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Processes controlling spatial and temporal variations in cockle *Cerastoderma edule* (L.) abundance and distribution

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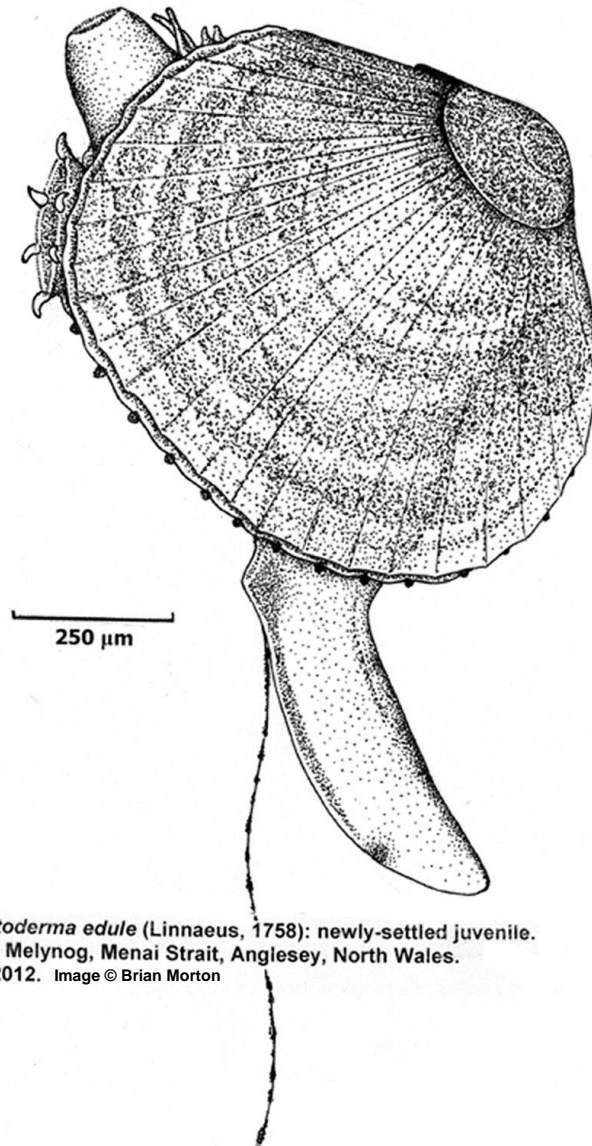
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**Processes controlling spatial and temporal variations in
cockle *Cerastoderma edule* (L.) abundance and
distribution.**



A thesis presented to Bangor University for the degree of Doctor of Philosophy

by Timothy A. Whitton

December 2013



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Summary

The common cockle *Cerastoderma edule* (L.) is an ecologically and commercially important species in Wales, and other North West European tidal flats and estuaries. Cockle populations often exhibit high inter-annual variability in recruitment strength but also spatially within population distributions. The post-larval stage is a critical period for determining recruitment strength in cockle populations, and likely to be a key period for creating spatial structure. This thesis aimed to record and quantify the spatio-temporal changes in post-larval cockle (newly settled 0-group individuals) density over large and small spatial scales, and investigate what processes may create or maintain these patterns. I found that over large scales (100s of metres) the initial settlement can be restricted to certain areas of the shore with high densities, but as time progresses, post-settlement redistributions move post-larvae across the shore. These redistributions recorded over two years extended post-larvae from initial low shore settlements into the mid and high shore, and resulted in a distribution similar to that of the adult population. Over small spatial scales (10s of metres) highly patchy and intense initial settlement of *C. edule* was also observed. Over time the spatial distribution of post-larvae became more dispersed resulting in significant positive correlation with the adult population post-settlement. To understand what is driving changes in density, experiments were conducted in the field and laboratory. Lugworms and adult cockles can be very abundant and disturb surface sediment during feeding and movements, which may kill or negatively affect *C. edule* post-larvae. Lugworms and adult cockles were excluded from areas of sediment in the field and the effect on post-larval densities over time investigated. Removing lugworms increased the densities of post-larvae but removing adult cockles decreased the post-larval density. Laboratory experiments looking at the possible role of lugworms and adults cockles causing direct mortality or increasing benthic and pelagic migrations showed no evidence that these were producing the results seen from the field experiment. I also tested if patchiness of post-larval cockles, compared to a uniform distribution, was beneficial for survival under predation from juvenile shore crabs *Carcinus maenas*, which experience a negative effect of interference competition on their foraging efficiency. Due to the behaviour of 'take away foraging' in the juvenile shore crabs, *C. edule* post-larvae still had higher mortality when aggregated. It is concluded in this thesis that spatial distribution and structure of post-larval *C. edule* change significantly during the post-settlement period, over large and small scales, and that macrofaunal interactions and predation may be a significant factor in producing these observations.

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Chapter 1: General Introduction

1.1 Background

The common cockle *Cerastoderma edule* (L.), formerly *Cardium edule*, is a commercially fished bivalve mollusc of soft-sediment shores from the western coast of Europe and the British Isles. The value of cockle fisheries in England and Wales has reached £19.05 million in 2003. Prices range from £600 – £1500 per tonne peaking at £3000 per tonne for large live cockles, which are often sold on the European (Hicken, 2008). North Wales currently has two to three commercially fished areas with the largest fishery located in the Dee Estuary. The Dee fishery is managed by the Environment Agency-Wales (now Natural Resources Wales – NRW) and is fished by fifty licence holders on up to four beds, depending on the abundance of harvestable (> 20 mm in shell length) cockles. The intertidal *C. edule* population in the Dee estuary is important due its ecological functions, the most referred to is its provision of food for overwintering birds. The *C. edule* beds are surveyed annually and the quota for the fishery is calculated, with the aim of leaving enough cockles to provide food for birds (set at 100 cockles m⁻²) and to ensure the sustainability of the population. The cockle population on the Dee beds is highly variable in abundance and distribution from year to year (unpublished data), with the driving processes largely unknown. Identifying the driving processes of high variation in recruitment magnitude and distribution is a key ecological question (Sutherland et al., 2013). The source of larvae to the intertidal beds is also unknown along with specific processes that are controlling distribution of the Dee Estuary population. There is an interest from the fishery managers to have a better understanding of the processes controlling the production and dynamics of the *C. edule* populations. This PhD was conducted to increase understanding of the processes that are spatially structuring *C. edule* populations on two local cockle beds. Often the priority for managing an exploited species is to understand and seek to quantify the processes controlling recruitment and standing stock. However the abundance of *C. edule* is likely to be influenced by processes affecting the spatial distribution which may limit the use of the habitats' carrying capacity. Understanding spatial distribution is important for investigating a species' ecology and appropriately designing surveys and experiments (Thrush, 1991). Identifying the processes that control the spatial distribution and structure of *C. edule* populations will better inform management, for either conservation or exploitation purposes, and our understanding of the common cockle's ecology.

Spatial distributions of marine invertebrates have been a source of scientific study for several decades from studying biogeography, distribution of a species, to population scale distribution patterns. Physical and biological factors can limit and structure population distributions of marine invertebrates with different factors being more or less important

through the life history of a species. The spatial structure of a population has important implications for its ecology and identifying causal processes is important for how we understand a population and how we investigate it (Thrush, 1991). This introduction will cover the processes acting on *C. edule*, pre- and post-settlement, which have the potential to create the observed distributional patterns in the juvenile and adult populations.

1.2 Patchiness in cockle populations

In many populations of *C. edule*, a high variability in the spatial distribution of adults can be observed (Bocher et al., 2007; Brock, 1979; Lindegarth et al., 1995; Moore, 2004). In a study of the abundance and distribution of molluscs across a latitudinal gradient from the Wadden Sea to the French Atlantic coast south of Brittany, it was found that *C. edule* distribution patterns were site specific and not species specific (Bocher et al., 2007). This indicates that the processes controlling the distribution of *C. edule* from location to location are variable in their presence and/or their magnitude. The majority of distributional data on adults is from grid design surveys in which stations are separated by hundreds of metres. Lindegarth et al. (1995) found that age class distribution differs between populations separated by 1 to 10 km in different bays, indicating small scale or site specific processes are important. This indicates that large scale conditions such as summer seawater temperature and primary production are unlikely to be determining year class strength for *C. edule* on an annual basis. Age class dominance did also differ on a scale of 100m between plots, and non-randomness in the number of year classes was evident on several spatial scales. Adult cockles have been shown to be patchy in their distribution (forming aggregations) both on small (Boldina and Beninger, 2013) and large (Kraan et al., 2009) spatial scales, and this spatial structure appears temporally stable. This indicates that population spatial structure is unlikely to be created or modified past adult maturity for a given cohort.

1.2.1 Processes potentially creating patchiness in cockle populations

Due to adult populations being temporally stable in their spatial distribution, it is likely that spatial patterns are created during earlier life stages. The most likely life stages are during larval settlement, when larvae may settle in aggregations, or by the modification of post-larval distribution through post-settlement processes.

1.2.1.1 Pre-settlement processes

1.2.1.1.2 Reproduction and larval stage

C. edule sexually reproduces through the release of gametes into the water column during the spawning season, which can range from May to August (Baggerman, 1954; Kingston, 1974; Lebour, 1938), with some veliger larvae still present as late as October (Jorgensen,

1946). *C. edule* overwinters in a spent condition and then rapidly develops to sexual maturity in the spring months, with most of the population in a ripe state by mid-summer (Seed and Brown, 1977). Maturity is reached after approximately 18 months with a shell length between 15 – 20 mm (Baggerman, 1954). The eggs of *C. edule* at a shell length of 30 mm are around 77 µm (40 µm excluding the gelatinous layer) with a fecundity of 700,000 female⁻¹ (Honkoop and van der Meer, 1998). The larvae of *C. edule* are planktotrophic and are considered to remain in the water column for 21 to 30 days after development begins (Dare et al., 2004; Lebour, 1938). The larval shell (prodissoconch) length at metamorphosis, and therefore settlement size of *C. edule*, appears to be quite variable from the literature. Lebour (1938) observed settlement at a length of 300 µm for individuals raised in aquaria. Jorgensen (1946) cites a range of observations from 150 – 345 µm, and that such size variation at metamorphosis is common in the Cardidae as noted by Loosanoff et al. (1966). Larvae which have metamorphosed and are ready to settle are recognisable by the loss of the velum and shell growth changing from the prodissoconch to the dissoconch (see Box 1), which displays growth rings and initial rib formation similar to that seen in the adults (Lebour, 1938).

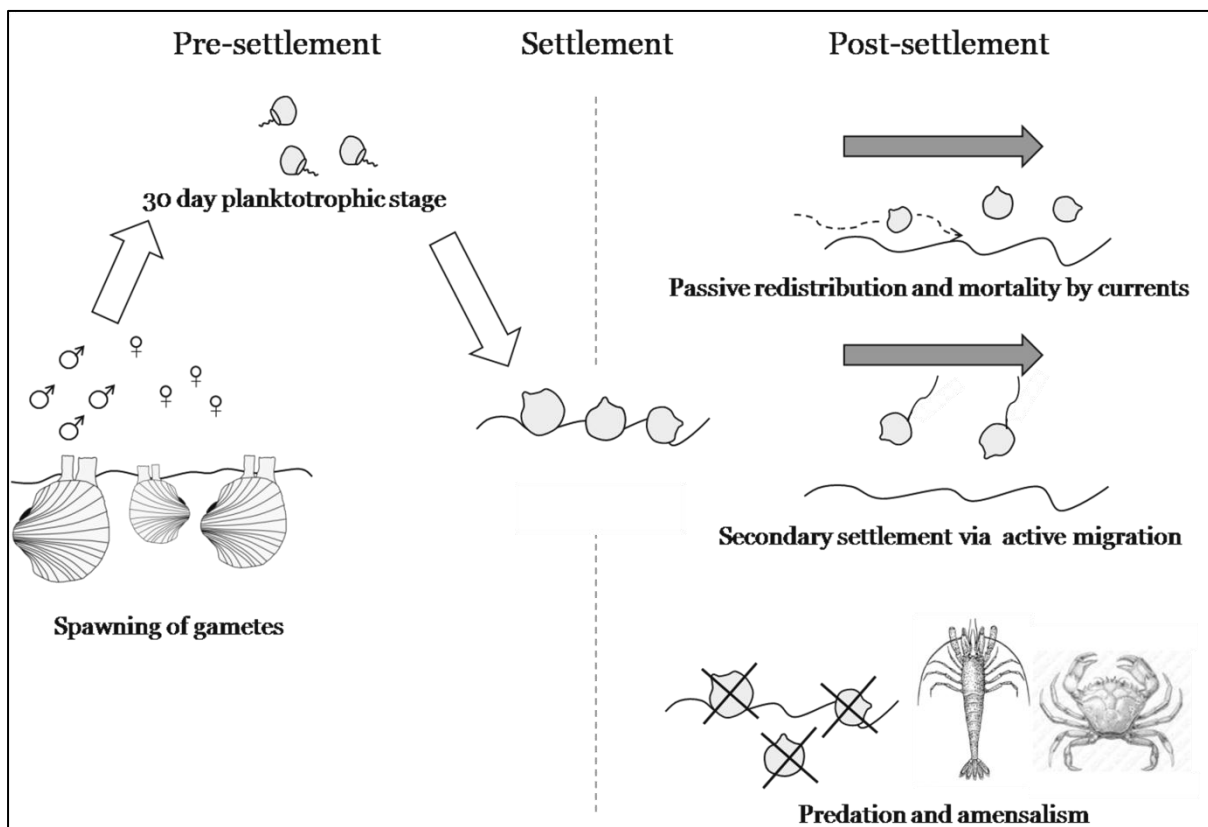


Figure 1.1. The main pre- and post-settlement processes which may affect the observed distributions of 0-group *Cerastoderma edule* in soft sediments. *Crangon crangon* drawing is by Pearson Scott Foresman and the *Carcinus maenas* drawing is by Timothy Sullivan Copyright 2003.

The number of larvae produced and surviving the larval stage was proposed as the key life stage in determining recruitment numbers for marine benthic species by Thorson (1950). He suggested that the potential losses at the larval stage far outweighed those that could occur post-settlement, and so pre-settlement processes (Fig. 1.1) must be the limiting factor in recruitment. This suggestion by Thorson (1950) produced further research into what we now call supply-side ecology (Lewin, 1986), to test this hypothesis and understand what limits recruitment in soft sediment populations.

1.2.1.1.3 Initial settlement processes

Initial settlement, in this thesis, is considered the first event of a metamorphosed larvae attaching or burying into the sediment after the planktonic larval stage. The term recruitment is often used to describe the number of post-larvae that have settled or the juvenile density, but is often observer-defined depending on the sieve mesh size used. There are two main hypotheses for the main processes controlling the spatial distribution of initial larval settlement onto the sediment. The first is the passive deposition hypothesis in which larvae behave the same as passive suspended particles of an equivalent density and sinking velocity, and are therefore deposited in the same areas as particles with similar characteristics (Butman, 1987). The second is active habitat selection by larvae, when they determine their settlement location by responding to cues such as sediment type, light or chemical cues. For example active settlement may be initiated by the presence of con-specifics and are then able to selectively swim to and remain in a certain patch of substratum, thereby reproducing the spatial structure of con-specifics. These two hypotheses are not considered to be mutually exclusive in light of observational and experimental observations (Butman, 1987; Lindegarth et al., 2002; Snelgrove, 1994; Snelgrove and Butman, 1994) and are considered to be operating at different spatial scales. Butman's (1987) review established through calculations based on past data that the limited swimming speeds of larvae and the velocities experienced in the boundary layer would limit swimming against a prevailing current to a few body lengths above the sediment surface in the viscous layer, and even then, this would be in extremely low current speeds. Active habitat selection has only really been demonstrated on scales of cm to tens of cm under laboratory conditions based on sediment characteristics, and so passive deposition of larvae is thought to be the most important process determining settlement patterns on the scale of tens of metres to kilometres (Armonies, 1996; Armonies and Hellwig-Armonies, 1992; Armonies and Reise, 2003). The magnitude at which a process is operational, be it active swimming or passive transport, will depend on the hydrodynamic regime of the shore (Bouma et al., 2001).

Box 1

Identification of post-larval *Cerastoderma edule*

The only published pictures of post-larval *Cerastoderma edule* that could be found were drawings made by Labour (1938), based on laboratory reared individuals from reared captured larvae and field collected post-larvae (Fig. 1.2).

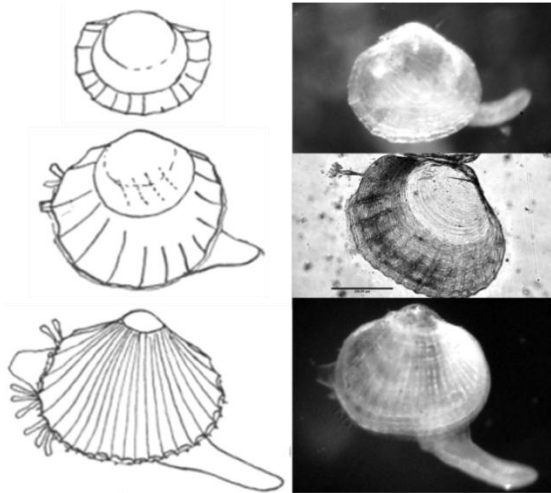


Figure 1.2. Drawings of post-larval *Cerastoderma edule* from Labour (1938) (left) and photomicrographs of live and empty shells collected for this thesis (right).

From these individuals she depicted the clear distinction that can be seen between the larval shell (prodissoconch) and the post-larval shell (dissoconch), with the former being smooth and the latter showing ribs. Identifying *C. edule* post-larvae from other bivalve post-larvae (Fig. 1.3) is helped by three distinctive morphological characteristics. 1) The transparent smooth larval shell has a clear transition to the ribbed post-larval shell which is orange to brown in colouration, especially towards the posterior. Other species do not show such an obvious transition, especially the abundant post-

larvae of *Macoma balthica*. 2) Through the clear larval shell a green mass (assumed to be algae in the stomach, but possibly lipid) can be clearly seen in individuals ranging from 400 – 700 μm in shell length. 3) The foot remains almost circular in cross-section as it explores and pulls a post-larva along a petri dish base. Abundant tellinid post-larvae, such as *M. balthica*, have a much flatter cross-section on the dorsal-ventral axis of the foot. It was also noted that the post-larval inhalant and exhalent siphons of *C. edule* differ significantly. The exhalent siphon has a long smooth valvular membrane which protrudes from the shell margin and mantle, whereas the inhalant siphon appears flush with the mantle and is fringed with tentacles. Post-larval *Mya arenaria* also show this variation in siphon morphology.

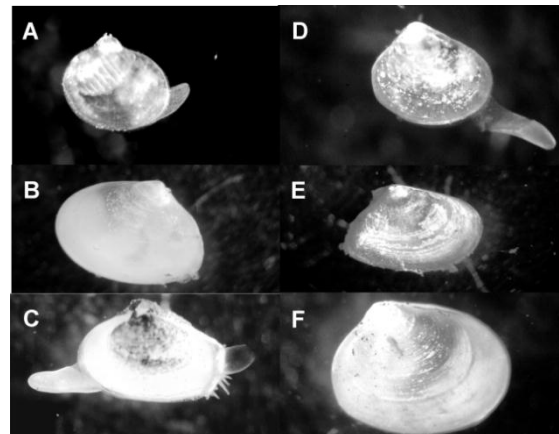


Figure 1.3. A selection of other small bivalve post-larvae species found associated with *Cerastoderma edule*. *Macoma balthica* (A), *Abra alba* (B), *Mya arenaria* (C), *Abra tenuis* (D), *Tellina* sp. (E) and *Kurtiella bidentata* (F). Specimens shown range from approximately 400 – 1500 μm in shell length (Whitton unpublished photographs).

1.2.1.1.4 Adult-larval interactions during initial settlement

High densities of adult *C. edule*, or other filter feeding organisms, may have negative effects on settlement (Andre and Rosenberg, 1991; Bachelet et al., 1992; Flach, 2003). This is due to the pumping of water into the pallial cavity during suspension feeding (Fig. 1.4) which will contain larvae and result in their subsequent mortality. Lindegarth et al (1991) used

fluorescent PVC microparticles to assess if passive deposition of particles with similar properties to those of metamorphosed *C. edule* larvae was influenced by the presence of adults. The particles had a mean size of 149 μm and a sinking speed of 4.5 mm sec^{-1} and were added to aquaria with live adult *C. edule* buried in the sediment (Fig. 1.4.A). Live larvae were also labelled by feeding them fluorescent pigmented particles of a size $< 10 \mu\text{m}$ which were visible through the transparent larval shell valves (Fig. 1.4.B). This technique allowed observations of settlement patterns to be made in real time, on a scale of tens of centimetres without invasive sampling. Lindegarth et al. (2002) used larval mimics and modelling to investigate if adult *C. edule* affected passive settlement and found that an area 2.5 cm^2

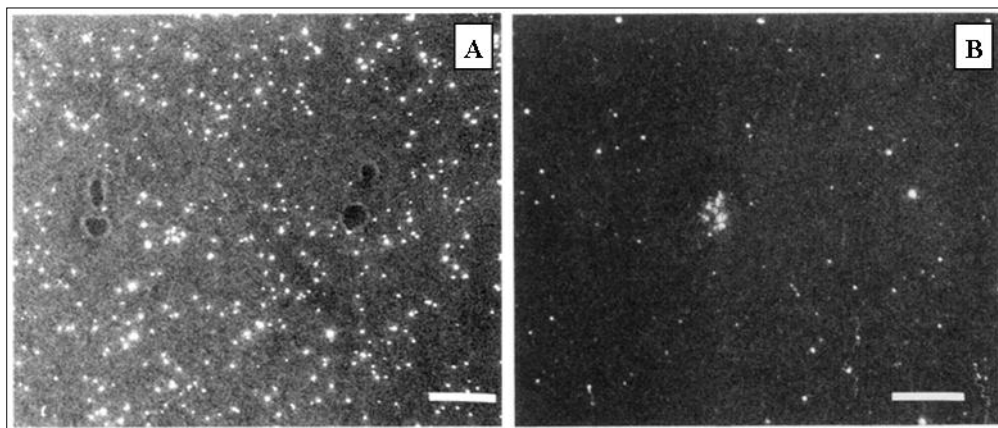


Figure 1.4. Photograph of fluorescent PVC microparticles which have settled around the siphons of two adult *Cerastoderma edule* (A) and fluorescently labeled *C. edule* larvae on the sediment surface (B). Note larvae which have accumulated in a depression (B). Scale bar = 1 cm. Taken from Lindegarth et al. (1991).

around the exhalent siphon of adult *C. edule* had a 20% reduction in settlement of the particles. Results from laboratory work are often more conclusive than those of field studies (Andre and Rosenberg, 1991; De Montaudouin and Bachelet, 1996), but with a less applicable spatial scale. However the evidence does indicate that high adult densities can reduce settlement through inhalation of larvae (Olafsson et al., 1994). In a flume study using live *C. edule* larvae Jonsson et al. (1991) observed that at current velocities greater than 5 cm sec^{-1} larvae became confined to less than 1 mm above the sediment surface with speeds over 15 cm sec^{-1} , resulting in larvae tumbling along the sediment surface and being susceptible to resuspension. This would increase their risk of inhalation from adults compared to those consumed at lower current speeds when they are fully mixed in the water column, however this observation has not been made in the field.

1.2.2 Post settlement processes

Post-settlement processes are considered important for driving recruitment variability and spatial distribution (Olafsson et al., 1994). There are two main types of post-settlement

processes which can alter the spatial distribution of *C. edule* post-larvae on the intertidal shore (Fig.1.1) which are redistribution, be it passive or active, and spatially heterogeneous mortality from predation or interactions with macrofauna (amensalism).

1.2.2.1 Redistribution of post-larvae

Post-larval redistribution after initial settlement may affect the recruitment into a population (De Montaudouin and Bachelet, 1996) and its distribution. Bivalve post-larvae from several families are known to migrate after initial settlement by means of byssus drifting (Armonies, 1996; Hiddink, 2002; Yankson, 1986). Byssus drifting refers to the use of a byssus thread, which is produced from a gland located at the base of the foot, to increase an individual's hydrodynamic drag and increase the chance of resuspension into the overlying water current. Turnover of *C. edule* post-larvae can be high with the whole population potentially changing location every night between June and September (Armonies, 1992). Drifting has been recorded in *C. edule* with a shell length between 0.5 & 3.5 mm (mean 1.26 mm) by Armonies (1992), with Yankson (1986) suggesting that the byssus structure of *C. edule* can only support post-larvae of 2 - 4 mm in shell length. A flume experiment indicated that post-larvae may be able to have a secondary settlement up to a size of 5.7 mm (De Montaudouin et al., 2003). However, applying the results from laboratory flume experiments to natural situations should be undertaken with caution.

Armonies (1996) used a grid system to sample settlement, post-larval abundance and sediment type along with the number of drifting individuals for several species of bivalves in a sheltered bay in the Wadden Sea. The study concluded that the patchiness and sandy sediment association of older *C. edule* is caused by post settlement mortality and/or active rejection of settlement areas, rather than spatial variation of initial settlement by metamorphosed larvae through active habitat selection. Buoma et al. (2001) surveyed early settlement and changes in 0-group *C. edule* distribution along with a series of physical parameters in an estuarine environment in the Netherlands. They concluded that sediment dynamics and bed-level height were important in initial settlement, and concluded that settlement was greatest in higher shore areas. Huxham and Richards (2003) conducted a field survey and experimental study on *C. edule* and *Macoma balthica* in the lower Forth estuary in Scotland with conclusions similar to that of Armonies (1996). They comment that there was no evidence that redistributing post-larvae could chose the sediment onto which they settled onto. They suggested that sediment related patterns observed in adult populations are most likely due to post-settlement processes. On highly dynamic shores data on the distribution of early bivalve recruitment and their correlation with sediment dynamics and hydrodynamics indicate that sediment stability may be controlling the distribution. However, biological post-settlement processes possibly are playing a more

influential role in sheltered locations (Bocher et al., 2007; Moore, 2004). This ability to change location after settlement across the intertidal shore has been demonstrated to potentially be advantageous in *Macoma balthica* allowing a reduction in predation, parasite infection and increased growth potential (Hiddink, 2003). The ability of *C. edule* post-larvae to redistribute is likely a key process that allows the population to utilise larger areas of available habitat, and be such a successful species on soft-sediment tidal flats (Armonies and Reise, 2003).

1.2.2.2 Predation

The extremely high abundances of *C. edule* post-larvae that can be found on an intertidal soft-sediment shore, over 16,000 m⁻² (Armonies, 1996) and 60,000 individuals m⁻² (Jensen and Jensen, 1985), represents a potentially highly abundant food source for many predators. Predation has the obvious potential to reduce the overall abundance of post-larvae and therefore recruitment magnitude in any given population. However, if predation pressure varies spatially, it has the potential to create patchy or graduated density patterns. Much of the earlier literature considers sources of predation on adult *C. edule* from birds such as the Oystercatcher *Haematopus ostralegus* (Horwood and Goss-Custard, 1977; Sutherland, 1982a) and flatfish and starfish (Hylleberg et al., 1978). Predators feeding on *C. edule* post-larvae include juvenile brown shrimp *Crangon crangon*, juvenile shore crabs *Carcinus maenas*, juvenile flatfish, the sand goby *Pomatoschistus microps* and various polychaete worms (Caron et al., 2004; Flach, 2003; Freitas et al., 2007; Hiddink, 2002; Jensen and Jensen, 1985; Pihl and Rosenberg, 1984). Sanchez-Salazar et al. (1987) described how predation could structure the distribution and size structure of adult cockles at different shore heights. This was controlled by size selection of cockles by the common shore crab *C. maenas* and Oystercatcher *H. ostralegus* and the different shore heights in which they are able to forage in. Studies into the influence of predation on 0-group *C. edule* have indicated that predation from epibenthic crustaceans has the potential to control population cohort strength, by limiting recruitment in years predator abundance is high (Beukema and Dekker, 2005). Beukema and Dekker (2005) suggest that predation pressure upon juvenile *C. edule* may only be expressed in the lower shore areas due to the limiting factor of emersion time in higher shore areas. Quantitative estimates of the potential predation pressure these species can place on *C. edule* and other bivalve species are significant. Pihl and Rosenberg (1984) estimate that *C. crangon* can consume up to 68 % of post-larval production of two bivalve species, and Jensen and Jensen (1985) have suggested that juvenile *C. maenas* are likely responsible for just over 26 % of the observed 0-group cockle mortality. Such predation was thought by Guillou and Tartu (1994) to be a large contributor to the rapid decrease in cockle density during the following spring after settlement, but after a potential size refuge was

reached at a shell length of 10 – 12 mm mortality decreased. However the mortality in post-larvae observed in autumn following the summer settlement was suggested to be caused by the transition from lipidic to glucidic metabolism where a glycogen deficit can occur (Sauriau, 1992 cited in Guillou and Tartu, 1994). It is important to remember that many of the predators mentioned have dynamic feeding behaviour which will change with prey availability, size of predator and prey, distribution and their own vulnerability to predation (Burrows and Gibson, 1995). Factors controlling the abundance and timing of arrival onto the shore of predators, such as *C. maenas* and *C. crangon*, have been shown to be affected by the preceding winter's temperatures and so predator abundance and temporal presence is also dynamic in nature (Beukema and Dekker, 2005; Freitas et al., 2007; Philippart et al., 2003). If predator species show varying preferences for certain sediment types or prey densities and spatial structure (aggregated to dispersed), then this also has the potential to modify post-larval distributions and create or maintain patchiness in populations.

1.2.2.3 Post-larvae and macrofauna interactions

Interactions between macrofauna and post-larval cockles are often seen as negative in consequence for the survival of the post-larvae. Negative interactions with other organisms where no advantage is gained for either party are often referred to as amensalism. Con-specific adults are also considered as macrofauna in this thesis. The feeding activity of deposit feeders, production of pseudofaeces and bioturbation from macrofauna have been suggested to cause mortality in *C. edule* post-larvae through smothering and frequent disturbance. Rhoads and Young (1970b) proposed that deposit-feeders inhibit suspension-feeders colonising muddy sediments by smothering and consuming settled larvae and through resuspending particles which would inhibit filter-feeding. Woodin (1976) further investigated and discussed this 'trophic-group amensalism' and proposed that adult-larval interactions can determine discrete communities dominated by suspension-feeders, tube builders and deposit-feeders. Whilst not being observed consistently in the field (Snelgrove and Butman, 1994) it does highlight that community structure across a shore may play an important role in habitat suitability and early post-settlement mortality in an area of sediment, and therefore may create patchy distributions of *C. edule*. However Compton et al. (2009) identified that across a wide geographical scale on several shores that adult and juvenile *C. edule* had overlapping distributions. The explanation for this could only be hypothesised but were considered to be resulting from either density dependant predation where adults provided a spatial refuge, or that despite the theory of adults suppressing settlement, areas with adults hold a fitness advantage over adjacent sediment (Compton et al., 2009). There remains some uncertainty about interactions between adults and early recruits.

1.3 Recording initial settlement

The most common methodology for sampling newly settled larvae and post-larvae is taking sediment cores to a shallow depth, and then sieving them in the laboratory over nested sieves after freezing or chemical preservation. Studying and recording the initial settlement of *C. edule* larvae has been recorded with limited success (Baggerman, 1954; Bouma et al., 2001; Butman, 1987). This is largely due to the problems of sampling newly settled larvae amongst the amount of residual sediment which is retained on fine mesh sizes ($\approx 150 - 350 \mu\text{m}$) which are required to retain the small new settlers (Bouma et al., 2001). Some studies have shown that simple methods such as swirling and decanting water from a sediment sample can remove up to 100% of the post-larvae present. Due to the often large size range (275 – 345 μm) of settling *C. edule* larvae (Jorgensen, 1946) and the high mobility of post-larvae (Armonies, 1994b), distinguishing between initial larval settlers and re-distributing post-larvae is often impossible. When post-larval distributions are recorded in the field, using the term colonisation to describe observations is often more accurate than stating larval settlement has been recorded (Gimenez, 2009) .

Identifying bivalve larvae is difficult, especially at the lower limit of the larval size range (Hendriks et al., 2005; Loosanoff et al., 1966), but modern techniques such as fluorescent labelling (Garland and Zimmer, 2002) and genetic techniques (Andre et al., 1999) can allow bulk identification of samples. The main source found for identifying bivalve post-larvae is Lebour (1938), however identification is helped in most post-larvae as they adopt some morphology seen in the adults (see Box 1). Taking sediment cores and sieving over stacked sieves remains a cheap and widely used technique for collecting bivalve post-larvae to record distributions. However Astro-turf mats have been used successfully to collect *Mya arenaria* post-larvae (Myrand et al., 2012). Methods for quantifying the spatial distribution of soft-sediment infauna range from pattern observations (Armonies, 1996), hierarchal ANOVA designs (Lindegarh et al., 1995; Morrissey et al., 1992a) and geostatistical approaches (Cole, 2009; Kraan et al., 2009). Geostatistical approaches allow quantification of spatial structure and is particularly useful if there is no previous knowledge of spatial structure (Fig. 1.5).

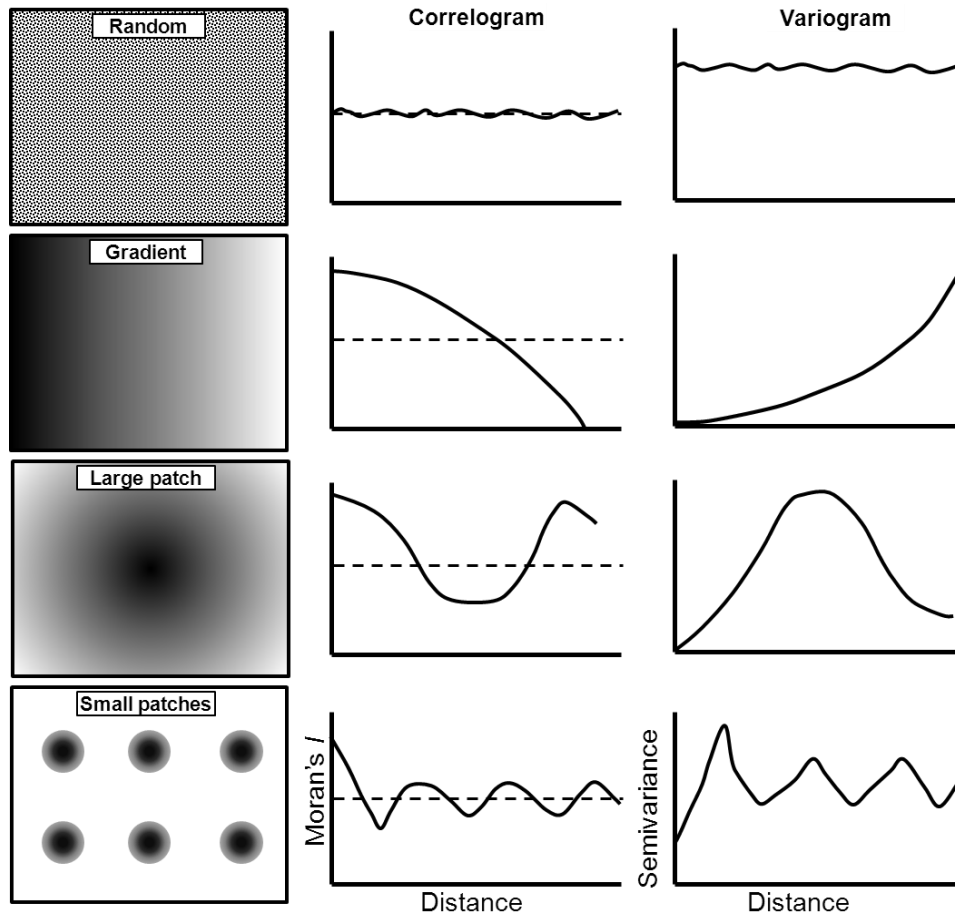


Figure 1.5. Illustration of how geostatistical analysis, such as correlograms and variograms, can be used to quantify spatial patterns of density. Correlograms use an autocorrelation index on the y-axis ranging from 1 to -1 with 0 (dashed line) indicating random structure. Variograms plot a measure of variance on the y-axis with lower values indicating higher similarity between samples. Both can be used to quantify how densities change at varying distances apart and the quantification of patch sizes and inter-patch distances. Darker shades illustrate higher densities. Adapted from Kraan et al. (2009).

1.4 Thesis outline

The overall aim of this thesis was to record spatio-temporal patterns in *C. edule* post-larval density, and identify the driving processes that create the spatial patterns often observed in adult populations. The two main objectives were to 1) measure the spatio-temporal patterns of post-larval density at various spatial scales to help identify what processes may be driving them and 2) to experimentally test these or other processes and how they may drive the observed spatial patterns of cockles. In Chapter 2 the large scale (10s – 100s of metres) spatial distribution and structure of *C. edule* post-larvae was investigated over two years. The main aim was to understand if and when post-settlement processes created the large scale spatial structure observed in the adult *C. edule* population at Traeth Melynog located on Anglesey in North West Wales. These observations would help identify processes that are operating and over what spatial scale. In Chapter 3 small scale (10's of metres) spatio-temporal changes in post-larval density were recorded at the Dee estuary (North East Wales)

/ North West England border) from settlement through to post-settlement. The main aim of this study was to quantify and understand how the spatial structure of post-larvae changes at small scales, and how this relates to the adult cockles and sediment properties. In Chapter 4 the potential effect(s) of macrofauna (lugworms *Arenicola marina* and adult cockles) to influence spatial patterns of *C. edule* post-larval density were experimentally tested. A combination of field and laboratory experiments were undertaken to investigate how density, survival and migration rates differed between macrofauna inhabited and macrofauna free sediment. In Chapter 5 the effect of small scale patchiness on the survival of *C. edule* post-larvae from epibenthic predation was tested. The main aim of this chapter was to discover if interference competition between juvenile shore crabs *Carcinus maenas* would produce a positive feedback for *C. edule* post-larvae being spatially aggregated. In my final chapter (Chapter 6) the findings of the previous chapters are discussed in the context of the overall thesis aim and previously published research. General conclusions and suggestions for future research are made.

Chapter 2: Population scale spatial and temporal patterns of common cockle (*Cerastoderma edule*) post-larval colonisation.

Timothy Andrew Whitton, Stuart Rees Jenkins, Christopher Allan Richardson, Jan Geert Hiddink

Abstract

The common cockle *Cerastoderma edule* is a commercially important bivalve species of soft-sediment intertidal shore communities of north western Europe. Cockle populations frequently show highly patchy distributions and the source of this spatial variability in distribution is currently considered to be generated through early post-settlement processes. In this study initial post-larval densities and how they change temporally between June 2010 and February 2012 were studied across Traeth Melynog tidal flat, North Wales, to inform possible processes creating spatial patchiness of post-larval and adult cockles. A staggered nested design was used to allow a spatially explicit analysis of cockles at 75 sample points distributed across 15 transects at exponentially increasing distances from 0.7 to 133.3 m over a 0.38 km² area. Settlement of post-larvae occurred in late May to early June in 2010 and 2011 and was restricted to the low shore with a peak post-larval density of 4,663 m⁻² with a mean shell length across all stations of 777 µm ± 43 (S.E.). After June, colonisation of the mid shore, and to a limited extent the high shore, took place resulting in a distribution similar to that of the adult cockle population. The spatial structure of post-larvae post-winter was a gradient distribution for the 2010 cohort and patchy in the 2011 cohort with adults having a gradient structure in both years. The low intertidal shore was important for initial benthic colonisation of cockles in 2010 and 2011 settlement. This indicates 'up shore' redistribution of post-larvae may be significant in determining the adult population structure and distribution, with the post-winter spatial structure being variable between years. This has implications for the understanding of post-settlement processes temporally as post-larvae redistribute after settlement either actively by bysso-pelagic migration, or passively via scouring currents. The possible outcomes of such redistributions are discussed and considered to be possibly advantageous for post-larval colonisation success.

2.1 Introduction

The distribution in space of individuals within a population is a key characteristic of a species and is often shaped by life history traits, ecology and the physical conditions experienced during life (Bergström et al., 2002; Legendre and Fortin, 1989; Marquet et al., 1993). Large scale spatial structure (tens of metres to kilometres) is considered to be created through physical processes, whilst small scale spatial structure through biological processes such as species interactions (Bergström et al., 2002; Legendre et al., 1997). The spatial structure of a population can have important implications for the survival of individuals and the long term stability of a population (Thrush, 1991). For example over small scales the patchiness of infaunal bivalves can affect the foraging efficiency of predatory crabs (Clark et al., 2000; Whitton et al., 2012), and on a larger scale the peak and trough wave pattern of soft sediment mussels beds optimises resistance against erosion but also reduces food competition (van de Koppel et al., 2005). It is therefore of value to accurately observe spatial structure in populations (Thrush, 1991), and then seek to understand the casual processes that generate them (Legendre et al., 1997). Such investigation cannot rely on correlative studies from a single snapshot in time (Snelgrove and Butman, 1994) because such processes are likely to be highly dependent on the phenology of the species under investigation. There are many examples of population distributions and within population spatial structure of marine species changing with phenology across several phyla (Armonies, 1994a; Corgos et al., 2010; Methven et al., 2003). The spatial distribution of many intertidal soft-sediment invertebrates can be highly patchy (Kraan et al., 2009; Olafsson, 1992) at different scales with the key driving processes still unknown for many species. In such systems the benthic post-settlement stage is supported as being highly significant in determining recruitment success (Olafsson et al., 1994). Processes such as predation (Beukema et al., 1998; Flach, 2003; Hiddink et al., 2002c) and post-settlement redistribution (Armonies, 1994b; Hiddink et al., 2002a; Jensen and Jensen, 1985) are documented as important for determining post-larval survival for soft sediment invertebrates such as bivalves. These processes are unlikely to be spatially homogenous in occurrence and intensity, and so it is logical to focus on this life stage when trying to understand the driving processes of population spatial structure in soft sediment invertebrates. The spatial structure of a population also has implications for the scientific investigation and management of a species and the communities in which they exist. Any spatial patchiness or other spatial trends can determine the appropriate spatial scale to design surveys and experiments, and the application of findings from one spatial scale to another (Thrush, 1991; Thrush et al., 1997). For example if a survey of a mussel bed was conducted with equally spaced sample locations (as many biological surveys are designed) at the same distances apart as the

peaks and troughs pattern mentioned above, the result would be either a significant over or under estimation of the population density. The commonly observed phenomenon in ecology that spatially close sample values are more similar than samples separated by larger distances is termed autocorrelation, and has implications for sample independence as well as gaining accurate variable measurements spatially (Legendre and Fortin, 1989; Tobler, 1970). Understanding the spatial structure of a population is essential in investigating large and small scale characteristics of a population accurately (Morrisey et al., 1992b), and it is accurate observational data that is critical for proposing well informed and appropriate ecological hypotheses (Underwood et al., 2000).

The common cockle *Cerastoderma edule* is an abundant commercially exploited and ecologically important species in most locations where it occurs (Cesar and Frid, 2009; Malham et al., 2012). Due to this importance it is often routinely surveyed for fishery and conservation purposes. Despite the visually homogenous sediment composition, shore height and food supply at large scales, densities of *C. edule* can vary from several hundred per square metre to complete absence (Bocher et al., 2007; Boldina and Beninger, 2013; Kraan et al., 2009; Lindegarth et al., 1995). *C. edule* has planktonic a larvae that settle onto the benthic environment as a post-larvae. Process such as bioturbation, sediment deposition and predation, amongst other possible processes, may vary spatially contributing to the spatial distribution of *C. edule* post-larvae. Potentially the most important process affecting distribution during the post-larval stage of *C. edule* is byssus drifting (Armonies, 1994b; 1996). This process is likely to dominate over other post-settlement processes with regard to determining the initial spatial distribution, predominantly due to its frequency soon after larval settlement. These distributional changes are facilitated through the production of a single byssus thread from a specialised organ in the heel of the post-larval foot (Yankson, 1986). This is currently understood as an active behavioural process by which *C. edule* post-larvae increase their hydrodynamic drag and are carried along by the current. This behaviour is termed bysso-pelagic migration, or byssus drifting hereafter. The maximum shell length at which byssus drifting has been observed is 5.7 mm (De Montaudouin et al., 2003). Within the first year of life *C. edule* attains a length of around 10mm, but growth is highly dependent on an individual's location on the shore, and so the first months of the post-larval stage are likely to be significant in determining the population spatial extent and structure while the ability to drift remains. This is illustrated in the Baltic tellin *Macoma balthica* which has a highly mobile early benthic phase like *C. edule*. Post-larval *M. balthica* undergo a series of migrations between different shore levels resulting in the observed adult distribution (Beukema and De Vlas, 1989). From a clear understanding of the spatial pattern and how it changed temporally, the most likely drivers of these changes in spatial structure have been investigated (Hiddink, 2003; Hiddink and Wolff, 2002). Therefore to understand the

population spatial structure observed in the adults, the processes involved in the early life stage need to be identified and their spatial and temporal attributes understood.

Recording spatial patterns of soft sediment organisms such as post-larval *C. edule* over time is difficult when individuals are less than 1mm in size, infaunal and highly mobile (Thrush, 1991). It is this high mobility of post-larvae that means the pattern of initial larval settlement cannot be confidently recorded in most studies, because you cannot identify if larval settlement or post-settlement migration have determined a small post-larvae's location. The rate of this redistribution is thought to be very high with the whole population potentially redistributing during a Spring tide (Armonies, 1994b). Due to this in reality only post-larval 'colonisation' is often measured and not initial larval 'settlement' (Bouma et al., 2001; Gimenez, 2009). Therefore there is a lack of knowledge about actual larval settlement patterns in *C. edule*. This lack of knowledge can be considered as little consequence due to the high turnover and ability of post-larvae to redistribute rapidly post-settlement, and so quantifying this was not an objective of this study. From current knowledge larval settlement is however considered by most to be a passive process governed by hydrodynamics (Bouma et al., 2001). Accurately recording spatial structure of an infaunal species is difficult. However, geostatistics is a tool being increasingly used in marine ecology to do this. Geostatistics incorporates the distance between samples and their variance, and so information can be gained on the autocorrelation (similarity of two sample values as a function of their separation distance) of data at varying distances within a population. By using geostatistical analysis, such as correlograms and variograms, to describe the change in autocorrelation with distance, along with mapping of densities, inferences can be made on spatial structure and some of its characteristics (such as patch size) quantified (Cole, 2009; Kraan et al., 2009; Legendre and Fortin, 1989).

In this study the spatial distribution and structure of *C. edule* post-larvae from initial benthic settlement through to their post-winter distribution using spatially explicit sampling with a high temporal resolution were investigated. The hypothesis was that during these initial months after larval settlement post-settlement processes would act upon the post-larvae, creating an increasingly patchy spatial distribution. This would occur during the transition from a large scale passively derived initial distribution with a higher spatial turnover, to a more stable distribution where small scale post-settlement processes become more significant for spatial structure. This was hypothesised to result in a distribution similar to that observed in the adult population within the first year of benthic life. The aim of the work was that by observing when and over what spatial scale these distributional and density changes occur, a better understanding would be gained of those processes involved in driving the observed spatial patterns.

2.2 METHODOLOGY

2.2.1 Study Site

Traeth Melynog is a south-east facing soft-sediment intertidal shore situated along the Menai Strait, Anglesey UK (Fig. 2.1). It forms a bay with a river outflow running along its eastern fringe which then flows into a channel running parallel with the low water level. The sediment is mainly fine sand (Mean D_{50} grain size of 242.4 μm , mean mud content of 8.7 %) and is moderately exposed to prevailing south easterly winds. The mean tidal range is approximately 3.5 metres for MLW to MHW. This site was chosen due to its close proximity, < 20 km from the School of Ocean Sciences, and it supports a well-documented population of *C. edule*. The site does not support consistent commercial harvesting of *C. edule* so the human impact on the population structure and ecology is assumed not to be significant. The site is well protected as an LNR (Local Nature Reserve) by the Countryside Council for Wales and the cockle stocks are annually assessed by Welsh Assembly Government fisheries scientists.

