Endler (1991) points out, as the probability of death increases, prey defences change from more generalized anti-predator tactics against all potential predators to highly specialized tactics to meet the immediate threat from one particular predator. In other words, the effectiveness of different anti-predator defences is determined by the stage of the predation sequence, where prey reaction must match predator action. However, the defences available to an animal, and their effectiveness during the predation sequence, also vary with the state of the prey throughout it's lifetime and may be greatly affected by periods of transition between life stages. In such cases, generalized defences (like crypsis or hiding) may prevail over specialized ones (like display or use of weapons) when the latter defences are impaired, non-existent or under development.

Animals at different life stages, such as egg, larva, juvenile and adult, are subject to a different array of species and sizes of predators and will have different defences, therefore experiencing different degrees of vulnerability. For healthy animals with continuous growth, vulnerability to predation usually decreases continually with size increase, as defences improve (Paine 1976, Arnold 1984, Palmer 1990). This is not the case for animals with discontinuous growth. Most arthropods experience abrupt size transitions at molt, during which defences are impaired. During periods of intermolt, the exoskeleton is hard and constitutes an effective armor. After molt, the newly formed exoskeleton is soft and offers little protection (Lockwood 1967, Skinner 1985). This period of transition may have profound implications for survival. Several decapod crustaceans and aquatic insects have been documented to be more vulnerable to predators during the molting period (Hines et al. 1987, Soluk 1990). For example, after molt, large *Ephemerella* (mayfly) larvae lose their size advantage and become more vulnerable to stonefly predators (Soluk 1990). Total handling time of freshly molted individuals is shorter, which could explain why soft individuals are selected over hard individuals (Soluk 1990). Crayfish also become preferred prey for fish after molt because of a decrease in handling time: mortality is higher for post-molt *Orconectes propinquus* crayfish compared to intermolt crayfish (Stein 1977).

The present study aimed to document patterns of predator-prey interactions and prey vulnerability in a system where both predator and prey were crab species. The goals were: (1) to provide data on the poorly known (Lawton 1989) predatory interactions between crustaceans; (2) to study how prey avoid detection and capture by the predator and how these behaviors are affected by the molt stage; and (3) to quantify the difference in vulnerability between post-molt and intermolt prey, for different size ratios of predator and prey.

The predator *Thalamita crenata* (family Portunidae) is a fast and agile swimming crab. The prey *Leptodius sanguineus* (family Xanthidae) is a relatively slow moving crab with a thick exoskeleton. Predators can reach 60 mm carapace width (George and Jones 1984) or more, whereas prey do not exceed 35 mm carapace width (see Chapter 3). Predator and prey share the same natural habitat: on Gravel Island, located in Kaneohe Bay, Oahu, Hawaii, they live buried in the sand and under or inside pieces of coral rubble.

Several predictions can be made about prey responses. Because of their cryptic exoskeleton, and because they move more slowly than the predator, xanthid prey should reduce movement in the presence of predators to enhance crypsis and avoid being detected. The greater the probability of detection by the predator, the more the crabs should reduce movement. On the other hand, the carapace of freshly molted prey is paler than intermolt prey which, at least to the human eye, makes them less cryptic. Assuming that movement is not impaired after molt, soft prey should try to hide more quickly than intermolt prey, since they may not be able to rely on crypsis. Indeed, post-molt prey should be more vulnerable to predators than intermolt prey due to the loss of the protective armor and because they may not be able to escape from the predator's grasp once captured. If prey use their claws to fight back, intermolt crabs should be able to escape the predator more easily, since their claws are stronger. Any size of freshly molted prey should be vulnerable to all sizes of predators, but predator-prey size ratio ought to be an important determinant of the outcome of an encounter between a predator and an intermolt prey. I tested this hypothesis by measuring handling times (from prey capture to death) of inter-molt and post-molt *L. sanguineus* by two sizes of *T. crenata*.

Methods

Experiment 1- Avoiding predator detection and capture

All experiments were conducted at the Hawaii Institute of Marine Biology, on Coconut Island, Kaneohe Bay, Oahu, Hawaii. Predators and prey were collected on Gravel Island, Kaneohe Bay.

Three behaviors were recorded, in the presence and absence of *T. crenata,* as indicators of the predator avoidance strategy of *Leptodius sanguineus*: (1) time the prey took to bury in the sand; (2) number of pauses prey made while burying; and (3) degree of burial when the crab stopped burying, or after 5 minutes of observation: totally buried, totally buried but with the antennae showing above the sand surface, a third of the body showing, half of the body showing, and not buried.

All trials were conducted in two 93 l aquaria. In the predation risk treatment, an individual *Thalamita crenata* predator was hidden in an opaque perforated Tupperware container (14 x 14 x 4 cm). The container was placed vertically, resting against the far narrow end wall of the aquarium and held down to the bottom with a rock. The predator was not visible to the prey and the container did not allow much movement. Sounds and vibrations therefore were likely minimal and any cue to the predator's presence probably olfactory. In the control treatment, an identical empty Tupperware container was located in the same fashion. Three predators of the largest carapace width (numbers 1-3; Table 2.1) were used on alternate days. Claw length was the maximum propodus length and claw height was the greatest propodus height. All measures were obtained with a digital caliper.

The bottom of each aquarium was covered with 6 cm of beige sand from Kaneohe Bay. Aquaria were surrounded on three sides with plywood sheets. The front narrow side facing the observer and the top were open. The water in the aquaria was not aerated during trials and was replaced

Table 2.1. Measures of carapace and claws (mm) of predatory crabs (*Thalamita crenata*) used in these experiments. S = predator classified in the small category, crusher claw < 11 mm in height; L = predator classified in the large category, crusher claw > 11 mm in height. * = crusher claw.

Predator # and category	Carapace Width	Left Claw		Right Claw	
		Length	Height	Length	Height
1 S	51.4	38.0	13.1	29.0	9.4*
2 L	44.3	38.6	14.3*	35.9	12.4
3 L	44.7	32.8	12.0	33.9	13.0*
4 S	36.7	24.5	8.8	24.8	10.0*
5 S	32.2	22.2	7.8	22.2	8.4*

after each trial. The water temperature was 28°C. Light was provided by ceiling-mounted fluorescent white tubes, set on a 15h day-9h night cycle.

During a trial a prey crab was introduced into one of the aquaria for observation, and then transferred to the other. About half of the crabs (n=46) were introduced to the predator treatment first; the others (n=54) were introduced to the control treatment first. Crabs were transferred by hand and released on the water surface near the front wall. Once the crab had reached the bottom, the observation period started. Observations of the 3 behaviors described above lasted for 5 minutes for each treatment. Trials were conducted both in the morning or the afternoon.

Prey introduced varied in carapace width from 10.3 to 18.6 mm and were all non-ovigerous females. Because of the risk of cannibalism on freshly molted individuals, crabs were held in individual jars. Prey of two molt stages were compared : crabs in intermolt and crabs who had freshly molted (less than a day old). Trials were constrained by the availability of freshly molted crabs. Crabs who had molted the previous night (usually 0 to 3) and a few intermolt crabs were tested the same day. Experiments were conducted during September and October 1991.

The analysis of 2x2 cross-over trials (Jones and Kenward 1989) was used to determine whether crabs behaved differently in the presence and absence of the predator, taking into account the potential influence of prey handling (period effect) and order of presentation to predator and control treatments (carry-over effect).

Experiment 2- Prey vulnerability once captured

Prey were introduced into aquaria containing an unrestrained predator. As in Experiment 1, prey were deposited on the water surface and after sinking through the water column, reached the sand. In most cases, predators attacked the prey immediately after it reached the sand surface, but in other cases the attack occurred later. One to three prey were presented to each predator every day, depending on their availability. Two indicators of prey vulnerability were measured once a prey was captured: (1) handling time of the prey; (2) number of escape events from the predator's grasp made while wrestling, fighting back, or autotomizing a claw. Handling time was measured from capture by the predator to prey death. Death was assumed when the predator started feeding on the internal body mass. See Appendix 1 for a detailed description of predator-prey interactions.

Prey vulnerability was assessed when the prey were hard, and one day after molt, when the prey were soft. Sizes of prey ranged from 11.0 to 17.2 mm (carapace width), resulting in a predator/prey size ratio varying between 2.2 and 4.2 (see Table 2.1 for sizes of predators). Carapace width and claw size were also used to establish predator/prey size ratios and to divide predators into two size categories to simplify analyses (see Appendix 2 for details). The same xanthid crabs used in Experiment 1 were used as prey here. If the prey was not immediately captured by the predator, behaviors were observed for 5 min, as described in Experiment 1, following which encounters were forced by pushing the prey towards the predator, using a glass rod.

Predators were kept individually in 93 l glass aquaria with aerated water. The bottom of each aquarium was covered with 6 cm of sand. The aquaria were separated with plywood sheets. A fluorescent light was installed behind the aquaria providing equal light to each one. The set-up was enclosed by a black plastic blind. Observations were made through 4 by 3 cm windows made in the blind in front of each aquarium. Predators were not fed outside trials. A total of 5 predators was used.

Results

1- Avoiding predator detection and capture

In the presence of a predator, crabs buried more slowly (Fig. 2.1), paused more often (Fig. 2.2) and did not bury as deeply in the sand (Table 2.2) compared to when the predator was absent. Crabs introduced first to the predation treatment buried on average 12 sec slower in the presence of the predator; but crabs introduced first to the control treatment buried on average 86 sec slower when the predator was present (Fig. 2.3). The analysis of 2X2 cross-over trials revealed no carry-over effect, but did reveal a period effect, since both control and treatment crabs' burial times were longer in the second period. Overall, the treatment effect was significant, which indicates that crabs buried more slowly in the presence of a predator.

Crabs introduced first to the predation treatment made on average 0.3 more pauses in the presence of the predator; crabs introduced first to the control treatment made on average 1.2 more pauses in the presence of the predator (Fig. 2.4). The analysis again revealed no carry-over effect, but did reveal a period effect, since the number of pauses for both control and treatment crabs was higher in the second period. The treatment effect was highly significant: crabs made more pauses in the presence of the predator.

After 5 min of observation, prey were rarely seen on the sand surface. Only in three instances (in the control treatment) did prey not bury, but walked around the aquarium instead. The combined data revealed that three quarters of the crabs buried totally (or almost) in the sand when the predator was absent, whereas only half of the crabs buried totally when the predator was present (Table 2.2). The analysis of 2X2 cross-over trials examines for each crab the changes in degree of burial attained in the presence and absence of the predator for the two trials (Table 2.3). To perform the analysis, the degrees of burial were scored 4, 3, 2, and 1, corresponding to totally buried, antennae showing, a third of the body showing, and half of the body showing,

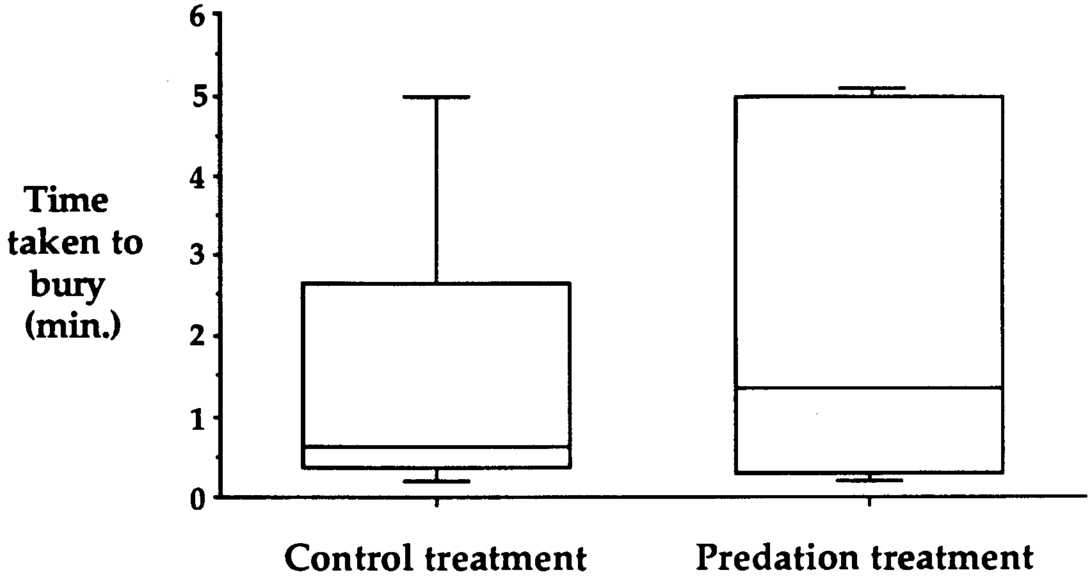

Figure 2.1. Whisker box plots (10th, 25th, 50th, 75th, and 90th percentile) of the time crabs took to bury in the sand, in the absence and presence of a predator. Molt stages are combined. Statistical analysis of these data reported in Table 2.5.

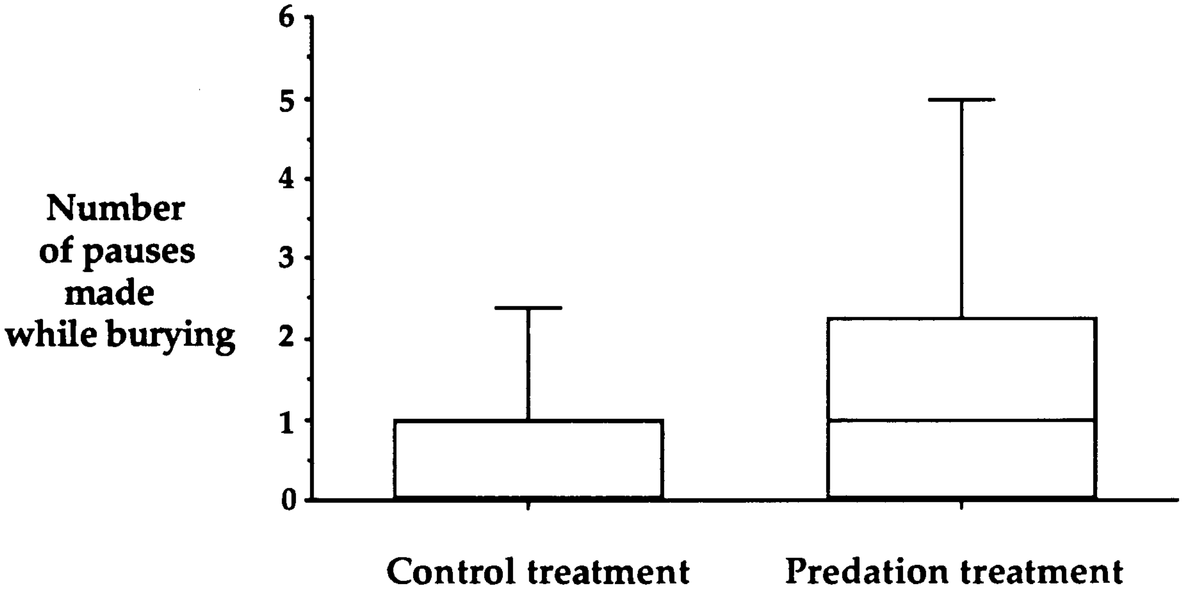

Figure 2.2. Whisker box plots of the number of pauses crabs made while they buried in the sand, in the absence and presence of a predator. Molt stages combined. Statistical analysis of these data reported in Table 2.5.

Table 2.2. Frequencies of the degree of burial in the sand crabs achieved after spending 5 minutes in the control treatment aquarium or the predation treatment aquarium (olfactory stimuli). G-test comparing all categories, p=0.009. G-test comparing body buried and body showing, p=0.002.

Degree of burial	Category	Control treatment	Predation treatment
Total	4	43	27
Antennae showing	3	31	26
Body buried		74	53
1/3 Body showing	2	17	35
1/2 Body showing	1	8	13
Body showing		25	48

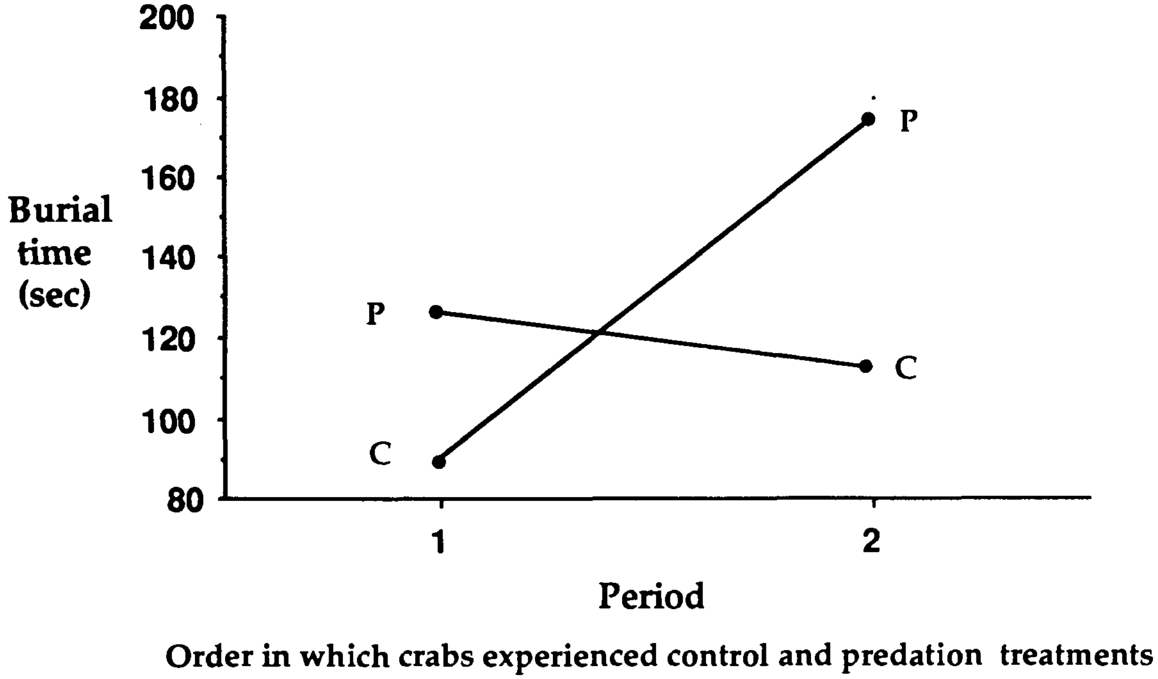

Figure 2.3. Group by period plot (analysis of 2X2 cross-over trials) showing the mean times crabs took to bury in the sand, in the absence and presence of predator, depending on the order in which they experienced the control and predation treatments. Wilcoxon test, carry over effect : p=0.4; period effect p=0.004; treatment effect : p= 0.02. P= Predation treatment; C= Control treatment. Lines connect trials on the same individuals.

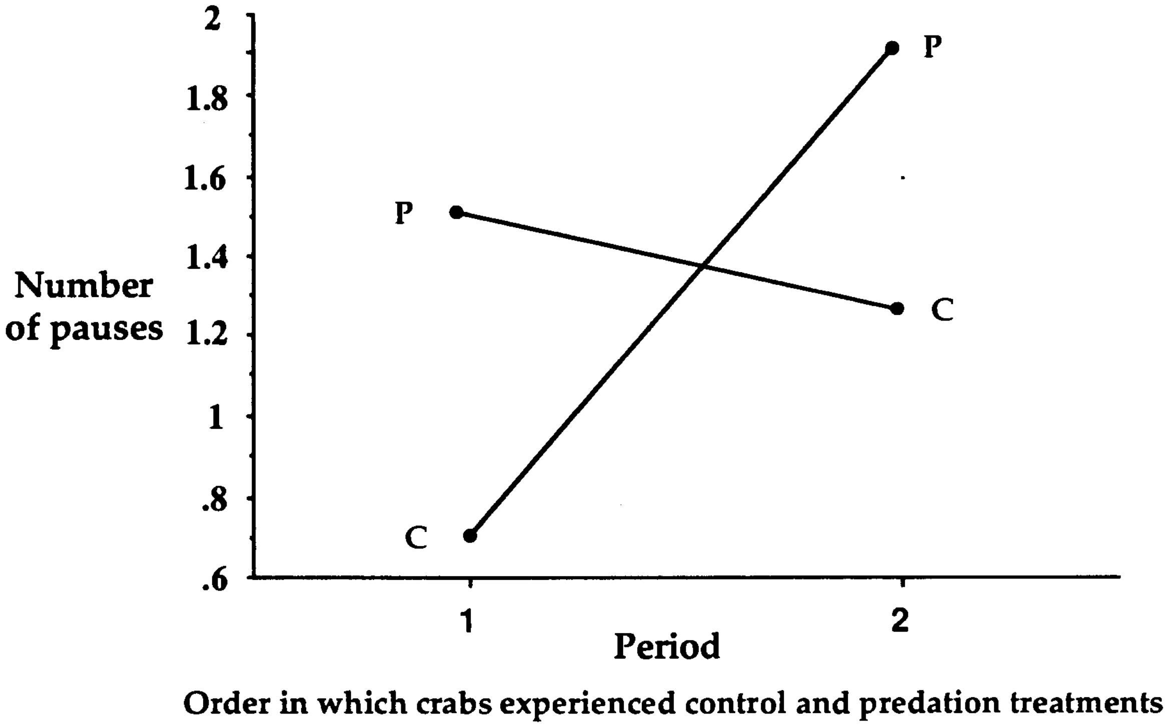

Figure 2.4. Group by period plot (analysis of 2X2 cross-over trials) showing the mean numbers of pauses crabs made while burying, in the absence and presence of a predator, depending on the order in which they experienced control and predation treatments. Wilcoxon test, carry over effect : p=0.8; period effect : p=0.01; treatment effect : p= 0.001. P= Predation treatment; C= Control treatment. Lines connect trials on the same individuals.

Table 2.3. Degree of burial crabs achieved when first experiencing the control treatment and then being transferred to the predation treatment and vice-versa. Numbers of crabs in each transition between two degrees of burial.

Degree of burial	Control to Predator	Predator to Control
Same burial		
T-T	7	11
A-A	7	8
2/3-2/3	5	4
1/2-1/2	2	4
Total	21	27
Deeper burial		
1/2-T	0	4
2/3-T	0	1
A-T	2	0
1/2-2/3	2	0
2/3-A	2	3
Total	6	8
Shallower burial		
T-A	6	7
A-2/3	4	3
2/3-1/2	3	0
T-2/3	13	0
Total	26	10

respectively. Crabs made thirteen different transitions from a degree of burial to another (or to the same degree). Moving from control to predation treatment, 21 crabs buried at the same level, 6 buried more deeply, and 26 buried less deeply when the predator was present. Moving from predation to control treatment, 27 crabs buried at the same level, 8 buried more deeply, and 10 buried less deeply when the predator was present. Overall, 48 (27+21) crabs buried in the same way, while 16 (6+10) crabs buried more deeply, and 34 (26+8) crabs buried less deeply in the predation treatment compared to the control treatment. The carry over effect was not significant (p=0.5), but both the period effect (p=0.03) and treatment effect (p=0.0003) were significant.

In instances where crabs were introduced into the predator's aquarium in Experiment 2, and not immediately captured, prey behaviors were also observed for 5 min. Since Experiment 2 followed Experiment 1, and the same individuals were used, it was possible to compare the degree of burial attained by crabs in a sequence of increasing predation risk treatments. Treatments were in the following order : (1) control (no predator); (2) predator present but restrained (olfactory cue only); (3) predator present and not restrained (visual and olfactory cues). Most prey buried totally when the predator was absent, buried two-thirds of their body when the predator cue was olfactory only, and did not bury and remained motionless when the predator was present and active before attack (Table 2.4). Since the crabs were killed after capture, it was not possible to conduct a cross-over trial. Therefore, carry over and period effects may have had an influence, but given the earlier findings the treatment effect most likely accounts for these results.

Freshly molted crabs buried more slowly than intermolt crabs (Table 2.5A), and made more pauses while burying (Table 2.5B) when the predator was present but not when it was absent. In the absence of the predator, freshly molted crabs buried more deeply than intermolt crabs, but in the presence of the predator, crabs of both molt stages buried to the same degree and less deeply than when the predator was absent (Table 2.5C). Degrees of burial were

Table 2.4. Contingency table of the degree of burial in the sand of crabs in three predation risk situations. Crabs were first introduced to the control treatment, transferred to the predation treatment, where the predator was restrained and not visible, and then transferred to the predation treatment where there was a free swimming hungry predator. All observations lasted five min. G-Test, p=0.0001, n=9.

Degree of burial	**Control treatment**	**Predation treatment: olfaction only**	**Predation treatment: vision and olfaction**
Total	4	0	0
Antennae showing	0	2	0
1/3 Body showing	2	6	1
1/2 Body showing	3	1	0
Zero	0	0	8

Table 2.5. Comparisons of burial behaviors for intermolt (n=80) and freshly molted (n=21) crabs, when a predator was present or absent. A. Mean burial time (s). B. Mean number of pauses. C. Mean depth of burial.

A	Predator present	Predator absent	2x2 cross-over trials
Intermolt	143.2	105.4	p=0.2
Freshly molted	179.3	96.1	p=0.03
Mann-Whitney U	p=0.04	p=0.8	

B	Predator present	Predator absent	2x2 cross-over trials
Intermolt	1.6	1.1	p=0.03
Freshly molted	2.2	0.8	p=0.04
Mann-Whitney U	p=0.02	p=0.2	

C	Predator present	Predator absent	2x2 cross-over trials
Intermolt	2.7	3.1	p=0.04
Freshly molted	2.7	3.4	p=0.04
Mann-Whitney U	p=0.4	p=0.04	

scored as 1, 2, 3 and 4 if crabs were half buried, two-thirds buried, buried but with the antennae above the sand, or totally buried, respectively. The influence of predator presence on burial time (Fig. 2.5), number of pauses (Fig. 2.6) and degree of burial (Fig. 2.7) did not vary with prey size. For this reason, all prey sizes were pooled in the analyses reported above.

2- Prey vulnerability once captured

Soft crabs that had recently molted were killed more rapidly by predators than were hard intermolt crabs (Fig. 2.8). However, larger predators could kill crabs rapidly regardless of the latter's molt stage (Fig. 2.8). Since hard crabs were killed by large predators as rapidly as soft crabs were killed by small predators, trials on soft crabs-large predators were not conducted, to avoid unnecessary sacrifice. Intermolt prey were killed faster as the predator/prey size ratio (carapace width) increased (Fig. 2.9A). For a ratio of 2.8 and greater, half the crabs were killed in less than 1 min; the rest were killed between 1 and 4 min. Below a ratio of 2.8, crabs were killed between 1 and 105 min. Crabs who had freshly molted were easily killed by the predators, regardless of predator or prey size (Fig. 2.9B). The same relationship was obtained using the predator crusher claw size to establish the predator/prey size ratio (Fig. A2.1, Appendix 2).

Of 30 crabs captured, 10 fell out of the predator's grasp; 6 escaped once, 3 escaped twice, and 1 escaped three times. 40% (8/20) of the intermolt crabs worked free of the predator's grasp, while only 20% (2/10) of the freshly molted crabs did so. But because they were all recovered by the predator, none of the crabs definitively escaped. The median size of prey that escaped momentarily was only 2.6 times smaller than their predator (Fig. 2.10). Escape never occurred when the predator was more than 3.2 times larger than the prey (Fig. 2.10). The two freshly molted crabs who managed to escape the predator temporarily were both relatively large; most of the soft-shelled crabs did not escape despite the fact that their predator was relatively small (Fig. 2.11).

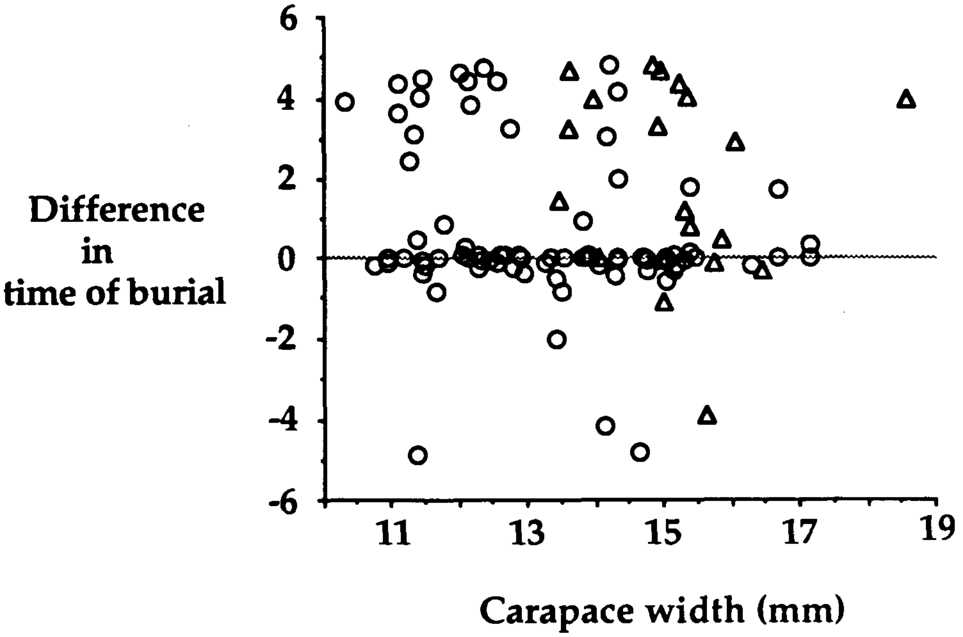

Figure 2.5. The influence of crab size on the difference between time (s) taken to bury by crabs in the presence and absence of predators. Positive values indicate that crabs buried more slowly when the predator was present. Negative values indicate that crabs buried more rapidly when the predator was present.

Legend: ○ Intermolt crabs (n=80),
△ Freshly molted crabs (n=21).

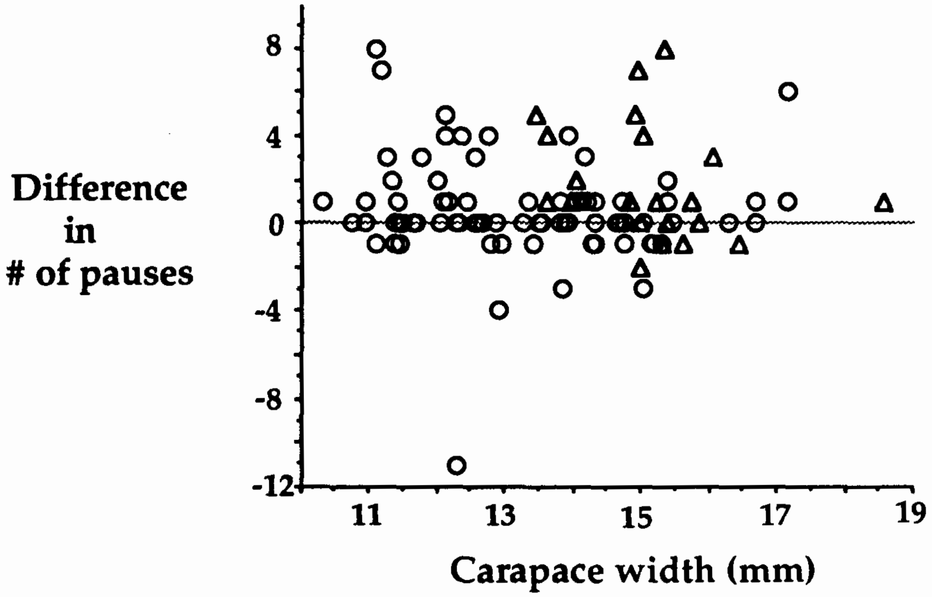

Figure 2.6. The influence of crab size on the difference between the number of pauses made in the presence and absence of predators. Positive values indicate that crabs made more pauses when the predator was present. Negative values indicate that crabs made fewer pauses when the predator was present.

Legend: ○ Intermolt crabs (n=80),
△ Freshly molted crabs (n=21).

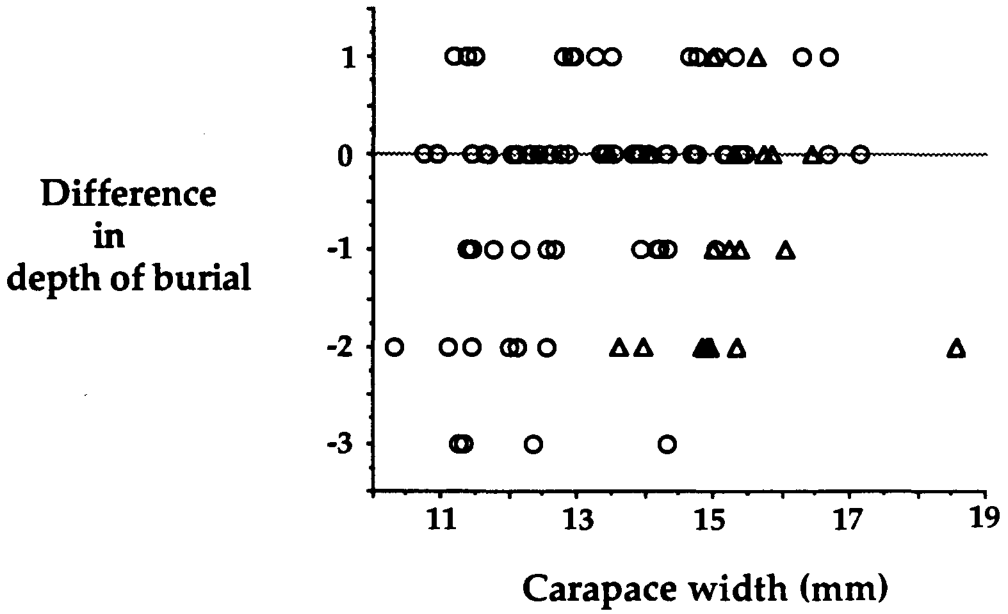

Figure 2.7. The influence of crab size on the difference between the levels at which crabs buried in the presence and absence of predators. The degrees of burial were scored from 1 to 4 and correspond to the categories in Table 2.2. Positive values indicate that the crabs buried less deeply into the sand when the predator was present. Negative values indicate that crabs buried more deeply when the predator was absent.

Legend: ○ Intermolt crabs (n=80),
△ Freshly molted crabs (n=21).

