# The potential of integrated open-water mussel *(Mytilus planulatus)* and Atlantic salmon *(Salmo salar)* culture in North West Bay, Tasmania.

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January 2001

# Abstract

**The potential of integrated open-water mussel *(Mytilus planulatus)* and Atlantic salmon *(Salmo salar)* culture in North West Bay, Tasmania.**

Tasmanian blue mussels *(Mytilus planulatus)* were cultured at four sites in the vicinity of an Atlantic salmon *(Salmo salar)* farm in North West Bay, Tasmania. The main objective of the study was to evaluate the potential of integrating mussel and salmon culture within a common multi-species marine farm. Filter-feeding bivalves cultured near open fish cages might obtain additional food supplies directly from particulate wastes (excess fish feed and faeces) and indirectly from enhanced phytoplankton production stimulated by dissolved nutrient wastes. Potential benefits of such integration include enhanced bivalve growth, increased productivity of a coastal marine fish farm, and reduced fish farm waste loadings and their associated environmental impacts.

Mussels were cultured for fourteen months, suspended from four longlines positioned at increasing distances (70, 100, 500 and 1200 meters) from the Aquatas Pty. Ltd. salmon farm in North West Bay. Mussels were monitored monthly for various growth and biochemical parameters including shell length, whole live weight, meat weights, total biomass, condition index, glycogen content, stable isotope ratios ($\delta^{13}C$ and $\delta^{15}N$), reproductive development and survival. Water quality and environmental parameters at each longline site were monitored weekly over the same period.

The performance of mussels grown within the fish farm lease (70 m and 100 m from the fish cages) was not appreciably different from that of mussels grown distant to the farm. Mussels spawned twice with no differences in biomass production among sites ($P > 0.05$). The final sample in May 1996 indicated no site differences for any parameter except for shell length ($P < 0.0001$) and condition index ($P < 0.01$). However, site differences were minor, with final mean shell lengths and condition (dry meat weight/internal shell cavity capacity) being within 2.0 mm and 17‰, respectively.

Similar mussel growth was likely due to similarities in environmental parameters among longline sites, most importantly food quantity and quality (POM, chlorophyll-a, %POM). Growth of mussels cultured within the fish farm was not enhanced due to several contributing factors: (a) solid waste loadings (feed particles and faeces) from the farm were too diluted to significantly increase particulate food concentrations above ambient levels; (b) phytoplankton production within the farm was not enhanced; (c) mussels may have been cultured too distant to intercept settling particulate wastes

emanating from the fish cages; and (d) ambient seston concentrations were above, or near, the pseudofaeces threshold concentration for most of the trial period. Therefore, mussels cultured within the farm site were physiologically limited in securing a significant quantity of additional food.

Increased bivalve growth through integration with open-water fish culture may only be achievable in coastal areas where ambient food concentrations are below the pseudofaeces threshold for extended periods, particulate fish farm wastes significantly increase particulate food concentrations above ambient levels, and bivalves are cultured in a suitable position to intercept these waste particles.

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# 1. General Introduction

The culture of fish in open-water sea cages, as with the aquaculture industry generally, has expanded considerably over the past few decades. The importance of fish culture will inevitably increase as the human population grows and wild fish stocks become more and more depleted. However, with the expansion of the aquaculture industry, so to have concerns regarding its impact on the environment. One of the major challenges for the sustainable development of the aquaculture industry is to ensure that increased environmental degradation does not accompany its projected expansion.

## 1.1 Environmental Impacts of Marine Fish Farming

Intensive fish cultivation generates significant amounts of organic and inorganic wastes. Open-water marine fish farms, therefore, represent point sources where nutrients and organic matter are introduced into coastal areas. All of the components in fish feed together with the by-products of fish metabolism are potential waste products. The quantity, composition, and physical nature of fish farm wastes are a reflection of the husbandry practices employed, the composition of fish food, and the digestibility of feed components. As such, environmental waste loadings may be highly variable among farms. However, as fish generally retain only a fraction of the total nutrients in feeds, the potential for environmental nutrient enrichment is clear. Estimates of feed nutrients ultimately lost to the environment from salmonid culture: 75-78% of feed carbon, or 878-952 kg C·tonne fish production$^{-1}$ (Hall et al., 1990); 67-71% of feed nitrogen, or 95-102 kg N·tonne fish production$^{-1}$ (Hall et al., 1992); and 81% feed phosphorus, or 21.9 kg P·tonne fish production$^{-1}$ (Holby and Hall, 1991); provide some insight into the waste loadings that may be expected.

The environmental impacts of finfish farming have been extensively documented in the literature. Waste produced by fish farms can be divided into two categories: particulate and soluble wastes. Particulate material is introduced to the marine environment as uneaten food, feed dust, and faeces, which eventually accumulates on the sea bottom. Within this material, organic carbon and nitrogen, as carbohydrates, proteins and lipids, are particularly important. The impact of particulate wastes is largely on the benthic environment; significant alterations of the sediment biogeochemistry and benthos have been well-documented (Brown et al., 1987; Gowen and Bradbury, 1987; Gowen et al., 1988; 1991; Kaspar et al., 1988; Ritz et al., 1989; Woodward, 1989; Wildish et al., 1990; Lim, 1991; Woodward et al., 1992; Johnsen et al., 1993; Johannessen et al., 1994; Krost et al., 1994; Findlay et al., 1995). Benthic

decomposition of sedimented organic material results in increased oxygen consumption and a decrease in sediment redox potential. Sediments can become anoxic; anaerobic decay processes may result in the generation of hydrogen sulphide, ammonia and methane - all detrimental to farmed fish. Low dissolved oxygen concentrations may also be transferred to the overlying water column and the fish cages. Benthic macrofauna in these anoxic sediments can become impoverished or absent. High sediment nutrient concentrations increase effluxes to the water column and together with fish metabolic wastes, of which ammonia excretion is most notable, may lead to hypernutrification. Dissolved nutrient wastes include ammonia, urea, bicarbonate, phosphate, silicate, vitamins, therapeutics and pigments. During periods of nutrient-limited growth, nutrient wastes have the potential to promote phytoplankton production and, under extreme conditions, eventually lead to eutrophication within a water body (Gowen and Bradbury, 1987; Frid and Mercer, 1989; Kaaetvedt et al., 1990; Persson, 1991). As well as affecting the natural environment, eutrophication might also pose a threat to farmed fish through depletion of dissolved oxygen or by enhanced growth of toxic or injurious species of phytoplankton.

## 1.2 Strategies to Reduce Environmental Impacts of Fish Farming

In many countries, concerns regarding the impact of aquaculture on the environment have led to the introduction of government legislation; which is likely to become increasingly important in regulating the aquaculture industry. Government regulations have limited farm production and set strict guidelines regarding feed composition; feed digestibility; maximum annual feed volume per fish farm (feed quota); food conversion ratios (FCR); and maximum pollution loadings for biological oxygen demand (BOD), suspended solids, total nitrogen, ammonia, and total phosphorus (Jensen, 1991; Kiarskou, 1991; Michelson, 1991). Monitoring programmes have been designed and implemented to keep operational activities, waste loadings, and impacts associated with aquaculture farms under surveillance (Wildish et al., 1990a; Caine and Castledine, 1991; Levings 1994; Ervik et al., 1997).

Research and development programmes have led to reductions in environmental waste loadings. These have ben achieved through improvements in feed formulations and processing technology (Cowey and Cho, 1991), a better understanding of feeding behaviours (Blyth et al., 1999) and refinements to feeding strategies and husbandry practices (Seymour and Bergheim, 1991; Blyth and Purser, 1992; Blyth et al., 1993; Talbot, 1993). Many of the approaches addressing the treatment of aquacultural wastes, in both land-based and open-water systems, are based on technical solutions

involving mechanical separation, sedimentation, filtration, or dispersion techniques (Gowen et al., 1991; Michelsen, 1991; Bergheim and Forsberg, 1993; Ackefors and Enell, 1994). Culturing fish in solid plastic bags, rather than mesh cages, has been proposed to contain particulate wastes in open-water systems (Fast, 1991); this type of system is currently being trialed by a salmon producer (Aquatas Pty. Ltd.) in southern Tasmania.

## 1.3 Integrated Aquaculture Systems

New and innovative aquaculture systems incorporating "ecological engineering" principles may be promising alternative strategies for reducing aquacultural wastes and environmental impacts. The basis of ecological engineering is the integrated culture of several species of organisms into an overall ecological system; waste discharges from one culture species become nutrient resources for other species at different trophic levels. Waste nutrients are ultimately removed from an integrated system upon harvest of the cultured species. Various combinations of integrated culture, including fish, seaweed, molluscs and shrimps, have been proposed, or trialed with varying degrees of success. Trials in land-based aquaculture systems have certainly demonstrated the potential benefits of integrated culture; cultured seaweeds and bivalves have been effective in reducing nutrient and particulate waste loadings in fish and shrimp farm pond effluent (Hopkins et al., 1993; Lin et al., 1993; Shpigel and Fridman, 1990; Shpigel et al., 1993a; 1993b; 1997; Buschmann et al., 1996; Jara-Jara et al., 1997; Jones and Preston 1999). However, the development of an effective integrated approach for open-water culture systems may be more challenging, as the dispersion of wastes is more difficult to control.

### The Folke and Kautsky Model

Many of the environmental and economic concerns regarding open-water marine finfish aquaculture, particularly salmon farming, have been addressed by Carl Folke and Nils Kautsky (Folke and Kautsky, 1989; 1992; Folke et al., 1994). They maintain that the development of a sustainable salmonid aquaculture industry requires a reduction in natural resource utilisation - while not exacerbating environmental degradation. Folke and Kautsky suggest: "the more a cultivation system recognises and mimics natural ecosystem functions, the fewer resource inputs are required, and reduced environmental impacts can be expected."

A model of coastal open-water aquaculture has been proposed by Folke and Kaustky; the model is based on ecological engineering principles and involves the integrated

culture of salmon, filter-feeding bivalves such as mussels, and seaweeds (Folke and Kautsky, 1989; 1992; Kautsky and Folke, 1989; 1990; Folke et al., 1994; Kautsky et al., 1997). They maintain that this model includes elements that potentially: reduces the dependency on external environments for food and energy, recycles waste nutrients among the culture components, assists in utilising nutrients from terrestrial sources, increases efficient utilisation of coastal marine resources, increases the stability of aquaculture systems by reducing environmental impacts, and produces multiple market products.

The Folke and Kautsky model contains several feedback loops whereby wastes generated from one component of the system may be utilised by others (Fig. 1.1). Fish culture directly contributes to increased mussel production, as mussels can utilise waste fish feed particles and faeces. Dissolved metabolic wastes from both fish and mussel culture may enhance seaweed and phytoplankton production. Increased phytoplankton abundance may further enhance the growth of mussels. The filtering activity of cultured mussels may reduce the severity of algal blooms and turbidity in the water, benefiting fish growth and survival. Therefore, some of the fish farm nutrient wastes, rather than being lost to the environment in traditional monoculture systems, are ultimately removed upon harvest of mussels and seaweed; these wastes have the potential to become economic resources, as they can provide additional cash crops or be partly recycled as fish feed. The higher the proportion of mussels and seaweeds recycled back into fish feed, the fewer are new nutrients introduced into the local area; less nutrients are imported from other areas (marine and terrestrial) in the manufacture of fishmeal and plantmeals, thereby reducing the potential for environmental degradation in the vicinity of fish farms. Folke and Kautsky propose that such a system could also transform excess nutrient inputs from domestic terrestrial sources into useful biomass and ultimately removed from the system at harvest. Therefore, they suggest such a culture system might actually improve environmental quality instead of contributing to environmental degradation, as is the case with the presently applied farming technology (Kautsky and Folke, 1989).

The Folke and Kautsky model of integrated marine aquaculture is certainly simple and, intuitively, appears promising. It views aquaculture from a more ecological perspective. Waste from one component is used as a resource input by others, leading to reduced environmental impacts and increased outputs of seafood and seaweeds. However, it is only a conceptual model; practical feasibility tests need to be carried out. It is necessary to determine the abiotic limitations and biological characteristics of the cultured species, the scale of the different cultures appropriate to the carrying capacity of the water body, and to assess the applicability of integrated systems under different environmental conditions.

Separate components of the Folke and Kautsky model have shown promise in some previous studies. As mentioned, bivalves and seaweeds have been successfully incorporated into land-based aquaculture systems where they fulfil a role as biofilters in ponds or effluent waters. In coastal environments, the potential of integrating seaweed culture with salmon farms appears promising (Petrell et al., 1993; Subandar et al., 1993; Troell et al., 1997). Enhanced growth of mussels (Wallace, 1980) and oysters (Jones and Iwama, 1990; 1991) cultured near salmon pens has also been demonstrated, indicating bivalves are capable of utilising particulate fish wastes and/or increased phytoplankton production in the vicinity of fish farms. However, there are other studies where little or no increased growth was displayed by bivalves cultured near fish cages (Taylor et al., 1992; Okumus, 1993; Stirling and Okumus, 1995; Gryska et al., 1996).

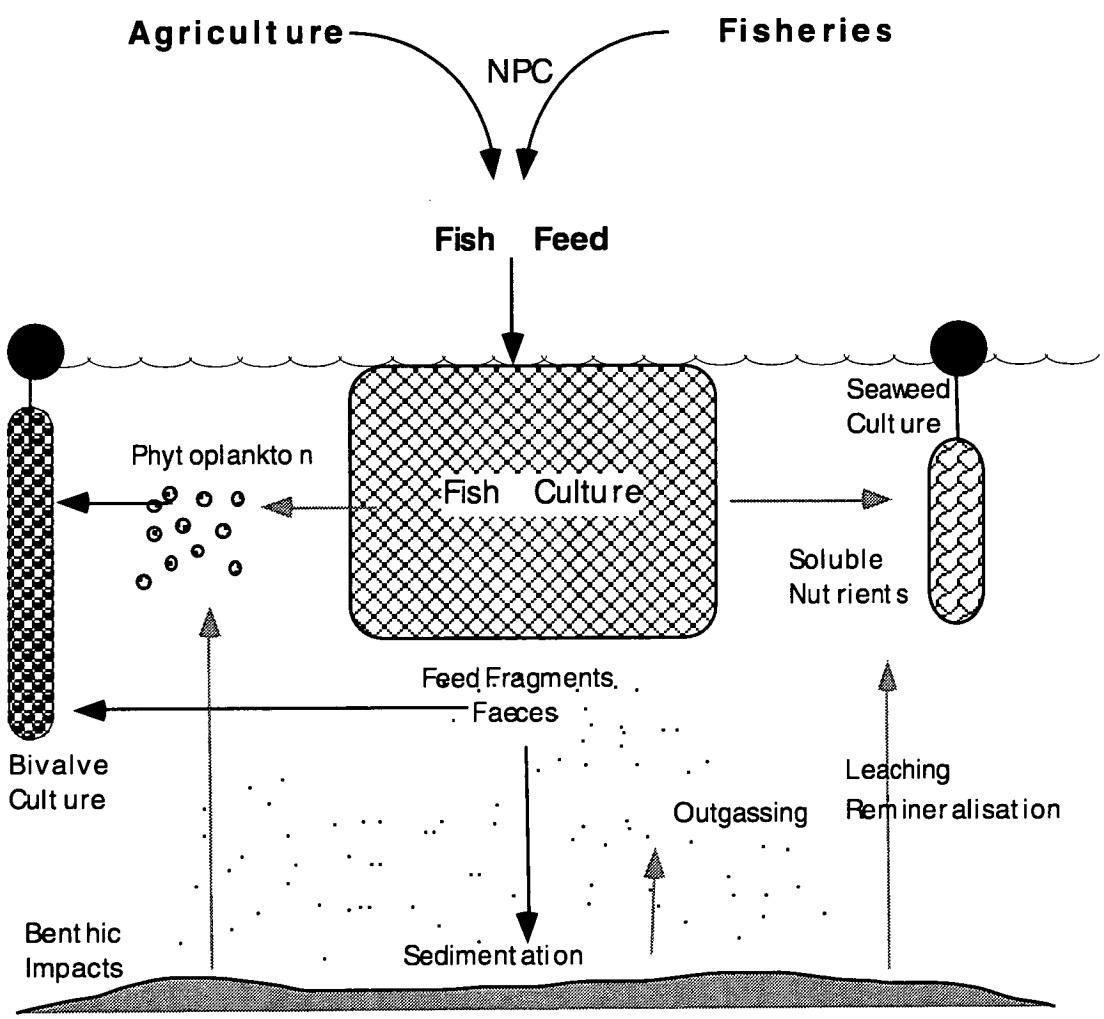


**Figure 1.1.** Nutrient flows within an integrated aquaculture system of seaweeds, mussels and salmon, where nutrient wastes from one culture are used as resources by the others. (N: nitrogen, P: phosphorus, C: carbon). Solid arrows represent nutrients in particulate form and stippled arrows represent dissolved nutrients. Modified from Kautsky et al. (1997).

An investigation of the feasibility of open-water integrated bivalve-fish culture has not been attempted in Australia. Further, research into these systems has been generally scarce elsewhere; studies that have been conducted have drawn conflicting conclusions regarding the merits of integrated bivalve-fish culture. Therefore, a further study of bivalve-fish culture was deemed appropriate. This study might shed additional light on the factors contributing to the success, or failure, of open-water integrated bivalve-fish culture systems.

## 1.4 Objectives of the Study

The primary objectives of this study were to:

1. Culture mussels *(Mytilus planulatus)* in close association with an Atlantic salmon *(Salmo salar)* farm to determine the potential benefits of integrated bivalve-fish seacage culture relative to mussels cultured distant to the farm. The possible benefits of most interest were enhanced mussel performance and reduced particulate waste loadings from the salmon farm into the coastal environment. Mussel performance will be assessed by a number of biological parameters, monitored concurrently, that are known to be influenced by food availability. These parameters include growth in shell length, live weight, meat weight and biomass, as well as condition, glycogen content, reproductive development and survival. However, any reductions in fish farm waste loadings, as a consequence of integrated culture, would only be implied by enhanced growth of mussels cultured adjacent to fish cages relative to control groups. Stable isotope analysis might prove to be a useful tool in determining whether fish farm wastes were actually consumed by mussels cultured on the farm.
2. Investigate the effect of depth on mussel growth. Different studies have reported increases, decreases, or no effect, on mussel growth with increasing depth.
3. Investigate temporal variation, as well as the above spatial variation, of mussel performance indices over the course of the trial period.
4. Monitor mussel condition and reproductive development, not only to assess differences between bivalve culture sites, but also to identify major spawning periods. This information would be beneficial to mussel growers, identifying optimum periods for harvest and deployment of spat collectors.
5. Monitor environmental parameters at mussel culture sites within North West Bay to assess the extent to which they may be influenced by the fish farm and to identify those factors most likely affecting mussel growth in North West Bay.

# 2. The Study Area

## 2.1 Characteristics of North West Bay

North West Bay (NWB), is a small trapezoidal-shaped bay, located approximately twenty-two kilometres southwest of Hobart, Tasmania, Australia (43°3'S:147°17'E, Fig. 2.1). Much of the bay is bordered by undeveloped land and the environs retain an essentially semi-rural character. The only significant centres of population are the small townships of Margate, Snug, Electrona, and Coningham, on the western shore, and Howden on the eastern shore. NWB has a small surface area, approximately nineteen square kilometres, and is protected from oceanic swells by Bruny Island to the south. The entrance to the bay, approximately three kilometres wide, does not link directly to the open sea. The mouth opens out into the northern section of the D'Entrecasteaux Channel, with links to Storm Bay, the mouth of the Derwent River estuary, in the north, and to the Tasman Sea in the south.

General background information of the environmental characteristics of North West Bay are provided in several studies conducted and reviewed by Matthews and Volframs (1978 and references within). Specific environmental conditions of NWB during the present study are provided in Section 4.

The climate of the region can be broadly classified as maritime temperate. Mean monthly air temperatures range from 4.4 - 11.4°C (minimum-maximum) in July to 11.8 - 21.5°C in February (93 year average). Rainfall is evenly distributed throughout the year; there is an average of fifteen rain days with 50 mm of precipitation per month and an annual mean rainfall of 634 mm.

The total watershed for the bay is fairly small, covering an area of about 290 square kilometres. The total annual freshwater discharge into North West Bay has been estimated to be in the order of $117.8 \times 10^6$ $m^3$. The bulk of freshwater inputs comes from the catchments of the North West Bay River (79% of total), Coffey Creek (1%), and Nierinna Creek (6%) draining into the northeast corner of the bay, and Snug River (11%) draining into the western corner of the bay.

Most of the bay is fifteen to twenty metres in depth, being formed as a system of drowned river valleys. The eastern shore drops off rapidly into a deep channel, which may be one of these drowned river valleys. In contrast, parts of the western shore are fairly shallow with numerous areas of sand and mudflats, these being most extensive in the northwestern corner of the bay.

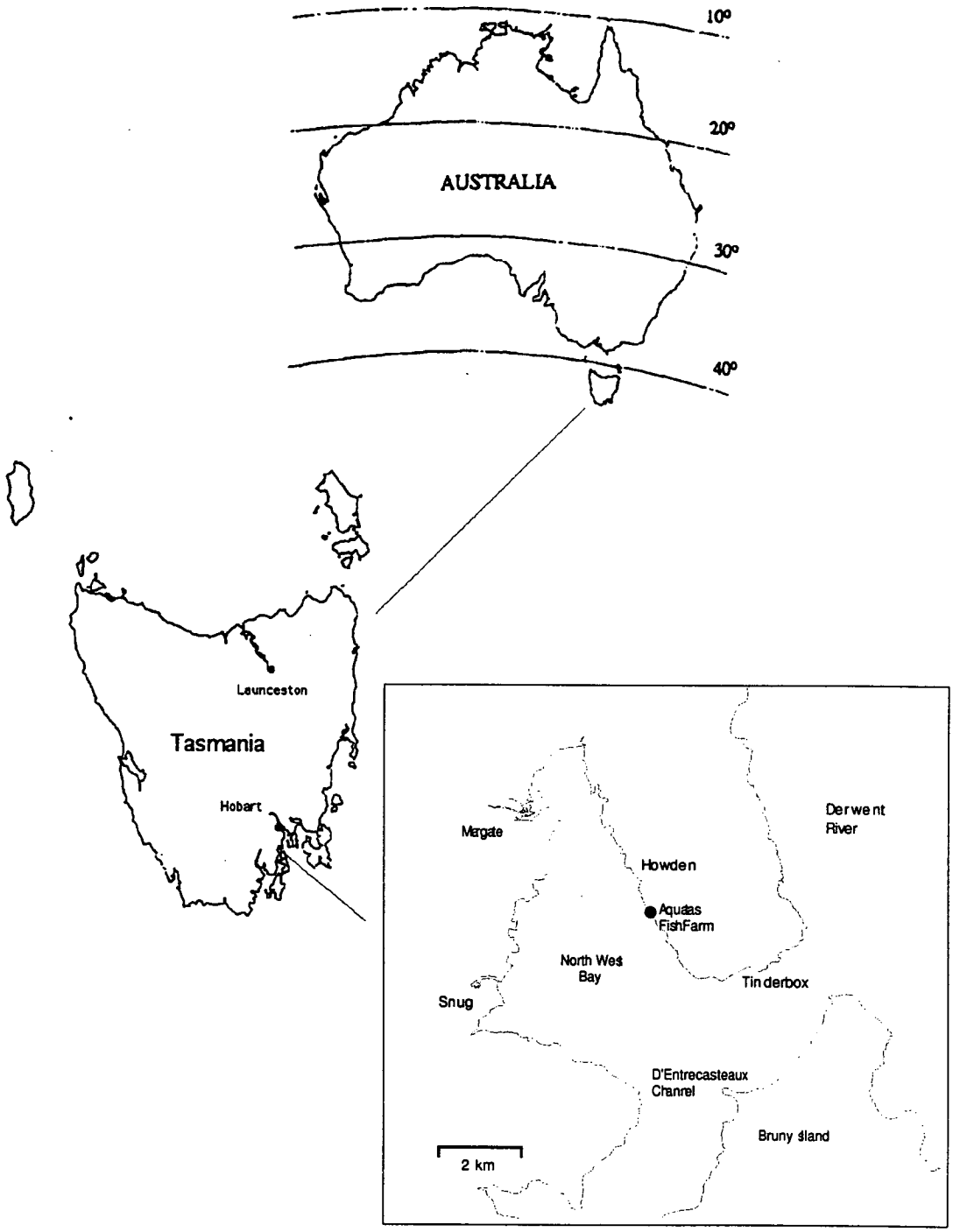


**Figure 2.1**. Location of the study area: North West Bay, Tasmania.

The circulation patterns that occur within North West Bay are a response to the combined effects of tides, meteorological forces and freshwater inflows. Northwesterly and westerly winds tend to predominate throughout the year, except for summer afternoons when winds are mainly southeast due to the influence of a seabreeze. Winds occasionally exceed 60 $km \cdot h^{-1}$, while sea-breezes rarely exceed 16 $km \cdot h^{-1}$. It has been shown that surface currents respond quickly to significant changes in wind speed and direction over a short period of time, with surface wind-drift currents varying from 1% to 4% of the wind speed. Wind stress is not significant at

depths below 5 m; its predominant influence on the bay is to act as a transient mixing mechanism.

Tides in the bay are semi-diurnal and correspond closely with tide tables that have been prepared for the Port of Hobart. Intertidal heights are fairly small with a maximum intertidal range of about one metre and a mean range in the order of half a metre. The total low-water volume of the bay is approximately 277 x $10^6$ $m^3$. For a 0.5 m mean tide, the volume contained in the tidal prism is about 9.52 x $10^6$ $m^3$, or 3.4 percent of the total low-water volume of the bay. Based on the assumptions that tide is the dominant exchange mechanism for the bay, and that complete mixing occurs on each tidal cycle, a mean tide of 0.5 m would indicate a mean flushing time of fifteen days. However, from observations of salinity and temperature fluctuations, Mathews and Volframs (1978) have suggested that complete flushing of bay water may be achieved in seven days. They have suggested that the flow regime in the D'Entrecasteaux Channel may establish a circulation pattern within North West Bay, producing a higher exchange rate than expected from tidal prism considerations alone. This circulation pattern probably enhances mixing throughout the bay.

Current velocities range up to 30 $cm \cdot sec^{-1}$ and, at times, sustained flows in excess of 10 $cm \cdot sec^{-1}$ may occur for periods longer than 12 hours. Current direction has been shown to be distinctly bimodal, displaying opposing flows with peaks at 120° and 335°.

Apart from a small estuarine region near the mouths of the North West Bay River and Nierinna Creek in the northwest corner, the bay is generally euhaline (marine) throughout the year. The bay is well-mixed with little vertical stratification. Salinities are similar to those of coastal waters, generally exceeding 34.8‰, although near surface salinities occasionally drop below 30‰ following periods of heavy rainfall. Without sustained rainfall this stratification decays fairly rapidly. Fluctuations in salinity, other than the extremes observed following heavy rainfall, have been attributed to external estuarine influences from the Derwent River to the north and possibility the Huon River to the south.

Surface sea water temperatures show a strong seasonal trend, with winter minima occurring in July (9°C) and summer maxima in February (19°C).

## 2.2 The Aquatas Fish Farm

The study of integrated mussel-salmon culture was conducted from March 1995 to May 1996 in the vicinity of the Aquatas smolt-rearing fish farm located at Gunpowder Jetty on the eastern shore of North West Bay (Fig. 2.1 and 2.2). This was the only fish farm within the bay. At the time of this investigation, standard practice was to introduce Atlantic salmon smolts to the farm during September and October and reared in Polarcirkel™ cages (60 m circumference and 7 to 10 m depths). After seven to ten months, these fish were transferred to a growout site in the D'Entrecasteaux Channel. Twenty-three cages were in use during the 1994-95 season and 17 cages during the 1995-96 season. Sixteen cages were anchored on either side of a central 'backbone' to form two parallel rows, with the remaining cages variously located within the lease area (Fig. 2.2). Additional cages of harvest fish were periodically towed onto the site and temporarily held until harvested by pressure-vacuum pump onto the farm jetty. The farm site was normally left fallow for approximately 3-4 months during the winter months (June through mid-September) until a new smolt year-class was introduced during September-October.

This study spanned approximately one and a third fish-growing seasons: the last three and a half months of the 1994-95 year class (March 1995 to June 1995) and the entire 1995-96 year class (September 1995 to May 1996). At the start of the mussel growth trial 243,429 fish were on-site with a total biomass of 247.8 tonnes. These fish remained on-site until April-June 1995 (410 tonnes) when they were progressively transferred to a growout site in the D'Entrecasteaux Channel. The farm site was left fallow for approximately 3.5 months from June-September 1995. Approximately 221,000 new smolts were introduced in September-October 1995, with an initial biomass of 22.5 tonnes. These fish remained on-site until April-May 1996 (350 tonnes), when they were progressively transferred to grow-out sites.

Fish were fed *ad libetum* by hand (new smolts) or feed blowers. Feed was distributed to cages on a rotating basis throughout the daylight hours, with each cage fed 3 to 6 times daily depending on time of year and appetite. Daily feed rations ranged from 1% to 2% of biomass with approximately 30-60% of the daily ration provided during the first one or two morning feeds. Feed composition was approximately 45:22:16:9:8% (protein:fat:carbohydrate:ash:moisture; Pivot Aquaculture, extruded Salmon Grower Diet) with an apparent digestibility of 0.83. Feed pellets ranged from 2 mm – 9 mm and the quantity of feed delivered ranged up to 123 tonnes per month, depending on fish biomass. Mean monthly FCR was approximately 1.3.

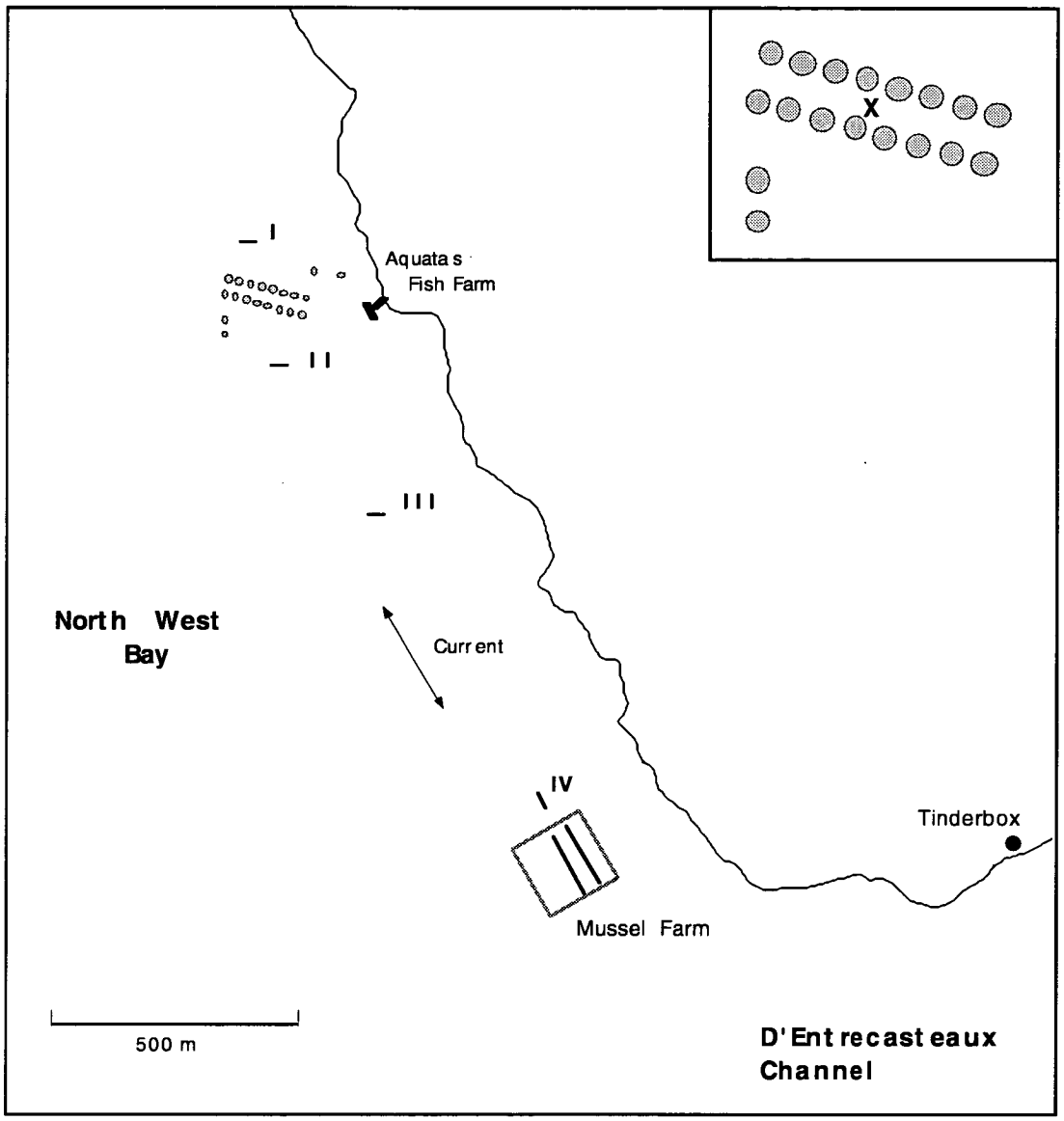


**Figure 2.2.** Location of the Aquatas fish farm in North West Bay and relative positions of fish cages and experimental mussel longline sites (I, II, III, IV). Site I was approximately 70 m north and sites II, III and IV were approximately 100, 500 and 1200 m south of the main fish cage unit. Water quality parameters were monitored at each longline site and at the centre of the main fish cage unit (inset, site X).

# 3. Experimental Design and Sampling Procedures

The primary objective of this study was to determine the effects of a salmon farm on mussel growth, condition and survival. In particular, the question of most interest was "is growth, condition and/or survival enhanced when mussels are cultured in an integrated mussel-salmon culture system?" Should this question be answered in the affirmative, it seems reasonable that the integrated mussels probably obtain a nutritional advantage from fish farm waste material. In so doing, the environmental loadings of organic and nutrient wastes from the fish farm might be reduced through integrated culture with mussels.

An experiment was designed to compare the growth of mussels cultured near fish farm cages with growth of mussels cultured more distant to the farm. The experiment also investigated the influence of depth on growth. It was desirable that the study incorporate materials and culture techniques used in current industry practices. The approach taken was to undertake a study that provided outcomes relevant to the Tasmanian mussel industry; that is, results which might be expected if mussels were to be cultured commercially at the Aquatas fish farm. Therefore, in accordance with industry practice, longline culture systems were chosen as the experimental units for mussel growout.

## 3.1 Experimental sites

Four sites for mussel culture were selected at increasing distances from the main fish cage unit (Fig. 2.2). All sites were in-line with the prevailing current passing through the farm to maximise the probability of particulate wastes, originating from the fish farm, passing through the mussel culture sites. Two sites (I and II) were located within the boundaries of the Aquatas farm lease on each side of the main fish cage unit in the directions of the ebb and flood current flow. These longlines were positioned as close to the cage system as practically possible so as not to interfere with the day to day operations of the farm. Therefore, they represent the two closest positions to the fish cages that would be acceptable to Aquatas for commercial mussel culture. Longlines at sites I and II were anchored approximately 70 m north and 100 m south, respectively, of the main fish cage unit. A site near the mouth of NWB, approximately 1200 m south of the fish farm and near a small subsurface mussel farm, was considered to be beyond the direct influence of the fish farm (site IV). The mussel farm comprised only two 100 m longlines of socked mussels suspended 5 m below the surface and was approximately 100 m south of site IV. A fourth site (site III), approximately 500 m south of the fish cages, was chosen as an intermediary between the two fish farm sites

and site IV. This site was considered as an "off-farm" site which might help establish the spatial influence of the fish farm on mussels cultured "downstream" of the farm. All longline sites were approximately 100 m from the eastern shoreline of NWB.

The orientation of the longlines at sites I, II and III was perpendicular to the current flow to reduce the possibility of within-longline growth differences due to downstream seston depletion or "shadowing" effects (Wildish and Kristmanson, 1985; Fuentes et al., 1994; Mueller, 1996). The orientation of the longline at site IV was parallel with the prevailing current to reflect the orientation of the two longlines at the nearby mussel farm. Therefore, mussel growth at site IV was expected to be a reasonable reflection of growth at the commercial mussel farm. Bottom depths were 20 m at sites I, II and III and 17 m at site IV. The maximum intertidal range in NWB is about 1 m with a mean range in the order of 0.5 m (Matthews and Volframs, 1978).

## 3.2 Stocking procedures

Mussels cultured in this study were blue mussels, *Mytilus planulatus*, common to Tasmanian waters (Richmond, 1990). However, it has been suggested that these mussels may actually be the species *Mytilus galloprovinciallis* (McDonald et al., 1991; Gosling, 1992a; 1992b). Mussels were stripped from the floating fishcage rings (Polarcirkel™) at the Aquatas smolt-rearing site in February 1995. These seed mussels were de-clumped and roughly graded into three size classes using a homemade grading machine. Intermediate-sized mussels from this grading were selected for growout and poured into 168 four-meter long mesh stockings (hereafter referred to as socks or droppers) after removal of some of the damaged and empty shells. Each sock comprised a central core of 10 mm polyethylene rope surrounded by a tube of cotton "mussock" material and reinforced by an outer layer of nylon mesh tubing (70 mm Netlon™). The cotton mussock decays after a few weeks in water during which the mussels attach to the central core rope, the Netlon mesh and each other. The Netlon mesh size was chosen to allow the mussels in the selected size range to grow through the mesh-opening over time.

Fourty-two socks were deployed to each of the four experimental longlines (22 m surface length), suspended 1 m below the surface at 0.5 m intervals. The process of mussel collection, grading, socking and deployment took approximately two weeks during February 1995. Mussel sampling commenced on 1 March 1995 to establish initial shell dimensions (length, height, width), live weights, meat weights, condition, sock densities and post-stocking survival.

## 3.3 Mussel sampling procedures

With relatively large experimental units (22 m longlines), unforeseen within-longline variation of environmental conditions (physical, chemical and/or biological factors) influencing within-longline mussel performance was considered a possibility. With this in mind, it was deemed prudent to ensure mussels were sampled from across the entire length of each longline, thereby providing the best estimate of mean mussel growth at each site. This was achieved by arbitrarily dividing each longline (42 mussel socks) into three equal sections (labelled A, B, and C) of fourteen socks (labelled 1 through 14) per section (Fig. 3.1). Two socks from each longline section were randomly selected each month (ie. six socks per site per month). To investigate the effect of depth on growth, mussels were sampled from the top and bottom of selected socks.

Socks were removed from the longline and lifted into a flat-bottomed skiff. Twenty-five centimeter sections from the top (1 m growing depth) and bottom (5 m growing depth) of selected socks were stripped of mussels. Mussel samples were placed in labelled plastic mesh bags, submerged in water (20 l plastic buckets) and transported to the Tasmanian Aquaculture and Fisheries Institute (TAFI), Marine Research Laboratory. Sampled socks were returned to their respective longline positions.

Sampled socks were deemed unavailable for sampling on subsequent months until all fourteen socks per longline section were sampled once (ie. after 7 months). The sequence of sock selection for the first seven months was repeated for the remaining seven months of the trial. The rationale behind this sampling regime was to prevent sampling the same sock on successive months and to prevent sampling a sock more than twice during the trial period. As mussels are disturbed when removed from the water, they are subjected to a certain degree of sampling stress due to handling and exposure. As such, subsequent growth may be compromised during the period immediately following each sampling event. A strictly random selection has the possibility of choosing the same sock on two or more successive occasions. This leads to the possibility of confounding treatment effects on observed growth with handling stress. Further, sampling the same sock more than twice during the trial period would eventually lead to the progressive removal of mussels intermediate to the 1 m and 5 m test depths, leading to a loss of definition between growing depths. Therefore, this sampling regime maximised the growing time that mussels were submerged in water without disruption (seven months), as well as ensuring that mussels would actually be sampled from the top (1 m growing depth) and bottom (5 m growing depth) of each sock.

Mussels were sampled at monthly intervals from March 1995 to May 1996 (excluding May 1995). The breakdown of sample collection each month was as follows:

- Four longline sites (I, II, III and IV).
- Three within-longline sections (A, B and C).
- Two droppers selected from each longline section.
- Two samples taken from each dropper (1 m and 5 m depth).
- Total of 6 samples per depth per site (12 samples per site).

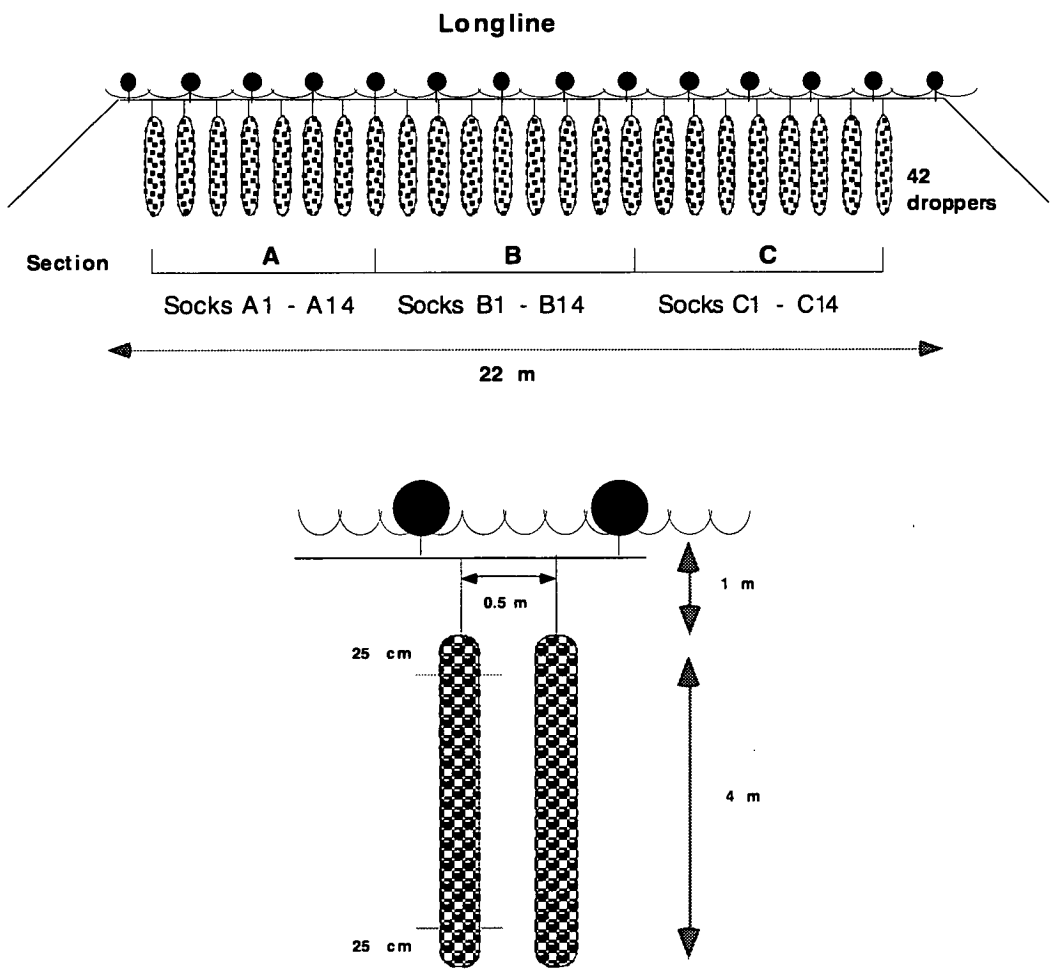


**Figure 3.1.** Each of the four experimental longline units comprised 42 mussel socks attached to a floating longline at 0.5 m intervals. The top of each sock was approximately 1 m below the water surface. Units were divided into three sections (A, B, C) of 14 socks per section. Two socks from each longline section were selected for sampling each month. Samples comprised all mussels stripped from the top and bottom 25 cm of selected socks.

## 3.4 Mussel sample preparation

Mussel samples were transported to the TAFI Marine Research Laboratory, Taroona and held in 500 l round tanks with continuous water exchange (natural seawater from the Derwent River estuary) until processed (usually within 1 to 7 days). Mussels were assessed for a number of performances indices including shell length, whole live weight, meat weight, condition, meat glycogen content, survival and reproductive development, as well as the presence of pea crabs in the mantle cavity.

Each sample was vigorously rumbled by hand in a bucket of seawater to de-clump mussels, remove debris and encourage valve closure. It was desirable that valves were closed while submerged to ensure the shell cavity was filled with water prior to exposure, providing accurate measurements of shell cavity capacity and condition index ($CI_{grav}$). A subsample of twenty mussels was selected and scrubbed clean of remaining foulants (eg. byssal threads and encrusting organisms such as algae, barnacles, tubeworms, ascidians and mussel overspat). The byssus of each mussel was removed with scissors by cutting the threads at the point where they emerged from the mantle cavity. The shell surfaces were blotted dry and the required measurements of whole live wet weight and shell linear dimensions immediately recorded before any water loss due to shell gaping occurred. Mussels were subsequently opened, presence of pea crabs noted, and meats removed and weighed. Shell and meats were dried to determine a number of different condition indices. Survival was assessed by the number of live mussels relative to the total number of mortalities + survivors in each sample.

## 3.5 Statistical Analyses

Mussel biological performance indices were assessed with analysis of variance (ANOVA) at four key months (March, August 1995, February, May 1996) with SITE and growing DEPTH as fixed factors ($\alpha = 0.05$). Prior to analysis, within-treatment distributions were checked for normality (Shapiro-Wilk test) and homogeneity of variances among treatment levels assessed (Cochran's test). Non-normal distributions were infrequently encountered and not considered to be of concern. ANOVA is sufficiently robust to non-normality, most types of non-normality likely to be encountered in practice should not seriously affect the outcome of the analysis, and therefore the assumption of normality may be relaxed under most circumstances (Underwood, 1981). Heterogeneous variances were indicated for length and weight data in February and May 1996. Transformations of the data (eg. square root, cube root, logarithmic, inverse) failed to stabilise these heterogeneous variances and

ANOVA proceeded with the original data. Non-significant outcomes of the analyses remain as reliable results. Although ANOVA is robust to many types and magnitudes of departures from homogeneity of variances (Underwood, 1981), significant differences at $P < 0.05$ for length and weight data on these two months were viewed with caution as the probability of a Type I error may be greater than 0.05. Significant treatment effects for all months were explored by comparing treatment means with Fisher's LSD.

## 3.6 Environmental monitoring

Monitoring of environmental parameters was conducted approximately weekly from May 30 1995 to May 21 1996 at each of the four mussel sites within North West Bay (I, II, III and IV) plus an additional site at the centre of the Aquatas fish farm (site X). These parameters included seawater temperature, salinity, dissolved oxygen, turbidity, suspended particulate matter, chlorophyll-a, and dissolved nutrients (nitrate, nitrite, phosphate and silicate). Zooplankton composition and abundance in the areas surrounding sites I, III and IV was monitored fortnightly over the same period. Current speed and direction was assessed from September to November 1995. Specific sampling procedures for each parameter are provided in the relevant sections.

The results from this investigation are divided into a number of sections. Environmental conditions and water quality parameters at each experimental site are presented in Section 4. Mussel growth, condition, reproductive development and survival are presented in subsequent sections (Sections 5 to 8). Additional data collected during the trial included the incidence of pea crab infestation (Section 9) and the growth of overspat that settled on the experimental mussels (Section 10). An attempt to distinguish fish farm-grown mussels from those grown distant to the farm using stable isotope analyses is described in Section 11. These experimental results are followed by analyses of theoretical scope for growth or mussel growth that might be expected under the prevailing environmental conditions (Section 12), and theoretical waste loadings from the Aquatas fish farm during the mussel growth trial (Section 13).

# 4. Environmental Parameters

## 4.1 Introduction

A comprehensive water quality monitoring program was conducted concurrently with the study of mussel growth in North West Bay. The purpose of the monitoring was to establish how environmental conditions vary seasonally, differ among the four mussel-growing sites, affect mussel growth, and were influenced by the Aquatas fish farm. Environmental and water quality parameters monitored included temperature, salinity, dissolved oxygen, turbidity, current speed and direction, suspended particulate matter, chlorophyll-a content, concentrations of selected dissolved nutrients, and zooplankton composition.

## 4.2 Methodology

### 4.2.1 In situ physico-chemical parameters: temperature, dissolved oxygen, salinity, transparency and current

Monitoring of all environmental parameters was conducted approximately weekly from May 30 1995 to May 21 1996 at each of the four mussel sites within North West Bay (I, II, III and IV) plus an additional site at the centre of the Aquatas fish farm (site X). At each site, single measurements of temperature, salinity and dissolved oxygen were taken *in situ* at two depths, 1 m and 5 m, corresponding to the top and bottom levels of the socked mussels. Measurements were taken at approximately 0800-0900 h. Temperature (± 0.05 °C) and dissolved oxygen concentration (± 0.05 $mg \cdot l^{-1}$) were measured using an electronic temperature/oxygen meter (OxyGuard Handy Mk III). Salinity was measured with a handheld refractometer (Atago SC-28; ± 0.5‰) or an electronic salinity meter (WTW LF320; ± 0.05‰). Transparency of the water column was measured with a standard black and white secchi disk lowered into the water with the depth (m) recorded at the point where the disk was no longer visible.

Current speed and direction were assessed from September to November 1995. One current meter (Aanderaa RCM 4S with a lower threshold of detectable current speed of 2.5 ± 1.0 $cm \cdot sec^{-1}$) was deployed at site I on Sept 18 and left undisturbed for six weeks. Another identical current meter was deployed at site IV on Sept 18 for two weeks, moved to site III for two weeks and finally to site II for another two weeks. Meters were positioned 3 m below the surface, with current speed (± 0.05 $cm \cdot sec^{-1}$) and direction (magnetic ± 0.5°) recorded on magnetic tape at 10 min intervals.

### 4.2.2 Water sampling procedures

Concurrent with *in situ* water quality measurements, the water column at each of the five sites was also sampled one day per week, commencing on May 30 1995. On sampling days, two replicate water samples were collected from each site. The first set of samples was collected at 0800-0900 h and the second set approximately 2-4 hours later. As sea bottom depths were approximately 20 m, samples of only the top 18 m were collected to avoid inclusion of sedimented material. An 18 m Nylex™ hose (internal diameter 12.5 mm) was lowered through the water column, a valve closed at the top-end, and the bottom-end retrieved with an attached line. The retained water samples (2 litres) were transferred to plastic containers, stored in an insulated cooling box and transported to the TAFI Marine Research Laboratory for analyses. The direction of current flow when water samples were collected was not monitored. All water samples were pre-screened through a 200 $\mu$m nylon mesh, removing large zooplankton and debris prior to subsequent analyses of particulate matter, chlorophyll-*a* and dissolved nutrients.

### 4.2.3 Chlorophyll-*a*

Indirect estimates of phytoplankton biomass have been routinely estimated by the concentration of chlorophyll-*a* in water samples. However, it is recognised that this methodology is not always an accurate reflection of phytoplankton biomass; chlorophyll-*a* concentrations may vary according to phytoplankton species as well as their nutritional status (Strickland and Parsons, 1972).

Chlorophyll-*a* concentrations were determined by modifications of the spectrophotometric method described by Lorenzen (1967) and Standard Method 1002G (APHA, 1985). This method corrects for the presence of chlorophyll degradation products, such as phaeopigments, which absorb light near the same wavelength as chlorophyll-*a*. All procedures in the filtration and extraction of pigments were conducted in subdued lighting to prevent photodegradation of chlorophyll. One liter of prescreened water was passed through a 47 mm Whatman GFC glass fibre filter under low vacuum pressure (- 20 kPa). The filter was folded, placed in a stoppered plastic test tube and immediately frozen at -20°C until analysed, usually within four weeks. The frozen filters were cut into small strips and placed into 25 ml glass centrifuge tubes with 11 ml of 90% acetone. Pigments were extracted from the filters ultrasonically (Labline Ultratip Labsonic System Model 9100-1) by immersing an ultrasonic probe (Microtip 9108) into the centrifuge tube for 60 seconds at 40 watts. Sonication aids in the extraction process by promoting cell lysis, releasing cell bound pigments into solution. After extraction, the tubes were centrifuged for 20

min at 10,000 rpm (Sorvall SS-4) and the supernatant pipetted into 15 ml plastic test tubes. Pigment extracts were then poured into a 4 cm path-length spectrophotometer cuvette and absorbances at 663 nm and 750 nm measured against a reference cell containing 11 ml of 90% acetone (Pye Unicam SP8-100 spectrophotometer). Extracts were then acidified using 1 drop of concentrated hydrochloric acid and absorbances at 663 nm and 750 nm re-measured against a reference cell with 11 ml of 90% acetone and 1 drop of concentrated HCl. Calculations of chlorophyll-*a* content were based on the equation given by APHA (1985):

$$\text{Chlorophyll a (ug l}^{-1}) = \frac{26.7\,(663_o - 663_a) \times V_1}{V_2 \times L}$$

where:

| | | |
|---|---|---|
| $V_1$ | = | volume of extract (ml) |
| $V_2$ | = | volume of water filtered (l) |
| L | = | light path of cuvette (cm), and |
| $663_o$ and $663_a$ | = | absorbance before and after acidification, respectively (after correction by subtracting absorbance at 750 nm) |

### 4.2.4 Suspended Particulate Matter

Measurements of suspended particulate material concentrations were based on the method described by Strickland and Parsons (1972). From each 2 l prefiltered sample, 800 ml was filtered through pre-ashed (480°C for 5 h in muffle furnace) and weighed (± 0.0.005 mg, Mettler AT261 DeltaRange balance) glass fibre filters (47 mm Whatman GFC) under low vacuum pressure (- 20 kPa). Salts were flushed from the filters with 7 ml of 0.9% ammonium formate solution. Filters were oven dried for 12 h at 80°C, allowed to cool in a desiccator and then weighed to determine total particulate matter (TPM or seston, $mg \cdot l^{-1}$). The filters were then ashed in a muffle furnace at 480°C for 5 h, cooled and re-weighed to determine, by difference, the combusted fraction or particulate organic material content (POM, $mg \cdot l^{-1}$). The difference between POM and TPM is the particulate inorganic matter (PIM, $mg \cdot l^{-1}$). Percentage of particulate organic matter was calculated as:

$$\% \text{ POM} = (\text{POM} \times \text{TPM}^{-1}) \times 100$$

Non-algal particulate organic matter (NAPOM), the fraction of POM not associated with live phytoplankton and including suspended fish farm wastes, was calculated by subtracting algal organic matter from total POM. Algal organic matter was estimated by multiplying chlorophyll-*a* concentrations by 67, which assumes that chlorophyll-*a* constitutes, on average, 1.5% of algal ash-free dry weight (APHA, 1985).

### 4.2.5 Nutrients

Water samples were analysed for concentrations of several common dissolved nutrients utilised by phytoplankton: nitrate, nitrite, total nitrogen (nitrate + nitrite), phosphate, and silicate. Unfortunately, lacking a reliable methodology, analysis of ammonia concentrations was not performed. All analyses were conducted using a Skalar segmented-flow autoanalyser following modifications of standard spectrophotometric methods (SAN, 1993).

Total nitrogen concentrations ($NO_x$, $\mu g \cdot l^{-1} = NO_3\text{-}N + NO_2\text{-}N$) were determined following Skalar modifications of Standard Method 418 C (APHA, 1985). The sample is diluted in an ammonium chloride buffer and pumped through a cadmium column where the nitrate is reduced to nitrite. A colour reagent (NEDH + sulphanilamide) is added to form a coloured-diazo complex with the nitrite ion. The extinction is measured at 540 nm and relates to the combined concentration of nitrate and nitrite. The procedure is repeated without passing through the cadmium column, to obtain the original concentration of nitrite ($NO_2\text{-}N$, $\mu g \cdot l^{-1}$) in the sample. Nitrate concentrations ($NO_3\text{-}N$, $\mu g \cdot l^{-1}$) are determined by difference between total nitrogen and nitrite.

Phosphate (as orthophosphate) concentrations ($PO_4\text{-}P$, $\mu g \cdot l^{-1}$) were determined following Skalar modifications of Standard Method 424 G (APHA, 1985). Phosphate reacts with ammonium molybdate in an acid medium to form phosphomolybdic acid. Reduction of this by ascorbic acid gives a phosphomolybdic blue complex. The extinction is measured at 880 nm and is in relation to the concentration of the phosphate.

Silicate concentrations ($SiSO_4\text{-}Si$, $\mu g \cdot l^{-1}$) were determined following Skalar modifications of Standard Method 425 E (APHA, 1985) and Babulak and Gildenberg (1973). The sample is acidified and mixed with an ammonium molybdate solution forming molybdosilicic acid. This is reduced with ascorbic acid to a blue dye and the extinction measured at 810 nm.

All analyses were conducted with a 60 second reaction time followed by a 60 second wash time (distilled water) between samples. Absorbances of test samples along with nutrient standards were recorded on continuous graph paper and peak heights measured manually with a ruler ($\pm$ 0.5 mm). A standard curve for each nutrient was obtained by regressing absorbance heights of nutrient standards against their known concentrations. Nutrient concentrations of test samples were calculated from the appropriate standard curve.

### 4.2.6 Zooplankton

Zooplankton composition, abundance and biomass was monitored fortnightly, from June 1995 to May 1996, adjacent to mussel longlines at sites I, III and IV. Details of sampling, identification, quantification, and statistical procedures are provided in Appendix 3.

### 4.2.7 Statistical analyses

Environmental variables were analysed by ANOVA with SITE and TIME as fixed factors. As there were only two replicates per sampling time normality of data cannot be tested. Heterogeneity of variances was checked with Cochran's test. Where variances were heterogeneous ($NO_2$ data), variances were stabilised by a log (X + 1) transformation of the data. Where treatment effects were significant, Fisher's LSD was employed to investigate the source of differences. Where site differences were not significant, data for appropriate parameters were pooled by depth to investigate depth x time relationships. Relationships among environmental parameters during the trial period were assessed by calculating the correlation coefficient (r) and the probability of r = 0 (Fisher r to z transformation) among each pair of environmental parameters. All analyses used Statview" 4.5 statistical software (Abacus Concepts, 1994). Unless otherwise specified, all mean values are expressed ± SEM.

## 4.3 Results

### 4.3.1 Temperature

Water temperature displayed a typical seasonal cycle, being lower in winter and higher in summer (Fig. 4.1). Initial temperatures of 17.9°C in March 1995 declined through the autumn-winter period to a low of 8.8°C in July. Temperatures subsequently increased through spring-summer, peaking at 17.8°C in February 1996, before declining over the final three months to 12.2°C in May 1996.

No significant difference in overall trial mean temperatures among sites ($P > 0.05$, mean 13.3 ± 0.1°C, n = 500 observations) nor any site x time interaction ($P > 0.99$) were indicated. However, a minor difference between depths was significant ($P < 0.0001$), where the overall mean temperature at 1 m was 0.143°C higher than at 5 m. Most of this difference was accounted for in the summer period, particularly January and February 1996, with the greatest difference between depths being 1.7°C on

January 2 1996. The remainder of the sample periods with significant temperature differences between depths were within 1°C. No differences in winter temperatures (June - August 1995) were significant ($P > 0.05$). Therefore, temperature differences between mussel growing depths were not considered to be important.

Daily photoperiod over the course of the trial ranged from winter minima of 9 h·day$^{-1}$ in July 1995 to summer maxima of 15.4 h·day$^{-1}$ in December 1995 (Fig. 4.1).

Significant correlations were indicated between temperature and photoperiod ($r = 0.65$, $P < 0.0001$), dissolved oxygen ($r = -0.55$, $P < 0.0001$), chlorophyll-*a* ($r = 0.50$, $P < 0.001$), POM ($r = 0.35$, $P < 0.05$), total nitrogen ($r = -0.85$, $P < 0.0001$) and phosphate ($r = -0.72$, $P < 0.0001$) (Table 4.1). Photoperiod was correlated with total nitrogen ($r = -0.77$), silicate ($r = -0.48$) and phosphate ($r = -0.75$, all at $P < 0.0001$).

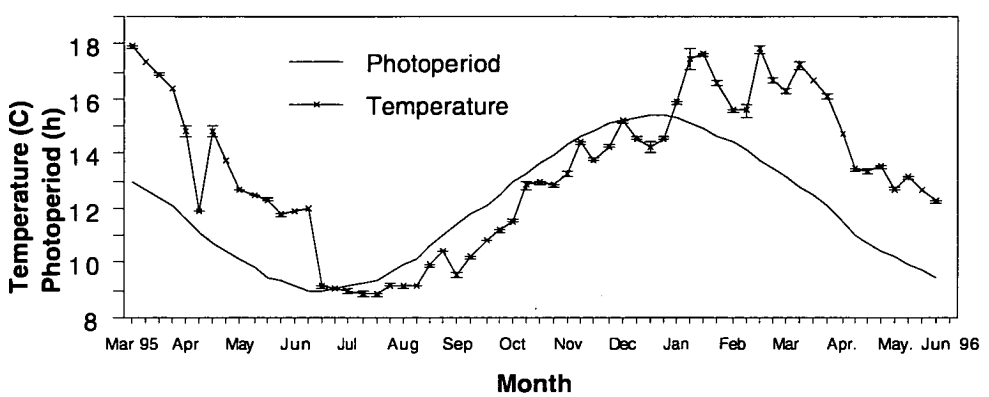


**Figure 4.1.** Cycle of seawater temperatures (mean ± SE °C) and corresponding daily photoperiod (h) from March 1995 to June 1996. Temperatures were taken at weekly intervals at two depths (1 m and 5 m) at each of five sites (n = 10 observations per week).

**Table 4.1.** Correlation coefficients (r) among environmental parameters (weekly mean values taken at 5 sites within NWB) from June 1995 to June 1996. Significant correlations ($r \neq 0$) are indicated by * ($P < 0.05$), ** ($P < 0.01$) and *** ($P < 0.0001$).

| Parameter | Temperature | Photoperiod | Salinity | DO | Secchi | Chlorophyll | TPM | POM | PIM | NOX | SiO4 | PO4 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Temperature | 1.000 | | | | | | | | | | | |
| Photoperiod | 0.654*** | 1.000 | | | | | | | | | | |
| Salinity | -0.274 | 0.047 | 1.000 | | | | | | | | | |
| DO | -0.550*** | -0.072 | 0.288 | 1.000 | | | | | | | | |
| Secchi | 0.029 | 0.315 | 0.734*** | 0.168 | 1.000 | | | | | | | |
| Chlorophyll | 0.500*** | 0.164 | -0.116 | -0.213 | -0.211 | 1.000 | | | | | | |
| TPM | 0.228 | -0.150 | -0.162 | -0.284 | -0.496** | 0.442** | 1.000 | | | | | |
| POM | 0.345* | -0.061 | -0.055 | -0.390** | -0.372* | 0.485** | 0.882*** | 1.000 | | | | |
| PIM | 0.037 | -0.206 | -0.253 | -0.090 | -0.523** | 0.274 | 0.856*** | 0.510** | 1.000 | | | |
| NOX | -0.848*** | -0.771*** | 0.235 | 0.402** | -0.149 | -0.257 | -0.064 | -0.148 | 0.045 | 1.000 | | |
| SiO4 | -0.067 | -0.478*** | -0.226 | -0.316* | -0.437** | 0.326* | 0.492*** | 0.440** | 0.414** | 0.347* | 1.000 | |
| PO4 | -0.715*** | -0.746*** | 0.139 | 0.230 | -0.186 | -0.181 | -0.010 | -0.089 | 0.080 | 0.901*** | 0.377* | 1.000 |

### 4.3.2 Dissolved Oxygen

Dissolved oxygen concentrations (DO, mg $O_2 \cdot l^{-1}$) also followed a seasonal cycle, tending to be higher during the winter period and lower during the spring-summer (Fig. 4.2). Mean DO increased from 7.7 $mg \cdot l^{-1}$ in March 1995 to 9.2 $mg \cdot l^{-1}$ during the winter period (July - August 1995). DO declined over the spring and summer to a minimum of 6.8 $mg \cdot l^{-1}$ in April 1996. Overall mean DO for the entire trial period was $8.53 \pm 0.04$ $mg \cdot l^{-1}$ ($n = 500$ observations) with no significant difference between depths ($P > 0.05$). A site x time interaction was indicated ($P < 0.05$); post hoc testing revealed lower DO concentrations at the centre of the farm for much of the period from mid-December 1995 to the end of the trial, whereby the DO on the fish farm ($7.5 \pm 0.1$ $mg \cdot l^{-1}$, $n = 36$) was slightly lower than the mussel sites (range $7.9 \pm 0.1$ $mg \cdot l^{-1}$ at site I to $8.4 \pm 0.1$ $mg \cdot l^{-1}$ at site IV, $n = 36$). Although differences in mean DO between the four mussel growing sites were significant for this period, they were minor; the greatest difference in mean DO was only 0.5 $mg \cdot l^{-1}$ between site I and site IV and not considered to be of importance.

Dissolved oxygen was significantly correlated with temperature ($r = -0.55$, $P < 0.0001$), POM ($r = -0.39$, $P < 0.01$), total nitrogen ($r = 0.40$, $P < 0.01$) and silicate concentrations ($r = -0.32$, $P < 0.05$).

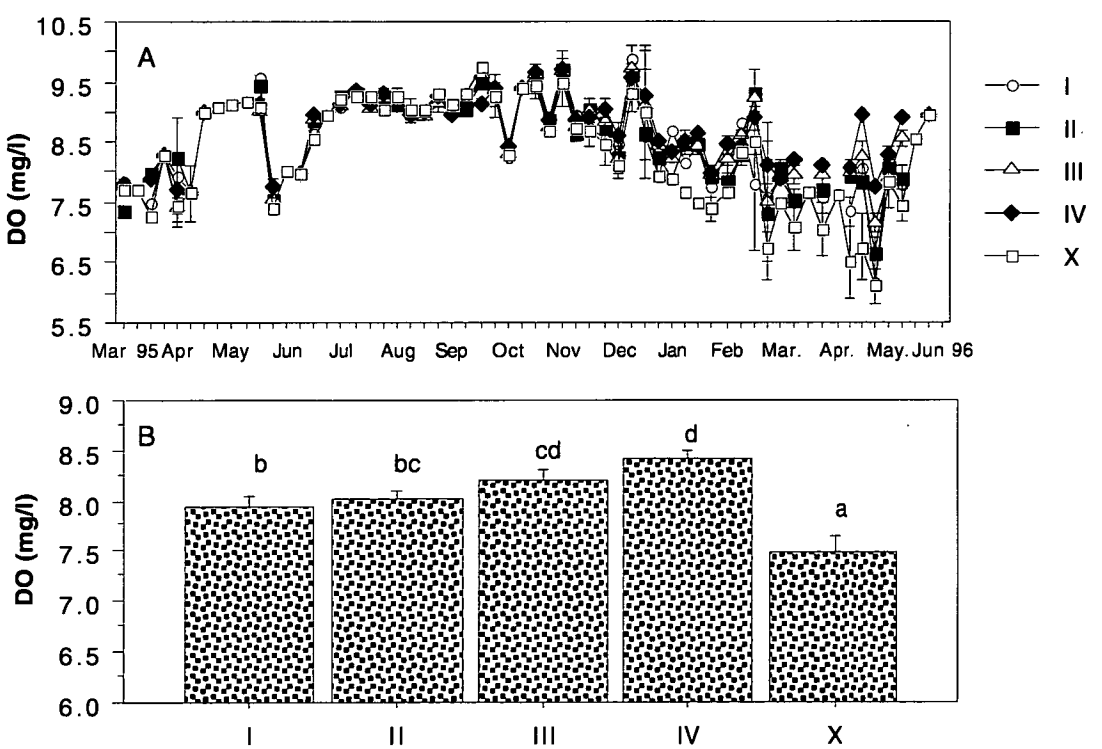


**Figure 4.2.** (A) Weekly dissolved oxygen concentrations (DO) at five sites in North West Bay (mean ± SE $mg \cdot l^{-1}$, $n = 2$ observations per site). (B) Mean DO from Mid-December 1995 to May 1996 (mean ± SE $mg \cdot l^{-1}$, $n = 36$ observations per site) was significantly different among sites ($P < 0.0001$); sites not sharing a common letter are significantly different ($P < 0.05$, Fishers LSD).

### 4.3.3 Salinity

The overall mean salinity from August 1995 to May 1996 was 34.5 ± 0.6SE‰, ranging from a low of 21‰ on December 22 1995 to a high of 40.0‰ on September 29 1995 (Fig. 4.3). No differences in overall mean salinity between sites were indicated ($P > 0.05$). A significant depth x sampling time interaction was indicated ($P < 0.0001$), whereby surface salinities (1m) were significantly lower than those at 5 m at specific times. This was most evident from December 1995 to February 1996, where stratification of the water column occurred following several major rainfall events. In particular, high runoff prior to the December 22 observations resulted in an obvious freshwater prism, where the salinity at 1 m was only 6‰ compared with 36‰ at 5 m.

There were no significant correlations between salinity and any other environmental parameters except for secchi depth ($r = 0.73$, $P < 0.0001$).

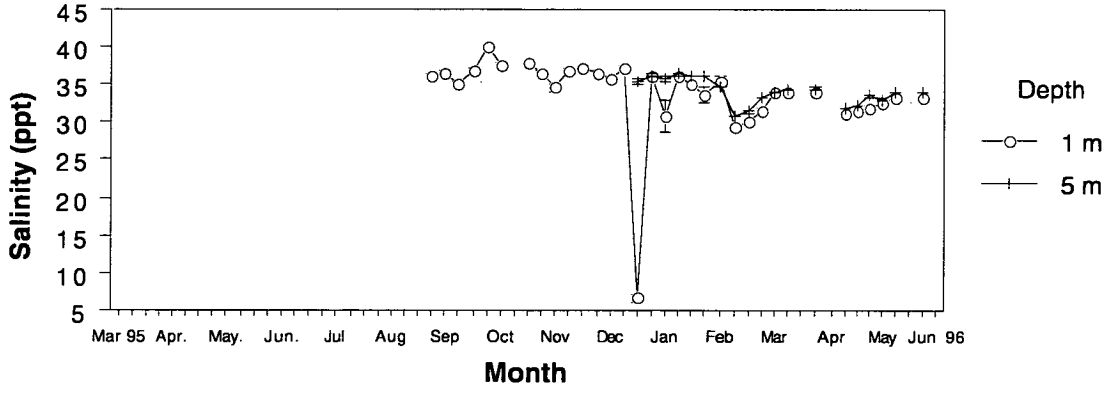


**Figure 4.3**. Salinity of North West Bay water samples taken from five sites at two depths (1 m and 5 m) from August 1995 to June 1996 (mean ± SE ‰, n = 5 observations per depth per week).

### 4.3.4 Transparency

Mean transparency (secchi depth, meters), ranged from 0.8 m to 10.1 m (Fig. 4.4). No obvious seasonal trends were apparent nor were any differences in overall mean transparency between sites significant ($P > 0.05$; overall mean 5.4 ± 0.2 m).

Secchi depth was significantly correlated with salinity ($r = 0.73$, $P < 0.0001$), TPM ($r = -0.50$, $P < 0.001$), POM ($r = 0.37$, $P < 0.05$), PIM ($r = -0.52$, $P < 0.001$) and $SiO_4$ concentrations ($r = -0.44$, $P < 0.01$).

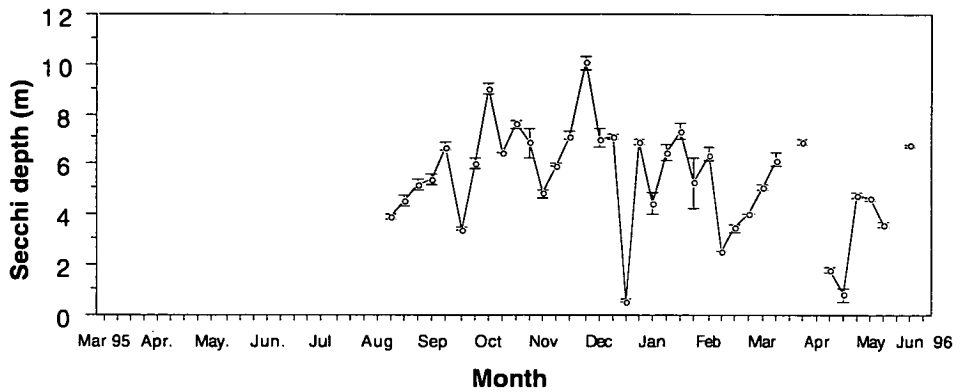


**Figure 4.4.** Transparency (Secchi depth, mean ± SE m) of North West Bay waters taken at weekly intervals from five sites, August 1995 to June 1996. n = 5 observations per week.

### 4.3.5 Current speed and direction

Current speeds and direction were measured at each of the four sites, although problems were encountered with the current meters deployed at sites I and II. Current speed data at site II was compromised due to algal fouling on the impellor and a magnetic battery in the meter at site I produced erroneous directional data. As data from these two sites were questionable, current speed and direction recorded at sites III and IV are presented (Fig. 4.5).

The modal current directions for the ebb tide, from Sept 18 to Oct 2 1995 at site IV and from Oct 2 to Oct 20 1995 at site III, ranged from 140°-160°, with an average speed of 5.2 cm·sec$^{-1}$. The modal current directions for the flood tide ranged from 310°-330°, with an average speed of 3.6 cm·sec$^{-1}$. The directional data from these two sites confirmed that the tidal currents ran parallel to the shoreline and the mussel longlines were located "inline" with the current passing through the fish farm. The maximum recorded speed was 24.7 cm·sec$^{-1}$; 78% of recordings were under 5 cm·sec$^{-1}$ with overall average current speed of 3.4 cm·sec$^{-1}$.

Although possible differences in current speed and direction among all four mussel sites cannot be tested, the data from sites III and IV closely agree with previous recordings at the Gunpowder farm site in May-June 1993 (Aquatas data) and with current speed/direction reported by Matthews and Volframs (1978). Due to the close proximity of the experimental sites, it seems unlikely that current speed and direction would have markedly differed among sites. However, the effect of the fishcages at sites I and II and variations in seabed topography differentially affecting current flow among sites cannot be discounted.

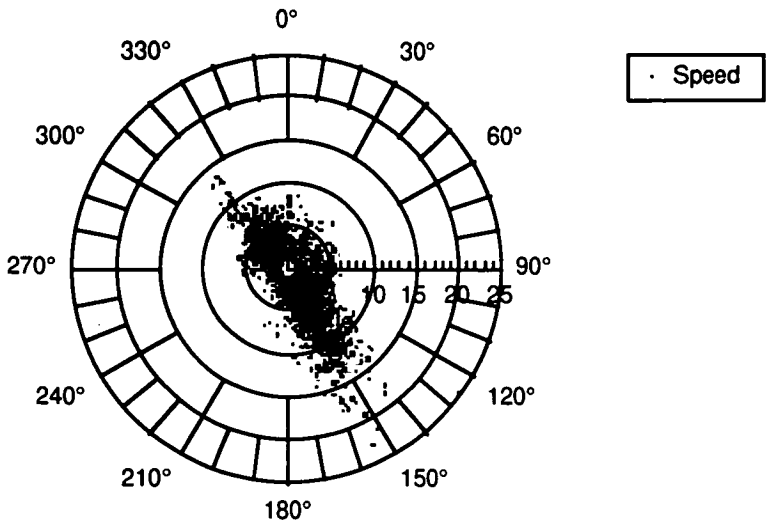


**Figure 4.5.** Summary of current speed ($cm \cdot sec^{-1}$) and direction at two sites (III and IV) in North West Bay from September 18 to October 20 1995. Each contour interval = 5.0 $cm \cdot sec^{-1}$. The maximum speed was 24.7 $cm \cdot sec^{-1}$ with 78% below 5 $cm \cdot sec^{-1}$ (n = 4687 observations).

### 4.3.6 Chlorophyll

Chlorophyll-*a* concentrations were generally low throughout most of the trial period, ranging from undetectable levels to a maximum of 5.8 $\mu g \cdot l^{-1}$, with an overall trial mean of 1.1 ± 0.1 $\mu g \cdot l^{-1}$ (n = 470 observations, Fig. 4.6). Chlorophyll content was below 0.5 $\mu g \cdot l^{-1}$ during the 1995 winter period before a short spring bloom in September, peaking at 2.9 $\mu g \cdot l^{-1}$. Chlorophyll concentrations remained below 1.5 $\mu g \cdot l^{-1}$ during spring-early summer, increased during January-February 1996 to 5.8 $\mu g \cdot l^{-1}$ and declined to 0.2 $\mu g \cdot l^{-1}$ in April. A short autumn bloom occurred in May, reaching levels of 3.0 $\mu g \cdot l^{-1}$. No significant differences in overall mean chlorophyll content between sites ($P > 0.05$) nor site x sample time interaction ($P > 0.05$) were indicated.

Significant correlations between chlorophyll concentration and temperature ($r = 0.50$, $P < 0.001$), TPM ($r = 0.44$, $P < 0.01$), POM ($r = 0.49$, $P < 0.001$) and silicate concentrations ($r = 0.33$, $P < 0.05$) were indicated.

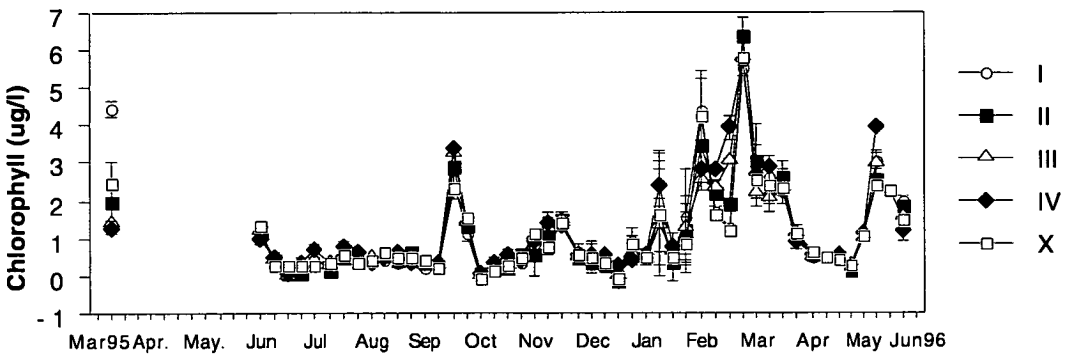


**Figure 4.6.** Chlorophyll-a concentrations at five sites in North West Bay (mean ± SE $\mu g \cdot l^{-1}$, n = 2 observations per site per sampling period).

### 4.3.7 Suspended Particulate Matter

Variations in suspended particulate matter concentrations (mg SPM·$l^{-1}$), comprising total particulate matter (TPM) or seston, particulate organic matter (POM) and particulate inorganic matter (PIM), are presented in Fig. 4.7; overall trial mean concentrations from each of the five sampling locations within North West Bay are summarised in Fig. 4.8. Particulate content did not change dramatically throughout the course of the trial. However, a period of approximately six weeks, from mid-November to the end of December 1995 displayed a slight depression in particulate levels as compared to the rest of the trial period.

Mean weekly TPM levels ranged from 3.7 to 9.7 mg·$l^{-1}$ with an overall mean of 6.6 ± 0.1 mg·$l^{-1}$ (n = 460 observations). A significant difference in overall trial mean TPM among sites was indicated ($P < 0.0001$), where mean TPM at site X (7.4 ± 0.2 mg·$l^{-1}$, n = 94 observations) was greater than at the four mussel growing sites ($P < 0.05$; mean 6.4 ± 0.1 mg·$l^{-1}$, n = 376 observations). No significant site x time interaction was indicated ($P > 0.05$).

PIM ranged from 1.8 to 5.9 mg·$l^{-1}$ with an overall mean of 4.29 ± 0.05 mg·$l^{-1}$ (n = 460). A significant difference in overall trial mean PIM between sites was indicated ($P < 0.0001$), where mean PIM at site X (4.8 ± 0.2 mg·$l^{-1}$, n = 94) was greater than at the four mussel growing sites ($P < 0.05$; mean 4.1 ± 0.1 mg·$l^{-1}$, n = 376). A site x time interaction was not indicated ($P = 0.99$). PIM displayed significant correlations with POM ($r = 0.51$, $P < 0.01$) and silicate concentrations ($r = 0.41$, $P < 0.01$).

POM was relatively constant throughout the trial period, ranging from a low of 0.82 mg·$l^{-1}$ to a high of 4.45 mg·$l^{-1}$ with an overall mean of 2.35 ± 0.05 mg·$l^{-1}$ (n = 460).

Mean POM from June to November 1995 was 2.2 $mg \cdot l^{-1}$ with peaks of 4.02 and 3.26 $mg \cdot l^{-1}$ in September and mid-November. POM levels fell to 1.0 $mg \cdot l^{-1}$ during November-December before increasing again in January 1996. Mean POM was 3.0 $mg \cdot l^{-1}$ from January through to June 1996. A significant difference in overall trial mean POM among sites was indicated ($P < 0.0001$), where mean POM at site X (2.6 $\pm$ 0.01 $mg \cdot l^{-1}$, $n = 94$) was greater than at the four mussel growing sites (mean 2.3 $\pm$ 0.04 $mg \cdot l^{-1}$, $n = 376$). No significant site x time interaction was indicated ($P > 0.05$). POM displayed significant correlations with chlorophyll ($r = 0.49$, $P < 0.01$), PIM ($r = 0.51$, $P < 0.01$), temperature ($r = 0.35$, $P < 0.05$), DO ($r = -0.39$, $P < 0.01$) and silicate ($r = 0.44$, $P < 0.01$).

Mean %POM ranged from 18.3 to 51.4%, displaying considerable variation from June 1995 to December 1995 (Fig. 4.7). From January 1996 to April 1996, %POM was relatively constant at approximately 40%, before decreasing in May. The overall mean for the entire trial period was 34.7 $\pm$ 0.4% with no significant site differences ($P > 0.05$) nor site x sample date interaction ($P > 0.05$).

It is of interest to note that differences in particulate concentrations among the five sampling sites were significant (TPM, POM and PIM all at $P < 0.05$) during the winter fallowing period when fish were not on the farm (July through September 1995). The mean TPM, POM and PIM concentrations at site X were significantly greater than at the other four sites ($P \leq 0.05$), but % POM was uniform across all sites (mean 35.1% $\pm$ 0.7%). This condition, higher particulate concentrations at site X but uniform %POM at all sites, prevailed after new smolts were introduced to the farm site and cultivated from late September 1995 to May 1996. However, particulate concentrations at site X displayed no relative increase over the four sites for the period when fish were on-site; mean TPM concentrations at site X were 19% greater than at the other four sites during the winter fallowing period and were 15% greater during the period when fish were on-site.

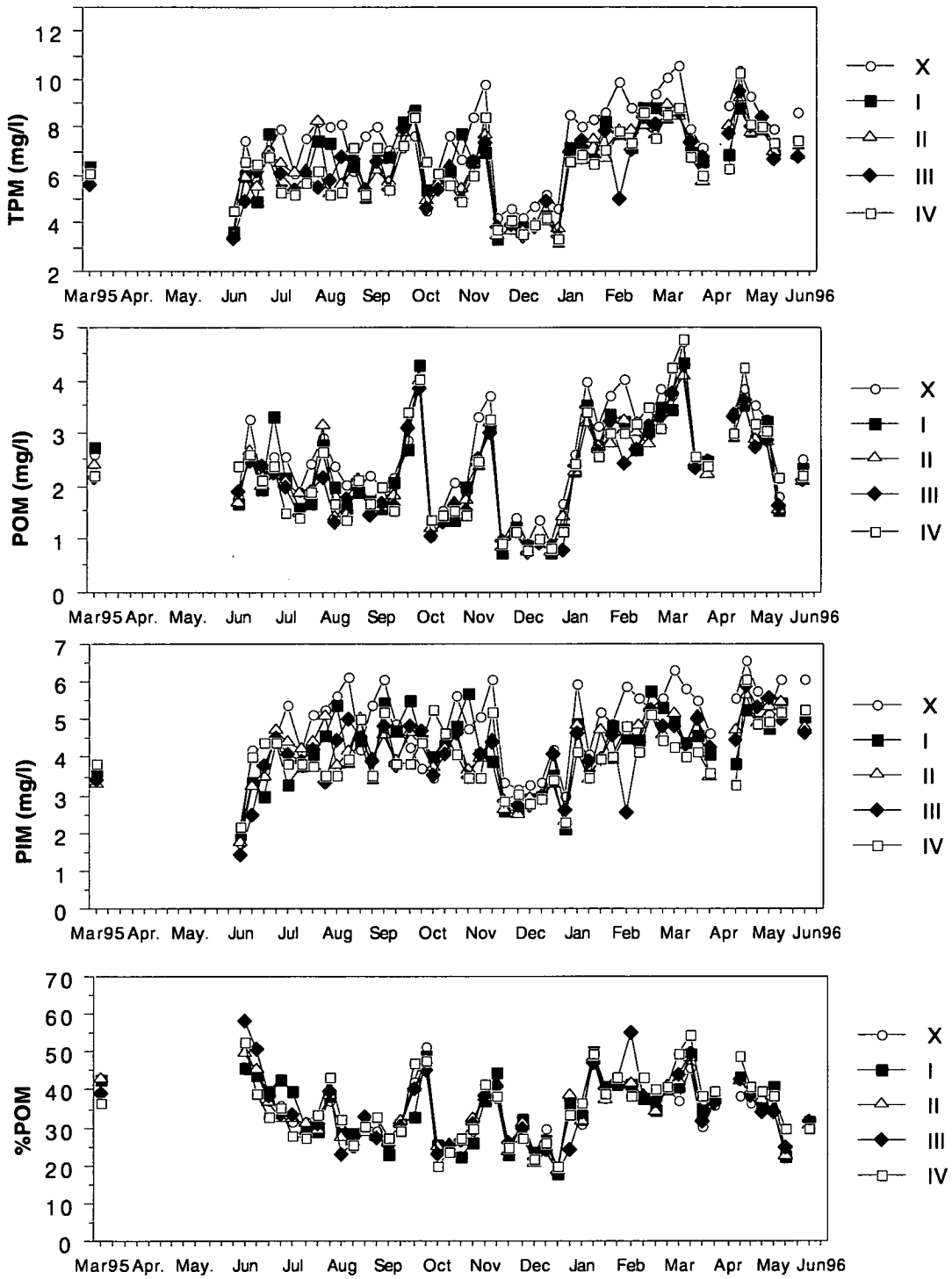


**Figure 4.7.** Suspended particulate concentrations (total particulate matter, TPM; particulate inorganic matter, PIM; particulate organic matter, POM and % POM) taken at weekly intervals from five sites (X, I, II, III, IV) in North West Bay (mean $mg \cdot l^{-1}$ or %, n = 2 observations per site per sampling period). For clarity, SE bars are not included.

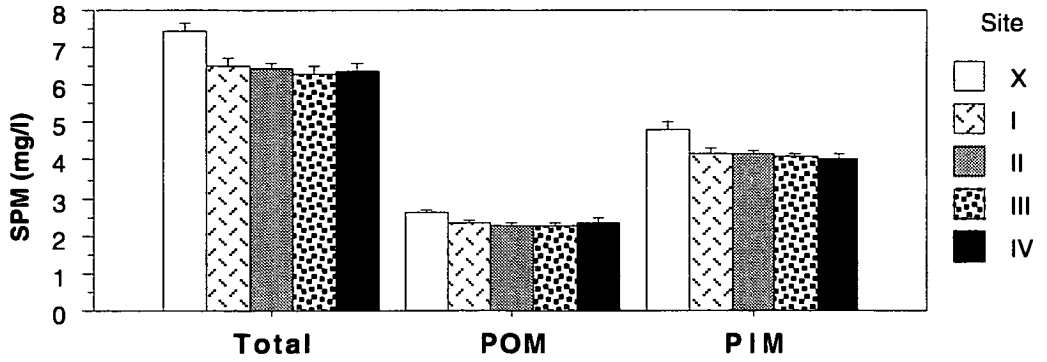


**Figure 4.8.** Overall trial mean suspended particulate matter (SPM) for each of five sites in North West Bay (± SE mg $l^{-1}$, n = 94 observations·site$^{-1}$). SPM is divided into total particulate matter, particulate organic matter (POM) and particulate inorganic matter (PIM). Significant differences between sites were indicated for all three parameters ($P < 0.0001$) where mean particulate concentrations at site X > I = II = III = IV ($\alpha = 0.05$).

Particulate organic matter was divided into algal and non-algal (ie. that not associated with phytoplankton and would be expected to include any suspended fish farm wastes) components (AP and NAP). The proportion of POM represented by non-algal biomass components (%POM-NAP) ranged from 88 to 100%, with an overall mean of 96.9% (Fig. 4.9). Significant reductions in %POM-NAP corresponded with phytoplankton blooms, as indicated by increased chlorophyll-a concentrations. No differences between sites in overall mean AP were significant ($P > 0.05$, overall mean $75.0 \pm 4.0$ μg·$l^{-1}$). Significant dfferences among sites were indicated for overall mean NAP ($P < 0.0001$), and % POM-NAP ($P < 0.01$) where mean NAP at site $X^b > I^a$, $II^a$, $III^a$ and $IV^a$ ($P < 0.05$) and mean %POM-NAP at site $X^c \geq I^{ab}$, $II^{bc}$, $III^{ab}$ and $IV^a$ ($P < 0.05$). However, the greatest difference in overall mean %POM-NAP, between sites X and IV, was less than 0.5%. No significant site x time interactions were indicated for either NAP or %POM-NAP ($P > 0.05$). As with total POM concentrations, site X displayed significantly higher NAP concentrations than the other four sites for the period June - September 1995 when no fish were on the farm ($P < 0.05$; mean NAP at site = 2.4 mg·$l^{-1}$ versus a mean of 2.1 mg·$l^{-1}$ at the other four sites). This represented a 14% higher concentration of NAP at site X over the NAP concentrations at the other four sites. After the introduction and growout of new smolts from the end of September 1995 to May 1996, the overall mean concentration of NAP during this growout period at site X, although again significantly higher than at the other four sites, did not display a relative increase compared with the NAP concentrations at the other four sites (ie. Sept-May mean at site X = 2.6 mg·$l^{-1}$ versus site I-IV overall mean of 2.3 mg·$l^{-1}$ or 13% higher NAP concentration, Fig. 4.10).

