OPTIMIZING CULTURED MUSSEL YIELDS: SECOND-SET DYNAMICS AND AVOIDANCE STRATEGIES

CENTRE FOR NEWFOUNDLAND STUDIES

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PATRICK SEAN MACNEILL







Optimizing Cultured Mussel Yields: Second-Set Dynamics and Avoidance Strategies

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By

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School of Fisheries Marine Institute Memorial University of Newfoundland December 2005

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Abstract

A major production constraint for some mussel farms in Canada and elsewhere is related to 'second-set' – an accumulation of unwanted mussel seed on mussel socks. Accumulated seed may originate from primary settlement (annual settlement of mussel larvae) or secondary settlement (post-settled spat that drift) and may be severe enough to decrease growth of production mussels, reduce harvest yields, and increase production costs (extra flotation, transportation and processing costs). The objectives of the present study were to identify the biotic and abiotic factors involved in second-set dynamics through environmental and biological monitoring, as well as current husbandry observations. A multifactorial field experiment was undertaken to examine the temporal (monthly) and spatial patterns (2 sites; 3 m, 6 m, 9 m depth) of larval and post-larval mussel settlement at two commercial mussel farms in Newfoundland in an attempt to understand second-set dynamics. Laboratory trials investigated mussel seed crawling behaviour under varying environmental conditions (food, temperature) with two seed sizes (5-10 mm and 15-20 mm) to explore a possible relationship with second-set accumulation. Finally, the influence of initial socking density (approximately 100, 200, 250, and 300+ mussels per 30 cm), sock deployment depth (4 m and 9 m), time of deployment (spring and autumn) and husbandry practices on the timing and intensity of second-set was examined.

Results indicated that environmental conditions influenced mussel spawning times, with seed collection heaviest during August. The seasonal thermocline may have led to heavy

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seed collection at a depth of 9 m, however, growth of seed was less than at 3 m or 6 m. There was evidence of secondary settlement of post-settled spat (byssal drifting) which may be a source of second-set accumulation. Crawling behaviour of seed mussels was influenced by temperature and seed size, with implications for optimal socking strategies. Second-set accumulation was significantly reduced with higher initial socking densities and with depth of deployment. Fouling was heavier on low density socks. Spring deployments showed the highest sock yields and least amount of second-set accumulation after one year. Socks deployed at 9 m yielded less marketable product per 30 cm of socking than socks deployed at 4 m, yet respectable yields of 70% of gross were attained at 9 m after 1.5 years deployed. Observations of present culture practices indicated a lack of understanding of the impact of environmental conditions and seed handling practices on sock quality. Poorly formed mussel socks had high accumulations of second-set. It is recommended that careful consideration be given to site conditions and mussel seed handling practices when socking. To avoid second-set, it is concluded that high sock quality (fullness, uniformity) be obtained, with consideration of environmental influences on mussel seed quality. For the present study, densities of 250+ per 30 cm of socking at 25-27 mm shell length socked in the spring, deployed in deeper water, at or below the seasonal thermocline, worked well in reducing unwanted accumulations of second-set mussels.

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1.0 Introduction

1.1 Newfoundland Mussel Industry

Mussel culture in Newfoundland occurs on all coasts of the island portion of the province, but is mainly concentrated on the northeast coast. Utilizing various adaptations of the longline system, the industry has experienced slow but steady growth, up to 1,700 tonnes in 2002 (Figure 1).

The south coast of Newfoundland is a relatively new area for mussel culture expansion in the province. To date the south coast remains a promising area with plenty of room to expand in its deep fjords. However, the region possesses unique environmental conditions that pose some technical challenges not experienced on the northeast coast. The south coast generally has warmer waters during the summer months, with frequent fluctuating temperatures (Clemens et al., 2000). Multiple mussel settlements occur throughout the year, leading to large accumulations of mussel seed on production gear. The accumulation of seed resulting from multiple settlements is a significant hindrance to the development of the industry on the south coast as in other areas where it occurs.

2.

1.2 Mussel Culture in Newfoundland

Unlike Europe, where bottom culture, raft culture and bouchot methods are the usual methods of cultivating mussels (Figueras, 1989; Gosling, 1992), mussel culture in

Newfoundland is based on suspended longline technologies, similar to those used elsewhere in Atlantic Canada (Smith and Goddard, 1988; Scarratt, 1993; Mallet and Myrand, 1995). The longline (also referred to as a mainline) essentially consists of a long header line, usually of poly rope (strengths depend upon site conditions, deployment duration, mussel end size – seed or market product) anchored at either end and suspended by plastic floatation. Longlines are typically 250-300 m long, but can be more than 400 m, depending upon the site. The lines can be held in position near the surface by the floats or sunk to a desired depth by adjusting flotation type and using weights (Mallet and Myrand, 1995). Mussel 'socks' – mesh tubes filled with mussels attached from a longline, or mussel seed collector ropes, are attached to the longlines (Figure 2).

The process of cultivating mussels involves three main steps: seed collection, socking and harvesting (Figure 3). Each mussel producing area has unique techniques, equipment and methods for each step, and is very often influenced by the species being cultured, the environmental conditions in which the mussels grow and the scale of the operation. As Mallet and Carver (1991) pointed out, the basic principles of longline culture are well established, however, there is continued development and evolution to techniques used that are often unique to an area.

1.2.1 Seed Collection - Larval Monitoring and Collector Deployment

Whether growers collect their own seed each year or purchase seed, it must be collected each season to supply the grow-out farm sites. In Newfoundland and elsewhere,

wild seed collection is practiced throughout the industry. There is abundant seed, which is relatively inexpensive to collect, as opposed to other means, such as hatchery production (Penney, 1993). However, proper technique is required to ensure reliable annual collection. Larval monitoring plays a crucial step in the mussel culture process. Mussels are monitored for signs of spawning (visual and meat yield tests) and plankton tows are carried out each week, from late June through to late August, to determine the relative mussel larval abundance, stage of development and readiness to settle on a substrate (Figure 4) (Macneill et al., 2000).

When mussels are determined to be at the settlement size (250 μ m in shell length – refer to section 1.3), collector ropes are deployed on the site. There are many different types of collectors, but generally they consist of poly rope of varying lengths hung from a longline at about 30-60 cm (1-2 feet) depth. Depending upon the number deployed and efficiency of the operation, collector deployment can take from one week to three weeks.

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1.2.2 Socking – Size Grading and Deployment

Under normal Newfoundland conditions, after 12-14 months of growth on the collector ropes, mussel seed is ready for socking. Socking involves several major steps: stripping the seed from collectors, size grading and deployment in 'socks'. Depending upon the site conditions, socking takes place from April through early June and again in late August through to November. Stripping of collectors can be done manually, but recent development of mechanical seed strippers has led to higher efficiencies in clearing

collectors. Stripped seed is then size graded using mechanical drum graders. Depending upon the size of the drum and bar width, there are 4 to 5 size grades, with the first very small grade (< 10 mm) usually discarded. Mussel seed graded to about 20-30 mm are most often preferred to be used on most farm sites.

It is important to pay particular attention to how seed reacts to the grading and socking process to minimize stress on the animal (Harding et al., 2004). While the temperature range for mussels is noted from -2°C to 25°C (Mallet and Myrand, 1995), some growers will not size grade their seed mussels if the water gets too warm (e.g., >18°C) because of high mortality. Once enough seed is graded into a particular size, the seed is socked.

There are two main types of socking methods used in Newfoundland. The first is called traditional socking – using mesh tubes that are filled with seed mussels and hung on a mainline and left to grow to market size (Figure 5A). Although an older method, this method is commonly used throughout Atlantic Canada (Mallet and Myrand, 1995). There are many adaptations to the traditional socking that make it fast, economical and high yielding.

The second method is continuous socking (Figure 5B), often referred to as New Zealand-style socking (Jenkins, 1985; Hickman, 1989). The continuous method utilizes a single continuous rope (usually fuzzy or used crab rope) as a central support or core for the mussels to attach to and it is wrapped with a dissolvable 100% cotton or cotton/poly mix mesh. As the 'sock' is filled, the rope is attached to the mainline in determined loop lengths. The continuous method generally produces a lower yield per unit (i.e., kg per 30 cm of sock) marketable product, however, it is very efficient and yields are often very

uniform looking. This technology has been in use on a small scale in Newfoundland but was introduced to Atlantic Canada during the early/mid 1990's and is found to be highly successful in deeper water sites (Darnell, 2000).

1.2.3 Grow-out and Harvest

After socking is completed, mussels are left to grow to market size. While market requirements differ from location, the grow-out time is usually 12 - 20 months 'in-sock' to produce a mussel 55-65 mm in shell length destined for the fresh live market. Grow-out times can vary with site, often being influenced by some combination of environmental conditions and overall biomass on the farm (McNeil, 2003). The standard methods for harvesting involve a harvesting barge and various sized containers to put mussels into. A typical single harvest is around 12,000 kg gross. Primary processed yields of 70% of gross weight are considered acceptable, but yields above 80% may be achieved.

1.3 Mussel Biology

In Atlantic Canada, there are two species that co-exist at commercial farming operations – *Mytilus edulis* and *Mytilus trossulus* (Koehn et al., 1984; Mallet and Myrand, 1995; Mallet and Carver, 1999, 2000; Landry and Tremblay, 2000). In Newfoundland, there have been several studies on the distribution and characteristics of the two species around the island (Innes et al., 1999; Penney and Hart, 1999; Struthers et al., 2002). Depending upon the location, either *M. edulis* or *M. trossulus* can be the dominant species, with some hybridization being noted (Bates and Innes, 1995; Innes and Bates, 1999; Innes et al., 1999; Penney and Hart, 1999; Toro et al., 2002). Similar in shape and colour, the shell of *M. trossulus* is slightly narrower than *M. edulis*, and there may be some differences in spawning and settlement behaviours (Innes et al., 1999; Freeman et al., 2002).

Mussels have separate sexes, however, one cannot distinguish them by external appearance. Gender is easily distinguished by the colour of the gonad tissue in reproductive adults – females are orange and males are creamy white (Figure 6). During late spring and early summer (April to June), phytoplankton blooms accompany a rise in water temperature and trigger reproductive development (Mallet and Myrand, 1995).

Spawning times vary with location and site condition but can typically start by late June when water temperatures exceed 5°C (Sutterlin et al., 1981; Bernard, 1997; Pryor, 2005). More than one spawning has been observed on many sites and most often occurs by late September through early October (Macneill et al., 1999). Mussels can spawn at temperatures lower than 10°C, occurring frequently in PEI (Bernard, 1997). Other factors, such as phytoplankton blooms (food abundance), spring tides, water currents, storm disturbances and salinity fluctuations may also influence spawning (Starr et al., 1990).

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Evidence of spawning at a farm site can be a sudden rise in flotation equipment, due to the release of eggs and sperm and subsequent weight loss in the mussels (Mallet and Myrand, 1995). Gametes are expelled and fertilization is external (Figure 7 – life cycle). Within a few hours, free swimming trocophore larvae develop (Bayne, 1965;

Rodhouse et al., 1984). Growth and survival of *M. edulis* larvae is reported to be both temperature and salinity dependent, with high mortalities occurring at temperatures above 25°C and 100% mortality at 30°C, within a 5-40 ppt salinity range, in one laboratory study. Growth was optimal at 25-30 ppt and 20°C water temperatures (Brenko and Calabrese, 1969).

Within a few days of fertilization, mussel larvae develop from trocophore larvae to a feeding veliger or 'D-stage' larvae. Veliger larvae of approximately 100 μ m in length were typically the first to be observed in the plankton tows under the NAIA mussel larval and spatfall monitoring program, which used 100 μ m mesh plankton nets (Macneill et al., 2000). After about 3 weeks, veliger larvae develop an umbone, a muscular foot for crawling and are subsequently called pediveliger larvae. These have a distinct appearance of shell - prodissoconch shell, and sizes range from 200-300 μ m shell length. When nearing settlement time, pediveligers develop paired eye spots (Bayne, 1965).

When pediveliger larvae reach ~250-300 µm in length, they seek a suitable location to settle. Pediveligers can delay primary settlement and metamorphosis for up to 5-6 weeks if a suitable substrate is not found (Bayne, 1965, 1976). Some factors influencing the length of time larvae remain planktonic include food abundance, water temperatures, salinity, currents and lack of suitable substrate (Gosling, 1992; Young et al., 1996; Snodden et al., 1997; Pernet et al., 2003). Mortality increases significantly with length of time mussels delay primary settlement (Widdows, 1991). Recent studies indicate that larvae prefer to settle on rough or filamentous surfaces, with greater settlement occurring in areas of higher current or water agitation (Young, 1983, 1985; • • •

Eyster et al., 1987; Pernet et al., 2003). Dobretsov and Wahl (2001) found preferential differences in spat settlement of blue mussels between artificial and natural substrata, as well as amongst microhabitat composition of natural substrata.

Once a suitable substrate is found, the larval velum disappears and secretion of byssal threads occurs. Larvae attach to the surface and metamorphose into juveniles by secreting the dissoconch, or adult shell, and at this stage are referred to as *spat*.

Spat that have settled can migrate or detach and re-attach several times, if conditions become unfavourable. This is referred to as byssal or byssus drifting (Lane et al., 1985). Spat sever their byssal attachments and go adrift with the water currents, using their byssal threads and foot as 'sails' (De Block et al., 1977; Newell et al., 1991). The detachment and byssal drifting may occur for up to 8 months after primary settlement has occurred and spat may reach a size of 3 mm before they settle again in a more permanent fashion. Sigurdsson et al. (1976) briefly described water current induced byssal thread secretion as a means of transport of young mussels by the currents. Reattachment of post-settled mussel spat is referred to as *secondary settlement*.

There has been considerable discussion on whether larval settlement, post-larval settlement and general dispersal is active (behavioural and have control over where they settle) or passive (larvae are under the direct influence of environmental conditions) (Scheltema, 1986; Pineda, 2000). Stage of development (Dobretsov and Miron, 2001), tides, currents (Levin, 1986; Newell et al., 1991) and boundary layers (e.g., thermoclines, haloclines and pycnoclines) (Mann et al., 1991; Raby et al., 1994; Manuel et al., 2000) have all been investigated as influences on larval distribution.

1.4 Second-Set Explained

The term 'second-set' refers to an accumulation of mussel seed on mussel production gear. The accumulation of seed may be light (sparse) and temporary, or quite severe (dense covering mussel socks) over the long-term such that by the time mussel socks are ready to harvest, they resemble mussel collector ropes being completely covered in seed (Figure 8).

Accumulating seed may originate from two main sources – *primary and secondary settlement* (described in previous section). Mussel socks can become coated with mussel spat from the annual settlement process, or *primary settlement*. In Newfoundland, generally there is one large primary settlement that occurs each season, usually from late June through early August, however, multiple primary settlement events (i.e., trickle spawning) have been known to occur throughout the year in some locations (Macneill et al., 2000; Pryor, 2005). This can lead to an extended period of primary settlement, often overlapping with prime socking conditions. The impact of *secondary settlement* (newly settled spat that detach and re-settle again – e.g., byssal drifting) as a sole source of accumulated seed has not been investigated.

Sites with or without a significant amount of gear deployed can have severe second-set problems. Current information from the NAIA mussel larval and spatfall monitoring program (Macneill et al., 2000), mussel extension program, line inspections and harvest yields indicate that some sites on the south coast of Newfoundland have a high occurrence of second-set as do some sites located in Notre Dame Bay of central Newfoundland. However, sites elsewhere in Atlantic Canada also experience second-set, such as farms in Gaspé, Québec, where studies have been carried out to understand the effects a second-set has on overall production yields (Bourque and Myrand, 2002).

