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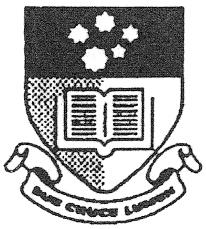

Project 94/091

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1. Executive overview - Non-technical summary

1.1. Background

At Port Lincoln (South Australia) farming of southern bluefin tuna (*Thunnus maccoyii*) has increased rapidly since humble beginnings in 1989 to become a major component of the local economy. Earnings from the industry have risen from $4 million in 1990 to around $40 million in 1994. In 1995 there were 46 sea-cages (from 30 - 50 m diameter) situated both within Boston Bay and seaward of Boston Island.

Sea-cage farming, requires stocking densities and feeding rates far in excess of natural populations and produces a constant stream of organic and non-organic waste (Gowen and Bradbury, 1987). As farming of this kind generally occurs in sheltered locations, the resulting enrichment of the seafloor in the immediate vicinity of the cage is inevitable (Brown *et al.*, 1987; Lumb, 1989). This waste is of particular concern to the industry (Gowen and Bradbury, 1987) which recognises the need to maintain a healthy environment for the fish thus ensuring the productivity and commercial viability of the farms. There is further concern in the wider community (Druff, 1987; Hammond, 1987) that degradation of the local environment should be minimised and farms should be closely monitored in order to determine the extent of impacts associated with the industry.

The Port Lincoln Aquaculture Management Plan (Bond, 1993) examined the possible effect of the waste from tuna farming on the waters of Boston Bay and defined the areas in which aquaculture could occur along with a series of guidelines which call for environmental monitoring.

Monitoring should enable the spatial extent and degree of seabed souring to be quantified. This will allow an assessment of the environmental quality of the coastal waters in which the farms are located which in turn impacts upon the health of caged fish and the productivity of farms. A knowledge of the spatial extent of souring and the rate of recovery of seabeds will also be an important factor in developing management strategies for cage rotation and siting.

A series of sampling methods were developed and analysed in order to determine their utility in quantifying the extent of seafloor souring in the vicinity of the Boston Bay tuna farms. In developing these methodologies a number of factors were considered. In particular the need to:

i) Have sufficient power to detect changes in seafloor communities both during souring and the subsequent recovery phases.

ii) Provide a basis for long term monitoring which allows comparisons of alternative farming methods and conforms with (and helps in the development of) regulatory requirements.

iii) Be cost effective so that they can be applied within the industry as a management tool.

1.2. Scope

The aims of our research were to develop a series of protocols for the ongoing environmental monitoring program and to use these protocols to investigate the impacts of tuna sea-cages. In order to achieve these outcomes the following objectives were agreed:

1) To develop and trial protocols for assessing epibenthic communities.

2) To develop and trial protocols for assessing infaunal communities.

3) To develop and trial a system for monitoring the changes in oxygen status of tuna cages.

4) To trial the use of a probe for making *in situ* measurements of REDOX potentials of the sediment under tuna cages.

5) To determine if potentially harmful gases such as methane (CH_4) and hydrogen sulphide (H_2S) were present (or likely to be present) in the organically enriched sediments under cages.

1.3. Approach

A series of survey protocols were developed to assess the infaunal and epibenthic communities in Boston Bay. These protocols were based on those being used to assess impacts on soft-bottom communities due to sand replenishment dredging (work by Cheshire *et al.* in collaboration with the Coast Protection Branch of South Australia). These protocols were trialed during 1994-1995 with data being collected from a number of sites within Boston Bay.

1.4. Summary of results

A remote suction sampler, designed to collect infaunal samples has been developed, trialed and proven to be a very effective tool for studies of benthic infauna. Video-transects of epibenthic communities have also been trialed and, although they are much cheaper than diver surveys, the quality of the data obtained is very low compared to information collected by divers.

Pilot studies using a fully automated photorespirometer were undertaken of the oxygen content within the sea-cages holding the tuna. These were further developed into an honours program (by Ms. E. Cronin, University of Adelaide) which is reported elsewhere.

Chemical studies of the sea-floor which looked at REDOX potential, and the production of methane (CH_4) and hydrogen sulphide (H_2S) were inconclusive and need further development before they will be of value as a management tool.

