Shore crabs (*Carcinus maenas* (Linneaus)) as predators of rough periwinkles (*Littorina saxatilis* (Olivi)): can crab feeding behaviour influence the distribution of rough periwinkles?

being a Thesis submitted for the Degree of Master of Science

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by

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Abstract

The feeding behaviour of the predator Carcinus maenas (L.) whilst foraging on the two distinct ecotypes, H and M, of the rough periwinkle Littorina saxatilis (Olivi) found on the North East coast of England was studied. The high shore (H) ecotype possesses a thin shell and the mid shore (M) ecotype, being generally larger, possessing a thicker shell.

Surveys were carried out at two sites on Filey Brigg during July and November of 2007 to establish the population distribution and size density of both the H and M ecotypes of L. saxatilis and C. maenas. The surveys showed that the frequency of individual L. saxatilis present within each zone decreased as the tidal height decreased. There were no C. maenas found at Site 1 during either survey and crabs were only found at Site 2 during July. The densities of both the H and M ecotype found during July were lower than those recorded in November, and overall there was a far higher incidence of shell damage in the M ecotype from the boulders at Site 2 than there was in the H ecotype at Site 1 where no C. maenas were observed. The H ecotype occupied a higher zone on the shore and there was no overlap of distribution with that of Carcinus maenas. The increased shell damage observed at Site 2 could be the result of damage incurred by living in a mobile substrate or could be the result of unsuccessful predation attempts made by C. maenas.

Preliminary feeding trials showed C. maenas chose individuals from the M ecotype of L. saxatilis from the size classes that were the most abundant at the same tidal height as that particular size class of crab. The H ecotype chosen proved to be a size class larger than that chosen from the M ecotype. The energy content per unit shell height for each ecotype, H and M, was determined for a sample of the snails found on the rocky shores at Thornwick Bay and Filey Brigg. There was no significant difference between the energy content of the H and M ecotypes found at Thornwick Bay, or between the M ecotype found at either shore. The H ecotype from Filey Brigg contained a higher energy per unit shell height when compared with the M ecotype from the same shore.

The foraging behaviours of C. maenas when predating upon the H and M ecotypes of L. saxatilis were recorded and analysed. A further analysis showed that there was no significant difference in the total handling times for each size class of C. maenas when feeding on the H and M ecotypes of L. saxatilis. A significantly higher
frequency of attempts to break into the shell of the M ecotype was made by all size classes of crab, except the largest, than those made to break into the H ecotype. Individuals from the M ecotype were resistant to predation attempts from all but the largest size class of crab, whereas all individuals encountered from the H ecotype were successfully predated leaving the shell crushed completely or with the majority of the shell having been broken away. However, despite there being no significant difference in handling times between the ecotypes, all the interactions with the H ecotype resulted in a successful foraging attempt whereas only the large crabs managed to forage on the M ecotype thus indicating that there is a difference in prey value for the crabs between the two ecotypes.

An ethogram of behaviours recorded was constructed for all trials involving *C. maenas* and individuals from the H and M ecotype of *L. saxatilis*. The analysis performed on the data showed that crabs foraging on the M ecotype performed more acts of behaviour when compared to individuals foraging on the H ecotype. Crabs attempting to forage on the M ecotype manipulated the shell far more frequently than those feeding on the H ecotype and had to make many more attempts to break into the shell. Again, this could possibly indicate that the H ecotype of *L. saxatilis* offered a higher prey value to *C. maenas* due to the ease with which the crabs managed to gain food from the interaction.

The alarm response time in the form of escape crawling speeds was investigated for the H and M ecotypes of *L. saxatilis* in the presence and absence of the *C. maenas*. Individuals from the M ecotype of *L. saxatilis* showed slightly faster crawling speeds in the absence of *C. maenas* when compared to those of the H ecotype. In the presence of *C. maenas* both the H and M ecotype of *L. saxatilis* produced faster escape crawling speeds than were recorded in the absence of the crab for each ecotype.

Overall the study suggests that the H ecotype would be a more profitable prey item to foraging *Carcinus maenas* as predation success is far higher and the amount of attempts taken to break into the shell far lower than the M ecotype with its thicker and stronger shell. The M ecotype would therefore have a higher survivorship in regions occupied by *C. maenas* (or a more unstable substrate) than the thin shelled H ecotype, which might explain the difference in distribution observed in the field.
Chapter 1

Introduction

1.1 The study of *Littorina saxatilis*

*Littorina saxatilis* (Olivi) is a widely distributed marine gastropod occurring on rocky shores, in estuarine habitats and on artificial substrates such as piers, groynes and harbour walls throughout north-western Europe, the Atlantic coast of North America, and locally in both the Mediterranean and South Africa (Hull *et al*., 1996).

The taxon *Littorina saxatilis* comprises of the four species of rough periwinkle found around British coastlines that were first erroneously placed into one species by Dautzenberg and Fischer (1912). *Littorina compressa* Gray, *L. neglecta* Bean, *L. arcana* Maton and *L. saxatilis sensu stricto* Jeffreys which may all occur on the same shore, though each species may occupy a different habitat (Heller, 1975). *Littorina saxatilis* is a highly polymorphic species and there is general disagreement amongst littorinid co-workers about whether morphologically and genetically varied populations should be considered as sub-species or ecotypes. Certain forms of *Littorina saxatilis* are also argued by some authors to be deserving of species status such as the barnacle dwelling *L. neglecta*, and the lagoonal *L. tenebrosa* (Wilding *et al*., 2000). Based on genetic and morphological evidence, Johannesson & Johannesson (1990) regarded *Littorina neglecta* as an ecotype of *L. saxatilis* where as Grahame *et al*., (1995) were of the opinion that it deserved specific species status. Evidence from more recent studies (Gosling *et al*., 1998) also suggested *L. tenbrosa* to be distinct from *L. saxatilis* where previous work had failed to resolve any differences between the two taxa (e.g. Janson & Ward, 1984).

*Littorina saxatilis sensu stricto* is a directly developing, ovoviparous species that inhabits the upper shore. As the females carry shelled embryos in a brood pouch they therefore have a very low juvenile dispersal capability accompanied by low mobility in the adults (Janson, 1983; Erlandsson *et al*., 1998). The combination of directly developing offspring and low mobility of mature individuals is one of the main reasons why *Littorina saxatilis* typically displays high genetic differentiation at local as well as at broader geographical scales (Ward, 1990) and lives under different
physiological and ecological conditions depending on tidal reach (Raffaelli & Hawkins, 1996).

Littorinid snails are predominantly herbivorous. Some feed largely on epilithic microalgae and the germlings of seaweeds or microflora on mud in mangroves whereas those inhabiting the upper reaches of rocky shores have incorporated lichens into their diet (Norton et al., 1990). Many snails browse on the surface of seaweed, seagrasses or mangroves consuming epiphytic algae, fungi or microorganisms such as diatoms, protozoa, cyanobacteria and bacteria. With maximum reported densities of *Littorina saxatilis* of well over 100 000 m$^{-2}$, it will likely compete with other snail species (Reid, 1996) and can also prey on newly settled barnacles (Carlton & Cohen, 1998).

Within the work carried out on the differing ecotypes of *Littorina saxatilis* occurring on European shorelines, differing patterns of distribution with respect to ecotype and location on the shore have been recorded.

Recent work carried out at Old Peak (Ravenscar) on the north-east coast of England indicated that the taxon *Littorina saxatilis* (Olivi) comprised of two distinct forms, inhabiting high (H morph) and mid-shore (M morph) boulders (Hull et al., 1996).

H and M ecotypes found on large boulders in the upper intertidal zone, and, medium size boulders in the mid-intertidal, were recorded on the North East coast of England (Hull et al., 1996). Here, the two distinct ecotypes are found at different tidal heights on the same shore. The findings of this investigation show that the H ecotype found in the upper intertidal zone is characterised by thin, predominantly dark coloured and ridged shells, whereas the M morph found in the mid-intertidal zone has a relatively smooth, pale coloured and thicker shell in comparison. Hull et al., (1999) summarised the patterns of reproduction observed at the different tidal heights the H and M ecotypes of *L. saxatilis* found on the North east coast of England. They found that the H ecotype, as it inhabits the relatively stable environment of the upper intertidal zone, maximised reproductive success by producing a small number of large embryos. With the directly developed crawl-aways, possessing characteristics like a large foot to adhere to the substrate, and a smaller surface area to volume ratio to reduce the risk of desiccation. These characteristics reflect an adaptation to life on an exposed shore. The M ecotype maximised reproductive success by producing a large number of small embryos within the brood pouch. These small crawl-aways are easily able to seek refuge in crevices within the boulders. Producing a large number of small embryos could well favour the
survival rate of juveniles against crab predation or being crushed by boulders (Yamada & Boulding, 1996).

Hull et al., (1999) also documented the seasonality of reproduction between the two ecotypes and found that the H ecotype reduced its reproduction during the winter months, possibly due to the prolonged exposure to the cooler air temperatures in the high shore environment. Conversely, the M ecotype population maintained a high level of reproductive activity throughout the year (Hull et al., 1999a).

A similar pattern, although ultimately the reverse of that demonstrated at Old Peak, regarding the distribution of *L. saxatilis* ecotypes has been recorded on the exposed rocky shores of Galicia, North West Spain (Johannesson et al., 1993; Carballo et al., 2005). Here, the ecotypes of *L. saxatilis* inhabiting the upper reaches of the shoreline in the barnacle zone were classified as ridged and banded (RB), and those inhabiting the lower intertidal mussel zone as smooth and unbanded (SU) (Johannesson et al., 1993). Although the relationship between shell thickness and the zone on rocky shores they occupy reflects that described by Hull et al., (1996), the morphology of *L. saxatilis* shells found on the Galician coast varies dramatically to those found in the equivalent rocky shore zones of North East England (see Rolan-Alvarez et al., 2004), with the ecotype found higher up the shore being the more robust and tolerant of crab predation and the ecotype found lower down on the shore possessing a small and relatively delicate shell. The reproductive strategies employed by the Galician populations of *L. saxatilis* with regards to position on the shore differ to those used by the populations in the same zones of the shore in England. The low shore ecotype in Galicia produces large embryos and the high shore ecotype small embryos (Rolan-Alvarez et al., 1996).

On the west coast of Sweden different ecotypes of *L. saxatilis* continuously replace each other when occupying the different rocky shore habitats along the coast i.e. replacement along a horizontal gradient of exposure from different shores, rather than different ecotypes from different tidal heights within the same shore (Janson, 1982). This pattern is, again, similar to that described by Hull et al., (1996) with small, thin shelled snails possessing a comparatively large foot inhabiting the exposed rocky cliffs being classified as the E-morph, and relatively large, thick shelled snails with a comparatively smaller foot occurring on the sheltered boulder shores being classified as the S-morph (Erlandsson et al., 1998). As well as shell morphology, the two distinct ecotypes of *L.*
saxatilis found on the Swedish west coast also differed in characteristics such as egg number and embryo size (Janson, 1983), and migratory behaviour (Janson & Sundberg, 1983).

In another Scandinavian example, Van Marion (1981) investigated shell variation of Littorina rudis (a synonym of L.saxatilis) Reid (1996) on a sheltered boulder shore near the Biological Station at Trondheim, Norway. Within the samples collected from this shore, two different shell types were easily distinguished. The ecotypes recognised from this investigation expressed a thin shell with a relatively narrow aperture and a slightly ridged shell surface (Type A) whereas Type C snails had a comparatively thick shell with a wide aperture and a smoother shell surface than Type A individuals (Van Marion, 1981).

Immediate ecotypes of Littorina saxatilis between the different morphs have also been recorded at their respective zone on the shores around Europe. Hull et al., (1996) found the intermediate forms of the H and M ecotypes in the same habitat as the M ecotype although these intermediates were found to make up less than 2% of the M population. The intermediates are rare on the north east coast despite the fact that the two distinct morphs are found on the same shore. However, hybrid zones are common place where the horizontal zonation of the moderate and wave exposed types of L. saxatilis is apparent in Sweden. On Sweden’s west coast, over a distance of 5- 20 m, high numbers of snails with intermediate shell morphology of the E and S morphs occur. This zone represents the change in dominance from the wave-exposed ecotype to the moderate ecotype (Janson & Sundberg, 1983). The Galician variants of L. saxatilis overlap in a zone of 1m width. Here, 11 - 29% of the snails present are intermediate hybrids of the RB and SU ecotypes inhabiting these shores (Johannesson et al., 1993).

Together with studies on the reproductive strategies employed by L. saxatilis, many investigations have been carried out into the processes of mate selection within the populations of the different ecotypes present on the shores. Within several species of Littorina, intrasexual and intersexual copulations are a consistent feature of the mating behaviour (Saur, 1990). Raffaelli (1977) reported that on the shore, males of L. saxatilis and L. nigrolineata (now known as L. compressa) copulated with intraspecific males or individuals of the other species to the same extent as with females of their own species.

Size assortative mating is a common invertebrate mating pattern and is usually accompanied by male and female sexual selection and these three behaviours can
contribute to reproductive isolation (Hull, 1998). The results produced by Saur (1990) provide evidence that males of *L. saxatilis* and *L. littorea* have the ability to distinguish between the sexes as well as species. The frequencies of intrasexual copulations differed between the two species which was by the low frequency of interspecific copulating pairs found on the shore. There was no relationship between the size of mate of intersexual copulations of *L. littorea* on the shore from Saur’s (1990) results. However, *L. saxatilis* showed a significant correlation between the shell heights of mates. For this species, large males were recorded to copulate more often with large females and small males with small females (Saur, 1990).

Erlandsson & Johannesson (1994) investigated *Littorina littorea* to determine if there was sexual selection for female size in this species. In the study, size assortative mating was seen to be persistent in the one out of the three populations of *L. littorea* observed. In another three out of five populations examined, there was found to be a pattern of sexual selection on size among the females. As in Saur’s (1990) findings, large females were being favoured over their smaller counterparts for mating partners by the males. No population showed any indication of selection on male shell size (Erlandsson & Johannesson, 1994). It was also noted that the larger females were copulating more frequently and for longer durations than the smaller ones which supported the authors’ field data in that the large females were favoured by most males as mates.

In Hull’s (1998) study into the mating patterns of two micro-allopatric populations of *Littorina saxatilis* found on the northeast coast of England, complete assortative mating to type was evident in the field. The males of both the H and M ecotypes, occupying different habitats on the same shore, demonstrated the same pattern of copulation as shown by Saur (1990) and Erlandsson & Johannesson (1994) in that they showed sexual selection for females approximately 10% larger than themselves, but not necessarily the largest, when presented with an assortment of female sizes indicating true size assortative mating in the males from both ecotype populations (Arnqvist et al., 1996). The M males showed complete assortative mating to type irrespective of the density of females presented from either of the H or M ecotypes. The H males did occasionally mate with M females when the densities were low. Size assortative mating patterns
were also witnessed in Galician populations of *L. saxatilis*, especially in the upper zone (Johannesson *et al.*, 1995).

Pickles & Grahame (1999) found that the same assortative mating patterns described by Hull (1998), was also present within H and M ecotypes of *L. saxatilis* from shores as far as 20 km apart. They found that the males appear to mate with the ‘right’ ecotype even if it was not from the same shore. This could have a profound effect on the demography and genetics of populations (Hull, 1998).

Copulations between individuals from different species can result in a reduction of fitness in non-discriminating individuals therefore the relative importance of species recognition systems depends upon the cost of species inter-specific mating to the non-discriminating individuals (Saur, 1990). Johannesson *et al* (1995) reported an example of incipient reproductive isolation within two sympatric populations of *Littoirna saxatilis*, in Galicia, Spain. There were overlapping distributions in the mid-shore where intermediate forms of the two ecotypes present accounted for approximately 20% of the total population (Johannesson *et al.*, 1993). Gene flow between the ecotypes was less than that within the ecotypes (Rolan-Alvarez *et al.*, 1996). This suggests that the two populations of *L. saxatilis* demonstrate incipient reproductive isolation. Rolan-Alvarez *et al.*, (1996) also noted that the hybrids between the two ecotypes were at a disadvantage as they mated less frequently than the pure ecotypes that were present where either of the pure ecotypes occurred in high frequencies. Rolan-Alvarez *et al.*, (1996) suggests that sexual selection acted to restrict gene flow between the two populations.

In all the above cases, there is some evidence that the taxon *L. saxatilis sensu stricto* is showing evidence of habitat specialisation. However, the mechanisms driving this divergence between the populations remains poorly understood.
1.2 The study of *Carcinus maenas*

*Carcinus maenas* (L.) is an intertidal brachyuran crustacean that is abundant on most northwest European shores and is the most familiar crab to be found in Northern Europe. Although predominantly intertidal, *C. maenas* can also be found in the shallow sublittoral zone, and due to wide tolerance of differing salinities, also range into estuaries and onto the upper reaches of salt marshes (Neil & Pizzolla, 2008).

*Carcinus maenas* (L.) can be considered a true omnivore as it consumes plants, algae, molluscs, arthropods (including its own species), annelids and carrion. For large individuals, *Mytilus edulis* is an important part of the diet. On rocky shores, juveniles consume *Semibalanus balanoides* whereas adults consume more gastropods (Rangeley & Thomas, 1987) especially the dog whelk, *Nucella lapillus*, and winkles, *Littorina Sp.* (Little & Kitching, 1996).

Crabs are common predators of marine snails in many parts of the world (Ekendahl, 1998). Together with birds and fishes they appear to be the most important predators on littorinid snails in the intertidal zone (Pettit, 1975; Cook & Garbett, 1992). For example, the green crab, *Carcinus maenas* (L.), is an important predator on *Littorina* species on both sides of the North Atlantic (Johannesson, 1986; Rangeley & Thomas, 1987; Lowell *et al.*, 1994).

Crabs are not visual hunters (Ekendahl, 1998). They cannot scan large areas to estimate prey availability. When foraging for food, prey is encountered one, or a few at a time, and detected by the walking legs when crawling over substratum. *Carcinus maenas* (L.) hunts for food using chemical and tactile stimuli received by sense organs on the antennae and walking legs (Crothers, 1968). The prey is detected at a distance in the water by chemoreceptors on the antennae. Funnel canals on the tops of the walking legs respond to the stimulus of prey beneath them and the prey is then grasped within the crabs’ chelae. Peak foraging occurs at night around high tide (Ropes, 1968), and predation rate is dependent on prey density (Ropes, 1968; Walton *et al.*, 2002) and environmental temperature (Ropes, 1968; Sanchez-Salazar *et al.*, 1987).

Work carried out on the diet of *Carcinus maenas* (L.) by Elner & Hughes (1978), confirms that manipulation with the chelae is an important behaviour shown by the predator before the acceptance or rejection of the prey item. Elner & Hughes (1978)
also provided data on the optimal prey size with respect to individual *C. maenas*. The prey items used in this investigation were blue mussels (*Mytilus edulis*). It was found that the optimal prey size increases with crab size and that suboptimal prey is incorporated into the diet in proportion to relative abundance, even when optimal mussels are present in excess. The optimal mussels were always taken when encountered. The suboptimal mussels were rejected at the first encounter following an encounter with an optimal mussel, but finally taken after a sequence of encounters. This result shows that *Carcinus maenas* (L.) must have a relatively short-term memory, or, as described by Cunningham & Hughes (1984) when investigating the predatory skills of *C. maenas* on mussels and dogwhelks, the skilfulness of a crab at any time represented the balance between a high rate of learning and a much lower rate of forgetting.

Along with the work that has been carried out on the diet of *Carcinus maenas* (L.), the way that the prey is attacked and manipulated has also been investigated.

When investigating the predation tactics of *Carcinus maenas*, and the morphological responses of *Nucella lapillus*, Hughes & Elner (1979) recognised that three distinct attack methods were being employed by the crab to get to the soft flesh within the shell.

These techniques were recognised again by Cunningham & Hughes (1984) when investigating the learning of predatory behaviour in shore crabs. They are as follows:

1). Crushing across the columella end along any radius; Shells were held by small chela and crushed by master chela. This method was largely successful when prey less than 10 mm in height was encountered. 2). Crushing across the apical end, usually across the last whorl; When larger shells were encountered, the shell was held vertically with the master chela around the apex. Whilst gripping the lip or columella with the smaller chela, the prey was steadied and the sub-apical whorls were crushed using the master chela. Often only partially successful, this method would leave the crab able to extract most of the flesh from the remainders of the broken shell. 3). Attacks around the lip of the shell, or along the collumella axis; Used during encounters with prey over 10 mm in height. Prey is often inverted so the apex is held by the smaller chela whilst the master chela attempts to crush the basal whorls.
1.3 Predation on *Littorina saxatilis* by *Carcinus maenas*

Eastwood *et al.*, (2007) used a series of factorial experiments to demonstrate that predation on *Littorina saxatilis* decreased with tidal height and that this species was preferentially predated upon by *Carcinus maenas* when the predator was offered *L. saxatilis* and *Littorina littorea*. The investigation also showed that predation rates on gastropods were significantly higher on sheltered shores than on wave exposed shores.

