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Bivalves for the remediation of prawn farm effluent: identification of some potentially useful species in Southern Queensland

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Abstract

Several species of oysters, clams and mussels are currently being used around the world to create extra profits and help remediate waste-waters from mariculture operations. To identify opportunities and potentially suitable species of bivalves for remediation of prawn farm effluent in Australia, recent literature dealing with bivalve filtration is reviewed, and species occurring naturally in a banana prawn, *Penaeus* (*Fenneropenaeus*) *merguiensis*, grow-out pond and effluent streams at the Bribie Island Aquaculture Research Centre (BIARC) were collected, identified and assessed in terms of their tolerance of high silt loadings over 3 months. Three bivalve species predominated in the BIARC case study. These were the mud ark, *Anadara trapezia*, the rock oyster, *Dendostrea folium*, and the pearl shell, *Pinctada maculata*. The mud ark demonstrated the highest tolerance of silt loading (99% survival), followed by pearl shells and rock oysters (88 and 63% survival respectively).

Keywords: prawn pond effluent, bivalves, silt tolerance

Introduction

Shellfish aquaculture is considered by many researchers around the world to be an ecologically sustainable activity (Shumway *et al*., 2003). Shellfish have been proposed and used for a variety of applications including the monitoring of ecosystem health (Tolley *et al*., 2003) and the recycling of domestic wastes (Ryther *et al*., 1972), but there is particular interest in their abilities to remediate mariculture effluents (Cheshuk *et al.* 2003; Jones *et al.* 1999, 2001; Lefebvre *et* al. 2000; Lin *et al.* 1993; Shpigel *et al.* 1991, 1993a, 1993b, 1997; Wang 1990, 2003). Their feeding actions and water filtration abilities are seen as a potentially profitable approach to the removal of fine particles such as bacteria, phytoplankton and suspended material from mariculture waste streams. This form of water treatment may be particularly useful for semi-intensive microalgal-based prawn farming operations. Laboratory scale trials have proven this concept and have facilitated projections to scale the process up to a broad-acre approach. However these studies have also highlighted some potential difficulties and economic considerations that need to be addressed before methods could be widely commercialised. For example, Jones and Preston (1999) estimated rates of oyster stocking necessary to significantly treat discharge from Australian prawn farms. They suggested that upwards of 12% of farm area could be necessary to usefully deploy oysters assuming particular exchange rates, stocking densities and treatment levels.

However, filter feeders like oysters have long been known to have difficulties in coping with suspended silt and high sediment loads, which reduce and even arrest feeding processes to effect their filtration capacities and survival (Loosanoff and Tommers, 1948). This is unfortunate since up to 72% of suspended material in prawn effluent can be made up of inorganic matter (Jones and Preston, 1999), as the result of pond wall and bottom scouring by aerators in earthen ponds. Many bivalve genera, as part of their natural feeding actions, sort the smaller digestible particles and expel larger undesirable particles as coagulated pseudofaeces (Jorgensen, 1990). Whilst this indiscriminately removes particles from the water column the process has its limitations. Continued feeding at high levels of suspended material can cost the animal more energy in separating digestible from indigestible particles than it derives from the digestible material, so that growth is reduced. Subsequent reductions in water processing actions during such adverse conditions can further cause growth to slow or become negative (Jorgensen, 1990).

Tolerance of suspended silt is likely to differ greatly between bivalve species depending on their evolutionary traits and natural habitat range. For example, it is generally accepted that estuarine species accustomed to living in turbid environments would have a higher tolerance of suspended solids than oceanic species. The following case study attempts to identify some useful bivalve species for prawn farm discharge remediation systems. To be useful they would need to be tolerant of conditions that occur in prawn production and settlement ponds where dense algal blooms and high levels of suspended particulates can prevail. The level of maintenance required to keep these shellfish working efficiently in a water remediation system is also an important factor in the suitability of species for this type of application. Our approach was to collect and identify local species that occurred naturally in prawn ponds at BIARC, and then subject them to high levels of sedimentation to assess survival and hence their potential for use as low-maintenance wastewater remediation species for prawn farms.

Case study - Materials and methods

Shellfish were collected from a high-density-polyethylene (HDPE) lined prawn culture pond (0.16 ha x 1.6 m deep) at harvest. The pond had initially been stocked with banana prawns (*Penaeus merguiensis*) at a commercial stocking density (30 m^{-2}), and was intensively managed for 11.5 months before harvest (7 January 2003). Shellfish were collected from the bottom and sides of the pond as the water receded and these remained out of water for no more than one hour.

Four shellfish tumblers (Tooltech Pty Ltd., Australia) made from HDPE were used to house the shellfish collected. Each was stocked with variable proportions of the live shellfish with almost total bottom surface coverage. These tumblers were suspended approximately 10 cm below the water surface in a rectangular concrete raceway (20,000 L) supplied with continuous aeration and flowthrough of unfiltered seawater (Figure 1). The raceway also contained several large (>1kg) sea mullet (*Mugil cephalus*), which maintained turbid conditions by continually re-suspending silt that had accumulated in the tank from artificial feeds and intake water. Shellfish inside the tumblers were left undisturbed for 3 months. They were then removed from tumblers and examined for sediment build-up and fouling, survival and the presence of other biota. All shellfish species collected were identified using photographic texts (Coleman, 1975; Rippingale and McMichael, 1961; Lamprell and Healy, 1998). Identifications were confirmed at the Queensland Museum.

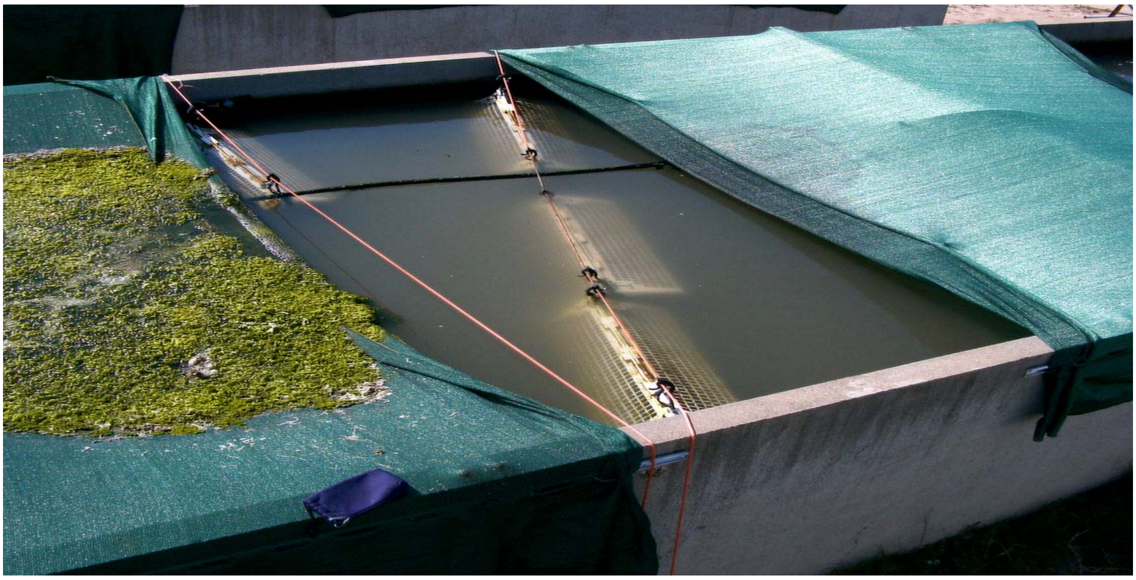

Figure 1. Concrete raceway used to hold shellfish in tumblers

Case study - Results

Four species of bivalves were found to survive and grow at notable levels in this prawn pond at BIARC. These were identified as follows: Sydney cockle or mud ark, *Anadara trapezia* (Deshayes, 1840); the rock oyster, *Dendostrea folium* (Linnaeus, 1758); the pearl shell oyster, *Pinctada maculata* (Gould, 1850); and the white hammer oyster, *Malleus albus* (Lamarck, 1819). Pictures of specimens of each of these species taken from the pond are shown in Figure 2. The internal shell and viscera for these specimens are shown in Appendix 1.

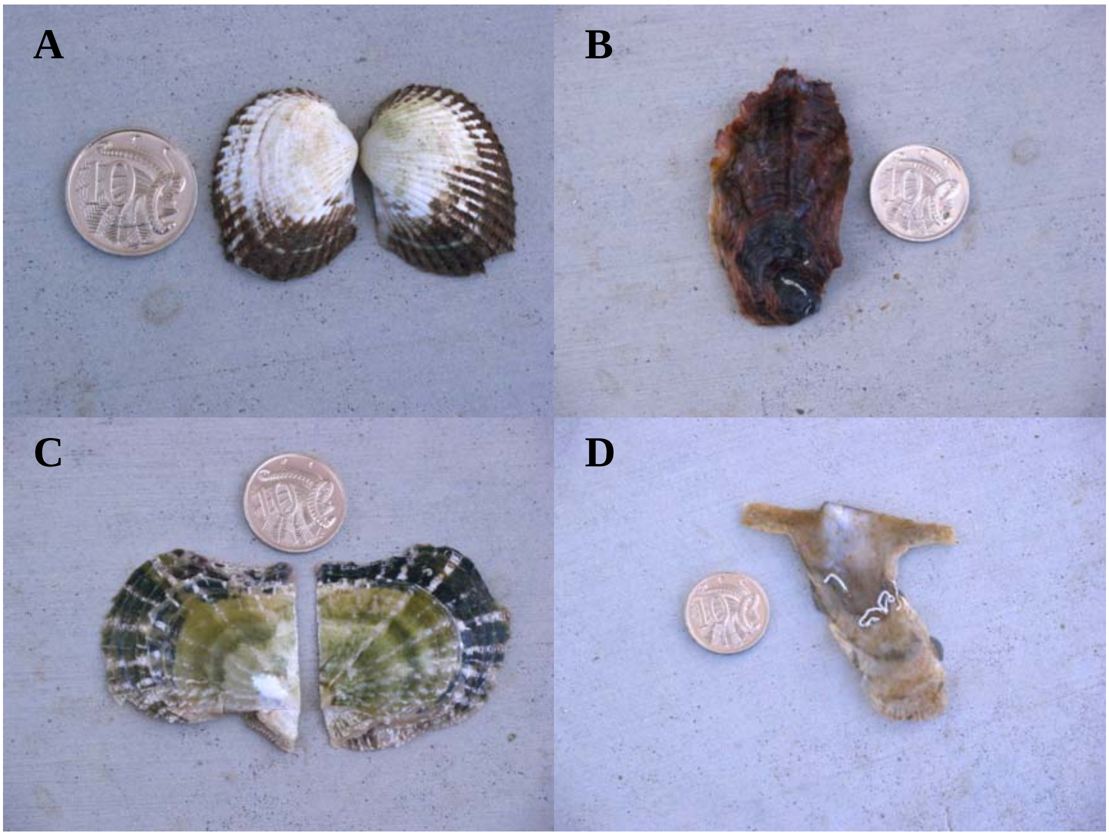

Figure 2. The four main bivalve species found in lined prawn ponds at BIARC. A - mud ark (*Anadara trapezia*), B - rock oyster (*Dendrostrea folium.*), C - pearl shell (*Pinctada maculata*), and D - white hammer shell (*Malleus albus*).

Although some colour differences were found (particularly in the pearl shell oysters – see Appendix 2), the majority of each species had very similar morphology and were assumed to be variations of the same species.

