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ScienceDirect
Aquaculture 272 (2007) 389–398
Aquaculture
www.elsevier.com/locate/aqua-online

Survivorship and growth of the sea cucumber *Australostichopus (Stichopus) mollis* (Hutton 1872) in polyculture trials with green-lipped mussel farms

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Received 6 March 2007; received in revised form 5 June 2007; accepted 24 July 2007

Abstract

The suitability of the common New Zealand sea cucumber *Australostichopus mollis* to polyculture with green-lipped mussels was investigated in a six-month field study. Sea cucumbers were caged at three densities (2.5, 5 and 15 ind m^{-2}) on the seabed beneath an operating mussel farm and survivorship and growth (weight change) monitored on a monthly basis. The sea cucumbers transplanted to below an operating farm showed excellent survivorship (91.7% overall) over the period of the study and exhibited growth at densities exceeding observed natural densities. Growth was density-dependent and at the highest densities appeared to be constrained by food limitation. *A. mollis* held at 2.5 and 5 ind m^{-2} gained 15.37%±5.33 (mean±SE) and 13.16%±3.42 of their pre-caged body weight, respectively, while those caged at a density of 15 ind m^{-2} showed a 0.21%±2.12 weight loss over the six-month trial. In addition, the acceptability of mussel farm-impacted sediment as a food source was investigated in tank-based feeding experiments with wild-collected *A. mollis*. Adult *A. mollis* readily consumed mussel farm-impacted sediment in laboratory feeding experiments, consuming 6.70 g±1.59 (mean±SE) wet weight mussel sediment d^{-1}. These results clearly indicate that *A. mollis* is an ideal candidate for polyculture with green-lipped mussel farms.

Keywords: *Stichopus mollis*; Polyculture; Sediments; Mussel farms; Sea cucumber; Waste conversion

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1. Introduction

Mussel farming causes large scale biodeposition as a result of mussel feeding. This deposition causes a suite of profound changes to the sedimentation regime and characteristics, in particular alteration of the sediment chemistry below the farm (Dahlbäck and Gunnarsson, 1981; Kaspar et al., 1985; Hatcher et al., 1994; Grant et al., 1995; Christensen et al., 2003). This alters the conditions experienced by benthic organisms and can lead to significant shifts in the composition of associated benthic communities (Kaspar et al., 1985; Grant et al., 1995; Hartstein and Rowden, 2004). These impacts are of concern as they represent degradation of large areas of coastal habitat and such concerns have stimulated interest in finding methods of reducing the impact of farming activities. One potential ecological solution is the polyculture of another species with mussel farms in order to reprocess or remove some of the waste produced. The environmental benefits of successful polyculture are decreased nutrient and waste output and

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doi:10.1016/j.aquaculture.2007.07.230

reduced feeding requirements within aquaculture systems (Neori et al., 1998; Lutz, 2003). Bivalve culture can improve water quality and reduce nutrient loads in both marine and freshwater finfish pond culture (Swingle, 1968; Shpigel and Blaylock, 1991). Zhou et al. (2006) also reported small reductions in organic content and nutrient concentrations in scallop lantern net waste in polyculture with *Apostichopus japonicus*.

There has been increasing interest in the potential of polyculturing deposit-feeding sea cucumbers that have the capability of consuming sediments impacted by aquaculture activities, thereby reducing the associated impact on the benthos and potentially producing a valuable secondary product. These organisms have previously been suggested as suitable candidates for polyculture with filter-feeding bivalves and finfish (Inui et al., 1991; Wu, 1995). They process large amounts of sediments and represent a high value food crop. There are several examples of successful polyculture trials involving sea cucumbers. Ahlgren (1998) reported increased growth of sea cucumbers in polyculture with salmon and consumption of salmon waste and fouling on cages. Kang et al. (2003) showed that *A. japonicus* grows well in polyculture with the charm abalone *Haliotis discus hannai*, while Zhou et al. (2006) reported that *A. japonicus* grows well and reduces organic waste indicators when polycultured with scallops in lantern nets. *A. japonicus* is also grown in polyculture on a commercial scale with shrimp in land-based ponds in China, although there is no information available regarding the ecological benefits of this practise (Yaqing et al., 2000).