The dynamics of second-set are just beginning to be understood. Depending upon the farm site, second-set may occur from the surface to depths exceeding 15 m and the severity of the settlement and impacts from this may vary with location within a site because of differences in tidal patterns, current directions, environmental conditions or settlement events.

1.4.1 Problems Associated with a Second-Set

Second-set can become a problem if the accumulation is dense enough. There are several major effects: on production gear itself (line stress and maintenance costs), reduced growth and performance of the mussels as well as increased processing costs compounded by lower primary processed yields. Second-set can greatly affect how many floats must be purchased and used, the timing and associated maintenance costs of adding extra flotation. Added weight can also affect stress load on mainlines. Traditionally, large eye bolts are secured into large boulders or cliffs. While they have tension strengths of up to 15,000 kg, the addition of second-set mussels on float ropes, floats, mainlines and socks can cause tremendous strain, resulting in loss of significant amounts of product if lines fail.

On mussel socks inundated by second-set mussels, the larger mussels are usually the first to fall off, thus leaving the sock with a high percentage of smaller mussels (unpublished observation). The high density per unit area suggests an eventual limitation of food and/or space, leading to high drop-off or 'thinning' of socks (Fréchette et al., 1992). Thus a sock covered in second-set will continuously shed larger product, resulting in low primary processed yields. Competition for food and space may lead to slower overall growth. One study indicated seeded beds of mussels at high densities show slower rates of growth (Beadman et al., 2003).

Perhaps the greatest impact of second-set is the increased production costs. Farmers see increases in production costs through equipment and labour, but processors also see increased costs through increased transportation costs of raw product, longer processing times, greater difficulty in removing unwanted, undersized product and poor end yields. In addition, there are the associated costs of dumping waste after processing.

1.4.2 Remedial Strategies

In areas where second-set has been problematic, mussel growers have attempted to remedy the problem by several methods. Strategies employed include grading harvests before shipping to the processing plant and re-socking the smaller grades. In New Zealand, seed mussels are frequently graded and re-socked using the continuous method, to ensure uniform product and to separate out unwanted blue mussels from the greenshell mussel (Jenkins, 1985; Hickman, 1989; Hearn, 2002).

In shallow water sites, such as in PEI, some growers will temporarily lower their lines down to the bottom. This will allow crabs, lobster and other predators to clean off

any fouling that has accumulated on the socks, including much of the smaller second-set mussels.

Removing undersized mussels on site initially seems like a logical proposition, saving money on transportation costs and processing costs at the plant. However, grading at harvest may have several negative effects. Increased stress on mussels through extra grading is possible, especially if mussels are near spawning or have just recently spawned. This may lead to potentially higher mortality or spawning en route to processing or market, as well as decreased shelf life of product. Stress levels are considerably elevated under warm weather conditions and increased handling (Harding et al., 2004).

On site grading may also increase the percentage of marketable product with fractured shells and/or abrasions, which may upon primary processing, lead to lower product yields. Mussels have been shown to have growth spurts, especially during the springtime, upon which the new shell growth is fragile. Extra handling during this time may increase breakage. The extra costs associated with grading out on-site may make this practice uneconomical.

1.4.3 Important Considerations in Developing an Avoidance Strategy

The best solution to second-set is avoidance. To do this, a better understanding and knowledge of the dynamics of second-set must first be gained. Where does secondset occur on any given site? When does it occur? Can it be avoided or controlled easily?

An avoidance strategy begins with a thorough knowledge of the physical and oceanographic site characteristics and effects they may have on mussel biology – in particular, the mussel spawning/recovery and settlement cycles. In addition, mussel behaviour, particularly early post-settlement and how mussels respond to handling and gear types that are to be used. For example, for single drop socking, proper sock formation is critical in producing a high quality product. A poorly formed sock (e.g., non-uniform fill) may be subject to heavy second-set coverage. Socked mussels are required to crawl out of the sock, thus understanding what effects the mobility of the seed is very important when considering a socking strategy.

There are surprisingly few studies in the literature where crawling behaviour of mussels has been examined. Most studies are on populations, predator/prey relationships and changes to population structure through competition. For example, for competition between species, juvenile *M. edulis* were observed to crawl to the exterior and form clumps over *M. californianus*, where both species exist in quiet waters. This behaviour is seen as an adaptive advantage of *M. edulis* over *M. californianus* as it seeks to remain free of silt that accumulates within the mussel bed (Shaw et al., 1988). As another example, chemical cues were viewed responsible for the clumping behaviour of mussels due to predation by lobster (Cote and Jelnikar, 1999). Competition for food and space, as described in the self thinning concept (Fréchette and Lefaivre, 1990; Fréchette et al., 1992) is another example of indirect mussel mobility behaviour investigations, in this case, movement of mussels from concentrated populations due to food and/or space limitations. Finally, Sullivan and Couturier (2004) examined crawling behaviour of *M*.

edulis and *M. trossulus*, as part of a species specific investigation of byssal thread production under varying salinity and food regimes.

It is important to consider mussel mobility when developing an optimal socking strategy. Seed size, mesh type and size, density to use for socking, time of year and how crawling behaviour is affected, to handling and site conditions (temperature, food, salinity, etc.) are all important considerations. Through good husbandry, knowledge of mussel biology, site characteristics, environmental change and how handling techniques affect behaviour and performance, a strategy for avoiding second-set can be developed. There are six potential options to explore:

(1) Sinking lines below the main mussel settlement depth. The literature supports the notion that shellfish larval settlement is not random, but occurs at specific depths and locale relevant to the species' life history and local environmental conditions (Sutterlin et al., 1981; Pennington and Emlet, 1986; Tremblay and Sinclair, 1990; Newell et al., 1991).

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- (2) Socking at a higher density to limit opportunity space for mussel larval settlement or any secondary settlement.
- (3) Socking *before* a major wave of settling mussel larvae or small spat occurs. This gives newly socked mussels ample time to migrate to the outside of the sock and grow enough so that settling larvae either have no space to settle or are consumed, instead of settling on socks.

- (4) Sock *after* a major wave of settling mussel larvae or spat has passed. Socked mussels will have ample time to migrate to the outside of the sock and grow to a size capable of consuming any future waves of settling mussel larvae.
- (5) Choosing a site with historically low mussel settlement for grow out.
- (6) Some combination of the above.

In order to explore the above options, a comprehensive set of trials was developed to evaluate all possible solutions.

1.5 Rationale

Observation of the current husbandry practices on the south coast operations indicated that a step by step protocol is followed in the seed collection-grading-socking processes. However, this protocol is not producing high yielding product, with second set being a major contributor to poor yields. There appears to be a general lack of understanding of the environmental and biological interactions in the area under investigation. To date, only trial and error adjustments to husbandry practices to mitigate the effects of second-set have been carried out.

The rationale behind the present series of experiments was to identify the biotic and abiotic factors involved in second-set dynamics through environmental and biological investigations, as well as current husbandry observations. By gaining a better understanding of second-set, proper husbandry techniques can be developed for operations prone to second-set. Field trials were carried out to determine which strategy works best. Recommendations were made to industry and the most economical and practical strategies were employed.

1.6 Objectives

Overall Objective:

To make recommendations to industry on strategies that help reduce or eliminate the negative impacts of second-set on single drop sock production gear.

Primary Objectives:

(1) To evaluate the crawling behaviour of different mussel spat sizes under varying temperatures and food conditions. This is to assess whether crawling behaviour of different sized spat increases with temperature and presence of food. The resulting observed behaviour may indicate how mussel sock arrangement can affect second-set accumulation on mussel socks.

- (2) To evaluate environmental conditions within the study area and relate to mussel spawning, larval settlement and growth as well as potential for second-set problems. It is hypothesized that the appearance of second-set on mussel socks is directly related to mussel spawning events.
- (3) To evaluate the influence of socking density, depth of sock deployment and time of year (spring versus autumn) of deployment on the amount of second-set
accumulation. It is hypothesized that the initial socking density, depth of sock deployment and timing of deployment will affect the amount of second-set accumulation, either independently or in some combination.

- (4) To evaluate the effect of changes in socking strategies on mussel sock yields. A strategy for avoiding second-set may influence mussel growth and thus impact upon harvest schedules of marketable product. It is hypothesized that sinking socks in deeper water and socking at higher initial sock densities per 30 cm will increase the length of time required to grow out to market size. It is hypothesized that deploying mussels in the spring will decrease the time required to produce a marketable product.
- (5) To evaluate husbandry practices and their potential influences on second-set accumulation. It is hypothesized that excess handling of mussels during the socking process will lead to increased second-set accumulation on mussel socks.

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2.0 Methods

2.1 Seed Crawling Behaviour in Response to Water Temperature and Food Supply

2.1.1 Experimental Set-up

The trial took place at the Ocean Sciences Centre (OSC), Logy Bay. Small rectangular plastic trays (observation trays) measuring approximately 40 cm x 30 cm x 10

cm (LxWxH) were placed in triplicate into two larger touch tanks. The water in the touch tanks served as a temperature control – warming through decreasing flow, or cooling by addition of ice. A total of twelve trial trays were used (Figure 9) – three replicates of four treatments: small seed fed and unfed as well as large seed fed and unfed. Each replicate tray had a grid drawn on the bottom consisting of numbers and letters that helped keep track of mussel movements (Figure 10). Small air stones were placed inside each replicate tray for aeration. A thermometer placed in each touch tank monitored temperature.

Before the experiment was started, a few days were spent observing mussel crawling behaviour in a tray. Preliminary observations showed that when many mussels were placed in the small observation tanks, they would form a tight aggregation and not move at all. Thus, it was decided to cut the number of mussels in each tank to five because it would be too hard to keep track of them individually with more present.

2.1.2 Animals

Mussel seed were obtained from Farewell Mussel Farms, located in Notre Dame Bay in January. Approximately 60 kg were harvested and brought to the lab for various student activities and were held in flow through holding tanks at the Ocean Sciences Centre. The mussels were held in flowing ambient sea water (at 0-4°C), salinity 30-32 ppt, unfiltered and aerated until used for the trials. They were not fed.

Mussels that were termed 'small' in the trials had mean shell lengths of 5.33 mm and 5.35 mm, with standard deviations of 0.84 and 0.44 mm, respectively (Table 1) and mussels termed 'large' had mean lengths of 19.1 mm and 19.2 mm, with standard deviations of 1.1 mm and 1.3 mm, respectively (Table 1). Each mussel was marked with quick drying nail polish of various colours to allow for easy identification in the observation tanks.

2.1.3 Temperatures

Three different water temperatures were used – 0°C, 5°C and 10°C. Temperature was controlled by increasing or decreasing flow to the water bath and allowed to warm before the trial started and/or by adding ice to water bath. Water used in the trials was filtered seawater (50 μ m) and 0-1°C ambient.

2.1.4 Algal Culture

In order to observe the effects of food on mussel crawling behaviour T-ISO (*Isochrysis* spp.) algae was added to each observation tray. The algae were cultured at the Ocean Sciences Centre and had a final cell count of 5.6×10^5 cells/mL when used. A small graduated cylinder of 10 mL of T-ISO was added a few minutes before the start of the trials to each of the trays.

2.1.5 Sampling Schedule

Mussels were placed at starting coordinates (e.g., A-3) of the grid-pattern marked on the bottom of the trays at time 0 (T_0). At 15 minute intervals (120 minutes total), the new coordinates of each mussel were recorded. Trials were repeated for each temperature, with and without the presence of food. At the end of the trials, the coordinates at each time interval were plotted on graph paper. Distances between coordinates were then measured (Figure 10) using a ruler and added up to get the total distance travelled over the 120 minutes.

2.1.6 Data Analysis

For each treatment, the total distance traveled by each mussel was determined. As a means of standardization, distances were expressed in mussel body lengths per hour. Two-way ANOVAs ($\alpha = 0.05$) were performed on each to examine the relationship of crawling rate to changes in water temperature and seed size. One-way ANOVAs ($\alpha =$ 0.05) were carried out on crawling rate versus food supply for each size separately. A Tukey's-b test was performed where necessary to determine any significance among treatments.

2.2 Mussel Spawning and Spat Settlement Dynamics at Salmonier Cove and The Tickle, Connaigre Bay

2.2.1 Study Sites

The sites chosen for studying second-set dynamics were Salmonier Cove and The Tickle, both in Connaigre Bay on the South Coast of the province. Both were relatively new areas of mussel culture and had poor yielding harvests with heavy second-set.

The mouth of Connaigre Bay is open to the Atlantic Ocean and subject to large swells and strong ocean currents. The farm sites used in the studies are located within fjords opening south-south west and have water depths of 15 m to more than 75 m in places (Figures 11 and 12). With such depths, water temperatures are greatly affected by wind. Warm surface waters may be displaced by wind, causing an upwelling of cold water (Tomczak and Godfrey, 1994; Stewart, 2003). Large fluctuations in temperature are common on the south coast (Clemens et al., 2000) and seasonal thermoclines may exist and shift depth with currents, tides and wind.

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2.2.2 Sampling Schedule

Field work for the project took place from late April/early May through November, 2000 and 2001. Environmental and biological information were recorded approximately once per month, with larval abundance information being recorded approximately every two weeks, with assistance from farm employees.

2.2.3 Environmental and Biological Monitoring

2.2.3.1 Water Temperature

Temperature data were recorded with thermographs (Vemco Ltd, Shad Bay, Nova Scotia). On each study site, three thermographs were attached to a length of poly rope at intervals of 3 m, 6 m and 9 m and then fixed to the mainline rope. The devices were set to automatically record temperature every hour for approximately one year. They were deployed during the period of 2000-2001 at both sites.

To determine the dynamics of water temperatures at the study sites, results of the thermograph data were cross-referenced with sea surface temperature satellite images for Atlantic Canada, created by the Department of Fisheries and Oceans (DFO), Canadian Hydrographic Service unit. Although only temperatures observed at the surface, the DFO data gave an overall description of changing (or stable) water temperatures for the entire region for the period of study.

2.2.3.2 Monthly Water Column Profiles

A Seabird SBE 25 CTD (Conductivity-Temperature-Depth) profiler was employed to gather additional environmental data at each of the study sites. The Seabird was lowered down through the water column, and provided a profile of environmental conditions at the site. The CTD probe recorded temperature ($^{\circ}$ C), salinity (ppt) and chlorophyll-a (µg/L) or total chlorophyll (µg/L). Three areas of each study site – inside, middle and outside – were visited once a month from May through November and depth profiles were recorded (Figure 12). All data were then summarized and plotted to give a monthly summary of the water column conditions.

2.2.3.3 Meat Yield and Larval Abundance

The timing of mussel spawning and larval settlement was monitored by performing meat yields and plankton tows at least once per month. Although a crude measure of the reproductive cycle, meat yields were quick to perform on-site and gave a general indication of the approximate time of spawning. Approximately 1 kg of marketable mussels was steamed for 10 minutes for each yield carried out (see Ibarra et al., 2000). Vertical plankton tows were carried out for depths of 10 m and larvae counted per mL of sample (see Macneill et al., 2001). Results were plotted to identify patterns in the timing of mussel spawning and mussel settlement.