2.2.2 Sampling Design

To gain an understanding of the spatial distribution and patchiness of the *C. edule* settlement post-larval population a staggered nested sampling design (Cole, 2001; Cole, 2009; Kraan et al., 2009) was used at the study site on 8 occasions from June 2010 to January 2011, to monitor the spatial distribution and abundance of the 2010 cohort (Table 2.1). A staggered nested design is based on a series of points along a transect at increasing distances apart from each other, with transects being the unit of replication (Fig. 2.2). This design allows changes in spatial variability and distribution to be measured over time more efficiently than more traditional approaches such as hierarchical nested designs (Cole et al., 2001). This design has the advantage of using inter sample distance as a continuous variable, as opposed to the hierarchical nested approach which treats distance as a categorical variable. This is made possible by knowing the exact geographical position (e.g. Latitude and longitude) of every sample location. By treating distance as a continuous variable in this way, the distance and value differences between any pair of samples in the design can be used to measure spatial variability. This is particularly advantageous as small distances can be replicated using the predefined samples along each transect, and larger distances replicated by analysing the variance between points positioned along different transects. Replication at small scales is often neglected in studies but is the scale over which many biological processes (such as predation and competition) operate, and so it is important to include allowing the ability of detecting small scale patchiness. Data used in this

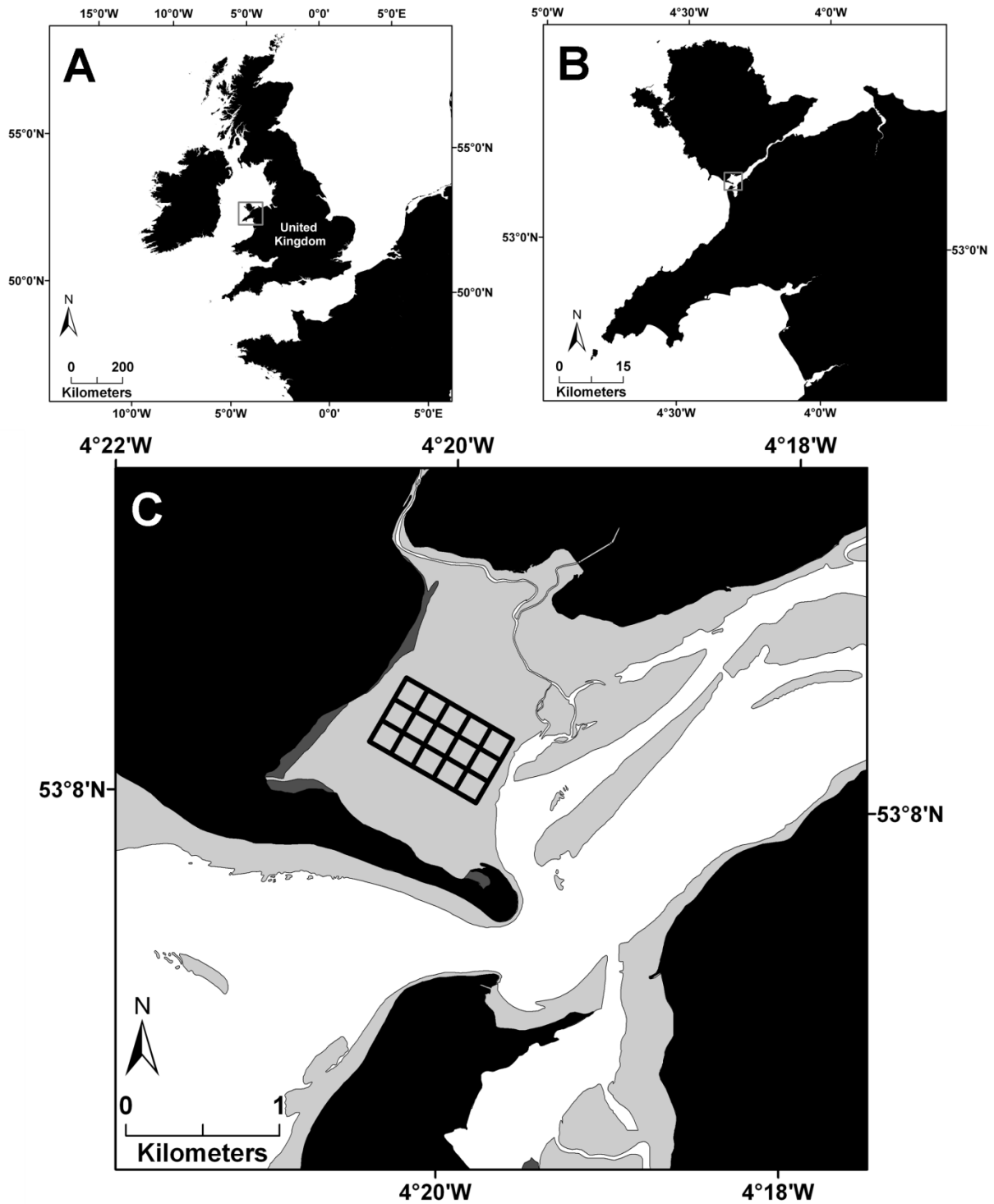


Figure 2.1. Location of the region in North Wales, UK (A) where the tidal flat of Traeth Melynog is situated at the southern end of the Menai Strait, which separates mainland Wales and the Isle of Anglesey (B). The Traeth Melynog *Cerastoderma edule* post-larval population was sampled across a 3 x 5 grid (pictured in black). Light grey areas show intertidal sediment, areas of saltmarsh depicted in dark grey and black areas (with the exception of the grid) is land above MHWS.

manner can be used to produce geostatistical outputs such as experimental semivariograms, which enables an estimation of spatial structure and distances of autocorrelation (Kraan et al., 2009). The distance at which autocorrelation breaks down can be useful for inferring patterns of patchiness and quantifying their grain (size) and lag (distance apart) which can be used to predict possible causative processes.

Table 2.3. Dates and specifics of sampling conducted to access the spatial structure over time of *Cerastoderma edule* post-larvae of the 2010 and 2011 cohort and collection of other variables at Traeth Melynog, Anglesey UK.

Date	Variables sampled	Sample unit
07.06.2010	2010 post-larvae	75 x Core 5.7 cm ² , depth 15 mm
16.06.2010	2010 post-larvae	75 x Core 32.17 cm ² , depth 15 mm
30.06.2010	2010 post-larvae	75 x Core 32.17 cm ² , depth 15 mm
09.07.2010	2010 post-larvae	75 x Core 32.17 cm ² , depth 15 mm
05.08.2010	2010 post-larvae	75 x Core 32.17 cm ² , depth 15 mm
12.08.2010	Adults, predators, <i>A. marina</i>	75 x 0.25 x 0.25 cm quadrat to 3cm depth, sieved over 500 µm mesh
16.09.2010	2010 post-larvae	75 x Core 32.17 cm ² , depth 15 mm
22.11.2010	2010 post-larvae	75 x Core 32.17 cm ² , depth 15 mm
20.01.2011	2010 post-larvae, sediment	150 x Core 32.17 cm ² , depth 15 mm
20.01.2011	Adults, 2010 post-larvae	75 x 0.25 x 0.25 cm quadrat to 3cm depth, sieved over 500 µm mesh
14.06.2011	2011 post-larvae	75 x Core 32.17 cm ² , depth 15 mm
10.02.2012	2011 post-larvae	75 x Core 32.17 cm ² , depth 15 mm
10.02.2012	Adults	25 x 25 cm quadrat to 2cm depth, sieved over 1mm mesh
23.03.2012	Topography	8555 x DGPS points

Five of the sampling events were during the summer of 2010 with some only separated by 10 days. This enabled the occurrence of post-settlement changes in distribution of the 2010 cohort to be accurately identified. The 2011 cohort was also sampled in June 2011 and February 2012 for comparison (Table 2.1). This was undertaken to identify if any general trends are repeated annually in post-larval distribution and spatial structure close to settlement and post-winter. The central area of the study site was selected (800 x 480 metres) from mean low water to mean high water and was divided into 15 square blocks (Fig. 2.1). The total sampling area covered by the 15 blocks was 0.38 km², which is approximately 23 % of the shore. Each block contained a 160 metre long transect with 5 exponentially staggered sampling points at 0, 0.7, 4.4 26.7 and 160 metres along its length (Fig. 2.2). Transects were randomly allocated one of six orientations (0, 90, 135, 180, 225, 270° relative to the high- low-shore axis) to prevent natural gradients biasing results and consideration of anisotropy (directional trends) in the data (Fig. 2.2) (Pettitt and McBratney, 1993).

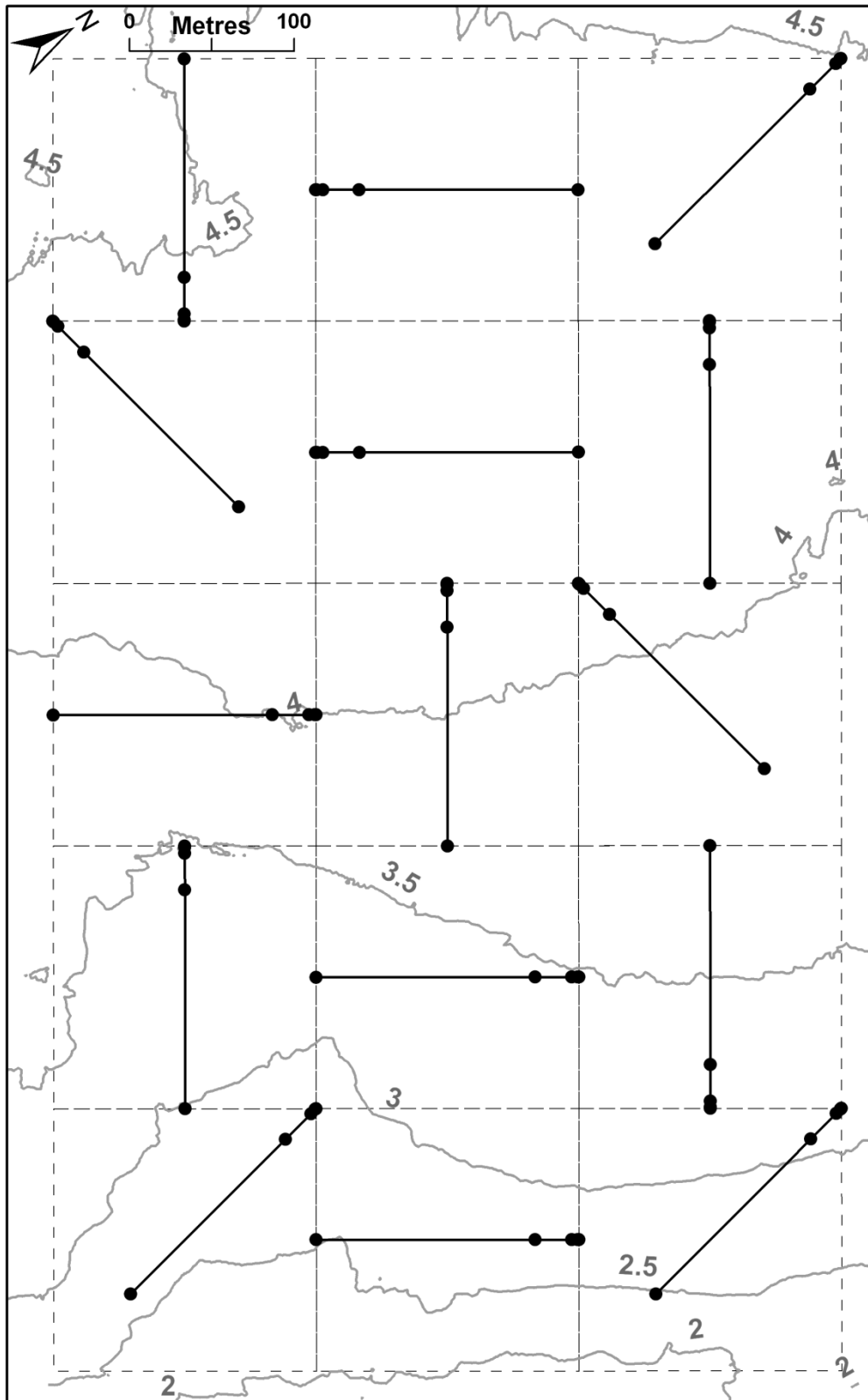


Figure 2.2. Sampling design at Traeth Melynog using 15 transects randomly allocated one of six possible orientations, and from 2 possible directions. Each 160 metre transect has 5 sample points along it at exponentially increasing distances of 0, 0.7, 4.4, 26.7 and 160 metres (closest 3 sample points overlap in this figure). Grey solid contours and labels show interpolated shore height above chart datum in metres based on point DGPS data. The dashed lines show the sample blocks for reference with figure 2.1.

2.3. Sampling approach

GPS was used to locate the first sampling point (0 metres) of each transect in the field and sampled. A tape measure and compass were used to ensure that the 0.7, 4.4 and 26.7 metre samples were taken at the exact predefined distance and bearing along the transect. The tape measure and compass were both used for these samples as the GPS used only had an estimated accuracy of up to 3 metres, which meant at this small scale the potential error would be considerable. The last sample location at 160 metres was located using the GPS. A single core (see Table 2.1 for core sizes) was taken at each sampling point to a depth of 15 mm. Cockles up to 10mm in shell length live in the top 15 mm of the sediment due to their siphon lengths (Zwarts and Wanink, 1989), and so all post-larvae should be retrieved within each core at the depth used. This method was repeated for each of the 15 transects resulting in 75 samples from each sampling event. Samples were placed in cold storage on return (5 °C) to reduce metabolism and oxygen consumption. A 27.7 mm diameter corer was used in the first sampling event in 2011 (June 6th 2011) and a 64 mm diameter corer was used in all subsequent sampling. The 64 mm diameter corer (1/310.8 m²) was considered suitable to ensure adequate numbers of post-larvae and not producing excessive amounts of residual sediment. Due to post-larvae reaching a larger size by the end of the winter after settlement the sample size and sieving was changed to increase efficiency. For the 2010 cohort post-winter sampling (on 20th January 2011) a 25 x 25 cm quadrat (1/16 m²) were used and each sample sieved over a 500 µm mesh in situ. For the 2011 cohort post-winter sampling (10th February 2012) a 1 mm mesh was used with the same quadrat size (Table 2.1).

2.4. Sample processing

Core samples were sieved over stacked sieves of 500, 350, 255 and 125 µm mesh size. The 125 – 255 µm fraction was checked for cockle larvae or post-larvae on the first few sampling occasions, however they were never seen in this fraction. Due to the settlement size of post-larvae (355 – 500 µm) it was decided that removing this fraction would not significantly affect recording newly settled post-larvae. The resultant fractions on each sieve were then sorted under a dissection microscope in filtered seawater, so that cockle post-larvae would remain alive aiding their detection. Bivalve post-larvae were extremely active and would be observed crawling across the petridish. *C. edule* post-larvae were distinguished from other bivalve post-larvae by the difference between the smooth clear prodissoconch (larval shell), and the ribbed brown dissoconch (post-larval shell) (Lebour, 1938) (Fig. 2.3). Other commonly encountered bivalve post-larvae were the abundant tellenid *Macoma balthica*,

Abra tenuis and *Mya arenaria* post-larvae all have a relatively smooth dissoconch and so easily distinguished from *C. edule*. The shell length (greatest distance between the anterior and posterior shell margin) of *C. edule* post-larvae was measured to the nearest 50 microns by dissection microscope fitted with a graticule eyepiece.

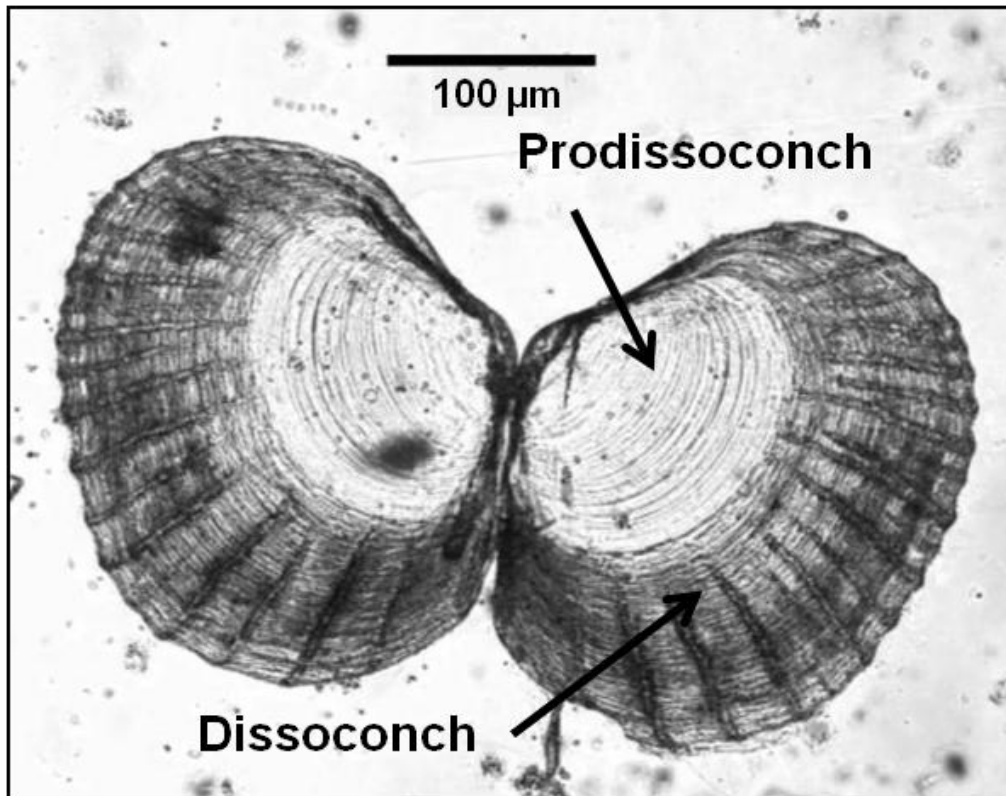


Figure 2.3. Shell of a *Cerastoderma edule* post-larva showing clear definition between the relatively smooth and transparent larval prodissoconch and the post-larval dissoconch which has well defined radiating ribs and orange to brown colouring.

2.2.5. Sediment and tidal level

During January 2011 post-larvae sampling, sediment was collected for particle size analysis using the same staggered nested design to understand how sediment properties vary across the sampling area. The sediment samples were frozen and then analysed using a laser particle size analyzer. Raw data from the laser particle analyser were analysed using GRADISTAT version 8 (Blott and Pye, 2001) to obtain % contribution and D50 values. The sediment is predominately fine sand (as defined by Folk and Ward) with a mean silt content of 8% and a D50 of 245 µm. The silt content was highest in the mid shore to low shore area with the highest silt content patch (21.72%) in the south-western section of the sampling area. A topographical survey was conducted across the sampling area in March 2012 using a DGPS to record how shore height changes across study area. Leica Geo Office 8 was

used to process and calibrate the DGPS data obtained during the topographical survey. Contours of shore height from the DGPS data were interpolated by using ordinary krigging with a 2nd order trend removal as this improved interpolation precision as indicated by a reduction in the Root-mean-squared error when compared to untreated data (Root-mean-squared error = 0.049). Sediment data could not be interpolated accurately by krigging (Root-mean-squared error = 7.11). Topography was not a consistent gradient from high shore to low shore. On the western side of the sample area the high shore extends downshore further than the eastern side by approximately 130 metres (Fig. 2.2.). The shore profile also became increasingly steep towards the low water (Fig. 2.4). On the western side of the sampling area a channel runs down from the mid shore to the low shore which is illustrated by the shore height contours (Fig. 2.2). The mean tidal range on the shore (height above chart datum at the mean low shore level of 2 m up to 4.5 m at the mean high shore level) was divided into 3 equal (equal in regards to height gain above chart datum) shore level categories (Fig. 2.4). These categories are High shore (>3.66 m above Chart Datum, n = 46), Mid shore (2.83 – 3.66 m above Chart Datum Blocks n = 18) and Low shore (< 2.83 m above Chart Datum Blocks n = 11). Both of these variables are of potential help in understanding the distribution and movements of post-larvae and adult *C. edule* at the study location.

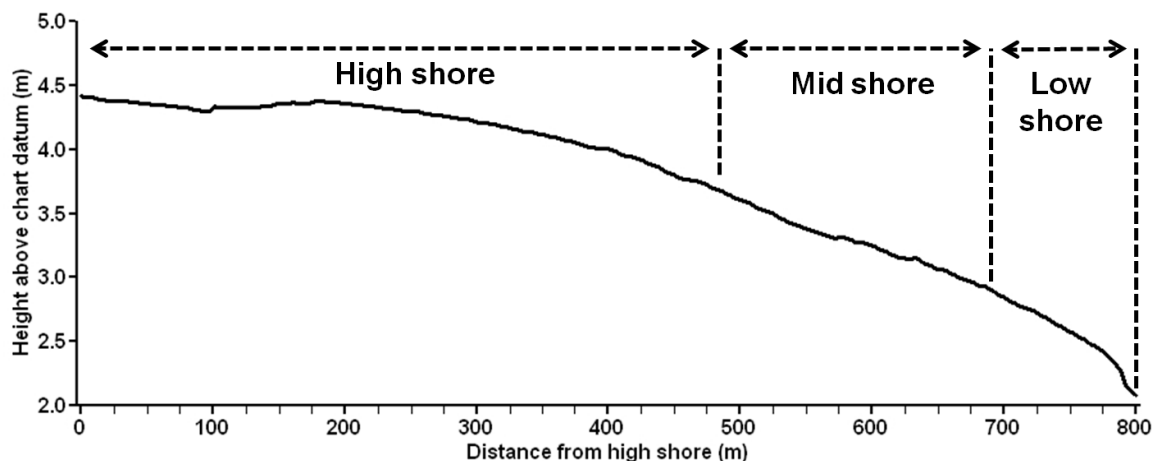


Figure 2.4. Shore height profile above admiralty chart datum along the centre of the sampling area at Traeth Melynog on Anglesey, UK. The shore height categories are based on the range in shore height equally divided into 3.

2.6. Data analysis

Semivariance and associated distance values, Moran's *I* statistics and range values for the semivariogram models were produced using ArcMap GIS software. Experimental semivariograms were then plotted using the statistical package SPSS. Experimental

semivariograms are a plot of the average semivariance (γ axis) against the sample separation distance class, known as a distance bin (x axis). The formula for calculating the semivariance between two sample points is shown by equation 2.1.

$$\gamma_{(a,b)} = \frac{1}{2} (Z_{(a)} - Z_{(b)})^2 \quad (2.1)$$

Half the difference of density values (Z) between location a and b are squared to give the semi-variance (γ) for that pair of samples (a,b). Semivariograms could unfortunately not be plotted for all post-larval data. This was due to a high number of zero counts amongst the 75 sample points on the majority of sampling occasions. High numbers of zero counts mean that autocorrelation will be overestimated, because two locations with zero counts have no variance between them. This masks any patterns in autocorrelation that are present within areas of post-larval presence (Legendre and Fortin, 1989). Samples with zero counts cannot simply be deleted from the data sets to remove this problem as a minimum thirty to fifty data values are needed for stable semivariogram creation (Burrough et al., 1998; Legendre and Fortin, 1989). The 0-year cohorts of post-larvae were identified by studying length frequency histograms of the animals collected. Where regression analysis was conducted the predicted and residual values were checked for equal variance.

2.3. RESULTS

2.3.1. *Cerastoderma edule* post-larvae density.

On the first sampling event (6th of June 2010) only six small cockles were found in all of the samples and their sizes ranged from 625 to 1000 μm . Post-larvae with a shell length below 500 μm , indicating recently settled post-larvae, were only observed on June 16th and September 16th from the 2010 cohort. These two events had clear size cohorts resulting from them in following sampling events (Fig. 2.5). This suggests two main periods of larval settlement occurred during the sampling period between June and September, with the most significant period being in June. The peak post-larval average density of 219 m^{-2} was recorded on June 16th 2010, with densities declining to 74.6 post-larvae m^{-2} by August 5th (averaged over all sampling points) (Fig. 2.6). By January 20th 2011 a final average density of 34 m^{-2} were observed. Adult densities averaged 83 m^{-2} and 67 m^{-2} across the same sampling points on 12th August 2010 and 20th January 2011 respectively. Density in the low shore decreased after the June settlement with densities for the period of the 16th of June to the 9th July decreasing by 83% from 1215 to 170 post-larvae m^{-2} . This decrease was greatly reduced after the July 9th sampling, reducing by only 57 post-larvae m^{-2} and levelling off when the autumn settlement was detected during September (Fig. 2.6). However, the mid

shore density more than doubled between 16th and 30th June, with no settlement in the mid shore indicated from the length frequency histograms during this period. The low shore post-larval density was over 7 times greater than the mid shore on the sampling event at peak settlement in June (974 and 124 m⁻² respectively) but from September onwards the mean densities differ by less than 100 m⁻² and by January 2011 the mean densities differed by only 11 m⁻². At the high shore there were no post-larvae present during the period of peak settlement, but afterwards post-larvae appeared in increasing densities through to September. Nevertheless cockle post-larval densities were always low in the high shore never exceeding 42 m⁻².

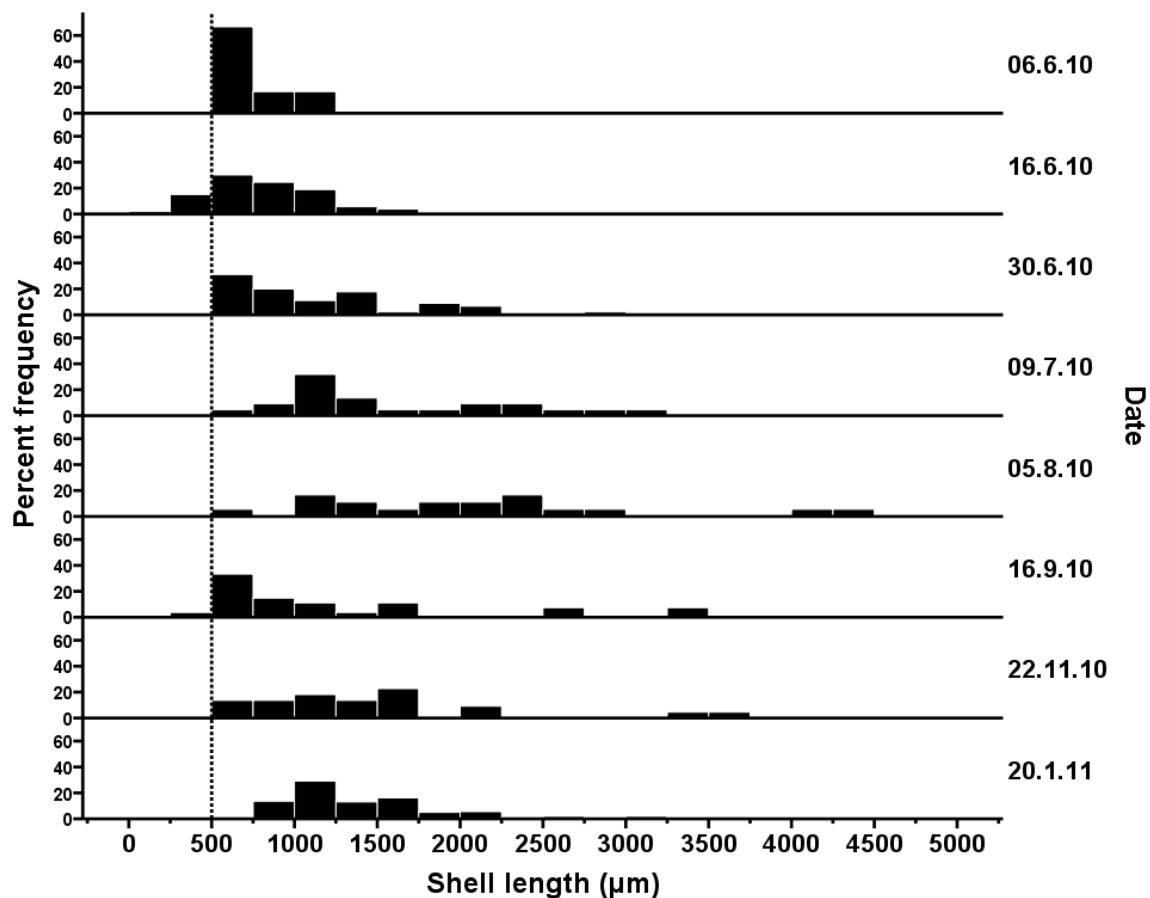


Figure 2.5. Length frequency distribution of *Cerastoderma edule* post-larvae shell lengths (all data) at each sampling date for the 2010 cohort at Traeth Melynog, UK. Dashed reference line shows the approximate 500 µm shell length upper range of newly settled post-larvae. Percent frequency equals 100% per panel.

3.2. Colonisation pattern of post-larvae

The main settlement events recorded in 2010 and 2011 occurred in the low-shore only (Fig. 2.7, 2.8 and 2.9). For both sampling occasions in June 2010 an average of 18 % of the

sampling stations had post-larvae present, compared to 61 % in January 2011 indicating more homogenous distribution in winter. From July 9th onwards post-larvae had a consistent

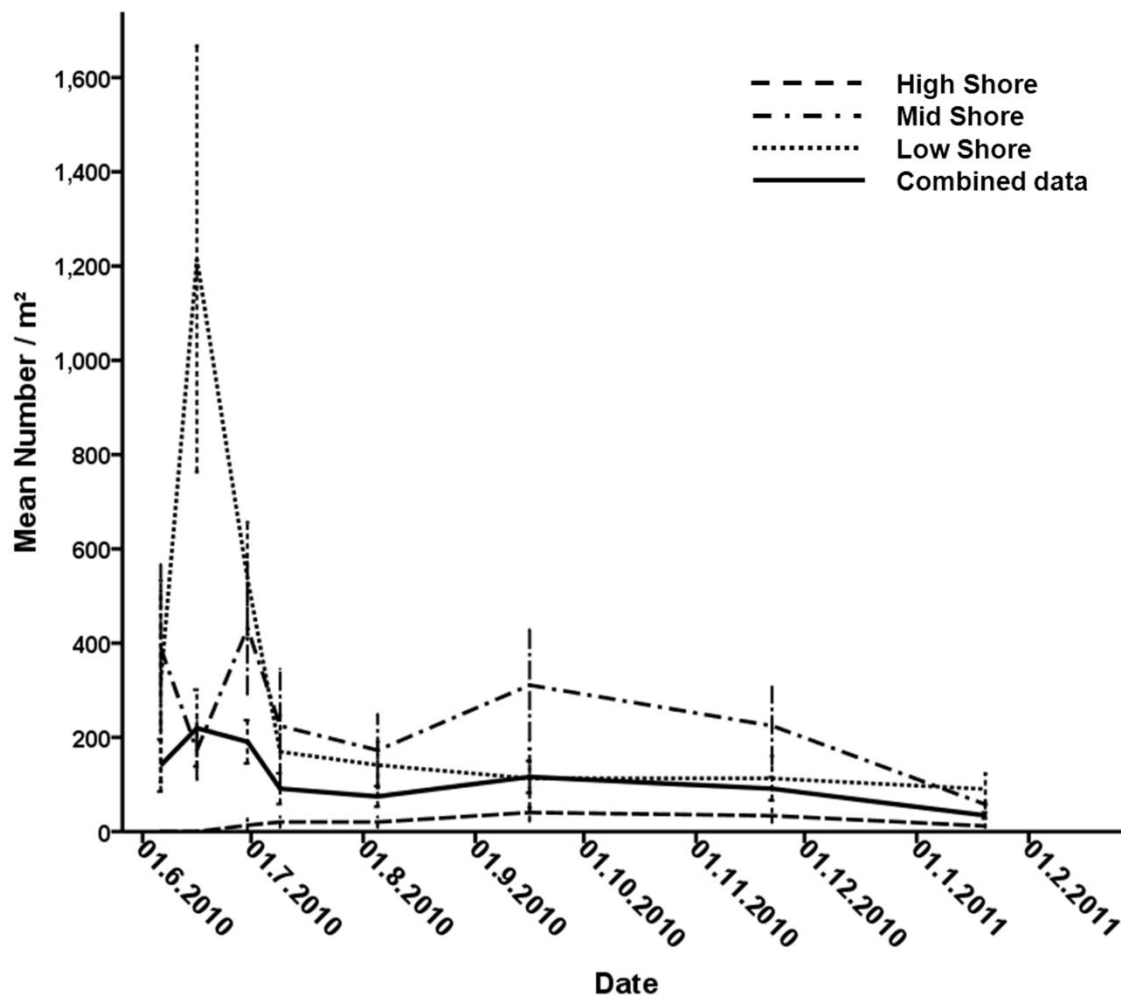


Figure 2.6. Density of *Cerastoderma edule* post-larvae shown at the 3 shore height categories and with all the data combined over time for the 2010 cohort at Traeth Melynog, UK. High shore n = 46, mid shore n = 18, low shore n = 11 and combined data n = 75 for each sampling event. Error bars are ± 1 standard error.

presence in mid and high shore areas as well as the low shore area (Fig. 2.7). Post-larvae presence at specific sampling stations was highly variable over time, with the station of highest density changing between all but one of the sampling events. However the lower eastern side of the low shore had a consistent post-larval presence during the first 4 months post-settlement. Size frequency histograms for the different shore categories further confirm that newly settled individuals (500 μm and below) were only found in the low shore area at one sampling location (apart from one individual) on June 16th 2010. The autumn settlement was indicated by <500 μm post larvae (Fig 2.8) found at the lower end of the mid shore

category on 16th September 2010 and cohort which followed that event (Fig. 2.5).

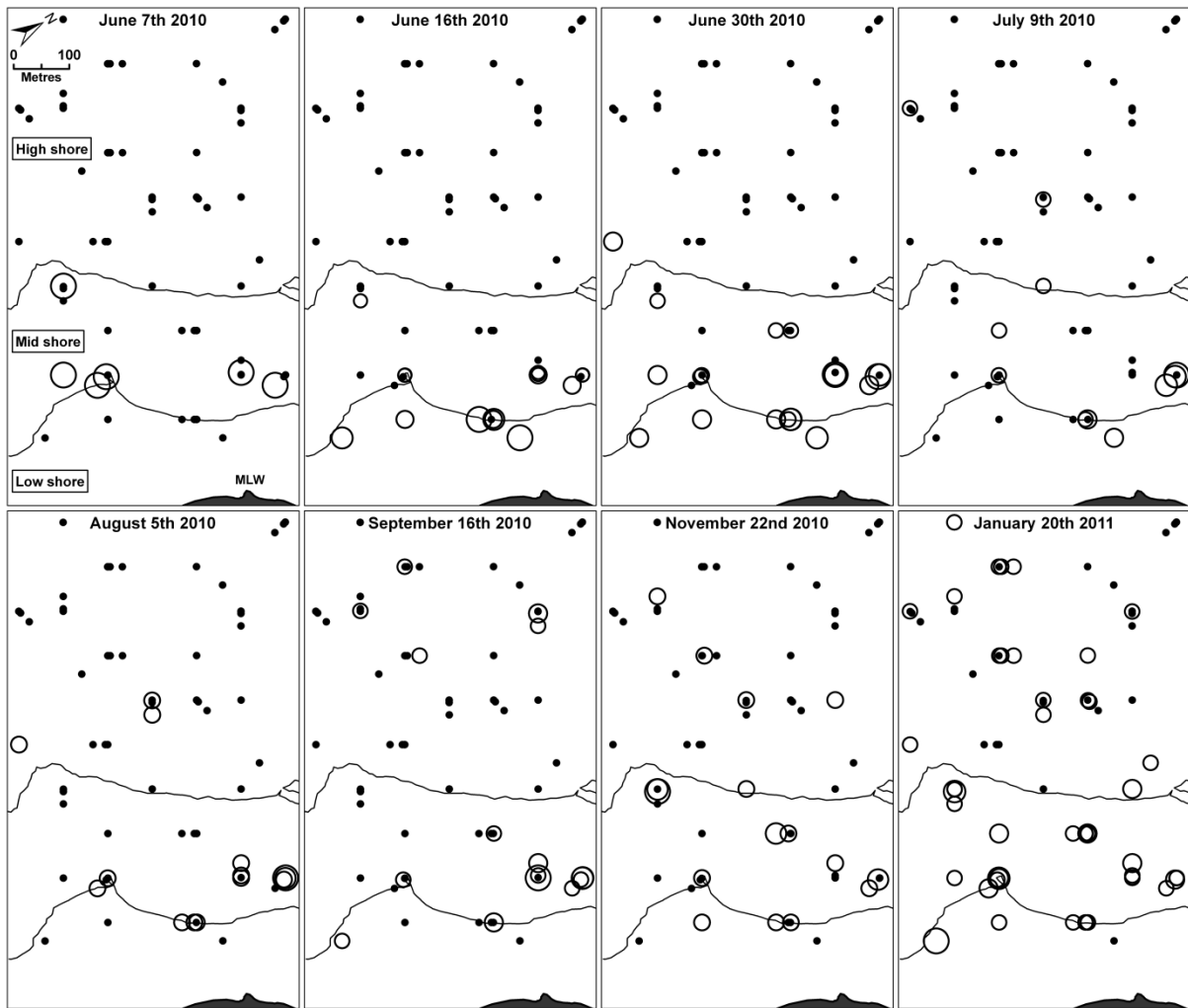


Figure 2.7. Density and distribution of *Cerastoderma edule* post-larvae from the 2010 cohort across the sampling points at Traeth Melynog, Anglesey. Black points indicate zero counts and open circle size is relative to the post-larval density per plot. Shore height categories shown by contour lines and are labelled in the first panel.

3.3. Post-larval distributions compared to the adult distribution over time

Soon after settlement in June 2010 the post-larvae and adults had a very different distribution. However, post-winter the distribution was more similar to that of the adults (Fig. 2.9). For the 2010 and 2011 cohort, correlation coefficients between the post-larval density for each sampling event and post-winter adult density showed a trend of increasing correlation over time (Fig. 2.10) from early summer settlement to post-winter, this indicates that post-larval distribution was becoming more similar to that of the adults over time ($r^2 = 0.45$, $P = 0.033$) (Fig. 2.10). On the 16th of June when post-larvae of a shell length $\leq 500 \mu\text{m}$ were first observed the distributions among stations showed a negative correlation coefficient with adult densities. After the first winter in 2011 and 2012 post-larvae densities

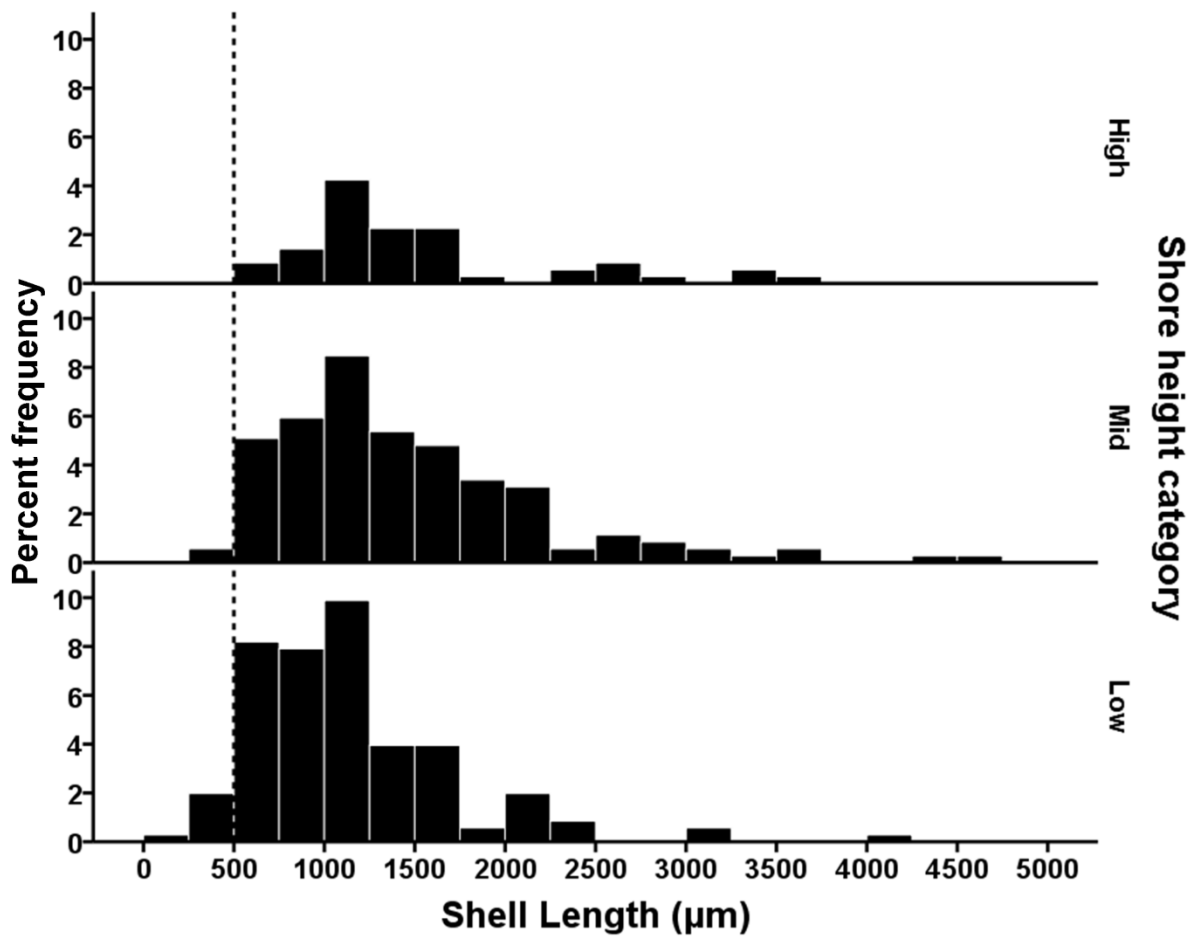


Figure 2.8. Shell length frequency distribution for *Cerastoderma edule* post-larvae for all sampling events by shore height category. Dashed reference line shows the approximate 500 μm shell length upper range of newly settled post-larvae. Percent frequency contribution of each bar is for the whole plot.

showed correlation coefficient values of 0.43 and 0.35 with the adult densities respectively. To put this into context the adult densities across the sampling points post-winter in 2011 and 2012 had a correlation coefficient of 0.44 with each other (Fig. 2.10).

3.4. Geostatistical analysis

Semivariogram plots were only made and used when 45 or more of the sampling points (out of the possible 75) were occupied by *C. edule* post-larvae or adults (Burrough et al., 1998), which excluded most sampling events except the post winter data for the 2010 and 2011 cohort. Semivariogram plots indicate that the post-winter spatial structure of post-larvae differs between the 2010 and 2011 cohort. The 2010 post-larvae cohort and adults both indicate an overall gradient distribution due to increasing semivariance with distance, with

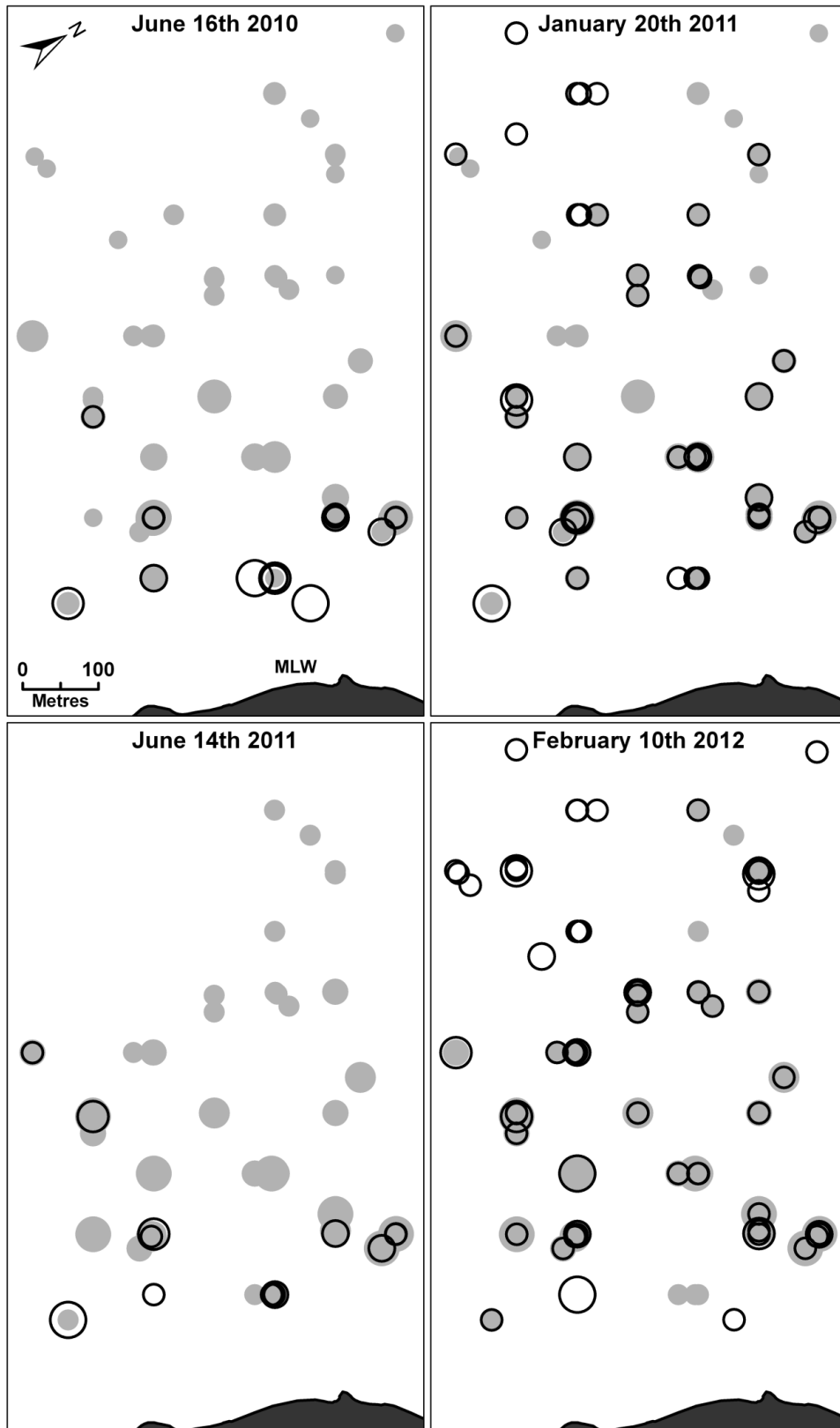


Figure. 2.9. Distribution of *Cerastoderma edule* post-larvae (black hollow circles) during settlement (June) and post winter (January and February) compared to adults (grey circles) across the sampling area at Traeth Melynog on Anglesey, UK. The top two panels show the 2010 cohort and the bottom two shows the 2011 cohort. Size of the circles indicates the relative abundance (not proportional between adults and post-larvae).