The common New Zealand sea cucumber *Australostichopus mollis* is relatively abundant around the New Zealand coast in a range of habitats from shallow rocky reef to mud seafloor at depths exceeding 100 m (Pawson, 1970). The species has recently been reclassified in the new genus *Australostichopus* on the basis of biochemical and morphological differences to other members of the genus *Stichopus* (Moraes et al., 2004). This species also feeds on organic-rich sediments and is a valuable food and food extract crop. *A. mollis* is included in the New Zealand fisheries quota management system (QMS) with a total allowable commercial catch of 22 tonnes for the entire fishery. Small scale export fisheries for *A. mollis* existed in Fiordland and Marlborough in New Zealand's South Island, and a "cottage" fishery exists to supply the small domestic market in New Zealand. The species can yield returns of up to US$12/kg greenweight if appropriately processed. Sea cucumbers are a valuable food item and dietary supplement in the People's Republic of China, Hong Kong, Taiwan, Singapore and Korea (Conand, 2000). The high value of sea cucumber in the international fish trade – up to US$60/kg at market for the premium species – ensures that fisheries are heavily exploited as part of the traditional and commercial fisheries which exist in many coastal nations. The species which are commercially fished show rapid depletion, species are generally fished out in descending order of value (Toral-Granda and Martínez, 2000; Uthicke, 2000). In the light of the promised financial rewards of harvesting, the current low exploitation of *A. mollis* appears unlikely to continue. It is the combination of factors such as commercial value and ability to feed on enriched sediments that make *A. mollis* an ideal candidate for a pilot polyculture system with green-lipped mussels.

As green-lipped mussel culture exclusively uses longline methods, any polyculture undertaking will involve maintaining the sea cucumbers directly on the seafloor below the farm. Consequently, polycultured animals will be exposed to enhanced sedimentation, altered nutrient availability, altered inorganic nutrient fluxes and possibly anoxia (Hatcher et al., 1994; Christensen et al., 2003; Hartstein, 2003). It is unknown how these conditions will affect the survival and growth of *A. mollis*.

This study investigated the potential of a novel polyculture system combining *A. mollis* and green-lipped mussels. The primary aim of the research was to investigate the initial potential of sea cucumber farming techniques for reducing or limiting the benthic impacts caused by mussel farms in New Zealand while producing an additional crop at low cost. Experimental work focussed directly on two aspects critical to any polyculture undertaking: 1) examining the survivorship and growth of *A. mollis* in the conditions beneath operating mussel farms, and 2) investigating suitable stocking densities at farm sites, as compared to observations of natural densities.

2. Materials and methods

2.1. Study area

Experiments were carried out in Kennedy Bay (S 36°40′ E 175°34′), Coromandel Peninsula, northeastern New Zealand. The bay has an area of approximately 3.9 km^2 and opens to the east through a 0.9-km mouth. The bay is relatively sheltered in all but direct Easterly weather conditions and has a low current regime (Gribben et al., 2004; Kelly et al., 2004). Situated at the northern end of the bay is a green-lipped mussel (*Perna canaliculus*), farm consisting of 5 farm blocks covering a total of 0.19 km^2 (19 ha). Two caging sites were established within the bay, an experimental caging site located in the centre of the green-lipped mussel farm and a control caging site situated approximately 0.7 km to the south of the mussel farm. The control caging site was established to isolate any effects of caging while excluding the effects of farm enrichment. The control cage area selected (not affected by mussel farming) is representative of the natural sub-tidal conditions in the bay and is characterised by mixed medium-fine sand. Sediment grain size is predominantly >125 μm and <250 μm (70%) at the control site but ranges up to 500 μm with little or no silt present, minimal shell debris and a uniform or semi-uniform facies. The sediment facies at the experimental site was initially surveyed to ensure that the site was within the farm's impact 'footprint', as indicated by direct observation of falling/accumulated mussel faeces/pseudofaeces and shell debris. The sediment upper layers consist of a mixture of fine silt, mussel faecal and pseudofaecal matter and medium-fine sand, along with large amounts of shell debris from both the NZ green-lipped mussel (*P. canaliculus*) and horse mussel (*Atrina zelandica*). Sediment grain size is predominantly between >125 μm and <250 μm (56%) at the farm site but ranges up to 1 mm. Grain size analysis showed 5.6% silt at the farm site. Four sediment cores (130 mm in diameter × 150 mm depth, upper 50 mm taken for total organic matter (TOM) analysis) were taken at both sites, these samples were used to determine TOM by loss on ignition. TOM in surface sediments at the caging site beneath the farm was 6.8% (±0.5, SE). Total organic matter (TOM) in surface sediments at the control site was 2.7% (±0.1, SE). Mussel-seeded longline droppers above the experimental site extended from backbone ropes at the surface approximately 5 m down into the water column, the longlines above the farm site were in full production for the duration of the experiment.

2.2. Caging location and experimental design

Cages (0.90 m × 0.90 m × 0.23 m, l × w × h), constructed of metal frames and covered with plastic mesh (10 mm roof and walls, 16 mm base), were placed at both locations in early April 2005. Low tide depth at both control and mussel farm sites was 7.0 m, water temperature ranged from 20.0 °C to 14.6 °C over the experimental period. At the mussel farm site two lines of five and one line of four cages were established parallel to, but between operating longlines. This arrangement avoided any potential damage to cages during movement of longline droppers in rough sea conditions. Cages were anchored to the seafloor, with ca. 1–2 cm of sediment covering the base of the cage. Within each cage a natural rock substrate (ca. 200 cm^2), was placed to provide sea cucumbers with a sediment refuge.