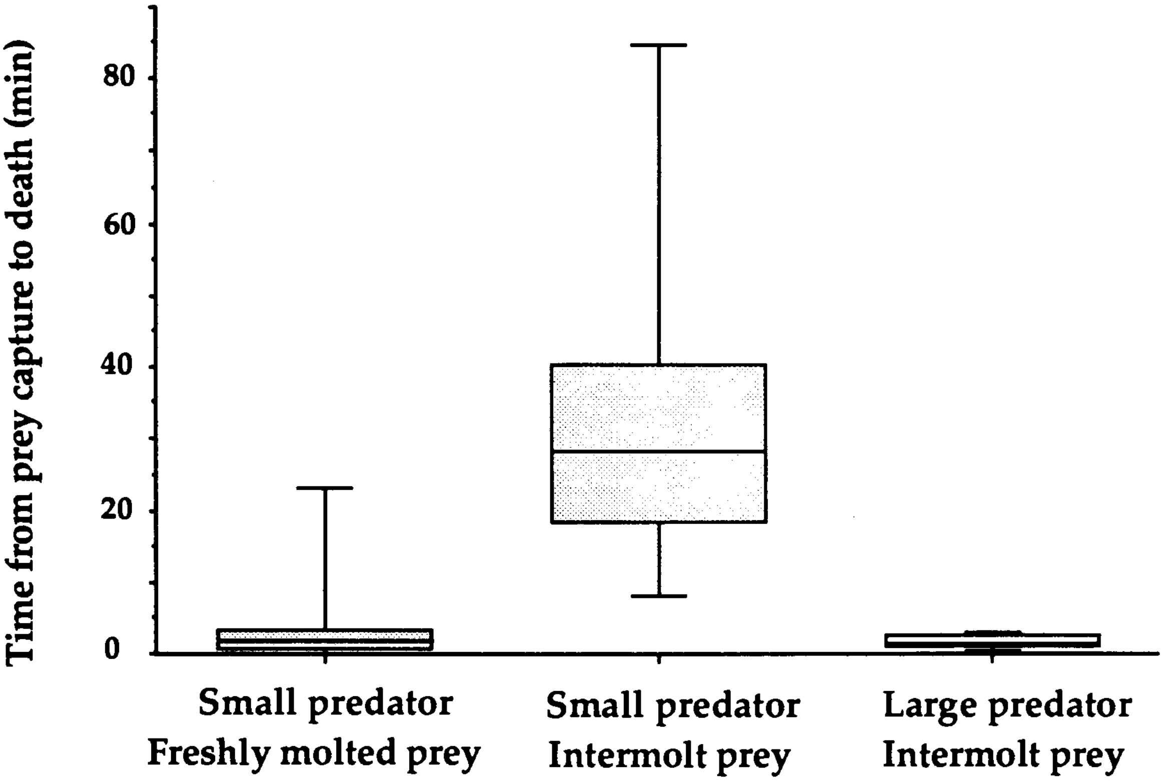

Figure 2.8. Whisker box plots of the handling time to prey's death in the grasp of small and large predators. Predators were given prey that had freshly molted and prey that were in the intermolt period. All prey sizes grouped. Kruskal-Wallis p=0.0005.

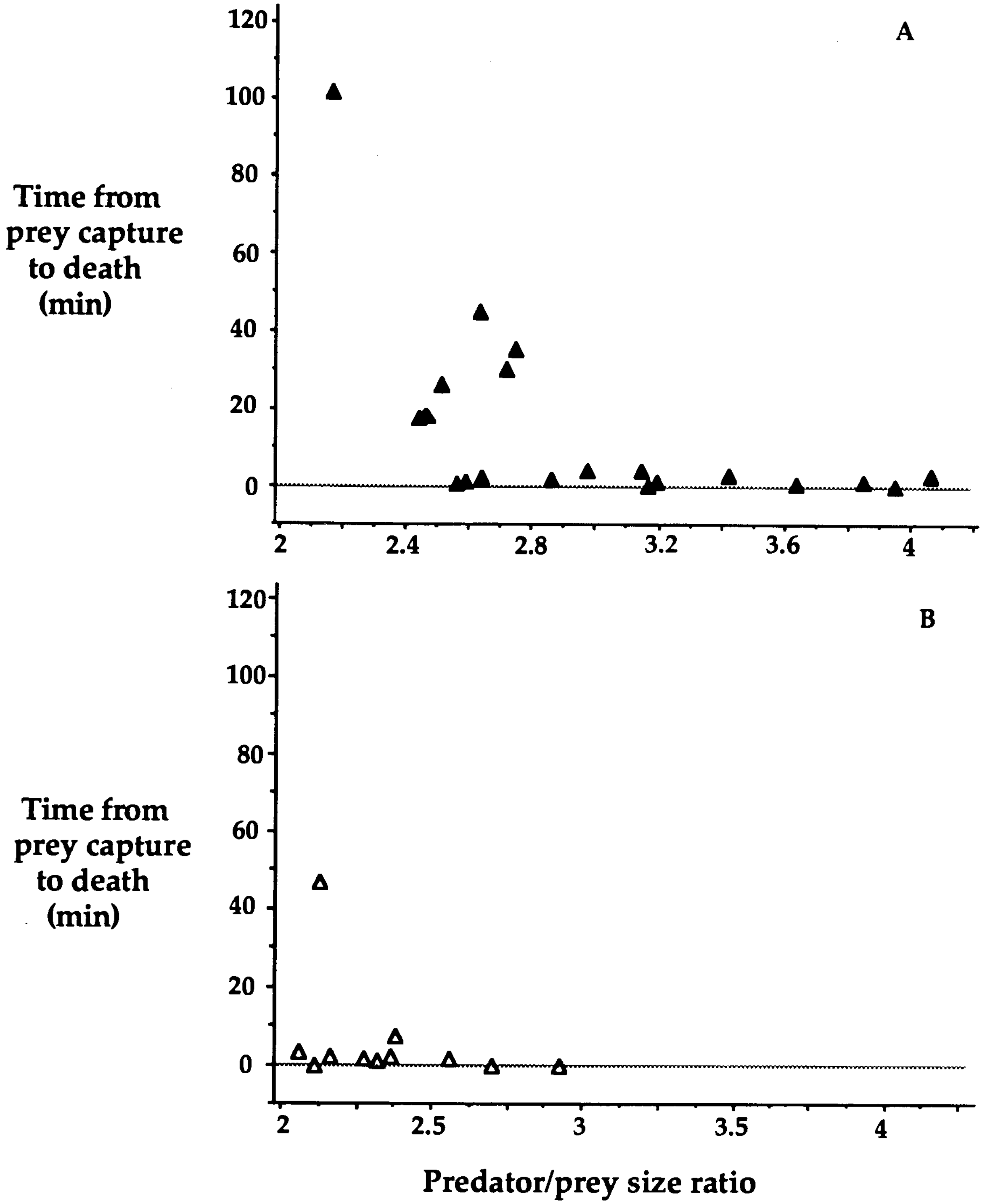

Figure 2.9. Influence of predator/prey size ratio on predator handling time. A. Prey at the intermolt stage. B. Prey who had freshly molted.

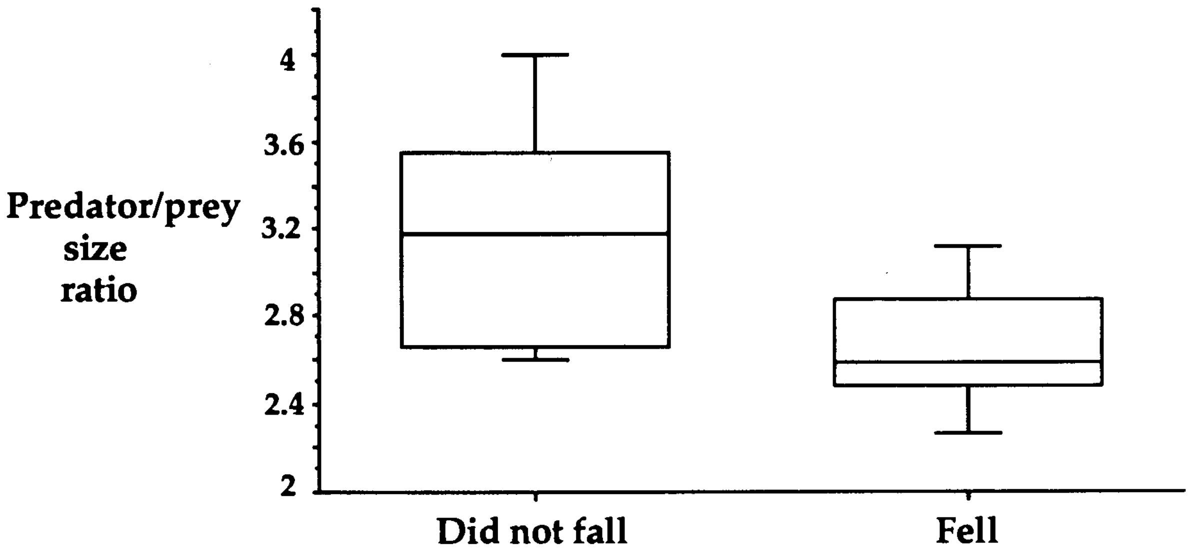

Figure 2.10. Relative sizes of intermolt prey who fell (n=8) or did not fall (n=12) out of the predator's grasp. Mann-Whitney U test, p=0.02.

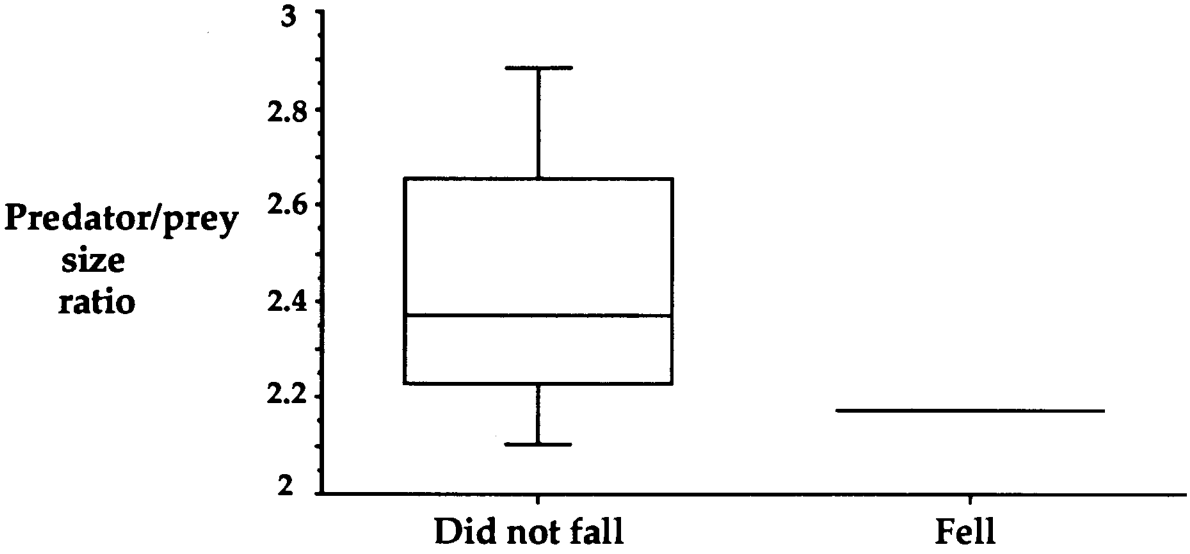

Figure 2.11. Relative sizes of freshly molted prey who fell (n=2) or did not fall (n=8) out of the predator's grasp. Mann-Whitney U test, p=0.3.

None of the 30 crabs autotomized their claws or legs to escape the predator. Crabs used their claws to protect their ventral surface (Fig. A1.1, Appendix 1). In two instances, they fought back, holding the predator by the claws, and keeping the predator from moving for 27 and 65 seconds (Fig. A1.1, Appendix 1).

Discussion

1- Avoiding predator detection and capture

Prey defences reflect the outcome of interactions between predator and prey over evolutionary time. Because risk may vary during an animal's lifetime, it is also advantageous for animals to assess the risk of predation and behave in a way that minimizes the risk of being consumed by predators (Lima and Dill 1990), taking into account their degree of vulnerability (Maynard Smith 1979).

Level of predator threat

Shortly after their introduction to the aquarium, prey started burying in the sand. In the presence of predators, prey buried more slowly and burial was interrupted with more pauses than when a predator was absent. Consequently, they buried less deeply into the sand in the same time period. Reduction of the prey's movements, combined with the observation that predators seem unable to locate a prey precisely unless the prey moves (Fig. A1.2, Appendix 1), support the hypothesis that crypsis in *Leptodius sanguineus* is important to avoid detection and attack by predators.

The fact that crabs buried at all indicates that simply being motionless is not enough. A trade-off may exist between being cryptic and being truly hidden, where being motionless in a predator's presence is the best of a bad situation. Immediate protection is enhanced by reducing movements, although hiding would provide better long-term protection. Getting to this more desirable position is risky (digging may give away the prey's position), and the risk may vary with the likelihood of attack by a predator. The speed of burial, the number of pauses and the depth of burial may therefore vary with the magnitude of predation risk. Observations of crab behavior in three situations of increasing predation risk suggest such a tendency. As risk intensified, crabs reduced their movements. In the extreme case, when the predator was actively searching, crabs remained motionless and did not bury (Table 2.4). The increased number of pauses made while burying may not only provide increased crypsis, but may also facilitate detection of the predator's whereabouts through visual, vibrational or chemical cues. Recent research (see Rittschof 1992 for a review) suggests that crabs can detect chemical signals and their duration. The antennules are analogous to the noses of vertebrates, flicking to sample odors which indicate the presence of food items. Olfactory recognition has also been shown in social contexts (commensalism and mating). Since the crabs' antennules increased their beating (flicking) frequency in the presence of predators (Fig. A1.1, Appendix 1), the prey may have been assessing the predator's movements and position through chemical cues. This observation supports the hypothesis that predator odors can be used to monitor changes in risk level.

When the risk of predation was higher crabs responded by reducing movements, but behaviors varied. Differences in crab burial behaviors may be due to a difficulty in assessing the level of predation risk in alien, rapidly changing artificial environments. Uncertainty about the risk may have made some crabs opt for a solution that could potentially bring the most benefits: hiding.

In natural situations, crabs are found not only buried in the sand but also hidden under cover or inside structures. During the experiment, crabs did not have the option of hiding under or inside coral rubble. This may have been important since prey initiate fleeing sooner when they are further away from a safe location (Dill and Houtman 1989, Lima and Dill 1990). Experiments in a more complex environment would help determine whether *Leptodius sanguineus* use different predator avoidance tactics in different habitats; fleeing may be preferred over crypsis (immobility) if cover is near, despite the fact that *L. sanguineus* are relatively slow moving.

Degree of vulnerability

Crayfish (*Orconectes propinquus*) reduce activities such as walking, climbing and feeding, and increase burrowing in the presence of a smallmouth bass predator, and small vulnerable crayfish reduce activities more than larger, less vulnerable individuals (Stein and Magnuson 1976). The present study with *Leptodius sanguineus* indicates that movement was reduced in relation to the magnitude of the threat to the point where even burrowing activity virtually ceased (Table 2.4). Reduction of movement in the xanthid species studied here did not correlate with vulnerability during intermolt: prey size did not influence the changes in the time taken to bury, the number of pauses made while burying or the depth of burial. On the other hand, the greater vulnerability during the molt stage (compared to the intermolt) influenced burial behaviors (Table 2.5): in the presence of the predator freshly molted crabs buried more slowly and made more pauses than intermolt crabs; in the absence of the predator, freshly molted crabs buried more deeply in the sand compared to intermolt crabs.

A soft carapace and reduction of muscular power may possibly impair capacity to hide in abrasive substrates. Soft-shelled lobsters (*Homarus americanus*) swam more slowly and accelerated less quickly compared to intermolt lobsters (Cromarty et al. 1991). Also, a high level of muscular activity could distort the carapace (Lockwood 1967). However, this may be problematic only for a few hours following the molt. In this experiment, the burying ability of crabs who had molted the previous day did not seem to be affected. Short burial times of around 10 s. were frequent and times as fast as 2 and 5 s. were recorded. Therefore, the state of the muscles and the carapace did not appear to constrain the burying behaviors observed.

A different strategy for avoiding predators was expected after molt since crab vulnerability is higher when soft (Exper. 2) and because crab color is less cryptic just after molt than during intermolt (personal observation). Therefore, not being buried would be more costly, and faster burial might be expected. Overall, the results show the opposite trend: while both types of crabs behaved similarly in the absence of predators, freshly molted crabs buried more slowly, and made more pauses than intermolt crabs in the presence of a predator; also the difference in the degree of burial crabs attained in the presence and absence of the predator was greater for freshly molted crabs compared to intermolt crabs. This supports the idea that defences deployed during a predation sequence vary with the state of the prey. In a comparable situation, several species of female scincid lizards trade-off fleeing and crypsis in the presence of predators. Normally, lizards outrun their predator, but when carrying eggs, females are more vulnerable to predators because they can't run as fast (Shine 1980). Bauwens and Thoen (1981) showed that gravid females do not suffer higher mortality because they reduce movement which is effective at avoiding predator detection. During periods of higher vulnerability, lizards and crabs seem to rely on more generalized modes of defence over specialized ones. Moreover, if movement is the most important give-away and the predator cannot detect differences in substrate color match, enhancing crypsis by reducing movements when a predator is present would be the optimal behavior when the animal is most vulnerable. Since crab predators use both mechanoreception and chemical cues to detect prey (Lawton 1989), freshly molted prey may be less detectable than intermolt prey: not only do they move less, but they may smell differently. What if freshly molted crabs are chemically "hidden" from crab predators, and therefore not easily recognized as edible prey? If visual and chemical recognition of prey is impossible or impaired, detection would only be possible through movement. This may have selected for reduced prey movement after molt compared to the intermolt period. The scenario may be different for visual predators such as fish, which may detect color differences, for example. Clearly, more research is needed to determine the cues available to predators from freshly molted arthropods.

2- Prey vulnerability once captured

Morphological, as well as behavioral, defences against predators are common (Harvell 1990). For example, the carapace and claws are used as armour and weapons (Stein 1977, Hudon 1987, Bildstein et al. 1989, Pillai 1990). However, their efficacy in defence will vary with the size and physiological state of both predator and prey. Capture may not end in prey consumption if the prey carapace and claws are hard or large, if prey can struggle or fight back, and if the predator abandons the predation sequence.

The role of the carapace

The vulnerability of a crab to predators varies with stage of the molt cycle: vulnerability is greatest at molt, the carapace being inefficient as an armour. Compared to intermolt crabs, post-molt crabs are killed faster regardless of their size and stand less chance of escaping the predator once captured. There are two reasons for a greater resistance to predators during intermolt : (1) the carapace is tough and serves as an armor; and (2) muscles are fully developed and functional (Skinner 1985). Any prey can potentially escape if the predator is disturbed, but a soft carapace combined with a weakened musculature would not permit the resistance and the strength needed to escape the predator's grasp. Figure 2.9 illustrates that during intermolt predator/prey size ratio was important in determining the outcome of an encounter, but relative body size was irrelevant when the prey had recently molted. At molt, all crabs were highly vulnerable to predation. If the predator was large relative to the prey (body size ratio over 2.8), the prey was at greater risk of being eaten regardless of the molt stage. On the other hand, when the predator was relatively small, crabs during intermolt were less vulnerable to predators, because they resisted for longer periods of time. This time gained may permit successful defence or the advent of some disturbance which may facilitate escape. Therefore, in a natural environment, relatively large crabs may escape death even if they are captured. As Palmer (1990) showed, predator attack may occur with equal frequency, but predation success will decrease with an increase in prey size. This size refuge is not absolute, since relatively small gastropod predators can feed on large barnacle prey (Palmer 1990). This is also true in this crab predator-prey system, especially since the size refuge is discontinuous due to the alternating molting and hardening periods.

The risk of molting may be inferred from the differences in vulnerability between intermolt and post-molt crabs, in relation to the relative sizes of predator and prey (Fig. 2.9). Vulnerability shortly after molt is always high. During intermolt, when the prey is relatively large compared to the predator, vulnerability is low (Fig. 2.12). Therefore, the difference in vulnerability between intermolt and post-molt stages is greater for a prey likely to encounter relatively small predators. The difference in vulnerability between stages decreases when predator/prey size ratio increases, because prey vulnerability becomes high in both cases. It follows that the increase in risk during molt will be higher for prey faced with a lower predator/prey size ratio and will decrease with increasing size ratio (Fig. 2.12).

While results show that *Thalamita crenata* handling time of intermolt xanthid crabs increased as the predator/prey size ratio decreased, it is still unclear why. There are three possibilities: (1) a xanthid's carapace may become tougher as it increases in size; (2) larger xanthids may be stronger and struggle more with the predator; and/or (3) relatively smaller predators are unable to crush the xanthid's carapace easily. Although these three factors are not mutually exclusive and could all play a role, results from Chapter 4 do not support hypothesis 1. It would be interesting to further test hypotheses 2 and 3. Hypothesis 2 has support from the fact that only relatively large crabs managed to escape the predator's grasp (Figs. 2.10 and 2.11). Hypothesis 3 is likely to be true also, since *T. danae* and *T. crenata* feeding on mussels may be limited in the size of prey they can break due to morphological and mechanical characteristics of the claws, with a crusher claw becoming more powerful with an increase in body size, allowing breakage of larger prey (Seed 1986).

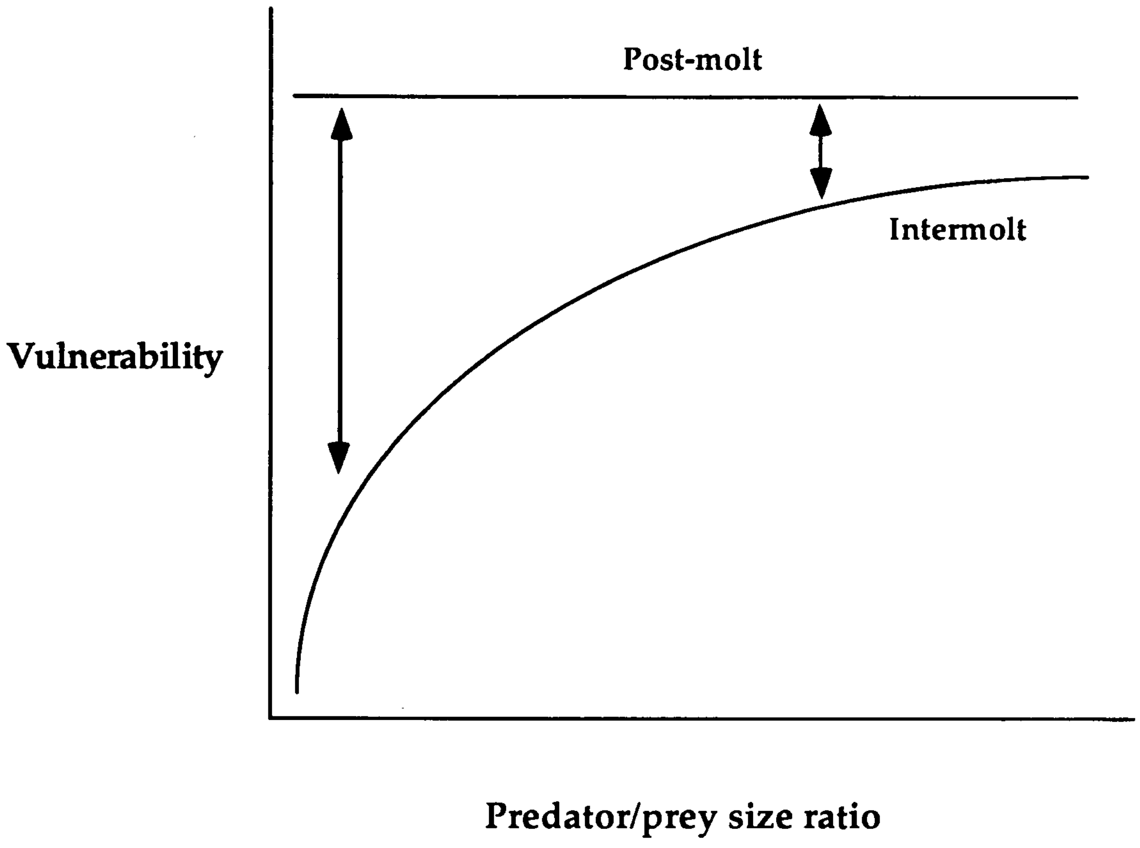

Figure 2.12. Variation in crab vulnerability during the post-molt and the intermolt stages, in relation to the size ratio between predator and prey. Arrows indicate the risk of molting.

The role of the claws

While the carapace provides protection, claws may be used in defence. Crayfish use a defensive claw display in the presence of a fish predator (Stein and Magnuson 1976). Fish will try to avoid a crayfish's claws during an attack and will also preferentially select individuals with smaller claws for the same body size (Stein 1977). *Leptodius sanguineus* also display to attacking fish (personal observation), but they never displayed to *Thalamita crenata,* nor did they autotomize claws in order to attempt to escape the portunid predator. *L. sanguineus* (or at least the females) may not use autotomy as a defence mechanism, or may use it in different circumstances. Autotomy can be seen as a mechanism of deflection of an attack, whereby a crab sacrifices a claw to fall out of the predator's grasp or to keep the predator busy with food while escaping. Even though damage and bacterial infection are minimal (Hopkins 1993), autotomy is costly in terms of growth, since the limbs lost must be regrown (McVean 1982, Skinner 1985, Smith 1990). Giving-up a claw may also be costly because the capacity to use it for defence in the future is lost.

Predator size and hunger level, along with the availability of a shelter nearby and familiarity with the surroundings, may be important considerations for evaluating escape probability and deciding whether to autotomize or not. If the predator is relatively small or hungry, it may be better to keep the claws to protect the body or even attack the predator, especially if a good place to hide is not available. Crabs were seen holding their claws tightly over their mouthparts and part of their abdomen while the predator was trying to remove the claws (Fig. A1.1, Appendix 1). Prey were also seen holding the predator's claws, impairing the latter's movements. This suggests that the prey's claws may play an important role in active defence and might be worth keeping. This would, however, only be true for intermolt crabs. After molt, claws were seen to be useless in defence since the predator could rapidly remove them (Fig. A1.3, Appendix 1).

Molt stage may also play an important role in the decision to autotomize. In the days following a molt, claw musculature must be restored before the claws are fully functional (Skinner 1985). Such an empty claw does not have the potential to keep the predator occupied for very long. An autotomized body part needs to increase the predator's handling time to facilitate the prey's successful escape (Dial and Fitzpatrick 1983). Observations of predators handling freshly molted prey showed that soft claws require little or no handling since they are mostly ignored (Section A1.3, Appendix 1). This supports the hypothesis that the autotomy of a soft claw would not increase the chances of escape. Moreover, if the animal survives, giving-up a claw at this early stage of the cycle must have a large cost. Autotomy may be profitable in the short term, but is followed by an increase in vulnerability which may result in death later on (McVean 1982). Autotomy should be most likely in the middle of the intermolt, when body growth has been completed and internal regeneration of the limb bud is possible before the next molt (Stages C and D; Bauchau 1966).

Since *Leptodius sanguineus* did not autotomize once during the experiments, regardless of the molt stage and relative size, it is also possible that claw autotomy may not be useful when facing such a rapid crab predator. It may be used against fish or birds, or against slower crab predators (*L. sanguineus* used autotomy against me). For example, larger *Porcellana platycheles* crabs often autotomize a claw to escape the slower crab *Cancer pagurus* (Lawton 1989). *Thalamita crenata* handled xanthid prey starting with the removal of the claws (Fig. A1.3, Appendix 1). Since the claws are less profitable than body mass (Lawton 1989), this behavior may be an adaptation to prevent the prey's escape by autotomy (or retaliation). While facing an agile "talented" crab predator, the strength of the carapace is likely to be the most important defence a xanthid crab possesses.

The impact of variation in predator and prey characteristics

For the same size ratio between predator and prey, there was variation in the time to the prey's death (Fig. 2.9). Factors other than predator and prey size may be responsible for the variation observed. For example, a more precise prediction of vulnerability would be obtained by knowing the exact period in the molt cycle of both predator and prey. Predators may not be a threat at all around their own ecdysis while prey may be vulnerable shortly before ecdysis as well as after. Hardening time lasts up to 20 days after molt (see Chapter 4). How vulnerability to predators varies during this period still needs to be determined. As well, individuals may be able to adjust their investment in carapace thickness. Calcium deposition continues throughout the entire molt cycle (Spivak 1988). Crabs could thicken or reduce their carapace according to perceived variation in predation risk (see Chapter 4).

Predator hunger level may also influence the length and outcome of an encounter. In the experiment, predators were only fed *Leptodius sanguineus* during trials, and their hunger level was likely high. This may have increased their willingness to spend time trying to break into a tough prey and prevent the prey's escape. In the natural environment, predator hunger level and prey choice are certainly different than in the laboratory. Predators are naturally subject to foraging trade-offs (Krebs and McCleery 1984). Soft-shelled crabs may be preferred prey due to their increased profitability. Recently molted prey contain twice as much digestible organic material as intermolt prey, at least in crayfish (Stein 1977). Handling time is reduced because a soft carapace is easier to break and the digestion of a hard carapace is avoided. *Cancer pagurus* crabs were observed regurgitating fine pieces of macerated exoskeleton up to 30 h after consumption (Lawton 1989). Because of their strong exoskeleton, intermolt *Leptodius sanguineus* may be such low profitability prey that predators may avoid them when given the choice.

Body size is commonly used to assess predator threat to prey. However, for crabs, claws are important in subduing prey and claw size is not always correlated with body size. Many studies have shown the importance of claw characteristics in determining the feeding habits of predatory crabs (reviewed by Seed 1986). Broken and worn down claws are less efficient at cracking open clams (Juanes and Hartwick 1990). Therefore, morphological and mechanical characteristics as well as condition of the claws are important to evaluate feeding performance. I observed in this crab predator-prey system that mouthparts are also used to break into the prey's carapace (Fig. A1.3, Appendix 1). Size of the mouthparts is probably closely related to body size. Therefore, a combination of both claw size and body size should be used to evaluate a crab predator's threat more accurately. Possibly, such factors may be important considerations for other arthropod predators as well. This suggests the need for a new approach to predator threat evaluation which would incorporate those variations in predator morphology and behavior (both between and within individuals) which potentially may have large effects on the outcome of predator-prey interactions. However, even if predator claw size determines the probability of escape after capture, this may be impossible for the prey to predict before being handled since the intensity of the olfactory stimulus emitted from a large crab predator will always be greater than from a small one, regardless of claw size.

The role of the environment

Environmental complexity plays an important role in predator-prey interactions. In the laboratory, only a few occurrences of escape were observed; none were definitive. This may be due to the fact that the aquarium was a very simple habitat and favored the predators. Different refuges and habitat topography offer different levels of protection and are important in determining the outcome of a prey's encounter with predators (Coen et al. 1981, Lipcius and Hines 1986, Wilson et al. 1990). Because crab predators are more efficient in sand than in harder substrates (Arnold 1984), sand alone is a poor refuge for xanthid crabs who have been detected by a crab predator (Section A1.2, Appendix 1). The presence of shelter provided by coral rubble could have helped crabs who had escaped from the predator's grasp to hide successfully.

The presence and absence of other species changes the dynamics of predator-prey interactions (Wilbur 1984). The absence of competitors, predators and other distractions allowed the predator to handle the prey with little disturbance. Such extended periods of prey handling may not be possible in nature. *Thalamita crenata* took up to 105 minutes to kill intermediate sized *Leptodius sanguineus* prey. Continual handling after failing to subdue the prey after 15-20 minutes may be unrealistic and artifactual. This may assure survival for most intermolt *Leptodius sanguineus* prey who encounter a *Thalamita crenata* predator smaller than 2.8 times their own size.

Concluding Remarks

Successful defences used during intermolt are lost at molt. Arthropods which molt throughout their life will only reach a temporary size refuges. These will be broken periodically and render the animal vulnerable to all sizes of predators. Post-molt individuals emphasize defences early in an encounter with a predator, such as crypsis, in order to compensate for increased vulnerability at the later stages of the predation sequence, where defences are impaired. But is this enough? Other studies have shown that freshly molted prey are not only preferred but they suffer greater mortality as well. If molting is so costly, why is it preserved as a growth process by natural selection throughout the life of some arthropods but not others? Examining the costs and benefits of molting will provide insight into this question. Molting puts the animals at risk, but this risk may be mediated by the timing of the molt. The question therefore becomes: When to molt?

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Chapter 3

Life History Plasticity in Crabs: Are Molt Timing and Size Increment Influenced by Predation Risk?

Introduction

Molt timing and size increment at each molt are important crustacean life history traits, since they determine the rate and pattern of growth. However, there is tremendous variation in these traits not only between species, but also between populations (Hartnoll 1982). There also exists the potential for phenotypic plasticity, i.e., a single genotype producing a range of phenotypes in different environments (Caswell 1983, Schlichting 1989, Harvell 1990, Lessells 1991, Via 1993). Phenotypic plasticity is a general term to describe all types of environmentally-induced phenotypic variation (Stearns 1989), but some authors have expressed concerns about whether all plasticity is adaptive (Caswell 1983, Schlichting 1986, Lessells 1991, Thompson 1991). Indeed, a few cases of plasticity have proven to be maladaptive; in other cases the adaptive value has proven to be hard to determine when the norm of reaction is still evolving (Via and Lande 1985, Stearns 1989). However, adaptive plasticities have been identified in response to the risk of predation (Stearns 1989).

To be adaptive, phenotypic plasticity must allow genotypes to produce a phenotype which is optimal in each different environment (Via 1993). This so-called reaction norm corresponds to a heritable set of instructions which, for example, govern behavior (a strategy). The genetic basis for plasticity is still controversial (Scheiner 1993, Schlichting and Pigliucci 1993, Via 1993). Via (1993) argues that phenotypic plasticity comes about as a by-product of selection toward different phenotypic optima in various environments, whereas Scheiner (1993) proposes that plasticity is itself a trait that can be selected. As Harvell (1990) points out, even without fully understanding the genetic basis of plasticity, it is possible to study whether phenotypes respond to selection. Measuring how different phenotypes respond to selection is beyond the scope of this thesis, but I attempt here to take the first step in identifying adaptive plasticity in crustacean growth: that is, whether a change in phenotype occurs in response to a specific environmental signal (Stearns 1989). I test the hypothesis that crustacean growth is plastic in response to predation risk.

Interactions with conspecifics and predators are often suggested as agents of selection favoring the correlation of molt timing with seasons (Lockwood 1967, Hartnoll 1982), the time of day or night (Lipcius and Herrnkind 1982, Paula 1989), full or new moon (Havens and McConaugha 1990), tidal cycle (Paula 1989) and habitat (DeGoursey and Stewart 1985, Hines et al. 1987, Dew 1990). However, direct evidence for such selection has seldom been obtained. Therefore, my goal in this Introduction is to explore variation in crustacean growth, to find evidence of adaptive plasticity, and to suggest the design attributes of a good molt strategy.

Variation in larval growth

Larval release, at the zoeal stage, often coincides with periods of high tides, likely facilitating dispersal, survival and growth (Sastry 1983, McConaugha 1992). Larval growth is thought to be determinate, because for most species the number of morphologically distinct larval instars is fixed (Hartnoll 1982). However, for some species, due to temperature changes, the number and length of instars may vary among populations (Barnes 1980). Shortage of food can reduce the size increment of some instars, but an increase in the number of instars can compensate for this, such that the size at metamorphosis need not be affected (Hartnoll 1982). On the other hand, compensation may not always be possible and consequently size at metamorphosis may vary. The timing of metamorphosis to benthic life may be influenced by factors such as presence of adult conspecifics and suitable sediment (Jensen 1989, O'Connor 1991), providing evidence of adaptive plasticity.

Variation in juvenile and adult growth

Post-larval growth is a series of stages, with periods of intermolt during which size is constant (although body mass may vary), and periods of molt, when size increases. In general, growth may be determinate (Hartnoll 1982, Havens and McConaugha 1990), with a fixed number of molts and with or without a terminal molt, marking the onset of sexual maturity (Skinner 1985); or indeterminate (Hartnoll 1982, Havens and McConaugha 1990), where molt and reproduction happen more or less regularly and repeatedly until death (Hopkins 1992).