The mud ark was the most common live species collected from the prawn pond (629), followed by the rock oyster (343), the pearl shell (192), and to a much reduced level the white hammer shell (4). Most of these shellfish were collected in and around the pond outlet monk, or on the ridges of folds in the plastic pond bottom. None occurred in the proximity of the anoxic sediment build-up in the centre of the prawn pond. Large numbers of live and dead rock oysters were also found attached to the plastic sides of the pond, just below the water level. Their considerable spat-fall had become apparent soon after commencement of the prawn production cycle, but mortalities in many of these was noted midway through the prawn cycle. At harvest, many of the live oysters on the sides of the pond could not be removed without damaging them so these were not used for further study.

The overall survival rates for shellfish in tumblers during the 3 month trial period is given in Table 1. Raw data for each of the 4 tumblers used is provided in Appendix 3. Mud arks had the highest survival closely followed by the pearl shells. Rock oysters had the lowest survival. The low number of white hammer shells in the study prevents meaningful comparisons.

Table 1. Overall survival of bivalves collected from a prawn culture pond and held in tumblers suspended in turbid conditions for 3 months.

Bivalve species	% survival
Mud arks	99
Pearl shells	88
Rock oysters	63
White hammer shells	75

Tumbler 1 mainly contained rock oysters, and appeared to be the most heavily fouled tumbler at the end of the sedimentation trial period (Figure 3). The oysters were covered in a thick layer of silt to the point that it was difficult to distinguish individual shells within the whole mass. Sprawling sponges, benthic algae and numerous small worms with soft mud casings also covered the shells. In some clumps of oysters there were pockets of mud that also supported large Nereid polychaetes. Many amphipods were also found sheltering between oysters and within dead shells. Upon opening, living oysters were found to contain considerable silt within the viscera.

Figure 3. Tumbler 1 showing the high degree of silt accumulation on rock oysters.

Tumbler 2 contained mainly mud arks and pearl shells with only a few rock oysters. The shellfish in this tumbler were similarly covered with a fine layer of silt and numerous worms with soft mud casings (Figure 4). Some pearl shells were also attached to each other and to the sides of this tumbler (Figure 5). Ascidians and sprawling sponges were also growing on the shellfish and tumbler in low numbers.

Figure 4. Tumbler 2 showing mud arks covered with silt and worms with soft mud casings.

Tumblers 3 and 4 contained a mix of all species collected. Again, a thick layer of silt prevailed with a similar community of fouling organisms. Some pearl shells had also again attached to each other and to the sides of the tumbler.

Figure 5. Tumbler 2 showing the attachment of pearl shells to each other and to the tumbler using byssal threads.

Literature review and discussion

Mechanisms of bivalve filtration

Jorgensen (1990) provides a detailed description of the filtration mechanisms of filter-feeding bivalves. In general, gravimetric, inertial and viscous forces act on suspended particles in ambient water as it is drawn through the mantle cavity and gills with cilia activity. Water entering the interfilament canals of the filter is strained with filaments that are coated with mucus so that suspended particles are retained on impact. Particle handling within these filters is facilitated by ciliary tracts, which produce water currents and collect and direct the passage of particles. Some tracts have fine cilia that retain small and light particles and lead to the mouth. Other tracts have course cilia, which retain larger heavier particles and carry it away for expulsion. Apart from this mechanical sieving of particles, electrical charge is also thought to assist in the capture of fine particles (see Shumway *et al.* 1985). Dissolved organic molecules can also be taken up through the epidermis (Jorgensen, 1990).

Shumway *et al.* (1985) studied the selective ingestion and digestion for six bivalve species in mixed algal suspensions, on the basis that at least 3 mechanisms of selection occur, namely: 1) preferential retention on ctenidial filters; 2) preingestive selection on the labial palps following transfer from the ctendia; and 3) differential absorption in the gut. This work demonstrated that such particle selection mechanisms act differentially on similarly sized particles, either in isolation or in combination, depending on the adaptive differences between bivalve species in their abilities to remove fine particles.

Bivalves are also thought to regulate their filtration rates according to prevailing algal concentrations (Winter, 1975), although the metabolic products of algal cells alone have also been shown to affect oyster feeding (Loosanoff and Tommers, 1948). Very low algal concentrations can cause starvation, and excessively high concentrations arrest growth by stimulating reduced filtration and increased material rejection as pseudofaeces. It is generally accepted that bivalves process available food particles to secure a relatively constant rate of food ingestion. This rate of ingestion is determined by their digestion rates. However, Jorgensen (1990) suggests that widely accepted views about the physiological control of water processing and particle clearances do not account for the high capacity of filter pumps, and that many previous experiments have not provided conditions necessary for demonstration of their full growth potential. Many environmental factors have been shown to affect bivalve filtration rates and efficiencies. These include temperature, oxygen levels, water flow rates, particle concentrations, animal size, biomass densities and species-specific characteristics (Shpigel *et al.*, 1997).

Nutrient cycling by filter-feeding bivalves

Shellfish are low order consumers in natural food chains. They are known to reduce water turbidity and filter organic matter, silt, bacteria and viruses from the water column to directly and indirectly remove nitrogen and other nutrients (Shumway *et al.* 2003). They generate particulate and dissolved materials, which impact on benthic and pelagic habitats. Dame (1996) provides a comprehensive examination of marine bivalve metabolism and nutrient cycling. Where dense aggregations exist they are known to have a pronounced effect on water qualities and nutrient cycling. They process and store both inorganic and organic carbon. Inorganic carbon is available in seawater as calcium carbonate, which is used in the matrix of bivalve's shell biomass. This has relevance to polyculture systems that may be prescribed for treatment and recycling of prawn farm effluent. For example, recent advice from a prominent shrimp farming company in Thailand (Charoen Pokphand Foods) provided at an Australian workshop in July 2003, suggested that when large numbers of mussels or snails are present in shrimp ponds, reduced calcium availability to shrimp can cause them to suffer from soft shells. This effect would be more pronounced in low water exchange or recirculated systems, where the addition of calcium as lime or dolomite may be required to increase calcium availability and maintain the health of shrimp.

Oyster reefs are considered highly heterotrophic estuarine processors, filtering large quantities of particulate organic carbon from the water column and releasing both particulate and dissolved organic carbon (Dame, 1996). Benthic bivalves can also rapidly recycle nitrogen in coastal waters. Particulate organic nitrogen is consumed mainly as phytoplankton, and released as particulate organic nitrogen, ammonium and dissolved organic nitrogen in the form of amino acids and urea (Dame, 1996). Positive feedback loops exist between phytoplankton and bivalve populations, since both benefit from this nutrient recycling, but areas beneath bivalve beds also receive nitrogen from bivalve deposits to cause an accumulation of nitrogen in sediments and influence biogeochemical processes. For example, Kaspar *et al.* (1985) found higher levels of nitrogen in the sediments under green-lipped mussel (*Perna canaliculus*) farms than at a control site. Although nitrite and nitrate levels were similar, the ammonium pool had been doubled. Decomposition of the organic nitrogen in deposits and a higher ion-exchange capacity of the sediments due to the increased organic matter content were thought to be responsible for this observed stimulation of remineralisation processes. The increased flux of pre-digested organic material supplied to sediment decomposers by bivalves is thought to greatly accelerate the cycling of nutrients through these ecosystems.

Grant *et al.* (1995) also documented much higher ammonium releases from sediments under mussel (*Mytilis* edulis) lines that had been used over 10 years, compared with a reference site. Whilst the impacts of biodeposits from bivalve aggregations on the benthos will greatly depend upon site-specific factors like localised hydrodynamics and sediment types, these studies demonstrate that a partial shift towards anaerobiosis can be expected in the long term.

Asmus and Asmus (1991) questioned the utility of bivalves to reduce phytoplankton biomass in another study of a mussel (*Mytilus edulis*) bed during summer in the North Sea. The results suggested that if all of the ammonium released by the mussel bed were taken up by phytoplankton, that the primary production induced by this nutrient release would be higher than the amount of phytoplankton consumed. The reason for this is thought to be due to the mussels consuming particulate organic matter other than phytoplankton, such as small zooplankton and detritus, or through an additional uptake of amino acids from the water column. Thus, nitrogen other than that incorporated in phytoplankton can be mobilised by bivalves to accelerate primary production. On the other hand, dense aggregations of bivalves have been shown in a number of studies to reduce chlorophyll concentrations in surrounding waters (eg: Shpigel and Blaylock, 1991; Jones *et al.*, 2002) and control phytoplankton production (see Jorgensen, 1990).

There has been comparably less research conducted into the effects of bivalves on phosphorus cycling, but the available evidence suggests that oysters such as *Crassostrea gigas* remove significant amounts from natural waters and deposit most of this in sediments (eg: Sorin *et al.*, 1986: cited in Dame, 1996). Dame *et al.* (1989) found that 8% of the total phosphorus uptake by dense *C. virginica* beds was released as orthophosphate, and that the uptake of phosphorus was proportionately higher than nitrogen and carbon uptakes predicted by the classical Redfield[1] ratio. Marine diatoms are a major component of

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[1] General atomic ratio for carbon, nitrogen and phosphorus in phytoplankton is 108:15.5:1 (Redfield *et al.* 1963).

the coastal phytoplankton communities utilised by bivalves and are also a major sink for other dissolved nutrients like silica, which are essential to coastal marine life. It is generally believed that the biodeposition of nutrients caused by filter feeding bivalves, and the resulting increased remineralisation in sediments provides increases in the productivity and stability of estuarine ecosystems (Dame, 1996).

Bivalve filtration rates and efficiencies

There has been considerable research over the last 50 years investigating the feeding efficiencies of various commercially important shellfish. Bivalves are generally thought to retain small particles (1µm) less effectively than larger particles (4µm). However different species have varying abilities to utilise fine particles (Shumway *et al*., 1985). Wisely and Reid (1978) studied the removal of different particle sizes and concentrations of cornflour and riceflour by Sydney rock oysters (*Crassostrea commercialis*). They found that particles of cornflour <5 µm appeared optimal for ingestion. Their work also suggested that riceflour particulate (<5 µm) concentrations of <2 mg L^{-1} produced faeces but very little pseudofaeces. At 18 mg L^{-1}, 50% of the oysters were also producing pseudofaeces, and at 35 mg L^{-1} all were producing faeces and pseudofaeces. Particle sizes of <5 µm are thought to be most suitable for this species (Wisely and Reid, 1978).

Tenore and Dunston (1973) investigated the effects of different phytoplankton concentrations on feeding and biodeposition for three species. These were the blue mussel *Mytilus edulis*, the American oyster *Crassostrea virginica* and the hard clam *Mercenaria mercenaria*. For all species, low algal concentrations caused low feeding rates, and clearance rates increased with algal concentrations, as did biodeposits. The clams were less efficient at removing particulate matter than the oysters, which had the most efficient assimilation of food, and the particle size of suspended material increased after oyster filtration. These bivalves were shown to be more efficient when exposed to natural phytoplankton levels, and these authors pointed to the resulting ecosystem shifts towards deposit-feeding pathways that could be expected when exposing bivalves to eutrophic systems in constructed ecosystems.

The effects of suspended silts on oysters' (and mussels') feeding activities were documented many years ago (eg: Loosanoff and Tommers, 1948; Lossanoff, 1962). These early investigations found that silt concentrations of 100 mg L^{-1} reduced average pumping rates (and therefore feeding) by over 50%, and concentrations of 3000-4000 mg L^{-1} reduced it by over 90%, or caused the bivalves to close their shells completely. Minute quantities of silt alone have been shown to stimulate pumping activity in *Crassostrea virginica*, possibly due to mechanical stimulation of the gills (Loosanoff, 1962), but at higher concentrations, shell movements become vigorous as gills are frequently cleansed, while large quantities of pseudofaeces are expelled. In contrast, the work by Winter (1975), with the mussel *Mytilus edulis,* suggested that suspended silt concentrations of 2.5-100 mg L^{-1} did not reduce tissue weight gain. In fact, a silt concentration of 12.5 mg L^{-1} increased the dry tissue weight by 32% when optimal algal cell densities were also supplied. Shell weight also increased with increased silt loading. This later study concluded that low levels of silt, which resulted in accelerated growth, stimulated the mussels' feeding activity.