Elner & Raffaelli (1980) found that *C. meanas* was more likely to predate upon *Littorina rudis* (*L. saxatilis*) than *L. nigrolineata* (*L. compressa*). Eastwood *et al.*, (2007) suggest that whilst behavioural responses are important in determining the upper most distribution of *L. saxatilis* (they will move higher up the shore once displaced), the lower limit of their distribution was controlled by predation by *C. maenas*, an invasive species in the Gulf of Maine where the studies took place.

Johannesson (1986) investigated the importance of shell morphology of *Littorina saxatilis* (Olivi) in withstanding predation by *Carcinus maenas*. In this investigation, samples of *L. saxatilis* were taken from three localities. The individuals taken from each locality showed a similar variation in shell morphology to the H and M morphs of *L. saxatilis* found at Old Peak (Ravenscar) and studied by Hull *et al*., (1996) in that the ecotypes found higher up the shore have thinner shells than those found lower down the shore.

Johannesson (1986) provided evidence that small female shore crabs attacked *L. saxatilis* using two different methods; ‘pulling’ and ‘crushing’. When the first method was employed by the crab, the shell of the prey was not damaged and the soft flesh was pulled out of the shell aperture. When the crushing tactic was employed, the shell was broken open by the master chela and the flesh removed. It was noted that these two main methods of attack used by *Carcinus maenas* could be further divided into five distinct techniques placed into two categories:-

A. Pulling techniques:

1). Prey held by the master chela whilst supported against the crab’s sternum. Pincher chela inserted into the shell aperture to grip the foot and operculum. When a firm grip is in place, the whole of the soft body is pulled out in one go. When larger, stronger snails
were encountered, or snails that had retreated deeper into their shell, the master chela was used instead of the pincher chela to pull out the soft body.

2). If insertion of both parts of the chela into the aperture was not possible, the crab used either the propus or dactylus of either chela is inserted behind the operculum. A wrestling motion away from the crab’s body would then push out the soft flesh until there is enough of the foot showing so a grip by the mouth parts or chela and pulled out.

Both of these techniques were only partly successful in that some of the posterior soft body parts were left in the shell, out of reach of the crabs (Johannesson, 1986).

B. Crushing techniques:

1). Small and weak snails steadied by pincher chela or mouthparts and crushed by master chela.

2). Larger individuals held vertically around apex with master chela. Small chela grasps the lip or columella of the shell, while the subapical whorls are crushed by the master chela often resulting in removal of the whole apex.

3). Less frequently, crabs were able to crush the columella by holding the shell vertically with the pincher chela around the apex and inserting the master chela into the shell aperture to grasp the columella. This resulted in the shell being broken into two pieces through the aperture (Johannesson, 1986).

Ropes (1968) studied the feeding habits of Carcinus meanas and observed behaviour similar to that recorded by Hughes & Elner (1978) and Johannesson (1986). Individual C. maenas, when encountering Mya arenaria ≥10 mm long, crushed or chipped pieces off the shell with the chelae, severed the adductor muscles then tore out pieces of flesh and ate them. Only the larger clams that were encountered were able to resist the attacks from C. maenas. Personal observations from Ropes (1968) during this investigation stated that some clams under 10 mm in length were picked up, crushed and then ingested nearly whole by the crab.
1.4 Aims of the thesis

At present we know very little about the mechanisms which may be driving the observed habitat differentiation in the two ecotypes of *L. saxatilis* found on the Yorkshire coast. Whilst adaptations to differing wave exposures (Davenport & Davenport, 2005), physiological constraints due to the occupation of different habitats on the shore (Hull *et al.*, 1999) may also be important, it is apparent the ability to withstand crab predation may also be a factor which regulates the distributions observed in the field (Johannesson, 1986; Elner & Hughes, 1978; Eastwood *et al.*, 2007). Therefore this thesis focuses upon an investigation of the effects of crab predation on the two ecotypes of *L. saxatilis* in order to determine if this may be important in regulating the distributions and driving the separation between them.

Chapter 2 presents the data collected about the distribution and density of both *Littorina saxatilis* (H and M ecotypes) and *Carcinus maenas* on the shore to establish if the predator occurred in the same area as the snails and could therefore possibly affect their distribution and density. The aims of this part of the study were two-fold.

Firstly, to determine the population density, shell size and amount of damage of both the H and M ecotypes of *L. saxatilis* with tidal height along a transect at two sites on the rocky shore at Filey Brigg in the July and November of 2007. Secondly, using the same transects and sampling periods, to record the density, carapace size and distribution of *C. maenas*.

Having established the fact that small *C. maenas* can often be found in the upper reaches of the tidal zone (though not usually within the H habitats) the remaining chapters of the thesis present the results of the laboratory based investigations of the interactions between *Carcinus* and its potential prey item *L. saxatilis*. Chapter 3 examines whether or not there is a difference in calorific value between the two ecotypes of snail and describes and quantifies the behaviours associated with crab foraging on both ecotypes. After establishing the calorific value of the snails the Chapter then determines whether or not the H and M ecotypes of *L. saxatilis* have differing prey value to foraging shore crabs of different sizes and also examines whether or not the crabs will exhibit a preference for either ecotype. Finally, Chapter 3 aims to determine if there is evidence of a reduction in shore crab handling time with experience over repeated foraging trials.
Chapter 4 aims to determine what magnitude of response in elicited in each ecotype of *L. saxatilis* in the form of escape times and crawl speeds when exposed to situations involving the absence and the presence of the predator *C. maenas*.
Chapter 2

Density and distribution of H and M *Littorina saxatilis* ecotypes and *Carcinus maenas* on the shore.

2.1 *Littorina saxatilis* density and distribution.

As previously stated in the introduction (Section 1.1) *Littorina saxatilis* (Olivi) is a widely distributed marine gastropod throughout north-western Europe, the Atlantic coast of North America, and locally in both the Mediterranean and South Africa (Hull et al., 1996).

Within the intertidal zone, the distribution of *L. saxatilis* varies between rocky shores extending from roughly mid-tide level up to above the extreme high-water mark of the spring tide where there is abundant spray on exposed shores, or to a level between that of mean high water mark of neap tides and that of spring tides on more sheltered shores (Berry, 1961). Previous work has also shown that the uppermost limit of *L. saxatilis* is often more obviously defined than that of the lower shore limit (Colman, 1933; Evans, 1947).

The population density of *L. saxatilis* also varies greatly between shores. Work carried out in Devon, observed up to 3000 snails per at Rum Bay and also numbers as low as 400 per were recorded in nearby localities (Moore, 1940). Densities of 1100 per were recorded by Spooner & Moore (1940) in the Tamar estuary, and Thorson (1941) collected 370 individuals from an area of just 0.1 on an Icelandic shoreline. The highest recorded density of *L. saxatilis* is well over 100 000 per (Reid, 1996) and the abundance within populations has been shown to be consistently higher towards the upper limits of their ranges than at more seaward levels (Berry, 1961).

Part of the investigation conducted by Berry (1961) was focussed on the fecundity levels of *L. saxatilis* in relation to their vertical height on the shore. After transferring individuals from their original level on the shore to an unfamiliar (higher or lower on the shore) level the conclusion was made that when the gastropods were coming into full egg production and the conditions were favourable, they rapidly acquired the fecundity characteristics of the zone to which they had been relocated. This was irrespective of the zone from which they had been transferred. Berry (1961) also concluded that the larger ovaries, higher brood pouch counts, and ultimately denser
populations observed at the higher shore levels, are all dependant on more favourable ecological conditions here than at lower shore levels.

2.2 Field density and distribution of *Carcinus maenas*.

*Carcinus maenas* (L.) is primarily an inshore intertidal species, but evidence from some studies has shown that it also undergoes migration at certain times of the year (Crothers, 1968, Ropes, 1968, Warman *et al.*, 1993). Not only seasonality (Naylor, 1962, Crothers, 1968), but crab size (Naylor, 1962) and moult stage (Warman *et al.*, 1993) have also been suggested as factors affecting the migratory behaviour *C. maenas*, usually occurring during the winter months (Hunter & Naylor, 1993).

Crothers (1968) concluded that some crabs remain hidden on the shore at low tide throughout the year. Other crabs were recorded to forage intertidally with the flood tide then retreat back down shore with the ebb. Some crabs were seen to remain permanently below the low water mark of the shore.

With respect to crab size, Warman *et al.*, (1993) suggest crabs with a carapace width of less than 35mm remain within the intertidal zone of the shore throughout the tidal cycle. Large, predominantly red crabs were recorded to remain in the sublittoral zone with the large, green crabs migrating up shore with the flood tide and down shore with the ebb.

Work carried out by Naylor (1962), Crothers (1968) and Warman *et al.*, (1993) confirmed that small crabs (<30mm carapace width) remain between tide marks over all states of tide and at all seasons. Data has also been presented that confirms the influx of intertidal migrants during the summer months and to a lesser extent the winter (Naylor, 1962; Crothers, 1968; Dare & Edwards, 1981)

Heller (1975) studied the changes in shell size and shape of *Littorina nigrolineata (=compressa)* and *L. rudis* (a synonym of *L. saxatilis*) in relation to wave exposure and the size of *Carcinus maenas* found at the same locality. In both species, small, more globose shells were favoured by individuals that inhabit an exposed shore with *L. rudis* possessing a relatively large aperture. On shores with similar levels of exposure, larger shells were favoured with the presence of large crabs with smaller shells being favoured in the presence of small crabs.
2.3 Aims

This part of the current study focuses upon obtaining information about the distribution and density of both *Littorina saxatilis* (H and M) and *Carcinus maenas* on the shore to establish if the predator occurred in the same area as the snails and could therefore possibly affect their distribution and density. The aim was twofold. Firstly, to determine the population density, shell size and amount of damage of both the H and M ecotypes of *L. saxatilis* along a vertical transect (upper to lower shore) at two sites on the rocky shore at Filey Brigg in the July and November of 2007. Secondly, using the same transect and sampling periods, to record the density, carapace size and distribution of *C. maenas*. 
2.4 Methodology

2.4.1 Site Description

Each site at Filey Brigg (British National Grid reference TA131815) was chosen for use within the investigation as both H and M ecotypes of *Littorina saxatilis* and *Carcinus maenas* are present on this shore.

Plate 2.1 Aerial view of Filey Brigg showing Site 1 and Site 2 (Google Maps, 2008)
Site 1 (Agony Point)

Plate 2.2 Site 1 (Agony Point) showing zones from which *L. saxatilis* of the H and M ecotypes were collected (photograph taken by author).

Site 1 comprises of a vertical rock face rising approximately 2.5 – 3 metres in height from the bedrock on the upper most limit of the shore. The rock face has large crevices and fissures occurring randomly throughout the outward surface. At the foot of the rock face, small boulders and temporary rock pools were present (Plate 2.2). During periods of high tides, the rock face is within the splash zone. Three definite zones were clear on the rock face: *Verrucaria maura* (7.3 – 6.5m above chart datum), the beginning of the *Fucus spiralis* zone (6.8 – 6m above chart datum) and the start of the *Semibalanus balanoides* zone (5.8m above chart datum)(Plate 2.2).
Site 2 (Carr Naze)

Plate 2.3 Site 2 (Carr Naze) showing zones from which L. saxatilis of the H and M ecotypes were collected (photograph taken by author).

Site 2 has a more horizontal zonation gradient down the shore to the sea than Site 1. Boulders of 20 – 50 cm length lie scattered across the bedrock approximately 2 metres further down the shore than a man made concrete maintenance strip which has little vegetation present as people use this strip as a footpath to get down to the end of the Brigg. Within Site 2 four very distinct zones can be seen: the *Pelvetia canaliculata* zone at the top of the boulder field (7 – 6.6m above chart datum), the *Fucus spiralis* zone (6.8 – 6m above chart datum), *Fucucs vesiculosus/Semibalanus balanoides* zone (5.5 – 4m above chart datum) and the *Fucus. vesiculosus/Fucus. serratus* zone (3.8 – 3.3m above chart datum). Although no data was collected on boulder size, it was apparent that size increased down the shore.
2.4.2 Sampling methodology

At the top of Site 1 a 50 metre transect was placed on the shore running from MHWS to the sea starting in the *Verrucaria* zone (0 m transect distance), then into the beginning of the *Fucus spiralis* zone (1m transect distance), and the start of the *Semibalanus balanoides* zone (2m transect distance. Sampling at Site 2 commenced in the *Pelvetia* zone at the top of the boulder field (20 – 23 m transect distance) as there were no snails any higher on the shore. Samples were also collected from the *Fucus spiralis* zone (26 – 30m transect distance) within the boulder field, the *Fucus vesiculosus/Semibalanus balanoides* zone (35 – 40 m transect distance), and ending in the *F. vesiculosus/F. serratus* zone (47 – 50 m transect distance).

In order to quantify the density and size of each *L. saxatilis* ecotype present within each zone at the two sites, ten replicate quadrats were placed in each zone described above along the transects. Quadrats have been used extensively in determining the distribution of slow moving invertebrates such as those found in leaf litter or in intertidal habitats. They are useful for identifying the species composition of a particular community and for making a quantitative assessment of the abundance of those species (Williams, 1987). As Site 2 comprised of loose small rocks, each rock was turned over to find the snails to find the snails then replaced in their original position.

The columellar height of all *Littorina saxatilis* found within each quadrat was measured to the nearest millimetre using callipers and the ecotype was noted for each individual. Any damage seen on the shells of *L.saxatilis* was recorded and the means of the damage was noted, i.e. crab damage or rock crushing damage etc (Blundon & Vermeij, 1983). Within the random quadrats in each zone of each site, any *Carcinus maenas* encountered had the width of their carapace measured and recorded. A survey of the rocky shore at Filey Brigg was carried out during July and November, 2007, for both Site 1 and Site 2.
2.4.3 Data Analysis

Size frequency histograms were produced in order to illustrate the population density and size distribution of *Littorina saxatilis* and *Carcinus maenas* for the rocky shore habitat of Filey Brigg where both the ecotypes of *L. saxatilis* occur in close proximity. Histograms of the damage seen on the shells measured in each zone at each site were produced to show the percentage of the total damage seen that can be credited to crab damage.

In order to test the null hypothesis that there was no significant difference in median *Littorina saxatilis* (H or M ecotype) density between the different zones within a site (either Site 1 or Site 2) during either July or November a Kruskal Wallis test was applied to the data. The Kruskal Wallis test was used as in all cases the data did not conform to a normal distribution (Kolmogorov-Smirnov test, $P < 0.05$) nor could the variances be considered equal (Levenes test $P < 0.05$ in all cases) and there were no more than two classes of the shell height variable therefore the Kruskal Wallis test was deemed appropriate (Dytham, 2003). The same procedure was used for the density data of each ecotype and the data collected for the M ecotype at Site 2 for each month.

In order to test the null hypothesis that there was no significant difference in *Littorina* density within each of the two sites between months a Mann-Whitney U test was applied to the data. As the data did not conform to a normal distribution (Kolmogorov-Smirnov test, $P < 0.05$) a Mann Whitney U-test was selected to prevent Type 1 errors (Dytham, 2003)
2.5 Results

2.5.1 Site 1

In July, there was a significant difference in median density of *Littorina saxatilis* H ecotype between the different tidal zones (Kruskal-Wallis \( H = 17.51, \ df = 2, \ P = < 0.001 \)). Overall densities were highest in the *Verrucaria* zone, followed by that found in the *Fucus spiralis* zone with the lowest densities found in the *Semibalanus* zone (Table 2.1).

Table 2.1 – Median *Littorina saxatilis* H density for each zone, Site 1, July 2007

<table>
<thead>
<tr>
<th>Zone</th>
<th>Minimum</th>
<th>Median</th>
<th>Maximum</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Verrucaria</em></td>
<td>15.00</td>
<td>39.50</td>
<td>77.00</td>
<td>15-77</td>
</tr>
<tr>
<td><em>F. spiralis</em></td>
<td>16.00</td>
<td>20.00</td>
<td>42.00</td>
<td>16-42</td>
</tr>
<tr>
<td>Barnacle</td>
<td>0.000</td>
<td>0.000</td>
<td>1.000</td>
<td>0 – 1</td>
</tr>
</tbody>
</table>

With respect to shell height, in the *Verrucaria* zone a total of 360 *L. saxatilis* of the H ecotype were found (smallest height of 1.5 mm and the tallest at a height of 11.5 mm). The modal groups are 2.5 mm and 3.5 mm with each group having 37 individuals at this shell height. There were three definite peaks of distribution on the size frequency histogram at 2.5, 3.5 and 6.5 mm (Fig 2.1).

In the *Fucus spiralis* zone, a total of 196 individuals were found and measured with the shortest at 1.5 mm and the tallest being 12 mm. The modal group with 20 individuals in this size class was 5.5 mm. On the height-frequency histogram two definite peaks can be seen, one at 5.5 mm and one at 8.5 mm (Fig 2.1) whereas, in the *Semibalanus balanoides* zone, only 6 individual *L. saxatilis* were found. From the six found, five were deemed to be individuals from the M ecotype. The only H ecotype had a shell height of 4.5mm.
Figure 2.1. Height-Frequency Histogram for H ecotype, Site 1, July 2007

Figure 2.2. Height-Frequency Histogram for M ecotype, Site 1, July 2007
Figure 2.2 shows that there were no M ecotype individuals found in the *Verrucaria* zone at Site 1 during the July survey where as four individual M ecotype snails were found in the *F. spiralis* zone and five were found in the *S. balanoides* zone.

Within the *L. saxatilis* collected from the *Verrucaria* zone in July, a total of 4.5% of the shells collected showed signs of external damage to the shell with 1.9% being credited to previous attempts to feed on the snails made by a crustacean predator (Fig 2.3). 5.6% of shells from the *F. spiralis* zone exhibited signs of damage with 1.5% being due to crab predation. The shell found in the barnacle zone did not show any signs of external shell damage (Fig 2.3).

**Figure. 2.3 Histogram showing percentage damage to H ecotype shells Site 1, July 2007**

The Kruskal-Wallis test for site 1, November, showed that the null hypothesis of no significant difference in median density of H morph *L. saxatilis* between the different zones could be accepted (Kruskal-Wallis (H = 0.22, df = 2, P = 0.896). Unlike the
sample from July, each of the three zones had similar median densities of the H morph (Table 2.2).

**Table 2.2 – Median *Littorina saxtilis* H density for each zone, Site 1, November 2007**

<table>
<thead>
<tr>
<th>Zone</th>
<th>Minimum</th>
<th>Median</th>
<th>Maximum</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Verrucaria</em></td>
<td>2.00</td>
<td>9.00</td>
<td>41.00</td>
<td>2 - 41</td>
</tr>
<tr>
<td><em>F. spiralis</em></td>
<td>1.00</td>
<td>12.00</td>
<td>25.00</td>
<td>1 - 25</td>
</tr>
<tr>
<td><em>S. balanoides</em></td>
<td>7.00</td>
<td>11.00</td>
<td>16.00</td>
<td>7 - 16</td>
</tr>
</tbody>
</table>

In the *Verrucaria* zone, a total of 142 *L. saxatilis* of the H ecotype were found with a range of shell heights from the shortest at 2mm to the tallest at 10.5mm. The modal group on the size frequency histogram was 7mm with 22 individuals in this size class. The peaks of size distribution occur at 4.5, 5.5 and 7mm (Fig 2.4).

In the *Fucus* zone, peaks in the size frequency histogram occur at 2.5mm, the modal size class of 3.5mm, 4.5mm, 6.5mm, 7.5mm and 8.5mm. 118 *L. saxatilis* were found (Fig 2.4) this height with a shell height range of 2.5mm to 11.5mm. There were 17 snails in the modal group of 3.5mm (Fig 2.4).

60 *L. saxatilis* were found in the *Semibalanus balanoides* zone with the shortest shell height being 2mm and the tallest shell height being 11.5mm (Fig 2.4). The modal size class was 2.5mm with 13 individuals with a shell of this height. The peaks of size distribution on the size frequency histograms occur at 2.5, 5.5, 6.5, and 7.5mm (Fig 2.4).
Figure 2.4 Height-Frequency Histogram for H ecotype, Site 1, November 2007

Figure 2.5 Height-Frequency Histogram for M ecotype, Site 1, November 2007
Figure 2.5 shows that there were no individuals from the M ecotype found in the *Verrucaria* zone and only one found in the *S. balanoides* zone with a shell height of 9mm. The highest numbers of M ecotype *L. saxatilis* found at Site 1 in the November survey were found in the *F. spiralis* zone.

1.3% of the shells found in *Verrucaria* zone at Site 1 during the November survey showed signs of external shell damage with exactly half (0.65%) being due to crab damage (Fig 2.6). None of the shells found in the *F. spiralis* or the barnacle zones showed any signs of shell damage.