Adult sea cucumbers were collected from natural rocky reef habitats located at the southeastern area of the bay, approximately 2–2.5 km from the mussel farm. Individuals were photographed for later identification based on their unique markings, weighed to ±5 g and randomly assigned to experimental cages by SCUBA divers. At both locations three cage densities (2, 4 and 12 sea cucumbers per cage, corresponding to 2.5, 5 and 15 ind m^{-2}), were established and replicated three times. Caging densities were determined based on the maximum observed natural densities. In addition three zero density cages were also established to provide controls for sedimentation measures. Collected sea cucumbers ranged in weight from 60 g to 220 g. Sea cucumbers caged at the farm site ranged in weight from 80 g to 160 g, average weight over all densities at the farm site was 109.1 g (±2.4, SE). Average weight at 2, 4 and 12 sea cucumbers per cage were 112.5 g (±6.6, SE), 104.6 g (±4.7, SE) and 110.3 g (±3.0, SE) respectively. Sea cucumbers caged at the control site ranged in weight from 60 g to 220 g, average weight over all densities at the farm site was 135.3 g (±4.0, SE). Average weights at 2, 4 and 12 sea cucumbers per cage at the control site were 125.8 g (±8.9, SE), 128.2 g (±8.0, SE) and 139.1 g (±5.2, SE) respectively.

2.3. Sediment traps

At each caging site four sediment traps (50 cm × 3.70 cm, l × d, trap area = 10.75 cm^2, aspect ratio = 13.86), opening 18 cm above sediment surface were placed a short distance (ca. 10 m), from cages, two further sediment traps were placed within the zero density cages. Traps were recovered after 89 days, the sediment removed, oven dried at 80 °C for 7 days and dry weight recorded.

2.4. Monitoring of growth and survivorship

Caged sea cucumbers were monitored on a monthly basis over a six-month period. Sea cucumbers were removed from cages by SCUBA divers, brought to the surface in 4-L watertight containers, individually photo-identified and weighed to the nearest 5 g, and returned to the assigned cages. The period of time outside cages did not exceed 25 min. Weight was chosen to assess growth over the more precise displacement method due to the practical limitations of measuring displacement at sea. The weighing method used was a variation of that recommended by Sewell (1987), weight variability was reduced as much as practically possible by weighing between 09:30 and 17:30, when the gut is most likely to be empty (pers. obs.). In addition fluid from the respiratory trees was removed by applying gentle pressure to the anterior end of the sea cucumber prior to weighing. This method reduced the weight variability to <±5% in a pilot study using repeated weight measurements in the laboratory. Survivorship was recorded as presence/absence of individual sea cucumbers in the assigned cage.

2.5. Observation of natural densities

In order to ascertain appropriate densities for caging, a total of 22 reef transects were undertaken. Sites were selected where high densities of sea cucumbers have previous been reported (Sewell, 1987; Archer, 1996), or where sea cucumbers were encountered during pre-transect dives, between one and three transects were carried out per site.

Transects were conducted by two divers counting specimens observed 1.5 m either side of a 50-m transect tape laid along a straight bearing, total area surveyed per transect was 150 m^2. Average depth and visual estimation of the sedimentation regime were also recorded. A five point qualitative scale (from 1—very low to 5—very high) was used to estimate

Table 1
Total *A. mollis* and *A. mollis* ind m^{-2} at natural reef sites in northeastern New Zealand as recorded in 150-m^2 reef transects

Transect site	Depth (m)	Sediment type	Sedimentation regime	Density (ind m^{-2})
Goat Island Sheltered Reef 1	8	Reef detritus	Low	0.02
Goat Island Sheltered Reef 2	7	Reef detritus	Low	0.01
Goat Island Sheltered Reef 3	7	Reef detritus	Low	0.05
Goat Island Outer Reef 1	14	Reef detritus	Low	0.01
Kaiarara Gt Barrier 1	7	Reef detritus/terrigenous	High	0.28
Kaiarara Gt Barrier 2	8	Reef detritus/terrigenous	High	0.13
Katherine Bay Gt Barrier 1	4	Reef detritus	Low	0.03
Katherine Bay Gt Barrier 2	4	Reef detritus	Low	0.01
Kiwiriki Bay Gt Barrier 1	4	Terrigenous	High	0.21
Kennedy Bay Outer Reef 1	8	Reef detritus	Very high	1.09
Kennedy Bay Outer Reef 2	8	Reef detritus	Very high	0.85
Kennedy Bay Southern Reef 1	6	Reef detritus	Medium	0.23
Kennedy Bay Southern Reef 2	6	Reef detritus	Low	0.05
Kennedy Bay Southern Reef 3	6	Reef detritus	Low	0.01
Kyle St Leigh Bay 1	5	Reef detritus	Medium	0.11
Kyle St Leigh Bay 2	7	Reef detritus	Medium	0.13
Kyle St Leigh Bay 3	8	Reef detritus	Medium	0.11
Leigh Harbour Mouth 1	9	Reef detritus	High	0.06
Leigh Harbour Mouth 2	9	Reef detritus	High	0.07
Ti Point 1	18	Reef detritus	Medium	0.03
Ti Point 2	18	Reef detritus	Medium	0.16
Ti Point 3	4	Reef detritus	High	0.31

sedimentation regime, the scale was based on visual estimation of average sediment cover on open raised areas.