More recent work by Shin *et al*. (2002) with the green-lipped mussel *Perna viridis*, showed that this species also has a high tolerance of suspended solids for short periods (1200 mg L^{-1} for 96 hr), but noted resultant permanent gill damage that was likely to exert sublethal effects in the long-term. The morphology of their ctenidia was affected by increased sediment loadings, and these mussels were suggested to temporarily increase their pumping rate to compensate for the impaired filtration and maintain their level of feeding. *M. edulis* has been suggested by Smit (2000) as the best species to remove particulates in some saltwater systems. Smit reports the development of algae and water quality control measures at a Dolfinarium in the Netherlands, and quotes 80% removal of all suspended organic matter in filter prototypes supplied with 25 L $hour^{-1}$ kg^{-1} mussel biomass.

Shpigel and Blaylock (1991) measured the filtration efficiency (FE) of the Pacific oyster *C. gigas* in fish-pond effluent by comparing the levels of particulate organic nitrogen (PON) in the inflow and outflow of their effluent treatment systems. These authors also measured filtration rates (FR) provided by *C. gigas* according to the following formula: $FR = [(PON_{in} - PON_{out})/W] \times$ (flow rate in L h^{-1}) where W = total oyster wet weight. They found that in 25 L conical tanks stocked with 20 g L^{-1} total oyster wet weight (ie: 500 g $tank^{-1}$), filtration efficiencies declined with increasing flow rates. Highest efficiencies were achieved during warmer (25-32°C) months with exchanges of 2 tank volumes h^{-1}, when PON levels were more than halved (51%). Doubling and trebling flow rates during summer reduced FE's to 41% and 17% respectively. FE's during winter (11-19°C) ranged from 34 to 12% with similar flow rates. Conversely, filtration rates increased with increasing exchange rates (2-6 tank volumes h^{-1}) during summer from 10.6 to 14.3 μg PON g^{-1} whole oyster h^{-1}. Filtration rates were not significantly affected by flow rates during cooler months. Oysters used in this work had mean sizes of 4.2 and 6.5 g in winter and summer respectively.

On the other hand, Bougrier *et al.* (1995) found that the maximum clearance rates for *C. gigas* were achieved at a temperature of 19ºC, whilst oxygen consumption increased with water temperatures (5-32ºC tested). However, the mortalities noted at 32ºC in this study, along with the oxygen consumption data, suggested that the acclimation period used may have been too short to reflect the species' tolerances of extreme conditions.

The growth of *C. gigas* in fish farm effluent was studied further by Lefebvre *et al.* (2000). They investigated the relative values of fish faeces (from *Dicentrarchus labrax*) and the diatom *Skeletonema costatum* as organic food sources for this oyster. Absorption efficiencies were high (56%) for the fish faeces but higher (66-70%) for the diatom. The results showed that the diatom was preferentially ingested in mixed suspensions. Clearance rates were not affected by the types of food offered, but were generally reduced when the oysters were reproductively mature. Under optimal conditions, the oysters demonstrated abilities to remove almost 100% of the faecal particles (4-5 µm) from the effluent, despite its dilution with inorganic clay particles from the earthen pond.

The filtration abilities of the pearl oyster, *Pinctada margaritifera* have also been studied by Pouvreau *et al.* (1999). They compared the retention efficiencies, clearance rates and gill areas of this species to a range of other bivalves. This species is commercially exploited in seemingly adverse-food conditions with low phytoplankton levels, typical for coral atoll lagoons of French Polynesia. It was found to have low (15%) retention efficiencies for small (1µm) algal particles (eg: cyanobacteria) but high efficiencies (98%) for slightly larger (5µm) algal particles (eg: *Chaetoceros gracilis* and *Isochrysis galbana*). The size of animals did not appear to affect these efficiencies, and nor did the type of experimental system used (closed verses flowing). The species was found to have high relative gill size and a high pumping rate making it well adapted to oligotrophic waters with low turbidity.

Integrating bivalves into biological systems like prawn farms

Molluscs form an integral part of natural macrobenthic assemblages (O'Hara, 2001), and are an important food source for many inshore species with high commercial value (eg: *Penaeus merguiensis*; Chong and Sasekumar, 1981). As discussed earlier, filter-feeding bivalves can greatly affect their environment through controls over primary production by phytoplankton grazing and biodeposition of silt and detritus (Jorgensen, 1990). Their abilities to remove suspended particles including microalgae from the water column and to reduce nutrients in waters to be discharged or recycled is well recognised. It is for these reasons that they have been proposed by many workers for use as remediators of nutrient rich mariculture effluent. Additionally, marine microalgae have been shown to effectively grow and remove inorganic nutrients from a variety of waste streams (eg: primary sewage effluent: Craggs *et al.*, 1995; land-based fish farm effluent: Lefebvre *et al.*, 1996), so linkages between phytoplankton and bivalve production systems may also have many future applications in other areas (domestic and industrial effluents).

Many authors have proposed the production of valuable secondary crops of bivalve molluscs from shrimp production ponds. Walker *et al.* (1991) for example cultured bay scallops, *Argopecten irradians concentricus*, under varying conditions in shrimp ponds in South Carolina, U.S.A. The results suggested that this was biologically feasible (up to 77% survival, good growth and limited fouling), but the economics of this activity were considered uncertain. Other bivalves have already been incorporated into shrimp-farm designs that employ complete recirculation in Taiwan and Thailand (Hopkins *et al.* 1995). Although the bivalves may not represent a proportionately large sink for nutrients in these systems, they can assist in removing phytoplankton and other non-settleable materials from the water column.

The high levels of bivalve production that may be possible within shrimp ponds is demonstrated in the work by Hopkins *et al.* (1993). They stocked a *P. vannamei* pond with hard clams *Mercenaria mercenaria* and oysters *C. virginica*. The clams were placed on the pond bottom (3.4×10^6 ha^{-1}) and the oysters were placed on trays and on the pond bottom (total of 1 x 10^6 ha^{-1}). Survival and growth of the shrimp (10.6 t ha^{-1}) was not affected by the substantial biomass of bivalves produced (approximately 7 t ha^{-1} each for clams and oysters). In terms of water quality differences between this polyculture, and a monoculture of prawns used as a control, suspended particulate matter including phytoplankton, BOD, nutrient levels and pH in the water column were measurably lower in the bivalve pond, whilst dissolved oxygen was higher.

More than 20 years ago, Scura *et al.* (1979) reported rapid growth and a reduction of the commercial grow-out period for oysters cultured in an intensive semi-closed raceway system. Three species of oysters were then under production, namely *Crassostrea virginica*, *C. gigas* and *Ostrea edulis*. Marine microalgae (*Thalassiosira* sp., *Chaetoceros* sp. and *Isochrysis* sp.) cultured in earthen ponds were recirculated through raceways where oysters were held in stacked trays. Good food chain efficiency was demonstrated and over 88% of the microalgae were removed from water in the oyster raceways (supplied with 3,000 L min^{-1}). *C. virginica* was reported to produce the best results, with mortalities in the order of 9.5% from stocking 2 mm seed to market size of 55 g in 10 months. However, it is believed that this and other attempts at land-based bivalve culture failed due to the excessive cost and low reliability of mass-producing the microalgae feed (Jakob *et al.*, 1993). Better prospects were seen in utilising the large quantities of microalgae wasted with discharge from shrimp ponds, although pond water flow rates and oyster biomass relationships would need to be optimised (Jakob *et al.*, 1993).

Wang (1990) provided a conceptual model of a shrimp/oyster co-production system that may produce two highly priced products, whilst reducing the level of system discharge. Although it was considered feasible to design a zero discharge system with these components, Wang emphasised that his group (in Hawaii) did not advocate a closed system due to perceived difficulties in balancing minor elements, and due to the inevitable need to add water periodically. Their focus was on removing algae and suspended solids from shrimp pond water, so that it could be reused in the shrimp ponds to dilute algal blooms. Other considerations given in this paper included mechanical sedimentation to remove suspended matter including oyster faeces, and the provision of oxygen to large oyster beds that would require continuous flow.

Sandifer and Hopkins (1996) have also provided a conceptual design for a sustainable shrimp culture system. The model consists of 6 components which incorporate almost total water recycling. The components of this proposed 4 ha production system are as follows:

1) 3 x 1 ha shrimp ponds (*Penaeus vannamei* or *P. setiferus* stocked at 100 m^{-2});
2) 1 x 1 ha oyster (*Crassostrea virginica* at 10^6 ha^{-1}) – mullet (*Mugil cephalus* at 7000 ha^{-1}) polyculture /water treatment pond;
3) A small phytoplankton inoculation pond used to culture desirable algal blooms to seed production ponds when necessary;
4) A solids settling basin used to settle solids removed from the ponds periodically as a water sludge mixture, with settled water returned to the production ponds;
5) Sludge dewatering beds, constructed with a perforated plastic underlayer;
6) Dewatered sludge usage on agricultural land.

The shrimp in this system would be produced on a 140-160 day cycle. Between shrimp crops (through winter) an aquatic pasture dominated by seaweed (*Enteromorpha* sp.) and invertebrates such as gammarid amphipods, copepods (*Acartia tonsa*) and polychaetes (*Capitella capitella, Polydora cornuta*) is allowed to develop in the ponds, being driven by residual nutrients. These organisms provide natural food for shrimp restocked in the next cycle. The oysters and mullet used in this design are maintained as 2 separate year classes, with half of the stock harvested each year. The oysters would be grown on racks suspended 60 cm above the pond bottom at a minimum density of 500 m^{-2}. Targeted production levels were 40 tonnes of shrimp per year (75% survival), 500,000 oysters per year (>95% survival), and >7 tonnes of mullet per year (≥90% survival).

Early work integrating bivalves with prawn ponds in Australia was conducted by Maguire *et al.* (1981) at Port Stephens in New South Wales. Those studies compared the growth and condition of the Sydney rock oyster (*Crassostrea commercialis*) on continuously submerged trays in estuarine ponds stocked with school prawns (*Metapenaeus macleayi*), and in an intertidal culture lease. Shell growth at the two sites was similar, but very high meat condition developed in the prawn ponds. Occasional problems in shell and meat growth in the prawn ponds were attributed to slow water flow past the oyster beds. Better meat condition was produced using low stocking densities (14,182 ha^{-1}), but even at high densities (347,273 ha^{-1}) significant fattening resulted in marketable condition. However, significant mortalities of oysters occurred in one large-scale polyculture trial. Maguire *et al*. noted that fouling of oysters with silts and spat-fall, and water current circulation were important aspects for integration into farm designs and management. Interest by this group of researchers also extended at that time to the fattening of oysters in fertilised ponds filled with seawater (Nell, 1985). The algal blooms that developed fed oysters placed in floating pontoon trays, so that when the water was >20ºC, poor oysters fattened to marketable condition in about four weeks. Although Sydney rock oysters can take 3 years to achieved market size (40-60 g), triploid oysters have recently been shown to grow faster in their second year, especially when they are produced from selected lines (3rd generation marketable 9 months earlier) (NSW Fisheries, 2002b).