![Figure 2.6 Histogram showing percentage damage of H ecotype shells, Site1, November 2007](image)

Although a small percentage of the shells found in the zones at Site 1 during the July and November surveys showed signs of crab damage, there were no *Carcinus maenas* found within the zones during the surveys.
There was no significant difference in median density for the H ecotype at Site 1 between the two sampling periods (Mann-Whitney U test, $W = 635.5$, $df = 24$, $P = 0.3315$), therefore the null hypothesis can be accepted. The densities were similar in July (median = 19.5; range = 0 - 77) to that of November (median = 11.50; range = 1 - 41). Also there was no significant difference in the median density of the M ecotype at Site 1 between the two months (Mann-Whitney U test, $W = 603.0$, $df = 24$, $P = 0.6838$). The densities were similar in July (median = 0.0; range = 0 - 5) and November (median = 0.0; range = 0 - 8).

### 2.5.2 Site 2

The Kruskal-Wallis test for site 2, July, showed that the null hypothesis that there was significant difference between the median densities of M morph *L. saxatilis* found in the different zones could be rejected (Kruskal-Wallis ($H = 15.03$, $df = 3$, $P = 0.002$)). Densities were the highest in the *Semibalanus balanoides* zone and lowest in the *Pelvetia* zone (Table 2.3).

Table 2.3 – Median *Littorina saxtilis* M density for each zone, Site 2, July 2007

<table>
<thead>
<tr>
<th>Zone</th>
<th>Minimum</th>
<th>Median</th>
<th>Maximum</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Pelvetia</em></td>
<td>0.000</td>
<td>0.000</td>
<td>3.000</td>
<td>0-3</td>
</tr>
<tr>
<td><em>F. spiralis</em></td>
<td>0.000</td>
<td>5.500</td>
<td>33.00</td>
<td>0-33</td>
</tr>
<tr>
<td><em>S. balanoides</em></td>
<td>3.000</td>
<td>15.00</td>
<td>28.00</td>
<td>3-28</td>
</tr>
<tr>
<td><em>F. vesiculosus</em></td>
<td>1.000</td>
<td>4.500</td>
<td>8.000</td>
<td>1-8</td>
</tr>
</tbody>
</table>

In the *Pelvetia* zone at Site 2 only 5 *L. saxatilis* were found in the ten quadrats. The shortest shell was 2mm and the tallest was 10.5mm with shell heights of 3, 4, and 7.9 mm recorded for the remaining individuals found at this shore height (Fig 2.7).

More snails were present in the *F. spiralis* zone; a total of 107 *L. saxatilis* were found, the shortest of which was 1.5 mm and the tallest was 15.5 mm. The size frequency histogram shows the modal shell height group to be 8.5 mm with 10 individuals in this size class. There are two definite peaks of distribution at this transect distance showing there are more *L. saxatilis* with shell heights of around 4 mm and 8.5 mm (Fig 2.7).
A total of 119 *L. saxatilis* were collected in the *S. balanoides* zone with the shortest shell height being 1.5 mm and the tallest at 11.5 mm (Fig 2.7). The modal size class for this distance was 4 mm with 13 individuals at this shell height. Two peaks are present on the height-frequency histogram at 4 mm and 7.5 mm.

The *L. saxatilis* from the *F. vesiculosus* zone had a shell height range from 1.5 mm to 10.5 mm with two size classes, 6.5 mm and 8 mm, being equally modal with 7 snails at each shell height (Fig 2.7). These modal groups are where the peaks on the height-frequency histogram occur for the 33 *L. saxatilis* found.
Figure 2.8 shows that only one *L. saxatilis* H ecotype with a shell height of 6mm was found in at Site 2 during the July survey.

Figure 2.9 shows that the no shells found in the *Pelvetia* zone of Site 2 expressed any external signs of shell damage. Of the 8.4% of shells found in the *F. spiralis* zone, 1.8% could be accredited to crab damage. 17.8% of the shells found in the *F. vesiculosus/S. balanoides* zone had damaged shells with 3.4% being due to crab damage. 21.8% of the shells found in the *F. vesiculosus/F. serratus* zone had damaged shells with 12.5% being put down to crab damage. Therefore the proportion of damaged shells increased with distance down the shore.
For the July data, the null hypothesis that there was no significant difference in median density of *L. saxatilis* M ecotype found in the different zones could be accepted (Kruskal-Wallis (H = 0.96, df = 3, P = 0.811). The density of the M morph was similar in all zones (Table 2.4).

### Table 2.4 Median *Littorina saxatilis* M density for Site 2, November 2007

<table>
<thead>
<tr>
<th></th>
<th>Minimum</th>
<th>Median</th>
<th>Maximum</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Pelvetia</em></td>
<td>1.000</td>
<td>5.50</td>
<td>53.00</td>
<td>1-53</td>
</tr>
<tr>
<td><em>F. spiralis</em></td>
<td>2.000</td>
<td>14.00</td>
<td>29.00</td>
<td>2-29</td>
</tr>
<tr>
<td><em>S. balanoides</em></td>
<td>2.000</td>
<td>7.00</td>
<td>36.00</td>
<td>2-36</td>
</tr>
<tr>
<td><em>F. vesiculosus</em></td>
<td>2.000</td>
<td>6.50</td>
<td>23.00</td>
<td>2-23</td>
</tr>
</tbody>
</table>

At 20-23m (*Pelvetia* zone), 155 *L. saxatilis* of the M ecotype were found with a range of shell heights from 2.5mm to 14.5mm (Fig 2.10). The modal groups were 10mm and...
11mm with 18 individuals in each size class. The peaks of size distribution in the size frequency histogram occur at 3, 4.5, 6.5, 8, 10 and 11mm. A total of 135 *L. saxatilis* were recorded at the *F. spiralis* zone with a shell height range of 2mm to 13.5mm with 7.5mm and 9.5mm being the modal size groups with 15 individuals with shells of these heights. The peaks occur in the height-frequency histogram at 6.5, 7.5, 9.5 and 11.5mm (Fig 2.10). In the *F. vesiculosus/S. balanoides* zone 77 *L. saxatilis* were found with a shell height range from 3.5mm to 13mm (Fig 2.10). The modal group was 7.5mm with 8 snails having a shell of this height. In the size frequency histogram there were two definite peaks occurring at 6 and 7.5mm. A total of 80 *L. saxatilis* were found at the *F. vesiculosus/F. serratus* zone with a shell height range from 2.5mm to 13.5mm. The modal group, with 9 snails with a shell of this height, was 6.5mm. Size distribution peaks in the size frequency histogram occur at 3.5 and 6.5mm (Fig 2.10). There were no H ecotype *L. saxatilis* found at Site 2 during the survey carried out in November.

Signs of shell damage were apparent in each zone at Site 2 during the survey carried out in November (Fig 2.11). 21% of shells from the *Pelvetia* zone and 23.7% from the *F. spiralis* zone were damaged with no signs of crab predation. Of the 20.7% of shells showing external signs of shell damage from the *F. vesiculosus/S. balanoides* zone, 7.8% of this damage was deemed to have been caused by crab predation along with 7.6% of the shells damaged from 10.1% of the shells showing damage from the *F. vesiculosus/F. serratus* zone (Fig 2.11).
Figure 2.10 Height-Frequency histogram for M ecotype, Site 2, November 2007

Figure 2.11 Histogram showing percentage damage of M ecotype shells, November 2007
As so few *L. saxatilis* from the H ecotype were found at Site 2, no valid statistical analysis could be performed on the data for the H ecotype at this site.

There was a significant difference in median density for the M ecotype at Site 2 (Mann-Whitney U test, $P < 0.05$, $W = 880.5$, $df = 32$ (July) 34 (Nov), $P = 0.0141$), therefore the null hypothesis can be rejected. The densities were lower in July (median = 4.00; range = 0 - 33) and November (median, 7.50; range = 1 - 53).
Figure 2.12 shows that the highest numbers of *C. maenas* were found in the *F. vesiculosus/S. balanoides* zone where 19 individuals with a carapace width range of 10 mm (1.5 – 11.5) were recorded. The zone supporting the next highest density of *C. maenas* was the *F. vesiculosus/F. serratus* zone where 16 individuals were found. The two largest individual *C. maenas* (carapace width = 29 & 50 mm) was found in the *F. spiralis* zone where the lowest density was recorded.
2.6 Discussion

2.6.1 Distribution of *Littorina saxatilis*

The data collected in both July and November 2007 regarding the size distribution of *Littorina saxatilis* on Filey Brigg revealed a very definite pattern with the highest number of individuals from the H ecotype being found at the limit of the population’s vertical distribution (see Table 2.1 and 2.2). This would suggest that snails of all shell heights exploit the space available within the cracks and crevices on the vertical wall. This further supports what Janson (1983) reported with the high shore, thin shelled E ecotype showing a preference for the ‘protected’ area of the tray used during the migration experiments as well as the aggregations of the E (H) ecotype seen in cracks and crevices in the field.

The higher densities of the H ecotype in the highest zones in Site 1 could be explained by the H ecotype maturing at a smaller size (Hull et al., 1996) therefore taking a shorter period of time to produce large crawlaways when compared to the M ecotype, ensuring continual replenishment of the population.

However, Svensson (1997) recorded that there were fewer numbers of *L. saxatilis* found as shore height increased although no differentiation was made as to which ecotype was occurring at each tidal height during the investigation. This contrasts with the frequencies of the different ecotypes found during the current investigation. The data collected during the July survey suggests that over twice as many *L. saxatilis*, regardless of ecotype, were found at Site 1 than were found further down the shore at Site 2. This could be explained by the numbers of *Carcinus maenas* present at Site 2 and the distinct lack of the predator at Site 1. A higher number of predators could have a negative effect on the population numbers of *L. saxatilis* during the summer months especially as the crabs will be feeding intensively in the summer months (Aagaard et al., 1995).

Another explanation for the differences in population density observed between the sites could be the difference in the substrate types. The H morphs were clumped in terms of their distribution with animals occupying the available crevices in the rock which may provide protection against desiccation (Rafaelli & Hughes, 1978). In a series of manipulative field experiments involving the creation of new crevices, Emson & Faller-Frisch (1976) clearly demonstrated that the density of populations of *Littorina rudis* (=*L. saxatilis*) increased with crevice availability. However, in a similar study, Hughes and Roberts (1981) showed that whilst an increased number of crevices did
affect the number of snails seeking shelter in a local population, the overall total population densities were not affected.

Whilst the H population density increased with shore height, the M population at Site 2 showed the reverse pattern during July. This supports the findings of Svensson (1997) who recorded numbers of *L. saxatilis* to decrease with an increase in tidal height on the shore. At Site 2, the M morphs were found under boulders or on the boulders themselves and would be prone to crushing as the boulders moved during storms (Hughes & Roberts, 1981). The mobility of the substrate could also be one of the main reasons for the low densities of the M morph observed higher up the shore at Site 2. Here the boulders were small and highly mobile whereas further down shore the boulders became larger and more stable. The fact that we could not sample under the larger boulders may also under-estimate the population density of the M morph (Hughes & Roberts, 1981).

At both sites, during July there was a higher proportion of smaller *L. saxatilis* found than during November. In the study of the reproduction of the H and M morphs at Ravenscar (25 mile away from the current site) the brood pouches of both the H and M morphs contained embryo’s throughout the entire year, but a higher proportion of the H morph were reproductively active in May/June which would account for a higher number of smaller snails observed during July (Hull *et al.*, 1999a).

When compared to the July survey, the M ecotype showed an increase in population density at each tidal height at Site 2 and at Site 1 (apart from the *F. vesiculosus/S. balanoides* zone) during November. This could be due to a similar pattern of migration described by Takada (1992) who documented the consistent seasonal migration of a sub-population of *Littorina brevicula* in the intertidal zone on a rocky shore in Japan. Takada noted that the same group of snails migrated downward every winter and the same group of snails stayed in the upper zone in the winter. Transplanted snails moved back towards their place of capture which would suggest the snails actively selected their tidal zone in winter. Takada (1992) also reported that most of the snails under investigation copulated within each sub-population which led to the conclusion that reproductive isolation between the two sub-populations had been partly established by the dimorphic migration behaviour. As Hull *et al.*, (1996) found there is evidence of reproductive isolation in populations of *Littorina saxatilis* on the rocky
shores of North East England. Takada (1992) summarised that the migratory behaviour seen in *L. brevicula* is determined separately for each member of the sub-population, the behaviour may be genetically controlled, and the behavioural dimorphism may be maintained by partial reproductive isolation between the two sub-populations. This pattern of migration could explain the increase in numbers of the M ecotype recorded during the November survey with returning snails increasing the numbers within that sub-population.

The higher numbers of the M ecotype found during the November survey could also be accounted for by the low dispersal rates of the species in question. Janson (1983) published literature stating distances of 1 – 4 metres travelled over 3 months. This distance is more than sufficient to alter the number of individuals found in a random quadrat at a given transect distance down the shore if *L. saxatilis* were leaving or returning to the area being sampled. The period between each survey provides ample time for the snails to travel back to the sample site. Also, due to the migration of the larger members of the *C. maenas* population into the deeper water for the winter months, there will be a decreased pressure from the predation of the snails from the crab. This will ultimately lead to an increase in numbers of the prey species.

During both survey periods only one H ecotype *L. saxatilis* was found at Site 2 with a shell height of 6mm (Fig 2.8) which could have possibly been moved between sites due to wave activity. However, no such pattern was observed in the H ecotype at Site 1 as there was no significant difference in snail abundance between months. A similar pattern in a cliff population of *Littorina rudis* (= *L. saxatilis*) was observed by Hughes & Roberts (1981) who demonstrated that overall population densities in the cliff populations remained relatively constant throughout the year.

The densities of both the H and M ecotypes of *L. saxatilis* recorded on the rocky shore of Filey Brigg were well below those recorded by authors in past investigations (Moore, 1940; Spooner & Moore, 1940; Thorson, 1941) during July and November, 2007, although the abundance within populations did fit in with the results from Berry (1961), with greater densities being found towards the upper limits of *L. saxatilis*’ range than at more seaward levels.
2.6.2 Distribution and density of *Carcinus maenas*

One of the most striking findings of the study was the total absence of *Carcinus* from Site 1 during the surveys. Personal observations made during visits to Filey Brigg on at least two occasions, saw a relatively small (<20mm carapace width) individual *C. maenas* up in the rock face at Site 1 close to the crevices the H ecotype inhabits. These individuals could have been placed in that area by visitors to the shore during the height of the tourist season as there was crevice or shelter near the *Carcinus*. As previously documented (Hull *et al.*., 1996) the H ecotype inhabits the upper reaches of the rocky shore environment. The high shore crevice environment might not be a suitable habitat for *Carcinus maenas* due to the lack of available shelter from predation (Emson & Faller-Fritsch, 1976). During the survey carried out in July 2007, there were also no *Carcinus maenas* found in the *Pelvetia* zone of the Site 2 boulder field, which could be explained by the very low abundance of *L. saxatilis* (Fig. 2.2) as a food source, or to a greater extent, a less favourable habitat for the shore crab due to fewer small rocks to use as shelter from predators such as sea birds.

Warman *et al.*, (1993) observed high numbers of *C. maenas* within the intertidal zone of the shore during the period of low tide and presumed the crabs were foraging around the area of the shore in which they shelter. During the investigation, Warman *et al.*, (1993) recorded that larger crabs were found no further up the shore than the *Fucus* sp. belt in the mid-shore, and that, unlike their smaller counterparts, crabs with a carapace over 35mm in width were scarcer in the shallow water on the ebb tide than the flood tide which is consistent with Naylor’s (1985) suggestion that endogenous tidal rhythmicity determines the migratory behaviour and cessation of locomotory activity in the anticipation of low tide. In the current investigation, the largest crabs were found in the *F. spiralis* zone at Site 2 with the smaller size classes being found equally within the *F. vesiculosus/S. balanoides* and the *F. vesiculosus/F. serratus* zones further down the shore.

The absence of *Carcinus maenas* during the survey carried out in November can be explained by the annual migration from the higher intertidal zone of the shore into the deeper water below mean low water (Naylor, 1962., Hunter & Naylor, 1993) which is a similar trait possessed by *Cancer pagurus* as described by Williamson (1900) and Meek (1913).
The results of Warman et al., (1993) illustrate that the behaviour of male and female crabs in the population vary considerably on both a seasonal and a tidal basis. Approximately 70% of the crabs observed during the investigation, either by diving or by capture in baited pots within the intertidal and the shallow sub-tidal zone, were male. With the reasonable assumption that there is a 50:50 sex ratio in the total population, the suggestion is that a high proportion of the females avoid the intertidal and shallow sub-tidal zones of the shore and generally forage less intensively than males. Previous work carried out by Edwards (1958), Naylor (1962) and Crothers (1968) support this suggestion stating that in general, adult female Carcinus maenas appear to undertake far less frequent and less intensive intertidal migrations than the males of the species. The results of these previous investigations can explain why the vast majority of the crabs collected from Filey Brigg for the current investigation were males found underneath rocks and in shallow intertidal pools.

The percentage of L. saxatilis shells showing external signs of damage at Site 2 during the July survey showed a definite pattern of increased amount of damage with increased distance down the shore. None of the shells found in the Pelvetia zone showed signs of damage with a steady increase in total percentage damage with an increase in the percentage of damage caused by C. maenas with the transition into each zone down the shore (Fig 2.9). During the survey carried out in November the Pelvetia zone produced shells that showed signs of damage but still no crab damage. Therefore some of the damage to the M ecotypes observed in the field is likely to be the result of boulder movement during storm events (Janson, 1983) rather than repairs to the shell after damage caused by crabs (Vermeij, 1982). However, it is very clear that the proportion of damaged shells is far higher at Site 2 (which contains the highest crab densities during the summer months) than at Site 1. The relatively low incidence of crab damage at Site 1 could be due to the lack of predators inhabiting that area or the possibility that the thin-shelled high shore H ecotype is easier to crush than a thick-shelled M ecotype (Hull et al., 1999a). Predators would possibly be less successful in breaking open the M ecotype shell due to its increased thickness (Hull et al., 1996) and studies have shown that thicker shelled littorinids often escape crab predation (Raffaelli, 1977; Johnannesson, 1986). Therefore, the very low incidence of shell injury at Site 1 may not just reflect a lower predation rate but may also reflect a higher predator success rate with the H ecotype as a prey item as the snails would not survive the attack.
Chapter 3

Optimality of feeding by *Carcinus maenas* on *Littorina saxatilis*.

3.1 Optimality

The feeding activities of any animal have consequences that ramify deeply into theoretical and conceptual developments of population, community and evolutionary ecology (Hughes, 1980).

A major factor in the energy budget of a consumer, especially a predator, is the energy expenditure entailed during feeding (Owen, 1980). Where some predators, along with most plant feeders, can feed whilst still being surrounded by their food source, many predators will have to find, stalk, chase and capture before the consumption of their prey. Such an attack may also prove to be unsuccessful. As all food gathering activities require the expenditure of energy, the calorific content of the food being gathered must be high enough to result in a net gain for the animal to ensure suitable fitness levels for daily activities and ultimately survival. To a certain extent, this can determine what a predator will accept as prey and partially explains feeding preference: the greater the energy yield in a single attack, the greater the gain in energy and nutrients (Owen, 1980).

Pyke (1984) states that the Optimal Foraging Theory is based on a number of assumptions combined in the attempt to predict the foraging behaviour of animals:

- The contribution (i.e. fitness) to the next generation from the individual when foraging, with the more favourable foraging traits being retained by future generations.
- There should be a heritable part of foraging behaviour, i.e. animals that forage in a certain way should produce offspring that forage in the same manner. The heritable part of the behaviour can be the rules by which the animal learns to make foraging responses, or the actual responses themselves.
- The currency of fitness is known. This is the relationship between foraging behaviour and fitness which will include a time scale. Fitness maybe assumed to be a function of a rate of some kind.
Evolution of foraging behaviour is not affected by genetic constraints and that mutations will eventually occur to avoid genetic linkages that prevent the progression of foraging behaviour.

Progression is determined by realistic functional constraints and that the information storing and processing capability of the animal must also be known as well as the morphologies and physical characteristics of the animal.

The foraging behaviour of an animal evolves at a quicker rate than the rate at which the conditions change. This would suggest that foraging behaviours change and maximise the animals’ expected fitness and in turn, forage optimally.

In the past, authors have produced conflicting opinions as to how animals solve their foraging problems and which mechanisms they employ for this purpose. Janetos & Cole (1981) and Myers (1983) argue that animals are equipped with ‘less than perfect’ rules which they follow when foraging. Take the largest, for example, may be an efficient method of foraging in a certain situation but may not be as beneficial in net gain to the animal as taking the optimal prey item would be.

On the other hand, ‘take the largest’ and other ‘rules’ the animals follow are seen by some authors (Krebs et al., 1983; Cheverton et al., 1985) to be the optimal method of foraging within a larger number of constraints e.g. Sensory, behavioural etc (Stephens & Krebs, 1986).

Stephens & Krebs (1986) suggested that if physiological and behavioural traits limit a forager’s ability to assess its environment, other ‘rules of thumb’ may be employed by the animal and ‘patch-leaving’ investigations carried out by various authors have produced descriptions of the different rules being followed in the situations that require them:

Firstly, Cowie & Krebs (1979) described a frequency rule; ‘leave after catching \( n \) prey’, Gibb (1958) described a duration rule; ‘leave after \( t \) seconds’, Krebs (1973) described a leaving time rule; ‘leave after \( g \) seconds of unsuccessful searching’, and Krebs et al., (1974) described a value rule; ‘leave when instantaneous intake rate drops to a critical value \( r \).