2.2.4 Spat Settlement

2.2.4.1 Monthly Spat Collector Deployments

To determine the timing of spat settlement, four spat collectors were deployed for a period of 4 weeks, retrieved and replaced with new collectors. While overlapping time sets would have been ideal (i.e., two week intervals), logistical difficulties prevented this option. They were deployed at approximately 30 cm depth (Figure 13). This was carried out from May through to November, 2000 and again during the same period for 2001.

2.2.4.2 Cumulative Spat Collector Deployments

To determine the cumulative effect of mussel settlement over time, 12 collectors per month, 4 each at 3 m, 6 m and 9 m, were deployed along an experimental mainline at Salmonier Cove and The Tickle (Figure 14). Given the currents and tides may differ from one side of the site to another, collectors were deployed in single sequence groups of 3-6-9 m to allow for within site variation in spatial distribution. Collectors were allowed to remain in the water the full field season, upon which three collectors from each depth, each month ($3x_3x_5 = 45$ collectors each site – 90 collectors total) were retrieved on Oct 29 – Nov 1, 2000, cleaned of organisms, weighed and frozen. These were brought to the lab, thawed and processed throughout the autumn and winter of 2000. Another 12 collectors were deployed on both sites in the late autumn and left over winter. Time and logistic constraints prevented a repeat of these particular trials in 2001.

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2.2.4.3 Sampling

Spat collectors were deployed for approximately one month and were stripped of seed, with estimates of spat collected on each collector determined by measuring spat per cm^2 and total length estimated per collector. Spat were also measured for shell length to the nearest millimeter in an attempt to determine growth for each month (n = 200).

Cumulative collectors were also stripped of seed, with estimates of spat per 30 cm of collector rope and full length determined. Fouling was observed and species identified where possible. Fouling was listed as very light, light, moderate or heavy, with numerical values assigned as follows:

	%		
No.	Coverage	Observation	Description
1	0-25% coverage	Very light	Sparse covering over collector rope with rope clearly visible, or containing mostly mussels.
2	26-50% coverage	Light	Partial cover by one or a few species, or a dense patch covering collector in one or more small spots.
3	51-75% coverage	Moderate	Several species covering most of collector, some patches without any mussels and/or mussels attached to fouling, not collector rope. Surface area of rope noticeably increased.
4	76-100% coverage	Heavy	Many species covering all of collector in dense concentrations. Mussel attachment restricted to fouling only. Surface area of rope increased greatly.

2.2.4.4 Data Analysis

For this part of the field trials, the month of greatest settlement was established for the sites by estimating accumulated spat numbers per collector. For the cumulative collection, a two-way ANOVA ($\alpha = 0.05$) was used to examine the relationship between spat growth rate (mm/day) for each month and depth of collector deployment. One-way ANOVA and Tukey's-b tests were used where necessary to determine significance among treatments. Mean spat size per month and depth was also determined for cumulative collectors deployed at each site.

2.3 Socking Trials

A series of socking trials were undertaken to determine the amount of second-set accumulation using three factors: mussel socking density per 30 cm of sock, depth of sock deployment (4 m and 9 m) and timing of deployment (spring 2000 and autumn 2000). The timeline for the field trials was approximately one full production cycle, or approximately 1 to 1.5 years in-sock.

2.3.1 Study Sites

Socking trials were carried at Salmonier Cove and The Tickle, Connaigre Bay (Figures 11 and 12). For each site, one line was chosen as an experimental line for deployments.

2.3.2 Experimental Set-up

A reinforced mesh square mesh material (Go Deep International) was used for the socking trials. The mesh size was approximately 1 cm² and by varying the sock diameter, mussel density could be varied easily. The sock materials used were: 4M - 4 cm diameter, 5M - 5.5 cm diameter, 6M - 6 cm diameter and $7M - \sim 7.5$ cm diameter and

TMM - 7.5 cm diameter. The 7M sock material had a reinforcing strip woven into each side of the sock, while the TMM did not. Table 2 shows the socking densities used for the socking trials. Densities ranged from approximately 100 mussels per 30 cm to just over 300 mussels per 30 cm of socking.

At Salmonier Cove and The Tickle, one single mainline was used near the outer end of the farm site. There were 80 socks deployed for the spring (May) 2000 trial, 40 socks deployed at 4 m depth and 40 socks deployed at 9 m depth. There were four sock densities used at each depth and 10 socks deployed at each density. As there may have been differences in site dynamics across the farm, the socks were deployed in 10 groups of 4 socks (40 socks, with each of the 4 socks representing a trial density) (Figure 15). Colour tags were used to identify the individual socks at each trial density (example – Red = 4M, Green = 5M, Blue = 6M, Yellow = 7M or TMM). The same set-up was used again for the autumn 2000 socking trials (end of September 2000/early October 2000). A large clear plastic strap was fixed to each sock for identifying the autumn deployments. Coloured plastic straps identified each trial density (example – Red = 4M, Green = 5M, Blue = 6M, Yellow = 7M or TMM).

2.3.3 Mussel Seed

Mussel seed used for the socking trials originated from Salmonier Cove and The Tickle (i.e., local stock). Seed was harvested from collectors and graded for size uniformity. For the spring 2000 trials, a mean seed shell length of 25 mm (n = 200) was

used. For the autumn 2000 trials, a mean seed shell length of 27 mm (n = 200) was used (Table 2).

Seed was graded using a vertical bar grader (Salmonier Cove – spring 2000) and a horizontal bar grader (The Tickle – spring 2000 and autumn 2000, Salmonier Cove – autumn 2000). For each site set-up, seed was harvested, graded and socked on the same day.

2.3.4 Sampling

Experimental socks were sampled at approximately 6 month intervals. The socks deployed during the spring 2000 were sampled during the autumn 2000, spring 2001 and autumn 2001. The socks deployed during the autumn 2000 socking were sampled during the spring 2001 and autumn 2001. At each sampling time, entire socks were weighed (kg) in the field, length measure (cm), a section cut out (approximately 30 cm), weighed (kg), placed in a heavy weight plastic bag and labelled. Triplicate samples (3 socks) were obtained for each sock density used, depth and deployment time. Sections were kept chilled until frozen at the Marine Institute or in Hr. Breton, until processing could be carried out.

At the Marine Institute, frozen sections were weighed (kg) and recorded. Fouling was removed and weighed (g) as well. Samples were divided into two size categories: small mussels (≤ 25 mm) and large mussels (≥ 25 mm). Each size separation was weighed and a percentage of the total sample weight calculated for each category. A random

sample size of 200 mussels was measured for length (mm) based on the proportion of mussels from each category. For example if there were 50% large and 50% small in the sock subsample, then 100 mussels from each size category were randomly measured.

The number of mussels per 30 cm of sock was calculated by first comparing the frozen subsample weight at the Marine Institute to the same sock subsample wet field weights. Mussels in the subsample were size separated, counted, weighed and individual mussel weights could be calculated. As frozen mussels drained when thawed, these individual mussel weights were then used to calculate the individual weights of mussels in the sample before they were frozen. Once wet weights of individual mussels were determined, the number of mussels per 30 cm of sock was calculated.

2.3.5 Data Analysis

Estimates of number of mussels per 30 cm of socking for each density, deployment depth and deployment times were made and graphed for comparison. Twoway ANOVAs ($\alpha = 0.05$) were carried out on sock density versus depth of deployment during spring versus autumn deployments and one-way ANOVAs ($\alpha = 0.05$) were carried out on mean mussel size with each sock density, deployment time and depth to determine the relationships of deployment treatments with mussel growth. The effect of each treatment on the amount of marketable (mussels >50 mm in shell length) product was assessed through a comparison of sock weight per 30 cm of socking. A two-way ANOVA ($\alpha = 0.05$) was used to determine any significance in amount of marketable

product among the treatments. For Salmonier Cove, spring 2000 deployment, only socks at 9 m remained, thus a one-way ANOVA ($\alpha = 0.05$) was carried out to determine any significance in sock weights and sock mesh type. Percent harvest yield (% by weight of mussels > 50 mm shell length) was calculated for each treatment.

2.4 Farm Husbandry Observations

Farm workers were observed in all aspects of the socking process - seed harvesting, handling practices, grading and socking itself (mesh type chosen, handling, etc). Summaries of procedures with possible links/contributors to second-set were made.

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3.0 Results

3.1 Seed Crawling Behaviour in Response to Water Temperature and Food Supply

3.1.1 General Observations

Throughout the trials, some qualitative observations were made regarding mussel crawling behaviour. The following describes some noteworthy observations. Mussel seed were observed crawling by first extending the foot and then contracting, pulling the body along. There was no apparent directional pattern to their movements as they moved in all directions. Occasionally, they attached themselves to the trays by producing new byssal threads. At the start of the trials, individuals were placed in a line, on known coordinates of the grid on the bottom of the trays. Throughout the 120 minute trials, no apparent clumping behaviour was observed or general movement toward aeration devices, with or without food present, or with changing temperatures. Some mussel seed moved up the side of the tray and off the grid, where they remained until the end of the trials. The total distances (mm) traveled under each condition for each individual for the 120 minute period was summarized (Figure 16). The mean body lengths per hour traveled by both small and large spat at each trial temperature, with and without food being present were summarized (Figure 17).

3.1.2 Seed Size and Water Temperatures – Absence of Food

There were significant differences between crawling rate (mean body lengths per hour) with spat size (Two-way ANOVA $F_{(1, 84)} = 64.92$, p < 0.001). Small spat averaged 16.4 body lengths per hour, while large spat averaged 1.59 body lengths per hour. There was no significant interaction of temperature and seed size ($F_{(2, 84)} = 2.95$, p = 0.057).

Water temperature was shown to significantly influence crawling rate of small spat (one-way ANOVA, $F_{(2, 44)} = 3.89$, p = 0.028), but not large spat (one-way ANOVA, $F_{(2,44)} = 1.18$, p = 0.318). Small spat at 0°C crawled an average of 13.34 body lengths per hour, but averaged 22.63 body lengths per hour at 10°C. Large spat averaged 1.23 body lengths per hour at 0°C and 2.04 body lengths per hour at 10°C.

3.1.3 Seed Size and Water Temperatures – Presence of Food

Visually comparing overall distances traveled (mm) by large and small mussel seed, both large and small spat appeared to travel further with the addition of food and in warmer water (Figure 16). However, a two-way ANOVA analysis showed that with food available in the water, there was a significant difference in mussel seed size and amount of crawling (F $_{(1,84)} = 50.77$, p < 0.001). However, there was no significant difference in crawling due to temperature change (F $_{(2,84)} = 1.73$, p = 0.183), as well as no significant interaction of temperature and seed size (F $_{(2,84)} = 1.61$, p = 0.206) with food present. Thus, crawling was independent of temperature when food is present.

To further investigate, one-way ANOVAs comparing crawl rate with food present in small seed and with large seed were carried out carried out separately. The results indicated that for small seed, adding food had no significant impact on crawling rate (p = 0.76) as was the same for large seed with food (p = 0.30).

3.2 Mussel Spawning and Spat Settlement Dynamics at Salmonier Cove and The Tickle, Connaigre Bay

3.2.1 Environmental and Biological Monitoring

3.2.1.1 Water Temperature

For Salmonier Cove, thermographs at 3 m and 9 m were lost. Poor weather prevented exchange of thermographs during the autumn 2000, so there was a period without data from January to June, 2001 (Figure 18).

Overall, water temperatures increased steadily throughout the summer, peaking at 18-19°C by late July/early August, for both 2000 and 2001. However, the warm-up and cool-down periods differed for each year. For example, temperatures for late spring (June) 2000 at Salmonier Cove were about 3°C higher than the same period for 2001, but early July temperatures were about 3°C cooler in 2001 than for the same period of 2000 (Figure 18). Late autumn 2001 temperatures were higher than the same period during 2000 for The Tickle site. Winter data were available for The Tickle site, and it was noted that temperatures did drop to -0.5°C during February (Figure 19).

Where depth is concerned, water temperatures at 9 m and 6 m were lower than at 3 m, but still climbed above 16°C during late July/early August for both years, at both sites. There were large fluctuations in temperature throughout the monitoring period, but were fewer during the winter period. Summarizing the 2000 data for The Tickle into 24 and 48 hr periods, the relationship of depth on temperature stability is made more clearer (Figure 20). Temperature change was clearly less pronounced at 9 m than at 6 m or 3 m.

To investigate whether placement of the thermographs may have impacted the temperature differences observed in 2000 and 2001, or due to some widespread pattern, satellite images showing sea surface temperatures were obtained from the Canadian Hydrographic Service unit at Fisheries and Oceans Canada (DFO). Figure 21 shows a 2-week composite of Atlantic Canada for the period of July 1st through 15th, 2000 (A) and the same period for 2001 (B). It clearly shows that waters were lower for 2001 than in 2000 during this period. Figure 22 shows the sea surface temperatures for the period of November 1st through 15th, for years 2000 and 2001. They show that during this period for 2001, water temperatures were higher than for the same period in 2000. This is consistent with the thermograph data shown in Figures 18 and 19 for the sites in the region and demonstrates that water warmed up later in 2001 and subsequently cooled off later than for the period in 2000.

3.2.1.2 Monthly Water Column Profiles

CTD profiles for Salmonier Cove were taken approximately once per month, for 2000 and 2001, respectively (Figures 23 and 24). Due to the nature of sampling, CTD

profiling provided a snapshot summary only at the time when sampling took place. However, when each monthly sampling was summarized together, the overall patterns of chlorophyll, temperature and salinity become more apparent (Figures 23 through 26). For Salmonier Cove, it was noted that the warmest months were experienced in July and August, for 2000 and 2001 (Figures 23 and 24). A temporary thermocline became evident by late July, both years at about 10-12 m depth. By October the thermocline disappeared as surface waters cooled and the water masses mixed in association with the autumn storms that passed through the region. As with Salmonier Cove, water temperatures at The Tickle were at their highest in July through August, for each year (Figures 25 and 26). A thermocline appeared for this site by late June, 2000 and early August 2001, at approximately 8-10 m depth. This thermocline disappeared by October of both years.

Salinity for Salmonier Cove was generally higher on a regular basis – at 31 ppt or higher for each month data were recorded, with the exception of August 2000, where the salinity was recorded at slightly less than 30 ppt (Figure 23). A small halocline appeared for Salmonier Cove at 10-12 m depth, for July and August, 2000, but was not apparent for the 2001 sampling season (Figure 24).

Salinity for The Tickle was influenced by river run-off at upper end of The Tickle (Figure 12). As with Salmonier Cove, salinity was generally higher than 31 ppt for each month data were recorded, with the exception of July and August samplings, during 2000, when surface salinity dropped to 27-29 ppt (Figure 25). For 2001, salinity was consistently above 30 ppt (Figure 26).

Chlorophyll concentration is a measure of available food supply. The CTD profile summaries depict total chlorophyll (μ g/L), with the exception of the June 21 sampling for 2000, where chlorophyll-a (μ g/L) was recorded (Figures 23 and 25). At Salmonier Cove during 2000, food concentration reached just under 5 μ g/L total chlorophyll in July and August (Figure 23). Food concentration decreased with depth during the summer months, but was nearly constant at 3-4 μ g/L throughout October 2000. Chlorophyll-a sampling in late June 2000 indicated increasing food concentration from 0 to 20 m depth.

For the 2001 season, sampling started in April (Figure 24) and total chlorophyll was recorded throughout. As shown, there was considerable food at depths from 8 m through 20 m, peaking at 15 μ g/L at 12 to 16 m. This compared to only 1-3 μ g/L near the surface. Food concentration dropped over the course of the sampling period, with approximately 1-3 μ g/L total chlorophyll recorded by November.