In general, molting is more frequent for juveniles (Klein Breteler 1975), and the intermolt period increases with increasing size (Hartnoll 1982, Siegel 1984), but for some species the length of successive molt cycles can also vary independently of body size (Davis et al. 1973). Size increment commonly decreases with increasing size (Donaldson et al. 1981, Hartnoll 1982, Spivak 1988, Skinner 1985, Robichaud et al. 1989) but it can also remain more or less constant (Hartnoll 1982). Pooled data, mostly for larvae, but also for juveniles and adults, have indicated a mean size increment of around 22.3%, but the variation is extreme: from decrements to 83% increments (see Hartnoll 1982 for a review). Between and within species, considerable variation also exists in maximum size (Hines 1989, see Conan et al. 1989 for review).

Slower growth can be a consequence of an increase in time spent in intermolt, or a decrease in size increment at molt, or both. These can be caused by a reduction in food quantity or quality (Wenner et al. 1974, Havens and McConaugha 1990), and by lower temperature (Steel 1980, Hartnoll 1982, Webster 1982, Anderson et al. 1985, Dew 1990) or high water hardness in freshwater species (Brown et al. 1991). Growth is also reduced when energy must be invested in regenerating appendages (Hartnoll 1982, Skinner 1985, Smith 1990), producing and caring for eggs (Wenner et al. 1974, Steel 1980, Bertness 1981, Hartnoll 1982, Webster 1982, Colby and Fonseca 1984, Anderson et al. 1985, Havens and McConaugha 1990) and guarding females (Robinson and Doyle 1985). Male and female *Gammarus pulex* can delay their molt for about three days in order to find a suitable mate, since the male must be hard-shelled and the female soft-shelled for mating to occur (Ward 1984b). Heavy metals (Weis et al. 1992) and parasites (Hartnoll 1982, Skinner et al. 1992) also delay molting. Some parasites can be partially or totally shed at molt (Ward 1984b, DeGoursey and Stewart 1985, Cash and Bauer 1993), and so parasitic loads could also be a factor triggering molt in some species.

Growth may be accelerated by an increase in temperature, which decreases molt interval and, for a few species, slightly increases size increment (Hartnoll 1982). On the other hand, crustaceans with a fixed number of instars that experience high temperature may molt faster, increase less in size at each molt, and reach a smaller size at the terminal molt (Hartnoll 1982). Temperature increase is often associated with longer days and both promote molting, but continuous light delays molting (Lockwood 1967). The loss of appendages has also been shown to hasten molting in several species, depending on how many are lost and when in the molt cycle this occurs (Hartnoll 1982, Skinner et al. 1992). The presence of abundant food may also trigger molting, and an increase in temperature may cue the start of a more productive season. For example, spring phytoplankton blooms cause synchronous molting in *Emerita analoga* (Siegel 1984).

Synchronous molting is also believed to be a mechanism for avoiding cannibalism in organisms living and foraging in groups (Morris 1985). Subordinate lobsters (*Homarus americanus*) will delay molt in the presence of a dominant individual (Cobb et al. 1982). Because the subordinate animals feed less, the delay may be controlled by energetic limitations. Female (and to a lesser extent, male) *Sicyonia ingentis* molt synchronously after the spawning season is over (Anderson et al. 1985). Synchronous molting may also be an adaptation to predator avoidance, not only through a dilution effect (the whole population is simultaneously vulnerable), but also as an escape tactic. When frightened, antarctic krill (*Euphausia superba*) shed their exoskeleton synchronously and swim away (Hamner et al. 1983). The authors hypothesized that this is a tactic to avoid pursuit from predators since it leaves the impression of a school, although the animals have vanished. This is the only case documented of an effect of predation risk on molting, and suggests that molting can be plastic.

Objectives and rationale

The objectives of this research were to further investigate variations in the timing and size increment of molting in crabs in response to stimuli from conspecifics and predators: do crabs exhibit adaptive phenotypic plasticity in growth?

The exoskeleton of most crustaceans serves as a defensive armor, but limits growth (Lockwood 1967). Shedding the exoskeleton to increase in size may be costly, since vulnerability increases (Chapter 2) and predation on the stages around molt is greater (Stein 1977, Robichaud et al. 1991). The benefits of molting are both indirect and direct. Molting yields indirect benefits since increase in size leads to increased reproductive output (Havens and McConaugha 1990, Hines 1992, Tomikawa and Watanabe 1992), may favor predator avoidance and escape (Sastry 1983, Hudon 1987) and may increase success in competition for refuges (Smith 1990). Molting may be directly beneficial since it permits repair or replacement of lost limbs or an injured carapace (Hartnoll 1982), removes parasites and epibiota (Ward 1984b, personal observation), gets rid of metabolites (Hartnoll 1982), and permits mating ((Birkhead and Clarkson 1980, Hartnoll 1982, Hines et al. 1987, Havens and McConaugha 1990). When the benefits outweigh the costs, animals obviously should molt. On the other hand, if the cost of molting is high, it may be advantageous to avoid molting. The cost of molting may increase with an increase in predation risk (or cannibalism) and may be size-dependent. Therefore, my predictions are: (1) that molting should be delayed when predation risk is high or increased, and (2) that larger animals (who have less to gain from molting and experience a relatively larger increase in vulnerability compared to smaller individuals; Chapter 2) should delay under relatively lower risks of predation, and delay longer.

A trade-off may exist between the number of molts and the size increment realized at molt (within the limits of what is mechanically and physiologically possible). Therefore, I predict (3) that to compensate for the growth opportunity lost while delaying, the size increment at the next molt should be larger than normal.

Energy intake is an important factor influencing the rate of growth. The risk of predation can limit an animal's foraging activity (Lima and Dill 1990), reducing energy intake. Though a reduced energy intake may result in molt delay, I predict (4) that plasticity in molt timing and size increment has evolved in response to the risk itself, not as a result of decreased foraging.

Finally, if delaying is not possible or is too costly, finding a relatively safe place to molt may be an alternative option to minimize the risk of predation: thus I predict (5) that molting under cover should be favored when the risk of being eaten by predators in the open is high.

Methods

The study system

The xanthid crab *Leptodius sanguineus* is a good subject for this study because it possesses a strong exoskeleton, which contrasts sharply with the soft freshly molted state, and because it co-occurs with a portunid crab predator, *Thalamita crenata*. Both species live among pieces of coral rubble and sand. *Leptodius sanguineus* were collected from their natural habitat on a coral rubble bar: Gravel Island in Kaneohe Bay, on the north shore of Oahu, Hawaii. Crabs were brought to the lab, at the Hawaii Institute of Marine Biology, located on Coconut Island in Kaneohe Bay. Predators were collected on Gravel Island and also around Coconut Island. Preliminary experiments were conducted in 1990, to see if *Leptodius sanguineus* delay molting when conspecifics or predators are present. For the main experiments in 1991, the goal was to learn more about the mechanism affecting molt timing and size increment under predation risk.

Preliminary experiments

Preliminary experiments were conducted in August and September 1990, in an indoor laboratory seawater facility. The daylight cycle was fixed at 15:9 (light:dark hours). A total of 40 xanthid crabs were kept individually in 19 l buckets. The following three treatments were randomly distributed in the spatial array: (1) Predator treatment = risk of predation: 14 crabs (6 males and 8 females, mean carapace width of 12.7 mm) received a predator in their bucket for an hour every day (due to a shortage of *T. crenata*, a snapper *Lutjanus fulvus* was used as predator every third day); (2) Conspecific treatment = risk of cannibalism: 13 crabs (8 males and 5 females, mean size of 13 mm) received a larger hard-shelled conspecific (20-25 mm) in their bucket for an hour every day; (3) Control treatment: 13 crabs (10 males and 3 females, mean size of 12.4 mm) did not receive visits, but water was disturbed.

The buckets were aligned in two rows on each of four water tables. Water flowed separately and continuously into each bucket and overflowed through a series of holes made 4 cm from the top. Water turnover time was 3-4 min. Each crab was provided with a piece of coral rubble as shelter. Predators and xanthids for the conspecific treatment were held individually in separate buckets. All xanthids were fed trout pellets every evening. All predators were fed fish every day. To reduce the likelyhood of losing crabs to predation, predators were fed to satiation before they were introduced to the buckets. The buckets were cleaned every day, to remove uneaten food and debris, by siphoning the bottom using a transparent plastic hose.

The date of every molt occurrence was noted. The exuviae were collected to measure the length and width of the carapace with digital calipers.

Main experiments

Experiments were conducted over four months, July through October 1991, in an outdoor seawater installation subject to natural light. Xanthid crabs were held individually (to prevent exchange of cues) in 9.5 l buckets. Crabs from 11 different treatment-groups (Table 3.1) were randomly distributed in a spatial array of 224 buckets. A total of 1770 crabs were collected in order to obtain crabs of the appropriate size ranges, and in good condition. About 2/3 of the females used in the experiments were carrying eggs when collected, but none were ovigerous when the experiments started, all the eggs having hatched. The treatments were divided into five groups (Table 3.1). (1) Controls: these crabs did not receive any cues from predators. The 32 small females received an empty basket an hour every day for the last two months of the experiment, and water disturbance was simulated for four months. (2) Predation risk 1: for the first two months of the experiment, crabs received water flowing from a predator holding tank (odor cue) for an hour every day. There was a ratio of one predator in the tank to seven prey receiving the water flow. For the final two months, the water was allowed to flow from the

Table 3.1. Description of the five treatments used in the 1991 experiments on molt timing and size increment. Legend: L=large; S=small; F=female; M=male; size (mm) =mean carapace width of *Leptodius sanguineus*.

Treatment	**Description**	n	**Size cat**	**Sex**	**Size (mm)**	**St. dev.**
Control	Crabs received no predator visits but appropriate water disturbance	16	L	M	18.7	0.97
		16	L	F	17.9	0.87
		16	S	M	11.7	1.88
		32	S	F	11.7	0.55
Predation risk 1	Crabs received predator odor for 1h/day for 2 months; then increased to 24h/day for another 2 months; cue strength also increased	16	L	M	19.1	0.74
		16	L	F	18.0	0.75
		16	S	M	11.9	2.21
		16	S	F	11.7	0.42
Predation risk 2	Crabs received 1h/day of a low concentration of predator odor for 2 months then a higher concentration for 2 more months	32	S	F	11.7	0.63
Constant predation risk	Crabs received a predator in their bucket for 1h/day for 4 months	16	S	F	11.6	0.49
Decreased predation risk (handled crabs)	Crabs weighed every day for 2 months then weighed for only 15 days following their next molt. Half of these crabs experienced control conditions; the other half, predation risk 1.	32	S	F	11.7	0.68

predator tanks to the prey's buckets for 24 h a day. Predators were also added to the holding tanks in order to increase the ratio to one predator to two prey. Therefore, the predation risk was assumed to be increased not only in time, but also in strength of the odor cue. (3) Predation risk 2: for the first two months, these crabs received the same predation risk as did the crabs from the predation risk 1 treatment. For the final two months, a predator was enclosed in a plastic screen basket and placed into the prey's bucket, for an hour every day. The predator-prey ratio was then one-to-one. The predation risk was the same in terms of time, but the strength of the odor cue was presumably greater. Tactile stimulation was also possible (through the mesh), but unlikely. (4) Constant predation risk: crabs received the visit of a predator in their bucket an hour every day for a period of four months. (5) Decrease in predation risk: these crabs were weighed every day for two months, at which point weighing was stopped until all the crabs molted. From then on, crabs were only weighed for the 15 days following their molt (the weighing experiment is described more fully in Chapter 4). Even though this "predation risk" is artificial, these data may be viewed in terms of a decrease in predation risk: the predation risk (human imposed) was high for the two first months, stopped for a while and then was reduced, since crabs were only handled for roughly half of their molt cycle.

A circular wooden platform mounted on six concrete blocks supported a group of 14 buckets in each of 16 large circular tanks (Figure 3.1). The tanks measured 1 m in diameter and were 1 m deep. A hole 6 cm in diameter at the bottom of each tank permitted the evacuation of overflowing water from the buckets. A predator holding tank was located at each of the four corners of the set-up. Fresh seawater was pumped from Kaneohe Bay and through a network of PVC pipes provided continuous water flow to each bucket. The water overflowed from holes perforated 4 cm from the bucket's rim, and turnover time was about 10 min.

Another network of PVC pipes was superimposed on the first one and used to distribute water from the predator holding tanks into the appropriate

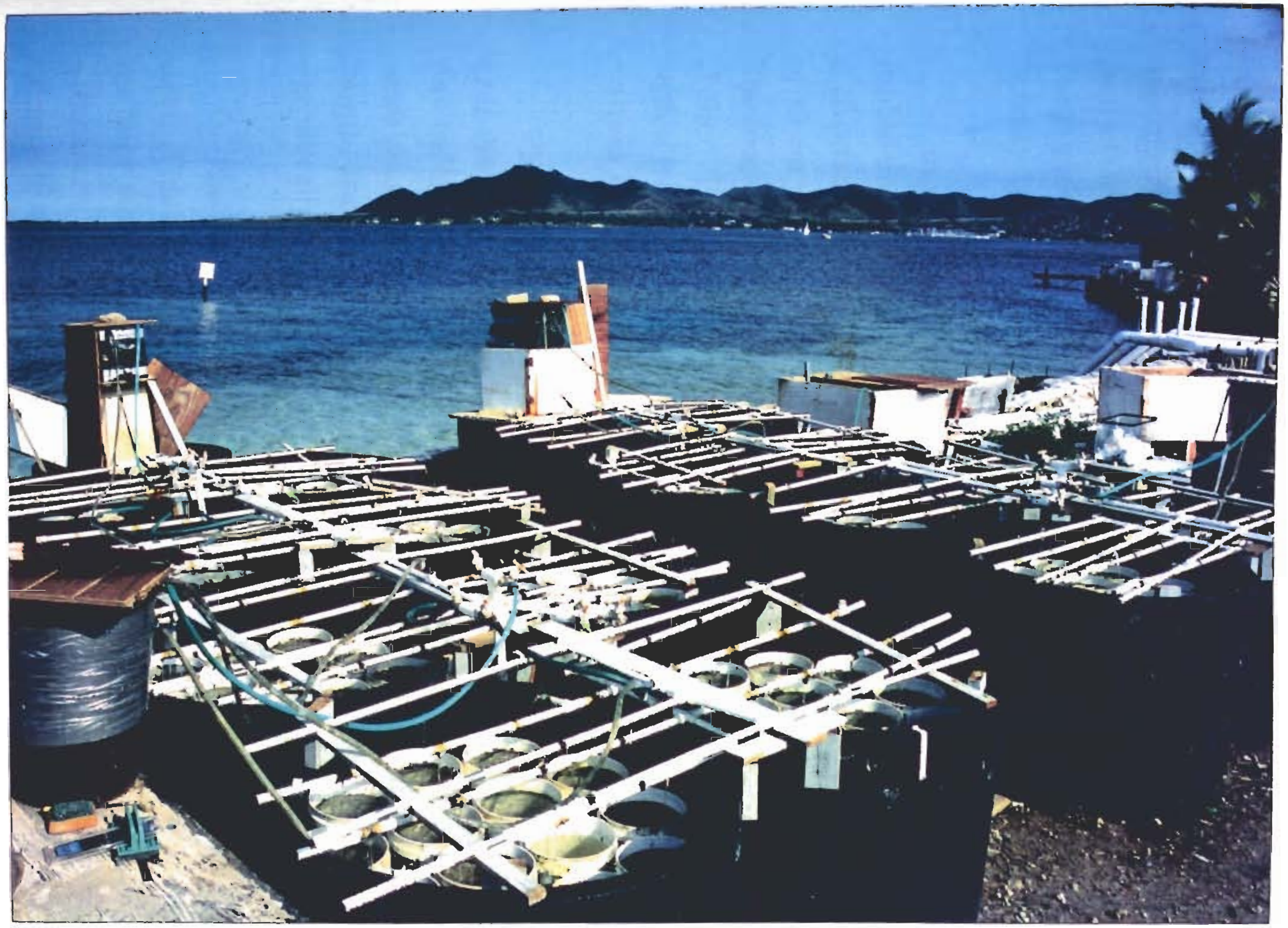

Figure 3.1. Experimental set-up (Hawaii Institute of Marine Biology, 1991), showing the 16 circular tanks containing 224 buckets, 4 predator holding tanks, and PVC pipes providing constant water flow or predator odor when needed.

predation risk treatment buckets. The predator tanks also received a continuous water supply and were allowed to overflow. This overflowing water was either directed into the experimental pipe system through a hose (during experimental periods), or into the adjacent circular tank otherwise. Inside the holding tanks, predators were kept individually in baskets to prevent fighting and cannibalism. The predators introduced in the prey's buckets had their claws restrained with electrical tape to prevent successful predation. These predators were kept in a separate tank, along with the predators in baskets used in the predation risk 2 treatment.

In the buckets, each xanthid crab was provided with a piece of coral rubble under which it was able to hide. Each piece of coral was selected so that it covered only a quarter to a third of the bucket's bottom, and was a maximum of 10 cm tall. Xanthid crabs were fed a single 1 cm cube of fish every afternoon. The food was deposited in the bucket as far away as possible from the crab's shelter. Predators were not fed. Every day, buckets were cleaned: siphoned with a transparent hose to remove uneaten food and debris, and scrubbed to restrain algal growth.

Every day, records were made of whether crabs had retrieved their food or if the piece of fish was still at the same location where it had been placed the day before. The date of every molt and the location of the exuviae in the bucket were also recorded. Exuviae were collected to measure carapace width and length, and the height and length of both claws (reported in Chapter 4), using digital calipers.

The data on molt timing were analyzed using an analysis of failure time data. This analysis (also called survival data analysis) is commonly used to monitor survival of lab animals exposed to different medical treatments or, in mechanics, to test the length of working life of different motors. The analysis takes into account both non-censored and censored observations, since the longer-lived animals or motors are more likely to be censored (still alive or working when the experiment ends). Applied to crab molt cycles, the analysis accumulates the percentage of molts through time, takes into account crabs that did not molt by the end of the experiment, and determines if some group of crabs molts later than others. To compare the treatments pairwise, several tests can be used. The p values given in the results are from a Wilcoxon test (which gives equal weight to early and late molts), unless noted otherwise. For more details, see Kalbfleisch and Prentice (1980).

Results

Preliminary experiments

Molt timing

Crabs in the presence of a conspecific or a predator for an hour every day spent more time in intermolt than did control crabs. For the first molt in the lab, control crabs molted sooner than the crabs in the conspecific and predation treatments (Fig. 3.2). For example, half the crabs in the control treatment had molted after 7 days, whereas half the crabs from the conspecific and predation treatments had molted only after 23 and 34 days, respectively. Overall, most crabs molted only once, but more than half of the control crabs molted a second time. In contrast, only two crabs from the conspecific treatment and none of the crabs from the predation treatment molted twice. Two crabs from the control treatment even molted a third time, but none of the crabs from the other treatments molted three times over a period of two months (Table 3.2). Note that the sample size is reduced in the predation treatment after the first molt, since crabs were lost to predation.

Growth increment

The percentage carapace width increment was compared between treatments for the first size increment. A Kruskal-Wallis test revealed significant heterogeneity among treatments (Fig. 3.3). Carapace width increments were lower for crabs under predation risk compared to crabs from control and conspecific treatments. Because too few crabs molted a second time, carapace width increment could not be compared between treatments for the second size increment.

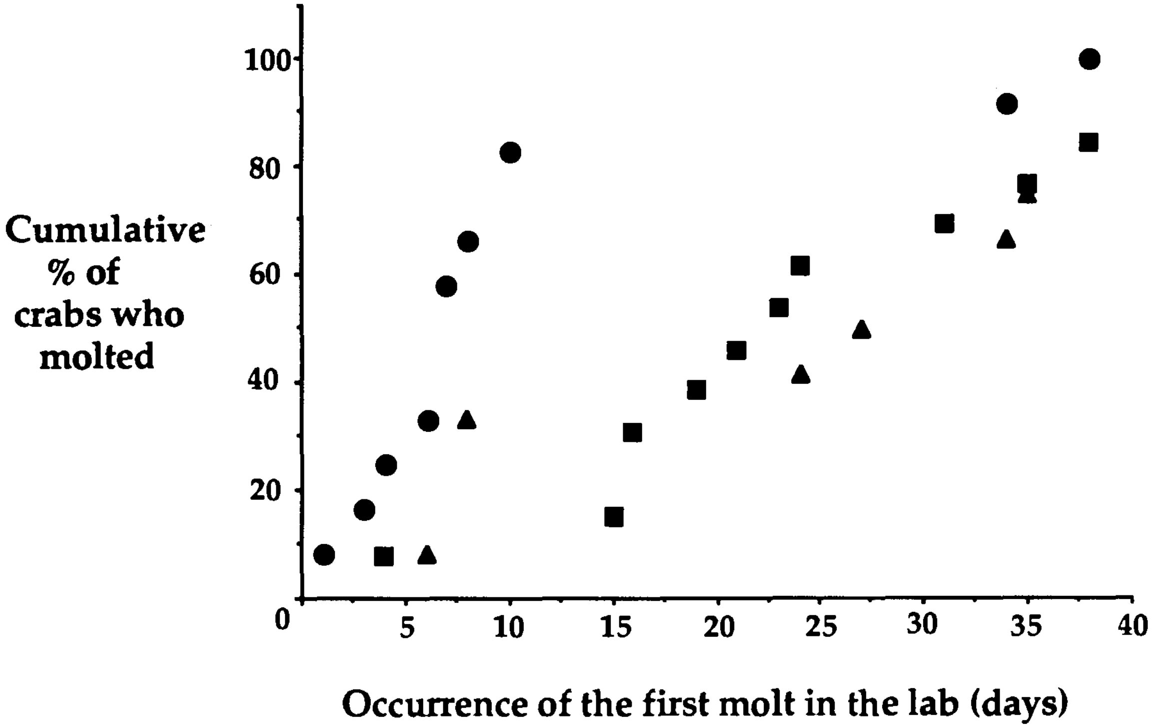

Figure 3.2. Cumulative percentage of crabs that molted in each of the three treatments over a period of 40 days from the beginning of the 1990 experiment. Analysis of failure time data, p=0.003 overall. Control vs Conspecific: p=0.003; Control vs Predator: p=0.01; Conspecific vs Predator: p=0.8.

Legend: ● Control n=12 ■ Conspecific n=13 ▲ Predation n=12

Table 3.2. Number of crabs from the control, conspecific and predation treatments that never molted (p=0.2), molted once (p=0.2), twice (p=0.01), or three times (p=0.2) during the preliminary experiment in 1990. p values are from G-tests comparing the three treatments.

	Number of molts			
	0	**1**	**2**	**3**
Control n=12	0	12	7	2
Conspecific n=13	2	11	2	0
Predation n=12 *n=6	3	9	0*	0*

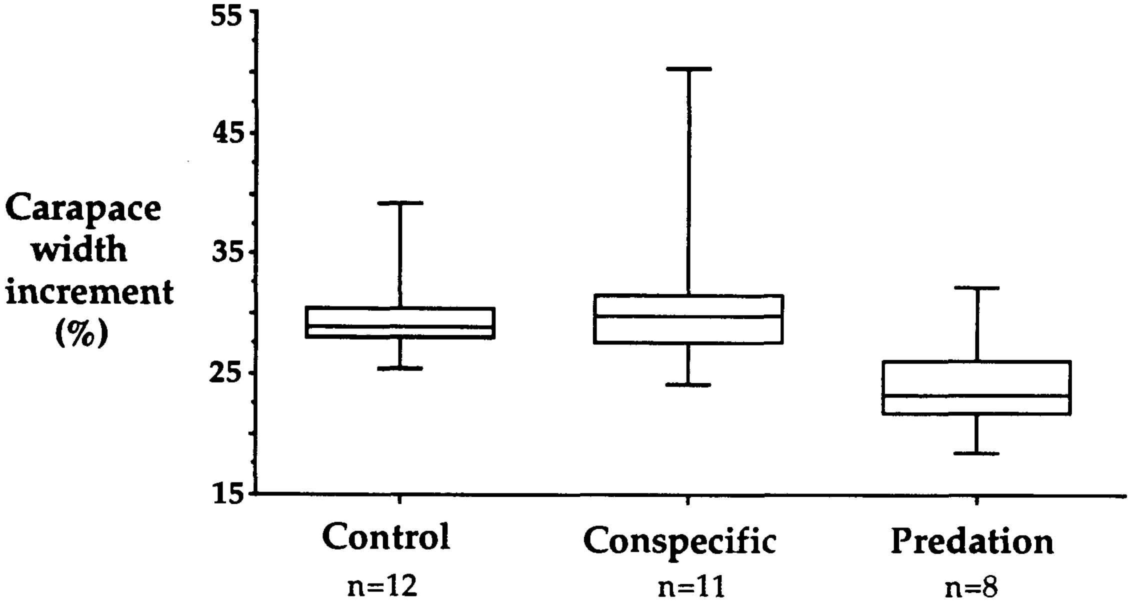

Figure 3.3. Distributions (10th, 25th, 50th, 75th, and 90th percentile) of crabs' first size increment in the lab (% size increase from previous size), for the three treatments in 1990. Kruskal-Wallis, p=0.02.

Main experiments

Molt timing

The timing of the first molt of the crabs from both the control treatment and the constant predation risk treatment (predator in the bucket) did not differ in 1991 (Fig. 3.4). The length of time crabs spent in intermolt during the first and the second complete molt cycles were also similar for control crabs and crabs under constant predation risk (Fig. 3.4). When the experiments ended, 8 of the 11 crabs under constant predation risk and 14 of the 26 control crabs had not completed the second molt cycle.

The timing of the first molt of the crabs from the control treatment and the crabs in the predation risk 1 and predation risk 2 treatments did not differ (Fig. 3.5). The length of time crabs from these three treatments spent in intermolt for the first complete molt cycle was also similar (Fig. 3.5). At the end of the first molt cycle, predation risk was increased, as described above, for predation risk treatments 1 and 2. The length of the second molt cycle was significantly longer for the crabs from the two predation risk treatments compared to the control crabs (Fig. 3.5). Also, a larger proportion of crabs under predation risk did not complete the second molt cycle compared to the control crabs: 16 of the 36 control crabs, 17 of the 27 crabs of the predation risk treatment 1, and 20 of the 27 crabs of the predation risk treatment 2 had not completed the second molt cycle when the experiment ended.

The timing of the first molt of the crabs from the control treatment and the crabs who were handled daily also did not differ (Fig. 3.6). Following this first molt, the crabs handled every day remained longer in intermolt than did the control crabs (Fig. 3.6). However, the increase in the length of the cycle is likely to have been underestimated, since I stopped handling the crabs about 35 days into their molt cycle, and molting resumed around 15 days later. The second molt cycle lengths were not statistically different (Fig. 3.6), although

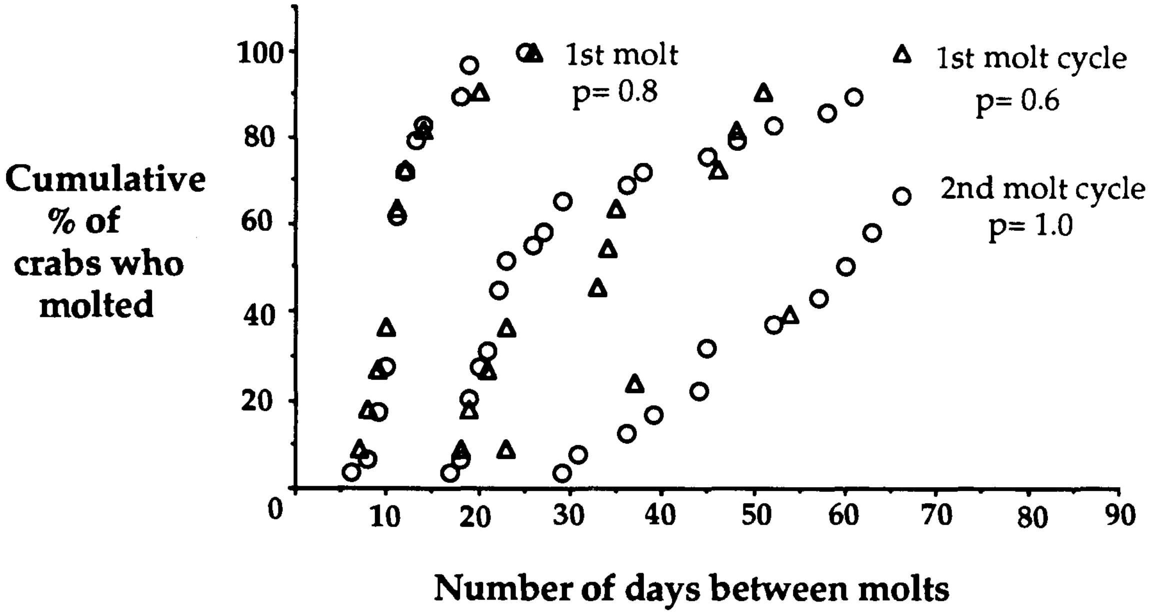

Figure 3.4. Cumulative percentage of female crabs that molted in control and constant predation risk treatments, for the first molt (measured from the day crabs were brought into the lab), and for the first and second complete molt cycles in 1991. p values from the analysis of failure time data between Control and Constant predation risk (see text).

Legend: ○ Control n=29 (1st molt and 1st molt cycle); n=26 (2nd molt cycle);

△ Constant Predation Risk n=11

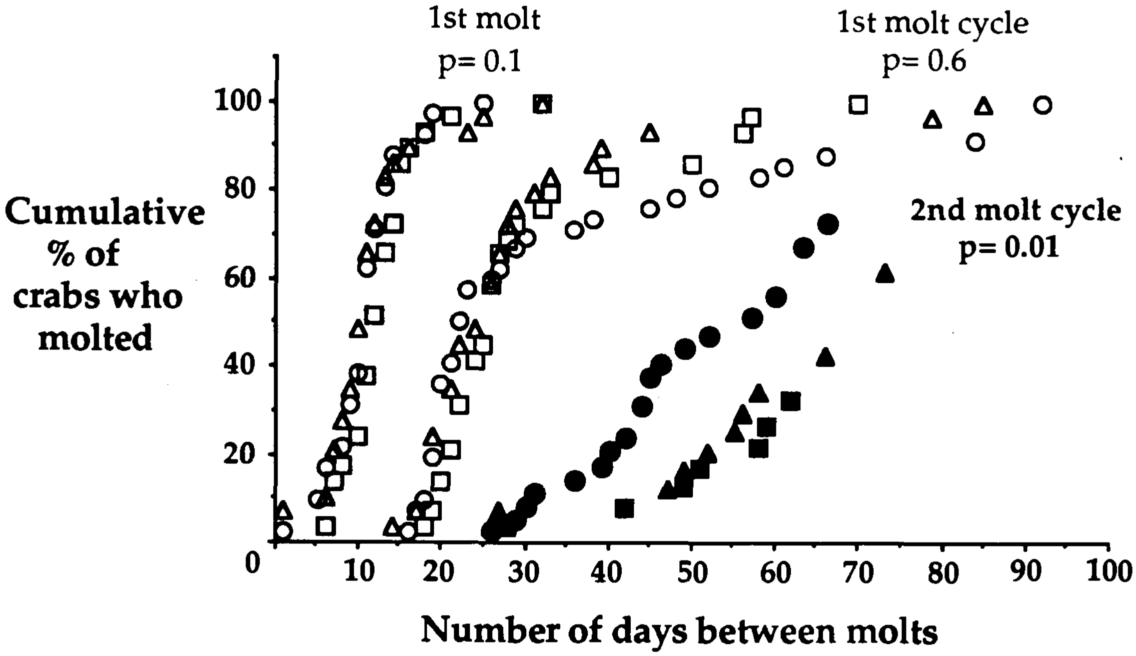

Figure 3.5. Cumulative percentage of crabs that molted in control and predation treatments, for the first molt (measured from the day crabs were brought into the lab), and for the first and second complete molt cycles in 1991. After the completion of the first molt cycle, predation risk was increased in the two predation risk treatments. p values from the analysis of failure time data, details on the second molt cycle: control vs. predation risk 1, p=0.04; control vs. predation risk 2, p=0.01; predation risk 1 vs. predation risk 2, p=0.5.

Legend: ○, ● Control n=42 (1st molt and 1st molt cycle),
n= 36 (2nd molt cycle);

△, ▲ Predation Risk 1 n=29 (1st molt and 1st molt cycle),
n=27 (2nd molt cycle);

□, ■ Predation Risk 2 n=29 (1st molt and 1st molt cycle),
n=27 (2nd molt cycle)

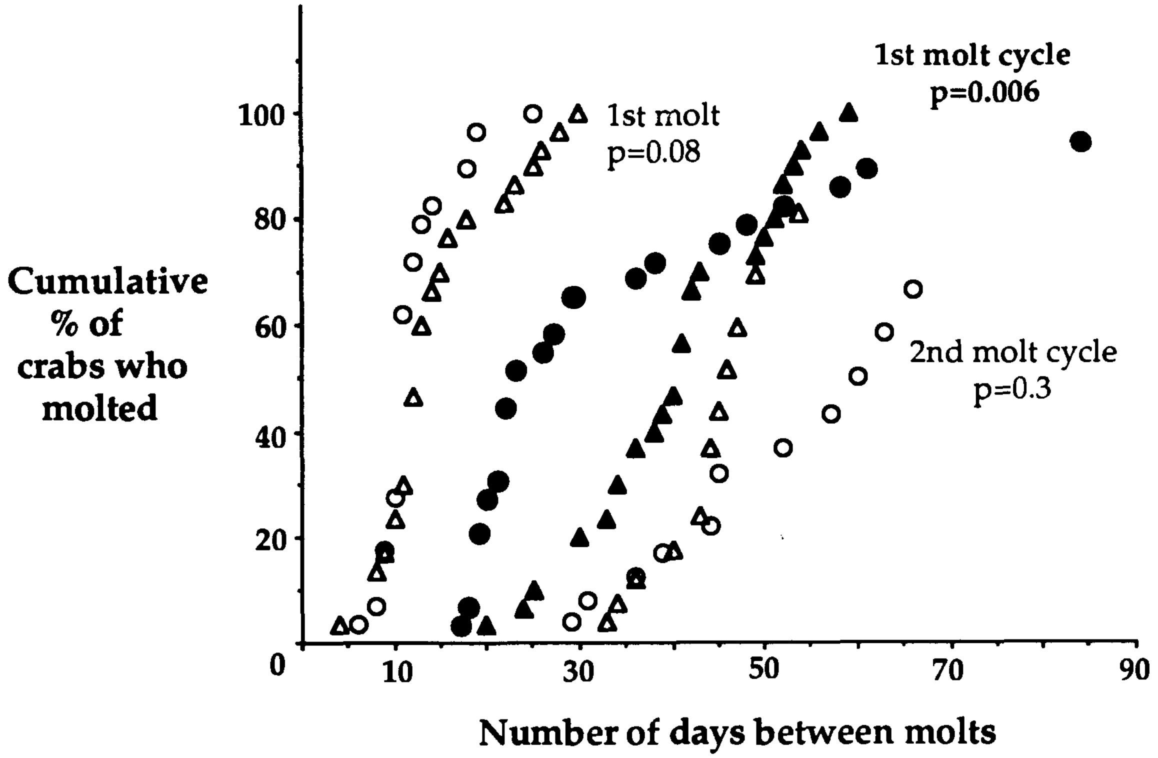

Figure 3.6. Cumulative percentage of crabs that molted in control and handled crab treatments, for the first molt (measured from the day crabs were brought into the lab), and for the first and second complete molt cycles in 1991. p values from the analysis of failure time data comparing Control and Handled crabs (see text).