Hughes & Elner (1978) worked using Carcinus maenas (L.) as the predator and Mytilus edulis (L.) as the prey item to determine if the predator took the most profitable
prey item during feeding trials. They found that, for a given crab size, there was an optimal mussel size at which the prey value was at a maximum and that the optimal mussel size increases with crab size. Crabs were observed to handle prey items for 1-2 seconds before accepting or rejecting an individual as a potential prey item and that, with unlimited prey available, crabs chose items close to the optimal. As optimal prey items decrease in number crabs chose the item next closest to the optimum size, eventually feeding on unprofitable prey after all profitable items had been removed.

3.2 Aims

The aims of this part of the investigation are:

1) To ascertain the calorific value of the H and M ecotypes of *L. saxatilis* so that their prey value could eventually be calculated.

2) To determine the order and frequency of handling techniques used by the predator *C. maenas* when feeding on the H and M ecotypes of *L. saxatilis*.

3) To determine if *C. maenas* develops a preference for either ecotype of *L. saxatilis* after the feeding trials.

4) To record whether there are changes in the handling time by *C. maenas* with increased experience of *L. saxatilis*.

For ease of interpretation, the Method and Results section of this Chapter has been divided into two sections. Section A presents the findings of the preliminary experiments and analysis the results of which formed the basis for Section B the main experimental methods and findings.
3.3 Section A Methodology

3.3.1 Determination of the size class of the two ecotypes of *Littorina saxatilis* to be presented to the different size classes of *Carcinus maenas* in future feeding trials.

From the samples of *L. saxatilis* collected from the rocky shores, the largest and smallest individuals, with respect to shell height (mm), from both ecotypes were identified and measured using digital callipers. The height of the shell was deemed to be the distance between the bottom of the lip of the aperture up to the top of the apex of the whorls, similar to ‘Columella length’ as described in Clarke *et al.*, (1999). The size classes were determined by dividing the difference between the smallest and the largest shell heights measured in the stock populations of H and M ecotypes of *L. saxatilis* into equal sized size classes (Table 3.1). All *L. saxatilis* were assigned to these size classes for the remainder of the investigation. Both the H and M ecotypes were divided into 5 size classes, spanning a total shell height range of 10 mm. Any snails encountered falling outside of these size classes were not used during the investigation.

Table 3.1 Size Classes of H and M ecotypes of *L. saxatilis*.

<table>
<thead>
<tr>
<th>H ecotype</th>
<th>M ecotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>Size class</td>
<td>Shell Height</td>
</tr>
<tr>
<td>1</td>
<td>2-3.9mm</td>
</tr>
<tr>
<td>2</td>
<td>4-5.9mm</td>
</tr>
<tr>
<td>3</td>
<td>6-7.9mm</td>
</tr>
<tr>
<td>4</td>
<td>8-9.9mm</td>
</tr>
<tr>
<td>5</td>
<td>10-11.9mm</td>
</tr>
</tbody>
</table>

A total of twenty-six individual *Carcinus maenas* were collected from Filey Brigg and taken back to the aquarium. The carapace width of each crab was measured using digital callipers and crabs were assigned to size classes (Table 3.2). Crabs in the same size class were kept separately in aquaria in order to try and decrease the chance of cannibalism. This was especially necessary for the smallest size class (see Moksnes *et al.*, 1998). The sizes below represent the sizes commonly found in the intertidal area at Filey Brigg. Larger crabs were not found in the area where *L. saxatilis* occurred on
the boulders. Whilst crabs may move up the shore at high tide (Hunter & Naylor, 1993) the current study focussed on the crab sizes found during the field surveys (Chapter 2).

Table 3.2 Size classes of *Carcinus maenas* used in feeding trials

<table>
<thead>
<tr>
<th>Size class</th>
<th>Carapace Width</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1</td>
<td>40 + mm</td>
</tr>
<tr>
<td>C2</td>
<td>30 – 39.9 mm</td>
</tr>
<tr>
<td>C3</td>
<td>20 – 29.9 mm</td>
</tr>
<tr>
<td>C4</td>
<td>&lt; 20 mm</td>
</tr>
</tbody>
</table>

A randomly selected individual from the *C. maenas* size class C1 was separated from the rest of that size class by a sheet of ‘Corri-flute’ and presented with a selection of 5 *L. saxatilis*, (one from each size class) of the H ecotype. In no particular order with regards to shell height, the five snails were placed with the aperture facing downwards, in a line directly in front of the crab parallel to the face of the Perspex tank in order to eliminate any bias as to which snail size was encountered first. At the first contact made with the shell by the chela, a stop watch was started. As soon as the first shell had been crushed, the flesh had been eaten, and the crab was no longer in contact with the shell, that trial was deemed to be over. This was seen to be an appropriate conclusion as, similar to Cunningham & Hughes (1984) where the trial involving *C. maenas* was over as soon as the breaking time of continuously replaced dogwhelk shells stopped decreasing. In the current investigation *C. maenas* was only receiving one prey item per trial which is ultimately over when the shell has been crushed with the flesh being consumed, or, in the case of an unsuccessful trial, *C. maenas* discards the shell and shows no further interest in the individual. The time taken and the size class of the snail eaten were recorded.

This process was repeated five times for each size class of crab (a new crab for each trial) and for the entire experiment repeated for the M ecotype of snail. If a trial ended with an unsuccessful attempt in breaking the shell and the crab moved onto a different snail, the size of snail the crab had spent the longest duration of time trying to break into was recorded as this would suggest that the crab had evaluated the shape and size of the *L. saxatilis* and prioritised it as the most suitable prey item (Mascaro & Seed, 2001). The size of snail that the crabs from each size class attacked the most frequently
or spent the most time trying to break into during an unsuccessful attack, were to be presented to that crab size for the remainder of the investigation. Histograms were produced to show the percentage of each size class of *L. saxatilis* attacked by each size class of *C. maenas* during the preliminary feeding trials.

In order to test the null hypothesis that there was no significant preference for a particular size class of *L. saxatilis* (either H or M ecotype) by a particular size class of *C. maenas* a Chi-square test was applied to the data. The Chi-square test was used the data was frequency based (Dytham, 2003). The same procedure was used for the data obtained from the preliminary feeding trials for both the H and M ecotypes.
3.3.2 Determination of the Energy Content of the two ecotypes of *Littorina saxatilis* (Olivi)

Samples of *Littorina saxatilis* of each ecotype (H and M) were collected from the rocky shore habitats of Filey Brigg (British National Grid reference TA131815) and Thornwick Bay at Flamborough (British National Grid reference TA226704). For site description of Filey Brigg see Methodology in Chapter 2.

The area at Thornwick Bay from which the samples of the H ecotype of *L. saxatilis* were collected comprises of a vertical rock face in the splash zone rising approximately 3 – 4 metres in height from the bedrock at the upper most limit of the high water mark. The rock face has large crevices running randomly throughout the outward surface. Temporary rock pools and small boulders are present at the foot of the rock face.

The samples of the M ecotype of *L. saxatilis* to be involved in the determination of the energy content phase of the investigation were collected from an area of Thornwick Bay with a more horizontal gradient beginning approximately 3 – 4 metres away from the foot of the vertical rock face described previously. Boulders between 20 cm and 50 cm in diameter lie scattered across the ground and occupy the majority of the available space on the bedrock down to the low water mark.

The samples of *L. saxatilis* collected from each shore were submerged in boiling water for a period of 5 minutes to kill the organisms as humanely as possible and to make sure the flesh came away from the inside of the shell for ease of extraction (Horton, 1980). After the submersion period, the samples were placed into a freezer and kept at -20°C for 7 days.

The samples were taken from the freezer and were allowed to thaw. Once thawing had occurred, the soft body of *L. saxatilis* was removed using watch-maker forceps and placed onto a sheet of aluminium foil. The height and width of the shell from which the body mass was removed was recorded using hand held digital callipers. This process was repeated for as many individuals of each ecotype from each shore as possible, but not more than 100. The aluminium foil holding the soft *L. saxatilis* bodies was placed in an oven at a temperature of 50°C for 24 hours, half as long as *Nucella lapillus* in Lawton & Hughes (1985) investigation as these prey items were 25 mm in
height which is almost twice the size of the largest individual *L. saxatilis* from the populations of H and M ecotype involved in the investigation.

The dry body masses of the snails were weighed using a Salter ER-180A electronic weigh-measure. The energy content of each snail was calculated using the correction factor of 25.94 kJ (g dry wt)$^{-1}$ (Grahame, 1970 in Elner & Raffaelli, 1980).

For each of the four populations collected (H and M from Filey Brigg and H and M from Thornwick Bay) scatterplots were produced to give an indication of the energy content of a snail at a given shell height. Analysis was performed on the log transformed data as this produced the highest values (coefficient of determination) (Fowler *et al.*, 1998). In all cases the data showed no significant departure from a normal distribution (Kolmogorov-Smirnov test, $P > 0.05$ in all cases) therefore Pearson’s Correlation coefficient was used to determine if there was a significant correlation between log shell height and log energy content for all four populations (Fowler *et al.*, 1998). As all correlations were significant, linear regression analysis was performed using the method of Least Squares in order to describe the relationship between the independent (log shell height) and the dependent (log energy per gram dry mass) variables (Zar, 1998). The regression analysis produced an equation in the form $y = mx + c$ which described the relationship between the independent and dependent variables in terms of the slope and intercept of the fitted line (Zar, 1998). Once regression equations had been produced for each population, the null hypothesis that there was no statistically significant difference in slopes and intercept between the regression lines generated for H and M populations within a shore was tested. The slopes of the two regression equations were compared using a modified Student t-test and elevations by calculating a common regression for the two data sets and then applying a modified Student t-test as shown in Zar (1998). The same procedure was also used to test the null hypothesis that there was no significant difference in slopes and intercept between H or M populations from different shores.
3.4 Section A Results

3.4.1 Determination of the size class of the two ecotypes of *Littorina saxatilis* to be presented to the different size classes of *Carcinus maenas* in future feeding trials

There was a significant difference between the sizes of the H ecotype of *L. saxatilis* attacked by each size class of *C. maenas* during the preliminary feeding trials (Chi = 41.029, df = 9, P < 0.001). Figure 3.1 shows that *C. maenas* from the C1 size class mostly attacked and ate individuals from the H5 size class (80%) and occasionally attacked individuals from the H4 size class (20%) whereas C2 size class crabs mostly attacked and ate individuals from the H4 size class (80%) and occasionally attacked individuals from the H3 size class (20%) (Fig 3.1). Crabs from the C3 size class attacked and ate individuals from the H3 size class (100%) whereas C4 crabs size class mostly attacked individuals from the H2 size class (80%) and occasionally attacked individuals from the H3 size class (20%).

![Figure 3.1 Histogram showing percentage of each *L.saxatilis* (H ecotype) size class chosen by different size classes of *C.maenas* during preliminary trials](image)

There was a significant preference for size class of the M ecotype of *L. saxatilis* attacked by each size class of *C. maenas* during the preliminary feeding trials (Chi = 32.8, df = 9, P < 0.001). Crabs from the C1 size class mostly attacked and ate individuals from the M4 size class (80%) and occasionally individuals from the M3 size class (20%) (Fig 3.2) whereas C2 size class crabs mostly attacked and ate individuals from the M3 size class (80%) and occasionally attacked individuals from the M2 size.
class (20%) (Fig 3.2). Crabs from the C3 size class mostly attacked and ate individuals from the M2 size class (60%) and occasionally attacked individuals from the M3 and M1 size classes (20% and 20% respectively). Crabs from the C4 size class mostly attacked and ate individuals from the M1 size class (80%) and occasionally attacked individuals from the M2 size class (20%) (Fig 3.2).

**Figure 3.2 Histogram showing percentage of each *L. saxatilis* (M ecotype) size class chosen by different size classes of *C. maenas* during preliminary trials**

Table 3.2 shows the size class of each ecotype of *L. saxatilis* that was to be presented to the different size classes of *C. maenas* during the feeding trials as a result of the preliminary investigation.
Table 3.2 Size of *L. saxatilis* to be presented to *C. maenas* size class in further studies

<table>
<thead>
<tr>
<th>Crab Size (Carapace Width)</th>
<th>H ecotype</th>
<th>M ecotype</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1 (&gt;40mm)</td>
<td>10-11.9mm (5)</td>
<td>9-10.9mm (4)</td>
</tr>
<tr>
<td>C2 (30-39mm)</td>
<td>8-9.9mm (4)</td>
<td>7-8.9mm (3)</td>
</tr>
<tr>
<td>C3 (20-29mm)</td>
<td>6-7.9mm (3)</td>
<td>5-6.9mm (2)</td>
</tr>
<tr>
<td>C4 (&lt;20mm)</td>
<td>4-5.9mm (2)</td>
<td>3-4.9mm (1)</td>
</tr>
</tbody>
</table>

3.4.2 Determination of the Energy Content of the two ecotypes of *Littorina saxatilis* (Olivi).

In all four populations, there was a significant strong positive correlation between log shell height and log energy per gram dry mass (P < 0.001 in all cases) (Table 3.3). There was also a significant relationship between log shell height and log energy content per gram dry mass and the % values indicated that most of the variation in log energy content could be accounted for by the regression of this variable on log shell height (Table 3.3).

Table 3.3 Summary of the results of the Pearson’s Correlation and linear regression analysis for all four *L. saxatilis* populations

<table>
<thead>
<tr>
<th>Population</th>
<th>df</th>
<th>Pearson’s Correlation Coefficient</th>
<th>Regression</th>
<th>ANOVA</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>P</td>
<td>df</td>
<td>F value</td>
</tr>
<tr>
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<td>983.71</td>
</tr>
<tr>
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<td>1.63</td>
<td>666.38</td>
</tr>
<tr>
<td>H Thornwick</td>
<td>93</td>
<td>0.942</td>
<td>1.93</td>
<td>747.23</td>
</tr>
<tr>
<td>M Thornwick</td>
<td>95</td>
<td>0.963</td>
<td>1.95</td>
<td>1210.87</td>
</tr>
</tbody>
</table>
The slopes and intercepts of the regression lines are also provided in Table 3.3 and Figure 3.3 is a scatterplot (with fitted regression line) for the H and M populations from Filey Brigg. It is apparent from Figure 3.3 that the H ecotype generally has a higher energy content per gram dry mass than the M ecotype. A comparison of the intercepts of the regression lines for the two ecotypes indicated that there was no significant difference in intercepts (modified Student t-test, \(t = 0.53\), \(df = 163\), \(P > 0.05\)) however there was a significant difference in the slope of the two regression lines (modified Student t-test, \(t = 5.49\), \(df = 155\), \(P < 0.001\)). The H ecotype therefore has a higher energy content per unit shell height than the M ecotype.

![Figure 3.3 Scatterplot showing log energy per gram dry mass versus log shell height (mm) for Filey Brigg H and M ecotypes.](image)

Figure 3.4 is a scatterplot (with fitted regression line) for the H and M populations from Thornwick Bay. Again it is apparent from Figure 3.4 that the H ecotype generally has a higher energy content per gram dry mass than the M ecotype. A comparison of the intercepts of the regression lines for the two ecotypes indicated that there was no significant difference in either the intercepts (modified Student t-test, \(t = 0.162\), \(df = 185\), \(P > 0.05\)) or the slope of the two regression lines (modified Student t-
test, \( t = 0.707, \) \( df = 184, P > 0.05 \). The H ecotype therefore does not have a higher energy content per unit shell height than the M ecotype.

![Graph showing log energy per gram of dry mass versus log shell height (mm) for Thornwick Bay H and M ecotypes](image)

**Figure 3.4 Scatterplot showing log energy per gram of dry mass versus log shell height (mm) for Thornwick Bay H and M ecotypes**

When the two M populations from the different shores were compared there was no significant difference in either the intercepts (modified Student t-test, \( t = 0.163, df = 185, P > 0.05 \)) or the slope of the two regression lines (modified Student t-test, \( t = 0.707, df = 184, P > 0.05 \)). The M ecotype from Filey Brigg therefore does not have a higher energy content per unit shell height than the M ecotype from Thornwick Bay. A similar pattern was seen when the H ecotypes from the two shores were compared. There was no significant difference in either the intercepts (modified Student t-test, \( t = 1.01, df = 191, P > 0.05 \)) or the slope of the two regression lines (modified Student t-test, \( t = 0.969, df = 190, P > 0.05 \)).

Subsequent data analysis therefore was performed on the data pooled by snail ecotypes as there was no significant difference in the slope or intercept of the regression lines between the ecotypes from the different shores.
3.5 Section B Methodology

3.5.1 Feeding Behaviour of *Carcinus maenas*.

After a two day starvation period to standardise hunger levels (Hughes & Elner, 1979), an appropriately sized individual *L. saxatilis* from the size ranges of the H ecotype was presented to an individual from the corresponding size class of *Carcinus maenas* determined during the preliminary feeding trials (see Table 3.2).

Each individual test crab was separated from the others in a large tank using four A4 size laminating envelopes stuck together to form a solid but flexible barrier to prevent any interaction between individuals that may affect the feeding behaviour. Once the crab had settled in the feeding area of the tank, an individual *L. saxatilis* from the corresponding size class was presented to the crab. The entirety of the attack, from first contact to the discarding of the shell, was recorded, from the front of the tank onto VHS and video cassette. The use of video cameras to record feeding behaviour has been widely used for different subject species; *Salvelinus alpines* (Malmquist, 1992), *Clupea harengus* (Batty et al., 1990), many copepods (Tiselius & Jonsson, 1990) and *Pleuronectes platessa* and *Solea solea* (Batty & Hoyt, 1995). For a given size class of crab, this procedure was repeated twenty times for the H ecotype followed by twenty individuals from the M ecotype. After the C1 sized crab had been involved in the first feeding trial with an individual from the appropriate size class of the H ecotype of *L. saxatilis* the smaller sizes of crab (C2 – C4) were presented with an individual from the H ecotype. As C1 was being recorded, C2 – C4 were left un-disturbed. When C2 was being recorded C1, C3 and C4 were left un-disturbed and so on. This rotation of feeding continued until *C. maenas* of each size class had experienced three individuals from the H ecotype. There was then a two day starvation period before the next bout of feeding trials involving three individuals from the H ecotype of *L. saxatilis*.

On completion of the feeding behaviour trials, the video recordings were played back and observed. During the play-backs, the behaviour and the actions displayed by the crabs were recorded as well as the duration of each behaviour.

As there was such a high frequency of unique behaviours displayed by *C. maenas* during the feeding trials, these were divided into a smaller number of behavioural terms for ease of analysis. The behavioural terms are listed below:
Pick-up: Any behaviour involving either the master or minor chela to pick up *L. saxatilis* during the feeding trial.

Minor Hold (MinHold): Any behaviour involving only the minor chela to hold *L. saxatilis* during the feeding trial.

Master Hold (MasHold): Any behaviour involving only the master chela to hold *L. saxatilis* during the feeding trial.

Mouth: Any behaviour involving *L. saxatilis* being held using only the mouth parts.

Taste: Any behaviour involving the aperture of *L. saxatilis* being held over the mouth of *C. maenas*. This can involve either the master or minor chela or both chela simultaneously.

Orient: Any behaviour involving *L. saxatilis* being manipulated into a different position. This can involve either the master or minor chela or both chela simultaneously.

Roll: Any behaviour involving either the master or minor chela to roll *L. saxatilis* up the sternum of *C. maenas*.

Crush: Any behaviour involving either chela to crush around the shell of *L. saxatilis*.

Pull: Any behaviour involving the flesh of *L. saxatilis* being held in the mouth of *C. maenas* and either, or both chela, gripping the shell and pulling away from the crab.
Master Minor Hold (MasMinHold): Any behaviour involving both chela to hold *L. saxatilis* during the feeding trial (does not include when both chela are involved in a ‘Pull’ behaviour).

Eat: Any behaviour involving the flesh of *L. saxatilis* being eaten by *C. maenas*.

Peel: Any behaviour involving the master chela breaking pieces of shell away from the whorls of the *L. saxatilis* shell in a peeling motion.

Discard: Any behaviour involving the mouth parts or either chela to drop or push away, broken or empty *L. saxatilis* shells.

Saw: Any behaviour involving the teeth on the articulatory dactylus of the master chela being positioned on the *L. saxatilis* shell and pushed backwards and forwards in a sawing motion.

For a full list of behaviours see Appendix I.