At The Tickle, the amount of food recorded during the 2000 season peaked at slightly above 5 μ g/L in July and August (Figure 25). Unlike Salmonier Cove, food levels fluctuated with each month and with depth, but in general the amount of food decreased with depth. For the 2001 season, sampling began April 30th. On this sampling date, food concentration increased rapidly with depth, from 1-3 μ g/L on the surface to slightly above 15 μ g/L at 20 m, indicating a recent phytoplankton bloom. Subsequent months showed total chlorophyll at 2- 5 μ g/L, with the November sample having only 1 μ g/L total chlorophyll.

For both Salmonier Cove and The Tickle, there were similar measures of temperature, salinity and food among sampling stations (i.e., inside, middle, outside – Figure 12).

3.2.1.3 Meat Yield and Larval Abundance

Meat yields were performed approximately once every two weeks. At the same time, plankton tows were carried out to determine the relative abundance of larval mussels. For both sites, meat yields remained at high levels for a longer period in 2001 than in 2000 (Figure 27). Larval abundance reached its peak in late July for Salmonier Cove and early/mid August for The Tickle, declining rapidly at both sites in October. Interestingly, the peaks in larval abundance were preceded (by about 4-6 weeks) by sharp declines in meat yields, indicating spawning events. This occurred in both sites, in both years (Figure 27).

3.2.2 Spat Settlement

3.2.2.1 Monthly Spat Collector Deployments

Spat collectors were set each month to determine the period of most intense settlement. For both Salmonier Cove and The Tickle, the most intense settlement period occurred during August in both years (Table 3). However, settlement was much greater during August 2000 than for the same period of 2001. Salmonier Cove yielded an average of 122,647 spat per collector rope in August 2000 but only 26,218 for the same period in 2001. Similarly, mean spatfall at The Tickle during August 2000 was 109,120 but only 31,131 during August 2001. Spatfall at Salmonier Cove during September was higher in 2001 than in 2000 (mean 5,534 and 14,745 spat, respectively).

Upon observation of collector ropes, newly settled spat of about 400 µm to about 4 mm were visible in the twists of the rope. Most spat collected were 2 mm or less after approximately 30 days (Figures 28 through 31). However, for Salmonier Cove, a number of larger spat between 5 and 10 mm in length were collected during August of 2000 (Figure 32) and again during the June 2001 collection and late autumn (October – November) of 2001. For The Tickle, larger spat between 3 mm and 10 mm were collected during September and October of the 2000 season. Unfortunately, most collectors for 2001 were destroyed in storms and were lost at this site.

3.2.2.2 Cumulative Collector Deployments

Collectors deployed for 130 days or more collected the most seed (May and June deployments), generally, more than 12,000 per 1.8 m long collector. Collectors deployed at the end of September collected very little seed (30 days deployed) (Figures 33 and 34).

For Salmonier Cove, there was no pattern of amount of seed collected with deployment depth, 130 days or more (Figure 34). A two-way ANOVA comparing amount of seed accumulated per collector with collector deployment depth and month of deployment indicated no significant difference in amount of seed collected per collector with depth of deployment ($F_{(2,44)} = 2.89$, p = 0.071), but was found to be significant with month of deployment ($F_{(4,44)} = 40.22$, p < 0.001) and interaction between depth and month ($F_{(8,44)} = 2.34$, p = 0.044). A one-way ANOVA (p = 0.564) with depth only did not show significance in amount of seed collected and depth of deployment. A one-way ANOVA with month was significant (p < 0.001), with Tukey's-b test indicating (p < 0.05) that June and July were significantly different from August, September and October's collection (Figure 34). Seed collection for the June and July deployments were the most intense and were also the longest deployed, 162 and 132 days, respectively.

A two-way ANOVA indicated that the growth rate (mm/day) of spat at Salmonier Cove was significantly different with depth *and* month of collector deployment ($F_{(2,44)} =$ 87.67, p < 0.001 – depth, $F_{(4,44)} =$ 71.42, p < 0.001 – month). Growth rate at 3 m was significantly higher than at 6 m and 9 m for July and August (Tukey's-b, p < 0.05) but only with 9 m for June, 6 m for September and no difference in growth rate with depth for October (Tukey's-b, p > 0.05) (Figure 35). Overall growth rate for August, September and October were similar, as were growth rates for June and July (Tukey's-b, p > 0.05) (Figure 35).

Collectors deployed at The Tickle at 9 m depth had more spat per collector (130 days or more) than at 6 m or 3 m (Figure 33). A two-way ANOVA comparing amount of seed accumulated per collector with collector deployment depth and month of deployment indicated a significant difference in amount of seed collected per collector with depth of deployment ($F_{(2,44)} = 8.75$, p = 0.001) and month of deployment ($F_{(4,44)} = 18.75$, p < 0.001) and interaction between depth and month ($F_{(8,44)} = 3.37$, p = 0.007). One-way ANOVA (p = 0.073) with depth only and Tukey's-b (p > 0.05) test did not

show significance in amount of seed collected and depth of deployment. A one-way ANOVA with month was significant (p < 0.001) and Tukey's-b (p < 0.05) indicated that June and July were significantly different from August, September and October's collection. Seed collection for the June and July deployments was the heaviest and also the longest deployed, 163 and 134 days, respectively (Figure 33). Seed collected at 9 m was smaller than seed at 3 m or 6 m, for all deployment periods.

A two-way ANOVA indicated that the growth rate of spat at The Tickle was significant with depth *and* month of collector deployment ($F_{(2,44)} = 30.24$, p < 0.001 – depth, $F_{(4,44)} = 48.61$, p < 0.001 – month). Growth rates at 3 m was significantly higher than at 6 m and 9 m for July (Tukey's-b, p < 0.05). Growth rate at 6 m and 9 m were not significantly different for June, August and September and no difference in growth rate with depth was observed for October (Tukey's-b, p > 0.05) (Figure 36). Overall growth rate for October was highest, while rates for June and July were similar, as were growth rates for August and September (Tukey's-b, p > 0.05) (Figure 36).

Generally, the longer the collectors were deployed, the greater the amount of fouling – especially for collectors deployed at 9 m compared to 3 m or 6 m (Tables 4 and 5). A percentage of overall fouling was estimated for collectors deployed at each depth and month. The most common fouling organisms were green and red filamentous algae, with more red algae occurring on collectors in shallower water, while hydrozoans were more abundant in deeper water. Other bivalves that settled on collectors were clams, *Hiatella arctica* (more at 3 m than 6 m or 9 m), common jingle shell (*Anomia simplex*) and scallop, both the sea scallop (*Placopecten magellanicus*) and Icelandic scallop (*Chlamys islandica*). Starfish (*Asterias vulgaris*) were also present in small numbers.

3.3 Socking Trial Results

3.3.1 Depth of Deployment and Second-set Accumulation

There were less second-set mussels collected on socks at 9 m than at 4 m, for both sites, for both spring and autumn deployments *and throughout every* sampling period (Figures 37-39). The mean number of mussels per 30 cm of socking for a 4M sock at Salmonier Cove, deployed in the spring 2000, fell from about 5,500 per 30 cm to about 500 per 30 cm, just by having the sock placed 5 m deeper. The effect of depth was significant at Salmonier Cove for the spring 2000 deployment – autumn 2000 sampling (ANOVA, p < 0.001) and for spring 2001 sampling (ANOVA, p = 0.041). No further comparison with depth could be made as socks at 4 m were lost before the autumn 2001 sampling could take place. In Figure 38, mean numbers per 30 cm after one year in sock (autumn 2000 deployment and autumn 2001 sampling) reached into the thousands per 30 cm at 4 m at all densities, but only into the hundreds, at 9 m at all densities. For the autumn 2000 deployment – spring 2001 sampling, there were significantly lower numbers of second-set at 4 meters compared to 9 meters at Salmonier Cove (p < 0.001), as there were for the autumn 2001 sampling (p < 0.001).

At the Tickle for the spring 2000 socking, 4 m (Figure 39) accumulation was less than for Salmonier Cove, but followed a similar pattern with density and depth. At 9 m and spring 2000 deployment accumulation was only into the hundreds across the trial densities, again following a similar pattern with Salmonier Cove. There were significantly lower numbers of second-set mussels at 9 meters than at 4 meters for the spring 2000 deployment – autumn 2000 sampling (ANOVA, p < 0.001), however, not significantly less for the spring 2001 sampling (ANOVA, p = 0.211). Unfortunately, no further data for the autumn 2000 deployments remained after a storm passed the region.

To further show the effect of depth of deployment on amount of second-set mussel accumulation, length-frequency histograms of mussels from sections of sock after one year deployment were plotted (Figures 40 through 42). For both sites, there was a clear settlement of smaller mussels < 25 mm (mean starting size of socked mussels) after one year in-sock at 4 m, but less at 9 m.

3.3.2 Timing of Deployment and Second-set Accumulation

While deploying socks in deeper water appeared to decrease the amount of accumulation, socking mussels in the autumn versus the spring, produced some interesting results. Seed deployed in the spring 2000 at both sites were observed to crawl out of the sock within a few days of initial deployment. For the autumn 2000 deployment, however, they were slower to crawl out, most having not crawled out after a week. For Salmonier Cove, socks deployed during the autumn 2000 did not yield high accumulation of second-set, at any density, at 4 m or 9 m during the spring 2001 sampling. However, in the autumn 2001 sampling (one year in-sock – autumn 2000 to autumn 2001), socks deployed at 4 m at Salmonier Cove had accumulated dense second-set across all trial densities but little at 9 m (Figure 38). This dense collection of second-set mussels was evident in length-frequency histograms of the four trial densities at 4 m,

which showed sizable amounts of mussels < 25 mm, even after one year in-sock, but little at 9 m (Figure 41). Unfortunately, socks were destroyed at The Tickle and no spring versus autumn deployment comparison could be made.

3.3.3 Initial Socking Density and Subsequent Mussel Densities

For each sampling period, the number of mussels per 30 cm of socking was determined for each of the socking densities at Salmonier Cove and The Tickle. Socks deployed in the spring 2000 at the lowest densities (4M and 5M) and at a depth of 4 m, showed a large spike in the average numbers of mussels per 30 cm of sock at the autumn 2000 sampling period (Figures 37 and 39). However, the highest socking densities (6M and 7M) showed much lower gain in densities through additional seed accumulation in the autumn following deployment (Figure 38).

After one year in sock (spring 2000 to spring 2001), mussel densities dropped over all mesh types, as indicated by the spring 2001 sampling for Salmonier Cove and The Tickle – 4 m, autumn 2000 sampling (Figures 37a and 39a). As evident by the length-frequency histograms, there were less second-set mussels with each increase in start density (Figures 40 and 42). Socks were lost at both sites during storms and did not permit an autumn 2001 sampling of socks deployed at 4 m.

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At 9 m deployment depth, initial sock density had some impact on subsequent sample densities (Figures 37 and 39), although at Salmonier Cove, mean sock densities per 30 cm of socking increased slightly at each subsequent sampling period. Length-

frequency histograms at Salmonier Cove showed that increasing the initial density reduced the amount of second-set mussels that accumulated on the socks (Figure 40).

For the autumn 2000 deployment period at Salmonier Cove, sock densities increased little by spring 2001 at 4 m or 9 m deployment depths. However, after one year in-sock (autumn 2000 to autumn 2001), second-set accumulation at 4 m deployment and all densities was heavy (Figure 38). There was evidence of heavy settlement of smaller mussels < 25 mm in shell length (Figure 41). A similar pattern was observed at The Tickle, however, only an increase in sock density was apparent in the 4M material, while remaining low in the 5M through TMM (Figure 38). Initial sock densities appeared to have less of an impact on subsequent second-set settlement on socks at 9 m depth, as sock densities remained low across all trial densities (Figure 41).

3.3.4 Mussel Growth, Depth and Sock Density

The effect of an avoidance strategy on normal production (length of production time and subsequent farm activities) was considered by determining the growth of the mussels under the various deployment conditions (Figures 43 through 45).

General observations indicated that mussels grew the fastest between May and November of the spring 2000 deployment. For the autumn 2000 socking, growth was good as well, with shell length nearly doubling on average from October 2000 to the end of May 2001. To determine whether the initial sock densities had any impact on growth of socked mussels, single factor ANOVAs were performed on a sample from each treatment (Tables 6 through 8). For Salmonier Cove, spring 2000 deployment at 4 m, there were significant differences in mean mussel size over the four sock densities by the autumn 2000 sampling (p < 0.001), however, these were not significantly different for the spring 2001 sampling (p = 0.48). Socks at 4 m were lost by the autumn 2001 sampling and no data were available. At 9 m deployment depth, however, mean mussel sizes across the four socks densities were significant at the autumn 2000 sampling (p < 0.001), spring 2001 (p < 0.001) and autumn 2001 (p < 0.001) sampling. At The Tickle, all densities showed significant differences in mean mussel sizes after each sampling period (p < 0.001) at both depths.

For the autumn 2000 deployments, only socks for Salmonier Cove were retrieved. A two-way ANOVA indicated that there were significant differences in mean mussel size with depth *and* initial sock densities at both the spring 2001 ($F_{(3,4799)} = 76.98$, p < 0.001 – depth; $F_{(1,4799)} = 76.33$, p < 0.001 – initial sock density) and autumn 2001 sampling ($F_{(3,4623)} = 15.21$, p < 0.001 – depth; $F_{(3,4623)} = 11.42$, p < 0.001 – initial sock density).

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3.3.5 Harvest Yields

The effects of the treatments on the overall sock yields were assessed by determining overall sock yields (kg) and the percentage of mussels > 50 mm after one year deployment. Because sock lengths varied, the yield (kg) of marketable mussels per 30 cm of socking was used as a standardized method of comparing yields. Fouling, calculated as a percentage by gross weight per sock was also assessed and summarized (Tables 9 through 11).

Generally, the higher the initial sock density, the heavier the socks were after one year deployment, spring and autumn, 4 m and 9 m deployments, both sites (Tables 9 through 11). The exception, however, was for a 6M sock (start density of 222 - spring, 247- autumn) at Salmonier Cove. These socks were on average the heaviest weight of the sock types used, at all depths, spring and autumn deployments. Even at the autumn 2001 sampling of a spring 2000 deployment (i.e., 1.5 years deployed), 6M socks deployed at Salmonier Cove at 9 m were heavier than the others deployed at that depth (Table 9). No socks deployed at 4 m remained for an autumn 2001 sampling.

Fouling on the socks was defined as any organism that was not mussels. This included red and brown filamentous algae, hydrozoans, worms and clams. Fouling after one year deployment at both sites, did not exceed an average of 10% of total sock weight, with the exception of the 4M socking, 9 m at The Tickle, where socks were heavily fouled and averaged 18% of total sock weight (Table 11). There was a pattern of lesser amounts of fouling on socks deployed at higher initial seed densities at Salmonier Cove,

especially for the spring 2000 deployment, but this was not so evident at The Tickle, or the autumn 2000 deployments (Tables 9 through 11).

Yields of marketable mussels at Salmonier Cove ranged from 0.798 kg to 1.203 kg per 30 cm of socking after one year deployed at 4 m, spring 2000 start and 0.291 kg to 0.728 kg per 30 cm of socking at 9 m, spring 2000 start (Table 9, Figure 46). A two-way ANOVA analysis indicated there was a significant difference in sock yields with depth (F $_{(1,22)} = 28.47$, p < 0.001), but not with sock density (F_(3,22) = 2.62, p = 0.082).