Legend: ○, ● Control n=29 (1st molt and 1st molt cycle),
n=26 (2nd molt cycle);

△, ▲ Handled Crabs n=30 (1st molt and 1st molt cycle),
n=29 (2nd molt cycle)

handled crabs seem to have molted faster (shorter intermolt) than control crabs, a tendency which is significant at p=0.03 when the Log-rank test (which gives more weight to later molts) is used instead of the Wilcoxon. When the experiment ended, 16 of the 29 handled crabs and 14 of the 26 control crabs had not completed the second molt cycle.

Considering only the crabs from the large crab treatment groups (Table 3.1), predation risk had no effect on molt timing: both female and male crabs in the predation risk 1 treatment first molted at the same time as control crabs in the lab (Fig. 3.7). Males were larger than females at the beginning of the experiment (Table 3.1), which probably accounts for the significant difference in molt timing between sexes (Fig. 3.7). Only three of the large crabs molted a second time. It was therefore impossible to compare molting behavior between large and small crabs after the increase in predation risk.

For the small crab treatment groups, females seemed to molt later than males in all molts (Fig. 3.8), but these differences were not statistically significant.

Growth increment

There was no consistent tendency for % width increment to vary with premolt boby size, at least within the narrow size range used in this experiment (Table 3.1). Data for small males and females were combined, where both were available. Percentage carapace width increments were compared for the first size increment with a Kruskal-Wallis test, revealing significant heterogeneity among treatments (Fig. 3.9). Percentage carapace width increments were lower for handled crabs compared to crabs from control, predation risk 1, predation risk 2, and constant predation risk treatments. For the second size increment, the Kruskal-Wallis test also revealed heterogeneity among treatments (Fig. 3.10). Carapace width increments were lower for crabs from the constant predation risk and handled crabs treatments compared to crabs from the three other treatments. There were no significant differences among treatments for

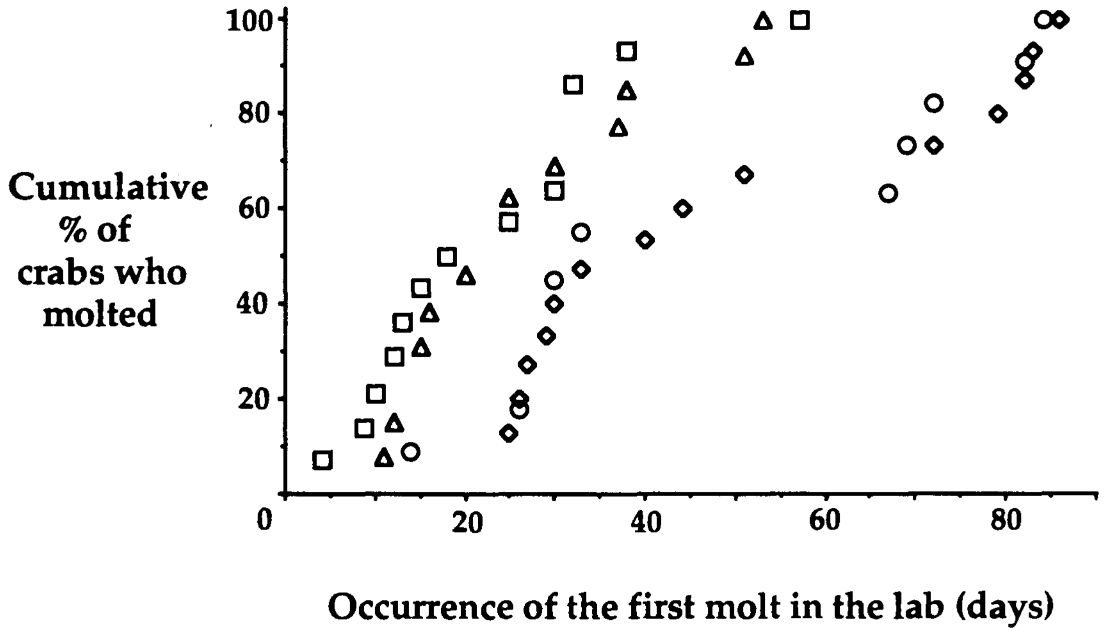

Figure 3.7. Cumulative percentage of crabs that molted in four treatment-groups (large males and females with and without predation risk), over a period of 90 days from the beginning of the experiment in 1991. Analysis of failure time data, p=0.01 overall. Control females vs. Predation risk 1 females,p=0.5; Control males vs. Predation risk 1 males, p=1.0.

Legend: △ Control females, n=16; □ Predation risk 1 females, n=16;

○ Control males, n=16; ◇ Predation risk 1 males, n=16

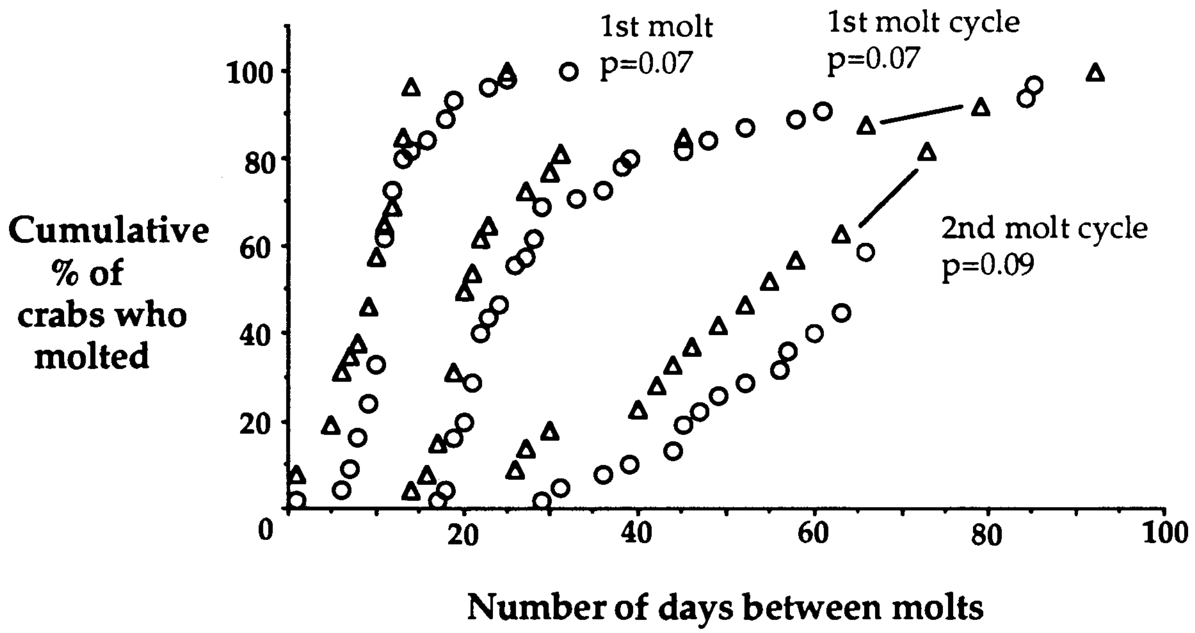

Figure 3.8. Cumulative percentage of male and female crabs that molted for the first molt and in the first and second complete molt cycles in 1991. Data from control and predation risk 1 treatments are combined, for the small crab treatment groups. p values from the analysis of failure time data.

Legend: ▲ Males n=26 (1st molt and 1st molt cycle),
n=22 (2nd molt cycle);

○ Females n=45 (1st molt and 1st molt cycle),
n=41 (2nd molt cycle);

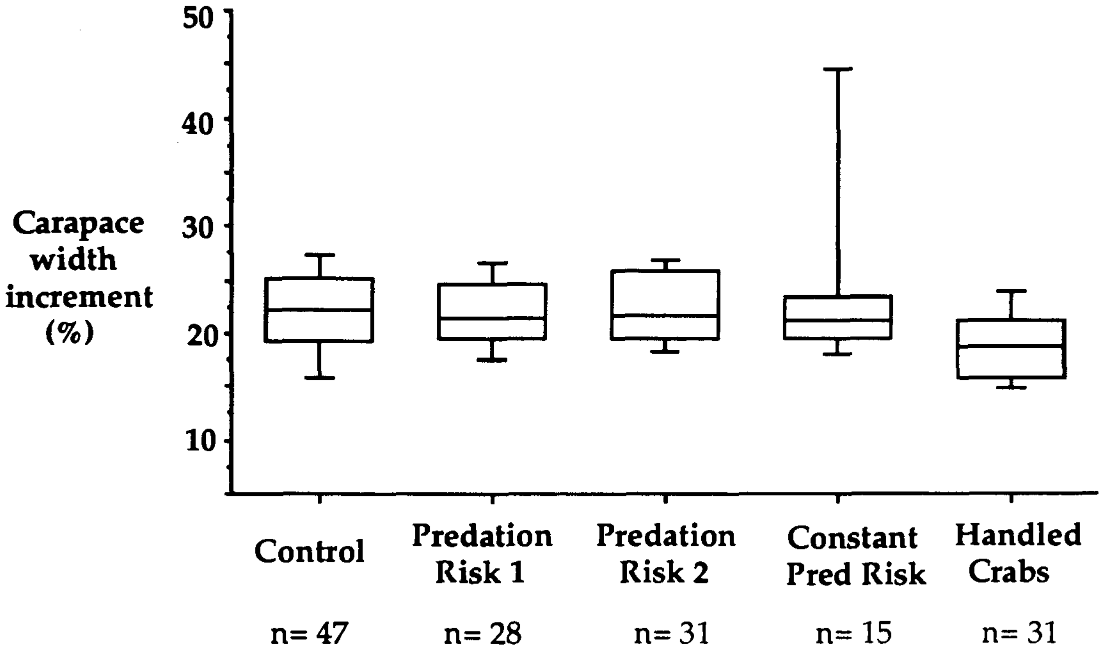

Figure 3.9. Distributions of the first size increment (% size increase from previous size) for the five treatments in 1991. Kruskal-Wallis, p=0.004.

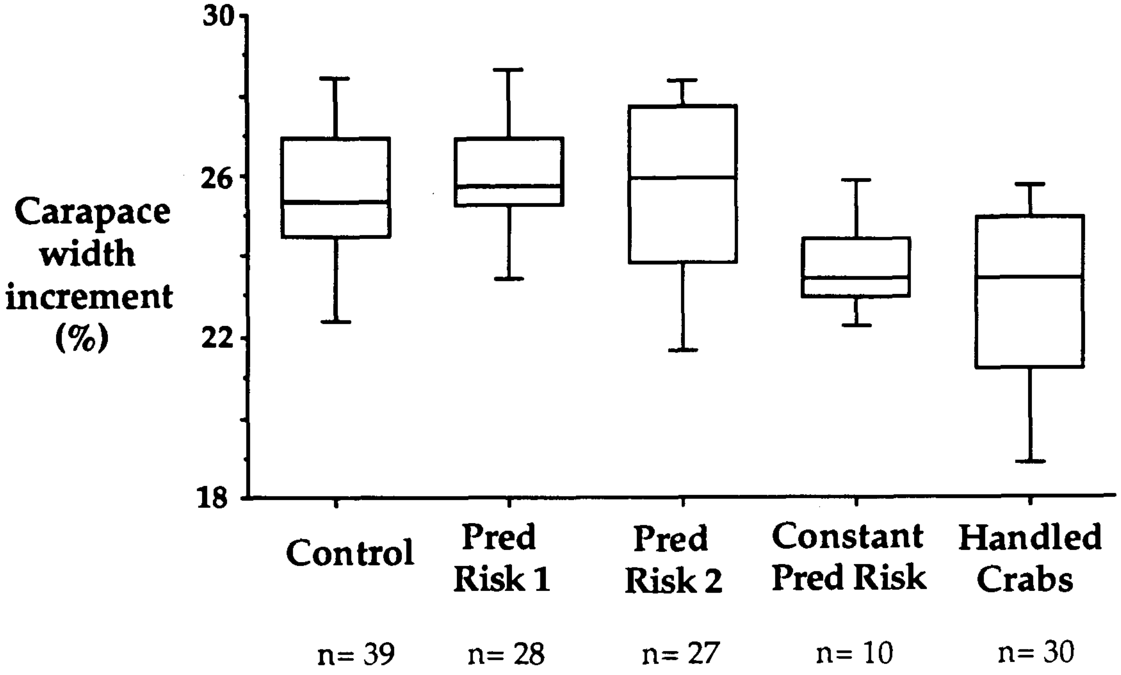

Figure 3.10. Distributions of the second size increment (% size increase from previous size) for the five treatments in 1991. Kruskal-Wallis, p= 0.0001.

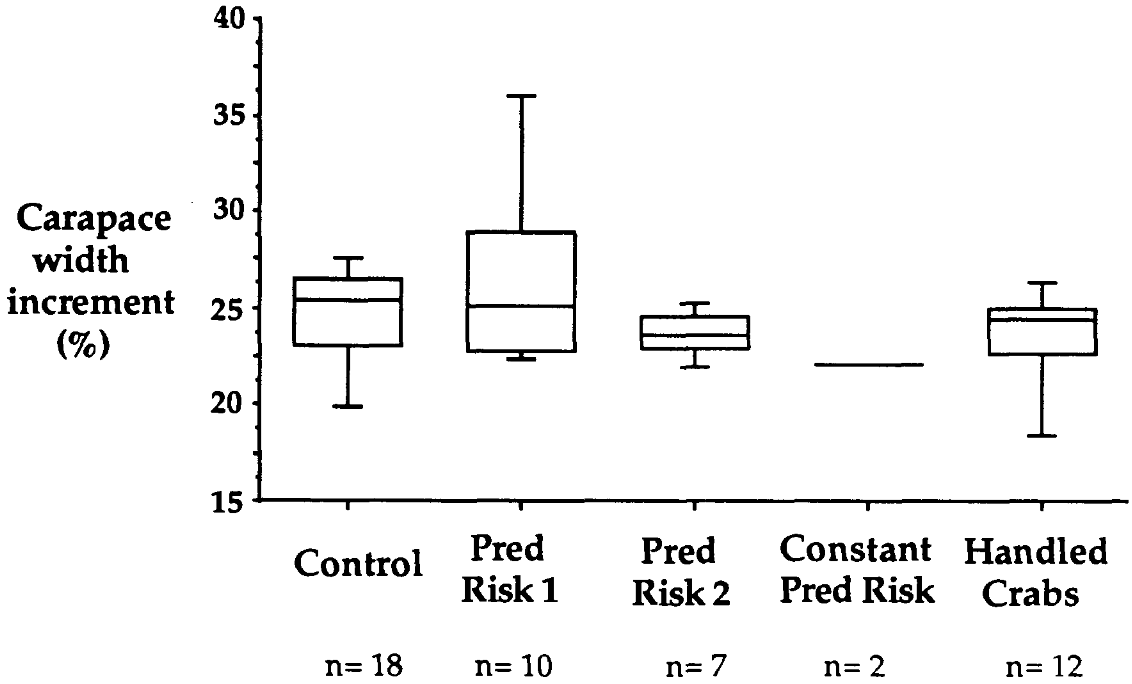

Figure 3.11. Distributions of the third size increment (% size increase from previous size) for the five treatments in 1991. Kruskal-Wallis, p= 0.2.

the third size increment (Fig. 3.11). The median % size increase was about 22% for the first molt (Fig. 3.9), 25% for the second molt (Fig. 3.10), and 25% for the third molt (Fig. 3. 11), considering only the data from the control, predation risk 1 and predation risk 2 treatments.

By combining data on the length of the intermolt with size reached after each molt, I constructed a growth curve for each of the five treatments (Fig. 3.12). Overall, growth seemed to be impaired for crabs under constant predation risk compared to crabs from the control, predation risk 1 and predation risk 2 treatments, which have similar growth curves. The handled crabs had a shorter third intermolt compared to their second, but they still experienced an overall growth loss compared to controls. Note that the last measures are censored and therefore underestimate the number of days spent in intermolt.

Comparisons of the first size increment for the large crab treatment groups showed that there was no significant treatment effect: both female and male crabs had a size increment in the predation risk 1 treatment comparable to that in the control treatment (Fig. 3.13). Although male size increment tended to be larger than female size increment, in both control and predation treatments, these differences were not significant. Since most large crabs did not molt twice, further comparisons between treatments and with smaller crabs were impossible.

For the crabs from the small treatment groups, males had a larger size increment than females for the first increment (Fig. 3.14). There were no significant differences in subsequent size increments between the sexes, although there was a tendency for males to have a larger size increment than females.

Foraging activity

Before the levels of predation risk and handling were modified, crabs from the control, the predation risk 1 and predation risk 2, and the constant

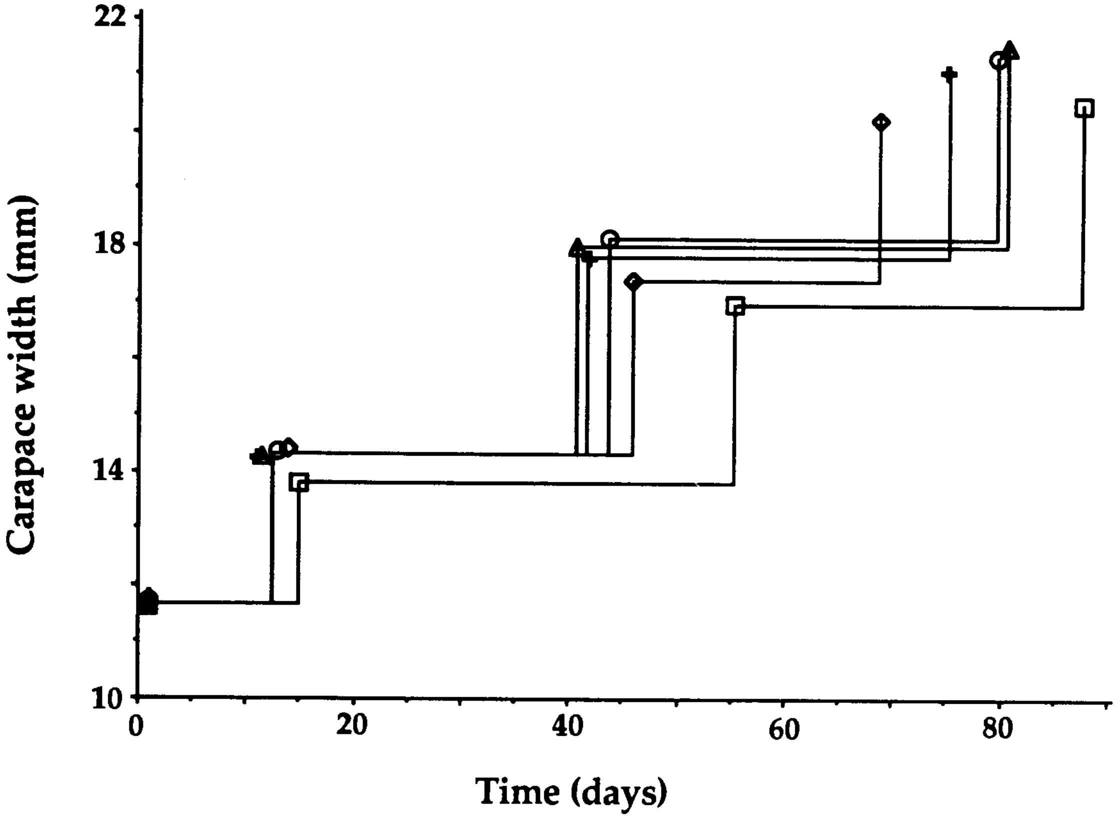

Figure 3.12. Mean growth curves for crabs from the five treatments in 1991.

Legend: ✚ Control
△ Predation Risk 1
○ Predation Risk 2
◇ Constant Predation risk
□ Handled Crabs

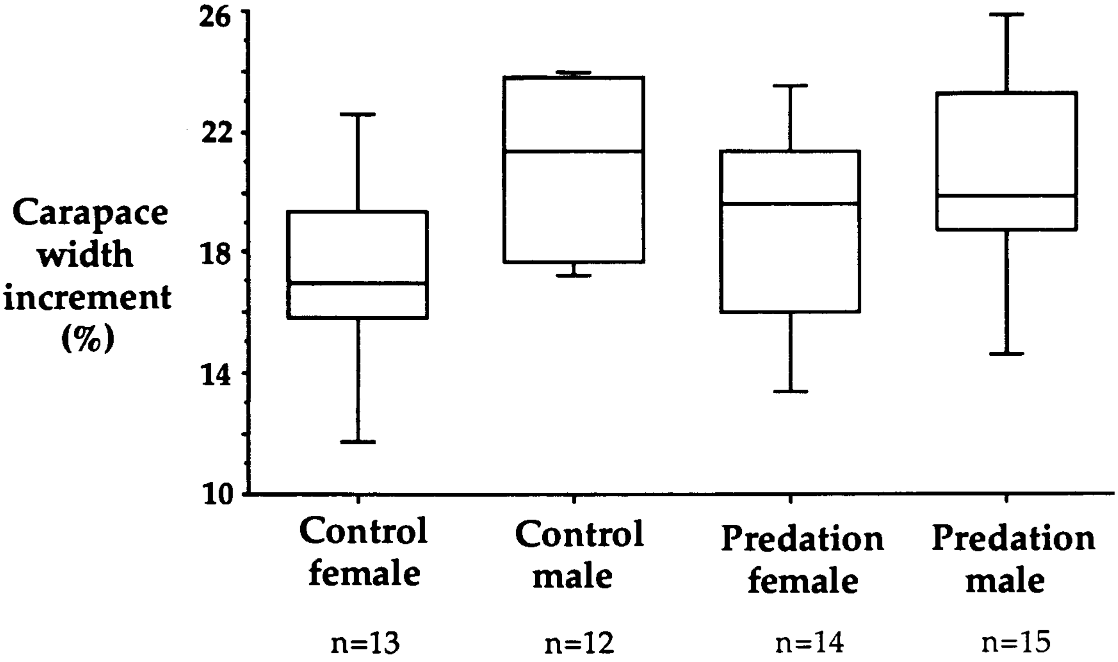

Figure 3.13. Distributions of the first size increment (% size increase from previous size) for the large crab treatment-groups in 1991. Kruskal-Wallis p=0.075. Predation=predation risk treatment 1,

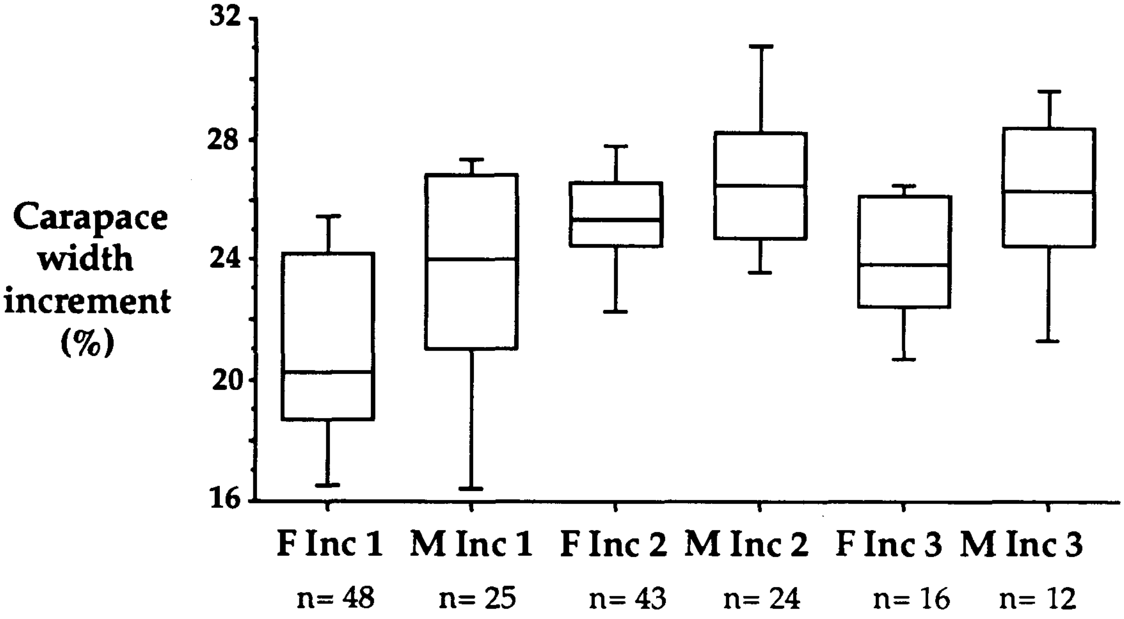

Figure 3.14. Distributions of the size increments of males and females from the small crab treatment groups, for the 1st, 2nd and 3rd size increments (% size increase from previous size) in 1991. Control and predation risk 1 treatments combined. Mann-Whitney U Test, Inc 1: p= 0.01; Inc 2 : p= 0.06; Inc 3 : p= 0.09. Legend: F=female, M=male, Inc=increment.

predation risk treatments foraged (retrieved pieces of fish) on a similar number of days: median % of days ranged from 80 to 87% (Fig. 3.15A). Only the handled crabs foraged significantly less frequently, with a median of 75% (Fig. 3.15A). After the levels of predation risk increased for the predation risk 1 and predation risk 2 treatments, and handling was reduced over half of the molt cycle, all crabs foraged with equal frequency, retrieving food a median of 80 to 85% of the days (Fig. 3.15B).

There was no relationship between the percentage of days crabs foraged during intermolt and the percent size increment achieved at the following molt in any treatment except one: crabs from the predation risk 2 treatment with a larger size increment at their second molt had foraged less in the first intermolt period (r^2=0.41, p=0.02). Overall r^2 = 0.03 and 0.005, for the first intermolt period and the following size increment, and for the last intermolt period and the last size increment, respectively.

For most crabs, there was no relationship between the percentage of foraging days and the length of time they spent in intermolt during the first molt cycle, except for two treatments: control and constant predation risk (Fig. 3.16). For these two treatments only, crabs who stayed longer in intermolt also foraged less. Such a relationship for the second molt cycle was only found in one treatment: predation risk 1 (Fig. 3.17). Overall, the correlations were weak: r^2=0.29 and 0.04 for the first and second molt cycles, respectively.

Molting location

Analysis of the location of the shed exoskeleton indicates that both female and male crabs from the control treatments molted more often in the open as opposed to under cover (Table 3.3). Females tend to molt preferentially in the open regardless of their size and the risk of predation (Table 3.3A). There was no significant difference in molt location for small males between control and predation treatments, but larger males molted more often under cover when under predation risk (Table 3.3B).

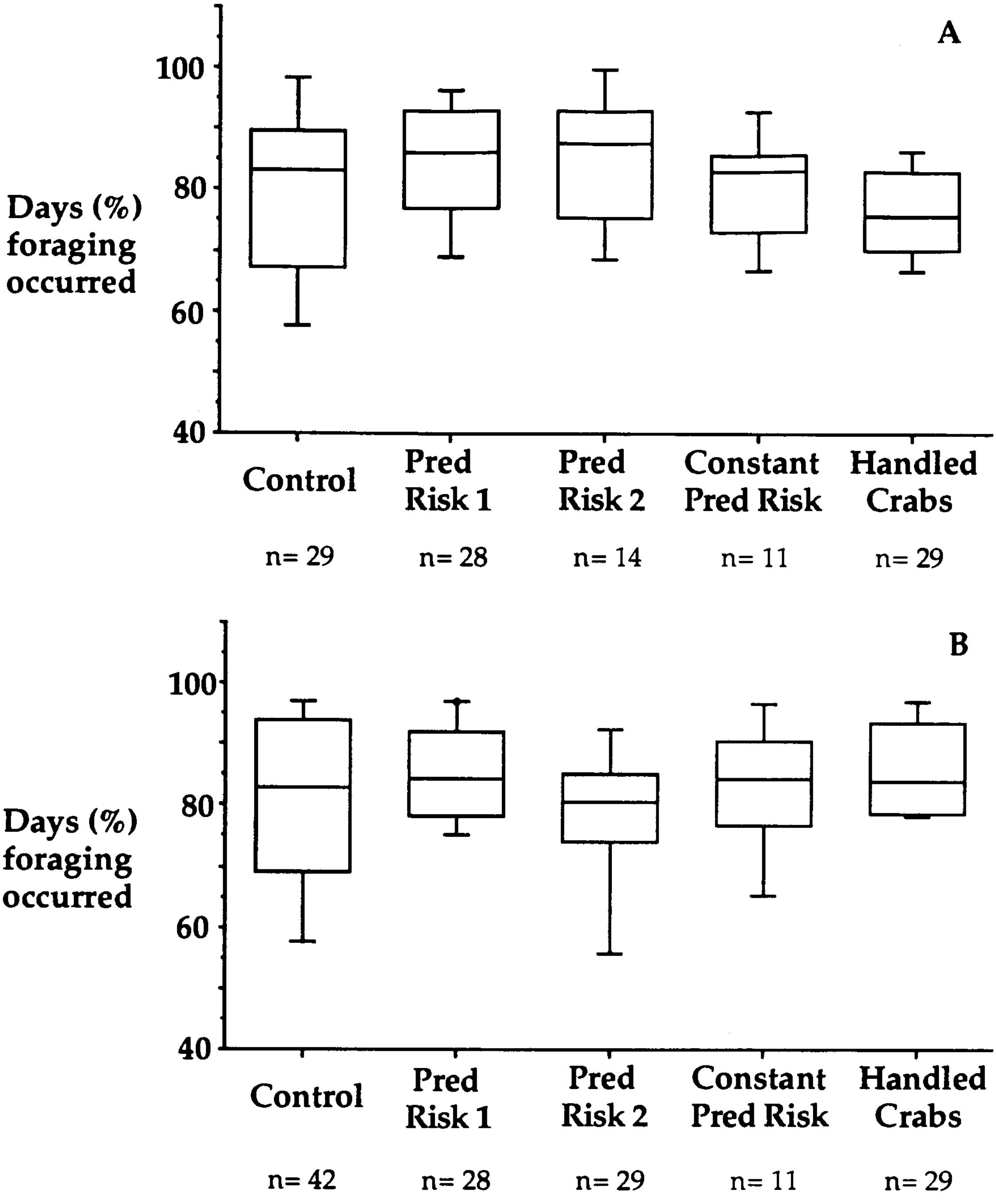

Figure 3.15. Percentage of days on which crabs retrieved pieces of food, for the five treatments in 1991. A) First two months. Kruskal- Wallis, p=0.03. B) Last two months. Kruskal-Wallis, p=0.2.

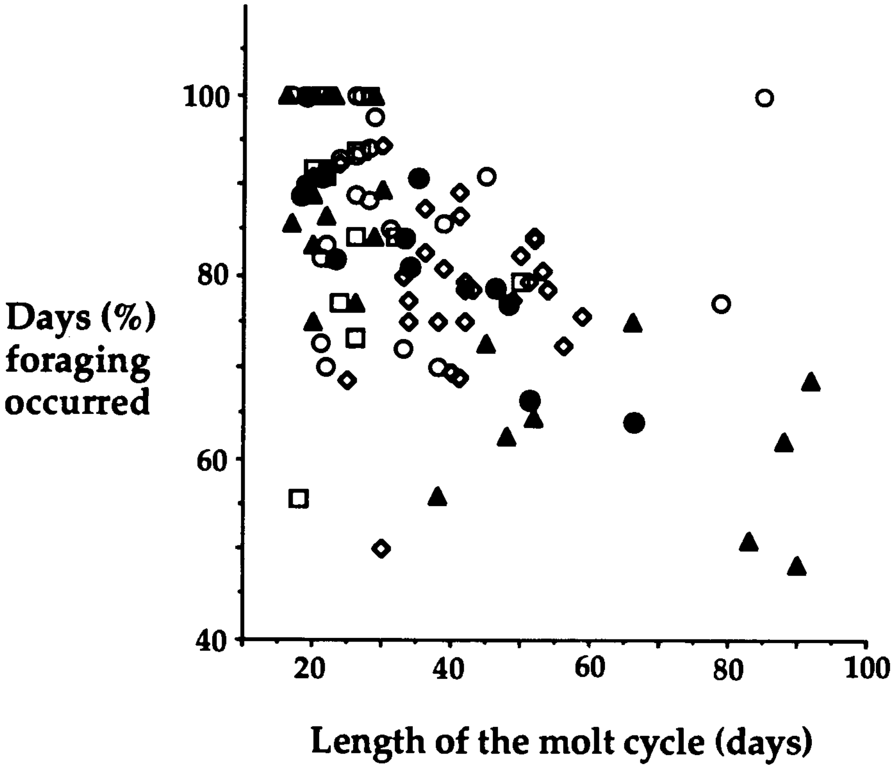

Figure 3.16. Correlation between the % of days spent foraging during the first complete molt cycle and the length of the cycle in 1991. Overall, $r^2 = 0.29$, $p=0.001$, $n=109$. The correlations for the five treatments are as follows: Control ($n=27$), $r^2 = 0.61$, $p=0.0001$; Predation risk 1 ($n=28$), $r^2 = 0.01$, $p=0.6$; Predation risk 2 ($n=14$), $r^2 = 0.001$, $p=0.9$; Constant predation risk ($n=11$), $r^2 = 0.76$, $p=0.0005$; Handled crabs ($n=29$), $r^2 = 0.006$, $p=0.7$.

Legend

▲ Control
○ Pred risk 1
□ Pred risk 2
● Const pred risk
◇ Handled crabs

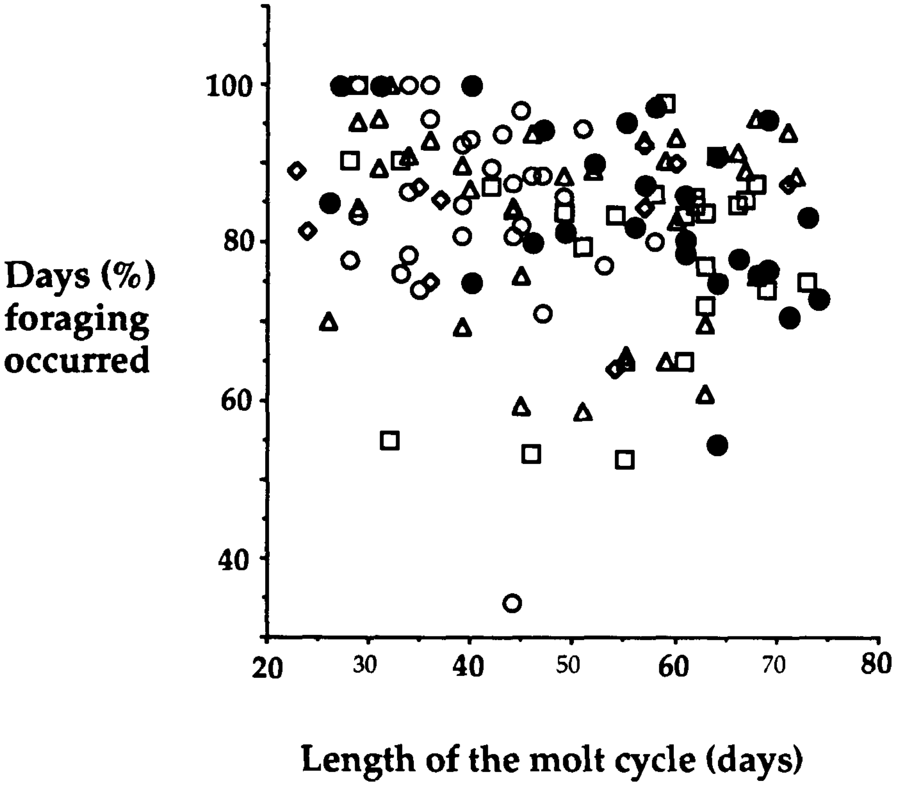

Figure 3.17 Correlation between % of days spent foraging during the second complete molt cycle and the length of the cycle in 1991. Overall $r^2=0.04$, $p=0.03$, n= 126. The correlations for the five treatments are as follows: Control (n= 34), $r^2= 0.015$, $p=0.5$; Predation risk 1 (n= 26), $r^2= 0.24$, $p=0.01$; Predation risk 2 (n= 27), $r^2= 0.003$, $p=0.8$; Constant predation risk (n= 11), $r^2= 0.01$, $p=0.7$; Handled crabs (n= 28), $r^2= 0.02$, $p=0.5$.