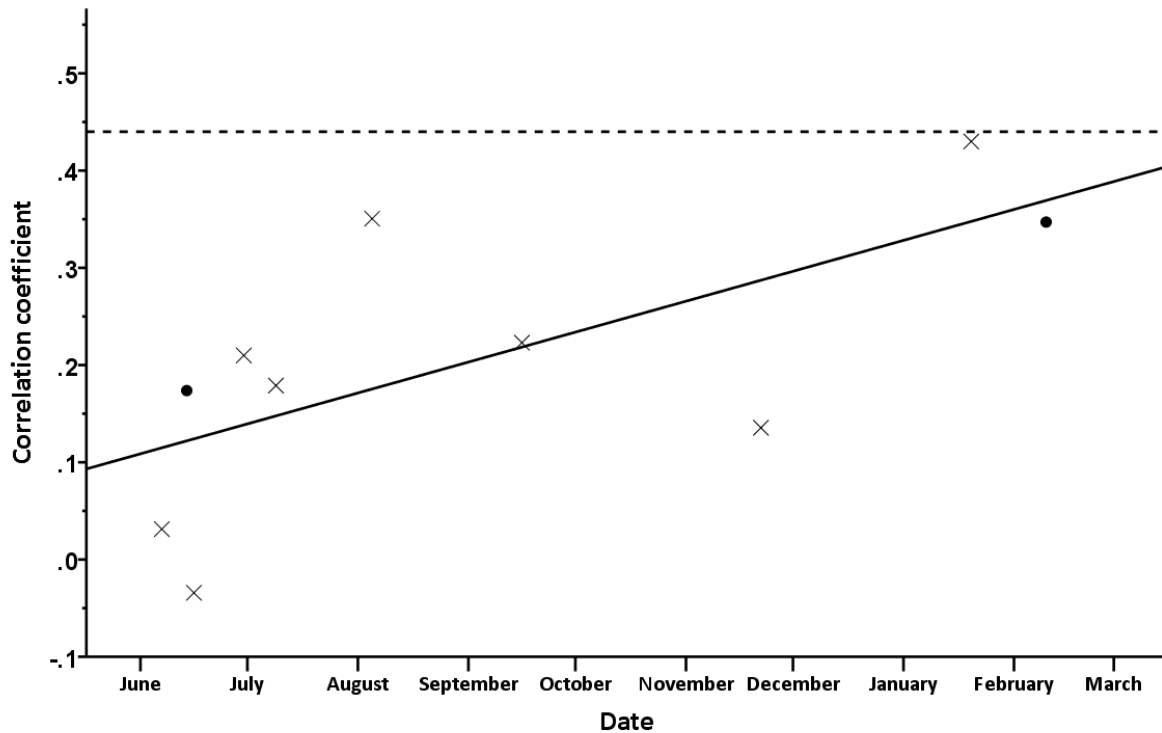


Figure. 2.10. Change in the correlation coefficient between *Cerastoderma edule* post-larvae and adult density over time for the 2010 (crosses) and 2011 (black circles) cohorts. Dashed line indicates the correlation coefficient between the adult distribution in 2010 and 2011, therefore representing the maximum correlation expected. Line of best fit shown for all data ($r^2 = 0.45$, $P = 0.033$).

adults showing an indication of patchiness within this gradient (Fig. 2.11). Moran's I statistic was calculated to help evaluate if densities are spatial clustered, dispersed or random, by using sample distances apart and associated values. The Moran's I for adults and 2010 cohort post-larvae both indicate a clustered distribution (Adults Moran's $I = 1.14$, $P = \leq 0.01$, post-larvae Moran's $I = 0.36$, $P = \leq 0.01$) showing that complete spatial randomness and a dispersed distributions can be confidently rejected. Variography on post-larvae and adult data collected on 20th February 2012 showed that despite the correlation coefficient showing good similarity between the density distributions across the sampling points post-winter (Fig. 2.10), the smaller scale spatial structure between them remains different (Fig. 2.11.C). The adult semivariogram indicates a gradient distribution while the post-larvae semivariogram indicates multiple patches of around 98 metres in size, as determined by a fitted spherical model range, the distance between repeating peaks and troughs (Fortin et al., 2006) and modelled data by Kraan (2010). Moran's I statistic on the same data also indicates a difference in the spatial structure of the post-larvae and adults. The Moran's I statistic for the

adults indicates a clustered pattern ($I = 0.64$, $P = \leq 0.01$) compared to an indicated random pattern ($I = 0.04$, $P = 0.727$) for the 2011 cohort post-larvae.

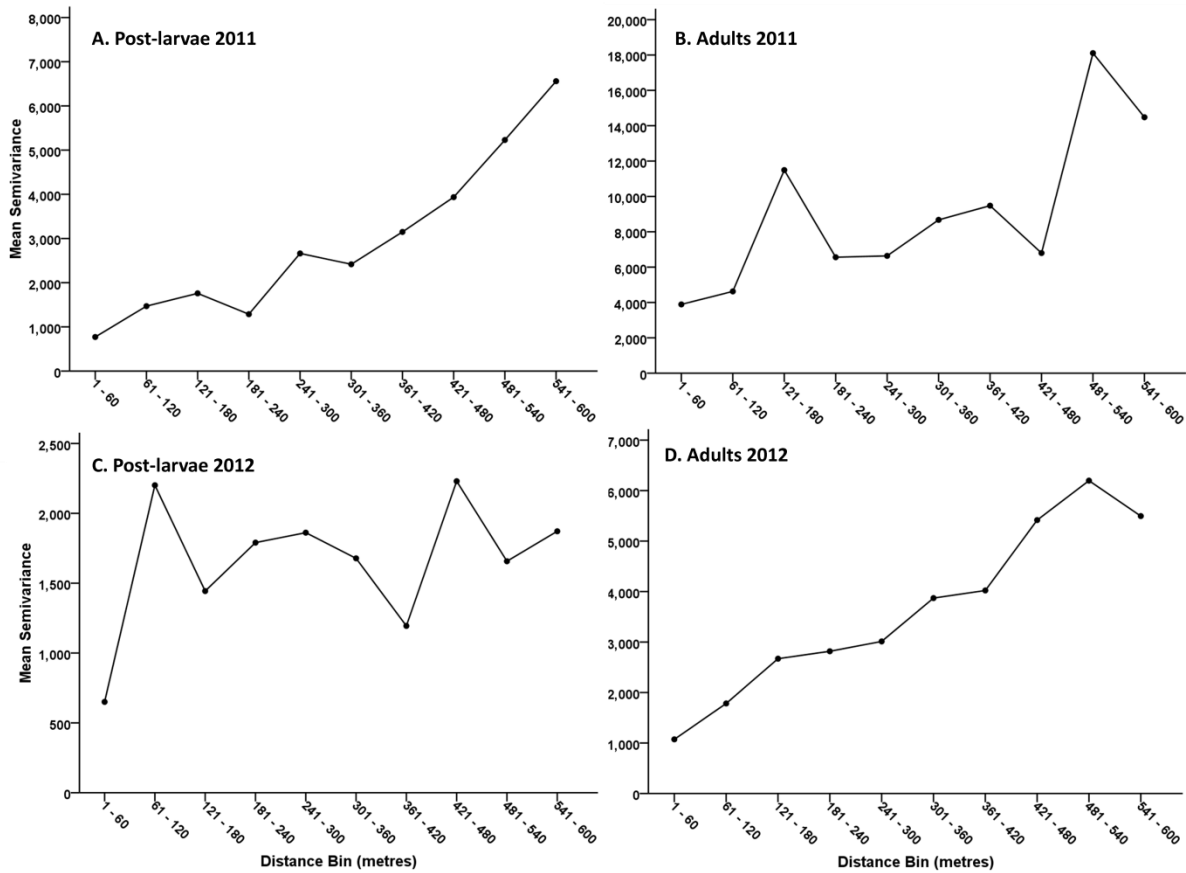


Figure 2.11. Experimental semivariograms of *Cerastoderma edule* post-larvae A) and adults B) post-winter on the 20th January 2011 and post-larvae C) and adults D) post-winter on 10th February 2012 at Traeth Melynog, UK.

2.4. Discussion

2.4.1. Trends in the colonisation pattern over time

Overall densities of post-larvae during and after settlement were lower than expected when compared to other studies (Bouma et al., 2001; De Montaudouin and Bachelet, 1996) and densities from other local shores (unpublished data), which often have 10s of thousands of post-larvae m⁻² at settlement. However, due to the good sample and temporal replication, trends can be identified with reasonable confidence. Disentangling mortality and redistribution is difficult when seeking to understand density changes when studying highly mobile post-larvae, like those of *C. edule*. However the combination of density, spatial distribution and size frequency data in this study allow some conclusions to be drawn with confidence. A hypothesis that spatial distributions of adult cockles are created at larval settlement can be rejected. The data collected over two consecutive years in 2010 and 2011

at Traeth Melynog show that initial benthic colonisation is spatially very different to that of the adults, and that early post-settlement processes may determine the population distribution on the shore. The starting hypothesis proposed that post-settlement processes are highly important for population spatial structure is supported. The finding that post-larval redistribution changes the distribution of post-larval from a low shore concentration to one similar of the adults within the first few months of benthic life is the main evidence in support of this hypothesis. Small post-larvae ($\leq 500 \mu\text{m}$ in shell length) found during the initial colonisation were almost exclusively found in an individual sample in the low shore during both settlement periods in 2010. Perhaps more persuasive is that post-larvae of any size were only found in the low shore areas during the period when the summer settlement would have been occurring, and so they would have to redistribute to colonise other areas of the shore. After initial colonisation in the low-shore there is evidence that post-larvae redistributing up-shore occurred during the summer, creating a distribution more similar to that of the adult population. We observed that densities increased in the mid-shore during the summer and outside the settlement period. Together this indicates post-settlement redistribution is likely to be a key process in spatially structuring this population. Similar studies seeking to measure initial settlement patterns have also noted low shore concentrations of cockle post-larvae (Armonies, 1996; Armonies and Hellwig-Armonies, 1992; Baggerman, 1954; Kreger, 1940), whereas others have noted high shore concentrations (Bouma et al., 2001). What these studies share in common is the observation of a concentrated initial post-larval distribution which changes over time into a distribution which is similar to the adult population. A study on distributions of post-winter juveniles compared to adults over a large geographic range for *Cerastoderma edule* noted that their distributions largely overlap, often in sandy sediments (Compton et al., 2009). This demonstrates that *C. edule* populations maybe largely spatially structured by post-settlement processes throughout their geographic range if settlement is concentrated as observed during this study. Also for the sub-tidal bivalve *Spisula ovalis* it was concluded that the spatial structure and distribution of a cohort is observed in the early juvenile stage, and likely determines that of the adults (Patric et al., 1997). Thus the conclusion is that the post-settlement stage is critical in spatially structuring adult populations.

2.4.2. Spatial structure

It was not possible to observe any changes in spatial structure over time from settlement to post-winter using geostatistical analysis, due to the very concentrated early colonisation of the post-larvae and subsequent redistribution period. However the limited geostatistical analysis that could be conducted on the post-winter data indicated that post-larvae spatial structure was different between that of the adults in the 2011 cohort, and also differed

between the years studied. Post-larvae for the 2010 cohort post-winter had a gradient structure which was similar to that of the adult population. This would support the observation that the large scale spatial distribution and finer structure are largely determined by post-settlement processes within the first 9 months of benthic life. However the 2011 cohort had a multiple patch structure of around 98 m in size post-winter, which was clearly different to the gradient structure of the adults. Due to the large scale distribution appearing close to that of the adults from the correlation analysis and density maps, cautiously it can be concluded that a patchiness creating process acting at a fairly large scale of around 100 metres has affected the finer scale spatial structure of the 2011 cohort. In line with what we have observed for the 2010 cohort post-winter Boldina and Beninger (2013) found that the spatial structure of juveniles and adults was positively correlated and inter-patch distances were similar to those found in this study. However, they compared cockles that are likely in their second winter with adults, and therefore not directly comparable with the post-larval observations in this study.

2.4.3. Possible causes of observed patterns

Initial larval settlement has been considered to be a passive process (passive deposition hypothesis) governed by hydrodynamics since the initial studies into cockle populations (Kreger, 1940; Orton, 1937) through to studies in recent decades (Armonies, 1996; Huxham and Richards, 2003). The predicted result of passive settlement is that a larger volume and longer immersion time of water over the low shore will result in the highest potential for larval settlement there during a tidal cycle (Armonies and Hellwig-Armonies, 1992; Kreger, 1940). Although larval settlement was not directly measured, the observations of initial post-larval distribution in the low shore would agree with this hypothesis. Another hypothesis may be that of higher larval mortality from adult cockle suspension feeding creates such patterns. Adult cockles are important suspension feeders at Traeth Melynog and are at their most abundant in the mid shore areas. They have been observed in the laboratory to kill conspecific larvae through their removing them from the water column during feeding (Andre et al., 1993). This has been supported as responsible for producing an initial negative correlation with adult densities from correlative field studies using settlement density versus adult densities (Andre and Rosenberg, 1991; De Montaudouin and Bachelet, 1996). However these field observations of a negative adult-larval settlement interaction have been seen to be rapidly reversed, most probably by post-settlement migration of post-larvae into areas inhabited by high adult densities (Armonies, 1996; De Montaudouin and Bachelet, 1996; Günther, 1992). Our results are also potentially compatible with this hypothesis, or a more indirect interaction with adults, of initial negative adult-larval interaction through the mismatch of initial settlement and highest adult density distribution. Secondary settlement

through active byssus drifting is largely supported as the mechanism by which cockle post-larvae can reject a settlement location and seek more suitable areas (Armonies, 1996; Huxham and Richards, 2003). However the causative cues, processes or behaviour that initiate post-larvae to byssus drift remain largely unidentified.

A \approx 83 % reduction of post-larval density was observed in the low-shore post-settlement. Whether this occurs due to mortality or redistribution could indicate that this area is likely to be unsuitable for long-term post-larval survival or growth. In the mid-shore area from June 16th to the 30th June the densities increased, independent of any identified larval settlement close to June 30th. This suggests that the mid shore area may have a lower rejection rate than those in the low shore by redistributing post-larvae, and therefore possibly more suitable for long term post-larval colonisation. This would need further experimental investigation to be supported. However densities did fall again between June 30th and July 7th in the mid shore, as did the low shore. During this same period high shore densities increased with no detected settlement supporting a suggestion that post-larvae are redistributing in an overall up-shore direction post-settlement. The observation of up shore redistribution may be supported by the adult *C. edule* population ecology. Adult densities are highest in the mid shore areas and extend into the high shore at Traeth Melynog (Unpublished data and Sanchez-salazar et al.(1987)). For the adult distribution pattern to persist over time at Traeth Melynog which appears to occur (Unpublished data), post-larval redistribution would be critical to the population persistence if settlement patterns found in this study are consistent over time.

2.4.4. Possible benefits of post-larval redistribution and observed spatial structure

In *Macoma balthica* migrations occur by post-larvae drifting from the settlement area in the lower parts of the shore to higher tidal levels during the summer (Gunther, 1991), although *M. balthica* migrates again in the winter back to sub-tidal locations. These migrations have been shown to be potentially costly for the population through increasing predation risk whilst suspended in the water column, despite occurring at night when predation risk is lower (Hiddink et al., 2002a), but have been suggested and modelled to have a probable benefit on fitness and reproductive output overall (Beukema, 1993; Hiddink, 2003). The mechanisms and possible advantages of migration have been studied less for *C. edule*, but some predictions can be made based on the following knowledge. Several studies have been conducted on the adult population at Traeth Melynog, with observations that growth potential increases with increasing immersion time (Richardson et al., 1980; Sutherland, 1982b). In addition densities of *C. edule* are highest in the mid shore, most likely due to effects of low-shore crab *Carcinus maenas* predation and high-shore oystercatcher

Haematopus ostralegus predation on juvenile and adult individuals of *C. edule* respectively (Sanchez-Salazar et al., 1987). These distributional trends at Traeth Melynog are shared for many other populations (Beukema and Dekker, 2009) but can vary across the species geographic distribution (Bocher et al., 2007). Based on the adult distribution and ecology an advantage can be seen of post-larvae redistributing from low to mid shore areas by avoiding marine derived predation in the low-shore which is restricted by tidal immersion. However this would come at a cost in a loss in potential growth rate from decreased immersion time and therefore feeding time. It must be remembered when making such comparisons with adult ecology that post-larval cockles are susceptible to different predators than adults. Juveniles of *Carcinus maenas*, *Crangon crangon* and *Pleuronectes* spp. are all important epibenthic marine predators whose predation opportunities are also likely to increase with immersion time (Beukema and Dekker, 2005; Jensen and Jensen, 1985; van der Veer et al., 1998). These predators themselves are open to predation from other macrofauna, as with the cannibalistic *C. maenas* (Moksnes et al., 1998), with a similar reduction in predation risk with decreasing immersion time however. Reaching a size refuge is important for *C. edule* post-larval survival against predation as their size and shell strength is linked and there is evidence that post-larvae can reach a size refuge which reduces predation risk (Jensen and Jensen, 1985). If growth potential and predation mortality are the dominant factors in post-settlement survival and recruitment success, then the mid-shore is likely to be the optimum location for both post-larvae and adults to colonise and remain during their life history. Therefore the redistribution of post-larvae in to the mid-shore may facilitate the population's persistence over time by ensuring recruitment in the optimal area for survival.

2.4.5. Conclusions

The focus of initial post-larval colonisation in the low shore and subsequent redistribution indicates that post-larval mobility is important for recruitment and distribution in the population of *C. edule* at Traeth Melynog. Our results show the importance of post-settlement processes within the first 9 months for determining the abundance and spatial distribution and structure of *C. edule* post-larvae settling into an established population. Another implication of our findings is that locations thought unimportant for supporting high adult densities, may be highly significant for early post-larval colonisation, and therefore recruitment success of a population. If cockle populations are managed for fishery or conservation objectives then identifying areas which are important for initial post-larval colonisation may be important for protecting the potential for recruitment success in to populations.

Acknowledgements

The authors would like to thank Leo Clarke, Enrique Gonzalez and Nicholas Jones for sampling assistance and John Leedham and Claudia Tanneberger for field and laboratory work assistance, The Countryside Council for Wales allowed access to Traeth Melynog and the Welsh Assembly Government Fisheries gave permission to sample the cockle population.

Chapter 3: Changes in small scale spatial structure of *Cerastoderma edule* post-larvae in the Dee estuary

Timothy Andrew Whitton, Stuart Rees Jenkins, Christopher Allan Richardson, Jan Geert Hiddink

Abstract

Understanding the spatial structure of an invertebrate population at different scales can help reveal key processes controlling abundance and distribution. Large scale changes of distribution in post-larvae of the common cockle *Cerastoderma edule* have been observed and are important for population structure (see Chapter 2). However knowledge on small scale changes is limited with small spatial scales being relevant for key process such as predation and other inter- and intraspecific interactions. It is predicted that spatial patterns of post-larvae over small spatial scales will change as post-settlement processes take effect. A small scale field survey over an area of 60 x 80 metres was conducted using a spatially explicit staggered nested design on three occasions to record the spatial structure of *C. edule* post-larvae from initial settlement in June 2011 to March 2012 at the Dee estuary, UK. To investigate changes in spatial structure empirical variograms, Moran's I correlograms and prediction mapping were used along with calculated correlations between variables. At the first sampling event in June 2011, when initial settlement was occurring, post-larvae were highly aggregated with patches interpreted at 12 -18 metres in size and 18 – 24 metres apart. By October 2011 the post-larvae had become more evenly dispersed with some small scale (< 6 metres in size) random patchiness and an underlying gradient. This spatial structure was maintained into March 2012 but with increased patchiness. At the time of initial settlement correlation showed no correlation with adult abundances or sediment mud content, but by October 2011 and March 2012 there was a strong positive correlation with adult abundances. It is concluded that at small spatial scales initial settlement of post-larvae is highly patchy and as they become recruited into the population before they become more dispersed and their distribution positively correlated with adults. These small scale changes are important in interactions between predators and adults and in understanding habitat use by *C. edule* populations.

1. Introduction

Understanding the spatial structure of a population is a fundamental part of understanding its ecology, and is also relevant in designing appropriate sampling strategies (Thrush, 1991; Underwood et al., 2000). By spatial structure I mean the pattern of how individuals are organised in space (e.g. uniform, random, patchy) and the quantification of that pattern (size of patches and their distance apart). The scale, characteristics and temporal variability of a population's spatial structure can indicate which processes may be important in creating or altering a population's distribution (Bergström et al., 2002; McArdle et al., 1997; Underwood et al., 2000). A failure to understand natural spatio-temporal patterns may lead to ill-informed and inappropriate identification and the testing of processes which determine basic population parameters. Without detailed observational data an understanding of the processes structuring communities and their ecological interactions cannot be progressed. Although unable to attribute causation directly, measuring the spatio-temporal patterns of a population, in conjunction with correlative analysis of variables identified as potentially important, can assist in elucidating which processes are operating and these can then be tested in future work. For example by observing small scale patchiness in soft sediment infauna with consistent interspecific positive and negative correlations between two sites with different hydrodynamic regimes, Bergström et al. (2002) hypothesized that the spatial structure was generated through biotic interactions.

The spatial structure of the common cockle *Cerastoderma edule*, a commercially and ecologically important infaunal soft-sediment bivalve, is often highly patchy (spatially aggregated) on large scales of thousands of metres (Kraan et al., 2009) and smaller spatial scales of tens of metres (Boldina and Beninger, 2013). The reasons for such biological spatial heterogeneity within a more spatially homogenous physical environment are not clearly understood (Kraan et al., 2009). However the post-larval stages of many soft sediment invertebrates are highly mobile and can have very high mortality rates, and so this life stage is a likely origin of this observed underutilisation of the available habitat (Armonies and Reise, 2003).

The post-larval stage (defined here as the period from larval settlement until the end of the 1st year) of *C. edule* is a critical period for determining a cohort's abundance and distribution, primarily through very high early post-settlement mortality (Beukema and Dekker, 2005; Jensen and Jensen, 1985) and redistribution (or secondary settlement) through bysso-pelagic migrations (Armonies, 1994b). The large scale ($\approx 1 \text{ km}^2$) spatial structure of *C. edule* post-larvae has been shown to change after initial larval settlement (Armonies, 1996; Bouma et al., 2001; Jung et al., 2006; Kraan et al., 2009; Lindegarth et al., 1995), and is spatially restricted when compared to the wider population, but post-larvae

often become more dispersed over a wider area post-settlement. The findings in chapter 2 add to and agree with these observations on large scales of hundreds to thousands of metres. From such observations of distribution changes over time, insights into key processes and the spatial scales over which the processes are operating may be gained. In the case of large scale observations there is good evidence that the initial patterns are created through passive deposition of larvae where currents are low or converge (Armonies and Reise, 2003). The subsequent post-settlement redistribution is facilitated by currents due to the distances involved of hundreds of metres, and previous knowledge of the ability of post-larvae to drift in currents over such distances. This is suggested due to the understanding that larval selection of sediment can only operate on very small spatial scales (cm) during settlement because of larval swimming speeds, and large scale correlations with fine sediment indicating a passive deposition of sinking larvae. Simultaneous recording of shear stress, sediment erosion and deposition and sediment content have been used in correlation analysis with post-larvae, and proved useful in interpreting pattern generating processes over these large scales (Armonies, 1996). However there is little information about spatial patterns, how they change over time and the processes driving them over smaller spatial scales (centimetres to tens of metres). Over a small spatial scale (<100 m) Boldina and Beninger (2013) concluded that juvenile *C. edule* (likely 1- and 2-group cohorts) were highly aggregated (patchy) with adults being more homogenous (random). However this work did not consider the structure of post-larval (0-group) individuals.

Over large scales the soft-sediment tidal flat can appear homogenous, but over smaller scales heterogeneity can be observed. Pools of standing water, loose algal mats (Arroyo et al., 2012; Norkko, 1998), meiofauna and microphytobenthos (Pinckney and Sandulli, 1990) are a few examples of variables which can be heterogeneous at scales of centimetres to metres on tidal flats. The importance of small scale patchiness is important to consider. The spatial structure of post-larvae at small scales may be highly influential on their survival and fitness. For example the juvenile shore crab *Carcinus maenas* and juvenile brown shrimp *Crangon crangon* have an important role in early post-settlement mortality of *C. edule* (Beukema and Dekker, 2005; Jensen and Jensen, 1985; van der Veer et al., 1998), and their ability to locate *C. edule* post-larvae and the efficiency with which they can feed on them is likely to be influenced by the spatial structure of the post-larvae (Andresen and van der Meer, 2010; Clark et al., 2000; Thrush, 1999). In chapter 5 I demonstrate that the survival of cockle post-larvae under predation threat from *C. maenas* can be considerably higher when aggregated in their spatial structure when tested against a uniform distribution at the same density. Predation has a large scale effect on post-settlement survival (Beukema and Dekker, 2005), but it will be the small scale spatial and behavioural components of this process which are driving its importance for the larger cockle population.

Other more indirect interactions can also operate on small scales such as amensalism. Intra-specific interactions with adults are often inferred to be important in settlement and post-settlement processes, largely in a negative context through larval ingestion (Andre et al., 1993) and disturbance that causes mortality through bioturbation (Flach, 1996). In Chapter 4 I introduce and investigate macrofauna interactions in more detail.

The post-larvae of *C. edule* recorded soon after initial settlement are likely to range in size from 350 - 1000 μm in shell length, and are infaunal. This makes measuring spatial structure technically difficult and time intensive due to sample processing requiring the microscopic location of post-larvae amongst similar sized sand particles. The cryptic nature of the post-larvae in the environment also means the sampling design used has a potentially significant influence on identification of patterns in spatial structure (Thrush, 1991). Due to the highly mobile nature of *C. edule* post-larvae and the recorded distribution changes that can take place at large scales, it is important to temporally replicate sampling to gain an accurate picture of the spatio-temporal structure in post-larval distribution. Geostatistics are increasingly used in marine ecology to quantify spatial patterns and structure in species distributions (Fortin et al., 2006; Legendre and Fortin, 1989; Liebhold and Gurevitch, 2002). This group of statistical analyses is of particular use for understanding spatial structure because it treats distance as a continuous variable and uses information from almost all of the samples taken (Cole, 2009). It can be a powerful tool if sampling is appropriately designed.

In this study it was hypothesized that the initial small scale spatial structure of post-larvae will be patchy, as suggested by large scale studies, and that structure will soon change due to large scale processes such as drifting which will reduce densities across an area and create a more uniform distribution. In this study the objective was to record and quantify how the spatial structure changes for a single cohort of *C. edule* post-larvae from initial settlement through to the following post-winter period over a small spatial scale (metres to tens of metres). Repeated use of a spatially explicit sampling design allowed the observation of spatial patterns over time, and an understanding of how they change. Similar observations on the sediment properties and distribution of adult *C. edule* may help to establish possible drivers for the observed patterns through comparing spatial structure and correlations with post-larval density.

2. Methods

2.1. Study site

Sampling was conducted on the Thurston cockle bed in the Dee estuary, UK. The Dee estuary drains the river Dee into Liverpool Bay (Irish Sea) and is macrotidal with a range of 7 – 8 metres at its mouth (Moore et al., 2009). The Thurston cockle bed is located on an intertidal mud and sand bank situated on the eastern side of the estuary which drains in a north westerly direction running parallel with the shore line (Fig. 3.1). This cockle bed is traditionally and intensively fished for > 20 mm cockles when densities are high enough from July until the total allowable catch has been harvested, but the bed was not fished during the entire study period.

2.2. Sampling design

2.2.1. Identifying a location for the small scale spatial sampling

The initial settlement of *C. edule* can be highly concentrated spatially as shown in Chapter 2. To determine a suitable location with post-larvae present in high densities for the small scale spatial sampling, a large scale survey of post-larval abundance was conducted across the Thurston cockle bed on May 3rd 2011 (Fig. 3.1.A.). A 64 mm diameter core was taken to a sediment depth of 15 mm at each of 18 stations distributed among 3 transects (each was 1.2 km long), which were placed parallel to the estimated low, mid and high shore levels of the shore (Fig. 3.1). This survey was then repeated on September 16th 2011 to reveal how post-larval abundances may have changed over a large scale and the relative importance of the small scale spatial sampling location for post-larval colonisation in the population. Two possible sample locations shared the highest abundance from the May sampling, the single location used was chosen as it was furthest from potential disturbance by human activity and a large drainage channel (not shown) (Fig. 3.1). This location was sampled with a 64 mm diameter core on 3rd May, 7th June, 21st July, 1st August, 16th September, 10th October 2011 and 7th March 2012 to monitor changes in size frequency and density of *C. edule* post-larvae during the study period. Stacked sieves with a mesh size 500, 350, 255 and 125 µm mesh size were used to help sort *C. edule* post-larvae from the sediment and other fauna.

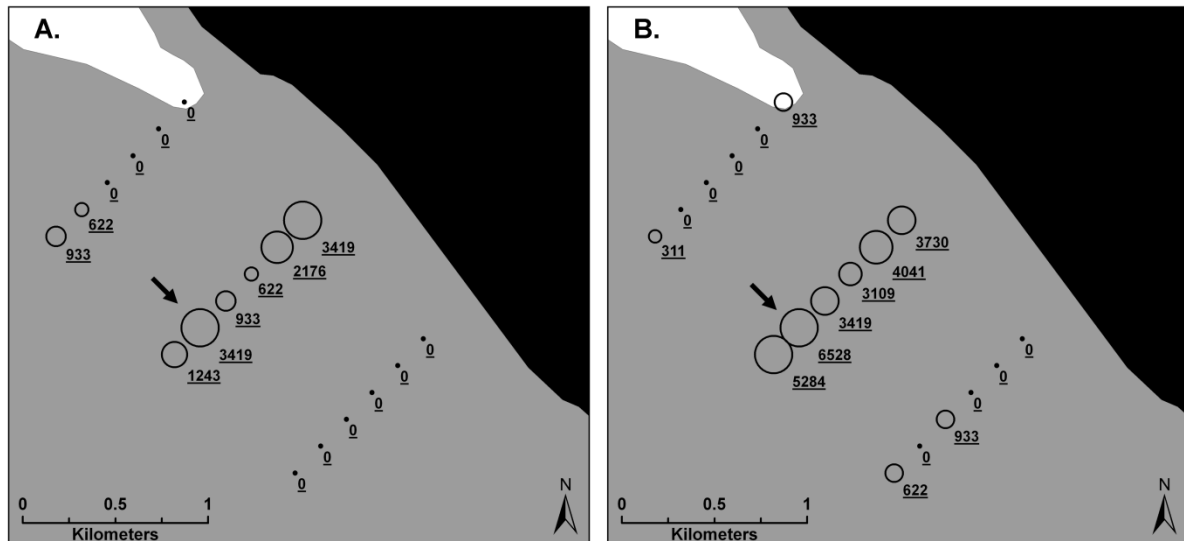


Figure 3.1. Densities (relative size of circle and underlined value of individuals) of *Cerastoderma edule* post-larvae m^{-2} from the large scale surveys on the Thurston cockle bed in the Dee estuary on A) 3rd of May and B) 16th September 2011. Shore height decreases from a south easterly to north westerly direction in parallel with the shore line. The black arrow in each plot indicates the station used for small scale spatial sampling. Black areas indicate land, grey intertidal sediment and white represents predominately subtidal areas.

2.2.2. Small scale spatial sampling design

To understand how the spatial structure of post-larvae changes temporally a small scale Staggered Nested Design (SND) was chosen to allow robust spatially explicit analysis (geostatistical analysis). A SND for the purposes of geostatistical analysis can be described as samples placed at various distances apart from each other nested along a transect or area which is then replicated (Cole, 2009; Cole et al., 2001). This approach was demonstrated by Cole (2009) with the bivalve *Austrovenus stutchburyi* and shown to have advantages over the more widely used hierarchical nested design for investigating spatial variability. Geostatistical analysis (utilising the XY locations of samples) such as variography and Moran's I correlograms allow the plotting and quantification of the spatial structure of a variable (Fortin et al., 2006; Kraan et al., 2009; Legendre and Fortin, 1989). Variograms and correlograms both plot a measure of variability between samples at different distances apart, which allows some insight into the spatial structure (e.g. is the variable spatially patchy, random, dispersed). For geostatistical analysis to be robust a high replication of different distances between samples is needed, particularly small distances which are often under-replicated. A SND approach allows this, as replication at small distances is determined along each transect with larger distances being replicated between samples on different transects. For all sampling events stations were sampled along twelve 20 m long transects placed within a 60 x 80 metre area. This was deemed as an appropriate spatial extent to investigate

spatial structure of metres to tens of metres. Transects were randomly placed at bearings of 45, 91, 135, 180, 225, 270, 315, 360° to avoid biasing the spatial structure through any natural gradients (Fig. 3.2). The initial design had five samples at exponentially increasing distances of 0, 0.1, 0.6, 3.4 and 20 metres along the transect totalling 60 sample stations for the whole survey (Fig. 3.2). This was to provide good replication at a variety of small distances (Fig. 3.3). After the first sampling event on June 7th 2011 it was decided to add two extra points along each transect at 7.55 and 11.70 metres for all subsequent sampling events (Fig. 3.2). This was carried out to increase the replication at small to medium distances and a 29% increase in sampling effort from the two extra points yielded a 50% increase in measurements of spatial variability (Fig. 3.3).

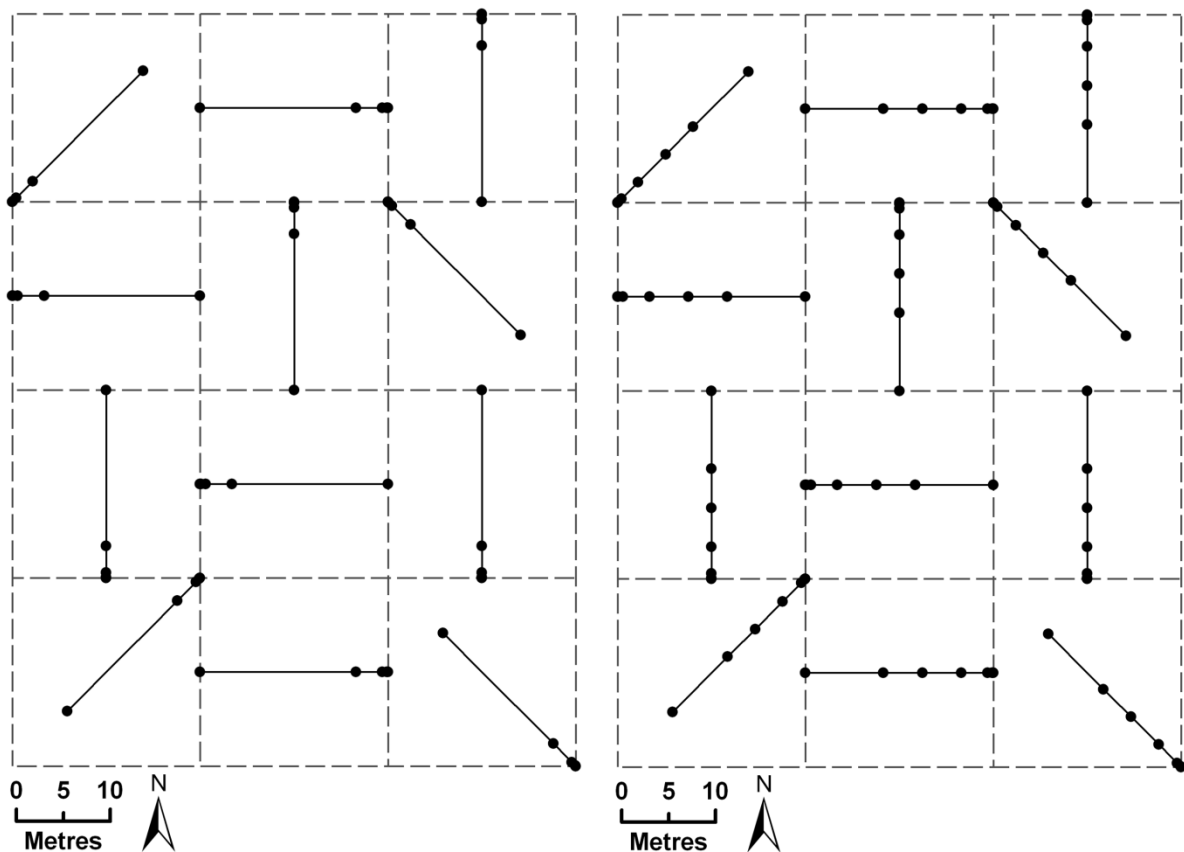


Figure 3.2. Staggered nested design used at the Dee estuary to sample small scale spatial structure of various variables. The design used on 7th June 2011 (left plot) used 5 points on each transect at 0, 0.1, 0.6, 3.4 and 20 metres along the transect totalling 60 sample stations. The design used on the 10th October 2011 and 7th March 2012 (right plot) used 7 samples along each transect at 0, 0.1, 0.6, 3.4, 7.55, 11.7, and 20 metres along the transect totalling 84 sample stations.

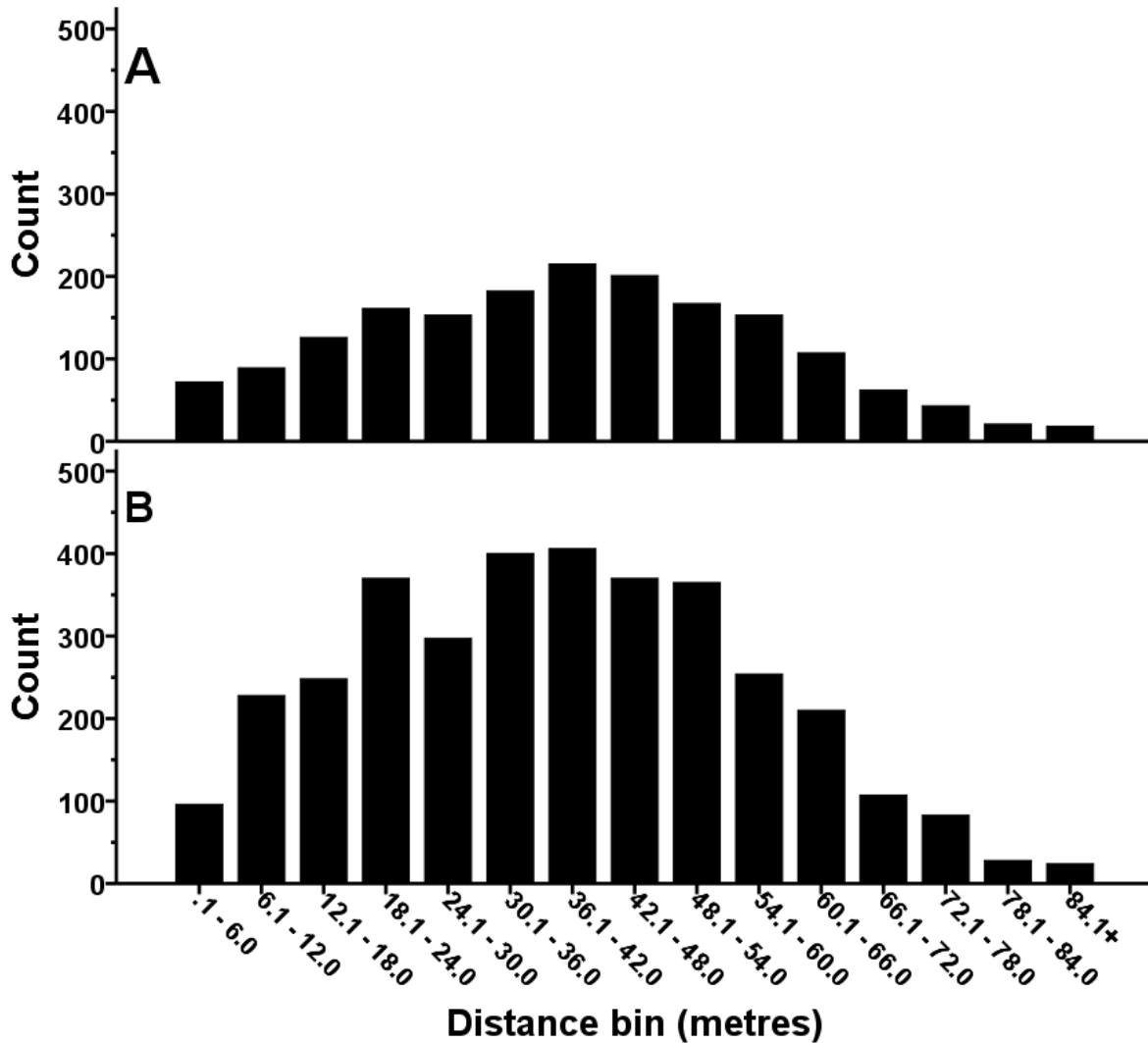


Figure 3.3. Replication of variance measurements for each 6m distance bin produced from the staggered nested design A) used on 7th June 2011 (n = 1,736) and B) the design used on the 10th of October 2011 and 7th March 2012 (n = 3,444).

2.2.3. Sampling procedure

A combination of a GPS unit, navigation compass and a tape measure were used to locate each transect and associated sample locations. Sampling of *C. edule* post-larvae was conducted to cover possible spatial structure changes during what were predicted to be important time periods. These were at settlement around June to a post-settlement period in October, and then through to post-winter in March (Table 3.1). Adults were also sampled in October and March along with sediment samples so that correlations with these could be investigated (Table 3.1). Samples were refrigerated at 4° C on return to the laboratory.

Table 3.1. Details of the sampling regime to investigate the spatial structure of *Cerastoderma edule* post-larvae in the Dee estuary, UK. A staggered nested design was used and all samples were taken to a sediment depth of 15 mm. n/a: not applicable.

Date	Variable sampled	Sample unit	n	Min mesh size
07/06/2011	<i>C. edule</i> post-larvae	64 mm corer (201.06 mm ²)	60	125 µm
10/10/2011	<i>C. edule</i> post-larvae	64 mm corer (201.06 mm ²)	84	125 µm
10/10/2011	<i>C. edule</i> adults	0.1 m ² quadrat	84	1 mm
10/10/2011	Sediment	27.7 mm corer (87.02 mm ²)	84	n/a
07/03/2012	All <i>C. edule</i>	0.1 m ² quadrat	84	1 mm

2.2.4. Sample processing

Cores taken to sample the post-larvae were sieved over stacked sieves of 500, 350, 255 and 125 µm mesh size. Under a dissection microscope each sieved fraction was searched through for any *C. edule* and their abundance and shell length (largest distance between the anterior and posterior margin) recorded. This was done immediately after sampling as samples were only refrigerated, as live post-larvae are easier to identify and separated from other fauna. Sediment samples were analysed using a Malvern laser particle size analyser. This produced the percentage contribution of sediment size fractions by volume, and the calculation of the D₅₀ (median particle size) for correlation analysis with the fauna data.

2.3. Data analysis

2.3.1. Data treatment

Due to evidence of a prolonged period of larval settlement in August, September and October 2011 it was decided that post-larvae belonging to these settlement events should be removed from the spatial analysis. This was possible as two clear cohorts each fitting a normal distribution could be seen in the size frequency distribution, identifying the early summer from the autumn settlement in the October 2011 spatial sampling data. Post-larvae from the early summer settlement were identified as being between 2.5 and 14 mm in shell length on 10th October 2011. Including the autumn settlement in the analysis would very likely mask the development in spatial structure of the main 2011 cohort settlement over time. This would occur because processes may be dependent on the time of settlement, ontogeny or size of the post-larvae and would create noise in the data inhibiting accurate analysis of the main spring/summer settlement. Adults were defined as individuals greater

than 18 mm on 10th October 2011 and greater than 19 mm in March 2012 from size frequency distributions from both sampling events.

2.3.2. Spatial analysis

Empirical variograms and Moran's *I* correlograms were the geostatistical methods used for analysing changes in the spatial structure of the *C. edule* post-larvae and both are based on autocorrelation coefficients (Fortin et al., 2006). Both were used to allow comparisons between the methods, as inferring spatial patterns from geostatistical outputs can be difficult with the two methods having subtle differences (Kraan et al., 2009). Empirical variograms and Moran's *I* correlograms group the distance between each sample into distance bins (distance classes) and plot the average semivariance or Moran's *I* value respectively for that bin. This means bin size and sufficient replication are important for a robust analysis. A bin size of 6 metres was chosen as a suitable compromise of being able to observe small scale spatial structure (smaller bin sizes) and ensuring sufficient replication (larger bin sizes). Data for inter-sample distances > 60 metres were excluded as this was the smallest dimension of the sampling area and replication reduced significantly with greater distances (Fig. 3.3). Both designs used were observed to produce very noisy patterns at distances > 60 metres when included, further justifying this decision which is a standard procedure (Fortin et al., 2006; Legendre and Fortin, 1989). Experimental variograms are a plot of the average semivariance (y axis) against the sample distance bin (x axis). The formula for calculating the semivariance between two sample points is shown by equation 3.1 which is adapted from Fortin et al. (2006).

$$y_{(a,b)} = \frac{1}{2} (Z_{(a)} - Z_{(b)})^2 \quad (3.1)$$

Half the difference of density values (*Z*) between location a and b are squared to give the semi-variance (*y*) for that pair of samples _(a,b). This allows the plotting of how the variance between samples changes with distance (termed autocorrelation). Typically semivariance increases with distance because samples are further apart, but the distance at which semivariance no longer increase (often called the range) can show the size of patches which may be present. If semivariance does not change with distance it indicates a random distribution. Moran's *I* computes the deviation of values from the mean and produces a coefficient between 1 (positive autocorrelation) and -1 (negative autocorrelation) (Fortin et al., 2006). A value of 0 would indicate a random distribution. When this is plotted against distance bins it is called a Moran's *I* correlogram and like the empirical variogram it can indicate the spatial structure as values change with distance (Fortin et al., 2006; Kraan et al., 2009; Legendre and Fortin, 1989). At small distances Moran's *I* is often positive and

decreases with increasing distances, this is termed autocorrelation. The distance at which this stops decreasing can indicate the patch size and the distance between the first two peaks can indicate the distance between patches (Kraan et al., 2009; Legendre and Fortin, 1989). If all distance bin values plotted along or close to zero then this would indicate a random distribution. Moran's I assumes little variance in the mean spatially (second-order stationary) so all counts were $\log(x+1)$ transformed to reduce deviation from this assumption (Boldina and Beninger, 2013; Kraan et al., 2009). Krigged prediction maps of variables were created by fitting a spherical model to the relevant variogram using the same bin size and maximum distance for the empirical variograms.

Raw data from the laser particle analyser were analysed using GRADISTAT version 8 (Blott and Pye, 2001) to obtain the percent contribution of silt and mud and D_{50} (median particle size) values.

2.3.3. Correlation analysis

To understand how the measured variables such as post-larval and adult density may relate to each other across the sampling points and how this relationship changes temporally, the Pearson's product-moment correlation was calculated for various combinations of data. Variables measured are often autocorrelated and therefore not independent, and so a Dutilleul corrected P-value was calculated which accounts for autocorrelation in the data (Rosenberg and Anderson, 2011). The correlation coefficient will range across -1 giving a perfect negative correlation to 0 indicating no correlation, and 1 would show a perfect positive correlation. The P-value produced is the probability of achieving the associated correlation coefficient if the null hypothesis of no correlation is true. For any data collected in October 2011 and March 2012 the two additional points along each transect were removed to allow a direct comparison with the 7th June 2011 data. Deviations from the normal distribution in the various variable data sets were checked before running Pearson's product-moment correlation.

3. Results

3.1. Non spatial routine sampling

Post-larvae were found on the first sampling event in the study area on 3rd May 2011 ranging from 350 to 500 μm in shell length (Fig. 3.4) at a density of 1,865 m^{-2} (Fig. 3.5). The size frequency data (Fig. 3.4) shows that the main larval settlement occurred from early May to early June 2011. The peak density of post-larvae was found on the 21st of July with an average of just over 15,000 m^{-2} (Fig. 3.5). The density then decreased as detected on the 1st August. The rate of decline then reduced due to more newly settled post-larvae (200 – 600

µm) appearing regularly in the sample area until October (Fig 3.4, Fig. 3.5). By October 2011 the post-larvae had attained an average shell length of 7.2 ± 0.8 mm. After their first winter on 7th March 2012 the 2011 cohort mostly ranged from 10 mm to 13 mm in shell length (Fig. 3.4).