2.6. Captive feeding experiments and feeding rates

Captive feeding experiments were undertaken in the laboratory in order to examine the acceptability of mussel farm-impacted sediment as a sole food source for *A. mollis* and determine approximate feeding rates. Mussel farm-impacted sediment was collected from beneath the farm for this purpose by SCUBA divers using a 'surface skim method' collection of the upper ca. 1 cm of sediment), sieved across a 2-mm mesh, frozen (−13 °C), and returned to the laboratory. Sediments were thawed, homogenised and divided into individual 75-g aliquots and refrozen.

Sea cucumbers for captive feeding experiments were collected by SCUBA divers from Matheson's Bay (S 36°18′ E 174°47′), in northeastern New Zealand, returned to the laboratory and housed in a flow-through seawater tank (2.0 m× 0.5 m×0.15 m, l×w×h), for approximately 2 days. Prior to feeding experiments sea cucumbers were removed from the holding tanking and held without food for 24 h in flowing filtered (50 µm), seawater to ensure gut contents were expelled. Individuals were then weighed to the nearest 5 g, as previously described, and placed in individual tanks with a base surface area of 0.20 m^2 (0.55 m×0.35 m×0.21 m, l×w×h), tanks were supplied with flow-through filtered seawater (50 µm). The mean weight of sea cucumbers used in the experiment was 116.3 g (±1.9 SE). Individual tanks were exposed to a daily 'sedimentation event' in which 75 g (wet weight), of farm-impacted sediment was added to the tank. Sediment was thawed and introduced into tanks as an even slurry over the entire water surface. The water supply was interrupted for ca. 35 min to allow the sediment to settle to the tank floor. The addition of 75 g of sediment to the tanks equated to 375 g m^{-2} d^{-1}, approximately that reported by Dahlbäck and Gunnarsson (1981) and Hartstein and Stevens (2005), following conversion to wet weight. Four tanks were subjected to a daily sedimentation event over a four-week period, while another four tanks were subjected to a daily sedimentation event for an eight-week period. Tanks were monitored daily for the following: 1) the presence of fresh faecal deposits, 2) physical disturbance of freshly added sediment, and/or 3) tentacular feeding on the sediment surface.

To determine rates of feeding on mussel impacted sediments four sea cucumbers (mean weight 116.3 g (±2.5 SE) were placed in one of each of four identical tanks and exposed to the sedimentation regime previously described for a period of 7 days. Feeding rates were determined by carefully siphoning faecal deposits from the base of each tank each morning (ca. 09:00), faecal deposits were then transferred onto sections of pre-weighed filter paper (1.2 µm), dried at 80 °C for 48 h and re-weighed to the nearest 0.005 g. The resulting dry weight was then converted to wet weight using the following formula; wet weight = dry weight/0.52, this relationship was previously determined in repeat pilot drying experiments. Faecal matter produced over the first 2 days was collected and discarded to allow sea cucumbers to adapt to the experimental conditions.

2.7. Statistical analyses

All data was tested for normality and homogeneity of variance using a Shapiro–Wilk and a Levene's test, respectively. To test the effect of density on growth at the farm site, a

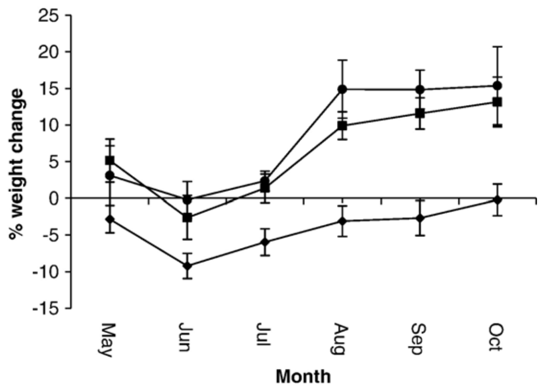

Fig. 1. Average percentage change in sea cucumber weight (100 * ((sample date weight − initial weight) / initial weight)) over six-monthly sampling periods. Sea cucumbers caged at densities of 2.5 ind m^{-2} (●), 5 ind m^{-2} (■) and 15 ind m^{-2} (◆) beneath a green-lipped mussel farm. Error bars indicate standard error of the mean.

one-way ANOVA test was applied to the six-month sample data from the farm comparing growth (calculated as percentage weight change of individual sea cucumbers (100 * ((sample date weight − initial weight) / initial weight)) averaged per replicate cage) across all densities.