Legend

- △ Control
- ● Pred risk 1
- □ Pred risk 2
- ◇ Const pred risk
- ○ Handled crabs

Table 3.3. Contingency tables of location, with respect to cover, of molt occurrences for the control, and predation risk 1 and 2 treatments (the latter two combined). A. First molt in the lab, for large and small females. B. First molt in the lab, for large and small males. G-tests.

A	Control Large	Predation Large	Control Small	Predation Small
Open	9	8	30	33
Under cover	6	6	4	13
Total	15	14	34	46
p	0.9		0.07	

B	Control Large	Predation Large	Control Small	Predation Small
Open	10	5	9	8
Under cover	4	10	2	7
Total	14	15	11	15
p	0.04		0.1	

Discussion

To molt or not to molt?

Crustaceans have planktonic larvae (McConaugha 1992). Because larvae are likely to settle in a different habitat from that of their parents, and because environmental conditions may vary temporally, their phenotypic optima may well differ, a necessary condition for the evolution of phenotypic plasticity (Via and Lande 1985). Molting behavior in *Leptodius sanguineus* is influenced by the risk of being preyed upon: in the presence of a conspecific or a predator, crabs delayed their molt, trading-off growth with survival. Delaying molt is therefore a defence against predation, and molt timing is phenotypically plastic. Prediction 1 was therefore upheld. However, it was not possible to determine if larger crabs were more sensitive to risk or delay longer (prediction 2). Prediction 3 was not supported: crabs did not compensate for growth losses, but this deserves further investigation. Predictions 4 and 5 were supported: molt delay is not mediated through reduced foraging activity, and crabs can molt preferentially sheltered under predation risk.

In the following discussion, I emphasize the potential mechanism responsible for molt delay. I also discuss further the likely nature of plasticity in molt timing when the environment fluctuates in space and time, and how the crab should decide when to resume molting. I also address how molt timing and size increment may be influenced by crab size, sex and stage in the molt cycle. Size increment is reduced under predation risk, which may also be an adaptation. However, since compensation for growth loss should be advantageous, I suggest conditions in which it could occur. I also discuss how the location of the molt may be important. Finally, I consider the consequences of phenotypic plasticity in growth for crab population dynamics.

Were energetic limitations responsible for molt delay?

Nutritional status influences crustacean growth by affecting molt frequency (Cobb et al. 1982, McCauley et al. 1990, Gurney et al. 1990). However, several observations in the litterature make it apparent that molting is not a simple consequence of food intake rate. In *Balanus balanoides*, water turbulence alone, in the absence of food, can stimulate molting (Barnes and Barnes 1982). Also, precocious molt is common among decapods, for example *Gecarcinus lateralis* usually molt once a year, but molt within 2 months after multiple autotomy (Skinner 1985). In several species, female's nuptial molt does not feature any size increment even though the energy invested in eggs is mostly yet to come (Hartnoll 1982, Havens and McConaugha 1990). Even more convincingly, molting can be followed by a reduction in body size in *Daphnia longispina* (Hartnoll 1982).

In the present study, predation risk caused *Leptodius sanguineus* to delay molting. Assuming that food retrieval is a good index of amount of food eaten, we may conclude that the delay was not mediated through energetic limitations, since foraging behavior was mostly unaffected. Even in cases where individual crabs had longer intermolt periods and retrieved pieces of food less often, the direction of cause and effect is unclear: were the crabs forced to stay longer in intermolt because they did not gather enough food, or were they gathering less food because they decided to remain in intermolt, a stage with lower energetic requirements? The data on food retrieval over the entire molt cycle suggest that crabs "foraged" less over time (as growth was completed), which would support the second hypothesis.

In the case of handled crabs, the number of days on which foraging occurred was significantly lower than for the control crabs. Therefore both predation risk and energetic limitations may have affected molting in this case. Otherwise, most crabs retrieved pieces of food between 70 and 100% of the days, regardless of the level, type or variation in predation risk. The most extreme case was a crab who foraged only 35% of the days and yet molted earlier than the mean for the other crabs. Therefore, molt delay was not mediated through energetic limitations.

Delaying molt: a response to the risk of being eaten

To date, cases of animals modifying their molt cycle in the presence of conspecifics have involved animals continuously in the presence of each other, resulting in a synchronized molt, or in a delayed molt for subordinate individuals. Continuous presence is not necessary to cause a delay. Alien *L. sanguineus* introduced for an hour every day caused the inhabitant to postpone it's molt. The fact that such a short exposure to a conspecific delayed molting suggests that the crabs responded directly to the risk of being cannibalized, rather than to permanent hormonal inhibition (due to continuous presence), or to reduced food access, two alternatives often suggested (Cobb et al. 1982). Crabs may have delayed due to a perceived increase in predation risk, since the larger conspecific I introduced was a better competitor for the single refuge provided and often left the owner in a more vulnerable position. But since they did not receive any cues from other predators, this explanation is less likely.

In 1990, xanthids also delayed their molt when a predator was introduced for an hour every day, and the delay may have been longer than in the presence of a conspecific. In 1991, crabs delayed molting when the presence of predator odor was increased in time from 1 to 24 h (with an increase in the number of predators), and they also delayed when the strength of the odor was increased but the time was kept constant (at an hour each day). The response seemed stronger in this last treatment, since fewer crabs had molted by the end of the experiment. But it is still unclear what exactly the crabs perceived: the presence of one large predator or several small ones, an increase in frequency of predator visitation, a change in the proximity of predators, or a combination of these and other cues. What cues are most reliable and whether they affect molting differently also need investigation. One thing is clear: a predator need not be present for a long period of time every day in order to cause a delay, and

Leptodius sanguineus can use a small subset of the possible cues to assess risk and delay molting.

Under what conditions should molting be delayed?

Since Darwin, plasticity has been thought to be favored in situations where species are subject to repeated and great changes in environmental conditions (Caswell 1983). Such variation will favor a generalist strategy, where a plastic response will maximize fitness over different environments compared to a non-plastic response (Thompson 1991). Many crustaceans have planktonic larvae that often disperse and settle in different habitats from the parents. These different habitats most likely vary in predation pressure. Therefore, a fixed pattern of molt would be less advantageous than a flexible one. The data from both years supported this hypothesis: in response to the risk of predation, *L. sanguineus* changes the timing of it's molt.

Plasticity in molt timing seems advantageous because of between-habitat variation in predation risk. However, even within a habitat risk is likely to vary temporally. Many predators trap-line several foraging grounds (so are not continuously present) and the size of predator cohorts may vary through time. Plasticity is especially advantageous in environments which change within the animal's lifetime (Lessells 1991). Therefore, the timing of the molt should not be set according to predetermined expectations of levels of predation risk, but should respond to variations in predation risk specific to the environment in which the animals live. Crustaceans should be especially sensitive to relative levels of risk as opposed to absolute levels, since growth must be persued to some extent even in the most dangerous environments. In this case, molt delay should not be a response to a threshold of risk, but a response to a perceived increase in predation risk.

In 1990 the presence of a predator in the bucket for an hour every day (constant predation risk treatment) caused a delay, while in 1991 the presence of the predator in the bucket for the same amount of time did not cause the crabs to delay molting. If crabs responded to an absolute level of risk considered high, they should have delayed molting in both years. Lab conditions being similar, the level of predation risk in the field might have been lower in 1990 compared to 1991, accounting for the results. In 1991, crabs who experienced an increase in predation risk midway through the experiment delayed their molt compared to crabs from control and constant predation risk treatments. It could be argued that these crabs delayed their molt because a threshold of risk was reached, but because the new level of risk in the predation risk 2 treatment most likely represented a lower risk than in the constant predation risk treatment, I argue that crabs responded to the change (increase) in predation risk. These results suggest that a rule of thumb exists, such as: delay if predation risk is greater than previously experienced, or molt otherwise.

How long to delay molting?

Since the experiment ended while most of the crabs were still delaying, the total time crabs delayed molting could not be determined. Nevertheless, the question deserves attention.

The adaptive value of plasticity depends on the existence of environmental cues that accurately predict future conditions (Lessells 1991). Is predation risk predictable? If a decrease in predation risk is predictable, then crabs should wait to resume molting until predation risk decreases. If predation risk is unpredictable, or shows no signs of a decrease, then molting should be unaffected, or could be delayed and resumed after a fixed period of time. It may also become a game between predator and prey, where predators must decide how long to forage in an area in order to encounter freshly molted prey (trade-off between encounter rate with profitable prey here vs. search time and encounter rate somewhere else), and prey must decide how long to delay molting in the presence of predators (trade-off between survival and growth), which may also be influenced by conspecific behavior. In 1990, most crabs eventually molted once: 16 days later than the control for the crabs from the conspecific treatment, and 27 days later for the crabs under predation risk (numbers based on the median). In 1991, crabs seemed to delay longer when exposed to higher concentrations compared to increased duration of predator odor. These results suggest that crabs delay molting different lengths of time according to the nature or the magnitude of the risk. Clearly, to assess the impact of predation risk on crustacean growth rate, more information needs to be gathered on variation in predator numbers, sizes, visitation rates and their predictability in the field.

Molting may also be triggered because the trade-off between the benefits and the costs of molting and delaying has changed through time. For example, the benefits of molting may increase if limbs have been lost or metabolites must be eliminated. In addition, molting may also be favored to gain opportunities for reproduction, to eliminate an increasing epibiotic or parasitic load, or to respond to increased competition, where larger size is favored.

When in the molt cycle to delay molting?

In 1991, the handled crabs started molting two weeks after handling ceased. How is this time partitioned between assessing the new level of risk (to predict the likelihood of a stable low level of risk), and physiologically preparing for molting?

The commitment to molt is believed to be irreversible at apolysis, which marks the start of the degradation of the exoskeleton at stage D1 (Skinner 1985). About 75% of the exoskeleton must be degraded before ecdysis (Skinner et al. 1992), and this takes about 24 days in *Gecarcinus lateralis* (Skinner 1985). That *Leptodius sanguineus* molted 15 days after predation risk (handling) stopped may indicate that this crab takes less time than *G. lateralis* to go through apolysis. This is not likely since tropical species spend most of their molt cycle in the D stages (Lockwood 1967), and this may mean that the molt delay occurred after the start of apolysis. We need to know exactly when molting becomes irreversible, and molt stages may be more flexible than believed. Delaying the molt after the beginning of apolysis may also be advantageous because in some circumstances a partially degraded exoskeleton, even though softer, may be better than a completely soft one (if the process were allowed to continue through to the molt itself). Cobb et al. (1982) suggested that subordinate *H. americanus* delay their molt by lengthening stage D0. Webster (1982) stated that the duration of stage D2 is increased in *Palaemon elegans* undergoing vitellogenesis, which delays molting. Skinner (1985), Hopkins (1992) and Chang et al. (1993) pointed out that a decrease in ecdysteroids seems necessary to permit exuviation, whereby water is absorbed to create a pressure which breaks the cuticle. If ecdysteroids inhibit molting and their production is stimulated under predation risk, a delay in molting may be possible all the way up to the point where water is absorbed, at the end of stage D4. If this is the case, molting could almost be instantaneous after the animal has assessed the environment as being safer. Rapid molt has been observed in antarctic krill (Hamner et al. 1983). Although conditions were reversed, since molting was used as an escape tactic, this observation suggests that molting or delaying are under fine and constant control.

Therefore, two events could potentially be delayed: 1) the onset of apolysis (pre-molt); and 2) the further resorption of calcium from the exoskeleton at any point during premolt. In the first scenario, the decision to delay molting is further removed from the actual molt, and potentially more costly in terms of growth losses, but offers better protection. In the second scenario, the benefits of delaying are potentially more immediate (at the extreme, ecdysis could be stopped). Delaying calcium resorption, preventing increasing vulnerability to predators, could be followed by a period of increased resorption or even the shedding of a carapace richer in calcium, in order to get through the ecdysis process sooner.

Were larger crabs more sensitive to risk?

In the present experiment, larger crabs did not complete a second molt cycle; consequently the effects of an increase in predation risk could not be determined. The plastic response to the same stress may vary throughout an organism's life cycle (Caswell 1983, Schlichting 1986), and because the cost of molting increases with size (Fig. 2.12), larger crabs may be more sensitive to changes in predation risk, and delay for a longer period of time relative to their already longer intermolt period (compared to smaller crabs). Therefore, crabs from the predation risk 1 and 2 treatments may not have delayed molting in the second molt cycle in response to changes in predation risk, but simply because they had become larger and were more sensitive to the risk of predation. It is indeed a confounding factor, but because the crabs from the constant predation risk treatment also grew larger and yet did not delay, the increase in predation risk was probably the factor most responsible for the delay observed.

Sex differences in molt timing

In the literature, crustacean females are often described as staying longer in intermolt due to the investment in egg-bearing (Hartnoll 1982). At comparable sizes and in similar situations, the intermolt of *L. sanguineus* females was comparable to males. Females may have stayed longer in intermolt than males, but the effect was not statistically significant. The slight difference may be due to the fact that females were making some investment in reproduction even though they were not carrying eggs, or were delaying molting a few days to see if a mating opportunity would arise.

How much to grow?

A few hours after the molt is completed, the new exoskeleton increases substantially in size, but true growth, the incorporation of new tissue, occurs during metecdysis, starting at stage C1 (Lockwood 1967). By stage C4, tissue growth and restoration of newly replaced limbs are completed, marking the start of the intermolt period (Table 1.1).

Since molt, and thus size increment of the carapace, is separated in time from true growth, what determines how much of a size step an animal takes?

The water uptake permitting exuviation begins in stage D4 and continues afterwards to expand the new exoskeleton to a larger size (Skinner 1985). Lockwood (1967) reported that several species of crabs go through this process in somewhere between 6 and 12 hours. Mangum (1992) found that the uptake of water in different populations of the blue crab lasts between 3 and 18 hours, and is not always explained by water salinity. Such variation in water uptake may result in differences in size reached, and because water uptake is not a passive process (Spivak 1988) some control may exist on the degree of expansion. However, expansion may be solely determined by the amount of cuticle present, if porosity of the material after expansion is not variable. In this case, size increment may be determined when the new cuticle is synthesized in stage D4, and could be influenced by predation risk and food availability encountered then.

Although energy intake is an important component of growth, the relationship between food intake and size increment is not straightforward. When a shortage in intake was shown to decrease size increment, it was usually only detectable at an extreme diet reduction (Hartnoll 1982, Havens and McConaugha 1990). Size in *Hemigrapsus oregonensis* (Hines 1989) and size increment in *Homarus americanus* (Havens and McConaugha 1990) were not affected by food availability. For *Leptodius sanguineus*, the number of days food was retrieved during intermolt was unrelated to size increment achieved at the following molt, except for the first size increment of one treatment, where size increment was larger when crabs foraged less. After the C stages, when growth is completed, food intake is used for maintenance and accumulation of reserves to be used during the next molt, when the animal cannot feed (Lockwood 1967, Skinner 1985). Therefore, in the absence of other factors, food eaten in the past may not influence size increment as much as the predicted availability of food in the future. This may explain why the increase in size of the crab *Carcinus maenas* gets smaller as they settle on the mud and sand flats later in the season (Klein Breteler 1975). Phytoplanktonic blooms cause molting in the sand crab *Emerita analoga* (Siegel 1984). It would be interesting to see if crabs take advantage of such a situation by molting more often, increasing to a larger size at each molt, or both. Colby and Fonseca (1984) suggested that female fiddler crabs (*Uca pugilator*) are able to catch up in growth after the reproductive period is over to reach a size comparable to males of the same cohort, but they did not indicate how growth was accelerated.

Can crabs compensate for losses in growth?

Leptodius sanguineus did not compensate for losses of growth opportunity by a larger size increment at the next molt following a delayed molt. On the contrary, crabs under constant predation risk in 1990 and 1991, and handled crabs who molted, increased less in size and were smaller than both control crabs and crabs from the two increased predation risk treatments. Unfortunately, too few crabs molted after the increase in predation risk in these latter two treatments to detect a potential effect. Nevertheless, molt increment is definitely plastic, and a reduced size increment could be an adaptation to molting under predation risk. Compensation in size increment could occur later on, after the risk of predation has decreased.

Compensation for growth loss while delaying should take the least costly path between molting more often and increasing in size more at each molt. The growth pattern of the handled crabs, who had a tendency to reduce their intermolt period (after having delayed), thereby accelerating growth (Fig. 3.12), might suggest that molting more often while making small size steps may be less dangerous than making large size steps less frequently. A smaller size increment may be an adaptation to reduce the time needed to expand the new cuticle and hasten the hardening process, reducing the time spent in a vulnerable state. Moreover, a larger carapace should take longer to fill up with tissue mass. Vulnerability may be greater with a larger half-filled shell than a small full one, in terms of maneuverability or defence. Therefore, a smaller size increment when there is risk of predation might be favored. Alternatively, increasing to a smaller size may be an adaptation to molting faster, or a consequence of molt delay. Less cuticle may be produced in stage D to accelerate passage through this stage, or simply to prevent further damage to the existing exoskeleton, in both cases resulting in smaller size increment at molt. But, it may also be a consequence of a stress-induced increase in metabolic rate. Even if a decrease in surface to volume ratio may be important in explaining why larger crabs have relatively smaller size increments, the costs of inflation may require such smaller size increments for larger crabs. Such hypotheses require further investigation.

Compensation by a larger size increment after a decrease in predation risk also seems likely. Multiple limb loss is followed by an intensive period of regeneration which usually decreases size increment (Skinner 1985). In the blue crab, the loss of many limbs affected the next size increment, which was smaller than normal, but by the second molt, crabs had caught up in size with the controls (Smith 1990). Recovery in size did not involve shortening subsequent intermolt periods (Smith 1990), which suggests that the compensation was made by an increase in size increment. It is unclear what caused the first size increment to be reduced. Were the blue crabs energetically limited, or did they perceive a high risk of predation (one way of losing limbs)? If growth compensation also occurs after crabs have delayed molting and predators have gone, then growth may only be temporarily affected. On the other hand, cuticle production and expansion must be constrained to some upper limit. If local patterns of predation pressure are such that considerable time has elapsed, compensation through a larger size increment, and recovery of growth opportunities lost while delaying, may not be possible.

Size differences between sexes

In the field (June 1991), the size distributions of *L. sanguineus* are similar for gravid and non-gravid females, but males are larger than females (Fig. 3.18). Smaller size for females has often been attributed to energy invested in reproduction which cannot be used for growth (Havens and McConaugha 1990). Larger male size is often believed to be the result of sexual selection. Females can promote sperm competition (spermatophores can be stored in the spermatheca for periods that can extend up to two years (Sastry 1983), or can be partitioned between present and future fertilization of different batches of eggs

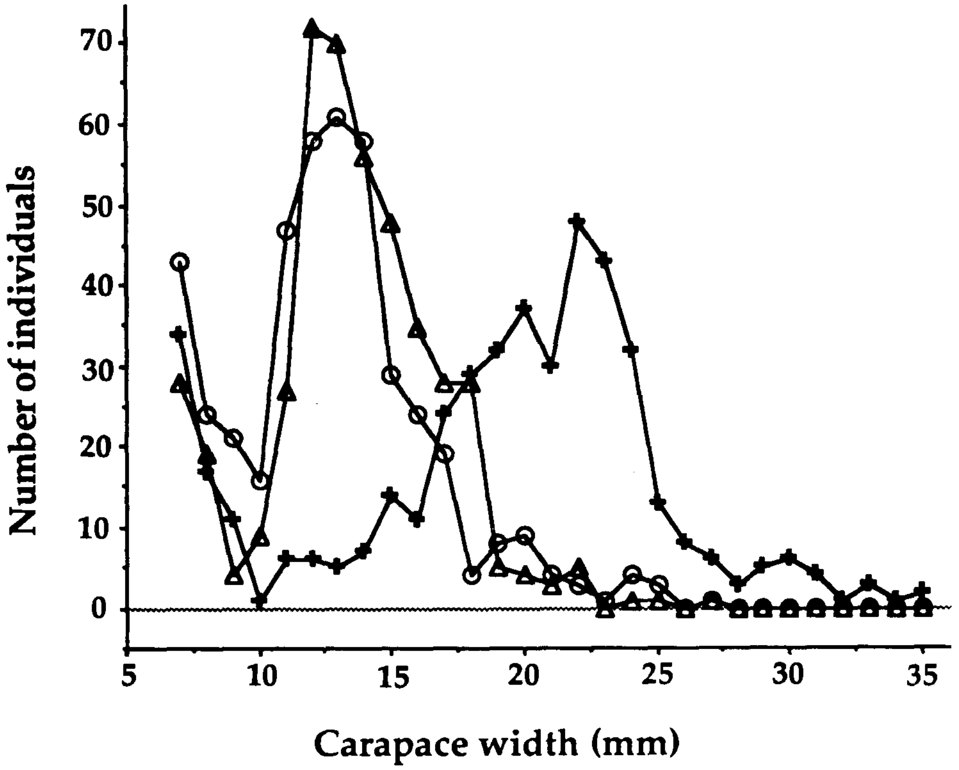

Figure 3.18. Size structure of the *Leptodius sanguineus* population (above 7 mm) randomly sampled on Gravel Island (Kaneohe Bay, Oahu, Hawaii) in June 1991.

Legend: ▲ Gravid females, n=444;

○ Non-gravid females, n=437;

✚ Males, n=439

(Elner and Beninger 1992)), therefore, males should mate with several females. While competing for females, larger males may be favored in their ability to fight and perform or resist take-overs (Ward 1983, 1988) and prevent the female from escaping (Robinson and Doyle 1985). Therefore, sexual selection may contribute to a larger size increment, which translates into more rapid growth for males.

Since females' size increments (and intermolt length) were mostly similar to males' in the lab, the size differences between the sexes in the field may be explained by the greater trade-off between growth and reproduction for females. It is noteworthy that females less than 7 mm were found to carry eggs. Since early maturity should be favored in fluctuating environments (Stearns 1976, Lessells 1991), this may be a sign that predation pressure in Kaneohe Bay is highly variable. The sex ratio seems biased towards females (2:1). It would be interesting to know if males are more susceptible to predation (maybe because they molt more often than females in the field) and whether this could explain their lower abundance in the population.

Where to molt?

Benthic species may use different habitats for feeding, mating and molting, where molting is performed in the safest habitat (Stein 1977, Hines et al. 1987). Brachyurans often need a shelter in which to hide until hardening is sufficient. For different species, molting takes place either in shelter, or in the open (Stein 1977, Tamm and Cobb 1978, Lipcius and Herrnkind 1982). In the latter case, the individual returns to the shelter after molt has been completed and mobility regained. *Leptodius sanguineus* molted either under shelter, or in the open, depending on sex, size and risk of predation.

Both small and large females mostly molted in the open, regardless of the risk of predation. Small males mostly molted in the open, but there was a (non-significant) tendency to molt under cover in the presence of predator odor. Large males molted in the open when there was no predator odor, but under cover when predator odor was present. Molting under cover may be effective at avoiding predation during ecdysis, especially for large individuals who may take longer to molt and who may be less likely to be challenged by intruders in their shelter. So why did females, even large females, molt in the open?

Hines et al. (1987) suggested that male blue crabs molt in sheltered areas of estuaries to reduce cannibalism, but females molt in the open water in order to find a mate. Mating usually takes place between a hard-shelled male and a freshly molted female (Birkhead and Clarkson 1980, Hines et al. 1987, Havens and McCaunogha 1990). Even though several species mate when both partners are in intermolt (Sastry 1983), or both mating strategies can be used among individuals of a single species (Elner and Beninger 1992, Elner and Stasko 1978), *L. sanguineus* females that molt in the open may be trading-off survival with the need for fertilization. Also, if males guard females as their molt approaches, they might offer protection that alleviates the need to molt under shelter. This protection seems important in avoiding cannibalism and predation for several species of *Gammarus* (Ward 1984a, Dick et al. 1990). These factors could explain why females molt in the open, but because small males also tended to molt in the open, despite the risk of predation, it is possible that molting in the open serves other functions. The process of exuviation may be facilitated due to space or oxygen availability, or it may be adaptive to conceal the exoskeleton from conspecifics who could use it as a cue to a neighbor's vulnerability. Male xanthid crabs *Menippe mercenaria* guard the female in the den during her molt. Once she has withdrawn from the old exoskeleton, the male carries it to the opposite side of the tank and returns to guard her (Wilbur 1989). In some species, the exuvia is eaten (Skinner 1966); or as a result of molting in the open it may be carried away by tides and currents.

Consequences of molt plasticity at the population level

The plastic response of individuals to the risk of being preyed upon during the vulnerable stage of the molt cycle may lead to differential schedules of growth between populations from different habitats. In environments where predation risk is high or highly variable, individual molting decisions, leading to more frequently delayed growth, may lower a population's growth rate and slow down reproductive output, since the number of eggs produced depends on female size (Hines 1982). Also, reproductive value of individuals, which characterizes different age classes as being worth more or less in terms of their contribution to the intrinsic growth rate of the population (Stearns 1976), may be overestimated. Since size and age would not necessarily be correlated, a smaller crab may have a lower reproductive value than expected since it may be classified in the wrong age class. With population estimates thus affected, erroneous harvesting policies may be implemented for commercial purposes, especially if generalizations are made from one population to others or from year to year.

Hartnoll (1982) has discussed the difficulty of determining age and growth rate in crustacean populations. Size frequency distributions are often analyzed and stages can be identified with some success, but not always, and it becomes increasingly difficult or impossible as the animals get older, especially for animals that molt and reproduce year-round. For fisheries management, a knowledge of age (Donaldson et al. 1981) and size-dependent molting probabilities (Mohr and Hankin 1989) are crucial. But, size does not necessarily accurately reflect past growth history: different combinations of molt intervals and size increments leading to a given size at age cannot be distinguished. *Leptodius sanguineus* growth data indicate that relationships between parameters of growth vary with the treatment applied (Appendix 3). Carapace width was a good predictor of the length of the intermolt period (Fig. A3.1) and size increment (Fig. A3.2) mostly in cases where predation risk was low. The length of the intermolt period was a poor predictor of size increment (Fig. A3.3). Therefore, models of growth should not assume that the length of the intermolt and the size increment vary only with size. Because the relationship between age, size, intermolt length and size increment is inconsistent, we have to be careful in setting the parameters of crustacean population models.

More studies are needed where the size frequency of populations is sampled at intervals to monitor decelerations and potential accelerations in growth. If growth compensation is possible, and frequently realized, many assumptions often made in models could be justified. In addition, we need to better understand the effect of the risk of predation (as opposed to predation events) experienced in the field over a long period of time: Who are the important predators? How often do they visit a prey population? Can they only affect parts of a population in a micro-habitat? How do their cohort sizes vary? Is predation risk predictable? Do predators particularly affect one size class of prey? Predictions of crustacean growth could be improved if behavior were to be integrated into models of population dynamics. Covariates such as sex and injury status have been incorporated in growth models to predict intermolt time (Hoenig and Restrepo 1989); predator behavior and responses to predation risk should also be included in such models to improve their utility.

Concluding Remarks

Leptodius sanguineus appear to control the timing of their molt, the location of their molt, and perhaps their size increment in ways that may decrease the risk of being preyed upon, thereby trading-off growth against survival. Growth in *Leptodius sanguineus* is indeterminate for both components: molt interval and size increment. Indeterminate growth may therefore be an adaptation to predation risk for animals whose growth is constrained by an exoskeleton. Of course, the important next step is to understand and measure the fitness consequences of molt plasticity. Differences in selection pressures may also cause different types of plasticity (Schlichting 1986), whereby even the opposite tactic could arise. For example, if mortality risk is high for small individuals, molting faster under predation risk (or at high density) could be selected to reduce time spent in that window of vulnerability. More research is therefore needed to understand the mechanisms and consequences of molt plasticity in crustaceans, and in arthropods in general.

Plasticity in molt timing affects only one type of defence against predators. Other defenses are likely to be induced under predation risk. The next chapter investigates whether crabs produce bigger claws and a stronger carapace under the risk of predation.

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Chapter 4

Inducible structural defences in *Leptodius sanguineus*

Introduction

Unpredictable environments favor inducible defences, because they are efficient at preventing predation and less energetically costly than permanent, constitutive defences in an environment where risk may be low (Walls et al. 1991). Inducible morphological defences have been reported in rotifers, bryozoans, and cladocerans (Harvell 1990), in snails (Appelton and Palmer 1988), and in barnacles and octocorals (Clark and Harvell 1992). Once induced, these defences usually behave as constitutive defences in protecting the animal against predators for the remainder of its life. However, fitness costs are associated with the development and maintenance of inducible defences (Harvell 1990), and usually involve shifts in allocation of time and energy from growth or reproduction. Antipredator structures are believed to increase energy expenditure or even reduce energy intake (Riessen and Sprules 1990); slower growth and a delay in the timing of reproduction are common (Harvell 1990, Riessen and Sprules 1990, Walls et al. 1991). In some species, clutch size is reduced (Harvell 1990, Riessen and Sprules 1990). Prey in variable environments must closely track predation pressure in their environment (Riessen and Sprules 1990), assess risk (Lima and Dill 1990), and invest in the production of defences, or intensify them, only when increased protection is necessary.

Many crustaceans have a heavily calcified exoskeleton that reduces vulnerability to predation (Palmer 1992, Wägele 1992), but around the molt period, vulnerability increases drastically (Chapter 2). If predation risk is variable, the timing of the molt then becomes important, and in *Leptodius sanguineus* a delay is induced when predation risk is increased (Chapter 3). The objective of this chapter is to examine whether predation risk also induces morphological change in *Leptodius sanguineus*. Predator-induced defences could include: 1) the production of larger claws; 2) faster carapace hardening after molt; and 3) the production of a tougher carapace. All are examined in this chapter.

Decapod chelipeds are used in feeding and communication, in courtship and territorial displays, in acquiring resources and defending them, in carrying a female during reproduction and guarding her, in burrowing, and in offensive and defensive actions against conspecifics (Stein 1977, Elner 1980, Hartnoll 1982, Berzins and Caldwell 1983, Seed 1986, Govind 1989, Pillai 1990, Homola et al. 1991, Weissburg 1991, Karplus et al. 1992). Chelipeds are often morphologically distinct from each other: the crusher claw is used to apply slow strong compressive force, and the cutter claw is used to perform faster more agile tasks (Seed 1986, Govind and Pearce 1989). Both chelipeds are flexible "instruments" articulated in a way to perform several different activities (Pillai 1990), but morphological characteristics that favor some functions may impair others. For example, larger claws are more effective in defence, but because they have a larger gape, make it more difficult to handle smaller prey (Seed 1986). Claws may also become disproportionately larger and more powerful as body size increases (Seed 1986). This allometry is often associated with the onset of maturity, but larger claws could also serve other functions such as defence. Even though chelipeds have often been suggested to play an important role in defence against predators, this has received little study.

Leptodius sanguineus use their claws to protect their mouthparts while being handled by a predator, and also to grasp and hold the predator's chelipeds; this halts the predation sequence (Chapter 2, Appendix 1.1). Larger claws may be more valuable in defence against predators: an increase in claw surface area could increase coverage of the mouthparts, and larger claws are stronger (Elner 1980, Seed 1986), thus they may inflict injury, and permit escape. But before such a defence can be effective, the crab may have to reach a certain size. Since small crabs were eaten readily, investing in a larger carapace rather than claws may be more beneficial when small. Since the commitment of resources to defence is likely to be traded-off with growth, the investment should vary with size and predation pressure.

The energy invested in claws for defence will not be available for growth of the body mass. Evidence for this comes from work on *Daphnia pulex,* in which neck spine production reduces growth (Riessen and Sprules 1990), and from work on *Uca pugnax,* where the regeneration of the walking legs causes up to a 67% decrease in weight of the major claw muscle (Ismail and Mykles 1992). Small crabs may not be able to afford to invest in claw defence, while mature individuals with a slower growth rate may be able to afford bigger claws, since investment in growth is not as critical, and more energy can be put towards defence. Producing big claws may also be a disadvantage if they take longer to fill with muscle tissue and remain only partially functional for a longer period of time, thus increasing the post-molt vulnerability period. As well, less energy may be available for reproduction if large claws are produced. Therefore, large claws should be an inducible defence sensitive to variations in predation risk.

The carapace plays a major role in defence as an armour. But after molt, crabs are vulnerable to predators at the time when carapace calcification and muscle formation take place. Since carapace strength relies on hardness, the rapidity of carapace hardening should be important when predation risk is high. Assuming a trade-off between energy or time used for calcium deposition and other functions, crabs molting under predation risk should put more effort into carapace hardening and consequently harden faster. Calcium and protein deposition could last for a longer period of time as well, and lead to the construction of a tougher carapace for the intermolt stage.

Inducing a stronger carapace may be analogous to snails developing a thicker shell: it reduces vulnerability to predation, but incurs an energetic cost and reduces growth rate (Appleton and Palmer 1988, Palmer 1990). Once again, the trade-off between growth or reproduction and defence should favor the induction of a tougher carapace by an increase in the risk of predation.

Methods

Investment in carapace and claws

In the course of experimentation on molt timing and size increment in 1991 (Chapter 3), carapace length and maximum width, and claw propodus length and maximum height, were measured on every exuvia of each crab. *Leptodius sanguineus* claws are dimorphic, differentiated as a crusher claw and a cutter claw. For each crab, I identified the largest claw as the crusher claw, and compared the percentage increments in crusher and cutter claw lengths and heights between predation and control treatments. Also, claw length and height were compared between treatments using a 2-way ANCOVA, with claw type and treatments as factors, and with carapace width as covariate, to determine the differential effect of predation risk on crusher and cutter claws, with carapace size controlled.

Treatment groups were: 32 small females from the control treatment, and the 32 small females from the predation risk 2 treatment combined with the 16 small females of the predation risk 1 treatment (refer to Table 3.1).

To determine the relative investment made in carapace and claws, the slopes and intercepts of the regressions of % claw size increment on % carapace width increment were compared between control and predation treatments. This was done separately for the first three molts. The effects of feeding and sexual maturity on claw morphology were minimized since crabs were fed soft pieces of fish and only females were used in this analysis. Here, the data for both claws were combined, and the total length (sum of both claw propodus lengths) and total height (sum of both claw propodus heights) were used in the analyses.

To estimate the error in the claw measurements, I measured 32 claws varying in length between 6.39 and 17.13 mm, and in height between 3.00 and 8.27 mm, a total of five times each. On average, there was less than 0.1 mm difference between repeated measures. The mean standard deviation was 0.05 mm for both length and height. The error is therefore relatively low.