Jones and Preston (1999) furthered this work in Australia by investigating the effects on water quality of the Sydney rock oyster as a means to filter prawn (*P. japonicus*) pond effluent. The study involved stocking 3 densities (low – 8, medium – 16, and high – 24) of oysters (55g) into 34-litre tanks, using an effluent retention period of 2 hours, and measuring resulting total suspended solids, organic and inorganic matter, total nitrogen, total phosphorous, bacteria and chlorophyll *a* concentrations. Dead shells were used as controls in the experiment, and the effluent contained 72% inorganic and 28% organic matter. All parameters tested were reduced significantly by oyster filtration, with the highest density having the most pronounced effects on water qualities. Suspended solids in this experimental system were reduced by up to 51%, bacterial numbers by up to 42%, total nitrogen by 20% and total phosphorous by 33%. Calculations based on 20% exchange rates per day suggested that 0.12 ha of treatment pond area containing 120,000 oysters of similar size and density (100 m^{-2}) would be necessary to treat discharge from a 1 ha prawn pond to a similar level.

Various other species of oysters have been proposed as biological filters for mariculture effluent. Shpigel and Blaylock (1991) demonstrated the use of the Pacific oyster *Crassostrea gigas* to reduce excessive phytoplankton from fish ponds, so that seawater usage was halved in the production of gilthead sea bream. This work conducted in Israel, produced commercial sized oysters in 14 - 18 months, but highlighted the need to consider the combined temperature and salinity tolerances of the species used. Stressed Pacific oysters in this study (>27°C and >41 ppt.) produced an increase in amino nitrogen (dissolved organic nitrogen), which represents a metabolic energy drain that lowers growth rates and can result in mortalities. Further work by this group showed that *C. gigas* grew faster and had better condition when grown in water supplied from an earthen sedimentation pond, than in water from a PVC-lined pond (Shpigel *et al*., 1991; 1993b). The reasons for this difference were thought to be higher algal diversity providing a better balance of nutrients, presence of benthic diatoms attached to suspended particulate inorganic matter, and more stable algal booms in the earthen pond.

Further biomechanical-filtration studies using two species of bivalves were undertaken by Shpigel *et al*. (1997) for fish-pond effluent. Different and complementary filtration capacities of two bivalve species (*C. gigas* and *Tapes philippinarum*) were combined with mechanical sedimentation in two flow-through reactors with different water mixing patterns. One was a laminar flow system described as a plug flow reactor (PFR), where longitudinal flow remained relatively unmixed. The other was a continuously stirred flow reactor (CSFR) that used aeration to maintain homogenous mixing of particulate matter between the intake and outlet ports. These reactors (15 cm wide x 80 cm long x 12 cm high) each contained 14.4 L of water and were stacked with trays of bivalves (1 kg biomass in each reactor using 3-7 g animals).

Better performance (10-20%) in terms of particulate matter and algae removal was demonstrated in the PFR compared with the CSFR. Comparisons between similarly sized *C. gigas* and *T. philippinarum* suggested that *C. gigas* was more efficient at total particulate removal (inflow concentrations of 5-35 mg L^{-1} dry weight). Small *C. gigas* (5-10 g) were also shown to be significantly more efficient at removing suspended particulate matter than large ones (20-25 g). Combining juveniles of both species was suggested as an optimal treatment approach, and predictive particle removal models for both reactor types are presented. Whilst the laminar flow reactor was shown in that study to be better at removing suspended particulates than the homogeneously mixed reactor, differential growth rates could be expected in filter-feeding bivalves within plug flow systems due to diminishing food resources in the water as it progresses along the laminar filtration system (Jorgensen, 1990). This could be especially pronounced if flow rates through such a reactor were slowed to provide maximum filtration levels. From a marketability perspective, variably sized bivalves are likely to reduce a crop's commercial value as a food grade product. Thus, a selective trade-off exists between the better economic returns that a CSFR may provide, and the higher environmental values inherent with a PFR.

Although research into recirculating systems for prawn farms has focussed recently on bacterial floc production systems (eg: Burford *et al.*, 2003), microalgal based systems may offer a number of advantages over such systems, as described recently by Wang (2003). These include the high nutrition that microalgae can provide to oysters for creation of additional profits, and control or reduction of bacterial and viral diseases of prawns from the microalgae's antibacterial properties. Wang (2003) states that a breakthrough in diatom production technologies at the University of Hawaii is providing controls over algal species continuously generated in open systems. This is facilitating the design of a recirculating oyster/shrimp system, which uses the marine diatom *Chaetoceros* sp. as an intermediary that incorporates excess nutrients from the shrimp's production.

An important aspect of this integrated design is the balance between resources needed by the shrimp, algae and oysters. Wang (2003) suggests that this balance can be best achieved by building in steady states for biomass and resource utilisation efficiency. In work so far conducted, this is achieved by dividing the facilities' production into many staged units. Frequent harvests and restocking of units within the facility maintains relatively consistent biomasses of the two animals, so that the rate of waste discharge can remain constant over time. Wang reports that two commercial systems using this concept have been developed recently in Hawaii. One produces shrimp and clam seeds and the other produces shrimp brood stock and clam seeds.

Combining bivalve and macrophyte culture systems may be particularly effective at removing nutrients from mariculture effluents (Hopkins *et al.* 1995). Following considerable background work with the isolated components, Shpigel *et al.* (1993a) proposed an integrated treatment system for fish culture effluent using bivalves and seaweed. The four components of this system are 1) phytoplankton-rich fish culture, 2) sedimentation facility, 3) bivalve (*C.* gigas and *Tapes semidecussatus*) culture, and 4) seaweed (*Ulva lactuca*) culture. The system operates sequentially as above with treated effluent flowing back to the sea, although water is also recirculated between the settlement and bivalve facilities. Nitrogen budgets developed from previous studies suggest that 63% of the nitrogen added as feed can be recovered in various harvested forms (26% fish, 14.5% bivalve and 22.4% seaweed), with the remainder wasted as settled faeces (32.8%) and discharge back to the sea (4.25%). For every 100m^2 of fish pond, the model requires 50 m^2 of sedimentation, 33 m^3 of bivalve troughs and 42 m^2 of seaweed culture area. This system was expected to produce 1kg of fish, 3 kg of bivalves (suitable for human consumption) and 7.8 kg of seaweed from 3 kg of fish food.

Shpigel and Neori (1996) later described an environmentally friendly integrated system for the culture of seaweed (*Ulva lactuca* or *Gracilaria* spp.), abalone (*Haliotis tuberculate*), clams (*T. philippinarum*) and fish (*Sparus aurata*). At least half of the nitrogen supplied to the system was incorporated in harvestable yields with a general approach of "controlled eutrophication". Projected revenues increased with the complexity of the modular systems, but figures generated were highly volatile depending on the value of the particular species utilised. The more complex designs incorporating all of these species required more land area, greater infrastructures, a much higher level of technical knowledge and labour, and was reliant on poorly understood microbial processes.

Enander and Hasselstrom (1994) have also reported an experiment utilising a bivalve/macrophyte combination to treat shrimp farm effluent in Malaysia. The bivalve used was the hairy cockle *Scapharca inaequivalvis*, and the macrophyte used was *Gracilaria* sp. These species were selected on the basis of their tolerance of dehydration, and of likely variations of water salinity, pH, and temperature. The cockles performed well, increasing their biomass by 27% in one month. However, the *Gracilaria* sp. did not grow well as it became fouled with the green algae *Enteromorpha* sp. Nevertheless, the system removed 83% of the phosphate, 61% of the total phosphorous, 81% of the ammonium, 19% of the nitrate, and 72% of the total nitrogen from the effluent stream.

Mussels have also been proposed as potential remediators of prawn farm effluent. Lin *et al.* (1993) suggests that economic, social and ecological values were realised in a pilot farm-scale trial where the green mussel (*Perna viridis*) was grown in the effluent stream of an intensive prawn (*P. monodon*) farm in Thailand. Additional seafood was produced and water qualities were reportedly improved (though not studied in detail). These mussels grew from 12.2 to 42.4 g during the 113-day culture cycle with 85% survival.

Cheshuk *et al.* (2003) deployed the Tasmanian blue mussel *Mylilus planulatus* on open-water salmon (*Salmo* salar) cages to determine whether enhanced mussel performance could be demonstrated. Such demonstration would indicate that integrating mussel culture practices could reduce nutrient loading of the environment surrounding the fish farm. However, no differences between mussels grown within the farm lease area and control sites were demonstrated. Reasons for this finding included several factors affecting the access of mussels to particulate waste emanating from the farm. Bodvin *et al.* (1996) proposed a similar and perhaps better system for open-ocean fish cages in sheltered localities. This modelled system utilises floating enclosed cages that direct discharge waters through floating enclosures containing mussels and seaweed. Theoretical calculations suggested that 112.5 tonnes of mussels are needed to completely filter 60 m^3 of water min^{-1}. Nitrogen budgets projected for this filtration suggested that 25% would be incorporated into mussel biomass, and the rest would be excreted as faeces (25-30%) or dissolved matter (45-50%).

Biologically integrated systems for nutrient removal can also be assisted by simple mechanical means. A good example of this is the approach taken by Jones *et al.* (2001). They recognised some of the difficulties presented by effluent from shrimp ponds and tested a sequential system of oyster (*Saccostrea commercialis*) filtration (24 hr) and macroalgae (*Gracilaria edulis*) absorption (24 hr), preceded by natural sedimentation (24 hr) to assist in suspended solids removal. The authors noted higher than normal suspended solids at the farm during the time effluent was sourced for testing, and that 95% of the suspended particles were in the size range 2-4 µm. Sedimentation reduced total suspended solids by 88%, total Kjeldahl nitrogen by 30%, total phosphorus by 53% and chlorophyll *a* by 28%. This increased the relative proportion of organic particles from 23 to 31%, thereby enhancing its food value to the oysters. Importantly, the results of several researchers, who have demonstrated a degree of tolerance or even improved growth of bivalves with low levels of suspended silt (eg: Loosanoff, 1962; Winter, 1975), suggests that not all of the inorganic suspended particles need to be removed from prawn farm effluent before filtration with bivalves.

Further work by Jones *et al.* (2002) demonstrated that recirculating the water through oyster beds provided significantly better filtration. That study confirmed that oysters may be more effective at filtering prawn farm effluent in a flowthrough system than in a still water system, and that multiple passes of water through the system improved the process further.

Hussenot *et al.* (1998) also conducted some interesting studies that incorporated the use of shellfish into integrated effluent treatment systems for extensive and intensive land-based fish farms. They compared three basic approaches, namely: 1) treatment by wastewater retention lagoons; 2) treatment by foam fractionation; and 3) treatment by microalgae and bivalve filter feeders. The microalgae/ bivalve system involved continuous stimulation of naturally occurring diatoms to reduce TAN levels, followed by removal of this phytoplankton with pacific oysters (*C.gigas*) or other filter feeders. These authors concluded that a combination of all of the basic approaches taken in the study were appropriate for inclusion because they each catered for different elements in the effluent. The retention lagoon removed particulate matter, foam fractionation removed dissolved organics, and the microalgae/bivalve reactor reduced inorganic nutrients and phytoplankton. Although the positioning of each of these components had not been optimised, a theoretical approach to their use to treat and recirculate wastewaters in intensive systems is discussed.

Mortalities and disease issues in bivalves

High levels of mortality in bivalves have been attributed to prolonged exposure to high concentrations of inorganic turbidity (Winter, 1975). Several studies have reported mortalities of oysters when they are suspended in heavy siltation zones (Jones *et al*., 2002) or placed on pond bottoms (Hopkins *et* al. 1993). Maguire *et al*. (1981) reported serious losses of this nature in trials with the Sydney rock oyster in turbid prawn ponds, and to avoid fouling Jones *et al*. (2002) recommend allowing prawn farm effluent to settle for 6 hours to reduce particulates prior to biofiltration with this oyster species.