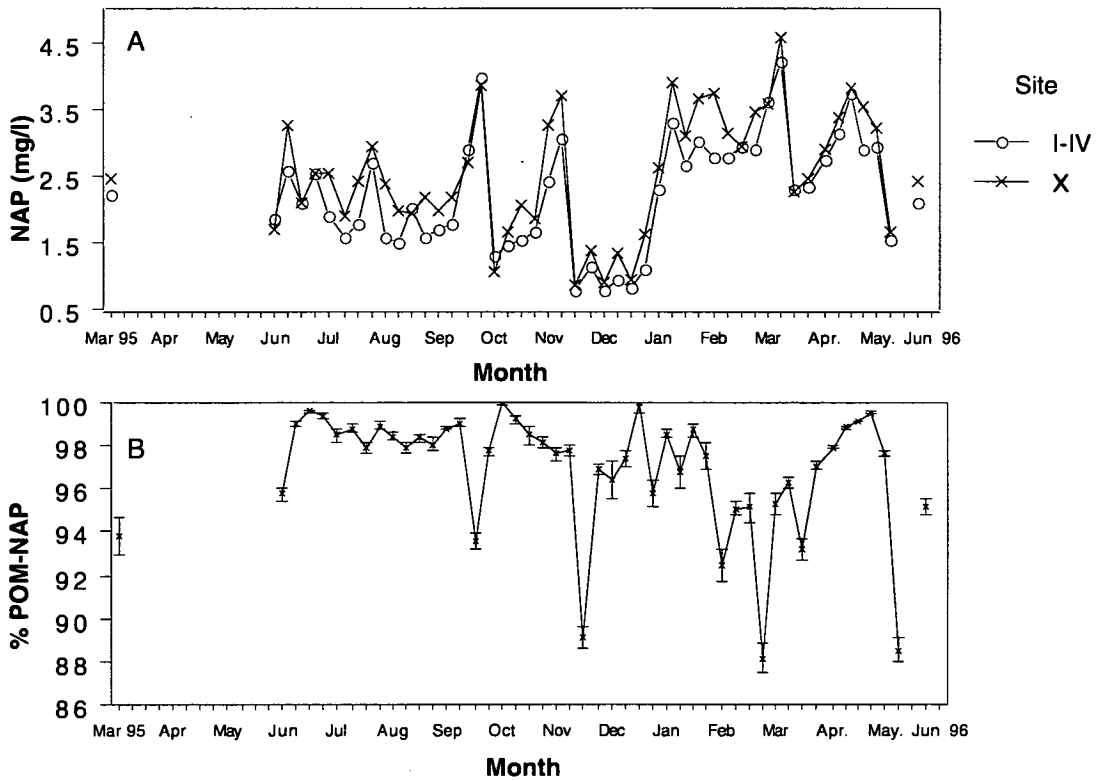


**Figure 4.9.** (A) Concentration of non-algal POM (NAP, mean ± SE $mg \cdot l^{-1}$) at site X (n = 2 observations per sampling date) and mussel longline sites (I - IV, n = 8 observations per sampling date) in North West Bay. (B) Percentage of POM represented by non-algal biomass (overall mean NAP ± SE %, n = 10 observations per sampling date).

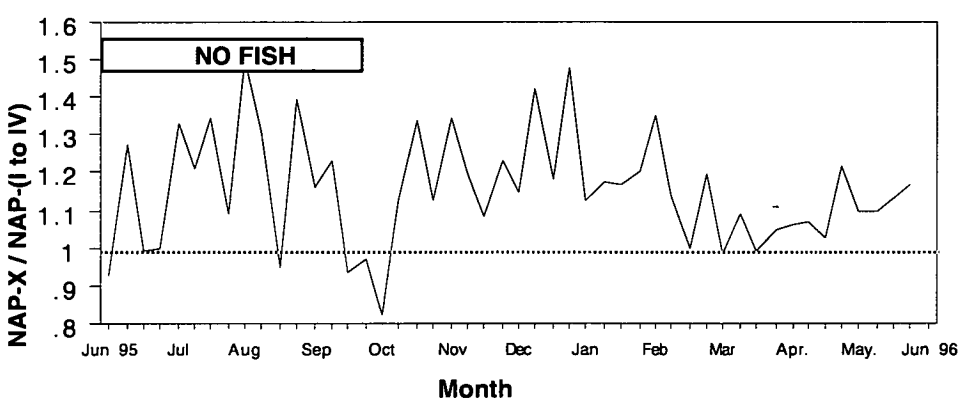


**Figure 4.10.** Concentration of non-algal POM (NAP) at the centre of the fish farm (site X) relative to the mean NAP at sites I - IV for the period June 1995 to June 1996.

### 4.3.8 Dissolved Nutrients

*Nitrogen*

Concentrations of total nitrogen, nitrate and nitrite achieved their highest levels in the winter and autumn months (June-September 1995 and April-May 1996) and their lowest levels during the spring and summer months (October 1995 to March 1996, Fig. 4.11).

Total nitrogen concentrations ($NO_X$-N, $\mu g \cdot l^{-1}$) ranged from 1.2 to 67.8 $\mu g \cdot l^{-1}$. $NO_X$ was highest during the first winter months (June to September 1995), ranging from 35 to 58 $\mu g \cdot l^{-1}$. These fell to 2 - 5 $\mu g \cdot l^{-1}$ during the summer before rising again in the autumn of 1996 to 24 $\mu g \cdot l^{-1}$. Overall trial mean $NO_X$ concentration was 20.5 ± 1.0 $\mu g \cdot l^{-1}$ (n = 460 observations). $NO_X$ displayed significant correlations with temperature ($r = -0.85$, $P < 0.0001$), photoperiod ($r = -0.77$, $P < 0.0001$), DO ($r = 0.40$, $P < 0.01$), silicate ($r = 0.35$, $P < 0.05$) and phosphate ($r = 0.90$, $P < 0.0001$).

Nitrate concentrations ($NO_3$-N, $\mu g \cdot l^{-1}$) ranged from 0.5 to 64.1 $\mu g \cdot l^{-1}$. These closely followed $NO_X$ values, as nitrate was the primary component of $NO_X$. The percentage of total nitrogen represented by nitrate ranged from 39 to 98%, averaging 83.2%. The overall trial mean nitrate concentration was 18.3 ± 1.0 $\mu g \cdot l^{-1}$ (n = 460).

Nitrite concentrations (N-$NO_2$, $\mu g \cdot l^{-1}$) were a minor component of total nitrogen, ranging from less than 1.0 $\mu g\ l^{-1}$ in the spring and summer months to 3 - 8 $\mu g \cdot l^{-1}$ during the autumn and winter months (Fig. 4.11). The overall mean nitrite concentration was 2.2 ± 0.1 $\mu g \cdot l^{-1}$ (n = 460).

No differences in overall trial mean $NO_X$, $NO_2$ or $NO_3$ concentrations among sites were significant (all at $P > 0.05$), nor were any significant site x time interactions indicated for $NO_X$ or $NO_3$ ($P > 0.05$). A significant site x time interaction was indicated for $NO_2$ concentrations ($P < 0.01$), whereby $NO_2$ at site IV was significantly less than the other sites on four occasions (June 22, September 22, September 29 and October 7, 1995).

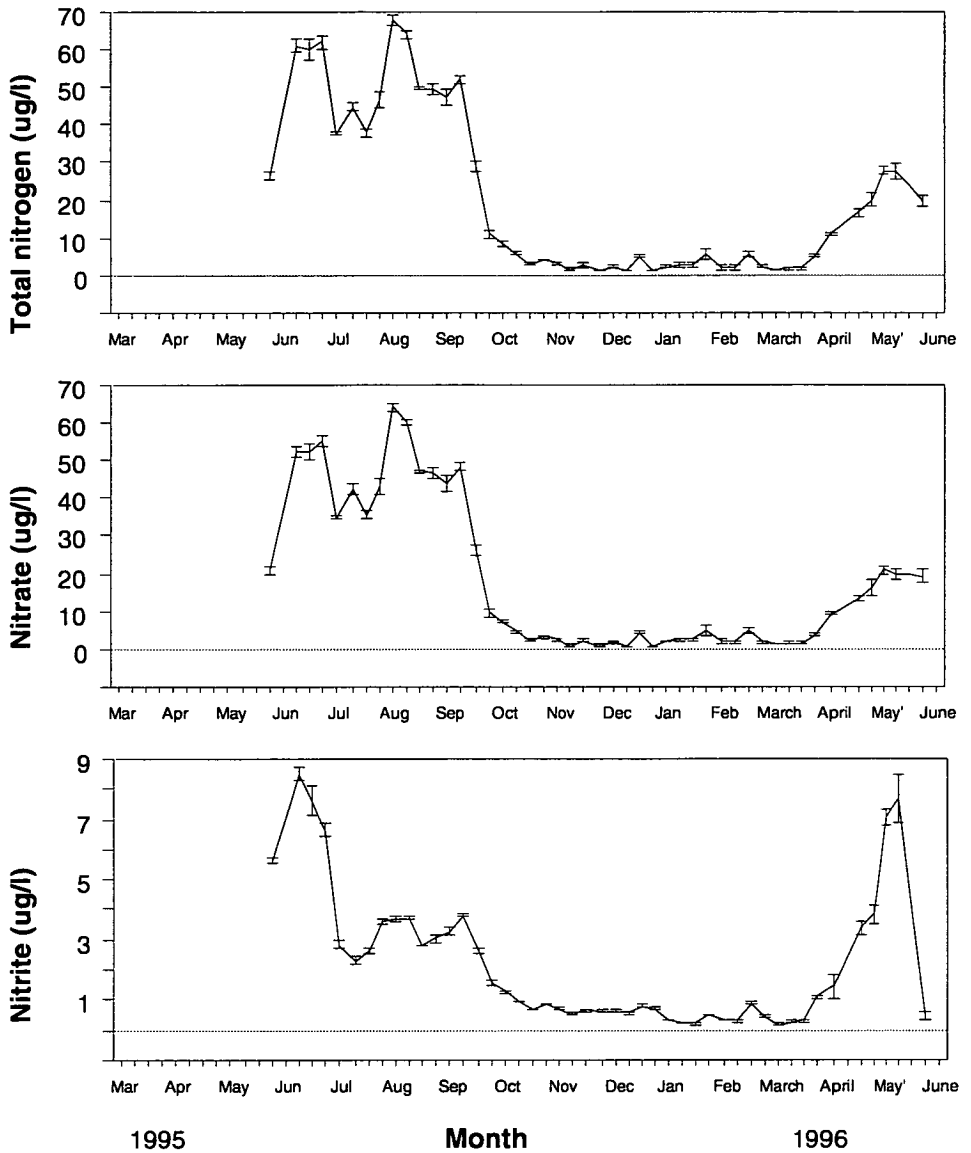


**Figure 4.11.** Total nitrogen ($NO_X$-N = $NO_3$ + $NO_2$), nitrate ($NO_3$-N) and nitrite ($NO_2$-N) concentrations (mean ± SE $\mu g \cdot l^{-1}$, n = 10 observations per nutrient per sampling date) of water samples collected at weekly intervals from five sites in North West Bay during 1995-96.

### *Phosphate*

Phosphate concentrations ($PO_4$-P, $\mu g \cdot l^{-1}$) also displayed an apparent seasonal cycle, whereby concentrations were higher in winter months and lower in summer (Fig. 4.12). Initial concentrations of approximately 18.2 $\mu g \cdot l^{-1}$ in June 1995 fell to ≈10 $\mu g \cdot l^{-1}$ during the summer months before rising to 14.4 $\mu g \cdot l^{-1}$ in May 1996. The overall trial mean was 13.1 ± 0.2 $\mu g \cdot l^{-1}$ (n = 460 observations) with no significant site differences nor site x time interactions indicated ($P > 0.05$).

Phosphate concentrations were correlated with temperature ($r = -0.72$, $P < 0.0001$), photoperiod ($r = -0.75$, $P < 0.0001$), $NO_X$ ($r = 0.90$, $P < 0.0001$) and $SiO_4$ ($r = 0.38$, $P < 0.05$).

Molar N:P ratios were derived from measured $NO_x$ and $PO_4$ concentrations. N:P ratios ranged from 6 - 8 during the winter (June - August 1995), falling to less than 1 during the spring-summer period and increasing to 4 by the end of the trial in May 1996 (Fig. 4.13). No significant differences among sites ($P > 0.05$) nor any site x time interaction ($P > 0.05$) were indicated for arcsine square root transformed ratios.

### *Silicate*

Silicate concentrations ($SiO_4$-Si, $\mu g \cdot l^{-1}$) also displayed a somewhat seasonal cycle (Fig. 4.12). Higher concentrations in the winter months from May to mid-September 1995 (range 96 to 152 $\mu g \cdot l^{-1}$) fell to 16 to 50 $\mu g \cdot l^{-1}$ during the spring. Summertime concentrations were punctuated by three distinct peaks in December 1995, February and April 1996 (135, 209 and 212 $\mu g \cdot l^{-1}$, respectively). A significant difference between sites in overall trial mean silicate concentration was indicated ($P < 0.0001$), whereby a trend of increasing overall mean silicate concentrations with increasing proximity to the fish cages was observed. However, a significant site x time interaction was also indicated ($P < 0.0001$). Post hoc examination of data from sites X and IV revealed mean Si concentrations at site X exceeded site IV by the critical difference (27.9 $\mu g \cdot l^{-1}$) on 10 of the 46 sampling dates; seven of the ten dates were during the final three months of the trial.

Silicate concentrations were correlated with photoperiod ($r = -0.48$, $P < 0.0001$), TPM ($r = 0.48$, $P < 0.0001$), transparency ($r = -0.44$, $P < 0.01$), POM ($r = 0.44$, $P < 0.01$), PIM ($r = 0.41$, $P < 0.01$), DO ($r = -0.32$, $P < 0.05$), chlorophyll ($r = 0.33$, $P < 0.05$), $NO_X$ ($r = 0.35$, $P < 0.05$) and $PO_4$ ($r = 0.38$, $P < 0.05$).

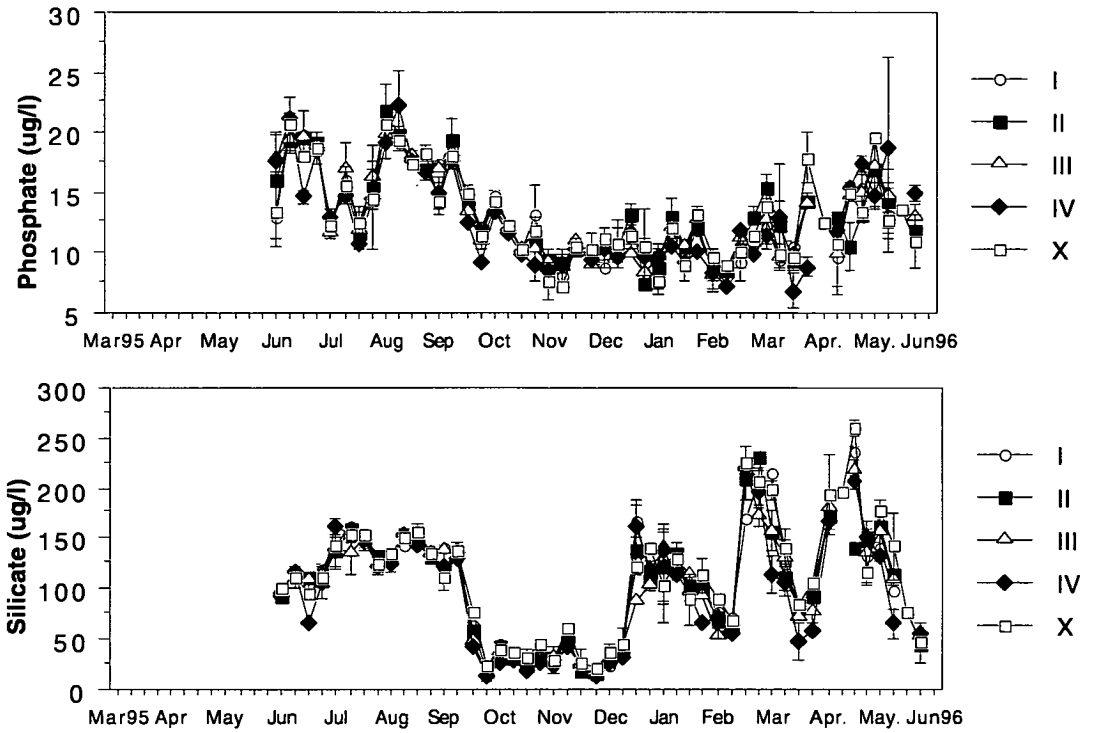


**Figure 4.12.** Mean phosphate ($PO_4$-P) and silicate ($SiO_4$-Si) concentrations taken at weekly intervals from five sites in North West Bay (mean ± SE $\mu g \cdot l^{-1}$, n = 2 observations per site per sampling date).

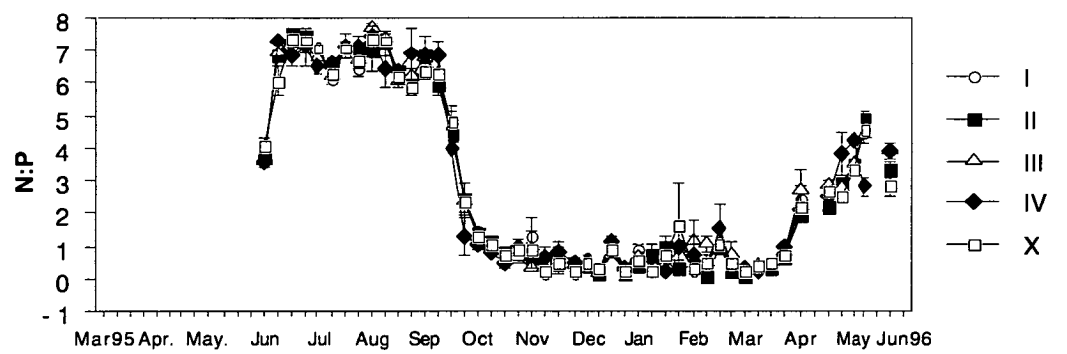


**Figure 4.13.** Weekly molar nitrogen:phosphorus ratios (N:P, mean ± SE, n = 2 observations per site per sampling period) of samples taken from five sites in North West Bay.

### *Correlations between dissolved nutrients and phytoplankton growth*

The seasonal cycle of dissolved nutrient levels was reflected by significant correlations with temperature and photoperiod (Table 4.1). However, only silicate displayed significant correlations with indicators of phytoplankton biomass (eg. chlorophyll and POM levels), although $NO_x$ and chlorophyll were close to being significant at P = 0.09. As a lag effect might be expected between fluctuations in nutrient concentrations and subsequent changes in planktonic biomass, correlations between biotic parameters

(eg. chlorophyll and POM) at each sampling period and nutrient concentrations from each of the previous four weeks were performed. Although a lag period of 3 - 4 weeks slightly improved the correlation between $NO_X$ and chlorophyll, no other significant improvements were evident (Table 4.2).

**Table 4.2.** Correlation coefficients (r) relating nutrient concentrations (total nitrogen $NO_X$-N = $NO_3$ + $NO_2$-N; silicate, $SiO_4$-Si; phosphate, $PO_4$-P, all $\mu g \cdot l^{-1}$) with chlorophyll-a ($\mu g \cdot l^{-1}$) and POM ($mg \cdot l^{-1}$) levels at time lags of 0 to 4 weeks. Significant correlations are indicated by * ($P < 0.05$) and ** ($P < 0.01$). n = 45 observations at 0 weeks.

| Time lag (weeks) | 0 | 1 | 2 | 3 | 4 |
|---|---|---|---|---|---|
| **Chlorophyll vs** | | | | | |
| NOX | -0.257 | -0.292 | -0.300 | -0.320* | -0.314* |
| $SiO_4$ | 0.326* | 0.171 | 0.234 | 0.159 | 0.019 |
| $PO_4$ | -0.181 | -0.290 | -0.277 | -0.286 | -0.271 |
| **POM vs** | | | | | |
| NOX | -0.148 | -0.163 | -0.236 | -0.244 | -0.225 |
| $SiO_4$ | 0.440** | 0.484** | 0.328 | 0.171 | 0.241 |
| $PO_4$ | -0.089 | -0.084 | -0.236 | -0.150 | -0.136 |

### 4.3.9 Zooplankton

Details of zooplankton composition, abundance and biomass are provided in Appendix 3. Although temporal variation in zooplankton community structure was significant (MANOVA, $P < 0.001$), no significant differences among sites nor a month x site interaction were indicated ($P > 0.05$). Similarly, temporal variation in each of the four most dominant species (Calanoid copepods, *Evadne nordmanni*, *Oikopleura dioica* and *Obelia* sp.) and biomass was also significant (all at $P < 0.0001$); however, no significant spatial variation among the three sampling sites nor site x time interaction was indicated (all at $P > 0.05$).

## 4.4 Discussion

All physical and chemical and biological parameters monitored during the course of the trial were similar to the seasonal ranges reported in a previous study of North West Bay (Matthews and Volframs, 1978) and studies of adjacent waters in southern Tasmanian, including the lower Derwent River estuary (DELM, 1995), Storm Bay (Clementson et al., 1989; Harris et al., 1991), Bruny Island (Crawford et al., 1996) and the Huon River estuary (Woodward et al., 1992).

### 4.4.1 Temperature-salinity

An obvious seasonal pattern of low winter and high summer temperatures was observed over the study period whereby temperature and photoperiod were strongly correlated. The range of temperatures (9°-18°C) was similar to those reported from an earlier study of North West Bay (Matthews and Volframs, 1978), from the mouth of the nearby Derwent estuary (DELM, 1995) and from Storm Bay (Clementson et al, 1989).

Salinity was fairly stable, maintaining levels characteristic of nearby marine waters for most of the study period (eg. annual salinity range in Storm Bay from 34 to 35.5‰, Clementson et al., 1989; overall mean salinity for the period August 1995 - June 1996, 34.5 ± 0.6SE‰). An anomalously high recording of 40‰ in September 1995 may have been erroneous. On a few occasions, marked declines in salinity, particularly at the surface in December 1995 and January 1996, were most probably the result of freshwater run-off from the catchment area following major rainfall events. On several of these occasions, a noticeable tongue of discoloured (brownish) freshwater was observed extending from the North West Bay River, at the northern limit of the bay, towards the mouth of the bay in the south. These periodic large-scale incursions of discoloured freshwater were previously noted by Matthews and Volframs (1978). North West Bay is also influenced by incursions of estuarine water, sourced from the Derwent and Huon Rivers, entering the bay via the D'Entrecasteaux Channel (Matthews and Volframs, 1978). Inputs of low salinity water from these sources, in addition to catchment runoff, may have contributed to the drop in salinity from 35 to 30‰, at both 1 m and 5 m, during February.

Matthews and Volframs (1978) summarised the general hydrographic characteristics of North West Bay. There is some slight vertical stratification present and at times the bay is very well mixed. Small but significant changes in the salinity-temperature structure can occur in less than a week with intense vertical stratification following

periods of heavy rainfall. Without sustained rainfall such stratification decays fairly rapidly. Also, there is no evidence of any longitudinal temperature-salinity gradient, suggesting that fairly rapid water exchange occurs (estimated flushing time of 7 days).

The results from this study support the above summary. In terms of overall mean salinity and temperature at the mussel growing sites, no significant differences were observed. Differences in salinity between 1 m and 5 m were significant on a few specific occasions where salinity at 1 m was less than at 5 m, but these were short-lived. These short-term declines in surface salinities were only slight (ie. less than 2‰, except for December 21 1995 when surface salinity dropped from 37 to 7‰) and unlikely to have significantly affected mussel performance (Shumway, 1977; Almada-Villela, 1984; Gruffydd et al., 1984). Temperature differences between growing depths in summer were also minor, with the mean temperature at 1 m being only 0.6°C higher than at 5 m (January to mid-March 1996). Again, it is highly unlikely that this small difference in water temperature would have any effect on differential mussel growth between the two depths.

### 4.4.2 Dissolved oxygen

Dissolved oxygen (DO) levels in estuarine waters are dependent on a number of factors, including temperature, salinity, pressure, biological activity, turbulence and mixing, and may fluctuate widely over a period of hours, weeks or months (Kennish, 1990). Oxygen dissolves in water more readily at low temperatures (Kennish, 1990), which was observed in this study with higher DO in winter (mean 9.2 $mg \cdot l^{-1}$, from July to September 1995) and lower DO in summer (minimum of 6.8 $mg \cdot l^{-1}$ in April 1996). Aquatic plants are net producers of oxygen during daylight hours but are net consumers at night. Therefore, DO levels also vary diurnally, with the lowest concentrations occurring around sunrise (DELM, 1995). DO monitoring in this study was not carried out over the full diurnal cycle; DO was measured only one day per week at approximately 0800 - 0900 hours. Therefore, the reported DO concentrations might have been near to the minimum concentrations on each sampling date. The annual range of observed DO in North West Bay was similar to reported levels in the adjacent waters of the lower Derwent estuary and Storm Bay (Clementson et al., 1989; DELM, 1995). The overall mean DO concentration for the entire trial period (8.5 $mg \cdot l^{-1}$) was slightly higher than the mean DO (7.35 $mg \cdot l^{-1}$) reported near Tinderbox at the entrance to the D'Entrecasteux Channel the previous year (DELM, 1995).

No difference in overall mean DO between the two sample depths (1 m and 5 m depths) was observed. However, differences among sites were significant, particularly during the latter period of the trial (mid-December 1995 to May 1996), whereby a gradient of increasing DO from the centre of the fish farm (site X) to the mouth of the bay (site IV) was observed. The DO levels at site IV were similar to those reported by Clementson et al. (1989) in Storm Bay over the same months (January - May) in previous years (1985 -1988). The lower DO levels in the vicinity of the fish farm may have been due to oxygen consumption by the farmed fish as well as the biochemical oxygen demand associated with decomposing organic fish farm wastes. Reduced DO levels associated with fish farms, particularly with near-bottom water, have been well documented (Rosenthal, 1983 cited in GESAMP, 1991; Brown et al., 1987; Gowen et al., 1988; citations in Levings, 1994).

The differences in DO levels among sites, although statistically significant, were not huge. Site mean DO concentrations from January to May 1996 were within 1 $mg \cdot l^{-1}$ (X = 7.5, I = 7.9, II = 8.0, III = 8.2 and IV = 8.4 $mg \cdot l^{-1}$), and even less among mussel sites (0.5 $mg \cdot l^{-1}$ difference between sites I and IV). The biological significance of such a small difference is probably nil; it is unlikely that differences in DO would have any significant impact on differential mussel growth among sites.

DO levels in healthy estuaries generally lie between 6.5 and 9.0 $mg \cdot l^{-1}$ (DELM, 1995). ANZECC (1992) guidelines suggest that DO levels in coastal marine waters should not fall below 6 $mg \cdot l^{-1}$, which also correspond to minimum DO levels (5 to 6 $mg \cdot l^{-1}$) appropriate for salmonid fish culture (Tarazona and Munoz, 1995). With the lowest recorded DO being 6.1 $mg \cdot l^{-1}$ (site X on April 21, 1996 compared with 7.75 $mg \cdot l^{-1}$ at site IV), DO levels never dropped below ANZECC (1992) guidelines, nor were levels achieved that would be considered stressful for fish (< 5 - 6 $mg \cdot l^{-1}$). Therefore, it appears unlikely that the fish farm had any deliterious effects on DO levels within the bay as a whole. These results are in agreement with GESAMP (1991), suggesting: "large-scale DO reductions in coastal waters by fish farms is unlikely, and though small short-term reductions within a fish farm are important to the farmer, it is probably not ecologically significant with the exception of low-energy coastal environments such as deep fjords and inlets." It has also been suggested that any significant effects of fish farming on DO is probably restricted to reduced DO levels in the underlying sediments and near-bottom water in the vicinity of a fish farm (Wildish et al., 1990; Gowen et al, 1991).

### 4.4.3 Transparency

Turbidity can play an important role in determining the eutrophic state of a body of water. The depth at which surface irradiance is reduced to 1% is considered to be the base of the euphotic zone. Beneath this there is insufficient light for photosynthesis and phytoplankton growth is light limited (Gowen, 1984 cited in Woodward, 1989). Any increase in turbidity due to humics, suspended solids or even due to an algal bloom itself will act to reduce the availability of light and so reduce algal production.

Gowen et al. (1988) reported that the euphotic zone was shallower at stations next to a fish farm than at a control station. Greater attenuation of light was attributed to the shading effect by the fishcages combined with more particulate material (ie. fish feed and faeces) in the water. On the other hand, Okumus (1993), comparing transparency between salmon farms and control sites, reported variations in secchi depth were more related to season, primarily due to variation in solar radiation and secondarily to PIM concentrations, rather than organic particulate wastes generated from fish farms.

In contrast to the results of Okumus (1993), suggesting secchi depth was primarily related to seasonal variation in solar radiation, no strong correlation with photoperiod was observed in the present study ($r = 0.315$), albeit close to being significant ($P = 0.06$); light intensity would be expected to be correlated with photoperiod. Transparency was significantly correlated with salinity ($r = 0.734$, $P < 0.0001$), TPM ($r = 0.500$, $P < 0.01$), POM ($r = 0.372$, $P < 0.05$) and PIM ($r = 0.523$, $P < 0.001$) but not chlorophyll ($r = 0.211$, $P > 0.05$). These results suggest that reduced transparencies may have been a consequence of influxes of particulate-laden freshwater rather than variations in primary productivity. It should be pointed out that secchi disk measurements were obtained under a variety of different lighting, weather and sea surface conditions. As such, using a secchi disk under nonstandardised conditions may be partly responsible for the poor correlations with particulate levels ($r = 0.5$) and chlorophyll-*a* ($r = 0.4$). As the parameters which influence transparency (eg. TPM, PIM, POM and chlorophyll levels) were regularly monitored at all sites, the secchi depth measurements recorded in this study are not particularly useful.

### 4.4.4 Current speed and direction

Current speeds and directions were not dissimilar to those reported by Matthews and Volframs (1978) nor to previous recordings undertaken by Aquatas (unpublished data). The range of current speeds considered to be adequate for suspended mussel culture is approximately 2 - 6 $cm \cdot sec^{-1}$ (Sutterlin et al., 1981; Larsson, 1985, both cited in Okumus, 1993). Although the overall average current speed of 3.4 $cm \cdot sec^{-1}$

fell within this range, approximately 40% of recordings were below 2 $cm \cdot sec^{-1}$. Under commercial culture situations where mussels are suspended from rafts or tiers of longlines, these low current speeds may result in localised seston depletion (ie. shadowing effects). Food particles are cleared from the water column by mussels at the leading edges of a culture system, significantly reducing the food supply and growth of mussels located downstream (Kirby-Smith, 1972; Rodhouse et al., 1985). At the other end of the scale, maximum current speeds of 25 $cm \cdot sec^{-1}$ are below those that might reduce mussel filtration rates and growth (Walne, 1972; Kirby-Smith, 1972; Wildish and Miyares, 1990).

Directional data confirms that the four longlines were appropriately positioned to intercept fish farm waste particles carried by the main current flow, assuming waste particles were still in suspension by the time they reached the longlines. The fate of particulate wastes will be discussed further in Section 13 - Fish Farm Wastes.

### 4.4.5 Suspended Particulate Matter

The primary objectives for monitoring suspended particulate matter (SPM) in this study were to determine: the quantity and, to some extent, the quality of SPM available for mussels at each site; the influence, if any, of the fish farm in raising SPM concentrations above ambient levels and; if fish farm particulate wastes were being transported to any or all of the mussel longlines (also see Section 13 - Fish Farm Wastes).

Mean weekly total suspended matter (TPM) or seston levels ranged from 3.7 to 9.7 $mg \cdot l^{-1}$ (overall trial mean of 6.6 $mg \cdot l^{-1}$), POM ranged from 0.8 to 4.5 $mg \cdot l^{-1}$ (overall trial mean of 2.6 $mg \cdot l^{-1}$), and %POM from 18.3 to 51.4% (overall trial mean of 34.7%). As there is a paucity of particulate data from Tasmanian waters in general and NWB in particular, it is unknown if the range of observed particulate concentrations are characteristic levels for North West Bay. However, a comparable annual average concentration of 8 mg $TPM \cdot l^{-1}$, which is considered to be moderate to low compared to other estuarine systems, with no discernable seasonal pattern, was reported for the adjacent Derwent estuary in 1993-94 (DELM, 1995). The overall mean and range of TPM, POM and %POM levels observed in the present study were also similar to samples taken from marine sites adjacent to three oyster growing embayments in southern Tasmania (Little Swanport Lagoon, Pipeclay Lagoon and Pittwater) over a comparable period in 1995-96 (I. Mitchell, Marine Fisheries Laboratory, Tasmanian Aquaculture and Fisheries Institute, unpublished data). Ms. Mitchell's data also displayed little seasonal variation in TPM and POM levels, although higher particulate

concentrations in December 1995 contrasts with the low concentrations observed in the same month in NWB. Therefore, it appears that the observed particulate levels and the general lack of any marked seasonal trends within North West Bay were not dissimilar to other coastal areas in southern Tasmania.

Although particulate concentrations (TPM, POM, PIM and NAP) were higher at the centre of the fish farm than at the four mussel longline sites, they were higher during the fallowing period when fish were not on the farm (June to mid-September 1995) and did not display appreciable increases, absolute or relative to the other four sites, after fish were introduced and cultivated from September 1995 onwards. It is possible that higher particulate concentrations during the fallowing period at site X may have been due to some degree of resuspension of fish farm waste sediments or contamination of the water samples from these sediments. However, after fish were re-introduced during September-October 1995, no appreciable increase in particulate concentrations, either in absolute terms or relative to the other four sites, was observed. This suggests that the quantity of particulate wastes generated by the fish farm (feed fines and fish faeces) was too low or diluted to achieve detectable concentrations. It is also possible that a significant fraction of faecal material may have remained bound as pellets, thereby not contributing to SPM concentrations.

Previous reports are inconsistent concerning the level of particulate matter associated with fishfarms relative to ambient concentrations. Significantly higher SPM levels, relative to control sites, were associated with a fish farm in British Columbia (Jones and Iwama, 1991). Okumus (1993) reported conflicting results, whereby seston and POM concentrations ($mg \cdot l^{-1}$) at salmon farms were higher than control sites but neither particle concentrations (no. $particles \cdot ml^{-1}$) nor particle size frequency distributions were significantly different among the same sites; perhaps this apparent contradiction indicates differences in particle densities. No significant association between SPM concentrations and distance from fishcages was observed by Taylor et al. (1992); it was speculated that net-fouling mussels were intercepting suspended wastes, thereby preventing particulates escaping from the fish cages.

Of particular interest is, contrary to expectations, that particulate levels at the two farm sites were not significantly higher than at sites III and IV. Perhaps particulate concentrations were higher at the farm sites, but were below detectable levels, or fish farm waste particles were not horizontally transported to sites I and II (70 m and 100 m from the cages). An examination of calculated particulate waste generation and distribution reveals that both factors are plausible: particulate fish farm wastes may have been too diluted to significantly raise ambient concentrations; and farm-site

mussels may have been too far removed from the sea cages to intercept suspended farm wastes before they sank below the level of the socks (see Section 13).

### 4.4.6 Chlorophyll

Phytoplankton biomass, as indirectly measured by chlorophyll-*a* concentrations, was generally low, with an overall trial mean of 1.1 ± 0.2 µg chlorophyll-*a*·l$^{-1}$. Negligible winter chlorophyll concentrations were followed by a short spring bloom in September 1995, achieving a peak of 2.9 µg·l$^{-1}$. Concentrations for the duration of the spring and early summer period remained low (< 1.5 µg·l$^{-1}$) but increased during February 1996 to peak at 5.8 µg·l$^{-1}$. The annual range of chlorophyll-*a* concentrations was not dissimilar to those previously reported for Storm Bay (Clementson et al., 1989), the D'Entrecasteaux Channel (Crawford et al., 1996) and the Lower Huon River estuary (Woodward et al., 1992). The mean summer-autumn (December 1995 to May 1996) concentration of 1.6 µg·l$^{-1}$ also compares with the reported mean concentration (1.1 µg·l$^{-1}$) from southern Tasmanian coastal waters over the same months in 1993-94 (Jameson and Hallegraeff, 1994). These concentrations are also comparable to those experienced in some Scottish lochs (Gowen et al., 1988), although levels up to 20 µg·l$^{-1}$ have been reported during bloom periods (Okumus, 1993). However, they are are low when compared to some studies in Canadian waters. Summer sub-surface maxima can range up to 20 µg·l$^{-1}$ in British Columbia salmon-farming waters (Haigh and Taylor, 1991; Jones and Iwama, 1991) and 32-90 µg·l$^{-1}$ in the Bay of Fundy, New Brunswick (Wildish et al., 1993).

Some studies have reported higher chlorophyll-*a* concentrations, at times, in waters associated with fish farms (eg. Mäkinenn and Pursianien, 1987 cited in Gowen et al., 1988; Aure et al., 1988; Jones and Iwama, 1991; Pridmore and Ruthorford, 1992). Increased primary production in waters adjacent to fish farms is held to be a consequence of nutrient enrichment, particularly excreted ammonia (Gowen et al., 1988). Holby and Hall (1994) found that high phosphorus and nitrogen loadings from a Swedish marine trout farm provided a good niche for the growth of diatoms. However, significantly higher chlorophyll-*a* concentrations were not associated with the fish farm in the present study, nor several other similar studies (Gowen et al., 1988; Taylor et al., 1992; Okumus, 1993; Wildish et al., 1993). Possible explanations for chlorophyll concentrations not being higher within the fish farm include: ambient nutrient concentrations were not at growth-limiting levels and waste nutrients from the fish farm would not have affected phytoplankton growth; nutrient wastes were insufficient or too diluted to stimulate growth; phytoplankton growth was stimulated, but was not detected due to dilution, zooplankton-grazing, or was

transported away from the farm and redistributed throughout the bay. In any case, according to measured chlorophyll-*a* concentrations, none of the four mussel longline sites provided mussels with any nutritional advantage over the others in terms of enhanced phytoplankton levels.

### 4.4.7 Nutrients

Seasonal cycles in nutrient concentrations, which typically result from various interactions between physical processes and phytoplankton growth, have been descibed by various authors (eg. Parsons et al., 1984; Valiela, 1984; Gowen et al., 1988). Generally during the winter months in temperate waters, light is the most critical factor limiting growth and there is little uptake of nutrients by phytoplankton. This, together with regeneration from sediments and the possible resupply from more open coastal waters, results in yearly-maximum nutrient levels. Due to vertical mixing in winter, nutrients tend to be uniformly distributed throughout the water column. During the spring and summer, stratification of the water column may occur and prevent mixing. As a result of increasing phytoplankton growth in the euphotic zone during spring and summer, surface waters tend to become depleted in nutrients. When phytoplankton growth slows in the autumn, regeneration exceeds utilisation and, in conjunction with vertical mixing, winter levels are re-established at all depths.

The seasonal cycles of all nutrients monitored during the present study followed the general pattern described above. Nitrogen levels were high during the winter of 1995, depressed during the spring-summer of 1995-96, and high again in autumn 1996. Phosphate levels were more stable over the study period; but did display reduced levels in summer compared with winter and autumn levels. Silicate concentrations were also high in winter and were followed by a depression in spring and early summer, although wide fluctuations with three distinct peaks were observed during the summer of 1996. The levels of all nutrients displayed a significant decline in September 1995, corresponding with a short spring phytoplankton bloom.

The range of observed $PO_4$ and $NO_3$ levels were comparable to those reported from Storm Bay (Clementson et al., 1989; Harris et. al., 1991), Derwent River estuary (DELM, 1993), D'Entrecasteaux Channel (Crawford et al., 1996), the lower Huon River estuary (Woodward et al., 1992), and are typical concentrations for "average" surface seawater (Valiela, 1984). It has been suggested that nitrites are typically short-lived in seawater as they are rapidly oxidised and are usually < 5% of nitrate concentrations (Clementson et al., 1989). This is reflected in the present results, whereby $NO_2$ was a minor portion of $NO_X$ and only in detectable quantities during the

winter and autumn periods, corresponding with higher nitrate levels. Silicate concentrations were also similar to those reported from other coastal locations in southeastern Tasmania (Crawford, 1996).

The extent to which measured nutrients influenced phytoplankton production within NWB during the period of study is difficult to assess. Algal growth and standing crop at any particular time are determined by numerous interacting factors including temperature, light intensity, photoperiod, nutrients (macro and micro), zooplankton grazing and toxicants. Also, different algal species have different requirements for optimum growth, which partly accounts for species succession over time. In this study, interactions among environmental parameters influencing algal growth appears likely; phytoplankton biomass, as chlorophyll-a, was not strongly correlated with any single environmental parameter (Table 4.1). These results agree with Boynton et al. (1982), who explored correlative relationships between phytoplankton production and biomass with several physical, chemical (including nutrients), and biological variables from a broad spectrum of estuarine systems. They reported that very few, if any, environmental variables alone were satisfactory predictors of phytoplankton production or biomass. As such, an in-depth analysis of environmental factors influencing phytoplankton biomass in NWB is beyond the scope of this study. However, a few general points regarding nutrient levels may be raised.

In coastal marine systems, nitrogen is considered to be the principal element limiting phytoplankton growth (McCarthy, 1980; Valiela, 1984). Nitrate is the predominant nitrogen source (80% of total N; Bougis, 1976), with the remainder comprised of nitrite, ammonia, urea and dissolved amino acids. It has been suggested that when ambient nitrate concentrations fall below 0.7μM (9.8 μg $NO_3$-N·$l^{-1}$) nitrogen-deficient cells are produced, unless ammonia is present, before cell division stops (Millero and Sohn, 1992). Total nitrogen ($NO_X$ = $NO_3$ + $NO_2$) was below this threshold concentration from October 1995 through to the end of March 1996, indicating that low nitrogen levels may have been a factor in limiting phytoplankton growth. However, it should be pointed out that nutrient deficiency does not imply that growth is necessarily retarded or ceases (Paasche, 1980). Although significant correlations between nitrogen (nitrates + nitrites) concentrations and chlorophyll-a have been reported (Gowen, 1994), no biologically significant correlations between chlorophyll and $NO_X$, at time lags of 0 to 4 weeks, were observed in this study. Possibly, alternative sources such as ammonia and dissolved organic nitrogen were available to satisfy phytoplankton nitrogen requirements.

Marine plankton preferentially absorb ammonia over nitrates and nitrites (McCarthy, 1980; L'Helguen et al., 1996; Lomas and Glibert, 1999). Although generally found in

small amounts relative to nitrate levels, ammonia concentrations display considerable temporal and spatial variability, even being the dominant nitrogen source in some cases (McCarthy, 1980). It has been found that when ambient ammonia concentrations are in the order of 7 - 14 $\mu g\ N \cdot l^{-1}$, nitrate utilisation is suppressed (McCarthy et al., 1977; McCarthy, 1980). Although ammonia concentrations were not monitored, studies of the nearby Derwent and Huon River estuaries may provide some indication of ammonia levels that may have been evident in North West Bay. Total ammonia in the estuarine section of the Huon River ranged up to 36 $\mu g\ TAN \cdot l^{-1}$ and up to 14 $\mu g\ TAN \cdot l^{-1}$ at marine sites in the D'Entrecasteaux Channel from 1996 to 1998 (HES, 2000). Ammonia concentrations exceeding those of nitrates+nitrites (annual means 24 $\mu g\ NH_4\text{-}N \cdot l^{-1}$ and 15 $\mu g\ NO_X\text{-}N \cdot l^{-1}$) were reported in adjacent waters of the D'Entrecasteaux Channel/lower Derwent River estuary (near Tinderbox) in 1993-94 (DELM, 1995). These high ammonia levels were attributed to inputs from sewage treatment plants, urban stormwater runoff and industrial effluents associated with the greater Hobart municipalities. Considering the close proximity of the Tinderbox sampling station (DELM, 1995) with the sites sampled in NWB in the present study (ie. within 3km) and that a significant portion of NWB water originates from the Derwent estuary (Matthews and Volframs, 1978), high ammonia concentrations may have also been evident in NWB. Ammonia inputs to NWB would have come from these incursions of Derwent estuarine water, effluent from two primary sewage treatment plants at Margate and Electrona, the surrounding watershed, as well as the Aquatas fish farm. However, as ammonia and other alternate sources of nitrogen (urea, amino acids) were not measured during this study, the total nitrogen available for algal growth in NWB during 1995-96 remains unknown.

At phosphate levels above a concentration of 0.3μM (9.3 $\mu g\ PO_4\text{-}P \cdot l^{-1}$), the rate of growth for many species of marine phytoplankton is independent of the concentration of P. Below 9.3 $\mu g\ PO_4\text{-}P \cdot l^{-1}$ cell division becomes inhibited, although this probably does not occur in most marine waters - $NO_3$ is usually exhausted before $PO_4$ reaches this critical level (Millero and Sohn, 1992). In the present study, $PO_4$ levels never fell below 10.0 $\mu g\ PO_4\text{-}P \cdot l^{-1}$, suggesting that phosphate was not limiting phytoplankton growth. This is reinforced, to some degree, by an examination of N:P ratios.

The 'Redfield ratio' has been applied as one approach for assigning a limiting role to major phytoplankton nutrients (Redfield, 1958; McCarthy, 1980). Redfield observed that the atomic ratio of N:P in both phytoplankton and nutrient-rich water approximates 16:1. It has been proposed that ratios above 20 or below 15 are indications of phosphorus or nitrogen-limited water, respectively (eg. DELM, 1995). If this generalisation is valid, then North West Bay waters were consistently nitrogen-deficient, as the N:P ratio of measured nutrients never exceeded 8. In particular, N:P

ratios never exceeded 1.0 from October 1995 through April 1996 which would suggest that phytoplankton production may have been nitrogen-limited. However, as ammonia and other available nitrogen sources are not accounted for, these ratios may be severely underestimated. For example, during the period where N:P ratios approximated 1.0, chlorophyll-a concentrations displayed several distinct peaks in November 1995 and January - March 1996 (Fig. 4.6).

Silicates were the only nutrient to have displayed any significant, albeit weak ($r = 0.33$; $P < 0.05$; Table 4.2), correlation with chlorophyll-a levels. Therefore, plankton growth may have been, at least partially, influenced by silicate concentrations. However, it has been suggested that silicon concentrations are rarely at growth-limiting levels in seawater, due to efficient silicon assimilation by diatoms and a fast redissolution rate of biogenic silica (Paasche, 1980). Nelson and Goering (1978 cited in Paasche, 1980) were unable to find clear cases of silicon limitation at ambient concentrations of 1.3 - 4.3 uM (36 -120 ug $Si\text{-}SiO_4 \cdot l^{-1}$) in Mexican and West African upwelling regions. Silicate concentrations in NWB were rarely below this range, generally exceeding 100 $\mu g\ Si \cdot l^{-1}$, except for the spring-early summer (September 22 to December 13 1995) when the mean concentration was $32.8 \pm 1.3\ \mu g\ Si \cdot l^{-1}$ (range 16 - 57 $\mu g\ Si \cdot l^{-1}$).

Although nutrients are often assumed to play a dominant role in algal growth, other environmental factors may be of equal or greater importance. For example, Hallegraph and Westwood (1994, cited in DELM, 1995) concluded that light limitation by turbid water, not nutrients, was the significant factor suppressing algal growth in the nearby Derwent River estuary. Therefore, plankton growth in NWB may have been similarly influenced by other factors that covaried with silicate concentrations, such as photoperiod and/or light intensity.

The questions of most relevance regarding nutrient levels in this study are: (i) was the salmon farm introducing nutrients at sufficient quantities to significantly increase nutrient concentrations above background levels; (ii) was phytoplankton growth enhanced in NWB as a consequence of nutrient enrichment from the fish farm; and (iii) was phytoplankton growth enhanced only in the vicinity of the fish farm, thereby providing fish farm mussels (sites I and II) with a greater food supply than mussels grown at the two distant sites (III and IV)?

Firstly, that the Aquatas fish farm is introducing dissolved inorganic nutrients to the NWB environment is not in question. Added nutrients come from fish excretory products and the leaching of various soluble compounds from feed and faeces. However, there were no significant differences in the concentrations of any of the

measured nutrients, except occasionally silicates, between the fish farm sites (X, I and II) and the other two sampling locations within NWB (III and IV). Similar results were reported by Gowen et al. (1988) and Wildish et al. (1993), whereby no significant increases in nitrates, phosphates or silicates were detected in waters associated with fish pens in Scotland or Nova Scotia. Due to the rapid nitrification of ammonia, the primary fish excretory product, to nitrite and nitrate in well-oxygenated waters (Valiela, 1984), elevated levels of nitrates and nitrites within the Aquatas fish farm was considered to be a definite possibility. Gowen et al. (1988) theorised that nitrogen and phosphorus compounds produced by a fish farm may be rapidly assimilated by phytoplankton and, therefore, gone undetected. However, they considered this possibility unlikely, as no differences in nutrient concentrations between fish farm and control sites were evident during winter months when plankton abundance was low. Similar comparisons of winter with summer nutrient levels cannot be tested in the present study as no fish were present on the farm site over the winter months. However, there were no significant differences in nutrient concentrations between the five sampled NWB sites and were similar to reported concentrations from other locations in southern Tasmania. It appears that the inputs of measured nutrients from the fish farm were either too low and/or too diluted to be detectable with the methodology employed.

The degree to which the Aquatas fish farm influenced ambient ammonia levels within NWB remains unknown. Elevated ammonia concentrations within fish farms have been reported but these appear to be largely limited to the immediate vicinity surrounding farms (Gowen et al., 1988; Wildish et al., 1993; HES, 2000). At a fish farm in Nova Scotia, Wildish (1993) reported considerable variation of ammonia concentrations among sampling dates (high in summer-autumn and low in winter), time of day (possibly due to variation in flushing rate and fish excretion), sample depths, and fish cages. Among 39 net pens sampled on the same day, ammonia concentrations varied by a factor of five (3 to 14 μM) with no identifiable pattern related to either the location or contents of net pens.

Despite the uncertainty regarding the quantity of ammonia excreted from the Aquatas farm, what is of most significance to this study is that the nutrient outputs from the salmon farm were not manifested in: (i) higher levels of primary production in NWB compared with those from other coastal embayments in southern Tasmania, or (ii) measurably higher levels of phytoplankton for mussels cultivated in the vicinity of the fish cages. The first conclusion, together with the results from the other measured environmental parameters (ie. generally no significant differences between sites in NWB), has important implications; the Aquatas fish farm appears to have a minimal impact on the water quality and level of primary production within NWB. The second

conclusion pertains more directly to this project. Based on particulate and chlorophyll-a data, which showed no differences among longline sites, there is no advantage in culturing mussels on the fish farm in order to provide an enhanced food supply. However, this is not to say that the quality of particulate material (digestibility or energy content) was the same at all sites. However, if the quality of food differed among sites, then a logical consequence might be differences in various mussel growth performance indices (eg. shell length growth, weight growth, condition, glycogen content, fecundity); these performance indices are addressed in the following sections. A more detailed discussion of dissolved nutrients, including ammonia, and the quantity and quality of solid wastes potentially discharged from the Aquatas fish farm, is provided in Section 13 - Fish Farm Wastes.

# 5. Mussel Growth

## 5.1 Introduction

The single most important factor influencing mussel growth is food supply (Seed and Suchanek, 1992). Enhanced growth of mussels (Wallace, 1980) and oysters (Jones and Iwama, 1991) cultured near fish cages has been attributed to elevated particulate food levels due to the discharge of uneaten fish feed and fish faeces, and increased phytoplankton production. Therefore, it was expected that any enrichment of the available food supply within the Aquatas fish farm might similarly be manifested by enhanced growth of mussels at sites I and/or II relative to the growth of mussels cultured at sites III and IV.

Mussel growth was assessed by monitoring the monthly changes in several parameters from March 1995 to May 1996, excluding May 1995. Monthly measurements included shell length (L), whole live weight (WLW), dry shell weight (SW), wet meat weight (WMW), dry meat weight (DMW), ash-free dry meat weight (AFDMW) and total individual biomass (IB).

Individual mussel biomass production during the trial period was also calculated. Production represents the net gain in body energy and occurs when the energy content of the absorbed food ration exceeds metabolic requirements. This energy surplus is then utilised for somatic growth and/or gamete production. If energy intake falls below what is required for maintenance, body reserves are utilised and negative growth may occur. Gross production of longline units was not determined due to the variation in the density of socks sampled over the trial period (see Section 8).

## 5.2 Methodology

Each month, twenty mussels were randomly selected from each sample collected from the four longlines (12 samples per site, 48 samples total per month). Shell length (L) was determined by measuring the maximum anterior - posterior axis (McDonald et al., 1991) of each mussel using sliding vernier callipers (± 0.05 mm). Each subsample (20 mussels) was bulk weighed (± 0.05 g, Sartorius IP65) to obtain an estimate of mean whole live weight per mussel (total wet weight/20), except for March 1995 and March 1996 when individual weights were measured.

Soft tissues from each subsample of twenty mussels were removed and any remaining byssal threads excised with scissors; therefore all meat weight data presented are "less

byssus". Meats were blotted with paper towelling and bulk weighed (± 0.05 g) to determine the mean WMW per mussel (total wet meat weight/20). Shells were also bulk weighed to determine mean wet shell weight per mussel (total shell weight/20). Dry shell weight (SW), dry meat weight (DMW) and meat moisture content (MC) were obtained after drying in an oven to constant weight (24 to 72 h at 80°C). Meat ash content (% DMW) and ash-free dry meat weight (AFDMW) were obtained by combusting 1 g subsamples of dried and ground meat in a muffle furnace at 480°C for 5 hours. AFDMW was calculated by subtracting the ash weight from DMW.

Individual biomass (IB, g·mussel$^{-1}$) is the total organic content of an individual. Monthly mean IB of each sample was calculated from the sum of sample mean ash free dry meat weight (AFDMW, g·mussel$^{-1}$) and the weight of shell organics. Shell ash and organic content were determined from samples collected at the start (March 1995) and conclusion (May 1996) of the trial. There were no significant differences in the percentage of shell ash or organic content between these two sampling dates nor among sites ($P > 0.05$, mean ash content 95.74 ± 0.04%, organic content 4.26 ± 0.04%, n = 60 samples). Therefore, shell ash and organic content were considered to have remained constant throughout the trial. Monthly estimates of total individual biomass incorporated the monthly mean weight of shell organic matter (ie. 4.3% x g dry shell weight·mussel$^{-1}$) with monthly ash-free dry meat weight (AFDMW, g·mussel$^{-1}$).

Net individual mussel production (IP) is defined in this study as the change in retained biomass within each mussel. Net IP was measured as monthly increments or losses in mean individual biomass. Net individual production only accounts for biomass retained within mussels and does not account for biomass lost as gametes. In order to estimate the total biomass produced per mussel during the course of the trial, including lost reproductive material, monthly biomass increments (mean individual biomass at each depth within each site minus mean individual biomass previous month) were summed to obtain gross individual biological production (GP). Similarly, biomass losses for those months where mean individual biomass decreased were summed to obtain an estimate of individual eliminated biomass (EB). These losses would be primarily reproductive tissue, but any metabolic losses would also be included. The difference between cumulative GP and EB at any particular month gives net retained biomass or net production at that month.

Length, weight and production data were subjected to ANOVA at four key sampling dates (March 1995, August 1995, February 1996 and May 1996) with SITE and DEPTH as fixed factors. The number of replicates used in the analyses was dependent on the parameter under investigation. Shell length analyses incorporated individual

mussel length measurements (n = 120 mussels·depth$^{-1}$·site$^{-1}$); weight and production analyses used the mean mussel weight (g·mussel$^{-1}$) derived from the bulk sample weight of 20 mussels·sample$^{-1}$ (n = 6 samples·depth$^{-1}$·site$^{-1}$). Normality of data was checked (Shapiro-Wilk test) and homogeneity of variances assessed (Cochran's test) prior to analyses. Heterogeneous variances were indicated for length and weight data in February and May 1996, transformations of the data failed to stabilise these heterogeneous variances and ANOVA proceeded with the original data with the understanding that any significant differences among treatments at an $\alpha$ level of 0.05 be viewed with caution.

Shell length growth was analysed further by fitting von Bertalanffy (VB) growth curves to individual shell length data from each depth within each site, pooled length data from each site and pooled length data from each growing depth. The general VB growth equation takes the form of:

$$L_t = L\infty \cdot (1 - e^{-k(t - to)}), \text{ where}$$

| | | |
|---|---|---|
| $L_t$ | = | Length (mm) at age t |
| $L\infty$ | = | Theoretical asymptotic length (mm) |
| e | = | Base of natural logarithm |
| K | = | Growth coefficient (month$^{-1}$), |
| t | = | Age (months) |
| $t_0$ | = | Theoretical age (months) when length is 0 mm. |

As the age of mussels was unknown, age was substituted with time (months) relative to the beginning of the growth trial. March 1995 was designated as 0 months and subsequent sampling months assigned a time from 1 (April 1995) to 14 (May 1996). Parameters for each VB growth equation ($L\infty$, K and $t_0$) were derived by iteration using least squares nonlinear regression (Systat 5.1). Analysis of residual sum of squares (ARSS, Chen et al., 1992) was used to determine if fitted VB curves differed among the four sites. This was followed by multiple comparison testing among the four curves using Likelihood Ratio tests (LR) to determine which curves differed and to identify the source(s) of difference (ie. VB parameters). LR tests were performed using a computer program developed by Malcolm Haddon (TAFI, Hobart) based on the procedures described by Kimura (1980).

## 5.3 Results

### 5.3.1 Shell length

The pattern of shell length growth during the trial period was similar at all sites, increasing from an initial overall mean length of 55.8 ± 0.2 mm (n = 960 mussels) in March 1995 to 85.0 ± 0.2 mm (n = 960 mussels) in February 1996 (Fig. 5.1). Shell growth was continuous during this period, although little or no growth was observed at sites II, III and IV in July 1995. Shell growth effectively ceased over the latter stages of the trial, with an overall final mean length of 83.9 ± 0.3 mm (n = 912 mussels) in May 1996.

Differences in shell length among sites were significant for each of the key sampling months (March 1995, $P < 0.01$; August 1995, $P < 0.01$; February 1996, $P < 0.0001$; May 1996, $P < 0.05$) and between growing depths on three key months (August 1995, $P < 0.05$; February 1996, $P < 0.0001$; May 1996, $P < 0.01$). However, differences among sites and depths were minor on each occasion, with mean shell lengths being within 2.9 mm. Differences were detected due to the large sample size and corresponding residual degrees of freedom (df = 952). These minor differences become insignificant if ANOVA is applied to sample mean lengths (residual df = 40) rather than individual shell length data. Shell length increments from initial mean lengths (55.8 mm) for the August 1995, February 1996 and May 1996 sampling dates were 18.1 ± 0.1 mm, 29.2 ± 0.6 mm and 28.0 ± 0.7 mm (n = 48 sample means), respectively, with no significant site differences ($P > 0.05$).

Estimated parameters of the von Bertalanffy growth equation (VB) for mussels grown at each depth within each site are presented in Table 5.1. $L_\infty$ ranged from 86.2 ± 0.6 mm (site IV - 1m) to 91.1 ± 0.2 mm (site I - 5m) and k ranged from 0.163 ± 0.012·month$^{-1}$ (site II - 1m) to 0.196 ± 0.011·month$^{-1}$ (site II - 5m). The eight VB growth curves are presented in Fig. 5.2, illustrating the similarity of shell length growth across all sites and depths. Predicted VB final shell lengths were within 4.0 mm across all site-depths (84.0 at IV-1m to 87.8 mm at I-5m). The slight overestimation of predicted final lengths compared with observed lengths is due to the slight decrease in mussel size over the last three months.

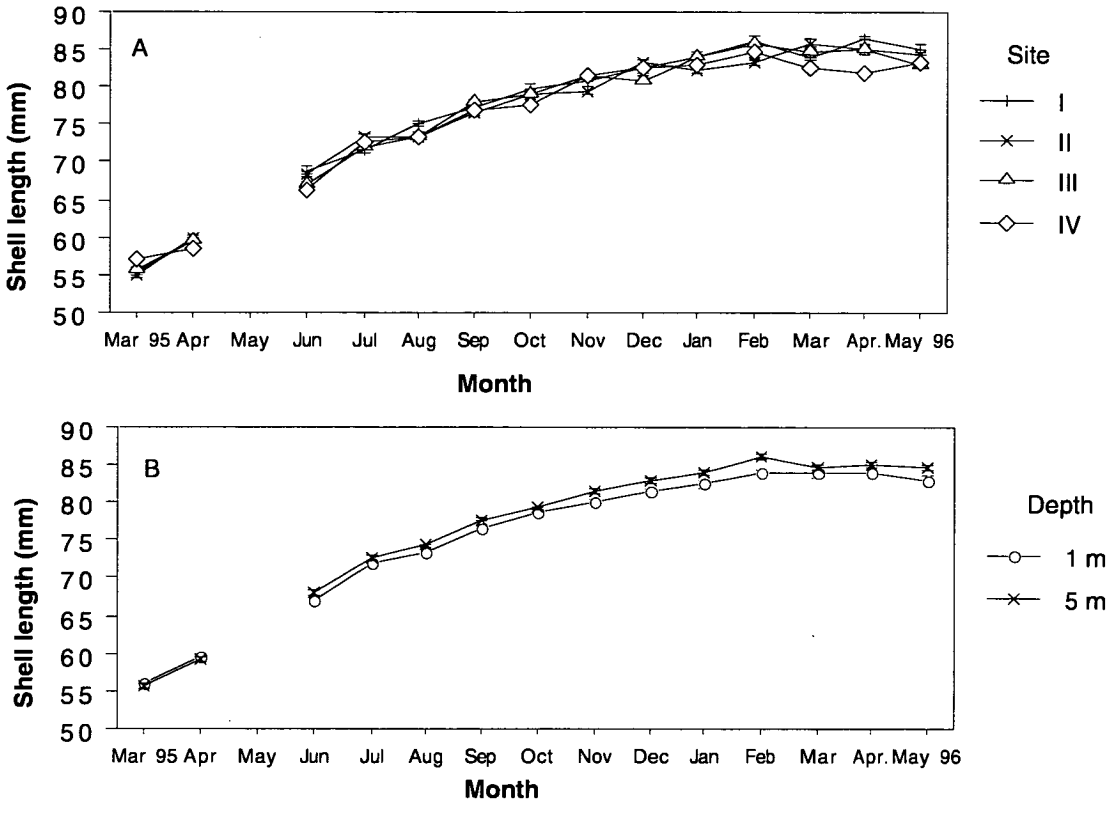


**Figure 5.1.** (A) Monthly shell length of experimental mussels at each site (mean ± SE mm, n = 240 mussels·site$^{-1}$) and (B) at each depth (n = 480 mussels·depth$^{-1}$). Error bars not visible lie within point boundaries.

**Table 5.1.** Shell length growth. Estimated parameters of the von Bertalanffy growth equation ($L\infty$, k and $t_0$ ± SE) for mussels grown at each depth (1m and 5m) within each site (I - IV). n = number of individual mussels measured from March 1995 to May 1996. Also included are the predicted and observed shell lengths at the conclusion of the trial in May 1996.

| Site | Depth (m) | $L\infty$ (mm) | k (month$^{-1}$) | $t_0$ (months) | n | Predicted Length May 1996 | Observed Length May 1996 |
|---|---|---|---|---|---|---|---|
| I | 1 | 87.56 (0.85) | 0.184 (0.013) | -5.37 (0.37) | 1657 | 85.1 | 84.3 |
| I | 5 | 91.09 (0.88) | 0.172 (0.011) | -5.26 (0.32) | 1670 | 87.8 | 85.9 |
| II | 1 | 88.44 (0.93) | 0.163 (0.012) | -5.95 (0.40) | 1676 | 85.0 | 83.9 |
| II | 5 | 88.40 (0.68) | 0.196 (0.011) | -4.87 (0.28) | 1665 | 86.2 | 84.7 |
| III | 1 | 88.39 (0.79) | 0.173 (0.011) | -5.56 (0.33) | 1675 | 85.4 | 82.1 |
| III | 5 | 88.77 (0.71) | 0.182 (0.010) | -5.26 (0.29) | 1668 | 86.1 | 84.0 |
| IV | 1 | 86.19 (0.64) | 0.190 (0.011) | -5.41 (0.31) | 1676 | 84.0 | 82.0 |
| IV | 5 | 87.67 (0.61) | 0.182 (0.010) | -5.36 (0.26) | 1680 | 85.1 | 84.4 |

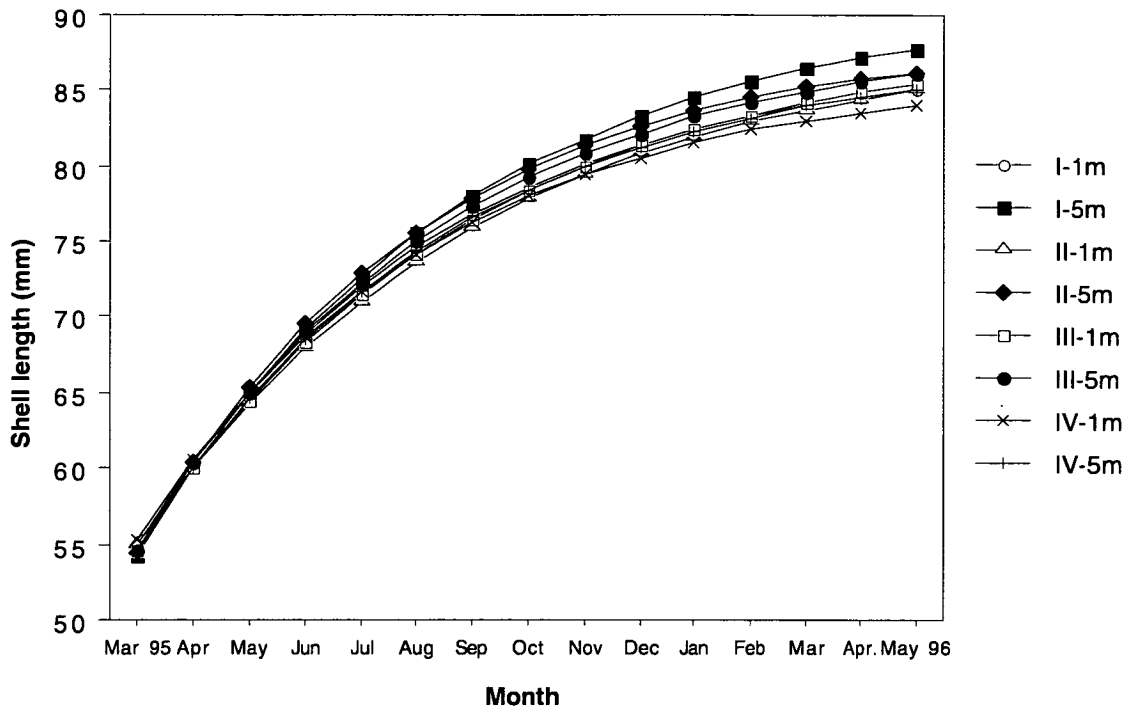


**Figure 5.2.** Von Bertalanffy growth curves modelling mussel shell length growth at each depth (1m and 5m) within each site (I - IV).

Analysis of residual sum of squares (ARSS) performed on VB curves generated from pooled lengths at each site indicated significant differences among the four curves ($P < 0.0001$). Multiple comparisons of VB curves using LR tests indicated that mussel growth was the same at sites II and III (ie. coincident curves, $P > 0.05$, Table 5.2). Although all other paired curves were significantly different the only differences in VB parameters lay in $L\infty$ at site IV (86.9 mm) being less than $L\infty$ at the other three sites ($P < 0.05$; range 88.3 to 89.4 mm). As the greatest difference in $L\infty$ was only 2.5 mm and with no significant differences in growth rates (k), shell growth was definitely similar among all sites. Although VB growth curves differed among depths ($P < 0.0001$), only a minor difference in $L\infty$ (88.59 mm at 1 m and 88.96 at 5 m) was shown to be significant ($P < 0.01$).

**Table 5.2.** Von Bertalanffy growth equation parameters ($L\infty$, k and $t_0$) for shell length at each site (I, II, III, IV) with probabilities of coincident curves (cc) and of common VB parameters between sites (Likelihood Ratio test). Where $P < 0.05$, VB curves and/or parameters are significantly different. ns = not significant ($P > 0.05$).

| Site | | | II | | | | III | | | | IV | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Parameter | | cc | $L\infty$ | K | $t_0$ | cc | $L\infty$ | K | $t_0$ | cc | $L\infty$ | K | $t_0$ |
| | | | | 88.343 | 0.180 | -5.350 | | 88.576 | 0.177 | -5.408 | | 86.914 | 0.186 | -5.382 |
| I | cc | | 0.03 | | | | 0.030 | | | | <0.0001 | | | |
| | $L\infty$ | 89.362 | | ns | | | | ns | | | | < 0.001 | | |
| | K | 0.177 | | | ns | | | | ns | | | | ns | |
| | $t_0$ | -5.334 | | | | ns | | | | ns | | | | ns |
| II | cc | | | | | | > 0.05 | coincident | | curves | < 0.001 | | | |
| | $L\infty$ | 88.343 | | | | | | ns | | | | 0.04 | | |
| | K | 0.180 | | | | | | | ns | | | | ns | |
| | $t_0$ | -5.350 | | | | | | | | ns | | | | ns |
| III | cc | | | | | | | | | | <0.0001 | | | |
| | $L\infty$ | 88.576 | | | | | | | | | | 0.01 | | |
| | K | 0.177 | | | | | | | | | | | ns | |
| | $t_0$ | -5.408 | | | | | | | | | | | | ns |

### 5.3.2 Whole Live Wet Weight

Initial overall mean whole live wet weight WLW of stocked mussels was $14.6 \pm 0.2$ g·mussel$^{-1}$ (n = 48 samples). WLW growth was similar among sites, steadily increasing from March 1995 to February 1996 (overall mean WLW $48.9 \pm 0.9$ g mussel$^{-1}$) after which no further increase was observed (Fig. 5.3). Final WLW in May 1996 was $49.3 \pm 1.1$ g mussel$^{-1}$ with no significant differences among sites or between growing depths ($P > 0.05$). No significant differences between sites or depths were indicated in August 1995. A difference between growing depths was indicated for the February 1996 sample period ($P < 0.05$), where WLW of mussels grown at 1m (mean $46.9 \pm 1.1$ g·mussel$^{-1}$, n = 24 samples) was slightly less than those grown at 5 m ($50.9 \pm 1.2$ g·mussel$^{-1}$). No significant site x depth interactions were indicated for any of the key sampling periods ($P > 0.05$).

WLW in August 1995, February 1996 and May 1996 represented overall mean increments of 17.1, 34.2 and 34.6 g·mussel$^{-1}$, respectively, from initial WLW. No significant increase in WLW was indicated for the last four months, February through May 1996 ($P > 0.05$, overall mean $49.2 \pm 0.5$ g·mussel$^{-1}$, n = 192 samples). Therefore, WLW attained by February 1996 represented a 235% increase relative to initial WLW.

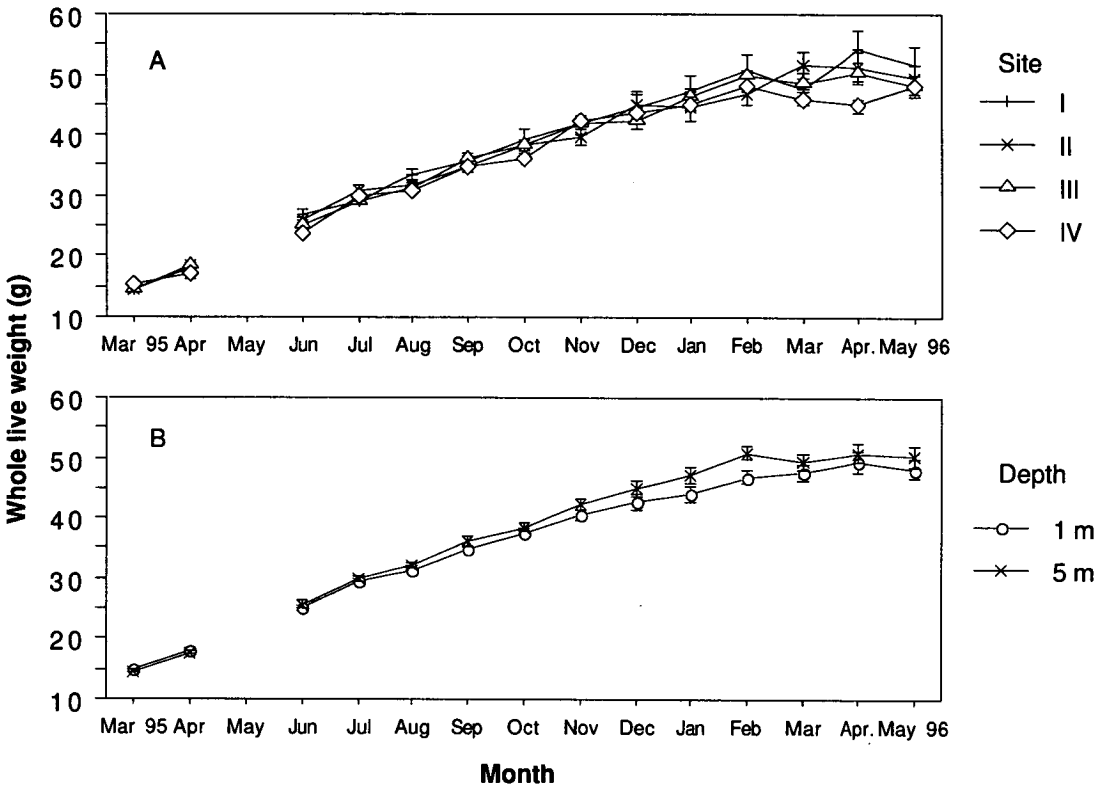


**Figure 5.3.** (A) Monthly whole live weight (WLW) of mussels at each site (mean ± SE g·mussel$^{-1}$, n = 12 samples per site) and (B) at each depth (mean ± SE g·mussel$^{-1}$, n = 24 samples per depth).

WLW was modelled from the shell length VB equations after applying a length-weight conversion ($WLW = aL^b$), where the constants a and b were derived from linear regression of ln (WLW) against ln (L). As mussels were not weighed individually, separate regressions of ln transformed sample mean WLWs against corresponding sample mean Ls were performed on data from each of the four sites. ANCOVA indicated no significant difference between regression slopes ($P > 0.05$; $F = 1.44$, df = 3 and 664) or intercepts ($P > 0.05$; $F = 1.67$, df = 3 and 667), so data were combined. The resulting regression using pooled data was highly significant, whereby ln (WLW) = - 8.401 (± 0.063) + 2.763 (± 0.015) ln(L) ($P < 0.0001$, $r^2 = 0.980$, n = 672 samples). The slope and transformed intercept from this regression can be used to model WLW growth from VB predicted shell lengths using either of the following conversions:

$$WLW_t\,(g) = 0.0002246 \cdot (\text{predicted VB-}L_t,\ mm)^{2.763}$$

or alternatively

$$WLW_t\,(g) = 55.71 \cdot (1 - e^{-0.16199\,(t + 5.76)})^{2.763}$$

where $W\infty$ (55.71 g) = $0.0002246(L\infty)^{2.763}$ and t is time in months relative to March 1995. This general WLW growth model provided a reasonable fit to observed mussel weight over the trial period, where $r^2 = 0.836$ (pooled WLW from all sites, n = 672

samples) and $r^2$ = 0.761, 0.824, 0.901 and 0.906 for sites I to IV, respectively (n = 168 samples·site$^{-1}$, Fig. 5.4).

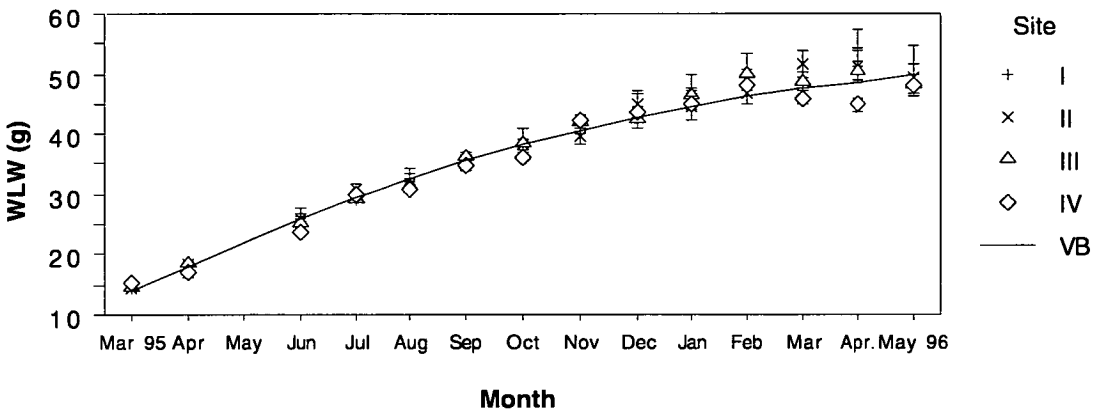


**Figure 5.4.** Observed monthly whole live weight at each site (WLW, mean ± SE g·mussel$^{-1}$, n = 12 samples per site per month) and general von Bertalanffy (VB) growth curve describing overall mussel growth $WLW_t$ (g) = 55.71 · (1 - e $^{-0.16199 (t + 5.76)}$) $^{2.763}$ ($r^2$ = 0.836, n = 672 samples).

### 5.3.3 Meat Weights

Meat weights (WMW, DMW and AFDMW) also displayed similar growth patterns among sites. Distinct peaks in meat weights were observed in August 1995 and February 1996 preceding significant tissue losses due to spawning (Fig. 5.5). Reduced meat growth was observed in July 1995 and December 1995. In March 1996, meat weights at sites III and IV were probably underestimates of true values. A delay in processing mussels from these two sites may have been responsible for some tissue loss through the utilisation of energy reserves while being temporarily held in a 500 l tank.

Wet meat weight (WMW) increased from an overall mean of 3.4 ± 0.1 g·mussel$^{-1}$ in March 1995 to 11.3 ± 0.2 g·mussel$^{-1}$ in August 1995 (n = 48 samples·month$^{-1}$). Following spawning during August, WMW fell slightly to 9.0 ± 0.2 g·mussel$^{-1}$ in September. WMW increased over the spring and summer period to 16.9 ± 0.4 g·mussel$^{-1}$ in February 1996. Due to a second spawning during February, WMW fell again to 14.3 ± 0.3 g before a slight recovery to 15.8 ± 0.3 g·mussel$^{-1}$ in May 1996. No significant differences in WMW between sites or depths were indicated for the key sampling dates March 1995, August 1995 or May 1996 ($P > 0.05$). In February 1996, no site differences were indicated but WMW of mussels grown at 5 m (17.8 ± 0.5 g·mussel$^{-1}$) was slightly greater than at 1 m (16.0 ± 0.6, n = 24 samples, $P < 0.05$). No significant site x depth interactions were indicated for any of the four key sampling dates.

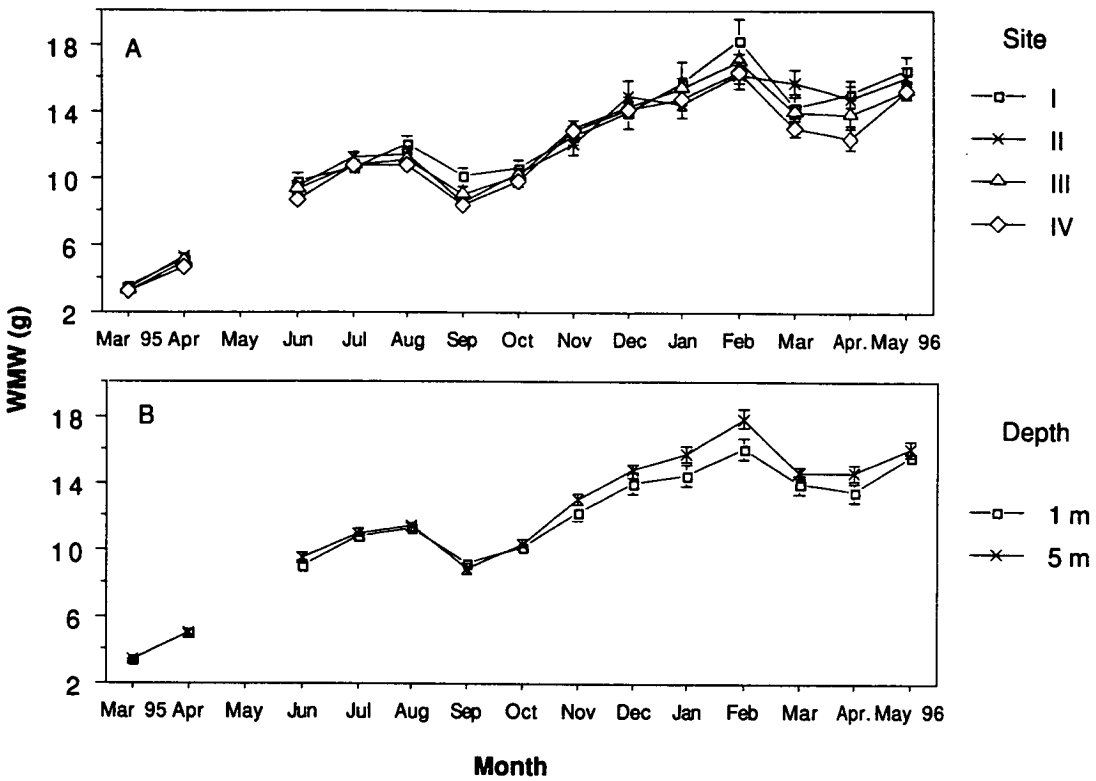


**Figure 5.5.** (A) Monthly wet meat weight (WMW) of mussels grown at at each site (mean ± SE g·mussel$^{-1}$, n = 12 samples per site) and (B) at each depth (mean ± SE g·mussel$^{-1}$, n = 24 samples per depth).

Overall mean ash-free dry meat weight (AFDMW) increased from 0.60 ± 0.01 g·mussel$^{-1}$ (n = 48 samples) in March 1995 to 1.97 ± 0.03 g·mussel$^{-1}$ in August, fell to 1.35 ± 0.04 g·mussel$^{-1}$ in September, increased again to 2.99 ± 0.10 g·mussel$^{-1}$ in February 1996 and finished at 2.07 ± 0.07 g·mussel$^{-1}$ in May 1996 (Fig. 5.6). Differences in AFDMW among sites were indicated for March 1995 ($P < 0.0001$; where $I^b = II^b > III^a = IV^a$, Fig. 5.7). Although site differences in AFDMW were significant in August 1995 ($P < 0.05$; where $I^b \geq II^{ab} = III^a = IV^a$) and February 1996 ($P < 0.05$; where $I^b > II^a = III^a = IV^a$), no differences in AFDMW increments for the periods March to August 1995 (mean increase 1.38 ± 0.03 g·mussel$^{-1}$) or September to February 1996 (mean increase 1.63 ± 0.09 g·mussel$^{-1}$) were significant among sites ($P > 0.05$). There were no differences in AFDMW among sites in May 1996 ($P > 0.05$). Differences between growing depths were significant only in February 1996 ($P < 0.05$) where AFDMW of mussels at 5 m (3.2 ± 0.1 g·mussel$^{-1}$) was greater than at 1 m (2.8 ± 0.1 g·mussel$^{-1}$). No significant site x depth interactions were indicated on any of the key sampling dates.

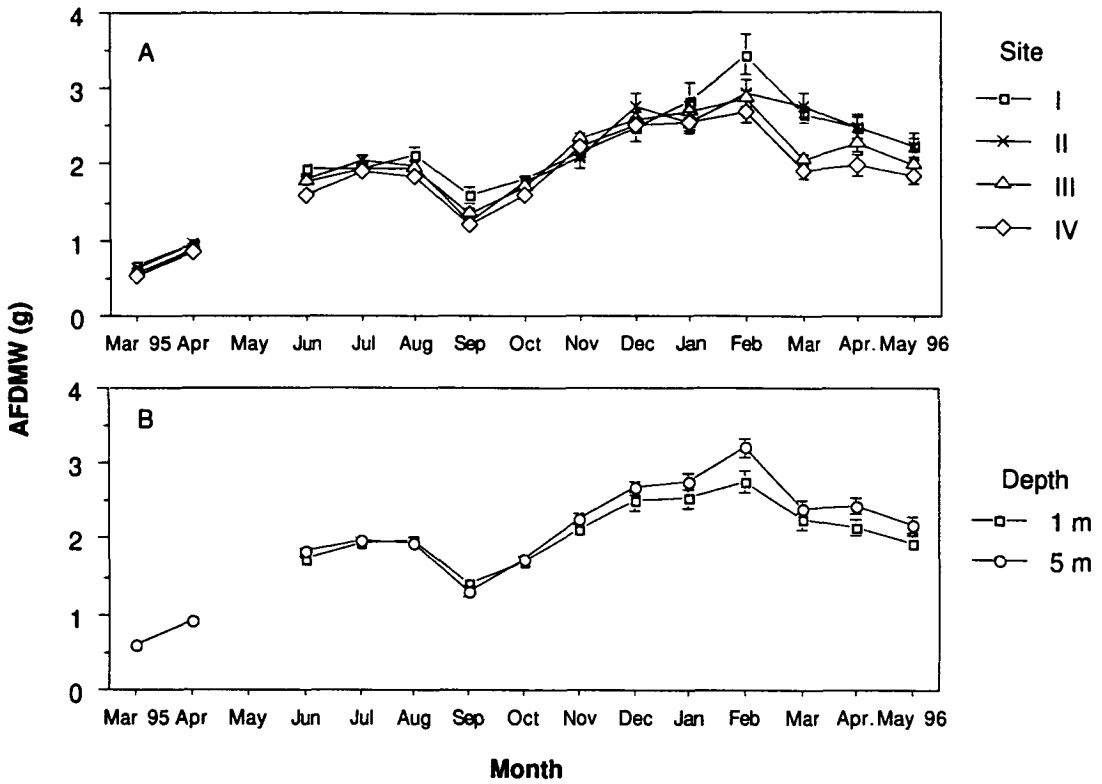


**Figure 5.6.** (A) Monthly ash-free dry meat weight (AFDMW) of mussels grown at each site (mean ± SE g·mussel$^{-1}$, n = 12 samples per site) and (B) at each depth (mean ± SE g·mussel$^{-1}$, n = 24 samples per depth).

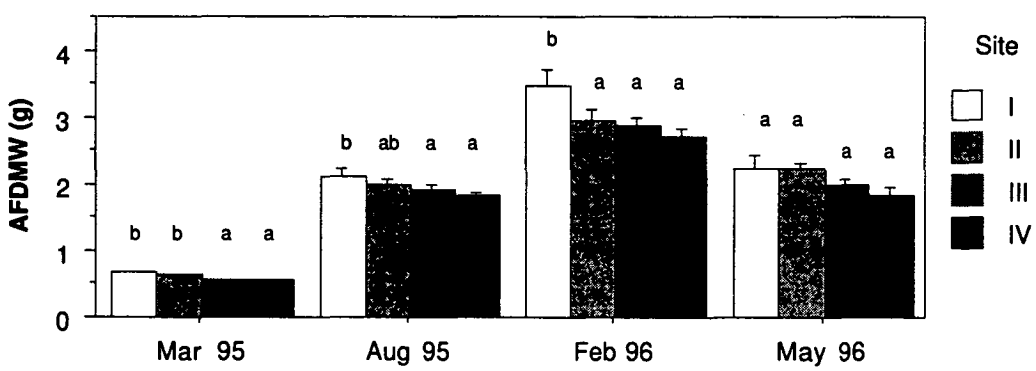


**Figure 5.7.** Ash-free dry meat weight (AFDMW) of mussels at each of the four key sampling months (mean ± SE g·mussel$^{-1}$, n = 12 samples per site). Significant differences among sites were indicated in March 1995, August 1995 and February 1996 ($P < 0.05$). Site means not sharing a common letter are significantly different ($P < 0.05$).

### 5.3.4 Individual Mussel Biomass and Production

AFDMW comprised the major portion of the total organic content, ranging from 73.5% to 83.4% of the total individual biomass, depending on mussel condition. As such, the annual cycle for IB was very similar to that of AFDMW; distinct peaks in biomass were displayed prior to spawnings in August 1995 (mean 2.39 ± 0.04 g AFDW·mussel$^{-1}$) and February 1996 (mean 3.60 ± 0.11 g AFDW·mussel$^{-1}$, n = 48 samples), each followed by declines due to the loss of reproductive tissue (Fig. 5.8). As with AFDMW, significant differences in IB among sites were indicated for the March 1995 ($P < 0.01$), August 1995 ($P < 0.05$) and February 1996 ($P < 0.05$) samples but not for the final sample in May 1996 ($P > 0.05$). A slight difference in IB between depths was also indicated for the February 1996 sample ($P > 0.05$), where IB at 1 m (3.4 ± 0.2, n = 24 samples) was less than at 5 m (3.9 ± 0.1, n = 24 samples).

As initial IB differed among sites, an analysis of biomass production during the course of the trial was deemed appropriate in order to assess the growth performance of mussels at the four sites. Monthly biomass increments and/or losses at each site are illustrated in Fig. 5.9-A. The cumulative sum of all biomass increments (gross production, GP) and biomass losses (eliminated biomass, EB) are illustrated in Fig. 5.9B. The difference in cumulative GP and EB is the biological production, or net production (NP), retained within mussels on any given date during the trial period (Fig. 5.9-C).