Unfortunately, socks at 4 m were lost and no depth comparison with sock yield could be made for the autumn 2001 sampling (i.e., 1.5 years deployed). However, sock yields at 9 m depth for the autumn 2001 sampling showed yields of 70% of gross or better for mesh sizes 5M, 6M through 7M (1.594 kg, 1.633 kg, 1.616 kg, respectively), with exception of the 4M sock, yielding only 0.859 kg or 57% of gross (Figure 46, Table 9). A one-way ANOVA comparison of sock types at 9 m indicated a significant difference in sock types ($F_{(3, 10)} = 6.18$, p = 0.022), with Tukey's-b test (p < 0.05) showing only the 4M sock yield significantly different from 5M through 7M.

For an autumn 2000 deployment, autumn 2001 sampling at Salmonier Cove, only the 4M and TMM socking had measurable amounts of mussels > 50 mm in length, at 2.2% and 8.7% of gross, respectively, at 4 m and 1% for 4M at 9 m (Table 10, Figure 47). Yields were less than 100 grams per 30 cm of socking for the treatments. A two-way ANOVA analysis indicated significant difference in sock yields with depth ($F_{(1,23)} = 4.6$, p = 0.045) but no significance with sock type ($F_{(3,23)} = 2.97$, p = 0.058). .

For The Tickle, after one year deployment (spring 2000-spring 2001), marketable yields ranged from 0.552 kg to 0.846 kg per 30 cm of socking at 4 m and 0.444 kg to

0.800 kg per 30 cm of socking at 9 m (Table 11, Figure 48). A two-way ANOVA indicated there was no significant difference in sock yields with depth (F $_{(1, 23)}$ = 4.55, p = 0.101), but was significant with sock type (F $_{(3,23)}$ = 4.55, p = 0.017). Further analysis with Tukey's-b test (P < 0.05) indicated that the 5M sock was significantly different from 4M, 6M and 7M sock material yields. There were no autumn 2000 sock deployments remaining for analysis.

3.4 Husbandry Observations

Throughout the socking project it was noted when and how various activities were carried out to determine if any husbandry activity may have helped contribute to secondset. Socking at the study sites was generally carried out in the autumn of the year. Seed was harvested into tote pans, for the most part reasonably quickly (example, a few hours to harvest 75+ tote trays). Tote pans were easily overfilled and when stacked, were often not 'locked', such that one tote pan slipped inside the one below, which led to a crushing of mussels. Excess mussels were often walked on and were often shovelled up and placed into totes with the harvested seed for socking. Seed was usually harvested in the early morning, or late in the evening. When enough tote pans were filled (for example 75 or so), then they were either taken to be graded and socked, or stored for the next morning. Harvested seed was not iced, but covered with a tarpaulin if the weather forecasted rain, otherwise was left uncovered overnight.

Seed was graded either on the barge or on shore. The grader hopper was often filled to capacity, with ample water flowing. Periodically, the grader was shut down and

cleared of byssal threads and fouling. The speed of the rotating drum was also observed to impact the grade. Spun too fast, mussels were not graded well and some tote pans of seed were graded more than once. Totes of graded mussels were stacked according to size.

There was a variety of socking materials available (different mesh types, tube diameters, etc). The choice of socking material selected (i.e., mesh type) was often determined by the order of the tote pan under the grader. For example, the second grade of seed was placed into a 5M square mesh sock or a blue 14 mm diamond mesh sock type. Interestingly seed from one area of a farm did not always grade the same as from another, especially when different year classes of seed were graded. For example, seed in the second or third grade pans were sometimes visually larger or smaller in shell length from a 1998 and 1999 year class. This sometimes posed a challenge to workers who traditionally used a particular sock type with a particular grade pan number/position under the seed grader.

The socking process was observed to be straight forward. A sock mesh type was chosen and a tube pipe to match was attached to the sock table. It was noted that when a small tube diameter sock material or inappropriate steel tubing was used, the pipe on the socking table often clogged up and a stick was used to free the pipe. Many mussels were crushed in the process. This was later assumed to be one of the causes of socks observed with many shells on the inside of the mesh material (Figure 49). The rate at which the sock was pulled away from the socking pipe greatly affected the fullness (and hence socking density) of the sock. Most socks were deployed within a few hours of being •...

readied, but on late days, socks were placed in totes, stacked and left until the next morning, sometimes covered, other times not covered.

Where initial sock density was concerned, the number of mussels per 30 cm was generally not counted or recorded, however, relative fullness of the sock was maintained by periodic comparisons between socks to ensure consistency. When socks were about to be tied on a mainline, the each sock was given a quick jerk, to tightly pack seed within the sock. If the socks were left in the tote pans too long, the mussels lost much water and when deployed, would float for some time, until the mussels re-hydrated.

Existing socks were periodically observed throughout the farm sites for signs of second-set. It was noted that socks that had second-set accumulation were often of low density and/or fouled. Mussels on these socks were generally arranged in no particular direction with socking material often visible. Clumps of newly settled seed were present mostly on spaces where there were no socked larger mussels (Figure 50). High density socks in contrast were free of second-set. Mussels were tightly packed together and arranged posteriorly-anteriorly, side by side for the most part (Figure 50). Upon close inspection, a cross-section of the sock often showed a uniform wheel shape, with little room for additional settlement (Figure 51).

4.0 Discussion

4.1 Seed Crawling Behaviour in Response to Water Temperature and Presence of Food Supply

Temperature and food levels are described frequently in the literature to play a role in the spawning behaviour of mussels (Bayne, 1965; Baird, 1966; Bayne et al., 1976; Kautsky, 1982; Thompson, 1984; Fell and Balsamo, 1985; Penney, 1993; Mallet and Myrand, 1995; Macneill et al., 2000). Apart from the physiological effects, how temperature and food affect behavioural attributes of mussels are not clearly understood. The literature provides little information to describe crawling behaviour, other than describing mussel bed density population dynamics in the competition for space and food (Shaw et al., 1988; Fréchette et al., 1990, 1992), or locomotion during the larval stage (Young, 1995). The trials at the Ocean Sciences Centre were an attempt to understand how temperature and food may affect the crawling behaviour of different sized mussels with potential implications for second-set accumulation.

The results indicated that smaller seed were more mobile than the larger seed, with and without food being present. This should come as no surprise, as post settlement larvae (young spat) and juvenile mussels have been shown to exhibit crawling behaviour as they move about to position themselves within a mussel bed (Shaw et al., 1988). Littorin and Gilek (1999) found that juvenile *M. edulis* mussels exhibited significantly higher downward movement to re-colonize cleaned areas of collector ropes than upward movement and on cleared rocky surfaces, mussels migrated towards the perimeter of the clearings. A study on the crawling behaviour of the green mussel, *Perna viridis* did not

show the same downward movement of mussels (Tan, 1975). In Newfoundland, many mussel growers report the small mussels migrate down collector ropes after the initial settlement period but once they reach about 30 mm in length, they do not move (personal communication, Alvin Hodder, Rick Pippy). However, even large mussels have been shown to move around (Harger, 1968; McGrorty and Gross-Custard, 1995; Urya et al., 1996) by extending the muscular foot and then contracting it, pulling the body along (Hickman et al., 1974).

In areas where second-set regularly occurs, knowledge of the crawling behaviour of different size seed and under varying environmental conditions is important when considering a second-set avoidance strategy, as it is crucial that sock uniformity be created and maintained to limit future settlement opportunity. Good sock quality highly depends upon choosing the proper mesh size for the various size grades so that they have ample room and time to migrate to the outside of the sock. Knowledge of how mussels behave under varying conditions should provide insight on choosing the correct sock materials that do not compromise proper sock formation.

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Increasing temperatures resulted in more crawling in small mussels than larger ones, without food added. With the addition of food however, crawling behaviour decreased as the temperature increased, but not significantly. The influence of temperature on metabolic rate has been reported in several papers, indicating that increases in temperature lead to increases in the standard metabolic rate in *M. edulis* (Bayne, 1973; Widdows, 1973). However, Thompson (1984) reported that even at low temperatures, mussels retained a high clearance rate and Loo (1992) found that at low
Mooney et al. (1999) determined that smaller mussels of *M. edulis/trossulus* exhibited a higher clearance rate than larger mussels. In the current study, a possible strategy is being played out by both large and small mussels to maximize their chances for survival in an ever changing environment. Perhaps the smaller seed depleted the food resource in the tanks sooner than the larger seed and began to exhibit foraging behaviour. If food becomes available (i.e., such as adding algae to the touch tanks), then mussels may start feeding rather than continue to search. Conversely, mussels may exhibit foraging behaviour with little food present. Highly speculative, this suggestion needs to be studied in more detail in order to draw firm conclusions.

In terms of a second-set avoidance strategy, this theory may prove useful as farm operators may be able to schedule socking in the autumn of the year, at lower water temperatures with little food present and still be confident that socked mussels will crawl to the outside of the socking material. This may provide ample time for proper sock formation that would not normally occur before a new wave of settling larvae passes through.

4.2 Mussel Spawning and Spat Settlement Dynamics at Salmonier Cove and The Tickle, Connaigre Bay

4.2.1 Environmental and Biological Monitoring

4.2.1.1 Water Temperatures

Monitoring water temperatures are important for operators of mussel farms, since water temperatures play a large role in mussel biology, which in turn can dictate many aspects of the production cycle (e.g., spawning, seed collection, seed quality, socking times, mussel growth and harvesting). Water temperatures on the farm sites did reach 19 °C, surpassing 10-12°C, often viewed as the critical threshold in temperature for triggering spawning (Bayne, 1976; Mallet and Myrand, 1995). Even at 9 m, waters reached 16°C, enough to trigger spawning. The sites may be considered under influence of the open ocean, since the fjords are deep and open to the expanse of Fortune and Connaigre Bays. As such, changes in temperature due to coastal upwelling would not be uncommon (Bourque et al., 1995). Indeed, as shown through the thermograph data, both The Tickle and Salmonier Cove showed periods of wide fluctuating temperatures, particularly during the period of June through November and temperature changes of 10°C in over a tidal cycle were observed. The changes in water temperature can be attributed to changes in wind direction and tidal influences, causing upwelling of colder deeper water to the surface (Stewart, 2003). Temperatures at 9 m were generally more stable than at 6 m or 3 m, however, during the winter months of December through

March, temperatures were the most stable, as surface temperatures were cold due to lack of daytime heating, and more frequent northerly winds.

Fluctuating temperature due to wind, tidal change, physical disturbances from storms, separately or in some combination, may play a role in the spawning of mussels (Bayne, 1976; Witherspoon, 1986; Newell et al., 1991). As an example, the California sea mussel (*Mytilus californianus*), a species adapted to attach firmly to heavy surf conditions, was found to spawn continually at low levels, while the blue mussel (*M. edulis*) was shown to have only one major spawning period per season, where it is found in quieter waters (Morris et al., 1980; Suchanek, 1981).

Knowledge of spawning in response to fluctuating temperatures has been adapted and used quite successfully in hatchery settings as an important tool in shellfish production. Manipulating temperature is common practice to spawn various species of shellfish, usually by increasing temperature over a short period of time often in conjunction with some other stimuli (Chew et al., 1987; Helm and Bourne, 2004). In the present study, increasingly unstable water temperatures in June (Figures 17 and 18) coincide with an increase in meat yield (Figure 27) up until temperatures are greater than 10°C at the top few meters, at which time there is a decrease in yields. It is quite possible that once near surface temperatures reached above 10°C, large fluctuations of much colder water from below could have triggered some spawning event(s), as evident by the drop in meat yields. Bonardelli et al., (1996) showed a similar phenomenon for the giant scallop (*Placopecten magellanicus*).

The event of spawning is significant to consider in a second-set avoidance strategy, not only because spawning produces the large amounts of unwanted seed, but

also because it is stressful event, most notably heightened around the pre and post spawn recovery times (Harding et al., 2004). The word 'stress' is defined by Bayne (1975) as a "measurable alteration of the physiological (behavioural, biochemical or cytological) steady state which is induced by an environmental change and which renders the individuals (or populations) more vulnerable to further environmental change. As mussels as small as 30 mm (approx. 10 months of age) are capable of spawning (Gosling, 1992), it is possible that seed from these areas being socked may be under a host of stressors - environmental stress (frequent temperature shocks), biological stress (spawning) and handling stress (grading, socking, etc.). These may impact not only mortality, but the crawling behaviour of the mussels, leaving the sock poorly formed. As earlier data have shown, large spat used in the present study moved slower than small spat, thus if on a commercial scale, these larger spat are poorly handled when socked, the increase in stress may further impede their movement to the outside of the socking material.

Good seed quality before and after socking is necessary for proper sock formation and recognizing that fluctuating temperatures may contribute to the chain of stressors on reproductive large mussel seed is helpful in building a strategy to avoid second-set. In this case, reducing stressors as much as possible (of which a stable environment is a contributing part) is important to ensure rapid crawling to the outside of a traditional drop sock to avoid becoming trapped inside. As water temperatures in Salmonier Cove and The Tickle were generally more stable at 9 m than 6 m or 3 m, deploying socks in deeper water and/or adjust socking times around spawning, or sock in cooler water temperatures might help minimize stressors and maintain seed quality and hence sock quality.

4.2.1.2 Monthly Water Column Profiles

Of significance from the CTD data was the appearance of a thermocline at both Salmonier Cove and The Tickle, for both 2000 and 2001 during mid/late July. Viewed as typical spawning periods for most of Newfoundland, the periods of late June through to August (Sutterlin et al., 1981; Macneill et al., 2000) saw temperatures above 10°C in the top 3 to 5 m, but remained 5 to 7°C cooler at depths of 10 m to 12 m during this time. Having gear deployed in deeper water under such conditions may have several benefits: controlled delay of spawning of production mussels and minimize high temperature induced stressors which may impact on mussel mortality that would eventually degrade sock quality (Harding et al., 2004).

Yet another significant finding related to the thermocline is the impact it may have on the dispersal and settlement of bivalve larvae. Generally, fish and shellfish that have pelagic larval stages are said to exhibit passive and/or active dispersal methods as a means of recruitment. Passive dispersal refers to larvae that drift about with the tides and currents, until arriving upon a suitable place to settle. Active dispersal refers to a more selective process where larvae exhibit some choice over where they end up and has also been studied quite extensively. Both methods are likely to occur at one point or another in the pelagic stages of bivalve larvae but have often been the focus of many reviews and discussion (Mann, 1986; Roegner, 2000; Sponaugle et al., 2002). The ability of bivalve larvae to actively move through boundary layers is often referred to as vertical migration. The reasons for vertical migration may depend upon the larval stage of development. Settling mussel larvae develop paired eyespots, which are said to be photosensitive and

may cue movement toward the surface (Bayne, 1965). Vertical migration has been described for the sea scallop, *Placopecten magellanicus*, as well (Kaartvedt et al., 1987; Manuel, 1996).

In the present study, the identification of seasonal boundary layers in temperature and salinity (at about 8-10 m) may yield an important clue in the attempt to avoid secondset. Previous work has shown that larvae of many species of shellfish can be limited to a particular locale in the water column by the presence of gradients (Mann et al., 1991; Manuel et al., 1996). If lines are deployed below the layers at key settlement periods then most of the second-set may be by-passed. Although logical, it was not proven in the present study as collectors were deployed (coincidently) on the boundary layer (refer to section 4.2.3.2).