In general the benthic survey protocols provide adequate power for the detection of impacts in both infaunal and epibenthic communities associated with sea-cage farming (based on an analysis of three environmental indices Taxa Richness, Shannon Diversity and Taxa Equitability). These methods have been costed and details of the relative costing have been provided on a per cage basis. This shows that the most cost effective methods for survey are those which look at the epibenthic community. Significant problems arise however in that the video surveys, which are the most cost effective, give results which are inconsistent with the diver surveys. This discrepancy needs resolution before the method can be uniformly adopted.

The survey techniques all utilise readily available equipment and procedures which can be applied within the industry.

1.5. Recommendations

1) Further studies should be undertaken to continue the development of the video monitoring system. This should concentrate on improving its resolution so that it can provide a similar level of taxonomic discrimination of the epibenthos to that achieved by divers.

2) Monitoring programs should be developed which utilize a combination of methodologies. Infaunal studies provide a higher level of discrimination than epibenthic community studies but are considerably more costly.

3) Work on seabed recovery should be undertaken using these methods to evaluate optimal cage rotation strategies.

4) A database should be setup within the framework of a Geographical Information System (GIS) which will facilitate studies of the longer term effects of sea-cage farming on benthic environments.

2. Background

Intensive farming of marine fish has grown considerably in the last decade (Frid and Mercer, 1989; Hall *et al.*, 1990), particularly in Europe (Gowen and Bradbury, 1987; Lumb, 1989), Canada (Aiken, 1993; Sylvain, 1993), Japan (Tsutsumi, 1995) and New Zealand (Kaspar *et al.*, 1988). In Australia there is a similar trend toward increased aquaculture. Atlantic salmon farming has been highly successful in Tasmania (Hodson and Burke, 1994) and, since 1990, the development of the southern bluefin tuna farms at Pt Lincoln has proven to be highly profitable with rapidly expanding potential.

2.1. Need

Bluefin tuna aquaculture requires a healthy local environment in order to maintain productivity and commercial viability (Gowen and Bradbury, 1987), however, intensive sea-cage farming produces a constant stream of organic and non-organic waste (Gowen and Bradbury, 1987). There is also community concern that degradation of the local environment should be minimised and that environmental health of areas surrounding farms should be closely monitored (Druff, 1987; Hammond, 1987).

To date the majority of research on seafloor souring under sea-cages has considered salmonid farming (eg Gowen and Bradbury, 1987; Lumb, 1989) which is the most commonly farmed fish in this type of aquaculture (Hall *et al.*, 1990). The research on tuna farming is restricted to the management plan developed by Bond (1993) which indicated that the environment of Boston Bay is likely to respond uniquely to the presence of tuna farms.

2.2. Objectives

With these issues in mind, a research program was initiated with the aims of a) developing a series of protocols for an ongoing environmental monitoring program and b) to use these protocols to investigate the impacts of tuna sea-cages on benthic environments in Boston Bay.

In order to achieve these outcomes the following objectives were agreed:

1) To develop and trial protocols for assessing epibenthic communities.

2) To develop and trial protocols for assessing infaunal communities.

3) To develop and trial a system for monitoring the changes in oxygen status of tuna cages.

4) To trial the use of a probe for making *in situ* measurements of REDOX potentials of the sediment under tuna cages.

5) To determine if potentially harmful gases such as methane (CH_4) and hydrogen sulphide (H_2S) were present (or likely to be present) in the organically enriched sediments under cages.

The applications of these methods to assessing the impact of sea-cage tuna farming on benthic environments are detailed in the companion reports (Cheshire *et al.*, 1996a; Cronin, 1995).

3. Methods

3.1. Survey methods

A number of methods were trialed which looked variously at i) the epibenthic flora and fauna of the bay, ii) infaunal communities, iii) changes in the oxygen status of the cage environments and iv) the chemistry of sediments.

3.1.1. Epibenthic studies

Two alternative procedures were trialed in surveying the epibenthos; diver surveys, which we have previously employed in an ongoing study on the effects of dredging at Pt Stanvac (20 km south of Adelaide), and remote video transects which were considered a viable alternative.