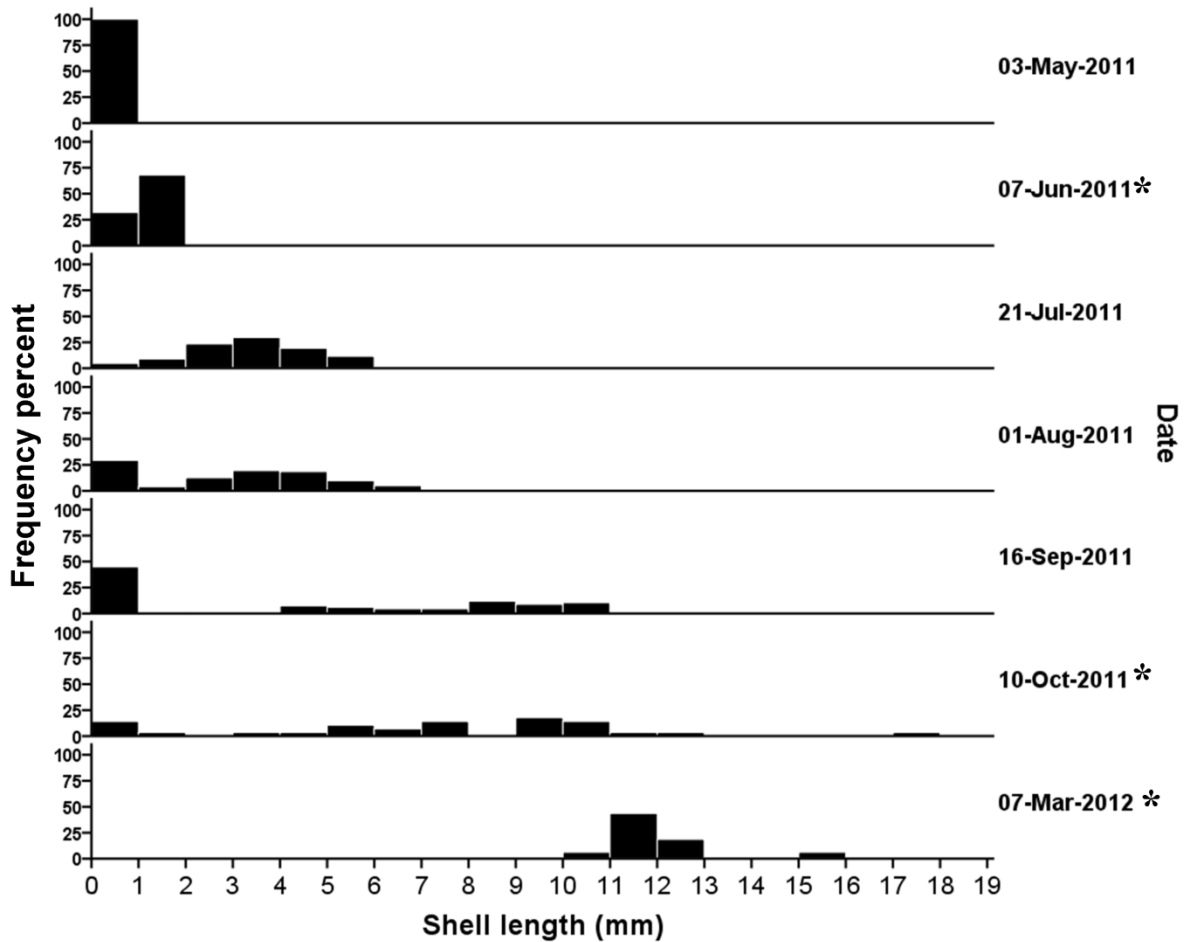


Figure 3.4. Percent frequency length histogram of *Cerastoderma edule* post-larvae collected at the location of the small scale sampling site from May 2011 to February 2012 on the Thurston cockle bed on the Dee estuary. Percentages calculated per sampling event and dates marked * indicate when the staggered nested design was used.

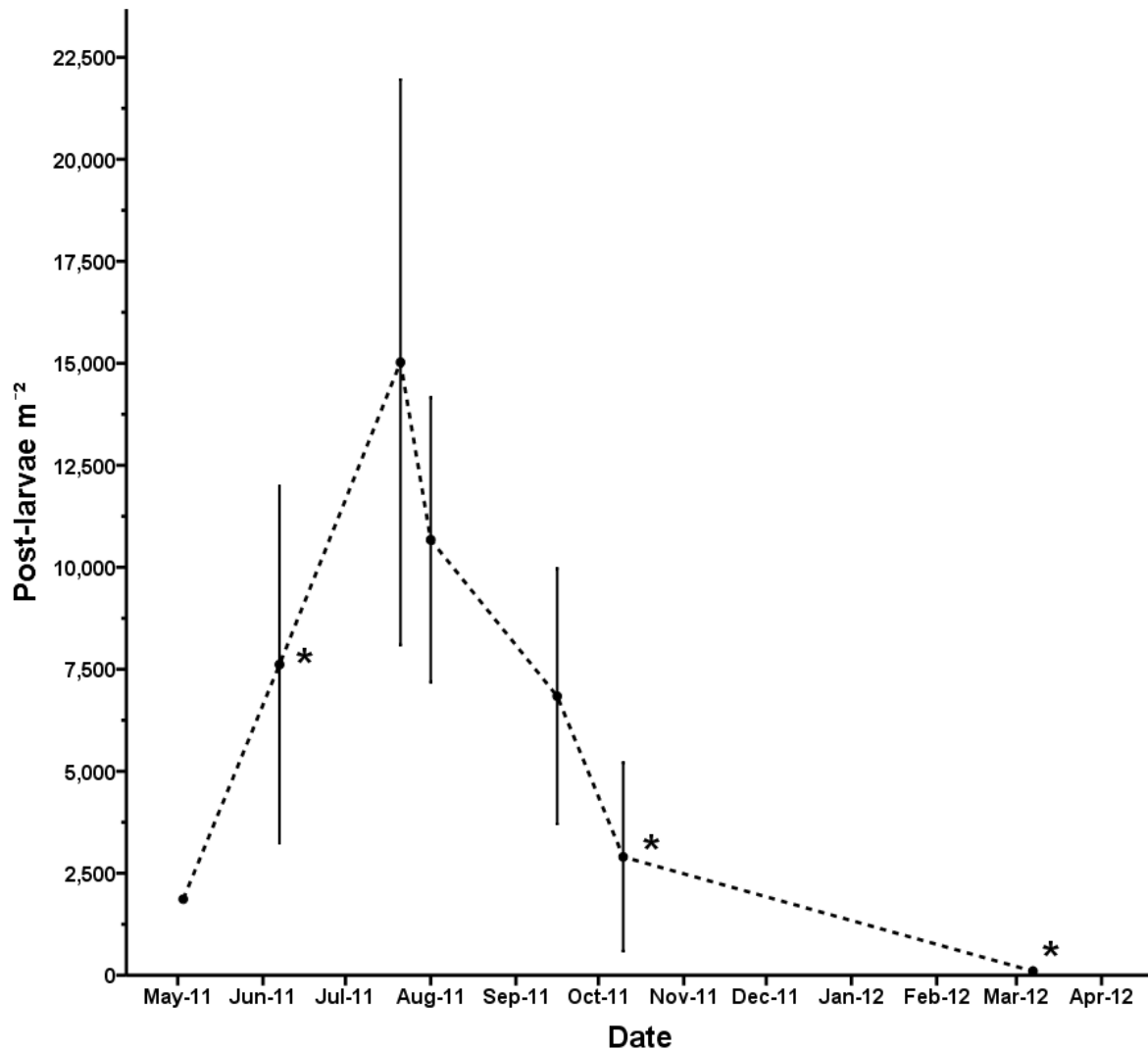


Figure 3.5. Density of post-larvae at the location of the small scale sampling site from May 2011 to February 2012 on the Thurston cockle bed in the Dee estuary. Error bars are ± 2 standard error and points marked * indicate when the staggered nested design was used.

3.2. Staggered nested design spatial sampling

3.2.1. *Cerastoderma edule* post-larvae

On the 7th June the newly settled post-larvae had a strongly patchy spatial distribution inside the 80 x 60 metre sampling area, as indicated by the clear autocorrelation at small spatial scales in the empirical variogram and correlogram (Fig. 3.6.A.). The empirical variogram had a lower semivariance between 0 - 6 metres which increased to the 6 – 12 metre bin, followed by other peaks and troughs in semivariance (Fig. 3.6.A.). This indicates several repeating patches of around 10 metres in size. The Moran's *I* correlogram also indicates several repeating patches of 12 - 18 metres in size seen as the fall in autocorrelation from the 0 – 6

metre size range (Moran's $I = 0.53$, $P = 0.01$) to the distance bin covering 12 - 18 metres, and a following peak at 18 - 24 metres indicating a possible inter-patch distance of 18 - 24 metres (Fig. 3.6.A.). There was also a significant autocorrelation peak at 48 – 54 metres further indicating multiple patches in the sampling area (Moran's $I = 0.27$, $P = < 0.01$) (Fig. 3.6.A.). The krigged prediction surface illustrates the patchy concentrations of post-larval density shown in the autocorrelation plots, but with no clear regularity or pattern evident (Fig. 3.6.A.). By the 10th October 2011 the spatial structure of the post-larvae had changed to become a more widespread random pattern, with some patchiness. The empirical variogram shows a spike in semivariance at the 12 – 18 metre scale with the overall pattern showing a random spatial structure (Fig. 3.6.B.). The Moran's I correlogram indicates some small scale autocorrelation at the 0 – 6 metre scale (Moran's $I = 0.24$, $P = 0.012$) but a random distribution dominates immediately after with little deviation from the zero line (Fig. 3.6.B.). The krigged surface supports this showing a more even distribution of densities across the sampling area than in June 2011, but with some density differences across the sample plot indicating a possible gradient in a south easterly to north westerly direction (Fig. 3.6.B.). Five months later in March 2012 the spatial structure changed again. The empirical variogram is very noisy and would suggest multiple small random patches (Fig. 3.6.C.). The Moran's I correlogram shows a largely random pattern with some positive autocorrelation between 0 – 6 m (Moran's $I = 0.26$, $P = < 0.01$) with negative autocorrelation at the largest scale of 54 – 60 metres (Moran's $I = -0.19$, $P = < 0.01$). The krigged prediction surface shows some small scale patches and a possible gradient in the same direction as in October 2011, but with a much more patchy structure (Fig. 3.6.C.).

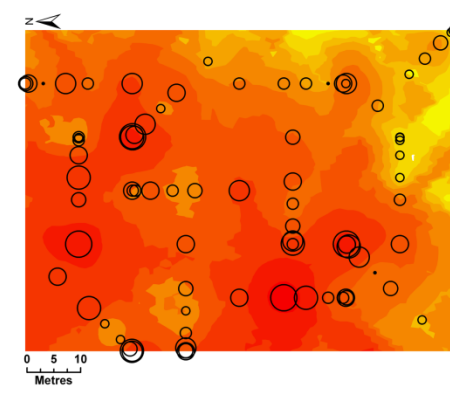
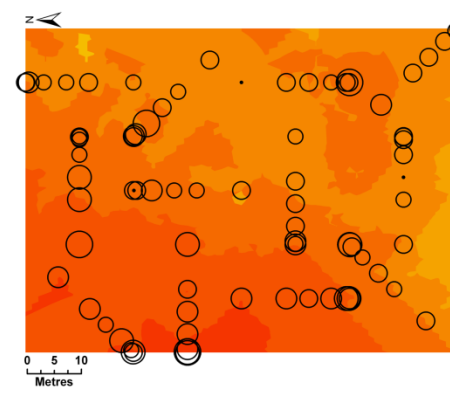
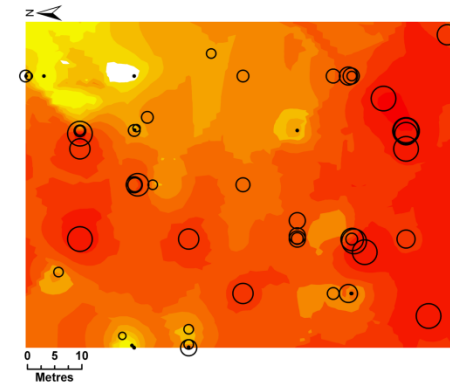
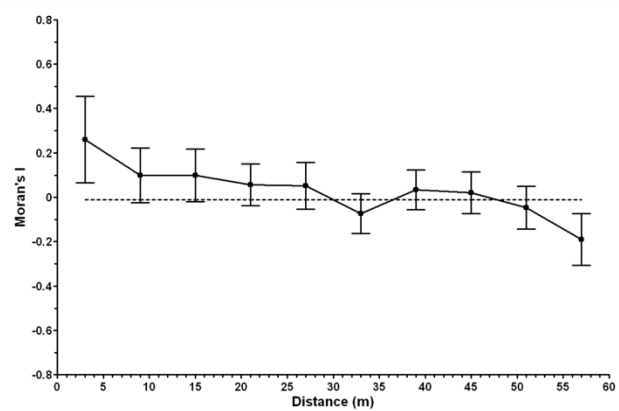
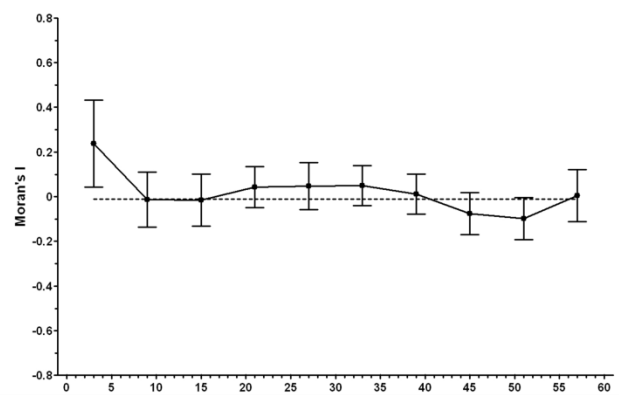
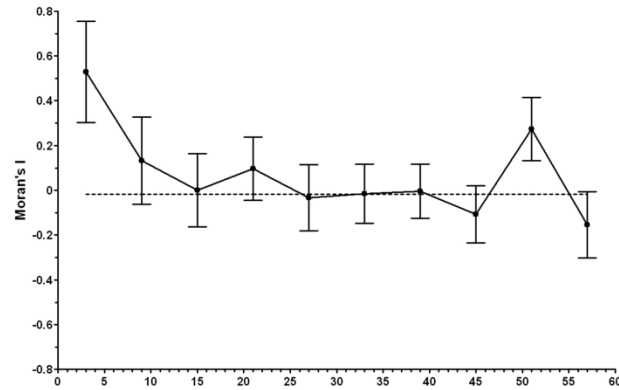
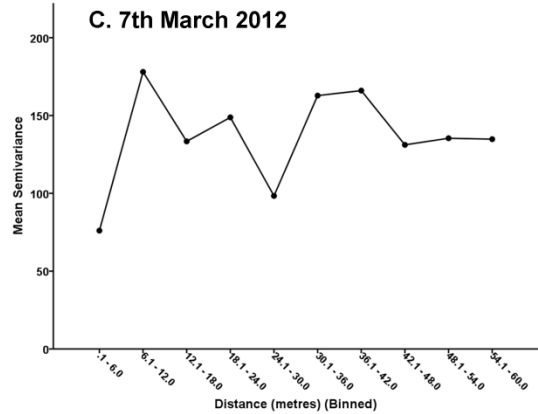
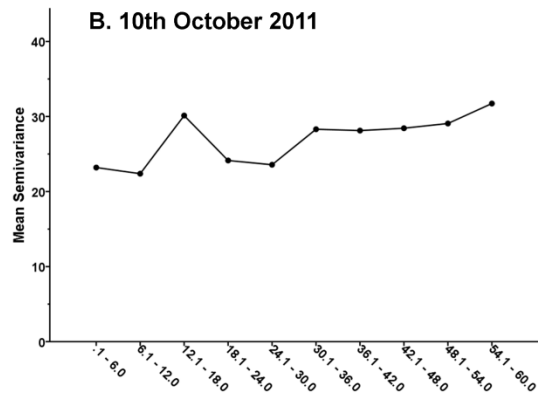
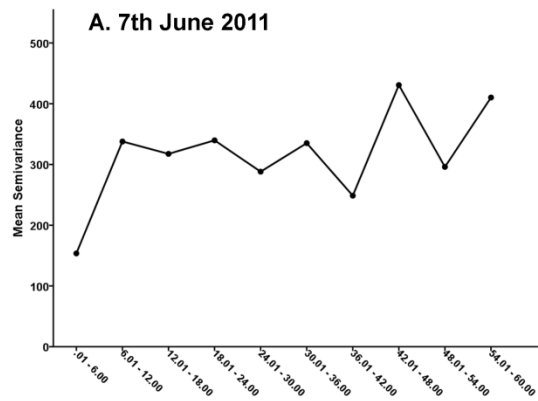


Figure 3.6. Spatial analysis of the 2011 *Cerastoderma edule* post-larvae cohort at the sampling area, Dee estuary UK. From left to right the empirical variogram, correlogram and krigged prediction surface are shown respectively for the A) post-larvae collected on 7th June 2011 B) on the 10th October (Shell length 2,500 – 14,000 μm) C) and on the 7th March 2012 (Shell length < 19 mm). A distance lag of 6m is used for the empirical variograms and correlograms. Bars in correlogram represent 95% CI and darker shades of red and larger circles in the Krigged surface indicate higher predicted densities in the krigged maps.

3.2.2. Adult *Cerastoderma edule*

Adult cockles were also sampled during collection of post-larvae so that a comparison could be made with the predicted final spatial structure that the post-larvae would attain, as the 2011 cohort could not be followed until maturity. The adult cockles sampled in October 2011 had a mean density \pm 95% CI of 39.83 ± 11.84 ranging from 0 to 170 m⁻², and showed weak random patchiness due to the small differences in autocorrelation between the peaks and troughs in both the empirical variogram and the Moran's *I* correlogram (Fig. 3.7.A.). The krigged prediction surface and the gradual decrease in autocorrelation in the correlogram indicate an underlying gradient in density, in a south easterly to north westerly direction (Fig. 3.7.A.). This is very similar to that seen in the October 2011 and March 2012 post-larvae krigged prediction maps (Fig. 3.6.B. and Fig. 3.6.C.).

3.2.3. Sediment

Analysis of the spatial structure of sediment properties showed fine sediment percent content (< 63 μ m) was strongly autocorrelated in the empirical variogram. This is shown by the increase in semivariance up to the 12 - 18 metre distance bin where it then levels off, which indicates patches of this size are present (Fig. 3.7.B.). The Moran's *I* correlogram shows decreasing autocorrelation to the 12 - 18 metre bin which has a negative autocorrelation value. This indicates that the main patches have a size of between 12 – 18 m and the peak at the 24 – 30 metre bin indicates patches are 24 - 30 metres apart. The fine sediment appears strongly patchy from the geostatistical analysis and supported by the krigged prediction surface (Fig. 3.7.B.). The D₅₀ (median particle size) shows a very different spatial structure to that of the fine sediment content, with little indication of significant patchiness by no clear increase in semivariance or fall in autocorrelation with distance in the empirical variogram and Moran's *I* correlogram respectively (Fig. 3.7.C.). This would indicate that D₅₀ of the sediment is randomly distributed in the sampling area.

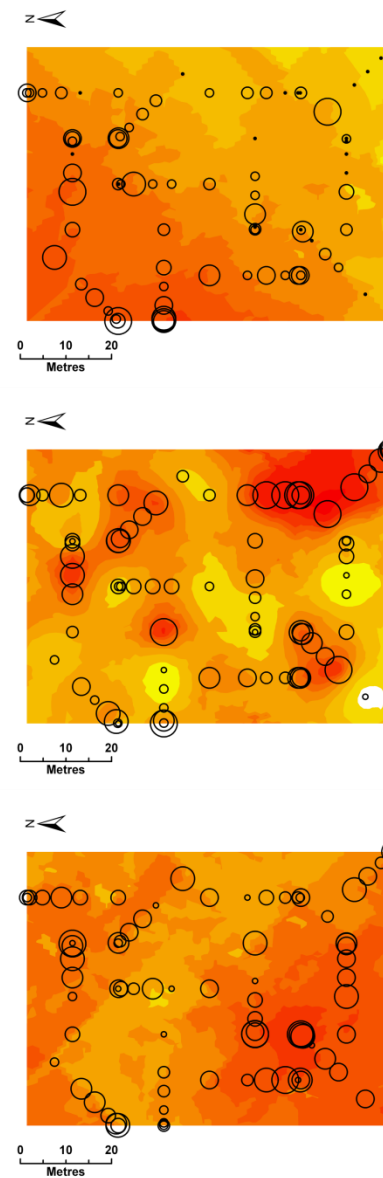
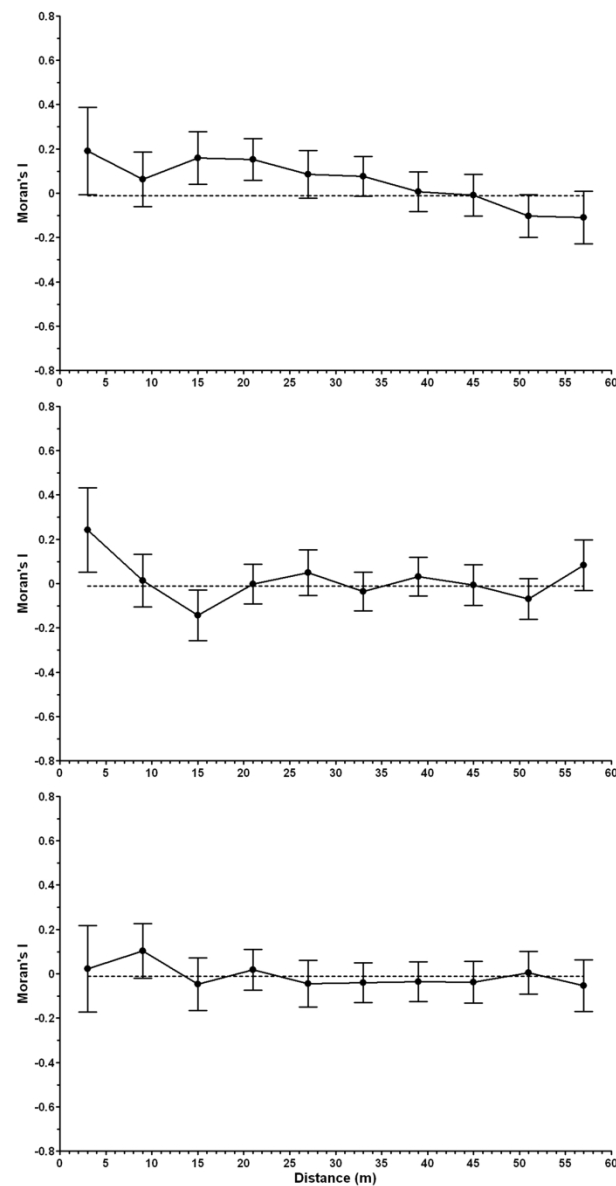
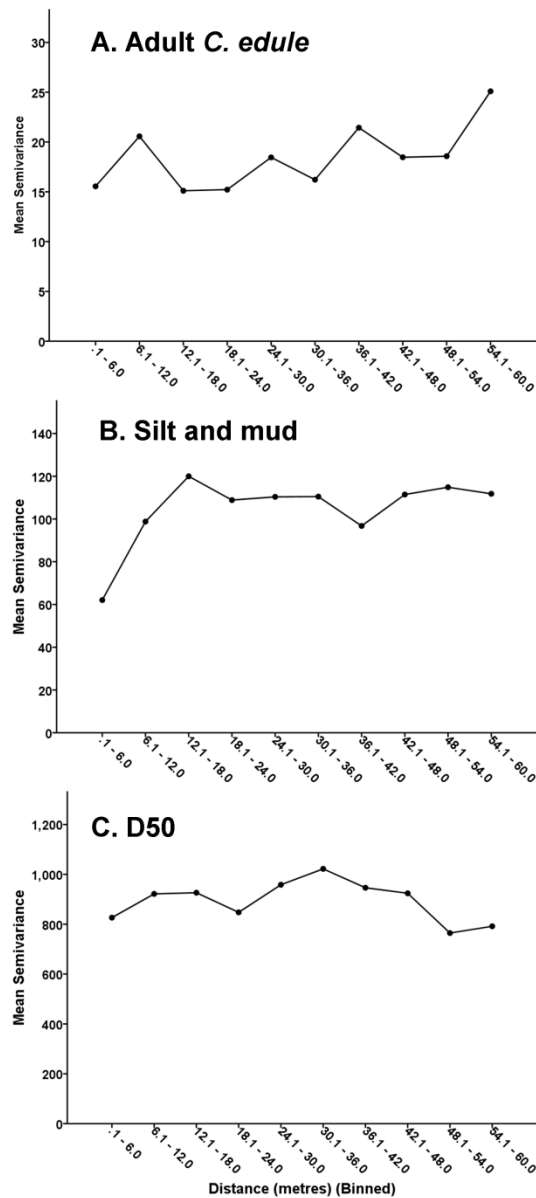


Figure 3.7. Empirical variogram, Moran's I correlogram and krigged prediction surface (from left to right respectively of A) adults *Cerastoderma edule* (shell length > 18 mm) B) fine sediment content (particles < 63 μ m) and C) D50 (median particle size) sampled using a staggered nested design on 10th October 2011 at the Thurston bed, Dee estuary. Bars in correlogram represent 95% CI and darker shades of red and larger circles in the Krigged surface indicate higher predicted densities in the krigged maps.

3.3 Correlations

By looking at correlations of variable values across the sampling area similarities and dissimilarities between the abundance changes over time for post-larvae can be assessed. Looking at correlations between different variables helps to indicate processes that may be operating and warrant further investigation.

3.3.1. *Post-larval density with adult C. edule density*

Post-larvae were predicted to have a negative correlation with adults at settlement and post-settlement due to larval ingestion and amensalism. There was no correlation of the post-larval density soon after settlement in June 2011 with that of the adults recorded in October 2011 (Fig. 3.8). However there was a significantly positive correlation of post-larval density in October 2011 and March 2012 with the October 2011 adult density (Fig. 3.8). When looking at the correlation between the June and October 2011 post-larvae with the March 2012 adult density there was no correlation. However, post-larval density in March 2012 had a positive correlation of 0.3 with the adults in March 2012, similar to that shown for the October 2011 adult data of 0.28 (Fig. 3.8).

3.3.2. *Post-larvae with post-larvae*

Post-larval densities in June 2011 correlated negatively with the densities in October 2011 and March 2012 but were non-significant. However the post-larval densities in October 2011 and March 2012 had a significant positive correlation with each other, indicating a similar density distribution across the study area (Fig. 3.8).

3.3.3. *Post-larval density and adult C. edule density with the sediment silt and mud content*

There were no significant correlations with sediment silt and mud content. However post-larval density in June 2011 had the highest positive correlation coefficient with silt and mud content from all the sampling events.

PL Jun		0.68	0.51	0.32	1.00	0.08
PL Oct	-0.05		0.01	0.04	0.84	0.51
PL March	-0.10	0.35		0.04	0.09	0.36
Ad Oct	-0.14	0.29	0.28		0.66	0.70
Ad March	0.00	-0.03	0.30	0.05		0.14
Sediment	0.22	0.10	0.14	-0.05	-0.20	
	PL Jun	PL Oct	PL March	Ad Oct	Ad March	Sediment

Figure 3.8. Correlation matrix of Pearson's coefficient (shaded grey) and associated Dutilleul corrected P-values (white boxes) between *Cerastoderma edule* post-larval (PL) density, adult (Ad) density and mud content (< 63 μ m) of the sediment across the staggered nested sampling design in the Dee estuary, UK. All sampling conducted in 2011 apart from the March data which was sampled in 2012. Significant results are indicated with a thick border.

4. Discussion

4.1. Patterns observed in post-larval density

The results of this study indicate that post-larval colonisation during the time of initial larval settlement is highly patchy at small scales, but then becomes more uniform with time post-settlement. These findings are in agreement with the original hypothesis. The results show a similar progression of spatial distribution on a small scale as seen in large scale studies. Soon after the initial larval settlement post-larvae were distinctly patchy with a similar structure to the fine sediment content, but no significant correlations were observed with adults or sediment. Over time the post-larval density became positively correlated with the adult density and obtained a similar spatial structure of a patchy gradient. This demonstrates that settlement patterns are very different in structure to the patterns of adults and of post-larvae observed later in the year, indicating the importance of post-settlement processes in spatially structuring populations of *C. edule*. Over small spatial scales it was observed that

initial settlement is spatially restricted to patches of very high post-larval densities. It further demonstrates the importance of post-settlement process which result in *C. edule* colonising more habitat and changing the spatial structure created at initial settlement. There is little evidence that the initial settlement of *C. edule* post-larvae effectively select for, or against, the silt content of sediment or the density of adult conspecifics. However there is evidence from this study (and later in Chapter 4) that *C. edule* post-larval density has a positive correlation with adult density post-settlement. It is important to acknowledge that there were some known, and likely to be many unknown, sources of small scale heterogeneity in the study area that were not quantified in this study. As with many tidal flats small scale heterogeneity in topography existed with some areas of standing water being visible. Other macrofauna may also have varying spatial structure at these small scales, such as the visible patches of mussels *Mytilus edulis* and other infaunal species such as *Macoma balthica*. It was not possible within the scope of this research, and is likely not possible, to collect all the potential variables that may influence *C. edule* post-larval density and is a recognised limitation of such observational studies. However the objective of this study, to record spatio-temporal changes in *C. edule* post-larval density, was met, and can inform future work to identify specific causative factors and their importance.

No study has looked at small scale changes in spatial structure of *C. edule* post-larval density over time before, so direct comparisons to previous studies cannot be made. Most studies, including Chapter 2, have examined large scale changes (100s – 1000s metres) in distributional patterns of *C. edule* post-larvae e.g. Armonies (1996) and Bouma et al. (2001). The only other study, apart from this one to the authors knowledge, on small scale (extent of a 35 x 35 metre grid, 5 metre sample lag) post-larval distribution of *C. edule* is that of Huxham & Richards (2003). This work, based on a single sampling event in a Scottish estuary in July 1998, suggested post-larval redistribution after settlement. Results from the variance / mean ratio indicated aggregation, but the Moran's *I* correlogram is reported as being 'non-significant', which I interpret as indicating a random structure (the correlogram is not shown). Huxham and Richards (2003) concluded from the pattern observed, along with an *in-situ* sediment selection experiment, that this early colonisation was likely governed by hydrodynamics and not behavioural selection of sediment properties. On a larger scale (extent ~1000 x 600 metre grid, sample lag of 100 metres) Armonies (1996) observed a spatially confined patchy post-larval distribution in the early summer close to settlement, which was associated with fine sediment. During the summer post-larval density become more uniform across the bay as they regularly redistributed, with the author also concluding that hydrodynamics determine the patchy initial settlement and that post-settlement redistribution was reducing the aggregation. In another large scale study (extent 700 x 800 metre grid, 100 metre sample lag) by Bouma et al. (2001) the conclusion was also

that initial settlement was governed by hydrodynamics as small post-larvae were found in the least dynamic areas with the highest content of fine sand grains, and that post-settlement redistribution was an important process for the population there. Chapter 2 showed similar changes in spatial distribution of *C. edule* post-larvae at a large scale as reported in the above studies. The emerging trend is that on large spatial scales the initial distribution of post-larvae is different to that after a post-settlement period. This pattern is concluded to have occurred from passive deposition of larvae with post-settlement redistributions.

Understanding the spatio-temporal dynamics of post-larval density can give important insights into causal processes during this important period for *C. edule* populations. Although conclusions made should acknowledge the inherent uncertainties that accompany them (Butman, 1987). In our small scale study the results indicate that the early post-larval spatial structure is highly patchy, and that post-larvae redistribute over small spatial scales. The initial patchiness observed in the spatial structure of post-larval density was similar to that of the silt content recorded in October, indicating that a similar process or scale of process are operating on both. It must be acknowledged that the sediment properties were measured in the October sampling and not in June when initial settlement of *C. edule* took place. There is reasonable confidence that the spatial structure of these sediment properties is unlikely to have changed (even if the location of the patches does); Cesar and Frid (2009) present data from the same location that shows little change (less than 5%) in the sediment silt content between June and October. From our observational data it is not possible to disentangle passive settlement and active habitat selection at larval settlement which could possibly create similar patterns. However it seems likely, given the well documented behavioural strategy of byssus redistribution (Armonies, 1992; 1994b) that post-larvae do not or attempt to, or cannot, remain in dense patches produced at settlement. Thus active selection for areas containing higher silt and mud content sediment at settlement, or any other abiotic or biotic variable, seems unlikely (Armonies and Hellwig-Armonies, 1992) given that they rapidly undergo dangerous migrations post-settlement (Hiddink et al., 2002a). Byssus drifting does appear to be a behavioural strategy to colonise new habitats post-settlement, and may enable post-larvae to escape the spatially restricted and numerically intense passive initial settlement (Armonies, 1996; Armonies and Hellwig-Armonies, 1992; Armonies and Reise, 2003; Hunt et al., 2003).

Predation is currently considered the main cause of high post-larval mortality in *C. edule* (Beukema and Dekker, 2005; Jensen and Jensen, 1985). Later in chapter 5 I demonstrate how *C. edule* post-larvae are likely to be more vulnerable to epibenthic predation from juvenile *Carcinus maenas* when present in dense patches, regardless of intraspecific interference competition among predators. A strategy for *C. edule* post-larvae to

change their spatial structure from that of dense patches to a more dispersed distribution may therefore reduce mortality from predation. Densities of post-larvae in these patches in the Dee estuary were so high ($> 10,000$ post-larvae m^{-2}) that if post-larvae remained in them intraspecific competition for space and food would likely occur (Jensen, 1993), giving another possible advantage of actively migrating away from dense aggregations.

The small scale spatial structure of post-larval density from October 2011 to March 2012 maintained a gradient, but became patchier within that gradient distribution according to the geostatistical analysis. The post-larvae collected during these later sampling dates were positively correlated with the adult *C. edule* density. This may indicate that the small scale spatial structure of adults is established within the first five months after initial settlement, but processes during the winter such as current scouring and freezing may create some small further scale patchiness. When observing the small scale spatial structure at a fishery impacted site of 1-group and 2-group cockles Boldina and Beninger (2013) concluded that spatial structure was maintained over time, demonstrating that post-larval distribution is dynamic compared to later life stages they examined. Juvenile *C. maenas* and *C. crangon* numbers in the intertidal fall during the autumn and are very low from winter until spring (Beukema, 1992; Hiddink et al., 2002c; van der Veer et al., 1998) with 0-group cockles likely to have reached a size refuge from predation by then. Larger *C. maenas* almost stop migrating into the intertidal at high tide in winter (Naylor, 1962) and the cockles are likely to remain too small for predation by overwintering wading birds (Sanchez-Salazar et al., 1987), and so epibenthic predation is an unlikely candidate for creating patchiness on a small scale during the winter period.

Some studies have suggested that adult cockles may depress settlement through larval ingestion on small spatial scales (Andre and Rosenberg, 1991; Andre et al., 1993; Lindegarth et al., 1991). This could be proposed as the reason for the difference in spatial structure between settled post-larvae and adults. The subsequent convergence of spatial structure and positive correlation with adults may be created when a refuge is reached from adult ingestion. If this process created the initial post-larval spatial structure it would be expected that a similar spatial structure (e.g. both weakly patchy like the adults but in opposite locations and with a gradient distribution, but in the opposite direction) and a strong negative correlation between post-larval density and adults in June 2011 would be observed when they are most vulnerable to ingestion by adults. However, very different spatial structure and no correlation between newly settled post-larvae and adults was observed. From flume and modelling investigations the effect of larval ingestion by adults appears to operate over very small spatial scales of cm around the inhalant siphons which should create a strong negative correlation, but it is unclear how this process would scale up spatially from these studies (Andre et al., 1993; Lindegarth et al., 2002). For these reasons it

is concluded that adult ingestion of larvae seems unlikely as a process creating the initial post-larval spatial patterns observed. In a study investigating patchy recruitment in *Spisula ovalis* it was noted that using correlation estimates that corrected for autocorrelation (as used in this study) was important for correctly interpreting negative adult-recruit interactions (Patric et al., 1997). In the same study they concluded that 0-group spatial patterns were independent of adult densities, but are formed by a similar process (Patric et al., 1997). This demonstrates how correlations between adults and earlier life stages should be interpreted with caution. Notable from the results in this study was the significant positive correlation and similar spatial structure of post-larval density with adult density in October 2011 and March 2012, with an underlying gradient in the same direction. This could suggest a positive association with adults post-settlement. Several hypotheses could be proposed to explain a positive association with adult density including 1) post-larvae actively associating themselves with adults during byssus migrations (by rejecting areas with a low density or absence of adults) 2) presence of adults increasing survival of post-larvae thereby producing an indirect positive correlation between the two 3) the small scale distribution of adults and post-larvae are formed under the same process (active or passive). Separating points 1 and 2 is not possible from my data, even though point 1 is based on active behaviour and point 2 is passively derived from survival. Both would depend on an advantage of being associated with adults. Adult-juvenile interactions can vary with sediment and habitat type for any given species (Thrush et al., 1996). A study in New Zealand by Legendre et al. (1997) where they examined the spatial structure of a venerid bivalve (*Austrovenus stutchburyi*), with a similar life history and ecology to that of *C. edule*, observed, on the scale of their study (250 x 3,500 m area), no juvenile-adult interactions whereas other studies on the same species did. These examples highlight the difficulty in using correlative observational data. For example a negative correlation between adults and post-larval density cannot be extrapolated to being caused by an adult-post-larval negative interaction as other covariates or indirect processes may be responsible. Experimental studies in the field and laboratory are required to address such hypotheses (with issues of scale and naturalness considered and acknowledged in their interpretation) to find the cause of observed patterns. No evidence for negative adult-larval or adult-post-larval interactions during initial settlement was found from the data, in fact the opposite may be likely with a positive 'interaction' post-settlement. This is investigated and discussed further for *C. edule* in the next chapter - Chapter 4.

4.2. Wider implications

The results obtained have potential implications for the ecology, management and monitoring of cockle populations. Close to settlement intense spatial variability (patchiness)

on a small scale was observed. In June post-larval density varied from zero to over 70 per core (zero – several thousand m^{-2}) within distances of 10 metres. This calls into question the reliability of point sampling for *C. edule* post-larvae over large spatial scales (sample lags of > 100 metres). To accurately sample post-larval densities on a large scale the replication at each point sample station would need to be separated by a distance greater than the inter-patch distance, which was calculated at between 18 - 24 metres based on our results. Within station replication is often performed but at very small spatial scales, often no more than a few metres from each other. Replicates taken close to each other may also not be independent due to autocorrelation at small spatial scales, and will not provide a good estimate of the local mean density. It is recommended that future large scale studies ideally perform a spatially explicit study like this to estimate the potential scales of patchiness (as recommended by other studies). Even without a pilot study to quantify potential patchiness, it will be beneficial to ensure that replication within-stations is not spatially aggregated. The spatial arrangement of within station replicates may be just as, if not more, important as the number of replicates for accurate estimates of the local density. This will reduce the chance of replicating only within or between patches, which may considerably over or under estimate the estimated densities at stations respectively, and thereby produce false distribution patterns, abundances and biomass estimates.

4.3. Conclusions

Density and spatial structure changed dramatically from the settlement period in June through to October, progressing from highly patchy to a more uniform distribution respectively. This small spatial scale study further supports post-settlement processes alter post-larval distribution and density (modified either through mortality or redistribution). This study demonstrates that the small scale (metres to tens of metres) spatial structure of *C. edule* post-larval density changes significantly during the most critical stage of benthic colonisation, with potentially significant implications for post-larval survival and intraspecific competition. It has also been demonstrated that on a small scale, spatial structure may be determined within the first year of benthic life, and so processes acting on small scales may be highly important for an entire population's structure and stability. I found no evidence of negative interactions with adult *C. edule* at settlement or post-settlement. This study along with the results and conclusions later in Chapter 4 will show that conspecific interactions may be positive at small spatial scales in the Dee estuary, which is contrary to what has been demonstrated and suggested for other areas. It is suggested that processes occurring during this early post-settlement period are critical in determining a species distribution and abundance within soft sediment tidal systems (Armonies and Reise, 2003). Therefore further

understanding of the post-settlement distributional changes and what proportions are active to passive, and what initiates active migrations are key areas for research. It is this process which structures *C. edule* populations over small and large scales. The effect of spatial structure on predation and conspecific interactions during the post-settlement period are likely to be highly relevant to understanding why recruitment has such high spatial variability. Understanding post-settlement processes at various spatial and temporal scales will give further insights into why species like *C. edule* do not appear to fully colonise what is currently understood as all the available suitable habitat at large spatial scales.

Acknowledgements

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Chapter 4: The effect of macrofaunal disturbance on *Cerastoderma edule* post-larval density.

Timothy Andrew Whitton, Stuart Rees Jenkins, Chris Allan Richardson, Jan Geert Hiddink

Abstract

Common European cockle (*Cerastoderma edule*) populations often have highly patchy distributions, which is likely to be determined during the settlement and post-settlement period. High densities of filter feeders and/or bioturbators are thought to reduce the success of larval settlement and the level of post-settlement survival. To further our understanding of how macrofauna influence the density and distribution of cockle post larvae, I tested the hypothesis that macrofauna decrease post-larval density due to ingestion and bioturbation. Post-larval densities were compared among treatments in which lugworms and adult cockles were excluded in field experiments in two locations (the Dee estuary and the Menai Strait respectively) from the time of initial larval settlement to late summer in 2012. Lugworm exclusion plots had significantly higher cockle post-larval densities than the control plots. Excluding adult cockles produced the opposite effect, the density *C. edule* post-larvae was lower in the adult cockle exclusion plots. The causal processes creating these differences in post-larval density are largely unconfirmed. In laboratory experiments we tested the hypotheses that lugworms and adult cockles would cause mortality and increase byssus drifting of cockle post-larvae when compared to macrofauna free controls. We found no evidence in support of these hypotheses from our results but discuss that they warrant further investigation. We conclude that the presence of adult cockles and lugworms has a positive and negative influence on post-settlement density of *C. edule* post-larvae respectively. However the causal processes remain unproven. We conclude the density, distribution and population dynamics of macrofauna may have significant implications on the success and spatial distribution of *C. edule* post-larval density and the resulting recruitment.

1. Introduction

Identifying the processes determining the success and spatial distribution of recruitment in populations is important for our understanding of ecosystems (Sutherland et al., 2013). Identification of the key processes driving recruitment in intertidal soft sediment systems is challenging, due to the highly mobile early life stages of many species and the importance of biological interactions in structuring these communities. The common European cockle *Cerastoderma edule* is an important species on North East Atlantic soft-sediment tidal flats, both commercially (Hicken, 2008) and ecologically (Flach, 1996). The cohort strength and distribution of adults is largely determined during the post-settlement stage, which is considered as the period from larval settlement through to survival of their first winter (Beukema and Dekker, 2005). The occurrence of high post-settlement mortality of *C. edule* due to predation (Jensen and Jensen, 1985) and their ability to migrate during this post-settlement period, is understood to determine adult densities and in addition is highly likely to determine spatial patterns (Armonies, 1994b). Previous studies on soft sediment systems have shown that biological interactions with other fauna play an important role in distribution and survival of infaunal macrofauna (Reise, 2002; Rhoads and Young, 1970a). These interactions can be direct trophic interactions, such as predation, or indirect through habitat engineering (Reise, 2002). For example habitat engineering through sediment surface disturbance by deposit feeders, burrowers and bioturbators may prevent larvae and post-larvae colonising affected areas, and thereby create discrete patches in abundance in some species (Woodin, 1976). The importance of such interactions in soft sediments, and the importance of the post-larval stage in *C. edule*, indicate that such processes could create spatial patterns in the adult population.

The adults of the lugworm *Arenicola marina* (a deep burrowing polychaete) and *C. edule* often make major contributions to the macrofaunal biomass in north western European intertidal flats (Beukema, 1976; Beukema et al., 1993). They are both considered bioturbators because they modify the physical and biological characteristics of sediment, through the physical disturbance of their surroundings (Flach, 1996; Flach and de Bruin, 1994). This bioturbation occurs when lugworms remove the surface sediment at their feeding depression and deposit onto the sediment surface when producing faecal casts. When lugworms are excluded from an area, the biological and physical habitat is likely to change. Abundances in microbes can rise (Lei et al., 2010), meiofauna have been shown to decrease (Reise, 1985a), and some macrofauna species (Flach, 1992; Volkenborn et al., 2009) and plants (Van Wesenbeeck et al., 2007) that depend on more stable sediment appear or increase in density. These biological responses are often due to the loss of the feeding burrows, oxygen penetration and sediment reworking (Reise, 1985a; Wendelboe et

al., 2013). Although less of an active bioturbator *C. edule* also disturbs the surrounding sediment when closing its valves and when making horizontal and vertical (Flach, 1996). The presence of these two macrofaunal species has been shown to affect the physical and biological environment in both field observations and experiments. Lugworms have been shown to reduce densities of post-larval *C. edule* when lugworms have been excluded from sediment (Flach, 1992; 2003). A similar effect on post-larval density by excluding adult *C. edule* has also been observed (Flach, 1996; Flach, 2003), but with more variable interaction outcomes within studies. Some of the observations in these studies show no effect or even an increase (De Montaudouin and Bachelet, 1996; Van Colen et al., 2013), have temporally variable outcomes (Andre and Rosenberg, 1991) and show little evidence of a linear relationship of adult cockle density on post-larval density. In some of these studies it was found that excluding adult cockles had nil or positive effects on post-larval density at the time of settlement (Flach, 1996; Flach, 2003; Van Colen et al., 2013). This interaction between macrofauna and *C. edule* post-larvae may be operating directly or indirectly, and has received limited experimental investigation to identify the causative processes. Direct interactions include post-larvae being ingested by macrofauna, or killed by sediment smothering from the macrofauna's bioturbation activities and increasing byssal migration rates. An indirect effect is not directly linked to the macrofauna. For example the mortality of the mud shrimp *Corophium volutator* increased in the presence of lugworms and adult *C. edule* due to increased swimming activity from the macrofauna disturbance, which in turn increased the predation risk from *Crangon crangon* (Flach and de Bruin, 1994). There is some evidence that an additive negative effect of disturbance from macrofauna and predation may also exist for post-larval bivalves (Flach, 2003).

The overarching question of this study was what effect macrofauna are having on *C. edule* post-larval densities, and how this contributes towards the observed spatial patchiness of *C. edule* populations. To address this question three experiments were conducted, one in the field and two based in the laboratory. The field experiment was conducted to test the effect of macrofauna on natural settlement and post-settlement densities, with the two laboratory studies seeking to investigate the causative processes of the patterns observed. In the field the hypothesis that macrofaunal presence decreases post-larval density compared to macrofaunal free areas and this difference will increase over time, was tested. We predict this as a consequence of disturbance from bioturbating *A. marina* and adult *C. edule* causing mortality or increasing migrations of post-larval *C. edule*. We predict the magnitude of effect to increase over time due to the increased chances of interactions with the macrofauna post-settlement. To test this, field experiments were conducted with *A. marina* and adult *C. edule* excluded from plots independently. To understand the potential causative processes two laboratory experiments were conducted. The first was an aquarium

based study to test the hypothesis that the presence of *A. marina* and adult *C. edule* will directly affect the survival and growth of *C. edule* post-larvae negatively. By confining post-larvae with *A. marina* and adult *C. edule* the direct effect of these macrofauna on the survival and growth of *C. edule* post-larvae could be evaluated. As part of this experiment I tested the hypothesis that survival and growth will be enhanced by provision of a spatial refuge from macrofauna. Finally I hypothesised that the presence of macrofauna increases the number of *C. edule* post-larvae byssus drifting compared to macrofauna free sediment. Byssus drifting has a high potential to create spatial patterns if certain conditions trigger migrations, such as the presence or disturbance of macrofauna.

2. Methods

2.1 Field experiment general approach

To observe the possible effect of lugworms and adult *C. edule* on colonisation of *C. edule* post-larvae, field experiments were set up at Traeth Melynog on the Isle of Anglesey in North Wales (53°08'22 N, 04°19'47 W) (Fig. 4.1.) and the Dee estuary on the English-Welsh border (53°20'52 N, 0310'38 W) respectively. In each experiment three treatments were randomly allocated amongst 18 plots (3 x 6 grid of 1 m² plots with a 10 metre separation between each plot). The experimental treatments were 1) a control with no disturbance to measure natural densities of post-larvae 2) an exclusion treatment where either lugworms or adult cockles were removed and 3) a procedural control to account for the disturbance of removing the macrofauna. The corners of each plot were marked with a bamboo stake that extended less than 10 cm above the sediment to minimise drag and the chance of algae collecting on the markers. The plots were then sampled from the detection of larval settlement onwards to understand how densities may change temporally among the treatments. To sample the plots for *C. edule* post-larvae a 64 mm diameter corer (201.06 mm²) was used to a depth of 15 mm. Randomly allocated replicate cores were taken from each plot and the number of post-larvae pooled.