Monthly biomass increments averaged 0.43 g·mussel$^{-1}$ across all sites during the initial stages of the trial (March - May 1995), declined in the early winter (June 1995 mean 0.24 g·mussel$^{-1}$) and were negligible in July 1995 (overall mean 0.02 g·mussel$^{-1}$, Fig. 5.9-A). In July 1995, biomass losses at sites II, III and IV contrasted with an increase in biomass at site I. Negative production at all sites during August was likely due to spawning losses. Production quickly resumed post-spawning through the spring and early summer (mean monthly production from September 1995 through January 1996 was 0.36 g·mussel$^{-1}$) although production in December 1995 was somewhat reduced (mean 0.10 g·mussel$^{-1}$). The decline in overall mean production observed in December was primarily due to the apparent biomass loss at site II. Production was negative at all sites in February 1996, due to a second major spawning, and continued to be negative or negligible through the final months of the trial. Biomass losses during March 1996 and April 1996 may be attributed to continued spawning, starvation and subsequent mobilisation of energy reserves, and/or mortalities of larger mussels.

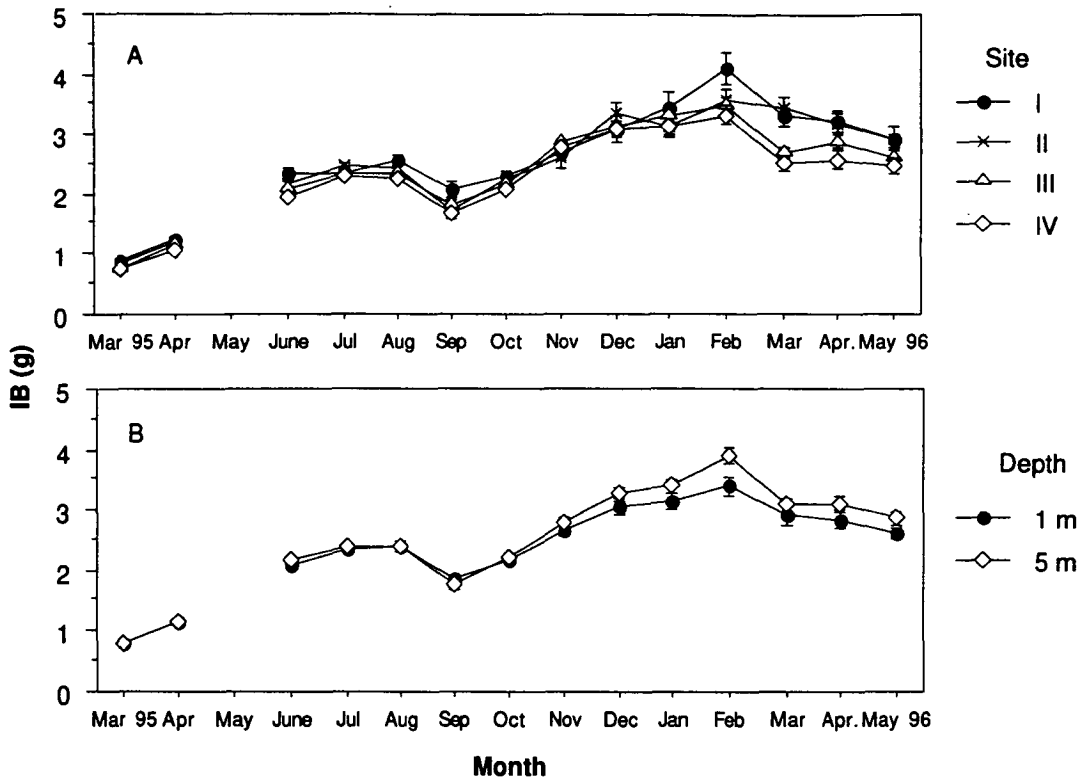


**Figure 5.8.** (A) Monthly individual biomass (IB) of mussels grown at each site (mean ± SE g·mussel$^{-1}$, n = 12 samples per site) and (B) at each depth (n =24 samples per depth).

At the conclusion of the trial, overall mean gross production was 3.60 ± 0.14 g·mussel$^{-1}$, total eliminated biomass was -1.68 ± 0.07 g·mussel$^{-1}$ and net retained production was 1.93 ± 0.08 g·mussel$^{-1}$ (n = 8 means, ranges presented in Table 5.3). No significant differences among sites for GP, EB or NP were indicated on any of the key sampling dates (August 1995, February or May 1996). Final net biological production retained by mussels at the conclusion of the trial represented only 54% of the total trial biomass production. Fourty-six percent of total production was lost primarily as gametes and possibly some metabolic losses.

**Table 5.3.** Gross production (GP), eliminated biomass (EB) and net retained production (NP) of mussels grown at each depth (1 m and 5 m) within each site (I - IV) from March 1995 to May 1996 (units as g ash-free weight·mussel$^{-1}$).

| Site | Depth | GP | EB | NP |
|---|---|---|---|---|
| I | 1 | 3.61 | -1.64 | 1.97 |
| I | 5 | 3.84 | -1.71 | 2.14 |
| II | 1 | 3.84 | -1.90 | 1.94 |
| II | 5 | 3.89 | -1.69 | 2.21 |
| III | 1 | 3.25 | -1.43 | 1.82 |
| III | 5 | 3.76 | -1.85 | 1.91 |
| IV | 1 | 2.80 | -1.33 | 1.48 |
| IV | 5 | 3.82 | -1.87 | 1.95 |

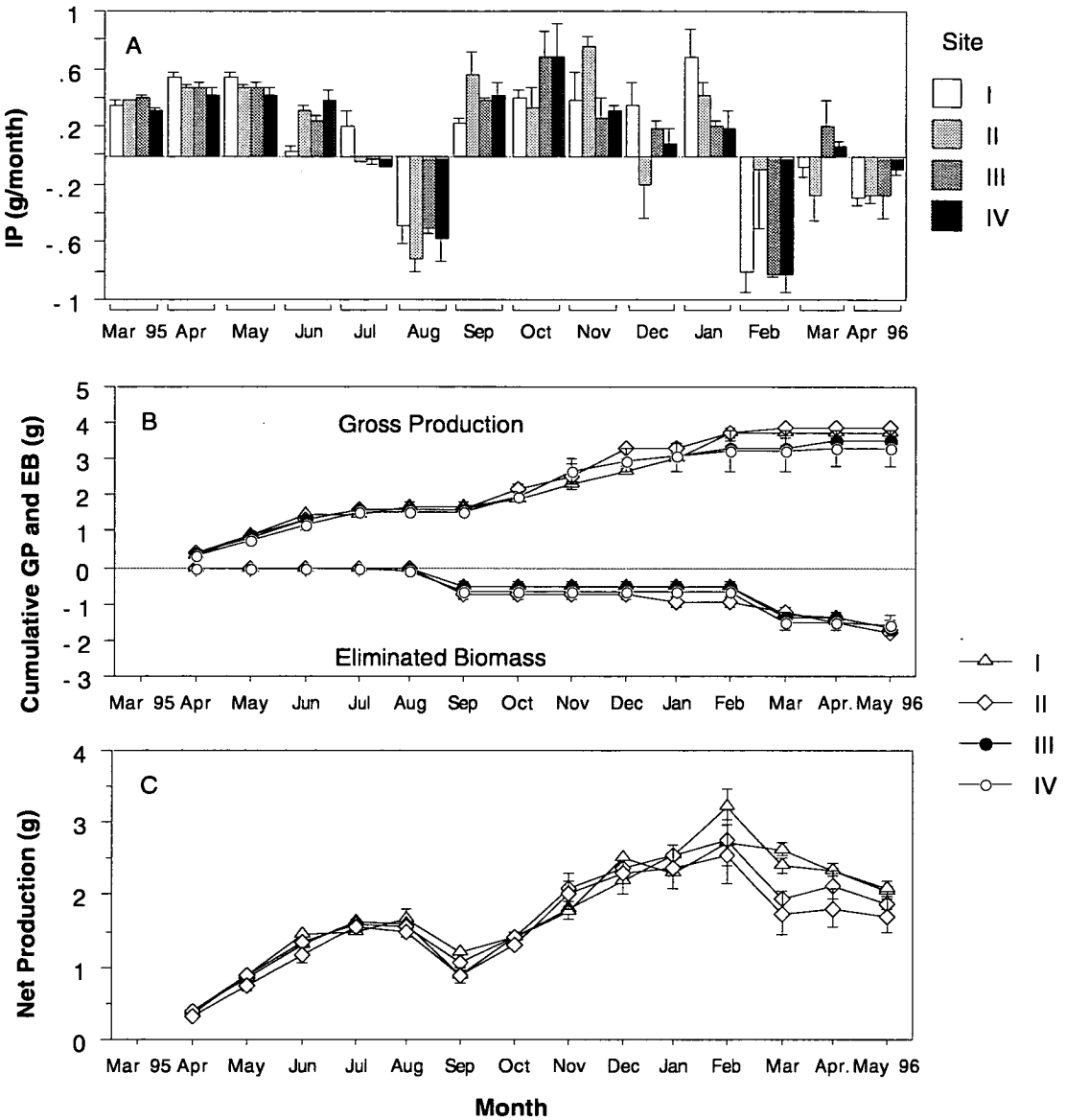


**Figure 5.9.** (A) Monthly biomass gains or losses at each site (mean ± SE g AFDW·mussel$^{-1}$). (B) Cumulative totals of gross production (GP) and eliminated biomass (EB) at the beginning of each month. (C) Net production (NP) at the beginning of each month (mean ± SE g AFDW·mussel$^{-1}$). n = 2 replicates per month at each site.

### 5.3.5 Influence of environmental factors on growth

A step-wise multiple regression analysis (F-to-enter 4.0, F-to-remove 3.99, Statview 4.5) was used to evaluate variables that may have influenced monthly absolute shell length growth rates (AGR, mm·month$^{-1}$ = monthly mean site length - mean site length previous month). These variables included mussel size (mean site shell length at the beginning of each month) and several environmental parameters (monthly mean temperature, salinity, dissolved oxygen, algal POM, non-algal POM, PIM and TPM). All environmental parameters were eliminated, with or without incorporating data from the final three months when AGRs were mostly negative. Shell length was the sole

independent variable influencing AGR. Disregarding the final three months, the relationship between growth and size is described by:

AGR ($mm \cdot month^{-1}$) = 11.55 (± 1.54) - 0.124 (± 0.021) L ($P < 0.0001$, $r^2 = 0.486$).

This suggests that shell length growth was independent of environmental factors and supports the continuous, albeit decreasing growth rates with increasing size or age (Fig. 5.10). Similarly, excluding the periods of mass spawning in August 1995 and February 1996, step-wise multiple regression also eliminated all environmental parameters that may have influenced monthly meat and biomass production.

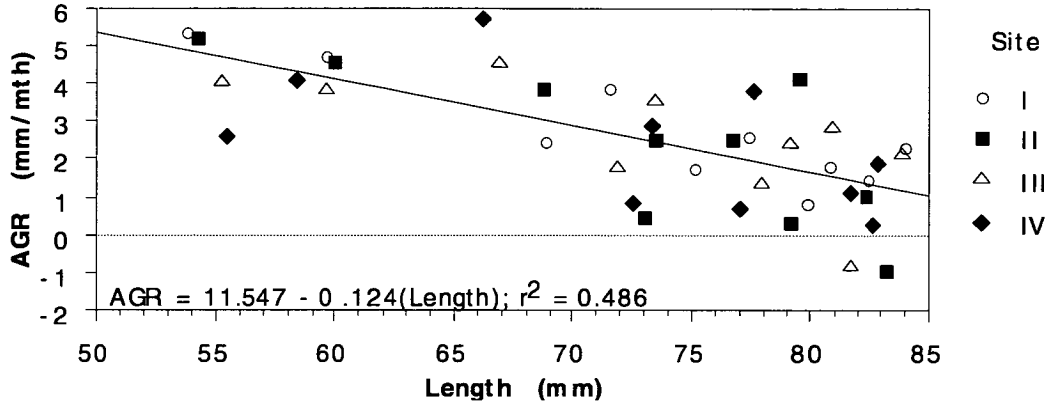


**Figure 5.10.** Regression plot of mean monthly absolute growth rate (AGR, $mm \cdot month^{-1}$) against mean shell length at the beginning of each month (mm) ($P < 0.0001$, n = 40 observations). Regression excludes growth rate data from February, March and April 1996 when AGRs were primarily negative.

## 5.4 Discussion

From the analyses of various mussel growth parameters during 1995-96 in North West Bay, several outcomes are apparent. Most notably, there were no major differences in mussel growth (eg. shell length, live weight, meat weights, total biomass, net individual production) among the four longline sites. Occasionally, statistically significant differences for some growth parameters were indicated. However, these differences were minor (eg. final mean site shell lengths and $L\infty$ were within 2.1 mm and 2.5 mm, respectively) and not considered biologically nor commercially significant. Therefore, it appears that the salmon farm had little or no influence on the growth performance of mussels cultured within the confines of the farm lease. Secondly, no effect of depth was apparent, whereby mussels at 1 m displayed similar growth to mussels grown at 5 m. Thirdly, no obvious seasonal variation in growth, such as extended periods of slow autumn-winter growth or increased growth during the spring-summer was observed. Fourthly, there appeared to be no significant correlations between environmental parameters and mussel growth. Lastly, mussel growth effectively ceased after February 1996.

### 5.4.1 Effect of site

The primary objective of this study was to compare the growth performance of mussels cultured at the Aquatas salmon farm with that of mussels grown at control sites. The concept of integrating suspended bivalve culture with cage culture of fin fish is intuitively promising. In theory, particulate wastes and localised plankton blooms are potential food items generated by the fish farm that may be utilised by cultured bivalves. The benefits of such integration might include increased bivalve growth rates and reduced environmental impacts through reductions in fish farm waste loadings.

Despite the possible advantages derived from integrating shellfish with salmon cultivation, no major differences in mussel growth were observed among the four test sites. As mussel growth on the fish farm was not enhanced, it follows that these mussels did not obtain any nutritional advantage, by way of farm waste products (fish feed, faeces, increased phytoplankton abundance), over mussels grown elsewhere in North West Bay. The question of most interest is "why not?"

It could be argued that an assessment of the Aquatas fish farm's influence on mussel growth might have been disadvantaged by the absence of fish for approximately three and a half months. Therefore, a significant portion of the early trial period (June to mid-September 1995) was devoid of supplementary feed, as particulate farm wastes or enhanced plankton production resulting from dissolved nutrient wastes. However, winter fallowing was standard practice at the Aquatas farm and, therefore, conferred conditions that would normally be experienced if mussel cultivation were introduced at this site. Despite the break in fish production, there were no significant differences in growth among the four mussel sites prior to the fallowing period; the period when fish biomass and associated waste production were at their highest levels and temperatures were favourable for growth. Further, if mussel growth at the farm sites was influenced by additional fish wastes, a noticeable reduction in growth rates might be expected during the fallowing period, followed by an increase after the re-introduction and subsequent grow-out of the new smolt year-class (September 1995 - May 1996). Mussel growth at either of the fish farm sites (I and II) did not display such a pattern.

Previous studies of integrated bivalve and fish cultivation have had mixed results (Table 5.4). In some cases, bivalves have grown much better adjacent to salmon cages (Wallace, 1980; Jones and Iwama, 1991). However, there are also reports showing no, or only minor, increase in growth from such co-cultivation (Taylor et al., 1992; Okumus, 1993; Gryska et al., 1996; this study).

**Table 5.4**. Summary of studies investigating integrated bivalve-salmon culture. (* indicates enhanced bivalve performance and/or available food levels at fish farm sites relative to non-fish farm sites).

| **Study**<br>**Species**<br>**Location**<br>**Culture Method** | **Culture Distances from Fish Cages** | **Bivalve Parameters Monitored** | **Comments** | **Food Parameters Monitored** | **Comments** |
|---|---|---|---|---|---|
| Wallace (1980)<br>*Mytilus edulis*<br>Norway<br>Fish-cage rings | Fish cage rings<br>Natural populations growing intertidally, subtidally and on mooring buoys/ropes | Shell growth* | Mussels growing on fishcage rings displayed greater growth during winter only | | Food levels assumed to be greater at fish farms due to waste feed and faeces |
| Jones and Iwama (1991)<br>British Columbia, Canada<br>*Crassostrea gigas*<br>Lantern nets | Station#/distance<br>1. 2 m (outside cages)<br>2.3.4. 0 m (inside cages)<br>5. 200 m<br>6. 500 m<br>Controls. 4 km and 6 km | Shell Growth*<br>Condition* | Oyster growth and condition was significantly greater inside fishcages.<br>Growth and condition at Stn 1=5=6.<br>Growth and condition at control sites was significantly less than all other sites associated with the fish farm. | POM*<br>Chlorophyll* | Although overall mean POM and chlorophyll of Stn. 1-6 was greater than Controls, mean POM and chlorophyll at Stn. 1 was similar to Controls. |
| Taylor et al. (1992)<br>British Columbia, Canada<br>*Mytilus edulis*<br>Grown on coiled rope inside mesh cylinders | Two farms (DB and GB)<br>1. 3 m<br>2. 15 m<br>3. 75 m<br>Controls (600 m, 800 m) | Shell Growth<br>Condition*<br>Glycogen*<br>Crude protein<br>Survival | <br>GB (ns). DB (Control<3m<15m=75m)<br>3 m > Controls | Seston<br><br>Chlorophyll | <br><br>DB (3m<15m<75m=control)<br>GB (3m=75m<15m=control) |
| Okumus (1993); Stirling and Okumus (1995)<br>Scotland<br>*Mytilus edulis*<br>Suspended socks (Trial 1)<br>Lantern nets (Trial 2) | 1. Suspended from walkways at 2 salmon farms in Loch Etive (ES1, ES2) and 1 salmon farm in Loch Leven, (LS).<br>2. One mussel farm in each loch (EM, LM).<br>EM-ES1 (2 km)<br>LM-LS (3 km) | Shell Growth*<br>WLW*<br>Meat Growth*<br>Condition<br>Glycogen<br>Net Production*<br>Survival | Minor enhanced growth associated with salmon farms only in Trial 2 and appears restricted to winter months.<br>Not tested<br>Not tested<br>ES1>EM>ES2 (Trial 1)<br>Higher mortality at fish farm sites (Trial 1) | Seston*<br>POM*<br>%POM<br>Non-algal POM *<br>Chlorophyll | In Loch Etive only.<br>In Loch Etive only.<br><br>In Loch Etive only. |
| Gryska et al., (1996)<br>*Placopecten magellanicus*<br>Maine USA<br>Pearl nets | unspecified | Shell growth<br>Adductor muscle | Growth similar to scallops grown commercially | | |
| This study (2000)<br>Tasmania, Australia<br>*Mytilus planulatus*<br>Socks suspended from longlines | I. 70 m<br>II. 100 m<br>III. 500 m<br>IV. 1200 m | Shell Growth*<br>Meat Growth<br>WLW<br>Condition*<br>Glycogen<br>Gonad stages<br>Stable isotopes<br>Survival | Final shell lengths within 2 mm<br><br>Final condition within 17‰ | Seston<br>POM<br>%POM<br>non-algal POM<br>chlorophyll | Overall mean seston, POM and non-algal POM concentrations were similar among mussel sites. These concentrations were higher at the centre of the fish farm than at the other four sites during both fish culture and fallowing periods. |

These differences in the effect of salmon-cage culture on bivalve growth may be due to a number of factors including fish biomass, feeding practices, feed type and digestibility, cultivation designs and ambient environmental conditions.

Firstly, upon examination of these and other studies investigating the effects of salmon farming on the environment, food availability within farm sites is not always enhanced. Additional food would be in the form of increased phytoplankton production and/or suspended POM in excess fish feed and faeces. Enhanced phytoplankton production has, at times, been associated with fish farms in some studies (eg. Mäkinenn and Pursianien, 1987 cited in Gowen et al., 1988; Aure et al., 1988; Jones and Iwama, 1991; Pridmore and Rutherford, 1992). Increased primary production adjacent to fish farms is held to be a consequence of nutrient enrichment, particularly from excreted ammonia (Gowen et al., 1988). However, higher phytoplankton (chlorophyll-a) levels are not always associated with fish farms (Gowen et al., 1988; Taylor et al., 1992; Okumus, 1993; Wildish et al., 1993; this study). Therefore, an increased and/or higher quality food supply, by way of increased phytoplankton production, is not necessarily available to fish farm mussels. The lack of additional phytoplankton may be due to a number of factors including: ambient nutrient levels are not limiting for phytoplankton growth production; low levels, or high dilution, of dissolved nutrients are not sufficient to stimulate phytoplankton growth; and phytoplankton growth is stimulated, but transported away from the farm, consumed by zooplankton, and/or diluted to undetectable levels.

Also, increased concentrations of suspended POM, which would include excess feed and faecal particles, are not always associated with fish farms (Taylor et al., 1992; this study). Of those that have demonstrated or implied that POM levels are enhanced within fish farms (Wallace, 1980; Jones and Iwama, 1991; Okumus, 1993), no information is presented on the feed, stocking density or husbandry practices of the experimental fish farms. Increased POM levels reported in two of these studies, conducted a considerable number of years ago, may be partially due to a number of unknown factors such as high fish biomass and feeding rates, poor feeding practices, high percentage of feed fines, highly soluble feed pellets, low current flow and/or poor feed digestibility with associated high level of faecal output. Where POM levels are not enhanced, may be due to converse circumstances (ie. low fish biomass and feeding rates, water-stable pellets, highly digestible feed, etc.). Certainly, the low fish biomass (2.5 – 3.7 tonnes salmon) and feed supplied (560 – 840 $kg \cdot month^{-1}$) in the study by Taylor et al. (1992), is not indicative of most fish farms. However, it is becoming more evident that particulate feed and faecal wastes, although considerable, may become highly diluted in the large volume of water flowing through the farm (Troell and Norberg, 1998). In the present study, particulate waste loadings may have

only contributed up to 0.03 $mg \cdot l^{-1}$ and 0.05 $mg \cdot l^{-1}$ of ambient POM and total SPM concentrations, respectively (calculated in Section 13). Assuming these calculations are reasonable, waste particulates would contribute little to the background concentrations within the fish farm (maximum of 2%).

The location of bivalve culture, relative to the fishcages, is also important. Certainly the Taylor et al. (1992) study was compromised by mussels at one farm not located in the direction of current flowing through the fishcages. Those studies that have shown a significant increase in bivalve growth cultured their animals directly next to, or inside of, fish cages (Wallace, 1980; Jones and Iwama, 1991; Okumus, 1993). In the present study mussels may have been located too far from the fishcages. It is possible that the majority of particulate wastes settled to the sea bottom before being horizontally transported to the mussel lines at sites I and II, 70 m and 100 m distant (see Section 13 for calculations used in modelling the distribution of solid wastes). However, the settling velocity of faecal material (4.0 $cm \cdot sec^{-1}$), used in calculating the horizontal transport of waste particles, is based on faecal pellets rather than discrete particles; salmon excretia is commonly voided as an 'explosion' of particles. The settling velocity of these particles would be considerably lower than intact faecal pellets and vary according to particle size and density. No investigations that identify the sinking velocities of these faecal particles have been reported to date. However, using the faecal settling velocities from a range of marine invertebrates as a guide (Robison and Bailey, 1981 and references within), it is conceivable that particulate salmon faeces might remain suspended for considerable distances, at least to the mussel longlines at site I and II, before descending below the depth of mussel socks.

Of particular interest from the studies showing increased bivalve growth, is the influence of ambient environmental conditions has on determining the potential for integrated culture to enhance bivalve growth. Although influenced by a variety of environmental factors (eg. temperature, salinity, light, current speed), the availability of organic food particles is considered the single most important factor determining growth rate of mussels (Seed and Suchanek, 1992). As such, the existence of both temporal and spatial variations in ambient food availability might partly explain the degree of success for integrated fish-bivalve cultivation.

In the studies by Wallace (1980) and Okumus (1993) growth of integrated mussels were enhanced only during the winter period when natural food levels were low; growth rates of fishfarm mussels and non-fishfarm mussels were similar in summer. Okumus (1993) also suggested significant differences in live weight and meat weights between test groups were a consequence of mussels grown on salmon farms not depleting their energy reserves over the winter period as much as mussels grown at

control sites. On reviewing the Jones and Iwama (1991) study, Troell et al. (1999) note that POM concentrations were under the threshold for pseudofaeces production 62% of the time at control sites compared with only 25% of the time inside the cages. Therefore, not only were POM concentrations higher at the fish farm but oysters would have been feeding at maximum capacity over twice as long as oysters at the control sites. It appears that only when ambient food levels are below the pseudofaeces threshold and particulate wastes significantly increase POM concentrations is growth of bivalves enhanced by integration with salmon farms.

Although the present study failed to demonstrate that particulate food levels were enhanced within the fish farm, it is possible that the sampling regime, where single water samples were collected at each site at approximately 0900 and 1200 h, was not comprehensive enough to identify peaks in faecal output. As sampling time was not replicated at each site, any significant site x time interaction in particulate concentrations cannot be tested. Increased particulate levels in the water column have been associated with feeding times (unpublished study, M. Troell, personal communication). Taylor et al. (1992) demonstrated that seston concentrations within a fish farm may vary considerably, up to 8-fold (0.5 – 4.0 mg seston·$l^{-1}$), during a 24 h period; this variation attributed to faecal output increasing from minimum values at 0400 to maximum values in the afternoon 1200 – 1800 h. For the sake of argument, let's assume that particulate waste output from the Aquatas farm was considerable, SPM and POM concentrations were substantially increased, possibly in distinct organic-rich pulses, and this waste material was transported to mussels at sites I and II. Why might mussels at these two sites not display greater growth relative to mussels at the two remote sites? It appears likely that the ambient particulate levels may have saturated the mussels' feeding capacity. Except for six weeks, from mid-November through December 1995, the measured seston concentrations within the farm site, and NWB generally, consistently exceeded the size-dependent threshold level for pseudofaeces production. The pseudofaeces threshold concentration for 55-80 mm mussels ranges from 5.1 to 5.44 mg·$l^{-1}$ (Widdows et al., 1979); mean monthly seston concentrations within NWB, excluding the six week period from mid-November through December 1995, ranged from 5.6 to 8.1 mg·$l^{-1}$. The mean seston concentration during November-December 1995 (4.0 mg·$l^{-1}$) was not far below the pseudofaeces threshold level for mussels during the same period (5.36 mg·$l^{-1}$). Therefore, ingestion rates at all four sites were probably at maximum levels throughout most of the year. Any additional fish waste particulates at sites I and II were unlikely to contribute much to the mussels' diet - their feeding capacity was already saturated by natural seston and extra food filtered would be voided as pseudofaeces. Troell and Norberg (1998) demonstrated the influence of ambient particulate levels on the ability

of bivalves to ingest additional food. A similar model, based on the ambient particulate concentrations observed in NWB, is presented in Appendix 5.

Therefore, several factors were responsible for the lack of enhanced growth of integrated mussels at the Aquatas farm. Localised phytoplankton production was not stimulated on the farm site. Waste feed particles and faeces may have been too low and/or too diluted to significantly enhance POM concentrations. Mussels were cultured too far from the fish cages. Ambient seston concentrations saturated the mussels' feeding capacity, hindering the ingestion of any additional food, even if it was available.

### 5.4.2 Effect of depth on growth

Mussels located at the top section of droppers (1 m depth) did not exhibit significantly different growth rates to mussels at the bottom (5 m depth). The effect of depth on mussel growth rates appears to be highly site-specific. Some studies have reported a general decrease in growth rate with increasing depth (Kautsky, 1982; Loo and Rosenberg, 1983; Rodhouse et al., 1984a) while others have reported the converse (Okumus, 1993). Page and Hubbard (1987) found mussel growth was higher at intermediate levels (9 m) than at shallower (2 m) and deeper depths (18 m). Some studies have found little or no differences in growth associated with depth (Mason and Drinkwater, 1981; Mueller, 1996; Sara et al., 1998).

The relationship between growth rates and water depth is generally thought to reflect variations in food availability within the water column (Seed and Suchanek, 1992). However, the effect of lower or fluctuating surface water salinities (Okumus, 1993) and lower temperatures with increasing depth (Kautsky, 1982) might also be implicated. The difficulty with correlating single environmental parameters with growth rates is that the correlation, although significant, need not be causal. Also, many environmental factors co-vary and isolating single environmental parameters is not always appropriate. As the above studies have not shown consistent growth trends according to depth, the factors influencing growth at various depths appear to be site-specific.

In the present study, the similarities in growth at 1 m and 5 m reflect similarities in environmental conditions. There was little difference in temperature and dissolved oxygen content between the two depths. Salinities were fairly stable for most of the trial period (32 to 35‰), although significant reductions at the surface layer were apparent on a few occasions following significant rainfall events. However, on only

one occasion did these surface salinities drop below 20‰, the suggested threshold salinity at which mussel growth might be affected (Shumway, 1977; Almada-Villela, 1984; Gruffydd et al., 1984), but this event was probably shortlived. Salinity fluctuations, generally low in amplitude and duration, were unlikely to have significantly influenced mussel growth.

As water samples were integrated whole water column samples, it is unknown whether particulate food or chlorophyll concentrations varied between growing depths. As observed mussel growth did not vary with depth, it seems unlikely that food availability was significantly different between 1 m and 5 m. However, as noted by Sara et al. (1998), differences in food concentration and/or food quality between growing depths may not always be manifested by different mussel growth rates, as mussels might adapt their feeding strategies according to the available food source.

### 5.4.3 Effect of Season and Environmental Parameters

No obvious seasonal growth variation in shell length or whole live weight was observed in this study, nor was growth significantly correlated with any measured environmental parameters. Shell growth was continuous, albeit at decreasing rates, from March 1995 to February 1996. After February 1996, no further growth was evident. The absence of seasonal variation contrasts with numerous field studies in temperate and boreal waters reporting rapid bivalve shell growth during spring-summer and little or no growth during winter (Baird, 1966; Seed, 1969b; Dare and Davies, 1975; Dare, 1976; Wallace, 1980; Mason and Drinkwater, 1981; Hilbish, 1986; Wilson, 1987; Brown and Hartwick, 1988; Kautsky et al., 1990; Roberts et al., 1991; Sukhotin and Kulakowski, 1992; Okumus, 1993; Sukhotin and Maximovich, 1994). Seasonal variation in growth rates has been attributed to temporal variation in environmental parameters, particularly temperature, salinity and food supply (Seed and Suchanek, 1992).

Significant correlations of mussel growth with temperature or degree-days has been demonstrated in some studies (Kautsky, 1982a; Wilson, 1987; Nielsen, 1988; Okumus, 1993). However, water temperature may strongly co-vary with phytoplankton abundance, making it difficult to generalise about the importance of temperature alone (Kautsky, 1982a). The effect of temperature on growth rates seems to be marginalised when food is abundant. Loo and Rosenberg (1983) found that low temperatures (< 5°C) did not seem to limit growth whenever these coincided with the spring phytoplankton bloom. Wallace (1980) found that mussels maintained summer

growth rates throughout the winter despite temperatures approaching 0°C. This winter growth was attributed to the availability of food provided by salmon farms.

Physiological studies of *Mytilus edulis* demonstrate that ambient sea water temperatures ranging from 10 - 20°C have little effect on "scope for growth" (Bayne et al., 1976b), confirming early investigations establishing this as the optimum temperature range for *Mytilus edulis* (Coulthart, 1929 cited in Seed 1976). This optimum temperature range is supported by the present study, where multiple regression and correlation analyses suggest water temperatures (range 9°- 18°C) had no significant effect on shell length growth rates. Several other mussel studies in Australia (Wilson and Hodgkin, 1967; MacIntyre et al., 1977; Dix, 1983) and California (Page and Hubbard, 1987; Page and Ricard, 1990) similarly reported a lack of seasonal growth or significant correlations with temperature. These results suggest that temperature has virtually no effect as a growth regulator over the temperature range, generally within 10°- 20°C, normally experienced by these mussel populations.

As previously discussed, salinity was relatively stable over the trial period. Although some minor fluctuations were observed, these were not considered to be extreme nor of significant duration to have affected mussel growth. This is supported by monthly mean salinities not being significantly correlated with monthly growth rates.

Although moderated by a number of environmental factors, food is probably the most important single influence on mussel growth (Seed and Suchanek, 1992). Many studies have reported a significant influence of food supply on bivalve growth rates (Kautsky, 1982a; Rosenberg and Loo, 1983; Brown and Hartwick, 1988; Okumus; 1993). However, multiple regression and correlation analysis suggest food supply (as measured by algal and non-algal POM concentrations) was not a significant factor influencing growth rates in this study. This observation is most likely due to the morphological and physiological restrictions on the experimental mussels' capacity to pump, filter, ingest and digest particles from the water column. Increased food availability does not necessarily lead to increased growth if an individual has a limited feeding and digestive capacity. As previously discussed, seston concentrations were above the pseudofaeces threshold (T) for most of the study period (ie. $> 5$ mg·l$^{-1}$), indicating that mussels were probably ingesting particles at maximum rates for most of the trial. At seston concentrations above T, particles are rejected as pseudofaeces (Foster-Smith, 1975). Based on physiological relationships derived from the literature and monitored environmental parameters during the study period, food particle ingestion and digestion rates were calculated (Section 12 - Scope for Growth). Despite fluctuating seston concentrations, particle filtration, ingestion rates and POM absorption rates were likely restricted to a more limited range (Fig. 5.11). Where

particulate concentrations exceed T, growth becomes a function of the energy content gained from POM actually absorbed balanced against energy losses (eg. costs of maintenance, filtration, digestion and absorption). Therefore, energy for growth is more regulated by food quality (eg. %POM or digestibility) rather than the quantity of seston when seston concentrations exceed T (Bayne and Widdows, 1978; Widdows et al., 1979). This conclusion is supported by Page and Ricard (1990) who reported growth and food supply were correlated only during periods when seston concentrations were below T.

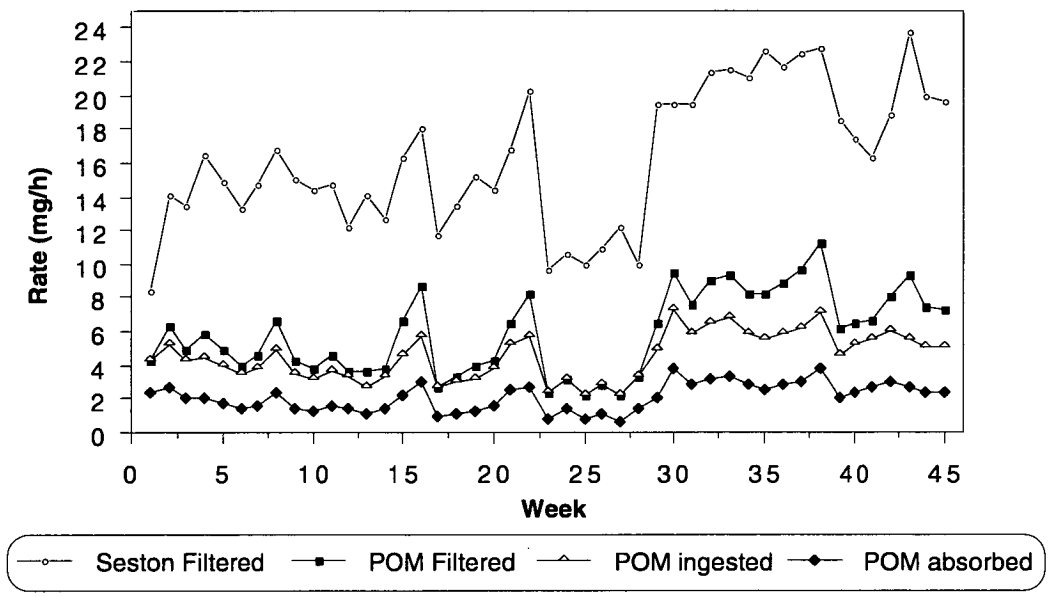


**Figure 5.11.** Estimated seston filtration and ingestion rates (mg particulate matter·h$^{-1}$) and ingestion and absorption rates of particulate organic matter (mg POM·h$^{-1}$) by mussels grown at four sites in North West Bay from June 1995 to May 1996. Rates based on physiological relationships in Page and Ricard (1990), mean weekly mussel DMW and environmental parameters (mean temperature, seston concentration and %POM) monitored at the four test sites (see Section 12.2 for details).

Ambient POM concentrations were adequate to sustain continuous growth throughout the trial. This is reflected by positive SFG estimates for most of the trial, except for approximately six weeks in late spring-early summer (November-December 1995), when SFG approximated 0.0 J·h$^{-1}$. Where mussel growth is reduced during periods of low temperatures and/or food concentrations, typically during winter months, reported SFG values are either low or negative (Bayne and Widdows, 1978; Thompson, 1984a; Deslous-Paoli et al., 1990; Page and Ricard, 1990). In contrast, although calculated SFG during November-December 1995 was negligible, shell growth continued. This could be due to erroneous SFG estimates that were based on literature values. Alternatively, mussels have the ability to buffer their shell growth during short term temporal variations in food availability by utilising glycogen reserves accumulated prior to, and during, gametogenesis (Bayne et al., 1983). Therefore shell

growth may continue, even during periods of submaintenance rations, despite the net loss of soft tissues (Hawkins and Bayne, 1992). This possibility is supported by a reduction in glycogen reserves observed from November 1995 to January 1996 (Fig. 6.4) and a reduction in meat growth in December 1995 (Fig. 5.6). However, this period also preceded the February 1996 spawning and whether glycogen reserves were being depleted due to submaintenance food concentrations, fuelling gametogenesis, or both, is speculative.

Although it has been suggested that, at seston concentrations above T, energy for growth is more regulated by food quality (eg. %POM) than quantity (Bayne and Widdows, 1978; Widdows et al., 1979), no such correlation between monthly length growth and %POM was significant in this study. Correlations between growth and food availability, or quality, may not be significant due to the ability of mussels to buffer growth, as previously mentioned. Also, %POM is not always a reasonable indicator of food quality; POM is a mixture of various substrates and a large proportion may be refractory and/or low in energy content (Widdows et al., 1979; Tenore, 1981; Tenore et al., 1982; Valiela, 1984; Grant and Bacher, 1998 and references within). Detrital particles, for example, have been shown, in many cases, to be a poor quality food source for bivalves (Winter, 1974; Williams, 1981; Stephenson et al., 1986; Langdon and Newell, 1990; Deslous-Paoli, 1990; Ruckelshaus et al., 1993; Yelenik et al., 1996, Duggins and Eckman, 1997). Therefore, alternative measures of particulate food quality have included the sum of carbohydrates, proteins and lipids in the seston (Widdows et al., 1979) and, perhaps most appropriate, the digestibility of the food material (Cheng et al., 1993 cited in Grant and Bacher, 1998). The nutritional value of fish feed and faeces to mussels, certainly an area of interest regarding the potential benefits of integrated mussel-salmon culture, has yet to be investigated.

The absence of significant correlations between growth and environmental factors may also have been partially due to the reproductive status of mussels. Page and Hubbard (1987) reported no correlation between growth of mussels over 50 mm and food supply; this was attributed to an increasing proportion of ingested food energy being allocated to gonad maturation. The physiological status of mussels undergoing gametogenesis and spawning is also somewhat different from the "resting" phase. Bivalves in advanced stages of gametogenesis and spawning display decreased clearance rates and higher respiration and ammonia excretion rates (Bayne et al., 1976a; Bayne and Widdows, 1978; Bayne et al., 1983; Prins et al., 1994; Smaal and Widdows, 1994; Lefebrvre et al., 2000). Based on histological examination of gonads, mussels were in advanced stages of gametogenesis or spawning for much of the trial period (Section 7). Therefore, shell growth may have been compromised to

some degree by the allocation of resources to gonad development, reduced ingestion rates and higher respiration rates.

It is also now being recognised that mussel feeding and digestion are a complex set of responses to fluctuating environmental conditions. Mussels can adapt to variations in food quantity and quality by changing their feeding behaviour (eg. filtration rates, preingestive selection) and efficiencies of digestion and absorption (Shumway et al., 1985; Willows, 1992; Bayne, 1993; Prins et al., 1994; Navarro et al., 1996; Wang and Fisher, 1996; Hawkins et al., 1997; Bayne, 1998). Therefore, the ambient food quality in this study, as indicated by %POM, may not necessarily be representative of the ingested nor assimilated ration and might explain its lack of correlation with observed growth rates.

The only parameter that was correlated with growth was size (shell length), whereby growth rates decreased with increasing size. Age or length is commonly reported as being a significant factor affecting bivalve growth rates (Baird, 1966; Seed, 1969b; 1976; Page and Hubbard, 1987; Mallet and Carver, 1993; Sukhotin and Maximovich, 1994; Maximovich et al., 1996). Age, which is associated with increasing size, however, is not always the primary cause of reduced growth. Transplantation of old non-growing mussels to a more favourable environment can often result in renewed growth, an indication of the growth-limiting conditions in the original habitat (Seed, 1969b). Decreasing growth rates with increasing size in a non-limiting environment is generally associated with decreased ingestion rates, reduced metabolic efficiency and increasing proportional allocations of total production to gamete synthesis (Jørgenson, 1976; Seed, 1976; Bayne et al., 1983; Hawkins and Bayne, 1992). Gamete production is considered to be the main "brake" on growth, with reproductive effort (RE) representing over 90% of total somatic production in large individuals (Bayne, 1976; Thompson, 1979; 1984b; Hawkins and Bayne, 1992). In this study, two major spawnings were identified and increasing RE with size/age may have influenced growth rates (see also Section 7 - Reproduction).

It is hypothesised that seasonal growth was not observed due to optimum growing temperatures, relatively stable salinities and an adequate food supply to support growth over most the trial period. However, growth in length and live weight actually ceased for the last three months (February - May 1996). Without any obvious predators, other than rarely-encountered sea stars, one might assume that mussels had attained their physiological limit to further growth, imposed by the ambient environmental conditions, particularly food supply. However, the final three months of the trial represented a period of optimum water temperatures (range 12°- 18°C, mean 14.9°C), maximum food supply (range 1.75 - 4.5 mg POM·l$^{-1}$, mean 3.06 mg POM·l$^{-1}$) and the

best potential for growth (SFG range 25 - 60 $J \cdot h^{-1}$, mean 41 $J \cdot h^{-1}$, Section 12). Several factors might have contributed to the cessation of growth. Firstly, the expected lifespan of *Mytilus planulatus* has been estimated to be only 2-3 years (Wilson and Hodgkin, 1967; MacIntyre et al., 1977). The exact age of experimental mussels is unknown but, using monitored growth of overspat as a guide, in February 1996 they may have been approximately 2 years and approaching the end of their expected lifespan. Therefore, the cessation of growth may be partly due to senility, which is supported by increasing mortality rates during the latter stages of the trial (Section 8). Secondly, although SFG was quite high during this period, the actual food available for the experimental mussels may have been much lower than measured concentrations due to competition from overlying mussel spat and other fouling organisms. Thirdly, mussels underwent a major spawning in February. Reproductive stress and a general lack of post-spawning fitness together with the effects of competition for food and increasing age may have all been associated with the cessation of growth and increased mortalities. Lastly, negligible growth may have been partly due to increasing incidences of dropoffs (mussels falling off droppers). Larger mussels from the perimeter of some droppers were being lost, leaving smaller individuals from the interior of droppers behind and, thereby, reducing overall mean shell lengths.

### 5.4.4 Meat Growth and Production

The growth of soft tissue (WMW, DMW, AFDMW) and total biomass (AFDMW + byssus and shell organics) closely followed the reproductive cycle, whereby increments preceded spawning losses in August 1995 and February 1996. Spawning losses during August were largely recovered by mid-October and soft tissue growth continued through until February 1996. Soft tissue weights failed to recover after the February spawning, with continued loss of meat weights and condition for the duration of the trial. As conditions were favourable for growth during this latter period, the failure to recover might be due to a number of factors including continued spawning, post-reproductive stress, senility, and increasing mortality (natural and dropoffs) of larger mussels.

Reduced meat production observed in July 1995 and December 1995 may have been due to different factors. In July, reduced production may have been due to a combination of lower temperatures and the advanced stage of gametogenesis (95% of sampled mussels were ripe, see Section 7). We have previously discussed the reduced feeding rates and higher respiration rates that may accompany this condition. Many mussels were also in an advanced stage of gametogenesis in December (45% ripe). However, their reproductive condition was accompanied by lower food

concentrations. This is the sole period when suspended particulate concentrations were below the pseudofaeces threshold and, therefore, may have been responsible for lower growth and production rates.

Similar production of soft tissue and total biomass among sites largely reflects the results provided by shell length growth. Together with the lack of significant correlations with environmental parameters, these results indicate that growing conditions (particularly food availability) were similar among sites and that food supplies were adequate throughout most of the trial period to sustain growth.

In summary, measures of mussel growth were similar at all experimental sites. These similarities are a reflection of the environmental conditions experienced by mussels cultured at each site. Most importantly, food concentrations (POM, chlorophyll) were similar, indicating that mussels grown on the fish farm did not derive any nutritional enhancement in the form of fish feed, faeces, or phytoplankton. This may have been due to fish farm wastes being too low or diluted to significantly enhance ambient food levels; mussels being too far removed from the source of fish farm wastes; and/or ambient particulate levels being consistently above T, limiting the mussels' ability to ingest a significant amount of additional fish farm organic wastes.

This and the preceding chapter have illustrated the similarities in environmental parameters, particularly suspended food concentrations, among experimental longline sites. These similarities in environmental conditions have been manifested in similar mussel growth among sites, suggesting no advantage of integrated culture in enhancing mussel growth. Concurrent with monitoring mussel growth, several other biological performances indices were also examined including condition index, meat glycogen content, reproductive development and survival. As the outcomes of the study did not show potential for enhanced mussel production near the Aquatas farm, alternative performance indices would also be expected to be similar among sites. However, the research outcomes regarding these other performance indices are sound, provide valuable information for comparison with subsequent studies at different locations, and are therefore presented in the following chapters.

# 6. Condition and Glycogen Content

## 6.1 Introduction

Condition indices (CIs) are generally regarded as useful measures of the nutritive status of bivalves. These indices may be used to follow seasonal changes in gross nutrient reserves, product quality and marketability, or the reproductive status of bivalve populations. Condition is also useful as an indicator of the general "health" of bivalves as it can be adversely affected by stress factors such as unfavourable environmental conditions, pollutants or disease. Various condition indices have been used in bivalve aquaculture for different purposes (Lucas and Beninger, 1985). Many of these indices relate meat weights (WMW, DMW, AFDMW, cooked MW) to some other variable (WLW, SW, internal shell cavity volume). As such, most of the common condition indices are measures of soft tissue "fatness" at any point in time.

It has been suggested that differences in CI between bivalve populations (Brown and Hartwick, 1988b; Hickman et al., 1991) or cultivation methods (Gabbott and Walker, 1971; Slabyj et al., 1978; Aldrich and Crowley, 1986) reflect differences in environmental parameters, including food availability. In British Columbia, the growth, condition and post-spawning recovery of Pacific oysters (*Crassostrea gigas*) cultivated within a salmon fish farm were all significantly enhanced compared with control groups (Jones and Iwama, 1991). These results were attributed to a higher quantity of food available to oysters grown on the fish farm (eg. uneaten fish feed and fish faeces).

The main objective of assessing condition in this study was to provide a relative measure of the nutritive status and market quality of mussels grown at the four sites in North West Bay. Comparisons were performed to assess if mussels grown within the Aquatas fish farm achieved a superior condition to those grown away from the farm. If so, a reasonable conclusion would be that mussels were utilising farm waste material as an additional food source. Secondarily, monitoring of CIs also provided useful information regarding the reproductive status of mussels, identifying spawning periods where condition and marketability might be unacceptable.

Glycogen is recognised as the principal energy storage product in juvenile and adult bivalves, utilised as an energy reserve under unfavourable environmental conditions and also in the formation of gametes. Therefore, the glycogen content (GC) of bivalves may vary according to environmental conditions, including food availability or quality, and the reproductive cycle. The proportion of glycogen in bivalve tissue is a good index of metabolic state and can be closely associated with condition (Gabbott

and Stephenson, 1974). High glycogen content has also been associated with improved flavour in *Crassostrea gigas* (Allen and Downing, 1991). Therefore, GC was included in this study as another criteria comparing mussel quality among the four test sites.

## 6.2 Methodology

To allow for comparisons with other studies and familiarities, three different indices were employed to describe temporal and spatial variation in condition or meat "fatness": $CI_{grav}$, $CI_{shell}$ and meat yield. The first of the three condition indices is a modified version provided by Lawrence and Scott (1982) and the CI recommended by Crosby and Gale (1990) for standardising CIs reported in the literature. This condition index ($CI_{grav}$), is based on the proportion of the internal shell cavity occupied by soft tissue and calculated as:

$$CI_{grav}\ (‰) = \text{dry meat weight (g) x 1000 / internal shell cavity capacity (g)}$$

where internal shell cavity capacity (CC) is determined gravimetrically by difference between total whole live weight (WLW, g) in air and dry shell weight in air (SW, g). This technique, much faster and easier than determining CC by displacement techniques (eg. Rodhouse, 1977), assumes no water loss due to gaping and the density of soft tissue approximates 1.0 $g \cdot ml^{-1}$.

The second condition index, $CI_{shell}$ (Walne and Mann, 1975), has been included in some bivalve studies (eg. Aldrich and Crowley, 1986; Brown and Hartwick, 1988; Taylor et al., 1992) and the CI recommended by Lucas and Beninger (1985) for most routine aquaculture applications. This index is the ratio of dry meat weight (DMW) to dry shell weight (SW) and is calculated as:

$$CI_{shell}\ (‰) = \text{DMW (g) x 1000 / SW (g)}$$

Meat yield (MY) is also included as it has been used in previous studies on mussel growth in Tasmania (Dix, 1980; Dix and Ferguson, 1984). MY is also the index most familiar to Tasmanian mussel growers and the CI recommended for commercial purposes by Hickman and Illingworth (1980). MY is the ratio of wet meat weight (WMW) to whole live weight (WLW), expressed as a percentage and calculated:

$$MY\ (\%) = \text{WMW (g) x 100 / WLW (g)}$$

Twenty mussels were randomly selected from each of the 48 samples collected monthly (12 samples·site$^{-1}$; sampling procedures have been previously described) and individually measured for shell length. These mussels were bulk-weighed to determine mean WLW·mussel$^{-1}$. Meats and shell were separated, dried to constant weight and bulk-weighed to determine mean DMW and SW·mussel$^{-1}$. Therefore, all three meat condition indices are the mean CI of 20 mussels per sample.

The three condition indices from all samples for the entire trial period were subjected to correlation analyses and each of the three pairings ($CI_{grav}$ vs $CI_{shell}$, $CI_{grav}$ vs MY and $CI_{shell}$ vs MY) analysed by least squares linear regression. This analysis was undertaken to determine the reliability of each condition index in predicting the other two CIs.

Glycogen content (GC) of dried meats was measured at the conclusion of the trial. Dried meats (stored at -20°C) from each depth (1 m and 5 m) at each site (I - IV) were split into two samples (approximately 20 mussel meats per sample) and finely ground with a geological rock-crushing mill for approximately twenty seconds. The analysis of dried meat GC (2 g subsamples) was modified from the enzymatic method of Keppler and Decker (1974). A full description of the methodology is provided in Appendix 2. All assays were performed in duplicate and GC expressed as mean glycogen (g) / 100g dry meat weight or %DMW.

As with most other mussel growth measurements, CI and GC data from four key sample dates (March 1995, August 1995, February 1996 and May 1996) were analysed by two-way ANOVA with SITE and DEPTH as fixed factors (n = 6 samples·depth$^{-1}$·site$^{-1}$). Normality of data was checked (Shapiro-Wilk test) and homogeneity of variance confirmed (Cochran's test) prior to analyses. Significant treatment effects were investigated using Fisher's PLSD.

## 6.3 Results

CIs increased from March to June 1995 prior to spawning in August. Condition increased again from September 1995 to February 1996 prior to a second major spawning. However, no subsequent post-spawning recovery was observed by the conclusion of the trial in May 1996.

### 6.3.1 $CI_{grav}$

Overall mean $CI_{grav}$ increased from 70.5 ± 1.2‰ (n = 48 samples) in March 1995 to a peak of 116.9 ± 0.8‰ in June 1995 (Fig. 6.1). $CI_{grav}$ declined slightly to 101.4 ± 0.7‰ in August and sharply to 62.5 ± 2.0‰ in September with the first major spawning. Condition increased through the spring and early summer to peak at 99.3 ± 1.7‰ in February 1996 before the second major spawning where $CI_{grav}$ fell to 79.5 ± 2.1‰ in March 1996. However, as previously noted, meat weights and condition of mussels at sites III and IV were probably underestimated in March 1996, as there was a delay in processing mussels from these two sites, and these values should be viewed with caution. Mussels continued to lose condition through the remainder of the trial, finishing with a mean $CI_{grav}$ of 69.3 ± 1.2‰ in May 1996.

$CI_{grav}$ was significantly different between longline sites on each of the key sampling dates, including the initial sample of March 1995 ($P < 0.0001$), where I > II > III > IV (Fisher's LSD, $P < 0.05$). In August 1995 a significant site x depth interaction was indicated ($P < 0.0001$), where $CI_{grav}$ of mussels at 5 m were similar among sites (mean 98.8 ± 0.4‰, n = 24 samples) but differed at 1 m, where $I^c = II^c > III^b > IV^a$ ($P < 0.05$). However, the greatest difference in $CI_{grav}$ at 1 m was only 9.5‰, between site I (107.3‰) and site IV (97.8‰). In February and May 1996, although the differences among sites were again significant ($P < 0.01$), differences were minor, being within 17‰ (ie. < 2%). Differences in $CI_{grav}$ between growing depths were not significant for any of the key sample dates other than the site x depth interaction in August 1995 already mentioned.

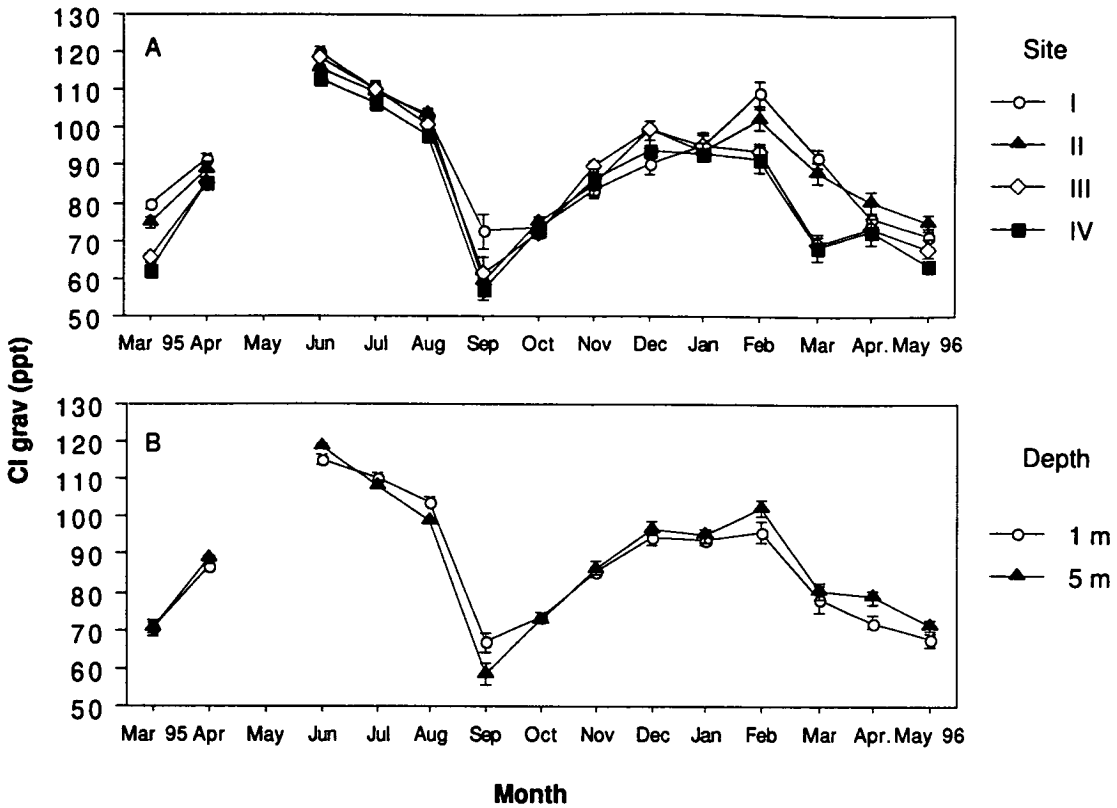


**Figure 6.1.** (A) Monthly condition index ($CI_{grav}$) at each site (mean ± SE‰, n = 12 samples per site) and (B) at each depth (n = 24 samples per depth). Error bars not visible lie within point boundaries.

### 6.3.2 $CI_{shell}$

Initial mean $CI_{shell}$ values of 134 ± 2‰ (n = 48 samples) increased to 241 ± 2‰ in June, fell slightly to 227 ± 2‰ in August and fell sharply to 141 ± 4‰ after spawning (Fig. 6.2). $CI_{shell}$ increased again to peak at 218 ± 4‰ in February before the second spawning event, where condition fell and continued to decline for the duration of the trial to finish at 147 ± 3‰ in May 1996.

Differences in $CI_{shell}$ between sites were significant for all four of the key sampling dates:

In March 1995 ($P < 0.0001$), $CI_{shell}$ at site I (146.9‰) > II > III > IV (119.8‰).
In August 1996 ($P < 0.05$), $CI_{shell}$ at site $I^{ab}$ (229.9‰) = $II^{a}$ = $IV^{a}$ ≤ $III^{b}$ (119.9‰).
In February 1996 ($P < 0.01$), $CI_{shell}$ at site I (240.8‰) > II = III = IV (202.1‰).
In May 1996, ($P < 0.05$) $CI_{shell}$ at site $I^{b}$ (150.6‰) = $II^{b}$ = $III^{ab}$ ≥ $IV^{a}$ (135.4‰).

Differences in $CI_{shell}$ between depths were significant in February 1996 and May 1996 (both at $P < 0.05$) where $CI_{shell}$ at 1 m was slightly less than at 5 m on both occasions

(17.8‰ and 11.5‰ less, respectively). Again, differences in $CI_{shell}$ among sites and between depths were considered to be minor. No significant site x depth interaction was indicated for any of the four key sampling dates.

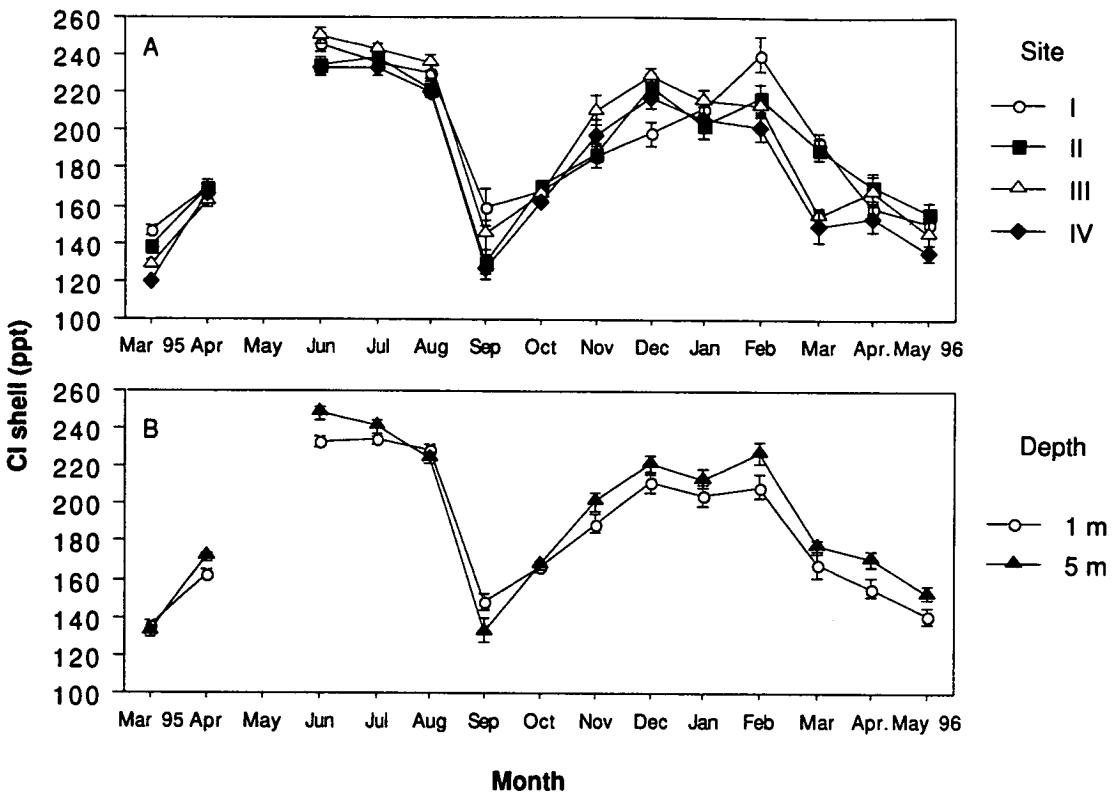


**Figure 6.2.** (A) Monthly condition index ($CI_{shell}$) at each site (mean ± SE‰, n = 12 samples per site) and (B) at each depth (n = 24 samples per depth). Error bars not visible lie within point boundaries.

### 6.3.3 Meat Yield

Meat yield (MY) increased from an initial mean of 23.5 ± 0.22% (n = 48 samples) in March 1995 to a peak of 36.6 ± 0.21% in June 1995 (Fig. 6.3). MY remained static through to August, whereby a sharp decline to 25.6 ± 0.5% was observed in September. MY subsequently increased, peaking at 34.4 ± 0.4% in February 1996, before declining again after a second spawning to 29.5 ± 0.4% in March. Post-spawning recovery was not evident, with MY continuing to decline to 26.2 ± 0.26% at the conclusion of the trial.

Significant differences in MY were indicated for March 1995 ($P < 0.0001$) and May 1996 ($P < 0.01$). The greatest difference in May 1996 was only 2.3% (where $II^{b} \geq I^{ab} = II^{ab} = IV^{a}$) and, again, of little biological interest. No differences in MY between

growing depths, nor a site x depth interaction, at any of the four key sampling times were significant ($P > 0.05$).

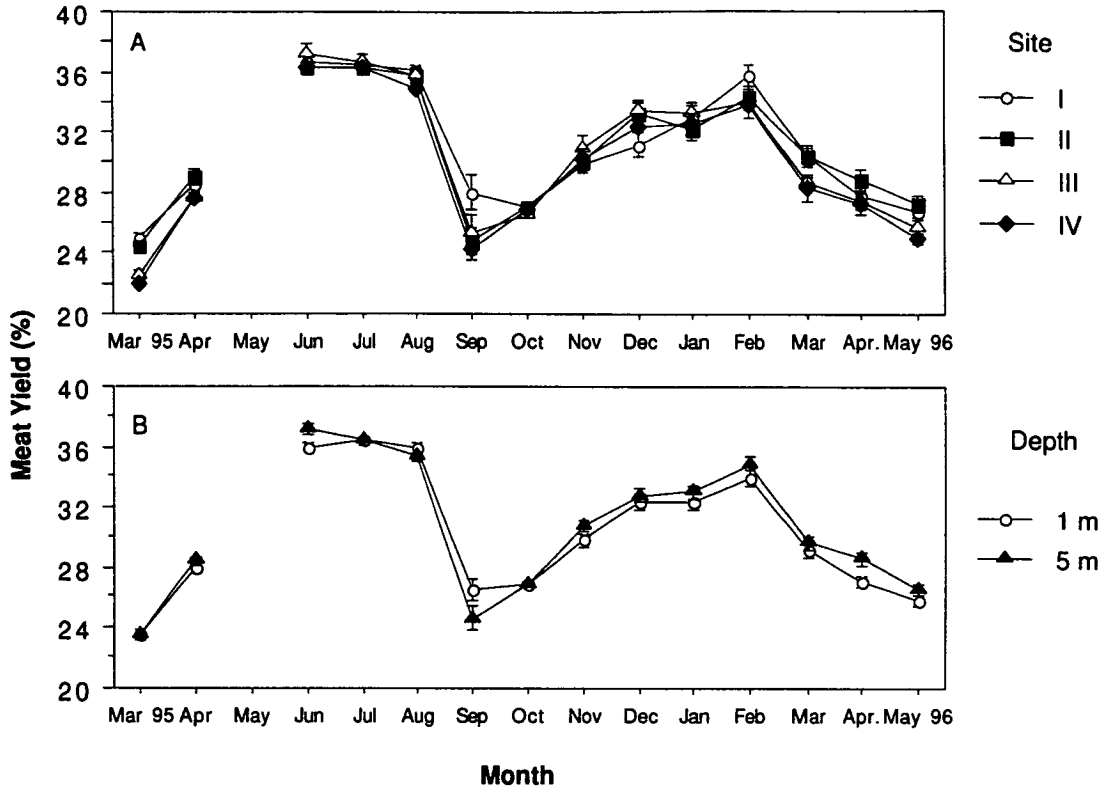


**Figure 6.3.** (A) Monthly meat yield at each site (mean ± SE%, n = 12 samples per site) and (B) at each depth (n = 24 samples per depth). Error bars not visible lie within point boundaries.

Correlations between the above three condition indices were highly significant ($P < 0.0001$). The three regression equations for combined CI data pooled across all sites (n = 672 samples) were:

$$CI_{shell} = 1.95 + 2.14\ CI_{grav} \quad (r^2 = 0.871;\ P < 0.0001).$$
$$MY = 9.53 + 0.24\ CI_{grav} \quad (r^2 = 0.867;\ P < 0.0001)$$
$$MY = 10.6 + 0.11\ CI_{shell} \quad (r^2 = 0.881;\ P < 0.0001)$$

These regressions suggest that any of the three condition indices may be converted to obtain reasonable estimates of the others.

### 6.3.4 Glycogen content

The variation in tissue glycogen content (GC) over the trial period, expressed as g glycogen / 100 g DMW or %DMW is presented in Fig. 6.4. GC displayed similar temporal variation among sites and between depths. The initial overall GC of 12.2 ±

0.21%DMW (n = 16 samples) in March 1995 increased to 16.8 ± 0.37%DMW in April. Thereafter, GC decreased through the winter months until September 1995, reaching a minimum of 3.9 ± 0.38%DMW, coinciding with minimum meat weights and condition. GC was quickly restored during September to 16.2 ± 0.19%DMW at the beginning of October and thereafter decreased slowly to 11.7 ± 0.22%DMW by February 1996. Contrary to the minimum levels achieved in September, this second "minimum" coincided with peaks in meat weights and condition. A major spawning event occurred in February and GC subsequently increased during the final three months of the trial. This increase in GC corresponded with decreasing meat weights and condition indices. Maximum GC levels of 18.9 ± 0.53%DMW were observed at the conclusion of the trial in May 1996. No differences in GC among sites or between growing depth were significant on any of the four key sampling dates, nor were any significant site x depth interactions indicated ($P > 0.05$).

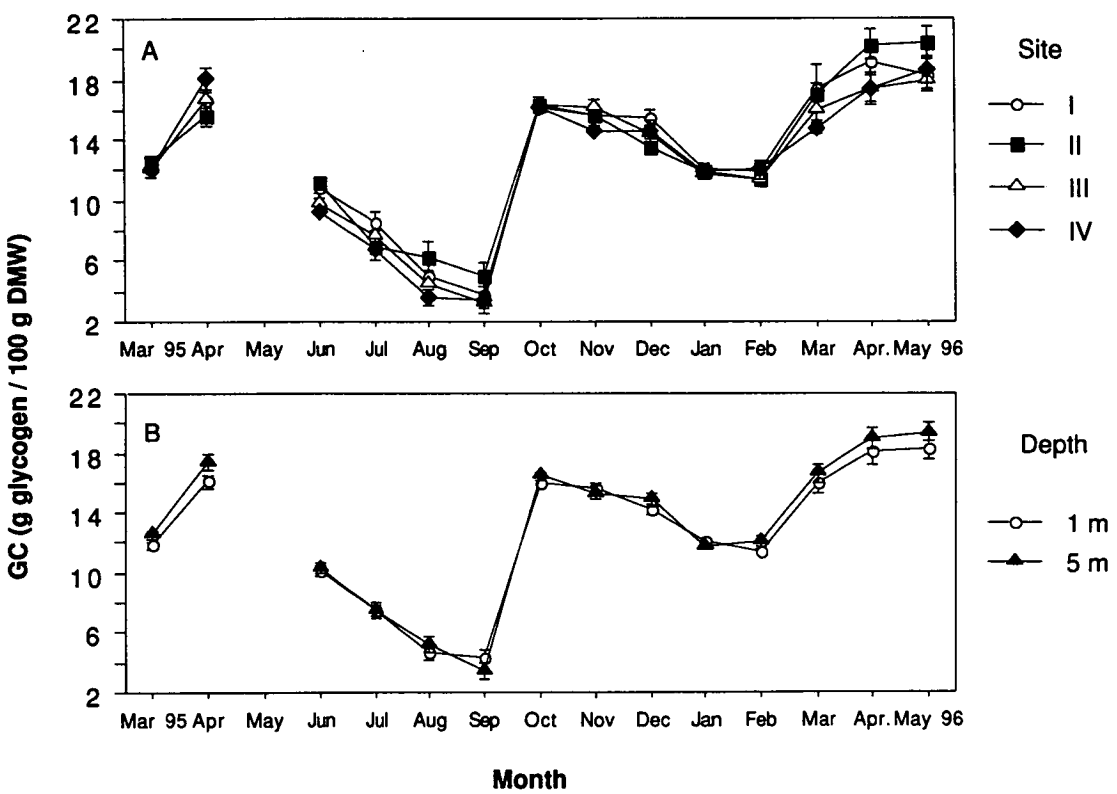


**Figure 6.4.** (A) Monthly glycogen content (GC) at each site (mean ± SE g glycogen/100g DMW, n = 4 samples per site) and (B) at each growing depth (n = 8 samples per depth). Each n is the duplicate mean glycogen content of a 2 g subsample from twenty pooled and ground mussel meats.

## 6.4 Discussion

Mussel meat condition indices ($CI_{grav}$, $CI_{shell}$ and MY) closely followed the cycle of reproductive development and spawning, whereby CIs gradually increased prior to two significant declines in August 1995 and February 1996 as a result of the loss of reproductive tissue (see also Section 7 - Reproduction). Similar condition cycles, with multiple spawnings within a year, have previously been reported for mussels in the northern hemisphere (Lutz et al., 1980; Incze and Lutz, 1980; Bressan and Marin, 1985; Emmett et al., 1987) as well as earlier studies of Tasmanian mussels (Dix, 1980; Dix and Ferguson, 1984). However, from these earlier studies, periods of peak condition and spawning are not always consistent from year to year, neither at the same location nor between locations within geographic areas. Temporal and spatial variation in condition and spawning most likely reflect variation in environmental conditions between habitats or periods. Particularly, differences in food availability, food quality and/or exogenous factors affecting mussels' capacity to assimilate nutrients, may alter the nutrient storage cycle and thus the timing of gametogenic events and spawning (Newell et al., 1982).

Temporal variation in condition is evident when comparing results from this study with those of Dix and Ferguson (1984). The annual mean meat yields (July 1979 - June 1980, July 1980 - June 1981) from three southern Tasmanian locations (including Margate in North West Bay) recorded by Dix and Ferguson (1984) were considerably less (18.4 - 22.7%) than the annual mean observed in the present study over a similar period (June 1995 - May 1996; mean 31.2 $\pm$ 0.2%, n = 576 samples). The differences may be attributed to different culture methods, environmental conditions and/or WMW measurements (a case for employing DMW rather than WMW in calculating condition indices).

It is of interest to note that all three meat CIs were at, or near to, their lowest values from the outset of the trial in March 1995. Low CIs, at the end of the Southern Hemisphere summer, suggests that mussels may have recently spawned prior to the start of the trial. This possibility is supported by the general appearance of meats in March 1995; it was difficult to distinguish gender from the colour of mantle tissue, as were mussels from the September 1995 and March 1996 post-spawn samples. It is not inconceivable that handling-induced spawning may have occurred as a result of the initial stripping, grading and socking procedures in February 1995. Physical stimulation will often cause ripe mussels to spawn (Lutz et al., 1980). It could also be argued that the stocked mussels, possibly less than one year old, might still have been immature, thereby making sex identification difficult. However, it seems unlikely that

condition would have been so low at the end of the 1994-95 summer-growing period if spawning did not occur.

It is also interesting to note that significant differences in condition were indicated for the initial sample of March 1995, with condition decreasing from sites I through to IV. As seed mussels were all sourced from the same location, no such differences would be expected. The gradient of decreasing condition from I to IV corresponds with the order of socking and deployment, where mussels at site I were in place approximately 2 weeks before mussels were deployed at site IV. Differences in condition may have been due to stress and/or spawning associated with the stocking procedures prior to deployment. Therefore, mussels at site I would have had two weeks recovery time prior to the first sample, compared with only a few days recovery time for mussels at site IV.

There appears to be no particular advantage in growing mussels on the Aquatas fish farm in terms of improving meat quality or marketability. Differences in meat condition were statistically significant between sites for some of the key sampling dates. However, CIs were not consistently greater at the fish farm sites throughout the trial. Statistically significant CI differences on key sampling dates were minor and would have no effect on the marketability of product. Although loss of condition during the August 1995 spawning appeared to be less severe at site I than at the other sites (ie. condition of September 1995 samples at site I > II, III and IV), there were no CI differences among sites in October 1995. This could suggest that spawning at site I had not been completed until after the September 1 1995 sampling. Monthly data clearly show the close coupling of condition indices at all sites over the entire trial period, with no trend of increasing condition for fish farm mussels (sites I and II) relative to off-farm mussels (sites III and IV). If condition is a reasonable reflection of nutritive status, it follows that fish farm mussels did not ingest a greater quantity, nor a higher quality, of food material than mussels grown distant to the farm. This might be expected, as no significant differences in particulate food availability (POM, chlorophll) nor quality (%POM) were indicated among the four longline sites.

A lack of detectable differences in available food supply and quality between longline sites is supported by the meat glycogen content analyses. As with meat condition indices, glycogen contents were very similar across all sites throughout the trial. As the accumulation of glycogen reserves is strongly associated with food availability and quality (Bayne, 1976; Gabbott and Stephenson, 1974; Bressan and Marin, 1985; Fidalgo et al., 1994) a reasonable conclusion is that diet was similar at all mussel longline sites.

In overseas studies, mussels grown adjacent to fish cages in Scotland (Okumus, 1993) and British Columbia (Taylor et al., 1992) similarly did not display significantly higher condition than mussels grown at control sites. These studies, together with the present study, contrast with Jones and Iwama (1991); condition and post-spawning recovery of Pacific oysters (*Crassostrea gigas*) grown within a salmon farm was significantly greater than mussels grown distant to the farm. Higher condition of fish farm oysters was attributed to an enhanced food supply from fish feed, faeces and elevated levels of phytoplankton. It appears that the conceptual advantages for integrating bivalves with fish culture systems, enhanced growth and condition as a consequence of a greater food supply, are not always realised; why not? The answer most likely lies with differences in food availability between culture locations, previously discussed in relation to growth but should be reiterated.

Firstly, although expected, food availability and quality are not necessarily higher at mussel longline sites adjacent to fishcages compared with control sites (Taylor et al., 1992; present study). The dilution of particulate waste matter in an open-water system, variously affected by the volume, current speed and mixing of the receiving water, may be such that increases in particle concentrations above ambient levels are either undetectable, or insignificant in terms of enhancing mussel growth or condition.

Waste particulate concentrations would also decrease with distance from the cages due to sedimentation. Jones and Iwama (1991) found the best growth and condition were displayed by oysters grown inside salmon cages. In the present study, the two longlines closest to the fishcages (70m and 100m distant) may have been too far from the cages, allowing suspended waste particles to settle to the sea bottom before reaching these two sites. Fouling organisms on the cages may also reduce fish waste particles reaching mussels grown on adjacent longlines. These organisms may either intercept particles emanating from the fish cages (eg. fouling bivalves, tunicates) or impede the current flow through the cage system (eg. fouling algae). Taylor et al. (1992) suggested that similar particle concentrations among test sites associated with fish farms was due to fouling mussels on the fish cage nets intercepting fish waste particles. In the present study, biofouling of nets was not significant. However, experimental droppers became increasingly fouled over the trial period, particularly by several settlements of overspat as well as tunicates, sponges and algae (Fig. 6.5).

Ambient suspended particle concentrations must also be considered when assessing the merits of an integrated fish-bivalve culture system. Bivalves grown adjacent to fish cages are unlikely to benefit from particulate waste loadings when ambient SPM concentrations exceed the threshold level for pseudofaeces production (Troell and Norberg, 1998). As previously discussed in other sections (eg. length and weight

growth) the SPM concentrations during this trial were consistently greater than the pseudofaeces threshold level (5 $mg \cdot l^{-1}$) for most of the trial period. When ambient SPM concentrations exceed this level, additional waste particulates in the water column constitute only a small fraction of filtered material and may be superfluous to a bivalve's total diet. Additional fish waste particles might only be beneficial if their quality (eg. digestibility, energy content) is higher than natural food sources. As pointed out by Troell et al. (1999) reviewing the Jones and Iwama (1992) study, particulate levels were below the threshold for pseudofaeces production (T) 62% of the study time at the control sites compared to only 25% inside the salmon cages; particulate wastes and increased phytoplankton production presumably raised ambient particulate levels. Therefore this is a case where bivalve growth and condition is enhanced when normally low ambient particulate levels are increased by particulate fish farm wastes.

As with most other measured growth parameters, depth did not significantly influence meat condition or glycogen content. This probably reflects similar environmental conditions, although this cannot be thoroughly tested, as some factors, particularly food supply, were not sampled at each depth.

Monitoring of bivalve condition can be useful in identifying periods of peak condition and marketability, as well as periods of low or unmarketable condition when harvesting should be avoided (Lutz et al., 1980; Zandee et al., 1980; Bressan and Marin, 1985; Hickman et al., 1991; Okumus, 1993). For example, the minimum marketable condition ($CI_{grav}$) of Tasmanian-grown Pacific oysters is 65‰ (Gardner, 1993). Monitoring the status of meat weight and condition is particularly important where product is sold and marketed as meat-only or in the half-shell. Peak mussel condition in NWB was achieved during the winter (June - August 1995) and to a lesser extent during the summer (December 1995 - February 1996) preceding each of the two major spawnings in August 1995 and February 1996. In commercial practice mussel condition has little relevance to Tasmanian mussel farmers. Production is relatively low (350 tonnes per annum), mussels are sold on a whole live weight basis (A$3.00 - A$3.50 / kg WLW, June 2000), no premium is paid for mussels in peak condition and demand is high throughout the year (G. Schroter, Secretary, Tasmanian Mussel Growers Cooperative, pers. com.). As long as harvested product is 70 - 80 mm minimum shell length with meat yield exceeding 20%, Tasmanian-grown mussels are marketable within Australia (G. Schroter, pers. com.). As such, mussel condition in this study never fell below marketable quality, even after spawning. In commercial practice, farmed mussels can fall below marketable condition and farmers generally avoid harvesting during spawning periods.

**Figure 6.5**. Example of algal *(Mycodia aciculare)* biofouling of experimental mussel socks (Site II, March 1996).

# 7. Reproduction

## 7.1 Introduction

Water temperature and food availability are among the main exogenous factors controlling the gametogenic cycle and fecundity of mussels, accounting for temporal and spatial variability of gonad development and reproductive output among different geographical areas (Seed, 1976). Differences in the reproductive cycle and reproductive effort among mussel populations from neighbouring sites have also been reported, explained by differences in food availability (Bayne and Worrall, 1980; Newell et al., 1982; Bayne et al., 1983; Rodhouse et al., 1984b; Villalba, 1995). As such, the gametogenic cycle and reproductive effort of mussels in this study were investigated to assess differences among longline sites. It was considered that any differences in gametogenesis, fecundity and/or reproductive effort might be indicators of a differential food supply (quantity and/or quality) among mussel longline sites; again, any differences would infer a nutritional advantage for mussels grown adjacent to the salmon fishcages.

## 7.2 Methodology

Mussel reproductive development was only monitored at site I (closest to the fishcages) and site IV (furthest from the fishcages); any site differences in reproductive development were expected to come from the two most distant longlines. Ten mussels per site (approximately 5 males and 5 females) were selected each month for gonad staging. A 1.0 x 0.5 cm section of mantle tissue was excised from each mussel, immersed in Davidson's fixative for 24 h, rinsed in freshwater and stored in 70% ethanol. Tissue samples were then dehydrated, embedded in paraffin, sectioned at 5 to 7 μm and placed on slides according to standard techniques (Luna, 1968). Tissues were rehydrated using serially increasing ethanol solutions and sections stained with haematoxylin-eosin (Humason, 1962).

Histological sections were examined microscopically, with each specimen assigned to one of six stages of reproductive development: Indeterminate, Developing 1, Developing 2, Advanced, Spawning and Regressive (modified from Dinamani, 1974) and a numerical factor (NF) ranging from 0 to 3 (modified from Seed, 1969a; see Appendix 1 for descriptions of the various stages and corresponding NF). The effect of longline SITE on gonadal development was analysed using Chi-squared tests on monthly staging data based on counts (n = 10 mussels·sites$^{-1}$, Statview™ 4.5, Abacus Concepts, 1994). For clarity of presentation, the number of reproductive stages was

condensed into three main categories: **D**eveloping (number of individuals in Developing 1 + Developing 2), **A**dvanced, and **S**pawning or spawned (number of individuals Spawning + Regressive + Indeterminate).

A monthly mean gonad index (GI) was calculated by multiplying the number of individuals in each gonadal stage by their corresponding numerical factor and dividing the sum of these products by the total number of individuals in the sample (Seed, 1969a). The resulting gonad index may vary from 0, if all individuals are in the resting or indeterminate condition, to 3 if all individuals are sexually mature. Gametogenesis leads to an increase in the GI, while a decrease indicates spawning activity.

Biomass losses due to spawning (fecundity) at all four sites were determined indirectly by the difference between ash-free dry meat weights (AFDMW) before and after identified spawning periods (Kautsky, 1982b; Bayne et al., 1983; Crisp, 1984). Reproductive effort (RE), the proportion of a mussel's total energy budget allocated to gamete production, was calculated from the expression

$RE\ (\%) = [(P_r) / (P_r + P_g)] \times 100$ , where:

$P_r$ (gamete production) = (spawning losses, g·mussel$^{-1}$) x 23.0 kJ·g AFDMW$^{-1}$

$P_g$ (somatic production) = (AFDMW increment prior to spawning - spawning losses, g·mussel$^{-1}$) x 21.8 kJ
(Bayne et al., 1983; Thompson, 1984b).

One-way ANOVA was used to test if fecundity or reproductive effort differed between sites for each major spawning period after confirmation of normality (Shapiro-Wilk test) and homogeneity of variance (Cochran's test, n = 12 samples·site$^{-1}$).

## 7.3 Results

### 7.3.1 Reproductive stages

The pattern of gonad development during the trial period was similar at both sites I and IV. No significant site differences in the distribution of gonad stages were indicated for any month (Chi-squared test, $P > 0.05$) except October 1996. Therefore, the total number of individuals in each reproductive stage for each month was pooled from both sites and presented in Table 7.1. The frequency (%) of mussels in each of the three condensed categories (D, A and S) is illustrated in Fig. 7.1.

There was evidence of individuals in a spawning condition throughout the trial, except June 1995, including the start of the trial period when the mussels were approximately one year old. A conspicuous lack of "resting" phase individuals was identified, with only one individual classified as "indeterminate".

**Table 7.1.** Distribution of gonad stages and gonad index (GI) in samples of *M. planulatus* from March 1995 to May 1996 (pooled data from sites I and IV). See Appendix 1 for description of gonad stages.

| | Male | | | | | | | Female | | | | | | Total | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Stage | I | D1 | D2 | A | S | R | n | D1 | D2 | A | S | R | n | I | D1 | D2 | A | S | R | N | GI |
| Mar | | | | 8 | | | 8 | | 1 | 9 | 2 | | 12 | | | 1 | 17 | 2 | | 20 | 2.9 |
| Apr | | | | 9 | | | 9 | | | 7 | | 4 | 11 | | | | 16 | | 4 | 20 | 2.6 |
| May | | | | | | | | | | | | | | | | | | | | | |
| Jun | | | | 7 | | | 7 | | 4 | 9 | | | 13 | | | 4 | 16 | | | 20 | 2.8 |
| Jul | | | | 10 | | | 10 | | 1 | 8 | | 1 | 10 | | | 1 | 18 | | 1 | 20 | 2.9 |
| Aug | | | | 10 | | | 10 | | 1 | 7 | | 2 | 10 | | | 1 | 17 | | 2 | 20 | 2.8 |
| Sep | 1 | | | 2 | 8 | | 11 | 1 | | 3 | 5 | | 9 | 1 | 1 | | 5 | 13 | | 20 | 2.1 |
| Oct | | | | | 9 | | 9 | | 5 | 2 | 1 | 3 | 11 | | | 5 | 2 | 10 | 3 | 20 | 2.0 |
| Nov | | | | 5 | 4 | 1 | 10 | | 7 | 1 | 1 | 1 | 10 | | | 7 | 6 | 5 | 2 | 20 | 2.2 |
| Dec | | | | 5 | 6 | | 11 | | | 4 | 2 | 3 | 9 | | | | 9 | 8 | 3 | 20 | 2.3 |
| Jan | | | | 9 | 2 | | 11 | | | 4 | | 5 | 9 | | | | 13 | 2 | 5 | 20 | 2.4 |
| Feb | | | | 8 | 2 | | 10 | | 5 | 4 | 1 | | 10 | | | 5 | 12 | 3 | | 20 | 2.6 |
| Mar | | 2 | | 2 | 6 | 2 | 12 | | 1 | 2 | 2 | 3 | 8 | | 2 | 1 | 4 | 8 | 5 | 20 | 1.9 |
| Apr | | 2 | 6 | 1 | | | 9 | 2 | 3 | 3 | 2 | 1 | 11 | | 4 | 9 | 4 | 2 | 1 | 20 | 2.0 |
| May | | 2 | 12 | 1 | | | 15 | | | 3 | | 2 | 5 | | 2 | 12 | 4 | | 2 | 20 | 2.0 |

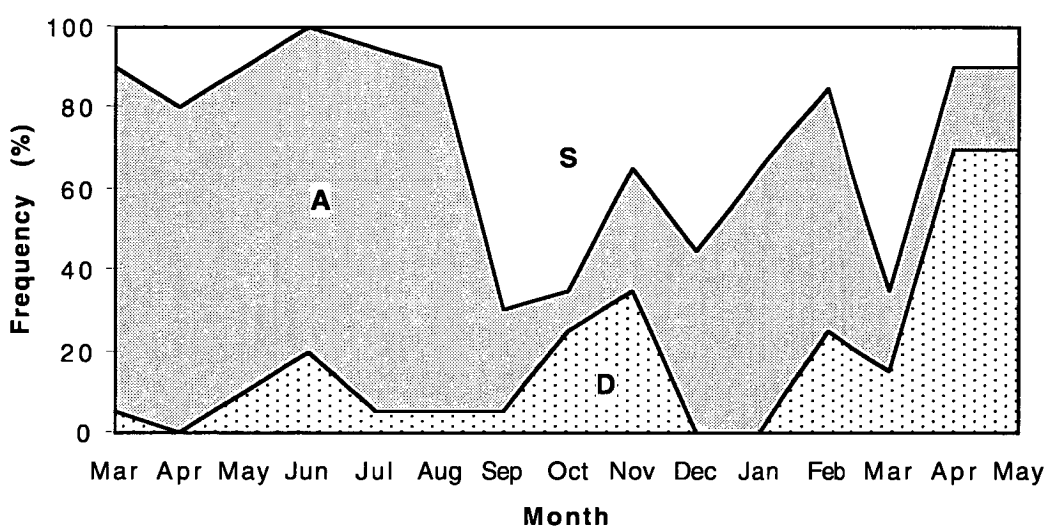


**Figure 7.1.** Frequency of the mussel population in each of the three main reproductive stages from March 1995 to May 1996. A = advanced or ripe, D = developing (stages D1 + D2), S = spawning and/or spent (stages S + R + I). n = 20 mussels per month (10 mussels from each of sites I and IV).

Two major spawning periods, August-September 1995 (late winter-early spring) and February 1996 (late summer), were identified by the increased percentage of S and R stage individuals (65%) in the September-October 1995 and March 1996 samples (also see Fig. 7.2). However, a significant number of spawning individuals was also

identified between these two major spawnings (spring through summer, average 35%), particularly in December when 55% of sampled mussels were in the spawning or spent phase.

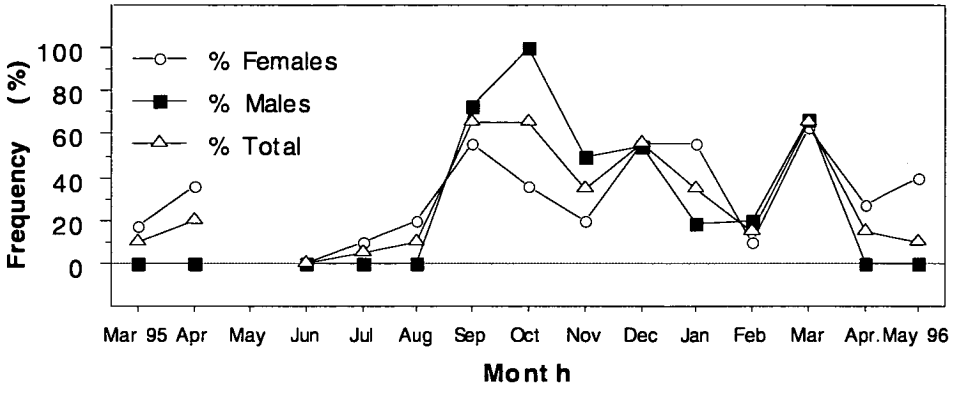


**Figure 7.2.** Frequency (%) of mussels in a spawning or post-spawning condition (stages S + R) during the period March 1995 to May 1996. Total n = 20 mussels per month, male n and female n as in Table 7.1 (pooled data from sites I and IV).