As the ponds at BIARC are lined ponds, lower levels of inorganic silt prevail than at commercial farms where aeration erodes pond bottoms and embankments. Nevertheless, significant mortalities occurred in rock oysters (*Dendrostrea folium*) during the prawn production cycle used for the present study. Whilst inorganic sediments were probably not responsible in this case, this species may be susceptible to other factors like elevated pH (up to 8.84 during the mid-crop period) or inappropriate phytoplankton diversity.

Mudworm (*Polydora* sp.) infections are well known to cause mortalities, and have a pronounced affect on the market value of oysters (Maguire *et al*., 1981; Beattie, 2002). However, infections do not immediately cause mortalities. Despite notable infection rates in some of the trials conducted by Maguire *et al*. (1981), mortalities of oysters grown in prawn ponds were not attributed to mudworms, because few of the oysters that died had shell blisters typically caused by *Polydora* sp. Mortalities in that study were thought to be more caused by high pond-water temperatures (up to 33ºC) and the accumulation of decaying material on the oysters. Nevertheless, oysters grown in prawn ponds are known to suffer high mud worm infection rates (eg: 75% in the work by Hopkins *et al*., 1993). Within the conceptual design of a shrimp/oyster/mullet integration proposed by Sandifer and Hopkins (1996), the oysters would be exposed to air regularly by pumping between ponds, so that oyster parasites like this boring mud worm and a boring sponge (*Chione* sp.) were controlled. Management regimes that may reduce or remove mudworm infections from oysters include regular cleaning of shells to remove settled silts, and routinely culturing them high in the water column away from bottom sediments, so that tidal fluctuations expose oysters to a regular dryout. More extreme treatments include periodic desiccation (7 - 10 days in cool air) or freshwater exposure (2 days), but care with these is recommended when oysters are not in optimal health (Beattie, 2002).

Other predatory species, which cause mortalities in cultured oysters, mussels and clams around the world, includes Stylochid flatworms from the primitive phylum of marine animals known as Platyhelminthes (see Jennings, 1996). Several new species have been identified in Moreton Bay, but *Imogine mcgrathi* is the main species known to cause problems to oyster growers in Queensland and NSW (Jennings, 1996; Beattie, 2002; NSW Fisheries, 2004b). It is reportedly more prevalent during times of prolonged high salinity, and more inclined to feast on young spat. Oyster drills or borers are also common pests in bivalve cultures along the east coast of Australia. There are several different species of this carnivorous gastropod, which breach the bivalve's shell by various means to digest and extract its meat. These include the mulberry whelk *Morula marginalba*, the tingle whelk *Xymene hanleyi*, and the hairy whelk *Monoplex australaisae* (Beattie, 2002).

Prevailing pond water temperatures are a critical limiting factor for selecting appropriate shellfish species, both in terms of optimising filtration activities and overall survival. *C. gigas* for example has been shown to be somewhat intolerant of tropical summer temperatures (32ºC: Bougrier *et al*., 1995). Heat mortality can be expected when oysters are exposed to the hot sun for five hours or more (Beattie, 2002). Potter and Hill (1982) investigated heat mortality in the Sydney rock oyster and methods to control overheating during prolonged exposure to the sun at low tide. Tissue temperatures were shown to rapidly rise during direct sun exposure (by 23ºC over 90 min) regardless of whether the oysters were clean or coated with mud. Lethal temperatures (upper median) were determined experimentally to be 41, 45 and 47ºC for 24-, 5- and 2-h exposures respectively. In sunlight, oyster temperatures were somewhat reduced by covering with shade cloth (2-4ºC) and spraying with water.

Winter mortality in the Sydney rock oyster is caused by the protozoan parasite *Mikrocytos roughleyi* (NSW Fisheries, 2004b). However this problem is not likely to affect oysters grown at prawn farms because it generally only affects areas south of Port Stephens where prawns are not normally grown. Whilst this problem has been reduced by more than half in triploid Sydney rock oysters, a small degree of brown discoloration of the gonad during warmer months has been observed in these treated animals (NSW Fisheries, 2002b).

QX disease in Sydney rock oysters is also caused by a protozoan parasite (*Marteilia sydneyi*) that has genetic variants from the Great Sandy Straits in Queensland to the Georges River in NSW (Kleeman *et al*., 2004). This pathogen has greatly affected oyster production in Queensland and NSW in the past, particularly those areas that are in close proximity to muddy substrates (Beattie, 2002). Outbreaks occur on a yearly cycle and tend to coincide with warmer months. An unknown intermediate host is thought to be involved in this parasite's life cycle.

Mortalities associated with viruses have also been documented for several bivalve species. For example, Lipart and Renault (2002) have detected a herpes-like virus in *Crassostrea gigas* spat. They developed DNA probes and an in situ hybridisation protocol that can be applied to confirm the presence of this ubiquitous family of viruses on histological sections.

Human consumption, health and translocation concerns

Oysters are used in many countries to monitor biological contaminants and the health of ecosystems (Tolley *et al.*, 2003). World-wide shellfish related food poisoning incidences have stimulated voluntary and enforced testing programs for culture environments and produce at national and international levels. Parameters of main concern include levels of human faecal indicator bacteria, heavy metals, petroleum hydrocarbons, organohalogens, and algal biotoxins (Rodgers, 2001). Shellfish harvested from areas with previously identified risks for human health are required to be cleansed of contaminants with approved depuration procedures prior to sale. Licenses are generally not issued to culture oysters in Queensland in high risk areas where water qualities are heavily influenced by sewage, domestic or industrial runoff (Beattie, 2002).

In Queensland, shellfish harvested from prawn farm settlement ponds have not been broadly proven suitable for human consumption. Conservative restrictions apply at present due to the chances of humans ingesting toxins accumulated in the shellfish during the grow-out phase. Many different types of algae can proliferate in these eutrophic systems and the accumulation of toxic algae is of major concern to health authorities. Further work is needed which identifies the risks, tests such produce to develop credibility ratings, and develops a code of practice that can be applied in practical terms (personal communication: J.B. Burke, Safe Foods Qld). The advantages of a food safety plan of this nature include an ability to recall product if problems are suspected, avoidance of litigation if unforeseen problems occur, better access to markets, and uniform culture and processing procedures (Burke, 2000). The National Health and Medical Research Council has a code of practice in place for commercial estuarine filter-feeding bivalves (oysters and mussels) that are grown in Australia for human consumption (NHMRC, 1987). With industry support, this could be used to guide future considerations regarding the sanitary and depuration requirements for shellfish grown in prawn farming systems.

The work by Shumway *et al.* (1985) provides insight into how toxic dinoflagellates can be concentrated in bivalves. The oyster *Ostrea edulis* selectively cleared the dinoflagellate *Prorocentrum minimum* from a mixed algal suspension. These authors point to the potential for preferential digestion of different algal species ingested, so that viable cells are egested. Concentration of these toxic algae can also occur on the bivalve's ctenidia. Arnott (1998) provides a summary of toxic marine microalgae, their biotoxins, and shellfish poisoning in the Australian context.

Bacterial contaminations are also of concern, particularly since humans often consume bivalves without cooking. Gram-negative facultative anaerobes like *Vibrio* and *Pseudomonas* commonly inhabit the digestive tract of bivalves, as mediated by the sources being the surrounding water and their food (Gatesoupe, 1999; Prieur *et al*., 1990). This is of particular concern in coastal areas affected by urban pollution, where potentially pathogenic bacteria (eg: *Escherichia coli*, *Salmonella*) can accumulate in the digestive tract (Prieur *et al*., 1990).

Lee *et al.* (2003) conducted investigative work aimed at modifying conditions in a mariculture sedimentation pond so that populations of the ubiquitous dinoflagellate *Amphidinium* sp. (known to produce organic solvent-soluble toxins) are minimised. This was particularly pertinent since this species becomes concentrated in close proximity to the sediments where some bivalves are feeding. This dinoflagellate was found to be out-competed by diatoms when the silica:nitrogen ratio was kept at 1:1 or greater. This approach of supplementing effluent treatment ponds with silica and other trace elements was suggested as one way to integrate molluscs for human consumption into mariculture operations. Further research, and vigilance regarding culture conditions was nevertheless recommended.

The potential also exists to internalise the use of bivalve-remediator biomass within establishments as additional food sources for stock. This creates production systems that are increasingly detached from wild resources, and avoids any dangers posed through direct ingestion of contaminated bivalves by humans. An example of this approach might be where waste nutrients from Penaeid cultures are taken up by microalgae, the microalgae are used to feed molluscs, which in turn are used as natural feed, conditioning or maturation diets for the Penaeids. This is seen in the work of Lombardi *et al.* (2001). They describe an experimental system for the brown mussel (*Perna perna*) grown on sea cages, which are used to feed prawns (*Litopenaeus vannamei*) in Brazil. Fresh mussel meat is used to supplement or replace expensive commercial pellets whilst live animals left on the mesh of cages assist in the clean up of organic wastes from the cages.

Translocation issues are also of concern where bivalves are introduced and proliferate in environments that are devoid of their natural predators. There are numerous accounts of mussels invading new areas, displacing native species, and becoming a pest through coating equipment in contact with seawater. The black-stripped mussel, *Mytilopsis sallei*, has destroyed pearl farms in other countries, and was discovered near the Port of Darwin in the Northern Territory in 1999 (Field, 2002). This and other species of problematic mussels (eg: Asian green mussel *Perna viridis*) have since been detected on the hulls of vessels entering the Port of Darwin. Costly monitoring and control programs have been implemented to safeguard this area against such infestations (Field, 2002). A risk assessment process should therefore be applied to the proposed introduction of species into farming operations. The use of locally occurring species with similar utility may be the preferred approach.

Potential bivalve species for use in Southern Queensland

Not all bivalve species will be suitable to integrate into prawn farming systems. One locally relevant example of this is the eugarie or beach pipi (*Donax deltoides*), which occurs in sandy substrates around Australia's high-energy coastlines. They are commonly used as food for prawn broodstock in Australia, and are routinely fed to such prawns at BIARC after nutritional enrichment with live cultured microalgae. These shellfish can quickly clear water containing high concentrations of several microalgal species of varying sizes, including *Chaetoceros mullerii*, *Nanochloropsis occulata* and *Tetraselmis chuii*. These algal species are typical of the different types and size ranges of phytoplankton that occur in prawn ponds, which can represent a large portion of the nutrients discharged in wastewaters. However, *D. deltoides* is never found in ponds at BIARC, despite there being healthy populations directly adjacent to the centre's intake pipes. As the free-living trochophore larvae from this naturally occurring bivalve have unrestricted access to these culture ponds, the reason for their absence on the site is most likely associated with their habitat preferences. Many other molluscan candidates for bioremediation uses also exist in Moreton Bay, and their natural occurrence in prawn culture ponds can be used as a guide to species that tolerate such conditions. These may be of use in wastewater remediation pursuits in the future, particularly if their larval stages can be stocked directly into farm conditions with reasonable survival.

Maguire *et al*. (1984) also studied the macro-benthic fauna of brackish water prawn ponds on the mid New South Wales coast. Of the 64 macro-benthic faunal species that occurred in close proximity to the pond water supply intake point, only 17 were recorded from any of the four ponds utilised. Three non-commercial bivalve species were recorded in core samples from one pond (mean of 0.6 m^{-2} and maximum of 31.8 m^{-2}), namely *Ennuncula* sp., *Notospisula trigonella* and *Wallucina assimilis*, and another three, *Laternula creccina*, *Sanguinolaria donaciodes* and *Tellina deltoidalis,* were noted as occurring in these ponds periodically, and sometimes at high densities (*L. creccina* up to 115 m^{-2}). These and other small bivalve species that occur regularly in prawn ponds could be useful as natural fodder after the prawns attain a size that facilitates predation. However, the advantages to prawn crops and potential problems that such bivalve species create in prawn ponds (eg: microalgae + calcium depletion and bloom management) have not been evaluated.