2.1.1 Lugworm exclusion experiment

The experiment was conducted at the mid to low shore of Traeth Melynog on Anglesey (Fig. 4.1). This location was chosen due to high numbers of lugworms which have a visible effect on the sediment surface topography. From the work in chapter 2, I also knew where *C. edule* post-larvae were likely to colonise and so I could choose a suitable location with high densities of lugworms and a high probability of post-larval presence during the experiment. Lugworms were excluded by burying a 1 m² square of plastic mesh (2 mm mesh) at 10 cm

depth in the sediment. This method has been successfully used in other studies to exclude lugworms from an area (Volkenborn and Reise, 2006; Volkenborn and Reise, 2007; Volkenborn et al., 2009) preventing them from maintaining and creating their burrows, which are often 10 – 20 cm deep (Fig. 4.2). The procedural control plots were created by digging the sediment to a depth of 10 cm in the same way as the exclusion plots, but without any mesh being placed before the sediment was returned. The experiment to exclude lugworms was set up on 18th April 2012. The plots were established at this date to allow enough time (29 days) for recovery from disturbance. The plots were sampled on five occasions on 16th May, 18th June, 20th July, 26th July and the 23rd of August 2012. On the first three sampling events three cores were taken per plot. However post-larval densities were low on the 20th July and so eight cores were taken per plot for the two following sampling events to ensure more precise density estimates. On the 20th of July the numbers of lugworms present in each plot were sampled using three 0.1 m² quadrats per plot, by counting the presence of the feeding depression and faecal cast produced at the sediment surface by each worm. A particular issue at this location was the large amount of drifting algae on the sand flat. The bamboo plot markers encouraged the build-up of algae and so any algal accumulations were removed at each sampling occasion.

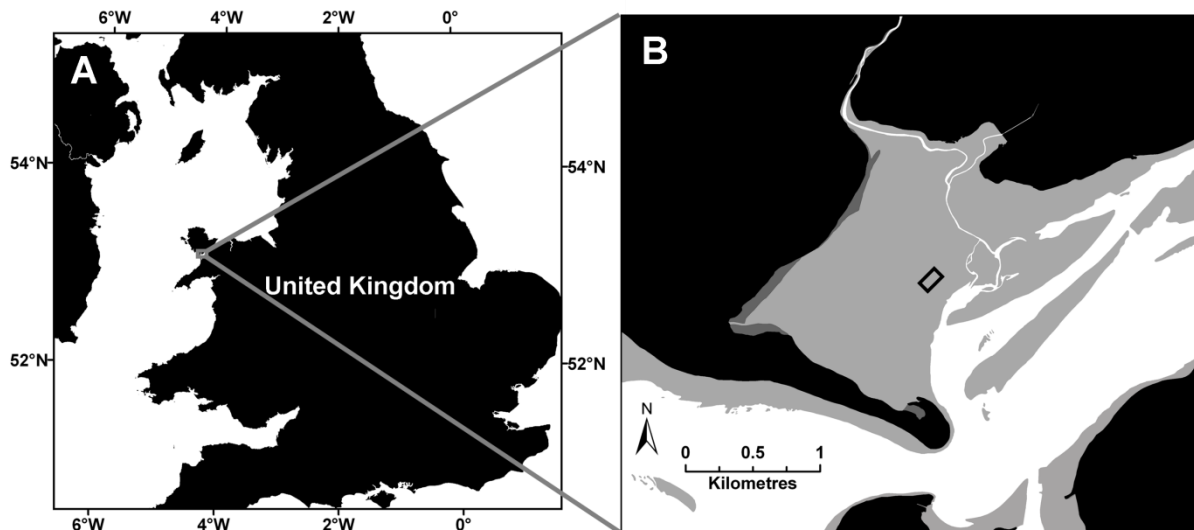


Figure 4.7. Location of *Arenicola marina* exclusion experiment at Traeth Melynog, Anglesey UK (A). The plots were located on the mid to low shore (black rectangle) with small fresh water input (Afon Braint) runs down the east side of the tidal flat which is situated at the southern end of the Menai strait (B). Grey areas show intertidal sediment, dark grey represents saltmarsh and black is land above MHWS.

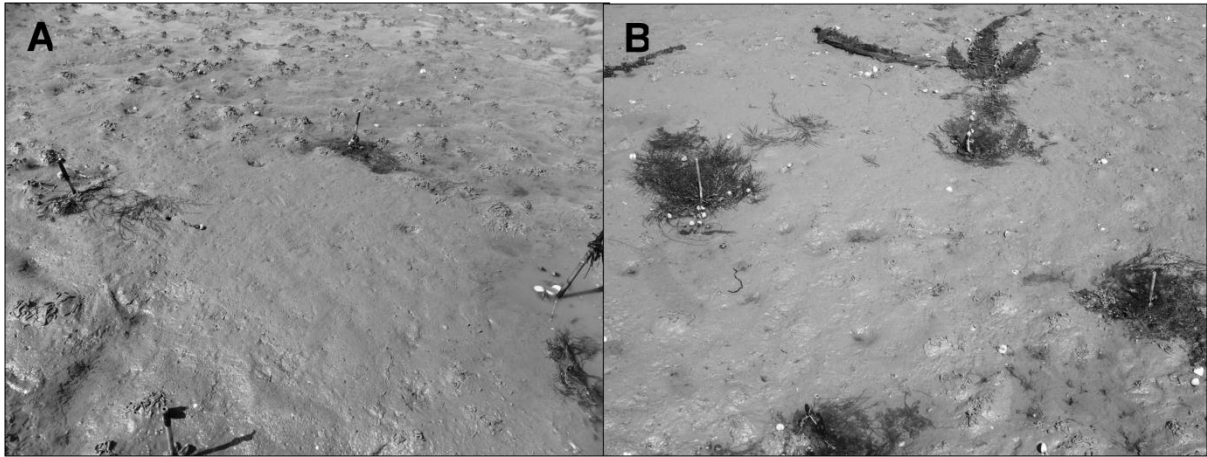


Figure 4.8. Examples of *Arenicola marina* experiment plots at Traeth Melynog, Anglesey UK. Where lugworms were excluded from the 1 m² plots the sediment surface becomes more homogeneous (A), compared to control plots where lugworm feeding depressions and faecal casts remained a dominant feature (B). Macroalgae can be seen accumulated on the bamboo stakes marking the plot corners.

2.1.2 Adult *C. edule* exclusion experiment

The experiment to exclude adult *C. edule* was conducted at mid to low shore level of Caldy Blacks, an area of intertidal mud-flat with good densities of adult *C. edule* at the Dee estuary UK (Fig. 4.3). Adult *C. edule* were removed by raking sediment from the exclusion plots into a mesh bag which allowed the sediment to pass through but retained adult cockles. The procedural controls were raked in exactly the same way but the mesh bag was omitted to allow adult *C. edule* to remain in the sediment. Control plots were left undisturbed (Fig. 4.4). The treatments were established on 9th May 2012 which allowed nine days for the plots to recover from the initial disturbance to when the first sampling took place on May 17th. The Dee estuary plots were sampled on three occasions using three pooled cores per plot on the 17th May, 19th June and the 2nd of July 2012. Sampling was not conducted past the 2nd of July because this marked the beginning of the cockle fishing season in the Dee estuary when, despite the cooperation of the cockle fishermen, the chance of disturbance of experimental plots was high. On the last sampling occasion a 0.1 m² quadrat was dug up to a depth of 5 cm in the centre of each plot and sieved over a 2 mm mesh to record the densities of adult *C. edule*. This was done to assess if the exclusion treatment plots had been successfully created. The patchy distribution of standing water pools and microphytobenthos amongst the plots was very variable. Due to the random distribution of the treatments this should not have biased any treatment results, but may increase variability in *C. edule* post-larval densities.

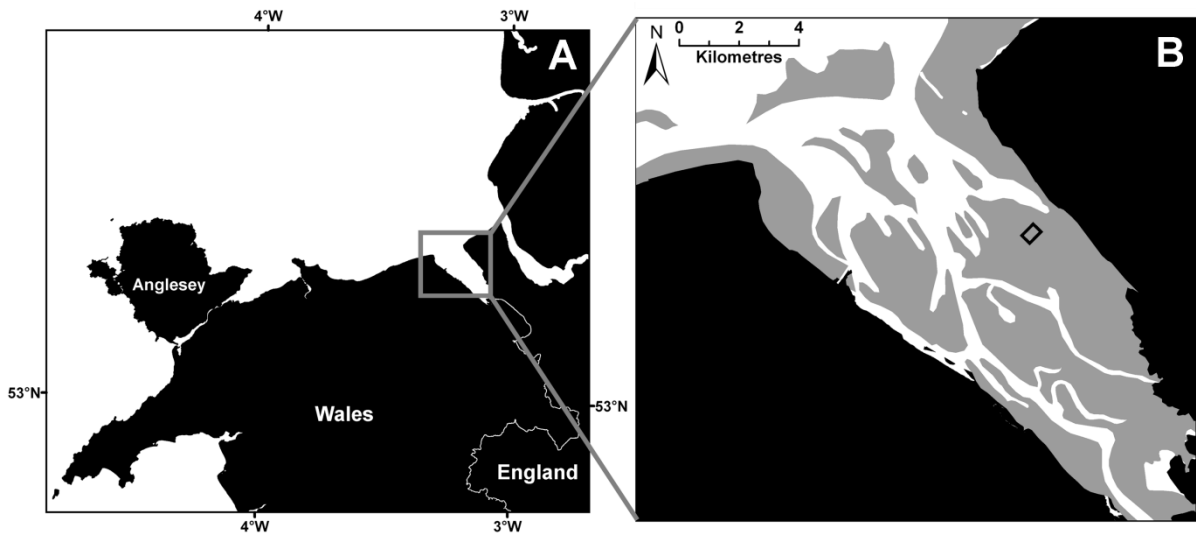


Figure 4.9. Location of the adult *Cerastoderma edule* exclusion experiment in the Dee estuary, UK (A). The area of the plot experiment (black rectangle) was located on the Thursaton cockle bed. The Dee estuary is macrotidal with intertidal areas shown in grey and land above MHWS in black.

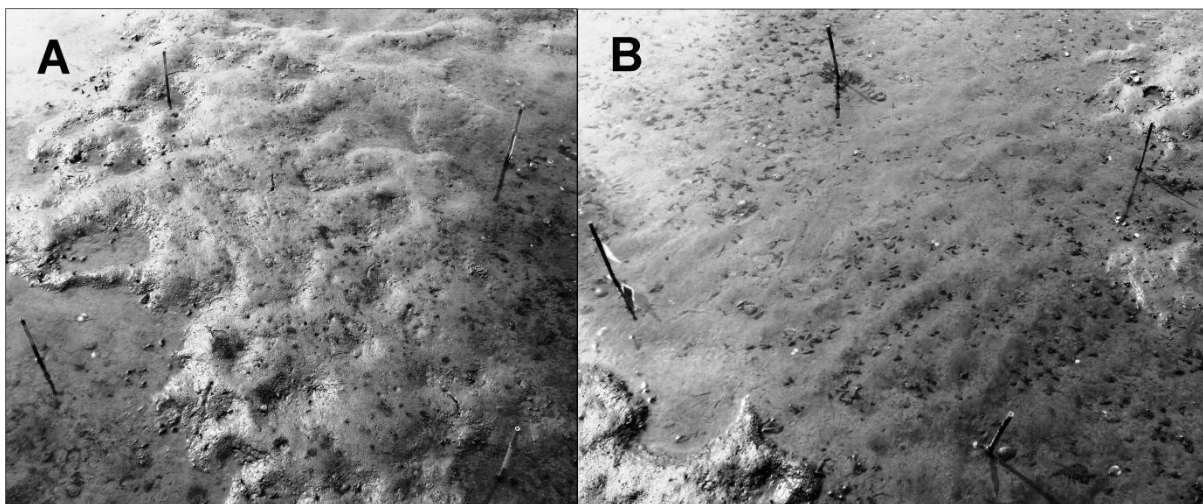


Figure 4.10. An example of control (A) and adult cockle *Cerastoderma edule* exclusion (B) plots from the field manipulation experiment in the Dee estuary, UK. Patchiness of standing water, sand ripples and microphytobenthos was observed across the experimental area with no difference in surface topography between treatments observed.

2.2. Laboratory study general approach

Two laboratory based studies were conducted to identify processes which may be driving the effects of adult cockles and lugworms on post-larval *C. edule*. An aquarium experiment was used to test the hypothesis that the presence of adult *C. edule* and lugworms has a direct negative effect on the survival and growth of *C. edule* post-larvae. An additional part of this aquarium study was to test the hypothesis that survival and growth will be higher when a spatial refuge from macrofauna is provided. The second experiment used a flume to test if the presence of adult *C. edule* or lugworms would increase rates if byssus drifting as a

possible means of migrating away from macrofauna, and therefore avoiding disturbance or potential mortality.

2.2.1. Aquarium study

To investigate the effect of macrofauna on survival and growth an aquarium study was conducted with three main factors. The first factor was macrofauna species with two levels (adult cockles present, and lugworms present). The second factor was the density of macrofauna which had two levels of full density (four lugworms, fourteen adult cockles) and half density (two lugworms, seven adult cockles). The third main factor was the presence or absence of a spatial refuge from macrofaunal disturbance which was only evaluated at 'half density' (two lugworms or seven adult cockles in one side of the tank only). Controls were also conducted with no macrofauna present. For post-larval survival a significant effect of the macrofauna species was expected, due to expected lower survival with lugworms compared to the cockle and then control treatments. Density and refuge presence was also expected to show a significant effect, with survival decreasing from the refuge treatment, low density and then high density treatment having the lowest survival. Due to the control treatments lacking macrofauna and therefore not expected to show differential survival between levels of the 'density and refuge' factor, a significant interaction was predicted. The 'full density' of macrofauna was calculated to represent the natural densities found in the locations of the plot studies (approximately 150 m⁻² for adult cockles and 25 m⁻² for lugworms). The 'half density' level was to help separate out the effect of a reduced macrofauna density from the provision of a spatial refuge on post-larval mortality. The spatial refuge level was added to identify if post-larvae would crawl away from close proximity to macrofauna, and associated disturbance, and thereby an increase in survival and growth is likely be observed in treatments with the spatial refuge provided. The refuge was established by placing a rigid plastic mesh (5 mm mesh size) into the sediment to the bottom of the aquarium and also protruding above the water surface (Fig. 4.5). This would allow post-larvae only to move through the divider and not the macrofauna. Two of the lugworm x refuge treatments failed to work as faecal casts were seen on both sides of the mesh divider, and so the provision of a spatial refuge with lugworm presence could not be tested. Control treatments were established to identify the survival rate and refuge use during the experiment independent of macrofauna presence. Each treatment had four replicates simultaneously conducted in plastic aquaria measuring 40 x 30 x 32.5 cm (height x width x length) in size. The aquaria were closed systems supplied with aeration, and 50% water changes every two days were conducted using filtered seawater (settled and passed through 45 µm mesh). Each aquarium was filled with kiln dried sand to a depth of 24 cm to allow the lugworms enough depth to establish burrows. The sediment was sieved through a 500 µm mesh before it was added, to

facilitate easier sampling of the post-larvae at the end of experiment as post-larvae would be > 500µm in shell length.

Adult cockles and lugworms used in the experiment were collected from Traeth Melynog with mean lengths of 29.0 ± 0.4 and 131.6 ± 4.3 mm ($\pm 95\%$ CI) respectively. The adult cockles and lugworms were added to the aquariums a week before the *C. edule* post-larvae were added to allow acclimation and for elevated crawling activity to stop, which often occurs when cockles are added to new sediment (Richardson et al., 1993).

Two hundred post-larvae were used in each aquarium with one hundred haphazardly distributed on each side of the dividing mesh. The number of post-larvae used in the tanks represented a density of 2,254 m⁻², which is at the upper end of density at Traeth Melynog and the lower end of the Dee estuary soon after settlement. The post-larvae were collected from the Dee estuary a month earlier and at the start of the experiment on the 18th July 2012 had a mean shell length of 1.8 ± 0.7 mm ($\pm 95\%$ CI). The organisms were fed three times a day with the microalgae *Isochrysis galbana* (T-Iso) with a total feeding of 1.3125×10^9 cells tank⁻¹ day⁻¹, based on calculated adult cockle feeding requirements (Helm et al., 2004) and the volume of microalgae culture available for use. This was used to maintain macrofauna and post-larvae condition during the experimental period, and prevent food being a limiting factor for growth and survival. Temperature and salinity were checked on a regular basis so that any dramatic changes would be observed; the mean temperature was 17.7 °C and salinity 32 over the course of the experiment. The experiment was run for two

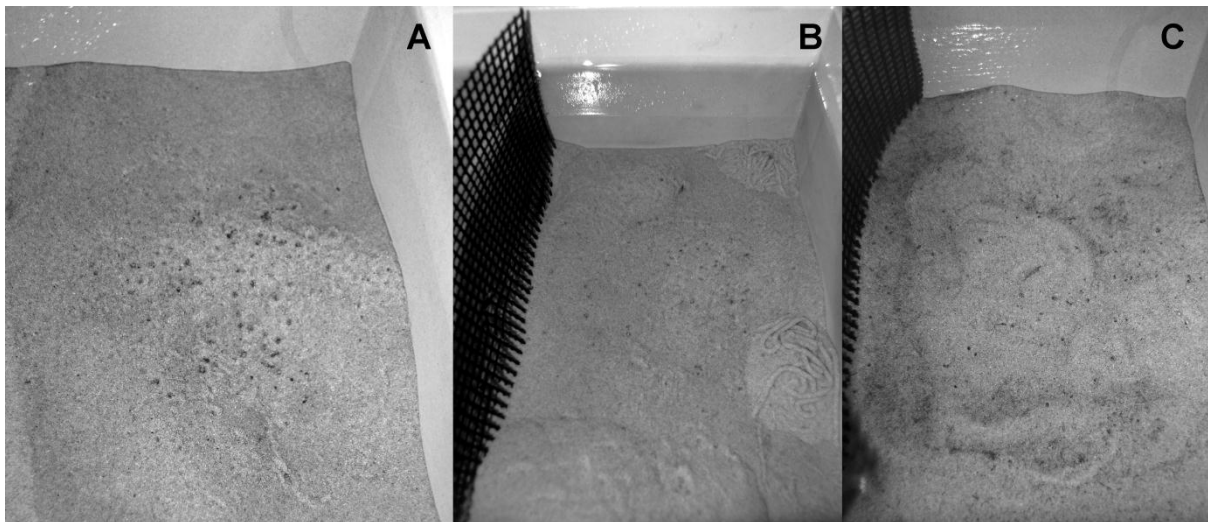


Figure 4.5. Examples of the sediment surface in control (A), lugworm *Arenicola maria* (B) and adult cockle *Cerastoderma edule* (B) experiment aquariums. Increased sediment heterogeneity can be seen in adult cockle and lugworm treatments. Eighty *C. edule* post-larvae (seen clearly in the picture of a control aquarium) were added to each aquarium for the two week experiment.

weeks, at the end of which *C. edule* post-larvae were recovered by siphoning out the sediment from each tank (each half separately) over a 500 µm sieve. Recovered post-larvae were counted, and shell length measured in a sub-sample from each replicate tank.

2.2.2. Flume study

To assess the effect of macrofauna disturbance on escape responses of post-larvae the number of byssus-drifting *C. edule* post-larvae were compared in the presence and absence of macrofauna using a laboratory based flume. Each of three treatments (adult cockles present, a lugworm present and a control with no macrofauna) was replicated five times. A recirculating flume (Fig. 4.6) was used to provide flow of 3 cm sec⁻¹, as measured by an ultrasonic doppler velocity profiler (UVDP) at the sediment surface. The flow speed was selected based on trials which balanced drifting potential of post-larvae and sediment erosion, and therefore potential passive scouring of the post-larvae and is typical of current speeds either side of slack water (Collins et al., 1998). The flume included a sediment box (24 x 11.5 x 25 cm, length x width x depth) which post-larval cockles and macrofauna could burrow into. The entire sediment box was filled with 2 parts < 500 µm kiln dried sand 1 part dried mud collected from the Dee estuary to encourage lugworm feeding activity. This sediment was made flush with the flume working area which had a thin layer of the same sediment covering it. This ensured the same bed roughness across the whole flume working area. A 500 µm net covered the flume's cross section downstream of the sediment box to retain any *C. edule* post-larvae carried away from the sediment box (Fig. 4.6).

At the start of each experiment the flume was filled with 40 µm filtered seawater. Adult cockles or a lugworm were added to the sediment box in the appropriate treatments in a density that was representative of the aquarium study (3 adult cockles or 1 lugworm which represents approximately 150 m⁻² adult cockles and 25 m⁻² lugworms). The flume was then turned on at 1 cm / sec to provide initial water circulation. Six hours after the macrofauna was added the flume was switched off and eighty *C. edule* post-larvae were added to the sediment surface of the sediment box at haphazardly selected locations. The *C. edule* post-larvae used had been collected from the Dee estuary and were maintained in mesocosms until being sorted for use in the experiment; they had a mean ± 95% CI shell length of 1.7 ± 0.02 mm. Two hours after the post-larvae were added the flume was switched on with a 0.5 cm sec⁻¹ min⁻¹ increase in flow velocity until 3 cm sec⁻¹ was reached. At the same time 1.75 x 10⁹ *Isochrysis galbana* (T-Iso) microalgae cells were added to provide food. The working area of the flume was covered with a black plastic sheet to block ambient light as drifting is more likely to occur in the dark (Armonies, 1992; Hiddink et al., 2002a). After a six hour period, which was chosen by conducting trial experiments, the flume was switched off, the

net removed from the flume and all post-larvae in the net counted and measured. All the sediment in the sediment box and on the working area downstream of it was then siphoned out and passed over a 500 μm mesh. All retained *C. edule* post-larvae were counted and a sub-sample from each replicate had their shell length measured to test for differences in size of drifters and those remaining in the sediment.

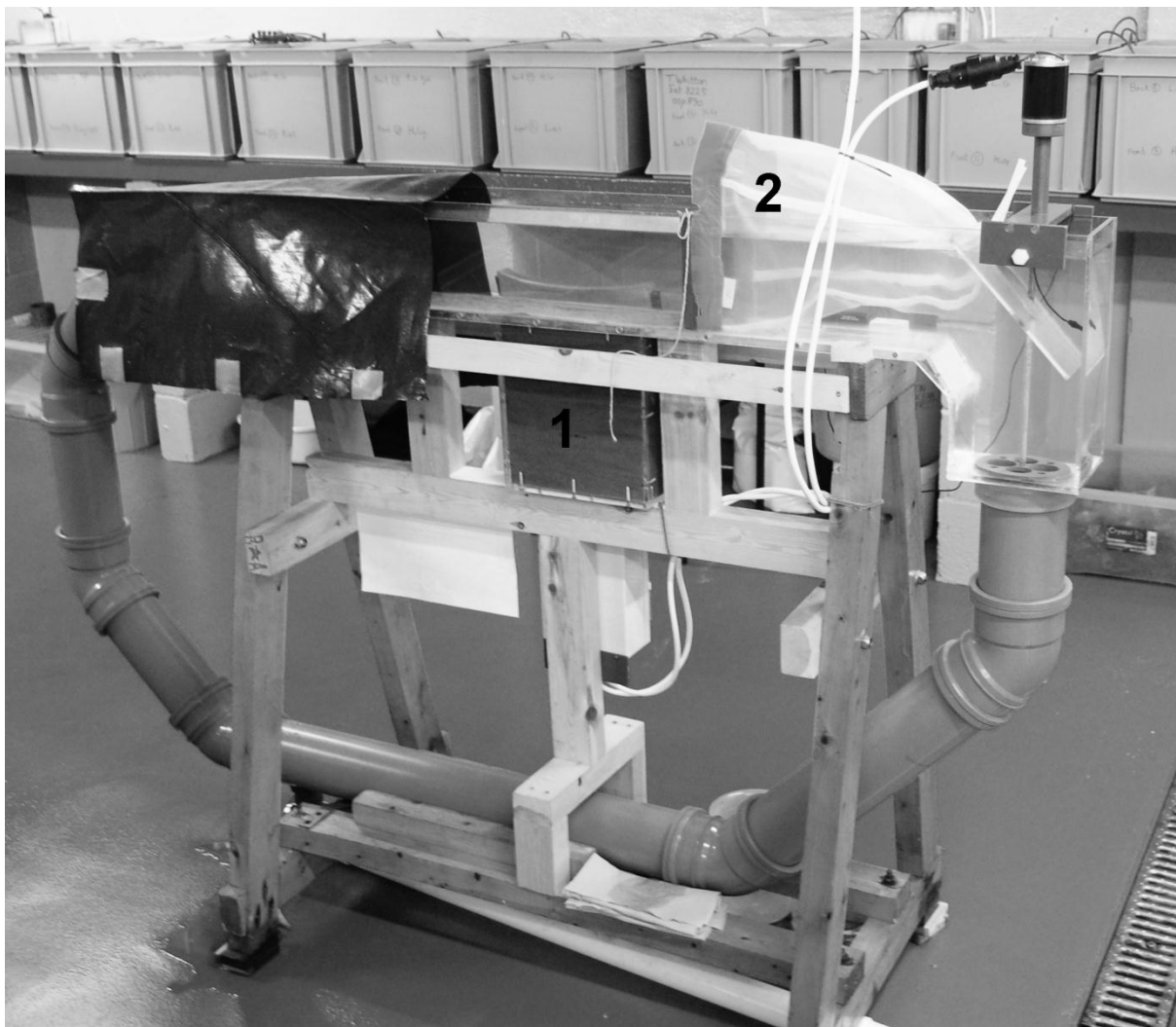


Figure 4.6. Recirculating flume used to record drifting rates of *Cerastoderma edule* post-larvae with and without the presence of macrofauna. Macrofauna and *C. edule* post-larvae were placed onto and burrowed into the sediment box (1) and any drifting post-larvae were caught in the 500 μm net downstream (2).

2.3. Data analysis

All mean values are reported and displayed \pm 95% Confidence Intervals (CI) unless stated otherwise. Data was checked for deviations from normality and equal variance before analysis of variance (ANOVA) was conducted. If the assumptions of ANOVA were not met a

log₁₀ transformation was conducted, failing that non-parametric Kruskal-Wallis tests were used. The field experiment plots were repeatedly sampled over time and therefore a repeated measures (mixed) ANOVA was used, as each sampling time cannot strictly be considered as independent. Sphericity was tested using Mauchly's test before the repeated measure ANOVA was conducted. Sphericity was not met for the lugworm plot data, and so the Greenhouse-Geisser adjusted degrees of freedom were used. The assumption of homogeneity of variance could not be met for shell length data in the aquarium study when conducting the factorial ANOVA, even after outliers were removed, hence differences for each level of the factor 'density and refuge' were analysed using separate one-way ANOVAs for each level of the 'macrofauna' factor.

3. Results

3.1 Field exclusion experiments

3.1.1 Lugworm exclusion experiment

The density of lugworms was sampled on the 20th July to determine if the exclusion had worked (Fig. 4.7). Some lugworms occurred in the exclusion plots but the mean density was less than 20% that in other treatment plots (Mean \pm 95% CI, Exclusion 5.6 ± 6.2 / m², Control 30.6 ± 10.3 and Procedural Control 26.1 ± 8.7 , $F_{(2,17)} = 16.20$, $p = <0.001$). The control and procedural control lugworm density were not significantly different (Tukey post-hoc test, $p = 0.619$).

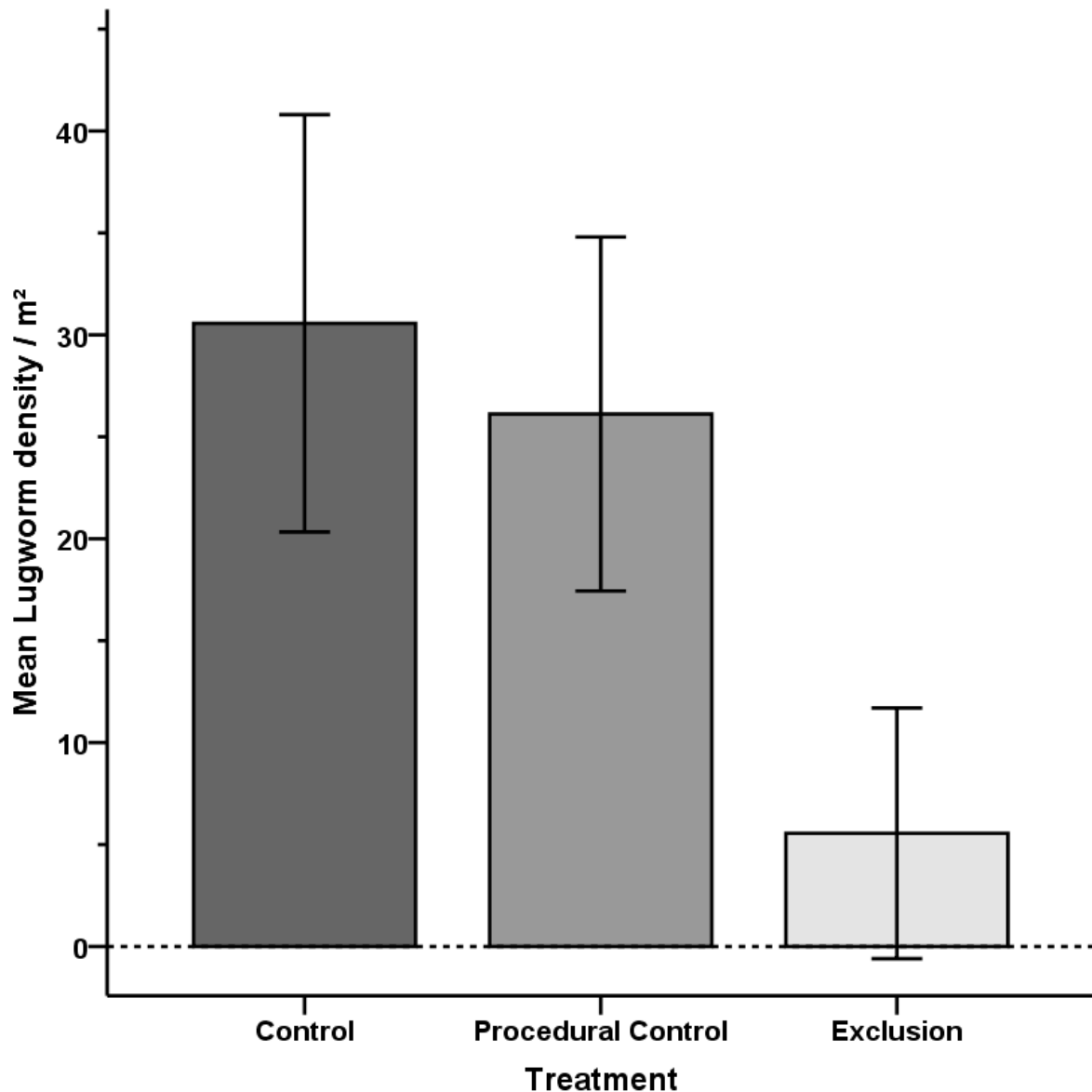


Figure 4.7. Mean density / m² of lugworms *Arenicola marina* for each treatment on the 20th July 2012 at Traeth Melynog, Anglesey. Error bars show the 95% confidence interval for the mean.

On the first sampling occasion 93% of the *C. edule* were below 1 mm in shell length, indicating larval settlement had occurred recently. The presence of individuals of 0.4 mm in shell length indicated settlement was still underway (Fig. 4.8). Small post-larvae with a shell length less than 1 mm were found on all sampling occasions. This suggests larvae were likely settling beyond the main settlement period in May and colonising the plots.

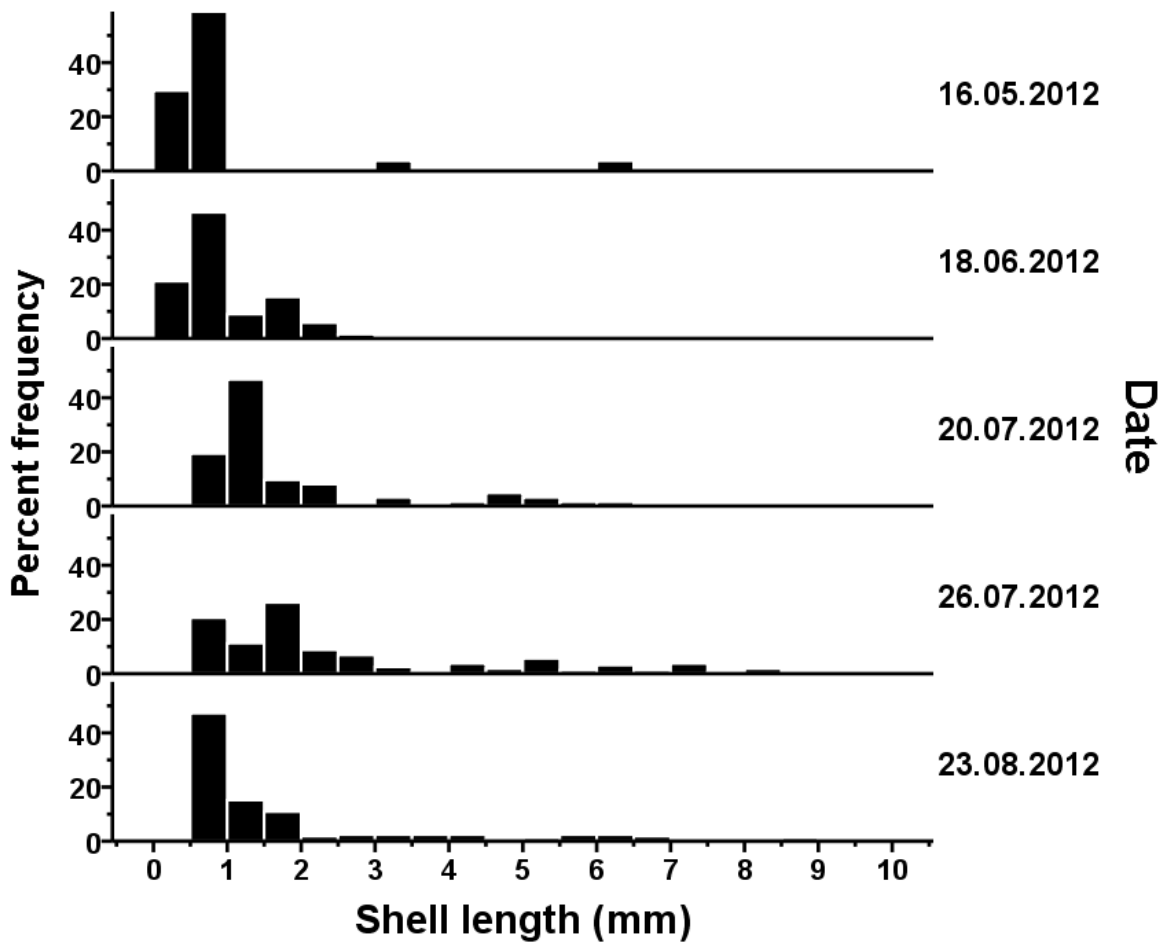


Figure 4.8. Size distribution of *Cerastoderma edule* post-larvae at Traeth Melynog, Anglesey UK. Percent contribution is calculated per sampling date.

When all treatments are considered repeated measures ANOVA indicated a significant main effect of sampling date on post-larval density (within-subject factor, $F_{(1.8, 27.1)} = 13.891$, $p < 0.001$) which would be expected as densities change from settlement into the late summer. The control and procedural control plots had similar densities throughout the experimental period (the procedural control had slightly lower densities for most sampling events) giving confidence that a direct or indirect effect of significantly reducing the lugworm abundance in the exclusion plots was responsible for the observed densities of post-larvae, and not the experimental disturbance of installing the mesh (Repeated measures ANOVA on control and procedural control only, $F_{(1,10)} = 0.740$, $p = 0.410$). Importantly the post-larval density was significantly higher in the lugworm exclusion plots during the experiment (between-subjects factor, $F_{(2,15)} = 4.868$, $p = 0.023$), with the exception of the first sampling event during the main settlement period (Fig. 4.9). The mean density of *C. edule* post-larvae in the lugworm exclusion plots was approximately double or more than measured in the procedural control plots, again with the exception of the May 16th sampling event. There was no interaction between the two factors ($F_{(3.62, 27.14)} = 0.848$, $p = 0.498$), validating that the higher post-larval density in the exclusion plots had a consistent temporal trend. This

provides strong evidence in support of the hypothesis that excluding lugworms increases post-settlement densities of post-larval *C. edule*.

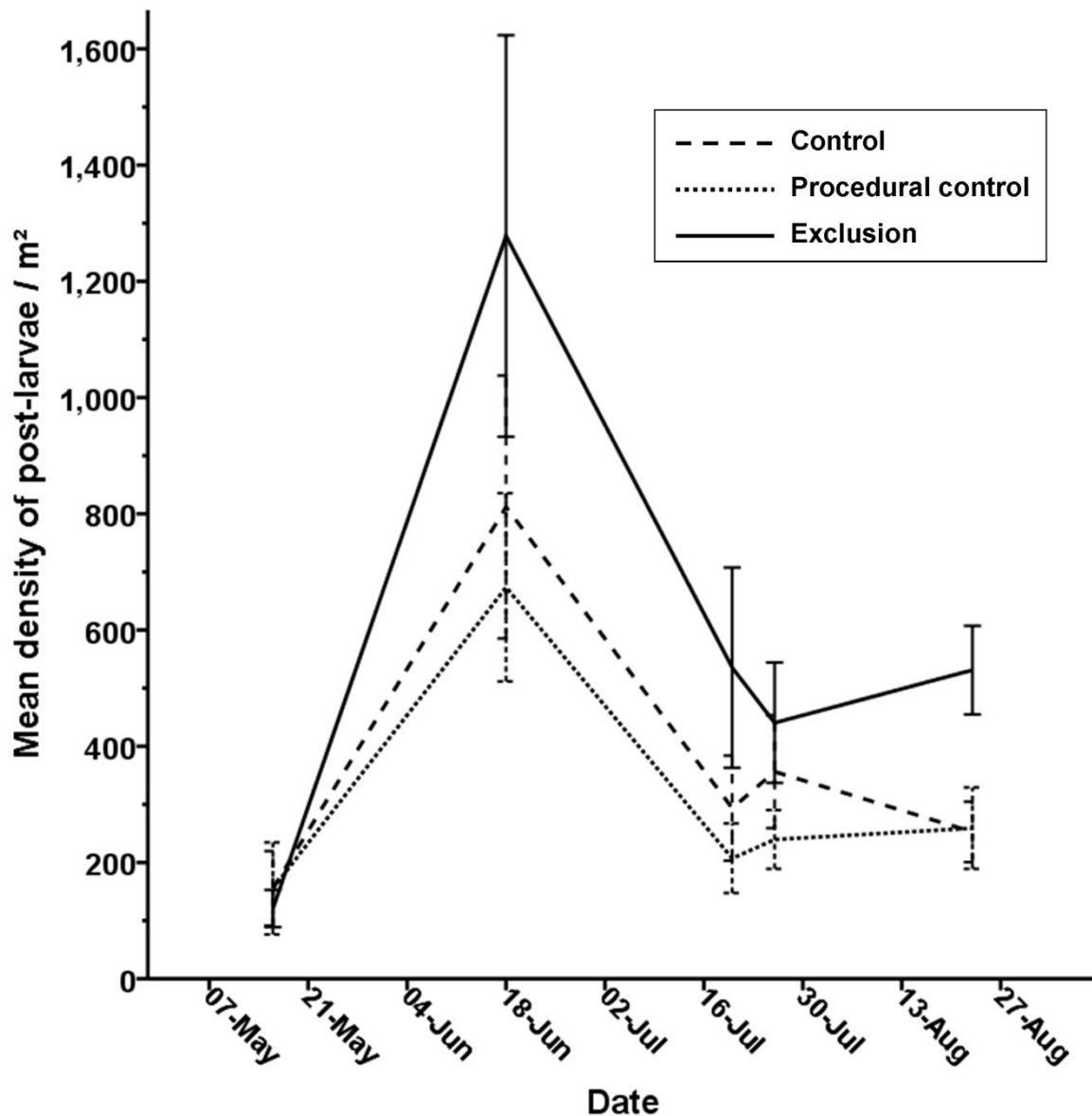


Figure 4.9. Mean number of *Cerastoderma edule* post-larvae / m² (n = 6 per data point) for each experimental treatment at Traeth Melynog, Anglesey. Error bars show one standard error for the mean.

3.1.2 Adult *C. edule* exclusion experiment

The adult *C. edule* exclusion plots had a much lower adult *C. edule* mean density of 17 ± 18 m⁻² ($F_{(2,15)} = 7.87$, $p = 0.005$, $(x+1)\ln_{10}$ transformed data) with smaller variability than the control and procedural plots (Fig. 4.10). The control and procedural plots had a mean adult *C. edule* density of 105 ± 92.6 and 160 ± 138.8 m⁻² respectively and were not significantly different from each other (Tukey post-hoc comparison $p = 0.019$).

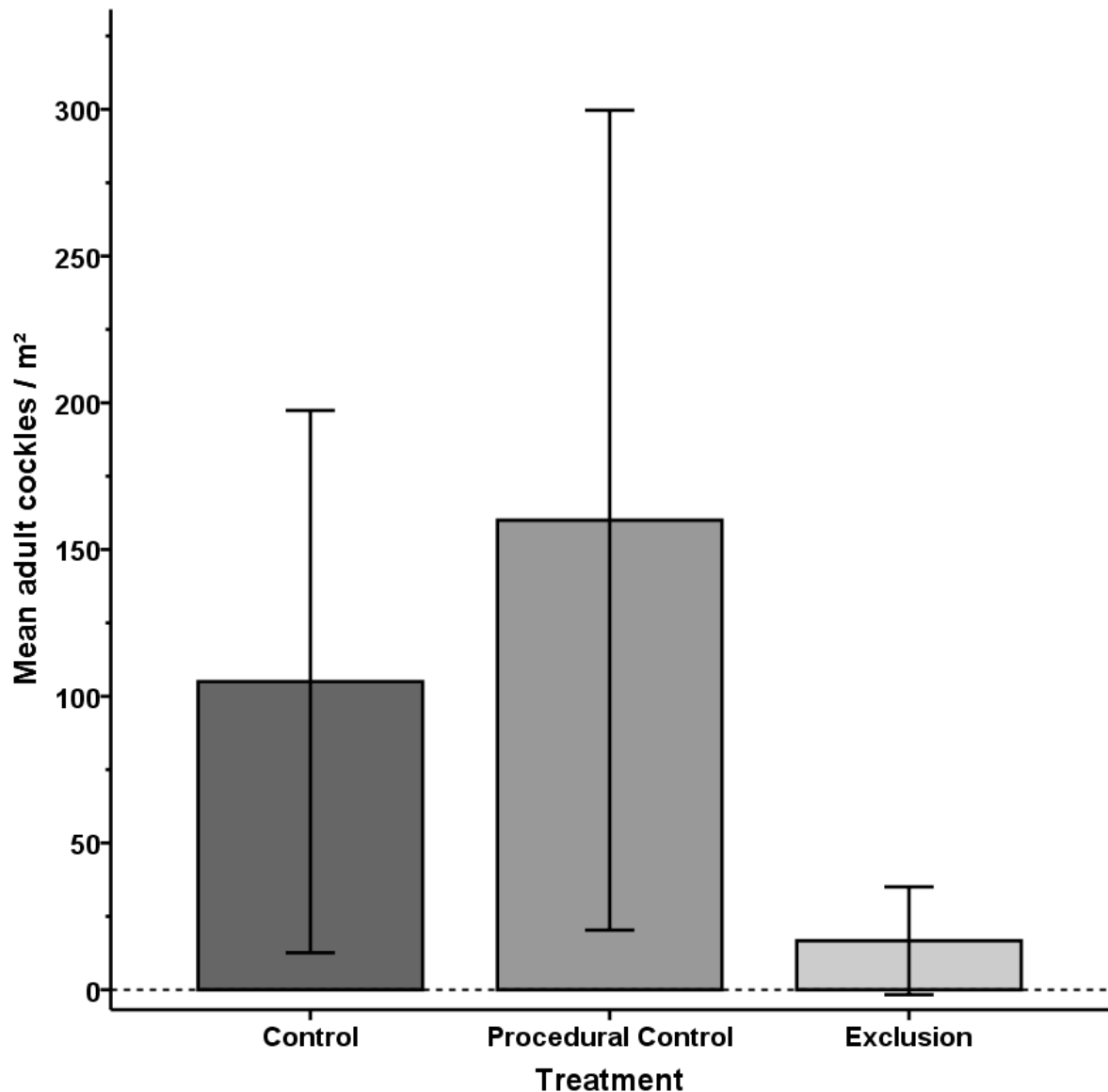


Figure 4.10. Mean density per m² of adult *Cerastoderma edule* in the field manipulation plots recorded on the 2nd July 2012 at the Dee estuary, UK. Error bars show the 95% confidence interval for the mean and n = 6 per treatment.

On the first sampling event on May 5th all *C. edule* post-larvae were below 750 μ m in shell length with over 70 % below 500 μ m, indicating that larval settlement was occurring at this time (Fig. 4.11). Unlike Traeth Melynog a clear single cohort can be followed from this settlement in May through to the last sampling event on the 2nd of July, in which the mean shell length reached was $2733 \pm 118 \mu$ m (2.7 ± 0.1 mm).

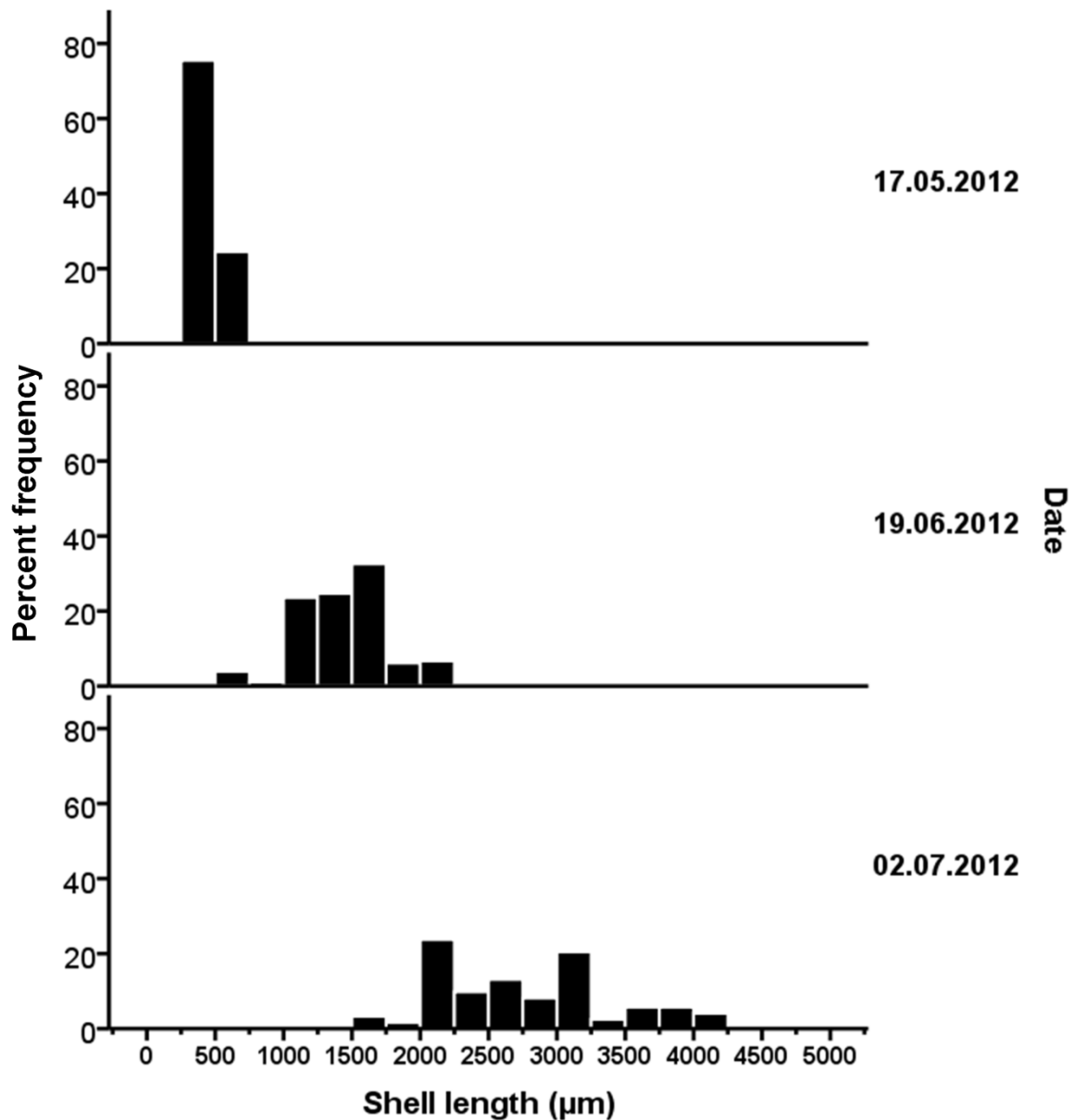


Figure 4.11. Length frequency histogram of shell lengths of *Cerastoderma edule* post-larvae from the Dee estuary experimental plots for each sampling event. Percent frequency is calculated per sampling date.