3. Results

3.1. Natural sea cucumber densities

Densities of sea cucumbers were highly variable across sites surveyed (Table 1), mean density was 27.05 (±8.70, SE), individuals per transect, this was equivalent to 0.18 (±0.06, SE), ind m^{-2}. The highest natural density observed was 1.09 ind m^{-2}. Highest densities were correlated with very high sedimentation sites. As the mussel farm site experienced substantial organic enrichment well in excess of that experienced in natural reef locations, the densities of sea cucumbers in experimental cages were deliberately selected to considerably exceed the highest natural densities observed. Caging densities chosen were 2.5 (2 ind cage), 5 (4 ind cage), and 15 ind m^{-2} (12 ind cage), fold above the highest density observed in natural habitats.

3.2. Mussel farm cages

3.2.1. Qualitative observations

Cages were repeatedly observed to contain fresh faecal casts during monthly sampling. When visibility permitted, it was possible to observe the effects of sea cucumber grazing at the base of the cage, with wide areas of sediment removed from the base of the cages, this effect was most evident in the highest density cages of 15 ind m^{-2}.

3.2.2. Growth measurements — mussel farm site

Sea cucumbers at the two lower caging densities of 2.5 and 5 ind m^{-2}, exhibited increases in mean body weight of 15.37% (±5.33, SE), and 13.16% (±3.42, SE), respectively, over the six-month period. In comparison the highest cage density of 15 ind m^{-2} showed a mean weight loss of 0.21% (±2.12, SE), of body weight over the six-month experimental period. The rate of weight gain appears to be dependent on caging density (Fig. 1). A one-way ANOVA was applied to the calculated change in body weight data to measure the effect of the density on growth. The one-way ANOVA was applied to data collected at the end of the experiment (sixth-month sampling). Density appeared to have an effect on data yet this effect was not statistically significant: ANOVA $F_{2,\,6}=3.5853$, $p=0.09$ for all groups. Applying the one-way ANOVA as described above to prior sampling dates revealed that the effect of density became apparent and significant at the third monthly sampling in July: ANOVA $F_{2,\,6}=13.998$, $p<0.01$ for all groups. This significant result appears, however, more likely to be due to the weight loss amongst sea cucumbers in cages at a density of 15 ind m^{-2} during early sampling (Fig. 1). Applying the same ANOVA to the fourth monthly sampling in August reveals equally

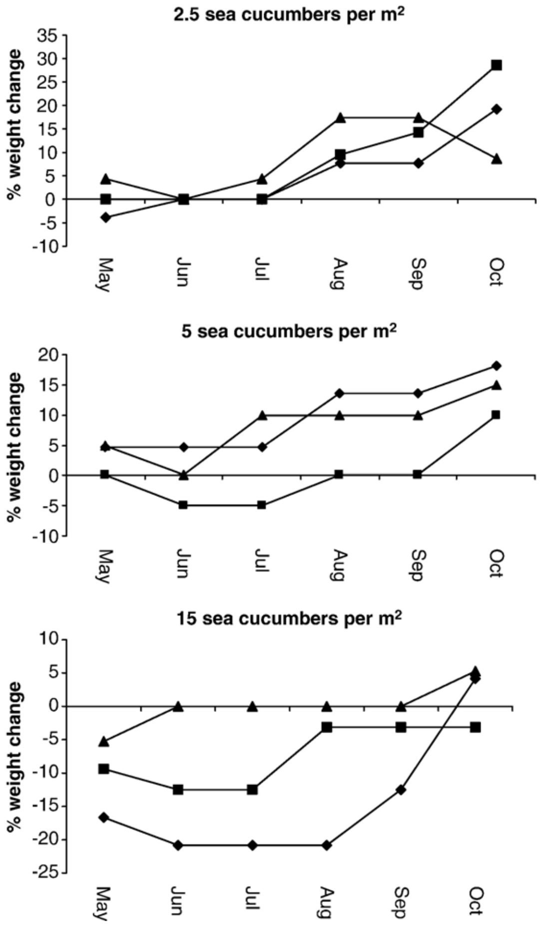

Fig. 2. Examples of individual weight change (100 * ((sample date weight − initial weight) / initial weight)) for three photo-identified *A. mollis* caged at densities of 2.5 ind m^{-2}, 5 ind m^{-2} and 15 ind m^{-2} beneath a green-lipped mussel farm.

significant results: ANOVA $F_{2,\ 6}=11.032$, $p<0.01$ for all groups, and this analysis appears to reveal the first apparent effect due to an increase in weight amongst sea cucumbers in cages at densities of 2.5 and 5 ind m^{-2} (Fig. 1).

The overall trend of weight change was similar across all cage replicates and densities. Three periods can be identified, an initial early phase characterised by weight loss, a middle phase characterised by rapid weight gain, which was then followed by a substantial slowing of growth (Fig. 1). The largest increase in average weight was observed between July and August with 2.5 and 5 ind m^{-2} cages recording 12.1% (±2.49, SE), and 8.69% (±1.45, SE), increases respectively. Photo-identification enabled the growth rates of individuals to be tracked over the course of the experiment (Fig. 2), and thereby the calculation of variances across individual cages. While the rates of weight gain over time were variable, individuals caged at densities of 2.5 and 5 m^{-2} showed a similar overall growth trend. In comparison sea cucumbers caged at 15 ind m^{-2} showed a more appreciable delay in weight recovery after an initial weight loss in May and June.