Sea mullet (*Mugil cephalus*) also have been proposed as potentially useful species for reducing organics in prawn farm sedimentation ponds (Erler *et al*., 1999). Their foraging activities over bottom sediments cause bioturbation in restricted habitats, and in the BIARC case study they provided the silty conditions in which shellfish harvested from the prawn pond were challenged. It is clear from the high survival rate that mud arks can survive under conditions of high silt loading with little maintenance. This may provide the option to seed directly onto pond bottoms, thus avoiding specially constructed facilities to accommodate their farm integration. This finding is not altogether surprising because they are often found naturally buried in mud flats in estuarine systems in New South Wales and Queensland (Coleman, 1975). Cockles may be of greater future potential in this regard than rock oysters, because they may require less pre-settlement of suspended solids before they are effective at removing phytoplankton and bacteria from the effluent. Mud arks also have a solid shell wall making them resistant to damage when handled.

At BIARC, tumblers containing mainly mud arks (Tumblers 2 and 4) appeared to have less silt accumulation, and were generally less fouled than tumblers with mainly rock oysters (Tumblers 1 and

3). This may simply have been due to the rounded shell shape of mud arks being less conducive to silt accumulation on the shell surface. Rock oysters, on the other hand, are cup-shaped, and those collected in this study were often attached to each other so they had a more convoluted surface and greater area for silt accumulation. In Tumblers 1 and 3 this occurred to such an extent that pockets of mud formed between adjacent shells, which enabled large Nereid worms (about 10 cm long) to live between the shells. Amphipods also seemed to prosper in these conditions, and were mainly found in tumblers with many rock oysters. Amphipods and polychaetes have also been found to predominate in studies of the sediments under bivalve farms, where biodeposits strongly influence the nature of bottom sediments (eg: Chamberlain *et al.* 2001).

Cockles and clams are harvested for human consumption in many parts of the world. In Galicia in North West Spain for example, about 16,000 tons of clams are traded each year (Jara-Jara *et* al., 1997). Effluent from an intensive turbot (*Psetta maxima*) culture in Spain has been evaluated by Jara-Jara *et al.* (1997) as a nutrient source for fattening and growing hatchery produced seed stock of the clam *Ruditapes decussates*. A starting density of 2.5 kg m^{-2} was used in trays in a series of tanks supplied with variable effluent-flow rates (100, 200 and 400 % h^{-1}). Differences in the growth of seed stock were not significant ($P>0.05$) between tanks with different flow rates and the condition index was also similar between tanks. However, the condition of clams grown in the effluent was higher than those grown in natural conditions, as were all biochemical components studied, including total carbohydrates, free reducing sugars, polysaccharides, total lipids, neutral lipids, phospholipids, proteins, ashes, and organic matter.

The cockle found to proliferate in the prawn culture ponds at BIARC, *Anadara trapezia*, is known to grow to 66 mm shell length, and occurs from southern Western Australia, through Tasmania, Victoria and New South Wales to northern Queensland (Lamprell and Healy, 1998). Along the Pacific North-west coast of Canada there has also been recent interest in the aquaculture potential of the heart clam or basket cockle (*Clinocardium nuttallii*). That work to date has shown *C. nuttallii* to be hardy following out-planting in a variety of habitats. Seed were successfully produced in hatcheries with studies including broodstock collection and conditioning, spawning, nursery and grow-out culture phases, and it is envisaged as a new commercial shellfish species for British Columbia (Clayton, 2003). Work conducted recently by Mebane *et al.* (2003) suggest that seedstock for hard clams like *Mercenaria mercenaria* can be over-wintered relatively easily using recirculating culture systems.

The pearl shell oyster, *Pinctada maculata*, also appeared to have a relatively high survival rate in the BIARC case study. This species grows to 55 mm shell length, and occurs from New South Wales to the Torres Strait (Lamprell and Healy, 1998). They did not seem to accumulate large amounts of silt, possibly because of their apparent ability to relocate themselves. Even though the pearl shells were stocked onto the bottoms of tumblers, later examination found them attached with byssal threads to the sides, up to 15 cm above the rest of the shellfish. This indicates that they have the ability to move around to find preferred habitats. Others had formed clumps of shells on the bottom of the tumblers but these did not appear to accumulate as much silt as the rock oysters. The southern pearl shell *Pinctada fucata*, is a similar species which occurs in estuaries from New South Wales to Queensland. It is known to prefer a muddy environment and is usually found attached to seagrasses or clumps of dead shells. It lives from low tide level to 30 m in depth, grows to 85 mm and is common (Coleman, 1975).

Pearl oysters (*Pinctada* sp.) have been proposed for profitable environmental remediation activities by Gifford *et al.* (2004). These authors' contention is that the cosmopolitan distribution of pearl oysters would provide opportunities to employ endemic *Pinctada* species in many parts of the world. This application is further supported by their high water pumping rates and high protein levels compared with other bivalves, offering good potential to remediate nutrient and toxic chemical loads. The accumulation of heavy metals and organopollutants such as pesticides in their shells and fatty tissues respectively, suggests they may be useful at removing low levels of such contaminants from natural waters. As their flesh is not generally consumed, the dangers to human health following ingestion are less of an issue, whereby the economics of such an operation is reliant rather on pearl production. However, their utility as a remediator of mariculture effluent is less clear, and the production of saleable pearls in the eutrophic environment of a prawn farm has not been demonstrated.

Several oyster species are farmed for pearls in Australia. These include the gold or silver lipped pearl oyster *P. maxima* (dominant northern-most species), the black lipped pearl oyster, *P. margaritifera*, the Shark Bay pearl oyster, *P. albina*, the winged oyster, *Pteria penguin,* and the Akoya pearl oyster, *P.*

imbricata (Love and Langenkamp, 2003). Most production occurs in waters off Western Australia, and this was reportedly worth \$175m in 2001-02 (NT = \$30m and Qld = \$0.49m in 2000-01) (Love and Langenkamp, 2003). The Akoya pearl oyster is a more temperate species that occurs widely around the world and along most of the NSW coast. Some encouraging pearl production trials have been conducted and are continuing with this species at Port Stephens (NSW Fisheries, 2002a).

Whilst pearl oysters appear to have a higher potential to accumulate nitrogen in their flesh than other bivalves that are grown for human consumption (reviewed in Gifford *et al*., 2004), large amounts of flesh would need to be harvested from the system regularly to remove appreciable quantities of nutrients from prawn farm discharges. The species which showed tolerance of prawn pond conditions in our case study is relatively small, and for pearl production to be realised, the oysters would probably need to be grown to a much larger size, and therefore possibly through several crops of prawns. Continuous on-farm prawn production cycles would be required to provide an uninterrupted culture environment, or alternatively, holding facilities would be needed to maintain them between crops where prawn production is seasonal. Pre-adaptions to oligotrophic environments as described for *P. margaritifera* by Pouvreau *et al.* (1999) may assist with maintenance in this second option. Nevertheless, the present study suggests that at least one species of pearl oyster (*P. maculata*) can tolerate prawn farm conditions to some degree, and therefore may be worthy of further investigations. Specially designed systems that facilitate their useful and profitable integration into prawn farms would be needed. Important considerations in such designs include low labour maintenance (eg: defouling), retaining or locking-up biodeposits for effective nutrient removal from the aquatic system, and an ability to blend clean seawater with effluent to avoid potentially catastrophic events with heavily invested stock. Depending on their inorganic silt tolerances, pre-settling of effluent may be necessary at farms utilising earthen ponds.

The rock oyster found in the BIARC prawn pond, *Dendostrea folium*, grows to 100 mm shell length, and occurs from Queensland to northern Western Australia (Lamprell and Healy, 1998). Like the other bivalves collected in the case study, it is edible, but it is not presently recognised as a commercial species. According to the results of this study and previously published work summarised above (eg: Jones *et al*., 2002), pre-settlement of prawn farm effluent and regular washing to remove fouling material may be necessary to achieve high survival of some edible oysters in prawn culture and treatment ponds. Sufficient evidence is certainly available to assert this for the Sydney rock oyster, but there are several other species of edible oysters that are commercially cultured in Australia which have not been tested in this regard. Edible oysters cultured in more temperate areas in Australia include the Sydney rock oyster, *Saccostrea glomerata* (also known as the Moreton Bay rock oyster and formerly known as *S. commercialis*), the Pacific oyster, *Crassostrea gigas* (prohibited in Queensland), and the flat oyster, *Ostrea angasi.* Species occurring in the subtropics are mainly harvested from wild spatfall and include the milky oyster, *S. cucullate* (previously referred to as *S. amasa*), and the blacklip oyster, *Striostria mytiloides* (previously referred to as *S. echinate*) (Beattie, 2002; Love and Langenkamp, 2003). *S. glomerata* is mainly produced in New South Wales and Queensland (worth \$29.6m and \$0.5m in 2001-02 respectively), *C. gigas* production is spread across South Australia, Tasmania, and New South Wales (worth \$13.3m, \$11.6m and \$2.0m in 2001-02 respectively), and the subtropical species *S. cucullata* and *Striostria mytiloides* are grown in Queensland from Hervey Bay to Mackay (worth \$56,700 in 2001-02). Culture of *O. angasi* is still in the start-up phase in New South Wales, and was valued at \$64,000 in 1999-2000 (Love and Langenkamp, 2003).

No mussels occurred in the prawn pond at BIARC. Nevertheless there are several subtropical endemic species that could be used like *Perna viridis* in Thailand (Lin *et al*., 1993, reviewed above). World production of mussels incorporating several species amounted to some 1,318,000 tonnes in 2000, but in Australia, the main mussel species commercially cultured in southern states is the native blue mussel *Mytilus galloprovincialis* (3,036 tonnes valued at about \$8m in 2001-02) (Love and Langenkamp, 2003; NSW Fisheries, 2004a). *M. edulis planulatus* is also commonly known in Australia as the blue mussel, and is thought to have been introduced into Western Australia and subsequently spread through all southern states (Aquaculture WA., 1995). These species would likely be unsuitable for prawn farm integration due to their preference of more temperate waters.

The white hammer oyster *Malleus albus* is a filter-feeding bivalve that lives on mud flats or muddy sand flats, and ranges from New South Wales and Queensland through the Northern Territory to northern Western Australia (Coleman, 1975; Lamprell and Healy, 1998). Although only small specimens were found in the pond at BIARC, it can grow up to 150mm in length. Whilst it survived in the turbid conditions of this case study, the low numbers studied make it difficult to assess their relative potential for wastewater remediation. The high surface area and fragile nature of the shell (see Figure 2) suggests handling damage could be an issue, and the low flesh yield apparent in this study would likely limit nutrient accumulation for removal.

Conclusion

The prawn farming industry in Australia currently relies almost entirely on miroalgal-based outdoor pond systems. These autotrophic systems produce large quantities of microalgae from waste nutrients, which is lost with discharges, to sometimes create nutrient loading concerns in receiving waters. All growth stages for molluscs and many other species in open mariculture food chains (eg: zooplankton: Brown *et al*., 1997) rely heavily on the high nutritional values of microalgae. Ecological systems with an abundance of microalgae, like prawn farm settlement ponds used in Australia to reduce nutrient levels prior to discharge, could be used to generate considerable profits if methods to convert such nutritional resources into marketable products could be reliably implemented.