C. edule post-larval densities were highest in the control plots with slightly lower densities in the procedural control plots, and the density of post-larvae in the adult *C. edule* exclusion plots were significantly lower (Repeated measures ANOVA on ln10 transformed data, between-subjects factor, $F_{(2,15)} = 4.634$, $p = 0.027$) (Fig. 4.12). On average across the three sampling events the densities of post-larvae in the exclusion plots were 50% lower than those in the procedural control (ranging from 59% lower in May to 44% lower in June). Densities of *C. edule* post-larvae in the plots showed a consistent trend on all sampling events between treatments (Fig. 4.12) validated by no interaction of the factors ($F_{(4, 30)} = 0.608$, $p = <0.66$). Density of post-larvae significantly increased with time (within-subjects

factor, $F_{(2, 30)} = 87.678$, $p = <0.001$) during the experiment, indicating net migration into the plots between June and July as no settlers were found at these events (Fig. 4.11).

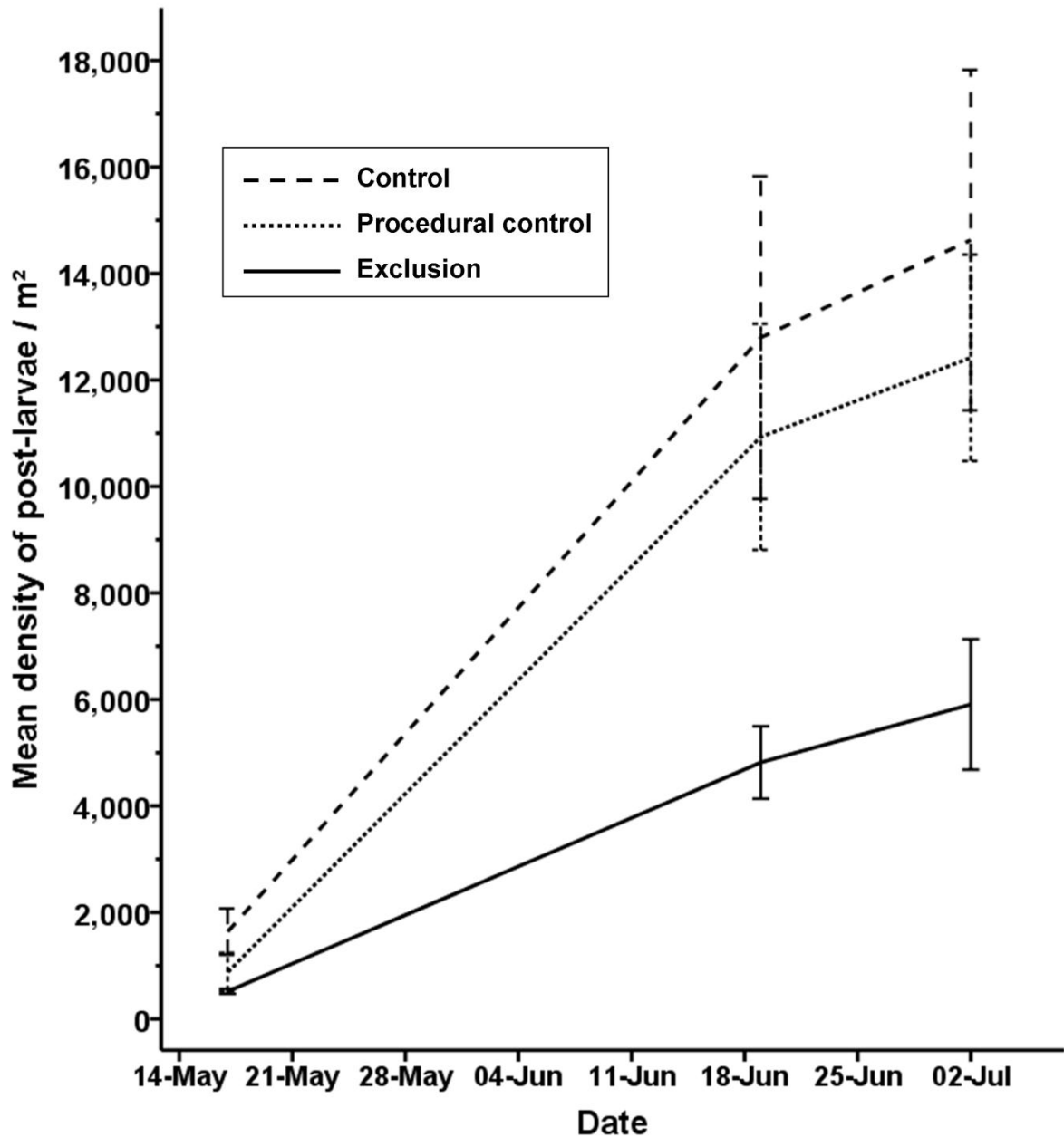


Figure 4.12. Mean density of *Cerastoderma edule* post-larvae / m² (n = 6) for each experimental treatment at the Dee estuary, UK. The exclusion treatment reduced the densities of the adult *Cerastoderma edule*. Error bars show one standard error for the mean.

3.2 Laboratory studies

3.2.1. Aquarium study

3.2.1.1 Survival of *C. edule* post-larvae in the presence and absence of macrofauna

Lower survival in all treatments that had macrofauna present was predicted. No significant effect was observed of macrofauna presence (arcsine transformed proportion, $F_{(2,27)} = 2.453$,

$p = 0.105$) on post-larval survival (Fig. 4.13). There was a non-significant trend for lower survival (and higher variability) in the control treatments and treatments with lugworms present compared to those with adult cockles present (Fig. 4.13). When the data point for each treatment replicate is plotted the upper limit of survival for each treatment was between 91 and 97% survival, with most points clustered above 85% survival (Fig. 4.14). However five replicates had noticeably lower survival of below 70% with 3 below 50%, which included a control replicate. This indicates that some of the replicates produced post-larval mortality regardless of any treatment level effect of macrofauna presence.

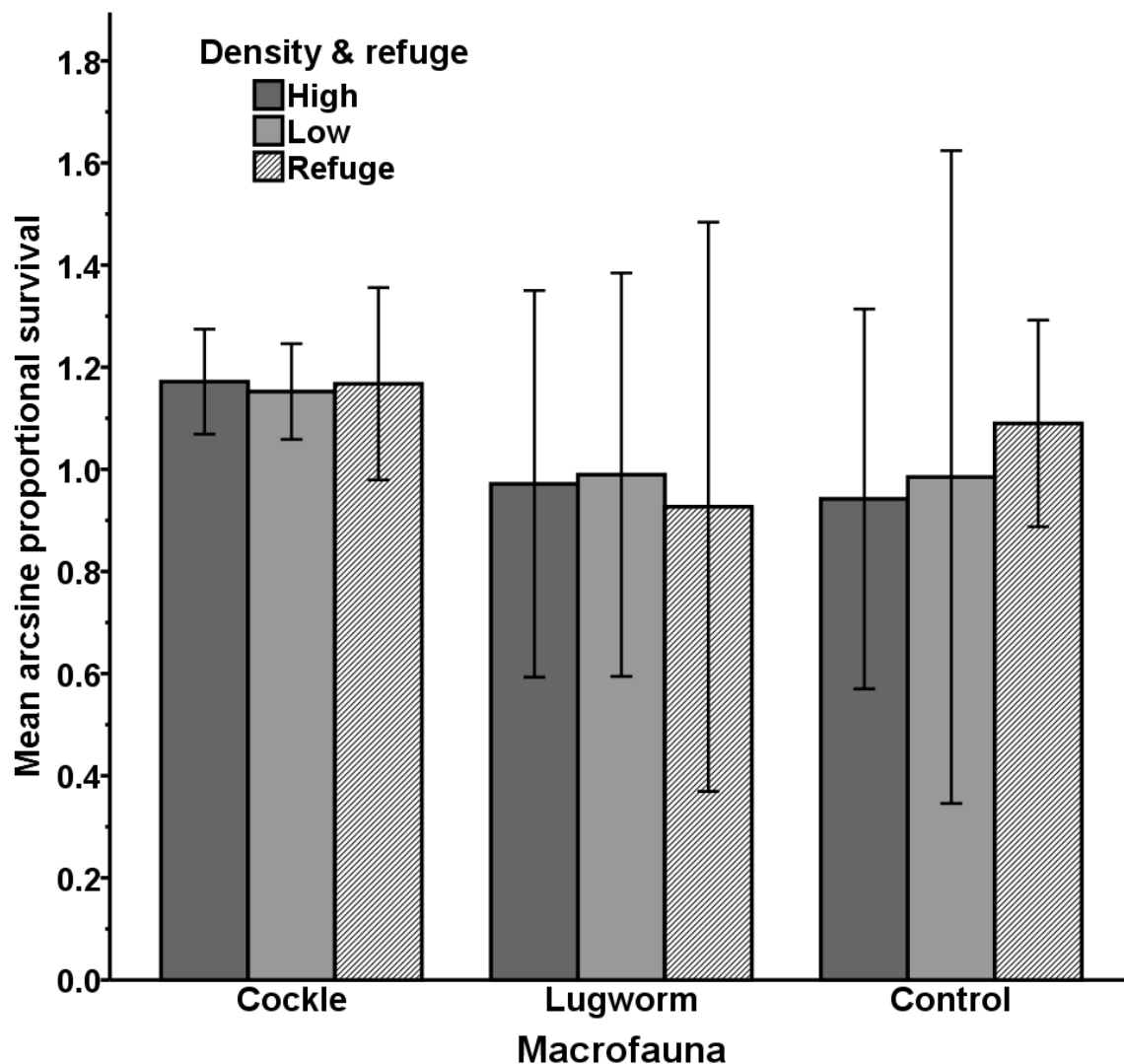


Figure 4.13. Mean survival of post-larval *C. edule* ($n = 4$ per treatment) when confined with adult cockles, lugworms *Arenicola marina* and in control (which had no macrofauna) factor levels across three treatments of high density, low density and the provision of a spatial refuge from macrofauna in one half of the experimental aquariums. Error bars show the 95% confidence interval for the mean.

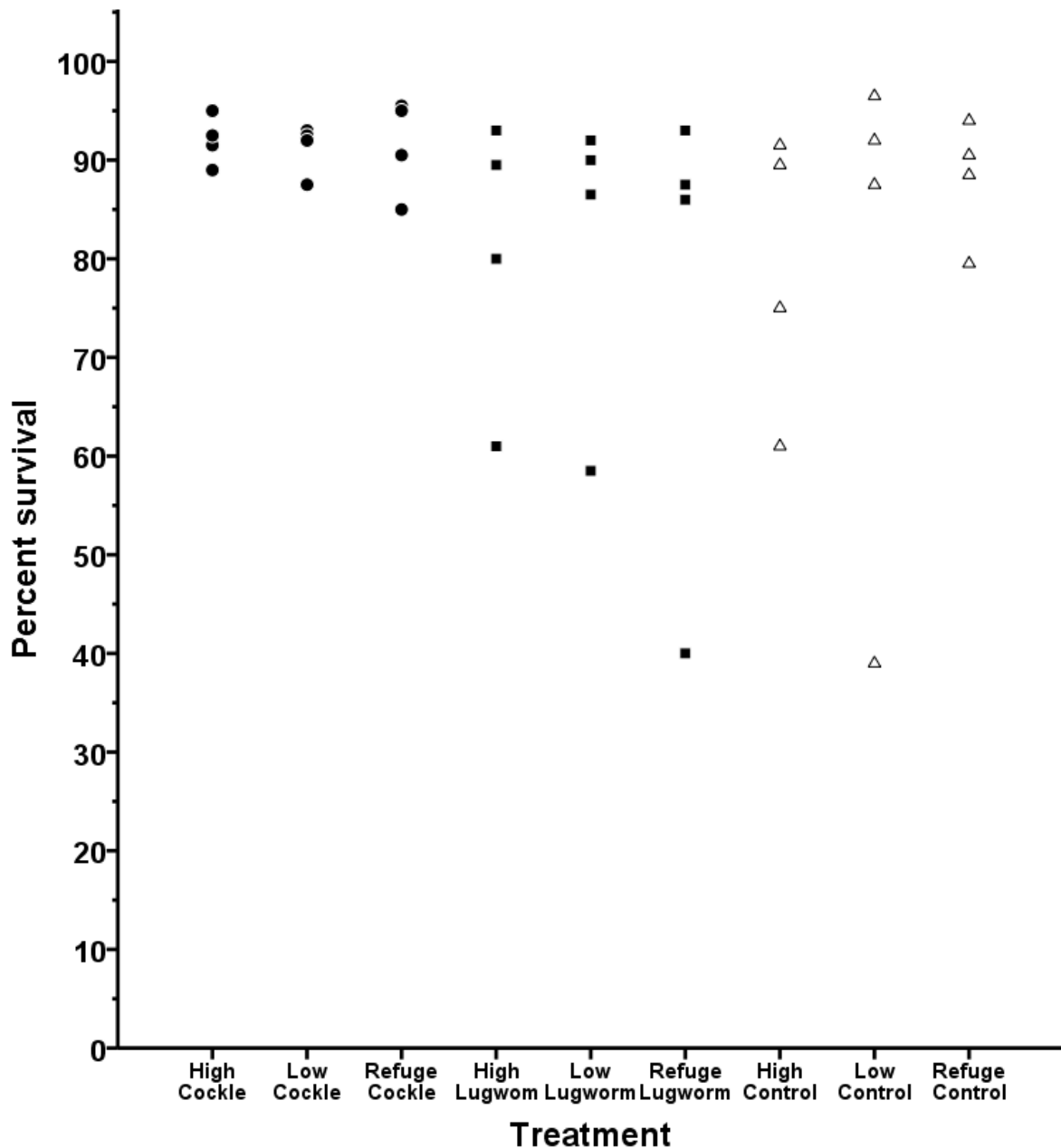


Figure 4.14. Survival data for each replicate aquarium of the two hundred *Cerastoderma edule* post-larvae in the experimental treatments (n = 4 / treatment).

3.2.1.2. Growth of *Cerastoderma edule* post-larvae

When macrofauna were present it was predicted that post larval growth would be lowest in high density treatments increasing with low density and then refuge treatments. The three control treatments were predicted to have the highest and similar growth during the experiment due to no disturbance. The mean shell length of *C. edule* post-larvae at the beginning of the experiment was $1801 \pm 66 \mu\text{m}$ ($1.8 \pm 0.6 \text{ mm}$). Factorial ANOVA could not be conducted as the assumption of homogeneity of variance could not be met. Therefore the density and refuge factor levels were tested for each macrofauna factor level individually. In

the adult cockle treatments the mean shell length at the end of the experiment was highest in the refuge treatment, with the high density treatment was significantly lower by 206 μm (one-way ANOVA, Log transformed data, $F_{(2,134)} = 5.737$, $p = 0.004$) with a mean of $1982 \pm 88 \mu\text{m}$ (Fig. 4.15). In the lugworm treatments there was a trend for a higher final shell length in the refuge treatment but differences were non-significant ($F_{(2,84)} = 1.400$, $p = 0.252$) (Fig. 4.15). The control treatments showed no significant differences between final shell length as well (Kruskal Wallis test, $\chi^2 = 4.948$, $p = 0.84$) (Fig. 4.15).

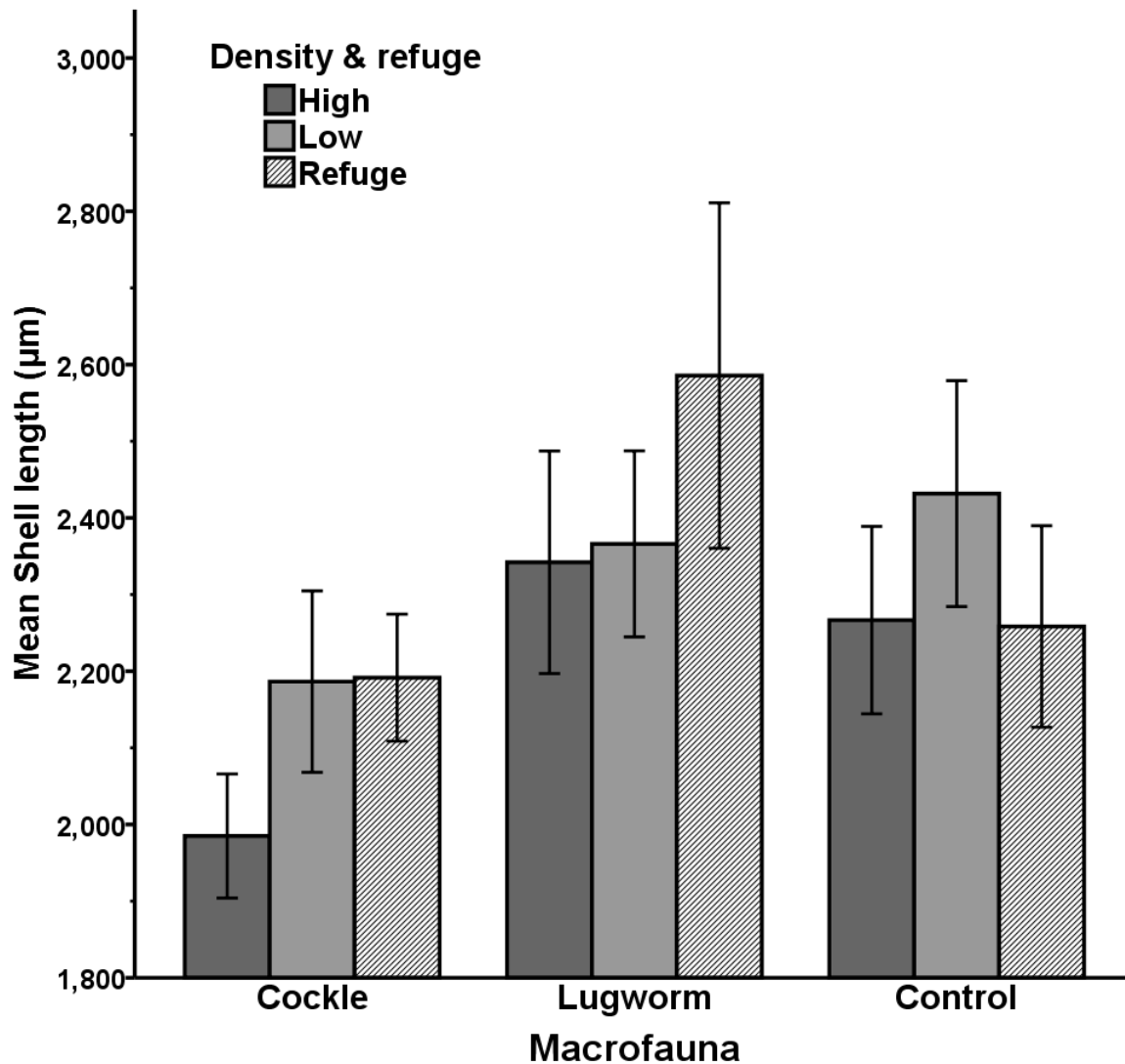


Figure 4.15. Mean (bars show \pm 95% CI) of *Cerastoderma edule* post-larvae shell length at the end of the 2 week aquarium study investigating the effect of macrofauna on survival and growth. At the start of the experiment *C. edule* post-larvae had a mean \pm 95% CI $1800 \pm 66.7 \mu\text{m}$.

3.2.1.3. Use of a spatial refuge

Two of the four lugworm refuge treatments failed due to lugworms managing to get past the mesh divider, and so disturb both sides of the aquaria and cannot be tested for refuge use.

For the adult cockle treatments where comparisons can be made of the number of live and dead cockles recovered between the refuge and the adult cockle side of the aquariums were tested. One-way ANOVA provided no evidence that less dead, or more live cockles, were recovered from the spatial refuge side ($F_{(1,6)} = 0.798$, $p = 0.406$ and $F_{(1,6)} = 0.002$, $p = 0.964$ respectively).

3.2.2. Flume study

The flume experiment was conducted to test the hypothesis that more post-larval *C. edule* would byssus drift in the presence of macrofauna when compared to macrofauna free sediment, due to disturbance. The number of post-larvae drifting was not significantly different between treatments (arcsine transformed proportion data, one-way ANOVA, $F_{(2,12)} = 0.743$, $p = 0.496$) (Fig. 4.16). Observations during the experiment indicated that in three out of the five lugworm treatments there was no evidence of feeding or bioturbation (no feeding funnel or faecal casts), which means the lugworm treatment may have little resemblance to the disturbance that lugworms may have in the field on *C. edule* post-larvae. Removal of these replicates did not change the analysis outcome. Post-larvae would quickly burrow after introduction to the flume. However at the end of the experiment some post-larvae could be observed on the surface with a byssus thread trailing behind (Fig. 4.17). The number of post-larvae drifting was relatively low in all the treatments and reached a maximum of 33 out of the original 80 post-larvae (41.3%) in one of the cockle treatment replicates. The size distribution of post-larvae did not differ between treatments but post-larvae recovered from the nets had a smaller size (lower minimum, maximum and median shell length) than those recovered from the sediment (Median length tested with a Kruskal Wallis test, $\chi^2 = 13.6$, $p < 0.001$).

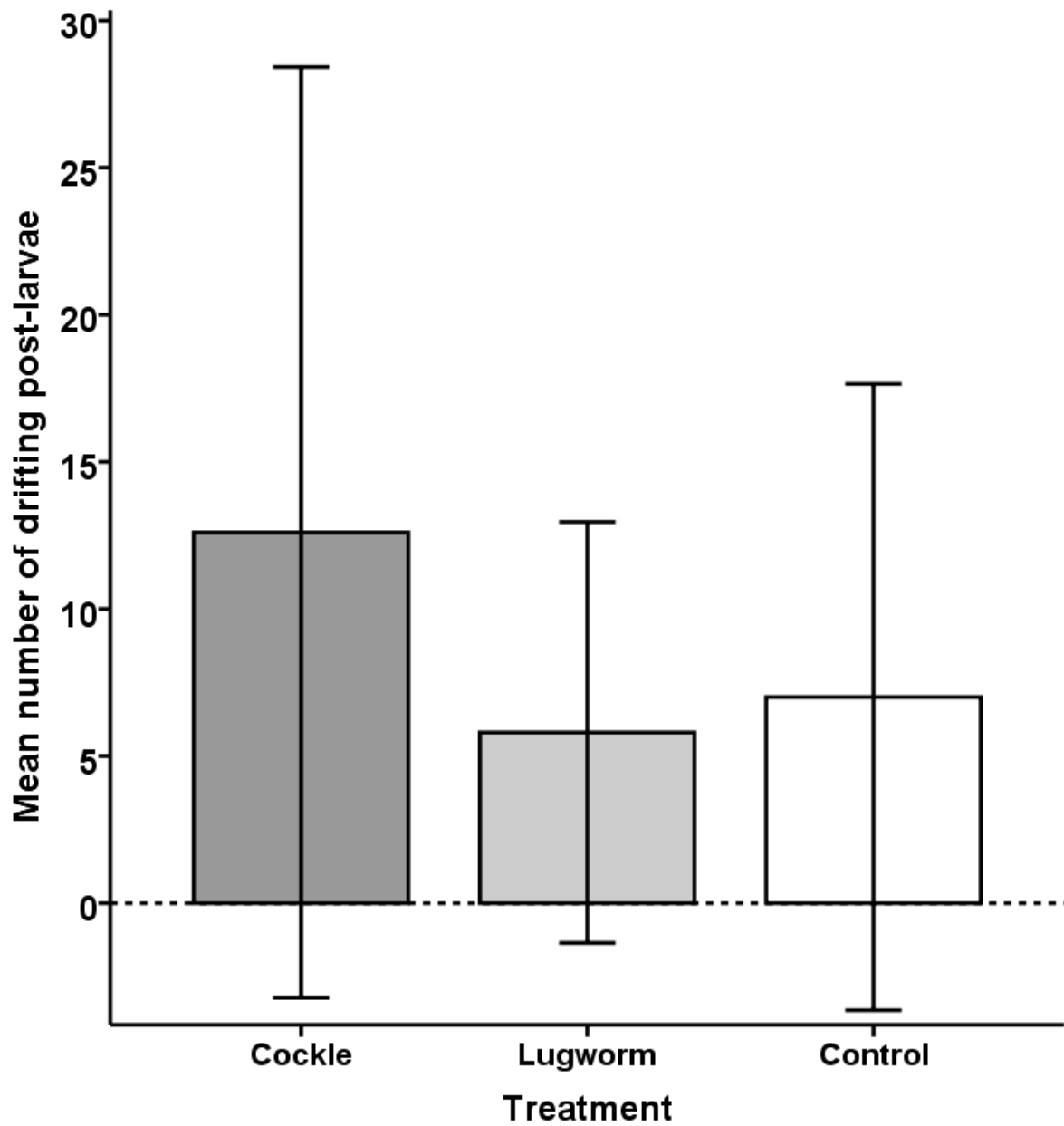


Figure 4.16. Number of *Cerastoderma edule* post-larvae drifting during the flume experiment with macrofauna (Adult cockle *C. edule* and the lugworm *Arenicola marina*) present in the sediment and devoid of macrofauna (Control). Error bars are 95% confidence intervals.



Figure 4.17. A post-larval *Cerastoderma edule* (circled) from the flume experiment with sand grains attached to its single byssus thread, it is being held by forceps at the top of the picture.

4. Discussion

The overarching question of this study was what effect macrofauna have on *C. edule* post-larval densities, and how this contributes towards the observed spatial patchiness of *C. edule* populations. I hypothesised that the presence of macrofauna would negatively affect the density of cockle post-larvae but field experiments revealed contrasting effects of the two bioturbating species; there was a negative effect of lugworms on cockle post larvae as expected but the presence of adult cockles caused an increase in post larval densities rather than a reduction. To understand what is driving such observations two laboratory experiments were conducted. In the aquarium experiment no significant difference was found in survival of post-larvae when confined with adult cockles, lugworms and in the absence of macrofauna regardless of the density or if a spatial refuge was provided. The only significant growth difference observed in the experiment was the high density of adult cockles reducing the post-larval growth when compared to the low density and refuge treatments. I found no significant advantage to post-larvae of providing a spatial refuge from macrofauna presence. Our final hypothesis tested was that more post-larvae would be recorded byssus drifting when in the presence of macrofauna due to the disturbance experienced. I found no significant evidence to support this from the flume experiment.

Clear effects of excluding lugworms and adult cockles on post-larval cockle densities were seen during the post-settlement period and were species specific, effecting densities positively and negatively respectively. The aquarium laboratory experiments indicate that the

observed effects in the field are not produced through direct interactions with macrofauna from ingestion or bioturbation disturbance and smothering. The flume experiment provide no significant evidence that macrofauna increase the rates of byssus migrations which may influence densities seen in the field. Macrofauna do affect post-larval densities significantly during the post-larval period and therefore may strongly influence spatial heterogeneity in cockle distribution. However the causal processes remains to be identified and proven.

In previous studies on soft sediment infauna the exclusion of lugworms shows there are species which benefit and those that are negatively affected (Reise, 2002). The post-larvae of *C. edule* benefit based on the results of the field experiment from this study. *C. edule* post-larvae have little benefit from the positive effects of lugworm presence that are experienced by some species, such as increased local oxygen penetration into the sediment and increased bacterial production. This is because cockle post-larvae are suspension feeders and shallow burrowers which occupy the top few mm of sediment which is sufficiently oxygenated by diffusion. Stable sediment is likely a desirable feature for *C. edule* post-larvae due to lower disturbance of feeding and risk of being smothered by the lugworm's faecal casts or entering into the feeding and burrow shaft. The sediment stabilisation in lugworm exclusion experiments often benefit species which prefer stable sediment, such as the tube building polychaetes *Pygospio elegans* and *Lanice conchelig* (Volkenborn et al., 2009). The presence of lugworms has been shown to result in a decrease in observed numbers of *C. edule* post-larvae on small spatial scales, on average by 52%, in other field experiments (Flach, 1992; 2003). However there does not appear to be any form of linear response of lugworm density and post-larval density when tested at five lugworm densities with a maximum of 44 m⁻² (Flach, 1992). In our study I also saw around a 50% reduction in *C. edule* post-larval density in plots with lugworms when compared to exclusion plots. From past studies and this one there is strong evidence that the presence of lugworms will reduce post-larval densities by around 50%, when compared to adjacent lugworm free sediment.

The exclusion of adult cockles caused a decrease in the density of *C. edule* post-larval, which is contrary to predictions on the interactions between *C. edule* settlement and conspecific adult presence. This is also in disagreement with our starting hypothesis, which I now reject, that excluding adult cockles will increase densities of *C. edule* post-larvae. Empirical evidence of a negative relationship between adults and post-larvae on small scales is less consistent than shown for lugworms from the literature. A longer period of recovery would have been better, but locating a suitable area for the plots took longer than expected due to low densities of adult *C. edule* in a historically high density area that was to be originally used.

During the first sampling event at the initial settlement period there was a reversed treatment effect and a smaller difference between treatments in the lugworm and cockle exclusions respectively on post-larval density. This suggests that at small scales ($\leq 1 \text{ m}^2$) the presence of lugworms and adult cockles has little effect on initial larval settlement success, despite being the period when post-larvae are at their smallest and therefore potentially most vulnerable to ingestion and smothering (Andre and Rosenberg, 1991). In agreement with Flach (1996; 2003) I conclude that a direct or indirect interaction with macrofauna is changing the post-larval densities during the post-settlement period, rather than at settlement itself. There is no apparent linear relationship between adult cockle density and post-larval density. In previous studies using adult cockle densities of 55 – 70 m^{-2} a positive effect or no effect at all has been observed on post-larval densities (Flach, 1996; Van Colen et al., 2013). At higher densities of adult cockles a notable reduction was seen in post-larval density. This indicates there may be density dependence of adult cockle presence being unimportant, beneficial, or negative with its effect on post-larval density. This is likely context dependant. In general cockles are seen as bioturbators and have been shown to increase the erosion of sediment (Andersen et al., 2010; Ciutat et al., 2007). However Donadi et al. (2013) recently showed that dense areas of adult cockles can actually stabilise sediment and increase the microphytobenthos production compared to cockle free or lugworm inhabited sediment. This may explain our results of higher post-larval density in the presence adult cockles. This is speculation as many hypotheses could be proposed to explain this result. For example predation is a key driver of post-settlement survival, and a hypothesis that adult cockles increase habitat complexity and therefore reducing predator foraging efficiency could be proposed. This is why experimental work to identify the causative processes of these field settlement experiments and observations are important.

To test the possible causes of the observations in the plot studies two laboratory studies were conducted. Adult cockles and lugworms may smother post-larval cockles when feeding and burrowing (Rhoads and Young, 1970a; Woodin, 1976), so I tested if this was a source of post-settlement mortality. After the two week aquarium study there was no evidence of increased mortality through direct ingestion or sediment smothering, with high survival in most experimental tanks across the treatments. Survival of the post-larvae across the treatments was on average high. The low survival in some replicates, including the control treatment, may be attributed to poor water quality due uneaten microalgae. This was suggested by comparisons with the adult cockle treatments where significant filter feeding was occurring and variability in survival was much lower. A previous preliminary study that was conducted by confining *C. edule* post-larvae with lugworms in aquaria found no difference in mortality with control lugworm free aquaria (Leedham, 2011). There was an indication that movement of *C. edule* post-larvae increased in the presence of lugworms

however (Leedham, 2011). Observation of *C. edule* post-larvae show that they were highly active and able crawlers when disturbed, and is likely that a post-larva may be able to return to the sediment surface if covered during lugworm defecation or adult cockle crawling activity. Adult bivalves and juvenile *Macoma* can vertically migrate to establish siphon surface contact and survive, from a depth of 2 – 3 times the shell length (Chang and Levings, 1978; Hinchey et al., 2006; Maurer et al., 1981). When investigating mortality of post-larval *Macoma balthica* Hiddink et al. (2002b) found that the presence of lugworms did reduce post-larval abundance in plots and found post-larvae in *A. marina* stomachs. No *C. edule* post-larvae were found in lugworm stomachs. Laboratory feeding experiments did show that direct ingestion by *A. marina* of *M. balthica* post-larvae is possible without alternative 'prey', but with other meiofauna present ingestion of post-larvae did not occur in the laboratory (Hiddink et al., 2002b). Post-larvae of *M. balthica* have been shown to survive complete burial in anoxic sediment for up to 10 days (Elmgren et al., 1986) and Hiddink et al (2002b) concludes for *M. balthica* that direct mortality from sediment smothering caused by bioturbation is unlikely to be a significant cause mortality for bivalve post-larvae, as I do for *C. edule*.

Differences in density observed in field experiment treatments may be due to differences in byssus migration rates. The flume study shows little evidence of differences in the number of cockle post-larvae drifting when in the presence of macrofauna. The drifting rates were generally low (1.3 – 11.3 %) but with some exceptions, particularly in the adult cockle treatment with one replicate having 41.3 % drifting. This experiment would merit further replication and refinement as the lugworms produced no, or very small faecal casts, which indicated little feeding activity was occurring. This may have been a poor representation of a natural bioturbation levels. A longer acclimation period and a higher organic content of the sediment may have stimulated more feeding and therefore representation of natural disturbance regimes. The flume study does not explain the differences seen in densities in the field study, but it illustrates the potential for up to 40% of post-larvae to migrate in a fairly short period of time and change densities post-settlement. Such changes in density therefore cannot be automatically attributed to macrofauna causing mortality. It is established that byssal drifting is an active behaviour in species such as *C. edule* and *M. balthica* (Armonies, 1994b; Hiddink, 2002). If post-larvae are in an unfavourable situation, such as the presence of bioturbators, then they may reject an area through initiating byssal-pelagic migrations with the gamble of finding more favourable habitat. Differences in density between field plots and flume experiments may also occur passively as lugworms and adult *C. edule* have been shown to alter the erodability of surrounding sediment and therefore increase or decrease the resuspension potential

compared to macrofauna free sediment (Andersen et al., 2010; Ciutat et al., 2007; Wendelboe et al., 2013).

4.1 Summary and implications

I observed opposite effects of the presence of lugworms and adult cockles on the density of *C. edule* post-larvae from settlement to two or three months post-settlement. Exclusion of lugworms increased post-larval densities and exclusion of adult cockles decreased the density of post-larvae. The exclusion of adult cockles at small spatial scales and at the natural density of that area resulted in lower densities than in areas with adults. This illustrates along with some previous studies that the interaction between post-larvae and adult *C. edule* is not necessarily a negative one. Depending on the scale at which this positive process or interaction occurs could have an important role to play in a population's recruitment success. It potentially means that depleting an area of adults during cockle fishing, or any other activity, could reduce that areas' recruitment in subsequent settlement events by up to 50%. Sustainability of cockle fisheries are massively dependant on post-larval survival up to the post-winter period which drives recruitment, and so understanding these processes affecting numerical and spatial colonisation success has important management relevance. The presence of macrofauna can play a role in spatial patchiness of post-larval densities and therefore recruitment, however the driving processes remain unclear. From this study mortality from smothering and ingestion or migration due to the presence of macrofauna appear unlikely factors driving these differences, but warrant further investigation. There are many other possible indirect effects of macrofauna presence such as modifying predation risk, either through disturbance of the post-larvae (Flach, 2003) or reducing predator foraging efficiency through habitat complexity. Further work is recommended on identifying indirect effects of species interactions on post-larval abundance, especially predation risk as this is a key post-settlement process controlling recruitment magnitude of *C. edule* and other intertidal bivalves.

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Chapter 5: Aggregated prey and predation rates: Juvenile shore crabs (*Carcinus maenas*) foraging on post-larval cockles (*Cerastoderma edule*)

Timothy Andrew Whitton, Stuart Rees Jenkins, Christopher Allan Richardson, Jan Geert Hiddink

Abstract

Many soft sediment invertebrates show highly patchy distributions within populations across small (centimetres – 10s metres) and large (10s – 100s kilometres) scales, despite relative large scale homogeneity in the physical environment. The common European cockle *Cerastoderma edule* is an example of a patchily distributed species. Predation during the post-larval period is probably critical in determining the recruitment success and potentially the spatial distribution of this species. We investigated if aggregation (being 'patchy') of *C. edule* post-larvae increases survival from predation from juvenile shore crabs *Carcinus maenas* when compared to a uniformly dispersed distribution. We hypothesised this will be caused by interference competition between *C. maenas*. Aquarium based experiments were conducted testing two factors of prey arrangement (aggregated and dispersed) and predator number (one or 5 crabs), and their interaction in controlling prey mortality and predator behaviour. Prey mortality was higher by 30 – 40 % for both predator treatments when aggregated, there was no interaction between factors. This indicates the presence of conspecifics has no more significant influence on foraging efficiency than prey arrangement. An individual crab spent 30% longer searching when prey were dispersed, indicating that prey encounter rates limit foraging efficiency when conspecifics are absent. When multiple crabs foraged together they engaged in antagonistic interactions (8.5 % of the total time) in both prey arrangements with no significant difference, and other behaviours did not explain clearly the difference in prey mortality. These results indicate that juvenile *C. maenas* are able to maintain a higher foraging efficiency on aggregated prey, despite the presence of conspecifics and associated interference competition. From this laboratory predator-prey system there is no evidence that aggregated cockle post-larvae are afforded greater protection from predation than uniformly distributed cockles. Therefore the migratory behaviour of *C. edule* post-larvae that may create or maintain dispersed distributions could be significant for post-settlement survival.

5.1. Introduction

The spatial distribution of many intertidal soft-sediment invertebrates can be highly patchy (Kraan et al., 2009; Olafsson, 1992; Omori and Hamner, 1982). The existence of patchiness within populations indicates that there may be advantages to patchy spatial arrangements for survival (Bergström et al., 2002). The alternative is that the processes creating patchiness are stronger or acting faster than migration and dispersion which will promote random or uniform distributions (Wiens, 1976). Population patchiness on large scales (100s – 1000s km) is often associated with habitat suitability or life history strategies, but the causes for smaller scale patchiness (10s of kilometres – centimetres) is not yet clearly understood.

The processes which control and create patchiness are potentially numerous, technically difficult to identify and rank in terms of their importance from field observations (Butman, 1987; Olafsson et al., 1994). Patchiness in soft sediment intertidal communities is of particular interest as the physical environment appears relatively homogenous over scales of several hundred metres, and yet species patchiness can occur on scales of metres to centimetres (Findlay, 1981; Kraan et al., 2009). In determining the relative importance of pre- and post-settlement processes in structuring soft-sediment communities, Olafsson et al. (1994) concluded that post-settlement processes were of great importance. Predation is thought to be one of the key processes that causes post-settlement mortality, and is unlikely to be spatially homogeneous in its impact (Beukema and Dekker, 2005; Flach, 2003; van der Veer et al., 1998). Any process or behaviour that modifies predation of a prey species spatially may be a causal mechanism for observed patchiness in prey species distributions (Andresen and van der Meer, 2010; Pinn and Ansell, 1993; Richards et al., 1999).

It is well documented that increased prey density increases foraging efficiency of predators, largely demonstrated through functional response studies (Kratina et al., 2009). This is attributed to the reduction in searching time needed to locate prey through increased prey encounter rates. However, prey density does not fully account for the chance of prey encounter rates by a predator. For a given density, or the spatial scale under investigation, the spatial arrangement of prey may vary from highly aggregated, multiple prey patches to uniformly dispersed. The effect of different prey spatial arrangements on foraging efficiency is less well understood than that of density. The ideal free distribution predicts that for the most efficient foraging more predators will be found where prey is distributed and abundance is highest. This infers that predators become patchy or aggregated when prey is patchy, which can potentially change their foraging efficiency (Fretwell and Lucas, 1969; Sutherland, 1983).

For a single predator foraging, the effect of spatially aggregated (or patchy) prey may be similar to that of the functional response with increasing prey density. This will be through

the same process of a higher foraging efficiency achieved through reduced searching time required for prey capture. The presence of conspecifics however can reduce the foraging efficiency of a predator through interference competition (DeLong and Vasseur, 2011). Interference competition between predators can result in behaviour such as increased vigilance, displacement, fighting and even cannibalism. Therefore foraging efficiency can be reduced through time spent in these interactions instead of searching for prey (Skalski and Gilliam, 2001), this can increase the survival chances of prey (Clark et al., 1999b; Kratina et al., 2009; Nachman, 2006).

The intensity (difference between high and low density areas) and lag of patches (distance between patches) can have important implications for population stability (Clark et al., 2000; Oaten, 1977) and vulnerability to predation of prey (Nachman, 2006; Sutherland, 1983; Yasuda and Ishikawa, 1999). Studies on the role of prey patch intensity and lag on predation efficiency in a crab-bivalve system in Chesapeake Bay, USA by Clark et al. (1999a; 1999b; 2000) and Hines et al. (2009) have demonstrated that the blue crab *Callinectes sapidus* foraged most efficiently on its infaunal bivalve prey when they were in two patches rather than in one patch. Furthermore, foraging efficiency was shown to be highest when prey patches had an intermediate lag (7 m apart) of the range tested. They reasoned that this was due to the balance between the positive effects of aggregative behaviour toward other crabs increasing prey detection, with antagonistic interactions between crabs reducing their foraging efficiency and so distancing individuals from one another. There is some evidence that the spatial arrangement of prey, independent of mutual interactions, can change foraging behaviour and subsequent prey survival. *Carcinus maenas* has been shown to reduce its handling time per prey item when feeding on *Mytilus edulis* clumps as a higher energy yield is gained (Burch and Seed, 2000). This occurs because it is time consuming to completely consume all the mussel flesh and glean the flesh from the shells before a new prey item is detected and attacked. By contrast individually presented mussels often had their flesh completely removed and eaten, which would have increased the handling time for a comparatively lower net energy gain. We hypothesise that for a given overall density, aggregated prey will have a higher survival rate than uniformly dispersed prey, when foraged upon by multiple predators. We expect this because an increased occurrence of interference competition is likely to occur as predators feed on spatially confined prey, thus reducing their foraging efficiency when compared to dispersed prey. The outcome is likely the creation of a refuge from predation and the maintenance of patchiness of a prey species.

The common cockle *C. edule* is an ecologically (Flach, 1994) and commercially important (Hicken, 2008) species on intertidal sand and mud flats on North western European shores. The key period for the successful recruitment and establishment of

juvenile bivalves is during the early post-larval benthic stage (0.35 – 5 mm shell length) (Armonies, 1996; Hiddink and Wolff, 2002). The monitoring and management of *C. edule* is made more difficult by its highly variable inter-annual recruitment into fisheries (defined as ≥ 20 mm shell length in UK fisheries) (Beukema et al., 2010), but also its highly patchy distribution within populations. Post-larval *C. edule* settle onto north eastern Atlantic shores during early summer in high densities and show patchiness in their distribution within the first year following settlement (Armonies, 1996; Bouma et al., 2001; Seed and Brown, 1977). The processes creating this patchiness are not clearly understood yet are potentially important for informed fisheries management, and determining the numerical abundance and structure of populations. If such processes are understood then predictions may be made on the effect of natural or anthropogenic perturbations on a population.

The post-larval stage is when *C. edule* is most vulnerable to predation from juvenile brown shrimp *Crangon crangon* (maximum length of 24 mm) and juvenile shore crabs *Carcinus maenas* (carapace width up to 20 mm). This is largely due to the coincidental appearance of high densities of these predators with larval settlement and maximum post-larval densities (Beukema, 1991; Olafsson et al., 1994; Strasser and Günther, 2001). Employing exclusion caging experiments Flach (2003), Reise (1978; 1985b) and Richards et al. (1999) demonstrated that epibenthic juvenile crustaceans reduce the survival of *C. edule* post-larvae over small scales ($\approx 1 \text{ m}^2$) and on large whole shore population scales ($\approx 50 \text{ km}^2$) (Beukema and Dekker, 2005). Due to the significance of predation on *C. edule* and other bivalve species such as *Macoma balthica*, it is important to establish how predation varies spatially and to understand its role in creating or maintaining juvenile and adult population patchiness. Juvenile and adult *C. maenas* are known to engage in interference competition from increased vigilance to cannibalism which can reduce foraging efficiency (Moksnes, 2004a; Smallegange et al., 2006). Smallegange and van der Meer (2007) found that foraging was greatly reduced when conspecifics were of an equal size and they also observed that crabs mostly avoided competitors when they were handling prey. Moksnes (2004b) and later Smallegange et al. (2010) demonstrated the importance of mutual interactions amongst juvenile *C. maenas* in controlling their distribution. They observed both in field and laboratory experiments that juvenile crabs actively migrate across open sand from patches of shelter and available food to avoid interference competition with conspecifics. Due to the strength of interference competition in *C. maenas* and our observations of spatial patchiness of *C. edule* we investigated how they may interact to influence *C. edule* post-larval mortality. We investigated the hypothesis that prey mortality will be lower for aggregated compared to uniformly distributed cockle prey due to interference competition. This was tested experimentally by creating aggregated and

uniformly dispersed *C. edule* post-larvae arrangements in laboratory aquaria, and comparing the mortality of *C. edule* in the presence of single or multiple foraging juvenile *C. maenas*.

5.2. Methodology

5.2.1 General outline

Experimental treatments of aggregated and dispersed cockle post-larvae were set-up and exposed to a single crab or five crabs in small aquaria (Fig 5.1). Each treatment (Aggregated x 1 crab, Dispersed x 1 crab, Aggregated x 5 crabs, Dispersed x 5 crabs) was replicated 5 times and the experiments for the four treatments, conducted for 25 minutes, were randomly distributed over an 8 day period.

5.2.2 Experimental animal collection and maintenance

5.2.2.1 *Cerastoderma edule* post-larvae

Post-larvae of *C. edule* were collected from the Dee estuary, UK 10 days prior to the beginning of the experiment by scraping off the top millimetres of sediment and sieving it coarsely over a 0.5 mm sieve *in-situ* to remove most of the sediment. In the laboratory the collected post-larvae and sediment were washed through two graded sieves, a 1mm sieve and then a 0.5 mm sieve and any retained material was then sorted under a dissection microscope and the *C. edule* post-larvae separated from the other fauna and sediment. Identification of *C. edule* post-larvae was aided by the description and illustrations in Lebour (1938) which showed representative size classes of recently settled *C. edule* post-larvae. Initial feeding experiments showed that *C. maenas* juveniles (10 – 20 mm carapace width) readily consumed post-larvae of this size range ($\leq 1\text{mm}$). Post-larvae were maintained on sieves in flow through aquaria supplied with ambient seawater and the illumination in the aquarium matched to the natural ambient photoperiod. Natural food present in the seawater was supplemented by the daily addition of microalgae *Isochrysis galbana* (T-Iso) culture. The shell length of the post-larvae used in the experiments ranged between 1600 & 3000 μm (mean $2413 \pm 20.3 \mu\text{m}$, ± 1 Std error). This was a suitable size class to represent recently settled post-larvae (Huxham and Richards, 2003).