The timing of each trial began when the first chelal contact was made with the *L. saxatilis* shell, and ended when either: the broken, empty shell had been discarded by the crab, or, if an attack was unsuccessful, the time had exceeded that of the longest duration of a successful attack for a crab of that size. During many trials involving the M ecotype, *C. maenas* dropped the snails, moved away from the shell and then returned and continued to attempt to break into the shell. In the event of this happening during the feeding trial, the time between dropping the shell and the next chelal contact was subtracted from the total handling time for that trial. This was seen to be acceptable as in Cunningham & Hughes (1984) the total handling time for dogwhelks was recorded as total breaking time plus eating time. Because *C. maenas* was neither breaking the shell nor eating the flesh of *L. saxatilis* during the periods when the shell had been dropped, it was not included in the calculations for the total handling time of the feeding trials. The variables recorded during the play back of the feeding trials were:
• The total handling time by *C. maenas* (the time in seconds from the first chelal contact with the shell until discarded with no further interest shown)
• The time when the first ingestion of the flesh of *L. saxatilis* by *C. maenas* took place,
• The time at which eating ceased (when the flesh had been consumed and the mandibles of *C. maenas* ceased moving)
• The total duration of eating (the time in seconds from first ingestion of the flesh of *L. saxatilis* to when *C. maenas* ceases eating)
• The number of attempts made by *C. maenas* to break into the shell (deemed to be when the crab performed an action similar to previously recorded actions that caused damage to the shell)
• The time taken until the first successful breaking of the shell (either completely crushed or a when a piece had been broken off).

In order to test the null hypothesis that there was no significant difference in mean total handling time of crabs foraging *L. saxatilis*, with either crab size (fixed factor with four levels) or with snail ecotype (fixed factor with two levels) a General Linear Model (GLM) analysis of variance (ANOVA) was applied to the data. The null hypothesis that there was no significant interaction between the size of the crab and the ecotype of the snail was also tested. GLM was deemed appropriate as the data conformed to a normal distribution (Kolmogorov-Smirnov test, $P > 0.05$ in all cases) and the variances could be considered equal (Levene's test, $P > 0.05$) (Underwood *et al.*, 1991). Where the result of the ANOVA was significant, a pair-wise Tukey test was applied to the data as a *post hoc* test in order to determine the source of the significance between the means (Fowler *et al.*, 1998).

As the data for both the number of attempts to break into the shell of a *Littorina saxatilis* and the time to the first break of the shell during a foraging bout did not conform to a normal distribution (Kolmogorov-Smirnov test, $P < 0.01$ in both cases and the variance of both the data sets could not be considered equal (Levene's test, $P < 0.01$ in both cases) a two-way GLM ANOVA could not be used to analyse the data. Instead the null hypothesis that there was no significant difference in the median number of attempts to break into the shell with crab size as a factor was analysed using the non-parametric Kruskal-Wallis test which is suitable for data which does not conform to a
normal distribution (Fowler et al., 1998). A Mann-Whitney U-test was used to test the null hypothesis that there was no significant difference in median number of attempts to break into the shell between ecotypes (Fowler et al., 1998).

Crabs of the size class C1 were the only ones able to break the shells of the M ecotype, the smaller crabs (C2-C4 size classes) all failed to break into the M shells during the experiment time period. Therefore only the data from the C1 size class of crab could be used to test the null hypothesis that there was no significant difference in median time to the first break for the C1 size class of C. maenas when feeding on either the H or M ecotype of L. saxatilis. As the data did not conform to a normal distribution (Kolmogorov-Smirnov test, P < 0.05) a Mann-Whitney U-test was used to test the null hypothesis as it is more applicable for the comparison of two populations of data that do not conform to a normal distribution (Fowler et al., 1998).

To test the null hypothesis that there was no significant difference between the order and frequency of behaviours used by C. maenas during the feeding trials on both ecotypes of L. saxatilis a modified Chi square test was applied to the data as described by Clark (1994). The preceding and following behavioural events were organised into a transitional probability matrix in which each cell represented the total acts of behaviour following behaviour (Table 3.4). The percent (P) for each transition can be calculated by dividing the total acts of behaviour b by the corresponding row total. The expected values for each cell were calculated by multiplying the column frequency by corresponding row total (example from Table 3.4: column frequency for Minor Hold = 0.274; row total for Pick-up = 97; expected for cell Pick-up/Minor Hold = 0.274 x 97 x 26.578, etc). Using Yates’ correction for estimating the individual χ² values for each cell in a row, the total row χ² value (with d.f. = 14) could be generated. Table 3.4 shows the sum of the row χ² values is equal to the χ² value for the entire matrix.
3.5.2 Prey value of *Littorina saxatilis* ecotypes

The shell height and energy g\(^{-1}\) dry mass data from the two shores was pooled by ecotype and a regression analysis performed as outlined in Section A. The new regression equations were then used to predict the energy g\(^{-1}\) dry mass for any size of H or M *Littorina saxatilis* used in the feeding trials. In order to calculate the prey value for each snail fed upon by the crabs, the energy g\(^{-1}\) dry mass data obtained from the regression model was divided by the handling time (Elner & Hughes, 1979). The C1 size class of crabs was the only size class which managed to break into and eat the flesh of the M ecotype. Where crabs did not successfully manage to forage on a snail during the experimental period, the prey value data was given a value of zero as no resources had been obtained.

In order to test the null hypothesis that there was no significant difference in mean prey value of the *L. saxatilis* attacked by the crabs with either crab size (fixed factor with four levels, C1-C4) or with snail ecotype (fixed factor with two levels, H and M) a General Linear Model (GLM) analysis of variance (ANOVA) was applied to the data. The null hypothesis that there was no significant interaction between the size of the crab and the ecotype of snail was also tested. The GLM was deemed appropriate as the data conformed to a normal distribution (Kolmogorov-Smirnov test, P > 0.05) and the variances could be considered equal (Levene’s test, P > 0.05) (Underwood *et al.*, 1991). Where the result of the ANOVA was significant, a pair-wise Tukey test was applied to the data as a *post hoc* test in order to determine the source of the significance between the means (Fowler *et al.*, 1998).
### Table 3.4 Transition matrix showing $\chi^2$ values for preceding and following behaviours during feeding trials involving the H ecotype of *L. saxatilis*

<table>
<thead>
<tr>
<th>H Preceding</th>
<th>Minor Hold</th>
<th>Master Hold</th>
<th>Mouth Hold</th>
<th>Taste</th>
<th>Orient</th>
<th>Roll</th>
<th>Pick-up</th>
<th>Crush</th>
<th>Pull</th>
<th>MasMin Hold</th>
<th>Eat</th>
<th>Peel</th>
<th>Discard</th>
<th>Saw</th>
<th>Row total</th>
<th>Pij</th>
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<td>0.16</td>
<td>4.5</td>
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<td>0</td>
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<td>1.044</td>
<td>0.048</td>
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<td>0</td>
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<td>0</td>
<td>0</td>
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<th>Master Hold</th>
<th>Mouth Hold</th>
<th>Taste</th>
<th>Orient</th>
<th>Roll</th>
<th>Pick-up</th>
<th>Crush</th>
<th>Pull</th>
<th>MasMin Hold</th>
<th>Eat</th>
<th>Peel</th>
<th>Discard</th>
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<td>15.68</td>
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<td>3.774</td>
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<td>2</td>
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<td>0.504</td>
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<td>3.774</td>
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</tr>
<tr>
<td>Taste</td>
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<td>0</td>
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<td>0</td>
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<td>0.0333333</td>
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<td>0.084</td>
<td>0.039</td>
<td>0.629</td>
<td>0.011</td>
<td>0.06</td>
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<td>0.225</td>
<td>3</td>
<td>0.001</td>
<td></td>
</tr>
<tr>
<td>MasMin Hold</td>
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<td>0.037</td>
<td>0.029</td>
<td>0.037</td>
<td>0.084</td>
<td>0.039</td>
<td>0.629</td>
<td>0.011</td>
<td>0.06</td>
<td>0.174</td>
<td>0.008</td>
<td>0.225</td>
<td>0.001</td>
<td>7</td>
<td>0.0023333</td>
<td></td>
</tr>
<tr>
<td>Peel</td>
<td>0.082</td>
<td>0.037</td>
<td>0.029</td>
<td>0.037</td>
<td>0.084</td>
<td>0.039</td>
<td>0.629</td>
<td>0.011</td>
<td>0.06</td>
<td>0.174</td>
<td>0.008</td>
<td>0.225</td>
<td>0.001</td>
<td>3</td>
<td>0.001</td>
<td></td>
</tr>
<tr>
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<td>0.3056667</td>
<td>0.196</td>
<td>0.091</td>
<td>1.46767</td>
<td>0.025667</td>
<td>0.14</td>
<td>0.406</td>
<td>0.0186667</td>
<td>0.525</td>
<td>3000</td>
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</tr>
<tr>
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<td>307</td>
<td>239</td>
<td>271</td>
<td>131</td>
<td>84</td>
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<td>0.02</td>
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<td>0.075</td>
<td>3000</td>
<td>0.0003</td>
<td></td>
</tr>
</tbody>
</table>

Total no. behaviours: 3000
3.5.3 Preference trial methodology

Using the experimental set-up as outlined in section 3.1, crabs from each size class of *C. maenas* were simultaneously presented with five individual snails from both the H and M ecotype of *L. saxatilis*. The trial began when *C. maenas* made first contact with a snail from the H or the M ecotype and ended when the flesh had been consumed after the successful breaking of the *L. saxatilis* shell. If an attack was unsuccessful, leading to a different shell being picked attacked, the shell that was broken into having the flesh eaten from inside was recorded. The ecotype chosen and consumed by *C. maenas* during each trial was recorded. The handling time of the preference trials was not recorded. This process was repeated five times for each size class of *C. maenas*. Histograms were created to show the frequency of each ecotype of *L. saxatilis* attacked by each size class of *C. maenas* during the preference trials and to show if the attacks made were successful or not.

To test the null hypothesis that there was no significant difference in the frequency of each ecotype attacked or the number of successful attacks made by the different sizes of *C. maenas* during the preference trials an rxc contingency Table Chi-squared test was applied to the data. The contingency Table Chi-Squared is often used to determine if there is a significant association (Dytham, 2003), therefore this test was applied to determine if there was a significant association between crab size and the ecotype of snail attacked or between attack success on each ecotype and crab size.

3.5.4 Changes in handling times with experience

A single crab of each size class was then used to determine if the crabs actually became more proficient at handling the prey items with experience. Using the methodology outlined in Section B 3.2, a single crab was repeatedly offered H ecotypes of *Littornia saxatilis* and the handling time or rate of abandonment of the snail was determined. The trials were then repeated with the M ecotype. After the C1 sized crab had been involved in the first feeding trial with an individual from the appropriate size class of the H ecotype of *L. saxatilis* the smaller sizes of crab (C2 – C4) were presented with an individual from the H ecotype. As C1 was being recorded, C2 – C4 were left un-disturbed. When C2 was being recorded C1, C3 and C4 were left un-disturbed and so on. This rotation of feeding continued until *C. maenas* of each size class had experienced three individuals from the H
ecotype. There was then a two day starvation period before the next bout of feeding trials involving three individuals from the H ecotype of *L. saxatilis*. After all twenty feeding trials had taken place and had been recorded, the same method was used for the feeding trials involving *C. maenas* and individuals from the M ecotype of *L. saxatilis*.

3.6 Section B Results

3.6.1 Behaviour of *Carcinus* feeding on *Littorina saxatilis*

Figure 3.5 shows the mean total handling time for each size class of crab feeding upon each ecotype of snail. There was no significant difference in total handling time between either snail ecotypes used for the investigation (*P* > 0.05; Table 3.4) nor crab size (*P* > 0.05; Table 3.5). The interaction term was also not significant indicating that the total handling time for the crabs of different sizes showed no significant difference between the ecotypes (Table 3.5).

Table 3.5 Summary of the results of the two factor GLM ANOVA examining the effect of snail ecotype and crab size on total handling time.

<table>
<thead>
<tr>
<th>Source</th>
<th>Df</th>
<th>Adjusted MS</th>
<th>F</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Morph</td>
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<td>7508</td>
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<td>0.606</td>
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<tr>
<td>Crab Size</td>
<td>3</td>
<td>25443</td>
<td>0.91</td>
<td>0.440</td>
</tr>
<tr>
<td>Morph * Crab Size</td>
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<td>60132</td>
<td>2.14</td>
<td>0.098</td>
</tr>
<tr>
<td>Error</td>
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<td>28098</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>159</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
There was a significant difference in the median number of attempts to break into a Littorina shell between the different crab size classes (Kruskal-Wallis, H) (adjusted for ties) = 9.60, df = 3, P = 0.022). The crabs from the smallest size class showed a greater number of attempts to break into the shells (Table 3.6).
Table 3.6 Median and range of the number of attempts made to break into *Littorina* shells of either ecotype with crab size

<table>
<thead>
<tr>
<th>Crab Size Class</th>
<th>Median number of attempts</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1</td>
<td>2</td>
<td>1-24</td>
</tr>
<tr>
<td>C2</td>
<td>5</td>
<td>1-31</td>
</tr>
<tr>
<td>C3</td>
<td>6.5</td>
<td>1-38</td>
</tr>
<tr>
<td>C4</td>
<td>8.5</td>
<td>1-33</td>
</tr>
</tbody>
</table>

When the number of attempts made to break into the shell was compared between ecotypes (across all crab sizes), there was a significant difference between the ecotypes (Mann-Whitney U-test, $W = 9170$, $df = 80.80$, $P < 0.0001$). The crabs made fewer attempts to break the shell of the H ecotype (median = 1, range 1-5) compared to the number of attempts made to break into the M ecotype (median = 15.5, range 1-38).

As previously stated, crabs from the smaller size classes (C2-C4) failed to break into the shells of the M ecotype during the experimental time period (Table 3.7). Therefore a comparison of the amount of time taken for the crab to break the shell of the two ecotypes could only be made for crabs from the C1 size class who had a success rate of 85% when attacking the M ecotypes (Table 3.7). There was a significant difference in the median time taken for the C1 crabs to break into the shells between the ecotypes (Mann-Whitney U-test, $W = 543$, $df = 20.20$, $P = 0.0003$). For C1 crabs, the time taken to break into an H ecotype was far quicker (median = 16.33s, range 11-27) than that taken to break into the M ecotype (median = 52.5s, range 1-202).
Table 3.7 Percentage of successful attacks for each *C. maenas* size class during feeding trials involving H and M ecotypes of *L. saxatilis*

<table>
<thead>
<tr>
<th>Crab size class</th>
<th>Percentage of attacks successful on H morphs (%)</th>
<th>Percentage of attacks successful on M morphs (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1</td>
<td>100</td>
<td>85</td>
</tr>
<tr>
<td>C2</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>C3</td>
<td>100</td>
<td>0</td>
</tr>
<tr>
<td>C4</td>
<td>100</td>
<td>0</td>
</tr>
</tbody>
</table>

3.6.2 Behavioural matrix analysis

The individual Chi-square results which contributed most to the overall significance of the Chi-squared analysis of the occurrence of the behavioural sequence analysis is presented in Table 3.8. Overall, the comparison of the sequence of the behaviours of the crabs feeding on the two ecotypes indicated that there were significant differences in the frequency of the behaviours exhibited in a particular sequence (Chi-Squared test, $\chi^2 = 8291.6$, df = 351, $P < 0.001$). A summary of the main similarities and differences in the behavioural sequences exhibited by *Carcinus* feeding on the two ecotypes is presented below. During attacks made on both the H and M ecotypes the behaviour ‘Pick Up’ was followed by ‘MinHold’ in frequencies significantly higher than expected indicating the crabs initially used the minor chela for manipulating the prey (Table 3.8 a) and b)). During attacks made on the H ecotype, ‘MinHold’ was followed by both ‘Eat’ and ‘Taste’ in frequencies higher than expected (Table 3.8 a) whereas for the M ecotype, ‘MinHold’ was followed by both ‘Orient’ and ‘Crush’ in frequencies significantly higher than expected (Table 3.8 b).

During attacks made on the H ecotype, ‘Roll’ followed by ‘MasMinHold’ as the crab re-positioned the snail and ‘Crush’ followed by ‘Eat’ and ‘Mouth’ as the crab started feeding all occurred in frequencies significantly higher than expected (Table 3.8 a). In contrast, a ‘Crush’ behaviour was followed by a ‘MinHold’ in frequencies higher than expected when crabs were feeding on the M ecotype whereas ‘Crush’ followed by both ‘Taste’ and ‘Crush’ occurred in lower than expected frequencies (Table 3.8 b). The crabs
followed an ‘Orient’ behaviour with a ‘Crush’ when feeding on the M ecotype in frequencies significantly higher than expected during attacks (Table 3.8 b).

A ‘Taste’ followed by the ‘Crush’ and the ‘Discard’ behaviour followed by a ‘Pick Up’ occurred significantly more frequently than expected during attacks made on the M ecotype as the crab tried to get into the shell and made repeated attempts even after dropping it. They very rarely exhibited the ‘Pull’ or ‘Peel’ behaviours when feeding on the H ecotype. Overall the pattern revealed was that of repeated attacks to gain access to the flesh of the M ecotype whereas after crushing the H ecotype the crab was able to feed.

Table 3.8 The significant adjusted Chi-square values calculated using the method of Clark (1994) when examining the frequencies of behaviours preceding and following a specific behaviour.

Table values calculated for crabs feeding on a) H and b) M *Littorina saxatilis*

### a) Feeding on H ecotype

<table>
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<tr>
<th>Preceding</th>
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<th>Actual Chi</th>
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<th>Expected</th>
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<tbody>
<tr>
<td>Pick Up</td>
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<td>60.536</td>
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<td>64.628</td>
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</tr>
<tr>
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<td>Taste</td>
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<td>64</td>
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</tr>
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<td>Roll</td>
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</tr>
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</tr>
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<td>MasHold</td>
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<td>7.4703</td>
</tr>
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<td>MinHold</td>
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Table 3.8 Continued

b) Feeding on M ecotype

<table>
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<th>Expected</th>
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<td>Orient</td>
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<td>69</td>
<td>24.453</td>
</tr>
<tr>
<td></td>
<td>Mouth</td>
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<td>115</td>
<td>44.613</td>
</tr>
<tr>
<td>Orient</td>
<td>Crush</td>
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<td>276.449</td>
<td>279</td>
<td>108.504</td>
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<td>4</td>
<td>35.772</td>
</tr>
<tr>
<td></td>
<td>Crush</td>
<td>56.439</td>
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<td>83.028</td>
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<tr>
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<td>45.484</td>
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<td>34.805</td>
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<td>Crush</td>
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<td>47</td>
<td>21.176</td>
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<td>MinHold</td>
<td>211.794</td>
<td>155</td>
<td>51.238</td>
</tr>
<tr>
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<td>Roll</td>
<td>160.01</td>
<td>14</td>
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<td></td>
<td>Peel</td>
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<td>5</td>
<td>0.093</td>
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<td>Discard</td>
<td>Pick Up</td>
<td>1035.097</td>
<td>22</td>
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<td>Pull</td>
<td>Peel</td>
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<td>MasMinHold</td>
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<td>0.008</td>
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<td>0.0023</td>
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<tr>
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<td>0.008</td>
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<tr>
<td></td>
<td>Saw</td>
<td>251</td>
<td>0</td>
<td>0.001</td>
</tr>
</tbody>
</table>

3.6.3 Prey value (J s⁻¹) of *Littorina saxatilis* ecotypes

The log shell height shell height (independent variable) and log energy g⁻¹ dry body mass (dependant variable) data for the H and M ecotypes from each shore was pooled and subjected to regression analysis. For the H ecotype, the regression of log energy g⁻¹ dry body mass upon log shell height was significant (Regression ANOVA, $F_{194} = 1227.5$, $P < 0.001$) and accounted for over 86% of the variation between the points ($= 86.4\%$). The
equation generated from the regression was as follows: \( \log \text{energy g}^{-1} \text{dry body mass} = -2.82 + 2.62 \log \text{shell height} \). A significant regression was also obtained for the M ecotype pooled data (Regression ANOVA, \( F_{1, 161} = 1608.5, P < 0.001 \)) and accounted for over 90% of the variation between the points ( = 90.9 %). The regression equation generated was as follows: \( \log \text{energy g}^{-1} \text{dry body mass} = -3.22 + 2.79 \log \text{shell height} \). These equations were then used to predict the log energy g\(^{-1}\) dry body mass for a known log shell height of snail predated upon by the crabs during the experiment. The log energy g\(^{-1}\) dry body mass values were then divided by the handling time to calculate the prey value (J s\(^{-1}\)) of the snail fed on by the crab. The calculated prey values were then used to test the null hypothesis that there was no significant difference in mean prey value of the \( L. \text{saxatilis} \) attacked by the crabs with either crab size or with snail ecotype.