3.2.3. Growth measurements — control site

The control site, which was less sheltered than the experimental mussel farm site, was heavily damaged during an intense easterly storm event in the third month of the experiment, with the loss of 5 cages. As survivorship was high at the mussel farm site, it was deemed that the control site was no longer required to test for negative effects due to caging. Despite this the 4 remaining cages were left in place and survivorship monitored, survivorship over the remaining four-month period was 94%. Average weight change at the control site over the first 2 months was negative for all densities, with sea cucumbers at 2.5 ind m^{-2} showing a 2.62% (±3.29, SE), loss on initial weight, and those at 5 ind m^{-2} and 15 ind m^{-2} showing 10.00% (±1.99, SE), and 13.91% (±1.41, SE), losses respectively.

3.3. Survivorship

Overall survivorship of sea cucumbers at the mussel farm site equated to 90.7%; 66.6% at 2.5 ind m^{-2}, 100%, at 5 ind m^{-2} and 91.7% at 15 ind m^{-2}. Recording survivorship as the number of individuals present in a cage, with those unaccounted for being assumed dead proved to be an inaccurate measure of survivorship. Sea cucumbers ≤ 80 g in weight were found to be able to escape cages when the sediment cover was eroded around the margins of the floor mesh. Consequently, losses were most likely escapes rather than deaths, sea cucumber carcasses were never observed in cages, and several of the smaller individuals were 'apprehended' with their bodies extending through the mesh. The assumption that losses were the result of escapes is also supported by the observation that the average initial weight of sea cucumbers that were lost 84.00 g (±9.14, SE, $n=5$), is substantially below the mean weight of individuals caged under the farm 106.76 g (±2.50, SE, $n=54$).

Table 2
Mussel farm-impacted sediment consumption rate for tank-held *A. mollis*

Treatment tank	Sediment consumed ww g d^{-1}	SE
1	5.37	0.38
2	10.82	0.82
3	7.28	1.21
4	3.34	0.69
Mean	6.70	1.59

3.4. Sediment traps

Two sediment traps from the western end of the mussel farm site were recovered, the remaining two sediment traps were not able to be relocated. Sedimentation rates in control cages averaged 1893.5 g m^{-2} d^{-1} (±162.0, SE), while rates outside cages were substantially lower 1376.3 g m^{-2} d^{-1} (±197.5, SE). Sedimentation rates appeared to be grossly overestimated by the traps, which appeared to collect large amounts of resuspended material.

3.5. Captive feeding experiments and feeding rates

Wild-collected sea cucumbers began feeding on mussel sediment within 12 h of being introduced to the tank. Feeding was recorded for all individuals on all days for the duration of the experiment, with no mortalities observed. The mean wet weight of faecal matter recorded per individual in feeding rate experiments was 6.70 g d^{-1} (±1.59, SE), the lowest mean wet weight faecal matter produced over the 7-day period was 3.34 g d^{-1} (±0.69, SE), and the largest was 10.82 g d^{-1} (±0.82, SE) (Table 2).

4. Discussion

This study investigated the suitability of the deposit-feeding sea cucumber *A. mollis* for polyculture with the green-lipped mussel (*P. canaliculus*). Determination of suitability involved two aspects; 1) a long-term (6 months), farm based transplant experiment that evaluated the response, growth and survivorship of *A. mollis* in the conditions prevailing directly beneath a mussel farm, and 2) laboratory based experiments that examined the acceptability of sediments impacted with mussel biodeposits as a potential food source.

4.1. Survival and growth

The high survivorship of *A. mollis* beneath the farm across all caging densities reflects the suitability of sea cucumbers to the benthic conditions prevailing beneath an operational green-lipped mussel farm. Initial concerns that the physicochemical sediment conditions which typically prevail beneath the farm (e.g. reduced oxygen availability, excessive organic sedimentation, excessive shell drop, nitrate release) may adversely affect survivorship proved unfounded. The high survivorship recorded over a prolonged period illustrates that *A. mollis* tolerates and exhibits growth in the conditions encountered beneath a typical green-lipped mussel farm. Zhou et al. (2006) also reported high survivorship of *A. japonicus* when co-cultured with bivalves. However, Zhou et al. (2006), co-cultured sea cucumbers in bivalve lantern nets above the seabed, as such, the sea cucumbers were isolated from the conditions prevailing beneath the farm. In the current study sea cucumbers were cultured on the seabed directly beneath the farm within the farm's impact footprint.

4.2. Growth is density and food resource dependent

Densities of *A. mollis* in natural reef environments can reach 1 ind m^{-2} if there is an appropriate amount of high organic matter sediment input. This was determined by measurement of densities of *A. mollis* at natural subtidal sites. Variability of density by site was independent of depth or other physical factors and appeared to be positively correlated with the observed level of sedimentation, and hence, food availability. The upper limit of natural densities provided comparative densities for the experimental work under mussel farms based on evidence of probable food limitation. As the rate of sedimentation and organic content of sedimenting particles below mussel farms far exceed that of natural reef environments, the maximum density observed (ca. 1 ind m^{-2}) was taken as the basis for the selection of caging densities.