### 7.3.2 Gonad Index

The pooled monthly mean gonad index (GI) from sites I and IV is provided in Table 7.1. Except for a suspiciously low GI for female mussels observed in April 1995, the overall mean GI was quite stable from March 1995 through to August 1995 (Fig. 7.3). An overall mean of 2.8 for this period indicates an advanced state of gametogenesis through the 1995 autumn and winter. Spawning activity in late winter-early spring is suggested from the decline in GI from 2.8 in August to 2.1 and 1.95 in September and October. GI subsequently increased through the spring and summer to 2.6 in February 1996 before declining to 1.9 in March 1996 with a second major spawning. Males tended to have a higher GI compared to the females, suggesting that males were in a more advanced or "ripe" condition for much of the trial.

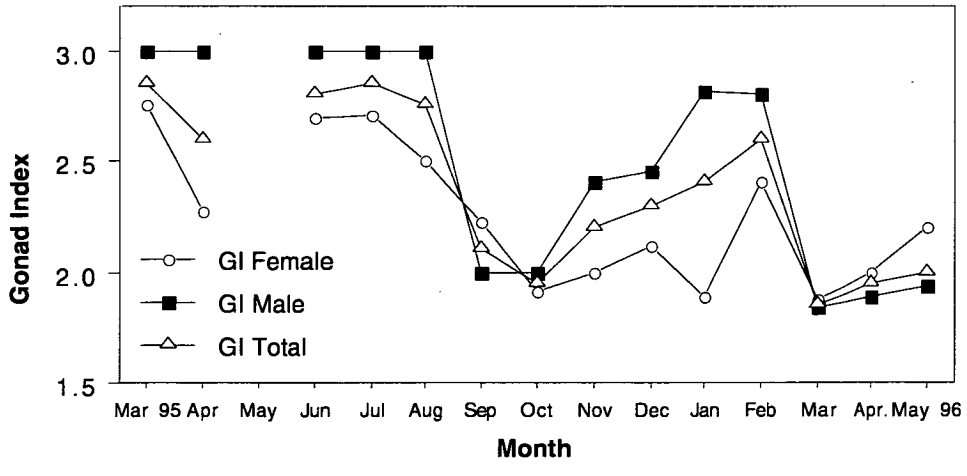


**Figure 7.3.** Monthly mean gonad index (GI) of mussels from March 1995 to May 1996 (pooled data from sites I and IV). GI Total n = 20 mussels per month; male n and female n as in Table 7.1.

The pattern of gonad development and spawning, as represented by the GI, followed that of soft tissue growth and condition for most of the trial (Fig. 7.4 and 7.5). Monthly changes in GI were significantly correlated with changes in mean ash-free dry meat weight from sites I and IV ($r = 0.817$, $P < 0.001$, $n = 13$ observations). Monthly GI was also significantly correlated with mean monthly condition index ($CI_{grav}$) from sites I and IV, ($r = 0.678$, $P < 0.01$, $n = 14$ observations).

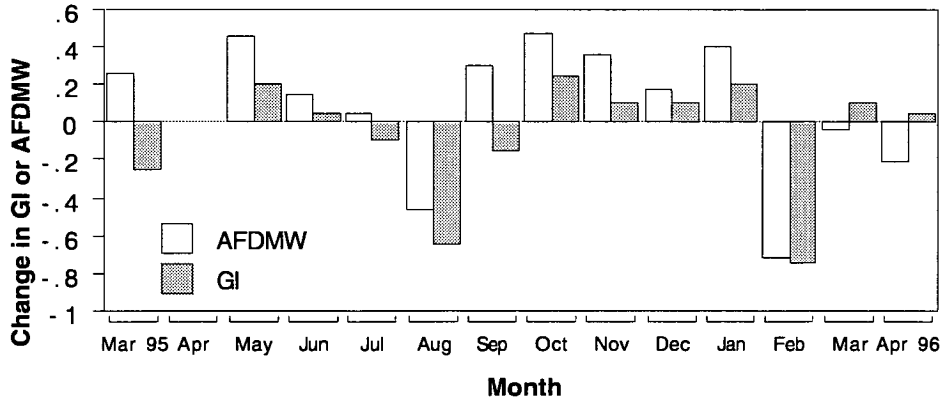


**Figure 7.4.** Monthly change in ash-free dry meat weight at sites I and IV (AFDMW, mean g·month$^{-1}$) and gonad index (GI) from March 1995 to May 1996.

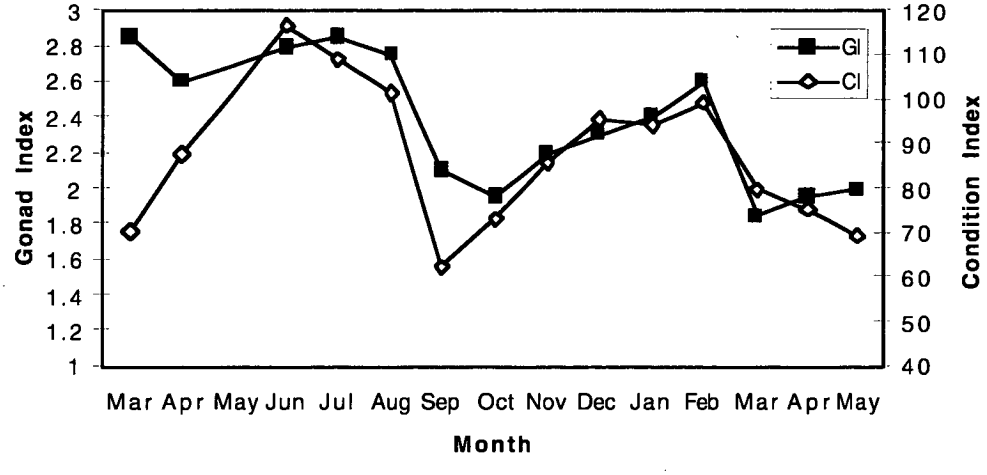


**Figure 7.5.** Monthly gonad index (GI) compared with mean gravimetric condition index ($CI_{grav}$ ‰, n = 24 samples per month).

A weak but significant negative correlation between GI and glycogen content (g glycogen/100 g dry meat weight) was also observed ($r = -0.54$, $P = 0.05$, $n = 14$ observations). The general trend of decreasing glycogen content during the periods of gametogenesis leading up to the two major spawnings indicates a diversion of, at least some, glycogen stores into gamete development (Fig. 7.6). The weak correlation was due to the dramatic decline in glycogen content prior to the spring spawning

(approximately 70% decline from April to August 1995) as compared to only a 28% decline from October 1995 to February 1996.

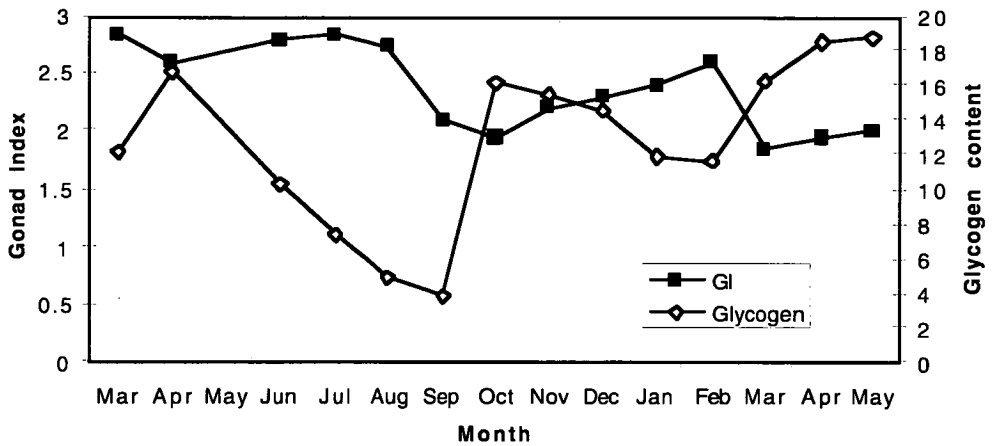


**Figure 7.6.** Monthly gonad index (GI) and overall mean glycogen content (g glycogen · 100 g dry meat weight$^{-1}$, n = 16 samples per month, each sample = the glycogen content of a 2.0 g subsample from 20 pooled mussel meats).

### 7.3.3 Spawning losses

The identification of spawning activity during August 1995 and February 1996, based on histological examination of gonads, was supported by corresponding losses in ash-free dry meat weight (AFDMW, Fig. 7.7).

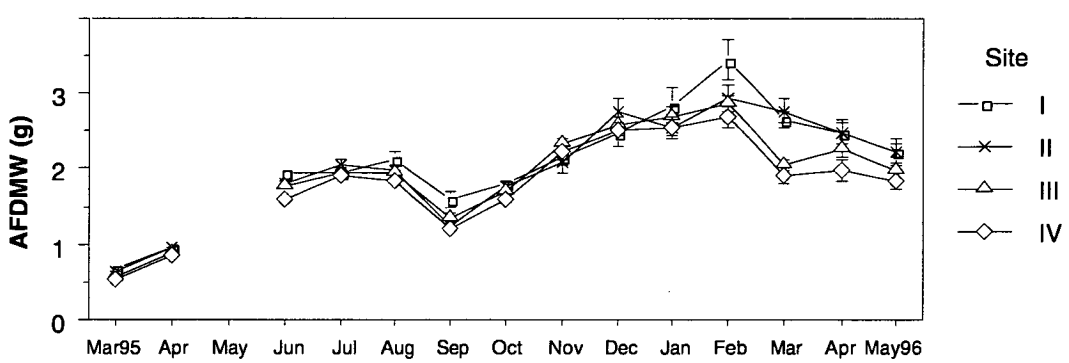


**Figure 7.7.** Monthly ash-free dry meat weight (AFDMW) at each of four sites in North West Bay from March 1995 to May 1996 (mean ± SE g·mussel$^{-1}$, n = 12 samples per site per month)

Overall mean spawning loss at all sites in August was 0.62 ± 0.05 g·mussel$^{-1}$ (difference between August and September 1995 mean AFDMW at each depth within each site, n = 8). This represented a mean loss of 31% of the pre-spawning AFDMW at the beginning of August. The high frequency of spawning individuals identified in October 1995 (65%) was not supported by biomass losses or static growth during September; an increase in AFDMW during September was observed. Biomass lost during August 1995 was quickly recovered by November and AFDMW production

continued until December, when AFDMW remained static. Meat growth increased again in January 1996 before another major spawning in February 1996, indicated by a sharp loss in AFDMW. A gradual decline in AFDMW until the end of the trial was observed.

As previously mentioned, the lower biomass values recorded at sites III and IV in March 1996 may have been partly due to the delay incurred in processing these mussels, and may not be totally attributable to spawning losses. Therefore, biomass losses attributed to the second spawning were calculated as the difference in AFDMW between February and April 1996; no AFDMW growth was observed in March 1996. It is recognised that biomass loss may be attributed to metabolic losses and mortality of larger mussels, as well as spawning losses; however, these three components cannot be differentiated. Therefore, based on the difference in AFDMW between February and April 1996, mean spawning losses were $0.69 \pm 0.10$ g·mussel$^{-1}$ ($n = 8$), or 23% of the pre-spawning AFDMW in February 1996.

Spawning losses in February 1996 were the same as in August 1995 ($P > 0.05$, mean $0.65 \pm 0.05$ g·mussel$^{-1}$, $n = 16$). Spawning losses were not significantly different between sites for either the August or February spawning periods, nor the combined spawning losses for both periods, nor was there a site x spawning period interaction ($P > 0.05$). February spawning losses relative to pre-spawning AFDMW levels (23%) were less ($P < 0.05$) than the August spawning losses (31%); but differences between sites or a site x spawning period interaction were not indicated ($P > 0.05$).

The August 1995 spawning losses represented a reproductive effort (RE) of $46.3 \pm 3.8\%$ ($n = 8$) of the longline production from March to August 1995. Spawning losses in February 1996 represented a RE of $43.4 \pm 5.1\%$ of the total production from September 1995 to February 1996. Total RE, based solely on biomass losses from these two spawning periods was $44.5 \pm 2.0\%$ ($n = 8$). It is recognised that spawning individuals were identified for most months other than the two major spawnings in August 1995 and February 1996, particularly during the September 1995 - February 1996 period. Therefore, overall reproductive effort for the trial period, based solely on the two major spawns, may be underestimated. However, reproductive effort was not significantly different between sites for either the March to August 1995 period, the September 1995 to February 1996 period, or for the total trial period ($P > 0.05$).

## 7.4 Discussion

### 7.4.1 Reproductive cycle

The observed gametogenic cycle compares favourably with previous studies of *Mytilus planulatus* in Tasmanian waters (MacIntyre et al., 1977; Dix and Ferguson, 1984). These earlier studies similarly reported evidence of some spawning individuals year-round, major spawnings during late winter-early spring and late summer-early autumn, and no well-defined "resting period" between spawnings. Although the general trends of the gametogenic cycles are similar, the timing of major spawnings of Tasmanian mussels is not consistent from year to year, or at all locations (Dix and Ferguson, 1984). Spatial and temporal variations in reproductive status are to be expected. Environmental variation between habitats or periods, differing in food availability/quality or other exogenous factors affecting a mussel's capacity to assimilate nutrients, will alter the nutrient storage cycle and, therefore, the timing of gametogenic events and spawning (Newell et al., 1982).

The observed reproductive cycle of Tasmanian *Mytilus planulatus* also compares with many temperate *Mytilus edulis* populations in the northern hemisphere. These populations typically display gametogenic development during the winter with extended or multiple spawnings during the spring and summer months (Seed, 1969a; 1976; Lutz et al., 1980; Lowe et al., 1982; Emmett et al., 1987; Page and Hubbard, 1987; Jamieson, 1989; Villalba, 1995; Honkoop and van der Meer, 1998). Protracted reproductive periods with repeated spawnings during the spring and summer is characteristic of many natural and cultivated mussels growing under particularly favourable nutrient conditions (Seed, 1969a; 1976; Seed and Suchanek, 1992). In contrast, some populations may exhibit only a single short spawning period, sometimes lasting only a few weeks (Seed, 1969a; Bayne and Worrall, 1980; Zandee et al., 1980; Kautsky, 1982b; Rodhouse et al., 1984a; 1984b; Thompson 1984a; Thorarinsdottir, 1996). Food shortages during the summer period may explain the lack of secondary spawnings in many of these instances (Kautsky, 1982b; Seed and Suchanek, 1992). Under extreme nutritive and/or temperature stress, some populations may actually fail to initiate gametogenesis or undergo resorption of mature gametes where spawning is foregone (Bayne, 1975; Schluter and Josefsen 1994). Other populations may slowly dribble gametes continuously over an extended part of the year; a pattern typically associated with less cyclical environments (Seed and Suchanek, 1992).

In contrast to Tasmanian mussels, *Mytilus planulatus* spawns earlier (late autumn-winter) in mainland Australia (Wilson and Hodgkin, 1967, MacIntyre et al., 1977).

Such a pattern is consistent with *Mytilus* populations in other parts of the world. Records summarised by Seed (1976) show, for northern hemisphere species, mussels from warmer southern waters generally spawn earlier than conspecifics living further north. Also contrasting with the Tasmanian situation, the summer months are a reproductively inactive period for mainland *Mytilus planulatus*, with a high percentage of individuals in the resting phase (Wilson and Hodgkin, 1967). This may be a negative response to high water temperatures, as gonad redevelopment in the mainland populations do not commence until the end of summer when water temperatures fall below 21°C. Low food availability during summer might also inhibit gonad development in mainland populations, but these details are not provided. Some authors (Lubet and Aloui, 1987 cited in Seed and Suchanek, 1992) have suggested that a 'temperature window' may exist. Outside this window, gametogenesis declines or does not occur. Inside, the reproductive strategy depends largely on food availability. This window presumably varies according to the temperature range normally experienced by a mussel population and to which it is therefore adapted.

Evidence of some spawning individuals at the start of the trial (March-April 1995) suggests that some individuals were sexually mature within the first year of settlement, at a size of approximately 55 mm. It is known that *Mytilus edulis* can become sexually mature in its first year; the size at which maturity occurs depends largely on local growth rates (Seed and Suchanek, 1992). It is unknown if the individuals identified as spawning in March-April 1995 represented the "tail-end" of a major spawning prior to March, or an asynchronous spawning of a few individuals. Past studies of reproductive development in Tasmanian mussels do not clarify this uncertainty, as they either examined adult mussels (65-80 mm shell length, Dix and Ferguson, 1984) or do not provide age/size structure corresponding to the observed gonad condition (MacIntyre et al., 1977). Overspat that settled on the mussel lines during the present study were sampled and measured for shell length and whole wet weight, but not dry meat weight or condition: parameters that may have identified a spawning event within the first year of settlement. However, a major spawning prior to the start of the trial is not improbable. Wilson and Hodgkin (1967) suggest the majority of young *Mytilus planulatus* in mainland Australia spawn when they are 11 months old and over 3.0 cm in length; both of these conditions were exceeded by stocked mussels in March 1995. Also, a significant increase in tissue glycogen content between March and April 1995 may have been indicative of a recent spawning event; significant increases in glycogen content were observed during the post-spawning periods in September 1995 and February - May 1996.

Male mussels tended to be in a more advanced stage of gametogenesis than females at any particular time. Seed (1969a) and Kautsky (1982b) reported a similar

observation. These authors suggest this apparent difference is probably a consequence of the arbitrary scheme of classifying gonad condition rather than any chronological difference in the rates of development, since females in their studies (and in the present study) often appeared to spawn before the males. However, Seed and Suchanek (1992) suggest that it is not improbable that egg production, with their large yolky reserves, occurs at a slower rate than sperm production. This is supported by Newell et al. (1982), reporting that male *Mytilus edulis* display a higher gamete volume fraction, a less subjective measure of gonad development, than females during the course of maturation.

### 7.4.2 Spawning cues

There were no obvious environmental cues that may have triggered spawning during this study. A wide range of exogenous factors influencing gametogenesis and spawning in *Mytilus* has been suggested, temperature and food supply being particularly important. Suggested spawning triggers include rising, falling, or fluctuating water temperatures; physical stimulation (eg. stormy weather, harvesting); lunar phase; tidal fluctuations; food supply; salinity changes; and the presence of gametes in the water column (Seed and Suchanek, 1992). It is possible any factor that is stressful to the mussel may be a spawning trigger (Ross and Nisbet, 1990). In field populations, the difficulty in demonstrating a causal relationship between single environmental variables and spawning is that many of these variables co-vary or interact, sometimes synergistically. Environmental cues may also interact with endogenous factors in a complex and perhaps synergistic fashion to control the initiation of the gametogenic cycle and synchronise spawning. Therefore, the relative contribution of any one factor in stimulating spawning remains uncertain (Seed and Suchanek, 1992).

In this study, the environmental conditions during the two main spawning events were markedly different. The August 1995 spawning was characterised by low water temperatures (July-August mean 9.3°C), low food levels ($\approx$ 2.0 mg POM$\cdot l^{-1}$ and 0.5 $\mu$g chlorophyll-a$\cdot l^{-1}$) and a short photoperiod ($\approx$ 10 h). No obvious fluctuations in measured environmental parameters were observed during this period except for a slight rise in temperature from 9°C in July to 10°C in August and photoperiod increasing from 9 h to 10.5 h. The August spawn preceded the spring phytoplankton bloom by several weeks. Therefore, it seems unlikely that temperature or food supply were significant spawning cues on their own. Environmental conditions during the February 1996 spawn were characterised by markedly higher water temperatures, higher food levels and a longer photoperiod than in August 1995. This second spawn

corresponded with slight temperature (17.5°C - 15.5°C) and salinity (36‰ - 32‰) fluctuations, higher food levels (≈ 3.0 mg POM·l$^{-1}$ and 3.5 µg chlorophyll-a·l$^{-1}$), decreasing photoperiod (15 h to 14 h) and stormy weather conditions. Spawning triggers may be complex, involving other exogenous (eg. heat-stable metabolites released from certain species of phytoplankton; Starr et al., 1990) and endogenous (eg. "ripeness" or minimum fraction of gamete biomass to gonadal or somatic biomass; Ross and Nisbet, 1990; Brylinsky and Sephton, 1991) factors; therefore, the environmental conditions mentioned are only observations and may not necessarily represent spawning cues.

### 7.4.3 Glycogen Content

The seasonal cycle of storage and utilisation of glycogen reserves in adult bivalves is closely related to the annual reproductive cycle and to the available food supply (Gabbott, 1975; Zandee et al, 1980; Bayne et al., 1982; Bressan and Marin, 1985; Barkati and Ahmed, 1990; Gabbott and Peek, 1991; Villalba, 1995). Gonad development involves intense metabolic activity, including the storage of large amounts of lipid in the developing eggs. The accumulation of lipids may take place at the expense of stored glycogen reserves, taken directly from ingested food, or both.

Two bivalve strategies of utilising energy to build up gonadal tissue have been described (Gabbott, 1975; Bayne, 1976; Honkoop and van der Meer, 1998). The "conservative strategy" uses energy from nutrients stored during the summer-growing season, mainly in the mantle, to build up the gonads over the winter months. Therefore, gametogenesis is buffered from periods of low winter food availability by nutrient reserves. This is followed by a spawning in the spring, giving both larvae and adults the opportunity to exploit the spring bloom, which is characteristic of temperate coastal waters. Spring spawning maximises the probability of successful recruitment of new larvae and allows for maximum rates of energy acquisition by adults for the following reproductive season. The "opportunistic strategy" is more closely linked with the current food supply, allowing individuals to capitalise on particularly favourable environmental conditions such as the spring to autumn period. During these favourable conditions, energy from ingested nutrients, temporarily accumulated in the digestive gland, is used directly for gametogenesis; stored nutrients are only used to survive periods of food scarcity.

Apparently, *Mytilus* species can utilise both reproductive strategies, depending upon the particular environmental regime (Lowe et al., 1982; Newell et al., 1982; Rodhouse et al., 1984a). Many populations follow a conservative strategy, whereby spring

spawnings allow the developing larvae to exploit the spring phytoplankton bloom. In some populations, the spring spawning is followed by further opportunistic spawnings, fuelled by energy resources accumulated concurrently with gametogenesis. Both spawning strategies are indicated in the present study, whereby glycogen reserves were depleted during the autumn-winter period leading up to the first major spawning in August 1995 (70% GC loss from April 1995). However, considerably less glycogen loss was observed during the spring-summer period leading up to the second major spawning in February (28% GC loss from October 1995 to February 1996). This suggests that food ingested during the spring-summer may have been directly utilised as an energy source to fuel gametogenesis, supplemented somewhat by glycogen stores. Most of the tissue glycogen loss in the inter-spawning period was in November-December 1995, corresponding to periods of low food supply (indicated by low chlorophyll-a and POM concentrations, see Section 4). The direct utilisation of absorbed nutrients from spring-summer feeding fuelling gametogenesis has been reported in several studies (Walne, 1970; Newell et al., 1982; Rodhouse et al., 1984a; Villalba, 1995). Also, the rapid recovery of glycogen stores observed after spawning has similarly been noted by others (Bayne, 1975; Dare and Edwards, 1975; Hickman and Illingworth, 1980; Emmett et al., 1987; Gabbott and Peak, 1991; Griffiths 1977 in Jaramillo and Navarro, 1995).

### 7.4.4 Spawning losses and reproductive effort

Estimates of biomass losses due to spawning can be measured directly by inducing individuals to spawn and weighing the dry gametes, or indirectly by determining the difference between meat or mantle dry weights before and after spawning (Bayne et al., 1983; Crisp, 1984). As the two methods have been shown to be comparable (Bayne et al., 1983), the results obtained by the indirect method in this study are probably reasonable estimates of spawning losses. However, it should be emphasised that the indirect method assumes biomass losses other than loss of gametes concurrent with spawning activity (eg. metabolic loss due to starvation) are minimal.

The absolute biomass losses incurred during each of the two major spawning events were not significantly different (mean loss 0.65 g AFDMW·mussel$^{-1}$). However, spawning losses relative to pre-spawning AFDMW in August 1995 (31%) were greater than in February 1996 (23%). This decrease in fecundity, relative to size, contrasts with studies reporting either an isometry between gamete production and soft tissue weight (Griffiths, 1977 in Kautsky, 1982b; Bayne et al., 1983) or an increase in size-relative fecundity (Thompson, 1979; Kautsky, 1982b). Perhaps the lower percentage reproductive loss in February 1996 may have been due to partial spawning.

Reproductive output is influenced by environmental variables such as temperature, food supply and tidal exposure, since these will broadly determine levels of net production (Seed and Suchanek, 1992). Therefore, fecundity estimates tend to display significant spatial and temporal variation. For example, spawning losses in this study (31% and 23% of pre-spawning AFDMW) were greater than those of six natural U.K. mussel populations studied by Bayne et al. (1983), which themselves displayed a ten-fold difference in gamete losses (2-20% weight loss on spawning). On the other hand, fecundity in this study was less than others, some reporting up to 60% spawning loss (Table 7.2).

**Table 7.2.** Reported spawning losses of *Mytilus* as a percentage of pre-spawning ash-free dry meat weight. Spring spawn[a], late summer-autumn spawn[b].

| Spawning weight (g) | Spawning loss (%) | Reference |
|---|---|---|
| 2.0[a]; 3.0[b] | 31[a]; 23[b] | This study |
| 0.5[a]; 2[b] | 20 - 30[a]; 40 - 60[b] | Thompson, 1979 |
| 0.1[a]; 0.14[b] | 38[a]; 52[b] | Kautsky, 1982b |
| 1.0 | 2 - 20 | Bayne et al., 1983 |
| 0.1 - 1.0 | 8 - 43 | Rodhouse et al., 1984a;b |
| 1.4 | 43 | Thompson, 1984a |

In general, the proportion of an organism's energy budget allocated to gamete production, referred to as reproductive effort (RE), increases asymptotically with age for many bivalve species (Bayne, 1976; Browne and Russell-Hunter, 1978; Griffiths and King, 1979; Thompson, 1979; Bayne et al., 1983; Thompson, 1984b; Rodhouse et al., 1984b). Estimates of RE range from 0% for 0 to 1 year old mussels to 94-100% for 4 to 12 year old mussels (Bayne, 1976; Thompson, 1979; 1984b). However, as with most growth parameters, reproductive effort can significantly vary between different populations, even those of similar size/age, according to habitat and the prevailing environmental conditions (Bayne and Worrall, 1980; Bayne et al., 1983; Thompson, 1984b). It is not known if the mussels in the present study conformed to the trend of increasing reproductive effort with age/size, largely because the pre-trial history of the mussels is uncertain (ie. spawning prior to March 1995 is suspected but remains unknown). Further, the calculation of RE from September 1995 - February 1996 cannot be determined precisely. Biomass losses, through asynchronous spawning individuals during this period, were likely masked by AFDMW growth of the larger percentage of non-spawning individuals.

Reproductive effort and the subsequent loss of biomass represented at least half of the total aquaculture production from March 1995 to May 1996. Of most relevance to this

study, no significant differences among sites were indicated in the allocation of energy resources to reproduction. If higher levels of available food increases reproductive effort (Bayne and Worrall, 1980; Newell et al., 1982; Bayne et al., 1983), the present results indicate that mussels growing closest to the fish cages (sites I and II) received no nutritional advantage over those mussels growing further away (sites III and IV)..

In summary, as the gametogenic cycle, fecundity and reproductive effort are largely determined by food availability, further evidence has been provided showing mussels cultured adjacent to the fish cages did not gain any nutritional advantage over those mussels grown distant from the farm. This conclusion is based on: no difference in gametogenic development between sites I and IV; all sites displayed two major spawnings with significant biomass losses in August 1995 and February 1996; and there were no significant differences in glycogen content, fecundity or reproductive effort among sites. The timing of major spawning events and subsequent loss of condition may have important commercial implications for mussel farmers. These include: choosing harvesting periods prior to spawning, ensuring mussels are in a marketable condition and circumventing biomass production lost to the environment as gametes; and timing the deployment of spat collectors, allowing for an appropriate period of "conditioning" in advance of major spawning periods.

# 8. Density and Survival

## 8.1 Introduction

Density is an important factor influencing bivalve growth and survival at various spatial scales, ranging from whole basins to individual culture units (Heral, 1993; Frechette and Bacher, 1998). As such, relationships of density with growth and survival are essential component in modelling carrying capacities of coastal areas for bivalve culture. On a smaller scale, bivalve culturing practices include ongoing density reductions throughout the various stages of hatchery and growout cycles for maximising growth and survival rates (Hickman, 1992). Density-growth relationships, such as biomass-density and production-density curves may be employed to determine optimal site-specific stocking densities to maximise production according to local environmental conditions (Frechette et al., 1996; Frechette and Bacher, 1998). Recognising that a density-growth/survival relationship may have been significant in this study, sample densities were monitored to determine if the stocking density of socks rather than the main treatment effects of site and depth might have influenced observed growth or survival.

Reduced levels of food consumption have been identified as a significant cause of bivalve mortality, particularly during post-spawning periods when nutrient reserves in the mantle are low (Inzce et al., 1980; Worral and Widdows, 1984; Seed and Suchanek, 1992). As such, survival rate was considered a useful criterion to assess the relative potential of the Aquatas salmon farm as a bivalve culture site. Superior survival rates of mussels grown adjacent to the fishcages (sites I and II) might be an indication of a nutritional enhancement over off-farm sites (III and IV).

## 8.2 Methodology

Samples of 25 cm sections of sock stripped of mussels were collected from the top (1 m depth) and bottom (5 m depth) of each selected sock. The total density (TD) and survival density (SD) of samples were determined by:

TD = the total no. survivors + mortalities

SD = the total no. survivors

The number of mortalities within each sample was estimated from the number of empty shells and shell fragments. To allow for comparisons with other studies, sample densities are expressed as the number of mussels per metre of sock (mussels$\cdot m^{-1}$). Sample densities were analysed using three-factor ANOVA, with SITE, DEPTH and TIME (Month) as fixed factors after confirmation of homogeneity

of variance (Cochran's test, Sokal and Rohlf, 1995). To determine if density had a negative effect on mussel growth, correlations between sample densities (TD and SD as the X variables) and their corresponding mean shell length, whole wet weight, individual biomass and survival rate (as Y variables) were performed on monthly data from April 1995 through May 1996.

With the large number of samples (672 samples) collected over the trial period from 168 mussel socks, considerable spatial and temporal variation in densities was apparent (see Results). It was concluded that droppers were not uniformly stocked (either between or within droppers) and the densities of droppers sampled in March 1995 were not representative of all socks subsequently sampled. Therefore, as different droppers were sampled on successive months, monthly survival rates could not be estimated from sample survival densities relative to the initial stocking densities of March 1995. As such, survival rates were estimated from the number of survivors in each sample relative to the total number of survivors plus mortalities (% survival = sample SD x 100 / sample TD). The disadvantage of this method is that it does not account for mortality losses due to "dropoffs" (mussels becoming dislodged and dropping off of socks during grow-out).

Two-factor ANOVA was performed to assess the effects of SITE and DEPTH (n = 6 samples·site$^{-1}$·depth$^{-1}$) on survival rates, after arcsine square root transformation and confirmation of normality (Shapiro-Wilk test) and homogeneity of variance (Cochran's test), at four key sampling dates (March 1995, August 1995, February 1996 and May 1996).

## 8.3 Results

### 8.3.1 Density

Although every effort was made to be consistent in the stocking, handling and sampling of mussel socks, sample densities were not uniform (Figs. 8.1 and 8.2). Sample total densities ranged from 44 to 256 mussels·m$^{-1}$ with an overall trial mean of 148 ± 1.2 mussels·m$^{-1}$ (n = 672 samples). Three-factor ANOVA (site x depth x time) revealed significant differences between sites, depths and sampling times (all at $P < 0.0001$). Although a site x time interaction was significant ($P < 0.01$), an in-depth analysis to identify all possible interactions was not warranted (1540 possible comparisons). However, a few general observations can be made. Total densities from site I and II displayed minimal temporal variation over the trial period Fig. 8.1-A. This indicates that droppers at these two sites were stocked uniformly and experienced

few dropoff losses. There were no significant differences in mean TDs between all four sites on five sampling occasions (March 1995, July 1995, January 1996, March 1996 and May 1996; $P < 0.05$). Densities of samples collected from April 1995 to December 1995 tended to be higher at site IV than at the other three sites, particularly sites I and II ($P < 0.05$).

Sample TDs collected from the top of socks (overall mean 136 ± 1.5 mussels·m$^{-1}$, n = 336 samples) were significantly lower ($P < 0.0001$) than TDs collected from the bottom of socks (overall mean 160 ± 1.6 mussels·m$^{-1}$, n = 336 samples).

The variation in survivor densities (Fig. 8.2) was similar to that of TDs, with significant site, depth and time differences ($P < 0.0001$) and site x time interaction ($P < 0.05$) indicated. A general decline in SDs was evident towards the final stages of the trial. It was not possible to distinguish *in situ* mortality from losses due to dropoffs.

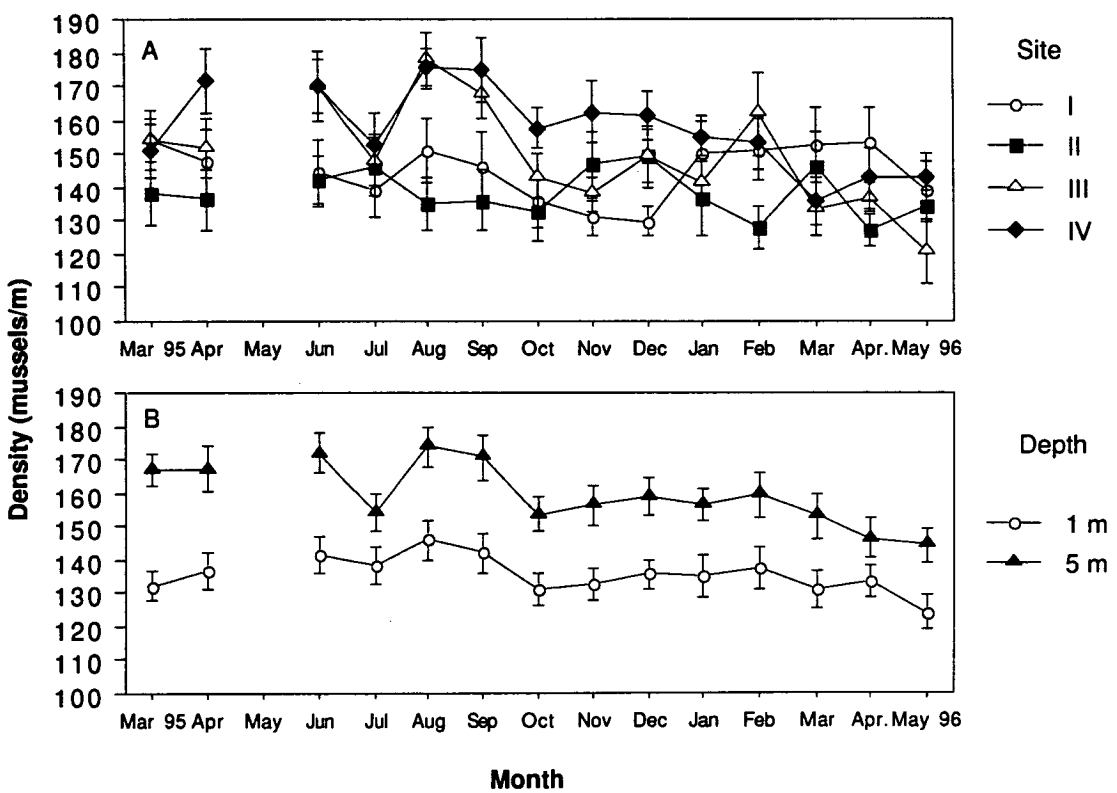


**Figure 8.1.** (A) Total density of mussel samples collected from 25 cm sections of droppers (mean ± SE mussels·m$^{-1}$) at each site (n = 12 samples per site) and (B) at each growing depth (n = 24 samples per depth). Total density = survivors + mortalities remaining on droppers.

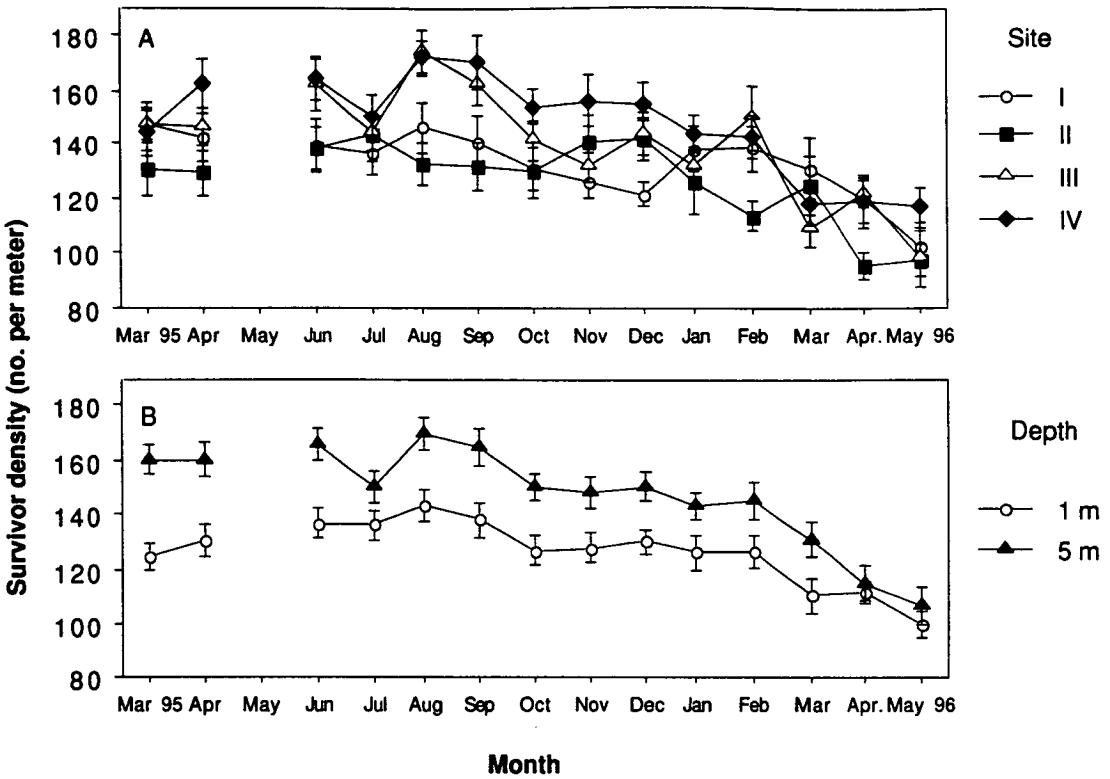


**Figure 8.2.** (A) Survivor density of mussel samples collected from 25 cm sections of droppers at each site (mean ± SE mussels·m$^{-1}$, n = 12 samples per site) and (B) at each growing depth (n = 24 samples per depth).

Correlation coefficients describing the relationships between monthly sample densities (TD and SD) and their corresponding sample mean shell length, whole wet weight, individual biomass and survival rate were generally poor; r-values were predominantly < 0.5, not consistently significant ($P < 0.05$), nor consistently negative (Tables 8.1 and 8.2).

**Table 8.1.** Correlation analysis of monthly sample total densities (TD, sum of survivors and mortalities per meter of sock) against corresponding mean shell lengths (mm), whole live wet weight (WLW), individual biomass and survival rates. r = correlation coefficient and P is the probability of r $\neq$ 0 (n = 48 samples per month).

| TD vs: | Length | | WLW | | Biomass | | Survival | |
|---|---|---|---|---|---|---|---|---|
| | r | Prob | r | Prob | r | Prob | r | Prob |
| April | -0.539 | <.0001 | -0.615 | <.0001 | -0.485 | 0.001 | -0.100 | 0.503 |
| June | -0.349 | 0.015 | -0.404 | 0.004 | -0.265 | 0.068 | 0.092 | 0.538 |
| July | 0.296 | 0.041 | 0.173 | 0.242 | 0.108 | 0.468 | -0.162 | 0.272 |
| August | -0.136 | 0.357 | -0.276 | 0.058 | -0.408 | 0.004 | -0.210 | 0.152 |
| September | 0.386 | 0.006 | 0.298 | 0.039 | -0.025 | 0.867 | 0.020 | 0.891 |
| October | -0.190 | 0.198 | -0.228 | 0.120 | -0.310 | 0.031 | 0.299 | 0.038 |
| November | 0.264 | 0.069 | 0.185 | 0.211 | 0.219 | 0.135 | 0.094 | 0.526 |
| December | -0.066 | 0.656 | -0.074 | 0.618 | 0.002 | 0.991 | 0.100 | 0.500 |
| January | 0.523 | <.0001 | 0.477 | 0.001 | 0.388 | 0.006 | 0.063 | 0.673 |
| February | 0.300 | 0.038 | 0.212 | 0.149 | 0.159 | 0.283 | 0.054 | 0.717 |
| March | 0.547 | <.0001 | 0.513 | 0.000 | 0.494 | 0.000 | 0.390 | 0.006 |
| April | -0.109 | 0.465 | -0.155 | 0.294 | -0.004 | 0.979 | -0.078 | 0.598 |
| May | 0.385 | 0.007 | 0.351 | 0.014 | 0.386 | 0.006 | -0.054 | 0.716 |

**Table 8.2.** Correlation analysis of monthly sample survival densities (SD, number of mussels per meter of sock) against corresponding mean shell lengths, whole live wet weight (WLW) and individual biomass. r = correlation coefficient and P is the probability of r $\neq$ 0 (n = 48 samples per month).

| SD vs: | Length | | WLW | | Biomass | |
|---|---|---|---|---|---|---|
| | r | Prob | r | Prob | r | Prob |
| April | -0.530 | <.0001 | -0.603 | <.0001 | -0.457 | 0.001 |
| June | 0.148 | 0.318 | 0.065 | 0.663 | 0.122 | 0.413 |
| July | -0.113 | 0.446 | -0.253 | 0.082 | -0.385 | 0.007 |
| August | -0.092 | 0.536 | -0.094 | 0.528 | 0.003 | 0.982 |
| September | 0.319 | 0.027 | 0.239 | 0.102 | 0.181 | 0.220 |
| October | 0.604 | <.0001 | 0.554 | <.0001 | 0.478 | 0.001 |
| November | 0.329 | 0.022 | 0.191 | 0.193 | 0.128 | 0.389 |
| December | -0.364 | 0.011 | -0.425 | 0.002 | -0.289 | 0.046 |
| January | 0.607 | <.0001 | 0.581 | <.0001 | 0.542 | <.0001 |
| February | 0.602 | <.0001 | 0.609 | <.0001 | 0.544 | <.0001 |
| March | 0.272 | 0.061 | 0.199 | 0.176 | 0.229 | 0.118 |
| April | -0.160 | 0.279 | -0.198 | 0.178 | -0.279 | 0.054 |
| May | 0.335 | 0.019 | 0.347 | 0.015 | -0.180 | 0.221 |

### 8.3.2 Survival

Survival rates, estimated from monthly sample SDs relative to their corresponding TDs, are presented in Fig. 8.3. The number of live mussels in the initial samples of March 1995 represented an overall mean survival rate of 95.0 $\pm$ 0.5% (n = 48 samples). No appreciable mortality was observed during the first nine months (March 1995 - December 1995), with an overall mean survival rate during this period of 96.4 $\pm$ 0.2% (n = 432 samples). Therefore, approximately 3.6% of mussels stocked in

February 1995 were assumed to be already dead. Survival declined slightly during December 1995 and January 1996 to 92.3 ± 0.7% (n = 48 samples) in February 1996. A marked decline in survival rates from February through May 1996 was evident, with overall mean survival rate in May 1996 of 77.5 ± 2.0% (n = 48 samples). This decline corresponded with the second major spawning of the trial and the period where negligible growth rates and declining condition were also observed.

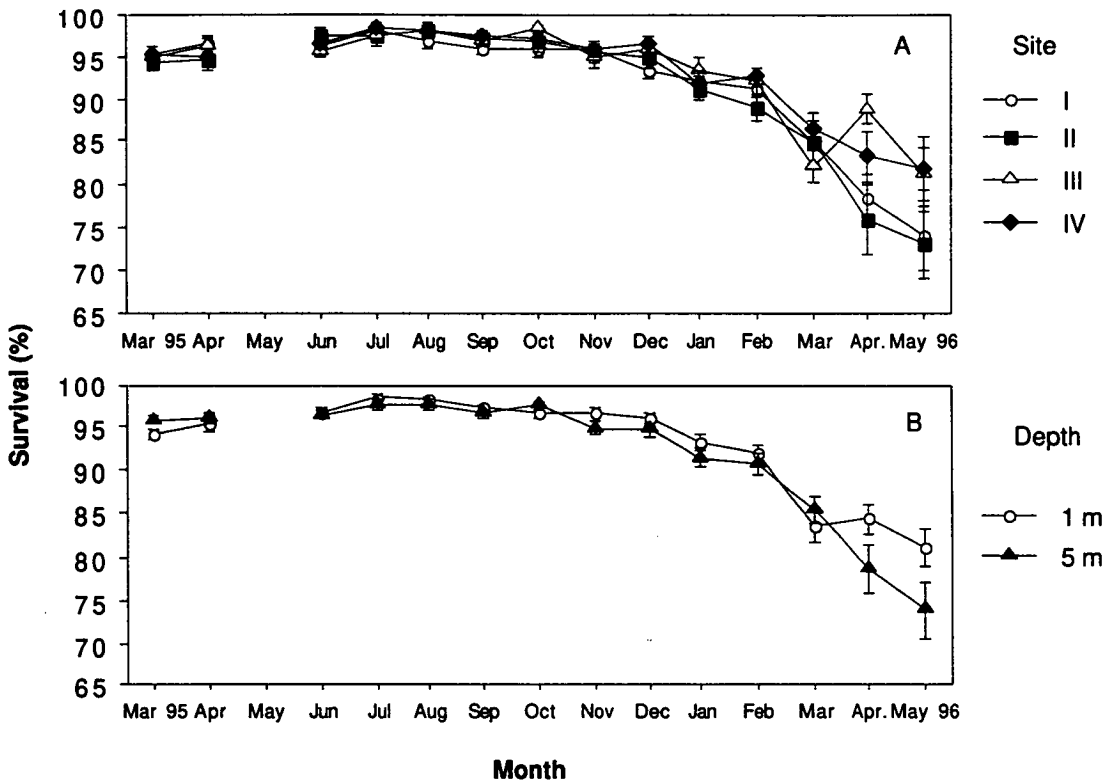


**Figure 8.3.** (A) Monthly survival rate of mussels at each site (mean ± SE%, n = 12 samples per site) and (B) at each depth (n = 24 samples per depth). Sample survival rate = SD x 100/TD.

No effects of site, depth or site x depth interactions on survival were indicated for any of the four key sampling dates ($P > 0.05$). Assuming an initial stocking mortality of approximately 3.6%, the number of survivors remaining at the end of the trial represented a net survival rate of about 81% for the trial period. However, losses due to dropoffs are not included in this figure. The actual number of survivors still attached to droppers in May 1996 relative to their initial stocking density in March 1995 (which cannot be determined with any certainty) would likely suggest a lower survival rate.

## 8.4 Discussion

### 8.4.1 Density

Sample total densities displayed considerable spatial and temporal variation. This indicates that the initial stocking densities of socks were not uniform, either between socks or within socks (depth). Variation in sample densities might be due to any or all of the following: (a) socks were not stocked with a consistent number of mussels in the first place; (b) the sections of droppers sampled (ie. 25 cm sections) might not have been measured consistently (sampling error); (c) in the latter stages of the trial, the presence of high densities of overspat may have resulted in larger-sized overspat included in some samples and/or obscured the selection of smaller experimental mussels in other samples; (d) sample sections may have been too small to "even out" variability; and (e) over time some mussels were lost due to "dropoff".

The differences in sample densities between the two depths, where both SDs and TDs at 5 m were significantly higher than at 1m, were evident from the start of the trial. Therefore, higher densities at 5m were probably due to the weight of overlying mussels forcing underlying mussels downwards into the bottom of the socks prior to attachment onto the core rope and Netlon mesh.

Although sample densities were not uniform, it appears that density had no significant effect on mean mussel growth or survival. No consistent correlations between sample densities and growth parameters (length, whole live weight, biomass) or survival were observed. Further, we have previously concluded that differences in growth (shell length, weight parameters, condition) and survival among sites and between depths were either minor or not significant. This is despite significant density differences between depths and, on many sampling occasions, among sites.

Density-dependent growth and survival rates have been reported for natural and cultivated bivalve populations (Wilson and Hodgkin, 1967; Seed, 1969b; Dare and Davies, 1975; Kautsky, 1982a; Peterson and Black, 1987; Boromthanarat and Deslous-Paoli, 1988; Peterson and Beal, 1989; Frechette et al., 1992; McGrorty and Goss-Custard, 1993; Rivonker et al., 1993; Mueller, 1996). The negative influence of density on growth is considered to be largely due to competition for the available food supply. The ability to effectively feed in crowded, anoxic conditions and the accumulation of metabolites and faecal wastes may also be implicated in impairing growth at higher densities.

Reductions in sock densities over time, through *in situ* mortalities and dropoffs, are normally observed in mussel culture studies (Loo and Rosenberg, 1983; Grant et al., 1993; Okumus, 1993; Rivonker et al., 1993; Maximovich et al., 1996) and in normal commercial practice. These reductions in survivor densities are due to competition for food but also for available space as the animals grow. Although reductions in survivor densities are expected, many studies show an eventual stabilisation of mussel densities (eg. McGrorty and Goss-Custard, 1993; Maximovich et al., 1996). Stabilisation suggests that, at some point, densities become appropriate to the available attachment area and the prevailing environmental conditions (eg. hydrographic conditions and available food). It is probably inappropriate to compare results from the above studies, as there are differences in species, age and size of stocked mussels, initial stocking densities, culture methods and environmental conditions. However, it is noteworthy that the density at which mussel numbers tend to stabilise in these different studies is in the vicinity of 300 - 600 mussels per meter of sock. With these "stabilised" sock densities in mind, the lack of a significant effect of mussel density on mean growth and survival in the present study might be due to stocking densities (TD < 256 mussels·m$^{-1}$ and SD < 228 mussels·m$^{-1}$) being below some "critical density" (eg. 300 - 600 mussels·m$^{-1}$). The critical density represents the maximum number of mussels per meter of sock that can be cultured in the prevailing environmental conditions without growth or survival being compromised. Therefore, it appears that stocking densities in this study did not exceed those that might impair mean growth or survival of mussels according to the environmental conditions (particularly available food supply) of North West Bay.

### 8.4.2 Survival

Severe drops in survival rates, due to both dropoffs and *in situ* mortalities, immediately after stocking, have been reported in several studies (Dare and Davies, 1975, Okumus 1993, Rivonker et al., 1993; Maximovich et al., 1996). However, *in situ* mortalities were not significant at all during the first nine months of this trial and obvious signs of dropoffs (clumps of mussels missing from droppers) were not encountered until February 1996. As mentioned, the initial stocking densities may have been below a "critical density" that might impair growth and survival. Declining survival did become evident after December 1995 and more pronounced after February 1996. The exact cause(s) for increasing mortalities during the latter stages of the trial is unknown. Natural mortality has been associated with senility, adverse environmental conditions, predators, parasites, disease, competition for available food and space, pollution and reproductive stress (Seed 1969b; Worral and Widdows, 1984; Seed and Suchanek, 1992; Smaal and Widdows, 1994).

Predation may have been a factor influencing mortality; but no obvious mussel predators were observed (eg. sea stars, birds, fish) except rarely-encountered sea stars. If predation was a factor, significant mortalities might have been expected throughout the trial, particularly during the earlier stages when mussels were smaller.

No adverse environmental conditions that might have influenced survival were apparent during the latter stages of the trial. In fact, temperature and POM levels were quite adequate for growth; scope for growth (SFG) estimates for January 1996 to May 1996, corresponding with the period of increasing mortality, were at maximum levels for the trial (see Section 12: Scope for Growth).

The increase in mortalities during the last few months may have merely been a consequence of "old age" or senility. Although the life span of *Mytilus edulis* has been estimated to be 17 - 24 years (Seed, 1969b; Thiesen, 1973) and *M. californianus* may live to 50 - 100 years (Seed and Suchanek, 1992), the longevity of *M. planulatus* is considerably less. The lifespan of *Mytilus planulatus* has been variously estimated to be 2 years (Dix, 1980), 2 to 3 years (Wilson and Hodgkin, 1967) and 4 to 5 years (Pregenzer, 1983). These variable lifespan estimates may reflect genetic differences between stocks. Genetic differences, observed within other *Mytilus* species, have been associated with differential mortality rates (Dickie et al., 1984; Mallet and Carver, 1989; Kautsky et al., 1990; Fuentes et al., 1994; Myrand and Gaudreault, 1995; Tremblay et al., 1998). The exact age of the experimental mussels is unknown. However, using spat growth as a guide (see Section 10), mussels in this study may have been approximately twelve months of age when stocked and approaching two years of age in February 1996 - corresponding with the expected life span suggested by Dix (1980) and the onset of increasing mortality. Poor mussel performance over the latter stages of the trial, such as declining growth rates, loss of biomass and declining condition, is often exhibited by mussel populations preceding natural senescence (Jamieson, 1989). Therefore, senility may partly explain the increased mortalities during the latter stages of the trial.

The onset of increased mortalities also corresponded with a major spawning event in February 1996 (late summer). However, no significant mortalities were observed following the August 1995 spawning (late winter). "Summer mortality" has been noted by others and is often associated with summer spawning activity (Incze et al., 1980; Worral and Widdows, 1984; Emmett et al., 1987; Jamieson, 1989; Taylor et al., 1992; Myrand and Gaudreault, 1995). Spawning is a time of metabolic stress; post-spawning mortalities may be due to reduced food rations and high temperatures at a time of high metabolic cost and low mantle nutrient reserves (Incze et al., 1980).

Tremblay et al. (1998) suggested that periodic outbreaks of 'summer mortality" may be the result of a synergistic interaction involving dietary deficiencies, high temperatures, post-spawning stress and the genetic characteristics (degree of heterozygosity) of the stock. Worral and Widdows (1984) found post-spawning mortalities were often most severe among larger mussels which have the highest reproductive effort. A higher mortality rate of larger-sized mussels with a higher reproductive effort might explain the slight decline in shell length from February 1996 to May 1996; however the overall mean loss of 1.3 mm was not statistically significant ($P > 0.05$). A higher percentage of larger mussels from the outside of the droppers falling off cannot be discounted as a partial cause of the decline in shell length.

As mentioned, the last few months of the trial corresponded with temperatures and levels of food availability that predicted maximum scope for growth. However, along with increasing mortality, mussel growth effectively ceased during this period. It is possible that several other features may have also contributed to, or exacerbated, the effects of senility and reproductive stress, resulting in reduced growth and increased mortality. Although the density of the socked mussels probably had minimal effect on growth and survival, additional competition for food may have come from overspat settlements, other biofouling organisms and possibly pea crabs.

There must have been considerable competitive pressure for the available food supply during the latter stages of the trial. At the conclusion of the trial, biomass of overspat was approximately tenfold that of the experimental mussels' biomass. Negative effects on mussel growth and survival due to oversettlements have been noted by others (Wilson and Hodgkin, 1967; Mason, 1972; MacIntyre et al., 1977; Hickman, 1992). As previously mentioned, the reduction in growth is largely the result of competition for food as well as the accumulation of metabolites and faecal wastes. Mussels within the centre core of droppers were trapped within an extremely fouled and anoxic environment of "faecal mud". This is where mussels tended to be stunted in size and where much mortality was found, possibly due to suffocation or starvation. Others have noted the negative effects of "mussel mud" on growth and survival (Dare, 1976; Ceccherelli and Rossi, 1984; Jamieson, 1989; Newell, 1990).

The presence of several species of filter-feeding fouling organisms (particularly ascidians and sponges) growing amongst the mussels would have also competed for a share of the suspended food supply. Epifauna overgrowing the shell gape and siphonal areas may also have restricted the ability of some mussels to feed. Many instances of shell gape occlusion were observed during sampling, particularly by an insidious species of sponge (identity unknown) that formed dense mats within the mussel assemblage.

Other fouling organisms that may have been detrimental to mussel growth and survival included two species of algae; a green filamentous alga, *Chaetophora linum*, and a red macrophyte, *Mycodia aciculare*, became extremely dense on many droppers during the latter months (Fig. 6.5). Although not competing for food, these algal foulants may have altered the flow of water around the droppers, limiting the penetration of food particles to the underlying mussels and impairing waste removal. Negative effects of epiphytic biofouling on mussel growth and fitness have been noted by others (Mason, 1972; Paine, 1976; MacIntyre et al., 1977). Normal husbandry practice includes the removal of ascidians and other fouling organisms from culture units during thinning operations (Mason, 1972; Hickman, 1992).

A variety of diseases and parasites have been associated with reduced mussel growth and survival (Pregenzer, 1983; Hickman, 1992; Bower et al., 1994). Some previous studies have not detected any incidence of disease outbreaks or pathogenic organisms in Tasmanian mussels (Wilson et al., 1993; Pethybridge, 1994). High incidences of trematode metacercaria (*Gymnophallus* sp.) in mussels from North West Bay have been reported (Pregenzer, 1983; MacIntyre et al., 1977); but these larvae are not considered to affect mussel growth (Bower et al., 1994). As pathogenic agents were not monitored during this study, disease as a factor influencing growth and survival cannot be tested; nor can it be discounted.

Pinnotherid crabs (pea crabs) live on the gills of host bivalves, feeding on food-laden mucous strings and may cause damage to the gills in the process (Pregenzer, 1978). Infestation by pea crabs has been implicated in reducing filtration rates, food intake, oxygen consumption, growth, condition and reproductive effort in bivalves (MacIntyre et al., 1977; Pregenzer, 1978; 1979; 1981; Bierbaum and Ferson, 1986; Haines et al., 1994; Tablado and Gappa, 1995; O'Beirn and Walker, 1999). In this study, the incidence of pea crabs (*Pinnotheres hickmani* Guiler 1950) increased from negligible levels at the start of the trial to maximum levels in February 1996 (overall mean 27%, range 13% at site I to 45% at site IV; Section 9): the period when spawning and increasing mortalities were observed. It is not possible to quantify the extent of pea crab-parasitised mortalities - there is no way of determining if mortalities harboured pea crabs prior to death. However, the additional stress of pea crabs on mussels, already in a stressed condition from spawning, may have contributed to reduced fitness, recovery and, ultimately, survival during the last few months.

A wide range of environmental contaminants (eg. petroleum hydrocarbons, organochlorines, pesticides, organo-metals and heavy metals), often at very low concentrations, have been associated with negative effects on the physiology, growth

and survival of mussels (Widdows et al., 1984; Widdows and Johnson, 1988; Viarengo and Canesi, 1991; Widdows and Donkin, 1992; Smaal and Widdows, 1994). However, as various contaminants in North West Bay were not monitored, the effect of any of these materials on mussel performance and survival cannot be tested nor speculated upon.

In summary, within-sock mussel densities were quite variable among sites and between depths. However, there is no evidence suggesting that growth or survival of mussels in higher density socks was significantly different than mussels cultured in lower density socks. Under the prevailing conditions of food availability and temperature, it appears that sock stocking densities may have been below levels that might negatively influence growth.

A combination of factors such as senility, reproductive stress during a period of higher water temperatures, and a restricted food supply due to competition or interference from fouling organisms and/or pea crab parasitism, may have been implicated in the lack of post-spawning recovery and increasing mortality during the last months of the trial. However, the observed mortality also appears quite similar to the "summer mortality" reported for *Mytilus edulis* in the Northern Hemisphere (Emmett et al., 1987; Taylor et al., 1992; Myrand and Gaudreault, 1995); which has been attributed to a post-spawning energy deficiency (Incze et al., 1980; Worral and Widdows, 1984). Uniformly distributed survival rates suggest that none of the culture sites provided mussels with a sufficient nutritional enhancement to overcome any deficiency. Therefore, in terms of survival, there appears to be no advantage in culturing mussels within the salmon farm over other sites within North West Bay; however, the extent to which the available food supply was restricted by overspat and other fouling organisms is unknown. Similar results are reported in other studies investigating salmon-mussel polyculture, whereby proximity to salmon cages has no effect on survival (Taylor et al., 1992; Okumus, 1993). Survival data from this study does reinforce the prudence of harvesting mussels prior to summer spawning activity. In so doing, farmers avoid stock losses due to post-spawning mortalities and harvest a higher quality product of mussels in peak condition.

# 9. Pea Crab Infestation

## 9.1 Introduction

Pinnotherid crabs (pea crabs) are a group of brachyuran crabs; following a planktonic larval stage they eventually occupy the mantle cavity of a bivalve host. There, they live on the host's gills, feeding on food-laden mucous strings destined for the labial palps. Although formerly considered to be commensals, there is increasing evidence that some species of pinnotherid crabs may produce adverse effects on hosts. Negative effects include: damage to gills, gonads and other tissues; decreased pumping, filtration and oxygen consumption rates; and therefore reduced growth, condition and reproductive effort (MacIntyre et al., 1977; Pregenzer, 1978; 1979; 1981; Bierbaum and Ferson, 1986; Bierbaum and Shumway, 1988; Haines et al., 1994; Tablado and Gappa, 1995; O'Beirn and Walker, 1999). Considering the deleterious effects of pea crabs on mussel growth and the high levels of infestation reported in some Tasmanian populations (eg. > 95% Pregenzer, 1983), pea crab incidence in experimental mussels was monitored. The incidence of pea crabs commonly inhabiting Tasmanian bivalves (*Pinnotheres hickmani*, Guiler 1950) was monitored in this study to assess: (a) pea crab distribution amongst the experimental sites and growing depths, and (b) the influence of pea crabs on mussel growth, condition and survival.

## 9.2 Methodology

Fourty-eight mussel samples were collected from the four experimental longlines each month (Section 3.3). Twenty mussels from each sample were selected for length, weight, and condition measurements. The presence of pea crabs was recorded as each mussel was opened and soft tissue removed. Sample pea crab incidence (PC) is expressed as the percentage of opened mussels harbouring one or more pea crabs:

PC (%) = no. mussels harbouring pea crabs x 100 / 20 mussels.

Two-factor ANOVA was performed to assess SITE and DEPTH differences of pea crab incidence (n = 6 samples·site$^{-1}$·depth$^{-1}$), after arcsine square root transformation and confirmation of normality (Shapiro-Wilk test) and homogeneity of variances (Cochran's test), at four key sampling dates (March 1995, August 1995, February 1996 and May 1996). Correlation analysis was employed on separate monthly data to examine the relationships between transformed sample %PC and sample growth parameters (shell length, whole live weight, ash-free dry meat weight, condition, total biomass) and survival.

## 9.3 Results

At the start of the trial few pea crabs were encountered, with an overall mean pea crab incidence (%PC) of less than one percent (Fig. 9.1). Infestation increased at all sites, peaking in January-February 1996. Differences among culture sites were evident for most of the trial period, with a trend of increasing %PC with distance from the fish cages. The highest percentage of crab-infested mussels occurred at site IV in February 1996 (mean 44.6 ± 4.0%, n = 12 samples), compared with 13.3 ± 3.3% at site I. A marked decline in crab infestation was observed at sites II and IV between February 1996 and March 1996, but this decline was largely recovered in April 1996.

Significant differences in %PC among sites were indicated for the August 1995 ($P < 0.01$), February 1996 ($P < 0.0001$) and May 1996 ($P < 0.01$) samples, but not between growing depths ($P > 0.05$). A general trend of increasing %PC with distance away from the farm was evident, whereby the greatest differences in %PC were between sites I and IV for all three key sample dates (Fig. 9.2). Although a significant site x depth interaction was indicated for the August 1995 and February 1996 samples (both at $P < 0.05$), the interaction was not consistent between the two sampling dates and not considered to be of any importance.

Correlation analysis did not provide evidence of any adverse effects of pea crabs on mussel growth or survival (Table 9.1). Correlations were rarely significant ($P < 0.05$) and correlation coefficients were generally low ($r < 0.5$).

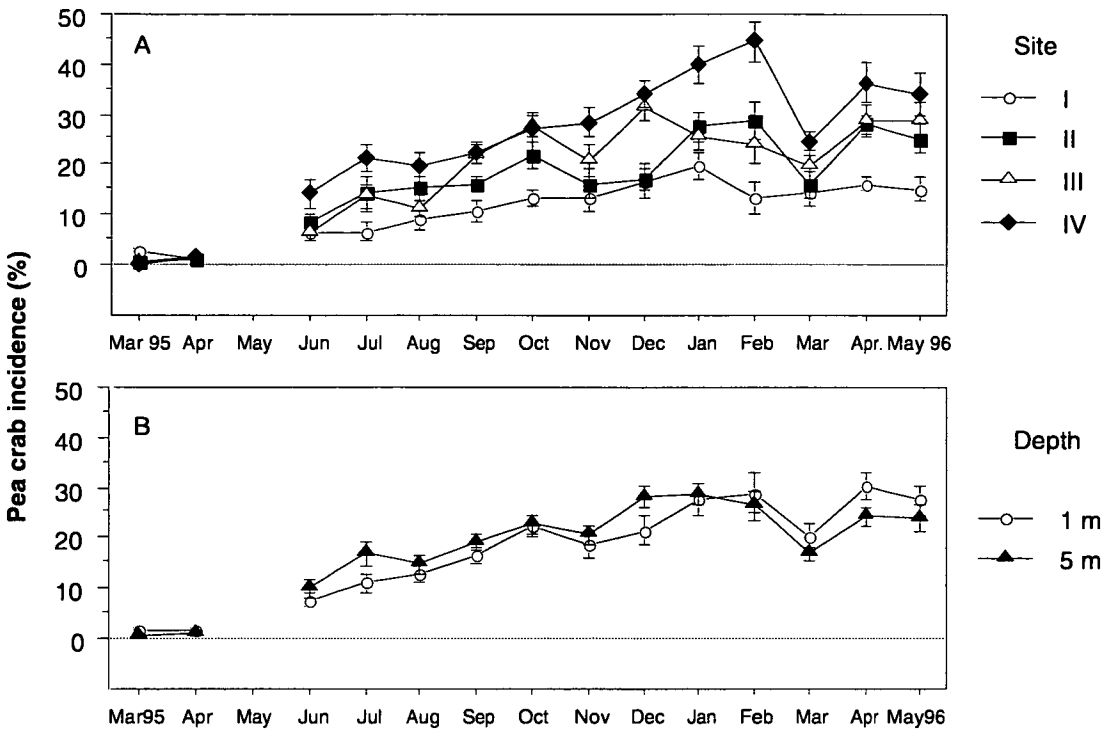


**Figure 9.1.** (A) Incidence of mussels harbouring one or more pea crabs at each site (mean ± SE%, n = 12 samples per site) and (B) at each growing depth (mean ± SE%, n = 24 samples per depth). Each sample = 20 mussels.

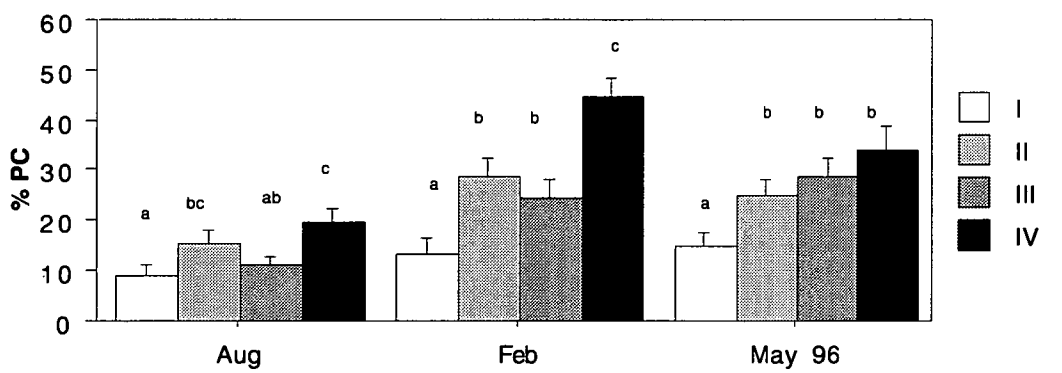


**Figure 9.2.** Pea crab incidence (%PC) of samples at each site (mean ± SE%, n = 12 samples per site per month) collected in August 1995, February 1996 and May 1996. Sites not sharing a common letter within each monthly grouping are significantly different ($P < 0.05$).

**Table 9.1.** Mean monthly pea crab infestation across all sites (± SE%, n = 48 samples per month) and monthly correlation coefficients (r) of sample %PC with sample mean length, whole live weight (WLW), biomass, condition (CIgrav), and survival rate (%). P is the probability of $r \neq 0$.

| Month | %PC | SE | Length | | WLW | | Biomass | | Condition | | Survival | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | r | P | r | P | r | P | r | P | r | P |
| April | 1.5 | 0.4 | 0.007 | 0.962 | 0.024 | 0.872 | -0.097 | 0.512 | -0.273 | 0.060 | 0.087 | 0.557 |
| June | 8.9 | 1.1 | -0.021 | 0.891 | 0.077 | 0.606 | 0.038 | 0.797 | -0.050 | 0.736 | 0.151 | 0.308 |
| July | 14.0 | 1.5 | 0.319 | 0.027 | 0.275 | 0.058 | 0.201 | 0.172 | -0.013 | 0.929 | 0.122 | 0.411 |
| August | 13.9 | 1.2 | 0.099 | 0.505 | 0.078 | 0.600 | -0.098 | 0.509 | -0.448 | 0.001 | 0.077 | 0.606 |
| September | 17.9 | 1.2 | 0.234 | 0.110 | 0.107 | 0.472 | -0.182 | 0.216 | -0.243 | 0.097 | 0.035 | 0.813 |
| October | 22.5 | 1.4 | 0.039 | 0.791 | 0.011 | 0.942 | -0.085 | 0.568 | -0.158 | 0.286 | 0.092 | 0.538 |
| November | 19.7 | 1.6 | 0.233 | 0.111 | 0.218 | 0.136 | 0.084 | 0.574 | -0.097 | 0.513 | 0.045 | 0.763 |
| December | 24.8 | 1.8 | 0.013 | 0.931 | 0.027 | 0.856 | 0.027 | 0.859 | 0.093 | 0.531 | 0.189 | 0.199 |
| January | 28.2 | 1.9 | 0.041 | 0.785 | 0.049 | 0.740 | -0.026 | 0.862 | -0.090 | 0.544 | -0.093 | 0.532 |
| February | 27.7 | 2.4 | 0.097 | 0.513 | 0.101 | 0.499 | -0.106 | 0.476 | -0.313 | 0.030 | -0.080 | 0.589 |
| March | 18.5 | 1.5 | 0.366 | 0.010 | 0.313 | 0.030 | -0.115 | 0.438 | -0.496 | 0.000 | 0.252 | 0.084 |
| April | 27.4 | 1.7 | -0.168 | 0.256 | -0.199 | 0.176 | -0.238 | 0.104 | -0.159 | 0.283 | 0.184 | 0.213 |
| May | 25.7 | 2.0 | 0.162 | 0.274 | 0.181 | 0.220 | 0.102 | 0.493 | -0.135 | 0.362 | 0.425 | 0.002 |

## 9.4 Discussion

Of all the biological parameters monitored in this trial, the only parameter displaying an obvious difference among longline sites was pea crab incidence. There was a general trend of increasing pea crab infestation with distance away from the fish farm. Although overall mean trial %PC was lower at the two fish farm sites (I and II) compared with sites III and IV, it seems unlikely that the salmon farm would have influenced the prevalence of pea crabs at sites I and II. Differences in %PC between sites, displaying a gradient from low levels at site I to higher levels at site IV, may be just a consequence of natural variation. It is noteworthy the the gradient of high to low pea crab incidence also coincides with longline positions relative to the mouth of NWB and the incoming current. Mussels at site IV, located towards the mouth of the bay and adjacent to the D'Entrecasteaux Channel, may have been in a position to intercept a greater number of planktonic crab post-larvae than mussels cultured at the other three sites. Irrespective of the differences between sites, %PC has little effect on the marketability of mussels in Australia; they are in demand regardless of whether or not they harbour pea crabs (G. Schroter, Secretary, Tasmanian Mussel Growers Cooperative, pers. com.). Therefore, there is no economic advantage in culturing mussels within a salmon farm solely for the purpose of obtaining product with a lower percentage of pea crab infestation.

The lack of any significant correlations between the incidence of pea crab infestation and mussel growth or survival is not surprising. We have already concluded from previous chapters that mussel growth and survival were similar among sites. This is

despite the differences in pea crab incidence, most significantly between sites I and IV. The lack of any significant growth differences among sites, nor significant correlations between %PC and growth, may be due to several factors. Firstly, it is possible that pea crabs genuinely had no effect on mussel growth. Secondly, pea crabs did have a negative effect on their hosts but the effect was not detectable. In this study, growth was monitored on bulk samples of 20 mussels rather on individual mussels. It is possible that any negative effects of pea crabs on individual shell or meat growth were too small to be detectable in bulk samples where the majority of individuals were not harbouring pea crabs. Thirdly, it has been suggested that using "presence" or "absence" of pea crabs as a criterion for investigating the negative effects of pinnotherids on bivalves may be inappropriate (Bierbaum and Ferson, 1986). "Presence" or "absence" provides no historical information of a mussel's prior association with pea crabs. For example, male and small female pea crabs are not really 'present' over long periods because they can, and do, move freely from host to host (Pregenzer, 1981; Bierbaum and Ferson, 1986). Also, the fact that an opened mussel is void of crabs provides no information to its recent history; a crab may have died or had recently vacated. Further, some studies have suggested that only larger mature female crabs have a negative effect on bivalve growth and condition (Pregenzer, 1981; Tablado and Gappa, 1995). If this is the case, then we can reasonably assume that the incidence of large female crabs was considerably lower than the overall pea crab incidence. Although pea crab sex, size, developmental stage and prior history of infestation of mussel samples are unknown, the lack of significant correlations in bulk samples may have been due to an extremely low incidence of mussels harbouring large female crabs and/or for only short periods of time.

In summary, significant differences in pea crab infestation were indicated between mussel sites. However, there is no evidence suggesting that pea crab incidence had any deleterious effects on overall growth, condition or survival of mussels. Therefore, the lower %PC incidence rates observed at site I provide no advantage in growing mussels within the Aquatas fish farm over any other experimental site within North West Bay.

# 10. Overspat

## 10.1 Introduction

Several months into the mussel growth trial, a settlement of natural mussel overspat was observed on some experimental droppers. Overspat settlements have been known to negatively affect the growth and condition of underlying mussels, largely through competition for available food (Wilson and Hodgkin, 1967; MacIntyre et al., 1977). Therefore, overspat settlements would be expected to have similar effects on mussel growth as would high stocking densities. Overspat compromising the growth of underlying experimental mussels and, therefore, masking the experimental effects of longline SITE and DEPTH was considered a possibility. Therefore, monitoring the growth of these recruits was deemed necessary, providing a backup dataset for assessing the influence of the Aquatas fish farm on mussel growth. Overspat monitoring also supplied information on the growth of younger mussels - possibly of greater interest to farmers than the growth of the larger experimental mussels as commercial stocks are typically cultivated from natural or hatchery-produced spat.

## 10.2 Methodology

Overspat on experimental mussel droppers at sites I and II were first noted in May 1995. A sparse settlement of overspat at sites III and IV was subsequently observed in June 1995. Spat shell lengths and weights were measured monthly from June 1995 through to May 1996. As only a few spat were observed at sites III and IV in June 1995, length and weight measurements at these two sites did not commence until July 1995.

Six droppers from each longline were sampled at two depths (1 m and 5 m) for length and weight measurements of the original experimental mussels (see previous descriptions of sampling procedures). Attached overspat within each sample were separated from the original experimental mussels. Within each site, spat were pooled according to longline section (A, B and C) providing 3 samples per site per month (12 samples total per month). Twenty individuals from each of these pooled samples were measured for length and bulk weighed to estimate individual whole live wet weight (individual WLW = bulk weight / 20). In April 1996 and May 1996 spat were not pooled; twenty individuals from each sample (12 samples per site) were selected for length and weight measurements. Individual shell lengths were measured with vernier callipers to the nearest 0.1 mm and individual WLW was estimated from the bulk weight of the twenty individuals in each subsample ($\pm$ 0.05 g, Sartorius IP65).

Two subsequent spatfalls were also observed during the trial (September 1995 and January 1996) and were, therefore, mixed in with the May 1995 settlement. In order to continue monitoring the progress of the May 1995 spatfall, a certain degree of sampling bias was introduced. Sampling bias took the form of selecting larger individuals from each mixed-cohort sample for length and weight measurements - it was assumed that the larger individuals in each sample were recruits from the May 1995 settlement. It is recognised that a certain degree of error may have occurred in the selection process. Firstly, monthly mean lengths and weights of the May 1995 cohort may have been overestimated through the selection of the larger individuals. Secondly, some overlap may have occurred, whereby larger individuals from the September 1995 cohort may have been selected for length and weight measurements in the later samples. However, as the selection process and, therefore, the sampling bias was consistent across all samples, comparisons of relative spat growth among sites were considered appropriate.

One-way ANOVA was used to assess mean shell length and whole live weight among sites for the July 1995 samples. Two-factor ANOVA was used to assess the effect of SITE and growing DEPTH on final shell length and whole live weight. Von Bertalanffy growth curves were fitted to shell length data from each site. Parameters for each VB growth equation ($L\infty$, K and $t_o$) were derived by iteration using least squares nonlinear regression (Systat 5.1). Analysis of residual sum of squares (ARSS, Chen et al., 1992) was used to determine if fitted VB curves differed among the four sites. This was followed by multiple comparison testing among the four curves to determine which curves differed and to identify the source(s) of difference (ie. VB parameters) using Likelihood Ratio tests (LR). LR tests were performed using a computer program developed by Malcolm Haddon (TAFI, Hobart) based on the procedures described by Kimura (1980).

In addition to monitoring the growth of the May 1995 spatfall, several growth parameters of the mixed-cohort spat population, which included individuals from all three identified spatfalls, were measured at the end of the trial (May 1996). A subsample of twenty-five mussels of mixed spat was randomly selected from each of the 48 dropper samples and individually measured for shell length and bulk weighed to estimate individual WLW. Sample spat densities were determined by counting the number of spat in a representative subsample (approximately 1/8 of each sample) and expressed as total spat per meter of dropper. Rough estimates of individual spat meat weight, shell weight, biomass, condition ($CI_{grav}$) and glycogen content were conducted on pooled subsamples (20 spat) according to each location (A, B, C) within each longline, providing three replicates per site. ANOVA was used to assess differences

among sites for each of the measured parameters. Correlation analyses, comparing sample spat densities (no. spat·m$^{-1}$), total spat weight (WLW·m$^{-1}$) and total spat + experimental mussel weight (spat WLW + experimental mussel WLW·m$^{-1}$) with sample mean length, biomass and survival of experimental mussels, were used to assess the influence of overspat and total mussel density on the growth and survival of the experimental mussels.

## 10.3 Results

Monthly measurements of shell length (L) and whole live weight (WLW) commenced in June 1995 at sites I and II but there were insufficient numbers of overspat for measurement at sites III and IV until July 1995. It is not known if spat settlement at sites III and IV occurred later than at sites I and II, or if only the droppers selected at II and IV in June 1995 were devoid of spat. As droppers were sampled across the entire length of each longline (ie. two droppers from each longline section A, B and C), a reasonable assumption is that settlement at sites III and IV occurred later than at sites I and II.

### 10.3.1 Spat Length

Mean L at sites I and II in June 1995 was 15.1 ± 0.5 mm and 13.5 ± 0.5 mm (n = 20 mussels per site), respectively, with significant differences among sites indicated ($P < 0.05$). Significant differences in L among all four sites were also indicated in July ($P < 0.001$), where length at site I (21.5 mm) > II (19.7 mm) > III (17.5 mm) = IV (16.5 mm) (n = 60 mussels at sites I, II and IV, n = 35 at site III, Fishers LSD, $P < 0.05$). Length increased steadily at all sites throughout the remainder of the trial with mean L at the two fish farm sites consistently greater than L at the two remote sites (Fig. 10.1). Final mean spat L in May 1996 was 64.4 ± 0.2 mm (n = 960 mussels). However, significant site differences were indicated ($P < 0.0001$), where a gradient of decreasing L from site I to IV was observed [site I (69.5 mm) > II (64.9 mm) > III (63.5 mm) > IV (59.5 mm), $P < 0.05$]. Final spat L at 1 m (63.5 ± 0.3 mm, n = 480 mussels) was slightly, but significantly, less than L at 5m (65.2 ± 0.4 mm, $P < 0.0001$).

Modelled von Bertalanffy growth curves and associated parameters at each site are presented in Fig. 10.2. Analyses of residual sum of squares (ARSS) indicated significant differences among curves ($P < 0.0001$). Likelihood ratio tests indicated no two growth curves were coincident ($P < 0.0001$, Table 10.1). However, only one

difference was indicated in multiple comparisons of VB parameters; the growth rate (k) at site II was marginally greater than at site III ($P < 0.05$).

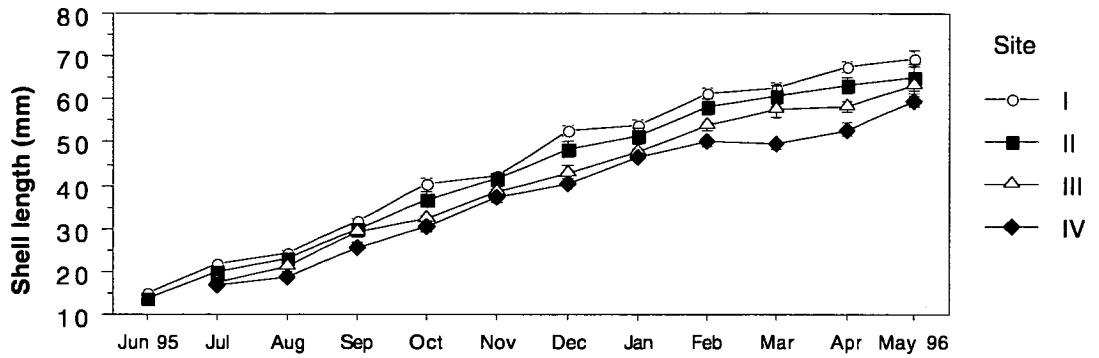


**Figure 10.1.** Monthly shell length of overspat at four sites in North West Bay from June 1995 to May 1996 (mean ± SE mm, n = 60 mussels per site). Error bars not visible lie within point boundaries.

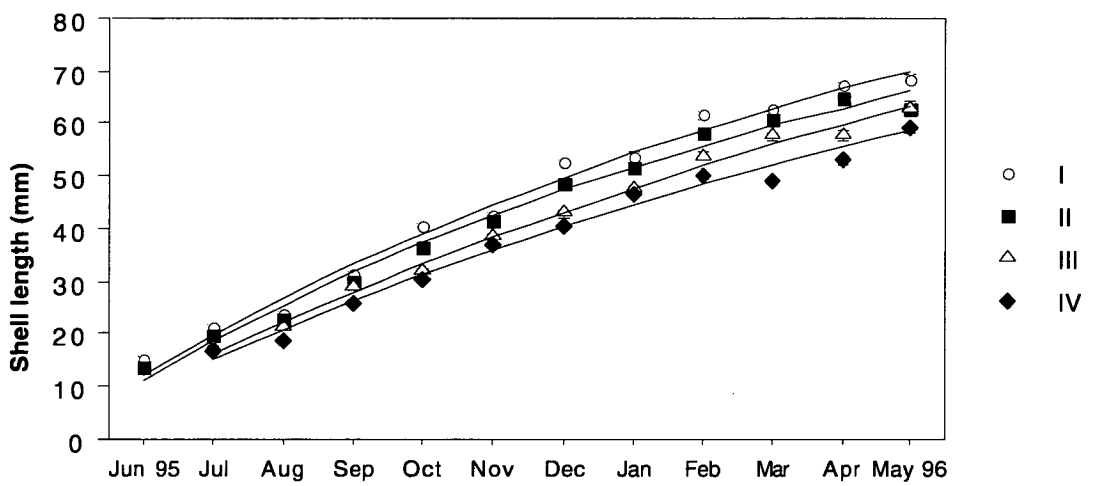


**Figure 10.2.** Monthly shell length at each site (mean ± SE mm) and their corresponding von Bertalanffy growth curves modelled by the equation $L_t = L\infty\ (1 - e^{-k(t - to)})$, where t at June 1995 = 0 months. VB curves describing growth from top to bottom are from sites I to IV, respectively. There were too few spat collected at sites III and IV in June 1995 for measurement. Estimated VB parameters ± SE:

| Site | L∞ (mm) | k (per month) | $t_0$ (months) | N |
|---|---|---|---|---|
| I | 112.1(5.1) | 0.078(0.006) | -1.49(0.13) | 860 |
| II | 100.0(5.3) | 0.087(0.009) | -1.30(0.16) | 860 |
| III | 121.2(9.5) | 0.060(0.007) | -1.38(0.18) | 815 |
| IV | 109.1(7.7) | 0.063(0.007) | -1.36(0.16) | 840 |

**Table 10.1.** Von Bertalanffy (VB) parameters ($L\infty$, K and $t_0$) describing spat growth at each site, probability of paired curves being coincident (cc) and probability of paired VB parameters being equal (Likelihood Ratio Test; Kimura, 1980). ns = not significant (P > 0.05).

| Site | | | II | | | | III | | | | IV | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Parameter | | cc | $L\infty$ | K | $t_0$ | cc | $L\infty$ | K | $t_0$ | cc | $L\infty$ | K | $t_0$ |
| | | | | 100.005 | 0.087 | -1.301 | | 121.232 | 0.060 | -1.375 | | 109.084 | 0.063 | -1.359 |
| I | cc | | <0.0001 | | | | <0.0001 | | | | <0.0001 | | | |
| | $L\infty$ | 112.128 | | ns | | | | ns | | | | ns | | |
| | K | 0.078 | | | ns | | | | ns | | | | ns | |
| | $t_0$ | -1.488 | | | | ns | | | | ns | | | | ns |
| II | cc | | | | | | <0.0001 | | | | <0.0001 | | | |
| | $L\infty$ | 100.005 | | | | | | ns | | | | ns | | |
| | K | 0.087 | | | | | | | 0.03 | | | | ns | |
| | $t_0$ | -1.301 | | | | | | | | ns | | | | ns |
| III | cc | | | | | | | | | | <0.0001 | | | |
| | $L\infty$ | 121.232 | | | | | | | | | | ns | | |
| | K | 0.060 | | | | | | | | | | | ns | |
| | $t_0$ | -1.375 | | | | | | | | | | | | ns |

Spat shell length growth in May 1996 represented a mean increase of 244% from July 1995 compared with only a 53% increase for the experimental mussels from March 1995 (Fig. 10.3). Significant site differences were indicated in relative shell length increments (P < 0.01), whereby relative growth at sites I and II (mean 225 ± 33% increase, n = 24 samples) were less than at III and IV (mean 258 ± 23% increase).

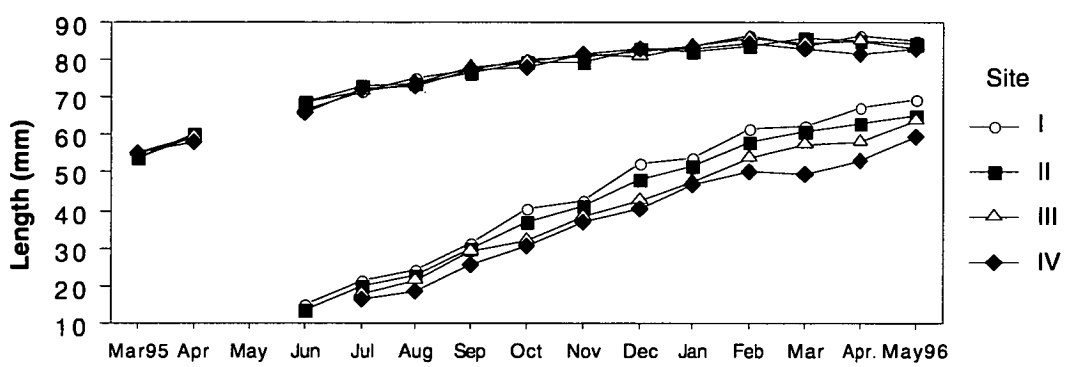


**Figure 10.3.** Monthly mean shell lengths of spat (lower curves) and experimental mussels (upper curves) at each longline site (I - IV).

## 10.3.2 Spat weight

Mean spat weight in June 1995 was 0.72 ± 0.04 g·spat$^{-1}$ at sites I and II. In July 1995, when all sites were sampled, the mean WLW was 0.95 ± 0.03 g·spat$^{-1}$ (n = 12 samples of 20 mussels per sample); differences among sites were significant (P < 0.001) where mean WLW at I > II > III = IV. The trend of lower WLW with increasing distance from the farm continued throughout the remainder of the trial (Fig. 10.4). By May 1996, overall mean WLW was 22.4 ± 0.9 g (n = 48 samples), with significant differences among sites indicated (P < 0.001, Fig. 10.5). The greatest

difference was 9.4 g·mussel$^{-1}$, between site I (26.9 ± 1.3 g, n = 12 samples) and site IV (17.5 ± 0.8 g, n = 12 samples). No differences between growing depths nor a site x depth interaction for final WLW were significant (P > 0.05).

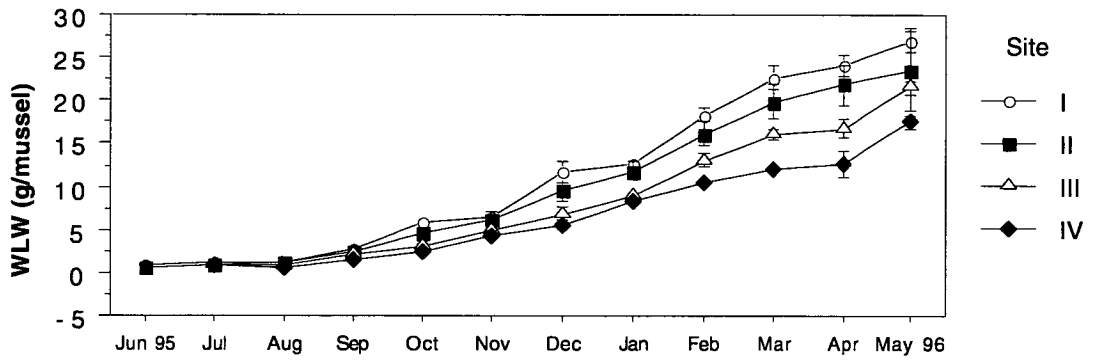


**Figure 10.4.** Monthly whole live weight (WLW) of overspat at each of four sites (mean ± SE g·mussel$^{-1}$, n = 3 samples per site except April 1996 and May 1996 where n = 12 samples per site). Each sample mean represents the individual WLW determined from the bulk WLW of 20 mussels.