Carapace hardening

During the 1991 experiment on molt timing and size increment, weights of 32 female crabs were measured daily for two months ('handled crabs" treatment; Table 3.1). Half of these crabs were controls and half were from the predation risk 1 treatment. These crabs molted once and then delayed their second molt (see Chapter 3). To allow molting to resume, handling was stopped until their next molt, following which crabs were only daily weighed for 15 days. In neither case did the measurement period cover the entire molt cycle, but the period covered is shorter in the second molt cycle. At molt, water uptake facilitates exuviation and expansion of the new cuticle. As growth of the tissue mass progresses, water is replaced by flesh. Because the specific gravities of flesh and water are similar, the rate of weight increase is assumed to be an estimate of the hardening rate of the carapace through calcium deposition. Weight increase during the first three days after the molt, and time to and weight at stabilization (determined when the change in weight was lower than 0.05 g from one day to the next) were compared between control crabs and crabs under predation risk. Since the exuvia of each crab was removed following the molt, it wasn't available to the animals for consumption after molt and thus could not be used as a potential source of calcium.

Carapace strength

On the last day of the 1991 experiment on molt timing and size increment (Chapter 3), the carapaces of the 32 small females from the control treatment, the 32 small crabs (16 females and 16 males) from the predation risk 1 treatment, the 16 small females from the constant predation risk treatment, and the 32 small females from the predation risk 2 treatment were pierced with a digital force gauge (Shimpo FG-5.0R) in order to measure resistance to breakage (lbs of pressure). Using a very fine (1 mm diameter) pointed metallic rod attached to the force gauge, two small holes were made in each crab's carapace: one in the anterior portion (called the gastric region), the other in the posterior portion (called the intestinal region). I restrained the crabs with my left hand while the force gauge was lowered onto the carapace in a regular motion controlled by a screw. The holes created did not cause sufficient injury to kill the crabs. The peak force applied (i.e. at breakage) was recorded. The toughness of the carapace was compared between the four treatments using ANCOVA, with carapace width and time since last molt as covariates. All measures are relative, since they depend on the diameter of the piercing rod; the apparatus was not calibrated, so absolute measures could not be obtained.

Results

Investment in carapace and claws

The % increment in carapace width was similar between treatments for the three size increments (unpaired t-tests: $0.2<p<0.4$). The % increments in claw length and width varied between 22 and 34 %. There were no differences between treatments in the % increments in crusher and cutter claw length or height for any of the three size increments (unpaired t-tests: $0.1<p<0.4$). However, there was a tendency for crabs under predation risk to have a larger third % size increment for both crusher and cutter claw (between 4 and 9 % longer and higher; t-tests). Crusher and cutter claw measures were combined for the analyses of the regressions of % increment .

The 2-way ANCOVA also showed no significant differences ($0.1<p<0.3$) between treatments for length and height of either crusher or cutter claws.

For the first (Fig. 4.1), and the second size increment (Fig. 4.2), there were no differences between treatments in the regressions of the % increment in claw length or height on the % increment in carapace width. Prior to the third size increment, predation risk was increased, but there were still no significant differences in the regressions between control and predation treatments (Fig. 4.3). However, in this case, the regression of the % increment in claw height on the % increment in carapace width is almost null, with a slightly negative slope and a higher intercept, for the predation treatment. This slope contrasts (although not significantly) with the positive regression for the control treatment. The reduced sample size ($n=10$) prevents confirming of the trend for both intercept and slope. In all other cases, the regressions are highly significant with slopes clustering around one.

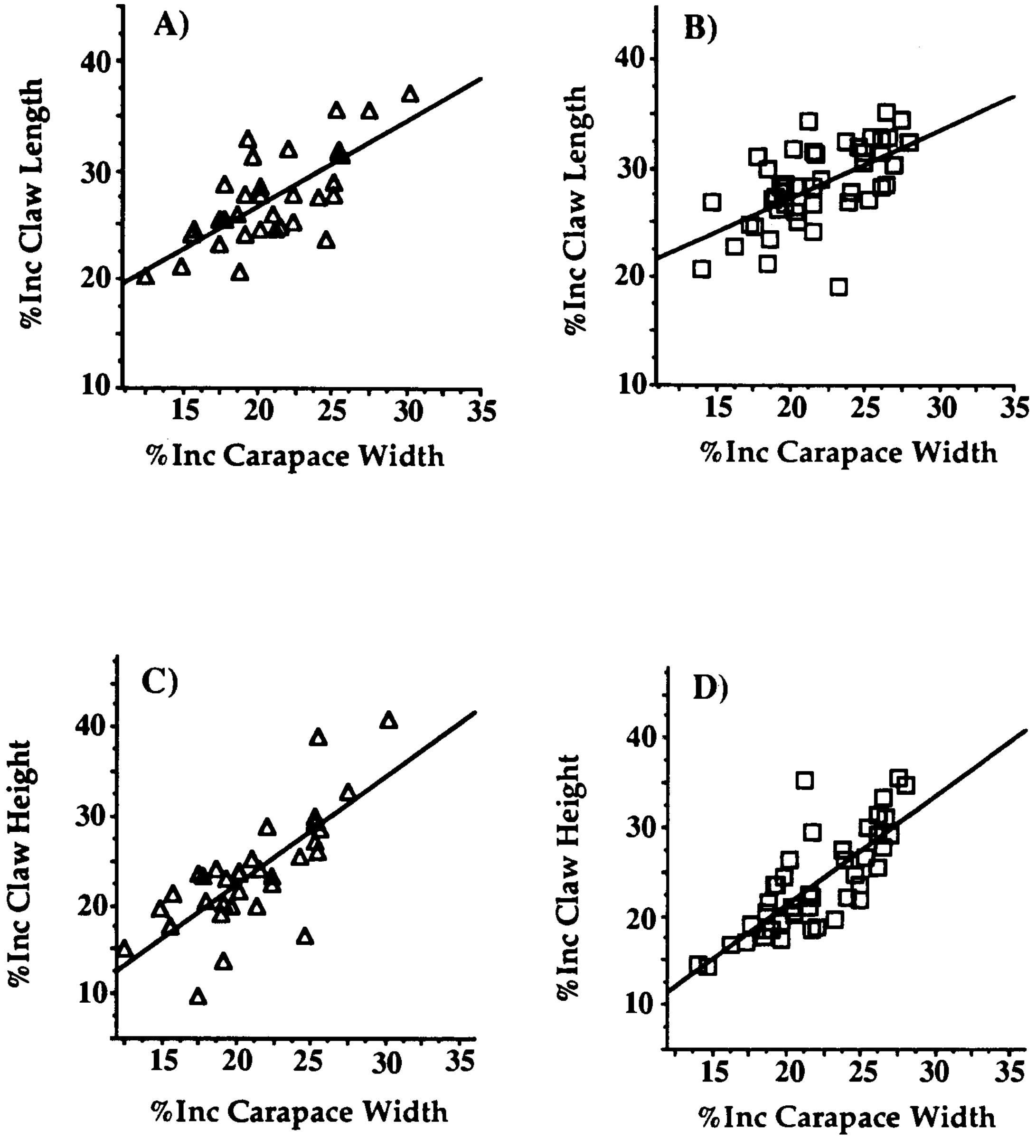

Figure 4.1. Regressions of the percentage increment in total claw length and height (i.e. the sum of the measurements of the two claws) on the percentage increment in carapace width for the first size increment in1991. A) Control: $y=0.78x + 11.09$, $r^2=0.53$. B) Predation: $y=0.62x + 14.89$,$r^2=0.36$. C) Control: $y=1.22x - 2.07$, $r^2=0.57$. D) Predation: $y=1.23x - 3.33$, $r^2=0.62$. ANOVA between slopes and intercepts of regressions A and B: $p=0.4$ and $p=0.3$. ANOVA between slopes and intercepts of regressions C and D: $p=0.9$ and $p=0.8$. The following outliers have not been plotted or analysed: B (21.9, 0); D (21.9, 0).

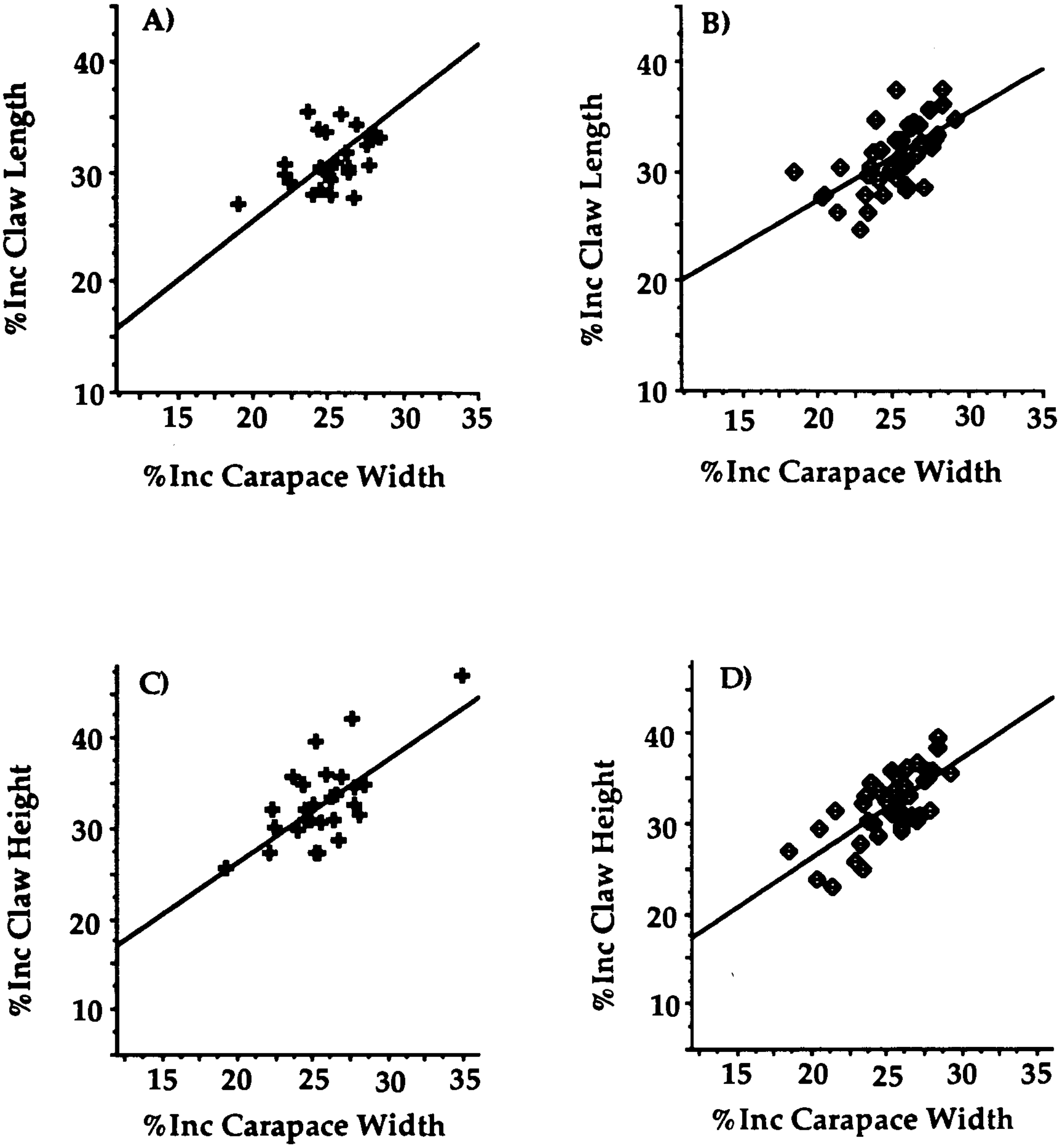

Figure 4.2. Regressions of the percentage increment in total claw length and height (i.e. the sum of the measurements of the two claws) on the percentage increment in carapace width for the second size increment in 1991. A) Control: $y=1.09x + 3.66$, $r^2=0.53$. B) Predation: $y=0.80x + 11.31$, $r^2=0.39$. C) Control: $y=1.15x + 3.24$, $r^2=0.47$. D) Predation: $y=1.11x + 3.92$, $r^2=0.50$. ANOVA between slopes and intercepts of regressions A and B: $p=0.3$ and $p=0.2$. ANOVA between slopes and intercepts of regressions C and D: $p=0.9$ and $p=0.9$. The following outliers have not been plotted or analysed: B (24.5, -17) and (51.2, 65.5); D (24.5, -14.2) and (24.5, -66.5).

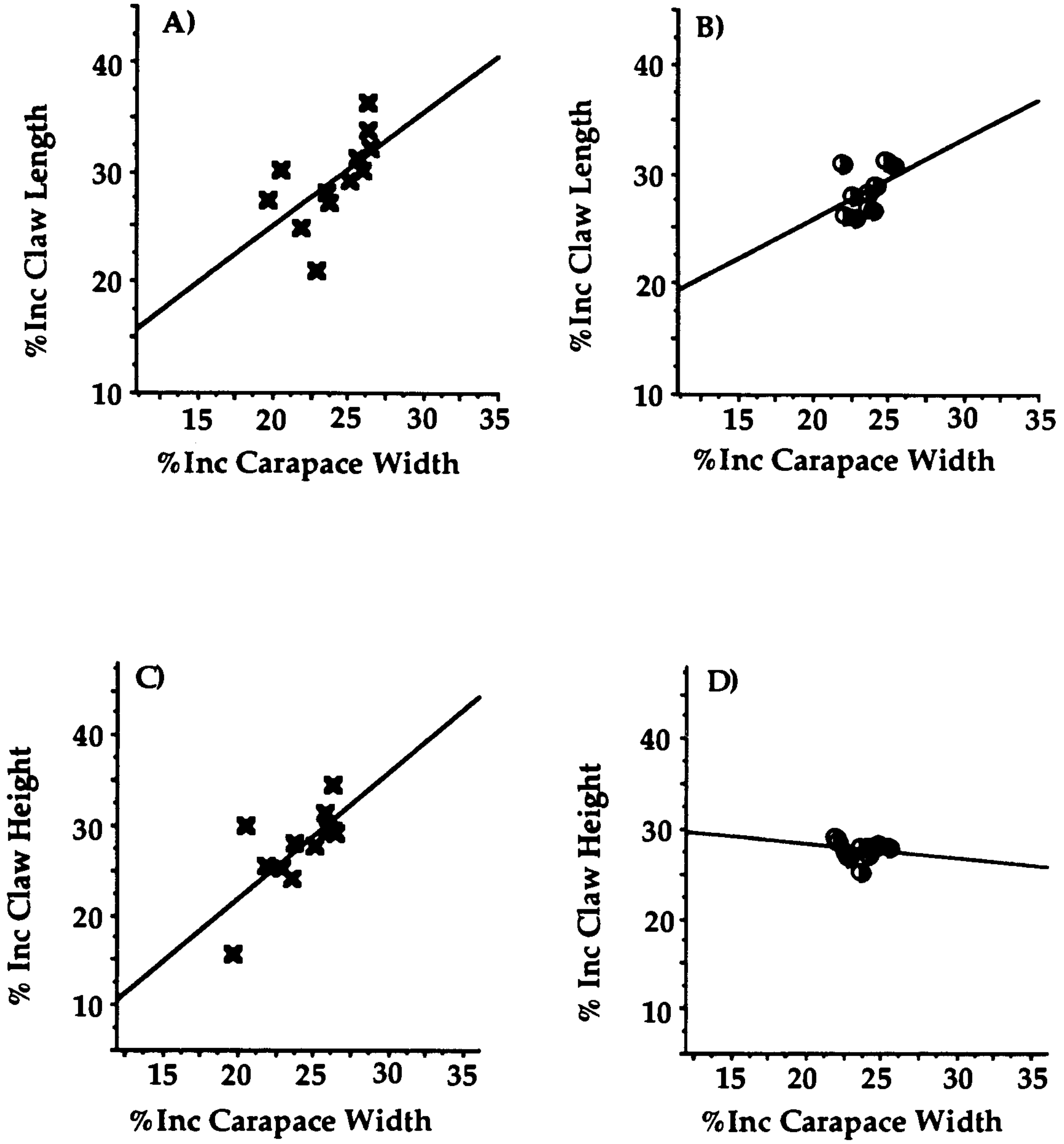

Figure 4.3. Regressions of the percentage increment in total claw length and height (i.e. the sum of the measurements of the two claws) on the percentage increment in carapace width for the third size increment in1991. A) Control: $y=1.05x + 4.09$, $r^2=0.37$. B) Predation: $y=0.73x + 11.43$, $r^2=0.16$. C) Control: $y=1.41x - 6.44$, $r^2=0.51$. D) Predation: $y=-0.17x + 31.64$, $r^2=0.029$. ANOVA between slopes and intercepts of regressions A and B: $p=0.7$ and $p=0.7$. ANOVA between slopes and intercepts of regressions C and D: $p=0.07$ and $p=0.06$. The following outliers have not been plotted or analysed: B (41.9, 72.8); D (41.9, 78.0).

Carapace hardening

For the three molts, there were no differences in the increases in weight between control and predation risk treatments for the first three days (Fig. 4.4).

Crabs increased in weight at each of the three molts, but there were no differences between treatments in weight either before or after molt (t-tests, $0.5>p>0.09$).

After the first molt, control crabs took 13 days on average to reach a stable weight; crabs under predation risk took significantly longer, 16 days on average (Fig. 4.5). After the second molt, crabs under predation risk also took longer to stabilize their weight compared to control crabs (12 days and 10 days, respectively), but this difference was not significant (Fig. 4.5). Since the measurements were not made over the entire molt cycle, the time to weight stabilization may have been underestimated. The length of time left for measurements after the third molt was not sufficient to determine weight stability.

The average of all the weights recorded for each crab after the first day when a crab reached a stable weight was used to estimate weight at stability. From this mean weight was subtracted the weight on the first day after molt in order to obtain a measure of weight increase. There were no differences between treatments in increase in weight for the first two molts (t-tests, $p=0.4$ in both cases).

Carapace strength

The carapace toughened substantially during the first twenty days following a molt, at which point it reached a plateau (Fig. 4.6). The gastric region reached higher values than the intestinal region. A total of only 22 crabs (from three treatments) were within the first 20 days postmolt when the measures were taken. It was therefore impossible to conduct meaningful

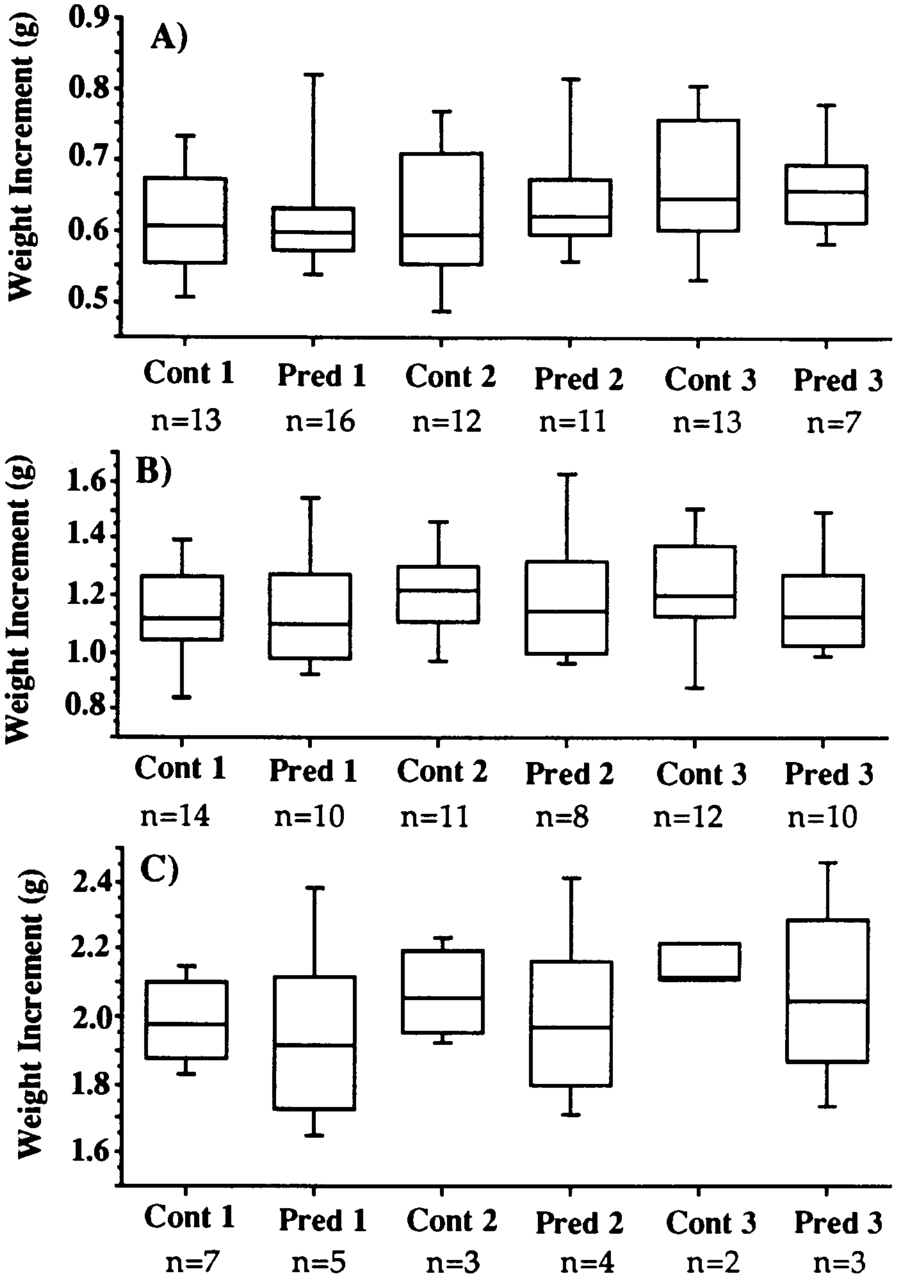

Figure 4.4. First three daily weight increments after molt (between day 1 and 4) for three molts, for control and predation treatments (10th, 25th, 50th, 75th, and 90th percentile). A) First molt: Inc 1, p=0.9; Inc 2, p=0.9; Inc 3, p=0.9. B) Second molt: Inc 1, p=0.9; Inc 2, p=0.9; Inc 3, p=0.54. C) Third molt: Inc 1, p=0.09; Inc 2, p=0.9, Inc 3, p=0.9. P-values are from unpaired t-tests with Bonferroni corrections, comparing control and predation treatments.

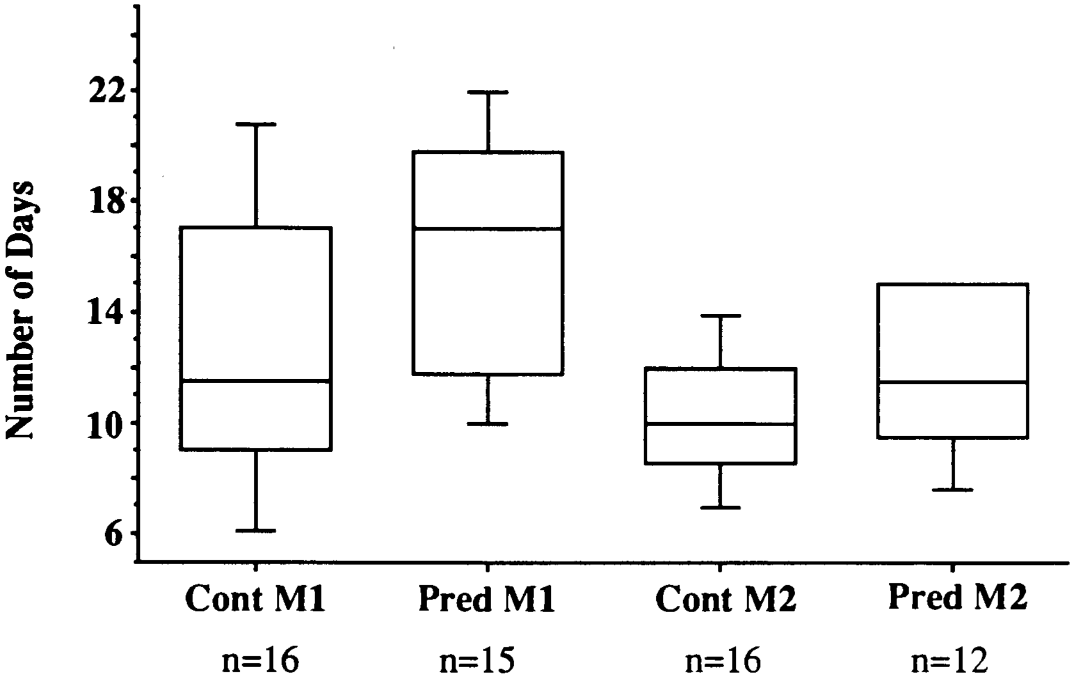

Figure 4.5. Distribution of the number of days crabs took to reach a stable weight, for control and predation treatments, for two successive molts. t-tests: M1, p=0.04; M2, p=0.06.

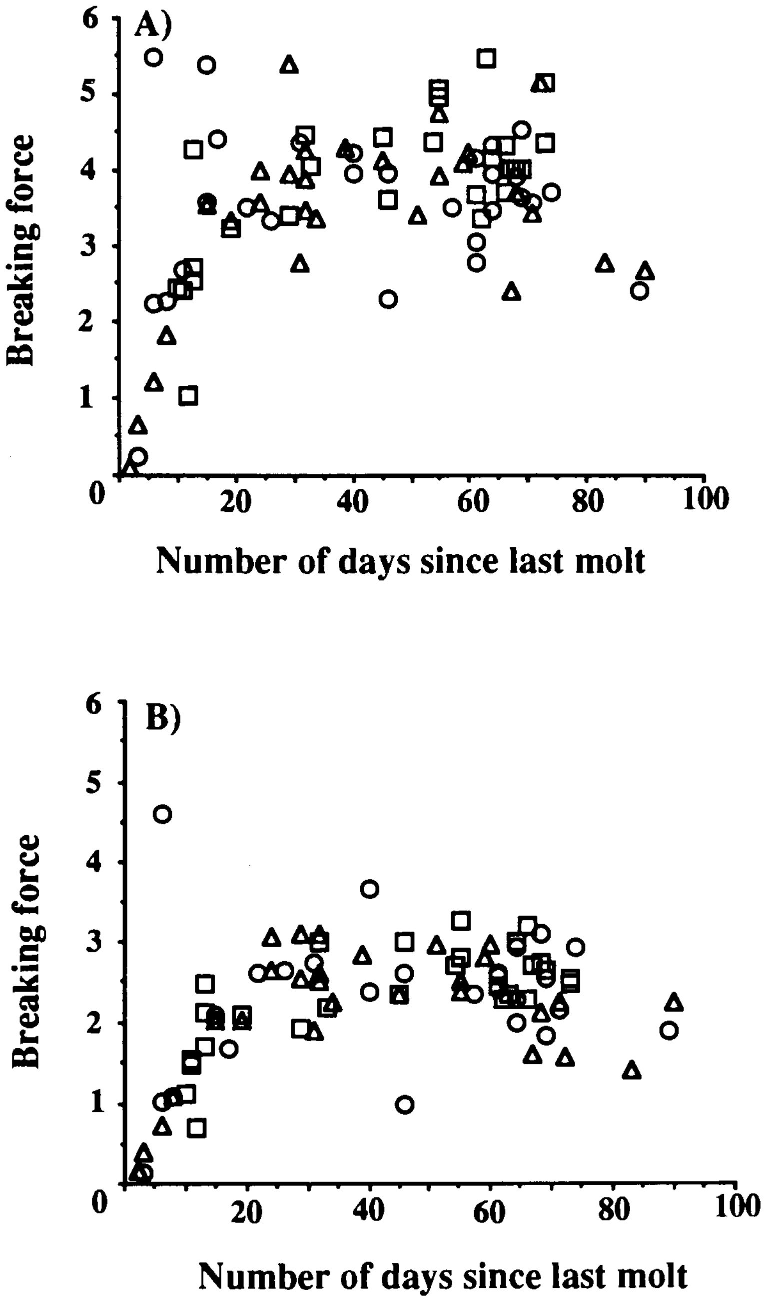

Figure 4.6. Relative force required to break the carapace of crabs from three different treatments, at different times postmolt. A) Gastric (front) region of the carapace. B) Intestinal (rear) region of the carapace.

Legend: △ Control
○ Predation risk 1
☐ Predation risk 2

analyses on the differences in toughness of the carapace between treatments for the first part of the cycle, but some interesting observations were nevertheless made. During the first two or three days following the molt, both posterior and anterior regions of the carapace seemed comparably soft; thereafter the anterior region became increasingly tougher than the posterior region in most cases, but less stongly for the predation risk 2 treatment crabs (Fig. 4.7). Even though the regressions for the control and predation risk 1 treatments were significant (i.e., breaking force increased with time), while the regression for the predation risk 2 treatment was not, there were no differences between treatments when the slopes were compared (ANOVA between regressions, control vs. predation risk 1: p=0.4; control vs. predation risk 2: p=0.3; predation risk 1 vs. predation risk 2: p=0.2).

Comparison of the toughness of the carapace 20 days or more after the molt (presumably, representing the end of stage C3 and stage C4) showed that crabs from predation risk treatment 2 had a tougher gastric region than crabs from the control, predation risk 1 and constant predation risk treatments (Fig. 4.8). The strength of the intestinal region did not differ between treatments, but was always less than the gastric region (paired t-tests, p=0.0001 for each treatment). The analysis of covariance was intended to correct for differences in size and days postmolt between crabs, but it revealed that these two factors had no influence on the toughness of the carapace, after 20 days postmolt.

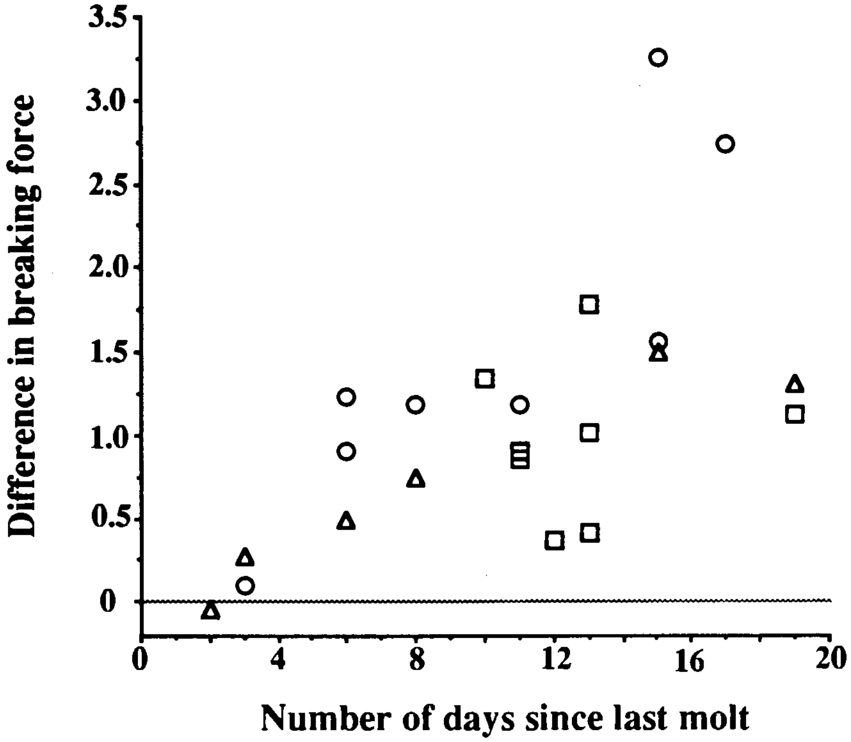

Figure 4.7. Difference between the gastric and the intestinal regions in the force required to break the carapace, over the first 20 days of the molt cycle, for three different treatments. Control: $y=0.08x - 0.02$, $r^2= 0.89$, $p=0.004$; Predation risk 1: $y=0.13x - 0.01$, $r^2=0.48$, $p=0.02$; Predation risk 2: $y=0.01 + 0.80$, $r^2=0.007$, $p=0.8$.

Legend: △ Control
○ Predation risk 1
□ Predation risk 2

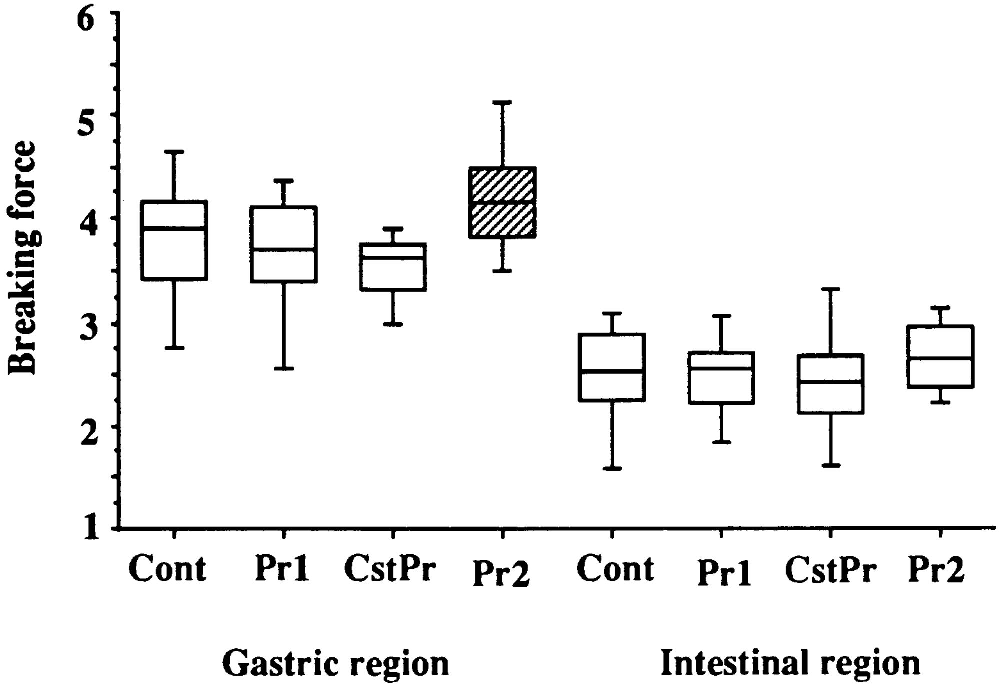

Figure 4.8. Distributions of measures of the toughness of the gastric and intestinal regions of the carapace for the control, predation risk 1, constant predation risk, and predation risk 2 treatments, for crabs 20 days postmolt or more. ANCOVAs, gastric region p=0.02; Pr1 vs Pr2, p=0.006; C vs Pr1, p=0.7; C vs Pr2, p=0.2; Pr1 vs Pr2, p=0.3, C vs CstPr, p=0.4; Pr2 vs CstPr, p=0.09.

Discussion

Investment in carapace and claws

Body parts can grow either isometrically (i.e., at the same rate) or allometrically (i.e., at different rates). Size allometry considers the patterns of variation and covariation of characters among individuals of a population during one stage of their life, and growth allometry compares the same patterns among ontogenetic stages (Hartnoll 1982, Klingenberg and Zimmermann 1992). Despite the fact that allometric growth is common (Gould 1971), it is still in many cases unclear whether the allometric parameters observed are the result of adaptations or constraints (Ebert 1988).