3.1.1.1.Diver survey

Four 200 m transects were run at locations in Boston Bay (Table 1; Fig. 1) in August 1994. A pair of divers swam along the transect line with a 2 m pole clipped at its centre to the transect rope. This pole defined a 1 m square quadrat on either side of the line which was surveyed by each diver. At distances of 0, 5, 10, 15, 20, 50, 100, 150 and 200 m along the transect line, the presence and number of different types of plants, animals and features within each quadrat was recorded. The transects radiated from the sea-cages and were placed so that they began as close to the outside of the predator net as was practical. A specimen of any animal or plant which could not be identified *in situ* was placed in a numbered plastic bag and brought to the surface. This bag number was used as a taxa label until properly identified in the laboratory (Appendix 2).

Diver surveys at Pt Lincoln, while providing high quality data, were problematical for a number of reasons. The sediment in areas of Boston Bay is fine and disturbance causes increased turbidity which obscured the seabed and made *in situ* counting and identification difficult. Thus, to be able to see the biota on the seafloor the divers had to work upside down and very carefully. This slowed the pace of the work and increased the bottom time. The depth of the work (15 - 18 m) limited dive times to less than 50 minutes (with 27 minutes allowable on the second dive after a two hour surface interval - DCIEM Tables) and the risk of sharks were also considered to limit the regular use of this method. When combined and with due consideration to the likelihood that future farming would be undertaken in deeper water, these problems were considered sufficient to warrant a different approach that removed the need for divers to enter the water.

3.1.1.2.Development of the video transecting technique

Benthic survey work using video cameras is not new (eg. Edmunds and Witman, 1991; Whorff and Griffing, 1992; Anderson, 1994; Parker *et al.*, 1994) in particular it has been used extensively in deep sea habitats (Edmunds and Witman, 1991) and on the Great Barrier Reef (Christie and Neale, 1995). The main advantage of the video method is that it permits the rapid collection of data that can be analysed in the laboratory (Leonard and Clark, 1993) and removes the need for divers.

A sled mounted video camera was used to film transects. The sled measured 1.2 X 1.9 m and was constructed from 75 mm diameter UPVC stormwater pipe. A sloping platform (45 °) was bolted onto the front of the sled. The camera was mounted on this platform so that it pointed forward

which allowed an image to be obtained which was relatively undisturbed by the passage of the sled. A triangular bridle was attached to the front of the sled to act as a towing point and a buoy line was mounted centrally. The central buoy was used to raise and lower the sled and to give an indication of its position along the transect line. This line would also serve as a safety mechanism for retrieval of the system should the towing line break.

For deployment of the sled at a tuna cage the camera was bolted in place and turned on. The sled was then lowered as close as practicable to the edge of the predator net and pointing in the direction the video transect was to take (roughly perpendicular to the cage). For the October 1994 survey a team member was left on the cage while a 200 m line (which was attached to the bridal of the sled) was laid out along the course of the transect. The boat was then anchored such that the full 200 m of rope was laid out. Depending on the prevailing wind/current, this often meant attaching extra rope to the transect line, dropping the anchor and then hanging off this point toward the cage or dropping the anchor before the line was fully layed and using this point to fix the boat's position.

The boat team then slowly pulled in the transect rope noting the time for retrieval of every 2 m. The person on the cage was required to observe the buoy line and to signal the boat team when the sled had begun to move. This was considered to be a distance of 0 m. The sled was then left at this position for thirty seconds and then slowly dragged closer to the boat. Further stops of thirty seconds were made at 2, 10, 20, 40, 60, 80, 100, 120, 140 and 150 m away from the 0 m point, using 2 m markings on the rope as a measure of distance. On completion of the transect the sled was retrieved and camera battery and tape replaced before moving on to a new site.