This study provides background information that may be useful in future considerations of bivalves for profitable bioremediation uses in Australia. It provides a preliminary assessment of some bivalve species that may be useful, particularly in Southern Queensland. There are many other potentially useful species that have been proposed for similar assessments during the course of this work. Those species, which occur naturally in the vicinity of farm effluent streams, may provide guidance to species selection in different locations.

Engineering and business plan modifications will be necessary to accommodate bivalve filtration and production systems in existing farm structures. Primarily, a desire to diversify into multi-species production systems will need to be realised. Once candidate species have been identified and screened for suitability to particular applications, systems optimisation for particular purposes will be necessary. Further, organisational developments will need to occur if seed supplies are to service industry uptake and expansion. Effective purging and depuration systems would also need to be proven and implemented as part of production activities for shellfish grown in prawn farm effluent for human consumption. These long-term initiatives are well within the capabilities of existing mariculture operations in Australia, and a strong case to pursue this direction is presented herein.

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References

Aquaculture WA. (1995). Blue mussels *Mytilus edulis planulatus. Publication by the Fisheries Department of Western Australia* **No. 5** ISSN 1323-8442.

Arnott, G.H. (1998). Toxic marine microalgae: a worldwide problem with major implications for seafood safety. Advancing Food Safety Technical Paper **February**: 24-34.

Asmus, R.M. and H. Asmus. (1991). Mussel beds: limiting or promoting phytoplankton? *J. Exp. Mar. Biol. Ecol.* **148**: 215-232.

Beattie, K. (2002). Oyster culture in Queensland. *Queensland Department of Primary Industries Information Series*. ISSN 0727-6273. 28pp.

Bodvin, T., M. Indergaard, E. Norgaard, A. Jensen and A. Skaar. (1996). Clean technology in aquaculture – a production without waste products? *Hydrobiologia* **326/327**: 83-86.

Bougrier, S., P. Geairon, J.M. Deslous-Paoli, C. Bacher and G. Jonquieres. (1995). Allometric relationships and effects of temperature on clearance and oxygen consumption rates of *Crassostrea gigas* (Thunberg). *Aquaculture* **134**: 143-154.

Brown, M.R., S.W. Jeffrey, J.K. Volkman and G.A. Dunstan. (1997). Nutritional properties of microalgae for mariculture. *Aquaculture* **151**: 315-331.

Burford, M.A., P.J. Thompson, R.P. McIntosh, R.H. Bauman and D.C. Pearson. (2003). Nutrient and microbial dynamics in high-intensity, zero-exchange shrimp ponds in Belize. *Aquaculture* **219**: 393-411.

Burke, J. (2000). Introduction to the oyster risk identification workshop. *Report on oyster risk identification workshop held at the Centre for Food Technology, 7 July 2000.* Centre for Food Technology, Hamilton, Brisbane. Queensland Government Publication.

Chamberlain, J., T.F. Fernandes, P. Read, T.D. Nickell and I.M. Davies. (2001). Impacts of biodeposits from suspended mussel (*Mytilus edulis* L.) culture on the surrounding surficial sediments. *ICES Journal of Marine Science* **58**: 411-416.

Cheshuk, B.W., G.J. Purser and R. Quintana. (2003). Integrated open-water mussel (*Mytilus planulatus*) and Atlantic salmon (*Salmo salar*) culture in Tasmania, Australia. *Aquaculture* **218**: 357-378.

Chong, V.C. and A. Sasekumar. (1981). Food and feeding habits of the white prawn *Penaeus merguiensis*. *Mar. Ecol. Prog. Ser.* **5**: 185-191.

Clayton, W.E.L. (2003). Cockle project provides aquaculture option for British Columbia First Nations. *Global Aquaculture Advocate* **October Issue**: 52-54.

Coleman, N. (1975). What shell is that? Lansdowne Press, Australia. 308 pp.

Craggs, R.J., V.J. Smith and P.J. McAuley. (1995). Wasterwater nutrient removal by marine microalgae cultured under ambient conditions in mini-ponds. *Wat. Sci. Tech.* **31 (12)**: 151-160.

Dame, R.F. (1996). Ecology of marine bivalves: an ecosystem approach. CRC Press. New York.

Dame, R.F, J.D. Spurrier and T.G. Wolaver. (1989). Carbon, nitrogen and phosphorus processing by an oyster reef. *Marine Ecology Progress Series* **54**: 249-256.

Enander, M. and M. Hasselstrom. (1994). An experimental wastewater treatment system for a shrimp farm. *INFOFISH International* **4**: 56-61.

Erler, D.V., P.C. Pollard, M. Burke and W. Knibb. (1999). Biological remediation of aquaculture waste: a combined finfish, artificial substrate treatment system. *In*: Kumar, M.S. (ed.) National Workshop on Wasterwater Treatment and Integrated Aquaculture Production. 17-19 September 1999. South Australian Research and Development Institute, West Beach, Adelaide, South Australia.

Field, D. (2002). Northern Territory vs marine invaders. *Austasia Aquaculture* **April/May 2002**.

Gatesoupe, F.J. (1999). The use of probiotics in aquaculture. *Aquaculture* **180**: 147-165.

Gifford, S., R.H. Dunstan, W. O'Connor, T. Roberts and R. Toia. (2004). Pearl aquaculture-profitable environmental remediation. *The Science of the Total Environment* **319 (1-3)**: 27-37.

Grant, J., A. Hatcher, D.B. Scott, P. Pocklington, C.T. Schafer and G.V. Winters. (1995). A multidisciplinary approach to evaluating impacts of shellfish aquaculture on benthic communities. *Estuaries* **18 (1A)**: 124-144.

Hopkins, J.S., R.D. Hamilton II, P.A. Sandifer and C.L. Browdy. (1993). The production of bivalve molluscs in intensive shrimp ponds and their effect on shrimp production and water quality. *World Aquaculture* **24 (2)**: 74-77.

Hopkins, J.S., P.A. Sandifer and C.L. Browdy. (1995). A review of water management regimes which abate the environmental impacts of shrimp farming. *In*: C.L. Browdy and J.S. Hopkins (eds.). Swimming through troubled water, Proceedings of the special session on shrimp farming, Aquaculture '95. World Aquaculture Society, Baton Rouge, Louisiana, USA.

Hussenot, J., S. Lefebvre and N. Brossard. (1998). Open-air treatment of wastewater from land-based marine fish farms in extensive and intensive systems: current technology and future perspectives. *Aquatic Living Resources* **11 (4)**: 297-304.

Jakob, G.S., G.D. Pruder and J.-K. Wang. (1993). Growth trial with the American oyster *Crassostrea virginica* using shrimp pond water as feed. *Journal of the World Aquaculture Society* **24 (3)**: 344-351.

Jara-Jara, R., A.T. Pazos, M. Abad, L.O. Garcia-Martin and J.L. Sanchez. (1997). Growth of clam seed (*Ruditapes decussates*) reared in the wastewater effluent from a fish farm in Galicia (N.W. Spain). *Aquaculture* **158**: 247-262.

Jennings, K.A. (1996). The reproductive biology of *Imogine mcgrathi* (Platyhelminthes: Polycladida) associated with oysters from Moreton Bay. University of Queensland Honours Thesis.

Jones, A.B. and N.P. Preston. (1999). Sydney rock oyster, *Saccostrea commercialis* (Iredale & Roughley), filtration of shrimp farm effluent: the effects on water quality. *Aquaculture Research* **30 (1)**: 51-57.

Jones, A.B., W.C. Dennison and N.P. Preston. (2001). Integrated treatment of shrimp effluent by sedimentation, oyster filtration and macroalgal absorption: a laboratory scale study. *Aquaculture* **193**: 155-178.

Jones, A.B., N.P. Preston and W.C. Dennison. (2002). The efficiency and condition of oysters and macroalgae used as biological filters of shrimp pond effluent. *Aquaculture Research* **33**: 1-19.

Jorgensen, C.B. (1990). Bivalve filter feeding: hydrodynamics, bioenergetics, physiology and ecology. Olsen and Olsen, Fredensborg, Denmark, 140 pp.

Kaspar, H.F., P.A. Gillespie, I.C. Boyer and A.L. MacKenzie. (1985). Effects of mussel aquaculture on the nitrogen cycle and benthic communities in Kenepuru Sound, Marlborough Sounds, New Zealand. *Marine Biology* **85**: 127-136.

Kleeman, S.N., R.D. Allard, X. Zhu and R.B. Gasser. (2004). Mutation scanning analysis of *Marteilia sydneyi* populations from different geographical locations in eastern Australia. *Molecular and Cellular Probes* **(In Press)**.

Lamprell, K. and J. Healy. (1998). Bivalves of Australia. Volume 2 Backhuys Publishers, Leiden.

Lee, J.J., M. Shpigel, S. Freeman, O. Zmora, S. Mcleod, S. Bowen, M. Pearson and A. Szostek. (2003). Physiological ecology and possible control of a toxic marine dinoflagellate, *Amphidinium* sp., from the benthos of a mariculture pond. *Aquaculture* **217**: 351-371.

Lefebvre, S., L. Barille and M. Clerc. (2000). Pacific oyster (*Crassostrea gigas*) feeding responses to a fish-farm effluent. *Aquaculture* **187**: 185-198.

Lefebvre, S., J. Hussenot and N. Brossard. (1996). Water treatment of land-based fish farm effluents by outdoor culture of marine diatoms. *Journal of Applied Phycology* **8**: 193-200.

Lin, C.K., P. Ruamthaveesub and P. Wanuchsoontorn. (1993). Integrated culture of green mussel (*Perna viridis*) in wastewater from an intensive shrimp pond: concept and practice. *World Aquaculture*, **24 (2)**: 68-73.

Lipart, C. and T. Renault. (2002). Herpes-like virus detection in infected *Crassostrea gigas* spat using DIG-labelled probes. *Journal of Virological Methods* **101**: 1-10.

Lombardi, J.V., H.L.A. Marques and O.J.S. Barreto. (2001). Floating cages in open sea water: an alternative for promoting integrated aquaculture in Brazil. *World Aquaculture* **September 2001**.

Loosanoff, V.L. and F.D. Tommers. (1948). Effect of suspended silt and other substances on rate of feeding of oysters. *Science* **107**: 69-70.

Loosanoff, V.L. (1962). Effects of turbidity on some larval and adult bivalves. *Proc. Gulf. Caribb. Fish. Inst.* **14**: 80-94.

Love, G. and D. Langenkamp. (2003). Australian aquaculture: industry profiles for selected species. ABARE eReport 03.8, Prepared for the Fisheries Resources Research Fund. Commonwealth of Australia.

Maguire, G.B., P.J. Gibbs and L.C. Collett. (1984). The macro-benthic fauna of brackish water prawn farming ponds at Port Stephens, New South Wales. *The Australian Zoologist* **21 (5)**: 445-458.

Maguire, G.B., B. Wisely and M.E. Skeel. (1981). Cultivation of the Sydney rock oyster *Crassostrea commercialis* (Iredale and Roughley) in prawn farming ponds. *Aquaculture* **24**: 63-75.

Mebane, B., Roberts, S. and Lindell, S. (2003). Researchers develop low-tech recirculating culture system for quahog clams. *Global Aquaculture Advocate* **December**: 35-36.

Nell, J.A. (1985). Pond fattening of oysters. *Agfact F2.1.2, first edition*. New South Wales Department of Agriculture. ISSN 0725-7759. 2 pp.

NHMRC. (1987). Code of hygienic practice for oysters and mussels for sale for human consumption. National Health and Medical Research Council, Australian Government Publishing Service, Canberra. 20 pp.