5.2.2.2 *Carcinus maenas* juveniles

Juvenile *C. maenas* (carapace width 10 – 17mm; mean 13.8 ± 2.1 , ± 1 Std. error) were collected from local intertidal shores 3 days before the experiment began. The size of crabs used did not differ significantly between treatments for single and multiple crab experiments. Juvenile *C. maenas* used were representative of 0-group crabs that are present on the intertidal shore and occur at the same time as post-larval *C. edule* are settling (Jensen and

Jensen, 1985; Klein Breteler, 1975). Crabs were maintained in several communal tanks supplied with flow through ambient seawater at a mean temperature of 17.7 °C. To prevent cannibalism and limb damage, aquaria were provided with sediment for burial, shelters and only similar sized crabs were housed together (Moksnes et al., 1998; Moksnes et al., 1997). During holding, the crabs were fed with crushed post-larvae and adult *C. edule* flesh. Twenty four hours before the start of an experiment crabs with all limbs intact were transferred from the communal aquaria to individual aquaria and deprived of food for 24hrs to ensure a consistent hunger level in all the experiments. After a feeding trial was



Figure. 5.11. Photographs showing experimental tank setup and behavioural film screenshots. Burrowed *C. edule* post-larvae can be seen as black specs in the sediment. (A) Aggregated arrangement of *Cerastoderma edule* post-larvae created in experimental treatments (B) Uniformly dispersed (grid) arrangement of *C. edule* post-larvae used in experimental treatments. (C) Screenshot from a film of five juvenile *Carcinus maenas* during an aggregated prey experiment. The crab on the furthest right can be seen feeding within the aggregated *C. edule* post-larvae area. (D) Screen shot from a film of five juvenile *C. maenas* during a dispersed prey experiment.

completed the crabs were monitored for 48 hours to ensure that they were not in proecdysis (Period of preparation before molting), as feeding stops during this process (Sneddon et al., 1997). All crabs were only used once in the experiments and then released. Out of twenty experiments two were repeated because the crabs moulted afterwards, indicating they were in proecdysis.

5.2.3 *Experimental aquaria set up*

All experiments were conducted using eight square plastic aquariums (30 X 30 cm) containing seawater to a depth of 18 cm at a temperature of 17 – 18 °C. The sediment used was purchased kiln dried sand, sieved to obtain a size fraction of 120 µm - 250 µm, rinsed before and after use to remove any residue or dust and distributed and placed at a depth of 10 mm. *Cerastoderma edule* post-larvae were added to the experimental tanks 6 hours before the start of an experimental trial to allow sufficient time for burial into the sediment and acclimation to the aquarium conditions. Post-larvae were recovered following the feeding trials by sieving all of the sediment over a 355 µm sieve. For both spatial arrangement treatments, 42 post-larvae were used to simulate a density of 500 m⁻² which is similar to local post-larval cockle densities in mid to late summer in North Wales, UK (pers ob.). At the same time as the post-larvae were added 6ml of *Isochrysis galbana* (T-Iso) phytoplankton culture was also added, so that the post-larvae had access to food and would extend their siphons and feed. Gentle aeration was supplied during the 6 hour acclimation period but removed during experiments to allow filming. All experimental trials were conducted in the dark as juvenile crabs feed more actively under these conditions (Rangeley and Thomas, 1987). When artificially illuminated crabs have been observed to seek refuge during daylight. The experiments were filmed using a SONY Handicam in 'Night Scene' mode with two dim red light sources providing illumination. The footage was recorded for later playback and behavioural analysis.

5.2.4 *Experimental treatments and procedure*

Aggregated prey arrangements were established by placing a tube (Ø 64 mm) gently onto the sediment surface randomly, using a visualized 3 x 3 grid (Figure 1A). Forty two post-larvae were introduced by pipette into the tube, and after 10 seconds the tube was gently removed from the water. A 64 mm diameter circle was used because it sufficiently aggregated the post-larvae in relation to the rest of the tank (i.e. 3.8% of the total tank area), but allowed sufficient room for the post-larvae to bury. The post-larvae burrowed into the sediment where they remained and did not crawl out of the circle during the 6 hour acclimation period (Figure 1A). Dispersed prey arrangements were established by pipetting

individual post-larvae evenly on a 6 x 7 grid (Figure 1B). The grid points were pre-marked by gently touching the sediment surface with a piece of wood with 7 nails hammered through it. Post-larvae were not observed to crawl from their original dispersed position during the acclimation period but would burrow into the sediment. For both prey arrangement treatments *C. edule* post-larvae were never placed closer than 2 cm to the tank edge, as pilot observations indicated that crabs would often crawl around the edges of the tank which may have biased results. A control procedure was also established where 5 aggregated and 5 dispersed prey treatments were conducted over seven hours with no predators present. This was undertaken to establish that the prey spatial arrangements did not cause different mortality or retrieval efficiencies of the *C. edule* post-larvae during the 25 minute experimental period. Any mortality in both experimental treatments can be confidently attributed to predation since in both prey arrangements only one post-larva was not recovered from one treatment and one died in the other.

All experiments started when the crabs were introduced into the aquaria and terminated after 25 minutes with their immediate removal. Upon removal the carapace widths of the crabs were measured with vernier callipers to 0.1mm. Post-larvae were then removed from the sediment (detailed above) and counted. The period of 25 minutes was chosen after several pilot experiments as a balance between ensuring sufficient mortality to determine treatment effects and preventing excessive prey depletion.

5.2.6 Data analysis

The proportion of cockle post-larvae eaten were arcsine transformed, the data checked for homogeneity of variance using Levene's Test of Homogeneity of Variance and a 2-way ANOVA conducted. Post-hoc Tukey's pairwise comparisons were conducted to further investigate differences between treatments. Films of 4 experiments from each treatment (16 films in total) were reviewed to assess crab behaviour (Figure 5.1C and 5.1D). In the multiple crab treatments a single focal crab was randomly chosen and its behaviour was noted resulting in a total of 16 behaviour data sets. During playback every time a new behaviour began the time was noted. The recorded behaviour categories (following the classifications in Smallegange et al. (2006)) were searching (walking over and probing of the sediment), handling (time a cockle was handled and consumed), avoiding/displacing another crab (changing movement to actively displace/avoid another crab), fighting (grasping or being grasped with the chelae of another crab) and resting (no movement or buried in sediment). From these timings, duration spent in each behaviour category was calculated within an experimental trial, and the proportional duration for the whole experiment calculated. All proportions were arcsine transformed before 2-way (prey arrangement and predator number as fixed factors) and 1-way ANOVAs (treatment as factor and proportion of time in behaviour

as dependant variable) were conducted with the data with homogeneity of variance. In addition the time taken to consume a single prey item was recorded from the same films with 26 observations from random crabs for each treatment (104 observations in total). A Kruskal Wallis and post-hoc Tamhane's T2 pairwise test were used to investigate significant differences of consumption time (seconds) per prey item (dependant variable) between treatments (factor). All means are reported ± 1 standard error.

5.3 Results

When crabs were moving within the experimental aquaria they showed a tendency to remain close to the edges facing inwards. The main methods of searching for the post-larvae prey were via the walking legs while walking and through probing the sediment with the chelae and the first two pairs of walking legs. Once found post-larvae were brought straight to the maxillipeds and consumed. Flesh was rarely observed on the recovered shell fragments after experiments. When a single crab was foraging with aggregated prey it would often spend the initial period searching until it found the aggregated prey zone. Once the first prey item in the aggregated prey zone was encountered the crab often remained there for the duration of the experiment feeding on consecutive post-larvae, which it detected almost instantaneously or even during feeding on another post-larvae (Figure 5.1C). When a crab was handling a prey item, it would avoid any contact with other crabs by actively moving away from other individuals. This was most noticeably observed in the aggregated prey treatment. When a crab had found prey in the aggregated prey zone it would remain feeding there. However, if approached by another crab it would rapidly move out of the area and continue to feed elsewhere in the experimental aquarium. When the prey item was consumed the crab would immediately begin searching again. Crabs did not appear to remember where the aggregated prey patch was located.

5.3.1 Mortality of the *Cerastoderma edule* post-larvae prey

Compared to dispersed prey, aggregated prey treatments had a significantly higher cockle post-larvae mortality (2-way ANOVA, $F_{1,16} = 30.27$, $P = <0.001$), which was on average 34.4 % higher across both predator treatments (Figure 5.2). The increased number of predators also predictably increased mortality (2-way ANOVA, predator number $F_{1,16} = 16.44$, $P = 0.001$) (Figure 5.2), although a fivefold increase in predator number only led to a twofold increase in mortality. There was no significant interaction between prey arrangement and predator number (2-way ANOVA (Arrangement x Predator number $F_{1,16} = 1.498$, $P = 0.239$)).

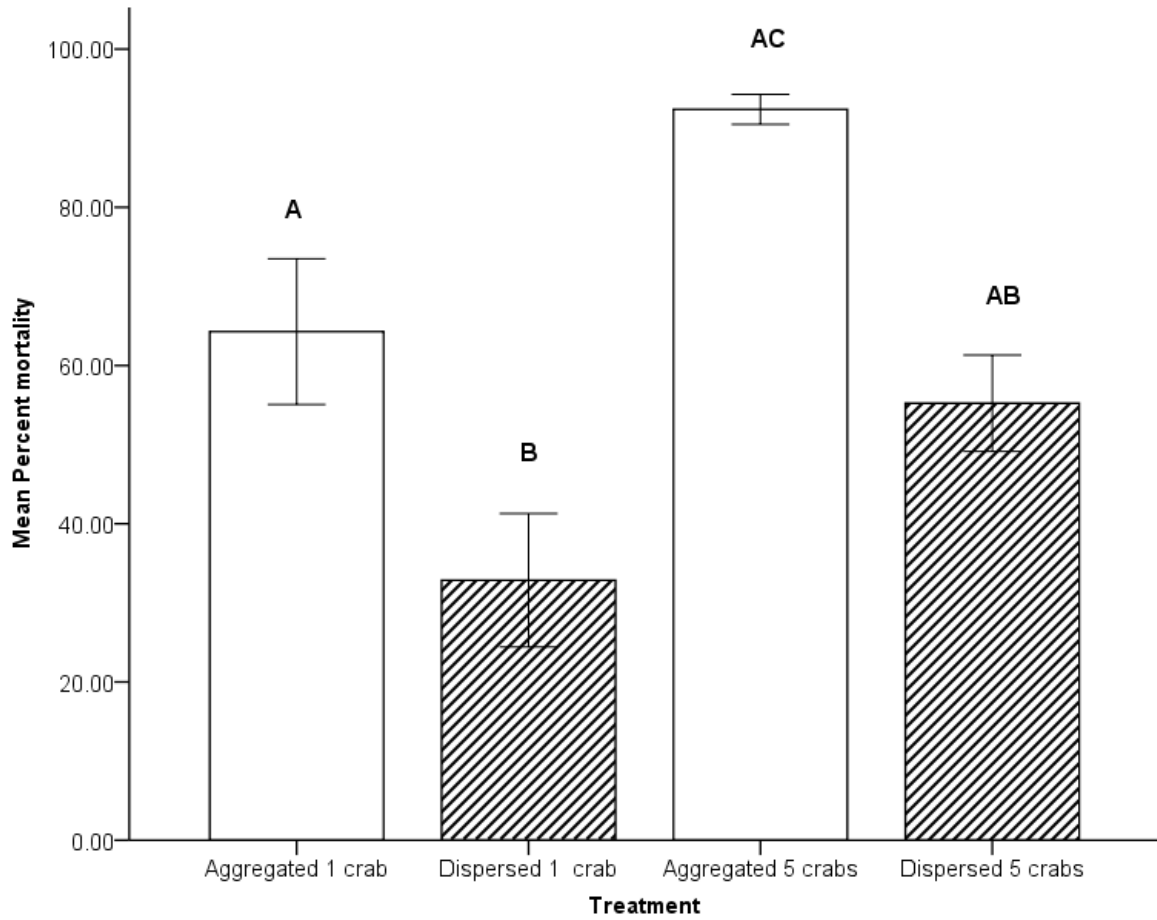


Figure. 5.2. Mean percent mortality of *Cerastoderma edule* post-larvae for the 4 experimental treatment combinations of prey arrangement and number of juvenile *Carcinus maenas*. When bars do not share a letter they are shown to be significantly different by Tukey's pairwise comparisons. Error bars are ± 1 standard error.

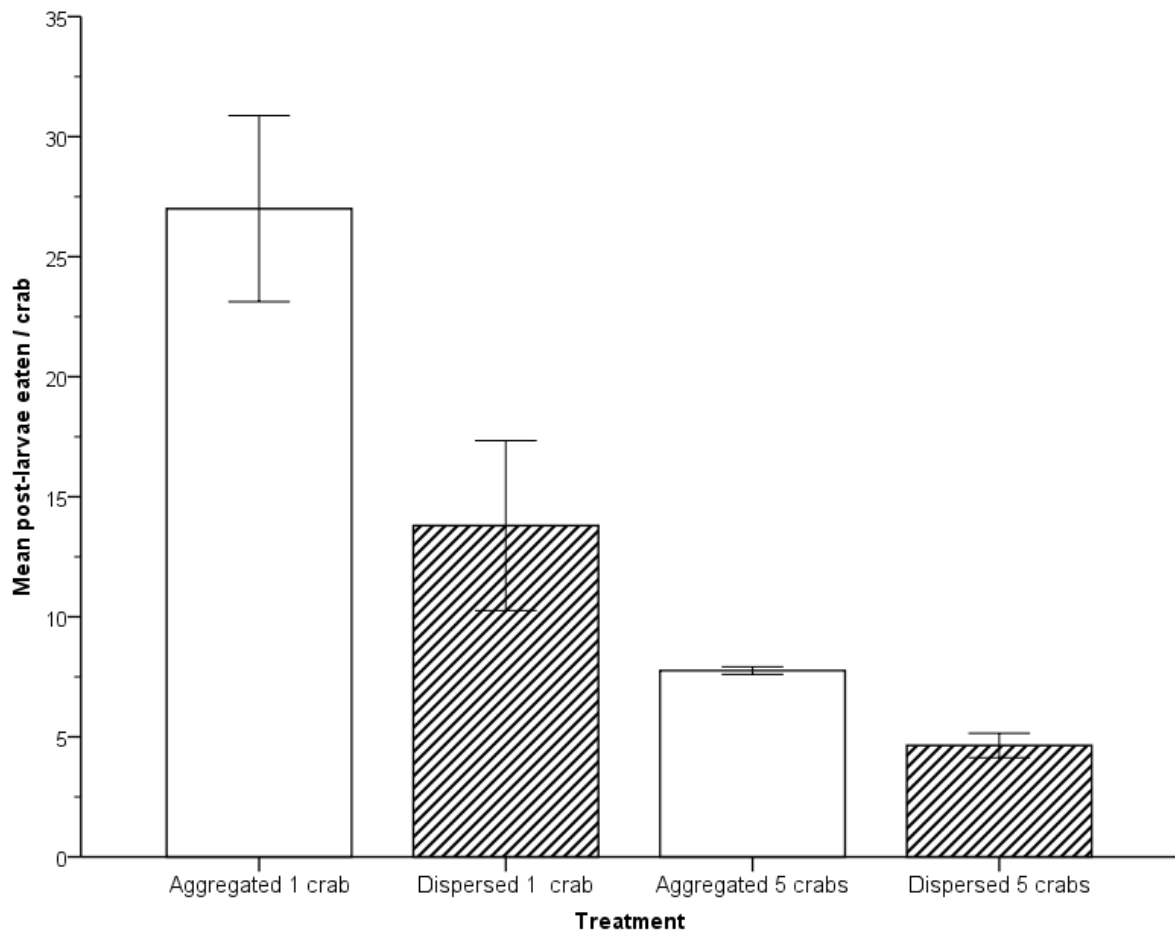


Figure. 5.3. Calculated mean juvenile *Carcinus maenas* per capita consumption of *Cerastoderma edule* post-larvae in each of the prey arrangement and predator number treatments. Experiments lasted 25 minutes. Error bars are ± 1 standard error.

The total mortality of *C. edule* post-larvae for a prey arrangement treatment with five crabs was not five times higher than with a single crab, as might have been expected. In fact it was only 20 – 30 % higher (Figure 5.2). Mortality of the aggregated prey with multiple crab treatments was >90% for 3 of the replicates, suggesting exploitative competition is highly likely to have occurred through prey depletion in these replicate experiments. The mean calculated per capita consumption of *C. edule* post-larvae for a lone crab feeding on aggregated prey was 27 ± 3.9 during a 25 minute experiment (Figure 5.3), amounting to an approximate feeding frequency of one post-larvae located and eaten per minute. For the dispersed prey the per capita consumption of *C. edule* post-larvae was 13.8 ± 3.5 indicating that prey capture was almost doubled when prey was aggregated for an individual crab foraging. The lowest calculated per capita consumption of *C. edule* post-larvae was in the dispersed prey treatment where five crabs were feeding, these only consumed a calculated mean of 4.6 ± 0.5 during the 25 minute experiments. The calculated per capita consumption

of *C. edule* post-larvae was 71.3 and 66.8 % lower for dispersed prey when conspecific predators were present, compared to a lone forager respectively (Figure 5.3).

5.3.2 *Carcinus maenas* predator behaviour

Two-way ANOVAs showed that prey arrangement and predator number interacted significantly on the proportion of time spent in searching and handling behaviour (Table 5.1).

Table 5.4. 2-Way ANOVA results of juvenile *Carcinus maenas* proportion time spent in different behaviours between the different treatments. All proportions arcsine transformed except where [†] = Log transformed . * denotes a significant result ($P \leq 0.05$).

	Searching		Handling		Resting [†]	
	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>	<i>F</i>	<i>P</i>
Prey						
arrangement	7.742	0.017*	5.544	0.036*	0.06	0.814
Predator						
number	10.667	0.007*	22.013	0.001*	2.272	0.175
Interaction	7.168	0.020*	7.968	0.015*	0.104	0.756

When prey were aggregated a single crab spent an average of 30 ± 6.6 % of the time searching for prey, compared to a significantly higher (Table 5.1) $61.1 \pm 3.4\%$ in the dispersed prey treatment (Tukey's pairwise comparison, significance level = Dispersed 5 crabs > Aggregated 5 crabs > Dispersed 1 crab) (Figure 5.4). Total handling time was significantly higher (Table 5.1) for a single crab feeding on aggregated prey (Tukey's pairwise comparison, significance level = Aggregated 5 crabs > Dispersed 5 crabs > Dispersed 1 crab) (Figure 5.4).

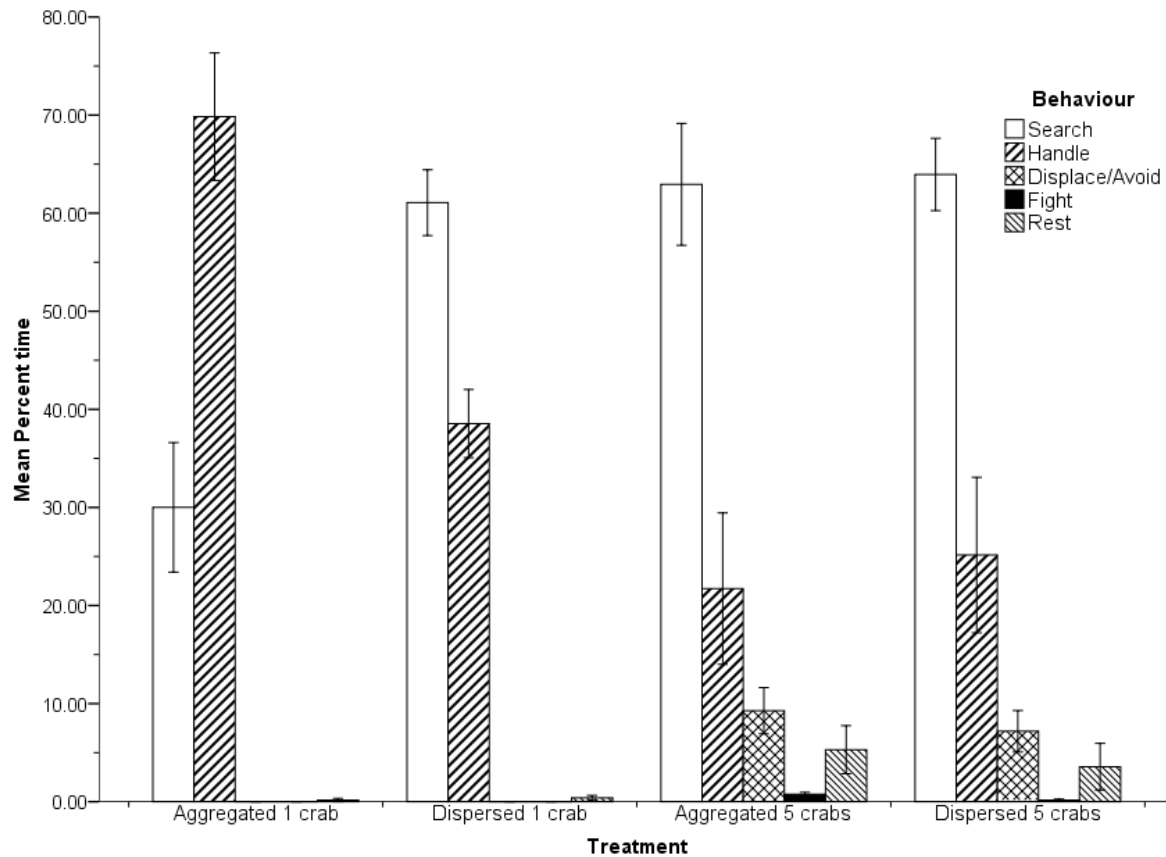


Figure. 5.4. Mean percent time spent in different behaviour of juvenile *Carcinus maenas* from each experimental treatment of *Cerastoderma edule* post-larvae arrangement and number of crabs. Error bars are ± 1 standard error.

When multiple crabs foraged together there was no difference in the total time spent searching for aggregated and dispersed prey (63 and 64% respectively). Similarly there was no difference in handling time between the same treatments. There was also no significant difference in the time spent in avoidance and displacing behaviour for the aggregated and dispersed treatments (10 and 7% respectively) for the same treatments. Crabs spent 10.05 ± 2.35 and 7.35 ± 2.11 % of their time in displacing and avoidance behaviour in the aggregated and dispersed treatments respectively, although this was not significantly different. Fights between crabs accounted for <1% of their activity in both prey arrangements. However, a significantly longer period was spent fighting in the aggregated treatments ($F_{1,6} = 7.574$, $P = 0.033$), whilst <1% of the time was spent resting in all treatments. The time taken to consume individual prey items indicated that crabs in the '5 crabs X dispersed prey' treatment took 21.13 seconds longer to eat a *C. edule* post-larvae than crabs in all other treatments (Figure 5.5) (Kruskal Wallis test, $\chi^2 = 11.3$, $P = 0.01$; Tamhane's T2 pairwise test, significance level = Aggregated 5 crab > Dispersed 1 crab > Aggregated 1 crab).

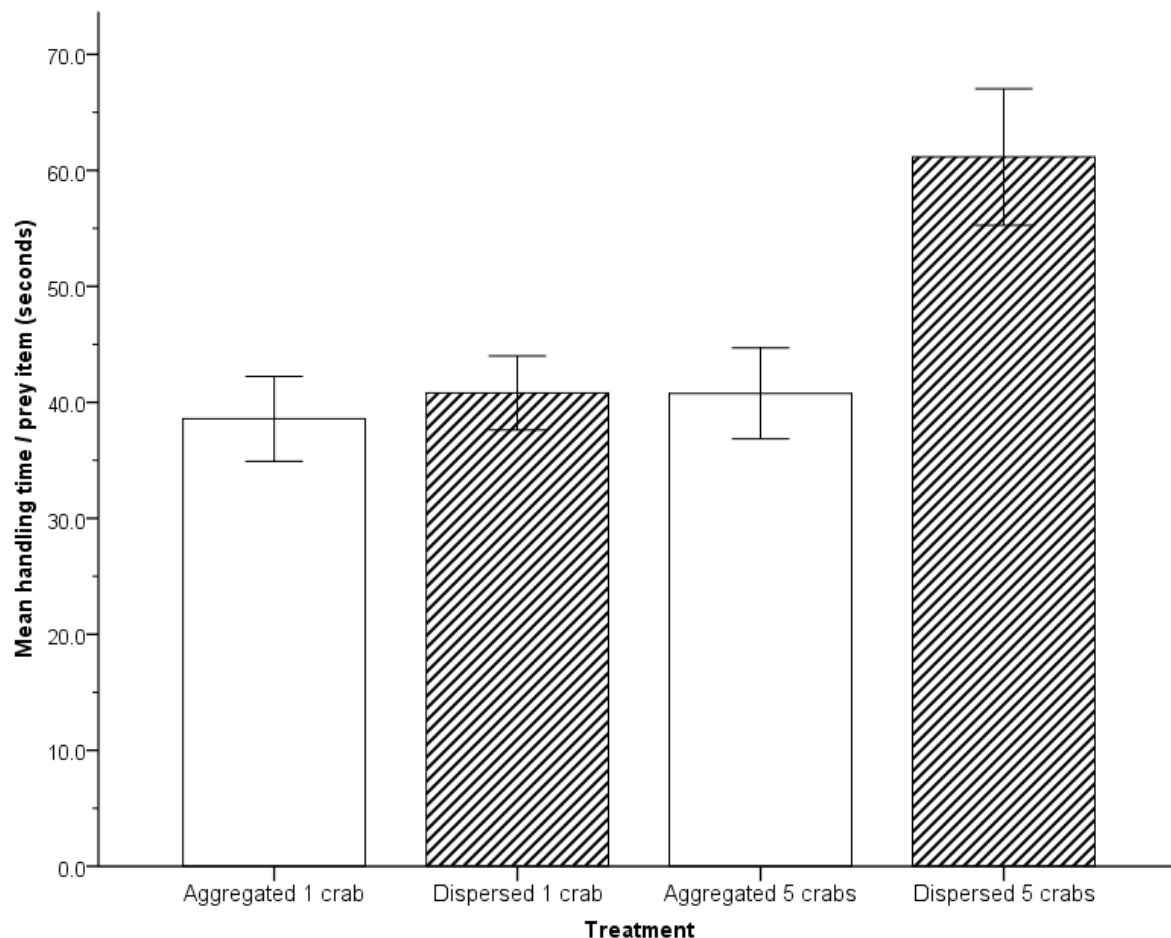


Figure. 5.5. Mean time taken (seconds) to consume individual *Cerastoderma edule* post-larvae by juvenile *Carcinus maenas*. Aggregated and dispersed refer to the arrangement of *Cerastoderma edule* post-larvae, and the number of crabs were a single individual or 5 crabs. Error bars are ± 1 standard error.

5.4. Discussion

This study aimed to investigate how the spatial arrangement of prey, may alter the risk of predation in the presence of single and multiple predators. We hypothesised that aggregated prey would have higher survival than dispersed prey due to antagonistic interactions between multiple predators increasing with spatially confined prey. This would be shown by an interaction between the number of predators and the spatial arrangement of prey. Contrary to our hypothesis this study found there was no interaction between these factors. Higher mortality in aggregated prey was found for both single and multiple crabs foraging. However, for a single crab foraging the results were explained by a significantly higher percentage of their time spent searching for the *C. edule* post-larvae when the prey were uniformly dispersed compared to the aggregated prey arrangement. It was predicted that interference competition would be higher for multiple crabs feeding on aggregated prey and therefore reduce foraging efficiency, but this was not observed. By contrast from the

mortality data it appeared that multiple crabs were able to feed more efficiently on aggregated prey, although behavioural observations did not clearly explain this pattern.

The behavioural data clearly explained the post-larval mortality results when a single crab was foraging. Searching was the dominant behavioural activity when foraging on dispersed prey, but prey handling dominated with aggregated prey. This indicates that prey detection (expressed in 'searching time') was limiting the food intake when prey were evenly dispersed throughout the tank, and therefore increased the chance of *C. edule* post-larvae surviving. This relationship was predicted in our experiment. However in nature juvenile *C. maenas* and many other predators rarely forage in complete isolation from conspecifics (Jensen and Jensen, 1985; Moksnes, 2004b).

When five crabs were foraging on *C. edule* post-larvae, cockle mortality was also higher in the aggregated prey treatments. However differences in the searching and handling time did not clearly explain this difference. The likely occurrence of exploitative competition in several of the aggregated prey experiments through near prey depletion is likely to have biased the behavioural data. Inevitably searching time would have increased as crabs searched for a rare resource, which was not the case in the other replicates. In this way a lower total searching time for aggregated prey may have been masked by prey depletion. Due to the burrowing behaviour of *C. edule* prey replacement similar to that used in other studies to remove exploitative competition (Smallegange et al., 2006) was not a viable option for the crab-cockle post-larva predator-prey system.

In the aggregated and dispersed prey treatments the juvenile *C. maenas* lost a similar amount of searching and handling time due to interference behaviour (7 - 10% of the total time). This is contrary to the prediction of a much longer time spent in antagonistic interactions for the aggregated treatment. There are two possible reasons for this. Firstly crabs avoided interactions when handling prey by 'take-away foraging', a term coined by Smallegange et al. (2010) to describe the observation that crabs carried prey items away from optimal prey patches to distance themselves from another crab. This behaviour may reduce the effect of the prey resource being aggregated. This behaviour will decouple the crab distribution from its prey-attack distribution, and therefore does not fit the assumption of the 'ideal free distribution' where predator distributions reflect prey profitability (Fretwell and Lucas, 1969; Smallegange et al., 2010). In this way predatory crabs may reduce the risk of kleptoparasitism, injury through fights and cannibalism during a particularly vulnerable period while handling food (McDonald et al., 2001; Moksnes, 2004a). This 'take-away foraging' can be assumed to have a much less energetic loss than losing food items or fighting, despite having to search again for prey in a different and possibly sub-optimal location. This behaviour observed in the laboratory suggests that retaining and consuming a food item is of greater benefit than defending a prey attack location. The consequences of this will depend

on the size of the prey patch and patch lag. If prey patches are large, or the lag is small, then this behaviour may have little impact on searching time for new prey. In this study the barrier of the experimental aquarium walls would have potentially reduced the disadvantage of this behaviour. The walls retained the crabs near to the prey patch and so even after 'take-away foraging' the crab would likely relocate the original prey attack location. This would not occur in nature and may have lessened the impact of interference competition in the aggregated treatments. Moksnes (2004b) showed in a mesocosm and field study that juvenile *C. maenas* will migrate from an area with abundant shelter and food, to avoid interference competition with other juvenile crabs. The observations in this study and those already documented show the importance of interference competition in shaping juvenile *C. maenas* behaviour and distribution.

The second explanation of no difference in antagonistic behaviour, is that the predator number was possibly too high for the experimental aquarium size. This would result in antagonistic interactions being already high for the dispersed treatment, creating similar encounter rates with other crabs irrespective of prey arrangement. This would potentially mean that the aggregated arrangement of prey would have little impact on the probability of antagonistic interactions. The issue of scale is a common problem in laboratory based experiments and is an important factor in interpreting results in the context of the natural phenomenon under investigation (Thrush, 1999; Thrush et al., 1997). However, due to the clear and significant differences in mortality between treatments in this study, this did not hide the key process determining foraging efficiency. Despite every effort to avoid exploitative competition as the result of prey depletion in this study, which can skew results of predation studies (Goss-Custard, 1980; Smallegange et al., 2006), mortality of *C. edule* post-larvae was >90% in three of the aggregated experiments when five crabs were foraging. This means exploitative competition will have taken place in these replicate experiments. This may have led to a higher searching time in multiple crab aggregated prey treatments than would have been expressed if more prey was present or had been replenished. It would have also reduced the estimated number of *C. edule* post-larvae consumed for the treatment. Again this does not compromise the result of higher mortality in the aggregated prey treatment. However, it does mean the total proportion of time spent in behaviours and per capita post-larvae consumed between aggregated and dispersed prey for multiple crabs cannot be directly compared with confidence. This makes understanding the causal processes of the observed difference in cockle post-larvae mortality between the two spatial arrangements difficult in this study.

The numbers of *C. edule* post-larvae eaten by juvenile *C. maenas* in this study were high when compared to previous studies. In studies conducted by Jenson and Jenson (1985) and Mascaro and Seed (2001) juvenile *C. maenas* feeding was calculated at 6 and 7

cockles day⁻¹ respectively. However when the mortality data for a single crab feeding on aggregated prey from this experiment is scaled up to 12 hours, (representing the daily high tide in the UK when predation is likely to occur) up to 777 cockle post-larvae could potentially be consumed. This figure, however, does not account for satiation, but nevertheless it illustrates the potential impact of juvenile crabs to significantly influence post-settlement mortality of cockles.

With multiple crabs the observation of a longer consumption time per prey item with dispersed prey could be seen to correlate with observations from other studies (Burch and Seed, 2000; Hughes and Elner, 1979). These studies observed that the detection of another prey item during feeding would decrease the time spent gleaning and consuming that prey item. However, this was not found with a single crab feeding in both prey treatments, which had similar consumption times to that of multiple crabs feeding on aggregated prey. With the data available it is difficult to explain why the presence of conspecifics did not change the consumption time of crabs feeding on aggregated prey, despite interference competition.

This study shows the importance of understanding predator behaviour when investigating how the spatial distribution of prey may alter chances of survival from predation. Interference competition clearly occurred, but the spatial distribution of prey and its effect on prey encounter rates was the dominant factor in determining foraging efficiency of the crabs. The ability of bivalve post-larvae such as *C. edule* to migrate post-settlement has often been cited (Armonies, 1994b; Huxham and Richards, 2003) as a strategy to colonise unoccupied sediment, re-colonise areas after predation or if another process has removed individuals. In light of our results from this simple laboratory system, the ability for *C. edule* post-larvae to maintain a dispersed population distribution through migration may have significant advantages in survival against post-settlement epibenthic predation. From the results of this study aggregation does not provide a refuge from *C. maenas* predation due to interference competition. The mobility and avoidance behaviour of juvenile *C. maenas* during feeding ensures they are able to exploit aggregated prey more efficiently than uniformly dispersed prey on the scale of this study, despite interference competition. This further supports that interference competition cannot be generalised as prey dependant (only prey availability is important), or ratio dependant (ratio of predator to prey) as discussed by DeLong and Vasseur (2011). Predators will often show complex behaviours in response to, or even to mitigate against as suggested in this study and by Smallegange et al. (2010), interference competition. Interference competition occurs on small spatial scales (Thrush, 1999), and so the findings of this study are valuable in understanding how patchiness in prey distributions can affect prey survival. We can reject, for a simple *C. edule* and *C. maenas* predator prey laboratory system, our hypothesis that aggregation (or patchiness) has a positive feedback in survival against predation. However juvenile *C. maenas* have many

other alternative prey species and post-larval *C. edule* have several other highly abundant predators, such as brown shrimp *Crangon crangon* and the common plaice *Pleuronectes platessa*. These other predators may have different mutual interactions and respond differently to prey aggregation when foraging, with the complication that alternative prey species are often available to these predators. To understand patchiness and its impact on foraging at natural scales (Thrush, 1999) more studies are needed. Particularly combined laboratory and field investigation using multiple patches (Oaten, 1977) informed by detailed spatial, temporal and behavioural information on prey and predator species. This knowledge will help move us to understand better spatial distributions of species and their causal processes.

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Chapter 6: General discussion

6.1 Introduction

A commonly observed feature in common cockle populations, and many other soft sediment invertebrates, is high spatial variability in density within populations. The spatial carrying capacity can appear underutilised by these species and the reasons for these observations are poorly understood (Armonies and Reise, 2003). Understanding processes controlling the spatio-temporal dynamics of populations is therefore a key objective in ecological research (Sutherland et al., 2013). The timing and processes driving patchiness in *Cerastoderma edule* populations were unclear and have relevance for the common cockle's ecology and management as an exploited species. This PhD has sought to identify processes which create and/or maintain within-population spatial patchiness of *Cerastoderma edule* (Fig. 6.1). It focused on the post-larval stage from larval settlement in spring through to post-winter, as this is currently understood as the critical period for determining recruitment strength. The approach of this PhD has been to 1) observe settlement patterns and how they change over time over both large and small spatial scales (Chapters 2 and 3) and 2) experimentally test the processes that may create and/or maintain the patterns observed in the field (Chapters 4 and 5) (Fig. 6.1).

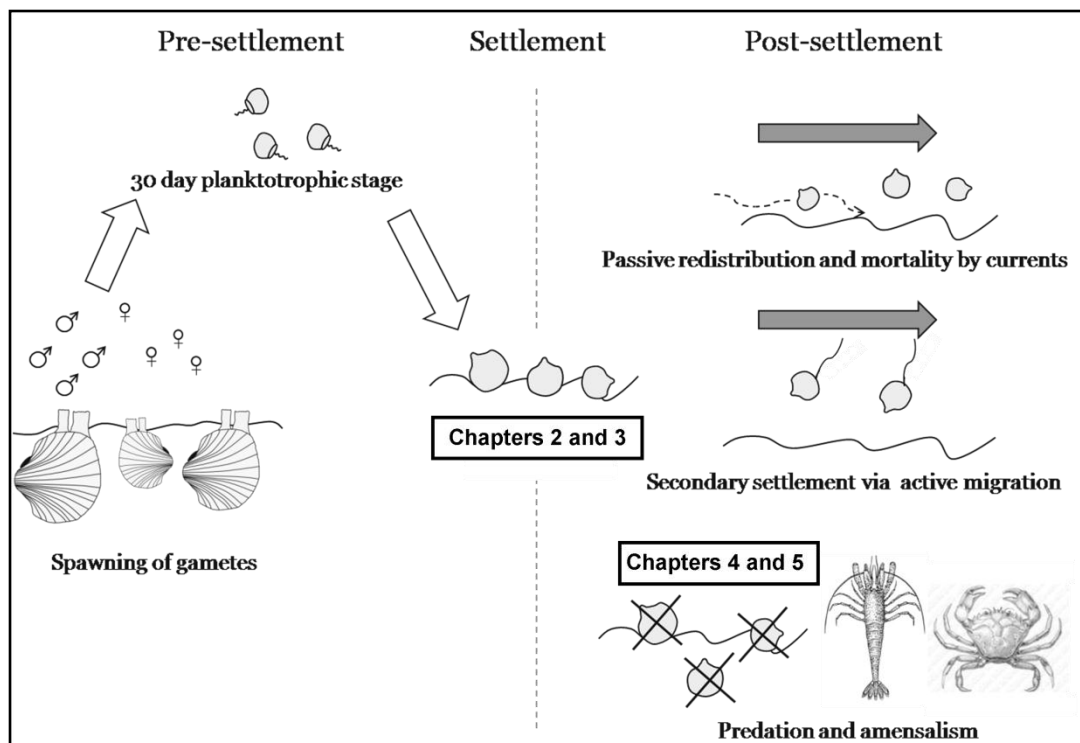


Figure 6.1. The main pre- and post-settlement processes which can affect the observed distributions of *Cerastoderma edule* post-larvae in soft sediments. Chapters of this PhD which address certain aspects are indicated.

6.2 Spatio-temporal patterns in post-larval density

The first main objective of this thesis was to observe and quantify, through geostatistical techniques, the development of within population spatial patterns from settlement to post-winter. Post-larval density of *C. edule* over time had been studied before, but not through spatially explicit techniques with high temporal replication. This observational work examined if spatial patchiness is created at settlement, post-settlement or even later during the juvenile or adult stage, to help identify what the key time periods or processes are in creating patchiness.

In Chapter 2 over a large scale (100s of metres) the main finding was that initial settlement was spatially restricted in areas of the low-mid to low shore at Traeth Melynog, compared to the more widely distributed adult population. After this highly focused and patchy settlement, post-larvae redistributed into new areas of the shore, resulting in a distribution similar to that of the adult population. We did not directly measure redistribution through net sampling or mark and recapture techniques. However due to size frequency and density being measured on a good temporal resolution there is high confidence that further colonisation up the shore was facilitated by redistribution and not later settlements of larvae. There is a growing body of evidence that indicates that initial settlement patterns in intertidal soft sediment bivalves are spatially restricted where hydrodynamic conditions are favourable for the passive deposition of competent larvae, and that post-settlement redistribution is a key process in large scale population distribution (Armonies and Reise, 2003; Günther, 1992; Hiddink, 2003; Morse and Hunt, 2013). The occurrence of such a phenomenon appears across several families of bivalve with several 0-group bivalve species being correlated in settlement patterns on some occasions (Armonies and Reise, 2003). This cannot be underestimated in its importance for whole intertidal systems, as it means discrete areas of a system are potentially the source of post-larvae. Although not investigated or tested in this thesis, the value in undertaking such migrations for *Macoma balthica* is beneficial for individual fitness (Armonies and Hellwig-Armonies, 1992; Hiddink, 2003). At Traeth Melynog optimal areas for adult survival (Richardson et al., 1980; Sanchez-Salazar et al., 1987), also inferred from 4 years of adult density data collected during the PhD (unpublished data), did not receive larval settlement but subsequently colonised by post-larvae. If post-larvae remained where they initially settled it would likely be disadvantageous for their long term survival. We discuss in Chapter 2 that distinct advantages exist in migrating from low shore areas both in regards to post-larval, but also adult, survival from predation. This post-larval mobility is suggested by Armonies and Reise (2003), and by this thesis, as a strategy that allows *C. edule* to colonise more of the available habitat than larval settlement alone.

Observations on a smaller spatial scale (10s of metres), described in Chapter 3, showed a pattern of spatially restricted initial settlement in patches of 12 -18 metres in size and 18 – 24 metres apart. There was a similar patch size to that of the fine sediment content which may indicate the same process or scale of processes is operating on these two variables, but no correlation was observed between them which would discount an active selection for fine sediment. This initial pattern of highly patchy post-larval density changed post-settlement with a more random distribution, with a significant positive correlation and similar spatial structure to that of the adult cockle population. Ideally all variables would have been measured on the every sampling event and other species recorded along with other abiotic variables, this was not possible due to time constraints in the field and laboratory.

Over large and small spatial scales similar spatio-temporal changes in post-larval density were observed (Fig 6.2). The post-larval distribution on large and small scales was similar to that of the adults by the autumn and particularly close post-winter. This indicates as seen in *Mya arenaria* by Morse & Hunt (2013) that the spatial distribution of a cohort's recruitment is likely to be established within the first few months of the benthic life stage. This further supports that drivers of recruitment processes occurring during the post-larval stage are very important in *C. edule* populations.

From the findings of Chapters 2 and 3 the following conclusions and predictions can be made.

- 1) Larval settlement is likely restricted spatially by physical conditions across the intertidal flat with specific areas receiving intense settlements of larvae.
- 2) The ability of post-larvae to survive and undertake passive and/or active migrations is an extremely important for structuring populations spatially on large and small scales.
- 3) Large and small scale population spatial structure is likely to be created within the first 10 months of benthic life.

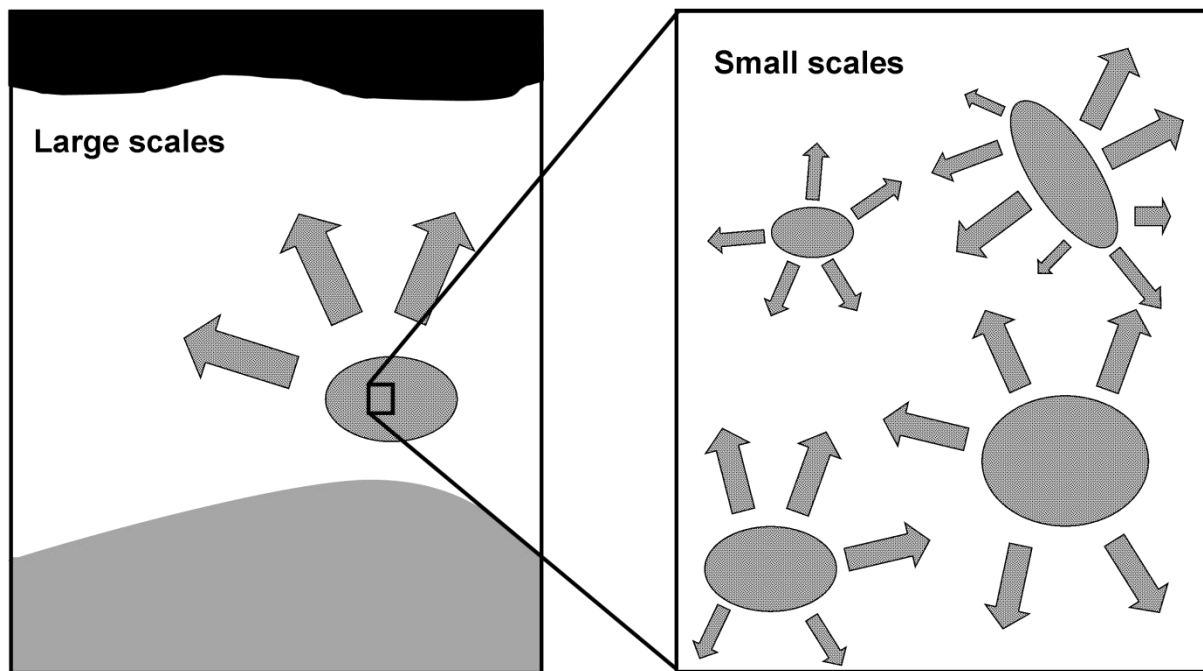


Figure 6.2. Schematic of distribution changes at large and small scales seen in chapters 2 and 3 of spatially restricted and patchy initial distributions becoming more dispersed post-settlement. White areas are intertidal sediment with grey arrows depicting the movement of post-larvae to become more dispersed.

6.3 The role of macrofaunal interactions in creating spatial patterns

Biological interactions are important in soft-sediment systems, and so the ability of macrofauna to cause variation in post-larval density and the causal processes were investigated. The presence of lugworms and adult cockles can alter the post-larval density in an area of sediment by 50% or more (Chapter 4). Importantly we observed that some macrofauna can have a negative effect on post-larval density (namely lugworms) and some a positive effect (adult cockles) at small scales (1 m² plots). This different interaction outcome indicates that different areas of sediment can be advantageous or disadvantageous for post-larvae depending on the macrofauna species. Therefore macrofauna can be a source of spatial heterogeneity in density of post-larvae. This thesis supports the hypothesis that clear advantages exist for a post-larva to be able to change its location after settlement. We saw no evidence from the laboratory experiments that direct mortality or increased byssus drifting rates occur in the presence of macrofauna. Repetition of the two laboratory studies with some refinements to the methodology would likely reduce variability in the data, but I do not think the main findings would differ. The observation of increased post-larval densities in the presence of adult cockles from the field experiment is supported by the large scale and small field studies that showed a positive correlation with adults after post-settlement redistribution. Evidence from this thesis supports a positive interaction between

post-larvae and adults existing at Traeth Melynog and the Dee estuary in Chapters 2 and 3. A negative relationship may well exist based on initial larval settlement from our data and other studies (Andre and Rosenberg, 1991); however a negative relationship does not seem to exist post-settlement. Although we cannot demonstrate a causative process for this positive interaction it is an important finding because it is largely thought to be negative. We speculate that the potential of adult cockles to increase habitat complexity and therefore reduce predator foraging efficiency may account for this observation. Adults increased survival of recruits from observations of natural recruitment and predation experiments for the bivalve *Katelysia* spp., including when adult mimics were used (Peterson and Black, 1993). The interaction between adult and post-larval bivalves (and subsequent recruitment) is more complex than perhaps thought and may be context dependant (Olafsson, 1989; Whitlatch et al., 1997). It was observed that the effect of macrofauna on post-larval densities is significant but further work is needed to identify the causal processes.