In crustaceans, allometric growth tends to be more marked with increased body size (Seed 1986). How differential investment in carapace and claws changes throughout animals' lives can often be correlated with particular events. For example, in several species, claws specialize for feeding. Both sexes develop heterochely, which is the differentiation of a crusher and a cutter claw (Hartnoll 1982, Seed 1986). This asymmetry occurs early in the lobster *Homarus americanus*, soon after the metamorphic molt (4th and 5th stages), and is stimulated by substrate and exercise (Govind and Pearce 1989). Claw strength is a function of the muscle in the propodus and the claw's lever system (Seed 1986), and is proportional to claw height, indicating that it may be advantageous to invest in large claws to exploit larger prey (Elner 1980).

Further allometric differentiation of claws in males corresponds to a change in reproductive status. Usually, the claws of males become proportionally larger and more powerful at the approach to and after maturity (Hartnoll 1982, Seed 1986, Clayton 1990), as they are used for courtship and territorial behavior (Hartnoll 1982, Homola et al. 1991). In some species, male claws are larger during the breeding season and smaller otherwise (Hartnoll 1982). Claw growth in females is more often isometric, but can also be positively allometric (Hartnoll 1982, Seed 1986).

Reproductive behaviors are not the only form of interaction between conspecifics. Social behaviors and communication may also play an important role in shaping claw development (Clayton 1990). A meral spread is an effective startle display and may make opponents flee (Berzins and Caldwell 1983). Other behaviors such as pushing, rubbing, embracing and extending the chelipeds are also important in displays (Clayton 1990, Karplus et al. 1992). Most aggressive acts between individuals involve the use of the claws (Karplus et al. 1992). Finney and Abele (1981) even suggest that selection to increase efficiency of agonistic displays can explain patterns of allometric growth in the xanthid crab *Trapezia ferruginea*. During conflicts, a wider, stronger claw can make it easier to crush an opponent's exoskeleton (Pillai 1990). For the same reason, strong claws could be important in defense against predators, and Weissburg (1991) has even suggested that larger chelipeds may have evolved to reduce predation risk.

The raptorial appendages of stomatopods are a good example of chelipeds which have specialized to become efficient in defence (Berzins and Caldwell 1983). *Homarus americanus* also defend themselves with their claws and successfully escape predators (Hudon 1987). Smallmouth bass consume crayfish in ascending order of chela size (Stein 1977). In *Uca pugilator*, an enlarged crusher claw seems to reduce vulnerability to predation by white ibises, since capture rates of intact males were four times lower compared to declawed males and females (Bildstein et al. 1989). Even more surprisingly, autotomized claws (simulated predation) in *Callinectes sapidus* grew back with larger teeth compared to the previous claws (Smith 1990), indicating a plastic investment in defence based on past experience. *Leptodius sanguineus* was also expected to invest in the production of larger claws under predation risk. Because claws serve so many different functions, and the production of larger claws is believed to be costly, increased claw size was expected to be a predator-induced defense, i. e., larger claws would only be produced when needed.

Mature female *Leptodius sanguineus* did not seem to invest in larger claws under predation risk. The relative investment made in carapace and claw was often equal. Because the magnitude of inducible defences may vary with an animal's age or size (Ebert 1988, Neill 1990), a greater investment in defence was expected at the third size increment: not only was there an increase in predation risk, but the crabs were larger as well. Both factors should have favored investment in defence, and larger crabs should have relatively more time and energy to invest in the production of bigger claws. However, contrary to the prediction, there was no significant effect, only a trend towards a greater investment in widening (but not lengthening) the claws.

These results suggest five hypotheses: 1) The measurements may have been too coarse and thus unable to detect an effect of the predation treatment. I kept the exuviae of each crab after the end of the experiments, to permit further analysis on carapace and claw size. Michelle Harrison (U. Alberta) is looking at these morphological changes through time using a computer program to digitize claw shape.. This technique might prove more fruitful, since claws can be measured at several places and more precise changes in shape can be detected. 2) If a trade-off exists between energy allocated to the claws and to the carapace, crabs may prefer to invest in carapace growth. A larger carapace may be more effective at avoiding predation, because it might be harder to break, and claws in *L. sanguineus* may be poor weapons. 3) If crabs prefer autotomy to fighting as a tactic, then it would be better to invest less energy in a limb which is likely to be lost. 4) The energy saved from the production of claws may be shifted to an investment in eggs under predation risk. 5) Functions such as food handling, burrowing or mate attraction may be the primary determinants of claw size and may not favor the morphological changes necessary for defensive purposes. This last point raises a crucial question: what are the selective pressures responsible for claw morphology and how do they interact?

Elner and Campbell (1981) proposed that the massive chelae of American lobsters may have evolved in response to selection related to sexual behavior, and that feeding and defense may be secondary functions. Weissburg (1991) suggested that ecological divergence may act in concert with sexual selection to explain claw size in fiddler crabs. For example, female *Uca pugnax* forage in safer habitats because they are more efficient at exploiting microalgae, since their claws are smaller and the number of setae on their maxillipeds is higher. Males, being inefficient and unable to compete successfully with females, forage in places where predation risk is higher, which may select for further increase in claw size (Weissburg 1991).

To be successful, allometry of body parts must balance energy intake and costs (Sebens 1987), within constraints (Hartnoll 1982). Trade-offs between conflicting functions would account for variations in relative size of body parts among species and also among individuals, according to which function must be favored. For example, chela propodus length in female *Cleistostoma kuwaitense* is usually isometric, but for ovigerous females there is a tendency for negative allometry (Clayton 1990). Energy may thus be shifted from the claws to reproductive output, in the same way as between claws and walking legs in other species (Hartnoll 1982, Ismail and Mykles 1992).

These shifts in investment between body parts suggest that the energy available to crustaceans is limited and bigger claws may be costly to produce if they need to be filled with tissue mass (= energy) which cannot be used elsewhere. At molt, crabs increase to a particular size, which will determine the allometric relationships between body parts for the rest of the molt cycle. During this intermolt period, environmental conditions may well change. It is therefore unclear what would determine a particular allometric relationship. Would it be triggered by environmental conditions before the molt, or at molt, and how well would a particular compromise do in a variable environment? What if predation risk subsequently increased or decreased? The investment made could either be insufficient or too great.

Should crabs favor an intermediate compromise or should they always produce larger claws? In other words should claw size be plastic or fixed?

Since the distribution of energy can change within the exoskeleton during a molt cycle, perhaps crabs should always produce large claws which could serve as energy reserves and be depleted according to other needs. Not only could this energy be used for reproduction, but since claw display can be an important predator deterrent (Stein 1977, Berzins and Caldwell 1983), the production of large claws, even if only partly filled with muscle, could conceivably bluff predators away. This may be the case for porcelain crabs, since they possess large claws and yet are filter feeders (Palmer, personal communication). However, producing large claws which may never be filled with muscles is probably selected against because a large empty claw may not be efficient for feeding or fighting. Claw size should therefore normally be an honest signal of claw strength, and determined by the trade-offs in allocation of energy and morphological features necessary to accomplish multiple functions. These requirements may deny the evolution of bigger claws as inducible defences in some species, and favor a fixed intermediate claw size.

Clearly, more studies are needed that attempt to identify for different species the selection pressures, constraints and trade-offs between feeding, reproduction and defense which dictate how energy is allocated between body parts, as well as the order of priority and the value of different combinations of investments throughout the animal's life in different situations.

Carapace hardening

The hardening of the exoskeleton is a complex process occurring over stages A, B, and C of the molt cycle (Table 1.1), and involves all layers of the cuticle. The cuticle is constructed of four layers, as described by Roer and Dillaman (1984), and O'Brien et al. (1991). The epicuticle is the thinnest, outermost layer, composed of tanned lipoprotein impregnated with calcium salts. The exocuticle, located under the epicuticle, is thicker and composed of chitin-protein hardened by quinone tanning and calcified with mineral crystals between the organic fibers. Below this lies the heavily calcified endocuticle, the thickest layer, made of chitin-protein but without tanning, and hardened solely by calcium salts ($CaCO_3$). Finally, the thin membraneous layer is made of chitin and protein, without minerals. Underneath the membranous layer is the hypoderm, which is responsible for cuticle formation, and is in close contact with the entire cuticle through numerous pore canals via which calcium travels.

The cuticle is continuously changing throughout almost the entire molt cycle, especially during pre- and postmolt. The beginning of premolt is marked by the degradation of the membranous layer, called apolysis; following this the new cuticle is secreted and tanned, and further degradation of the old exoskeleton takes place (Roer and Dillaman 1984, Fiber and Lutz 1985, Skinner 1985, O'Brien et al. 1991, Skinner et al. 1992). After molt (stage A), tanning continues and calcification begins with active transport of calcium by the hypodermis (Roer 1980, Mangum 1992).

The timing of calcification is often hard to determine (Greenaway 1985). Calcification of the epi- and exocuticle starts immediately after molt in small species like *Gammarus pulex,* half an hour after molt in *Heterocypris* (Greenaway 1985), one to four hours after molt in *Cancer pagurus* (Welinder 1975, Greenaway 1985), ten hours after molt in *Carcinus maenas* (Welinder 1975, Roer and Dillaman 1984), and not until the second day in *Panulirus argus* (Greenaway 1985). Usually, deposition of calcium, chitin and protein reaches a peak the second day after molt (Welinder 1975, Roer and Dillaman 1984). There is large variation in calcium deposition apparent among species and possibly among individuals. *Callinectes sapidus,* for example, responds to low calcium concentration by increasing the number of calcium transporting sites in the gills (Greenaway 1985). Crabs under predation risk might also increase calcium transport, resulting in faster hardening of the epi- and exocuticles. However, changes in hardening rate between treatments were not detected, at least by the crude technique used here.

Weight increase did not differ between treatments for the first three days after each of the three molts, suggesting that the rate of carapace hardening was similar. How calcium transport is controlled is still unknown (Greenaway 1985), but we know that movements of calcium are accomplished by Ca-ATPase and a Na/Ca exchange mechanism, and also depend upon the presence of HCO_3^- in the water (Roer and Dillaman 1984). In *Leptodius sanguineus* (or this population, at least), the pressure to harden as fast as possible early on (stage A) may always be high, and the pump may always work at full capacity. However, handling of the crabs (for weighing) may itself represent some sort of predation risk, and may have equalized the treatments (see Chapter 3).

Another possible avenue to promote faster hardening after molt could be to use calcium which had been reabsorbed and stored during premolt. Calcium resorption during premolt is necessary to permit exuviation (Greenaway 1985), but the amount of calcium reabsorbed varies among species: in *Gecarcinus lateralis, Palunirus argus,* and *Carcinus* 75%, 20%, and 15-20%, respectively of the mineral content is reabsorbed during premolt. This calcium can be stored in several organs, most often in gastroliths of the cardiac stomach and in the hepatopancreas (Greenaway 1985, Skinner 1985, Sardà et al. 1989). Marine crustaceans depend little on storage of calcium compared to terrestrial forms, since it is readily available in the water (Roer and Dillaman 1984). Consequently, the calcium is either excreted or left in the carapace and shed (Greenaway 1985). The resorption and accumulation of calcium reserves in highly calcified species may be energetically costly and time consuming, potentially resulting in longer premolt (Sardà et al. 1989), and limited by the space available for storage (Greenaway 1985). For these reasons, larger *Macrobrachium rosenbergii* reabsorb proportionally less calcium compared to smaller prawns (Brown et al. 1991). On the other hand, depending on the species' metabolism, stored calcium may be a valuable asset after molt since it may speed up the hardening process and even allow the preferential hardening of certain parts (Greenaway 1985). Storage may therefore be advantageous in the face of predation risk because it may promote faster hardening, if this process is faster than uptake from the water.

If crabs under predation risk had stored more calcium, they would presumably have been heavier than controls the first day after their molt, but this was not the case. Once again, the handling effect may have impaired the treatment effect. It would be interesting to test this hypothesis more adequately.

During stage B, the endocuticle is formed and mineralized as organic and inorganic materials are deposited at the same rate. In *Astacus fluviatilis*, deposition of calcium salts and chitin reaches a fairly constant level between days 4 and 14, and protein deposition decreases to almost zero after 12 days (Welinder 1975). Stage C3 may last up to 6 weeks (Welinder 1975), at the end of which the membranous layer is deposited and growth of the cuticle is completed; thickness and composition don't change thereafter (Welinder 1975, Roer and Dillaman 1984, Fiber and Lutz 1985, O'Brien et al. 1991, Skinner et al. 1992).

Crabs under predation risk took longer to stabilize their weight. It is tempting to conclude that they were building a tougher carapace, which takes longer to make, possibly due to a limitation in the rate of calcium deposition (Palmer 1981). If so, these crabs should have been heavier as well, but they weren't. Several explanations are possible. For example, the periods of weight measurement lasted about 30 days after the first molt, and 15 days after the second molt. Thus, the A stage was probably over in both cases, but the plateau observed may correspond to stage B or early stage C, and most of the growth of the endocuticle in late stage C was probably missed. To support this, consider that percent calcium is still rising after 14 days in *Astacus fluviatilis* (Welinder 1975), that the exoskeleton of *Macrobrachium rosenbergii* increases in dry weight from 19% body weight in stage A to 29. 8% in stage B, in about a week (Fiber and Lutz 1985), and that in *Cancer pagurus* and *Maia squinado* stage C3 itself can last 2-6 weeks (Welinder 1975). The weights of *Leptodius sanguineus* were taken for a maximum of 30 days, at which point crabs were shown to delay their molt (Chapter 3). Stage C3 may thus not have been completed when weighing stopped, and predation risk might have led to a more calcified, heavier exoskeleton later on in that stage.

Calcium is the major element responsible for the hardness of the crustacean carapace, since most of the proteins are similar to those which create soft exoskeletons in insects (O'Brien et al. 1991). But, the common assumption that calcium is the most important material for carapace toughness might be incorrect. Unbound proteins are believed to inhibit calcification and the cross-linking of proteins with chitin after molt creates the configuration needed to promote calcification (O'Brien et al. 1991). These proteins may be even more limiting than calcium, since they are costly to produce. This is the case in molluscs, where the organic matrix accounts for 50% of the cost of shell production, although comprising only 5% of the shell material (Palmer 1992). Also, many of the large proteins are shed at molt (O'Brien et al. 1991), which means they have to be resynthesized. In addition to their role in calcification, the matrix of proteins, along with tannins and chitin, may also offer resistance to breakage, whereas calcium alone would be brittle. Therefore, the combination of chitin-protein and calcium would determine the toughness of a carapace, and strengthening it further may require adding as much or more organic as inorganic material. The production of organic material would require more time, but would not lead to an immediate increase in weight. This might help explain why weight differences were not detected when measurements stopped.

Carapace strength

Inducible defenses can reduce vulnerability to predation, and thereby increase fitness (Riessen and Sprules 1990). Snails (*Nucella*) develops larger apertural teeth in the presence of predators, reducing vulnerability to shell-peeling predators (Appleton and Palmer 1988, Palmer 1990). Similarly, females*Leptodius sanguineus* produced a tougher (anterior) carapace when predation risk was raised. As was the case for molt delay (Chapter 3), the induction of a tougher carapace seems to respond to an increase in predation risk, as opposed to a threshold of risk, since the crabs under constant predation risk did not develop a tougher carapace although the crabs for which predation risk was raised (predation risk 2 treatment) did so. It also appears that different magnitudes of risk increase are necessary to trigger molt delay and carapace toughening. Molt delay was triggered by smaller risk increments, since crabs from the predation risk 1 treatment delayed their molt but did not invest in a tougher carapace.

This tougher carapace may increase the effectiveness of defence against predators, but may be costly to carry (if heavier) or costly to produce. In *N. lamellosa*, the deposition of calcium has an energetic cost and also limits the rate of growth because of constraints on the rate of calcification (Appleton and Palmer 1988). Therefore, mature animals are more likely to develop the defense because they grow more slowly than juveniles who may have to trade-off defense for growth in order to grow more rapidly (Palmer 1990). The situation is somewhat different with crustaceans since the carapace is shed at intervals, but if developing a tougher carapace lengthens postmolt, it could translate into a longer period of time between molts and slower growth. It may therefore be less costly for larger crabs to invest in a tougher carapace, since they molt less often.

However, carapace toughness did not vary with *L. sanguineus* size (between 13 and 23 mm carapace width). Since the carapace must be retoughened at each molt, toughness may not only vary among individuals, but within the same individual from one cycle to the next, independent of body size. The strength of the carapace could be controlled at each molt by deposition of more or less layers of material. Greater material deposition would mean that more time would have to be spent in postmolt. Consequently, by toughening the carapace crabs are trading-off growth with defence.

Which layer should be toughened and when?

The endocuticle may be the preferred area for carapace strengthening, which occurs during the C stage, especially in C3 when the endocuticle is known to thicken (Davis et al. 1973). Four pieces of evidence support this hypothesis: (1) The epi- and exocuticle are elaborated during premolt, which may limit their size or thickness after molt; (2) Strength may be determined by a combination of protein and calcium, which are more abundant in the endocuticle; (3) Weight did not differ between treatments for the first 15 to 30 days after molt; and (4) The frontal and lateral areas of the carapace are the last parts to mineralize (Lockwood 1967). However, because the epicuticle (believed to be thoroughly calcified after stage A) contains more than three times as much calcium-binding protein as the exocuticle and the endocuticle (O'Brien et al. 1991), an acceleration of protein formation under predation risk could promote faster hardening. In Xanthidae, the cuticle is thicker than in other families and possesses particular cone shaped regions between the exo- and endocuticle, with larger mineral spherules (Roer and Dillaman 1984). It would be interesting to investigate whether such structures play a role in increasing carapace toughness under predation risk.

The cuticle is believed to be completed at the end of C3 and to be stable in intermolt (C4), with the membranous layer impairing more calcium deposition (Roer and Dillaman 1984, Greenaway 1985). But because carapace repair is made during intermolt, when amorphous $CaCO_3$ is deposited with atypical tissue lamellae (Roer and Dillaman 1984), further investigation is required to determine if the carapace can be toughened during intermolt. The capacity to adjust carapace toughness throughout the entire molt cycle would be an advantage since it would allow the crabs to respond to short term variations in the level of predation risk.

Differential toughness of the exoskeleton

Different parts of the crustacean body are mineralized to different extents. For example, in *Astacus astacus* claws contain denser mineral matter than the rest of the exoskeleton (Huner et al. 1990); for other species, legs, claws or carapace may be more highly mineralized (Greenaway 1985). The carapace was not toughened homogeneously in *L. sanguineus*. The gastric region of the carapace was always tougher than the intestinal region, and the gastric region was toughened more under predation risk. However, the intestinal region of crabs in the predation risk 2 treatment may also have tended to toughen, since for the first 20 days after molt these crabs seemed to invest more equally in the two regions (Fig. 4.7).

Since the gastric region covers the stomach, the preferential toughening of this area was most likely not meant to protect essential internal organs (such as the heart), which are located in the center of the body. In semiterrestrial burrowing forms, the exoskeleton is also most heavily calcified at the anterior end, supposedly to resist abrasion during fossorial life (Greenaway 1985). In limpets, the thickness of the shell margin contributes more to shell strength than thickness in more apical regions, and is correlated with localized forces generated by crab predators (Lowell 1986). The anterior portion of a crab's carapace may also be such a critical region. *Leptodius sanguineus*, which live among coral rubble, usually sit with the posterior portion of their body lodged in the coral, leaving the anterior portion more exposed and therefore more likely to be grabbed by predators. Also, once captured, the anterior region may receive more stress than the posterior region during handling by a predator. These factors may explain why the toughening of the anterior region of the carapace takes priority.

Concluding Remarks

Larger claws do not appear to be an inducible defense in female *Leptodius sanguineus*. The role of the claws as weapons for defence against predators might be negligible compared to the role of the carapace, but this needs further investigation. Existing claw size favored by natural selection may already be effective in defense and unable to be improved. Defences are important when faced with a predator, but they are costly to produce. Morphological changes that favor particular defences may not be selected for if they impair other functions and consume energy required elsewhere. Trade-offs and priorities between functions of different body parts are likely to influence their growth and, within constraints, dictate allometric relationships. Therefore, relative claw size will be the result of compromises, and a plastic response to predation risk may thus be limited.

Contrary to the ambivalent interpretation of claw function, the role of the carapace as armor is well established. However, the protection offered by the carapace varies throughout the molt cycle. The strength of the cuticle decreases around molt, but after molt the carapace toughens. This toughening can be increased to enhance protection from predation, but more research is needed to understand how this inducible defence operates. Physiologists have discovered a great deal about the dynamic characteristics of the crustacean cuticle. This knowledge needs to be combined with behavioral responses to different environments (such as changes in predation risk), to investigate what controls the formation of the cuticle, its strength and flexibility throughout the molt cycle.

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Chapter 5

General Discussion

An animal's size will affect its energetics, life history, competitive ability, and susceptibility to predators (Sebens 1987). In turn, predation risk will affect the animal's behavioral decisions, influencing life history, and perhaps size, in a feedback loop. This is especially true for organisms with an exoskeleton. The exoskeletal armor is efficient at preventing predation. However, arthropods such as *L. sanguineus* who molt throughout their life only reach a temporary size refuge, periodically broken by periods of muscle breakdown, ecdysis, and re-hardening (Skinner 1985), rendering them vulnerable to all sizes of predators.

Indeed, crab molt stage and relative predator/prey size had a major impact on the outcome of an encounter. Below a predator/prey size ratio of 2.8, intermolt crabs resisted the predator from 1 to 105 minutes. For freshly molted crabs, predator/prey size ratio was irrelevant: most crabs were killed in under a minute of predator handling. Escape was possible (though, because trials were performed in aquaria without refuges, prey were ultimately always consumed) for relatively large prey (below a ratio of 2.6) and mostly during the intermolt stage. After molt, even relatively large prey could not escape. *Leptodius sanguineus* used their claws to protect their body once captured, and to fight with the predator, but did not use autotomy as an escape mechanism. After molt, claws were useless in defence. Since defences used during intermolt are ineffective around the time of molt, this period of transition increases prey vulnerability to predators. Consequently, for each molt cycle, *Leptodius sanguineus* must decide how much time and energy to invest in defences and which ones to use. There are several possibilities. Behavioral options such as reduction in activity and choice of a safe location in which to molt can help alleviate the increase in vulnerability in stages D through C, but more costly defences may also be necessary, such as a change in molt timing and size increment, or strengthening of the exoskeleton.

Under predation risk, crabs traded-off the benefit of being hidden with the cost of breaking crypsis. To avoid detection, *Leptodius sanguineus* buried more slowly in the sand, made more pauses while burying, and did not bury as deeply in the presence of the predatory crab *Thalamita crenata*. Reduced movement in the presence of the predator varied with molt stage: freshly molted crabs could still bury themselves, but buried more slowly, and made more pauses compared to intermolt crabs. The magnitude of the risk of predation also affected crab burial decisions. The greater the threat, the less crabs buried; at very high risk levels they remained motionless on the sand surface.

Males, especially large ones, molted in their shelter under predation risk, whereas control crabs molted in the open. Molting under cover may therefore be effective at avoiding predation during ecdysis. However, the tendency to molt in the open still begs an explanation. Molting in the open may be advantageous for females in search of a male, in which case they would be trading-off survival with the need for fertilization. However, we also need to investigate whether the process of exuviation is facilitated in the open, or if crabs choose to molt away from conspecifics who can also represent a threat.

Crabs also reacted to the risk of predation or cannibalism by delaying their molt. Following the predictions of a dynamic model (not presented in this thesis) and infering from the results, I suggest that a rule of thumb exists: delay if predation risk is greater than previously experienced; molt otherwise. In this case, molt delay would not be a response to a threshold risk level, but a response to an increase in predation risk, adjusted to the environment in which the animal lives. Inducible defences are believed to respond to lower and upper thresholds of risk to trigger their activation and determine the intensity of the response; I suggest that researchers consider the magnitude of increase in risk when interpreting their data. For *L. sanguineus,* this presupposes that crabs can track predator visitation rate.

L. sanguineus behaviors in the presence of a predator suggest the prey may have been assessing the predator's movements and position through chemical cues. But what do they really perceive in the wild? Can they determine the size of the predator and how many there are? Which cues are most reliable, and whether they affect molting differentially also deserve investigation. We also need to ask: How do crabs assess future conditions and resume molting? How long to delay may be a fixed response to any risk level or may vary with the perceived level of risk or the magnitude of the increase in risk. Molting may only resume when the risk of predation has decreased, but crabs may become habituated to predator presence, or their state may change through time, to the point where an increase in the benefits of molting allows ecdysis to proceed.

Because the strength of the exoskeleton varies from stage D1 to C3, vulnerability to predators should vary accordingly. Therefore vulnerability should be quantified for each stage of the molt cycle. This knowledge would help determine the probability of molt being delayed at different stages of the cycle. It is also appropriate to ask what cues are available to predators to detect the stage in the molt cycle of their prey. Is prey localization only possible through prey movement or can the predator be attracted by the odor emitted by a freshly molted prey? More research is needed to determine the cues available to predators from freshly molted arthropods, in order to further understand the risks of molting. Field observations of handling and giving up time in the complex natural habitat are also necessary. The effect of the stage of the molt cycle of the predator should be taken into account, since the strength of the predator's exoskeleton should play a role in its ability to subdue prey.

To understand the crustacean growth process, we need to know why and how molt increment varies (Hartnoll 1982). Size increment in crustaceans is not straightforward, since it is independent from tissue growth. In these experiments, energetic status (feeding rate) likely did not play a role in the decision to delay molting and did not affect size increment either. I hypothesize that size increment may be determined by the predicted availability of food in the future. Also, the expected predation risk may influence size increment, and this may be controlled before ecdysis, through the synthesis of a new cuticle of a desirable size. The next question becomes:

What is a desirable size increment? Most *L. sanguineus* had a similar size increment regardless of the treatment. *Leptodius sanguineus* did not compensate for losses of growth opportunity by a larger size increment at the next molt following a delayed molt. On the contrary, some crabs increased less in size under predation risk. Reduced size increment may be a result of a stress-induced increase in metabolic rate, but may also be an adaptation to molting under predation risk. This could be true, for example, if a smaller exoskeleton expands and hardens faster, or if a smaller size increase allows the crab to regain maneuverability sooner since less muscle tissue needs to be formed. In either case, the animal would regain full usage of its defences more rapidly, reaching stage C4 faster. On the other hand, a smaller size increment could be the consequence of a decision to shorten the last stages of proecdysis (after a delay or not), so less cuticle material would have been synthesized. Different strategies need to be considered, evaluated and tested. Compensation in size increment could occur later on, after the risk of predation has decreased, and should take the least costly path between molting often and increasing in size more at each molt. All these ideas require future testing. I also suggest that under predation risk crabs might be induced to produce larger claws, and that this would have to take place at the expense of an investment in carapace increment.

Weissburg (1991) proposed that larger chelipeds may have evolved to reduce predation risk, and claws are used in display and defense against predators in several species (Hudon 1987, Bildstein et al. 1989, Weissburg 1991). However, mature female *Leptodius* did not invest in larger claws under predation risk. Despite their role in defence, optimal claw size may be a compromise between many conflicting functions which would not favor the additional morphological changes necessary for defensive purposes. As well, a modification in claw shape must be initiated before the crab encounters future living conditions (size of body parts may be determined when the cuticle is formed in premolt); this may limit the usefulness of any extreme adjustment. The selective pressures responsible for claw morphology and their interactions must be investigated further. For *Leptodius sanguineus* facing a crab predator, which begins handling crabs by removing their claws, the strength of the carapace is likely to be the most important defence.

Indeed, *Leptodius sanguineus* produced a tougher (anterior) carapace when predation risk increased in a particular way. I suggest that crabs control their carapace toughness, for every molt cycle independently, by spending more or less time in postmolt (stages A through C3), depositing layers of material, thereby trading-off growth with defence. The fortification of this region as opposed to others may be prioritized because crabs are more likely to be grabbed by predators in the anterior region and this area may also receive more stress during handling by a predator. But more study is needed to observe differential investment in different regions of the carapace and the limbs as well. Also, investment at different stages of the molt cycle should be examined. For example, there may be a tendency for crabs under predation risk to invest in the posterior region of the carapace more than usual during the first few days after molt. Therefore, early emphasis on hardening may be advantageous, and storage of calcium before the molt may promote faster hardening. However, changes in hardening rate between treatments were not detected. It is possible that the pressure to harden as fast as possible early on (stage A) is always high and maximized, but I believe that the handling imposed on the crabs for this experiment equalized the treatments and prevented any significant differences from being detected. Questions related to hardening therefore still require adequate testing, and many of the ideas for future research proposed so far should be conducted concurrently with physiological studies.

Lipcius and Herrnkind (1982) stated that despite the bulk of studies on physiology on the one hand, and predation risk on the other, there is still a crucial need for quantitative studies that relate physiology and behavior in order to identify ecologically adaptive features. A decade later, a great deal of progress has been made in understanding hormonal and molecular aspects of crustacean molting, but there is still a need to tie in behavior and investigate such questions as what triggers molting. Hopkins (1992) suggests that external and internal cues should act together to cause molting in the best possible conditions, and that one external cue such as daylength may be responsible for the decrease in molt inhibiting hormone allowing proecdysis to proceed. I believe that features of the biotic environment may be at least as influential in stimulating or inhibiting the progress of stages of the molt cycle (especially in the tropics where molt is not tightly tied to the seasons). I also propose that environmental factors could play a role right up to the time of ecdysis. We need to investigate exactly when molting becomes irreversible, and what minimum amount of time is required to prepare for the molt at each stage of proecdysis. I believe that molting or delaying are under fine and constant control. The hormonal pathway could be examined by looking at the effect of predation risk on the production of molt inhibiting hormones and ecdysteroids.

Similarly, physiological studies on the strengthening of the exoskeleton after molt will help understand how this inducible defence operates. It is likely that the endocuticle is the preferential area for carapace strengthening, and that this occurs especially during stage C3, but other possibilities need to be investigated. If the carapace could be toughened during intermolt, this would allow crabs to respond to short term variations in the level of predation risk. We need to test what makes a carapace strong, what controls cuticle formation, and how flexible it can become throughout the molt cycle.

At this point, one of the major conclusions of this thesis can be drawn: molt timing, investment in carapace strength, and possibly size increment are plastic responses to environmental change. In fact, predator-induced defences are among the adaptive reaction norms that have been identified (Stearns 1989). Inducible defences may be quantitative characters under the control of many genes because there are several components to the response (Harvell 1990). Phenotypic plasticity is believed to be selected if fitness averaged over different environments is greater than the fitness associated with a non-plastic response (Thompson 1991). Via (1993) believes the process of evolution toward different optimum phenotypes in each environment produces phenotypic plasticity, but others believe there are genes for plasticity. The genetic basis for plasticity and the object of selection are therefore controversial issues (Scheiner 1993). The proximate cause of molt plasticity is unknown.

A question that must be addressed, is the influence of different genotypes and their reaction norms on molt timing. For example, Harvell (1990) showed that each bryozoan has its own threshold and amplification trajectory of a response to an increased risk of predation. Such differences in plasticity between genotypes could be responsible for different lengths of molt delay for example. On the other hand, one could argue that the variability could be explained because the crabs all had different previous experience with predators and thus the magnitude of the increase in risk was different for each individual, resulting in a different response. The trait undergoing selection could therefore be the amount of increase in the environmental inducer needed to elicit a response. More research is obviously needed to understand the mechanisms and consequences of molt plasticity in crustaceans, and in arthropods in general.

Evidence presented in this thesis suggest that *Leptodius sanguineus* can control the timing of their molt, the location of their molt, strength of their exoskeleton, and perhaps their size increment in order to decrease the risk of being preyed upon, thereby trading-off growth with survival. Ultimately, phenotypic plasticity in events related to molting and defences that increase survival is adaptive, since the criterion to identify adaptation is a change in phenotype that occurs in response to a specific environmental signal (Stearns 1989). However, to conclude this with certainty, the relationship must prove to result in an improvement in growth, survival or reproduction (Stearns 1989). Therefore, the most important next step is to understand and measure the fitness consequences of molt plasticity. This must be done by comparing lifetime reproductive output and growth rate for plastic and non-plastic individuals (Harvell 1990), and in different environments (Via 1993). Avoiding predators is important and must often be prioritized, otherwise fitness could equal zero. However, any investment in defence is expected to be costly.

For example, reduced activity will have lost opportunity costs, since other activities cannot be performed. Both delaying molt and producing a tougher carapace also have lost opportunity costs since they reduce growth rate and may delay reproduction. Also a female who delays molting may lose opportunities for mating and fertilization if she must be freshly molted to mate. A stronger carapace may be more costly to carry (if heavier) or more costly to produce. A change in the allometric relationship between body parts that increases defence ability may also reduce the ability to perform other activities. Behavioral decisions made under the risk of predation affect growth and have a direct effect on life history, potentially affecting fitness through the consequences for reproduction.

When survival is uncertain, reproduction dominates growth and sexual maturity occurs at an earlier age (Stearns 1976, Horn and Rubenstein 1984, Skinner 1985, Lessells 1991). This may be the case for *L. sanguineus,* since females less than 7 mm in carapace width were found to carry eggs. Since larger females produce larger egg masses (Robinson and Doyle 1985, Havens and McConaugha 1990, Hines 1992), crabs that reproduce early may be constrained by their vulnerability (low survival probability through molt) to adopt a r-strategy but, because of their small size, suffer a lower reproductive output. Investment in inducible defences has been shown to have negative consequences in several species (see Harvell 1990 for a review). For example, in *Daphnia pulex,* spine production reduces growth (Riessen and Sprules 1990), and lowers reproductive rate because animals take longer to mature (Dodson 1989). When the barnacle *Chthamalus anisopoma* adopts a bent-over form that is resistant to predation, its clutch size is reduced by up to 50% (Lively 1986). However, in the case of organisms molting throughout their lives, the investment in defences are not permanent. Adjustments in allometry, hardening and carapace strength may be done and undone at each molt cycle. If growth compensation also occurs after predators have gone, then growth may only be temporarily affected. Therefore, costs and negative effects on fitness may be compensated for, or at least not compounded. However, cuticle formation or expansion must be constrained within some upper limit. If local patterns of predation pressure or other environmental condition are such that recovery of growth opportunities lost while delaying and producing other defences is not possible, some fitness cost may result. It would be interesting to quantify how different delay times, investment in morphological defences and size increments would translate into differential fitness for animals of different sizes. Larger crabs molt less often and increase less in size at molt. This may be because they require more time to build up tissue mass, as volume: surface ratio increases, or because of mechanical constraints. Alternatively, because the risk of molting increases with body size increase, larger crabs may make a greater investment of time and energy in defences, delaying molt longer, increasing less in size to promote faster hardening, and producing a tougher carapace.

The cost of molting may also be responsible for the evolution of different patterns of life history in different species. For species in which mortality risk is highest for small individuals, molting faster under predation risk could be selected to reduce time spent at smaller stages. Differences in vulnerability and survival through the molt period may have selected for a variety of different strategies, including species which molt and reproduce throughout their life, and these which mature at a true terminal ecdysis (where the y-organs degenerate) after growth is completed. Selection for plasticity in molt timing may be less likely for species that molt only once (or a few times) a year, since delaying may be too costly in terms of fitness. Other strategies such as molt synchrony may represent a better solution for these species.