The CTD profiles also indicated food levels present throughout the water column, however, more food is present in near surface waters. The amount of food in the water depends upon many factors, however a good rainstorm, windstorm and/or mixing of water, longer day length, along with warming water temperatures in late spring can trigger a phytoplankton bloom. By relating the CTD profiles with meat yields used as a gonad index, one can better understand the triggers for gonad development and mussel spawning, making prediction easier to determine. At both experimental sites, a bloom was evident in April and has been reported to be quite common for sites around Newfoundland for this period (Clemens et al., 2000). During the ensuing weeks and months, a rapid growth in reproductive products occurs which is fuelled by the spring bloom (Thompson, 1984).

In developing a strategy for reducing the amount of second-set on sock gear, water column chl-a profiles are invaluable because they indicate a time frame in which ample food is present (springtime mostly). In theory, a socking time schedule could be developed to make sure socked mussels benefited from the food resources, while at the same time be deployed at a depth that avoids major aggregations of bivalve larvae (i.e., deployed below thermoclines) and remain in a cool, stable environment that minimizes stressors. At the sites, there was ample water depth to sink gear to potentially avoid settlement, however, there was no knowledge of how deep mussel settlement would occur and what impact deploying gear in deep water would subsequently have on growth, spawning and recovery periods in the mussels.

4.2.1.3 Meat Yield and Larval Abundance

In the present study, rather than monitor to gain seed, meat yields and larval abundance were monitored so that a timeframe to avoid second-set accumulation could be determined. Overall, seed collection success can be linked to deploying collectors at periods of high pediveliger larval abundance (Macneill et al., 1999, 2000). Pineda (2000) argued that settlement does not always correlate directly with larval supply and to some degree this appears correct, as, in some cases, seed collection can be very successful with relatively few mussel larvae per volume sampled (e.g., The Tickle during the 2000 collection season). In addition, the monitoring of meat yields as a tool to judge spawning times, was a useful and simple method to predict the timing of larval appearance.

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As the data showed, spawning appeared somewhat delayed in 2001 compared to 2000 and may be linked to cooler water temperatures in the Atlantic region during 2000. Assuming that the larval abundance for the area during the period of study came from non-cultured populations, then this delay in spawning in cultured mussels held true for wild ones as well, with larval abundance following overall similar patterns for both years, but was later for 2001 than 2000. Regardless of the source, the appearance of mussel larvae within a predictable time based on declining meat yields, provides growers with a workable time schedule to engage in socking activities, or plan socking schedules outside of the main seed collection times to avoid second-set.

4.2.3 Spat Settlement

4.2.3.1 Monthly Collector Deployments

The most intense settlement period was found to be August. As mentioned in the previous section, the colder water appeared to impact the timing of settlement and the intensity, as spatfall numbers were lower for the same months of 2001 than 2000 and settlement appeared to extend for a longer period during that year. In forming a strategy for second-set avoidance, one can probably exclude the month of August and perhaps the latter two weeks of July for socking because of the likelihood of high settlement. But in addition, water temperatures during this time are high and the stress of handling product during socking may lead to poor quality socks through delayed mortality. Logically then,

a good strategy for second-set avoidance would be to determine the most intense period of settlement and deploy socks before or after.

There have been numerous studies and reviews of population dynamics in both plants and animals - with food and space limitations being the most common controls of a population per unit area (Drew and Flewelling, 1979; Fréchette et al., 1990; Elliot, 1993). Determining whether food resource or space is the critical factor has been subject of much discussion. Lawton (1989) argued that per capita use of resource is less in small bodied individuals, hence a finite food source would support more small individuals than large ones. Mooney et al. (1999) noted that smaller mussels had higher specific clearance rates than larger mussels, and as such, limited food resources may get depleted quickly at high densities. Ardisson and Bourget (1991) concluded after an 11 year survey of mussel recruitment on navigation buoys, that during the first few months of growth on clean collectors, mussels rarely saturate the available space, thus suggesting a food limitation. It certainly is possible, as in the current study, the CTD profiles showed decreasing food levels from the April/spring bloom to low levels during the late summer. Density on recently settled collectors can be very high (Macneill et al., 1999). Rapid growth by the settled spat would likely limit food resource and space, leading to the maximum sustainable biomass to be exceeded, causing many to leave by byssal drifting. When spat growth was analyzed, it was interesting to note the appearance of larger spat, some >10mm in length, on collectors deployed for approximately 30 days. While it is possible that some spat may have exhibited rapid growth, it is more likely that the spat detached from elsewhere and resettled on the collectors (Lane et al., 1985).

The result of larger spat settling on new collectors supports the phenomenon of byssal drifting observed for a number of post-larval bivalve species, including mussels (Bayne, 1965; Lane et al., 1985; Beaumont and Barnes, 1992; de Montaudouin, 1997) either as active transport or as a result of storms (passive) shaking seed from the ropes (Caceres-Martinez et al., 1994). Thus it is shown here that secondary settlers do have the potential to settle on production gear and help contribute to second-set accumulation on mussel socks. To what extent this may occur and what impact it may have, is unknown from the present study.

4.2.3.2 Cumulative Collectors

Determining where larvae are located in relation to boundary layers is often difficult in field work, but has been investigated for a numbers of species, with interesting results. Raby et al. (1994) found an aggregation of *Mytilus edulis* (among others) larvae directly above and below a halocline. Similar results have been reported for polychaete larvae (Thiébaut et al., 1992). Several studies have been conducted on the distribution of scallop (*Placopecten magellanicus*). In an investigation on the George's Bank, Tremblay and Sinclair (1990) found assemblages of scallop larvae above the pycnocline. Other field investigations on vertical distribution of marine invertebrate larvae include urchins (Pennington and Emlet, 1986). The effect of thermoclines on settlement depth has also been investigated using scallop veligers in large mesocosms in the laboratory (Pearce et al., 1998; Manuel et al., 2000). The shifting depths of the thermocline and to a lesser extent halocline layers in the current study are significant when keeping the data of the monthly collection trials in mind, as seed was obviously settling in June. Depending upon the timing of socking, settling larvae may be present at the boundary layers, and if not careful, socks may be deployed at the optimum depth for collection. For 2001, the thermocline again appeared and reached a maximum depth of about 8 m to 10 m, so this appears to be a stable boundary layer depth for the area. Coincidently, the collector maximum depth was about 11 m (9 m deployed + 1.3 m collector rope length + some line sagging). Thus for these trials, some of the collectors deployed at 9 m may have been in the threshold area of the thermocline and not 100% in one layer at all times. As with the previously mentioned investigations, if larvae aggregate just above and below the boundary layer, this may in part explain why there were just as many or more mussel spat per collector observed at 9 m as at 6 m or 3 m, especially for The Tickle and to a lesser extent, Salmonier Cove.

Further to this, recent studies have shown populations and species differences in settlement depth. Manuel et al. (1996) confirmed differences in the vertical distribution of veligers of the giant scallop, *Placopecten magellanicus*, in a 10 m deep mesocosm with a thermocline induced deep water tank. For mussels, Freeman et al. (2002) conducted a mesocosm study using *M. edulis* and *M. trossulus* and showed that *M. edulis* settled in deeper water than *M. trossulus*, under mixed (warm and cold) water conditions but both species settled below 6 m. Their investigations were applied to the field by Kenchington et al. (2002), who determined that more *M. edulis* settled at depths of 5 m than *M. trossulus*. If these results are applicable to other areas, it would be significant for the south coast, where both species co-exist (Innes et al., 1999). Mussel species

identification was not part of the scope of the current project, although it would be an important future project to complement the puzzle of the seed collection in the region.

Another explanation for the high accumulation of mussel seed over time in deeper water is the accompanying accumulation of fouling. Collectors deployed at 9 m for the longest periods (June and July through November) had heavy algal fouling. This creates surface area available for settlement, although not necessarily favorable, as Penney (1993) demonstrated that fouling of *Polysiphonia* spp. actually deterred mussel settlement.

Where spat size is concerned, spat at 9 m were smaller than at either 6 m or 3 m depths. CTD profiles (Figures 23 through 26) indicated more food present near the surface and warmer temperatures and, as such, it is likely that spat grew quicker than spat in deep water. Self-thinning models described by Fréchette and Lefaivre (1990, 1995) suggest that in any population, a threshold in space and/or food will be reached and then followed by a reduction in the population to sustaining levels. In the present study, fewer spat at 3 m and 6 m collectors may be a result of the threshold in space and/or food having already been met, causing the collectors to shed biomass over time.

As discussed, there are a few possible reasons for the observed findings. Seed collection depth may follow the seasonal thermocline and, as such, a good strategy for reducing second-set may be to deploy socks below the main collection depth. However, based on the results presented here, it is not proven so. Further research needs to be conducted to better understand seed settlement in this area – species preferential distribution, settlement patterns in stratified waters on the south coast and the role of fouling on seed collection and growth.

4.3 Socking Trials

4.3.1 Depth of Sock and Second-set Accumulation

The results showed deploying socks in deeper water should be very encouraging to operators in areas where second-set has been a problem. All initial sock densities performed well when deployed at 9 m (spring or autumn), accumulating far fewer mussels than socks held at 4 m depth.

It is interesting to note that the results here are in sharp contrast with the results from the cumulative seed collection section (refer to 4.2.3.2). There, more mussel seed collected at 9 m than at 3 m or 6 m for several key months (July and August, most notably). Based on these results alone, socking during this time would not be recommended. However, with evidence indicating that collected seed held at shallower water grew more rapidly, the likelihood of mussels in shallower water self-thinning to support the biomass is plausible. In addition, with increased surface area due to fouling algae at the depth (8 – 10 m) where a thermocline boundary layer was recorded, more spat may have settled, and can in-part explain more mussels at 9 m. Another explanation of the differing results stems from available literature indicating the ability of mussels to filter bivalve larvae and other materials as food sources (Davenport et al., 2000; Robinson et al., 2002). As shown in the next section, increased initial density resulted in less second-set, even at 9 m depth, suggesting there may be an element of larval bivalve filtering occurring, although not investigated.

4.3.2 Initial Sock Density and Second-set Accumulation

The results indicated that increasing initial sock density can have a significant impact on the amount of seed accumulated on production gear. At the size of seed used here, 100 mussels per 30 cm did not result in a 'full' sock, but rather a sparsely filled sock, leaving ample room for attachment of mussels and foulers (Figure 50). By increasing the density, the sock becomes 'full', leaving little room for attachment of new spat.

In a way, suspended mussel socks can be viewed as communities of mussels. With this in mind, the behaviour of socked mussels becomes important, more specifically, how does the 'population' of socked mussels react to initial disturbance (e.g., handling) stressors, its environment (food, temperature, etc.) and how does the population changes as it reacts to competition for space and food. Initially, socked mussels are all inside the sock and the natural behaviour for them is to start crawling and compete for position to obtain enough food and room to grow. A properly formed sock can be viewed as one that has high enough density to force mussels to compete for position on the outside, thus, they form a solid column, mussels positioned side by side, with siphons exposed to the passing food supply. Socks formed like this remain clean and free from heavy fouling and second-set. Apart from little room for drifting larvae to settle, one suggestion is the filtering activity of the mussel reduces settlement. It is noted in the literature that adult mussels filter out zooplankton (Davenport et al., 2000), with mussel larvae being found in the stomachs of adults (Robinson et al., 2002). Lehane and Davenport (2004) reported in laboratory experiments that about 90% of bivalve larvae made available to mussels were ingested and apparently fully digested. The suggestion is that out on a mussel farm, adult mussels have the capability to reduce bivalve larvae significantly. In essence, a properly formed sock becomes a good filtering system, enough to discourage fouling organisms, including settling mussel larvae, as they are ingested. This contrasts with Commito (1987), who suggested that high density of adults did not inhibit the settlement of spat onto a mussel bed.

A low density sock is often sparse with mussels poorly oriented to the current as there is little competition. It has been noted among growers that with a low density sock, mussels tend to be less active, do not crawl out as fast and can become trapped inside when they grow too large, leaving the sock to become a giant collector, densely coated in second-set (personal communication, Alvin Hodder, Terry Mills). This may be possible if there is food arriving and with little competition, mussels *may* more likely stay where they are and start feeding. The present study does not support the notion of foraging behaviour by seed mussels. As observed, crawling rates of large and small spat was not affected by the presence of food. It is more likely that the wrong choice of sock type (i.e., mesh size or tube diameter), poor handling of seed or inappropriate socking density leads to a sparse sock.

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There has been much discussion in the industry on an effective socking density to use in order to yield a high harvest. As Mallet and Carver (1991) pointed out, the yield is often a function of site, grow-out depth in addition to seed density. In terms of second-set accumulation, we can add several key factors to this list; the seed size being socked, sock mesh type, seed handling procedures and environmental considerations. Stating for example, 400 mussels per 30 cm by itself would be misleading, without including the

seed size and mesh size/tube diameter. Finding the correct density will take some practice, but it is much easier if consideration is given to the environmental conditions of the site at the time of socking and condition/size of the seed. For this particular project, with an average seed size of 25-27 mm, sock densities of at least 250 mussels per 30 cm, using a medium square mesh sock with 50-60 mm diameter was effective in reducing the amount of second-set. It is speculated that using the same mesh socking, but smaller seed (~20 mm) would have produced more uniform looking socks, with densities 275+ per 30 cm being acceptable for reducing second-set.

4.3.3 Spring vs Autumn Deployments and Second-set Accumulation

Perhaps the most notable thing about the results of the spring and autumn socking trials is that even though the socks deployed in the spring 2000 had to endure two full settlement periods and the autumn 2000 deployments had to endure only one full settlement period, the spring socks did better, in terms of lesser amounts of second-set accumulation. Even at 9 m, where seed accumulation was far less over all the trial densities (both spring and autumn deployments), the spring socks had only slightly higher numbers of mussels per 30 cm than those socked in the autumn at 9 m.

It was also interesting to note that increasing initial sock density appeared to have no impact on accumulation at 4 m, for socks deployed in the autumn 2000. This observation is important in formulating a second-set avoidance strategy as it stresses the importance of deploying socks during optimal conditions and the need to understand how

the environmental conditions affect the biology and behaviour of mussels. Some growers may decide to sock in the autumn after the pulses of mussel larvae have passed and the water is cooling down, however, as the results have shown, this strategy may not work. The best scenario appears to be to sink the gear in deeper water to reduce the amount of settling mussels.

Do the results eliminate an autumn socking altogether? Often farm activity is very busy during the spring – line maintenance, harvesting, collector preparation, etc. Adding a major socking operation in April and May might not be possible all the time. The trials conducted here suggest spring socking, sunk deep and at high densities for the best result. However, autumn socking may be possible, if different socking types are used and close attention paid to mussel condition, their behaviour, handling and environmental conditions. An example would be to switch to the continuous socking method during the autumn. Food supply and temperatures are decreasing, mussels may be in post-spawn conditions, thus it is likely mussels will not react very well to stressors and may not crawl out of the socks very quickly if socked under such conditions. But as Thompson (1984) pointed out, under 'normal' winter conditions, metabolic rate is surprisingly high, so they will filter any food out and as such will grow at least a little. If this assumption is true, then there is a greater risk of mussels deployed in single drop socks in the autumn becoming trapped inside the sock. By the time the spring bloom arrives and there is a burst of growth (Mallet and Carver, 1993), the mussels would be too large to crawl out. With continuous socking methods, however, mussels can remain inside the material, which will dissolve over time. When the bloom arrives during the spring, mussels would be firmly attached and proper arrangement attained. Thus, it is

possible to sock during the autumn, however different socking methods must be used to ensure proper sock formation under the varying environmental conditions.