Rate of growth among farm caged animals was density-dependent with the highest growth rates observed at the two lowest caging densities. At the maximum density attempted in this study, ca. 15 times the highest observed natural density, the net zero observed weight change was most likely due to food limitation. In this instance, the benthos (TOM 6.83%, ± 1.59, SE) below the green-lipped mussel farm used in this study appears to have an optimal sea cucumber stocking density between 5 and <15 ind m^{-2}, significantly higher than densities observed in natural reef locations.

4.3. Growth compared to other holothurians

Comparisons of sea cucumber growth rates recorded in the current study with those from previous studies should be interpreted with caution due to the variety of weight measures applied, differences in the developmental stage of animals studied and the wide range of culturing methods. Nevertheless, the growth rates observed in this study compare acceptably with other studies; growth of individuals at the two lowest densities (ca. 220 g and 550 g sea cucumber biomass m^{-2}), averaged 0.065 g d^{-1}. Tank-reared juvenile sandfish *Holothuria scabra* fed fresh and powdered algae (Battaglene et al., 1999), achieved growth rates of 0.2 g d^{-1}, although growth slowed once densities exceeded 225 g of sea cucumber biomass per square meter. Pitt et al. (2004) reported similar growth rates (max 0.3 g d^{-1}) for *H. scabra* fed shrimp starter food, with growth limitation occurring at 300 g m^{-2} while Zhou et al. (2006) report growth rates between 0.15 and 0.26 g d^{-1} for *S. japonicus* when co-cultured at varying densities in scallop lantern nets.

Adult sea cucumbers have been shown to exhibit negative growth when food limited in both natural and tank conditions (Uthicke and Benzie, 2002). Nevertheless, adult sea cucumbers made considerable gains during June and August in the farm caging experiment. This can be partly explained by the coincidence of this period of accelerated growth with the time of maximal gonad development in *A. mollis* (Morgan, 2004). Despite this, gonad growth does not exceed 2% of wet body weight per month and mean total gonad weight would not exceed 2.5 g wet weight during the period of this experiment (Sewell, 1987; Morgan, 2004). Future research into the growth potential of juvenile *A. mollis* feeding on mussel waste would be of particular interest following the identification of reliable sources of juveniles for collection and ongrowing.

4.4. Feeding rates and food conversion

The recorded sea cucumber feeding in laboratory experiments and subsequent growth in the laboratory and caging experiments indicates that sediment heavily impacted by mussel biodeposits is an acceptable food source. An average of ca. 6.70 g wet weight sediment was consumed daily by *A. mollis* in experiments investigating feeding rate. Combining the wet weight consumed and the mean weight gain from the feeding experiments (0.19 g/day) allows the approximate calculation of a feed conversion ratio of 38.95:1 or 2.57% conversion. On average, animals consumed less than 10% of their body weight in sediment per day. This is not in agreement with Lopez's (1987) review which showed that majority of deposit-feeding marine life consumes several times their own body weight per day. The total weight of sediment consumed by *A. mollis* does not approach the large amounts (ca. 40% body weight/day) reported by Uthicke (1999) for *Holothuria atra* and *Stichopus chloronotus* in similar tank experiments or the feeding rate values compiled by Roberts et al. (2000) for a number of deposit-feeding holothurians. The amount consumed by *A. mollis* may be lower due to the high organic matter content of the mussel farm-impacted sediments or the sediment's high algal detrital content (Lofty, 1974) as compared to the sand substrate used by Uthicke (1999). Da Silva et al. (1986) raised the possibility of an overestimation of expected temperate feeding rates by comparison with values for species feeding on tropical sand substrates, with lower organic matter content. Uthicke (1999) also allowed a two-week period of adjustment before measuring faecal matter production, as compared to 2 days in this study, which may partly explain the comparatively low values produced in our feeding experiment. Despite this, measures of sediment consumption are useful for basic estimates, as no comparative measurements of *A. mollis* feeding in the field are available.

4.5. Potential application with other forms of aquaculture

Sea cucumber polyculture with other aquaculture systems, such as other bivalve farms or finfish cages, may also be equally viable. The high organic waste output from finfish cages may allow even greater densities of *A. mollis* to be farmed below them. Conversely, the conditions below finfish cages represent a massive increase in the level of organic enrichment and chemical pollution in comparison to those of mussel farms. Furthermore, excess feed accumulation (high protein) and inputs from antibacterial and antiparasite treatments will lead to differences in the composition of impacted sediments below finfish farm. These combined differences in benthic conditions may adversely affect sea cucumber survivorship (Brown et al., 1987; Haya et al., 2001; Pitt et al., 2004). Equally interesting is application to the New Zealand oyster farming industry. Organic nutrient output from oyster farms is slightly lower than from green-lipped mussel farms (Forrest, 1991), but may be sufficient to support considerable densities of *A. mollis*.