NSW Fisheries. (2002a). Akoya Pearl Farming, Aquaculture Research Information Sheet. Sydney November (www.fisheries.nsw.gov.au/aquaculture/oysters/pearloyst.htm).

NSW Fisheries. (2002b). Triploid Sydney rock oyster farming, Aquaculture Research Information Sheet. Sydney July (www.fisheries.nsw.gov.au/aquaculture/oysters/triploid.htm).

NSW Fisheries. (2004a). Aquaculture prospects for the blue mussel. Sydney. January (www.fisheries.nsw.gov.au/aquaculture/seawater/blue_mussels.htm).

NSW Fisheries. (2004b). Diseases of Sydney rock oysters, Aquaculture Research Information Sheet. Sydney January (www.fisheries.nsw.gov.au/aquaculture/oysters/diseases.htm).

O'Hara, T.D. (2001). Consistency of faunal and floral assemblages within temperate subtidal rocky reef habitats. *Mar. Freshwater Res*. **52**: 853-63.

Prieur, D., G. Meval, J.L. Nicolas, A. Plusquellec and M. Vigneulle. (1990). Interactions between bivalve molluscs and bacteria in the marine environment. *Oceanogr. Mar. Biol. Annu. Rev* **28**: 277-352

Potter, M.A. and B.J. Hill. (1982). Heat mortality in the Sydney rock oyster, *Saccostrea (Crassostrea) commercialis* and the effectiveness of some control methods. *Aquaculture* **29**: 101-108.

Pouvreau S., G. Jonquieres and D. Buestel. (1999). Filtration by the pearl oyster, *Pinctada margaritifera*, under conditions of low seston load and small particle size in a tropical lagoon habitat. *Aquaculture* **176**: 295-314.

Redfield, A.C., P.H. Ketchum and F.A. Richards. (1963). The influence of organisms on the composition of seawater. *In*: Hill, M.N. (ed.) *The Sea. Vol. II. The Composition of Seawater*. Wiley, London. p. 26-77.

Rippingale, O.H. and D.F. McMichael. (1961).Queensland And Great Barrier Reef Shells.. The Jacaranda Press Pty Ltd., Australia. 210 pp.

Rodgers, C.J. (2001). The NSW shellfish quality assurance program: an operational review. Final Report commissioned by Safe Food Production NSW. January 2001. 120 pp.

Ryther, J.H., W.M. Dunstan, K.R.Tenore and J.E. Huguenin. (1972). Controlled eutrophication – increasing food production from the sea by recycling human wastes. *Bioscience* **22 (3)**: 144-152.

Sandifer, P.A. and J.S. Hopkins. (1996). Conceptual design of a sustainable pond-based shrimp culture system. *Aquacultural Engineering* **15 (1)**: 41-52.

Scura, E.D., A.M. Kuljis, R.H. York Jr. and R.S. LeGoff. (1979). The commercial production of oysters in an intensive raceway system. *Proc. World Maricult. Soc.* **10**: 624-630.

Shin, P.K.S., F.N. Yau, S.H. Chow, K.K. Tai and S.G. Cheung. (2002). Responses of the green-lipped mussel *Perna viridis* (L.) to suspended solids. *Marine Pollution Bulletin* **45**: 157-162.

Shpigel, M. and R.A. Blaylock. (1991). The Pacific oyster, *Crassostrea gigas*, as a biological filter for a marine fish aquaculture pond. *Aquaculture* **92**: 187-197.

Shpigel, M., A. Gasith and E. Kimmel. (1997). A biomechanical filter for treating fish-pond effluents. *Aquaculture* **152**: 103-117.

Shpigel, M. and A. Neori. (1996). The integrated culture of seaweed, abalone, fish and clams in modular intensive land-based systems: I. Proportions of size and projected revenues. *Aquacultural Engineering* **15 (5)**: 313-326.

Shpigel, M., A. Neori and H. Gordin. (1991). Oyster and clam production in the outflow water of marine fish ponds in Israel. *Spec. Publ. Eur. Aquac. Soc.* **14**: 295.

Shpigel, M., A. Neori, D.M. Popper and H. Gordin. (1993a). A proposed model for “environmentally clean” land-based culture of fish, bivalves and seaweeds. *Aquaculture* **117**: 115-128.

Shpigel, M., J. Lee, B. Soohoo, R. Fridman and H. Gordin. (1993b). Use of effluent water from fish-ponds as a food source for the Pacific oyster, *Crassostrea gigas* Thunberg. *Aquaculture and Fisheries Management* **24**: 529-543.

Shumway, S.E., T.L. Cucci, R.C. Newell and C.M. Yentsch. (1985). Particle selection, ingestion and absorption in filter feeding bivalves. *J. Exp. Mar. Biol. Ecol.* **91**: 77-92.

Shumway, S.E., C. Davis, R. Downey, R. Karney, J. Kraeuter, J. Parsons, R. Rheault and G. Wikfors. (2003) Shellfish aquaculture – In praise of sustainable economies and environments. *World Aquaculture* **December 2003**: 15-18.

Smit, M.G.D. (2000) Water treatment using mussels and macro-algae. TNO Environment, Energy and Process Innovation, M.Smit@mep.tno.nl

Tenore, K.R. and W.M. Dunstan. (1973). Comparison of feeding and biodeposition of three bivalves at different food levels. *Marine Biology* **21**: 190-195.

Tolley, S.G., A.K. Volety and M. Savarese. (2003). Shellfish research and adaptive resource management in Southwest Florida: oysters as sentinels of ecosystem health. *World Aquaculture* **December 2003**: 64-66.

Walker, R.L., P.B. Heffernan, J.W. Crenshaw Jr. and J. Hoats. (1991). Effects of mesh size, stocking density and depth on growth and survival of pearl net cultured bay scallops, *Argopecten irradians concentricus*, in shrimp ponds in South Carolina, U.S.A. *Journal of Shellfish Research* **10 (2)**: 465-469.

Wang, J.-K. (1990). Managing shrimp pond water to reduce discharge problems. *Aquacultural Engineering* **9**: 61-73.

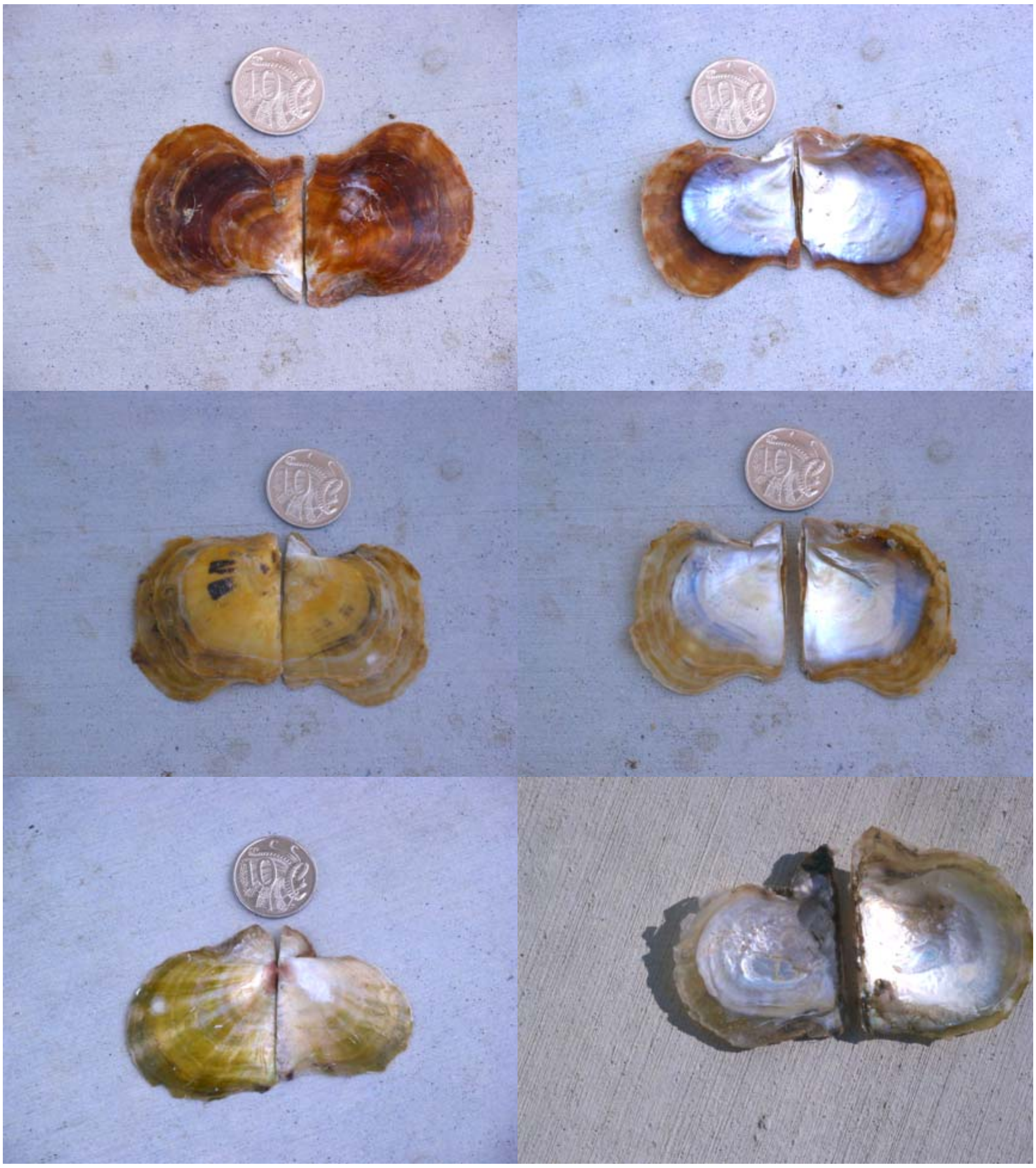
Wang, J.-K. (2003) Conceptual design of a microalgae-based recirculating oyster and shrimp system. *Aquacultural Engineering* **28**: 37-46.

Winter, J.E. (1975). Feeding experiments with *Mytilus edulis* at small laboratory scale. II. The influence of suspended silt in addition to algal suspensions on growth. In: *Proceedings of the 10th European Symposium on Marine Biology* (ed. G. Persoone and E. Jaspers). Universa Press, Watteren. p. 583-600.

Wisely, B and B.L. Reid (1978). Experimental feeding of Sydney Rock Oysters (*Crassostrea commercialis = Saccrostrea cucullate*) I. Optimum particle sizes and concentrations. *Aquaculture* **15**: 319-331.

Appendix 1. Internal shell and viscera of the four species found at BIARC in the study.
Top row. Mud ark (*Anadara trapezia*).
2nd row. Rock oyster (*Dendrostrea folium*).
3rd row. Pearl shell (*Pinctada maculata*).
Bottom row. White hammer oyster (*Malleus albus*).

Appendix 2. Different colour morphologies found in the pearl shell (*Pinctada* sp.)



Appendix 3. Raw data: Stocking and survival of shellfish in tumblers

	Mud arks			**Pearl shells**			**Rock oysters**			**White hammer shells**		
Receptacle	**Tot**	**Liv**	**%**	**Tot**	**Liv**	**%**	**Tot**	**Liv**	**%**	**Tot**	**Liv**	**%**
Tumbler 1	0	0	0	0	0	0	198	145	73	0	0	0
Tumbler 2	287	282	98	81	74	91	10	9	90	2	1	50
Tumbler 3	70	69	99	43	39	91	131	60	46	0	0	0
Tumbler 4	272	269	99	68	55	81	4	2	50	2	2	100
Total	629	620	99	192	168	88	343	216	63	4	3	75

Tot = Total number of shellfish stocked
Liv = Number of living shellfish remaining
% = Percentage survival from initial stocking