**Table 3.9 Summary of the results of the two factor GLM ANOVA examining the effect of snail ecotype and crab size class on average prey value**

<table>
<thead>
<tr>
<th>Source</th>
<th>df</th>
<th>Adjusted MS</th>
<th>F</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Crab Size</td>
<td>3</td>
<td>0.0000165</td>
<td>10.03</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Ecotype</td>
<td>1</td>
<td>0.0003229</td>
<td>196.3</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Crab Size x Ecotype</td>
<td>3</td>
<td>0.000023</td>
<td>14.01</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Error</td>
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<td>0.0000016</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>159</td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

There was a significant difference in calculated prey value between the different crab size classes \( (P < 0.001; \text{Table 3.8}) \) The mean prey value obtained by the crabs whilst foraging on both ecotypes of \( L. \text{saxatilis} \) was significantly higher for both the C1 (mean prey value = 0.00248 Js\(^{-1}\), s.d. = 0.0017) and C4 size class (mean prey value = 0.00241 Js\(^{-1}\), s.d. = 0.0029) than that obtained by size classes C2 (mean prey value = 0.00124 Js\(^{-1}\), s.d. = 0.0014) and C3 (mean prey value = 0.00145 Js\(^{-1}\), s.d. = 0.0014) (Tukey, \( P = 0.05 \)). All other pair-wise comparisons were not significant. There was also a significant difference in mean prey value obtained by crabs of all sizes when foraging on the two different ecotypes \( (P < 0.001; \text{Table 3.9}) \). Overall, crabs of all sizes gained a significantly higher prey value when foraging on the H ecotype (mean prey value = 0.00334 Js\(^{-1}\), s.d. = 0.0019) than the M ecotype (mean prey value = 0.0005 Js\(^{-1}\), s.d. = 0.001). There was also a significant interaction (Table 3.9) indicating that crabs of different
size classes did not show the same pattern of energy gained from the two different ecotypes. Figure 3.6 shows the mean prey value gained by crabs of different size classes when foraging on the two different ecotypes of *Littorina saxatilis*. Whilst the larger crabs (C1 size class) managed to forage on both ecotypes, smaller crabs (C2-C4 size classes) did not manage to break the shells of the M ecotype, therefore the M ecotype had a prey value of zero for small crabs.

Figure 3.6 Mean prey value (standard deviation) for each size class of *Carcinus* when feeding on the two different ecotypes of *Littorina saxatilis*
3.6.4 Preference of *Carcinus maenas* for the different ecotypes

There was a significant difference in the frequency of each ecotype attacked by the different sizes of *C. maenas* during the preference trials (Chi-Squared, \( \chi^2 = 30.973, \) df = 9, \( P < 0.001 \)).

During the preference trials C1 attacked a total of 16 individuals from the H ecotype and 9 from the M ecotype (Fig 3.7) whereas the C2 – C4 size classes of *C. maenas* attacked the H ecotypes in higher frequencies than the M ecotype.

![Histogram showing numbers of *L. saxatilis* attacked by *C. maenas* during preference feeding trials](image)

**Figure 3.7 Histogram showing numbers of *L. saxatilis* attacked by *C. maenas* during preference feeding trials**

There was a significant difference in the frequency of successful attacks made by *C. maenas* when foraging on the two different ecotypes of *L. saxatilis* during the preference trials (Chi-Squared = 59.341, df = 9, P < 0.001)
During the preference trials C1 successfully broke into the shell and ate the flesh of 100% of the 16 individuals from the H ecotype and the 9 individuals from the M ecotype which it attacked (Fig 3.8). C2-C4 crabs were 100% successful in breaking the shell and eating the flesh of H ecotype snails with none of the attacks made on the individuals from the M ecotype being successful (Fig 3.8).

Figure 3.8 Histogram showing numbers of successful attacks made on each ecotype of *L. saxatilis* by each *C. maenas* size class during preference feeding trials

3.6.5 Experience and handling time

Figure 3.9 shows the first trial for C1 involving the H ecotype took over twice as long (399 seconds) to break into, eat the flesh, and discard the shell as was taken during the second trial (177 seconds) with the third trial’s handling time (279 seconds) being almost exactly half way between trial one and trial two. Trials eighteen and nineteen produced comparable handling times to each other (114 and 112 seconds respectively) with trial twenty producing a total handling time of 166 seconds (Fig 3.9).

During the feeding trials involving C1 and individuals of the H ecotype of *L. saxatilis* 100% of the trials ended with the successful breaking of the shell and the consumption of
the flesh. A similar pattern was observed when the crab fed on the M ecotype. Trial one produced the longest total handling time (1290 seconds) but by trial eleven the time was down to 116 seconds, the shortest time measured. During the feeding trials, 85% of encounters ended with the successful breaking of the shell and the consumption of the flesh of the M ecotype of *L. saxatilis* (Table 3.7).

![Histogram showing total handling time and trial number for C1 sized crab foraging on H and M ecotypes](image)

**Figure 3.9 Histogram showing total handling time and trial number for C1 sized crab foraging on H and M ecotypes**

Figure 3.10 shows the first trial involving the crab of the C2 size class and the H ecotype of *L. saxatilis* produced a total handling time of 579 seconds. By, trial eleven the handling time had dropped to 118 seconds, however, subsequent trials produced longer handling times (Fig 3.10). These were still lower than the initial times recorded during the trial. All the H ecotype *L. saxatilis* presented during the trials were broken by the crab and the flesh consumed.
Figure 3.10 Histogram showing total handling time and trial number for the C2 sized crab and the H and M ecotypes.

The M ecotype of *L. saxatilis* produced an initial total handling time of 520 seconds but by trial eleven this had declined to 182 seconds with trial twelve producing the shortest total handling time for C2 trials with the M ecotype of 126 seconds (Fig 3.10). Despite there being variation in total handling time throughout the trials, the final times were still shorter than the initial ones. All the M ecotypes were discarded by the crabs without ever breaking the shells. A similar pattern was obtained from the trials involving both the C3 (Fig 3.11) and C4 (Fig 3.12) crabs, with all H ecotypes being consumed by the crabs and all the M ecotypes being discarded without the crabs breaking the shells. In Figure 3.11 most of the trials involving crab attempts to break into the M ecotype had shorter times than those where the crab was foraging on the H ecotype. The M ecotype was discarded quite quickly whereas the crab successfully gained a meal from the H ecotype.
Figure 3.11 Histogram showing total handling time and trial number for the C3 sized crab and the H and M ecotypes

Figure 3.12 Histogram showing the total handling time and trial number for the C4 size crab and the H and M ecotypes
Figure 3.11 shows that the first and second trials involving C3 sized crabs and the H ecotype produced total handling times of 255 seconds and 270 seconds respectively whereas the third trial produced a total handling time of 1267 seconds after which, no trial produced a total handling time greater than 303 seconds. Trial eleven produced the shortest total handling time of 102 seconds (Fig 3.11). The feeding trials involving the C3 sized crab and the M ecotype all produced total handling times below 400 seconds with the exception of trial sixteen which produced the longest total handling time (656 seconds) (Fig 3.11). Only six of the twenty trials involving C3 sized crabs and the M ecotype produced total handling times greater than those involving the H ecotype for C3 sized crabs. All of the H ecotypes encountered were successfully broken into and consumed whereas none of the M ecotypes were attacked successfully and were discarded in times shorter than those taken by the crabs to consume the flesh of the H ecotypes.

Figure 3.12 shows that the trials involving C4 sized crabs and individuals from the H ecotype of *L. saxatilis* produced the longest total handling time (518 seconds) during the third trial with no other trials producing total handling times longer than this. By trial twelve the total handling time had declined to 73 seconds, the shortest measured total handling time for C4 crabs feeding on the H ecotype (Fig 3.12). All encounters involving the H ecotype of *L. saxatilis* and C4 sized crabs ended with the successful breaking of the shell and consumption of the flesh. None of the encountered M ecotype were successfully broken into during the trials involving C4 sized crabs and only three out of the twenty trials produced total handling times shorter than those involving H ecotype snails. With the majority of encounters involving the M ecotype producing total handling times longer than those recorded during the trials involving the H ecotype, it shows that the C4 sized crabs persisted with their attempts at breaking into the M ecotype shells for longer than the crabs in the larger size classes (C1-C3) (Figs 3.9 – 3.12).
3.7 Discussion

3.7.1 Preliminary experiments

The preliminary trials, to determine the size of each ecotype to be used during the feeding trials, can possibly help explain the presence of crab damage seen on some of the H ecotype snails in an area which is deemed to be free from *Carcinus maenas* (Chapter 2). During the preliminary trials, *Carcinus* did not necessarily attempt to break into the eventual size class chosen immediately. On a number of occasions a larger shell than was possible for the crab to successfully break into was chosen and attempts were made to crush the shell. Unsuccessful attacks could ultimately leave the shell with a piece broken away from the lip or a scar on the shoulder before the crab eventually gave up and concentrated its efforts on breaking into a shell that was more manageable for a crab of that size due to the mechanics of the crabs’ chelae similar to findings by Hughes (1989) on the tropical crab, *Ozius verreauxii* when feeding on the gastropod *Nerita scabricosta*. If, as seen in personal observations, *Carcinus maenas* does find itself in the vicinity of the H ecotype on the rocky shore, unsuccessful attack attempts may be made in eating a larger snail than is possible for the crab to attack successfully and therefore leaving the snail with a damaged shell. The risk of chelal damage and the possibility of future impairment to foraging as described by the chelal wear model (Hughes & Seed, 1995) could help explain why *C. maenas* chose a smaller size shell from the M ecotype to prey upon as handling larger size classes of hard-shelled prey incurs a risk of chelal damage. There are however, models that support intermediate-sized prey items to be the most profitable (Hamilton, 1976; Elner & Hughes, 1978) as well as times when the most profitable prey items are predicted to be the smallest (Hughes & Seed, 1981). As there are arguments for all sizes of prey to be the most profitable, the chelal wear model does not provide conclusive reasoning for why the smaller size class of snail from the M ecotype of *L. saxatilis* were chosen in comparison of the their H ecotype counterparts during the preliminary feeding trials. The fact that *C. maenas* chose a smaller size class of the M ecotype than was chosen from the H ecotype during the preliminary feeding trails, could be due to the assessment of prey vulnerability (Hughes, 1989) being poor by the crab, but could also be explained by *C. maenas* not preferring the most profitable prey item as in Van Der Meer & Smallegange (2003).
Elner & Raffaelli (1980) reported that the energy content of snails is a function of their size, but if attack success rate is taken into account with handling time, smaller snails become more valuable than larger snails. This can explain why the crabs chose snails from the M ecotype in a size class smaller than the size class from the H ecotype. In their natural habitat, spending less time, and therefore energy, breaking into a large number of smaller M ecotype snails would eventually yield a greater profit than attempting to break into a low number of larger snails. Although the larger snails contain more energy per gram of flesh, they possess a larger and therefore stronger shell. Taking more time, therefore using more energy, attempting to break into large shells will not be the optimal choice for the crabs in this situation.

The scatterplots for log energy versus log shell height, for Filey Brigg and Thornwick Bay (Fig 3.3 and 3.4, Table 3.3), suggest the H ecotype has a higher energy content and therefore calorific value than the M ecotype at Filey Brigg but there is no difference between the energy content of the H or M ecotype per unit shell height at Thornwick Bay. This fact alone means it will be more beneficial for *Carcinus maenas* to attack and eat the H ecotype rather than a smaller, harder to break, M ecotype for the same net energy gain.

Figure 3.3 clearly shows the clustering of the H and M ecotypes with the H ecotypes possessing higher energy content per unit shell height than the M ecotype. There are a small number of what appear to be individuals from the H ecotype within the same plots as those from the M ecotype. These can be accounted for by the possibility that intermediates of the two ecotypes were included with the H ecotype when the energy per gram of dry flesh conversion was being carried out. Intermediate forms of the H and M ecotypes of *L. saxatilis* regularly express the characteristics of the M ecotype as described by Hull et al., (1995) and could have possibly been misidentified and included in the data produced for this ecotype.

The H ecotype of *L. saxatilis* possessing a thinner shell than the M ecotype, will have invested less energy in shell production as thinner shells are less expensive to produce energetically (Palmer, 1981). Individuals from the H ecotype will therefore possess a higher calorific value per gram of dry flesh than individuals from the M ecotype with a similar
shell height as less of the energy has been invested in producing the thick shells possessed by the M ecotype (Hull et al., 1996).

3.7.2 Behaviour during feeding trials

The ethogram from the feeding trials shows that *Carcinus maenas* utilises many different tactics and sequences of tactics in order to break into *Littorina saxatilis* shells of differing sizes from both the H and M ecotype. Hughes & Elner (1979) and Johannesson (1986) collectively described eight very broad methods to break into shells of *Nucella lapillus* and *Littorina saxatilis*. The attack methods described by these authors only illustrates the general methods used by *Carcinus maenas* not the entire repertoire of individual movements and action made by the master and minor chela. The behaviours seen and described in the current investigation include the previously reported actions documented by Hughes & Elner (1979) and Johannesson (1986) along with more complex patterns and frequencies of more intricate and precise movements. After watching the tactics employed by each crab during the feeding trials involving both H and M ecotypes, a total of 75 actions were recorded documenting the manipulation and orientation of the shell with the exact location of the shell’s morphology each chela was positioned to perform the action needed within the complete procedure for the shell being investigated by the crab. The acts of behaviour recorded in the current investigation could be combined with the sequence of crab behaviour produced by Hadlock (1980) to create a detailed picture of the ‘thought’ processes *C. maenas* goes through during an encounter with *L. saxatilis*.

The total number of attempts made to break into the shell of *Littorina saxatilis* from the H ecotype was only significantly different during attacks made by the C1 size class of crab. The attempts made by the C1 crab were significantly lower than expected. There was no significant difference between the number of attempts to break the shell of the H ecotype snails during attacks made by the C2, C3 and C4 crabs. As C1 was the largest specimen used in the trials and it was able to crush the shells of the H and M ecotype from the size classes chosen in the preliminary trials, a lower number of attempts were needed to get to the flesh of *L. saxatilis* as, due to the strength of the crab, a single crushing pressure applied in the correct place would have been sufficient to break the shell leaving the flesh exposed. The smallest size class of *C. maenas* made the highest number of attempts to break into the shells of *L. saxatilis* from either ecotype (Table 3.6). As *C. maenas* is an
omnivore with less specialised chelae (Hughes, 1989) the smaller crabs may have had to persist with attempts to break into the shells of *L. saxatilis* for longer than it perhaps would in its natural environment where it could forage for a different food source after the first rejection of *L. saxatilis* whereas in the aquarium, there were no other prey items to be foraged. As critical prey size is determined by the strength of the shell relative to the strength of the chela (Hughes, 1989) the smaller crabs used in the investigation may simply have not been as strong (Sneddon & Swaddle, 1999) and not exerted a force strong enough to break the shells as readily as the largest crabs.

The C1 sized crab was the only crab able to break into and eat the size of snail from the M ecotype that the preliminary trials had determined. There was no significant difference in the time to the first break during attacks made by C1 on the H and M ecotypes of *Littorina saxatilis*. As the shells of the snails in the size classes of the M ecotype chosen by *Carcinus maenas* during the preliminary trials were slightly smaller in comparison to the corresponding size classes chosen for the H ecotype, the crab would spend the same amount of time breaking into a thicker shell to ultimately gain a lower calorific value from eating a snail from the M ecotype rather than the net gain from eating the H ecotype. As crabs are not visual predators (Ekendahl, 1998) they may not be able to differentiate between the H and M ecotypes of *L. saxatilis* upon first contact with the shells. Trussell (2000) provided evidence that when raised in the presence of crabs *Littorina obtusata* produced significantly thicker and heavier shells than conspecifics raised without crabs. As Chapter 2 shows, *C. maenas* was not found at Site 1 but was present at Site 2 where the M ecotype of *L. saxatilis* was abundant. The predator-induced plasticity expressed by the M ecotype of *L. saxatilis* in reaction to the presence of *C. maenas* could help to explain why the smaller size classes of the M ecotype proved more difficult to break into than the larger individuals of the H ecotype that were readily broken into. Ash (1988) reported that *Nucella lapillus* and *Littorina littorea* could both grow into a size refuge where after they will be immune from the maximum breaking load (0.5kN) exerted by *C. maenas*. As the smallest crab used in Ash’s (1988) investigation were as large as the largest crabs used in the current investigation, it is acceptable to assume that the smallest crabs will have a smaller maximum breaking load and will therefore be unable to break into smaller sizes of gastropod shells, thus the smaller size of M ecotype of *L. saxatilis* chosen by *C. maenas* in
the preliminary feeding trials may have already exceeded the critical size that size class of crab could feasibly break.

When presented with five pairs of *L. saxatilis* (pairs consisting of H and M ecotype from the predetermined size classes), every individual of the H ecotype presented in the preference trials was attacked and eaten (100%) compared to only 22.5% of the M ecotype individuals (Fig 3.8). *Carcinus maenas* from the largest size class were the only crabs able to break into the shell, and therefore eat the flesh, of the M ecotype from the predetermined size classes. During the preference trials, a number of individuals of the M ecotype were initially picked up and numerous attempts were made to break into the shell. As the attempts made to break in to the shell of the M ecotype by the smaller size classes of *C. maenas* were unsuccessful, and there was the option of discarding the shell to be replaced by an H ecotype, *C. maenas* desisted with unnecessary energy expenditure and concentrated its efforts on breaking into the thinner shelled H ecotype that was also present.

Regardless of declarations made to the contrary (Elner & Hughes 1978; Seed & Hughes 1995), prey choice in shore crabs does not seem to be based on maximizing prey profitability alone (Smallegange & Van Der Meer, 2003), since *Carcinus maenas* systematically have a preference for smaller prey than those that yield the highest profitability (Elner & Hughes 1978; Elner 1980). Moreover, like most molluscivorous crabs, *C. maenas* forage preferably on prey that fall well below the critical size that can be opened (Juanes 1992).

The time spent by *Carcinus maenas* handling both the H and M ecotypes of *Littorina saxatilis* does appear to decrease with increased experience. The trials that produced the longest handling time period during attacks made on the H ecotype occurred within the first three trials for crab sizes 2, 3 and 4 and the handling time for the remaining trials never exceeds this maximum time. The shortest handling time recorded for each size class of *C. maenas* during the feeding trials involving the h ecotype of *L. saxatilis* was achieved by trial eight for C1 crabs, trial eleven for C2 and C3, and trial twelve for C4 (Figs 3.9 – 3.12). This echoes what Cunningham & Hughes (1984) reported with the acquisition of predatory skills being shown in the decrease of handling time with experience. The necessary tactics and behaviours needed to break into the prey item are honed by *C. maenas* when experience levels increase. During Cunningham & Hughes’
(1984) investigation, the skills were seen to be acquired after the predator had experience of 5 – 6 prey items with the resulting tactics being partially retained for two days and lost after four. During the feeding trials for the H and M ecotypes, the acquisition of tactics and skills necessary to break into *L. saxatilis* shells appears to happen after experiencing 3 individuals from each ecotype (Fig 3.9 – 3.12). Cunningham & Hughes (1984) did not incorporate a starvation period between feeding trials, but instead maintained hunger levels in the predator using the relevant flesh from the prey item. During the two-day starvation period used in the current investigation, the tactics and skills utilised and mentally retained by *C. maenas* may have been somewhat forgotten. This would lead to the trial immediately following the starvation period taking longer than the previous trial as the forgotten skills needed to break into the shell would have to be re-learned. This can help in explaining the pattern of handling times and trial number during attacks made on H and M ecotypes. The first three trials, when the crab has the least experience of the prey item, produced the longest handling times for both the H and M ecotypes for both C2 and C4, the M ecotype for C1 and the H ecotype for C3 size class of *C. maenas*. The mean handling times decrease with increased experience but after a decrease in handling time occurs, there is an increase in handling time. This could be linked to the persistence of learned skills fading (Cunningham & Hughes, 1984) in *C. maenas* during the two day starvation period.

The handling times during attacks made on the M ecotype were generally longer than those seen during the trials involving the H ecotype although the majority of trials involving individuals from the M ecotype of *L. saxatilis* ended with an unsuccessful attack (Table 3.7). As previously mentioned the fewest number of trials needed to achieve the shortest handling time for any size class of *C. maenas* involving the H ecotype of *L. saxatilis* came from the C1 size class of crab taking eight trials (Fig 3.9) and the most trials needed to produce the shortest handling time for the H ecotype was twelve by the C4 size class of crab (Fig 3.12). As the feeding trials process (outlined in Section B Methodology) states, after being presented with three *L. saxatilis* from the H ecotype, *C. maenas* was then subjected to a two-day starvation period. With this in mind, it will have taken approximately seven days for the C1 size class of crab to achieve the minimum handling time involving the H ecotype of *L. saxatilis* with all other size classes of crab (C2 – C4) achieving this in approximately ten days at the most. Cunningham & Hughes (1984) described the acquisition of skills to be apparent after *C. maenas* had had experience of 5 –
6 prey items which were continually being replaced, with these skills being partially retained for up to two days after feeding and lost after four. As the crabs in the current investigation were being presented with half as many prey items per bout of feeding as in Cunningham & Hughes (1984) investigation, it would be acceptable to assume that the acquisition of the skills needed to break into the shell of *L. saxatilis* would possibly require more experience of the prey item as the skills gained by *C. maenas* during experience with the first three shells may have been partially lost during the two day starvation period before the next bout of feeding trials.