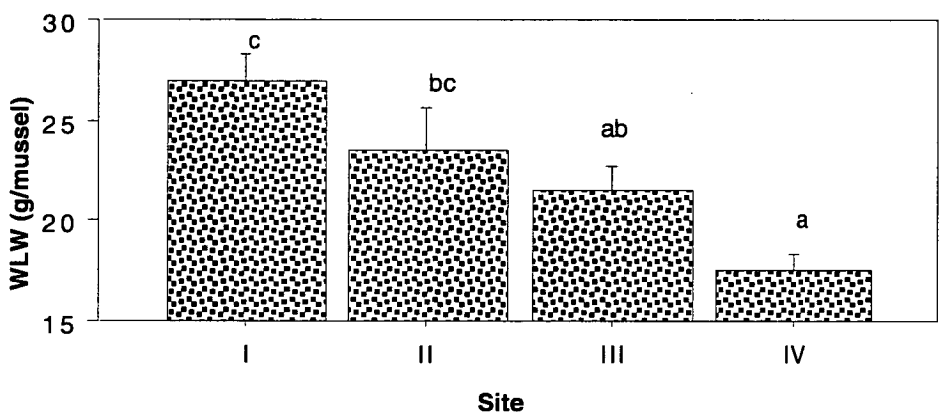


**Figure 10.5.** Whole live weight (WLW) of overspat in May 1996 (mean ± SE, n = 12 samples per site with each sample being the mean WLW of 20 individuals).

Although differences in the absolute WLW among sites in May 1996 were significant, the percentage increase in WLW from July 1995 to May 1996 was not different among sites, growing depths nor a site x depth interaction (P > 0.05, overall mean increase 2250 ± 75%, n = 48 samples).

### 10.3.3 Spat density

Sample densities of the May 1995 spatfall were only estimated once, in September 1995. A wide range of densities (approximately 100 to 2000 spat per meter) was observed with an overall mean of 490 ± 55 spat·m$^{-1}$ (n = 48 samples). Significant differences among sites were indicated (P < 0.0001), but not between depths nor a site

x depth interaction ($P > 0.05$). Spat densities decreased with distance from the farm cages (Fig. 10.6), from 873 ± 147 spat·m$^{-1}$ at site I to 241 ± 18 spat·m$^{-1}$ at site IV (n = 12 samples per site).

There were no significant correlations of sample spat densities (no. spat /m or kg WLW/m) with mean shell length ($P = 0.98$, $r = 0.004$, n = 48 samples) or biomass ($P = 0.24$, $r = 0.174$, n = 48 samples) of the experimental mussels in September 1995. This suggests that spat density probably had no effect on growth of the underlying experimental mussels as of September 1995.

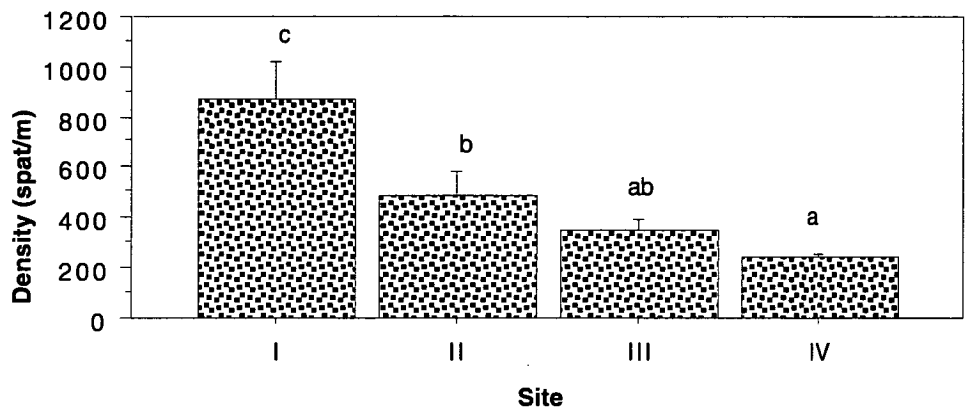


**Figure 10.6.** Spat density at each site in September 1995 (mean ± SE spat·m$^{-1}$, n = 12 samples per site). Significant differences in density between sites were indicated ($P < 0.0001$). Site densities not sharing a common letter are significantly different ($P < 0.05$, Fishers LSD).

### 10.3.4 Mixed cohort sample

Subsamples of all spat, including those that settled after May 1995, from each sample collected in May 1996 were measured for length and bulk weighed (25 spat·sample$^{-1}$; 300 spat·site$^{-1}$, 1200 spat in total). Considerable overlap in size-frequency prevented identification of the three cohorts and, therefore, a more unbiased estimate of mean shell length of the May 1995 cohort (Fig. 10.7). A small number of individuals ranging from 8 mm to 20 mm in length (30 individuals) suggested a fourth spatfall may have occurred between February 1996 and May 1996.

Overall mean length of the mixed cohorts in May 1996 was 46.0 ± 0.4 mm (n = 1200 mussels) with significant differences between sites indicated ($P < 0.0001$). Spat L at site I (51.4 ± 0.8 mm, n = 300 mussels) was significantly larger than spat L at the other three sites (site means ranged from 43.2 to 45.5 mm, Fig. 10.8). Mussels grown at 1 m (45.1 ± 0.5, n = 600) were slightly smaller than those grown at 5 m (46.8 ± 0.5; $P < 0.05$).

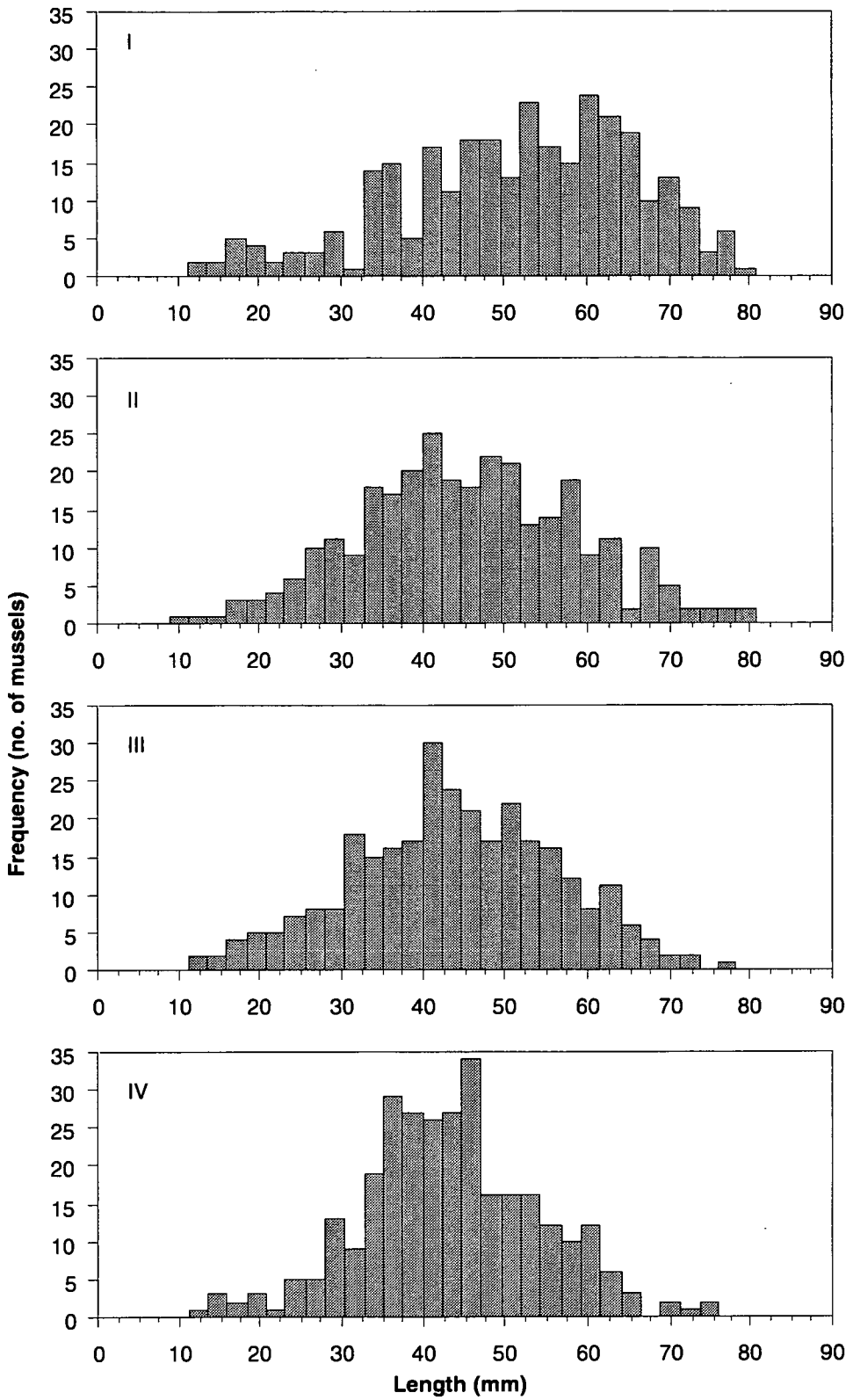


**Figure 10.7.** Frequency histogram of spat lengths (mm) from mixed cohort samples collected from four sites (I, II, III and IV) in May 1996 (n = 300 spat per site). Although three, and possibly four, cohorts are represented, considerable overlap prevents identification of each cohort.

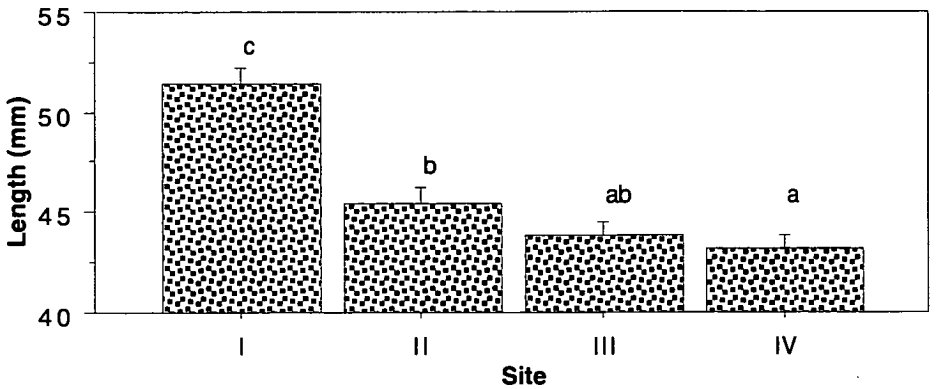


**Figure 10.8.** Overall mean shell length of mixed-cohort spat collected in May 1996 (mean ± SE, n = 300 mussels per site). Site means not sharing a common letter are significantly different ($P < 0.05$, Fishers LSD).

Overall mean whole live weight of the mixed spat was 10.2 ± 0.5 g·spat$^{-1}$ (n = 48 samples), with spat WLW from site I (13.8 ± 1.0 g·spat$^{-1}$, n = 12 samples) greater than at the other three sites ($P < 0.001$, mean WLW from sites II, III and IV = 9.0 ± 0.4 g·spat$^{-1}$, n = 36 samples).

Due to the mixture of overspat, it was not possible to estimate the density of the oldest cohort (ie. May 1995 settlement) still remaining in May 1996. Mixed cohort densities were similar at all sites ($P > 0.05$; overall mean 1889 ± 102 spat·m$^{-1}$, n = 48 samples, range 528 to 3296 spat·m$^{-1}$) but differed between depths ($P < 0.001$). Densities at 1 m (2217 ± 117 spat·m$^{-1}$, n = 24 samples) were higher than at 5 m (1561 ± 139 spat·m$^{-1}$).

Spat density, expressed as spat per original experimental mussel, was 15.0 ± 1.1 spat·mussel$^{-1}$ (n = 48 samples), approximately five times greater than the density estimates in September 1995. Significant differences between sites ($P < 0.05$) indicated the highest number of spat per original mussel was at site III (19.0 ± 2.4 spat·mussel$^{-1}$, n = 12 samples) and the lowest at site I (13.0 ± 1.8 spat·mussel$^{-1}$). Significant differences between growing depths ($P < 0.0001$) indicated density at 1 m (19.0 ± 1.4 spat·mussel$^{-1}$) was greater than at 5 m (11.0 ± 1.1 spat·mussel$^{-1}$).

Spat WLW per metre of dropper was 18.9 ± 1.2 kg·m$^{-1}$. Site differences were indicated ($P < 0.05$), where spat WLW decreased with distance from the fish farm (Fig. 10.9). The combined overall mean WLW of spat and experimental mussels was 25.6 ± 1.3 kg·m$^{-1}$; again total WLW decreased with distance from the fishcages. Spat dominated dropper production, representing approximately 72% and 87% of the total dropper WLW and biomass (ash-free dry weight), respectively.

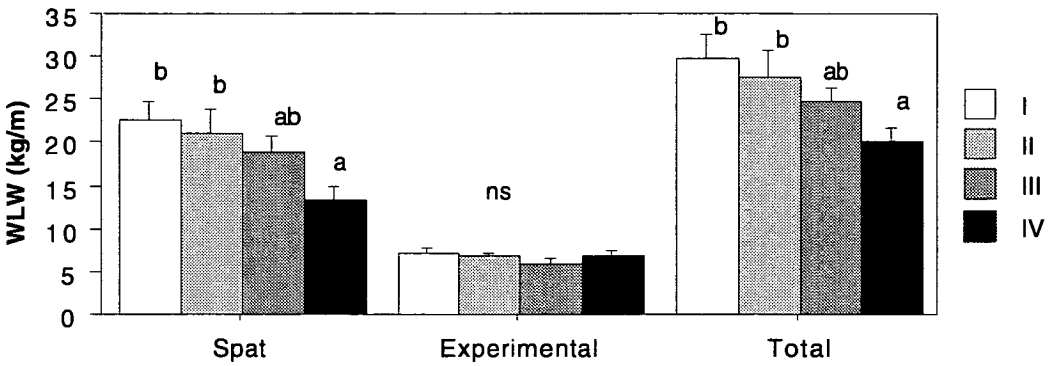


**Figure 10.9.** Whole live wet weight (WLW) components of mussel socks at each of four sites in May 1996, comprising experimental mussels' WLW and overspat WLW (mean ± SE kg·m of sock$^{-1}$, n = 12 samples per site). Site means not sharing a common letter within each grouping are significantly different ($P < 0.05$).

No significant differences between sites were indicated for any other measured parameters ($P \geq 0.05$). These included wet meat weight (mean 6.5 ± 0.3 g·spat$^{-1}$, n = 3 samples per site), dry meat weight (mean 1.3 ± 0.1 g·spat$^{-1}$), shell weight (mean 6.3 ± 0.3·spat$^{-1}$), condition ($CI_{grav}$ mean 100.3 ± 1.3‰), meat moisture content (mean 79.7 ± 0.1%), individual spat biomass (mean 1.46 ± 0.06 g AFDW·spat$^{-1}$), spat biomass per meter of sock (mean 2.76 ± 0.18 kg AFDW·m$^{-1}$) or glycogen content (mean 23.3 ± 0.6 g glycogen·100g DMW$^{-1}$).

There were no significant correlations between sample spat densities (no. spat·m$^{-1}$) and final shell length ($P = 0.99$, $r = 0.001$, n = 48 samples), individual biomass ($P = 0.43$, $r = -0.116$, n = 48 samples) condition ($CI_{grav}$, $P = 0.15$, $r = -0.210$) or survival ($P = 0.97$, $r = -0.005$) of the experimental mussels. Nor were correlations significant between spat densities, expressed as kg WLW·m$^{-1}$, and condition ($P = 0.59$, $r = -0.078$) or survival ($P = 0.25$, $r = 0.170$) of experimental mussels. Although spat density (kg WLW·m$^{-1}$) did show significant correlations with final shell length ($P = 0.001$, $r = 0.449$) and individual biomass ($P = 0.02$, $r = 0.345$) of experimental mussels, correlations were neither negative nor particularly strong (ie. r values were below 0.5).

## 10.4 Discussion

The settlement and subsequent growth of overspat certainly dominated the biological production of droppers, particularly during the latter stages of the trial. Overspat represented approximately 72% of the total wet weight and 87% of the total biomass of droppers in May 1996. Two questions regarding overspat are most pertinent to this study. Firstly, did overspat impair the growth of the underlying experimental mussels? Secondly, was spat growth on the fish farm greater than at sites III or IV?

Did spat affect the growth of the underlying experimental mussels? The intent of monitoring overspat during the trial was to obtain a backup dataset describing mussel growth at each of the four longline sites. This data was collected in case the growth of experimental mussels was compromised by the fouling overspat, thereby masking any effect of longline site relative to the fish farm cages. Negative effects on mussel growth due to oversettlements or high stocking densities are recognised (Wilson and Hodgkin, 1967; Mason, 1972; MacIntyre et al., 1977; Hickman, 1992; van Erkom Shurink and Griffiths, 1993; Frechette and Bacher, 1998). The reduction in growth is considered to be largely the result of competition for food, but the accumulation of metabolites and faecal wastes may also be implicated. Husbandry practices can address this problem by grading and thinning culture units during growout.

Spat densities and biomass attained extremely high levels towards the end of the trial. With such high levels of fouling biomass overlying the experimental mussels, considerable competitive pressure for available resources (eg. food and oxygen) would be expected. However, correlation analyses suggest no significantly negative effect of overspat density or biomass (May 1996 mixed cohort sample) on the final length, condition or survival of the underlying experimental mussels. This is despite overspat densities ranging from 500 to 3300 spat$\cdot m^{-1}$ or 4 to 38 kg WLW$\cdot m^{-1}$ and biomass ranging from 0.8 to 4.9 kg$\cdot m^{-1}$. This could mean that overspat had no influence on the growth of underlying experimental mussels; or alternatively, overspat did affect experimental mussel growth, but growth was affected similarly across all droppers. Reduced growth and survival rates during the final stages of the trial being, at least partially, due to increasing spat biomass and competition for food has previously been acknowledged (Sections 5.4.3 and 8.4.2). However, as spat densities were not monitored throughout the trial, this relationship cannot be tested. Further, the degree to which overspat may have reduced experimental mussel growth and survival relative to other possible factors (eg. size, age, reproductive stress, other types of biofouling, water contaminants) cannot be determined. All we can say is that overspat may have influenced mussel growth and survival, but their influence was probably similar across all droppers and sites. If we assume that overspat influence on mussel growth was similar among sites; the potential of the Aquatas fish farm for enhancing mussel growth, using the results from the experimental mussel data analyses, can be assessed with increased confidence.

Did the proximity of longline to the fish cages influence spat growth? On the surface one might suspect that spat growth was enhanced with increasing proximity to the fish cages. Shell lengths and live weights were consistently higher nearer the fish cages throughout the trial period. At the end of the trial, spat at site I were 10.0 mm longer

and 9.4 g heavier than spat at site IV. These differences in growth among the four sites might be indicative of increasing food availability (eg. as fish farm particulate wastes) with increasing proximity to the fish cages. However, there are a number of uncertainties concerning the data that renders any definite conclusions questionable.

Firstly, it is unknown if the original spat settlements occurred at the same time at all four sites. The lack of sufficient numbers of measurable spat at sites III and IV in June and July 1995 suggests settlement was not simultaneous among sites. Also, differences in shell length between site I (15.1 mm) and II (13.5 mm) were already significant in June 1995. This could suggest that conditions were more favourable for growth at site I (closest to the fish cages) or, alternatively, spat settled earlier at site I than at site II. The same argument can be applied to sample data on subsequent months when all sites were represented and where shell length increased with proximity to the fish cages. Further, the effect of differential experimental mussel densities on spat growth may also be implicated in differential spat growth among sites. Just as spat can influence the growth of underlying mussels, competition for food from older established mussels may similarly restrict the growth of new recruits (Seed and Suchanek, 1992). We have previously discussed that experimental mussel densities in socks allocated to the two fish farm sites were less than at sites II and IV (Section 8). Therefore, it is unknown if increasing spat size with increasing proximity to the fish cages was due to: increasing food availability, as a consequence of extra fish farm wastes; reduced competitive pressure from a lower density of experimental mussels; different settlement times among sites; or a combination of factors. Different settlement times as the primary factor in explaining differences in spat sizes is supported by the parameters measured in the May 1996 mixed cohort sample and by monitored food concentrations. Similar spat condition indices and meat glycogen contents from the May 1996 mixed cohort sample indirectly suggest similar food availability among sites. Also, measured concentrations of particulate food concentrations (eg. POM, %POM, algal POM, non-algal POM) were all similar among mussel longline sites (Section 4.3.7).

It is unknown why spat might have settled at sites I and II in May 1995 but not at sites III and IV. Free-swimming mussel larvae probably occur as aggregations in the water column. Perhaps such an aggregation was carried through the farm from the north, settling on the droppers at sites I and II, and probably on the fishcages as well. The current direction at the time may not have directed this aggregation to sites III and IV. Alternatively, the current direction was appropriate, but the aggregation was depleted by the settlement of individuals onto longlines I and II and the fish farm structures.

Analysis of von Bertalanffy growth curves does not clarify the uncertainty of differential growth among sites. Due to several spat settlements over the trial period, the observed growth of the original settlements at each site was estimated from biased samples, where the larger individuals were selected for measurement from October 1995 to May 1996. High spat $L\infty$ values (100 to 121 mm) compared with experimental mussels' $L\infty$ (86.9 to 89.4 mm) might be a possible consequence of this bias; however, it could also be argued that the lower experimental mussel $L\infty$ was biased because mussels were graded prior to stocking. Although ARSS indicated VB growth curves differed among sites ($P < 0.0001$) and LR tests suggest no two curves were coincident ($P < 0.0001$), no differences in VB growth parameters were indicated (except a minor difference in K between sites II and III). This is interesting, considering the wide range of $L\infty$ values among sites. With large sample sizes, minor differences among growth curves can be statistically significant but biologically trivial. The absence of significant differences in VB growth parameters could suggest that sample variation was probably too extreme to detect any differences. Therefore, the discrepancy between significantly different growth curves and non-significant VB parameters does not clarify whether growth was actually similar among sites or if the variation was just too great to detect differences in the VB parameters. The only way to really assess the growth of smaller mussels cultured on the fish farm is to run another similar experiment using smaller mussels with uniform stocking densities.

In summary, the degree to which overspat affected the underlying experimental mussels remains undetermined. Some of the reduced mussel growth and survival over the final three months might be attributed to increasing competitive pressure from overspat for the available food supply. However, there is no statistical evidence to support this claim. Overspat tended to be larger with increasing proximity to the fish cages. However, it is unclear to what extent these size differences can be attributed to differences in available food, different ages and/or sampling bias. Therefore, enhanced growth of spat within the Aquatas fish farm cannot be confidently concluded nor refuted. As such, the assessment of site differences must rely on experimental mussel data with some caution - growth may have been influenced to some extent by overlying spat. However, similar final spat length and weight at the two sample depths does support the lack of any significant depth effect on experimental mussel growth (Section 5)

# 11. Stable Isotope Analyses

## 11.1 Introduction

The analysis of stable isotopes (SI) within animal tissues has been incorporated in many biological studies; for identifying food sources of animals, and tracing food and energy flows through food chains (eg. Fry et al., 1984; Fry and Sherr, 1984; Gearing et al., 1984; Peterson et al., 1985; 1986; Dunton and Schell, 1987; Peterson and Fry, 1987; Fry, 1988; Lochmann and Philips, 1996; Yelenik, 1996; Loneragan et al., 1997). The underlying basis for this type of investigation is "you are what you eat", with an animal's SI composition largely refecting the SI composition of the material consumed.

Stable isotopes commonly used in these investigations are those of carbon ($^{13}C$ and $^{12}C$) and nitrogen ($^{15}N$ and $^{14}N$). Typically, consumer SI compositions are not identical to their food source(s) because the ratios of both $^{13}C/^{12}C$ and $^{15}N/^{14}$ tend to increase (ie. become more enriched in the heavier isotope) at each successive trophic level. Isotopic enrichment is a consequence of metabolic fractionation within consumer organisms by way of the selective respiration of $^{12}C$ (McConnaughey and McRoy, 1979) and excretion of $^{14}N$ (Owens, 1987; Peterson and Fry, 1987). These physiological processes result in the retention of greater proportions of the heavier isotopes ($^{13}C$ and $^{15}N$). The extent of isotopic enrichment tends to vary according to species and the particular tissue being analysed (De Niro and Epstein, 1978; Stephenson and Lyon, 1982).

The original purpose of incorporating stable isotope analyses into this study of mussel and salmon polyculture was to ascertain if fish farm wastes (fine fish feed particles and faecal material) actually constituted a significant portion of the diet of mussels grown near the fish cages. If so, differences in the SI compositions of fish farm mussels and non-fish farm mussels might support any growth differences as being due to fish farm wastes being consumed by mussels grown on the fish farm (assuming the composition of fish wastes was isotopically distinct from natural food particles normally consumed by mussels).

However, as we have seen in preceding sections, growth was not significantly different among mussels cultured on or off the fish farm. Perhaps the analyses of SI composition might prove to be useful in explaining the lack of growth differences among mussel sites.

## 11.2 Methodology

Variations in stable isotopic composition have been found in different bivalve body tissues (Stephenson and Lyon, 1982). In order to reduce the variability caused by differential fractionation among organs and storage products, mantle tissue was chosen for SI analyses. This tissue has been found to display consistent isotopic enrichment and is considered to be the most favourable tissue for discriminating dietary compositions (Ruckelshaus et al., 1993).

It was assumed, not unrealistically, those mussels closest to, and furthest from, the fish cages (sites I and IV, respectively) would display the greatest differences in SI composition. Therefore, tissue sampling and SI analyses were limited to mussels from these two groups. Each month, sampled mussels that were not selected for length-weight-condition measurements were pooled according to growing depth. From each pool, mantle tissue from one valve was removed from ten males and ten females. Mantle tissues from each set of 10 individuals were combined, dried to constant weight, ground to a fine powder, and stored at -20°C until analysed for SI composition.

A supplementary sampling was undertaken after the conclusion of the growth trial (April 1997) to determine if mussels grown within the fish farm could actually be distinguished from mussels grown away from the farm by analysing their respective SI composition. Mussels that were growing directly on fishcage rings for over one year (length 60 - 70 mm) were sampled along with mussels grown at site IV over the same period (overspat of similar length). Four samples were taken from two fishcages and from four different droppers at site IV. Ten mussels were selected from each sample, mantle tissue removed and pooled for analysis.

Samples of fish feed (Pivot Aquaculture, Salmon Grower Diet: 45% protein: 22% lipid content; 2 mm to 9.5 mm pellet size) used on the farm site during and after the study period were similarly dried, ground and stored until analysed for SI composition.

Suspended particulate matter in the water column during the study period was not sampled and analysed for stable isotope composition. However, subsequent to the trial period, seston samples in North West Bay were collected with a plankton net (20 μm mesh towed at slow speed at approximately 5 m depth for 3 min) in April 1997 and December 1997. Samples were centrifuged, dried and stored at -20°C until analysed. These samples provided an estimate of the possible range of seston isotopic composition during the mussel growth trial (1995-96). In April 1997, three replicate samples were taken from each of three locations: within the fish farm, approximately

1.5 km north, and 1.5 km south, of the fish farm. The December 1997 samples did not include the within-farm location. It was assumed that seston samples were a mixed assemblage of food particles available to mussels including phytoplankton, zooplankton, detritus, fish feed and faeces, as well as inorganic particulate material.

Faecal material, a possible source of mussel food, was extruded from the intestines of six freshly killed salmon in April 1997. The SI composition of faeces was compared with the ingested feed to obtain an estimate of faecal isotopic fractionation (ie. the extent of SI enrichment or depletion in faeces relative to ingested feed).

Subsamples of approximately 2 to 3 mg of dried and finely ground material (mantle tissue, fish feed, faeces and suspended particulate matter) were sent to the CSIRO Division of Water Resources (Adelaide, South Australia). Analysis of stable carbon and nitrogen isotope composition, as well as percentages of total carbon and nitrogen content in each sample, was performed using an isotope ratio mass spectrometer following the procedure of Barrie and Prosser (1996). Isotopic compositions are expressed in standard differential notation:

$$\delta X = \left[ \left( \frac{R_{sample}}{R_{standard}} \right) - 1 \right] \times 1000$$

where $\delta X$ is the isotope ratio in delta units relative to a standard (‰), and $R_{sample}$ and $R_{standard}$ are the isotopic ratios of the heavy to the light isotope measured for the sample and the standard, respectively (Ehleringer and Rundel, 1989). In this study the R ratios represent $^{13}C/^{12}C$ and $^{15}N/^{14}N$. Sample carbon and nitrogen R ratios were measured relative to the R ratio of a PeeDee belemnite standard (PDB) and atmospheric nitrogen, respectively.

Mussel SI ratios were analysed with three-way ANOVA, with TIME (month), SITE (I & IV) and SEX (male & female) as fixed factors ($n = 2$ samples·sex$^{-1}$·site$^{-1}$·month$^{-1}$) after confirmation of homogeneity of variances (Cochrans test). With only two replicates per treatment level, normality could not be assessed. Where differences among, or interactions between, factors were significant, post hoc tests (Fishers LSD) were performed to identify the source of these significant differences or interactions.

## 11.3 Results

### 11.3.1 Mussel $\delta^{13}C$

All samples of mussel mantle tissue produced negative $\delta^{13}C$ values as they are isotopically light ($^{13}C$ depleted) compared to the $^{13}C_{PDB}$ standard. The $\delta^{13}C$ ratios displayed significant monthly variations ($P < 0.0001$) with three distinct peaks: July 1995, October 1995 and April 1996 (Fig. 11.1). Mussel $\delta^{13}C$ values at both sites were closely coupled throughout the trial, which included both fish culture and fallowing periods. A trend of progressive $^{13}C$ enrichment at both sites was observed, from initial values of -19.4 ± 0.1‰ (n = 8 samples) in March 1995 to -17.7 ± 0.1‰ at the conclusion of the trial. No differences in overall trial mean $\delta^{13}C$ between sites, nor any site x time interaction, were significant ($P > 0.05$). $\delta^{13}C$ values of both males and females became increasingly enriched during the course of the trial with a significant time x sex interaction indicated ($P < 0.0001$). Post hoc testing revealed male $\delta^{13}C$ values were significantly higher ($P < 0.05$) than female $\delta^{13}C$ values (ie. slightly more enriched in $^{13}C$ and therefore less negative) for all months except April 1996 and May 1996 ($P > 0.05$, Fig. 11.1).

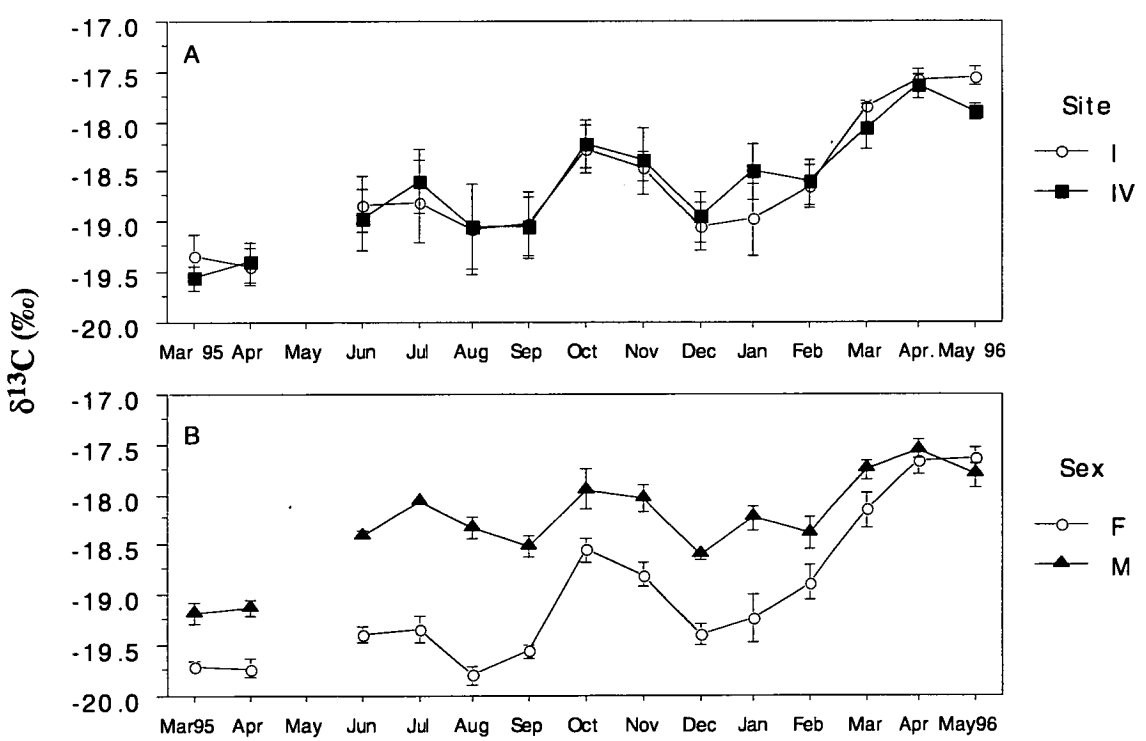


**Figure 11.1.** (A) Monthly $\delta^{13}C$ values of mussel mantle tissue at sites I and IV (mean ± ‰ SE, n = 4 samples per site) and (B) according to sex (n = 4 samples per sex). Each n is the composite $\delta^{13}C$ value of a subsample taken from 10 pooled, dried and ground mantle tissues. Although overall mean $\delta^{13}C$ changed over time ($P < 0.0001$), no site differences or site x time interactions were significant ($P > 0.05$). Significant differences in $\delta^{13}C$ between males and females were indicated for all months ($P < 0.05$) except April 1996 and May 1996 ($P > 0.05$).

### 11.3.2 Mussel $\delta^{15}N$

Mantle tissues were isotopically heavy ($^{15}N$ enriched) compared to the $^{15}N$ standard (atmospheric nitrogen) and displayed less temporal variation than $\delta^{13}C$ ($P < 0.01$, variance ratio test on log-transformed data; Zar, 1984). Monthly mean $\delta^{15}N$ values ranged from a low of $9.1 \pm 0.1‰$ in February 1996 to a high of $9.7 \pm 0.2‰$ ($n = 8$ samples) in April 1995. Although significant temporal variation was indicated ($P < 0.05$), there was no obvious trend of $^{15}N$ enrichment or depletion over the trial period (Fig. 11.2). There was no difference in overall mean $\delta^{15}N$ between sites ($P > 0.05$) but a significant site x time interaction was indicated ($P < 0.05$). However, nothing meaningful is obvious from this interaction, where differences in monthly $\delta^{15}N$ values between sites oscillated between significance and non-significance. Post hoc multiple comparison testing revealed no significant difference between $\delta^{15}N$ values at the start and end of the trial at either site ($P > 0.05$). $\delta^{15}N$ displayed a marginally significant site x sex interaction ($P = 0.04$). Post hoc testing revealed that overall mean $\delta^{15}N$ of site I males$^{a}$ < site I females$^{b}$ = site IV males$^{b}$ = site IV females$^{b}$ (Fig. 11.3). A sex x time interaction was not significant, although marginal ($P = 0.06$).

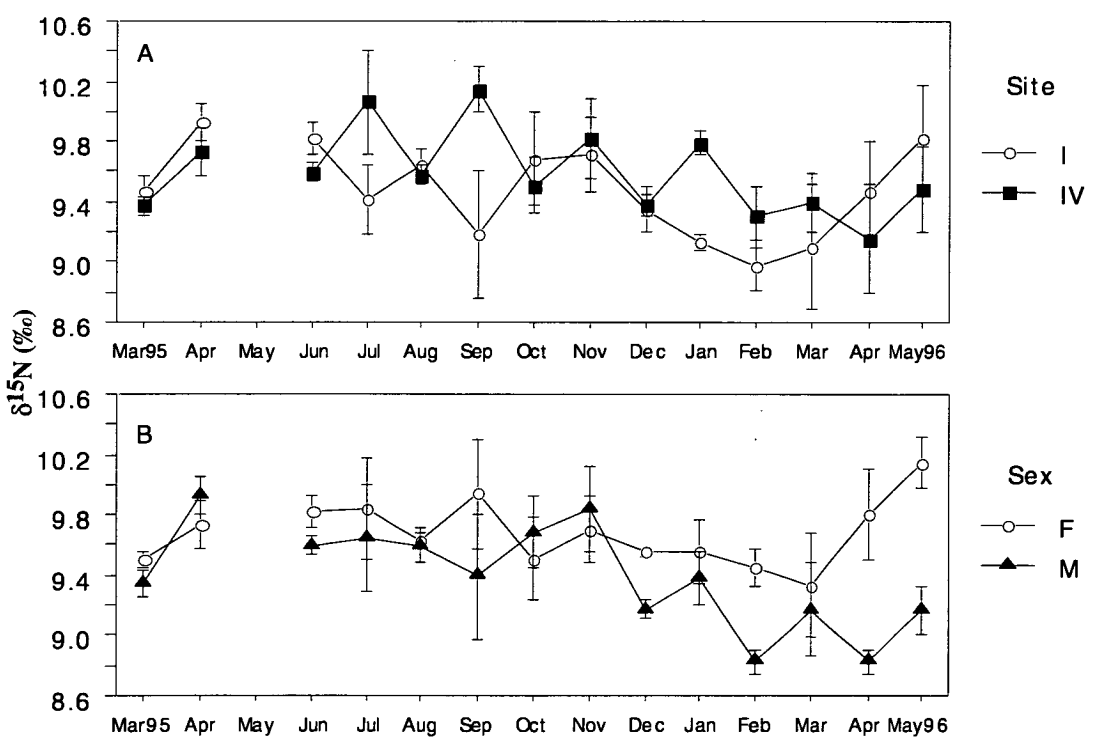


**Figure 11.2.** (A) Monthly $\delta^{15}N$ values of mussel mantle tissue at sites I and IV (mean ± SE‰, n = 4 samples per site) and (B) according to sex (n = 4 samples per sex). Each n is the composite $\delta^{15}N$ value of a subsample taken from 10 pooled, dried and ground mantle tissues. A site x time interaction was significant ($P < 0.05$).

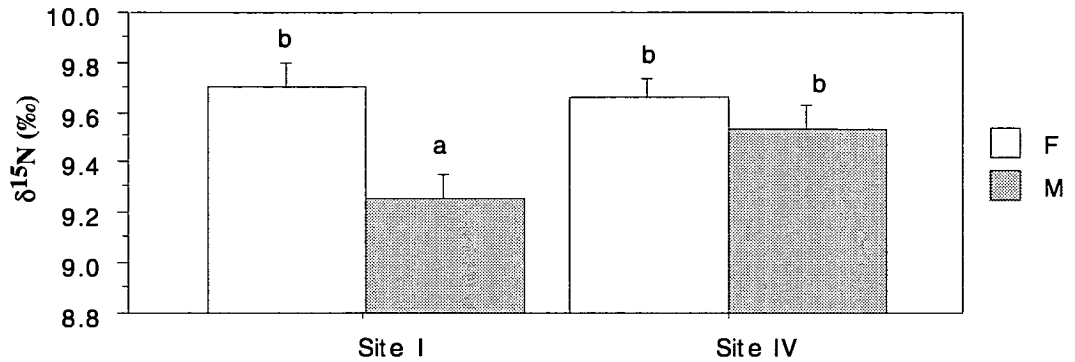


**Figure 11.3.** Overall trial $\delta^{15}N$ values of mussel mantle tissue according to sex within each site (mean ± SE‰, n = 28 samples). Mean $\delta^{15}N$ values not sharing a common letter are significantly different ($P < 0.05$).

### 11.3.3 $\delta^{13}C$ and $\delta^{15}N$ of mussel feed sources

Twenty-five fish feed samples collected during and after the conclusion of the trial, displayed considerable variation in SI composition (Fig. 11.4). However, the significance of this variation cannot be tested, as replicate samples from each feed batch were not analysed. Feed $\delta^{13}C$ ranged from -18.7‰ to -21.5‰ (mean -20.6 ± 0.1‰, n = 25 samples) and $\delta^{15}N$ ranged from 10.4 - 15.1‰ (mean 12.3 ± 0.2‰, n = 25 samples).

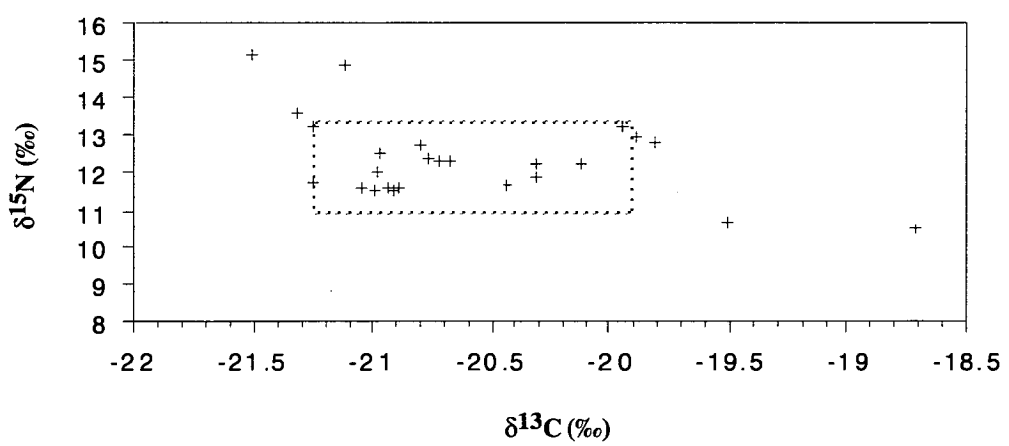


**Figure 11.4.** Stable isotopic composition ($\delta^{13}C$ and $\delta^{15}N$, ‰) of twenty-five samples of fish feed (Pivot Aquaculture Salmon Grower Diet) manufactured on various dates over a two year period (1995-1997). Boxed area represents the mean ± 1 standard deviation.

Stable isotope analyses of salmon faeces were only performed on samples collected on one occasion (April 1997) and compared with the SI values of the feed fed to the fish on the same day (one feed sample only). The $\delta^{13}C$ and $\delta^{15}N$ values of the feed were -20.1‰ and 12.2‰, respectively. The corresponding mean faecal $\delta^{13}C$ and $\delta^{15}N$

values from six fish were -20.7 ± 0.3‰ (range - 21.4 to - 19.9‰) and 11.5 ± 0.2‰ (range 11.2 to 12.2‰), respectively. These δ values indicate that faeces were slightly depleted in both $^{13}C$ and $^{15}N$ compared with the feed (both $P < 0.05$).

Seston samples showed no significant spatial differences in $\delta^{13}C$ or $\delta^{15}N$, either among the three areas of NWB sampled in April 1997 or between the two areas sampled in December 1997 ($P > 0.05$). However, significant temporal differences was indicated between the two sampling periods for both $\delta^{13}C$ and $\delta^{15}N$ ($P < 0.0001$). Seston $\delta^{13}C$ and $\delta^{15}N$ values in April 1997 ($\delta^{13}C$ = -18.3 ± 0.1‰ and $\delta^{15}N$ = 8.7 ± 0.1‰, n = 9 samples) were more enriched compared with December 1997 ($\delta^{13}C$ = -20.4 ± 0.1‰ and $\delta^{15}N$ = 7.7 ± 0.1‰, n = 6 samples). Comparisons of SI composition between seston and feed samples revealed significant differences in $\delta^{15}N$ values ($P < 0.0001$); but only seston collected in April 1997 differed from feed in $\delta^{13}C$ values ($P < 0.0001$). The range of $\delta^{13}C$ and $\delta^{15}N$ values for seston and salmon feed/faeces along with trial mussel mantle tissue SI values are displayed in Fig. 11.5. The scatter of feed SI composition is most evident, particularly with 4 samples that might be considered outliers (2 samples with high $\delta^{13}C$/ low $\delta^{15}N$ and two samples with high $\delta^{15}N$).

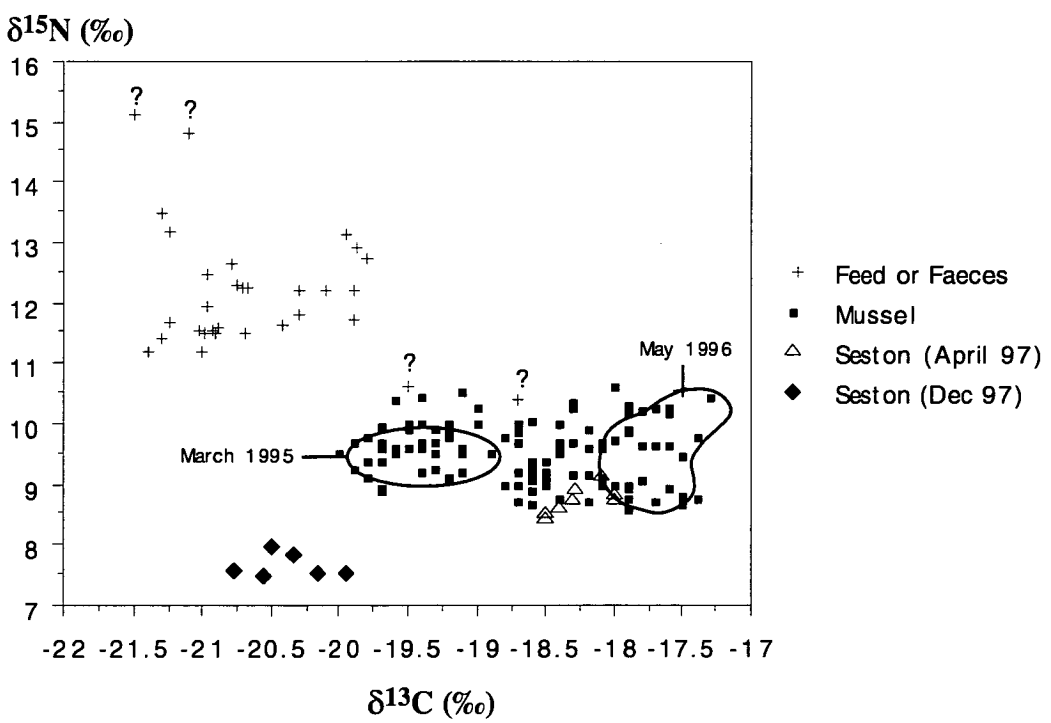


**Figure 11.5.** Stable isotopic composition ($\delta^{13}C$ and $\delta^{15}N$) of mussel mantle tissue at sites I and IV over the entire trial period. The range of mussel $\delta^{13}C$ and $\delta^{15}N$ values at the start (March 1995) and conclusion of the trial (May 1996) are encircled. Mussel mantle tissues displayed a significant enrichment in $\delta^{13}C$ from March 1995 to May 1996; but no significant change in $\delta^{15}N$. Also displayed are SI signatures of possible feed sources for mussels: fish feed/faeces and seston (from samples collected in North West Bay in April 1997 and December, 1997). ? identifies four feed samples that might be considered outliers.

The supplementary sample of mussels collected in April 1997 indicated mussels grown directly on fishcages for approximately one year were slightly enriched in $^{13}C$ (mean $\delta^{13}C = -19.28 \pm 0.10‰$, n = 4 samples) relative to mussels grown at site IV (-19.68 ± 0.05‰, n = 4 samples) over the same period ($P < 0.05$, Fig. 11.6). However, the difference in $\delta^{15}N$ between the two groups was not significant, although marginal ($P = 0.08$, mean $\delta^{15}N = 9.81 \pm 0.07$, n = 8 samples).

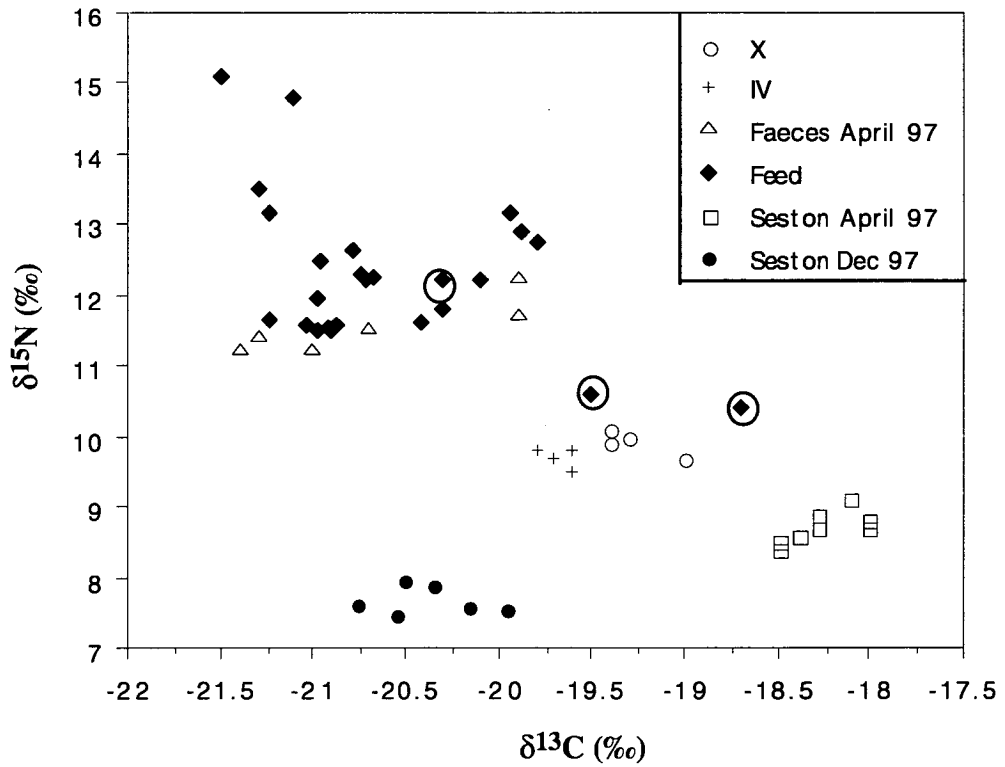


**Figure 11.6.** Stable isotope composition ($\delta^{13}C$ and $\delta^{15}N$) of mantle tissue samples taken from mussels growing directly on fishcages (X) and on a longline 1.5 km from the fishcages (site IV) in April 1997. For comparison, the SI composition of seston samples collected on two dates (April 1997 and December 1997) and fish feed and faecal samples are also included. The SI compositions of three feed samples given to the fish in April 1997 are circled. Mean $\delta^{13}C$ values were significantly different between the two mussel groups ($P < 0.05$) but mean $\delta^{15}N$ values were similar ($P > 0.05$).

## 11.4 Discussion

### 11.4.1 Site differences

The primary objective of SI analyses was to determine if mussels grown on the fish farm were actually consuming and assimilating fish farm waste material, inferred by a different isotopic signature than mussels grown distant from the farm. As expected, the SI signatures of both groups of mussels were similar at the start of the trial - all test mussels were harvested from fishcages at the same farm. Therefore, SI compositions reflected a similar diet consisting of natural sestonic particles as well as any assimilated fish feed or faecal particles. After stripping mussels from the fish cages and deployment to their respective longline sites, SI signatures of the on-farm and off-farm groups were expected to diverge over time. Site I mussels might retain their pre-trial SI composition if fish feed and faecal particles continued to be a significant part of their diet. Site IV mussels might display a shift in their SI composition due to the loss of fish farm waste particles as a food source, reflecting a diet of only natural sestonic particles. However, no such divergence was observed. Further, no obvious deviations in the SI composition of site I mussels were observed between fish culture and fallowing periods that might indicate any significant shifts in food sources (ie. fish farm wastes from March to June 1995 $\Rightarrow$ natural seston from July to September 1995 $\Rightarrow$ fish farm wastes from October 1995 through to May 1996).

The results of both the $\delta^{13}C$ and $\delta^{15}N$ analyses indicate that both mussel groups displayed similar trends of increasing $\delta^{13}C$ over time, but with no significant change in $\delta^{15}N$. The close coupling of SI composition and lack of any significant site x time interaction suggests that both groups were feeding on similar particulate material throughout the trial period (including fish culture and fallowing periods), raising the possibility that mussels at site I were not consuming fish wastes. This supposition is not unreasonable; it has been shown previously that the POM and %POM concentration at site I did not differ significantly from site IV during the trial period (see Section 4). Therefore, due to low current speeds, site I may have been too far from the fishcages for fish waste particulates to reach the suspended mussels before settling to the bottom. However, there are other possible factors that might also explain the lack of differences between the two groups.

Despite their utility, problems may arise when stable isotopes are used to determine sources of diet. These problems arise when: (i) food materials of different origins or composition have similar SI signatures; (ii) the SI content of a particular food material is not unique; (iii) more than two sources of feed are being consumed; (iv) the SI composition of a particular food source does not remain relatively constant; or (v) the

degree and direction (depletion or enrichment) of the fractionation within a consumer is not known (Fry and Sherr, 1984; Owens, 1987; Peterson and Fry, 1987). Unfortunately, many of these problems, and others, were apparent in this study. As a result, SI analyses does not provide unequivocal evidence to conclude that fish farm wastes were, or were not, consumed by mussels at site I. Some of the problems in resolving this question are addressed in the following discussion.

As filter feeders, mussels filter and ingest a variety of suspended particulates including bacteria, phytoplankton, microzooplankton and detritus (Seed and Suchanek, 1992). SI composition varies between, and also within, each of these food sources (Owens, 1987). Sestonic SI composition may display significant spatial and temporal variation according to phytoplankton species composition and growing conditions, zooplankton composition, as well as the type and origin (ie. marine or terrestrial) of detrital components (Fenton, 1981; Fry and Sherr, 1984; Mariotti et al., 1984; Mitchell et al., 1996). Temporal variation in sestonic SI signatures was evident in this study, where both sestonic $\delta^{13}C$ and $\delta^{15}N$ differed between samples collected on two occasions (April and December 1997). Further, other than particulate organic matter, mussels may also acquire a significant proportion (13-40%) of their metabolic requirements from dissolved organic matter (DOM) such as free amino acids and sugars (Siebers and Winkler, 1984; Hawkins and Bayne, 1992 and references within). Therefore, the SI signature of omnivorous animals, such as mussels, is a composite of many particulate and dissolved SI pools assimilated during their feeding history. SI composition would be expected to vary over time according to the temporal variations in sestonic and DOM SI composition. With such a wide range of available nutrient sources, the ability to unequivocally identify individual food items such as fish wastes in mussel tissues may be limited.

Another problem in assessing if fish farm wastes were being assimilated comes from the isotopic composition of the fish feed, where considerable variation in SI composition amongst samples was apparent; it follows that faecal SI composition would also vary with the feed SI composition. Feed SI variation may have been due to differing sources of ingredients or formulations between different feed batches, or merely an artefact of the sample preparation and analyses. Due to the limitations of the analytical technique, subsamples of each feed batch were extremely, but necessarily, small (approximately 2 mg dry weight). With such small subsamples, even minor contaminations might significantly affect the SI analysis. Also, subsample SI values may not necessarily have been representative of the feed if samples were not adequately ground into a fine and homogeneous consistency. The uncertainty as to the cause of the observed variation amongst feed batches underlines the importance of replication, although financial considerations were also involved. If the feed SI

composition did actually vary as the analyses suggest, it follows that fish farm mussels' SI composition might also vary - assuming a significant portion of waste feed and faecal material was assimilated. Neither feed nor seston SI composition was consistently monitored throughout the trial; therefore variations in mussel SI composition reflecting variations in feed or seston SI composition cannot be tested.

It should be reiterated that only a few feed samples (8) were collected during the period of the mussel growth trial. The remaining 17 samples were from feeds manufactured after the conclusion of the trial and are only indicators of the possible range of feed SI content during the trial. Secondly, faecal samples being depleted in $^{13}C$ and $^{15}N$ relative to ingested feed is contrary to reports indicating significant enrichment in animal faeces (citations in Peterson and Fry, 1987; Ye et al., 1991). Therefore, the faecal $\delta$ values may be suspect and certainly suffer from a lack of adequate replication. However, assuming that the range of analysed feed/faecal SI contents are representative of the waste particulates released from the farm during the study, feed and faecal $\delta^{13}C$ values (range -18.7‰ to -21.5‰; mean -20.6 ± 0.1‰) were not particularly distinct from sestonic $\delta^{13}C$. The seston $\delta^{13}C$ values of the two samples taken in April 1997 and December 1997 (range -18.0‰ to -20.7‰) lie within the range of $\delta^{13}C$ typical of temperate marine phytoplankton (-18‰ to -24‰ with a mean of 21‰ used for mixing models; Fry and Sherr, 1984) and are similar to those previously reported from adjacent waters of Storm Bay in the winter months (-18.6‰ to -20.5‰; Fenton, 1981; 1985). Due to the overlap in $\delta^{13}C$ values between seston and feed/faeces, carbon isotopes are of limited value in determining if mussels at site I were consuming fish farm waste particles, unless one can identify periods where waste $\delta^{13}C$ and seston $\delta^{13}C$ were distinct. $\delta^{13}C$ may have been more useful if regular seston sampling was employed and the results revealed seston $\delta^{13}C$ values were significantly different than fish feed/faeces for a significant portion of the trial period.

For the sake of discussion, let's assume:

1. the mean seston $\delta^{13}C$ (-19.4 ± 0.5‰), derived from samples collected in April 1997 and December 1997 along with three values reported from seston samples collected in adjacent waters in August-September 1981 and July 1982 (Fenton, 1981; 1985), was representative of the 1995-96 seston $\delta^{13}C$ values during the study period,
2. the mean fish feed/faeces $\delta^{13}C$ (-20.6 ± 0.1‰) was representative of the fish feed/faeces signatures over the same period, and
3. mantle tissue enrichment is approximately 1‰ relative to the dietary $\delta^{13}C$ (DeNiro and Epstein, 1978; although 2‰ enrichment has been proposed by Ruckelhaus et al., 1993).

ANOVA suggests that the mean fish feed/faeces $\delta^{13}C$ is significantly lower than the mean seston $\delta^{13}C$ ($P < 0.01$). Therefore, the overall mean mussel $\delta^{13}C$ (-18.6‰) represents an enrichment factor of approximately 1‰ relative to seston (-19.4‰); 1‰ is the assumed mean $\delta^{13}C$ enrichment factor and is therefore appropriate if seston was the primary source of food. If mussels at site I were obtaining a significant part of their diet from fish wastes they should have displayed slightly lower $\delta^{13}C$ values, representative of a higher portion of dietary $^{13}C$ in ingested fish feed/faeces, than mussels at site IV. However, lower $\delta^{13}C$ values of mussels at site I were not observed.

Significant differences between seston and fishwaste $\delta^{13}C$ might explain why the mussel $\delta^{13}C$ values at the start of the trial became progressively more enriched after being transplanted to the study longlines. The shift in mussel $\delta^{13}C$ suggests a change in diet towards food source(s) with a higher proportion of enriched $^{13}C$ (seston) and away from sources with lower $\delta^{13}C$ values (fish feed/faeces). Since mussel $\delta^{13}C$ values were not significantly different between sites I and IV, one might conclude that mussels at site I were not obtaining a significant proportion of their diet in the form of fish feed or faeces.

Stable nitrogen isotopes may be more useful in determining if site I mussels were incorporating fish farm wastes. The SI content of feed/faeces are distinguishable from seston using $\delta^{15}N$ values; feed/faeces $\delta^{15}N$ values were consistently higher (range 10.4‰ to 15.1‰) than sestonic $\delta^{15}N$ (range 7.5‰ to 9.1‰). The seston $\delta^{15}N$ values in this study were within the ranges of 6‰ to 10‰ for SPM, and 3‰ to 12‰ for phytoplankton reported in other studies of marine environments (reviewed in Owens, 1987). Therefore, if we assume:

1. Mean seston $\delta^{15}N$ values did not exceed 10‰ through the trial period and the mean fish feed/faeces $\delta^{15}N$ of 12.1‰ was representative of the feed used during the trial;
2. Mussels at site IV did not obtain any dietary contribution from fish feed/faeces; and
3. Mussel mantle tissue is enriched by 3‰ to 6‰ relative to dietary $\delta^{15}N$ (Incze et al., 1982; Yelenik et al., 1996);

then it appears unlikely that mussels at site I were obtaining a significant proportion of their dietary intake from fish feed or faeces. If so, they should have been more enriched in $^{15}N$ than mussels at site IV. Mussels at site IV would be expected to become more depleted in $^{15}N$ due to the loss of fish feed/faeces as a food source. As $\delta^{15}N$ values did not display any clear differences between the two longline sites, it appears unlikely that mussels growing at site I obtained any significant dietary contribution from fish farm wastes.

If mussels derived the bulk of their diet from natural seston, the range of mussel $\delta^{15}N$ values (9.1‰ to 9.7‰, mean 9.5‰) indicates that their primary ration was suspended organic matter in the range of 3.1‰ to 6.7‰ $\delta^{15}N$ (assuming that enrichment was in the order of 3‰ to 6‰, Incze et al., 1982; Yelenik et al., 1996). As sestonic $\delta^{15}N$ values (7.5‰ to 9.1‰) did not fall into this range several possibilities for the observed mussel $\delta^{15}N$ values can be suggested: (i) sestonic $\delta^{15}N$ values of the two 1997 samples were not representative of the sestonic $\delta^{15}N$ during the course of the study, ie. overall sestonic $\delta^{15}N$ values were actually much lower than the two sestonic $\delta^{15}N$ values measured in 1997; (ii) the 1997 sestonic $\delta^{15}N$ values were representative of the trial $\delta^{15}N$ values, but mussel mantle enrichment is less than 3 to 6‰ relative to their diet; (iii) mussels were selectively feeding on specific sestonic particulates with a lower $\delta^{15}N$ than the mean sestonic $\delta^{15}N$; or (iv) mussels were selectively feeding on different particulates with a wide range of $\delta^{15}N$ values, resulting in tissues with an intermediate $\delta^{15}N$.

Without a consistent SI signature of the fish feed or conscientious monitoring of feed and seston SI compositions, and with the likelihood of multiple food sources being consumed, the relative contribution of fish feed and faeces in the diet of mussels cannot be evaluated. However, even if the SI compositions of feed, faeces and seston were isotopically distinct, how realistic is the expectation of actually detecting feed and faecal stable isotopes in mussel tissue?

It would be expected that mussels growing within a fish farm, although exposed to fish farm wastes, would derive the bulk of their dietary intake from the ambient seston. Although particulate wastes from a fish farm may be considerable, they may actually constitute a minor fraction of the total particulate concentration in the water column. Theoretical calculations of particulate wastes generated from the Aquatas farm during the study period suggest, due to the effect of dilution, fish farm waste particles may only contribute less than 2% of the total available particulates *within the fish cages* (see Section 13). Therefore, the maximum contribution to mussel diets from fish farm wastes would be less than 2%. This contribution might actually be less for mussels at site I due to a significant portion fish farm waste particles settling out of the water column before reaching the suspended mussels. Also, mussels may display pre-ingestive selection of a specific size range of particles or certain components of the seston (eg. selecting algal cells over detritus), which might further reduce the contribution of farm waste isotopes in mussel tissue. Although based on theoretical calculations involving a number of assumptions (and POM concentrations are of whole water column samples rather than concentrations at discrete depths), it appears that the contribution of fish farm wastes to total POM is probably minor. It seems unlikely that such a small contribution of waste particles to the diet (< 2%) would alter a

mussel's SI signature to detectable levels. Therefore, mussel tissue SI composition would largely be a reflection of sestonic SI composition. This conclusion is supported from the mussel samples collected in April 1997, where mussels living directly on fishcages for over one year were compared with mussels grown at site IV for the same period. Albeit a small sample of mussels, there was no difference in $\delta^{15}N$ between the two groups and only a slight, but significant ($P < 0.05$), difference in $\delta^{13}C$ (fishcage mussels $\delta^{13}C$ -19.28 ± 0.10‰ vs site IV mussels -19.68 ± 0.05‰, n = 4 samples). The difference in $\delta^{13}C$ does not make much sense. If fish farm mussels were consuming some fish feed, their $\delta^{13}C$ values would be expected to be slightly lower than mussels at site IV. The site difference in $\delta^{13}C$ becomes negligible ($P > 0.05$) when differences in lipid content between sexes are accounted for and $\delta'^{13}C$ values are compared (see discussion on sex differences and lipid normalisation in 11.4.2). Therefore, fish waste particulates might be expected to be a significant fraction in mussel diets only during periods of low ambient particulate levels.

Whether or not mussels grown within a fish farm actually consume a significant portion of fish waste particles in their diet and whether these particles can be detected in mussel tissue using SI analysis might be resolved in a more controlled experiment. Mussels could be grown in tanks under different combinations of dietary sources to determine if fish feed/faecal nutrients are incorporated in detectable quantities and to determine if addition of fish feed/faeces to natural seawater enhances mussel growth. Examples of treatments might include: (A) filtered water - control group, (B) Filtered water with ground fish feed, (C) filtered water with faeces (probably unrealistic), (D) filtered water with mixed fish feed/faeces (could be collected in sediment traps beneath fish cages), (E) natural water with fish feed/faeces, and (F) natural seawater. The experiment could closely monitor stable isotopic content of the diets and mussel tissue to determine if mussels were actually assimilating fish feed or faeces and estimate their contribution to the total mussel diet. The experiment might also show differences in growth rates according to diet.

### 11.4.2 Sex differences

The only unambiguous observation from the SI analyses appears to be the differences in $\delta^{13}C$ isotopic signatures between males and females. Females were significantly lighter in $^{13}C$ than males except for the last two months of the trial period. Why the difference? It has been suggested that lipids normally possess lower $\delta^{13}C$ values than protein and carbohydrate and, therefore, animals with a higher fat content tend to be isotopically lighter than "leaner" animals (McConnaughey and McRoy, 1979). To facilitate comparisons between animals with different fat contents, McConnaughey and

McRoy (1979) suggested the use of "lipid normalised" $\delta^{13}C$ values ($\delta'^{13}C$) calculated from $\delta^{13}C$ and carbon: nitrogen ratios. Using this approach, the difference in mantle lipid levels as the source of variation between sex $\delta^{13}C$ values was tested.

Lipid content (LC) and $\delta'^{13}C$ were calculated from equations derived by McConnaughey and McRoy (1979):

$$LC = 93 / [1 + (0.246\ C/N - 0.775)^{-1}]$$
$$\delta'^{13}C = \delta^{13}C + D\ [-0.207 + 3.90 / (1+287/LC)]$$

where D is the isotopic difference between protein and lipid (assigned a value of 6‰ by McConnaughey and McRoy, 1979). Percentage of carbon and nitrogen content in mantle tissue was provided from the stable isotope analysis and used to calculate C/N ratios.

Overall mean C/N ratios and calculated lipid contents were higher in females than in males ($P < 0.001$ and $P < 0.0001$, respectively). Month x sex interactions (both $P < 0.0001$) and post hoc tests suggest that sex differences in C/N and lipid contents were significant for all months up to February 1996, after which differences were negligible (Fig. 11.7A). This supports Murphy and Abrajano (1994) who reported a lower abundance in fatty acids in male *Mytilus edulis* and *Modiolus modiolus* compared with females. A higher lipid content in females may be due to the production of lipid vitelline reserves from glycogen during oogenesis (Zwaan and Mathieu, 1992).

After lipid normalisation, no significant difference in overall mean $\delta'^{13}C$ values between sexes nor sex x time interactions were observed ($P \geq 0.05$, Fig. 11.7B and C). This provides a strong case for $\delta^{13}C$ differences among sexes being due to higher fat contents in females. It is very interesting to note that the variation in $\delta'^{13}C$ values correspond very closely with the reproductive cycle and pattern of tissue glycogen content, where gametogenic activity preceded major spawning events in August 1995 and February 1996 (see Sections 7 and 8). It is, therefore, possible that the observed variation in mussel $\delta^{13}C$ values during the trial period was also influenced by their reproductive development rather than solely by temporal variation in sestonic $\delta^{13}C$. Again, without detailed knowledge of the sestonic $\delta^{13}C$ variation during the trial, this possibility cannot be tested. However, an important consideration for future investigations of SI composition in bivalves is the real possibility of differences in stable isotope signatures between sexes, due to differences in lipid content. If not accounted for, these sex differences may influence experimental treatment effects. This was apparent in the April 1997 mussel samples, where $\delta^{13}C$ values between fishcage and site IV mussels were significantly different; however, after lipid

normalisation, mean $\delta^{13}C$ values were similar. Future studies should incorporate equal proportions of males and females in all samples or at least be aware of differences in sex ratios among samples. Alternatively, lipid normalised $\delta'^{13}C$ values should be used to avoid the discrepancy in $\delta^{13}C$ values between sexes.

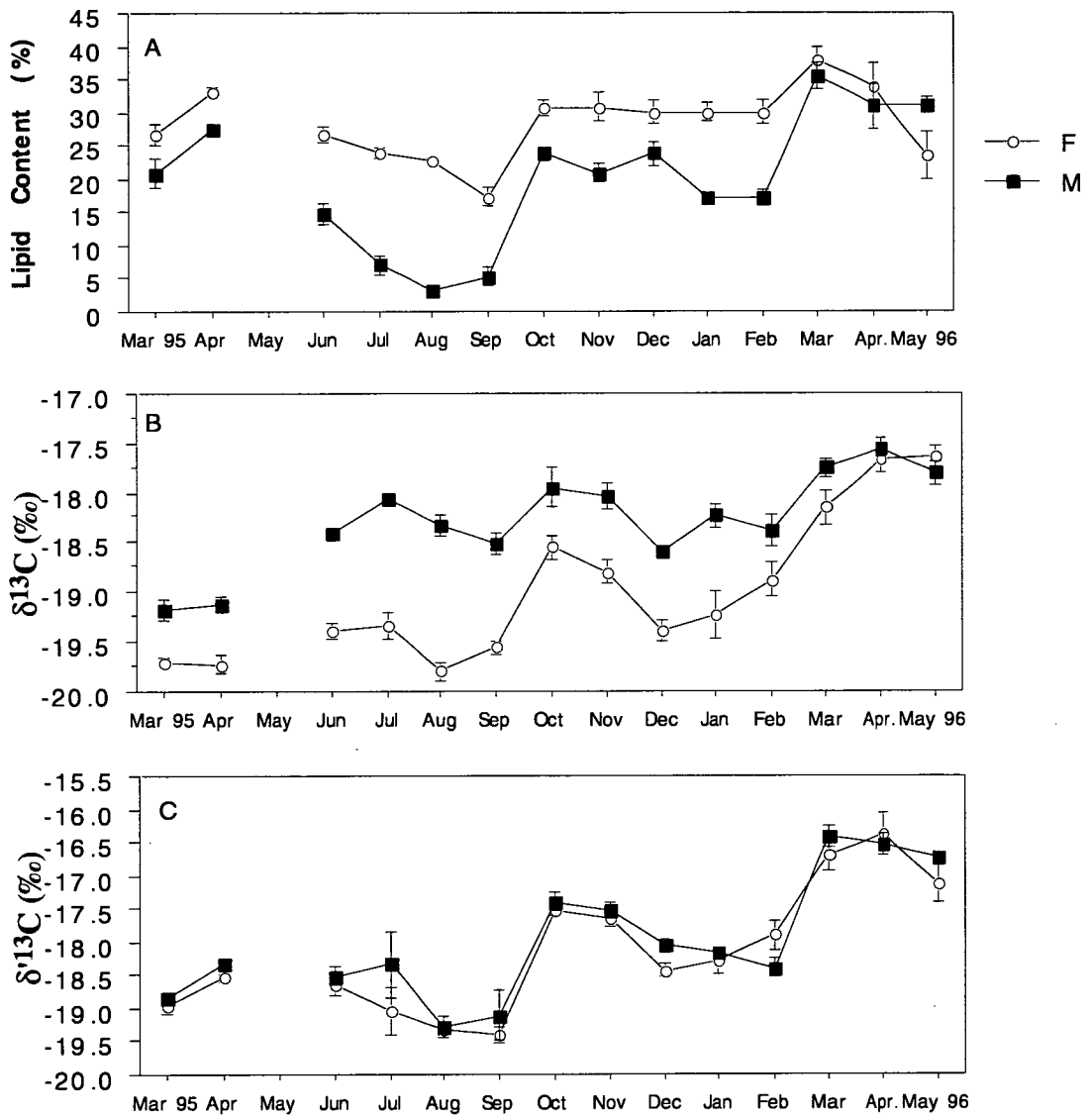


**Figure 11.7.** (A) Monthly lipid content (mean ± SE% dry meat weight), (B) analysed $\delta^{13}C$ unadjusted for lipid content (mean ± SE‰) and (C) "lipid-normalised" $\delta'^{13}C$ values (mean ± SE‰) of mussel mantle tissue according to sex (monthly n = 4 samples per sex). Lipid content was significantly different between sexes for all months except March 1996 to May 1996. Significant differences in $\delta^{13}C$ between males and females were indicated for all months except April 1996 and May 1996. After lipid normalisation, no monthly sex differences were significant in $\delta'^{13}C$ values. Significance level = 0.05.

# 12. Scope for Growth

## 12.1 Introduction

Previous sections have outlined many of the similarities in monitored performance indices (shell and soft tissue growth, whole live weight, condition, biomass production, reproduction, survival) between mussels cultured within the Aquatas fish farm (sites I and II) and mussels cultured at non-fish farm sites (sites III and IV). These results suggest there is little advantage in culturing mussels within the fish farm, at least for enhancing mussel growth. However, there are questions regarding some factors that may have influenced the performance of mussels during this trial. As such, an unambiguous conclusion, that integrated mussel-salmon culture at the Aquatas fish farm in North West Bay has no potential for enhancing mussel performance, may be premature.

Some factors that may have possibly influenced the growth of experimental mussels include variable sock densities (Section 8), pea crab infestation (Section 9) and, most notably, overspat settlements (Section 10) and other forms of biofouling. Fouling of socks by mussel overspat, sponges, ascidians and macroalgae became increasing severe during the course of the trial and may have significantly reduced the suspended food supply to the underlying experimental mussels. As biofouling, other than overspat, was not quantified nor monitored, it is unknown if this component differed among sites. Increases in dissolved nutrient concentrations adjacent to fish cages have been implicated in the enhanced growth of macroalgae cultured within fish farms (eg. Troell et al., 1997). It is possible that biofouling may have been more severe at the two fish farm sites; thereby reducing available food levels for mussels more so than at the other two sites and, thereby, negating any potential nutritional advantage derived from particulate fish farm wastes.

No evidence for any negative effects of sock density, overspat density or pea crab infestation on mussel growth was detected. However, the author was not totally confidant that these factors, algal fouling, or combination of factors, had no influence on the growth of experimental mussels. As there were uncertainties regarding the effects of these factors on mussel growth, it was desirable to evaluate the potential for mussel growth at each site, independent of sock density, overspat density, pea crab infestation, and biofouling density. If this could be realistically achieved, a more confident conclusion of the relative merits of integrated mussel-salmon culture for enhancing mussel growth might be drawn.

Secondly, no obvious seasonal variation in mussel shell growth was observed during this trial. Temporal variation in growth has been reported elsewhere, whereby many temperate and boreal mussel populations display slow growth during the autumn-winter period and increased growth during spring-summer (Mason, 1972; Okumus, 1993). However, elevated growth rates during winter periods have been observed by others (Wallace, 1980; Page and Hubbard, 1987). Physiological and ecological evidence indicates that in many situations, temporal variations in food availability, which includes the concentration and composition of phytoplankton and other suspended particulates, may be the single most important factor regulating mussel growth (Seed, 1976; Widdows et al., 1979; Incze et al., 1980; Rodhouse et al., 1984b; Frechette and Bourget, 1985). In the present study, shell growth was continuous throughout most of the trial, albeit at decreasing rates, until the final three months when growth effectively ceased. Was the observed growth pattern representative of the prevailing environmental conditions or was growth also influenced by other factors such as the increasing age of mussels and/or the increasing density of overspat and other biofoulants?

Thirdly, it was also desirable to assess the potential for mussel growth closer to the salmon cages than the positions of the within-farm experimental longlines (sites I and II). Enhanced growth of bivalves has been reported in some studies where bivalves were cultured within, or immediately adjacent, to fish cages (Wallace, 1980; Jones and Iwama, 1991; Okumus, 1993). Although sites I and II were within the bounds of the fish farm lease, they were 70 m and 100 m away from the fish cages. It was considered that these two longline locations may have been too far from the fish cages to intercept a significant fraction of suspended waste particles (see Section 13). Therefore, an assessment of the available food immediately next to the fish cages might reveal a greater potential for enhancing mussel growth than sites I and II.

In order to address the above questions, "scope for growth" was estimated over the trial period to evaluate the temporal and spatial variation of environmental factors on the potential for mussel growth in North West Bay. The concept of scope for growth (SFG), as applied to mussels, has been reviewed by others (eg. Bayne et al., 1976b; Bayne, 1998). SFG is an instantaneous measure of the potential energy in ingested food particles that is available for mussel growth and reproduction, after accounting for energy losses through respiration and excretion. SFG, measured in energy units (eg. $J \cdot h^{-1}$), can range from maximum positive values, under optimum environmental conditions, to negative values when feed is scarce or the animals are severely stressed and utilising body reserves for maintenance. Therefore, negative SFG is associated with conditions that lead to losses in soft tissue biomass. In many studies, SFG has been an effective tool for evaluating and comparing the suitability of bivalve culture

sites and for explaining temporal variation in observed growth patterns (Bayne and Widdows, 1978; Bayne et al., 1979; Widdows et al., 1984; Kautsky et al., 1990; Frechette and Grant, 1991; Okumus and Stirling, 1994).

Inputs of physiological responses to several key environmental conditions are required to estimate the quantity of ingested food and the potential energy available for growth. These responses include particle filtration, ingestion and absorption rates, to estimate potential energy gains; and respiration and excretion rates, to estimate energy losses. Also, estimates of assimilated energy require measures of the energy content and digestibility of suspended food particles. As these physiological parameters and particulate energy contents were not measured in this study, literature values were employed in the calculation of SFG. Therefore, all estimates of the temporal variation in SFG at each experimental site (sites I-IV) and at the centre of the main fish cage unit (site X) are strictly hypothetical. In order to assess the validity of SFG estimates, the actual growth of experimental mussels biomass was compared with the growth predicted from SFG calculations. Differences between predicted and observed growth are discussed.

## 12.2 Methodology

SFG calculations incorporate several physiological responses to environmental parameters for estimating potential biomass production. The general energy equation for calculating SFG is:

$$SFG = A - (R + U)$$

where SFG is the potential energy available for growth of soft tissue (dry weight of meat, gametes, byssus and shell organics), A is the absorbed food energy, R is the respiratory heat loss and U is the energy lost by excretion (energy units = $J \cdot time^{-1}$). The physiological response parameters required for the estimation of SFG were not directly measured in this study. These parameters were estimated from relationships derived from the literature (outlined in Page and Ricard, 1990), incorporating environmental factors (seston concentration, %POM and temperature) measured during the course of this investigation, where:

Clearance rate (CR, $l \cdot h^{-1}$) $= 1.73 \cdot W^{0.413} - 0.006 \cdot (seston)$

Seston filtered (SF, $mg \cdot l^{-1}$) $= CR \cdot (seston)$

Pseudofaeces threshold (T, $mg \cdot l^{-1}$) $= 3.81 \cdot \log L - 1.93$

% pseudofaeces (%Ps) $= 100 - [(86.4\ e^{0.288\ T}) \cdot (seston)^{-(0.489\ T\text{^}0.329)}]$

Seston ingested (SI, $mg \cdot h^{-1}$) $= SF - (SF \cdot \%Ps/100)$

Absorption efficiency (AE) $= 0.5 \cdot \log(\%POM) - 0.32$

POM absorbed (A, $mg \cdot h^{-1}$) $= (SI \cdot \%POM/100) \cdot (AE/100)$

Respiratory rate (R or $VO_2$, ml $O_2 \cdot h^{-1}$)$= a\ W^{0.782}$, where $a = 0.117 \cdot (10^{0.044\ Temp})$

Scope for Growth (SFG, $J \cdot h^{-1}$) $= (A \cdot 23.5) - (VO_2 \cdot 20.3)$

W is mussel dry meat weight (DMW, g), L is shell length (mm), (seston) is total particulate matter or seston concentration ($mg \cdot l^{-1}$), % POM is percent of seston represented by particulate organic material and Temp is water temperature (°C). Water temperature, seston concentration and %POM were measured approximately weekly. Absorbed food was estimated from measurements of mussel size, seston concentration, %POM, and from published values for size-specific clearance rates, ingestion rates and absorption efficiencies. When possible, clearance rates, ingestion rates and absorption efficiencies were based on studies using "natural" POM rather than cultured algae. Clearance and ingestion rates were assumed to be independent of water temperature, but dependent on mussel size and seston concentration (Foster-Smith, 1975; Bayne et al., 1976a; Widdows, 1978a; Widdows et al., 1979). The fraction of filtered seston rejected as pseudofaeces (%Ps) was determined from the filtration rate and the size-specific critical seston concentration for rejection (eg. 4.6 $mg \cdot l^{-1}$ for a 55 mm mussel, Widdows et al., 1979). Metabolic expenditures were estimated according to mussel size, water temperature and published data on oxygen consumption rates (Thompson, 1984a; Widdows et al., 1984). Oxygen consumption and respiratory losses were assigned an energetic value of 20.3 $J \cdot ml\ O_2^{-1}$ (Widdows, 1978b).

The energy content of absorbed POM was assigned a constant value of 23.5 $J \cdot mg\ POM^{-1}$ at all sites (ie. sestonic energy content suggested by Widdows et al., 1979). As total POM and non-algal POM concentrations did not differ among longline sites I to IV (Section 4.3.7), it was assumed that fish farm wastes did not significantly contribute to the available food supply at these sites. Overall trial mean POM concentration at site X was 15% greater than at the other sites. The extra POM was assumed to be excess fish feed fines and faecal particles. Based on the gross and digestible energy content of the fish feed, 22.0 $J \cdot mg\ feed^{-1}$ and 19.4 $J \cdot mg\ feed^{-1}$ respectively (Pivot Aquaculture), and the average farm suspended solid waste loadings (fish feed fines and faeces, Section 13), the energy content of suspended matter at site X was calculated as 87% x 23.5 $J \cdot mg\ POM^{-1}$ (natural seston) + 13% (0.94 x 25.75

J·mg dry faeces$^{-1}$) + 13% (0.06 x 26.45 J·mg dry fish feed fines$^{-1}$). The resulting energy content of the suspended POM at site X (23.8 J·mg POM$^{-1}$) was within 1% of natural POM. It is possible that suspended waste particles comprised a much smaller fraction of the total suspended matter available for mussels than 15% (ie. possibly less than 2.0% of total suspended POM; the difference may have been due to contamination of water samples from bottom sediments; see Section 13). Therefore, the gross energy content of suspended POM at site X would probably not be any greater than natural seston and was allocated the same value as the other sites (23.5 J·mg POM$^{-1}$).

Energy losses due to excretion are considered to be minor (less than 5% of absorbed ration, Kiorboe et al., 1981; Page and Ricard, 1990; Smaal et al., 1997) and were ignored in SFG calculations.

As comprehensive monitoring of all environmental parameters required for SFG calculations did not commence until June 1995 (ie. seston and %POM), SFG for the first three months of the growth trial (March - May 1995) was not determined. Weekly theoretical SFG for a "standard" mussel of 1.0 g (DMW) and 59 mm shell length was calculated for each experimental mussel longline site (I - IV) and for the centre of the fish farm (site X), incorporating the required environmental parameters (seston, %POM, temperature) measured weekly at each site.

Weekly SFG was also determined to account for the increasing size of the experimental mussels by incorporating observed mean shell lengths and DMWs into the calculations. As test mussels were only sampled monthly, weekly mussel lengths and DMW were estimated from mean site specific growth rates (length and DMW) achieved each month, where:

$$\text{specific growth rate } (\%\cdot\text{day}^{-1}) = [\ln (L_n) - \ln (L_{n-1})/30 \text{ days}] \times 100$$

and $L_n$ and $L_{n-1}$ represent the site mean length (or DMW) each month and previous month, respectively.

The experimental mussel SFG was also calculated on a monthly basis where
SFG (kJ·month$^{-1}$) = mean monthly SFG (J·h$^{-1}$) x 24 h x 30 days x 0.001.
This allowed for comparisons of the potential for growth (based on monthly theoretical SFG estimates) with the actual growth (ie. monthly energy increments) of the test mussels. Monthly mussel energy increments were calculated for each site as the change (increase or decrease) in mussel soft tissue energy content + increase in shell energy content + monthly byssus production. Soft tissue energy content was calculated for each month from the relationship Y (kcal·g DMW$^{-1}$) = 0.0559 x % organic content (Winberg, 1971). Kilocalories were converted to kilojoules by multiplying by 4.184. As the variation in monthly soft tissue organic content and

energy contents were minor (range 20.5 - 21.2, mean 21.0 kJ·g $DMW^{-1}$), the change in energy content at each site was calculated as Δ mean DMW x 21.0 kJ·g $DMW^{-1}$. The increase in shell energy content at each site was calculated by Δ mean dry shell weight x 0.75 kJ·g dry shell $weight^{-1}$ (Sukhotin, 1992) and monthly byssus production was taken as 0.08 kJ·$month^{-1}$ (Hawkins and Bayne, 1985).

Theoretical SFG for overspat that settled on the experimental droppers in May 1996 was also calculated. Mean shell lengths, measured monthly at each site, were used to estimate DMW by the allometric relation DMW (g) = 0.0058*$L(cm)^{2.8}$, where the constants a and b are the annual means from monthly regressions reported by Thompson (1984a). Monthly spat SFG (kJ·$month^{-1}$) was calculated and compared with estimated monthly DMW energy increments as previously described for experimental mussels.

Theoretical SFG over the period June 1995 to May 1996 was assessed by ANOVA, with SITE and MONTH as fixed factors using the four weekly SFG estimates per month at each site as replicates. Normality (Shapiro-Wilks test) and homogeneity of variances (Cochran's test) were confirmed prior to analysis. ANOVA was also used to determine if monthly mussel growth (mean biomass energy increment at each site, n = 4) differed from predicted growth (monthly SFG at each site, n = 4). Correlation analysis was used to assess the relationship of monthly SFG with monthly biomass energy and shell length increments.

## 12.3 Results

Environmental and physiological parameters used in calculating the 1.0 g standard mussel SFG, averaged by month, are summarised in Table 12.1. Standard mussel (1.0 g DMW) SFG over the period June 1995 to May 1996 displayed significant temporal (monthly) variation ($P < 0.0001$), but no significant differences among sites (overall mean 21.9 ± 0.7 J·$h^{-1}$, n = 225 estimates) nor any site x month interaction ($P > 0.05$). SFG displayed a general decline from high levels in winter (1 June 1995, mean 33.0 ± 3.2 J·$h^{-1}$, n = 5 estimates) to minimum levels in late spring-early summer (21 December 1995 mean - 0.1 ± 0.3 J·$h^{-1}$, n = 5 estimates), although two distinct peaks occurred in September (43.6 ± 1.4 J·$h^{-1}$) and November (29.6 ± 1.4 J·$h^{-1}$) (Fig. 12.1); these peaks corresponded with short-term peaks in POM content. SFG increased in January 1996 and remained at relatively high levels throughout the summer-autumn period (January - May 1996 mean 27.4 ± 0.8 J·$h^{-1}$, n = 85 estimates). Post hoc testing revealed mean SFG for six of the eleven months were statistically equivalent ($P > 0.05$, Fig. 12.2).

The pattern of experimental mussel SFG was similar to the standard mussel SFG, although experimental mussel SFG was slightly greater for most of the trial due to their larger size and, therefore, filtration and ingestion rates. Mean weekly SFG values ranged from a low of - 8.1 ± 1.3 $J \cdot h^{-1}$ (n = 4 estimates) in the first week of December 1995 to maximum values above 40 $J \cdot h^{-1}$ at various times throughout the trial period (Fig. 12.2). As with the standard mussel SFG, experimental mussel SFG displayed significant temporal variation ($P < 0.0001$); but no differences in overall mean SFG among sites (29.4 ± 1.3 $J \cdot h^{-1}$, n = 180 estimates) nor a site x month interaction were significant ($P > 0.05$). Again, post hoc testing revealed SFG for six of the eleven months were statistically equivalent ($P > 0.05$, Fig. 12.3). Environmental and physiological parameters used in calculating experimental mussel SFG are summarised in Table 12.2.

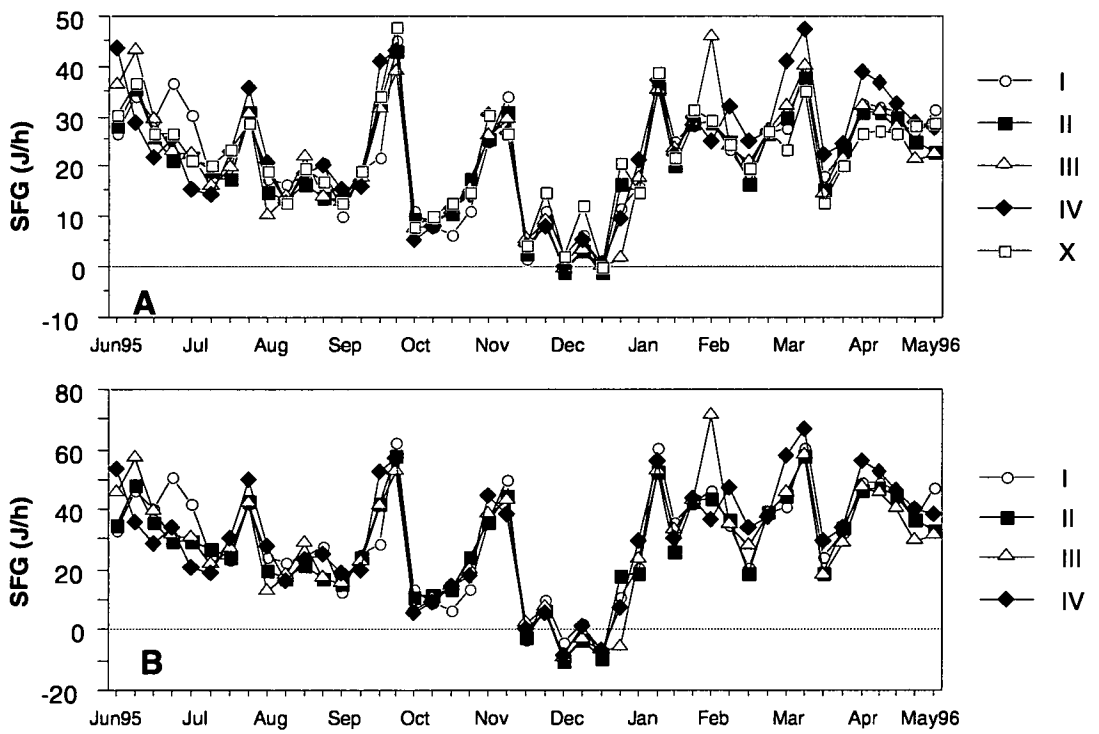


**Figure 12.1.** (A) Theoretical scope for growth (SFG, $J \cdot h^{-1}$) of a standard 1.0 g dry meat weight mussel and (B) of actual experimental mussels based on weekly measurements of environmental factors (seston, %POM and temperature) at each sampling site (I, II, III, IV and X) from June 1995 to May 1996.