This study shows that behavioral decisions about molting may have profound effects on life history. The consequences of these effects can extend to the population level. For example, investment in defences and early reproduction under predation risk may influence the size structure and sex ratio of a population if decisions are specific to a particular sex and sizes. Obviously, delayed individual growth in some habitats may lower a population's growth and reproductive rates. Intrinsic growth rate of the population may be overestimated by attributing too high a reproductive value to organisms who are older than they appear. Because the relationships between age, size, intermolt length and size increment are inconsistent, we have to be careful in setting the parameters of crustacean population models. Predator behavior and prey responses to predation risk should be included in models of population dynamics, since anti-predator behavior may sometimes be more important than predation events *per se* in influencing population dynamics of both predator and prey.

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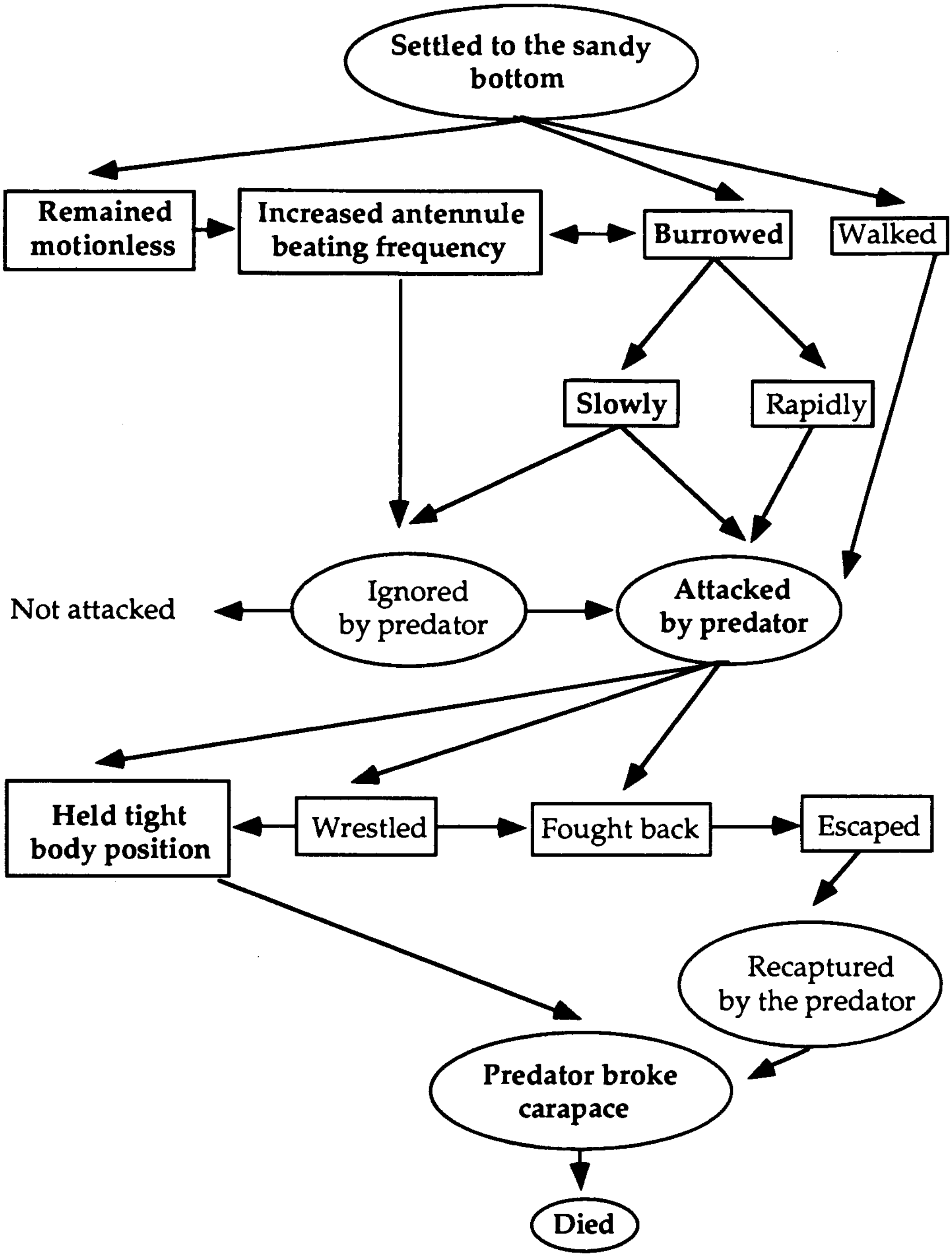
Appendix 1

Predator-prey interactions observed between *Thalamita crenata* and *Leptodius sanguineus*

Figure A1.1. The prey's behavior .

Leptodius prey were introduced into the predator's aquarium: once deposited on the water surface, they slowly settled onto the sand. On the sand surface, three behaviors could be observed : (1) the prey remained motionless; (2) the prey started burrowing; or (3) the prey walked on the sand. The first behavior was the most frequent. While the animal remained motionless, the beating frequency of the antennules (flicking rate) increased. Burrowing sometimes started as the prey touched the sand, but usually occurred after a period of immobility which lasted from a few seconds to over 15 minutes. When the prey burrowed, the antennules continued to beat rapidly. Burrowing was usually slow, involving leg movements of small amplitude, and interrupted with several pauses. The body remained horizontal. In some cases, prey walked away from the place where they landed and stopped moving after a few steps or when they reached the side of the aquarium. The predator by this point may or may not have located the prey. In some cases the prey was ignored. Usually the prey was attacked. More attacks occurred when the prey was moving, either digging or walking.

Once captured, crabs tried to wrestle out of the predator's grasp by using their claws and legs to push on the predator's claws and mouthparts. On two occasions, both opponents engaged in a spread-grasp action pattern. This pattern has been described in *Hemigrapsus nudus* (Jacoby 1981) when conspecifics spread their claws and grasp each other's chelae. These actions lasted 27 and 65 s. After wrestling and fighting back, escape sometimes occurred, but prey were all recaptured after they had escaped. In most cases, prey handled by a predator held a tight body position: the legs were folded onto the abdomen with the claws covering the mouth parts and part of the abdomen. Prey death was recorded when the carapace was broken by the predator.



Legend : Bold statements indicate most frequent actions.

Actions not under prey's control

Prey's actions

Figure A1.2. The predator's behavior.

Before prey introduction into the aquarium, the predator was usually resting on the sand surface, or partly buried. In some cases, the prey was ignored. In most cases, the predator detected the prey during it's descent through the water column: the predator either waited before attacking, remaining oriented towards the prey, or attacked promptly. In other cases, detection is assumed to have been chemical, since the predator increased the beating frequency of it's antennules, then raised it's body high on legs that were closer together than usual (stalking; see Lawton 1989) and started walking around the aquarium, apparently randomly. In some instances, the predator even walked over a half-buried prey without detecting it. The predator eventually concentrated it's searching in the area where the prey was located.

Most attacks were made after the prey had moved. The prey were either captured with the claws, the predator extending both claws to grab the prey with it's chelae closing at the right moment, or with the legs (pouncing; see Lawton 1989). In the latter case, the predator stood over the prey, surrounding it with the walking legs. The legs were used to dig up the prey and trap it into a net formed by the legs. If the prey was caught with the legs, it was moved forward and transferred to the claws for further handling.

In most cases, the predator handled the prey without interruption until consumption was over, and remained immobile, but, in some instances, the predator walked around the aquarium while handling the prey. At such times, the predator was not stalking, and walked normally with the legs further apart than previously described and consequently with the body closer to the sand. Sometimes predators stopped handling the prey crab for short periods of time, remaining immobile for no apparent reason, and resumed feeding. Pauses were also made for walking, pivoting, and cleaning mouthparts. In some cases, the predator made meral spreads (Jacoby 1981) and even buried into the sand while still holding a prey (alive or dead) and did not complete consumption during the observation period. When consumption of the prey was completed, the predator raised it's body high on it's legs and made a series of up and down flexions. The predator then groomed it's mouth parts and antennules before going back to rest.

Legend : Bold statements indicate most frequent actions

Predator's actions Prey's action

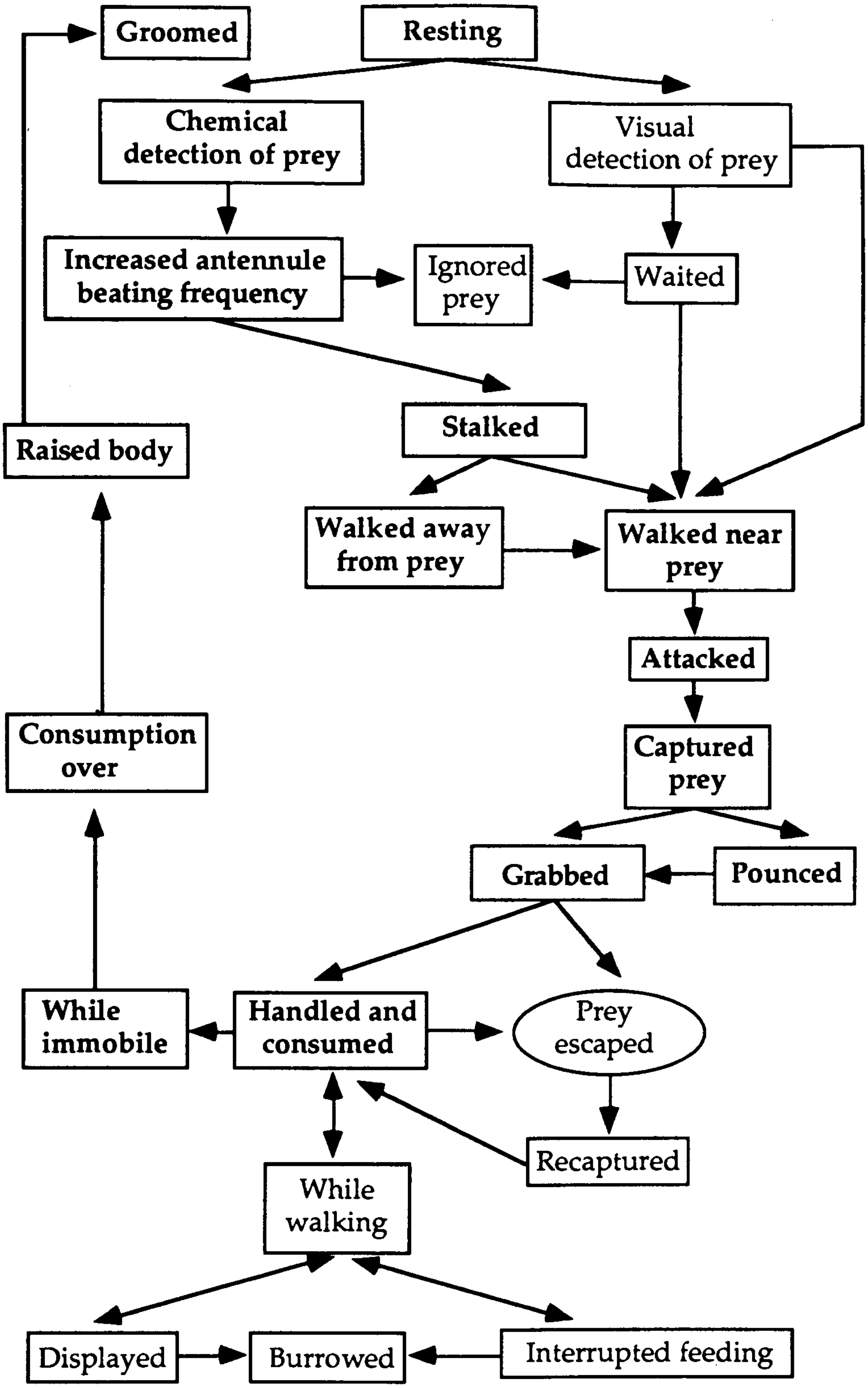

Groomed
Resting
Chemical detection of prey
Visual detection of prey
Increased antennule beating frequency
Ignored prey
Waited
Stalked
Raised body
Walked away from prey
Walked near prey
Attacked
Consumption over
Captured prey
Grabbed
Pounced
While immobile
Handled and consumed
Prey escaped
Recaptured
While walking
Displayed
Burrowed
Interrupted feeding

Figure A1.3. What happened in the "handle and consume" box: details on prey handling .

Once the predator was holding the prey, the first thing it attempted to do was to remove the prey's claws. Often, the predator would remove one or two claws and proceed to eat them before further handling of the live prey. On other occasions, the claws were dropped on the sand and the predator continued handling the prey and consumed the claws later, either while feeding on the body mass or after consumption of the body was complete. When the prey were freshly molted, claws were removed very easily and rapidly, but were dropped on the sand and always remained untouched. On some occasions, especially with smaller predators, legs were removed and eaten or dropped on the sand. After removing the claws, predators proceeded to handle the crab itself.

There were several ways in which the predators subdued the prey. When the predator was large, or the prey was soft, the prey was rapidly torn into two pieces. Usually, the prey was rolled around in the predator's claws until the predator managed to break into the prey. It may have done so by cracking the carapace with the crusher claw, by loosening the connection between the carapace and the abdomen, or by using it's mouth parts to patiently saw into the exoskeleton, creating a hole. Both claws and mouth parts were used to kill and manipulate the prey.

Once the body was consumed, the predator searched in the sand, and ate any edible parts that were left over. When the predator was through eating, only a few little bits of prey exoskeleton remained. Often, the frontal segment of the carapace of intermolt prey was left intact, while the rest of the exoskeleton was consumed.

Legend

☐ using claws

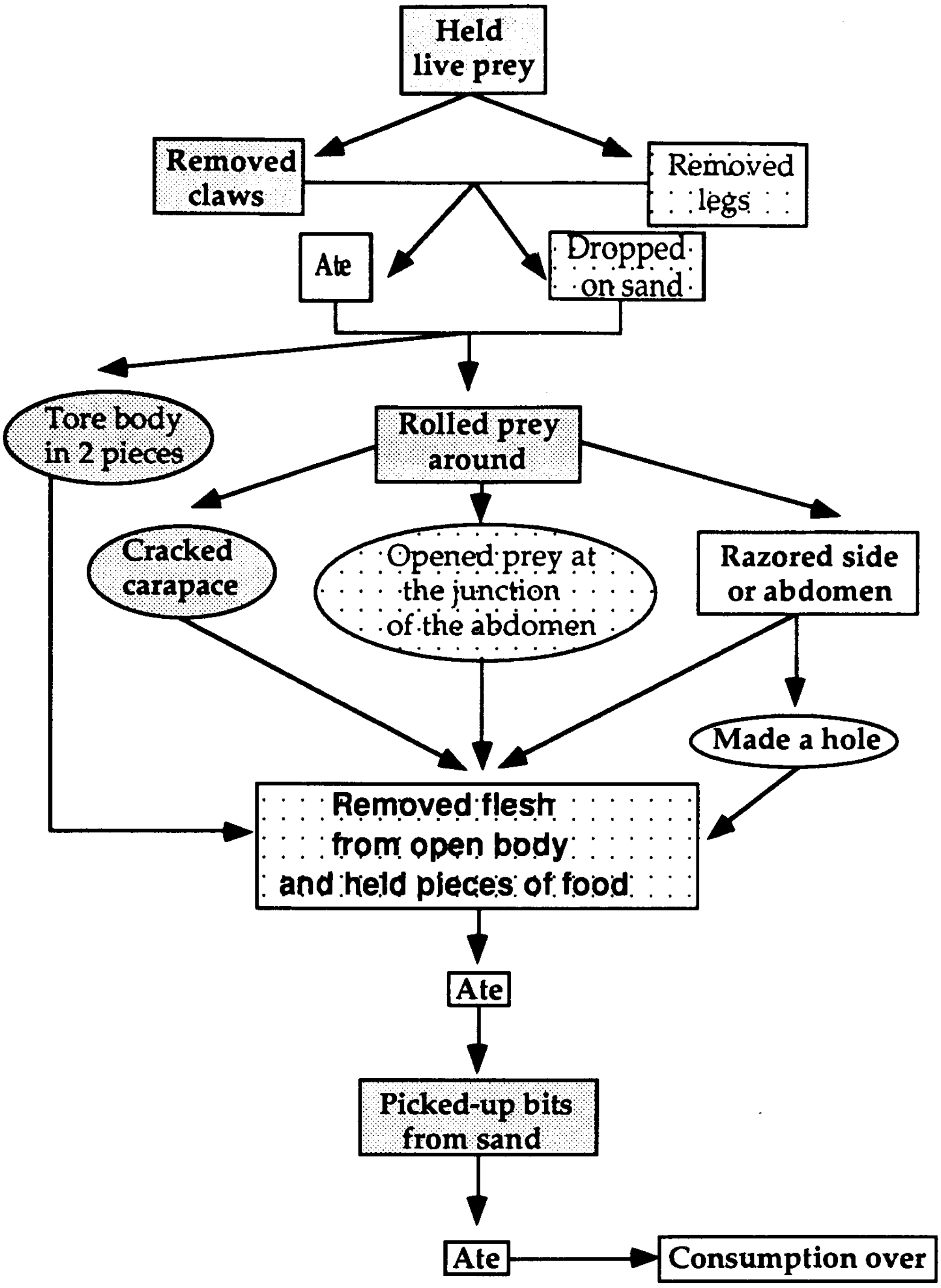
☐ using mouth parts

░ using both

Bold statements indicate most frequent actions.

Actions

Marks prey death


Held live prey
Removed claws
Removed legs
Ate
Dropped on sand
Tore body in 2 pieces
Rolled prey around
Cracked carapace
Opened prey at the junction of the abdomen
Razored side or abdomen
Made a hole
Removed flesh from open body and held pieces of food
Ate
Picked-up bits from sand
Ate
Consumption over

Literature cited

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Appendix 2

Predator Category, Body Size and Claw Size

To simplify analysis, predators were placed into two categories. To establish the members of each predator category, body size and claw size were considered. Body size and claw size are not always correlated in crabs: large crabs can be found with one or two small claws. This situation arises when a claw has been lost or autotomized in the past and the regenerated claw has not yet reached normal size, which may require up to three molts (Skinner 1985, Smith 1990). Even during normal growth, *Thalamita crenata*'s claws are asymmetrical. The crusher claw is normally larger and more powerful than the cutter claw, possesses molar-like teeth, and is used to apply compressive force to crush hard-shelled prey (Seed 1986). The cutter claw is used for cutting or shearing the flesh of the prey (Seed 1986). Because claws appear to be more important in subduing prey than mouth parts (personal observation), predator size categories were based on the size of the crusher claw (Table 2.1), but to comply with convention, the results are presented in terms of body size (carapace width).

The ratio between predator and prey carapace width could not be used directly to express predator threat for predator #1, because this large predator had a small crusher claw (Table 2.1). Crusher claw size has been shown to correlate better than body size with mussel size eaten (Elner 1980). To establish the predator/prey size ratio, this predator was given an effective carapace width of 35.2 mm. This value was derived from comparison with the claw proportions of the smaller predators. The ratio of the predator crusher claw size compared to the prey's carapace size was also used to categorize encounters. When plotted against time to prey death, the ratio of predator crusher claw size to prey size (Fig. A2.1) gives the same relationship as when using predator and prey body size, with the correction made for predator #1 (Fig. 2.9). This result validates the correction. Because both claws and mouthparts are used to subdue prey, both claw size and body size should be examined to evaluate an individual predator's threat to prey.

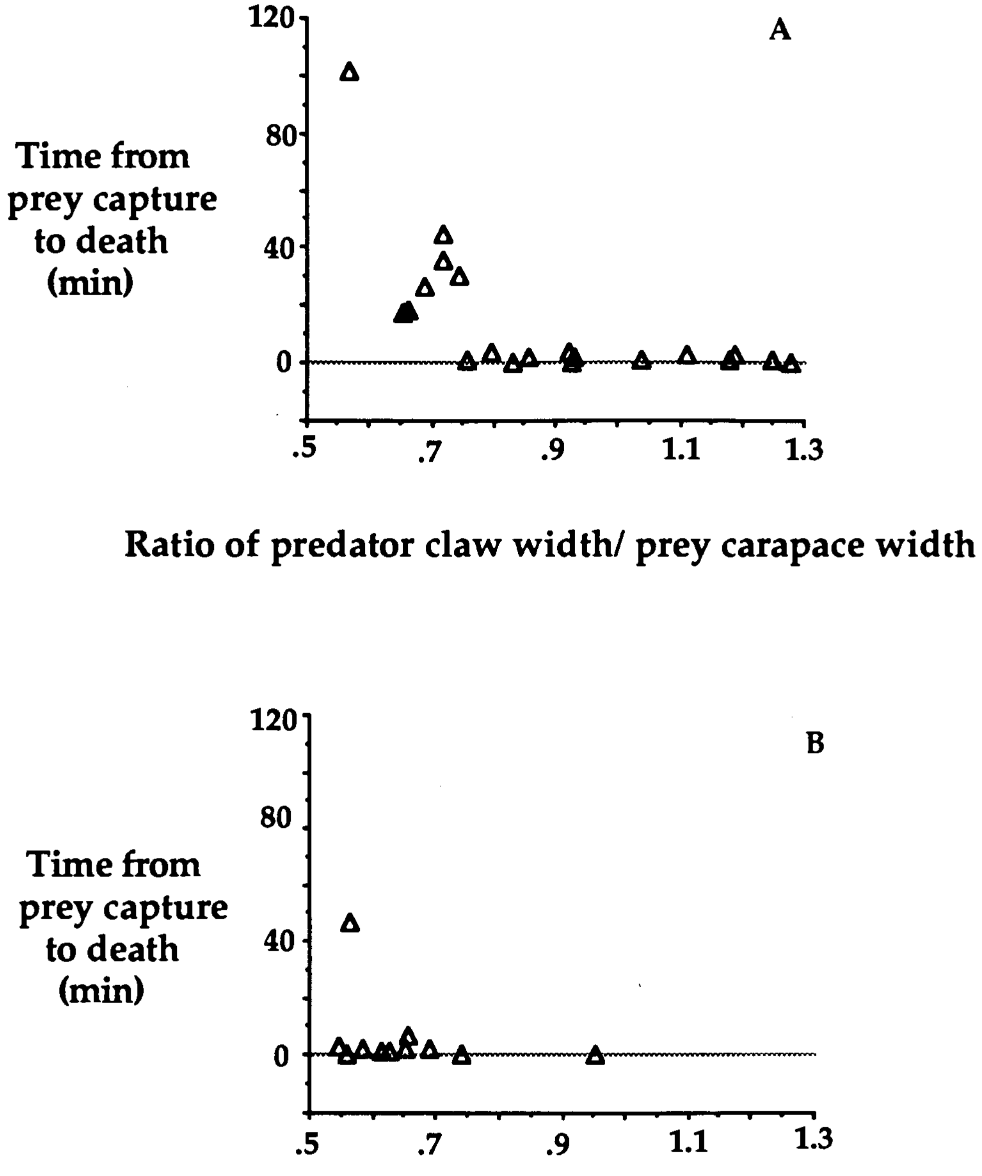

Figure A2.1. Influence of the ratio of predator crusher claw size to prey carapace width on predator handling time. A. Prey at the intermolt stage. B. Prey that had freshly molted.

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Appendix 3

Relationships between size, size increment and intermolt length

The size of a crab was a good predictor of the time spent in intermolt for the control and the predation risk 1 and 2 treatments: as the crab got larger, the length of the intermolt period increased (Fig. A3.1). There was no such relationship for the crabs under constant predation risk or the handled crabs. Carapace width was a good predictor of the next size increment for the crabs from the control treatment: as size increased the size increment decreased (Fig. A3.2). The relationship was weaker (p values lower, but still significant) for the predation risk 2 and the constant predation risk treatments. There was no relationship for the crabs from the predation risk 1 treatment or the handled crabs. The time spent in intermolt was a poor predictor of the subsequent size increment, except for the handled crabs, who increased more in size after spending more time in intermolt (Fig. A3.3). The slopes for all other treatments tended to be negative.

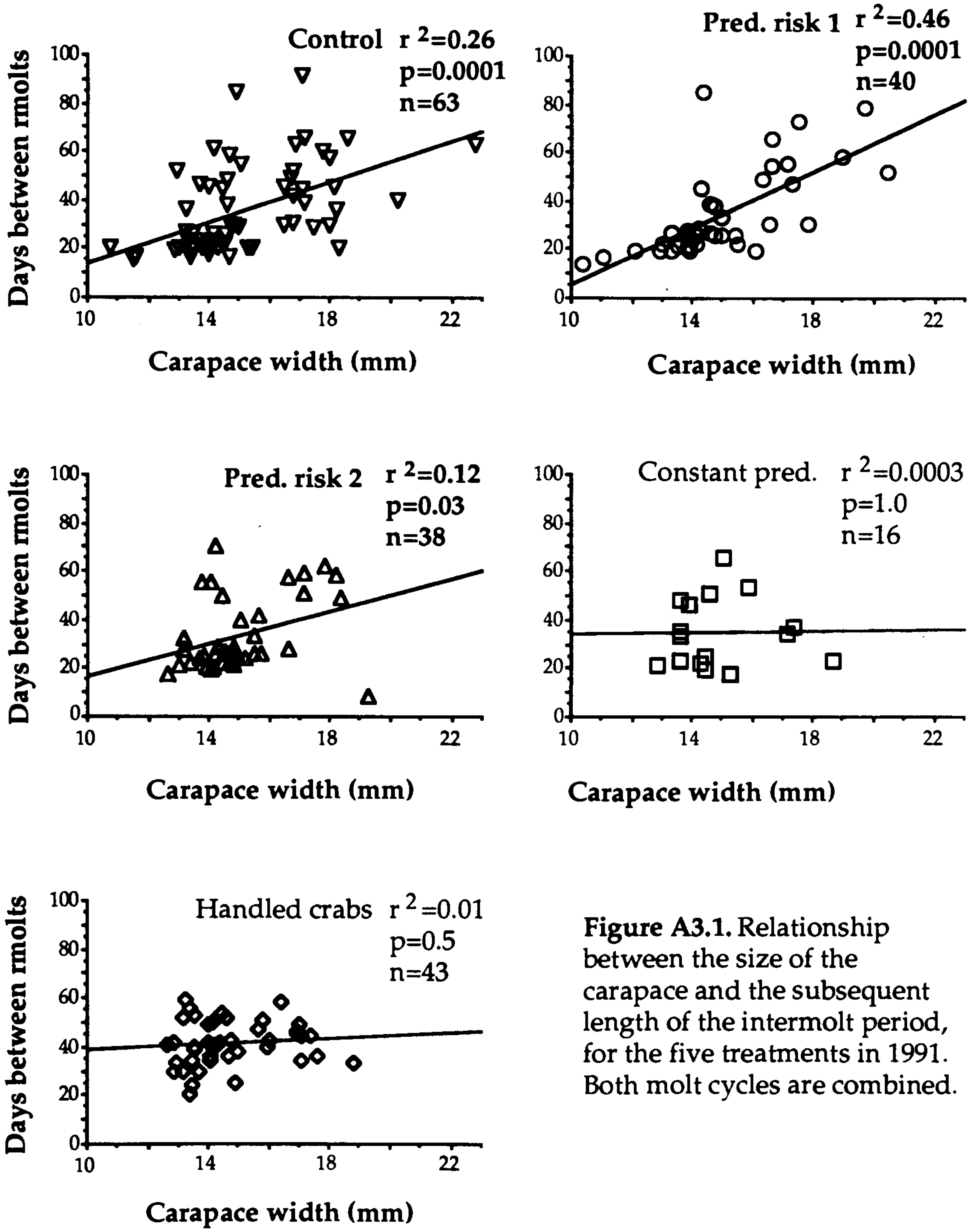

Figure A3.1. Relationship between the size of the carapace and the subsequent length of the intermolt period, for the five treatments in 1991. Both molt cycles are combined.

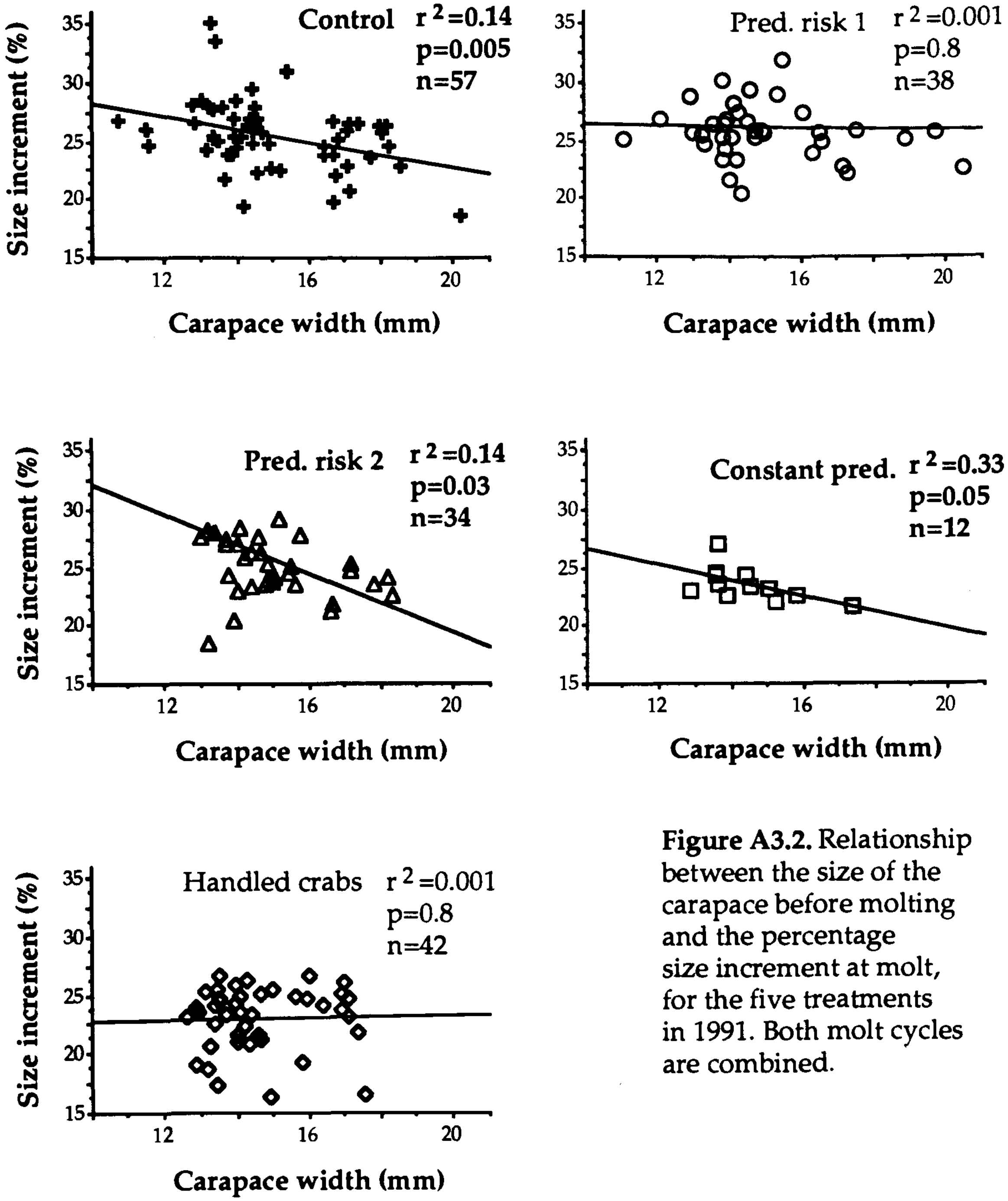

Figure A3.2. Relationship between the size of the carapace before molting and the percentage size increment at molt, for the five treatments in 1991. Both molt cycles are combined.

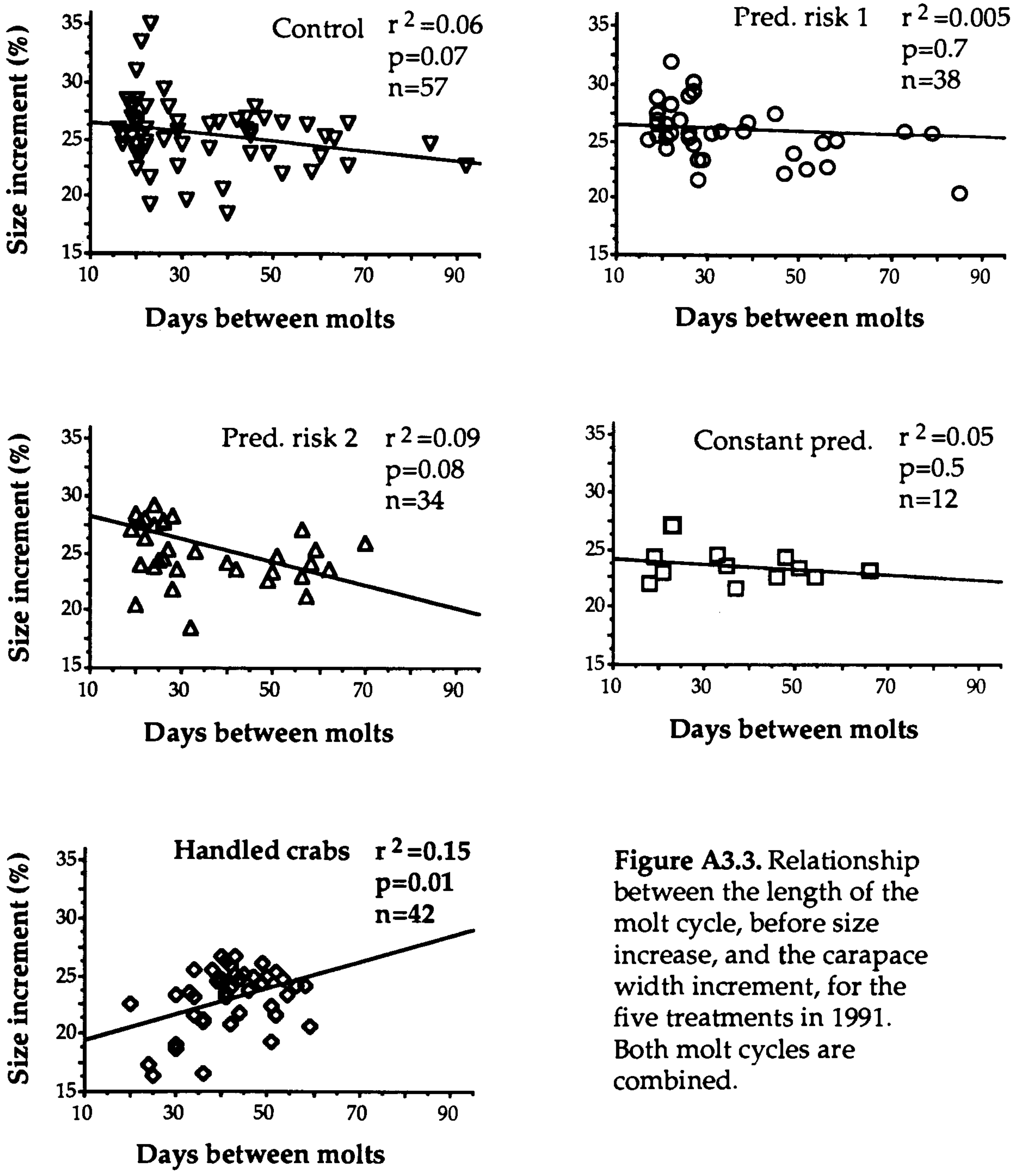

Figure A3.3. Relationship between the length of the molt cycle, before size increase, and the carapace width increment, for the five treatments in 1991. Both molt cycles are combined.