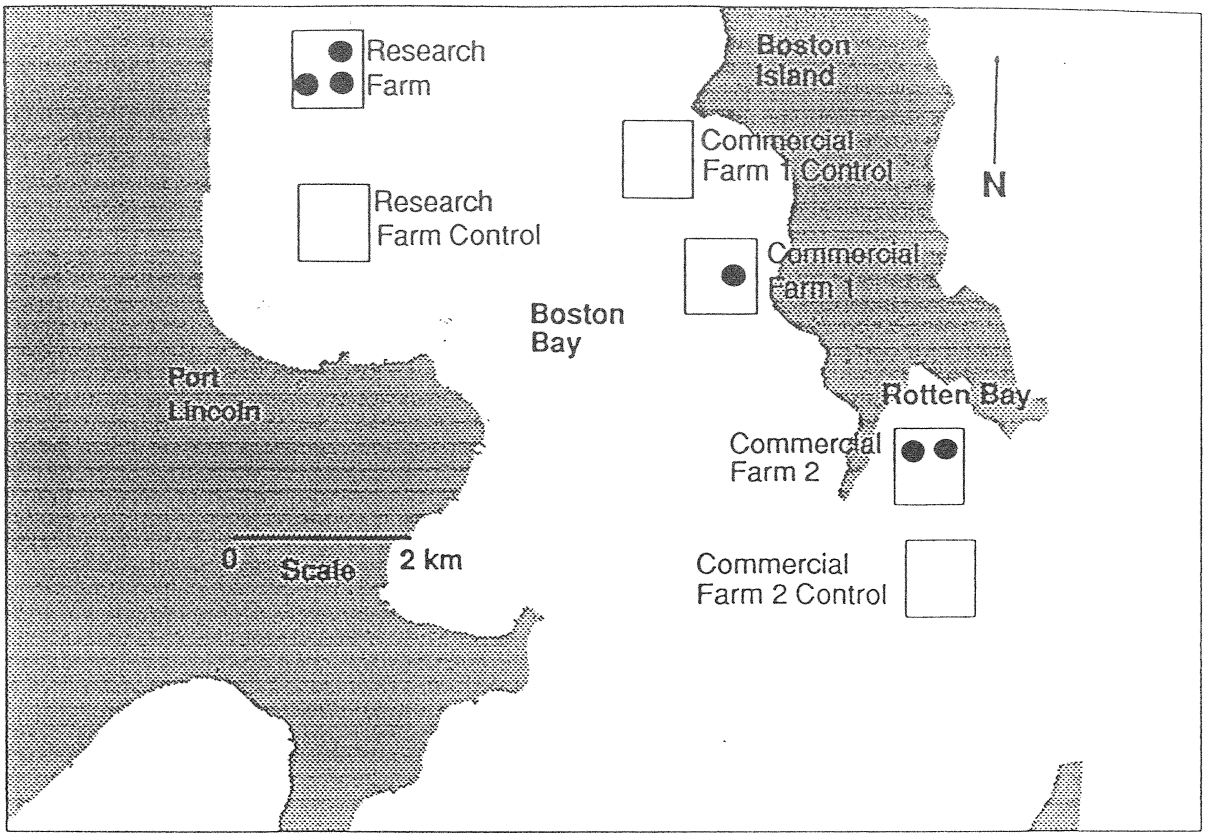
For the February 1995 survey no team member was placed on the cage. Instead the buoy line was hooked over the railing of the tuna cage at such level that a minimum of movement of the sled would be easily observed from the boat. Once this movement was seen the sled was considered to be at 0 m (distances were as before). Time spent at each stop was increased from thirty seconds to one minute. It was also necessary to pay out transect line from the boat at each of the stops. This prevented wave action on the boat from levering the sled out of position and making the stop points hard to detect, a problem encountered in the October videos. Longer stops also helped alleviate this problem.

Distance measurement using marks on the transect line was not as exact as those of diver surveys, however, for the scale of changes in the system, this method was considered accurate enough.

Control transects were managed in a similar manner. The sled was dropped in open water and once settled on the bottom the transect line was deployed. As there was no indication as to when the sled would begin moving, the boat was used to pull the slackness from the line for a very short distance. Once anchored about 20 metres of transect line was retrieved before the assumption was made that the sled had begun moving. This was considered to be the starting point.

For all transects a check was made that the total time as recorded on the surface corresponded to the total time recorded by the camera.

Figure 1. Map of Boston Bay, Pt Lincoln showing the cages and the areas where the survey work was undertaken.