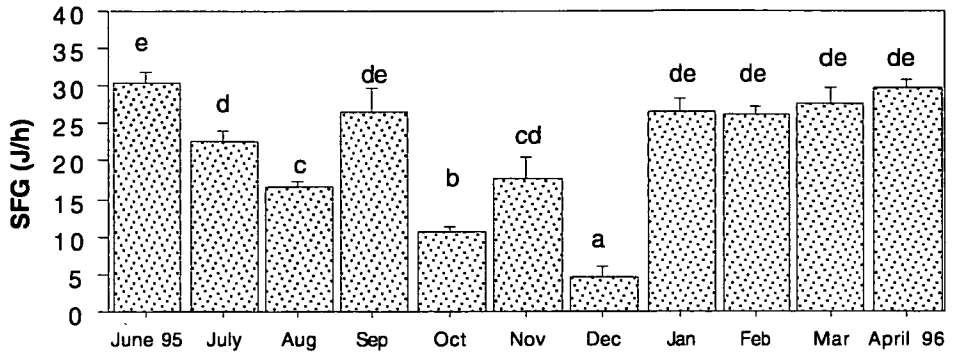


**Figure 12.2.** Mean monthly theoretical scope for growth (SFG, $J \cdot h^{-1} \pm SE$, n = 20 observations per month, 4 from each site I, II. III, IV and X) of 1 g standard mussel in North West Bay from June 1995 to May 1996. Months not sharing a common letter are significantly different ($P < 0.05$).

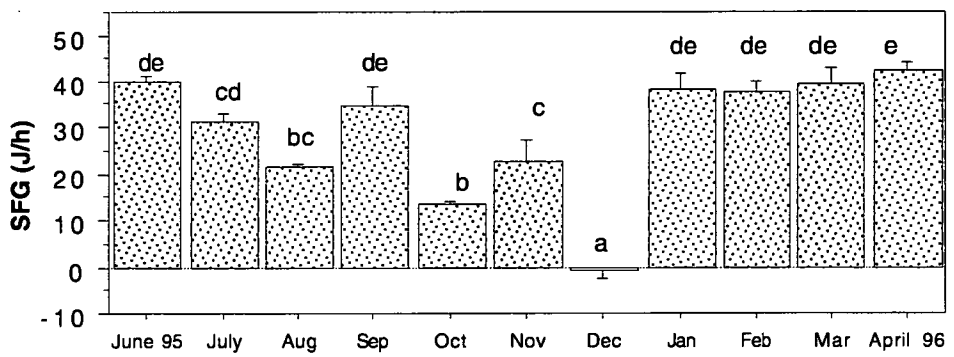


**Figure 12.3.** Mean monthly theoretical scope for growth (SFG, $J \cdot h^{-1} \pm SE$, n = 16 observations per month, 4 from each site I, II. III, IV) of experimental mussels from June 1995 to May 1996. Months not sharing a common letter are significantly different ($P < 0.05$).

SFG closely followed the variation in POM over the trial period, with SFG significantly correlated with POM concentration ($P < 0.0001$, $r = 0.89$, n = 176 observations). Except for six weeks, from mid-November to December 1995, the seston concentrations within North West Bay consistently exceeded the threshold level for pseudofaeces production. This indicates that mussels were probably ingesting suspended particles at maximum rates throughout most of the year. Positive SFG for most of the trial suggests that food availability (POM) and quality (%POM) in NWB were at levels that would promote growth for most of the year. SFG was negligible for six weeks from November to December 1995, corresponding with low POM levels (mean 0.99 mg $POM \cdot l^{-1}$) and low dry meat growth rates (+ 0.07 g $DMW \cdot month^{-1}$) during this period. Despite lower food levels, ingested POM was still adequate to maintain basal metabolic requirements; but not sufficient for growth, as suggested by December SFG $\approx 0.0$ $J \cdot h^{-1}$ and supported by negligible growth of mussels.

**Table 12.1.** Summary of monthly mean environmental parameters (seston, %POM, temperature), theoretical physiological parameters and scope for growth (SFG) for a standard 1.0 g DMW mussel at each longline site in North West Bay (sites I - IV) plus the centre of the fish farm (site X) from June 1995 to April 1996 (averaged by month from weekly estimates). % Ps = percent pseudofaeces, POM = particulate organic matter, T = pseudofaeces threshold and $VO_2$ = respiratory rate.

| Month | Seston (mg/l) | % POM | Temp (C) | Clearance rate (l/h) | Seston filtered (mg/h) | T (mg/l) | % Ps | Seston ingested (mg/h) | Absorption efficiency (%) | POM absorbed (mg/h) | $VO_2$ (ml $O_2$/h) | SFG (J/h) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Site 1** | | | | | | | | | | | | |
| June | 5.6 | 43.0 | 10.5 | 1.70 | 9.42 | 4.90 | 14.46 | 7.73 | 49.7 | 1.65 | 0.34 | 31.73 |
| July | 6.0 | 34.2 | 9.0 | 1.69 | 10.10 | 4.90 | 17.72 | 8.19 | 44.5 | 1.26 | 0.29 | 23.80 |
| August | 6.1 | 30.2 | 9.7 | 1.69 | 10.34 | 4.90 | 19.21 | 8.22 | 41.9 | 1.04 | 0.31 | 18.11 |
| September | 7.7 | 33.9 | 10.4 | 1.68 | 12.94 | 4.90 | 33.58 | 8.52 | 43.7 | 1.32 | 0.34 | 24.06 |
| October | 6.3 | 24.4 | 12.5 | 1.69 | 10.65 | 4.90 | 21.31 | 8.26 | 37.3 | 0.75 | 0.42 | 9.25 |
| November | 5.2 | 34.2 | 13.9 | 1.70 | 8.79 | 4.90 | 13.42 | 7.25 | 44.1 | 1.18 | 0.48 | 17.97 |
| December | 3.9 | 26.1 | 14.6 | 1.71 | 6.58 | 4.90 | 0.00 | 6.58 | 38.1 | 0.65 | 0.52 | 4.88 |
| January | 7.3 | 41.3 | 16.9 | 1.69 | 12.28 | 4.90 | 30.86 | 8.45 | 48.5 | 1.71 | 0.65 | 27.08 |
| February | 8.1 | 38.6 | 16.4 | 1.68 | 13.68 | 4.90 | 36.80 | 8.59 | 47.3 | 1.57 | 0.62 | 24.29 |
| March | 7.7 | 40.7 | 16.5 | 1.68 | 12.92 | 4.90 | 33.36 | 8.52 | 48.3 | 1.69 | 0.63 | 27.10 |
| April | 7.8 | 40.7 | 13.8 | 1.68 | 13.18 | 4.90 | 34.73 | 8.54 | 48.4 | 1.68 | 0.47 | 29.99 |
| **Site II** | | | | | | | | | | | | |
| June | 5.4 | 40.8 | 10.5 | 1.70 | 9.22 | 4.90 | 14.80 | 7.64 | 48.2 | 1.48 | 0.34 | 27.92 |
| July | 6.8 | 32.7 | 9.0 | 1.69 | 11.40 | 4.90 | 25.86 | 8.35 | 43.6 | 1.20 | 0.29 | 22.38 |
| August | 5.8 | 27.4 | 9.7 | 1.69 | 9.90 | 4.90 | 16.57 | 8.17 | 39.9 | 0.89 | 0.31 | 14.66 |
| September | 7.0 | 36.5 | 10.4 | 1.69 | 11.81 | 4.90 | 27.56 | 8.39 | 45.5 | 1.44 | 0.34 | 26.97 |
| October | 5.6 | 27.0 | 12.5 | 1.70 | 9.45 | 4.90 | 13.51 | 8.11 | 39.5 | 0.87 | 0.42 | 12.00 |
| November | 5.3 | 33.6 | 13.9 | 1.70 | 8.96 | 4.90 | 14.42 | 7.22 | 43.9 | 1.14 | 0.48 | 16.99 |
| December | 3.8 | 25.3 | 14.6 | 1.71 | 6.56 | 4.90 | 0.00 | 6.56 | 37.2 | 0.64 | 0.52 | 4.64 |
| January | 6.9 | 39.9 | 16.9 | 1.69 | 11.72 | 4.90 | 28.25 | 8.40 | 47.8 | 1.62 | 0.65 | 24.90 |
| February | 7.9 | 38.6 | 16.4 | 1.68 | 13.25 | 4.90 | 35.42 | 8.56 | 47.3 | 1.57 | 0.62 | 24.25 |
| March | 7.6 | 40.4 | 16.5 | 1.68 | 12.83 | 4.90 | 32.20 | 8.50 | 48.1 | 1.67 | 0.63 | 26.58 |
| April | 8.4 | 39.8 | 13.8 | 1.68 | 14.13 | 4.90 | 38.67 | 8.64 | 48.0 | 1.65 | 0.47 | 29.19 |
| **Site III** | | | | | | | | | | | | |
| June | 5.3 | 45.3 | 10.5 | 1.70 | 8.99 | 4.90 | 13.20 | 7.57 | 50.3 | 1.70 | 0.34 | 32.87 |
| July | 5.8 | 33.2 | 9.0 | 1.70 | 9.76 | 4.90 | 16.19 | 8.16 | 43.9 | 1.20 | 0.29 | 22.24 |
| August | 6.1 | 27.4 | 9.7 | 1.69 | 10.41 | 4.90 | 20.28 | 8.24 | 39.7 | 0.90 | 0.31 | 14.91 |
| September | 7.1 | 35.2 | 10.4 | 1.69 | 12.00 | 4.90 | 28.15 | 8.41 | 44.8 | 1.37 | 0.34 | 25.23 |
| October | 5.3 | 26.1 | 12.5 | 1.70 | 9.02 | 4.90 | 9.95 | 8.04 | 38.7 | 0.82 | 0.42 | 10.77 |
| November | 5.4 | 33.6 | 13.9 | 1.70 | 9.19 | 4.90 | 14.30 | 7.49 | 43.9 | 1.16 | 0.48 | 17.53 |
| December | 3.9 | 21.8 | 14.6 | 1.71 | 6.70 | 4.90 | 1.38 | 6.58 | 34.7 | 0.49 | 0.52 | 1.10 |
| January | 7.2 | 40.8 | 16.9 | 1.69 | 12.17 | 4.90 | 30.38 | 8.44 | 48.4 | 1.68 | 0.65 | 26.29 |
| February | 7.2 | 42.8 | 16.4 | 1.69 | 12.08 | 4.90 | 28.00 | 8.41 | 49.3 | 1.79 | 0.62 | 29.39 |
| March | 7.8 | 40.5 | 16.5 | 1.68 | 13.14 | 4.90 | 34.43 | 8.54 | 48.1 | 1.69 | 0.63 | 26.98 |
| April | 8.5 | 38.9 | 13.8 | 1.68 | 14.26 | 4.90 | 39.08 | 8.65 | 47.4 | 1.60 | 0.47 | 28.02 |
| **Site IV** | | | | | | | | | | | | |
| June | 6.1 | 40.0 | 10.5 | 1.69 | 10.34 | 4.90 | 19.18 | 8.19 | 47.7 | 1.57 | 0.34 | 30.00 |
| July | 5.6 | 33.0 | 9.0 | 1.70 | 9.50 | 4.90 | 14.19 | 8.12 | 43.5 | 1.19 | 0.29 | 22.11 |
| August | 5.7 | 30.2 | 9.7 | 1.70 | 9.72 | 4.90 | 14.99 | 8.14 | 41.9 | 1.03 | 0.31 | 17.99 |
| September | 7.1 | 37.9 | 10.4 | 1.69 | 11.90 | 4.90 | 27.88 | 8.40 | 46.2 | 1.52 | 0.34 | 28.97 |
| October | 5.8 | 25.1 | 12.5 | 1.70 | 9.88 | 4.90 | 16.53 | 8.17 | 37.7 | 0.78 | 0.42 | 9.85 |
| November | 5.6 | 33.1 | 13.9 | 1.70 | 9.47 | 4.90 | 14.50 | 7.60 | 43.5 | 1.16 | 0.48 | 17.42 |
| December | 3.8 | 25.2 | 14.6 | 1.71 | 6.47 | 4.90 | 0.00 | 6.47 | 37.6 | 0.62 | 0.52 | 3.99 |
| January | 6.8 | 42.1 | 16.9 | 1.69 | 11.42 | 4.90 | 26.71 | 8.36 | 49.1 | 1.74 | 0.65 | 27.68 |
| February | 7.8 | 40.8 | 16.4 | 1.68 | 13.20 | 4.90 | 35.04 | 8.55 | 48.5 | 1.69 | 0.62 | 27.24 |
| March | 7.5 | 45.5 | 16.5 | 1.68 | 12.67 | 4.90 | 31.64 | 8.48 | 50.7 | 1.98 | 0.63 | 33.93 |
| April | 8.3 | 43.5 | 13.8 | 1.68 | 13.85 | 4.90 | 36.40 | 8.59 | 49.9 | 1.87 | 0.47 | 34.28 |
| **Site X** | | | | | | | | | | | | |
| June | 5.7 | 42.1 | 10.5 | 1.69 | 10.09 | 4.90 | 19.60 | 7.79 | 48.9 | 1.58 | 0.34 | 30.18 |
| July | 6.3 | 33.3 | 9.0 | 1.69 | 12.51 | 4.90 | 31.52 | 8.47 | 44.1 | 1.25 | 0.29 | 23.50 |
| August | 6.3 | 28.8 | 9.7 | 1.69 | 12.63 | 4.90 | 32.13 | 8.49 | 40.9 | 1.00 | 0.31 | 17.25 |
| September | 7.3 | 36.1 | 10.4 | 1.69 | 12.60 | 4.90 | 32.47 | 8.49 | 45.7 | 1.49 | 0.34 | 28.28 |
| October | 5.8 | 25.8 | 12.5 | 1.69 | 10.55 | 4.90 | 20.07 | 8.19 | 39.1 | 0.85 | 0.42 | 11.54 |
| November | 5.7 | 33.5 | 13.9 | 1.69 | 11.41 | 4.90 | 21.26 | 8.15 | 43.5 | 1.23 | 0.48 | 19.09 |
| December | 4.0 | 25.0 | 14.6 | 1.70 | 7.98 | 4.90 | 2.62 | 7.76 | 38.5 | 0.83 | 0.52 | 9.01 |
| January | 7.3 | 40.9 | 16.9 | 1.68 | 14.10 | 4.90 | 38.70 | 8.64 | 48.0 | 1.70 | 0.65 | 26.74 |
| February | 8.0 | 39.9 | 16.4 | 1.68 | 15.26 | 4.90 | 42.57 | 8.74 | 47.4 | 1.60 | 0.62 | 25.14 |
| March | 8.0 | 40.9 | 16.5 | 1.68 | 14.95 | 4.90 | 40.63 | 8.70 | 46.3 | 1.52 | 0.63 | 22.94 |
| April | 8.0 | 40.0 | 13.8 | 1.67 | 16.05 | 4.90 | 45.01 | 8.80 | 46.9 | 1.56 | 0.47 | 27.13 |

**Table 12.2.** Summary of monthly mean shell length, dry meat weight (DMW), environmental parameters (seston, %POM, temperature), theoretical physiological parameters and scope for growth (SFG) for experimental mussels at each of four sites (I - IV) in North West Bay from June 1995 to April 1996 (averaged by month from weekly estimates). % Ps = percent pseudofaeces, POM = particulate organic matter, T = pseudofaeces threshold and $VO_2$ = respiratory rate.

| Month | Length (mm) | DMW (g) | Seston (mg/l) | % POM | Temp (C) | Clearance rate (l/h) | Seston filtered (mg/h) | T (mg/l) | % Ps | Seston ingested (mg/h) | Absorption efficiency (%) | POM absorbed (mg/h) | $VO_2$ (ml $O_2$/h) | SFG (J/h) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Site I** | | | | | | | | | | | | | | |
| June | 69.9 | 2.2 | 5.6 | 43.0 | 10.5 | 2.35 | 13.08 | 5.10 | 12.04 | 11.08 | 49.7 | 2.36 | 0.63 | 42.54 |
| July | 72.9 | 2.3 | 6.0 | 34.2 | 9.0 | 2.39 | 14.25 | 5.17 | 13.41 | 12.16 | 44.5 | 1.87 | 0.55 | 32.87 |
| August | 76.0 | 2.2 | 6.1 | 30.2 | 9.7 | 2.34 | 14.33 | 5.24 | 13.81 | 12.11 | 41.9 | 1.53 | 0.57 | 24.41 |
| September | 78.3 | 1.9 | 7.7 | 33.9 | 10.4 | 2.19 | 16.87 | 5.29 | 28.86 | 11.89 | 43.7 | 1.85 | 0.55 | 32.20 |
| October | 80.2 | 2.2 | 6.3 | 24.4 | 12.5 | 2.34 | 14.75 | 5.32 | 14.69 | 12.37 | 37.3 | 1.13 | 0.76 | 11.07 |
| November | 81.4 | 2.5 | 5.2 | 34.2 | 13.9 | 2.51 | 12.96 | 5.35 | 10.30 | 11.23 | 44.1 | 1.84 | 1.00 | 23.01 |
| December | 83.0 | 2.9 | 3.9 | 26.1 | 14.6 | 2.67 | 10.29 | 5.38 | 0.00 | 10.29 | 38.1 | 1.02 | 1.19 | -0.15 |
| January | 84.8 | 3.4 | 7.3 | 41.3 | 16.9 | 2.83 | 20.61 | 5.42 | 24.03 | 15.57 | 48.5 | 3.16 | 1.69 | 39.88 |
| February | 85.3 | 3.5 | 8.1 | 38.6 | 16.4 | 2.86 | 23.21 | 5.43 | 30.63 | 16.03 | 47.3 | 2.93 | 1.65 | 35.34 |
| March | 84.8 | 2.9 | 7.7 | 40.7 | 16.5 | 2.64 | 20.27 | 5.42 | 26.83 | 14.64 | 48.3 | 2.91 | 1.44 | 39.26 |
| April | 85.8 | 2.7 | 7.8 | 40.7 | 13.8 | 2.55 | 19.95 | 5.44 | 28.15 | 14.25 | 48.4 | 2.81 | 1.02 | 45.35 |
| **Site II** | | | | | | | | | | | | | | |
| June | 70.4 | 2.1 | 5.4 | 40.8 | 10.5 | 2.33 | 12.70 | 5.11 | 12.23 | 10.87 | 48.2 | 2.10 | 0.62 | 36.79 |
| July | 73.2 | 2.3 | 6.8 | 32.7 | 9.0 | 2.39 | 16.16 | 5.17 | 22.00 | 12.45 | 43.6 | 1.79 | 0.55 | 30.88 |
| August | 74.6 | 1.9 | 5.8 | 27.4 | 9.7 | 2.23 | 13.01 | 5.21 | 11.48 | 11.40 | 39.9 | 1.25 | 0.52 | 18.76 |
| September | 77.6 | 1.6 | 7.0 | 36.5 | 10.4 | 2.07 | 14.56 | 5.27 | 22.46 | 11.02 | 45.5 | 1.92 | 0.50 | 34.99 |
| October | 79.3 | 2.1 | 5.6 | 27.0 | 12.5 | 2.32 | 12.96 | 5.31 | 7.29 | 11.93 | 39.5 | 1.29 | 0.75 | 14.97 |
| November | 80.9 | 2.6 | 5.3 | 33.6 | 13.9 | 2.54 | 13.29 | 5.34 | 11.57 | 11.24 | 43.9 | 1.78 | 1.02 | 21.08 |
| December | 82.9 | 3.0 | 3.8 | 25.3 | 14.6 | 2.71 | 10.41 | 5.38 | 0.00 | 10.41 | 37.2 | 1.02 | 1.22 | -0.96 |
| January | 82.7 | 3.0 | 6.9 | 39.9 | 16.9 | 2.69 | 18.70 | 5.37 | 21.68 | 14.62 | 47.8 | 2.82 | 1.54 | 35.03 |
| February | 84.2 | 3.2 | 7.9 | 38.6 | 16.4 | 2.76 | 21.75 | 5.41 | 29.33 | 15.37 | 47.3 | 2.81 | 1.55 | 34.68 |
| March | 85.6 | 3.0 | 7.6 | 40.4 | 16.5 | 2.68 | 20.44 | 5.43 | 25.33 | 14.87 | 48.1 | 2.93 | 1.48 | 38.83 |
| April | 84.7 | 2.7 | 8.4 | 39.8 | 13.8 | 2.55 | 21.48 | 5.41 | 32.91 | 14.36 | 48.0 | 2.75 | 1.03 | 43.72 |
| **Site III** | | | | | | | | | | | | | | |
| June | 68.8 | 2.1 | 5.3 | 45.3 | 10.5 | 2.31 | 12.27 | 5.07 | 11.00 | 10.62 | 50.3 | 2.36 | 0.61 | 43.13 |
| July | 72.5 | 2.2 | 5.8 | 33.2 | 9.0 | 2.37 | 13.62 | 5.16 | 11.92 | 11.96 | 43.9 | 1.75 | 0.54 | 30.30 |
| August | 75.1 | 1.9 | 6.1 | 27.4 | 9.7 | 2.24 | 13.77 | 5.22 | 15.29 | 11.56 | 39.7 | 1.26 | 0.52 | 19.09 |
| September | 78.4 | 1.7 | 7.1 | 35.2 | 10.4 | 2.10 | 14.99 | 5.29 | 22.84 | 11.24 | 44.8 | 1.84 | 0.51 | 32.93 |
| October | 80.1 | 2.2 | 5.3 | 26.1 | 12.5 | 2.36 | 12.53 | 5.32 | 5.01 | 11.81 | 38.7 | 1.21 | 0.77 | 12.87 |
| November | 81.3 | 2.7 | 5.4 | 33.6 | 13.9 | 2.59 | 14.01 | 5.35 | 11.20 | 11.99 | 43.9 | 1.87 | 1.06 | 22.51 |
| December | 82.0 | 3.0 | 3.9 | 21.8 | 14.6 | 2.69 | 10.57 | 5.36 | 0.00 | 10.57 | 34.7 | 0.79 | 1.21 | -6.09 |
| January | 84.6 | 3.1 | 7.2 | 40.8 | 16.9 | 2.73 | 19.70 | 5.41 | 23.53 | 15.01 | 48.4 | 2.98 | 1.58 | 38.01 |
| February | 85.4 | 2.9 | 7.2 | 42.8 | 16.4 | 2.64 | 18.80 | 5.43 | 21.67 | 14.34 | 49.3 | 3.05 | 1.42 | 42.94 |
| March | 84.7 | 2.4 | 7.8 | 40.5 | 16.5 | 2.44 | 19.04 | 5.42 | 28.07 | 13.58 | 48.1 | 2.68 | 1.25 | 37.70 |
| April | 84.2 | 2.4 | 8.5 | 38.9 | 13.8 | 2.45 | 20.77 | 5.41 | 33.47 | 13.75 | 47.4 | 2.55 | 0.95 | 40.70 |
| **Site IV** | | | | | | | | | | | | | | |
| June | 68.6 | 1.9 | 6.1 | 40.0 | 10.5 | 2.24 | 13.72 | 5.07 | 17.23 | 11.14 | 47.7 | 2.12 | 0.57 | 38.14 |
| July | 72.8 | 2.1 | 5.6 | 33.0 | 9.0 | 2.34 | 13.07 | 5.17 | 9.64 | 11.78 | 43.5 | 1.72 | 0.53 | 29.86 |
| August | 74.7 | 1.8 | 5.7 | 30.2 | 9.7 | 2.18 | 12.45 | 5.21 | 9.76 | 11.09 | 41.9 | 1.41 | 0.49 | 23.02 |
| September | 77.2 | 1.5 | 7.1 | 37.9 | 10.4 | 2.02 | 14.30 | 5.26 | 22.89 | 10.77 | 46.2 | 1.98 | 0.47 | 36.81 |
| October | 79.1 | 2.1 | 5.8 | 25.1 | 12.5 | 2.31 | 13.39 | 5.30 | 10.53 | 11.92 | 37.7 | 1.15 | 0.74 | 11.84 |
| November | 82.0 | 2.7 | 5.6 | 33.1 | 13.9 | 2.56 | 14.23 | 5.36 | 11.39 | 11.98 | 43.5 | 1.84 | 1.03 | 22.29 |
| December | 82.7 | 2.9 | 3.8 | 25.2 | 14.6 | 2.66 | 10.06 | 5.38 | 0.00 | 10.06 | 37.6 | 0.96 | 1.18 | -1.45 |
| January | 83.6 | 3.0 | 6.8 | 42.1 | 16.9 | 2.67 | 18.04 | 5.39 | 19.68 | 14.48 | 49.1 | 3.01 | 1.51 | 39.95 |
| February | 83.9 | 2.7 | 7.8 | 40.8 | 16.4 | 2.57 | 20.13 | 5.40 | 28.99 | 14.25 | 48.5 | 2.82 | 1.35 | 38.96 |
| March | 82.3 | 2.2 | 7.5 | 45.5 | 16.5 | 2.35 | 17.63 | 5.37 | 25.57 | 12.86 | 50.7 | 3.00 | 1.16 | 47.14 |
| April | 82.3 | 2.2 | 8.3 | 43.5 | 13.8 | 2.34 | 19.26 | 5.37 | 30.91 | 12.98 | 49.9 | 2.82 | 0.87 | 48.66 |

Predicted growth, based on monthly SFG values (kJ·month$^{-1}$), was largely not reflected by observed mussel energy increments (Fig. 12.4) or by shell length growth. No significant correlations between monthly SFG and mussel energy increments ($r = -0.189$, $P = 0.24$; $n = 44$ observations) nor shell length increments ($r = -0.138$, $P = 0.48$; $n = 44$ observations) were indicated. However, after eliminating data from the last three months (February - April 1996), a weak, but significant, relationship between SFG and shell length growth was revealed ($r = 0.346$, $P = 0.05$). Except for October to December 1995, SFG was significantly greater than observed energy increments ($P < 0.05$ in November, $P < 0.001$ for all other months).

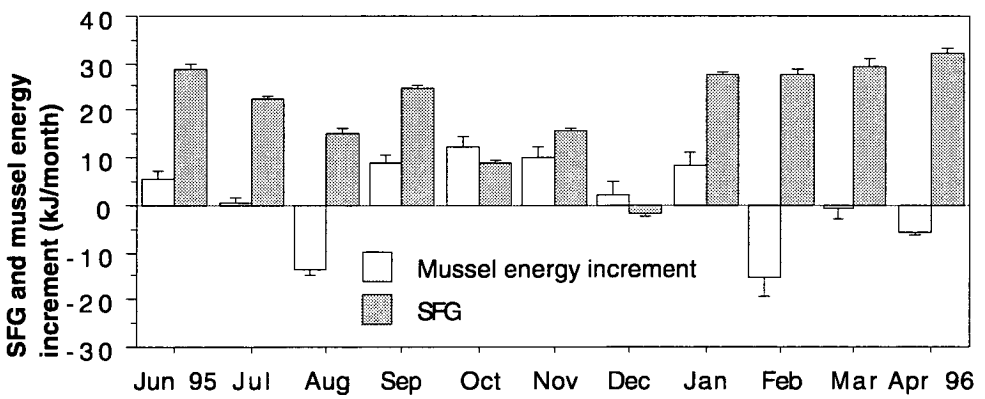


**Figure 12.4.** Mean monthly experimental mussel SFG and mussel energy increments (mean ± SE kJ·month$^{-1}$, n = 4 estimates per month, each estimate being the mean monthly SFG or energy increment from each of the four experimental sites). SFG was significantly greater than mussel energy increments for all months ($P < 0.05$) except October, November and December 1995 ($P > 0.05$).

SFG during the three-month period of October to December 1995, where SFG and actual growth coincided, was analysed separately. No significant differences in SFG among the four test sites were indicated (ANOVA, $P > 0.05$).

Theoretical SFG for the May 1995 overspat cohort was fairly stable at approximately 5 J·h$^{-1}$ from June 1995 through December 1995, except for two peaks in September and November when SFG achieved levels of 18 J·h$^{-1}$ (Fig. 12.5). SFG increased in January 1996, fluctuating between 17 and 40 J·h$^{-1}$. Again, although significant temporal variation was indicated ($P < 0.0001$), no differences among sites, nor a month x site interaction, were significant ($P > 0.05$).

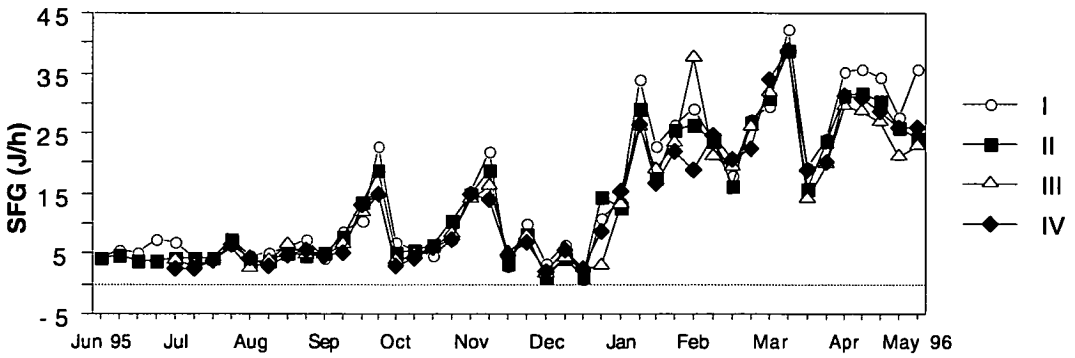


**Figure 12.5.** Weekly theoretical scope for growth (SFG, $J \cdot h^{-1}$) of overspat at each longline site (I, II, III, IV) from June 1995 to May 1996.

As with experimental mussels, mean monthly spat energy increments were significantly less than those predicted by SFG calculations except for December 1995 ($\alpha = 0.05$, Fig. 12.6). However, a weak correlation between spat energy increments and SFG was significant ($P < 0.05$, $r = 0.33$, $n = 44$ observations).

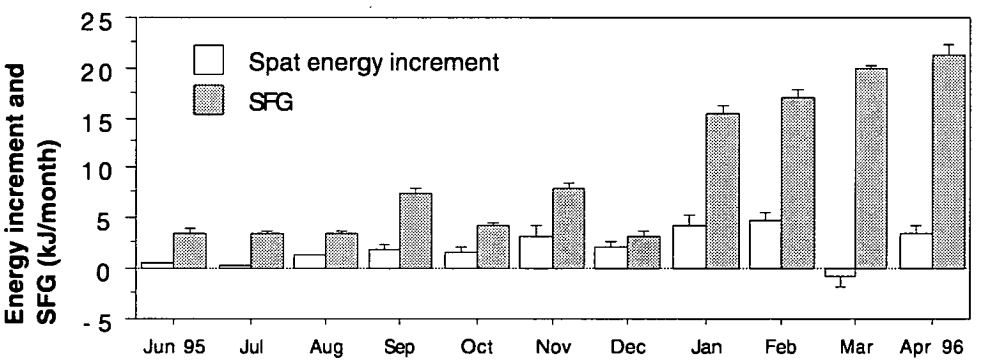


**Figure 12.6.** Mean monthly spat scope for growth (SFG) and spat energy increments (mean ± SE $kJ \cdot month^{-1}$, $n = 4$ estimates per month, each estimate being the mean monthly SFG or energy increment from each of the four experimental sites). Spat energy increments were significantly less than SFG for all months except December 1995 ($\alpha = 0.05$).

## 12.4 Discussion

### 12.4.1 Temporal variation.

Temporal variation in SFG was significant in this study, whereby SFG generally followed variations in suspended POM content. The analysis of "seasonal variation" requires a somewhat arbitrary time scale classification defined by calender months, and therefore pooling SFG results according to "season" is somewhat ambiguous. However, standard mussel SFG showed little difference between pooled "winter" (June to August 1995, mean SFG = 23.2 ± 1.2 $J \cdot h^{-1}$, n = 48) and "summer-autumn " (January to April 1996, mean SFG = 27.9 ± 0.9 $J \cdot h^{-1}$, n = 64) values. SFG during the "spring-early summer" (September to December 1995, mean SFG = 14.5 ± 1.5 $J \cdot h^{-1}$, n = 64) period was significantly less than both the winter and summer-autumn periods ($P < 0.0001$), despite the two peaks in POM concentrations in September and November 1995. Although being somewhat lower in the spring-early summer period, theoretical SFG estimates suggest that North West Bay is a suitable location for mussel culture, with a sufficient food supply to sustain growth throughout most of the year.

These results contrast with studies reporting low, and sometimes negative, SFG in autumn-winter and higher SFG in spring-summer for mussels (Bayne and Widdows, 1978; Thompson, 1984a; Rodhouse et al., 1985; Deslous-Paoli et al., 1990; Page and Ricard 1990; Okumus and Stirling, 1994), scallops (MacDonald and Thompson, 1986) and cockles (Smaal et al., 1997). Seasonal variation in these studies is primarily due to seasonal differences in food availability, whereby winter POM and phytoplankton levels were generally lower than in spring-summer.

As a historical record of total particulate and POM levels in NWB is not available, it is unknown if the low particulate levels in November-December 1995 were characteristic of this period. No discernible seasonal trends in particulate concentrations are reported from the nearby Derwent estuary (DELM, 1995), nor from two other bays in southern Tasmania (Little Swanport Lagoon and Pittwater; I. Mitchell, TAFI, unpublished data). However, significantly higher TPM and POM levels were found in December 1995 at Pipeclay Lagoon compared with other months (I. Mitchell, unpublished data).

It appears that seasonal variation in SFG is largely site-specific, determined by seasonal variations in food availability, but moderated by the maximum ingestion rate and respiratory losses of mussels. In this study, although POM content was higher in the summer months than during winter, theoretical summer SFG was similar to winter SFG. Summer SFG was constrained by the inability of mussels to ingest POM above the pseudofaeces threshold and by higher respiratory rates during the warmer summer

months. On the basis of theoretical SFG calculations, it appears that NWB is a suitable site for bivalve culture, maintaining a positive SFG throughout the majority of the year. Although monthly SFG estimates displayed little resemblance to the actual growth of experimental mussels, an adequate supply of food did support shell length growth over most of the trial. Growth in biomass was fragmented by the two spawning periods in August 1995 and February 1996. Negligible growth of either shell or biomass, along with increasing mortalities, was observed during the post-spawning period of March-April 1996 when SFG was at maximum levels. However, overspat growth was observed during the same period. Perhaps the growth of the underlying experimental mussels was compromised by a combination of factors such as senility, post-reproductive stress, and competition for food from overspat and other biofouling organisms.

### 12.4.2 Spatial variation

The analyses of all SFG estimates (1.0 g standard, experimental mussels and overspat) for the period June 1995 to May 1996 revealed no significant differences among the four mussel longline sites. This might be expected, as environmental parameters (ie. temperature, seston, %POM), upon which SFG calculations are dependent, were all similar among sites. The only factor relevant to SFG that might have differed among sites is the energy content of food particles, which was not measured. However, as non-algal POM concentrations were similar among sites, any increase in non-algal POM levels from fish feed and faeces at sites I and II was not detectable. Therefore, as both algal (ie. chlorophyll) and non-algal concentrations were similar among sites, it is unlikely that gross sestonic POM energy contents would have significantly differed among sites.

Incorporating physiological studies and SFG estimates with growth studies, allows the exploration of differences in production potential by individuals of the same species at different sites. Where growth differences are significant between sites, physiological studies may suggest a number of factors likely causing the observed differences. Differential growth among mussel populations have been attributed to various environmental factors affecting physiological responses such as clearance, respiration and excretion rates, and absorption efficiencies (Bayne and Worrall, 1980; Widdows et al., 1984). In the present study, this exercise helps to explain the similar growth observed among the four longline sites. As environmental parameters were similar among longline sites, the difference between assimilated food energy and respiratory energy losses (the energy balance) was also similar among sites. As both SFG and observed growth were similar among sites, it seems reasonable that any negative

effects of differential stocking densities, pea crab infestations, overspat settlements or other biofoulants on mussel growth were either minimal or, if significant, were similarly manifested among sites.

It is noteworthy that the standard mussel SFG estimates at the centre of the fish farm (site X) were never significantly different from any of the four experimental longline sites, despite site X displaying significantly higher particulate levels. This assumes that the energy content of available food particles within the farm, comprising mostly natural POM along with some fish feed fines and faeces, was not significantly higher than the energy content of natural POM. Total POM energy content at site X would have to be above 26.5 J·mg $POM^{-1}$ in order for SFG to exceed other sites. It is unlikely that this level would be attained; the estimated gross energy contents of fish feed and fish faeces are 26.45 and 25.75 J·mg dry matter, respectively, and only comprise a small fraction of the total suspended matter available to mussels. However, as POM energy was not actually measured, similar POM energy content among all sites remains speculative. It is probable that SFG at site X did not exceed other sites because %POM was not higher and ambient particulate levels consistently exceeded the pseudofaeces threshold. Consequently, calculated POM ingestion and absorption rates were similar among sites ($P > 0.05$) and at, or near, maximum levels for most of the trial. Therefore, mussels growing within the fishcage system would not be physiologically capable of taking full advantage of any extra particulate loadings offered in the form of fish farm wastes. Fish farm conditions that might favour enhanced mussel growth over other sites within NWB might include: (a) significantly higher %POM levels, (b) significantly higher POM concentrations together with ambient particulate levels consistently below the pseudofaeces threshold concentration, or (c) the total energy content of suspended particulates, comprised of natural seston and particulate fish wastes, is consistently higher than ambient POM at other sites.

Theoretical SFG values suggest that North West Bay has excellent potential for mussel culture. The annual mean mussel SFG of $21.8 \pm 0.8$ J·$h^{-1}$·g $DMW^{-1}$ (n = 176 estimates from sites I - IV, excluding site X) exceeds reported SFG at many culture sites in temperate waters of the Northern Hemisphere (Table 12.3). The lower SFG values quoted in some of these studies are a consequence of extended winter conditions with low temperatures and POM levels. However, as we have previously discussed, due to the lack of physiological and energetic measurements, the SFG values in this study are only theoretical and were rarely reflected by actual growth. Therefore, comparisons with other studies, where SFG has been calculated from direct physiological and particulate energy measurements, require caution.

**Table 12.3.** Summary of reported scope for growth (SFG) of mussels (1 g DMW) at various locations. *Mytilus edulis* except for *M. galloprovincialis*[a] and *M. planulatus*[b].

| SFG | | | | |
|---|---|---|---|---|
| Min | Max | Mean | Location | Reference |
| 0 | 47 | 21.8 | Tasmania [b] | This study |
| -4 | 28 | 8.5 | Lynher, U.K. | Bayne and Widdows (1978) |
| -11 | 40 | 8.5 | Cattewater, U.K. | Bayne and Widdows (1978) |
| -2 | 6 | - | Lynher, U.K. | Bayne et al. (1979) |
| -2 | 10 | 1.3 | Swale, U.K. | Bayne et al. (1979) |
| -4 | 7 | 0.1 | Kings Dock, U.K. | Bayne et al. (1979) |
| 2 | 21 | 10.5 | Newfoundland, Canada | Thompson (1984a) |
| 5 | 25 | 15.0 | Tamar, U.K. | Widdows et al. (1984) |
| 0 | 25 | 6.3 | Swansea, U.K. | Widdows et al. (1984) |
| -2 | 15 | - | Spain [a] | Navarro et al. (1991) |
| 0 | 35 | 20.0 | Netherlands | Smaal et al. (1997) |

### 12.4.3 Theoretical SFG versus observed growth

Although SFG estimates for NWB are higher than those reported in other studies, monthly experimental mussel SFG (predicted growth) was only matched by observed growth from October to December 1995. Theoretical monthly SFG ranged up to a maximum of 32 kJ·month$^{-1}$ in April 1996. However, the highest actual energy increment was only 12.3 kJ·month$^{-1}$ in October 1995. Several possibilities for the discrepancy between SFG and observed growth may be suggested, all of which may have varying degrees of influence on the calculated values of SFG.

Firstly, was observed growth less than predicted growth as a consequence of biofoulants (overspat, tunicates and algae) reducing the food available to the underlying mussels? Perhaps not - observed growth fell far short of predicted growth even during June and July 1995 (Fig. 12.4) prior to overspat and other biofoulants becoming heavily established. Therefore, it seems unlikely that food levels were significantly reduced by fouling organisms, at least during the first few months. It is more probable that the discrepancy between observed and predicted growth was the failure to directly measure the temporal variation in mussel physiological response parameters and the energy content of the available food supply (seston, fish feed, faeces). Estimates of physiological parameters, based on mathematical relationships and values derived from the literature, may not have been representative of the true physiological status of the experimental mussels. Feeding and assimilation rates may not have agreed with literature values due to genetic differences (*Mytilus planulatus* vs *Mytilus edulis*), reproductive condition, age, and field conditions that are usually not replicated in lab studies. Secondly, the gross energy content of food particles may

have varied over time; therefore not always agreeing with the constant POM energy value (23.5 J·mg $POM^{-1}$) incorporated into SFG calculations. A more detailed exploration of the sources of variation that may be responsible for variation in SFG estimates is presented in Appendix 6.

As there is disagreement between theoretical SFG and actual mussel growth, has this exercise been worthwhile in answering the questions posed at the outset? Unfortunately, without complete confidence in the accuracy of the physiological response parameters or POM energy content incorporated into SFG calculations, no unequivocal conclusions can be drawn. However, the analysis of SFG does provide some support to the relative growth of mussels observed in this trial.

Similar SFG estimates among the longline sites do reflect the observed similarities in mussel growth. Similar SFG and actual growth among sites would be expected - environmental conditions were similar among sites. Despite the physiological response parameters used in SFG calculation being in question, it is not unrealistic to assume that the physiological responses of mussels to similar environmental conditions among sites would also be similar. Secondly, although the energy content of suspended food material is uncertain, similar algal and non-algal POM concentrations suggest any significant difference among sites is unlikely. Therefore, the prevailing environmental conditions within North West Bay were not so dissimilar as to significantly differentiate feeding, digestion, absorption or respiratory processes of mussels at the four longline sites. Consequently, growth was similar.

Similar SFG among sites supporting the similarities in experimental mussel growth suggests: variable sock densities; biofouling densities; or pea crab infestation; either did not significantly impair mussel growth, or did impair growth, but any impairment was equally manifested among sites (ie. negative effects were independent of sock density, overspat density, biofouling density or percentage of pea crab infestation). Therefore, SFG estimates do support the experimental results, suggesting that the Aquatas fish farm has little potential for enhancing the growth of cultured mussels. However, Riisgard and Randlov (1981) suggest: "only when the relationship between estimated and actual growth has been convincingly described does it seem proper to use 'scope for growth' as a physiological integration in quantifying the physiological condition and the suitability of the environment". With this recommendation in mind, SFG during the three-month period of October to December 1995, where SFG and actual growth were in agreement, was analysed separately. No significant differences in SFG among the four test sites were indicated, supporting the conclusion that mussel growth is unlikely to be enhanced at the two test sites within the Aquatas fish farm.

It also appears unlikely that culturing mussels immediately next to the fish cages would improve growth. Although observed POM levels were higher, food quality (%POM and energy content) was not significantly higher than at other sites. The ambient particulate levels, consistently above the pseudofaeces threshold, restrict mussels from ingesting a significant portion of additional POM released from the fish cages. Therefore, growth next to the fish cages would be expected to be similar to the other experimental sites. For mussels to attain higher growth rates within the fish farm, relative to other sites in NWB, requires:

- significantly higher %POM levels on the fish farm,
- significantly higher POM concentrations on the fish farm together with ambient particulate levels consistently below the pseudofaeces threshold concentration, or
- total digestible energy content of suspended POM on the farm, comprised of natural seston and particulate fish wastes, to be consistently higher than ambient POM at other sites.

Sustained growth during the winter months observed in this study was due to an adequate supply of food and supported by high SFG estimates. Continued growth through the spring and early summer was also supported by positive SFG. However, the lack of growth during the final stages of the trial is not supported by SFG projections. SFG was at maximum levels during this period. As overspat continued to grow during this period, experimental mussel growth may have been constrained by several factors including old age, post-reproductive stress, and biofouling organisms (including overspat) reducing available food levels.

In conclusion, accurate modelling of bivalve growth and confident assessments of the potential for growth at various locations requires reasonable measures of available food and the physiological responses of mussels to site-specific environmental conditions. Relying on other studies, conducted under different environmental conditions and using different species of mussels, to provide these values may not be appropriate. Unless a consistent relationship between observed and predicted growth from literature values is established, frequent measurements of physiological parameters (clearance rates, absorption efficiencies, respiration rates, excretion rates) and ambient particulate energy levels are required to provide realistic estimates of SFG. Although difficult to achieve, physiological measurements should preferably be conducted in the field or under simulated field conditions that account for as many environmental variables as possible. The various physiological relationships required for the assessment of SFG have not been established for *Mytilus planulatus*; therefore, this is an area that requires further investigation.

# 13. Fish Farm Waste Loadings

## 13.1 Introduction

The purpose of this exercise was to quantify the loadings of major waste products from the Aquatas fish farm into North West Bay during the period of the mussel growth trial. Fish farm wastes of most interest to this study were particulate wastes, such as faeces and fish feed particles, and dissolved nutrients that may promote phytoplankton production. These wastes are of interest not only from a mussel's point of view; they have been implicated in having deleterious environmental impacts, and therefore are of interest to both the fish farmer as well as to the broader community (Cowey and Cho, 1991; Pillay, 1992; Talbot and Hole, 1994). Waste loadings were estimated largely from relationships derived from the literature and digestibility coefficients supplied by the feed manufacturer; therefore, they are only theoretical waste loadings. However, this exercise provides some insight into the possible waste discharges from the Aquatas fish farm into the local environment of North West Bay and their potential influence on the growth of mussels cultured within the farm site. The potential of theoretical waste discharges to increase ambient particulate and nutrient levels in North West Bay will be compared with the actual conditions monitored during the course of the mussel growth study.

## 13.2 Particulate Wastes

### 13.2.1 Methodology

***Fish Biomass and Feeding Practices***

Quantification of particulate and nutrient waste loadings is dependent on the level of fish production as well as husbandry practices. The mussel growth study spanned approximately one and a half fish-growing seasons at the Aquatas smolt-rearing fish farm (Howden, North West Bay): the last three months of the 1994 year-class (March 1995 to May 1995) and the entire 1995 year-class (September 1995 to May 1996).

Smolts from the 1994-year class were introduced from August 1994 to October 1994 into twenty-three 60 m circumference fishcages (Polarcirkel™). At the start of the mussel growth trial (March 1995), 243,429 fish remained on-site with a total biomass of 247.8 tonnes. Beginning at the end of April 1995, these fish were progressively transferred to a grow-out site in the D'Entrecasteaux Channel. The Howden farm site was left fallow for approximately 3.5 months from June to mid-September 1995. In

September-October 1995, approximately 221,000 new smolts with an initial biomass of 22.5 tonnes were introduced into seventeen cages. All fish remained on-site until the end of April 1996, except for 17,000 fish that were removed during April 1996 (Fig. 13.1).

Fish were fed *ad libetum* by hand (new smolts) or feed blowers. Feed was distributed to cages on a rotating basis, with each cage fed 4 to 6 times daily. Daily feed rations ranged from 1% to 2% of biomass with approximately 60% of the daily ration provided during the morning. Feed composition was approximately 45:22:16:9:8% (protein:fat:carbohydrate:ash:moisture; Pivot Aquaculture, Salmon Grower Diet). The quantity of feed delivered ranged up to 123 tonnes per month, depending on fish biomass (Fig. 13.1). Mean monthly FCR was approximately 1.3.

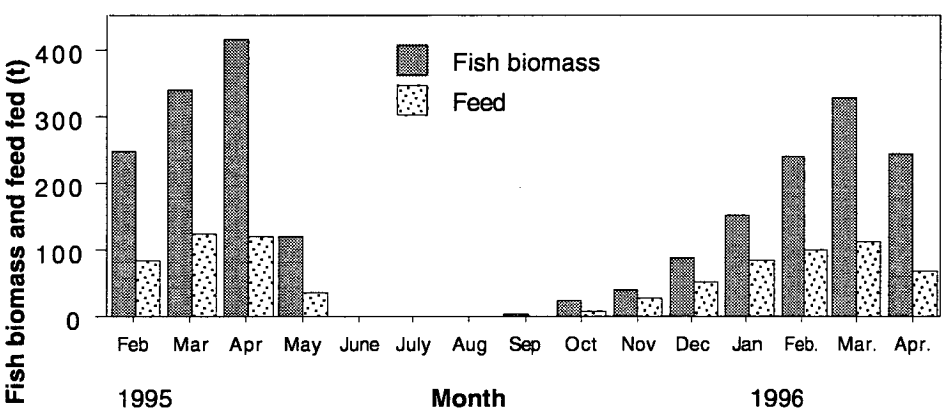


**Figure 13.1.** Inventory of fish biomass (tonnes) at the end of each month and quantity of feed fed (tonnes) each month at the Aquatas fish farm from February 1995 to April 1996.

### *Calculation of Particulate Waste Discharge*

Calculations of solid waste loadings (dry weight) were based on the following assumptions:

| | |
|---|---|
| Feed moisture content | 8% |
| Uneaten feed pellets | 4% of feed delivered |
| Feed fines (< 1 mm) | 1% of feed delivered |
| Fish faeces | 17% of feed (dry weight) consumed |

Feed moisture content and percentage of fines were provided by the feed manufacturer (Pivot Aquaculture, Hobart, Tasmania). Faecal waste production, as a percentage of feed consumed, is related to diet digestibility. An estimate of whole diet digestibility of 83% was derived from the dry component digestibility coefficients also provided by

the feed manufacturer. Therefore, faecal waste of 17% of feed consumed (dry weight) is less than other estimates exceeding 25% (eg. Beveridge et al., 1991 in Bergheim and Asgard, 1996). Estimates of uneaten feed range from 1-2% to 40% of delivered feed (Gowen and Bradbury, 1987; Troell and Norberg, 1998). Without on-site investigation, the amount of uneaten feed wastage is difficult to quantify (no data available from Aquatas); therefore an estimate of 4% feed wastage was applied (Cho et al., 1991; Bergheim and Asgard, 1996). As feed wastage is largely determined by husbandry and feeding practices, which can vary between farms and even among staff, it is recognised that 4% feed loss may be a substantial underestimate.

The contribution of suspended particulate fish farm wastes (feed fines + faeces) to ambient suspended particulate matter concentrations (SPM) within the farm was estimated for the period June 1995 to May 1996. In order to determine the expected whole water column concentrations of suspended fish farm wastes during the times when water sampling was carried out (approximately 0900 and 1200 h), the following assumptions were made:

1. 60% of daily feed, including feed fines, was delivered between 0800 and 1200 h. In practice this feed is delivered in pulses, but for this calculation continuous feeding is assumed.
2. 22% of daily faecal output occurred between 0800 and 1200h (Taylor et al., 1992).
3. Faeces were voided as suspended particulates rather than intact faecal pellets.
4. Feed fines and faecal wastes were uniformly distributed among and within all cages.
5. Feed fines and faeces had a settling speed of 4 $cm \cdot sec^{-1}$ (Gowen et al., 1988), requiring 8.3 minutes to reach the sea bottom (20 m). Therefore, the quantity of waste SPM in the water column during sampling is the dry weight of feed fines and faeces released in 8.3 minutes.
6. No current. This maximises the expected fish waste SPM concentrations at slack tides. Any water movement will variously dilute the fish waste SPM concentration according to current speed.
7. No resuspension of bottom sediments.
8. Only feed fines and faeces contributed to SPM concentrations; waste feed pellets were considered to precipitate to the bottom without breaking up and did not contribute to SPM or POM concentrations.
9. Theoretical waste particulate concentrations per cage were calculated by the mass of waste particles (feed fines and faeces) released or delivered in 8.3 minutes into a cylindrical water column bounded by the cage plus the volume directly below the cage to the sea bottom (volume = 6,280 $m^3$). This extra volume was included to allow comparisons with measured particulate concentrations (integrated samples of the total water column).

Monthly farm feed input (data supplied by Aquatas) was back-calculated to estimate the mean quantity of feed supplied daily and feed supplied hourly from 0800 to 1200 h (corresponding to the times when water samples were collected). The quantity of particulate waste material (feed fines + faeces) discharged in 8.3 minutes was estimated to calculate the expected concentration of waste particulates within each fishcage when sampling occurred. As calculations were based on monthly feed

delivery (daily inventory was not available), the waste concentrations are the mean expected concentrations during water sampling for any day within each month. The SPM and POM concentrations at mussel longline sites I through IV were not significantly different over the trial period ($P > 0.05$, Section 4.3.7). Therefore, the monthly mean concentrations at these four sites were taken as being representative of the ambient particulate concentrations within NWB. The monthly mean waste particle concentrations (theoretical) were added to the monthly mean ambient particulate concentrations (observed) to give monthly total particulate concentrations that might be expected on the farm site. These theoretical concentrations were compared with actual particulate concentrations from water samples collected at the centre of the fish farm (site X) during the trial.

### *Distribution of particulate wastes*

The distribution of solid waste particles was modelled to determine if these suspended particles could have actually reached the mussels suspended within the farm site (longlines I and II) and to estimate the spread of organic fish farm wastes onto the sea bottom. The calculation of horizontal particle displacement (D) required inputs of current speed (s), bottom depth (d) and particle settling velocities (v), where $D\ (m) = d\ (m) \cdot s\ (m \cdot sec^{-1}) / v\ (m \cdot sec^{-1})$ (Gowen and Bradbury, 1987). Current speeds recorded at 10 minute intervals from September to November 1995 were applied to the above equation and bottom depth was set at 20 m. Settling velocities of 0.13 $m \cdot sec^{-1}$ and 0.04 $m \cdot sec^{-1}$ for feed pellets and faeces, respectively, were applied (Gowen et al., 1988; Coyne et al., 1994). The resulting distributions of faeces and feed pellets assumed continuous feeding and faecal discharge throughout the period of current meter deployment (September - November 1995); therefore they are only indicators of the potential displacement of solid fish farm wastes onto the surrounding seabed during a complete fish-growing season (typically September through May). As current speed and direction were monitored over at least one lunar cycle, it is unlikely that modelled solid waste distribution would significantly differ from a model incorporating data collected over a longer period.

## 13.2.2 Results

### *Particulate waste loadings*

Depending on fish biomass and quantity of feed delivered, daily total solid waste discharge ranged up to 783 kg dry weight·day$^{-1}$ and organic loading (as protein, lipid and carbohydrate) ranged up to 571 kg dry weight·day$^{-1}$ (Table 13.1). Eighty-seven tonnes of solid waste, comprised of 63 tonnes of organic matter, was potentially released from the farm in the 1995-96 season. Eighty-one percent of the solid waste loadings were in the form of feed fines (5%) and faeces (76%), with the remaining 19% made up of uneaten feed pellets. Mean monthly solid waste discharge was 263 kg·tonne fish production$^{-1}$.

### *Particulate waste concentrations*

Waste particulate concentrations that might have been expected during water sampling varied with the quantity of feed delivered, ranging up to maxima of approximately 0.03 mg POM·l$^{-1}$ and 0.05 mg total SPM·l$^{-1}$ (Table 13.2). These theoretical particulate waste concentrations are extremely low, are below detectable levels, and would elevate ambient POM and SPM concentrations by only 1.7% and 0.6% (maxima), respectively. The low theoretical concentrations sharply contrast with observed fish farm particulate levels that were consistently higher than ambient concentrations. Overall mean site X concentrations were 0.30 mg POM·l$^{-1}$ and 1.04 mg total SPM·l$^{-1}$ higher than overall mean ambient concentrations (Fig. 13.2). Monitored fish farm concentrations were up to 32% and 26% higher than ambient POM and SPM concentrations (mean 15% higher and POM 17% higher SPM).

**Table 13.1.** Estimates of mean daily feed input, total solid and organic waste loadings (kg dry weight·day$^{-1}$) at Aquatas fish farm from February 1995 to April 1996. Assumptions: uneaten pellets = 4%, feed fines = 1% and solid faecal production (dry weight) = 17% of feed consumed, POM faecal discharge = 13% of feed organic content consumed. No fish were on-site in June, July and August 1995.

| | Feb-95 | Mar-95 | Apr-95 | May-95 | Sep-95 | Oct-95 | Nov-95 | Dec-95 | Jan-96 | Feb-96 | Mar-96 | Apr-96 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Monthly Fish Production (tonnes) | 73 | 94 | 83 | 27 | | 4 | 16 | 48 | 67 | 86 | 91 | 51 |
| **Total solid wastes (daily)** | | | | | | | | | | | | |
| Feed delivered dry wt (kg/day) | 2810 | 3650 | 3703 | 1059 | 108 | 235 | 866 | 1535 | 2475 | 3250 | 3296 | 2027 |
| Feed consumed (kg/day) | 2670 | 3468 | 3518 | 1006 | 103 | 223 | 823 | 1458 | 2351 | 3087 | 3131 | 1926 |
| Losses (kg/day): | | | | | | | | | | | | |
| Waste feed pellets (kg/day) | 112 | 146 | 148 | 42 | 4 | 9 | 35 | 61 | 99 | 130 | 132 | 81 |
| Fines (kg/day) | 28 | 37 | 37 | 11 | 1 | 2 | 9 | 15 | 25 | 32 | 33 | 20 |
| Faeces (kg/day) | 454 | 590 | 598 | 171 | 17 | 38 | 140 | 248 | 400 | 525 | 532 | 327 |
| Total suspended particulates (kg/day) | 482 | 626 | 635 | 182 | 19 | 40 | 149 | 263 | 424 | 557 | 565 | 348 |
| Total solid wastes (kg/day) | 594 | 772 | 783 | 224 | 23 | 50 | 183 | 325 | 523 | 687 | 697 | 429 |
| Monthly solid waste (kg/tonne fish production) | 246 | 247 | 283 | 245 | | 358 | 353 | 202 | 235 | 240 | 229 | 253 |
| **Waste organic matter (daily)** | | | | | | | | | | | | |
| Feed delivered organic dry wt (kg/day) | 2524 | 3278 | 3325 | 951 | 97 | 211 | 778 | 1378 | 2222 | 2918 | 2960 | 1820 |
| Feed consumed (kg/day) | 2398 | 3114 | 3159 | 904 | 92 | 201 | 739 | 1309 | 2111 | 2772 | 2812 | 1729 |
| Losses (kg/day): | | | | | | | | | | | | |
| Waste feed pellets (kg/day) | 101 | 131 | 133 | 38 | 4 | 8 | 31 | 55 | 89 | 117 | 118 | 73 |
| Fines (kg/day) | 25 | 33 | 33 | 10 | 1 | 2 | 8 | 14 | 22 | 29 | 30 | 18 |
| Faeces (kg/day) | 307 | 399 | 404 | 116 | 12 | 26 | 95 | 168 | 270 | 355 | 360 | 221 |
| Total suspended POM (kg/day) | 332 | 431 | 438 | 125 | 13 | 28 | 102 | 181 | 292 | 384 | 389 | 240 |
| Total organic wastes (kg/day) | 433 | 562 | 571 | 163 | 17 | 36 | 133 | 236 | 381 | 501 | 508 | 312 |
| Monthly organic waste (kg/tonne fish production) | 179 | 180 | 206 | 178 | | 261 | 257 | 147 | 171 | 175 | 167 | 184 |

**Table 13.2.** Theoretical concentrations of total suspended particulate material (SPM) and particulate organic material (POM) of farm wastes (faeces + feed fines) that might be expected within the Aquatas fish farm compared with measured ambient concentrations of North West Bay (monthly means from sites I - IV) and measured fish farm concentrations (monthly means from site X) from water samples collected over the same period (mg dry weight $SPM \cdot l^{-1}$ or $POM \cdot l^{-1}$). Water sampling did not commence until June 1995. No fish were on the farm in June, July and August 1995.

| | Feb-95 | Mar-95 | Apr-95 | May-95 | Jun-95 | Jul-95 | Aug-95 | Sep-95 | Oct-95 | Nov-95 | Dec-95 | Jan-96 | Feb-96 | Mar-96 | Apr-96 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **SPM** | | | | | | | | | | | | | | | |
| Ambient total SPM (mg/l) | | | | | 5.6010 | 6.0180 | 5.9590 | 7.2140 | 5.7520 | 5.3720 | 3.8550 | 7.0510 | 7.7560 | 7.6570 | 7.8470 |
| Theoretical farm total SPM wastes (mg/l) | 0.0292 | 0.0379 | 0.0385 | 0.0169 | 0.0000 | 0.0000 | 0.0000 | 0.0065 | 0.0033 | 0.0122 | 0.0216 | 0.0348 | 0.0457 | 0.0463 | 0.0323 |
| Expected total SPM (ambient + theoretical farm waste SPM, mg/l) | | | | | 5.6010 | 6.0180 | 5.9590 | 7.2205 | 5.7553 | 5.3842 | 3.8766 | 7.0858 | 7.8017 | 7.7033 | 7.8793 |
| Expected increase over ambient (%) | | | | | **0.00** | **0.00** | **0.00** | **0.09** | **0.06** | **0.23** | **0.56** | **0.49** | **0.59** | **0.61** | **0.41** |
| Observed farm total SPM (mg/l) | | | | | 6.0 | 7.4 | 7.5 | 7.5 | 6.2 | 6.8 | 4.7 | 8.4 | 9.1 | 8.9 | 9.2 |
| Observed farm increase over ambient (%) | | | | | **6.45** | **23.43** | **25.81** | **3.63** | **8.41** | **26.14** | **21.71** | **19.03** | **17.48** | **16.59** | **17.11** |
| **POM** | | | | | | | | | | | | | | | |
| Ambient POM (mg/l) | | | | | 2.2950 | 2.0110 | 1.7020 | 2.6540 | 1.4680 | 1.9140 | 0.9290 | 2.8790 | 3.0670 | 3.2490 | 3.1590 |
| Theoretical farm POM wastes (mg/l) | 0.0208 | 0.0270 | 0.0273 | 0.0120 | 0.0000 | 0.0000 | 0.0000 | 0.0046 | 0.0023 | 0.0087 | 0.0153 | 0.0247 | 0.0325 | 0.0329 | 0.0230 |
| Expected POM (ambient + farm waste POM, mg/l) | | | | | 2.2950 | 2.0110 | 1.7020 | 2.6586 | 1.4703 | 1.9227 | 0.9443 | 2.9037 | 3.0995 | 3.2819 | 3.1820 |
| Expected increase over ambient (%) | | | | | **0.00** | **0.00** | **0.00** | **0.17** | **0.16** | **0.45** | **1.65** | **0.86** | **1.06** | **1.01** | **0.73** |
| Observed farm POM (mg/l) | | | | | 2.4 | 2.5 | 2.1 | 2.7 | 1.6 | 2.4 | 1.2 | 3.4 | 3.5 | 3.3 | 3.4 |
| Observed farm increase over ambient (%) | | | | | **6.19** | **22.33** | **26.09** | **3.28** | **11.85** | **22.99** | **31.86** | **16.71** | **15.00** | **3.05** | **8.99** |

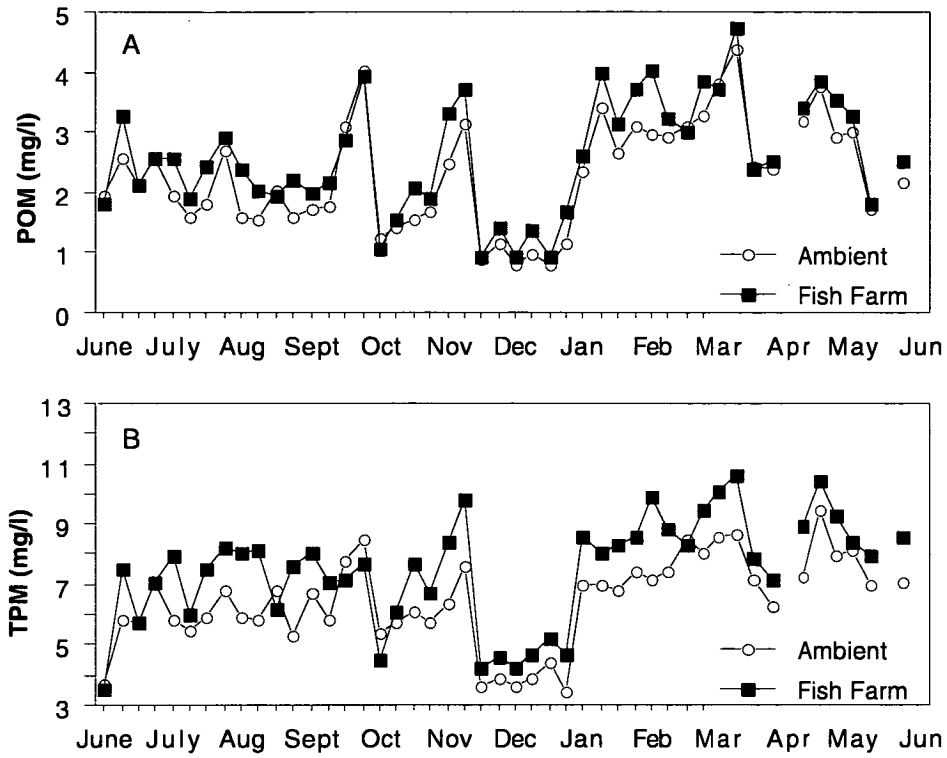


**Figure 13.2.** (A) Particulate organic matter (POM) and (B) total suspended particulate matter (TPM) concentrations ($mg \cdot l^{-1}$) at the centre of the fish farm (mean at site X, n = 2 samples) and ambient concentrations (mean of sites I - IV, n = 8 samples). For clarity, standard error bars are not included.

### *Distribution of solid wastes*

Assuming laminar water flow, no variation in current speed with depth, a mean settling speed of 13 $cm \cdot sec^{-1}$ and a bottom depth of 20 m, it requires 153 seconds for uneaten feed pellets to reach the bottom. Based on measured current speed and directional data, 60% of uneaten feed pellets would accumulate on the sea bottom within 5 m and 99% within 23 m of the fish cages. Under similar circumstances of water flow and assuming a setting velocity of 4 $cm \cdot sec^{-1}$, faecal material and fine feed particles require 500 seconds to reach the bottom. Fourty percent of this material would be deposited on the bottom within 10m, 70% within 30 m, and 90% within 50 m of the fish cages. The greatest horizontal displacement of faeces would be 123 m at the maximum observed current speed of 25 $cm \cdot sec^{-1}$, which occurred only rarely. The displacement of faecal material might be less; faeces would not be released from the surface, but at variable distances between the top and bottom (10 m) of the cages. The bulk of waste material would be deposited in a NW-SE direction (330° - 150°) from the cages, corresponding with the modal current directions (Fig. 13.3). The surface area of the sedimented material would be in the order of 38,000 $m^2$ (Fig. 13.4), although a wider area of dispersal is possible due to turbulent diffusion (Silvert, 1992) and/or lower faecal settlement rates (eg. 2.0 $cm \cdot sec^{-1}$; Findlay and Watling, 1994).

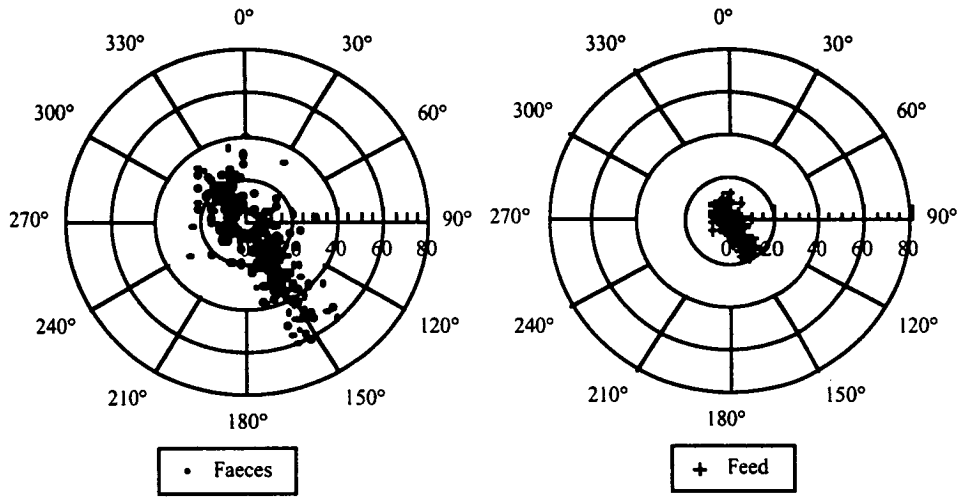


**Figure 13.3.** Distribution (direction in degrees° and distance in m) of settled faecal and fine feed particles (left) and uneaten fish pellets (right) originating from a single cage at the Aquatas fish farm. Modelled from current speed and direction data collected from September to November 1995 and assuming a settling speed of 4 $cm \cdot sec^{-1}$ for faeces, 13 $cm \cdot sec^{-1}$ for feed pellets and a bottom depth of 20 m. Each circle radiating from the centre represents 20 m.

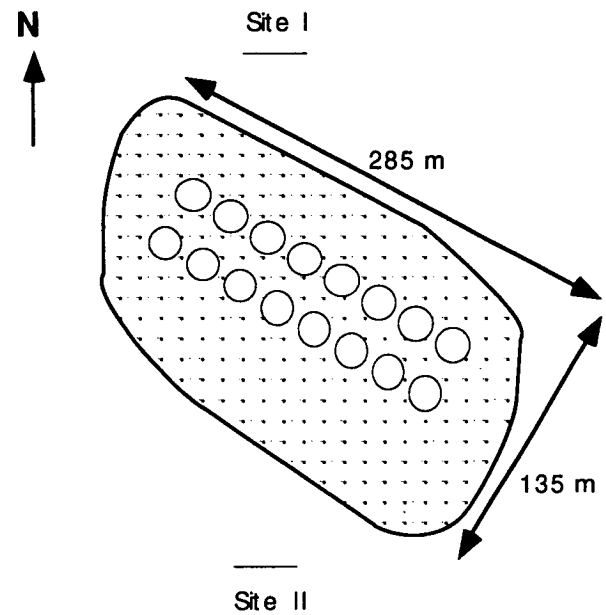


**Figure 13.4.** Predicted bottom distribution of 90% of particulate wastes from the Aquatas fish farm and the positions of the two farm-site mussel longlines, I (70 m north) and II (100m south).

## 13.2.3 Discussion

***Particulate concentrations***

Theoretical SPM and POM concentrations attributable to fish farm particulate wastes were extremely low compared with ambient concentrations within the bay. Due to the dilution of waste particles in the water column, the maximum theoretical waste particle concentrations (0.03 mg POM·$l^{-1}$ and 0.05 mg SPM·$l^{-1}$) would be undetectable (balance measured to ± 0.05 mg), only elevating ambient levels within 1.7%. Theoretical particulate concentrations would be even lower if a significant fraction of faeces were intact pellets, or if current speeds were included the calculations. These concentrations are in sharp contrast to the observed particulate levels at the fish farm, displaying up to 26% and 32% higher SPM and POM concentrations than ambient concentrations. Why then, was there such a discrepancy between theoretical and observed particulate values?

It is recognised that these calculations of theoretical waste loadings are based on a considerable number of assumptions, increasing the probability of modelling error. Sources of error include under-estimating feed fines and faecal discharge, over-estimating digestibility, and assuming continuous feeding and faecal discharge. However, in modelling fish waste loadings at a Chilean fish farm, Troell and Norberg (1998) reported similarly low faecal waste concentrations, ranging from 0.02 to 0.2 mg POM·$l^{-1}$, lending some support to the calculated particulate waste concentrations at the Aquatas farm.

Although particulate concentrations (including total SPM, POM, PIM and non-algal POM) were higher at the centre of the fish farm (site X) than at the other four mussel longline sites, they were: (a) also significantly higher during the fallowing period (June to mid-September 1995) when fish were not on the farm and waste particulates would not have been added to the water column; and (b) did not display any appreciable increases, absolute or relative to the other four sites, after fish were introduced and cultivated from September 1995 onwards. Therefore, it seems unlikely that the higher observed particulate concentrations at the fish farm were due to high levels of suspended feed and faecal waste particles. It is not known why particle concentrations on the farm were higher than ambient concentrations, particularly during the fallowing period. However, as the bottom depth was approximately 20 ± 0.5 meters (depending on tide) and water was sampled with a hose to approximately the same depth, it is possible that water samples at site X may have included resuspended material from the flocculant sludge-layer beneath the cages. No appreciable increases in particulate concentrations, after fish were re-introduced in

September 1995, would be expected if the theoretical waste concentrations are realistic. The absence of an observable increase in particulate concentrations suggests that the quantity of particulate wastes generated by the fish farm was too diluted to achieve detectable concentrations, lending support to the theoretical concentrations (maxima of 0.03 mg POM·$l^{-1}$ and 0.05 mg total SPM·$l^{-1}$). Unfortunately, due to the uncertainty regarding the reliability of observed fish farm particle concentrations, as bottom sediments may have contaminated them, we cannot really test the validity of the theoretical concentrations. However, if these theoretical waste particulate concentrations are realistic, the minimal rise in ambient food concentrations is unlikely to have a significant impact on increasing growth rates of mussels cultured within the farm.

Previous reports are inconsistent concerning the level of particulate matter associated with fish farms relative to ambient levels. Significantly higher SPM levels, relative to control sites, were associated with a fish farm in British Columbia (Jones and Iwama, 1991). In contrast, Okumus (1993) reported conflicting results, where seston and POM concentrations (mg·$l^{-1}$) at monitored salmon farms were higher than at control sites; but neither particle concentrations (numbers of particles·$ml^{-1}$) nor particle size frequency distributions were significantly different between the same sites. This discrepancy might indicate differences in the density of particles or possibly the disintegration of aggregates by the Coulter counter. Taylor et al. (1992) observed a lack of a significant association between SPM concentrations and distance from fishcages. This was attributed to net-fouling mussels intercepting suspended fish feed and faecal material that reduced the quantity of waste particles escaping the farm site. However, their conclusion was only speculative.

### *Distribution of particulate wastes*

Of particular interest, regarding the cultivation of mussels within the bounds of the Aquatas fish farm, is that particulate levels at sites I and II were not significantly different from sites III and IV. This suggests: particulate concentrations were higher at these two sites, but were below detectable levels; or waste particles were not horizontally transported to sites I and II (70 m and 100 m from the cages). It seems both factors are plausible. Low waste particle concentrations due to dilution has been previously discussed.

The modelled horizontal displacement of suspended waste particles suggests 90% of all particulate wastes would settle to the sea bottom within 50 m of the cages. These estimates agree with previous studies, suggesting the distribution of solid wastes is

largely restricted to the immediate area surrounding fish cages (Brown et al., 1987; Gowen and Bradbury, 1987; Gowen et al., 1988; Frid and Mercer, 1989; Lim, 1991; Ye et al., 1991; Woodward et al., 1992; Johnsen et al., 1993; Coyne et al., 1994; Krost et al., 1994; Findlay et al., 1995). Although variations in feed, feeding practices, feed and faecal settling velocities, current speed, and bottom depth would be expected among different farms, the consensus from these studies is that farm-derived sedimentation is largely concentrated within 10 m, and not significant beyond 50 - 60 m, of the fishcages. The modelled waste particle distribution in the present study indicates the two mussel longlines within the farm site (sites I and II) were located in the appropriate directional positions from the fishcages. However, they may have been located too distant from the cages for mussels to effectively intercept the bulk of suspended waste particulates (site I was 70 m north; site II was 100m south). Even at the maximum observed current speed of 25 cm·sec$^{-1}$, particulate wastes would still have settled to below 11 m by the time they reached the mussel longline at site I (Fig. 13.5). At a current speed of 25 cm sec$^{-1}$, a faecal settling rate of less than 1.8 cm sec$^{-1}$ would be required for mussels at the bottom of the dropper to intercept any particulates. Therefore, based on a faecal settling velocity of 4 cm·sec$^{-1}$ and the low current speeds at this site, it appears that the two within-farm mussel groups may not have been in ideal positions to obtain much additional feed from particulate wastes originating from the fishcages. A more appropriate location for longlines would be immediately adjacent to the cages. However, this might create a logistical problem for many fish farms (eg. boat access to cages, moving cages, net changes etc.).

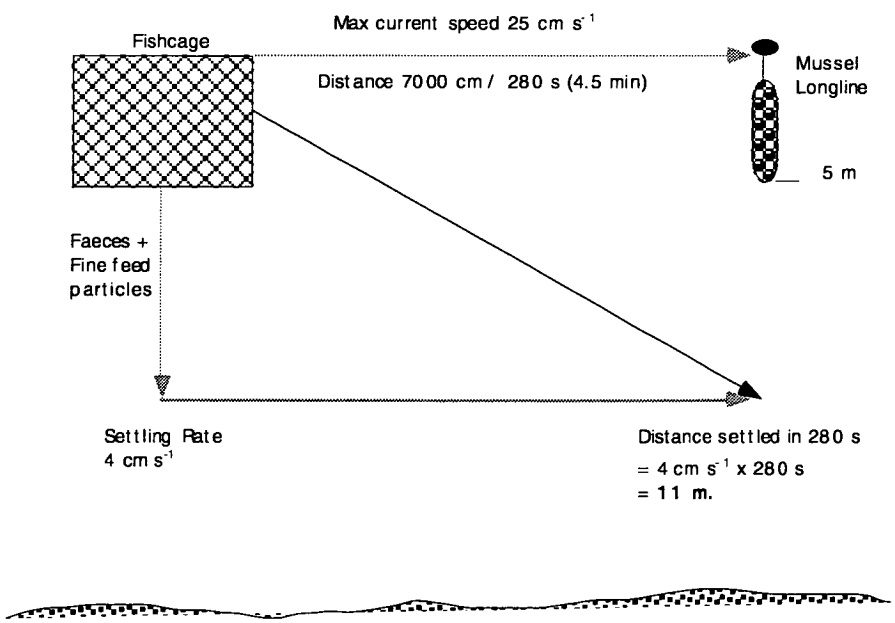


**Figure 13.5.** Vector diagram illustrating the displacement of particulate wastes (faeces and feed fines) at a current speed of 25 cm·sec$^{-1}$ and a particle settling speed of 4 cm·sec$^{-1}$. At this current speed, particulates transported to the mussel longline at site I (70 m from cages) would have settled to a depth of 11 m or 6 m beneath the bottom level of the mussel droppers (drawing not to scale).

It could be argued that the settling velocity used to determine the horizontal displacement of faeces and feed fines (4 $cm \cdot sec^{-1}$) was too high. This settling rate is an intermediate-estimate compared with some other rates reported in the literature, ranging from 2 $cm \cdot sec^{-1}$ (Findlay and Watling, 1994) to 6 $cm \cdot sec^{-1}$ (Chen et al., 1999). However, all reported settling velocities of salmon faeces are of intact faecal pellets; settling velocities may vary according to faecal pellet size and density, water temperature and salinity. Importantly, salmon also void faecal material as bursts of mucoid strings and particles. The fraction of total faecal output voided in this manner and the settling velocities of faecal particles and mucoid strings has not been investigated. Let us assume that a significant fraction of faecal material is particulate in nature with a much lower sinking velocity (eg. invertebrate faecal pellet settling speeds range from 0.002 to 1.0 $cm \cdot sec^{-1}$; Robison and Bailey, 1981 and references within). It is not unreasonable to suggest that: (a) the horizontal distribution of particulate fish farm wastes might be more extensive than the modelled distribution, and (b) a significant fraction of these particulate wastes might be transported to sites I and II before settling to the bottom. If this was a likely scenario during the experimental period, why weren't SPM and POM concentrations higher at sites I and II compared with sites III and IV? It is unlikely, at the current speeds measured in NWB, that suspended faecal wastes would be equally distributed among all longline sites, at least not as far as site IV (1.2 kilometers from the farm). Therefore, we must come back to our original explanation for similar POM and SPM concentrations among longlines: suspended fish farm wastes were too diluted to be detectable.

## 13.3 Nutrient Wastes

Dissolved nutrients are released into the water column directly from fish as excretory wastes and indirectly through the leaching or decomposition of solid wastes. Most attention has been given to nitrogen and phosphorus compounds, as these have been implicated in promoting primary production and eutrophication problems (Persson, 1991; Talbot and Hole, 1994). Nitrogen excretion products from catabolised proteins include ammonia and urea. Excretion rates are directly related to the nitrogen consumed in the feed. If there is surplus protein or the amino acid profile does not correspond to the requirements of the fish, the excess becomes deaminated, and nitrogen, primarily as ammonia, is excreted. Ammonia is the preferential source of nitrogen for phytoplankton growth.

In freshwater, enhanced levels of dissolved inorganic phosphate have caused eutrophication problems; phosphate released by fish farms in marine waters may cause similar effects (Persson, 1991). Nitrogen is considered to be the limiting nutrient for primary production in coastal waters, but the role of phosphorus may also be important (Valiela, 1984). On one hand, it has been suggested that, except for some low salinity coastal environments, phosphate is not influential in controlling algal growth, and is not an important waste product in seawater (Gowen and Bradbury, 1987). On the other hand, there is some evidence that soluble phosphorus, or deviations from normal N:P ratios, may influence the formation of toxic phytoplankton blooms, or increase the toxicity of certain species (Smayda, 1990; Holby and Hall, 1991; Kaartvedt et al., 1991; Folke et al., 1994; Johansson and Graneli, 1999). Therefore, phosphorus concentrations may be more important in coastal waters than previously believed.

Little attention has been paid to the release of silicates from fish farms. This waste product is also of interest, as it can be the limiting nutrient in diatom production (Raymont, 1976). Also, it has been suggested that sustained imbalances in nutrient availability, with large amounts of N and P relative to Si, may favour the development of toxic dinoflagellate blooms with the potential to cause large-scale destruction of mariculture operations (Smayda, 1990; Holby and Hall, 1994).

As nutrient wastes released by fish farms have the potential to significantly alter the natural productivity within coastal embayments, dissolved nutrient waste loadings from the Aquatas fish farm into NWB were estimated. These theoretical loadings were compared with actual concentrations measured during the mussel growth trial and their potential for increasing ambient nutrient concentrations are discussed.

## 13.3.1 Methodology

### *Calculation of nutrient wastes*

Monthly environmental loadings of nitrogen and phosphorus were estimated from feed and production data supplied by Aquatas Pty. Ltd. and incorporated into the following equations:

Total nutrient loading ($kg \cdot month^{-1}$) = nutrient in feed - nutrient retained in fish

or, alternatively = sum of nutrient in wastes (nutrient in uneaten feed+faecal wastes+excretion)

where,

nutrient in feed = feed out ($kg \cdot month^{-1}$) x 0.072 (N) or 0.016 (P)

nutrient retained in fish = fish production ($kg \cdot month^{-1}$) x nutrient conc. in fish (%)

and

% N in fish = 0.096 x 17.4 x Fish wet weight $(g)^{0.099}$ (Hall et al., 1992)

% P in fish = 0.45 (Einen et al., 1995)

nutrient in uneaten feed = 0.05 x nutrient in feed (ie. 1% fines + 4% uneaten pellets)

Solid faecal nitrogen ($SF_N$) and dissolved excretory ammoniacal nitrogen (DN) discharges per kg fish production were calculated according to Nijhof (1994) by the following formulae:

$$SF_N = FC\,(1 - AD_N) \times F_N$$

$$DN = [(F_{CP} \times FC \times AD_{CP}) - CP_{fish}] / 6.25$$

where,

$SF_N$ = solid faecal discharge of N ($g \cdot kg$ fish production$^{-1}$)

FC = feed conversion (kg feed *consumed* per kg fish produced)
feed consumed = 0.95 x feed delivered (assumption)

$AD_N$ = apparent digestibility of N/crude protein (0.9, Pivot Aquaculture)

$F_{N\,(CP)}$ = dietary content of N (72) or crude protein (450) ($g \cdot kg$ feed$^{-1}$)

DN = Dissolved ammoniacal nitrogen release per kg production ($g\ N \cdot kg^{-1}$)

= 90% of total excreted N; balance is excreted urea-N (Bergheim et al., 1996)

$CP_{fish}$ = crude protein content of fish produced; %N in fish x 62.5 ($g \cdot kg^{-1}$)

According to the relationship between fish weight and %N, nitrogen content of fish varied with size, ranging from 2.5% to 3.5% N, corresponding to 161 to 220 g CP·kg fish$^{-1}$.

Solid faecal phosphorus ($SF_P$) and dissolved excretory phosphorus (DP) discharges were calculated by:

$$SF_P = FC\,(1 - AD_P) \times F_P \qquad \text{(Nijhof, 1994)}$$

$$DP = \text{(Feed-P consumed)} - \text{(P-retained in fish)} - SF_P$$

where:

| | | |
|---|---|---|
| $SF_P$ | = | solid faecal discharge of P (g·kg fish production$^{-1}$) |
| $AD_P$ | = | apparent digestibility of P (0.67, Pivot Aquaculture) |
| $F_P$ | = | dietary content of P (1.6%, Pivot Aquaculture) |
| DP | = | dissolved excretory phosphorus (kg) |
| Feed-P consumed | = | Feed out (kg·month$^{-1}$) x 0.95 x 0.016 |
| Fish-P retained | = | biomass gain (kg·month$^{-1}$) x 0.0045 |

Silica waste loadings and silica content of fishfeed and fish were based on Holby and Hall (1994) who investigated waste Si loadings at a marine rainbow trout farm. As silica content in the feed used at the Aquatas farm was unknown (no data on Si content available from feed manufacturer), the Si content was set at 1200 mg Si·kg feed$^{-1}$ (dry weight) according to the Si content in commercial feeds analysed by Holby and Hall (1994). Si content in fish was set at 5 mg·kg$^{-1}$ (wet weight), also suggested by Holby and Hall (1994). These authors propose that loss of dissolved reactive silicate ($SiO_4$) by fish excretion and leaching from sinking particulates (feed and faeces) is insignificant and can be discarded from calculations. Therefore silicate loadings are entirely in the particulate form and can be estimated by the difference between Si contents in the feed and fish. Subsequently, Si loading into the water column is due to the flux of dissolved reactive silicate from the sediment. Annual benthic flux rates were estimated as 0.3% of the biogenic silica present in the farm sediment (Holby and Hall, 1994), suggesting a very small return of silicates to the water column.

## 13.3.2 Results

### *Nitrogen*

The calculated environmental loading of nitrogen for the period February 1995 through to May 1996 ranged up to 5.8 tonnes N·month$^{-1}$, 190 kg N·day$^{-1}$, or 56 - 61 kg N·tonne fish production$^{-1}$ (Fig. 13.6, Table 13.3). Approximately 36% of the total nitrogen input in the feed was retained by the fish with the remaining 64% lost to the environment (15% solid and 49% dissolved). Nitrogen in uneaten pellets, fish feed fines and faecal discharge, approximately 23% of the total N loss, precipitates to the sea floor. Of the sedimented N, approximately 89% accumulates in the sediments with

the remainder released back into the water column over time, mostly as dissolved organic nitrogen and small quantities of urea and ammonia (Hall et al., 1992). Of the total N loss, 77% was in the dissolved form (ammonia and urea) which would be directly available for phytoplankton production.

### *Phosphorus*

Total phosphorus loadings ranged up to 1.5 tonnes P·month$^{-1}$, representing a 78% loss of P in the feed or 16 kg P·tonne fish production$^{-1}$ (Fig. 13.6; Table 13.4). Fifty-three percent of the calculated total P loss was excreted in dissolved form, which can be as organic P and orthophosphate (Lall, 1991).

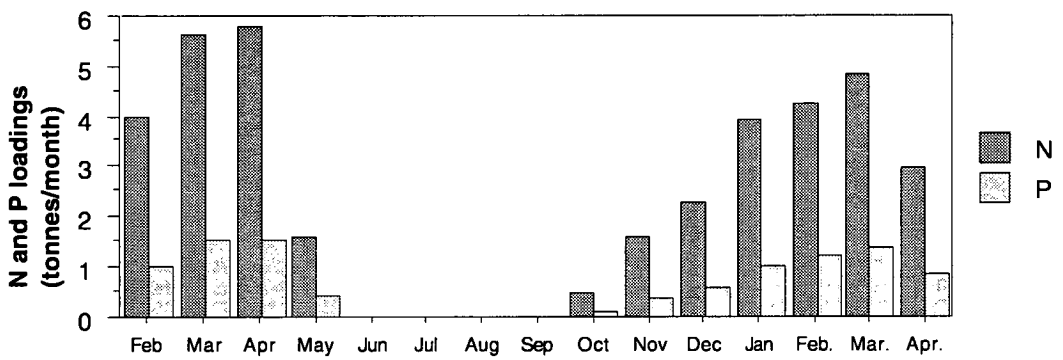


**Figure 13.6.** Total theoretical fish farm nitrogen and phosphorus loadings (tonnes N and P·month$^{-1}$) into North West Bay for the period February 1995 to April 1996.