4.6. Sea ranching

The current research employed a caging method which would be impractical on a commercial scale. Installing large caging structures beneath an operating mussel farm is likely to be both expensive and disruptive to the mussel farming cycle. Submerged cage structures are also likely to be damaged during normal farming operations, to obstruct or tangle farm structures such as droppers, or to be overwhelmed by shell debris, all of which would result in animal losses and intensive maintenance requirements. The option of sea ranching appears to be considerably more practical. It can be expected that the progressive decrease in organic matter (hence low food-value) concentration in sediment towards and beyond the edge of the farm footprint will act as a habitat border for sea cucumbers, essentially keeping seeded sea cucumbers within the immediate proximity of the farm. Specific physical structures on the seabed or seabed profile types may also be useful to delineate habitat boundaries (Massin and Doumen, 1986). The need for substrate, such as rock piles or similar to provide a refuge for sea cucumbers in such a system will also need to be studied. It is unknown how sea cucumbers react to storm events and displacement during such events may remove sea cucumbers seeded beneath the farm.

4.7. Stocking densities relate to farm impact

An additional tentative calculation can be made as to the required stocking rate for effective processing of mussel waste. Green-lipped mussels have been shown to have widely variable seston filtration rates but an average of 20 mg h (dry weight) has been determined for small mussels (37 mm shell length) at medium to high chlorophyll concentrations (3–6 $\mu g\, l^{-1}$) with 30–50% of the filtered material being rejected as pseudofaeces (Hatton et al., 2005). Average chlorophyll *a* concentrations at New Zealand green-lipped mussel farms are 1.4 $\mu g\ l^{-1}$. Under conservative assumptions (James et al., 2001) of linear feeding reduction with decreased chlorophyll *a*, that 40% of filtered material is rejected and that 25% of the ingested material is assimilated, a single small green-lipped mussel would produce ca. 127 mg d^{-1} dry weight (233 mg wet weight) per day of faeces and pseudofaeces combined. A sea cucumber consuming 6.7 g wet weight of mussel waste per day would grow effectively on the waste produced by 29 mussels. An average green-lipped mussel farm is stocked at a density of 10 kg wet weight mussels per m^2 (entire farm area) at harvest, which equates to ca. 150 mussels per m^2 assuming an average wet weight of 65 g per mussel when harvested at 90 mm (Inglis et al., 2000). On the basis of this calculation, a stocking density of 5.2 sea cucumbers per m^2 would be required to reprocess the majority of mussel waste below an operating farm. Or viewed conversely, the carrying capacity below an operating green-lipped mussel farm would be expected to average 5–6 sea cucumbers per m^2. Interestingly, the sea cucumbers in the present study exhibited highest growth at densities of 2.5 and 5 animals per m^2, with growth limitation occurring between 5 and 15 per m^2, although there is no information available regarding the effect of the cages on sedimentation for the current study. This estimate does not take into account variations in mussel biodeposit supply, dispersal and erosion which will vary both with season, site, hydrodynamic regime and mussel diet (Hartstein, 2003; Giles and Pilditch, 2004).

4.8. Density, remediation and physicochemistry

Growth was recorded for sea cucumbers caged below mussel farms at densities well in excess of the natural densities measured at comparable depths and habitats in northeastern New Zealand. A strategy of pursuing maximal growth in future co-culture will require that the initial stocking density of sea cucumbers be closely aligned with the severity of impact of individual farms. In this regard stocking density will most likely scale with organic enrichment, with higher impact farms being capable of supporting higher stocking densities. Alternatively, stocking density may also depend on whether a strategy of remediation or maximal growth is pursued. Exceeding recommended stocking densities may improve the potential remediation of sediments below the mussel farm but may compromise overall sea cucumber biomass yield or time to maximum biomass yield. It remains to be seen whether sea cucumbers have a positive effect on the physicochemical attributes of mussel farm-impacted sediments. If their co-culture does remediate sediments, they can be directly cultured on the impacted sediment as a biological tool to either constrain impact during farm operation or to expedite remediation during farrowing or rotation periods. Insight into the sediment chemistry effect of sea cucumber feeding on mussel farm-impacted sediments is required to estimate the sediment remediation potential of polyculture with mussels and other aquaculture systems.

5. Conclusion

The study of *A. mollis* suitability to culturing with a mussel farm has shown the species to be capable of surviving and growing at high densities in the conditions prevailing below mussel farms. Results also show the acceptability of mussel farm-impacted sediments as a food source for this species. These results strongly indicate that *A. mollis* is a suitable candidate for polyculture with mussel farms in New Zealand. Larger scale piloting of polyculture methods, including farm seeding and ranching, will be required to develop practicable farming methods. In addition, insight into the effect of *A. mollis* grazing on the chemistry of mussel farm-impacted sediments is recommended to understand the potential ecological benefits of sea cucumber polyculture with mussel farms.

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