Control transects were located 1 to 2 km away from the tuna cage on which the cage transect was taken and run in the same direction. Cages were examined in both summer and winter with one transect per cage and one per control collected in winter and two transects for each cage and two per control collected in summer (Table l). Throughout all surveys control transects were always run in the same direction as the cage transects to ensure that the influence of environmental gradients (other than cage effects) were the same.

Each tape was transcribed from the Hi 8 format to high quality VHS tapes. This served both as a backup and made for more convenient viewing. Data was collected from the tapes by watching them. At each of the stops the area of seafloor observed by the camera was treated as a quadrat and the number of organisms counted and classified. As with the diver transects some components were scored only for presence/absence. Larger macro-algae (which intermixed with itself and other algae making a discrete count difficult) were considered in terms of an estimate of the area of cover (to the nearest 5%). A total of 47 different field codes were identified which were analysed in 12 groups (Appendix 2).

Table 1 Cages and numbers of transects conducted from August 1994 to February 1995.

Site	Date	Type
Commercial Farm 1	10/10/1994	Diver
Commercial Farm 1	18/10/1994	Video
Commercial Farm 1	14/02/1995	Video
Commercial Farm 1	18/02/1995	Video
Commercial Farm 1 Control	18/10/1994	Video
Commercial Farm 1 Control	14/02/1995	Video
Commercial Farm 1 Control	21/02/1995	Video
Research Farm	20/10/1994	Video
Research Farm	13/02/1995	Video
Research Farm	15/02/1995	Video
Research Farm (2 transects)	9/10/1994	Diver
Research Control	9/10/1994	Diver
Research Control	18/10/1994	Video
Research Control	13/02/1995	Video
Research Control	15/02/1995	Video
Commercial Farm 2	21/10/1994	Video
Commercial Farm 2	14/02/1995	Video
Commercial Farm 2	23/02/1995	Video
Commercial Farm 2 Control	18/10/1994	Video
Commercial Farm 2 Control	14/02/1995	Video
Commercial Farm 2 Control	23/02/1995	Video

3.1.2. Infauna survey

3.1.2.1.Development of a remote suction sampling system

The method of sampling sediment for infauna analysis with suction devices is not new (eg Christie, 1976) and the data obtained can be more informative than epibenthic surveys although the sorting and counting of the animals in sediment samples is time consuming (and hence costly). Field work with suction samplers has always been very diver intensive. Owing to the difficulties outlined in the epibenthic survey with respect to diving in Boston Bay, it was decided to use a remote sampling approach.

The basic design of a suction sampler can vary considerably although the basic components are similar. We used a 75 mm diameter PVC pipe approximately 1.7 m long with an elbow at the exhaust end. A valve in the side of the pipe allowed for the injection of compressed air from a Hookah system located on the surface (Fig. 2). The venturi effect of this air passing up the pipe creates the suction necessary to suck sediment through the pipe and into a mesh bag that was tied over the exhaust. This bag had a mesh size of 1 mm. Fine sediments pass through the bag and everything larger than 1 mm was trapped.