### *Silica*

The total silica loadings to the environment are supplied from uneaten feed and faeces, amounting to 99.7% of the silica supplied in the feed, with fish retaining the remainder. Monthly theoretical Si loadings ranged up to 135 kg Si·month$^{-1}$, averaging 1.5 kg Si·tonne fish production$^{-1}$ (Fig. 13.7 and Table 13.5). At a benthic flux rate of only 0.3% from sedimented-Si·year$^{-1}$ (Holby and Hall, 1994), biogenic silica would accumulate over time with only a small amount of dissolved silicate returned to the water column.

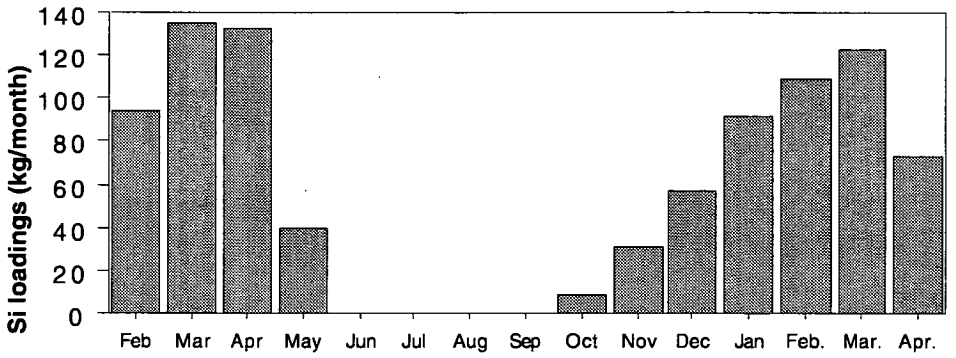


**Figure 13.7.** Total theoretical sedimentary silica loadings (kg Si·month$^{-1}$) from fish feed and faeces for the period February 1995 to April 1996.

## 13.3.3 Discussion

### *Nutrient waste loadings*

Comparison of nutrient loadings with other studies can be somewhat confusing, as results are often expressed in different formats, suggesting a standardised format would be beneficial. Differences in diet formulations, nutrient levels, digestibility, feeding practices, estimates of uneaten feed, fish (species, sizes, ages), and experimental conditions (tanks, raceways, cages, freshwater, saltwater) have all contributed to a wide variation in reported waste loadings. Although calculated waste loadings from the Aquatas farm are only theoretical and based on a number of assumptions, mean loadings of 56 - 61 kg N and 16 kg P per tonne fish production agree with the environmental losses claimed by the feed manufacturer (56.4 kg N and 13.9 kg P·tonne fish production$^{-1}$ at an FCR of 1.2, Pivot Aquaculture). The mean total nitrogen environmental loading of 56 - 61 kg N·tonne fish production$^{-1}$ compares favourably with 60 kg N·tonne fish production$^{-1}$ reported by Ackefors and Enell (1994) under similar conditions of feed protein content and feed conversion. Mean N loadings of 64% of feed-N (56 - 61 kg N·tonne fish production$^{-1}$) are lower than 72-80% (86 - 104 kg N·tonne fish production$^{-1}$) reported in some earlier studies (references in Hall et al., 1992). Lower N loadings in this study are due to a higher feed digestibility and a lower rate of uneaten feed losses incorporated into the calculations. Ammonia excretion rates of approximately 34 g $NH_4$-N·kg feed$^{-1}$ are similar to 10-40 g N·kg feed$^{-1}$ (Bergheim et al., 1991) but higher than 12-20 g N·kg feed$^{-1}$ (Fivelstad et al., 1990). In relation to ingested N and P, estimated fish retention rates of 38%(N) and 23%(P), faecal losses of 10%(N) and 33%(P), and soluble wastes of 51%(N) and 44%(P) are within the ranges typical for salmonid culture (Talbot and Hole, 1994). Mean P loadings of 79% of feed-P or 16 kg P·tonne fish

production$^{-1}$ are also within the range of 77-88% of feed-P (references in Holby and Hall, 1991), 8.1-40.2 kg P·tonne fish production$^{-1}$ (references in Lall, 1991), and 19.6-22.4 kg P·tonne fish production$^{-1}$ (Holby and Hall, 1991). However, 53% of total P losses as dissolved phosphorus is considerably higher than some other studies, such as: 25-30% (Holby and Hall, 1991), 16% (Ackefors and Enell, 1990), and 9% (Johnsen et al., 1993 in Bergheim and Asgard, 1996). Higher levels of theoretical excreted P may due to several factors: lower fish-P content (0.45%), compared with 0.5 to 0.8% (Lall, 1991; Ackefors and Enell, 1994); higher levels of feed phosphorus (1.6%), compared with levels under 1.0% in other diets (Wiesmann et al., 1988; Ackefors and Enell, 1990; 1994); and a high phosphorus digestibility coefficient (0.67), which can vary significantly between diets according to chemical form, diet digestibility, particle size, type of fishmeal, and feed processing (Ackefors and Enell, 1994). Therefore, dissolved P loadings may have been overestimated. Silicate loadings, approximately 1.5 kg Si·tonne fish production$^{-1}$, compares reasonably with 2.5 kg Si·tonne fish production$^{-1}$ reported by Holby and Hall (1994).

### Effect of the Aquatas fish farm on nutrient concentrations in North West Bay

No differences in measured dissolved nitrogen concentrations (nitrate, nitrite or nitrate + nitrite) between the five sampling locations were significant during the trial period. The extent to which a fish farming operation influences the concentrations of nitrate and nitrite in the water column is probably minor. As ammonia and urea are well recognised as being the primary salmonid excretion products, no direct input of nitrate or nitrite from fish into the water column would be expected. Additional quantities of these compounds might possibly come from the decomposition of organic matter in the sediments and nitrification of excreted ammonia. Several studies of land-based fish farms have failed to identify significant increases in nitrate or nitrite concentrations in farm effluent (Bergheim and Selmer-Olsen, 1978; Butz and Vens-Cappell, 1982; Bergheim et al., 1984). Significant increases in nitrate concentrations have not been detected within some salmon farms (Gowen et al., 1988; Wildish et al., 1993); minor elevations of nitrite concentrations were detected by Aure et al. (1988). In a study investigating nitrogen enrichment at a marine farm in western Sweden, nitrate fluxes *into* bottom sediments from the overlying water column were observed (Hall et al., 1992). Therefore, it appears unlikely that fish culture would significantly elevate nitrate or nitrite concentrations at the Aquatas fish farm, or within NWB generally.

Unfortunately, due to the lack of an appropriate methodology, the nutrient of most interest, ammonia, was not monitored. Significant increases in ammonia concentrations in the vicinity of seacage farms, at times, have been reported (Aure et

al., 1988; Gowen et al., 1988; Falconer and Hartnett, 1992; Wildish et al., 1993; HES, 2000). Temporal and spatial variation in measured ammonia concentrations within fish farms would be due to a number of factors such as fish biomass, feed-N digestibility, excretion rates, time of day, ammonia methodology, rate of ammonia uptake by phytoplankton, and water flow, flushing rates and subsequent dilution in the water column. Using literature values of ammonia production (Nijof, 1994), theoretical estimates of water column ammonia concentrations due to fish excretion on the farm site were calculated. As with the estimations of solid waste concentrations, monthly ammonia production was back-calculated to determine the output from a single cage. The expected residence time of the water column passing through a cage would be approximately 9 minutes at a mean current speed of 3.5 $cm \cdot sec^{-1}$; therefore the quantity of ammonia-N excreted from one cage in 9 minutes was estimated to determine the expected increase in nitrogen concentrations in the water column.

Calculated total ammoniacal nitrogen (TAN) excretion ranged from 0.6 to 6.3 kg TAN per cage per day, varying with fish biomass and feed output (Table 13.3, C). The contribution of TAN from benthic flux was not included in the calculation, as this component was considered to be minor compared with excreted ammonia (less than 1% of excreted N, Gowen et al., 1988; Hall et al., 1991). Therefore, the corresponding theoretical water column TAN concentrations due to fish excretion at the farm site range from 0.6 to 6.0 $\mu g\ TAN \cdot l^{-1}$. Based on these calculations, excreted ammonia could potentially raise ambient nitrogen concentrations (based on measured $NO_x$ concentrations only) two to three fold on the farm site in the summer months from November 1995 to March 1996. However, the actual total nitrogen concentrations within NWB are unknown, as ambient ammonia and dissolved organic nitrogen concentrations were not monitored. Reported ammonia levels from predominantly marine waters of the lower Derwent River estuary, adjacent to NWB, ranged up to 75 $\mu g\ NH_4\text{-}N \cdot l^{-1}$ in 1993-94 (DELM, 1995). If these levels are representative of those that may be expected in NWB, it appears that the additional ammonia input from fish excretion might not have significantly increased the total nitrogen concentration within the farm-site, nor within the bay. It is recognised that the ammonia levels within the Derwent estuary would be influenced by industrial and domestic discharges from the Hobart municipality. In contrast, reported 1997-98 ammonia levels in the lower Huon River estuary, approximately 25 km south of NWB, were generally below 16 $\mu g\ TAN \cdot l^{-1}$; TAN concentrations near some fish farms appeared to be occasionally higher than other sites within the estuary (HES, 2000). However, ammonia concentrations reported in the literature should always be viewed with some caution; accurate seawater ammonia measurements are infrequently achieved, particularly using dated methodologies (Holmes et al., 1999). Without

reliable measurements of ammonia concentrations within NWB, no definite conclusion regarding the contribution of fish farm ammonia to ambient levels can be drawn.

Elevated phosphate concentrations within the Aquatas fish farm was not indicated; this agrees with some studies (Wildish et al., 1993), but slightly elevated phosphate concentrations within fish farms have been reported by others (Aure et al., 1988). The theoretical output of dissolved phosphate from the Aquatas farm into the water column ranged from 2 to 28 kg $PO_4$-P·day$^{-1}$, varying with fish production (Table 13.4, B). As with ammonia, the daily output of excreted $PO_4$-P was back-calculated to estimate the fish farm's contribution to ambient phosphate concentrations. The contribution of $PO_4$ from benthic flux was not included in the calculation, as this component was considered to be minor (2.5 - 4.5% of the sedimented P, Holby and Hall, 1991). Theoretical enrichment by excreted phosphate ranged from 0.1 to 1.4 μg $PO_4$-P·l$^{-1}$ (Table 13.4, B). These levels are low compared with the ambient $PO_4$ levels during the same time period (9.3 to 13.1 μg $PO_4$-P·l$^{-1}$). Although accounting for a maximum of 13% of the measured $PO_4$-P concentrations, the fact that no measurable differences were observed among sites suggests that the calculated levels of phosphate excretion may be overestimated.

There was evidence of elevated silicate levels at the fish farm, where overall mean silicate concentrations increased with increasing proximity to the centre of the farm (Section 4). Monthly mean farm-site Si concentrations (site X) exceeded ambient (site IV) by up to 32% (Table 13.5, B). However, a significant site x time interaction was indicated; subsequent post hoc testing revealed differences in Si concentrations between sites X and IV on only 10 of the 46 sample periods, primarily during the final three months. Fish farms have not been implicated in elevating silicate concentrations in previous studies (Aure et al., 1988; Guildford, 1993; Wildish et al., 1993), probably due to the low levels of silica in feed, insignificant Si excretion by fish, and low benthic flux rates from sedimented Si (Holby and Hall, 1994). Therefore, it is interesting that Si concentrations at the Aquatas site were significantly higher than ambient levels at certain times.

Assuming, unrealistically, all sedimented feed-Si over several growing seasons remained on the farm site, the benthic flux of silicate into the overlying water column would still be negligible. For example: if the Si content of the feed was constant; the quantity of feed delivered, fish production and waste loadings of the 1995-96 season is representative of past years; and the annual benthic flux rate of Si is 0.3% (Holby and Hall, 1994); then the theoretical quantity of biogenic Si in waste fish feed and faeces accumulating on the sea floor approaches 3.5 tonnes after 8 years (Fig. 13.8). Benthic flux rates after 8 years of Si accumulation would only be 10 kg Si·month$^{-1}$ or

300 g $Si \cdot day^{-1}$ from the entire farm site (Fig. 13.8). Clearly, this quantity of Si flux would not elevate ambient silicate concentrations to detectable levels. Possibly, some of the assumptions used in the calculations of theoretical silicate loadings have been underestimated, including feed silica content, solubility, benthic flux rates, and fish excretion. Also, water samples may been contaminated with resuspended bottom material arising from the flocculant layer beneath the cages. This material would include silica originating from fish feed and possibly external sources of sedimentary silica, such as terrestrial particulates and phytoplankton deposition. Holby and Hall (1994) estimated 55-80% of the biogenic silica in sediments at a Swedish fish farm was of diatomaceous origin.

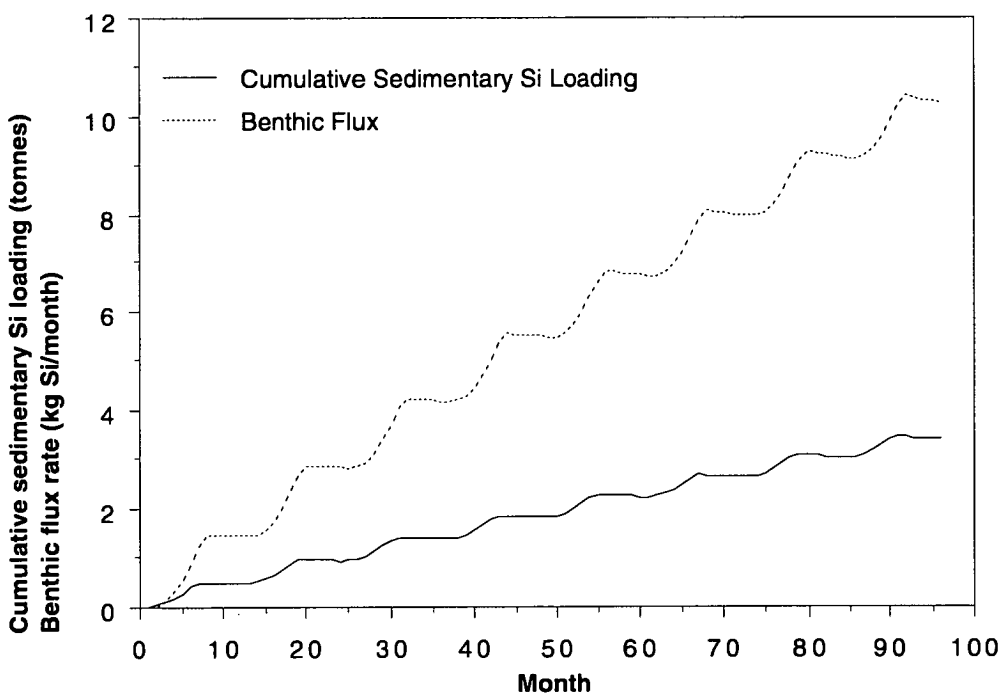


**Figure 13.8.** Theoretical sedimentary silica loadings (tonnes) and benthic flux rates (kg $SiO_4$-$Si \cdot month^{-1}$) over an eight year period at the Aquatas fish farm based on a benthic flux rate of 0.3% of sedimented Si loadings per year.

Holby and Hall (1994) suggested fish farms, with their associated nitrogen and phosphorus loadings, may create a good niche for the growth of Si-utilising diatoms. Thus fish farms actually take up silicate from the surrounding water as a result of diatom growth. It has also been proposed that sustained imbalances in nutrient availability, with large amounts of available N and P relative to Si, create conditions favouring the development of toxic dinoflagellate blooms, potentially affecting mariculture operations (Smayda, 1990; Maestrini and Graneli, 1991; Schollhorn and Graneli, 1993; 1996). Therefore, Holby and Hall (1994) propose that the excess supply of N and P from fish farms need to be balanced with inputs of Si in order to

sustain a healthy population of diatoms. Minimising conditions that might promote toxic dinoflagellate blooms represents another reason, other than oxygen supply and dispersal of wastes, for locating cages in areas receiving adequate amounts of fresh seawater and deliberately adding biogenic silica to fish food (Holby and Hall, 1994). In the present study, realistic Si:N ratios cannot be calculated as, with ammonia and other nitrogenous compounds unaccounted for, total nitrogen concentrations are unknown. Si:P ratios were significantly higher at the salmon farm sites X and I (mean $9.4 \pm 0.4$, n = 184 samples) than at the other three sites (mean $8.6 \pm 0.3$, n = 276 samples; $P < 0.05$). It would be tempting to suggest that the farm site, with its higher Si concentrations, might actually be promoting the growth of diatoms over dinoflagellates. However, differences in Si:P ratios among sites were minor and this suggestion seems unlikely. A comparison of Si:P ratios between sites X and IV is illustrated in Fig. 13.9.

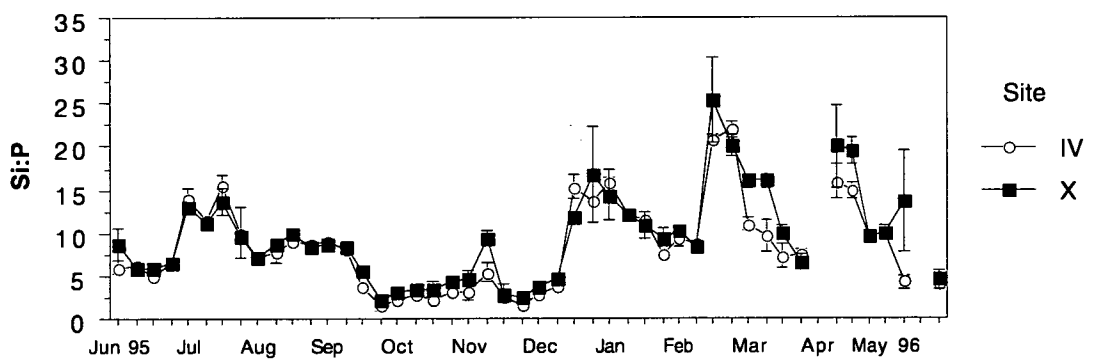


**Figure 13.9**. Si:P ratios at the Aquatas fish farm (site X) compared with ambient Si:P ratios (site IV) from June 1995 to May 1996.

In Nova Scotia, no differences in seawater nutrient concentrations (total inorganic nitrogen, phosphate and silicate) or chlorophyll-a were observed between sites associated with aquaculture activities (mussel strings and salmon pens) and control sites (Guildford, 1993). However, there was a significant difference in the nutritional status of the resident phytoplankton; phytoplankton associated with aquaculture activities were less N-deficient and more Si-deficient than those at control sites. Although there may be no detectable difference in nutrient concentrations between aquaculture sites and other sites, there may be a change in the nutritional status of plankton associated with aquaculture sites. Toxin production in some species of flagellates has been associated with an altered physiological nutrient status (ie. Si:N:P ratios; references within Maestrini and Graneli, 1991 and Guildford, 1993; Johansson and Graneli, 1999). Therefore, nutrient inputs from fish farms, as well as other anthropogenic sources, have the potential to alter the natural nutrient balance of coastal

waters; possibly promoting toxic dinoflagellate blooms or increasing the toxicity of certain species.

It is apparent that the Aquatas fish farm is responsible for releasing dissolved nutrients into North West Bay, ammonia being most notable. From an environmental point of view, the questions of interest in this study are:

1. are the fish farm nutrient loadings significantly increasing the ambient nutrient concentrations within NWB?
2. what are acceptable levels of nutrient enrichment within NWB?
3. are the nutrient loadings significant in terms of promoting nuisance algal blooms?
4. are the nutrient loadings stimulating localised phytoplankton production, thereby providing mussels with an increased food supply?

### *1. Are fish farm nutrient loadings significantly increasing the ambient nutrient concentrations within NWB?*

The degree of hypernutrification (defined in this instance as a detectable increase in nutrient concentrations over ambient levels) of selected nutrients within coastal bays can be estimated from the equilibrium rise in nutrient concentration (Gowen et al., 1988; Turrell and Munroe, 1989; Aure and Stigebrandt, 1990; Silvert, 1992; Gowen, 1994). Assuming complete mixing of the fish farm excretion products within North West Bay, a flushing rate of the bay of 7 days (Matthews and Volframs, 1978), no recirculation of flushed water back into the bay, and discounting the contribution of nutrient benthic flux from farm sediments, the nitrogen and phosphorus equilibrium concentrations were calculated as:

$$E_c = (W \times F) / V$$

where $E_c$ is the equilibrium concentration of N or P, W is the mean daily output of soluble nutrient wastes (N or P), F is the flushing time in days and V is the volume of NWB ($2.8 \times 10^{11}$ litres; Matthews and Volframs, 1978).

Monthly nitrogen equilibrium concentrations were calculated from estimates of excreted ammonia-N + urea-N only; nitrogen inputs from benthic flux were disregarded as they were considered to be minor relative to excretory losses (< 1-3%, Gowen et al., 1988; Aure and Stigebrandt, 1990; Hall et al., 1992). Monthly nitrogen $E_c$ varied with fish production, with the highest level of 3.8 $\mu g\ N \cdot l^{-1}$ occurring in April 1995 (Table 13.3-B). This means that the Aquatas fish farm might be responsible for raising soluble nitrogen levels within NWB by 0.3 to 3.8 $\mu g\ N \cdot l^{-1}$, depending on the

time of year. Monthly phosphorus $E_c$ values, calculated from estimated soluble phosphate excretion, were less than 0.7 μg $P \cdot l^{-1}$ (Table 13.4-C). Silicate $E_c$ would likely be negligible; less than 0.0000085 μg $Si \cdot l^{-1}$ after 8 years of accumulated sediment Si, assuming a benthic flux rate of 0.3% of sedimented Si per year.

Are the $E_c$ concentrations, attributable to nitrogen and phosphorus discharges from the fish farm, significantly increasing the nutrient levels within North West Bay? Although fish farm nitrogen discharges might double N concentrations within NWB during January-March 1996 (based solely on measured $NO_x$), the total dissolved N concentrations within NWB would still be less than 6 μg $N \cdot l^{-1}$ (Table 13.3-B). As mentioned previously, the total dissolved nitrogen concentrations are unknown, as ammonia and dissolved organic nitrogen were not measured. However, if the background ammonia concentrations within the bay are comparable to those reported in the lower Derwent River estuary (up to 75 μg $NH_4$-$N \cdot l^{-1}$; DELM, 1995), then the level of nitrogen discharge from the fish farm may be insignificant in raising the total dissolved nitrogen concentrations within the bay. The overall mean phosphorus $E_c$ from fish excretion was 0.35 μg $PO_4$-$P \cdot l^{-1}$ for the 1995 - 96 growing season. This rise is insignificant compared with ambient $PO_4$-P concentrations ranging from 9 to 19 μg $PO_4$-$P \cdot l^{-1}$.

What is the fish farm's contribution to the total nutrient inputs to NWB? In order to achieve a balanced perspective upon the environmental effects of fish farms, the emission of dissolved nutrients from fish farms should be compared with other anthropogenic emissions and inputs from natural horizontal and vertical nutrient flows within and between water bodies. Any rise in nutrient concentrations within NWB, attributable to the Aquatas fish farm, might be insignificant relative to other sources of nutrient inputs. However, estimating the farm's relative contribution to the nutrient budget of NWB requires a comprehensive assessment of all other nutrient sources entering the bay from the surrounding catchment area, domestic and industrial inputs and from adjacent water bodies; an assessment which is beyond the scope of this study.

Some nutrient inputs would be sourced from the catchment area of North West Bay (260 square kilometres), comprising natural bushland, agricultural properties, and numerous residential areas drained by the North West Bay River and several small creeks. Stormwater from several small towns, two sewage treatment plants, residential septic systems, several fish processing plants, a refuse disposal site and a marina, all contribute to the nutrient loading within North West Bay. Nutrient inputs from marine sources and adjacent estuarine systems (Derwent and Huon rivers) might also contribute substantially to the total nutrient budget of NWB. A significant

contribution to the annual nitrogen and phosphorus budgets of the Huon River estuary comes from nutrient-rich marine water of the Southern Ocean (HES, 2000). Similarly, Aure and Stigebrandt (1990) suggest N and P from oceanic waters comprise over 90% of the total nutrients inputs into Norwegian fjords; this compares with only 3.5% of nutrient inputs from fish farms. As all possible nutrient inputs into NWB were not assessed, the relative contribution of fish farm N and P loadings to the total nutrient budget cannot be evaluated.

### *2. What are acceptable levels of nutrient enrichment within NWB?*

In relatively closed systems, characteristic of many freshwater aquaculture systems, relationships between nutrient loading and primary production have provided a basis for establishing allowable levels of hypernutrification, but these are not as well established for marine systems (ANZECC, 1992; Silvert, 1992). Nutrient enrichment is not always synonymous with eutrophication; additional nutrients only stimulate phytoplankton growth when a particular nutrient is limiting (Gowen, 1994). At present, there are no specific recommendations for acceptable levels of nutrient enrichment that might prevent nuisance algal blooms within Australian coastal waters. This is largely due to the number of site-specific factors (poor light, high turbidity, temperature, high grazing rates, salinity) which can also limit the development of algal blooms. Also, there appears to be no consensus of what constitutes a "nuisance growth" of algae; although in the UK, chlorophyll-a concentrations regularly exceeding 10 ug·l$^{-1}$ during the summer may be indicative of eutrophic conditions (Gowen, 1994). The following table provides ranges of concentrations, at or above, where "problems" have been known to occur in Australian waters (ANZECC, 1992).

| | Estuaries and embayments | Coastal waters |
|---|---|---|
| $NH_4$-N | < 5 µg l$^{-1}$ | < 5 µg·l$^{-1}$ |
| $NO_3$-N | 10 - 100 µg l$^{-1}$ | 10 - 60 µg·l$^{-1}$ |
| $PO_4$-P | 5 - 15 µg·l$^{-1}$ | 1 - 10 µg·l$^{-1}$ |
| chlorophyll-a | 1 - 10 µg·l$^{-1}$ | < 1 µg·l$^{-1}$ |

### *3. Are the nutrient levels within the bay significant in terms of promoting nuisance algal blooms?*

Nutrient and chlorophyll concentrations sampled within North West Bay during 1995-96 generally fell within the ranges indicative of eutrophication problems reported in Australian coastal waters (ANZECC, 1992; see table above). Chlorophyll-a concentrations within NWB were not dissimilar to levels reported in adjacent waters

without salmon farms (DELM, 1995; Crawford et al., 1996). However, no appearance of a significant algal bloom, indicative of eutrophic conditions, occurred during the trial; peak chlorophyll concentrations were below 6 μg chlorophyll-a·$l^{-1}$.

Although nutrients are often assumed to play a role in algal growth, other factors may be of equal or greater importance. In Tasmania, sustained blooms of the toxic dinoflagellate *Gymnodinium catenatum* require the interaction of several factors: high water temperatures (>14 °C), a significant rainfall event carrying high levels of organic and inorganic nutrients, a stable water column for 5 or more days, and possibly an unknown limiting micronutrient (Hallegraeff et al., 1989;1995; DPIF, 1994). Therefore, specific nutrient concentrations that might promote nuisance algal blooms are not appropriate for all situations. Research into site-specific factors influencing phytoplankton growth is required to determine appropriate guidelines for acceptable nutrient concentrations within different coastal systems.

### *4. Are nutrient wastes from the fish farm contributing to enhanced phytoplankton productivity and thereby providing mussels with an increased food supply?*

As phosphorus is not generally considered to be limiting for phytoplankton production in marine waters, ammonia rather than phosphate excretion is probably the most important fish farm waste nutrient. If the calculated ammonia production from the 1995-96 season is averaged over the seven month growing season, a typical fish cage produces approximately 12000 mmol ammonia-N·$h^{-1}$. As ammonia is the preferred nitrogen source for marine algae (Lomas and Glibert, 1999), phytoplankton is a potential sink for the flux of ammonia from fish farms. The relationship between phytoplankton growth and dissolved nitrogen can be assessed by using the subsistence quota: the minimum cellular content of a nutrient (eg. 0.05 mol nitrogen:mol carbon; Tett and Droop, year of publication unspecified, cited in Gowen et al., 1988). A carbon to chlorophyll ratio of 50:1 gives an estimate of 0.2 mmol nitrogen per mg chlorophyll. Thus, assuming all of the excreted ammonia is used for phytoplankton growth, 1 mmol of nitrogen could support a chlorophyll biomass of 5 mg (Gowen et al., 1988). Therefore, the mean rate of ammonia excretion at the Aquatas fishfarm (4 kg TAN per cage daily or 11 mol TAN per cage per hour) could theoretically support phytoplankton production at a rate of about 60 g chlorophyll-a·$cage^{-1}$·$h^{-1}$. This corresponds to a concentration rise of approximately 1.4 μg chlorophyll-a·$l^{-1}$, assuming a water column residence time of 9 minutes within each cage. An increase of 1.4 μg chlorophyll-a·$l^{-1}$ over ambient levels during the 1995-96 fish-growing season would have resulted in an overall mean of 2.7 μg chlorophyll-a·$l^{-1}$ at the farm site, compared with the observed overall mean of 1.3 μg·$l^{-1}$. This hypothetical level of

phytoplankton biomass would certainly have been detectable and significantly higher than background levels within the bay.

As ammonia levels were not measured, it is unknown if excreted ammonia significantly increased ambient concentrations to levels that might promote phytoplankton production. However, as there was no significant differences in measured chlorophyll-a concentrations among the five sampling sites, it may be that: (a) any additional ammonia-N was not utilised by phytoplankton; (b) ammonia excretion was overestimated and actual levels of additional nitrogen were insufficient to stimulate phytoplankton growth; (c) excreted ammonia was too diluted to stimulate phytoplankton growth; (d) some other factor other than nitrogen was controlling phytoplankton growth; or (e) excreted ammonia stimulated phytoplankton production, but this growth was delayed, dispersed, or consumed as rapidly as it was produced.

Gowen et al. (1988) suggested the most probable explanation for the lack of increased phytoplankton growth in the vicinity of a fish farm in Scotland was related to the short residence time of water around the fish cages. Firstly, despite low current speeds (less than 16 $cm \cdot sec^{-1}$), the water flow through the farm was probably sufficient to disperse much of the excreted ammonia. Secondly, additional nitrogen need not necessarily be utilised by phytoplankton. Thirdly, some of the additional ammonia could have been absorbed, as the time scale for uptake is in the order of 1 - 2 minutes; but the time-scale for phytoplankton growth can be in the order of days, depending on various factors such as light, temperature and species. Gowen et al. (1988) concluded that any additional phytoplankton biomass would be transported away from the farm, dispersed within the loch and masked by the greater proportion of ambient phytoplankton. These arguments may also apply to this study, explaining the similarities in phytoplankton biomass among experimental sites. As a consequence, mussels cultured within the fish farm (sites I and II) did not gain any nutritional advantage by way of increased phytoplankton.

The effect of the Aquatas fish farm on the environmental conditions within North West Bay, based on theoretical waste loadings, may be summarised as:

1. Theoretical suspended solid waste loadings from the farm, although substantial (up to ≈700 kg total SPM·day$^{-1}$ and 462 kg POM·day$^{-1}$), would only increase ambient suspended particulate concentrations by a maximum of 1.7%. However, observed particulate concentrations within the fish cage system were significantly higher (up to 32%) than at other sites within NWB. This suggests that theoretical waste loadings are inaccurate and/or water samples from the farm site were contaminated with sediments from beneath the fish cages.
2. Based on measured current speeds and settling velocities of 13 cm·sec$^{-1}$ (feed pellets) and 4 cm·sec$^{-1}$ (faeces and feed fines), the distribution of solid wastes would likely be restricted to the immediate vicinity of the farm site; 90% of solid wastes would settle to the bottom within 50 m of the cages.
3. It appears unlikely that mussels grown at sites I or II had a significantly greater food supply (fine fish feed particles, faeces, phytoplankton) than those grown away from the farm. SPM, POM and phytoplankton biomass were not significantly higher at sites I or II than at the two remote longline sites (III and IV). Longlines positioned within the farm site may have been too far from the fish cages for mussels to intercept a significant portion of suspended waste particles. Due to low current speeds, waste particles may have settled to the bottom before reaching the longlines. Also, solid waste loadings may have been too diluted to achieve detectable concentrations.
4. Any increase of nutrient (nitrate, nitrite, phosphate, and silicate) concentrations within North West Bay, attributable to nutrient loadings from the fish farm, was probably minor. The maximum rise in dissolved nitrogen levels due to fish excretion (ammonia and urea) within NWB was in the order of 3.8 μg·l$^{-1}$; however, lacking details of ambient ammonia concentrations, the influence of fish excretory products on dissolved nitrogen levels within the bay is unknown.
5. Although theoretical levels of ammonia excretion had the potential to enhance primary productivity within the immediate vicinity of the fish farm, increased phytoplankton biomass was not observed. Chlorophyll-a levels on the farm, and within the bay as a whole, were not dissimilar to those reported in adjacent waters without fish farms. Ambient nitrogen concentrations may not have been limiting for phytoplankton production, or phytoplankton production was stimulated but dispersed and ultimately flushed from the bay.

**Table 13.3.** Summary of theoretical nitrogen loadings from fish production at Aquatas fish farm from February 1995 to April 1996. (A) Total monthly nitrogen loadings. (B) Monthly equilibrium concentrations (Ec) of excreted total ammoniacal nitrogen (TAN) + urea-N. (C) Theoretical excreted TAN concentrations per cage compared with measured $NO_X$ concentrations.

| | Feb-95 | Mar-95 | Apr-95 | May-95 | Cumulative (Feb-May) | Oct-95 | Nov-95 | Dec-95 | Jan-96 | Feb-96 | Mar-96 | Apr-96 | Cumulative (Oct-Apr) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **A** | | | | | | | | | | | | | |
| Fish Biomass (kg) | 247815 | 339745 | 414198 | 118988 | | 22548 | 38000 | 86000 | 152000 | 237529 | 328000 | 244000 | |
| Fish Production (kg) | 72524 | 93786 | 83104 | 27458 | 276872 | 4169 | 15573 | 48173 | 66704 | 85848 | 91358 | 50852 | 362677 |
| Feed out (kg) | 85534 | 122999 | 120757 | 35696 | 364986 | 7922 | 28243 | 51707 | 83381 | 98907 | 111055 | 66107 | 447322 |
| Feed consumed (kg) | 81257 | 116849 | 114719 | 33912 | 346737 | 7526 | 26831 | 49122 | 79212 | 93962 | 105502 | 62802 | 424956 |
| FCR (feed out/production) | 1.18 | 1.31 | 1.45 | 1.30 | 1.32 | 1.90 | 1.81 | 1.07 | 1.25 | 1.15 | 1.22 | 1.30 | 1.23 |
| FC (feed consumed/production) | 1.12 | 1.25 | 1.38 | 1.24 | 1.25 | 1.81 | 1.72 | 1.02 | 1.19 | 1.09 | 1.15 | 1.23 | 1.17 |
| Feed out - N (kg) | 6158 | 8856 | 8695 | 2570 | 26279 | 570 | 2033 | 3723 | 6003 | 7121 | 7996 | 4760 | 32207 |
| Feed consumed - N (kg) | 5851 | 8413 | 8260 | 2442 | 24965 | 542 | 1932 | 3537 | 5703 | 6765 | 7596 | 4522 | 30597 |
| Fish - N retained (kg) | 2405 | 3211 | 2908 | 978 | 9502 | 109 | 433 | 1424 | 2069 | 2866 | 3152 | 1794 | 11846 |
| Feed waste - N (kg) | 308 | 443 | 435 | 129 | 1314 | 29 | 102 | 186 | 300 | 356 | 400 | 238 | 1610 |
| Faeces - N (kg) | 585 | 841 | 826 | 244 | 2497 | 54 | 193 | 354 | 570 | 677 | 760 | 452 | 3060 |
| Ammonia -N (kg) | 2574 | 3925 | 4073 | 1097 | 11670 | 341 | 1175 | 1583 | 2758 | 2901 | 3316 | 2048 | 14122 |
| Urea - N (kg) | 286 | 436 | 453 | 122 | 1297 | 38 | 131 | 176 | 306 | 322 | 368 | 228 | 1569 |
| Total N loss (kg) | 3753 | 5645 | 5786 | 1592 | 16777 | 462 | 1600 | 2298 | 3935 | 4255 | 4844 | 2966 | 20361 |
| | | | | | Mean | | | | | | | | Mean |
| Total N loss/Feed out-N (%) | 61 | 64 | 67 | 62 | 64 | 81 | 79 | 62 | 66 | 60 | 61 | 62 | 63 |
| kg N loss/ t fish production | 52 | 60 | 70 | 58 | 61 | 111 | 103 | 48 | 59 | 50 | 53 | 58 | 56 |
| g TAN/kg fish production | 35 | 42 | 49 | 40 | 42 | 82 | 75 | 33 | 41 | 34 | 36 | 40 | 39 |
| g TAN/kg feed consumed | 32 | 34 | 36 | 32 | 34 | 45 | 44 | 32 | 35 | 31 | 31 | 33 | 33 |
| **B** | | | | | Mean | | | | | | | | Mean |
| Daily TAN + urea-N (kg) | 102.2 | 140.7 | 150.9 | 39.3 | 108.3 | 12.2 | 43.5 | 56.7 | 98.8 | 115.1 | 118.9 | 75.9 | 74.4 |
| Ec (TAN + urea-N, ug/l) | 2.6 | 3.6 | 3.8 | 1.0 | 2.7 | 0.3 | 1.1 | 1.4 | 2.5 | 2.9 | 3.0 | 1.9 | 1.9 |
| Ambient $NO_3 + NO_2$ (ug/l) | | | | | | 5.3 | 2.2 | 2.5 | 3.3 | 3.0 | 2.5 | 13.9 | |
| Total N concentration (ug/l) | | | | | | 5.6 | 3.3 | 3.9 | 5.8 | 5.9 | 5.5 | 15.8 | |
| **C** | | | | | Mean | | | | | | | | Mean |
| Daily TAN excretion (kg) | 91.9 | 126.6 | 131.4 | 35.4 | 96.3 | 11.0 | 39.2 | 51.1 | 89.0 | 103.6 | 107.0 | 68.3 | 67.0 |
| Daily TAN excretion per cage (kg) | 4.0 | 5.5 | 5.7 | 2.4 | 4.4 | 0.6 | 2.3 | 3.0 | 5.2 | 6.1 | 6.3 | 4.6 | 4.0 |
| Theoretical excreted TAN per cage (ug/l) | 4.0 | 5.5 | 5.7 | 2.3 | | 0.6 | 2.3 | 3.0 | 5.2 | 6.1 | 5.9 | 3.6 | |
| Measured Ambient NOx-N (ug/l) | | | | | | 5.3 | 2.2 | 2.5 | 3.3 | 3.0 | 2.5 | 13.9 | |
| Theoretical Total-N (ug/l) = $NO_X$-N + TAN | | | | | | 6.0 | 4.5 | 5.5 | 8.5 | 9.1 | 8.4 | 17.4 | |
| Excreted-TAN/Total-N (%) | | | | | | 10.8 | 50.6 | 54.7 | 61.0 | 66.6 | 70.1 | 20.5 | |

**Table 13.4.** Summary of theoretical phosphorus loadings from fish production at Aquatas fish farm from February 1995 to April 1996. (A) Total monthly phosphorus loadings. (B) Theoretical excreted $PO_4$-P concentrations within each cage compared with measured ambient $PO_4$-P concentrations. (C) Monthly equilibrium concentrations (Ec) of $PO_4$-P.

| | Feb-95 | Mar-95 | Apr-95 | May-95 | Cumulative (Feb-May) | Oct-95 | Nov-95 | Dec-95 | Jan-96 | Feb-96 | Mar-96 | Apr-96 | Cumulative (Oct-Apr) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **A** | | | | | | | | | | | | | |
| Fish Biomass (kg) | 247815 | 339745 | 414198 | 118988 | | 22548 | 38000 | 86000 | 152000 | 237529 | 328000 | 244000 | |
| Fish Production (kg) | 72524 | 93786 | 83104 | 27458 | 276872 | 4169 | 15573 | 48173 | 66704 | 85848 | 91358 | 50852 | 362677 |
| Feed out (kg) | 85534 | 122999 | 120757 | 35696 | 364986 | 7922 | 28243 | 51707 | 83381 | 98907 | 111055 | 66107 | 447322 |
| Feed consumed (kg) | 81257 | 116849 | 114719 | 33912 | 346737 | 7526 | 26831 | 49122 | 79212 | 93962 | 105502 | 62802 | 424956 |
| FCR | 1.18 | 1.31 | 1.45 | 1.30 | 1.32 | 1.90 | 1.81 | 1.07 | 1.25 | 1.15 | 1.22 | 1.30 | 1.23 |
| FC | 1.12 | 1.25 | 1.38 | 1.24 | 1.25 | 1.81 | 1.72 | 1.02 | 1.19 | 1.09 | 1.15 | 1.23 | 1.17 |
| Feed out - P (kg) | 1369 | 1968 | 1932 | 571 | 5840 | 127 | 452 | 827 | 1334 | 1583 | 1777 | 1058 | 7157 |
| Feed consumed - P (kg) | 1300 | 1870 | 1836 | 543 | 5548 | 120 | 429 | 786 | 1267 | 1503 | 1688 | 1005 | 6799 |
| Fish - P retained (kg) | 326 | 422 | 374 | 124 | 1246 | 19 | 70 | 217 | 300 | 386 | 411 | 229 | 1632 |
| Feed waste - P (kg) | 68 | 98 | 97 | 29 | 292 | 6 | 23 | 41 | 67 | 79 | 89 | 53 | 358 |
| Faeces - P (kg) | 429 | 617 | 606 | 179 | 1831 | 40 | 142 | 259 | 418 | 496 | 557 | 332 | 2244 |
| Dissolved - P (kg) | 545 | 831 | 856 | 240 | 2471 | 62 | 218 | 310 | 549 | 621 | 720 | 444 | 2923 |
| Total P loss (kg) | 1042 | 1546 | 1558 | 448 | 4594 | 108 | 382 | 611 | 1034 | 1196 | 1366 | 829 | 5525 |
| | | | | | Mean | | | | | | | | Mean |
| Total P loss/ Feed out-P (%) | 76 | 79 | 81 | 78 | 79 | 85 | 84 | 74 | 78 | 76 | 77 | 78 | 77 |
| kg P loss/ t fish production | 14 | 16 | 19 | 16 | 17 | 26 | 25 | 13 | 16 | 14 | 15 | 16 | 15 |
| **B** | | | | | Mean | | | | | | | | Mean |
| Daily dissolved-P excretion (kg) | 19.5 | 26.8 | 27.6 | 7.7 | 20.4 | 2.0 | 7.3 | 10.0 | 17.7 | 22.2 | 23.2 | 14.8 | 13.9 |
| Daily dissolved-P per cage (kg) | 0.8 | 1.2 | 1.2 | 0.5 | 0.9 | 0.1 | 0.4 | 0.6 | 1.0 | 1.3 | 1.4 | 1.0 | 0.8 |
| Theoretical excreted-P concentration per cage (ug/l) | 0.8 | 1.2 | 1.2 | 0.5 | | 0.1 | 0.4 | 0.6 | 1.0 | 1.3 | 1.4 | 1.0 | |
| Measured Ambient $PO_4$-P (ug/l) | | | | | | 11.8 | 9.3 | 10.3 | 10.6 | 9.7 | 11.8 | 13.1 | |
| Excreted-P/ambient-P (%) | | | | | | 1.0 | 4.6 | 5.7 | 9.8 | 13.4 | 11.5 | 7.5 | |
| **C** | | | | | | | | | | | | | |
| Ec (ug P/l) | 0.5 | 0.7 | 0.7 | 0.2 | 0.5 | 0.1 | 0.2 | 0.3 | 0.4 | 0.6 | 0.6 | 0.4 | 0.4 |

**Table 13.5.** (A) Summary of monthly theoretical silica loadings from fish production at Aquatas fish farm from February 1995 to the end of April 1996. (B) Comparison of silicate concentrations at the fish farm and ambient silicate concentrations (monthly means at site X and IV from weekly water samples, respectively. n = approximately 8 samples per site per month)

| | Feb-95 | Mar-95 | Apr-95 | May-95 | Cumulative (Feb-May) | Oct-95 | Nov-95 | Dec-95 | Jan-96 | Feb-96 | Mar-96 | Apr-96 | Cumulative (Oct-Apr) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **A** | | | | | | | | | | | | | |
| Fish Production (kg) | 72524 | 93786 | 83104 | 27458 | 276872 | 4169 | 15573 | 48173 | 66704 | 85848 | 91358 | 50852 | 362677 |
| Feed out - Si (kg) | 94 | 136 | 133 | 39 | 403 | 9 | 31 | 57 | 92 | 109 | 123 | 73 | 494 |
| Feed consumed - Si (kg) | 90 | 129 | 127 | 37 | 383 | 8 | 30 | 54 | 87 | 104 | 116 | 69 | 469 |
| Fish - Si retained (kg) | 0.36 | 0.47 | 0.42 | 0.14 | 1.4 | 0.02 | 0.08 | 0.24 | 0.33 | 0.43 | 0.46 | 0.25 | 2 |
| Feed waste - Si (kg) | 4.72 | 6.79 | 6.67 | 1.97 | 20 | 0.44 | 1.56 | 2.85 | 4.60 | 5.46 | 6.13 | 3.65 | 25 |
| Faeces - Si (kg) | 89 | 129 | 126 | 37 | 381 | 8 | 30 | 54 | 87 | 103 | 116 | 69 | 467 |
| Total Si loss (kg) | 94 | 135 | 133 | 39 | 402 | 9 | 31 | 57 | 92 | 109 | 122 | 73 | 492 |
| | | | | | Mean | | | | | | | | Mean |
| Total Si loss / Feed out-Si (%) | 99.6 | 99.7 | 99.7 | 99.7 | 99.7 | 99.8 | 99.8 | 99.6 | 99.6 | 99.6 | 99.6 | 99.7 | 99.7 |
| kg Si loss/ t fish production | 1.3 | 1.4 | 1.6 | 1.4 | 1.4 | 2.1 | 2.0 | 1.2 | 1.4 | 1.3 | 1.3 | 1.4 | 1.5 |
| **B** | | Jun-95 | Jul-95 | Aug-95 | Sep-95 | Oct-95 | Nov-95 | Dec-95 | Jan-96 | Feb-96 | Mar-96 | Apr-96 | May-96 |
| Theoretical Fish farm $SiO_4$ (ug/l) | | 105.4 | 144.5 | 144.9 | 87.0 | 38.5 | 35.0 | 86.7 | 108.9 | 148.0 | 133.0 | 180.5 | 163.7 |
| Measured Ambient $SiO_4$ (ug/l) | | 95.9 | 146.3 | 137.9 | 76.3 | 25.5 | 25.6 | 84.9 | 103.3 | 136.4 | 81.6 | 159.7 | 107.4 |
| Fish Farm-Si/Ambient-Si (%) | | 103.4 | 100.8 | 103.8 | 119.8 | 123.9 | 132.1 | 114.7 | 103.0 | 109.8 | 127.7 | 112.3 | 132.9 |

# 14. General Summary and Discussion

## 14.1 Environmental/Water Quality Parameters

A summary of the environmental conditions monitored at each mussel longline site (I, II, III and IV) and at the centre of the Aquatas fish farm (site X) follows.

1. Water temperatures were similar among sites, displaying a typical seasonal cycle ranging from 9°C in winter to 18°C in the summer. At each sampling time, temperatures at 1m and 5 m were within 1°C.
2. Salinities were similar among sites with an overall trial mean of 34.5‰. Surface salinities at 1 m depths were significantly lower than at 5 m on a few occasions following periods of high rainfall.
3. Dissolved oxygen concentrations (DO) ranged from 6.8 to 9.2 mg $O_2 \cdot l^{-1}$, generally being lower in warmer months and higher in the cooler months. DO was significantly lower at the centre of the fish farm than at the four mussel longline sites from mid-December 1995 to May 1996. DO concentrations among the four mussel sites and the two growing depths were within 0.5 mg $O_2 \cdot l^{-1}$.
4. Current speeds ranged up to a maximum of 24.7 $cm \cdot sec^{-1}$ with 78% of recordings below 5 $cm \cdot sec^{-1}$. The predominant current directional flows were 150° and 320°.
5. Dissolved nitrogen concentrations ($NO_3$-N and $NO_2$-N) were similar among sites, ranging from negligible levels during the summer to maximum levels of 64 µg $NO_3$-$N \cdot l^{-1}$ and 8 µg $NO_2$-$N \cdot l^{-1}$ in the winter. Dissolved phosphate concentrations were similar among sites, ranging from spring-summer lows of approximately 10 µg $PO_4$-$P \cdot l^{-1}$ to 15-20 µg $PO_4$-$P \cdot l^{-1}$ during the autumn and winter months. N:P ratios ($NO_X$-N:$PO_4$-P, µg $atoms \cdot l^{-1}$) were also similar among sites, ranging from 6-8 during the winter months to less than 1 during the summer. Silicate concentrations ranged from minima of 20-50 µg $SiO_4$-$Si \cdot l^{-1}$ during spring months, 150 $\mu g \cdot l^{-1}$ during winter months, to several distinct peaks of 150-250 $\mu g \cdot l^{-1}$ during the summer and autumn period. Silicate displayed a trend of slightly increasing concentrations with increasing proximity to the fish cages during the latter three months of the trial.
6. Significant correlations of phytoplankton production (chlorophyll-a concentrations) with $NO_X$ or $PO_4$ concentrations were not indicated. A weak correlation between chlorophyll-a and silicate concentrations was significant ($r = 0.326$, $P < 0.05$).
7. Chlorophyll-a concentrations were similar among sites, ranging from negligible levels during the winter months (June - July 1995) to 5.8 $\mu g \cdot l^{-1}$ in February 1996. Short-lived spring and autumn blooms were indicated by two peaks of approximately 3.0 µg chlorophyll-$a \cdot l^{-1}$ in September 1995 and May 1996.

8. Suspended particulate concentrations (TPM, POM, PIM and non-algal POM) were similar among mussel sites but higher at the centre of the fish farm over the entire trial, including the period of winter fallowing. %POM was similar among all sites, including the centre of the fish farm, ranging from 18.3 to 51.4% (overall mean 34.7%). Following the winter fallowing period (June - September 1995), the percentage of total POM represented by non-algal POM (which would include waste fish feed and faeces) did not increase on the farm after the introduction and growout of spring smolts from September 1995 to May 1996.
9. Based on measured currents speeds and settling velocities of 13 $cm \cdot sec^{-1}$ (feed pellets) and 4 $cm \cdot sec^{-1}$ (faeces and feed fines), the distribution of solid wastes would likely be restricted to the immediate vicinity of the farm site; 90% of solid wastes would settle to the bottom within 50 m of the cages.
10. Zooplankton community structure and biomass displayed significant temporal variation; but spatial variation among sampled sites (I, III and IV) in NWB was similar (see Appendix 3).

Most of the environmental/water quality parameters were similar among all five monitored sites within North West Bay. The only factors that displayed any significant differences among sites were dissolved oxygen, silicate and suspended particulate concentrations. Reduced dissolved oxygen concentrations, observed in the summer-autumn months, were slightly but significantly lower at the centre of the fish farm compared with DO concentrations at the four mussel longline sites. This period coincides with warmer seawater temperatures, increased fish biomass, and when salmon respiration and biochemical degradation of organic sediments would be expected to be at their highest levels. The differences in DO levels between sites, although statistically significant, were minor, with site mean DO concentrations from January to May 1996 being within 1 mg $O_2 \cdot l^{-1}$ and never falling below ANZECC (1992) guidelines (< 6 mg $O_2$ $l^{-1}$), nor those considered stressful for fish (< 5-6 mg $O_2 \cdot l^{-1}$; Tarazona and Munoz, 1995). Silicate concentrations tended to be sporadically higher on the farm site during the same period. The extent to which these differences can be attributed to fish excretion and benthic flux of dissolved silicate from solid farm wastes is unknown. Although suspended particulate concentrations (SPM) were higher at the centre of the farm site, they were higher throughout the year, including the winter fallowing period. This suggests that these higher concentrations may have been due to sample contamination from bottom sediments rather than a significant increase in SPM concentrations from suspended fish feed or faecal particles. Although there is some uncertainty as to the cause of elevated farm-site SPM concentrations, these higher concentrations were restricted to the fish cage system and not observed at the two closest longline sites (I and II). Ammonia concentrations were not monitored but the release of dissolved ammonia from the fish farm would likely have been

significant. It is unknown if excreted ammonia increased ambient concentrations within the farm or North West Bay. However, there is no evidence suggesting that the release of dissolved nutrient wastes stimulated any localised primary production in the vicinity of the farm, nor within North West Bay generally. Chlorophyll-a concentrations were similar among sampling sites and with those reported from other coastal areas of southern Tasmania. Similarities in zooplankton composition and abundance among sites supports the conclusion that localised phytoplankton production near the fish farm was not enhanced. Environmental conditions were such that mussel growth was encouraged throughout most of the year, excluding a six week period from mid-November to the end of December 1995 when suspended POM concentrations were low. However, food availability appeared to be adequate during this period for maintaining basal metabolic requirements.

## 14.2 Mussel Performance

The primary objective of this study was to assess the merits of culturing mussels and salmon within a common farm lease. The potential benefits of integrated mussel-salmon culture include improved growth and condition of mussels. In past studies, improved performance of bivalves (mussels and oysters) has been associated with an enhanced food supply from fish farm particulate wastes (feed fines and faeces) and/or increased phytoplankton biomass (Wallace, 1980; Jones and Iwama, 1991; Okumus, 1993).

The following is a summary of the biological performance of mussels cultured on longlines at four sites within North West Bay from March 1995 to May 1996. Two longlines were located within the Aquatas fish farm lease area, 70 m north (site I) and 100 m south (site II) of the main fish cage unit (16 cages). Two reference sites were located 500 m (site III) and 1200 m south (site IV) of the fish cages.

1. Shell length growth was similar at all sites with no obvious seasonal variation (ie. reduced growth in winter and increased growth in summer). Although statistically different, final mean shell lengths among sites were within 2.0 mm. No differences in trial shell length increments (May 1996 mean length - March 1995 mean length) were significant among sites. Calculated $L_\infty$ differed statistically between sites I and IV, but the difference (2.5 mm) was minor. Shell length growth was similar among the two growing depths despite minor statistically significant differences for final shell length (1.6 mm difference) and $L_\infty$ (0.3 mm difference).

2. Whole live weight growth was similar among sites and depths with no significant differences in final WLW or WLW increments.
3. Variation in meat weight was similar among sites and depths. Two major spawnings, August 1995 and February 1996, were indicated by significant meat weight losses. There were no significant differences among sites or depths for final wet meat weight or ash-free dry meat weight.
4. Net biological production retained by mussels in May 1996 represented only 54% of the total production during the trial period. The remainder of the total trial production was lost as gametes during spawning. Gross biological production, eliminated biomass and net retained production were similar among sites.
5. Variation in mussel condition during the trial, reflecting the reproductive cycle rather than changes in environmental conditions (eg. food availability), was similar among sites and depths. Significant declines in condition in August 1995 and February 1996 was due to gamete losses during spawning. Although statistically significant, differences in final condition ($CI_{grav}$) among sites were minor (within 17‰).
6. Reproductive development, spawning, fecundity, reproductive effort and mantle glycogen concentrations were all similar among sites and depths. Although two major spawnings were identified in August 1995 and February 1996, individuals in a spawning condition were also observed throughout most of the trial period.
7. The density of mussels within socks varied among sites and depths. However, there is no evidence suggesting higher densities negatively influenced mussel growth or survival.
8. Survival was similar among sites and depths (mean 81% at the end of the trial).
9. Incidence of pea crab infestation increased during the trial period, with significant differences among sites. The highest percentage of infested mussels was observed in February 1996; 45% of mussels at site IV harboured pea crabs compared with only 13% at site I. However, there was no indication of any negative effects of pea crab infestation on mussel growth, condition or survival.
10. Although not entirely conclusive, stable isotope analysis ($\delta^{13}C$ and $\delta^{15}N$) of mussel mantle tissues suggests that a significant portion of waste fish feed particles or faeces was not assimilated by mussels grown closest to the fish cages (site I). Sex differences in mantle $\delta^{13}C$ values were likely due to higher lipid content in females.
11. Temporal variation of monitored environmental parameters was not reflected by coincident variation in mussel growth or condition.
12. Theoretical scope for growth (SFG) was similar among mussel longline sites; reflecting the similar environmental conditions among sites, particularly temperature and the available food supply. However, actual mussel growth rarely matched calculated SFG predictions. This discrepancy was likely due to literature-derived physiological response parameters (clearance rate, absorption efficiency, respiration

rate) used in SFG calculations rather than parameters directly measured. Theoretical SFG suggests that mussel growth would not be enhanced at the centre of the fish farm. Despite higher suspended particulate levels within the fish cage system, ambient SPM concentrations consistently exceed the psuedofaces threshold and, therefore, limit the ability of mussels to ingest additional food particles.

Mussels in this study did not display any obvious seasonal variation in growth (ie. a pronounced reduction in growth during winter relative to spring and summer growth), as has been demonstrated by populations cultured in more temperate and boreal waters. Therefore, environmental conditions within North West Bay, particularly temperature, salinity and food supply, were adequate to promote growth throughout the year.

The overall performance of mussels cultured within the Aquatas fish farm (sites I and II) was not dissimilar to mussels grown at the two reference sites (III and IV). None of the monitored biological parameters (shell, live weight, meat weight growth, production, condition, survival, reproductive development, fecundity) were indicative of an enhanced food supply for mussels cultured within the fish farm.

The lack of any significant enhancement of available food and, therefore, growth and condition of mussels cultured at sites I and II may be attributed to several factors. Firstly, the locations of the two fish farm longlines may have been too far removed from the fish cages. At distances of 70 m and 100 m, most of the particulate fish farm wastes (fish feed fines and faeces) released from the fish cages might have settled to the bottom before reaching these two longlines. Secondly, suspended particulate wastes from the fish farm, although substantial, were likely too dispersed and diluted in the water column to significantly increase POM concentrations above background concentrations. Theoretical waste loadings from the farm suggest a maximum possible increase in POM of 0.03 $mg \cdot l^{-1}$ (< 2.0% of background concentrations), which is below the level of detection. Thirdly, enhanced phytoplankton productivity, a possible consequence of dissolved nutrient waste loading from fish excretion and benthic flux from farm sediments, was not observed at the two fish farm sites. Phytoplankton concentrations within NWB were similar to those reported from other areas in southern Tasmania. Primary production may not have been stimulated by the levels of dissolved nutrient wastes, or if so, was dispersed within the bay and masked by the greater proportion of ambient phytoplankton.

## 14.3 Implications for integrated mussel-salmon culture

The results from this study provide little incentive for the integration of mussel culture with the Aquatas salmon farm in North West Bay. The two most pertinent objectives of integration, increased mussel growth and reduced particulate loadings, are unlikely to be achieved. Mussel growth was not significantly enhanced at the farm during this study, nor would improved growth be likely even if longlines were positioned immediately next to the fish cages. Although in a better position to intercept suspended fish wastes than at sites I and II, waste particulate loadings are too diluted to significantly increase the quantity or quality of food available for mussels above ambient levels. This conclusion is supported by SFG estimates; suggesting mussel growth would not be significantly enhanced at the centre of the fish farm over growth at other sites in North West Bay.

It could be argued that this fish farm might be inappropriate for the integration of mussels. As this farm is utilised primarily as a smolt-rearing site, particulate waste loadings would be lower than at grow-out farms with higher fish biomass, feed delivery and faecal output. However, in addition to the quantity of particulate waste loadings that might be exploited by mussels, ambient particulate concentrations are an important factor in determining the suitability of bivalve integration with a fish farm. In North West Bay, ambient particulate conditions largely exceed the pseudofaeces threshold concentration (T, approximately 5.0 mg TPM·$l^{-1}$) throughout the year. At these concentrations, mussel physiology limits the ability to fully exploit additional fish farm waste particles. Above T, a significant portion of filtered fish waste particles is voided as pseudofaeces rather than ingested and incorporated into mussel tissue. As a consequence, mussel growth within a fish farm would not be expected to be any different than at other coastal areas experiencing similar environmental conditions. Although not monitored, ambient particulate concentrations at the Aquatas growout farms, located in the D'Entrecasteaux Channel adjoining NWB, would likely be similar to those recorded in this study. Therefore, mussel growth may not be significantly enhanced at these farm sites unless food quality (eg. %POM, energy content, digestibility) is substantially superior to alternative culture areas in the region.

What about some of the ecological benefits of integration, proposed by Folke and Kautsky, such as reducing fish farm solid waste loadings and their impacts on the benthic environment? With low current speeds relative to feed and faecal sedimentation rates and multi-directional current flow, mussel lines would need to be suspended very close to and surrounding the fish cages, with socks extending much deeper than 5 m to effectively intercept a significant portion of suspended wastes. The necessary scale and proximity of mussel lines to fish cages may not be acceptable to

the practical management of this farm. Mussel lines may interfere with the normal day to day activities. Perhaps raft culture would be a more flexible culture system, allowing mussels to be moved about the farm as required. However, it is unlikely that a significant reduction in particulate wastes could be achieved at the NWB farm regardless of the culture method. As ambient particulate levels consistently exceed the pseudofaeces threshold concentration within North West Bay, a significant fraction of filtered wastes would be voided as pseudofaeces. The ambient temperature and particulate concentrations within North West Bay, together with the filtration, ingestion, and absorption rates presented in Section 12.2, indicate that mussel culture might actually increase rather than decrease the particulate loading within the farm site. Biodeposition (faecal and pseudofaecal POM) of both filtered ambient seston and fish farm wastes would exceed the quantity of fish farm wastes ingested and assimilated into harvestable mussel tissue. Additionally, mussels dropping off of socks would also contribute to the organic loading of the sea bed. Therefore, mussel culture might just redistribute or even extend the areal distribution of organic sedimentation around the farm. Organic loading from suspended mussel culture has similar negative impacts on the benthic environment as do fish farm wastes (Dahlback and Gunnarsson, 1981; Mattsson and Linden, 1983; Kaspar et al., 1985; Gowen et al., 1988; Folke and Kautsky, 1989; Burnell and Cross, 1995). Integrated mussel culture at the Aquatas farm might exacerbate the organic loading already occurring beneath the fish cages; increasing the potential for methane and hydrogen sulphide outgassing and detrimental effects to both salmon and mussels.

Folke and Kautsky (1989) have suggested that mussels grown on fish farms would benefit by feeding on increased phytoplankton production stimulated by fish farm nutrient loadings. In so doing, mussels counter this increased production and reduce the ecological impact of fish farms. Although intuitively reasonable, the assumptions of (i) increased primary productivity due to fish farm nutrient waste loadings actually occurring, and (ii) increased phytoplankton biomass being available for mussels to consume, are both questionable. Firstly, a key phrase that is frequently used by Folke and Kautsky and others reviewing the environmental impacts of fish farming is "the potential of fish farm nutrient loadings to increase localised phytoplankton production and eutrophication effects". However, actual increases in primary production leading to localised eutrophication are rarely documented in the literature, and were not observed in this study. Why not? As previously mentioned, ambient nutrient levels within a water body may not be at limiting concentrations, or dissolved nutrient output from farms may be too low or diluted to stimulate increased production. Secondly, if production is stimulated, phytoplankton is probably well dispersed by water currents, become undetectable from background levels, and is ultimately flushed from the bay.

Therefore, any phytoplankton production is likely to be either unavailable or too low to enhance growth of mussels cultured within a fish farm.

Many reviews of the impact of salmon farming on increased phytoplankton production refer to the study by Jones and Iwama (1991) who reported high phytoplankton concentrations associated with a British Columbia salmon farm. Their results imply that phytoplankton production within the farm, and particularly within the fish cages, was stimulated by fish waste nutrients. It is noteworthy that chlorophyll concentrations inside fish cages were up to three times greater than outside of the cages. The reasons for this apparent discrepancy can only be speculated upon as fish feed delivery, feed digestibilty, water exchange rates and nutrient conditions are not detailed. The farm may have been located in stagnant water, thereby restricting the horizontal transport of phytoplankton out of the fish cages, but this possibility seems unlikely. Possibly their methodology did not correct for chlorophyll-a degradation products, such as phaeopigments retained within fish feed or faeces, resulting in higher "chlorophyll" concentrations inside the fish cages. If this was the case, chlorophyll levels, attributed to phytoplankton biomass, may not have been so different among sites.

As most salmon farms are located in areas of high water exchange rates, it seems reasonable that dissolved waste nutrients and any phytoplankton production stimulated by these nutrients would be well dispersed beyond the bounds of a fish cage and farm lease. Therefore, the removal of waste dissolved nutrients, by way of mussels filtering and ingesting phytoplankton stimulated by these wastes, is largely dependent on ambient nutrient levels and the hydrography of specific sites. It is highly unlikely that any phytoplankton production stimulated by excretory wastes would remain resident within most marine farms long enough to be consumed by integrated mussels. The only nutritional benefit that mussels are likely to obtain from excretory wastes is from increased phytoplankton biomass within a localised area, such as the bay in which the farm is located. However, this benefit could be obtained by culturing mussels anywhere within the local area - not necessarily on the fish farm. Caution is recommended if increased phytoplankton production, either within a fish farm or within a bay generally, is the sole basis for integrating mussels onto a fish farm.

Perhaps a more realistic viewpoint of the ecological benefits of mussel culture in reducing primary production stimulated by fish farm wastes is one with a wider, more regional perspective. Let's assume that phytoplankton production is stimulated by excreted fish farm nutrients, but this production is well dispersed and eventually flushed from the bay. Firstly, the ecological impacts, if any, from this production are uncertain. Secondly, it would be near impossible to attribute increased production

directly to fish farm waste loadings. Other agricultural, domestic, industrial and natural nutrient inputs can be significant. Perhaps integrated mussels, although not directly benefiting from any enhanced phytoplankton abundance within the fish farm, might still be ecologically beneficial in reducing phytoplankton production, regardless of the sources of nutrient enrichment. Mussels would still remove some phytoplankton transported through the farm, thereby offsetting to a certain extent any production stimulated by fish farm wastes and/or other anthropogenic nutrient inputs. In order to achieve negligible net phytoplankton production, the production stimulated by waste nutrient inputs and the scale of mussel culture required to remove an equivalent amount of phytoplankton would need to be evaluated. However, mussels need not be cultured immediately next to the fish cages - they could be grown anywhere in the local area to achieve the desired reduction in primary production. So again, there is no particular advantage for integration of mussels onto fish farms.

The potential of mussel culture actually reducing phytoplankton production may be questioned. It has been suggested that mussel culture may promote, rather than limit, phytoplankton production. Asmus and Asmus (1991) suggest that algal blooms caused by anthropogenic sources (which would include fish farms) are unlikely to be combated by the introduction of mussels. As with fish farms, mussel culture is also responsible for releasing large amounts of dissolved nutrients (ammonia, phosphate and silicate) through excretion and benthic flux from sedimented biodeposits. Primary production stimulated by the nutrients released from mussel cultures may actually exceed phytoplankton removal under certain environmental conditions (Tenore et al., 1985; Asmus and Asmus, 1991; Gibbs et al., 1992). Conditions that might stimulate phytoplankton production are high seston concentrations with a high non-algal POM component, and growth-limiting dissolved nitrogen concentrations. Mussel culture would have the potential to stimulate phytoplankton growth beacuse (a) mussels feed upon and remineralise other material (eg. detritus, zooplankton, dissolved organic nitrogen) other than phytoplankton alone, and (b) due to the digestive processes of mussels, nutrients in consumed phytoplankton are recycled back into the water column more rapidly than natural processes of phytoplankton sedimentation and decomposition. As a result, the nutrient loading from a mussel culture system may exceed the removal of nutrients in ingested phytoplankton.

It could be fairly argued that mussel culture, unlike salmon culture, does not introduce new nutrients into a system. Therefore, the harvesting of mussels does result in a net removal of nutrients from coastal waters. However, it is also clear that mussel culture, similar to salmon culture, has the potential to concentrate particulate and dissolved nutrients that otherwise might be distributed over a much wider area. Therefore, combined with the dissolved nutrient loadings from caged salmon, the potential of an

integrated mussel-salmon farm for (i) promoting rather than limiting primary production, and/or (ii) significantly altering the nutrient status (ie. N:P:Si ratio) of the receiving water body, possibly promoting the growth or toxicity of harmful algal species, might be increased. Either of these consequences of mussel culture seems plausible in NWB. As phytoplankton represents such a minor component of the available POM (< 12% of total POM), nutrients released from a mussel culture system might exceed the uptake and assimilation of phytoplankton-bound nutrients.

Contrary to the above argument, it has been suggested that mussel culture may help to alleviate altered dissolved N:P ratios in the vicinity of salmon farms (Troell and Norberg, 1998). However, this potential benefit of integrated bivalve-salmon culture would require a thorough investigation of the total nutrient loadings of a fish farm together with the net nutrient loadings (balance between nutrient uptake and nutrient release through excretion and remineralisation from biodeposits) from a suspended mussel culture. Site-specific parameters requiring quantification include: dissolved N and P output from a farm (both from excretion and benthic flux), retention of particulate-bound N and P by mussels (which would depend on the scale of culture, particulate concentrations, algal and non-algal concentrations, feeding and assimilation rates), mussel N and P excretion rates, remineralisation rates of N and P from mussel biodeposits and flushing time of the bay. However, assuming that mussel culture might aid in reducing altered N:P ratios, and with the general conclusion that mussel culture has a limited capacity of reducing the particulate wastes released from fish cages, this benefit might be achieved by culturing mussels anywhere within a bay where fish farms are located – not necessarily cultured within a fish farm.

The release of dissolved nutrients, from either a salmon monoculture or integrated salmon-mussel culture, and their potential for increasing phytoplankton production presents a case for integrating seaweeds into a culture system. Seaweeds act as a sink for removing a portion of these dissolved nutrient wastes. Again the scale of seaweed culture required to remove the desired quantity of dissolved nutrient wastes from salmon and/or mussel culture would need to be evaluated.

Although dissolved nutrients released from fish farms can be quantified, the actual impact of these wastes on the environment, such as stimulating phytoplankton production, is not so well defined. Solid organic waste loadings are a more tangible commodity and their effects on the environment, particularly the benthos, are more defined and measurable. We have discussed the limitations of integrated mussel culture within the Aquatas fish farm in removing solid organic wastes emanating from the fish cages. However, mussels would filter particulates from the water column regardless of their source. Perhaps mussels could be environmentally beneficial.

They could be cultured to counterbalance fish farm organic waste loadings by filtering and assimilating an equivalent amount of organic matter, most of which would be natural sestonic particles. Therefore, the net organic loading from a mussel-salmon duoculture would be negligible. Intuitively, this suggestion seems reasonable. However, what scale of culture would be required and what might be the consequences?

An intensive mussel cultivation system with the potential to absorb 321 kg POM·day-1 was modelled (Appendix 4). This quantity of POM is equivalent to the mean daily solid organic waste loadings from the Aquatas fish farm (Section 13.2). The model is based on the carrying capacity model of Inzce et al. (1981) using mean current speed, temperature and seston concentrations of NWB and the feeding rates of mussels (Page and Ricard, 1990). Using 70 mm mussels at stocking densities of 200,000 mussels·100 m longline$^{-1}$, 3.8 km of longlines (7.6 million mussels) are required to absorb an equivalent amount of POM as the daily fish farm waste loadings. Clearly, this scale of culture would be too extensive to incorporate into the Aquatas lease. Secondly, with natural seston in NWB exceeding the pseudofaeces threshold concentration (mean 6.42 mg·l$^{-1}$), the organic waste loadings from this mussel system would be nearly twice those of the fish farm. Although not adding new organic nutrients into the environment (as does the fish farm), mussel culture would concentrate existing organic particles, deposit them on the sea bed, and exacerbate the benthic loading and environmental impacts from the fish cages. Therefore, it is unlikely that this scale of culture within the farm would be acceptable. The desired effect, counterbalancing the organic loading of the farm, could be achieved by a mussel culture system located anywhere - not necessarily, nor desirably, within the Aquatas lease. Again we have no incentive for integrating mussels with salmon in NWB.

The Folke and Kautsky model of integrated culture also suggests there are ecological and economic benefits from recycling mussel meat back into fish feed, reducing the dependence on pelagic fish stocks for fishmeal production. The energy required for pelagic fish production, capture, transport and processing, and any environmental or economic impacts arising from depleted prey-fish stocks, could be reduced (Folke and Kautsky, 1991; 1992). However, the efficiency of resource utilisation and subsequent ecological/economic benefits of integrated culture depends upon (i) the quantity of waste fish feed/faeces actually incorporated into farm-grown mussels, and (ii) the quantity of mussel meat recycled back into the fish feed. In the case of the Aquatas fish farm, it appears unlikely that mussels could assimilate a significant amount of fish feed/faeces (maximum of 2% of waste POM). Therefore, the quantity of nutrients recycled within this integrated system would be negligible. Still, replacing fishmeal

with mussel meat would be more ecologically and energetically efficient. Mussels are produced at a lower trophic level and require less energy inputs for their production, harvesting, transport and processing than pelagic fish stocks. Incorporating cultured mussel meat into fish feed would reduce the fishing pressure on pelagic fish as well as reduce any detrimental ecological and economic impacts arising from diminished fish stocks. The feasibility of replacing fishmeal with mussel meat would certainly require evaluation by fish nutritionists and feed technologists. However, assuming mussel meal is a viable alternative to fishmeal, some of the ecological and economic benefits proposed by Folke and Kautsky could be achieved by culturing mussels anywhere - not necessarily integrated with a salmon farm. With little evidence that mussels could remove a significant portion of farm wastes in NWB, there is no strong case for integrated culture.

How realistic is the notion of replacing fishmeal with mussel meat? The quantity of feed used at the Aquatas farm during 1995-96 was approximately 414 tonnes (dry weight). Assuming a fishmeal content of 35%, 145 tonnes of dried mussel meat would be required to totally replace the fishmeal in feed. Using the dry meat weight of mussels in peak condition in February 1996 (3.36 g DMW·mussel$^{-1}$) and corresponding mussel densities (approximately 135 mussels per meter of sock or 22,680 mussels per 20 m longline), fishmeal replacement would require the total mussel harvest from approximately 38 km of cultured longlines! Clearly, the notion of fishmeal replacement by integrated mussel culture is unrealistic.

From a theoretical perspective, this discussion illustrates that many of the ecological benefits of integrated mussel-salmon culture, as suggested by Folke and Kautsky, may not be realised. Rather than ameliorating some of the negative impacts of salmon farms on the environment, by reducing particulate loadings and phytoplankton production, integrated mussel-salmon culture has the potential to actually exacerbate these impacts.

Certainly, integration would increase the productivity of the lease site by providing another marketable product. Capital costs for longlines or rafts would not be exorbitant and mussel seed could be partially obtained from natural settlements on the fish cages. Any maintenance, re-socking and harvesting tasks could be undertaken by existing farm staff. With a current market price in Australia of AUS$3.00 - $3.50 per kilogram live weight, the potential revenue from mussels would be in the order of $240 - $280 per meter of longline (stocking density of 800 mussels·4 m sock$^{-1}$ and harvest weight of 50 g·mussel$^{-1}$). However, if heavily-fouled, mussels may have little or no value. At the conclusion of this growth trial, mussel socks were so fouled with overspat and algae that mussels were disposed, rather than harvested, cleaned and

sold. Capital expenditure for appropriate processing equipment may be required. Therefore, any decision to integrate mussel culture requires an evaluation of the potential increase in revenue and ecological/economic advantages weighed against expenditures and the potential disadvantages of integration.

We have seen that there may be limited ecological advantages (reduced particulate loads, reduced phytoplankton production, minimal nutrient recycling) in culturing mussels in NWB. What are some of the disadvantages of integrating mussels with open-water salmon culture? We have already discussed the potential of mussel culture to exacerbate existing waste loadings under high natural seston concentrations. These and other disadvantages of integrated bivalve-fish culture integration have largely been ignored in the literature, with most reviewers accentuating the positive aspects. Ready access to fish cages might be a problem, but a mussel culture system could be appropriately designed to minimise disruptions to fish farming activities. However, there are more important concerns regarding integrated culture that need to be addressed. Particularly, how would an extensive mussel culture system affect the growth and survival of salmon? Suspended mussel culture might possibly alter the water flow through a farm - increasing sedimentation, reducing the supply of oxygenated water, and restricting dissolved and solid waste removal. Oxygen demands of cultured mussels themselves may reduce available DO for salmon, particularly in the warm summer months when DO levels can approach stressful levels. Mussels also represent a potential reservoir of bacterial and viral finfish pathogens that could be transmitted to caged fish (Meyers, 1983). Although bacterial and viral diseases are not of major significance in Tasmania at present, they may be in the future and are certainly of concern elsewhere. The accumulation of chemical/heavy metal residues in bivalve tissues, as a consequence of therapeutic treatments and/or where antifoulants (eg. TBT) are in use, creates a public health and safety issue that would need to be addressed. What about consumer acceptance and marketability of fish farm-grown mussels? The public may not readily adopt a product that feeds on fish faeces and/or exposed to therapeutics. All of these issues need to be factored into any decision to incorporate mussels onto a fish farm - in the case of Aquatas, a farm that appears to have no major impact on the water quality of North West Bay and where benthic impacts are likely restricted to within the lease area. As mussel culture at this site has limited potential for enhancing growth and reducing solid waste loadings from the fish cages, integrating mussels on a large-scale has little to offer Aquatas.

After reviewing the growing number of practical and theoretical investigations of integrated bivalve-salmon culture, Folke and Kautsky have recognised some of its limitations (Troell et al., 1999). They also agree that increased growth of mussels,

accompanied by a reduction in fish farm particulate wastes, may not always be achieved. The suitability of cultivating mussels with salmon is largely dependent upon local environmental conditions, farming practices and the levels of waste generation. Clearly, the flow-through and open characteristics of sea-cage salmon farming results in solid wastes becoming highly diluted and dispersed throughout the receiving water body. As a result, solid wastes may not increase available POM concentrations to levels that significantly improve mussel growth. However, some farms may generate wastes that do increase POM concentrations. Further, it may be argued that the sampling regime in this study, two samples taken one day per week, may have failed to adequately detect peaks in POM concentrations resulting from faecal wastes discharged in pulses. Despite these possible increases, and depending on ambient particulate loads, mussels may not always be able to fully exploit additional food. For example, Troell and Norberg (1998) suggest feeding fish in pulses may significantly increase short-term SPM concentrations within a farm; however, the pseudofaeces threshold limits the particle retention from a rich pulse. The short duration of a pulse and saturation of mussel feeding makes the long-term ambient seston concentration more important for mussel growth than fish farm wastes (Troell and Norberg, 1998). Similarly, the present investigation has demonstrated that extended periods of ambient seston concentrations exceeding T also limit the ability of mussels to fully exploit fish farm waste particles. Therefore, integrated suspended mussel-salmon culture may only significantly improve mussel growth and reduce solid waste loadings on farms that satisfy a number of conditions:

- coastal regions that experience extended periods of low natural POM concentrations (ie. below the pseudofaeces threshold),
- waste feed fragments and faecal particles are of a filterable size,
- suspended particulate wastes significantly increase suspended POM above ambient levels, and
- mussels are appropriately positioned to intercept a significant fraction of particulate wastes emanating from the fish cages.

It would be expected that new developments in feed technology, improved feed digestibility, and feeding practices, will reduce solid waste loadings from fish farms, further limiting additional feed for mussels and the viability of integrated mussel-salmon culture. Dependent upon local environmental conditions, increased phytoplankton biomass is unlikely to be characteristic of most salmon farms. Any increase in phytoplankton biomass stimulated by nutrient wastes would be dispersed well beyond the bounds of a farm lease. Therefore, a farmer considering the inclusion of mussels onto his fish farm should not rely on increased phytoplankton production within his lease to enhance mussel growth. Mussels could be grown anywhere within

the local area to counter any phytoplankton production that may be stimulated. Culturing mussels distant to the fish farm would alleviate many of the concerns associated with integrated culture. These concerns include access to fish cages, altered water flow, reduced oxygen levels, increased particulate loading, therapeutic or heavy metal residues in bivalves, and the transmission of viral and bacterial pathogens to caged fish.

Newkirk (1996) and Brzeski and Newkirk (1997) have suggested extending the Folke and Kautsky model to a more complex multiple-species system. They have proposed incorporating benthic species of fish and/or invertebrates to feed upon some of the solid waste loadings generated by the bivalve-fish duoculture. Examples of possible benthic species that might be appropriate for integration onto a farm include flounder, sea cucumbers, sea urchins, bait worms, clams and lobsters. The selected species that would utilise the food falling to the benthos may, or may not, be confined to cages. The link to the benthic food chain is one step in what may be a further elaboration of the Folke and Kautsky model to an integrated Sustainable Coastal Production System (SCPS). Such a system might also include wild fish, seaweeds and various grazing species of fish and invertebrates to help control biofouling on fish cages and mussel socks (Newkirk, 1996). Although Newkirk's SCPS model is interesting, the viability of such an elaborate system would certainly require investigation and recognise some of the limitations of integrated culture that have been addressed in this discussion.

In view of some of the limitations and potential disadvantages of large scale bivalve-fish culture, it is my opinion that in many instances, largely indiscriminate filter-feeding bivalves, such as suspended mussels and oysters, may not be appropriate species for integration with salmon farms. Particulate wastes are often too diluted to enhance suspended food levels and are scattered in all directions according to the vagaries of water current and turbulence. With the majority of their diet coming from natural seston rather than farm wastes, suspended bivalves would be adding to the particulate loading of a fish farm. Perhaps a more effective biological control of fish farm solid wastes would be benthic detritivores, feeding more exclusively on farm wastes and cultured where these wastes are most available and of most concern - on the sea bed underneath the fish cages. An advantage of incorporating benthic detritivores is that they would be able to utilise larger waste particles such as sedimented feed and faecal pellets which are not directly available to suspended filter feeders. If the detritivores were free-range, they could move to where food was most plentiful. Faecal output from detritivores would come mostly from consumed fish farm wastes rather than natural food. Consequently, detritivores would not be adding to the fish farm waste loadings, as would filter feeders, and solid waste loadings

would be reduced. The cultured species, scale and design of such a system, and the efficiency of reducing solid waste loadings, would of course require investigation.

Although this discussion has largely focussed on the negative aspects of integrated mussel-salmon culture, the author does not intend to be overly critical of the Folke and Kautsky model. Nor are other integrated culture models, regardless of their complexity, totally without merit. However, for the past decade the potential benefits of integrated bivalve-fish culture in coastal waters may have been significantly overestimated. As some practical investigations, including this one, have failed to demonstrate significantly improved bivalve performance, some of the limitations of incorporating indiscriminate filter-feeding bivalves onto salmon farms need to be highlighted. The existing model of integrated bivalve-fish culture is unlikely to be a highly-effective method of addressing waste loadings from open-water sea cage fish culture, as some have suggested. Certainly, bivalve growth may be enhanced and fish farm wastes reduced in some circumstances. However, these circumstances may be limited to more temperate and boreal climates with extended periods of low natural POM levels, as is frequently the situation during the winter months.

Rearing salmon in impermeable plastic bags (SEA System™, FutureSEA Systems, Inc., Nanaimo, Canada) that contain and concentrate solid wastes is currently being trialed by Aquatas and some farms overseas. Should this technology prove to be a viable alternative to cage culture, it would likely be a more effective method of managing solid fish wastes than mussel cultivation. However, bag culture might also provide more potential for integrated culture than sea-cage culture, as waste materials would be more easily contained and controlled. An integrated system could be designed similar to those of land-based systems. Effluent from fish bags, with high concentrations of particulate and dissolved nutrient wastes, could be directed to a series of "treatment bags" that contain suspended bivalves and/or seaweeds. Solids would be filtered by the bivalves and dissolved nutrients absorbed by the seaweeds. Remaining solid wastes from treatment bags could be then removed and composted as they are at present. Such a system would enhance the opportunities for reducing solid and nutrient emissions from salmon farms while providing a supplementary income. However, the potential benefits of such a system relative to the capital expenditure and running costs would require evaluation.

Whether or not effective biological solutions, which address the concerns associated with open-water fish farm wastes, can be developed is uncertain. As waste dispersion from sea cages is so difficult to control, perhaps technical solutions to fish farm waste management may be more effective. The development of highly digestible, low polluting diets (possibly incorporating cultured bivalve meat rather than fishmeal), continued improvements in feeding techniques and technologies, impermeable bag culture, and/or a major shift to off-shore cage farming, may further limit the applicability of large-scale biological waste control systems, such as integrated bivalve-fish culture, in the future.

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# 16. Appendices

## Appendix 1. Gonad Staging

Criteria used for staging gonads in Section 7 were modified from Dinamani (1974). Each stage was also assigned a numerical factor (NF) according to Seed (1969a).

**I. Indeterminate.** Indeterminate sexual phase. Follicles generally contracted. Any germ cells present are not sexually differentiated. Phagocytes characteristically present. NF = 0.

**D1. Developing 1.** In the male, all stages of spermatogenesis are present. However the vast majority of germ cells consist of spermatogonia, with decreasing numbers of primary and secondary spermatocytes. Females are characterised by the presence of oogonia, and primary oocytes. A small number of secondary oocytes attached to follicle wall by an elongated peduncle will also be present. NF = 1.

**D2. Developing 2.** Gonad area as a percentage of total cross sectional area is increased substantially from D1. In the male all stages of spermatogenesis are present. Primary and secondary spermatocytes form a loose band at the periphery of the follicle with the central follicle area consisting primarily of spermatids and some spermatozoa. In females, gametes within the follicle primarily consist of advanced secondary oocytes attached to the follicle wall and some ova lying free in the lumen. NF = 2.

**A. Advanced.** Gonad occupies a relatively large proportion of overall cross sectional area. Male follicles are densely packed with spermatozoa, with some spermatids and secondary spermatocytes at the periphery. The females consist primarily of large numbers of mature ova free in the lumen, however some secondary oocytes attached to the follicles may still be present. NF = 3.

**PS. Post Spawned.** Spawning, partial or complete, has occurred. Follicles tend to have a disorganised appearance with some showing evidence of rupture. Small numbers of phagocytes often present. Developing germ cells will still be present lining the follicles. NF = 2.

**R. Regressive.** This stage is characterised by large numbers of phagocytes. Discharged follicles have collapsed. Female follicles which have not discharged are characterised by gametes with a granular/porous texture indicative of deterioration. In males, follicles will generally only contain residual spermatozoa. NF = 1.

# Appendix 2. Tissue Glycogen Analysis

The method used for analysis of soft tissue glycogen content was modified from the method of Keppler and Decker (1974). Modifications included the use of a lower amyloglucosidase (AMP) concentration (0.05 mg/ml instead of 10 mg/ml), a shorter glycogen hydrolysis time (30 minutes instead of 2 hours), NAD instead of NADP and automation of the glucose spectrophotometric assay procedure with a Technicon RA-1000 autoanalyser..

## Sample Preparation

Dried meats from each sampling period were stored at -20°C until the conclusion of the trial. Dried meats from each site were pooled according to growing depth, split into two samples per depth and finely ground with a geological rock-crushing mill for approximately twenty seconds.

## Extraction of glycogen from mussel tissue

Two gram subsamples of dried ground mussel tissue was rehydrated with 8 ml of distilled water and one drop of Triton X 100 wetting agent. This mixture was homogenised in a Waring™ commercial blender with 50 ml of 0.6M perchloric acid ($HClO_4$) for three minutes.

## Hydrolysis of glycogen

Homogenate (0.2 ml) was pipetted in duplicate to assay tubes, followed by 0.1 ml potassium bicarbonate ($KHCO_3$), 2 ml of amyloglucosidase reagent (0.05 mg AMP·$ml^{-1}$) and incubated in a water bath for thirty minutes at 40°C with occasional agitiation. Glycogen present in the sample was converted to glucose with amyloglucosidase. The reaction was stopped by addition of 1 ml of 0.6M perchloric acid. Blank samples of 0.2 ml homogenate were prepared as for test samples, replacing the AMG reagent with 2 ml of acetate buffer. Samples were then centrifuged for fifteen minutes at 3000 rpm. Approximately 1 ml of the resulting supernatant was decanted from the centrifuge tubes and analysed spectrophotometrically using a Technicon RA - 1000 autoanalyser. Blanks were also assayed to provide a measure of glucose already present in the samples.

## Spectrophotometric assay for glucose

The RA-1000 is an automated spectrophotometer where a hexokinase reagent (hexokinase + G6PDH + ATP + NAD + buffer) is added to a cuvette and the absorbance measured at 340 nm. The test sample is then added at the ratio of 7 µl sample to 350 µl hexokinase reagent and incubated at 37°C for 5 minutes. Glucose present is phosphorylated with ATP in a reaction catalysed by hexokinase. The phosphorylated glucose is then oxidised to 6-phosphogluconate in the presence of NAD. The resultant reduction of NAD to NADH is measured spectrophotometrically. NAD does not absorb at 340 nm whereas NADH does and so the increase in absorbance is proportional to the concentration of glucose in the sample. The change in sample absorbance is compared to the change in absorbance measured for a glucose standard (50 mg glucose/100 ml). The results were then printed out as mg glucose/100 ml homogenate as calculated by:

$$[\text{Sample glucose}]\ (\text{mg/dL}) = \frac{\text{change in absorbance of sample}}{\text{change in absorbance of standard}} \times [\text{Standard}](50\text{mg/dL})$$

Test samples were then corrected for glucose concentrations of the blanks. Tissue glycogen concentrations (g glycogen/100 g dry tissue weight) were calculated as:

$$[\text{Glycogen}]\ (\text{g/100 g dry wt}) = [\text{Glucose}]\ (\text{mg/dL}) * 0.146 * (\text{dw} * 0.03) * (100/\text{dw})$$

where 0.146 = conversion factor (mg glucose/dL to mg glycogen/ml)
dw = dry weight of ground mussel used

(dw*0.03) converts [glycogen] (mg/ml) to [glycogen] (g/dw)
(100/dw) converts [glycogen] (g/dw) to [glycogen] (g/100g dry weight)

## Reference

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# Appendix 3. Zooplankton Composition and Abundance

## Introduction

Concurrent with the monitoring of physical and chemical environmental parameters within North West Bay (NWB), a zooplankton monitoring program was undertaken. The seasonal variation in zooplankton species composition and abundance was of interest to the fish farm management team (Aquatas Pty Ltd), to complement its ongoing water quality and phytoplankton monitoring program, as well as to the present investigation. This monitoring program might determine if the fish farm had any effect on zooplankton abundance or community structure. Through the addition of dissolved waste nutrients (ammonia, phosphate, silicate), fish farms have been implicated in stimulating localised phytoplankton production (Jones and Iwama, 1991). Should phytoplankton production be stimulated in the vicinity of the Aquatas fish farm, an indirect influence on the zooplankton community was considered a possibility. Therefore, this monitoring program was undertaken to assess zooplankton community structure, dominant species, and zooplankton biomass at several sites in the vicinity of the Aquatas fish farm.