The trial that produced the longest period of handling time on the H ecotype for crab size 1 was trial number 17 (Fig 3.9). As the preliminary feeding trials determined, each crab ‘chose’ a snail from each ecotype and size class that was proportional to their body size. It would therefore be acceptable to assume that the handling times for each crab when attacking the H ecotype would be proportional to the size of the crab. Personal observations made during the trials saw that the dactyls, both articulatory and inarticulate, of the master and minor chelae of C1 were slightly damaged and somewhat worn after a moult that occurred shortly after the introduction of *C. maenas* to the aquarium. These worn dactyls could have meant that the sure grip could not be established as the fine points of the dactyls were missing. The handling times for this crab may have been increased as it may have had to perform a higher frequency of behaviours to compensate the loss of the natural shape and mechanics of the chelae and an impaired foraging capability (Hughes & Seed, 1995). Cunningham & Hughes (1984) mention that the methods they witnessed being used by the crabs to break into *Nucella lapillus* appeared to be determined by the grip offered by the geometry of the shell. Not being able to take advantage of this grip offered by the shell could alter the process normally involved in the breaking of the shell.

The results from this part of the investigation show there is no difference in the energy content per unit shell height between the H ecotype from Thornwick Bay or Filey Brigg, nor is there a difference in the energy content between the M ecotypes from these shores. Individuals from the H ecotype found at Filey Brigg appear to have a higher energy content per unit shell height than the M ecotypes found on this shore. Despite the apparent difference in energy content between the H and M ecotypes of *L. saxatilis* found at Filey Brigg it is the fact that shells from the M ecotype are more resistant to predation attempts
from *C. maenas* that ultimately means it offers a lower prey value than the H ecotype to crabs that are able to break into the shells, and a prey value of zero to the crabs that are unable to break into the shells of this ecotype. There is no difference in the total handling time by the different size classes of *Carcinus maenas* recorded during the feeding trials involving the different ecotypes of *Littorina saxatilis*, H or M. *Carcinus maenas* will readily attack both the H and M ecotype of *L. saxatilis* and attempt to break into the shells with the predator having a greater success rate when foraging on the H ecotype than upon the M ecotype. All size classes of *C. maenas* were successful in breaking into the shells of the H ecotype but only the largest size class of crab was successful when foraging on individuals from the M ecotype. During the unsuccessful foraging attempts, *C. maenas* persists in the attacks made on *L. saxatilis* until a ‘giving up’ time is reached. This giving up time differs for each size class of *C. maenas* involved in the feeding trial. The H ecotype of *L. saxatilis* offers a higher value as a prey item as the shell is more readily broken into by *Carcinus maenas* meaning less energy is expended during foraging. Individuals from the M ecotype of *L. saxatilis* are often rejected by foraging *C. maenas* for a size of snail present at the same height on the shore.
Chapter 4

Alarm Responses in *Littorina saxatilis*

4.1 Alarm Responses

Many aquatic organisms rely on chemical senses to detect predators. Often avoidance behaviours are elicited by distance or contact chemoreception of predator ‘odour’ or ‘taste’ (Mackie & Grant, 1974).

The alarm responses of *Littorina littorea* have been investigated a number of times by different authors. Jacobsen and Stabell (1999) studied the alarm responses of *L. littorea* in laboratory experiments over the course of two consecutive summers and an intermediate autumn season. In this investigation, the alarm responses were detected as crawl-out responses which equate to the frequency of snails moving from out of the water. The snails were exposed to extracts of injured conspecifics, extracts of *Modiolus modiolus* (L.) and water conditioned by the predator *Carcinus maenas* (L.) that had been maintained on different diets.

The findings of the investigation show that during the summer months, the chemical stimuli from the injured conspecifics elicited a significantly greater crawl-out response in *L. littorea* than did the extracts of the mussel *Modiolus modiolus* (L.) or the filtered seawater. The water conditioned by *C. maenas* fed on *L. littorea* induced more crawl-outs compared to the water conditioned by crabs fed on a diet of fish. This finding suggests that the source of the chemical stimulus is contained within the snails themselves and that they are not reacting to the ‘odours’ given off by other marine organisms (Jacobsen & Stabell, 1999).

The investigation was performed in both light and dark conditions and the tests carried out during the dark hours evoked a higher crawl-out response from the snails than those performed in the light. These results may indicate an adaption made by the snails to nocturnal predators (Jacobsen & Stabell, 1999). The results suggest that a diet of *Littorina littorea* may mark the predators with snail alarm substances and that the predator-induced responses of *L. littorea* are in fact responses to conspecific alarm substances released from the crab maintained on a diet of *L. littorea* (Jacobsen & Stabell, 1999).
Hadlock (1980) also conducted investigations into the alarm responses of *Littorina littorea* (L.) along with field and laboratory observations of *Carinus maenas* (L.) predation on *L. littorea*.

Field experiments were conducted in tidal pools using a 60 cm diameter octagonal grid as a control area to determine the time taken for the snails to disappear from sight after the introduction of crushed conspecifics. The results of this investigation showed the number of visible snails in the 60 cm grid decreased significantly in the 10 minute period following the addition of crushed conspecifics or snail juice when compared to the change in numbers in the grid when tested with intact snails or changes in seawater. Laboratory observations made on the predation by *C. maenas* on *L. littorea* showed that it took longer for the crab to begin attacking snails in crevices (sheltered) when compared to those in exposed environments.

In a non-related study, Janson (1983) performed mark and recapture experiments to determine the survival and migration rate of two distinct ecotypes found on the west coast of Sweden. The results from Janson’s (1983) investigation show that the ecotype from the exposed areas of the rocky shore (E ecotype) had a mean migration distance after release that was generally higher than the distance migrated by the S (sheltered) ecotype from the more sheltered areas of the shore. The migration distance was between 1 – 4 metres over a period of three months, similar to that of *L. irrorata* Say which has been recorded to migrate average distances of four metres over a period of seven and a half months in the Gulf of Mexico (Hamilton, 1978). During Janson’s (1983) investigation the E ecotype was seen to be more mobile on average than the S ecotype snails and Janson believed both ecotypes were capable of crawling speeds greater than those described by Hamilton (1978).

### 4.2 Aims

The aims of this part of the investigation are to determine what magnitude of response is elicited in each ecotype of *L. saxatilis* in the form of escape times and crawl speeds when exposed to situations involving the absence and the presence of the predator *C. maenas*. 
4.3 Methodology for escape trials

An individual *L. saxatilis* from the smallest size class of the H ecotype was placed onto a piece of waterproof paper which was submerged in 2 litres of saltwater in a Perspex tank (300 (l) x 200 (h) x 205 (w)). Following the format of Hadlock’s (1980) investigation, a control area, in this case a circle, 80 mm in diameter, was marked out on waterproof paper and the *L. saxatilis* was placed at the centre of this circle with the aperture facing downwards towards the paper. After relocation to the new tank, and the antenna of *L. saxatilis* had been fully extended, a stopwatch was started. When the front of the shell had crossed the outer line of the circle the stopwatch was stopped and the time was recorded. This was repeated five times for 5 individual snails from each size class and ecotype of *L. saxatilis* without the presence of *Carcinus maenas*. Each trial was carried out on a fresh piece of waterproof paper to avoid trail following behaviours which is common in species of the marine prosobranch family *Littorinidae* (Erlandsson & Kostylev, 1994). A video camera was set up directly above the Perspex tank and each trial was recorded onto VHS video cassette. The trials took place in a dark room with the tank being lit by a microscope illuminator (Hadlock, 1980).

Plate 4.1 Showing alarm response tank set up for predator absence trials (Photograph taken by author)

Plate 4.2 Showing alarm response tank set up for predator present trials (Photograph taken by author)
After conducting the trials without the presence of *C. maenas*, the whole process was repeated using five different snails from each size class of snail of each ecotype in the presence of *C. maenas* the size of which was the size used in the previous feeding behaviour investigation (see Chapter 3). *C. maenas* was separated from the *L. saxatilis* by a four A4 laminate envelopes stuck together with a series holes in so the effluent from the predator could circulate freely within the tank. Before the trial began, the crab was removed, the water was mixed using a glass rod (five anticlockwise rotations on the *C. maenas* side of the tank), and then the crab was replaced. *L. saxatilis* was put in position after the crab had been replaced. Each trial was recorded onto VHS video cassette.

On completion of the trials, the recordings on VHS were observed. The route taken by the snail was marked down on a circle with a diameter of 80mm. To determine the total distance travelled by each *L. saxatilis* a length of string was laid down on the route taken and then measured in millimetres. The speed of the snail was calculated by dividing the distance travelled (mm) by the time taken (seconds). The direction travelled by the different *L. saxatilis* during each trial was also recorded. The direction was recorded as A, B, C or D depending on where *L. saxatilis* crossed the boundary of the test area.

![Diagram of Alarm response tank format showing position of predator and directions marked on control area.](image_url)

**Figure 4.1** Diagram of Alarm response tank format showing position of predator and directions marked on control area.
4.4 Data Analysis

In order to test the null hypothesis that there was no significant difference between alarm response escape speed for individuals from either the H or M ecotype of *L. saxatilis* in the presence and absence of the predator *C. maenas*, a paired Student t-test was applied to the data. The paired Student t-test was used as in all cases the data showed no significant departure from a normal distribution (Kolmogorov-Smirnov test, \( P > 0.05 \) in all cases) and the data was paired (Dytham, 2003).

To test the null hypothesis that there was no significant difference in the frequency of direction (A, B, C or D) travelled by either ecotype of *L. saxatilis* in response to the stimulus of the presence of *C. maenas* a contingency Table Chi-squared test was applied to the data. The Chi-square test was used as the data was discontinuous and frequency based (Dytham, 2003).

4.5 Results

There was significant difference in mean escape speeds between the H ecotype of *L. saxatilis* in the absence and presence of the predator (Paired Student t-test \( (df = 19, t\text{-Value } = -13.66, P < 0.001) \)). Overall the escape speeds recorded in the presence of the predator *C. maenas* were between two and a half and three and a half times quicker than the escape speeds recorded during the trials with the predator absent (Fig 4.1).

![Figure 4.2 Histogram showing Alarm Response Speed for H ecotype](image-url)
There was also significant difference in escape speeds between the M ecotype of *L. saxatilis* in the absence and presence of the predator (Paired Student t-test (df = 19, t-value = -23.51, P < 0.001). Overall the escape speeds recorded in the presence of the predator *C. maenas* were approximately two and a half times quicker than the escape speeds recorded during the trials with the predator absent (Fig 4.2)

![Graph showing escape speeds](image)

**Figure 4.3 Histogram showing Alarm Response Speed for the M ecotype**

There was significant difference in the frequency of direction travelled for the H and M ecotypes during the trials involving the absence and presence of *C. maenas* (Chi square = 25.812, df = 3, P < 0.001). Overall, directions A and B were used the most during the trials involving the presence of *C. maenas* compared to the trials involving the absence of the predator.
There was a significant difference in the frequency of direction travelled during the alarm response trials. During the trials when the predator was absent, *Littorina saxatilis* from each size class of each ecotype (H and M) travelled in every direction (A, B, C and D) towards the boundary of the control area. When the predator was introduced into the
Perspex tank, the snails only travelled in directions A or B (Chi-Sq = 25.812, df = 3, P < 0.001). The predator was situated behind a perforated plastic separation sheet in the middle of the tank and directions A and B were the directions towards the predator (Plate 4.2) C and D were the opposite direction to the predator (Fig 4.1)
4.6 Discussion

4.6.1 Alarm Responses (Escape speeds)

During the alarm response trials involving the absence of the predator for the H ecotype, all size classes of *L. saxatilis* had a similar escape speed (mean = 0.2206 mm per second, s.d. = 0.0611) with the speed decreasing slightly as the shell height increases (Fig 4.2). These speeds are less than half as fast as the quickest mean speeds recorded by Svensson (1997) of 3.25 cm/minute which equates to 0.5416 mm/second. The M ecotype produced speeds slightly faster than the H ecotype and showed the same pattern as the H ecotype with respect to the largest snails being slightly slower than the smallest (Fig 4.3).

With the addition of a predator, both ecotypes increased escape speed. The smallest increase in speed during trials with the H ecotype came from H4 with an increase of approximately 0.3 mm per second. The remaining size classes had an overall increase of escape speed of approximately 0.5 mm per second (mean = 0.6282 mm per second, s.d. = 0.1303). The second largest size class in the M ecotype, M3, also showed the smallest increase in escape speed with an increase of approximately 0.3 mm per second whereas the remaining size classes produced an increase in speed of approximately 0.4 mm per second (mean = 0.6442 mm per second, s.d. = 0.0750).
During the alarm response trials the H ecotype, although possessing a slower escape time than the M ecotype when the predator was absent, appeared to express a greater increase in escape speed with the presence of the predator.

As a characteristic of the thinner shelled H ecotype is a large aperture and a large foot for adhesion to the wave-exposed substrate, the H ecotype will hypothetically have more muscular tissue in the foot which will enable a quicker escape speed when it is needed (Johannesson, 1986). The quicker escape speeds in the smaller size classes for each ecotype can be explained by the fact that they will be more susceptible to having their shell crushed by a predator so they move away as quickly as possible whereas the larger snail will stand a better chance against crab predation. This is more relevant to the M ecotype as they have thicker, stronger shells. The quicker escape speeds expressed by the smaller size classes from the H and M ecotypes of *L. saxatilis* could also be due to the reserves of water being held in the shells. Chapman (1997) described the reserves of water held in the shell of *Littorina unifasciata* Gray increased with the increase in mass of the shell itself. Larger, therefore heavier shells will hold larger reserves of water which will ultimately lead to a relatively higher mass being carried by a larger *L. saxatilis*. This increased mass could make the snail move more slowly than a smaller-shelled, lighter counterpart.

During the escape trials, the slowest escape speed in the absence of *C. maenas* recorded by either ecotype was slightly over 0.2 mm/sec and the quickest was slightly under 0.3 mm/sec (Fig 4.2 & 4.3). At these speeds, it would take approximately 222.22 minutes to travel four metres in straight line distance at a speed of 0.3 mm/sec and approximately 333.33 minutes to travel four metres at a speed of 0.2 mm/sec. These times are much faster than the reported four months taken to travel 1 -4 metres during Janson’s (1983) mark and recapture experiment and the seven and half months taken to travel four metres by *L. irrorata* in the Gulf of Mexico (Hamilton, 1978). As the distances moved by *L. saxatilis* in Janson (1983) and Hamilton (1978) were measured in a straight line distance, and over a period of up to a month between release and capture, not a continually monitored random crawl as in the current investigation, these data support Janson’s theory that the snails are capable of travelling distances at quicker speeds than previously reported if travelling at a constant speed.
As *Carcinus maenas* is not usually present in the zones of the rocky shore that the H ecotype of *Littorina saxatilis* inhabit (see Chapter 2), the snails will have very little, if any, experience of crab effluent with the scent of previously consumed *L. saxatilis*. This could explain the greater increase in escape times in the H ecotype when compared to the M ecotype. A new and very real predation threat posed to the H ecotype could induce a massive response in the gastropod as they do not want to underestimate the apparent danger and risk shell damage or death. Chapter 2 shows that there was a higher frequency of damage shown in the shells of the M ecotype of *L. saxatilis* found in the same environment as *C. maenas*. The scars on the shells of individuals from the M ecotype may have been caused by a rock or a failed attack made by a predator which the characteristic thicker shell helped the snail to survive. Higher frequencies of unsuccessful predation attempts may desensitise the M ecotype of *L. saxatilis* to the danger posed by *C. maenas*. As there were higher numbers of crabs found in the same environment as the M ecotype of *L. saxatilis* the snails may have already become more familiar with the chemical signals given off by *C. maenas* therefore eliciting a less dramatic response to that of the H ecotype when in the presence of a predator.

### 4.6.2 Response direction

What is confusing is that the prey organism moved closer to the predator during the trials. It would be expected that the snail could detect the chemical signals produced by the crab in the water and take the appropriate evasive action. One explanation for this unnatural pattern can be seen in Marko and Palmer’s (1991) investigation where *Nucella lamellosa* (Gmelin) was seen to be attracted to the scent of *Hemigrapsus nudus* (Dana) with a random response being expressed in the presence of a common prey item. Marko and Palmer (1991) suggested that *N. lamellosa* can assess the risk posed by different species of crab and then initiate the appropriate response. They suggested that the attraction to *H. nudus* could be due to the association of this crab species with crevices and the undersides of boulders where *N. lamellosa* would be less vulnerable.

There may also be methodological reasons why *L. saxatilis* moved in the direction of the predator. As there was no current present in the Perspex tank during the escape trials,
the effluent with the scent of previously consumed *L. saxatilis* may have settled in the corners, or the side, of the tank that were closest to the directions C and D leading to higher concentrations of effluent in this area. If this was the case, *L. saxatilis* may have made the assumption that the predator was on the other side of the tank than it was, therefore leading to the response of escaping in the ‘wrong’ direction.

Also, as *L. saxatilis* is negatively phototactic (Mitsukuri, 1901), it would move away from any source of bright light. During the escape trials, a microscope illuminator was positioned directly above the centre of the Perspex tank (Plate 4.1 and 4.2) where the separation sheet was positioned. If there was an effect caused by the light source on the direction travelled by *L. saxatilis* it is likely the snail would move in the direction of C or D rather than A or B as the intensity of light will have been slightly stronger towards the centre of the tank.
Chapter 5

5.1 Discussion

The field data presented in Chapter 2 showed that the H ecotype occurred in the upper shore at Site 1 on Filey Brigg. The densities were greatest at the maximum vertical height of the distribution of the population and were not significantly different between July and November. All size classes were represented and were present on all surfaces of the rock face and within the cracks and crevices or protected areas available which reflects the findings of Janson (1983). The clumping behaviour of the H ecotypes may provide protection against desiccation (Raffaelli & Hughes, 1978) as they have a relatively larger aperture than the M ecotype and may be more prone to desiccation (Hull et al., 1996). The populations are composed of smaller individuals than those found in the M ecotype which may also enable them to seek shelter in the available crevices (Emson & Faller-Frisch, 1976). During both survey periods, only one individual from the H ecotype of *L. saxatilis* was found at Site 2 which could have possibly been moved between sites due to wave activity) (Hull et al., 1999a). However, the M ecotype occurred at both sites. As conditions must be suitable for the survival of the *L. saxatilis* taxon at Site 2 (due to the presence of the M ecotype), it does not suggest that there is some factor limiting the H ecotype to the upper shore at Site 1. This is unlikely to be the result of adaptation to wave exposure (Boulding et al., 1999) as both populations are on the same shore (and the M ecotype occurs at Site 1 albeit in a lower zone, see Figure 2.5). There could also be differences in physiological tolerances (Hull et al., 1999b) that will enable the H ecotype to survive in the higher reaches of the shore then the M ecotype. However, it was noticeable that no *C. maenas* were found at this site during the survey periods.

The frequency of *L. saxatilis* shells showing signs of damage caused by predation attempts from crabs was extremely low at Site 1 (less than 5% of the shells) and decreased with tidal height during the surveys (Figs 2.3 and 2.6). As the shell had been repaired with further growth continuing from the point of damage it would suggest that predators such as *Carcinus maenas* make migrations further up the shore than previously thought. However, there could be different predators which attack the high shore populations and leave similar signs of damage on a fragile shell after an unsuccessful attack (Ekendahl, 1998). Therefore, the very low incidence of shell injury at Site 1 may not just reflect a lower predation rate.
but may also reflect a higher predator success rate with the H ecotype as a prey item as the snails would not survive the attack.

Whilst the most abundant ecotype on the upper reaches of the shore at Site 1 was the H ecotype of *L. saxatilis* (as seen at Ravenscar by Hull *et al.*, 1996) the M ecotype was the most abundant ecotype on the boulders of the mid-shore at Site 2, and individuals from the M ecotype were also found in the barnacle zone of Site 1 (Fig 2.5). At Site 2, the M morph density increased down the shore reaching a maximum in the *Semibalanus* zone during July a pattern consistent with the findings of Svensson (1997) who noted that the numbers of *L. saxatilis* decreased with an increase in tidal height on the shore. The mobility of the substrate could also be one of the main reasons for the low densities of the M morph observed higher up the shore at Site 2. Here the boulders were small and highly mobile whereas further down shore the boulders became larger and more stable. The fact that we could not sample under the larger boulders may also under-estimate the population density of the M morph (Hughes & Roberts, 1981).