4.3.4 Mussel Growth and Harvest Yields

An important consideration in a second-set avoidance strategy is what impact, if any, will changes in growing techniques have on overall production. Initial socking density was expected to impact greatly the overall growth of the mussels held at 4 m and 9 m, as it was hypothesized that mean mussel size would be smaller with each increase in initial seed density used, due to food and/or space limitations (Fréchette et al., 1990, 1992). Generally speaking, there were significant differences among socks deployed at differing start densities, but it did not always follow the hypothesis for each deployment and at each site. At Salmonier Cove for example, the lower density socks showed the smallest mean size at 4 m and 9 m for a spring 2000 deployment. Coincidently, these socks often had the highest accumulation of second-set and fouling. It is likely that the increased biomass and/or fouling had an impact on growth, not the socking density itself. Langan (2001) compared growth of mussels in an open ocean, submerged long line system and found no comparable difference in growth using 500 per meter (150 per 30 cm) and 800 per meter (240 per 30 cm) at 20-25 mm start size. The experimental sites used here are in an open ocean like setting and as such perhaps ample food was available to negate any major effect of the high initial sock densities on growth used in the present study.

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At The Tickle, the spring 2000 deployment, mean mussel size was larger for lower density socks in the autumn of 2000 and spring 2001. But in this case, the amount of second-set accumulation was much less than at Salmonier Cove and fouling was less. In addition, starting densities for The Tickle at spring 2000 was a little higher per 30 cm of socking. One can speculate that all these factors combined may have contributed to the differences observed between the results of the two sites. It is interesting that the highest amount of fouling at The Tickle was for the lowest density sock (4M), 9 m (18% - Table 11). These socks also accumulated the most second-set at 9 m of all the trial socks, both sites after one year deployed, showing that surface area available for settlement can influence seed collection.

For the autumn 2000 deployment at Salmonier Cove, there were no clear patterns with mussel sizes and initial sock densities, other than high seed accumulations on all socks deployed at 4 m, then a gradual thinning of seed after one year. Despite these different explanations, one can conclude that increasing initial sock density may not have as negative an impact on growth as having socks covered in second-set, and as a strategy to avoid high accumulations, increasing sock density is a good choice to make, to avoid fouled overset socks.

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One clear finding from the study was the effect of depth on the weight of marketable product after one year of deployment. Overall, socks at 4 m weighed more than at 9 m and was reflected in the amount of product > 50 mm per 30 cm of socking. Considering the temperature dynamics of the study sites, it was shown that the seasonal thermocline forms at around 8 m to 12 m. It is possible then that socks at 9 m may have been just in the midst of the thermocline boundary layer, in enough colder water to slow

growth. This would however contrast with Thompson (1984) and Loo (1992), who showed that even at cold temperatures, mussels were able to exhibit high metabolic rates and some growth and in the results of the present study, a high rate of seed crawling behaviour at 0° C.

From the environmental section of the study, food appears to be abundant in deep water at least in late spring to early summer, but this did not appear to lead to higher yields in deeper water. Ogilvie et al. (2004) investigated growth of the greenshell mussel, *Perna canaliculus* in water depths of 5 m and 17 m to determine if growth can be sustained or enhanced in the deep water chlorophyll layer, but found no significant difference. Despite this, mussels appeared to take advantage of a bloom(s), as harvestable yields per 30 cm were higher for the spring 2000 deployed socks than the same year class deployed in the autumn 2000. The difference being that spring socks after one year endured two spring blooms (spring 2000 and spring 2001) under a much reduced density, while the autumn socks endured only one bloom (spring 2001) under the controlled density (i.e., socks at a desired density per 30 cm) and one on seed collector ropes under high density. Market yields at 9 m after 1.5 years deployed were in the 70% range in Salmonier Cove, thus it can be concluded that high yields free of second-set can be attained. At The Tickle where second-set accumulation was less at 4 m and 9 m for the spring deployments, sock market yields were 50-70% after one year deployed.

It is concluded that respectable product yields can be attained by deploying socks in deeper water, but the timing of deployment and depth may influence the length of time required to reach an acceptable market percentage. The initial sock density may have some impact on growth, but in areas where site conditions permit, a higher biomass per

unit area may be supported. Depth does influence amount of second-set accumulation, with socks deployed in deeper water being cleaner and more acceptable to processors.

4.4 Husbandry Observations

Observing existing farm husbandry practices was an invaluable supplement to the field and lab experiments. The effect of husbandry practices on existing marketable crop (in terms of poor yields, uniformity of socks, second-set accumulation, fouling, etc.,) was assessed speculatively with the common expression of "poor product in equals poor product out" kept in mind. Two important questions arose from the field observations. First, how might the natural environment affect the behaviour and seed quality? Second, what contribution (positive or negative) to seed quality is husbandry related? The answer to the first question has been shown through the various field and lab studies. The following is a short discussion of the husbandry observations.

It was noted that on many existing socks coated in second-set, many adult mussels were trapped inside the sock and were too large to get out. To say that the wrong mesh size was used is an easy answer, however, from the results of all the field investigations, it is possible to piece together another scenario. The correct mesh size might have been used, however, if mussels were under stress, either by being mishandled, or in post-spawn condition, or socked during bad environmental conditions, their behaviour may be altered such that they stay inside the sock, either eventually dying, or growing just enough to become trapped. Maintaining seed quality is paramount and a full understanding of the

biology, behaviour and the effects of stress has on mussels is necessary to optimize husbandry practices (Harding et al., 2004).

Other socks covered in second-set had few mussels trapped inside. As noted earlier, staff rarely checked initial densities, thus it is likely that not enough mussels were deployed per 30 cm yielding a poor sock, or, if enough were there initially, they may have fallen out, suggesting the wrong sock mesh size used for the seed size at hand. Density control was one factor noted that heavily influenced sock quality, especially during the socking process. Depending upon the person working on the socking table, socks were either full, sparse, loose or tight. When deployed, either of these may lead to different amounts of mussel staying inside or falling out. As shown, a low density sock was subject to heavy second-set and fouling, so maintaining consistent sock quality is important.

5.0 Implications for Industry and Concluding Remarks

The series of lab, field observations and trials conducted here were a broad attempt to understand the dynamics of second-set on the south coast of Newfoundland. By piecing together environmental conditions, mussel biology and reaction to site conditions, as well as using various socking techniques, a recommended starting point would be to sock during the spring, using smaller seed (hence higher densities) and sink gear deep, near or below where the seasonal thermocline appears. If an autumn socking must take place, then it is recommended that the gear type be changed to account for the negative impact environmental conditions and handling during that time of the year may have on mussel crawling behaviour. As such, continuous cotton socking in the autumn appears to be suitable if conditions (either environmental or seed quality) produce doubt on whether uniform socks can be formed.

Where sock densities are concerned, a reference to seed size *must* be considered when a number is quoted. For the current study, an average seed size of 25-27 mm, sock densities of at least 250 mussels per 30 cm, using a medium square mesh sock with 50-60 mm diameter was effective is reducing the amount of second-set. One may consider socking smaller seed (~20 mm), as it may produce a more uniform sock. Using the same tube diameter or mesh type, densities of 275+ per 30 cm would be acceptable for reducing second-set accumulation.

A poorly created sock is often a target for second-set accumulation, no matter what the density. Proper farm husbandry practices are required, with a thorough understanding of the site being operated, including all aspects of environmental conditions and mussel biology. Growers are encouraged to take note of their socking practices to make sure proper mesh sizes are used. Avoid walking on mussel seed during seed harvest and scooping the broken ones into the pans waiting to be socked. Proper water flow in socking tables is required to prevent clogging of sock pipes and reduces the necessity of using a stick to free the pipe, causing crushed mussels to be socked.

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The project also highlighted the lack of understanding of mussel crawling behaviour under various environmental and biological conditions by the average farm employee and how important this knowledge is in terms of creating a uniform, high quality sock. There is a necessity for industry to undertake more comprehensive

investigations involving husbandry activities and resulting effects on mussel behaviour, health and growth. For areas where mixed *M. edulis* and *M. trossulus* populations exist, there is a need to better understand the settlement patterns of each species and how they impact second-set accumulations. Finally there has been much emphasis on mussel seed and sock quality throughout the present study. It is hoped that employees of these farms can use and build upon the information gathered from the project and pass it on to new employees and other farm sites in the area.

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Tables

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Table 1. Mean mussel seed sizes used for movement behaviour trials. SSNF –
Small seed no food added, SSF – Small seed food added, LSNF – Large seed no
food added, LSF – Large seed food added.

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Treatment	Mean Size (mm)	±S.D	±S.E.
SSNF	5.3	0.84	0.217
SSF	5.4	0.44	0.113
LSNF	19.1	1.10	0.282
LSF	19.2	1.30	0.326

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Table 2. Mean starting mussel seed densities and mean number of mussels per 30 cm of socking used for the sock trials. *TMM 7.5 was used for the autumn 2000 trials because 7M was not available. Overall average seed size is given for each socking time and site (n = 200 mussels).

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Mesh Type	Salmor	nier Cove	The Tickle		
	Spring 2000	Autumn 2000	Spring 2000	Autumn 2000	
4M	94	115	151	129	
5M	164	165	193	173	
6M	222	247	266	276	
7M (TMM 7.5*)	291	278	333	296	
Overall Mean Size (mm)	25.4 mm	27.2 mm	25.2 mm	27.9 mm	

Table 3. Mean number of mussel spat per collector rope for Salmonier Cove and The Tickle, 2000-2001. No collectors were deployed for over-wintering in the autumn, 2001. Collection period = approximately 30 days however, number of spat per collector rope standardized for equal number of deployment days. (n = 3 collectors)

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Collection Period	Salmonier Cove					The	Tickle	
	2000 2001			2000)	200)1	
	mean/ collector	mean/ collector ±S.E		±S.E	mean/ collector	±S.E	mean/ collector	±S.E
June	1	0.5	21	14	12	5	n/a	n/a
July	11,592	925	5,976	2,196	3,751	391	38,475	12,277
August	122,647	8,968	26,218	2,681	109,120	8,297	311,131	2,948
September	5,534	576	14,745	1,664	3,438	1,117	n/a	n/a
October	122	37	1,454	138	87	31	n/a	n/a

Table 4. Fouling types and relative amounts on collectors deployed at Salmonier Cove throughout the Summer of 2000 at depths of 3 m, 6 m and 9 m. mod = moderate fouling, BFA = brown filamentous algae, GFA = green filamentous algae, RFA = red filamentous algae, Crustaceans = skeleton shrimp (*Caprella* sp.).

Start Collection Date	3 meters	6 meters	9 meters
	type	type	type
June (162 days)	Hiatella – light Starfish – light Jingle shells – light RFA – light/mod Crustaceans - heavy	RFA – light Starfish – light Crustaceans – heavy	Hiatella – light Jingle shells – light Periwinkles – light BFA & RFA- mod Crustaceans – heavy
Overall % Fouling	0-25%	26-50%	51-75%
July (132 days)	BFA & RFA – light Jingle shells – light Starfish – light (7) Crustaceans - mod	GFA & RFA – light Starfish –light (3) Crustaceans - mod	<i>Hiatella</i> – light Starfish – mod (17) RFA – mod Crustaceans – heavy
Overall % Fouling	0-25%	0-25%	26-50%
August (103 days)	GFA & RFA – light Crustaceans - light	GFA – light Jingle shells – light Hydrozoans - light Crustaceans – light Starfish –light (4)	BFA – light Jingle shells – light Crustaceans – mod Starfish – mod (16)
Overall % Fouling	0-25%	0-25%	0-25%
September (70 days)	Jingle shells – light Periwinkles - light RFA - mod	Periwinkles – light RFA – light BFA - moderate	Scallop – light Jingle shells – light RFA – light Periwinkles – light
Overall % Fouling	0-25%	26-50%	0-25%
October		DEA y light	DEA y light
(30 days)	BFA – v. light	DFA – v. light	DFA – v. light

Table 5. Fouling types and relative amounts on collectors deployed at The Tickle throughout the Summer 2000 at depths of 3 m, 6 m and 9 m. mod = moderate fouling, BFA = brown filamentous algae, GFA = green filamentous algae, RFA = red filamentous algae, Crustaceans = skeleton shrimp (*Caprella* sp.).

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Collection Start Date	3 meters	6 meters	9 meters	
	type	type	type	
June (163 days)	Hiatella – light Periwinkles – light Crustaceans – mod GFA & RFA -mod	Starfish – light Hiatella – light Sea anemones –light Starfish – light (17) Periwinkles - light Crustaceans – mod Hydrozoans – heavy GFA & RFA- heavy	Starfish – light (12) Crustaceans - light RFA- mod Sea anemones –heavy Hydrozoans – heavy	
Overall % Fouling	26-50%	75-100%	51-75%	
July (134 days)	GFA & RFA – light Crustaceans - light	GFA & RFA – light Crustaceans - light	Hydrozoans – light Starfish – light (7) RFA – light/mod Crustaceans – mod	
Overall % Fouling	0-25%	0-25%	26-50%	
August (104 days)	GFA & RFA – light Periwinkles – light/mod Jingle shells – light	BFA & GFA – light Jingle shells - light	Hydrozoans – light BFA – light Periwinkles – light Jingle shells – light Scallop - light	
Overall % Fouling	0-25%	0-25%	26-50%	
September (72 days)	Hydrozoans – light Jingle shells – light Periwinkles - light RFA - mod	Hydrozoans – light Periwinkles – light Scallop – light RFA – mod/heavy	Scallop – light RFA – light Hydrozoans – light/mod Periwinkles – mod	
Overall % Fouling	0-25%	26-50%	26-50%	
October (30 days)	BFA – v. light	BFA – v. light	BFA – v. light	
Overall % Fouling	0-25%	0-25%	0-25%	

Table 6. ANOVA ($\alpha = 0.05$) comparing sock types (4M, 5M, 6M and 7M) for mean seed size (mm) at start of trials and each subsequent sample period – 4 m and 9 m deployment depths, Salmonier Cove, spring 2000 deployment.

Sample Times	4 meter of	deployment depth	9 meter deployment depth		
Sample Times	p-value	F-value	p-value	F-value	
Spring 2000					
(Start)	0.13	1.87	0.13	1.87	
Autumn 2000	< 0.001	10.47	< 0.001	8.87	
Spring 2001	0.48	0.82	< 0.001	11.20	
Autumn 2001	N/A	N/A	< 0.001	20.32	

Table 7. ANOVA ($\alpha = 0.05$) comparing sock types (4M, 5M, 6M and 7M) for mean seed size (mm) at start of trials and each subsequent sample period – 4 m and 9 m deployment depths, The Tickle, spring 2000 deployment.

Sample Times	4 meter dep	loyment depth	9 meter deployment depth		
Sample Times	p-value F-value		p-value	F-value	
Spring 2000					
(Start)	< 0.001	39.01	< 0.001	39.01	
Autumn 2000	< 0.001	19.99	< 0.001	16.78	
Spring 2001	< 0.001	20.37	< 0.001	25.55	
Autumn 2001	N/A	N/A	N/A	N/A	
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Table 8. ANOVA ($\alpha = 0.05$) comparing sock types (4M, 5M, 6M and TMM) for mean seed size (mm) at start of trials and each subsequent sample period – 4 m and 9 m deployment depths, Salmonier Cove, autumn 2000 deployment.