6.4 The role of predation in creating spatial patterns

Post-settlement predation is understood to be the main driver of recruitment success for intertidal bivalve populations (Beukema and Dekker, 2005), and so may also drive or maintain spatial distributions. Density-dependent predation and the ideal free distribution theory would work together so that patchiness in prey will be reduced over time under predation. In the field we do observe patchiness in prey populations inferring other processes may be important, and so a predication of a positive feedback between spatial aggregation and survival was made. In Chapter 5 we observed that predation risk is reduced for post-larvae when they are in a more dispersed and uniform spatial distribution, when compared to being spatially aggregated. Higher mortality of *C. edule* post-larvae was still observed when post-larvae were aggregated compared to dispersed. Interference competition of juvenile of juvenile *Carcinus maenas* was not higher when prey was aggregated, despite foraging efficiency being significantly reduced across both prey spatial arrangements. We concluded that the behaviour of 'take-away foraging', where crabs moved away from the prey patch (Smallegange et al., 2010), mitigated interference competition when feeding on aggregated or patchy prey. This would infer an advantage for post-larvae in the change from the highly aggregated spatial structure to a more dispersed distribution seen at the Dee estuary in Chapter 3, with the acknowledgement that spatial scales in patchiness were considerably different in the predation experiment compared to the field observations. Juvenile plaice *Pleuronectes platessa*, another predator of 0-group *C. edule*, are also able to feed effectively on patches of prey, when in isolation, by remaining in the patch area (Hill et al., 2000). However a similar behaviour to 'take-away foraging' was also

observed in juvenile *P. platessa* as they leave a prey attack location due to the disturbance created when feeding, which may attract predators (Burrows and Gibson, 1995), rather than due to interference competition or cannibalism seen in juvenile *C. maenas*. This strategy of moving away from prey patches may be disadvantageous if predators cannot relocate the prey patch however, which would be advantageous for prey but was not accounted for in our small scale experiment. It is clear that the spatial arrangement of prey, such as post-larval *C. edule*, has consequences on predation risk, but whether being patchy is beneficial for survival may depend on spatial scales of prey patches (Hines et al., 2009) and variable predator behaviours (Scharf et al., 2012).

6.5 Management implications for cockle populations

In Chapter 2 and 3 it was demonstrated that initial settlement of larvae can be highly concentrated. This means that individual sites may be extremely important for sustaining the entire exploited population, which in turn supports many ecosystem services and the overwintering birds which are highly protected in the Dee estuary. As suggested by Armonies & Reise (2003) we also conclude that sites that appear homogeneous may serve very different functions in that ecosystem. Identifying and understanding the properties of these locations and protecting them should be a high priority for those concerned with cockle recruitment. Indeed some areas in the Dee estuary receive good recruitment while other do not from year to year with no obvious reason, and so understanding processes creating spatial variation in 0-group cockles should not be ignored and incorporated into management. The finding of Chapter 4 may have important implications for management as we observed that removal of adults, using the same technique as cockle fisherman in the Dee, reduced the post-larval density. There is little evidence of a stock-recruitment relationship existing for species like *Cerastoderma edule* caused by reproductive output, however it may actually exist through an indirect interaction post-settlement in some locations. These results imply that exploiting areas where adult densities are significantly reduced may directly reduce the potential post-larval densities in the following settlement period. Identifying density dependence and the cause of this interaction between adult and post-larvae, may be highly relevant to fishery managers. Finally our findings on predation also have management relevance. It was observed that the juvenile crabs could consume a large number of post-larvae in a short period of time, especially when post-larvae are aggregated. Post-larval cockles can be predated on by a number of juvenile fish and crustaceans, which in turn are prey of adult conspecifics and other species. For example work done in Morecambe bay found that flatfish abundance correlated best with cockle recruitment (Cesar, 2009). Activities which may modify abundance of these adult or juvenile

predators may have effects on cockle post-larval survival and therefore recruitment. The management of the other fisheries in or outside the Dee may potentially impact cockle recruitment through complex food web linkages. It is clear that the post-larval stage is critical for recruitment and therefore adult populations and so increasing our understanding of processes acting upon post-larval cockles will benefit the stewardship of this important species.

6.6 Perspectives for future research

In this PhD we have seen the important role post-settlement redistributions in the early life history of *C. edule*, and the effect it has on population distribution. However the conditions that drive post-larvae to possibly reject or accept its physical and biological surroundings, if the process is active, are not fully understood. An expansion of the flume study in combination with field experiments, although technically difficult, to investigate further possible biotic (such as predator presences or chemical cues) and abiotic effects (such as emersion time or disturbance) that influence post-larval migrations may help identify why some areas have no or low densities of post-larvae. At Traeth Melynog post-larvae redistributed from the low shore, where predation pressure is greatest. Testing the hypothesis that redistribution may be a direct response to predator presence would be beneficial. Are post-larvae migrating actively from dense aggregations of conspecifics, like those seen in the Dee? Or is the small scale change to a more uniform distribution caused by passive transport or density dependent predation and predator aggregation? There are many other possible processes which could be hypothesised. An experimental approach to understand what is or is not causing or effecting redistributions is worthwhile, in light of the importance of redistribution to the cockle populations studied in this thesis and other documented bivalve populations. It was also seen that studying post-larval densities in single locations may provide inaccurate conclusions. For example if only the low shore area at Traeth Melynog was sampled it would have indicated a rapid increase and decrease in post-larval density immediately after settlement. However the large scale post-larval data including all shore heights shows that the rapid reduction in density was not representative of post-larval densities across the whole shore due to redistribution, and so future work on post-settlement processes should account for this spatial variation and the process of redistribution. Another area of research is the importance of direct and indirect interactions with other fauna, and is identified as a current key ecological question (Sutherland et al., 2013). Gaps in knowledge remain on how macrofauna interactions affect other important processes such as predation. Testing the effect of habitat engineering on predator foraging rates may explain patterns seen in the field and can be tested with simple laboratory

experiments, such as looking at predator foraging efficiency on post-larvae with adults present, absent and with adult mimics to separate the effect of the physical presence of adults from other factors. In the Dee estuary highly variable recruitment can be seen on large scales between beds (unpublished data) which could be assumed to have similar larval supply and predator recruitment, and so identifying the source of this variation is ecologically interesting and potentially commercially important.

6.7 Conclusion

In this thesis I reported that redistribution of post-larval *C. edule* from initial settlement locations were significant for large scale distributions. On small scales I quantified the spatial structure and change from patchy to more uniform in structure. Positive correlations in distribution with adults are seen after these changes in distribution. Experiments confirmed that a positive interaction can exist between post-larval and adult *C. edule*, but are negative with lugworms. Further experiments did not reveal a reason for this. The experiment assessing juvenile shore crab predation revealed cockle post-larvae have higher survival when they are more uniform in distribution. It is concluded that redistribution is important for habitat use and population recruitment, and that the end location or spatial arrangement a post-larvae finds itself in after redistribution has significant consequences for the outcome of macrofaunal interactions and predation risk.

References

- Andersen, T.J., Lanuru, M., van Bernem, C., Pejrup, M., Riethmueller, R., 2010. Erodibility of a mixed mudflat dominated by microphytobenthos and *Cerastoderma edule*, East Frisian Wadden Sea, Germany. *Estuarine, Coastal and Shelf Science* 87, 197-206.
- Andre, C., Rosenberg, R., 1991. Adult-larval interactions in the suspension-feeding bivalves *Cerastoderma edule* and *Mya arenaria*. *Marine Ecology-Progress Series* 71, 227-234.
- Andre, C., Jonsson, P.R., Lindegarth, M., 1993. Predation on settling bivalve larvae by benthic suspension feeders - The role of hydrodynamics and larval behavior. *Marine Ecology-Progress Series* 97, 183-192.
- Andre, C., Lindegarth, M., Jonsson, P.R., Sundberg, P., 1999. Species identification of bivalve larvae using random amplified polymorphic DNA (RAPD): differentiation between *Cerastoderma edule* and *C. lamarcki*. *Journal of the Marine Biological Association of the United Kingdom* 79, 563-565.
- Andresen, H., van der Meer, J., 2010. Brown shrimp (*Crangon crangon*, L.) functional response to density of different sized juvenile bivalves *Macoma balthica* (L.). *Journal of Experimental Marine Biology and Ecology* 390, 31-38.
- Armonies, W., 1992. Migratory rhythms of drifting juvenile molluscs in tidal waters of the Wadden Sea. *Marine Ecology-Progress Series* 83, 197-206.
- Armonies, W., 1994a. Drifting meio- and macrobenthic invertebrates on tidal flats in Königshafen: A review. *Helgoland Marine Research* 48, 299-320.
- Armonies, W., 1994b. Turnover of postlarval bivalves in sediments of tidal flats in Königshafen (German Wadden sea). *Helgoland Marine Research* 48, 291-297.
- Armonies, W., 1996. Changes in distribution patterns of 0-group bivalves in the Wadden Sea: Byssus-drifting releases juveniles from the constraints of hydrography. *Journal of Sea Research* 35, 323-334.
- Armonies, W., Hellwig-Armonies, M., 1992. Passive settlement of *Macoma balthica* spat on tidal flats of the Wadden Sea and subsequent migration of juveniles. *Netherlands Journal of Sea Research* 29, 371-378.
- Armonies, W., Reise, K., 2003. Empty habitat in coastal sediments for populations of macrozoobenthos. *Helgoland Marine Research* 56, 279-287.
- Arroyo, N.L., Aarnio, K., Mäensivu, M., Bonsdorff, E., 2012. Drifting filamentous algal mats disturb sediment fauna: Impacts on macro-meiofaunal interactions. *Journal of Experimental Marine Biology and Ecology* 420, 77-90.
- Bachelet, G., Desprez, M., Ducrottoy, J.P., Guillou, J., Labourg, P.J., Rybarczyk, H., Sauriau, P.G., Elkaim, B., Glemarec, M., 1992. The role of intraspecific competition in regulating recruitment in the cockle, *Cerastoderma-edule* (L.). *Annales De L Institut Oceanographique* 68, 75-87.
- Baggerman, B., 1954. Spatfall and Transport of *Cardium edule* L. *Archives Neerlandaises de Zoologie* 10, 315-342.
- Bergström, U., Englund, G., Bonsdorff, E., 2002. Small-scale spatial structure of Baltic Sea zoobenthos-inferring processes from patterns. *Journal of Experimental Marine Biology and Ecology* 281, 123-136.
- Beukema, J.J., 1976. Biomass and species richness of the macro-benthic animals living on the tidal flats of the Dutch Wadden Sea. *Netherlands Journal of Sea Research* 10, 236-261.
- Beukema, J.J., 1991. The abundance of shore crabs *Carcinus maenas* (L.) on a tidal flat in the Wadden Sea after cold and mild winters. *Journal of Experimental Marine Biology and Ecology* 153, 97-113.
- Beukema, J.J., 1992. Dynamics of juvenile shrimp *Crangon crangon* in a tidal-flat nursery of the Wadden Sea after mild and cold winters. *Marine ecology progress series. Oldendorf* 83, 157-165.

- Beukema, J.J., 1993. Successive changes in distribution patterns as an adaptive strategy in the bivalve *Macoma balthica* (L.) in the Wadden sea. *Helgoland Marine Research* 47, 287-304.
- Beukema, J.J., De Vlas, J., 1989. Tidal-current transport of thread-drifting postlarval juveniles of the bivalve *Macoma balthica* from the Wadden Sea to the North Sea. *Marine ecology progress series*. Oldendorf 52, 193-200.
- Beukema, J.J., Dekker, R., 2005. Decline of recruitment success in cockles and other bivalves in the Wadden Sea: possible role of climate change, predation on postlarvae and fisheries. *Marine ecological progress series* 287, 149-167.
- Beukema, J.J., Dekker, R., 2009. The intertidal zoning of cockles (*Cerastoderma edule*) in the Wadden Sea, or why cockle fishery disturbed areas of relatively high biodiversity. *Helgoland Marine Research* 63, 287-291.
- Beukema, J.J., Honkoop, P., Dekker, R., 1998. Recruitment in *Macoma balthica* after mild and cold winters and its possible control by egg production and shrimp predation. *Hydrobiologia* 375-376, 23-34.
- Beukema, J.J., Dekker, R., Philippart, C.J.M., 2010. Long-term variability in bivalve recruitment, mortality, and growth and their contribution to fluctuations in food stocks of shellfish-eating birds. *Marine Ecology Progress Series* 414, 117-130.
- Beukema, J.J., Essink, K., Michaelis, H., Zwarts, L., 1993. Year-to-year variability in the biomass of macrobenthic animals on tidal flats of the Wadden Sea: How predictable is this food source for birds? *Netherlands Journal of Sea Research* 31, 319-330.
- Blott, S.J., Pye, K., 2001. GRADISTAT: a grain size distribution and statistics package for the analysis of unconsolidated sediments. *Earth surface processes and Landforms* 26, 1237-1248.
- Bocher, P., Piersma, T., Dekinga, A., Kraan, C., Yates, M., Guyot, T., Folmer, E., Radenac, G., 2007. Site- and species-specific distribution patterns of molluscs at five intertidal soft-sediment areas in northwest Europe during a single winter. *Marine Biology* 151, 577-594.
- Boldina, I., Beninger, P.G., 2013. Fine-scale spatial structure of the exploited infaunal bivalve *Cerastoderma edule* on the French Atlantic coast. *Journal of Sea Research* 76, 193-200.
- Bouma, H., Duiker, J.M.C., de Vries, P.P., Herman, P.M.J., Wolff, W.J., 2001. Spatial pattern of early recruitment of *Macoma balthica* (L.) and *Cerastoderma edule* (L.) in relation to sediment dynamics on a highly dynamic intertidal sandflat. *Journal of Sea Research* 45, 79-93.
- Brock, V., 1979. Habitat selection of two congeneric bivalves, *Cardium edule* and *C. glaucum* in sympatric and allopatric populations. *Marine Biology* 54, 149-156.
- Burch, A., Seed, R., 2000. Foraging behaviour of *Carcinus maenas* on *Mytilus edulis*: the importance of prey presentation. *Journal of the Marine Biological Association of the UK* 80, 799-810.
- Burrough, P.A., McDonnell, R.A., McDonnell, R., 1998. Principles of geographical information systems. Oxford university press Oxford.
- Burrows, M.T., Gibson, R.N., 1995. The effects of food, predation risk and endogenous rhythmicity on the behaviour of juvenile plaice, *Pleuronectes platessa* (L.). *Animal Behaviour* 50, 41-52.
- Butman, C.A., 1987. Larval settlement of soft-sediment invertebrates - The spatial scales of pattern explained by active habitat selection and the emerging role of hydrodynamical processes. *Oceanography and Marine Biology* 25, 113-165.
- Caron, A., Desrosiers, G., Olive, P.J.W., Retiere, C., Nozaisa, C., 2004. Comparison of diet and feeding activity of two polychaetes, *Nephtys caeca* (Fabricius) and *Nereis virens* (Sars), in an estuarine intertidal environment in Que'bec, Canada. *Journal of Experimental Marine Biology and Ecology* 304 225 - 242.
- Cesar, C., 2009. The roles of the cockle *Cerastoderma edule* L. on ecosystem functioning: cockle comings and goings. University of Liverpool.

- Cesar, C.P., Frid, C.L.J., 2009. Effects of experimental small-scale cockle (*Cerastoderma edule* L.) fishing on ecosystem function. *Marine Ecology* 30, 123-137.
- Chang, B.D., Levings, C.D., 1978. Effects of burial on the heart cockle *Clinocardium nuttallii* and the Dungeness crab *Cancer magister*. *Estuarine and Coastal Marine Science* 7, 409-412.
- Ciutat, A., Widdows, J., Pope, N.D., 2007. Effect of *Cerastoderma edule* density on near-bed hydrodynamics and stability of cohesive muddy sediments. *Journal of Experimental Marine Biology and Ecology* 346, 114-126.
- Clark, M.E., Wolcott, T.G., Wolcott, D.L., Hines, A.H., 1999a. Foraging and agonistic activity co-occur in free-ranging blue crabs (*Callinectes sapidus*): observation of animals by ultrasonic telemetry. *Journal of Experimental Marine Biology and Ecology* 233, 143-160.
- Clark, M.E., Wolcott, T.G., Wolcott, D.L., Hines, A.H., 1999b. Intraspecific interference among foraging blue crabs *Callinectes sapidus*: interactive effects of predator density and prey patch distribution. *Marine Ecology Progress Series* 178, 69-78.
- Clark, M.E., Wolcott, T.G., Wolcott, D.L., Hines, A.H., 2000. Foraging behavior of an estuarine predator, the blue crab *Callinectes sapidus* in a patchy environment. *Ecography* 23, 21-31.
- Cole, R.G., 2001. Staggered nested designs to identify hierarchical scales of variability. *New Zealand Journal of Marine and Freshwater Research* 35, 891 - 896.
- Cole, R.G., 2009. Staggered nested designs to assess scales of variability: the advantages of a spatially explicit analysis. *Environmental Monitoring and Assessment* 153, 427-434.
- Cole, R.G., Healy, T.R., Wood, M.L., Foster, D.M., 2001. Statistical Analysis of Spatial Pattern: A Comparison of Grid and Hierarchical Sampling Approaches. *Environmental Monitoring and Assessment* 69, 85-99.
- Collins, M.B., Ke, X., Gao, S., 1998. Tidally-induced Flow Structure Over Intertidal Flats. *Estuarine, Coastal and Shelf Science* 46, 233-250.
- Compton, T.J., Troost, T.A., Drent, J., Kraan, C., Bocher, P., Leyrer, J., Dekinga, A., Piersma, T., 2009. Repeatable sediment associations of burrowing bivalves across six European tidal flat systems. *Marine Ecology-Progress Series* 382, 87-98.
- Corgos, A., SÁnchez, N., Freire, J., 2010. Dynamics of the Small-Scale Spatial Structure of a Population of the Spider Crab *Maja brachydactyla* (Decapoda: Majidae). *Journal of Shellfish Research* 29, 25-36.
- Dare, P.J., Bell, M.C., Walker, P., Bannister, R.C.A., 2004. Historical and current status of cockle and mussel stocks in The Wash. CEFAS Lowestoft, 85.
- De Montaudouin, X., Bachelet, G., 1996. Experimental evidence of complex interactions between biotic and abiotic factors in the dynamics of an intertidal population of the bivalve *Cerastoderma edule*. *Oceanologica Acta* 19, 449-463.
- De Montaudouin, X., Bachelet, G., Sauriau, P.G., 2003. Secondary settlement of cockles *Cerastoderma edule* as a function of current velocity and substratum: a flume study with benthic juveniles. *Hydrobiologia* 503, 103-116.
- DeLong, J.P., Vasseur, D.A., 2011. Mutual interference is common and mostly intermediate in magnitude. *BMC Ecology* 11, 8pp.
- Donadi, S., Westra, J.I., Weerman, E., Heide, T., Zee, E., Koppel, J., Olf, H., Piersma, T., Veer, H., Eriksson, B., 2013. Non-trophic Interactions Control Benthic Producers on Intertidal Flats. *Ecosystems*, 1-11.
- Elmgren, R., Ankar, S., Marteleur, B., Ejdung, G., 1986. Adult Interference with Postlarvae in Soft Sediments: The *Pontoporeia-Macoma* Example. *Ecology* 67, 828-836.
- Findlay, S.E.G., 1981. Small-scale spatial distribution of meiofauna on a mud- and sandflat. *Estuarine, Coastal and Shelf Science* 12, 471-484.
- Flach, E.C., 1992. Disturbance of benthic infauna by sediment-reworking activities of the lugworm *Arenicola marina*. *Netherlands Journal of Sea Research* 30, 81-89.

- Flach, E.C., 1994. The influence of the cockle, *Cerastoderma edule*, on the macrozoobenthic community of tidal flats in the Wadden Sea, 29th European Marine Biology Symposium. Blackwell Wissenschafts-Verlag GmbH, Vienna, Austria, pp. 87-98.
- Flach, E.C., 1996. The Influence of the Cockle, *Cerastoderma edule*, on the Macrozoobenthic Community of Tidal Flats in the Wadden Sea. *Marine Ecology* 17, 87-98.
- Flach, E.C., 2003. The separate and combined effects of epibenthic predation and presence of macro-infauna on the recruitment success of bivalves in shallow soft-bottom areas on the Swedish west coast. *Journal of Sea Research* 49, 59-67.
- Flach, E.C., de Bruin, W., 1994. Does the activity of cockles, *Cerastoderma edule* (L.) and lugworms, *Arenicola marina* L., make *Corophium volutator* Pallas more vulnerable to epibenthic predators: a case of interaction modification? *Journal of Experimental Marine Biology and Ecology* 182, 265-285.
- Fortin, M.J., Dale, M.R.T., Hoef, J., 2006. Spatial analysis in ecology, *Encyclopedia of environmetrics*.
- Freitas, V., Campos, J., Fonds, M., Van der Veer, H.W., 2007. Potential impact of temperature change on epibenthic predator-bivalve prey interactions in temperate estuaries. *Journal of Thermal Biology* 32, 328-340.
- Fretwell, S.D., Lucas, H.L., 1969. On territorial behavior and other factors influencing habitat distribution in birds. *Acta Biotheoretica* 19, 16-36.
- Garland, E.D., Zimmer, C.A., 2002. Techniques for the identification of bivalve larvae. *Marine Ecology Progress Series* 225, 299-310.
- Gimenez, L., 2009. Uncertainty relationships in the estimation of settlement rates of mobile marine organisms. *Marine Ecology-Progress Series* 377, 103-111.
- Goss-Custard, J.D., 1980. Competition for food and interference among waders. *Ardea* 68, 31-52.
- Guillou, J., Tartu, C., 1994. Post-larval and juvenile mortality in a population of the edible cockle *cerastoderma edule* (L.) from northern Brittany. *Netherlands Journal of Sea Research* 33, 103-111.
- Günther, C.-P., 1992. Dispersal of intertidal invertebrates: A strategy to react to disturbance of different scales? *Netherlands Journal of Sea Research* 30, 45-56.
- Gunther, C.P., 1991. Settlement of *Macoma balthica* on an intertidal sandflat in the Wadden Sea. *Marine Ecology-Progress Series* 76, 73-79.
- Helm, M.M., Bourne, N., Lovatelli, A., Food, Agriculture Organization of the United, N., 2004. Hatchery culture of bivalves: a practical manual. Food and agriculture organization of the United Nations.
- Hendriks, I.E., van Duren, L.A., Herman, P.M.J., 2005. Image analysis techniques: A tool for the identification of bivalve larvae? *Journal of Sea Research* 54, 151-162.
- Hicken, V., 2008. Fisheries and Nature Conservation Issues of the Cockle Fishery in the Welsh District of the North Western and North Wales Sea Fisheries Committee. North Western & North Wales Sea Fisheries Committee, Lancashire, pp. 36.
- Hiddink, Kock, Wolff, 2002a. Active pelagic migrations of the bivalve *Macoma balthica* are dangerous. *Marine Biology* 140, 1149-1156.
- Hiddink, J.G., 2002. The adaptive value of migrations for the bivalve *Macoma balthica* Marine Biology research group. University of Groningen, Groningen, pp. 172.
- Hiddink, J.G., 2003. Modelling the adaptive value of intertidal migration and nursery use in the bivalve *Macoma balthica*. *Marine Ecology Progress Series* 252.
- Hiddink, J.G., Wolff, W.J., 2002. Changes in distribution and decrease in numbers during migration of the bivalve *Macoma balthica*. *Marine Ecology Progress Series* 233, 117-130.
- Hiddink, J.G., ter Hofstede, R., Wolff, W.J., 2002b. Predation of intertidal infauna on juveniles of the bivalve *Macoma balthica*. *Journal of Sea Research* 47, 141-159.
- Hiddink, J.G., Marijnissen, S.A.E., Troost, K., Wolff, W.J., 2002c. Predation on 0-group and older year classes of the bivalve *Macoma balthica*: interaction of size selection and

- intertidal distribution of epibenthic predators. *Journal of Experimental Marine Biology and Ecology* 269, 223-248.
- Hill, S., Burrows, M.T., Hughes, R.N., 2000. Increased turning per unit distance as an area-restricted search mechanism in a pause-travel predator, juvenile plaice, foraging for buried bivalves. *Journal of Fish Biology* 56, 1497-1508.
- Hinchey, E., Schaffner, L., Hoar, C., Vogt, B., Batte, L., 2006. Responses of Estuarine Benthic Invertebrates to Sediment Burial: The Importance of Mobility and Adaptation. *Hydrobiologia* 556, 85-98.
- Hines, A.H., Long, W.C., Terwin, J.R., Thrush, S.F., 2009. Facilitation, interference, and scale: the spatial distribution of prey patches affects predation rates in an estuarine benthic community. *Marine Ecology Progress Series* 385, 127-135.
- Honkoop, P.J.C., van der Meer, J., 1998. Experimentally induced effects of water temperature and immersion time on reproductive output of bivalves in the Wadden Sea. *Journal of Experimental Marine Biology and Ecology* 220, 227-246.
- Horwood, J.W., Goss-Custard, J.D., 1977. Predation by the Oystercatcher, *Haematopus ostralegus* (L.), in Relation to the Cockle, *Cerastoderma edule* (L.), Fishery in the Burry Inlet, South Wales. *Journal of Applied Ecology* 14, 139-158.
- Hughes, R.N., Elner, R.W., 1979. Tactics of a Predator, *Carcinus maenas*, and Morphological Responses of the Prey, *Nucella lapillus*. *Journal of Animal Ecology* 48, 65-78.
- Hunt, H.L., McLean, D.A., Mullineaux, L.S., 2003. Post-settlement alteration of spatial patterns of soft shell clam (*Mya arenaria*) recruits. *Estuaries* 26, 72-81.
- Huxham, M., Richards, M., 2003. Can postlarval bivalves select sediment type during settlement? A field test with *Macoma balthica* (L.) and *Cerastoderma edule* (L.). *Journal of Experimental Marine Biology and Ecology* 288, 279-293.
- Hylleberg, J., Brock, V., Joergensen, F., 1978. Production of sublittoral cockles, *Cardium edule* L., with emphasis on predation by flounders and sea stars [Denmark] *Natura Jutlandica* (Denmark), 20 p. 183-191.
- Jensen, K.T., 1993. Density-dependent growth in cockles (*Cerastoderma edule*): evidence from interannual comparisons. *Journal of the Marine Biological Association of the UK* 73, 333-342.
- Jensen, K.T., Jensen, J.N., 1985. The importance of some epibenthic predators on the density of juvenile benthic macrofauna in the Danish Wadden Sea. *Journal of Experimental Marine Biology and Ecology* 89, 157-174.
- Jonsson, P.R., Andre, C., Lindegarth, M., 1991. Swimming behavior of marine bivalve larvae in a flume boundary-layer flow - evidence for near-bottom confinement. *Marine Ecology-Progress Series* 79, 67-76.
- Jorgensen, C.B., 1946. Lamellibranchia. In: Thorsen, G. (Ed.), *Reproduction and larval development of Danish marine bottom invertebrates: with special reference to the planktonic larvae in the Sound (Øresund)*, Copenhagen, pp. 523.
- Jung, K., Stelzenmüller, V., Zauke, G.-P., 2006. Spatial distribution of heavy metal concentrations and biomass indices in *Cerastoderma edule* Linnaeus (1758) from the German Wadden Sea: An integrated biomonitoring approach. *Journal of Experimental Marine Biology and Ecology* 338, 81-95.
- Kingston, P.F., 1974. Studies on the reproductive cycles of *Cardium edule* and *C. glaucum*. *Marine Biology* 28, 317-323.
- Klein Breteler, W.C.M., 1975. Food consumption, growth and energy metabolism of juvenile shore crabs, *Carcinus maenas*. *Netherlands Journal of Sea Research* 9, 255-272.
- Kraan, C., 2010. Spatial ecology of intertidal macrobenthic fauna in a changing Wadden Sea, Faculty of Mathematics and Natural Sciences. University of Groningen, Groningen, pp. 200.
- Kraan, C., Van der Meer, J., Dekinga, A., Piersma, T., 2009. Patchiness of macrobenthic invertebrates in homogenized intertidal habitats: hidden spatial structure at a landscape scale. *Marine Ecology Progress Series* 383, 211-224.

- Kratina, P., Vos, M., Bateman, A., Anholt, B.R., 2009. Functional responses modified by predator density. *Oecologia* 159, 425-433.
- Kreger, D., 1940. On the Ecology of *Cardium edule* L. *Archives Néerlandaises de Zoologie* 4, 157-200.
- Lebour, M.V., 1938. Notes on the breeding of some lamellibranchs from Plymouth and their larvae. *Journal of the Marine Biological Association of the UK* 23, 119 - 144.
- Leedham, J., 2011. Improving the sustainability of cockle fisheries :interactions between *Arenicola marina* and cockle (*Cerastoderma edule*) spat School of Ocean Sciences. Bangor University, Bangor, pp. 43.
- Legendre, P., Fortin, M.J., 1989. Spatial pattern and ecological analysis. *Plant Ecology* 80, 107-138.
- Legendre, P., Thrush, S.F., Cummings, V.J., Dayton, P.K., Grant, J., Hewitt, J.E., Hines, A.H., McArdle, B.H., Pridmore, R.D., Schneider, D.C., Turner, S.J., Whitlatch, R.B., Wilkinson, M.R., 1997. Spatial structure of bivalves in a sandflat: Scale and generating processes. *Journal of Experimental Marine Biology and Ecology* 216, 99-128.
- Lei, Y., Stumm, K., Volkenborn, N., Wickham, S., Berninger, U.-G., 2010. Impact of *Arenicola marina* (Polychaeta) on the microbial assemblages and meiobenthos in a marine intertidal flat. *Marine Biology* 157, 1271-1282.
- Lewin, R., 1986. Supply-Side Ecology. *Science* 234, 25-27.
- Liebold, A.M., Gurevitch, J., 2002. Integrating the statistical analysis of spatial data in ecology. *Ecography* 25, 553-557.
- Lindegarth, M., Jonsson, P.R., Andre, C., 1991. Fluorescent Microparticles: A New Way of Visualizing Sedimentation and Larval Settlement. *Limnology and Oceanography* 36, 1471-1476.
- Lindegarth, M., Andre, C., Jonsson, P.R., 1995. Analysis of the spatial variability in abundance and age structure of 2 infaunal bivalves, *Cerastoderma edule* and *C. lamarcki*, using hierarchical sampling programs. *Marine Ecology-Progress Series* 116, 85-97.
- Lindegarth, M., Jonsson, P.R., Andr, eacute, C, 2002. Physical and numerical modeling of the role of hydrodynamic processes on adult-larval interactions of a suspension-feeding bivalve. *Journal of Marine Research* 60, 499-516.
- Loosanoff, V.L., Davis, H.C., Chanley, P.E., 1966. Dimensions and shapes of larvae of some marine bivalve mollusks. *Malacologia* 4, 351-435.
- Malham, S.K., Hutchinson, T.H., Longshaw, M., 2012. A review of the biology of European cockles (*Cerastoderma* spp.). *Journal of the Marine Biological Association of the United Kingdom* 92, 1563-1577.
- Marquet, P.A., Fortin, M.J., Pineda, J., Wallin, D.O., Clark, J., Wu, Y., Bollens, S., Jacobi, C.M., Holt, R.D., 1993. Ecological and evolutionary consequences of patchiness: a marine-terrestrial perspective. *Patch dynamics*. Springer-Verlag, New York, New York, USA, 277-304.
- Mascaro, M., Seed, R., 2001. Foraging behavior of juvenile *Carcinus maenas* (L.) and *Cancer pagurus* L. *Marine Biology* 139, 1135-1145.
- Maurer, D., Keck, R.T., Tinsman, J.C., Leathem, W.A., 1981. Vertical migration and mortality of benthos in dredged material--part I: Mollusca. *Marine Environmental Research* 4, 299-319.
- McArdle, B.H., Hewitt, J.E., Thrush, S.F., 1997. Pattern from process: it is not as easy as it looks. *Journal of Experimental Marine Biology and Ecology* 216, 229-242.
- McDonald, P.S., Jensen, G.C., Armstrong, D.A., 2001. The competitive and predatory impacts of the nonindigenous crab *Carcinus maenas* (L.) on early benthic phase Dungeness crab *Cancer magister* Dana. *Journal of Experimental Marine Biology and Ecology* 258, 39-54.
- Methven, D.A., Schneider, D.C., Rose, G.A., 2003. Spatial pattern and patchiness during ontogeny: post-settled *Gadus morhua* from coastal Newfoundland. *ICES Journal of Marine Science: Journal du Conseil* 60, 38-51.

- Moksnes, P.-O., 2004a. Self-regulating mechanisms in cannibalistic populations of juvenile shore crabs *Carcinus maenas*. *Ecology* 85, 1343-1354.
- Moksnes, P.O., 2004b. Interference competition for space in nursery habitats: density-dependent effects on growth and dispersal in juvenile shore crabs *Carcinus maenas*. *Marine Ecology Progress Series* 281, 181-191.
- Moksnes, P.O., Pihl, L., Montfrans, J.v., 1998. Predation on postlarvae and juveniles of the shore crab *Carcinus maenas*: importance of shelter, size and cannibalism. *Marine Ecology Progress Series* 166, 211-225.
- Moksnes, P.O., Lipcius, R.N., Pihl, L., van Montfrans, J., 1997. Cannibal-prey dynamics in young juveniles and postlarvae of the blue crab. *Journal of Experimental Marine Biology and Ecology* 215, 157-187.
- Moore, J., 2004. Surveys of cockle and mussel stocks on three Welsh sand flats. . In: Sanderson, W.G. (Ed.), CCW Marine Monitoring Report. CCW, pp. 29.
- Moore, R.D., Wolf, J., Souza, A.J., Flint, S.S., 2009. Morphological evolution of the Dee Estuary, Eastern Irish Sea, UK: A tidal asymmetry approach. *Geomorphology* 103, 588-596.
- Morrisey, D.J., Howitt, L., Underwood, A.J., Stark, J.S., 1992a. Spatial variation in soft-sediment benthos. *Marine Ecology-Progress Series* 81, 197-204.
- Morrisey, D.J., Underwood, A.J., Howitt, L., Stark, J.S., 1992b. Temporal variation in soft-sediment benthos. *Journal of Experimental Marine Biology and Ecology* 164, 233-245.
- Morse, B.L., Hunt, H.L., 2013. Impact of settlement and early post-settlement events on the spatial distribution of juvenile *Mya arenaria* on an intertidal shore. *Journal of Experimental Marine Biology and Ecology* 448, 57-65.
- Myrand, B., Chevarie, L., Tremblay, R., 2012. Benthic Spat Collection of Softshell Clams (*Mya arenaria* Linnaeus, 1758) using Mats. *Journal of Shellfish Research* 31, 39-48.
- Nachman, G.s., 2006. The Effects of Prey Patchiness, Predator Aggregation, and Mutual Interference on the Functional Response of *Phytoseiulus persimilis* Feeding on *Tetranychus urticae* (Acari: *Phytoseiidae*, *Tetranychidae*). *Experimental and Applied Acarology* 38, 87-111.
- Naylor, E., 1962. Seasonal Changes in a Population of *Carcinus maenas* (L.) in the Littoral Zone. *Journal of Animal Ecology* 31, 601-609.
- Norkko, A., 1998. The impact of loose-lying algal mats and predation by the brown shrimp *Crangon crangon* (L.) on infaunal prey dispersal and survival. *Journal of Experimental Marine Biology and Ecology* 221, 99-116.
- Oaten, A., 1977. Transit Time and Density-Dependent Predation on a Patchily Distributed Prey. *The American Naturalist* 111, 1061-1075.
- Olafsson, E., 1992. Small-scale spatial distribution of marine meiobenthos: the effects of decaying macrofauna. *Oecologia* 90, 37-42.
- Olafsson, E.B., 1989. Contrasting influences of suspension-feeding and deposit-feeding populations of *Macoma balthica* on infaunal recruitment. *Marine ecology progress series*. 55, 171-179.
- Olafsson, E.B., Peterson, C.H., Ambrose, W.G., 1994. Does recruitment limitation structure populations and communities of macroinvertebrates in marine soft sediments - The relative significance of presettlement and postsettlement processes. *Oceanography and Marine Biology* 32, 65-109.
- Omori, M., Hamner, W.M., 1982. Patchy distribution of zooplankton: Behavior, population assessment and sampling problems. *Marine Biology* 72, 193-200.
- Orton, J.H., 1937. Some Interrelations between Bivalve Spatfalls, Hydrography and Fisheries. *Nature*, 505-506.
- Patric, D., Patrick, B., Philippe, N., Philippe, J., 1997. Patchy recruitment patterns in marine invertebrates: A spatial test of the density-dependent hypothesis in the bivalve *Spisula ovalis*. *Oecologia* 111, 331-340.

- Peterson, C.H., Black, R., 1993. Experimental tests of the advantages and disadvantages of high density for two coexisting cockles in a Southern Ocean lagoon. *Journal of Animal Ecology*, 614-633.
- Pettitt, A.N., McBratney, A.B., 1993. Sampling Designs for Estimating Spatial Variance Components. *Journal of the Royal Statistical Society. Series C (Applied Statistics)* 42, 185-209.
- Philippart, C.J.M., Aken, H.M.v., Beukema, J.J., Bos, O.G., Cadace, G.C., Dekker, R., 2003. Climate-Related Changes in Recruitment of the Bivalve *Macoma balthica*. *Limnology and Oceanography* 48, 2171-2185.
- Pihl, L., Rosenberg, R., 1984. Food selection and consumption of the shrimp *Crangon crangon* in some shallow marine areas in western Sweden. *Marine Ecology-Progress Series* 15, 159-168.
- Pinckney, J., Sandulli, R., 1990. Spatial autocorrelation analysis of meiofaunal and microalgal populations on an intertidal sandflat: Scale linkage between consumers and resources. *Estuarine, Coastal and Shelf Science* 30, 341-353.
- Pinn, E.H., Ansell, A.D., 1993. The Effect of particle size on the burying ability of the brown shrimp *Crangon crangon*. *Journal of the Marine Biological Association of the United Kingdom* 73, 365-377.
- Rangeley, R.W., Thomas, M.L.H., 1987. Predatory behaviour of juvenile shore crab *Carcinus maenas* (L.). *Journal of Experimental Marine Biology and Ecology* 108, 191-197.
- Reise, K., 1978. Experiments on epibenthic predation in the Wadden Sea. *Helgoland Marine Research* 31, 55-101.
- Reise, K., 1985a. Tidal flat ecology. An experimental approach to species interactions. *Ecological Studies*, i-x, 1-191.
- Reise, K., 1985b. Predator control in marine tidal sediments. *European Marine Biology Symposium* 19, 311-321.
- Reise, K., 2002. Sediment mediated species interactions in coastal waters. *Journal of Sea Research* 48, 127-141.
- Rhoads, D.C., Young, D.K., 1970a. The influence of deposit-feeding organisms on sediment stability and community trophic structure. *Journal of Marine Research* 28, 150-178.
- Rhoads, D.C., Young, D.K., 1970b. The influence of deposit-feeding organisms on sediment stability and community trophic structure. *Journal of Marine Research* 28, 150 - 178.
- Richards, M.G., Huxham, M., Bryant, A., 1999. Predation: a causal mechanism for variability in intertidal bivalve populations. *Journal of Experimental Marine Biology and Ecology* 241, 159-177.
- Richardson, C.A., Crisp, D.J., Runham, N.W., 1980. Factors influencing shell growth in *Cerastoderma edule*. *Proceedings of the Royal Society of London. Series B. Biological Sciences* 210, 513.
- Richardson, C.A., Ibarrola, I., Ingham, R.J., 1993. Emergence pattern and spatial distribution of the common cockle *Cerastoderma edule*. *Mar Ecol Prog Ser* 99, 71-81.
- Rosenberg, M.S., Anderson, C.D., 2011. PASSaGE: Pattern Analysis, Spatial Statistics and Geographic Exegesis. Version 2. *Methods in Ecology and Evolution* 2, 229-232.
- Sanchez-Salazar, M.E., Griffiths, C.L., Seed, R., 1987. The interactive roles of predation and tidal elevation in structuring populations of the edible cockle, *Cerastoderma edule*. *Estuarine Coastal and Shelf Science* 25, 245-260.
- Scharf, I., Ovadia, O., Foitzik, S., 2012. The advantage of alternative tactics of prey and predators depends on the spatial pattern of prey and social interactions among predators. *Population Ecology* 54, 187-196.
- Seed, R., Brown, R.A., 1977. A comparison of the reproductive cycles of *Modiolus modiolus* (L.), *Cerastoderma (=Cardium) edule* (L.), and *Mytilus edulis* L. in Strangford Lough, Northern Ireland. *Oecologia* 30, 173-188.
- Skalski, G.T., Gilliam, J.F., 2001. Functional responses with predator interference: viable alternatives to the Holling type II model. *Ecology* 82, 3083-3092.
- Smallegange, I.M., van der Meer, J., 2007. Interference from a game theoretical perspective: shore crabs suffer most from equal competitors. *Behavioral Ecology* 18, 215-221.

- Smallegange, I.M., Van Der Meer, J., Kurvers, R.H.J.M., 2006. Disentangling interference competition from exploitative competition in a crab–bivalve system using a novel experimental approach. *Oikos* 113, 157-167.
- Smallegange, I.M., van der Meer, J., Sabelis, M.W., 2010. 'Take-away' foraging spatially uncouples predator and prey-attack distributions. *Journal of Animal Ecology* 79, 769-776.
- Sneddon, L.U., Huntingford, F.A., Taylor, A.C., 1997. The influence of resource value on the agonistic behaviour of the shore crab, *Carcinus maenas* (L.). *Marine & Freshwater Behaviour & Physiology* 30, 225-237.
- Snelgrove, P.V.R., 1994. Hydrodynamic enhancement of invertebrate larval settlement in microdepositional environments: colonization tray experiments in a muddy habitat. *Journal of Experimental Marine Biology and Ecology* 176, 149-166.
- Snelgrove, P.V.R., Butman, C.A., 1994. Animal sediment relationships revisited - cause versus effect. *Oceanography and Marine Biology*, Vol 32 32, 111-177.
- Strasser, M., Günther, C.P., 2001. Larval supply of predator and prey: temporal mismatch between crabs and bivalves after a severe winter in the Wadden Sea. *Journal of Sea Research* 46, 57-67.
- Sutherland, W.J., 1982a. Spatial Variation in the Predation of Cockles by Oystercatchers at Traeth Melynog, Anglesey. II. the Pattern of Mortality. *Journal of Animal Ecology* 51, 491-500.
- Sutherland, W.J., 1982b. Spatial Variation in the Predation of Cockles by Oystercatchers at Traeth Melynog, Anglesey. I. The Cockle Population. *Journal of Animal Ecology* 51, 481-489.
- Sutherland, W.J., 1983. Aggregation and the 'Ideal Free' Distribution. *Journal of Animal Ecology* 52, 821-828.
- Sutherland, W.J., Freckleton, R.P., Godfray, H.C.J., Beissinger, S.R., Benton, T., Cameron, D.D., Carmel, Y., Coomes, D.A., Coulson, T., Emmerson, M.C., Hails, R.S., Hays, G.C., Hodgson, D.J., Hutchings, M.J., Johnson, D., Jones, J.P.G., Keeling, M.J., Kokko, H., Kunin, W.E., Lambin, X., Lewis, O.T., Malhi, Y., Mieszkowska, N., Milner-Gulland, E.J., Norris, K., Phillimore, A.B., Purves, D.W., Reid, J.M., Reuman, D.C., Thompson, K., Travis, J.M.J., Turnbull, L.A., Wardle, D.A., Wiegand, T., 2013. Identification of 100 fundamental ecological questions. *Journal of Ecology* 101, 58-67.
- Thorson, G., 1950. Reproductive and larval ecology of marine bottom invertebrates. *Biological Reviews* 25, 1-45.
- Thrush, S.F., 1991. Spatial patterns in soft-bottom communities. *Trends in Ecology & Evolution* 6, 75-79.
- Thrush, S.F., 1999. Complex role of predators in structuring soft-sediment macrobenthic communities: Implications of changes in spatial scale for experimental studies. *Australian Journal of Ecology* 24, 344-354.
- Thrush, S.F., Hewitt, J.E., Pridmore, R.D., Cummings, V.J., 1996. Adult/juvenile interactions of infaunal bivalves: contrasting outcomes in different habitats. *Marine Ecology Progress Series* 132, 83-92.
- Thrush, S.F., Cummings, V.J., Dayton, P.K., Ford, R., Grant, J., Hewitt, J.E., Hines, A.H., Lawrie, S.M., Pridmore, R.D., Legendre, P., 1997. Matching the outcome of small-scale density manipulation experiments with larger scale patterns: an example of bivalve adult/juvenile interactions. *Journal of Experimental Marine Biology and Ecology* 216, 153-169.
- Tobler, W.R., 1970. A computer movie simulating urban growth in the Detroit region. *Economic geography* 46, 234-240.
- Underwood, A.J., Chapman, M.G., Connell, S.D., 2000. Observations in ecology: you can't make progress on processes without understanding the patterns. *Journal of Experimental Marine Biology and Ecology* 250, 97-115.

- Van Colen, C., Thrush, S.F., Vincx, M., Ysebaert, T., 2013. Conditional Responses of Benthic Communities to Interference from an Intertidal Bivalve. *PLoS ONE* 8, e65861.
- van de Koppel, J., Rietkerk, M., Dankers, N., Herman, P.M.J., 2005. Scale dependent feedback and regular spatial patterns in young mussel beds. *The American Naturalist* 165, E66-E77.
- van der Veer, H.W., Feller, R.J., Weber, A., Witte, J., 1998. Importance of predation by crustaceans upon bivalve spat in the intertidal zone of the Dutch Wadden Sea as revealed by immunological assays of gut contents. *Journal of Experimental Marine Biology and Ecology* 231, 139-157.
- Van Wesenbeeck, B.K., Van De Koppel, J., Herman, P.M.J., Bakker, J.P., Bouma, T.J., 2007. Biomechanical warfare in ecology; negative interactions between species by habitat modification. *Oikos* 116, 742-750.
- Volkenborn, N., Reise, K., 2006. Lugworm exclusion experiment: Responses by deposit feeding worms to biogenic habitat transformations. *Journal of Experimental Marine Biology and Ecology* 330, 169-179.
- Volkenborn, N., Reise, K., 2007. Effects of *Arenicola marina* on polychaete functional diversity revealed by large-scale experimental lugworm exclusion. *Journal of Sea Research* 57, 78-88.
- Volkenborn, N., Robertson, D., Reise, K., 2009. Sediment destabilizing and stabilizing bio-engineers on tidal flats: cascading effects of experimental exclusion. *Helgoland Marine Research* 63, 27-35.
- Wendelboe, K., Egelund, J.T., Flindt, M.R., Valdemarsen, T., 2013. Impact of lugworms (*Arenicola marina*) on mobilization and transport of fine particles and organic matter in marine sediments. *Journal of Sea Research* 76, 31-38.
- Whitlatch, R.B., Hines, A.H., Thrush, S.F., Hewitt, J.E., Cummings, V., 1997. Benthic faunal responses to variations in patch density and patch size of a suspension-feeding bivalve. *Journal of Experimental Marine Biology and Ecology* 216, 171-189.
- Whitton, T.A., Jenkins, S.R., Richardson, C.A., Hiddink, J.G., 2012. Aggregated prey and predation rates: Juvenile shore crabs (*Carcinus maenas*) foraging on post-larval cockles (*Cerastoderma edule*). *Journal of Experimental Marine Biology and Ecology* 432, 29-36.
- Wiens, J.A., 1976. Population Responses to Patchy Environments. *Annual Review of Ecology and Systematics* 7, 81-120.
- Woodin, S.A., 1976. Adult-larval interactions in dense infaunal assemblages - patterns of abundance. *Journal of Marine Research* 34, 25-41.
- Yankson, K., 1986. Observations on byssus systems in the spat of *Cerastoderma glaucum* and *Cerastoderma edule*. *Journal of the Marine Biological Association of the United Kingdom* 66, 277-292.
- Yasuda, H., Ishikawa, H., 1999. Effects of prey density and spatial distribution on prey consumption of the adult predatory ladybird beetle. *Journal of Applied Entomology* 123, 585-589.
- Zwarts, L., Wanink, J., 1989. Siphon size and burying depth in deposit- and suspension-feeding benthic bivalves. *Marine Biology* 100, 227-240.