The percentage of *L. saxatilis* shells showing external signs of damage at Site 2 during the July survey showed a definite pattern of increased amount of damage with increased distance down the shore. None of the shells found in the *Pelvetia* zone showed signs of damage with a steady increase in total percentage damage with an increase in the percentage of damage caused by *C. maenas* with the transition into each zone down the shore (Fig 2.9). Some of the damage to the M morphs observed in the field is likely to be the result of boulder movement during storm events (Hull *et al.*, 1999a) rather than repairs to the shell after damage caused by crabs (Blundon & Vermeij, 1983). However, it is very clear that the proportion of damaged shells is far higher at Site 2 (which contains the highest crab densities during the summer months) than at Site 1. The relatively low incidence of crab damage at Site 1 could be due to the lack of marine predators inhabiting that area or the possibility that the thin-shelled high shore morph is easier to crush than a thick-shelled M ecotype (Hull *et al.*, 1999a). Predators would possibly be less successful in breaking open the M ecotype shell due to its increased thickness (Hull *et al.*, 1996) and studies have shown that thicker shelled littorinids often escape crab predation (Raffaelli, 1989; Johnannesson, 1986).
The lower frequencies of the M ecotype of *L. saxatilis* found within the zone closest to the sea can be explained simply by the conditions found in the *F. vesiculosis/F. serratus* zone not being as favourable to *L. saxatilis* as the zones further up the shore due to longer periods of submersion which minimise the time available for foraging, higher levels of exposure to wave energy increasing the likelihood of becoming dislodged, and a greater risk of predation from predators such as wading birds and fishes (Pettit, 1975; Cook & Garbett, 1992). As the frequency of shells from the M ecotype of *L. saxatilis* showing signs of shell damage was higher than that of the H ecotype, it could suggest a higher degree of resistance to predation attempts. The low frequency of shell damage caused by crabs recorded in the H ecotype of *L. saxatilis* could simply be because crabs are not found in the same area of the shore as the snails (Naylor, 1962; Crothers, 1968 and Warman et al., 1993) therefore the thin-shelled H morph managed to survive in the higher reaches of the shore due to the lack of predation, If the H morph occurred lower down the shore they may succumb to predatory attacks or if they inhabited the mobile substrate at Site 2 they may not be able to withstand the crushing forces of the mobile boulders.

The July survey carried out at Site 2 recorded *Carcinus maenas* to be present within the same tidal zones as the size class of the M ecotype of *L. saxatilis*. The lack of *C. maenas* on the shore during the November survey could possibly be due to the fact that the move further off shore to deeper waters during the winter months (Naylor, 1962; Hunter & Naylor, 1993).

Overall the findings of Chapter 2 illustrate that the H ecotype occupies a high shore habitat where no *C. maenas* were found during the survey periods. The M ecotype has a more extensive distribution down the shore, and shares this habitat with juvenile *C. maenas* during the summer months. The lack of individuals from the H ecotype at Site 2 could possibly be explained by the fact that their thin shells would make them more vulnerable to predation and crushing by boulders.

In Chapter 3, the preliminary feeding trials could possibly help explain the presence of crab damage seen on some of the H ecotype snails in an area which is deemed to be free from *Carcinus maenas*. On a number of occasions a larger shell than was possible for the crab to successfully break into was chosen and attempts were made to crush the shell. Unsuccessful attacks could ultimately leave the shell with a piece broken away from the lip
or a scar on the shoulder before the crab eventually gave up and found a new prey item more manageable due to the mechanics of the crabs’ chelae similar to findings by Hughes (1989) on the tropical crab, *Ozius verreauxii* when feeding on the gastropod *Nerita scabricosta*. If, as seen in personal observations, *Carcinus maenas* does find itself in the vicinity of the H ecotype on the rocky shore, unsuccessful attack attempts may be made in eating a larger snail than is possible for the crab to attack successfully therefore leaving the snail with a damaged shell. The risk of chelal damage and the possibility of future impairment to foraging as described by the chelal wear model (Hughes & Seed, 1995) could help explain why *C. maenas* chose a smaller size shell from the M ecotype to prey upon as handling larger size classes of hard-shelled prey incurs a risk of chelal damage. There are however, models to support intermediate-sized prey items to be the most profitable (Hamilton, 1976; Elner & Hughes, 1978) as well as times when the most profitable prey items are predicted to be the smallest (Hughes & Seed, 1981).

Elner & Raffaelli (1980) reported that the energy content snails is a function of their size, but if attack success rate is taken into account with handling time, smaller snails become more valuable than larger snails. This may explain why the crabs chose smaller snails from the M ecotype than the H ecotype. In their natural habitat, spending less time (therefore energy), breaking into a large number of smaller M ecotype snails would eventually yield a greater profit than attempting to break into fewer large snails. Although the larger snails contain more energy per gram of flesh, they possess stronger shell. Figure 3.3 clearly shows the clustering of the H and M ecotypes with the H ecotypes possessing higher energy content per unit shell height than the M ecotype. The H ecotype of *L. saxatilis* have invested less energy in shell production as thinner shells are less expensive to produce energetically (Palmer, 1981). Individuals from the H ecotype will therefore possess a higher calorific value per gram of dry flesh than individuals from the M ecotype with a similar shell height as less of the energy has been invested in producing the thick shells possessed by the M ecotype (Hull *et al.*, 1996).

The ethogram from the feeding trials shows that *Carcinus maenas* utilises many different tactics and sequences of tactics in order to break into *Littorina saxatilis* shells of differing sizes from both the H and M ecotype. Hughes & Elner (1979) and Johannesson (1986) collectively described eight methods used to break into shells of *Nucella lapillus*.
and *Littorina saxatilis*. These attack methods described by the authors illustrate the general methods used by *Carcinus maenas* not the entire repertoire of individual movements and action made by the master and minor chela. The behaviours seen and described in the current investigation include the previously reported actions documented by Hughes & Elner (1979) and Johannesson (1986) along with more complex patterns and frequencies of more intricate and precise movements. After watching the tactics employed by each crab during the feeding trials involving both H and M ecotypes, a total of 75 actions were recorded documenting the manipulation and orientation of the shell with the exact location of the shell’s morphology each chela was positioned to perform the action needed within the complete procedure for the shell being investigated by the crab. The acts of behaviour recorded in the current investigation could be combined with the sequence of crab behaviour produced by Hadlock (1980) to create a detailed picture of the ‘thought’ processes *C. maenas* goes through during an encounter with *L. saxatilis*.

Despite there being no significant different in handling times for crabs feeding on the two morphs, feeding trials showed that only the largest *C. maenas* could break into the shell and consume the flesh of the M morph. The smallest size class of *C. maenas* made the highest number of attempts to break into the shells of *L. saxatilis* from either ecotype (Table 3.6). As *C. maenas* is an omnivore with less specialised chelae (Hughes, 1989) the smaller crabs may have had to persist with attempts to break into the shells of *L. saxatilis* for longer than it perhaps would in its natural environment where it could switch prey after the first rejection of *L. saxatilis*. As critical prey size is determined by the strength of the shell relative to the strength of the chela (Hughes, 1989) the smaller crabs used in the investigation may simply have not been able to generate sufficient force to break the shells as readily as the largest crabs (Sneddon *et al.*, 2000).

As crabs are not visual predators (Ekendahl, 1998) they may not be able to differentiate between the H and M ecotypes of *L. saxatilis* upon first contact with the shells. Trussell (2000) provided evidence that when raised in the presence of crabs *Littorina obtusata* produced significantly thicker and heavier shells than conspecifics raised without crabs. As Chapter 2 shows, *C. maenas* was not found at Site 1 but was present at Site 2 where the M ecotype of *L. saxatilis* was abundant. The predator-induced plasticity expressed by the M ecotype of *L. saxatilis* in reaction to the presence of *C. maenas* could
help to explain why the smaller size classes of the M ecotype proved more difficult to break into than the larger individuals of the H ecotype that were readily broken into. Ash (1988) reported that *Nucella lapillus* and *Littorina littorea* could both grow into a size refuge where they will be immune from the maximum breaking load (0.5kN) exerted by *C. maenas*. As the smallest crab used in Ash’s (1988) investigation were as large as the largest crabs used in the current investigation, it is acceptable to assume that the smallest crabs will have a smaller maximum breaking load and will therefore be unable to break into smaller sizes of gastropod shells. Therefore the smaller size of M ecotype of *L. saxatilis* chosen by *C. maenas* in the preliminary feeding trials may have already exceeded the critical size that size class of crab could feasibly break. During the feeding trials involving the smaller size classes of *C. maenas* (C2 – C4) and the M ecotype of *L. saxatilis* that ended in unsuccessful foraging, along with the total handling times for these feeding trials, support claims that animals follow certain rules. Gibb (1958) described a duration rule; ‘leave after t seconds’ and Krebs *et al.*, (1974) described a value rule; ‘leave when instantaneous intake rate drops to a critical value r’. Both of these rules are followed by *Carcinus maenas* during foraging on the M ecotype of *L. saxatilis*.

The resistance to crab predation of the M ecotype of *L. saxatilis* is also illustrated by the number of attempts made by *C. maenas* to break into each size class of *L. saxatilis* being significantly higher during feeding trials than the trials involving the H ecotype (Chapter 3). The H ecotype of *L. saxatilis* was broken into after significantly less crushing attempts made by *C. maenas*, whilst the M ecotype was able to withstand the multiple crushing attempts made by the crab. The increased vulnerability of the H morph to predation was highlighted by the fact that every individual of the H ecotype presented in the preference trials was attacked and successfully eaten compared to only 22.5% of the M ecotype individuals (Chapter 3). *Carcinus maenas* from the largest size class were the only crabs able to break into the shell, and therefore eat the flesh, of the M ecotype. During the preference trials, M ecotype were initially picked up and numerous attempts were made to break into the shell however the crab eventually switched prey items and concentrated its efforts on breaking into the thinner shelled H ecotype that was also present. Regardless of declarations made to the contrary (Elner & Hughes 1978; Seed & Hughes 1995), prey choice in shore crabs does not seem to be based on maximizing prey profitability alone (Van Der Meer & Smallegange, 2003), since *Carcinus maenas* systematically have a
preference for smaller prey than those that yield the highest profitability (Elner & Hughes 1978; Elner 1980). Moreover, like most molluscivorous crabs, *C. maenas* forage preferably on prey that fall well below the critical size that can be opened (Juanes 1992).

The time spent by *Carcinus maenas* handling both the H and M ecotypes of *Littorina saxatilis* does appear to decrease with increased experience. The trials that produced the longest handling time period during attacks made on the H ecotype occurred within the first three trials and the shortest handling time by trial eight for C1 crabs, trial eleven for C2 and C3, and trial twelve for C4 (Figs 3.9 – 3.12). This echoes what Cunningham & Hughes (1984) documented with the acquisition of predatory skills being shown in the decrease of handling time with experience. During Cunningham & Hughes’ (1984) investigation, the skills were seen to be acquired after the predator had experience of 5 – 6 prey items with the resulting tactics being partially retained for two days and lost after four. During the feeding trials for the H and M ecotypes, the acquisition of tactics and skills necessary to break into *L. saxatilis* shells appears to happen after experiencing 3 individuals from each ecotype (Figs 3.9 – 3.12). The mean handling times decrease with increased experience but after a decrease in handling time occurs there is an increase in handling time. This could be linked to the persistence of learned skills fading (Cunningham & Hughes, 1984) in *C. maenas* during the two day starvation period.

Overall the findings of the prey value and feeding choice experiments show that the H morph would be a more profitable prey item to the different sizes of foraging *Carcinus* as it would take the crabs a fewer number of attempts to break into the shell, they had a 100% success rate at obtaining food from an individual of the H ecotype. Whilst only the larger crabs could successfully forage on the M ecotype, smaller crabs could not manage to break the shell therefore wasted energy in attempting to predate upon this ecotype.

The results from the alarm response trials show that each size class of the H ecotype of *L. saxatilis* had a similar escape speed to each other in the absence of *Carcinus maenas*. The M ecotype showed a slightly quicker escape speed than the H ecotype in the absence of *C. maenas* which may be due to the higher threat of predation in the natural environment of the mid shore than that posed to the H ecotype when inhabiting the high shore. The escape speeds recorded during the current investigation in the absence of the predator were less than half as quick as the quickest mean speed recorded by Svensson (1997) although the
time period over which the distance travelled was recorded was much longer than during the current investigation.

The H ecotype of *L. saxatilis* showed a greater increase in escape speed with the introduction of *C. maenas* than the M ecotype did when compared to the escape speeds recorded in the absence of the predator.

5.2 Further Investigations

The results of this investigation have suggested various opportunities for further work on the behaviours exhibited by *Carcinus maenas* whilst foraging on the H and M ecotypes of *Littorina saxatilis*.

For example, future studies could determine if there was a difference in handling times and, behaviours used, during foraging on the two ecotypes, between male and female *C. maenas* as they are said to forage with differing intensities (Edwards, 1958; Naylor, 1962; Crothers, 1968; Warman *et al.*, 1993). Also, the foraging behaviour of the red and green colour phases of *C. maenas* could also be examined as crabs expressing different colour variations have been recorded to possess differing levels of strength (Kaiser *et al.*, 1990).

Whilst *C. maenas* would feed on *L. saxatilis* in the laboratory, little is known about the intensity at which *C. maenas* forages on *L. saxatilis* when there are other suitable prey species available should be looked at as the predation pressure may be less intense on *L. saxatilis* when there are different species for the crab to concentrate its energies on.

Field distribution and population density surveys like those conducted in Chapter 2 could be performed during each month of the year to create a more detailed profile of the rocky shore at Filey Brigg (and the other shores where the marked polymorphism in *L. saxatilis* occurs e.g. Ravenscar, Hull *et al.*, 1996: Flamborough, Grahame *et al.*, 2001). The study could be extended to not only survey *Carcinus* and the H and M ecotypes of *L. saxatilis*, but also the different prey species present on the shore to determine the fluctuations, if any, in the density and distribution on the shore for each species throughout
the year. This will determine if the predation pressure faced by the prey species creates changes within the population dynamics of the rocky shore habitat.

The feeding trials could be repeated using higher replicates including higher frequencies of *C. maenas* from a wider range of size classes in order to create a more detailed picture of the handling times and acts of behaviour used by a greater sample size of the predator. Also, different species of crustacean predator such as the velvet swimming crab, *Necora puber*, found on the rocky shore habitat of Filey Brigg could be used in separate feeding trials to determine the pressures posed by different predators. The trials could also be run using the H and M ecotypes from the different shores in the area to determine if the same pattern of predation success occurs between shores.

Further studies into the strength of *Littorina saxatilis* shells of individuals from the H and M ecotypes should be made. Samples of *L. saxatilis* should be from a wider area around the British Isles where the H and M ecotypes occur both simultaneously and individually.

The methodology applied by Blundon & Vermeij (1983) could be used to determine if there are any differences in shell strength between ecotypes and different shores by applying force to the dorsal surface of the body whorl using a steel bar. The physical environment of the rocky shore could also be analysed at the locations where both ecotypes of *L. saxatilis* occur. The environment could differ from rocky shore to rocky shore which could ultimately lead to differing shell structure and strength (Brandwood, 1985) of the H and M ecotypes found at the different locations around the British Isles.

Investigations into the magnitude of the chelal forces applied by different size classes of *Carcinus maenas* could be coupled with the results from data collected from investigations into the shell strength of the H and M ecotypes of *L. saxatilis*. Foraging scenarios could be simulated (Elner, 1978) using a strain gauge embedded in a shell of the H and M ecotypes of *L. saxatilis* to determine the forces applied by *C. maenas* during feeding and ultimately the necessary forces needed to break into the shells of the available prey species found on the rocky shores. This could reveal to a greater extent, the risk posed to the H and M ecotypes of *L. saxatilis* by *C. maenas* and provide more evidence of the
predation pressure from the predator being a driving force behind the speciation of *L. saxatilis*.

The survivorship of the H and M ecotypes of *Littorina saxatilis* in the different regions of the shore needs investigation to determine if individuals from the H ecotype have a lower survivorship in the boulder habitat of the M ecotype or are predominantly removed by predators such as *Carcinus maenas* (Raffaelli & Hughes, 1978). This may determine which factors are important in creating the observed degree of divergence between the two ecotypes of *L. saxatilis*. The resistance to predation attempts shown by the M ecotype and the susceptibility to predation and shell crushing injuries observed for the H ecotype could possibly lead to a predominance of the M ecotype on rocky shores with abundant predators or unstable substrates whilst the H ecotype could be better adapted to wave exposed environments where predation occurs at lower intensities.
Appendix I. List of behavioural acts recorded during feeding trials and terms given to each act for use in ethogram.

<table>
<thead>
<tr>
<th>Description of Behavioural Act Term</th>
<th>Behavioural Term</th>
</tr>
</thead>
<tbody>
<tr>
<td>Master picks up shell around back of body whorl.</td>
<td>Pick Up</td>
</tr>
<tr>
<td>Master picks up shell around apex.</td>
<td>Pick Up</td>
</tr>
<tr>
<td>Master picks up shell at base of columella and lip.</td>
<td>Pick Up</td>
</tr>
<tr>
<td>Minor picks up shell around 2nd whorl.</td>
<td>Pick Up</td>
</tr>
<tr>
<td>Minor picks up shell by lip.</td>
<td>Pick Up</td>
</tr>
<tr>
<td>Master raises aperture of remaining shell to mouth.</td>
<td>Pick Up</td>
</tr>
<tr>
<td>Minor raises broken shell fragments to mouth.</td>
<td>Pick Up</td>
</tr>
<tr>
<td>Master picks up shell at bottom of aperture.</td>
<td>Pick Up</td>
</tr>
<tr>
<td>Minor holds apex.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds around 2nd whorl.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds around top of body whorl.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds back of body whorl.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds bottom of aperture.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds broken columella.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds lip.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds round back of body whorl from lip.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds side of aperture.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor holds top of aperture.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor inserted into top of aperture.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor supports back of body whorl.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor supports bottom of lip.</td>
<td>Minor Hold</td>
</tr>
<tr>
<td>Minor grips apex and lowers shell to ground.</td>
<td>Minor Hold</td>
</tr>
</tbody>
</table>
Master extracted then re-inserted into top of aperture.  
Master grips from lip and round back of body whorl.  
Master holds across aperture.  
Master holds apex.  
Master holds around 2nd whorl.  
Master holds around body whorl.  
Master holds base of columella.  
Master holds bottom of lip.  
Master holds lip at columella.  
Master inserted into bottom of aperture.  
Master inserted into top of aperture.  
Master moved up and down length of aperture.  

Apex held in mouth.  
Bottom of lip held in mouth.  
Mouthparts break pieces of shell from lip.  
Mouthparts hold on to lip of aperture.  
Mouthparts test shell.  

Aperture held over mouth.  

Back of body whorl over mouth.  
Shell manipulated so apex is pointing downwards.  
Shell turned approx 180 degrees anti-clockwise.  
Shell turned approx 180 degrees clockwise.  
Shell turned approx 90 degrees anti-clockwise.
Shell turned approx 90 degrees clockwise.
Orient

Minor aids rotation of shell.
Orient

Master rolls shell up sternum towards mouth.
Roll

Minor presses shell against sternum and rolls up to mouth.
Roll

Master crushes across aperture and around body whorl.
Crush

Master crushes across aperture from top of 2nd whorl.
Crush

Master crushes across aperture.
Crush

Master crushes apex.
Crush

Master crushes around 2nd whorl.
Crush

Master crushes around back of body whorl.
Crush

Master crushes around shoulder of body whorl from aperture.
Crush

Master crushes around top of aperture and shoulder of body whorl.
Crush

Master crushes columella.
Crush

Master pulls down and away from body holding side of aperture.
Pull

Minor holds 2nd whorl and pulls down and away from body.
Pull

Master and Minor hold sides of aperture and pull down and away from body.
Pull

Master and Minor hold shell on floor.
Master Minor Hold

Master and Minor release grip in order to establish a better one.
Master Minor Hold

Flesh from aperture held in mouth.
Eat

Mouthparts discard shell fragments as flesh is eaten.
Eat

Master 'peels' away shell on body whorl.
Peel
Minor breaks away remaining body whorl.  
Mouthparts push shell away from mouth.  
Master discards shell.  
Master pushes discarded shell away.  
Minor discards shell.  
Mouthparts discard shell fragments.  
Master 'saws' around back of 2nd whorl.  
Master 'saws' around side of body whorl.  
Walks away from attack site  
Peel  
Discard  
Discard  
Discard  
Discard  
Discard  
Saw  
Saw  
Used to signify end of trial
Appendix II – Diagrams showing direction and distance travelled by H and M ecotypes of *Littorina saxatilis* during the alarm response trials in the presence and absence of *Carcinus maenas* (not to scale) (for directions see Chapter 4).
Appendix III Pictures showing the remains of Littorina saxatilis shells after successful foraging attempts by Carcinus maenas.

1 – Evidence of the body whorls being partly broken away. Master and Minor would have held either side of the intact aperture and pulled away from the mouth. The flesh of the snail would have been held in the mouth and pulled from the shell.

2 – Evidence of the body whorls being completely broken away. The apex of the shell, or part of the body whorl, would have been crushed with the remaining shell being peeled away by either the Master or Minor chela as the spare chela held the lip of the aperture. The flesh from the shell would have been eaten as this process was taking place.
3 – Evidence of the lower whorls being broken. The aperture would have been either chipped away by the Master chela as the Minor chela held the apex for stability, or crushed across the entire opening causing a large piece of the shell to break away.

![Image](image1)

4 – Evidence of the body whorls being peeled away leaving only the columella and the bottom of the aperture lip.

![Image](image2)

5 – Evidence of the body whorls being peeled away as well as the aperture being partially broken.

![Image](image3)
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