## Methodology

Three locations in North West Bay were sampled for zooplankton at approximately fortnightly intervals from 22 June 1995 to 21 May 1996. These sampling locations were adjacent to three experimental mussel longline sites (sites I, III and IV). Oblique water column samples were collected by hauling in a weighted 300$\mu$m conical zooplankton net (300 mm diameter mouth x 1 m length) from 20 m depth while being towed behind a small skiff at low speed. Tows followed a circular path adjacent to each of the mussel longlines and were of 3 - 4 minutes duration (timed with a stopwatch). Zooplankton was concentrated in a 50 ml cod-end bucket and immediately preserved in a 4% hexamine-buffered formaldehyde-seawater solution (Steedman, 1976).

In the laboratory, samples were transferred to a 50 ml round-bottomed flask and agitated to ensure a homogeneous distribution of zooplankton. Six 1 mL subsamples were removed with a Stempl pipette, placed in a petri dish and observed under a dissecting microscope for zooplankton identification and quantification. Zooplankton were classified as close as possible to species level. However, in many cases identification to genus, family, or order levels was only possible.

After identification and quantification, each sample was washed with distilled water, filtered on pre-combusted and pre-weighed Whatman GFC filters, dried at 60°C for 24 h and ashed at 480°C for 5 h to determine sample dry weight and ash-free dry weight (AFDW or biomass).

A flow meter was not available to determine the volumes of water sampled at each tow. Therefore, all estimates of zooplankton abundance and biomass are relative, standardised to the number of individuals or milligrams ash-free dry weight (biomass) per 3 minutes of towing time. Relative species abundance was determined for each site as the mean number of individuals (from the six 1 ml subsamples) multiplied by 50 and standardised to the number of individuals per 3 minutes of towing time by:

$$\text{Relative abundance}_{\text{(no. per 3 min tow)}} = \text{mean no.}_{\text{(from 6 subsamples)}} \times 50 \times \text{tow time (sec)} / 180 \text{ sec}$$

All abundance data (counts) were ln (x + 1) transformed prior to statistical analysis as recommended by Zar (1984). Differences in community structure were analysed by two-factor non-parametric multivariate analysis of variance (NP-MANOVA computer program; Anderson, 1999) with SITE and MONTH as fixed factors. Species abundance data from the two fortnightly samples per site collected each month were used as replicates. Taxa that were rarely encountered (overall trial mean < 2 individuals per 3 minute tow) were omitted from the analysis, leaving 29 taxa as variables. The analysis was based on Bray-Curtis dissimilarity distances between each pair of samples and 4999 permutations of the ln (x + 1) transformed raw data.

Two-factor ANOVA was performed on each of four of the most common species encountered during the trial as well as biomass data (SITE and MONTH as fixed factors), again using the two fortnightly samples collected at each site per month as replicates.

## Results

Although temporal variation in zooplankton community structure was significant (MANOVA, $P < 0.001$), no significant differences among sites nor a month x site interaction were indicated ($P > 0.05$).

### Major Zooplankton Species

The major groups/species of zooplankton, their overall trial mean abundance and the range of species abundance encountered in individual tows are listed in Table 16.1. Taxonomic classification, observations and references used in identification follow. Relative abundances (mean number of individuals per 3 minute tow, n = 3 tows per sampling date) of individual groups or species over the sampling period are provided in Fig. 16.1 to 16.4.

Phylum Coelenterata/Cnidaria
- Class Hydrozoa
  - Order Leptomedusae
    1. *Obelia* sp.; possibly *O. australis* or *O. geniculata* (Shepherd and Thomas, 1982)
    2. *Tiaropsis* sp. (Newell and Newell, 1977)
  - Order Anthomedusae
    3. *Rathkea octopunctata* (Shepherd and Thomas, 1982)

Free-swimming coelenterates were primarily represented by the above three small hydromedusae, of which *Obelia* sp. was the most predominant (Fig.16.1). *Obelia* sp. medusae were found throughout the year but most abundant during the summer months from November 1995 through February 1996. Significant fluctuations in *Obelia* abundance between successive sampling periods were evident during this period (eg. fluctuations between 17 and 1400 individuals per tow).

Phylum Mollusca
- Class Bivalvia
  4. Bivalve veliger larvae

Bivalve larvae were only encountered twice during the trial, achieving abundance levels of 517 and 41 veliger larvae per tow during January 1996, representing only 8% and 1% of total zooplankton abundance on these two occasions. Shells ranged from 150 to 200 $\mu$m in length and were light brown in colour. Larvae were most probably *Mytilus* sp., as they were superficially similar to descriptions of several authors (Newell and Newell, 1977; Lutz and Kennish, 1992; Cameron, 1997). However, detailed examination of such diagnostic structures as provinculum dentition (see Lutz and Kennish, 1992) was not undertaken. Therefore, the exact species to which these larvae belong remains uncertain.

**Table 16.1.** Zooplankton collected in North West Bay from June 1995 to May 1996, their overall trial mean abundance (mean number of individuals per 3 minute tow, n = 24 sampling dates) and the range of abundances encountered in individual tows (n = 72 tows).

| Species or Taxonomic group | Overall trial mean abundance (No. per 3 min tow) | Range |
|---|---|---|
| Calanoid copepods (mixed assemblage) | 1168 | 28 - 4100 |
| *Evadne nordmanni* | 714 | 0 - 4883 |
| *Oikopleura dioica* | 251 | 1 - 1228 |
| *Obelia sp.* | 225 | 0 - 1371 |
| Fish eggs | 47 | 0 - 429 |
| *Podon polyphemoides* | 40 | 0 - 222 |
| *Nyctiphanes australis* (calyptopsis and furcilia larvae) | 28 | 0 - 245 |
| Bivalvia (veliger larvae) | 23 | 0 - 517 |
| *Membranipora membranacea* (cyphonautes larvae) | 21 | 0 - 377 |
| *Pathkea octopunctata* | 21 | 0 - 178 |
| *Lucifer hanseni* | 21 | 0 - 210 |
| *Tiaropsis sp.* | 14 | 0 - 87 |
| Majiidae (zoea larvae) | 13 | 0 - 68 |
| Echinoidea (echinopluteus | 9 | 0 - 115 |
| Fish larvae | 6 | 0 - 35 |
| *Calliannassa sp.* (zoea larvae) | 5 | 0 - 29 |
| *Pagurus sp.* (zoea larvae) | 4 | 0 - 37 |
| *Balanus sp.* (naupius/metanauplius larvae) | 4 | 0 - 72 |
| *Astropecten irregularis* (bipinnaria larvae) | 4 | 0 - 54 |
| Leucosidae (zoea larvae) | 4 | 0 - 38 |
| *Munida sp.* (zoea larvae) | 4 | 0 - 33 |
| *Porcellana sp.* (zoea larvae) | 3 | 0 - 30 |
| Ophiuroidea (ophiopluteus | 3 | 0 - 35 |
| *Crangon sp.* (post-larvae) | 2 | 0 - 23 |
| *Jaxea sp.* (zoea larvae) | 2 | 0 - 43 |
| *Sagitta sp.* | 2 | 0 - 23 |
| Polychaeta (spionid | 1 | 0 - 18 |
| *Phoronis australis* (actinotroch larvae) | 1 | 0 - 7 |
| *Vibilia sp.* | < 1 | 0 - 2 |
| Ascidiacea (appendicularia-type larvae) | < 1 | 0 - 3 |
| *Conchoecia sp.* | < 1 | 0 - 1 |
| *Jasus sp.* (phyllosoma larva) | < 1 | 0 - 1 |
| *Caprella sp.* | < 1 | 0 - 1 |

Phylum Annelida

Class Polychaeta

5. Unidentified larva; similar to late spionid larva (Newell and Newell, 1977)

Polychaete larvae were rarely encountered. The highest abundance was observed in January and March 1996 at 18 and 10 larvae per tow.

Phylum Arthropoda

Class Crustacea

Subclass Ostracoda; Order Myodocopa

6. *Conchoecia* sp. (Watson and Chaloupka, 1982); rarely encountered.

Subclass Branchiopoda; Order Cladocera

7. *Evadne nordmanni* (Della Croce, 1974; Watson and Chaloupka, 1982)

8. *Podon polyphemoides* (Dakin and Colefax, 1940; Della Croce, 1974)

Cladocerans were the second most abundant zooplankton group after the calanoid copepods, representing 19% of the total abundance during the trial. *Evadne nordmanni*, the dominant species of this group, was found throughout the year, with peaks in October-November, January and March-April (5000, 2000, and 700 individuals per tow, Fig. 16.1). *Evadne* was the dominant species in samples collected in October-November and March-April (Fig. 16.5). *Podon polyphemoides* was primarily encountered in the spring and summer, achieving peaks of 150 and 220 individuals per tow in November 1995 and February 1996 (Fig. 16.1).

Subclass Copepoda

9. Order Calanoida (mixed assemblage)

Calanoid copepods were the dominant zooplankton group in North West Bay. Copepods represented an overall average of 51% of the total zooplankton abundance ranging from 3% in October 1995 to 92% in July 1995. Identification of the various species of copepods was beyond the scope of this study. For reviews of the common copepods found in Tasmanian waters see Taw, 1975; Taw and Ritz, 1979; Watson and Chaloupka, 1982. Copepods were encountered throughout the year with the greatest abundance observed during the summer months (Fig. 16.1). Abundance levels from June to November were less than 500 per tow, peaking to 2000-4000 during the summer months, declining to low levels from February to April (40 - 800, mean 260 per tow) before increasing in May to 3000 individuals per tow. Copepods dominated samples for most of the study period. However, a significant inverse relationship with *Evadne* was observed in the percentage of total zooplankton abundance ($r = - 0.841$, $P < 0.0001$). When copepod percentages fell, a corresponding increase in *Evadne* dominance, and vice versa, was evident (Fig.16.5). This relationship was explored further. Although % dominance was significant, no significant correlation between the actual abundance of the two groups were indicated at time lags of 0, 1, 2 or 3 weeks ($P > 0.05$), nor was abundance of either group correlated with chlorophyll levels at the same time lags. A few harpacticoid copepods were encountered on rare occasions but their abundance levels were not enumerated.

Subclass Cirripedia

10. *Balanus* sp. nauplius/metanauplius larvae
(Newell and Newell, 1977)

Barnacle larvae were encountered at low densities (< 7 larvae per tow) in August-September, February and May (Fig. 16.2). Peak abundance of 72 larvae per tow was observed in March 1996.

Subclass Malacostraca

Order Amphipoda

11. Suborder Gammaridea, eg. *Vibilia* sp. (Watson and Chaloupka, 1982); rarely encountered
12. Suborder Caprellidea, eg. *Caprella* sp. (Edgar, 1997); rarely encountered

Order Euphausiacea

13. *Nyctiphanes australis* - calyptopsis and furcilia larvae
(Dakin and Colefax, 1940; Mauchline, 1971)

Larval stages of the euphausid *Nyctiphanes australis* were primarily encountered during the summer months, achieving a maximum abundance of 245 larvae per tow at the end of February 1996 (Fig. 16.2).

Order Decapoda
Suborder Natantia
Section Caridea

14. Family Crangonidae - larvae eg. *Crangon* sp. (Dakin and Colefax, 1940; Williamson, 1960; Watson and Chaloupka, 1982)

A few caridean postlarvae, less than 25 per tow, were identified during the summer months between December and February (Fig. 16.2).

Section Penaeidea

15. Family Sergestidae, *Lucifer hanseni*
(Dakin and Colefax, 1940; Taw, 1975; Watson and Chaloupka, 1982)

The sergestid shrimp, *Lucifer hanseni*, was found in most samples throughout the year at low densities (Fig. 16.2). Peak abundance of 210 individuals per tow was observed in March 1996.

Suborder Reptantia
Section Palinura

16. Family Scyllaridae - phyllosoma larva of *Jasus* sp.
(Dakin and Colefax, 1940)

Only one specimen of lobster larvae was collected during the entire sampling period (November, 1995).

Section Anomura

Superfamily Thalassinidea - larvae (Dakin and Colefax, 1940)
17. Family Callianassidae, eg.*Callianassa* sp.
18. Family Laomediidae, eg. *Jaxea* sp.

*Callianassa* sp. zoea were mainly encountered during the summer months at low densities, peaking at 30 larvae per tow at the end of February. *Jaxea* sp. zoea were only encountered on three occasions during December and January, with the highest abundance of 40 larvae per tow at the end of January (Fig. 16.2).

Superfamily Galatheidea/Family Galatheideae - zoea larvae
19. *Munida* sp. (Pike and Williamson, 1972)
20. *Porcellana* sp. (Pike and Williamson, 1972)

Low densities of galatheid zoea, probably belonging to *Munida* sp., were found from June through to December with the highest levels of abundance in July and August (32 zoea per tow, Fig. 16.2). Porcelain crab zoea, possibly *Porcellana* sp. or *Petrolisthes* sp., were collected during the summer months (December to February) achieving maximum densities of 30 zoea per tow in December.

Superfamily Paguridea/Family Paguridae
21. *Pagurus* sp.- like zoea larvae (Pike and Williamson, 1958)

Hermit crab larvae were collected in the summer months (December to February) with the highest density of 37 zoea per tow observed at the beginning of December (Fig. 16.3).

Section Brachyura

22. Family Majiidae (Watson and Chaloupka, 1982)
23. Family Leucosidae (Watson and Chaloupka, 1982)

Brachyuran zoeas, characterised by the presence of a rostral and a dorsal spine on the carapace, classified as Family Majiidae (Watson and Chaloupka, 1982), were encountered from September 1995 through to March 1996, achieving a peak abundance of 70 per tow in December (Fig. 16.3). Brachyuran zoeas lacking a rostral and dorsal spine on the carapace, classified as Family Leucosidae, were mainly encountered from December to February with a peak abundance of 38 zoea per tow at the end of January (Fig.3). Brachyuran megalopa were rarely encountered, represented by only a few specimens from November to February.

Phylum Phoronida

24. *Phoronis australis* larvae (Forneris, 1957; Barnes, 1968; Edgar, 1997)

Small numbers (one or two per sample) of phoronid worm actinotroch larva, probably *Phoronis australis*, were found in samples from June to September and February to April (Fig. 16.3).

Phylum Bryozoa/Ectoprocta

25. *Membranipora membranacea* larvae (Ryland, 1965)

Bryozoan cyphonautes larvae, probably belonging to *Membranipora membranacea* which are commonly found in Tasmanian waters (Edgar, 1997), were collected during January and February. Peak abundance of 375 larvae per tow was observed on January 31 1996 (Fig. 16.3).

Phylum Echinodermata

Class Asteroidea

26. *Astropecten* sp. - bipinnaria larvae (Newell and Newell, 1977)

Class Ophiuroidea

27. Unidentified ophiopluteus larvae (Newell and Newell, 1977)

Class Echinoidea

28. Unidentified echinopluteus larvae (Newell and Newell, 1977)

Three echinoderm larval forms, sea star bipinnaria (eg. *Astropecten* sp.), brittle star/basket star ophiopluteus and sea urchin/sand dollar echinopluteus were observed in small numbers. Bipinnaria larvae were only encountered in significant numbers in December and January (50 larvae per tow), echinopluteus larvae in November, February and March (25, 114 and 30 larvae per tow, respectively) and ophiopluteus larvae in February and March (30 larvae per tow).

Phylum Chaetognatha

29. *Sagitta* sp. (Dakin and Colefax, 1940; Watson and Chaloupka, 1982)

Chaetognaths were collected in low numbers (< 20 per tow) primarily during the summer months (Fig. 16.4).

Phylum Chordata

Class Ascidiacea

30. Unidentified appendicularia-type larvae

Only a few specimens of tunicate larvae were collected, restricted to the July to September 1995 period.

Class Appendicularia/Larvacea

31. *Oikopleura dioica* (Dakin and Colefax, 1940; Watson and Chaloupka, 1982)

Appendicularians comprised the third largest zooplankton group by number, representing 11% of the overall abundance. *O. dioica* was collected year round, achieving a maximum abundance of 1200 individuals per tow in November (Fig. 16.4).

Class Osteichthyes

32. Fish eggs and larvae (unidentified)

Pelagic fish eggs were collected during the summer achieving maximum abundance in November (160 eggs per tow) and January (430 eggs per tow). Fish larvae were collected between November and January, achieving a maximum abundance of 35 larvae per tow (Fig. 16.4).

Miscellaneous

In January 1996 a large number of cream-coloured, spherical, single-celled organisms were collected. Cells were approximately 200 $\mu$m diameter, some with fine, almost invisible spines, emanating from the body. Identification was not established but was superficially similar to a radiolarian (eg. *Thalassicolla* sp.; Barnes, 1968) or a foraminiferan (eg. *Globigerina* ; Newell and Newell, 1977). Abundance of these organisms were not enumerated but were relatively dense in January, after which only a few individuals were identified per sample.

Significant algal blooms during the trial were reflected by the collection of several species of phytoplankton in the zooplankton tows. Specifically, on Feb 28 and Mar 13 1996 several species of diatoms *(Pseudonitzschia pseudodelicatissima, Chaetaceros affinis and C. didymus)* and dinoflagellates *(Ceratium tripos and C. furca)* were collected in significant numbers. Extensive rainfall and runoff occurred in the south-east of Tasmania during early February, which may have contributed to conditions favouring the observed dinoflagellate bloom. A subsequent bloom of *Pseudonitzschia* was observed in May 1996.

Although temporal variation in each of the four most dominant species (Calanoid copepods, *Evadne nordmanni*, *Oikopleura dioica* and *Obelia* sp.) was significant ($P < 0.0001$), no significant spatial variation among the three sampling sites nor site x month interactions were indicated (all at $P > 0.05$).

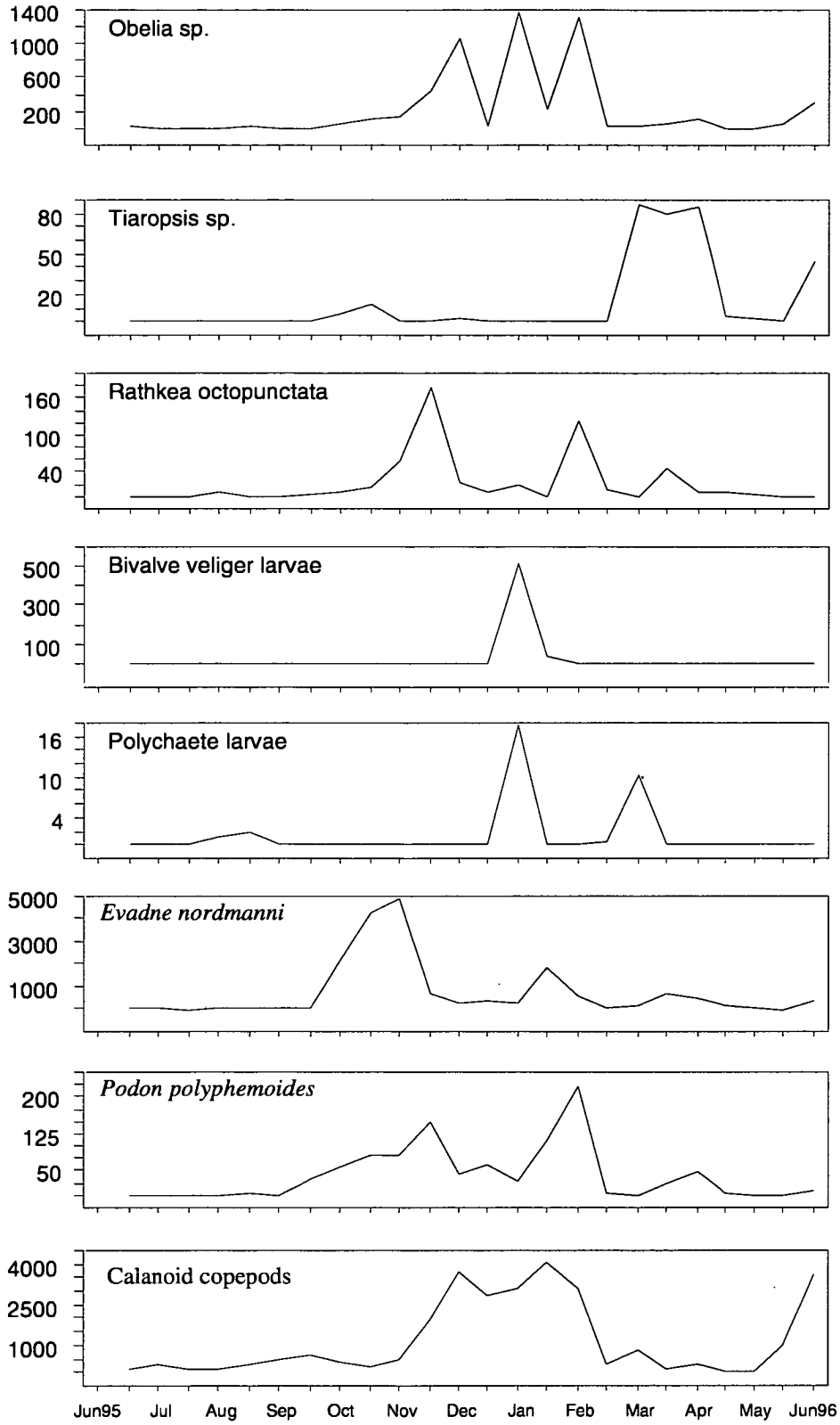


**Figure 16.1** Zooplankton abundance (mean number per 3 min tow, n = 3 tows per sampling date) in North West Bay from June 1995 to June 1996.

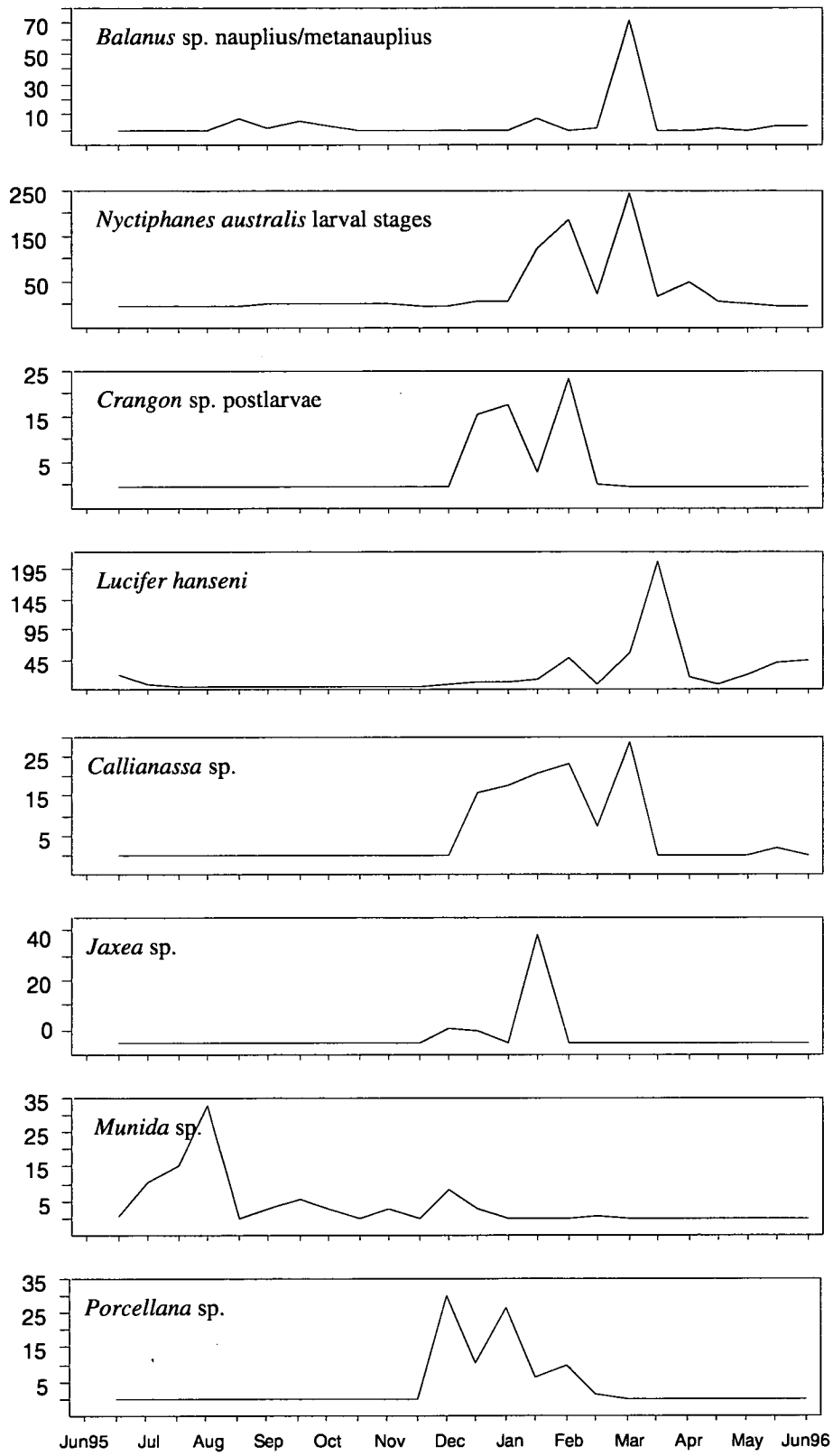


**Figure 16.2** Zooplankton abundance (mean number per 3 min tow, n = 3 tows per sampling date) in North West Bay from June 1995 to June 1996.

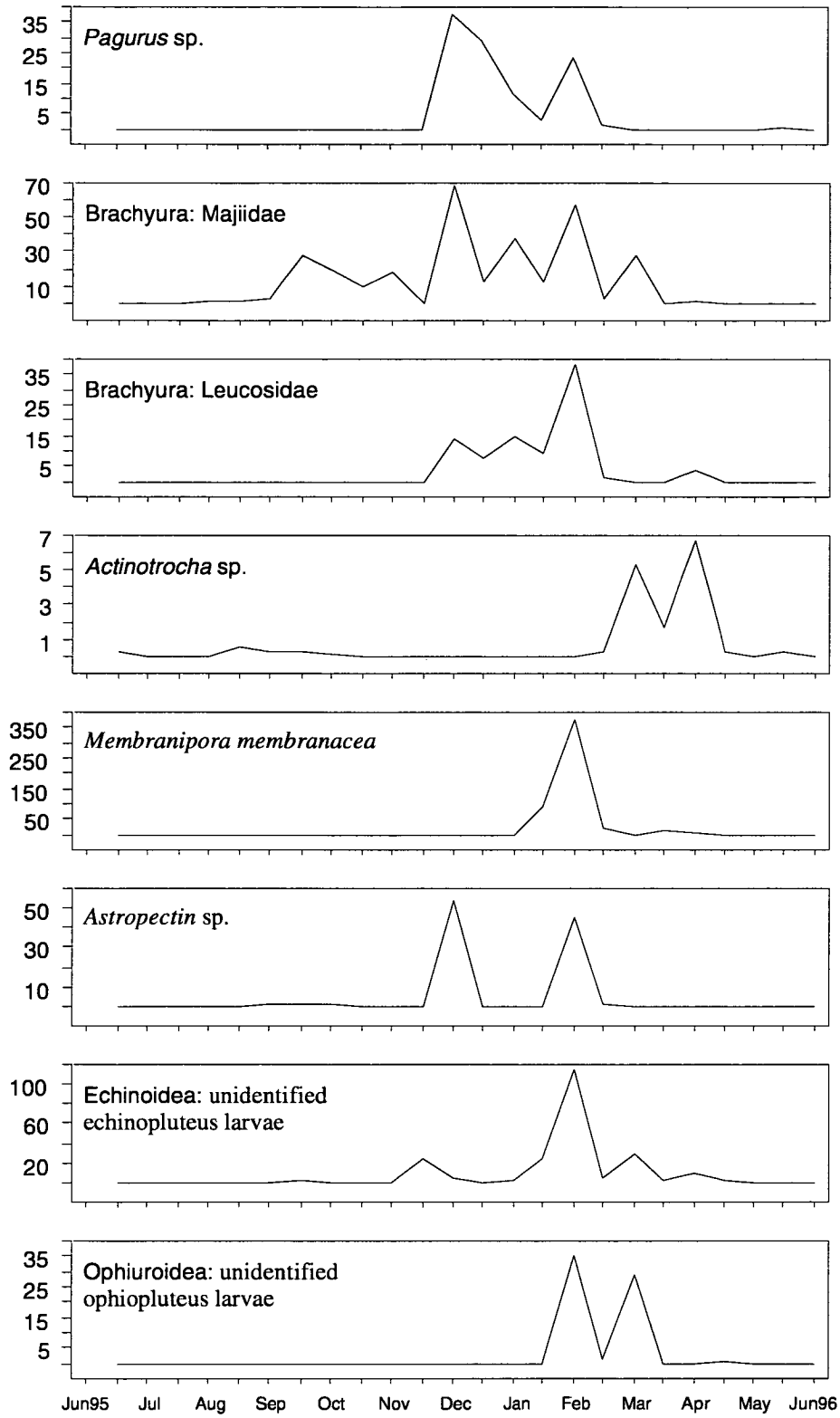


**Figure 16.3.** Zooplankton abundance (mean number per 3 min tow, n = 3 tows per sampling date) in North West Bay from June 1995 to June 1996.

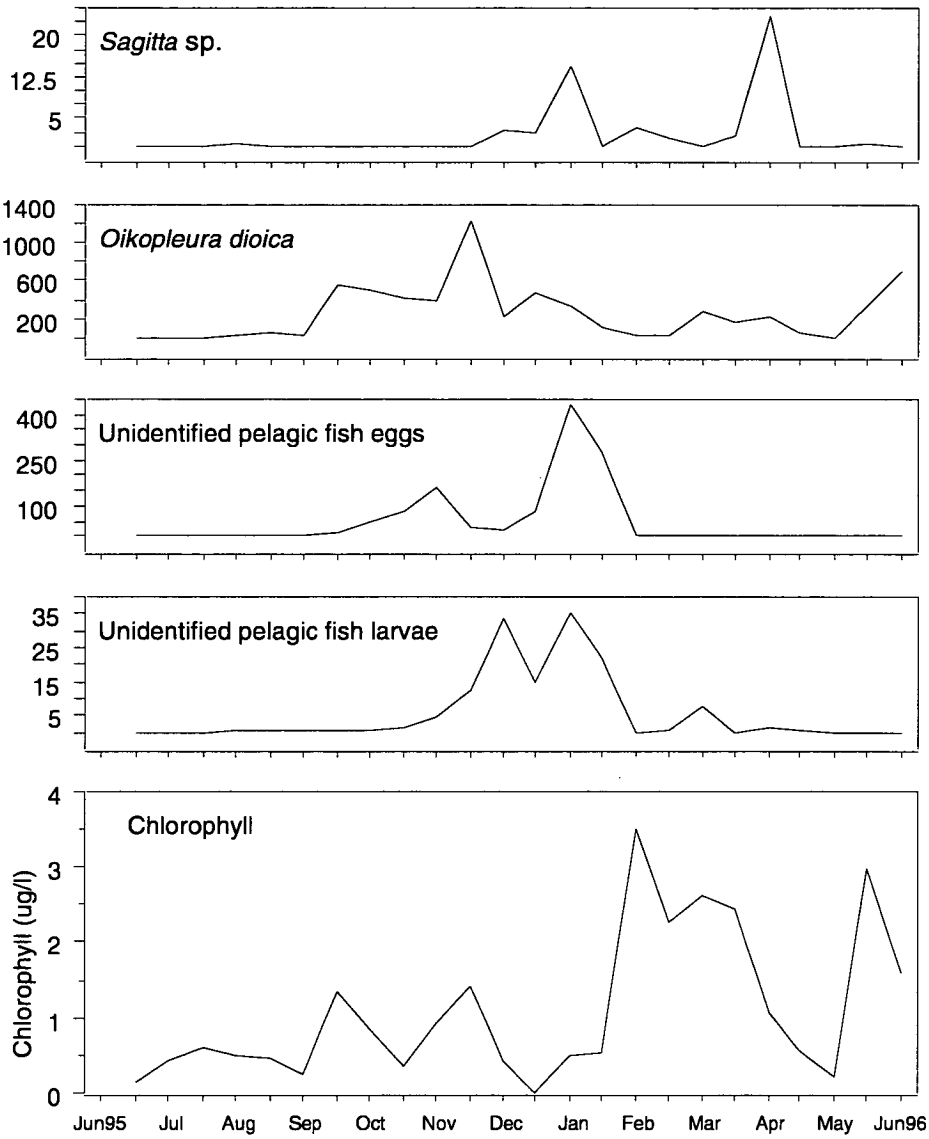


**Figure 16.4.** Zooplankton abundance (mean number per 3 min tow, n = 3 tows per sampling date) and mean chlorophyll content ($\mu g \cdot l^{-1}$) in North West Bay from June 1995 to June 1996.

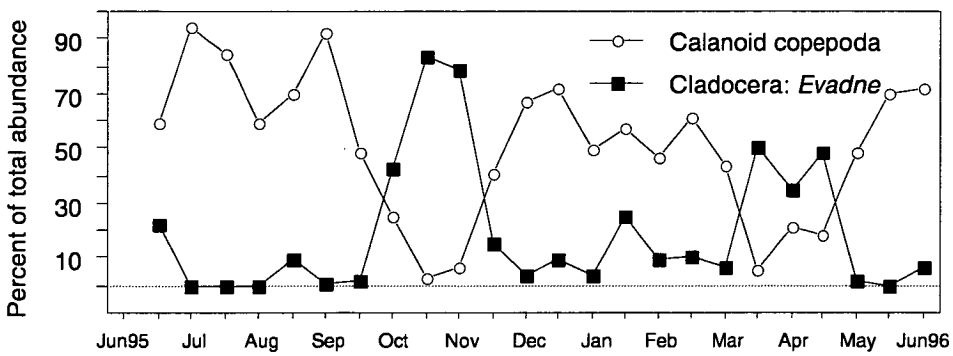


**Figure 16.5.** Annual cycle of copepod and cladoceran (*Evadne nordmanni* ) abundance in North West Bay, expressed as a percentage of total numbers of individuals per sampling date.

### Zooplankton Biomass

Zooplankton biomass remained relatively low (< 5 mg AFDW per tow) over the winter period (June to September 1995), gradually increasing over the spring to peak at 95 mg AFDW per tow in December 1995 (Fig.16.6). Biomass displayed significant fluctuations over the summer period, declined to low levels during autumn (2.0 mg AFDW per tow, May 1 1996) and increased to 60 mg AFDW per tow at the conclusion of the trial. Variations in biomass were largely due to changes in abundance of three species, with biomass positively correlated to numbers of copepods ($r = 0.693$, $P < 0.0001$), *Obelia* sp. ($r = 0.613$, $P = 0.001$) and *Oikopleura dioeca* ($r = 0.57$, $P = 0.003$). Significant temporal variation in zooplankton biomass was indicated ($P < 0.0001$). There were no significant differences among the three sampling locations (I, III and IV) in overall trial mean biomass (mean = $31.4 \pm 3.7$ mg·tow$^{-1}$, n = 72 samples; $P > 0.05$) nor a significant site x month interaction ($P > 0.05$).

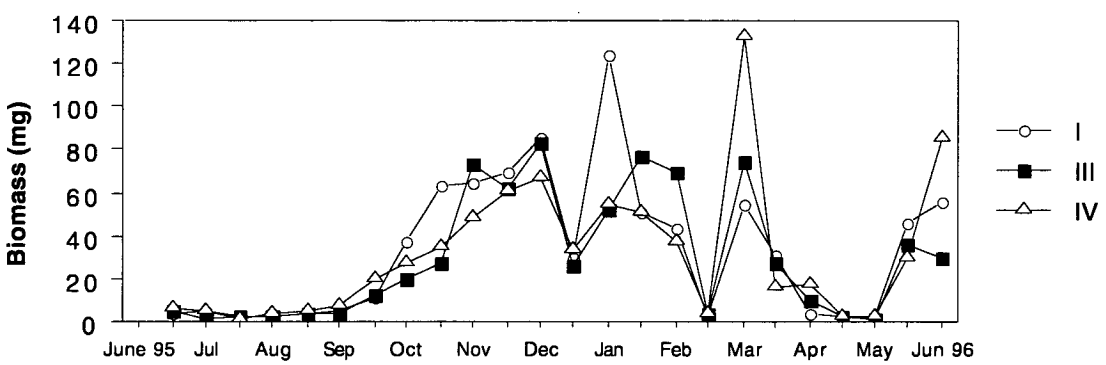


**Figure 16.6.** Zooplankton biomass (mg ash-free dry weight per 3 min tow) collected fortnightly at three sites in North West Bay from June 1995 to June 1996.

## Discussion

Zooplankton composition and abundance in southern Tasmanian coastal waters has not been extensively investigated. However, Taw (1975) and Taw and Ritz (1979) have provided detailed accounts of some common species, including: the seasonal abundance of numerous species of copepods (64 species of calanoids, 6 species of cyclopoids and 1 harpacticoid), euphausids (8 species) chaetognaths (11 species, including 9 species of *Sagitta*), and tunicates (8 species) found in southern Tasmania. The present study has identified a number of zooplankton species found in southern Tasmanian near-shore coastal waters in addition to those reported by Taw (1975) and Taw and Ritz (1979). Notable additions include several species of hydromedusae, bivalve larvae, two cladoceran species, several species of crustacean larvae, the

appendicularian *Oikopleura dioica,* as well as phoronid, ectoproct and echinoderm larvae.

The absence of any significant spatial variation in zooplankton community structure, dominant species, or biomass among sites, suggests that the Aquatas fish farm may have no significant influence on these parameters. These results support those in Section 4, whereby water quality and environmental conditions were similar among the experimental sites. In particular, no differences were significant for suspended particulate matter or chlorophyll. It could be suggested that dissolved nutrient wastes generated by the fish farm were not sufficient to stimulate phytoplankton production or, if so, may have been dispersed throughout the bay. It has also been suggested that phytoplankton production stimulated by fish farms may be grazed upon by zooplankton as fast as it is produced (Gowen et al., 1988). If so, the effect of fish farms on phytoplankton production could be masked by the grazing activity of zooplankton. Any localised increases in phytoplankton production might be reflected by localised increases in zooplankton abundance (eg. Ryan et al., 1986). The inclusion of a zooplankton monitoring program, therefore, might help identify some possible environmental impacts of fish farms that might otherwise be overlooked. As zooplankton composition and abundance was similar among sites, it seems reasonable to suggest that no localised increases in phytoplankton production was stimulated by nutrient wastes from the Aquatas fish farm.

Although, copepods and *Evadne* demonstrated an inverse relationship in % dominance of zooplankton samples, it is unknown if any real relationship might exist between the two groups. Although, significantly correlated, % dominance was not similarly reflected by any relationship between their actual abundance at time lags of 0 to 3 weeks. Further no correlation of abundance with chlorophyll concentrations were indicated at similar time lags. It may be that the two groups displayed rhythms characteristic of their normal life cycle and % dominance was just a manifestation of these natural cycles. Any relationship between the two groups would be highly speculative, particularly as copepods were a mixed assemblage of species, possibly displaying a range of food preferences. Therefore, without further investigation, the significant relationship of % dominance must remain as an interesting observation.

Although not a specific objective, the zooplankton-sampling program may have helped to identify specific periods during the year when mussel larvae were present in the water column. Periodically high concentrations of veliger larvae would be indicative of a major mussel spawning in the preceding weeks. Therefore, this type of information might be used to confirm spawning periods observed in the mussels cultured in this study, inferred by significant losses of meat weight and condition.

However, the appearance of veliger larvae was restricted to only two sampling dates in January 1996, neither coinciding with the two major spawnings identified by significant losses of soft tissue weight and condition (August 1995 and February 1996). The presence of larvae in January 1996 suggests that they may have originated from other mussel stocks or asynchronous spawning of experimental mussels prior to the major spawning in February 1996.

The general absence of larvae throughout the year, particularly following the two major spawning events in August and February, may be due to several factors. Firstly, the magnitude of the natural mussel population in the region and their spawning activity (ie. was spawning of natural mussels coincident with that of cultured mussels) is unknown. As such, the source(s) of the planktonic larvae remains unknown. The number of spawning adults in NWB, both natural and cultured, and the subsequent dilution of larvae within NWB, may have been insufficient to produce collectable numbers of larvae. Secondly, with a flushing time of NWB being approximately 7 days, significant numbers of larvae may have been transported out of the bay between zooplankton sampling (approximately every two weeks). Lastly, low abundance or absence of larvae in samples would certainly be exacerbated by the large mesh size of the zooplankton net (300 $\mu$m) with low retention efficiencies for smaller larvae (eg. shell lengths of 'D'-shaped veliger are approximately 100 to 120 $\mu$m, Lutz and Kennish, 1992). The cumulative effect of all three of these factors might have resulted in the general absence of planktonic bivalve larvae.

Despite the inappropriate mesh size to adequately sample bivalve larvae, low mussel larvae abundance and settlement in North West Bay was also reported by Cameron (1997) during the same period. However, Cameron (1997) did note a significant number of planktonic larvae in March 1996 and May 1996 (1178 and 853 larvae·m$^3$, respectively) using an 85 $\mu$m mesh plankton net. A modest spat settlement on collectors was also observed in March-April 1996. Planktonic larvae in March 1996 and settlement in March-April 1996 coincides with, and may have originated from, the major spawning event observed in the cultured mussels during February 1996. However, both larval abundance and spat settlements within NWB were notably poorer than other regions in southern Tasmanian waters and most sites surveyed exhibited spat settlements well below the commercial threshold levels of 200 mussels per metre of collector (Cameron, 1997).

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# Appendix 4. Model Mussel Longline System

## Introduction

Mussels cultured within the Aquatas fish farm probably have a limited capacity to ingest and assimilate a significant portion of solid organic wastes released from the farm. This limited capacity is due to three main factors: (i) suspended mussels are probably located too far distant from the fish cages to be in a suitable position to intercept a significant fraction of suspended wastes before they settle to below the level of mussel droppers; (ii) through the effect of dilution, particulate wastes constitute only a minor fraction of the total available suspended POM in the water column; and (iii) the ambient seston concentration, being consistently above the pseudofaeces concentration, limits the ability of mussels to ingest a significant fraction of additional suspended POM. However, perhaps integrated mussel-salmon culture may still be environmentally beneficial. Although not assimilating a significant fraction of solid organic wastes from the fish farm, mussels could be cultured to remove natural suspended POM, a quanity that is equivalent to the solid waste loadings of the farm. Mussel culture could counteract the organic waste loadings (fish feed and faeces) from the fish farm by removing suspended particulates from the water column (primarily phytoplankton and detritus but would also include some fish farm wastes); theoretically, the net outcome of such an integrated salmon-mussel culture system would be negligible solid organic waste loadings into NWB. What is the scale of mussel culture required to achieve this outcome? Would there be any negative repercussions from such a system?

## Methodology

An intensive mussel cultivation system was modelled to remove a quantity of organic particulate matter equivalent to the organic waste loadings of the Aquatas salmon farm. Mean theoretical organic waste loadings from the Aquatas salmon farm in 1995-96 approximated 321 kg total solid organic matter·day$^{-1}$ (dry weight); 246 kg organic matter·day$^{-1}$ was fish feed fines and faecal wastes and 75 kg organic matter·day$^{-1}$ was uneaten fish pellets (see Section 13.2 for details of feed delivery, feed digestibility and assumptions made for estimates of solid waste loadings). Two models of the scale of mussel culture required to remove fish farm waste loadings were made: one for the removal of feed fines and faecal wastes, and one for the removal of total organic wastes (feed fines + faecal wastes + uneaten fish feed pellets). Two models were made because feed fines and faecal wastes were more precisely quantified (details of feed digestibility and feed fine content were provided by the manufacturer); the

quantity of uneaten feed pellets was unknown, allocated a value of 4% of feed delivered, and may have been underestimated.

The required scale of intensive mussel culture to neutralise the fish farm solid waste loadings was based on the carrying capacity model of Incze et al. (1981). The model consists of tiers of longlines, oriented perpendicular to the current flow, with mussels cultured on suspended socks. The quantity of suspended matter absorbed by each tier is a function of the seston concentration and the feeding and digestive processes of mussels. The concentration of seston as it enters each tier is a function of water flow rates, original seston concentration (before entering the first tier) and the filtration of particles by all mussels in the up-current tiers (Incze et al., 1981). In this modified model, POM absorption by each tier of mussels was estimated using the physiological feeding responses of individual mussels described in Page and Ricard (1990) and in Section 12.2.

This particular model consists of longlines (tiers) stocked with mussels of 70 mm shell length (L), 30 g whole live wet weight (WLW) and 2.0 g dry meat weight (DMW); this is the approximate size of experimental mussels in peak condition in June-July 1995. These are cultivated in 5 m socks, at a density of 200 mussels·$m^{-1}$, suspended at 0.5 m intervals from 100 m longlines. These sock densities (200 mussels·$m^{-1}$) are slightly higher than those of the present study (mean survivor density of socks from March to December 1995 was 145 mussels·$m^{-1}$). However, this density is in the range of grow-out densities encountered in normal commercial practice (Incze and Lutz, 1980; Hickman, 1992; G. Schroter, pers. comm.). This density provides for a potential harvest production of 10 kg WLW·m of $sock^{-1}$ or 10 tonnes·100 m $longline^{-1}$, assuming mussels are harvested at the maximum size attained during the present experiment (85 mm L and 50 g WLW prior to spawning in February 1996).

The model has the following assumptions:

1. The concentration of particles per litre is homogeneous as it enters each tier.
2. Current flow is normal to the face of each tier, laminar and with a constant velocity through the length of the system (4.0 cm·$sec^{-1}$; the mean current velocity measured from September-November 1995).
3. Seston concentration ($n_1$) entering the first tier ($T_1$) = 6.421 mg·$l^{-1}$ and 34.9% POM (mean trial concentrations from sites I-IV).
4. Clearance rate (CR, $l \cdot h^{-1}$) of mussels in each tier
   $CR = 1.73 \cdot DMW^{0.413} - 0.006 \cdot$ seston concentration of water entering that tier, where DMW = 2.0 g.
5. Pseudofaeces threshold concentration (T) of 70 mm L mussels = 5.01 mg·$l^{-1}$.
6. Absorption efficiency = 45.2%
7. No pre-ingestive selection of organic particles in preference to inorganic particles (ie. %POM of ingested ration = %POM of seston).

Terms used in calculating the seston concentration flowing into each tier:

a: ($m^2$), area of each tier normal to the current flow (= 100 m · 5m = 500 $m^2$).
V: ($m \cdot h^{-1}$), current velocity (= $4cm \cdot sec^{-1}$ = 144 $m \cdot h^{-1}$).
N: ($liters \cdot h^{-1}$), volume of water flowing through each tier per hour.
$N = V \cdot a \cdot 10^3 = 7.2 \cdot 10^7$ $liters \cdot h^{-1}$.
$n_k$ ($mg \cdot l^{-1}$), seston concentration flowing into tier ($T_k$), k = 1, 2, 3,...
M: number of mussels suspended in each tier
M = 200 mussels·m of $sock^{-1}$ ·x 5 m socks x 200 $socks \cdot longline^{-1}$ = 200,000.

The seston concentration (n) flowing into tier 1 ($T_1$) is $n_1 = 6.421$ $mg \cdot l^{-1}$ and $n_k$ entering each subsequent tier ($T_k$) was calculated:

$$n_k = n_{k-1} \cdot [(N - CR_{k-1} \cdot M)/N]^{k-1}, \quad k = 2, 3, 4, \ldots$$

The quantity of POM absorbed per mussel ($A_m$, mg $POM \cdot h^{-1}$) in each tier was calculated according to the physiological parameters described by Page and Ricard (1990). The daily quantity of POM absorbed per tier ($A_T$) = $A_m \cdot M \cdot 24$ h. Longlines were successively added to the model until the number of tiers required to absorb 246 kg $POM \cdot day^{-1}$ (quantity of fish feed fines and faeces only) and 321 kg $POM \cdot day^{-1}$ (total solid organic wastes including uneaten fish pellets) was attained (ie. where $\sum A_T$ = 246 and 321 kg $POM \cdot day^{-1}$).

Scope for growth was estimated for each tier based on the physiological relationships described by Page and Ricard (1990). This was included to determine if the seston depletion by the leading tiers of mussels might significantly reduce the potential for growth of mussels in "downstream" tiers (ie. a shadowing effect).

Organic waste loadings from the longline system were also estimated. Organic matter lost in pseudofaeces (Ps-POM) and intestinal faeces (faecal-POM) were calculated:
Ps-POM (mg $POM \cdot h^{-1}$) = seston filtered (SF, $mg \cdot h^{-1}$)·%POM·%Ps
Faecal-POM (mg $POM \cdot h^{-1}$) = POM ingested - POM absorbed (mg $POM \cdot h^{-1}$)
Total waste POM per mussel ($mg \cdot h^{-1}$) was the sum of Ps-POM and faecal-POM; daily total wastes per tier = total waste $POM \cdot mussel^{-1}$ x M x 24 h.

## Results

Twenty-nine tiers of suspended mussels (approximately 5.8 million mussels) are required to absorb a quantity of POM matching the daily solid organic particulate waste loadings (fish feed fines + faeces) from the Aquatas fish farm (246 kg POM·day$^{-1}$). Thirty-eight tiers (approximately 7.6 million mussels) are required to remove an equivalent quantity of POM as the total waste organic loadings (fish feed fines + faeces + uneaten feed pellets) from the farm (321 kg POM·day$^{-1}$, Table 16.2). Thirty-eight tiers of mussels would only reduce the original seston concentration of 6.42 mg·l$^{-1}$ to 5.05 mg·l$^{-1}$ in the water column passing out of the last tier; a concentration still exceeding the pseudofaeces threshold. The density of this system does not dramatically affect SFG. Individual mussel SFG in the leading tier, 131.3 kJ·day$^{-1}$, is reduced by 5% to 124.3 kJ·day$^{-1}$ in the last tier. However, these are SFG values if current flowed only in one direction; as current flow reverses, seston depletion through the tiers is also reversed (ie. the leading tier becomes the last and vice versa). The net effect is that daily SFG is about the same for all tiers at 127.8 kJ·day$^{-1}$, a reduction of only 2.7% from that of a sole mussel longline.

Although the modelled culture system has the potential to remove an equivalent quantity of POM from NWB as is produced by the fish farm, it also has its own waste loadings. At the modelled densities of 29 and 38 longlines, approximately 403 - 507 kg total waste organic matter·day$^{-1}$ could be expected (Table 16.2).

**Table 16.2.** (A) Estimates of physiological feeding parameters of individual mussels in a series of longline units (tiers), the number of tiers required to remove 246 and 321 kg POM·day$^{-1}$ and the scope for growth (SFG) of each tier. Estimates are based on 100 m longlines stocked with 70 mm mussels (2.0 g DMW, 30g WLW), sock density of 200 mussels·m$^{-1}$, 5 m socks suspended at 0.5 m intervals. Physiological parameters calculated according to those described in Page and Ricard (1990). CR (clearance rate), SF (seston filtered), Ps (pseudofaeces), SI (seston ingested), POM-In(gested), POM-Ab(sorbed). Not included in table but incorporated into calculations are pseudofaeces threshold concentration = 5.01 mg seston·l$^{-1}$, absorption efficiency = 0.45 and respiratory rate = 0.759 ml $O_2$·l$^{-1}$. (B) Estimated waste loadings for each tier.

**A**

| Tier ($T_K$) | Seston (mg/l) | CR (l/h) | SF (mg/h) | Ps (%SF) | SI (mg/h) | POM-In (mg/h) | POM-Ab (mg/h) | POM-Ab (kg/tier/day) | Σ Tier POM-Ab (kg/day) | SFG (kJ/day/tier) |
|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 6.42 | 2.265 | 14.54 | 20.7 | 11.54 | 4.03 | 1.82 | 8.74 | 8.7 | 131.3 |
| 2 | 6.38 | 2.265 | 14.45 | 20.3 | 11.53 | 4.03 | 1.82 | 8.73 | 17.5 | 131.2 |
| 3 | 6.34 | 2.265 | 14.36 | 19.8 | 11.51 | 4.02 | 1.82 | 8.72 | 26.2 | 131.0 |
| 4 | 6.30 | 2.266 | 14.27 | 19.4 | 11.50 | 4.02 | 1.81 | 8.71 | 34.9 | 130.8 |
| 5 | 6.26 | 2.266 | 14.19 | 19.0 | 11.49 | 4.01 | 1.81 | 8.70 | 43.6 | 130.6 |
| 6 | 6.22 | 2.266 | 14.10 | 18.6 | 11.48 | 4.01 | 1.81 | 8.69 | 52.3 | 130.4 |
| 7 | 6.18 | 2.266 | 14.01 | 18.1 | 11.47 | 4.01 | 1.81 | 8.69 | 61.0 | 130.2 |
| 8 | 6.14 | 2.267 | 13.92 | 17.7 | 11.46 | 4.00 | 1.81 | 8.68 | 69.7 | 130.0 |
| 9 | 6.10 | 2.267 | 13.84 | 17.3 | 11.45 | 4.00 | 1.81 | 8.67 | 78.3 | 129.8 |
| 10 | 6.07 | 2.267 | 13.75 | 16.8 | 11.44 | 4.00 | 1.80 | 8.66 | 87.0 | 129.6 |
| 11 | 6.03 | 2.267 | 13.67 | 16.4 | 11.43 | 3.99 | 1.80 | 8.65 | 95.6 | 129.4 |
| 12 | 5.99 | 2.267 | 13.58 | 15.9 | 11.42 | 3.99 | 1.80 | 8.65 | 104.3 | 129.2 |
| 13 | 5.95 | 2.268 | 13.50 | 15.5 | 11.41 | 3.98 | 1.80 | 8.64 | 112.9 | 129.1 |
| 14 | 5.91 | 2.268 | 13.41 | 15.0 | 11.40 | 3.98 | 1.80 | 8.63 | 121.6 | 128.9 |
| 15 | 5.88 | 2.268 | 13.33 | 14.6 | 11.39 | 3.98 | 1.80 | 8.62 | 130.2 | 128.7 |
| 16 | 5.84 | 2.268 | 13.25 | 14.1 | 11.38 | 3.97 | 1.79 | 8.61 | 138.8 | 128.5 |
| 17 | 5.80 | 2.269 | 13.17 | 13.7 | 11.36 | 3.97 | 1.79 | 8.61 | 147.4 | 128.3 |
| 18 | 5.77 | 2.269 | 13.08 | 13.2 | 11.35 | 3.97 | 1.79 | 8.60 | 156.0 | 128.1 |
| 19 | 5.73 | 2.269 | 13.00 | 12.8 | 11.34 | 3.96 | 1.79 | 8.59 | 164.6 | 127.9 |
| 20 | 5.69 | 2.269 | 12.92 | 12.3 | 11.33 | 3.96 | 1.79 | 8.58 | 173.2 | 127.7 |
| 21 | 5.66 | 2.269 | 12.84 | 11.8 | 11.32 | 3.95 | 1.79 | 8.57 | 181.7 | 127.5 |
| 22 | 5.62 | 2.270 | 12.76 | 11.4 | 11.31 | 3.95 | 1.78 | 8.57 | 190.3 | 127.3 |
| 23 | 5.59 | 2.270 | 12.68 | 10.9 | 11.30 | 3.95 | 1.78 | 8.56 | 198.9 | 127.2 |
| 24 | 5.55 | 2.270 | 12.60 | 10.4 | 11.29 | 3.94 | 1.78 | 8.55 | 207.4 | 127.0 |
| 25 | 5.52 | 2.270 | 12.53 | 10.0 | 11.28 | 3.94 | 1.78 | 8.54 | 216.0 | 126.8 |
| 26 | 5.48 | 2.271 | 12.45 | 9.5 | 11.27 | 3.94 | 1.78 | 8.53 | 224.5 | 126.6 |
| 27 | 5.45 | 2.271 | 12.37 | 9.0 | 11.26 | 3.93 | 1.78 | 8.52 | 233.0 | 126.4 |
| 28 | 5.41 | 2.271 | 12.29 | 8.5 | 11.25 | 3.93 | 1.77 | 8.52 | 241.5 | 126.2 |
| 29 | 5.38 | 2.271 | 12.22 | 8.0 | 11.24 | 3.93 | 1.77 | 8.51 | 250.0 | 126.0 |
| 30 | 5.35 | 2.271 | 12.14 | 7.5 | 11.23 | 3.92 | 1.77 | 8.50 | 258.5 | 125.8 |
| 31 | 5.31 | 2.272 | 12.07 | 7.1 | 11.22 | 3.92 | 1.77 | 8.49 | 267.0 | 125.6 |
| 32 | 5.28 | 2.272 | 11.99 | 6.6 | 11.20 | 3.91 | 1.77 | 8.48 | 275.5 | 125.4 |
| 33 | 5.25 | 2.272 | 11.92 | 6.1 | 11.19 | 3.91 | 1.77 | 8.48 | 284.0 | 125.3 |
| 34 | 5.21 | 2.272 | 11.84 | 5.6 | 11.18 | 3.91 | 1.76 | 8.47 | 292.5 | 125.1 |
| 35 | 5.18 | 2.272 | 11.77 | 5.1 | 11.17 | 3.90 | 1.76 | 8.46 | 300.9 | 124.9 |
| 36 | 5.15 | 2.273 | 11.70 | 4.6 | 11.16 | 3.90 | 1.76 | 8.45 | 309.4 | 124.7 |
| 37 | 5.11 | 2.273 | 11.62 | 4.1 | 11.15 | 3.90 | 1.76 | 8.44 | 317.8 | 124.5 |
| 38 | 5.08 | 2.273 | 11.55 | 3.5 | 11.14 | 3.89 | 1.76 | 8.44 | 326.2 | 124.3 |

**B**

| Ps POM (mg/h) | Faecal POM (mg/h) | Total waste POM (mg/h) | Total waste POM (kg/tier/day) | Σ Tier Total waste POM (kg/day) |
|---|---|---|---|---|
| 1.05 | 2.24 | 3.29 | 15.78 | 15.8 |
| 1.02 | 2.23 | 3.26 | 15.64 | 31.4 |
| 1.00 | 2.23 | 3.23 | 15.50 | 46.9 |
| 0.97 | 2.23 | 3.20 | 15.35 | 62.3 |
| 0.94 | 2.23 | 3.17 | 15.21 | 77.5 |
| 0.91 | 2.23 | 3.14 | 15.07 | 92.6 |
| 0.89 | 2.22 | 3.11 | 14.94 | 107.5 |
| 0.86 | 2.22 | 3.08 | 14.80 | 122.3 |
| 0.83 | 2.22 | 3.05 | 14.66 | 137.0 |
| 0.81 | 2.22 | 3.03 | 14.53 | 151.5 |
| 0.78 | 2.22 | 3.00 | 14.39 | 165.9 |
| 0.76 | 2.21 | 2.97 | 14.26 | 180.1 |
| 0.73 | 2.21 | 2.94 | 14.12 | 194.3 |
| 0.70 | 2.21 | 2.91 | 13.99 | 208.2 |
| 0.68 | 2.21 | 2.89 | 13.86 | 222.1 |
| 0.65 | 2.21 | 2.86 | 13.73 | 235.8 |
| 0.63 | 2.20 | 2.83 | 13.60 | 249.4 |
| 0.60 | 2.20 | 2.81 | 13.47 | 262.9 |
| 0.58 | 2.20 | 2.78 | 13.34 | 276.2 |
| 0.56 | 2.20 | 2.75 | 13.22 | 289.5 |
| 0.53 | 2.20 | 2.73 | 13.09 | 302.5 |
| 0.51 | 2.19 | 2.70 | 12.96 | 315.5 |
| 0.48 | 2.19 | 2.67 | 12.84 | 328.4 |
| 0.46 | 2.19 | 2.65 | 12.71 | 341.1 |
| 0.44 | 2.19 | 2.62 | 12.59 | 353.7 |
| 0.41 | 2.19 | 2.60 | 12.47 | 366.1 |
| 0.39 | 2.18 | 2.57 | 12.35 | 378.5 |
| 0.37 | 2.18 | 2.55 | 12.23 | 390.7 |
| 0.34 | 2.18 | 2.52 | 12.10 | 402.8 |
| 0.32 | 2.18 | 2.50 | 11.99 | 414.8 |
| 0.30 | 2.17 | 2.47 | 11.87 | 426.7 |
| 0.27 | 2.17 | 2.45 | 11.75 | 438.4 |
| 0.25 | 2.17 | 2.42 | 11.63 | 450.0 |
| 0.23 | 2.17 | 2.40 | 11.52 | 461.5 |
| 0.21 | 2.17 | 2.37 | 11.40 | 472.9 |
| 0.19 | 2.16 | 2.35 | 11.28 | 484.2 |
| 0.16 | 2.16 | 2.33 | 11.17 | 495.4 |
| 0.14 | 2.16 | 2.30 | 11.06 | 506.5 |

## Discussion

The modelled mussel culture system indicates that 38 tiers of mussel longlines, at a density of 200,000 mussels·longline$^{-1}$, could remove 321 kg POM·day$^{-1}$; the scale of mussel culture system required to off-set the organic solid waste loadings from the Aquatas farm. The number of tiers would vary according to several factors including: size of seed mussels, within-sock stocking densities, within-longline sock densities, current speed, ambient seston concentrations and %POM, and discrepancies between theoretical and actual physiological parameters incorporated into the calculations (see Section 12.4.3). The number of tiers would also vary with different estimates of waste loadings; in particular, the quantity of uneaten fish pellets, at 4% of feed delivery, may be underestimated. However, the model is simple and can be easily manipulated with more precise data.

Incze et al. (1981) recommended in their carrying capacity model that bivalve cultures should not deplete more than 50% of the available food supply, otherwise detrimental environmental impacts and a decrease in production per unit area might be expected. In the present model, seston was depleted by only 21%, with negligible effects on mussel production; a 50% reduction of ambient suspended particulate concentrations would require at least 110 longlines. Therefore, the scale of the modelled culture system, 38 longlines, appears to be well within the carrying capacity of NWB to support mussel growth without significantly affecting the marine environment. However, this scale of culture is quite extensive and is unlikely to be incorporated within the limited area of a salmon lease. Secondly, the organic waste loadings from such a culture may be double that of the fish farm. Although not adding new organic nutrients into the environment (as does the fish farm), the quantity of solid wastes deposited on the sea bed, together with the pre-existing fish farm solid waste loadings, would likely render this scale of mussel production as highly unacceptable to Aquatas. The desired effect, counteracting the organic loading of the fish farm, could be achieved by the mussel culture system located anywhere - not necessarily, nor desirably, within the Aquatas lease. Again we have no incentive for integrating mussels with salmon.

The model can be used to answer a number of questions given different scenarios. For example: if a fish farm was significantly increasing the organic content of the water column and mussels could be grown in a suitable position to intercept these waste particles, how many tiers of mussels would be required to reduce the POM concentration of the water column passing out of the farm to background levels? If we provide some dummy variables, we can provide an estimate: ambient seston concentration = 3.0 mg·l$^{-1}$ with 35% POM (1.05 mg POM·l$^{-1}$); and fish farm organic

wastes raise this concentration by 2.0 mg POM·l$^{-1}$ to 5.0 mg seston·l$^{-1}$ at 61% POM (ie. the water column concentration flowing out of the farm). Although this is a high level of organic enrichment, it is not unrealistic; a mean increase of 2.5 mg POM·l$^{-1}$ was reported in the vicinity of a salmon farm in British Columbia compared with control sites (Jones and Iwama, 1991). At the current speed, stocking densities and clearance rates given previously, a reduction in POM concentration of the water column flowing out of the fish farm (3.05 mg POM·l$^{-1}$) back to ambient levels (1.05 mg POM·l$^{-1}$) requires 174 tiers of mussels! This is but one hypothetical example; but again it seems that the potential for bivalve culture to realistically address the issue of fish farm wastes is limited.

# Appendix 5. Effect of ambient seston concentrations on POM ingestion

Troell and Norberg (1998) have demonstrated the relative importance of ambient seston levels on a mussel's ability to ingest additional food (eg. organic fish farm wastes). A similar model is presented here, incorporating some of the environmental parameters experienced within NWB. An example of the relative insignificance of increased POM levels on mussel (1.0 g DMW) ingestion and POM absorption rates above the pseudofaeces threshold is illustrated in Fig. 16.7 and Fig. 16.8 (physiological responses taken from Page and Ricard, 1990). In the first example (Fig. 16.7), mussels are grown under low ambient seston concentration of 1.0 $mg \cdot l^{-1}$, 35% POM (overall trial mean from this study), and exposed to 1.0 mg increases in seston levels due to extra POM loadings (eg. fish farm wastes). These increases in SPM and POM concentrations are reflected by increased filtration, ingestion and POM absorption rates, and would be manifested in significant increases in mussel growth. However, after the pseudofaeces threshold concentration (T) is exceeded (approximately 5.0 mg $seston \cdot l^{-1}$), ingestion becomes saturated. Above T, an increasing quantity of filtered material is voided as pseudofaeces; further increases in POM levels have a minor effect in increasing the quantity of organic material ingested and absorbed.

An alternative scenario, illustrating the importance ambient particle concentrations have on mussels' capacity to utilise additional food material, is where mussels are grown in two different ambient seston concentrations: one below and one above T (eg. 1.0 and 6.4 mg $seston \cdot l^{-1}$, both at 35% POM) with both groups exposed to increases of 0.2 mg $POM \cdot l^{-1}$ (Fig. 16.8). Again, filtration, ingestion and absorption rates increase with increasing POM concentrations (Fig. 16.8-A). However, compared with initial ambient concentrations of 1.0 $mg \cdot l^{-1}$, an increase of 0.2 mg $POM \cdot l^{-1}$ nearly doubles the quantity of POM absorbed (Fig. 16.8-B). An increase of 2.0 mg $POM \cdot l^{-1}$ provides ten times the quantity of POM absorbed as under initial ambient conditions. In contrast, when mussels are grown in ambient seston conditions above T, increases in POM have only a minor effect on increasing absorption rates. Therefore, it is clear that additional food from fish faeces and waste feed particles have the potential to significantly enhance mussel growth rates primarily in coastal areas of low (< T) ambient particulate concentrations. In the present study, where ambient seston concentrations exceeded T for most of the trial period, mussels cultured within the farm are unlikely to benefit from fish farm organic wastes.

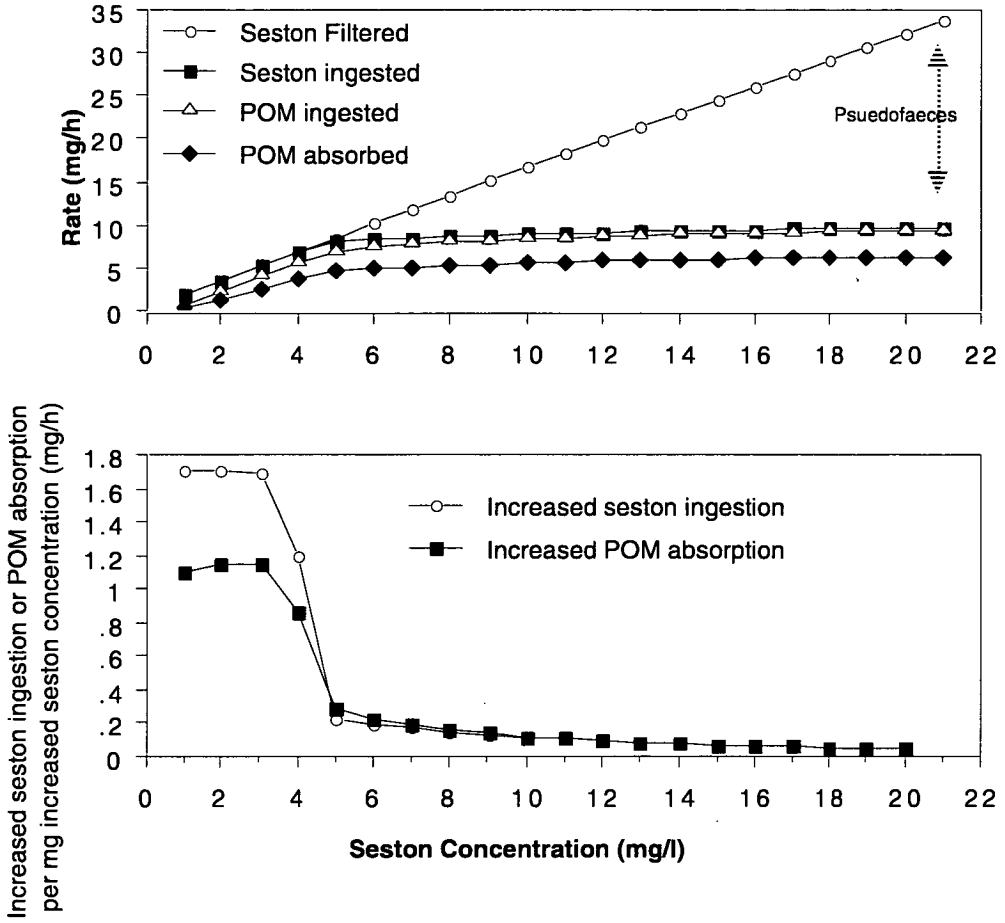


**Figure 16.7.** (Top) Particle filtration, ingestion and absorption rates ($mg \cdot h^{-1}$) of a 1.0 g DMW mussel at increasing seston concentrations ($mg \cdot l^{-1}$) due to 1.0 mg increases in POM concentrations. (Bottom) Increased seston ingestion and POM absorption ($mg \cdot h^{-1}$) per 1.0 mg increase in POM concentrations at increasing seston concentrations ($mg \cdot l^{-1}$). Initial seston concentration = 1.0 $mg \cdot l^{-1}$ at 35% POM. Pseudofaeces threshold concentration = 5.0 mg $seston \cdot l^{-1}$.

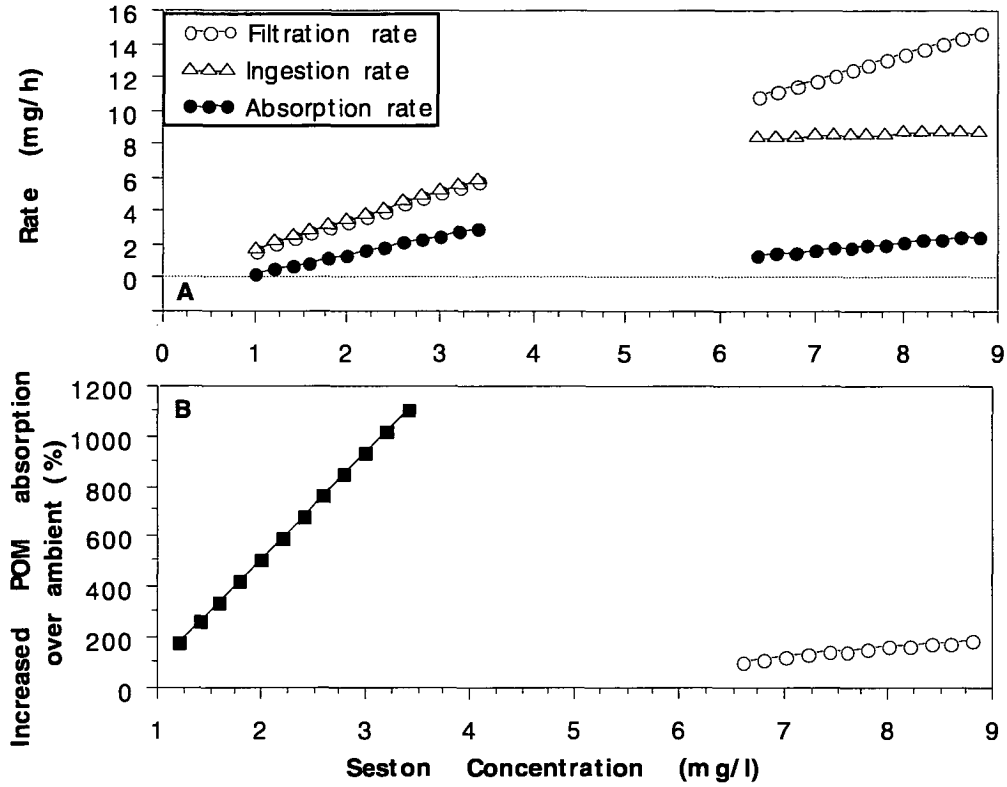


**Figure 16.8.** (A) Seston filtration and ingestion rates and POM absorption rates ($mg \cdot h^{-1}$) of a 1.0 g dry meat weight mussel cultured in ambient seston concentrations of 1.0 and 6.4 mg $seston \cdot l^{-1}$ (35% POM) and exposed to increases of 0.2 mg $POM \cdot l^{-1}$. (B) Corresponding increases in POM absorbed with increasing POM relative to ambient conditions (%). T = 5.0 mg $SPM \cdot l^{-1}$

# Appendix 6. Sources of variation in SFG calculations

Section 12 (Scope for Growth) revealed that calculated SFG values were often greater than the actual growth displayed by mussels during this study. An exploration into the possible causes for this discrepancy was warranted. Conceivable sources of error or variation in SFG calculations responsible for discrepancies between predicted and observed growth include:

- Physiological parameters were derived from literature values, based largely upon laboratory studies using *Mytilus edulis*, which may not be universally applicable to all mussel species, such as *Mytilus planulatus*.
- The reproductive status of mussels may have a significant influence on physiological response parameters.
- Constants used to describe the energy content of mussel biomass and/or food (POM) may not always be appropriate.
- Pre-ingestive and digestive selection are variables often not incorporated in the estimation of ingested and absorbed rations.
- Gross POM energy contents may not reflect "biologically-available" POM.
- SFG calculations do not account for miscellaneous factors that arise in the field such as: competition, biofouling, environmental stressors (eg. pollutants), disease, parasites, or natural aging, all factors that may reduce food availability or the ability of mussels to feed.

### *Physiological Parameters*

The determination of monthly SFG values was based on published physiological relationships rather than measured directly. A wide range of energy budgets and growth models has been presented, based on variable estimates of physiological responses to environmental parameters. Some of these studies suggest different physiological relationships between various intrinsic variables within mussels (eg. size, DMW, condition, age, genotype) and various environmental parameters (eg. temperature, salinity, water circulation, food quantity and quality). Variable physiological relationships reported in the literature include: feeding rates (ie. clearance, filtration and ingestion rates), which have been shown to vary not only with seston concentration but also on the composition of suspended particles (Foster-Smith, 1975; Incze et al., 1980; Lucas et al., 1987; Newell et al., 1989; Navarro et al., 1991; 1996; Riisgard, 1991; Willows, 1992; Bayne et al., 1993; Prins et al. 1994; Hawkins et al., 1998); absorption efficiencies (Widdows 1978a; 1978b; Kiorboe et al., 1981; Thompson, 1984a; Nielsen, 1985; Bayne et al., 1987; 1993; Deslous-Paoli et al., 1990; Willows, 1992; Navarro and Thompson, 1994; Smaal and Widdows, 1994;

Wang and Fisher, 1996; Hawkins et al., 1998); respiration rates and oxygen energy equivalents (Vahl, 1973; Bayne and Widdows, 1978; Riisgard and Randlov, 1981; Bayne et al., 1983; Hamburger et al., 1983; Widdows et al., 1984; MacDonald and Thompson, 1986; Bayne et al., 1987; Tendengren et al., 1990; Sukhotin, 1992; Schluter and Josefsen, 1994; Smaal et al., 1997).

The variation in published physiological relationships may be due to a number of factors: differences in experimental methods, environmental conditions, energy conversion factors, and possibly genetic variation between and within species (Doering and Oviatt, 1986; Gosling, 1992b; Seed and Suchanek, 1992; Widdows et al., 1984). Physiological response parameters determined in laboratory studies may not necessarily reflect those of mussels in the field. Clearance rates, determined in the laboratory using cultured algae, might overestimate *in situ* clearance rates (Cranford and Hill, 1999). The number of possible permutations in calculating theoretical SFG from literature-derived physiological relationships are obviously endless, providing variable SFG estimates according to the chosen relationships incorporated into the calculations.

The variation in physiological responses to environmental conditions underlines the necessity for site-specific studies rather than relying on published values. As the validity of some or all physiological parameters used in the calculation of SFG is uncertain, there is a need for further investigation of the physiological responses of *Mytilus planulatus* to Tasmanian conditions. Without this information, theoretical SFG estimates are suspect, unless validated by observed growth. It seems likely that SFG in NWB is actually much lower than the calculated range. Despite the uncertainty of SFG estimates, the environmental conditions of NWB did promote winter growth of both experimental mussels and overspat; contrasting with many temperate and boreal areas in the Northern Hemisphere.

### *Reproductive status*

The lack of correlation between SFG and actual growth is certainly partially due to the reproductive status of mussels. The most obvious and expected discrepancy between SFG and actual growth occurred during spawning in August 1995 and February 1996, when significant losses of biomass coincided with high SFG values. However, the reproductive condition leading up to spawning may also affect SFG. Bivalves in advanced or ripe stages of gametogenesis may display: higher respiration and ammonia excretion rates (Bayne et al., 1976b; Bayne and Widdows, 1978; Bayne et al., 1983; Smaal and Widdows, 1994), reduced clearance rates (references in Prins et al., 1994; Lefebvre et al., 2000), and reduced efficiencies of nutrient utilisation (Hawkins and

Bayne, 1991). Therefore, it has been suggested that periods of active reproductive development and spawning be avoided when measuring SFG physiological parameters (Seed and Suchanek, 1992; Widdows and Donkin, 1992; Smaal and Widdows, 1994). This recommendation has some support from this study; actual growth and predicted growth corresponded only during the inter-spawning period (September to December 1995). Therefore, the literature-derived physiological relationships may have appropriately described SFG during this inter-spawning period. Prior to spawning (June-July 1995, January 1996), theoretical SFG may have been overestimated due to reproductively advanced or spawning mussels displaying lower clearance rates and higher respiration rates than the literature-derived rates. It is interesting that mussel growth never recommenced after the second spawning as it did after the first spawning. SFG during these last few months of the trial were at maximum levels. Perhaps other factors may have influenced mussel growth during this period, such as post-reproductive stress, higher water temperatures, senility, pea crab infestation, and/or biofouling; factors that may have limited the ability of mussels to feed and grow.

### *Energy Content of Mussels*

It is possible that mussel energy content may have been underestimated, partly accounting for the lower growth increments compared with SFG estimates. However, the mussel soft tissue energy content of 21.0 kJ·g $DMW^{-1}$ used in SFG calculations is within the range (19.5 to 23.7 kJ·g $DMW^{-1}$) of measured *Mytilus* energy values reported in other studies (Winberg, 1971; Dare and Edwards, 1975; Chambers and Milne, 1979; Bayne and Worrall, 1980; Okumus, 1993; Beukema, 1997). Therefore, incorporating 21.0 kJ·g $DMW^{-1}$ into SFG calculations was unlikely to have incurred serious error. The energy diverted into shell and bysuss production may also have been underestimated. However, these energy components are relatively minor compared with meat growth and have typically been ignored in many studies of bivalve production. Therefore, it seems unlikely that an under-estimation of biomass energy content was a major component in the difference between mussel energy increments and theoretical SFG in this study.

### *Pre-ingestive selection*

The calculation of absorbed POM (A) assumed that seston ingested (SI) was in the same proportions of POM/TPM as the ambient sestonic ratio. This assumption does not account for pre-ingestive particle selection, an important component of mussel feeding behaviour (Beninger et al., 1995; Grant, 1996; Hawkins et al., 1998). Particle selection has been observed or implied in numerous studies. Efficiencies of

particle retention and ingestion have been related to a number of possible particle characteristics including size, shape, motility, density, chemical composition, surface properties, and chemical cues such as algal ectocrines (Vahl, 1972; Mohlenberg and Riisgard, 1978; Incze et al., 1980; Jorgensen, 1981; Kiorboe and Mohlenberg, 1981; Shumway et al. 1985; Lucas et al., 1987; Brown, 1988; Sidari et al., 1998; Bayne et al., 1989; Ward and Targett, 1989; Langdon and Newell, 1990; Hawkins and Bayne, 1992; van den Enden, 1994; Hawkins et al., 1996; 1997; Barille et al., 1997; Bougrier et al., 1997; Defossez and Hawkins, 1997; Cheryaev et al., 1998; Ward et al., 1998; Lefebvre et al., 2000). The general consensus from these studies is that mussels and oysters do have some capacity to preferentially select certain particles over others: organic particles over inorganic, algal cells over non-algal particles, and certain algal species over others. Therefore, particle selection may impose qualitative and quantitative modifications to the ambient seston; the characteristics of the ingested ration can be significantly different than the material filtered (eg. higher POM and algal content than seston). As such, the energetic content of the ingested ration may be different to that of the prevailing seston. A generalised term accounting for particle selection has been factored into some models of bivalve feeding behaviour (Willows, 1992; Hawkins et al., 1998). However, mussels may also display compensatory mechanisms; the ability to alter particle selectivity and absorption efficiency according to fluctuations in food quality and quantity (Bayne, 1993; 1998; Bayne et al., 1993), further complicating estimates of selectivity and the characteristics of ingested food particles.

Particle selectivity was not incorporated in the calculations of SFG. However, if this extra component were included, an increase in the percentage of POM ingested would likely have resulted, due to the pre-ingestive selection of organic particles over inorganic particles. This increase in ingested POM would be reflected by higher absorption efficiencies, higher POM absorption rates and, consequently, higher SFG estimates than those calculated without a selectivity component. Therefore, it is certainly unlikely that the discrepancy between observed and predicted growth was due to a missing selectivity factor in SFG calculations. However, this does identify another source of variation among different methods of estimating SFG.

### *Energy content and quality of food*

Particulate energy content was not measured; therefore, the two assumptions regarding food energy content being 23.5 J·mg dry $POM^{-1}$ (Widdows et al., 1979) and constant throughout the year, may be questioned. This value has been used as a energetic conversion factor for POM in several other studies (Widdows and Johnson, 1988;

Page and Ricard, 1990; Smaal et al., 1997) and is similar to other estimates of POM energy content (Widdows, 1985 in Okumus, 1993; Bayne et al., 1987; Okumus and Stirling, 1994). However, dissimilar and/or temporally variable values of POM energy content have been reported in other studies (Vinogradova, 1956 in Winberg, 1971; Bayne and Widdows, 1978; MacDonald and Thompson, 1985; Laane et al., 1987 in Van Haren and Kooijman, 1993; Heral et al., 1993 in Hawkins et al., 1996).

Variations in POM energy content may be expected due to variations in the sestonic composition. POM composition, nutritive value and energy content may vary temporally and spatially according to: algal species and their physiological condition (Platt et al., 1969; Winberg, 1971; Platt and Irwin, 1973; Bougis, 1976; Parsons et al., 1984; Shumway et al., 1987); and the variable proportions of non-algal material such as bacteria, zooplankton (Slobodkin, 1962; Bamstedt, 1981; Morris and Hopkins, 1983), detritus and its origin (Tenore, 1977; Tenore and Hanson, 1980; Tenore; 1981; Tenore et al., 1982; Valiela, 1984; Berg and Newell, 1986); and organic-mineral aggregates (Grant and Bacher, 1998). Therefore, the quality of POM may vary between sites and over various time scales, from tidally to seasonally, according to its composition. It is conceivable that variation in POM composition and overestimates of POM gross energy content during the trial period may partly account for some of the differences between observed and predicted growth. Also, we have previously discussed the unknown influence of selectivity on the characteristics of the ingested ration; as well as altering the type of food ingested, it follows that pre-ingestive particle selection might also alter the gross energy content of the ingested ration from that of seston.

Probably of more practical significance than the gross energy content of the sestonic and ingested POM, which may be temporally variable in themselves, is the actual quality or "biologically-available" energy content of the different components of POM. Variations in available energy content may be due to differences in digestibilities of different food particles; some algae possess highly refractory cell walls and some detrital materials are composed of refractory structural carbohydrates (Kristenson, 1972; Tenore, 1981; Shumway et al., 1985; Langdon and Newell, 1990; Sidari et al., 1998). The selective digestion of certain particles over others has also been demonstrated (Lopez and Levinton, 1987; Shumway et al., 1985; Hawkins and Bayne, 1992). Therefore, the gross energy of sestonic POM may not necessarily be reflected by the energy content of absorbed POM. Due to variable digestibilities, nutrient contents, and a particle sorting mechanism, differential absorption efficiencies (Wang and Fisher, 1996) and growth (Stromgren and Cary, 1984) have been associated with different algal species and mixed diets.

As phytoplankton species (Aquatas data) and nutrient concentrations varied throughout the year, and the detrital fraction of POM may have been substantial (88% to 100% of the POM was estimated to be non-algal in nature); significant variations in POM energy content, nutrient content, and/or digestibility may have been evident. Therefore, it is possible that the POM energy content and/or "biologically-available" energy content may have varied from the constant 23.5 J·mg dry $POM^{-1}$ used in the SFG calculations - possibly accounting for some of the differences between observed and predicted growth.

Mussels, therefore, display considerable variability in the relative utilisation of different suspended particles. This variability stems from preferential retention of particles on the gills, pre-ingestive selection at the labial palps, and differential processing within the gut. Consequently, mussels may impose qualitative as well as quantitative changes on suspended particles in the water column; the relative composition and gross energy content of particles in the seston is not necessarily reflected by the material actually ingested and assimilated. As such, the nutritional significance of mixed seston assemblages may not necessarily be inferred from simplistic categorisations of available food particles such as "POM", "%POM," or "phytoplankton carbon". Grant and Bacher (1998) suggest "the nature of bivalve food particles and their selection is poorly understood and the sensitivity of descriptive models to food quality and feeding requires better knowledge of the variance in suspended food sources and the ability of bivalves to deal with natural particle mixtures."

It is certainly notable that calculated SFG estimates were highly correlated with absorption efficiency ($r = 0.937$, $P < 0.0001$, $n = 176$ observations). Therefore, calculated POM absorption rates (A) and SFG would be extremely sensitive to any error in AE estimates. Considering the variable digestibilities and absorption efficiencies of sestonic particles (Wang and Fisher, 1996), perhaps refinements of AE (not solely a function of %POM in the ingested ration) and absorption rates, are required to produce more realistic estimates of SFG.

### *Other factors*

Several other factors, some discussed in previous sections, may have contributed to limiting mussel growth below levels predicted by SFG calculations. Most of these factors are related to various field conditions that may have reduced the capacity of mussels to obtain food and, therefore, limit their growth. For example, competition for the available food resource from overspat and other filter-feeding organisms, and a reduced food supply within socks due to algal fouling may have been significant.

Fouling was certainly severe during the final months of the trial. Fouling by overspat, tunicates and macroalgae may have been responsible for localised depletion of food particles, reduced water flow, reduced oxygen supply, inhibiting waste removal and, ultimately, limiting mussel growth.

Senility may also have been a factor in limiting mussel growth during the final stages of the trial. Poor mussel performance may be a consequence of reduced filtration rates and metabolic activity in older individuals (Jorgenson, 1976). Declining growth rates, loss of biomass and declining condition, all of which were observed in this study, are often exhibited by mussel populations preceding natural senescence (Jamieson, 1989).

Many other variables have been shown to influence mussel growth. Disease, parasitation, exposure to contaminants, or other variable environmental conditions (eg. salinity, light, current speed, wave action) not accounted for in SFG calculations, may have affected the experimental mussels ability to obtain and utilise food. As there are questions regarding the reliability of the physiological response parameters incorporated into SFG calculations, the degree to which biofouling, senility, or any other factor may have limited growth remains unknown.

***Previous studies***

Studies where mussel physiological response parameters to prevailing environmental conditions were directly measured have generally shown good agreement between observed and predicted growth (Bayne et al., 1979; Bayne and Worral. 1980; Riisgard and Randlov, 1981; MacDonald and Thompson, 1986; Okumus and Stirling, 1994). Therefore, some, or all, of the parameters used in calculating SFG were probably inappropriate and certainly require empirical validation.

One study that calculated theoretical SFG solely from literature-derived physiological parameters is that of Page and Ricard (1990), reporting significant correlations between theoretical SFG and mussel shell growth rates, but ignoring biomass growth. However, these correlations were significant only for time lags of 1 to 4 weeks, with the strongest correlations at time lags of 2 and 3 weeks. The present author has two problems with their results. Firstly, it is not clear how they could realistically compare monthly shell growth rates with weekly SFG values. Assuming growth is continuous in small 20 mm mussels, each monthly shell growth increment would be the cumulative product of food availability and environmental conditions experienced during the entire month. Therefore, it seems erroneous to relate monthly growth with conditions measured on one specific day in a month. Secondly, Page and Ricard (1990) suggest that significant correlations of shell growth rate with theoretical SFG at

time lags of 1 to 4 weeks reflects the time required for metabolic conversion of absorbed energy and nutrients into the shell. However, mussels respond quite rapidly to ingested food rations; assimilated food is usually channelled directly into shell and soft tissue growth within 12 to 24 h of ingestion (Stromgren and Cary, 1984; Nielsen, 1985; Nielsen and Stromgren, 1985). Therefore, any meaningful biological relationship between observed monthly growth and SFG estimated from environmental conditions that occurred in previous weeks seems questionable.

In summary, a number of factors may be responsible for discrepancies between calculated SFG estimates and actual growth displayed by mussels. The exploration of these has highlighted the necessity for direct measurements of physiological parameters rather than utilising values provided in the literature. These measurements need to encompass temporal variations in environmental conditions and the physiological state of cultured bivalves to more accurately assess the potential for bivalve growth over extended periods.