Sample Times	4 meter dep	oyment depth	9 meter deployment depth		
	p-value	F-value	p-value	F-value	
Autumn 2000					
(Start)	0.004	4.51	0.004	4.51	
Spring 2001	< 0.001	33.97	< 0.001	52.10	
Autumn 2001	< 0.001	12.31	0.020	3.30	

Table 9. Mean mussel sock yield for each mesh type at 4 m and 9 m deployment at Salmonier Cove, after one year in-sock, spring 2000 deployment. *Sock weight after % fouling removed. Note: No socks at 4 m remained for autumn 2001 sampling.

Depth	Sock Type	Sock Weight (kg)*	% Foul*	% Mussels < 50 mm	% Mussels > 50 mm	Yield (kg) per Sock >50 mm	Yield (kg) per 30 cm	± S.E
Spring 2000 Deployment – Spring 2001 Sampling								
	_4M	14.35	8.42	48.4	51.6	7.39	0.798	0.14
4	5M	21.33	4.40	52.1	48.6	10.2	1.079	0.05
meters	6M	23.55	2.18	43.8	56.2	10.4	1.044	0.19
	7M	22.72	3.01	48.3	51.7	11.9	1.203	0.25
	4M	9.53	6.59	70.7	29.3	2.29	0.291	0.09
9	5M	13.22	5.61	74.6	25.4	3.39	0.343	0.06
meters	6M	17.49	3.77	58.8	41.0	7.27	0.728	0.12
	7M	17.01	2.86	66.4	33.7	5.762	0.559	0.09
Spring 2	2000 De	ployment	– Autur	nn 2001 Sa	mpling			
	4M	16.00	3.71	42.7	57.3	9.09	0.859	0.14
9	5M	21.11	4.36	26.5	76.5	16.14	1.594	0.05
meters	6M	24.33	3.47	29.6	70.3	17.17	1.633	0.19
	7M	23.77	3.19	26.5	73.7	17.74	1.616	0.14

Table 10. Mean mussel sock yields for each mesh type at 4 m and 9	Horizon m m m m m m m m m m m m m m m m m m m
Salmonier Cove, after one year in-sock, autumn 2000 deployment.	*Sock weight after %
fouling removed.	

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Depth	Sock Type	Sock Weight (kg)*	% Foul*	% Mussels < 50 mm	% Mussels > 50 mm	Yield (kg) per Sock >50 mm	Yield (kg) per 30 cm	± S.E
Autumn 2000 Deployment – Autumn 2001 Sampling								
	4M	11.22	7.08	98.6	2.2	0.25	0.023	0.01
4	5M	12.86	9.03	100	0	0	0	0
meters	6M	17.15	5.72	100	0	0	0	0
	TMM	14.85	8.44	91.3	8.7	1.14	0.099	0.40
	4M	8.79	6.45	99	1	0.061	0.006	0.06
9	5M	12.59	4.45	100	0	0	0	0
meters	6M	17.49	3.77	100	0	0	0	0
	TMM	17.01	2.86	100	0	0	0	0

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Table 11. Mean mussel sock yield for each mesh type at 4 m and 9 m deployment at The Tickle, after one year in-sock, spring 2000 deployment. *Sock weight after % fouling removed. Note: No socks remained for autumn 2001 sampling.

Depth	Sock Type	Sock Weight (kg)*	% Foul*	% Mussels < 50 mm	% Mussels > 50 mm	Yield (kg) per Sock >50 mm	Yield (kg) per 30 cm	± S.E
Spring 2000 Deployment – Spring 2001 Sampling								
	4M	8.99	2.97	39.9	60.0	5.39	0.552	0.10
4	5M	12.11	2.30	31.5	68.7	8.32	0.846	0.02
meters	6M	14.32	3.76	43.2	56.7	8.11	0.799	0.11
	7M							
	4M	6.02	18.12	37.6	62.7	4.08	0.444	0.16
9	5M	10.77	8.25	28.5	72.0	7.69	0.800	0.09
meters	6M	12.48	2.93	62.9	37.2	4.76	0.486	0.09
	7M	13.68	3.50	42.9	57.0	7.73	0.725	0.003

Figures

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Figure 1. Canadian and Newfoundland cultured mussel production and values, 1990-2003. Source: Department of Fisheries and Oceans Canada Aquaculture Statistics (2003).



Figure 2. Example of the longline system employed in Newfoundland.

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Figure 3. Major steps, tasks and general timeline to mussel culture in Newfoundland.



Figure 4. First step in mussel culture – larval monitoring. (A) Plankton tow jar contents poured into bucket. (B) Contents filtered using 500 µm and 80 µm screens. (C) Plankton funnelled into sample jar. (D) Final rinse of screens into sample jar. (E) Label sample jar. (F) Analyze under light microscope. (Source: Macneill et al., 2001)



Figure 5. (A) Traditional single drop socking. (B) New Zealand style continuous socking.



Figure 6. Distinguishing colouration of male and female mussels. Female – top, male –bottom. Approximate size 50 mm.



Figure 7. Mussel life cycle.



Figure 8. Examples of second-set accumulation on mussel socks. (A) Sock free of second-set. (B) Dense second-set on sock. Note only a few commercial sized mussels protruding (area encircled).



Figure 9. Schematic for seed behaviour study carried out at the Ocean Sciences Centre (OSC). Replicate tanks (SNF1, LSNF1....) were aerated with air stones. SNF = small seed no food, SSF = small seed food, LSNF = large seed no food, LSF = large seed food.

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Figure 10. Example of how distance travelled by mussel is measured. Grid marked on the bottom of each replicate tank help to track individual mussel movements over each time period. T_0 = location at start, T_1 = location at first 15 minute increment, up to T_8 (120 minutes total). Distance is measured in mm in a straight line.

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Figure 11. Mussel farm sites used for the second-set experiment. (A) Salmonier Cove (B) The Tickle. Approximate latitude and longitude, (A) 47.5839°, 55.7907° (B) 47.5889°, 55.7523°, respectively.



Figure 12. Sampling station locations for CTD profiling. Salmonier Cove – (A) Inside (lat/long – 47.5925°, 55.7725°), (B) Middle (lat/long – 47.5889°, 55.7835°), (C) Outside (lat/long – 47.5821°, 55.7892°). The Tickle – (A) Inside (lat/long – 47.5953°, 55.7458°), (B) Middle (lat/long – 47.5899°, 55.7513°), (C) Outside (lat/long – 47.5856°, 55.7519°).



Figure 13. (A) Set-up of monthly collector deployments. (B) Deploying collectors at Salmonier Cove in May, 2000.



Figure 14. Set-up for cumulative collector deployments. Deployments arranged in groups (A group contains one collector deployed at 3 m, 6 m and 9 m). Four groups of collectors (4 groups x 3 coll. per group = 12) were deployed each month from May until October (i.e., 12 collectors per month x 5 months = 60 collectors per site). At the end of October, three groups were retrieved from each month (3 groups x 3 coll. per group = 9 x 5 months = 45 collectors total). Another group of collectors were deployed at the last sampling and remained out over the winter but was not used in the trials. One group for each month was a spare.



Figure 15. Set-up for socking trials at Salmonier Cove and The Tickle. Rep 1 - represents a group of four socks (D1-4). Ten groups in all. D1-4 = Sock types in sequence representing increasing density per 30 cm: 4M, 5M, 6M, 7M (TMM*). *TMM square mesh was used for a autumn 2000 deployment instead of 7M square mesh. Both had same tube diameter and mesh size.



Figure 16. Individual total distances traveled (mm) by small and large mussel spat under varying temperature regimes and presences/absence of food. Duration of trials was 120 minutes. (A) small spat, no food. (B) small spat, food. (C) large spat, no food. (D) large spat, food. n=15 per treatment.



□ Small Spat, No Food ■ Small Spat, Food □ Large Spat, No Food ☑ Large Spat, Food

Figure 17. Mean mussel spat movement expressed as body lengths per hour for small and large spat, with and without food being present. Bars represent \pm S.E., n = 15.



Figure 18. Temperatures recorded for Salmonier Cove, April 2000 – November 2001, at depths of (A) 3 m, (B) 6 m and (C) 9 m.

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Figure 19. Temperatures recorded for The Tickle, April 2000 – November 2001, at depths of (A) 3 m, (B) 6 m and (C) 9 m.



Figure 20. Sample temperature changes over (A) 48 hours and (B) 24 hours at The Tickle, during the 2000 monitoring season. Thermographs were deployed at 3 m, 6 m and 9 m. Note that temperatures during these example periods are more stable at 9 m than at 3 m or 6 m.

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Figure 21. Sea surface temperatures during the first half of July 2000 and 2001. Water temperatures on the South Coast averaged 11-14°C during July 1-15, 2000, but only 10-12°C for the same period in 2001(area encircled on map corresponds to temperature bar highlighted. (Fisheries and Oceans Canada - www.mar.dfo-mpo.gc.ca/science /ocean/ias/seawifs/seawifs_1.html)



Figure 22. Sea surface temperatures during the first half of November 2000 and 2001. Water temperatures on the South Coast averaged 4-8°C during November 1-15, 2000, but were 9-12°C for the same period in 2001(area encircled on map corresponds to temperature bar highlighted. (Fisheries and Oceans Canada - www.mar.dfo-mpo.gc. ca/science/ocean/ias/seawifs/seawifs 1.html)



Figure 23. CTD profiles for Salmonier Cove, 2000 field season – inside, middle and outside stations of site. *Note that for June 21, chl-a readings were taken (*), the rest are total chlorophyll.



Figure 24. CTD profiles for Salmonier Cove, 2001 field season – inside, middle and outside stations of site.



Figure 25. CTD profiles for The Tickle, 2000 field season – inside, middle and outside stations of site. *Note that for June 21, chl-a readings were taken, the rest are total chlorophyll.



Figure 26. CTD profiles for the Tickle, 2001 field season - inside, middle and outside stations of site. No CTD profile was done in October.


Figure 27. Mussel meat yields and mussel larval abundance for 2000 and 2001 for (A) Salmonier Cove and (B) The Tickle. Sampling period from April through to November of each season.



Figure 28. Shell length frequency distribution for newly collected spat at Salmonier Cove, 2000 field season. Not graphed is the period May 23 – June 21. Only 2 spat were observed, 2.08 mm and 1.32 mm in shell length.

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Figure 29. Shell length frequency distribution for newly collected spat at Salmonier Cove, 2001 field season.



Figure 30. Shell length frequency distribution for newly collected spat at The Tickle, 2000 field season.



Figure 31. Shell length frequency distribution for newly collected spat at The Tickle, 2001 field season. Collectors deployed in June and after September were destroyed in storms.



Figure 32. (A-B) Newly settled spat from Salmonier Cove – July 2000 deployment, August collection period. (approximately 30 days). Note the high density. Spat are about 400 µm to about 4 mm in length. (C) Mussel collector rope showing primary (I) and secondary (II) settlement mussels.



Depth of Deployment in Meters

Figure 33. Cumulative collector results for The Tickle – mean spat number and size (mm) per collector at 3 m, 6 m and 9 m. Number of days deployed are noted on each figure along with month at start of collection period - a, b denote months where cumulative spat collection were significantly different for month indicated. Note the scale differences of mean spat numbers for each deployment month.



Depth of Deployment in Meters

Figure 34. Cumulative collector results for Salmonier Cove – mean spat number and size (mm) per collector at 3 m, 6 m and 9 m, \pm S.E. Number of days deployed are noted on each figure along with month at start of collection period. Note the scale differences of mean spat numbers for each deployment month – a, b denote months where cumulative spat collection were significantly different for month indicated.











Sample Fellou

Figure 37. Mean number of mussels per 30 cm of socking at Salmonier Cove, (A) 4 meters (B) 9 m – spring 2000 deployment, sampled autumn 2000 and spring 2001. All socks were lost at 4 m during storm by the autumn 2001 sampling period. Bars represent means \pm S.E., n = 3.



Figure 38. Mean number of mussels per 30 cm of socking at Salmonier Cove, (A) 4 m (B) 9 m – autumn 2000 deployment, sampled spring 2001 and autumn 2001. Bars represent means \pm S.E., n = 3.



Sample Period

Figure 39. Mean number of mussels per 30 cm of socking at The Tickle, (A) 4 m, (B) 9 m – spring 2000 deployment, sampled autumn 2000 and spring 2001. All socks were lost before the autumn 2001 sampling could take place. Bars represent means \pm S.E., n = 3.



Mussel size (mm)

Figure 40. Length (mm) frequency of mussels sampled from each trial sock type (4M-7M) deployed at Salmonier Cove at 4 m and 9 m depth, spring 2000 deployment – spring 2001 sampling (n = 600).



Figure 41. Length (mm) frequency of mussels sampled from each trial sock type (4M-TMM) deployed at Salmonier Cove at 4 m and 9 m depth, autumn 2000 deployment – autumn 2001 sampling (n = 600).



Mussel size (mm)

Figure 42. Length (mm) frequency of mussels sampled from each trial sock type (4M-7M) deployed at The Tickle at 4 m and 9 m depth, spring 2000 deployment – spring 2001 sampling (n = 600).



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Figure 43. Mean socked mussel size (mm) for Salmonier Cove, spring 2000 sock deployments, sampled in the autumn 2000, spring 2001 and autumn 2001. (A) 4 m, (B) 9 m. Note all socks from 4 m were lost at the time of the autumn 2001 sampling. Sizes represent means \pm S.E, n = 600, except n = 200 for spring 2000 starting sizes.



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Figure 44. Mean socked mussel size (mm) for Salmonier Cove, autumn 2000 sock deployments, sampled in the spring 2001 and autumn 2001. (A) 4 m, (B) 9 m. Sizes represent means \pm S.E, n = 600, except n = 200 for autumn 2000 starting sizes.



Figure 45. Mean socked mussel size for The Tickle, spring 2000 sock deployments, sampled in the autumn 2000 and spring 2001. (A) 4 m (B) 9 m. Note all socks for an autumn 2001 sampling were lost. Bars represent means \pm S.E., n = 600.



Figure 46. Mussel sock yields for Salmonier Cove, spring 2000 deployment, 4 m and 9 m depth. (A - B) One year in sock. (C) 1.5 years in sock. Weights are standardized to kg per 30 cm of socking. 4M - 7M are socking mesh types used. Bars represent means \pm S.E., n = 3. Note, no data for autumn 2001 at 4 m are available.



Figure 47. Mussel sock yields for Salmonier Cove, autumn 2000 deployment – autumn 2001 sampling. (A) 4 m (B) 9 m. Weights are standardized to kg per 30 cm of socking. 4M – TMM are socking mesh types used. Bars represent means ± S.E., n = 3.



Figure 48. Mussel sock yields for The Tickle, spring 2000 deployment – spring 2001 sampling. (A) 4 m (B) 9 m. Weights are standardized to kg per 30 cm of socking. 4M - 7M are socking mesh types used. Bars represent means \pm S.E., n = 3.



Figure 49. Example of excessive broken mussel shells inside a mussel sock. Poor sock quality is a major contributor to second-set accumulation.



Figure 50. (A) Low density sock showing second-set accumulation. (B) High density sock free of second-set. Note arrangement of mussels and lack of opportunity room for additional settlement on the high density sock.