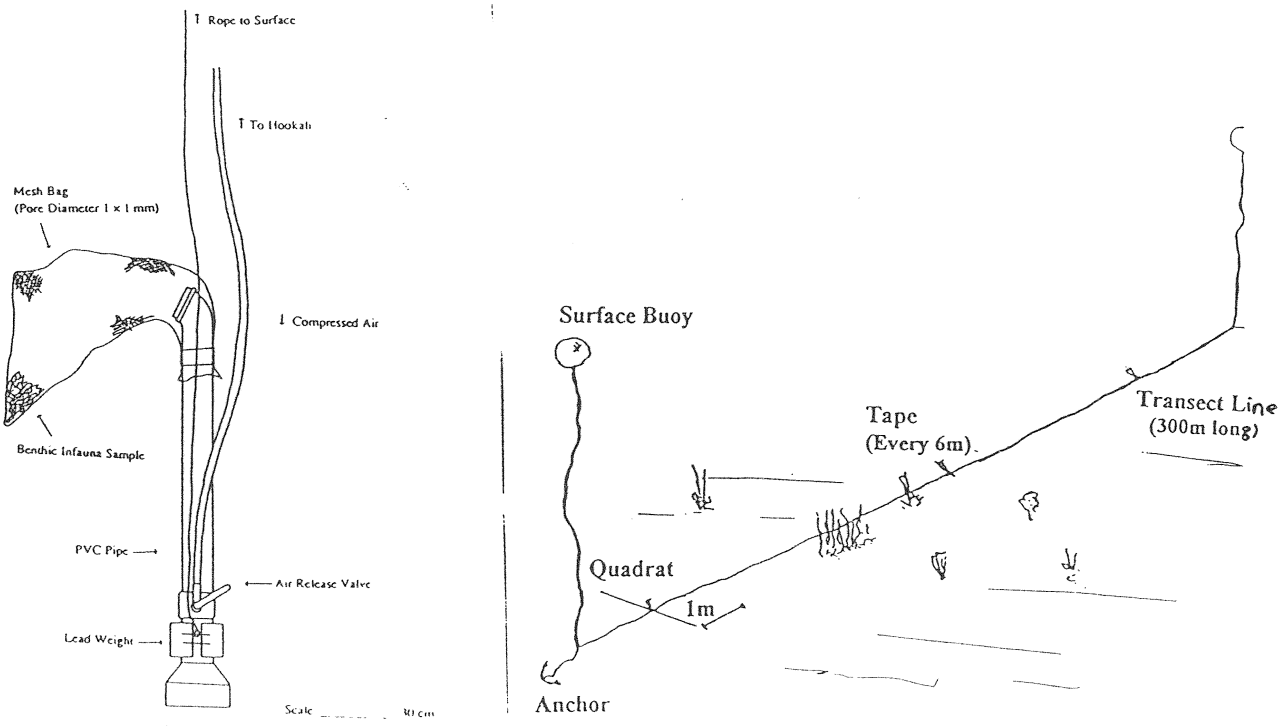
The bag, with the trapped sediment, would be sent to the surface and emptied into labelled plastic jars. The sample was then fixed in a solution of 4% formaldehyde in seawater to which a biological

stain, Phloxine B, was added. This stain increased the accuracy of the sorting process by staining living animal tissue bright pink.

Modification of the sampler so that it could be operated remotely was relatively simple. However, some aspects of sample collection could not be controlled as strictly as when a diver is using the device. The main problems related to the positioning of the sampler on the seafloor and the volume of sediment collected. A length of rope was attached to the front of the PVC tube and ran along its length. This served to keep the sampler upright on the bottom. A PVC funnel (200 mm diameter) was glued to the front of the sampler. This end was also strapped with a considerable quantity of lead weight. The funnel acted to increase the area of substrate available to the sampler and the weight ensured that the sampler struck the bottom upright and with some force. Testing of the sampler off the coast at Pt Stanvac, 20 km south of Adelaide, where diver operated surveys had been performed provided a favourable comparison (Miller, 1995). The sampler very quickly embedded in the course sediment of this site and a considerable quantity was collected in the bag.

Three cages associated with two commercial farms and the research farm and three control sites were surveyed (Fig. 1). At each cage the suction sampler was dropped between the cage and the predator net with the hookah running. The moment the sampler struck the bottom timing was started. The sampler was left undisturbed for twenty seconds, then agitated gently for five seconds and left again for a further ten seconds. The sampler was then lifted to a height of approximately 4 m above the substrate and then dropped again. The sequence of times (twenty seconds in place, five seconds agitation, ten seconds in place) was then repeated before the sampler was brought to the surface. This ensured that the upper 10 to 15 cm of sediment where the vast majority of infaunal animals are found was collected.

Figure 2. Diagram of the remote suction sampler employed in the infauna survey and the layout of the epibenthic transect for diver surveys.



Five samples were collected at each location (0, 2, 20, 100 and 200 m distance from the cage). The 0 m distance was taken to be samples taken from between the predator and cage nets. Each sample was placed in a labelled jar to which the staining fixative was added (4% formaldehyde/seawater with Phloxine B). At 0 and 2 m the samples were collected while working from the cage. For the rest a transect line, attached to the cage, was used as a measure of distance. This involved laying off from the anchor and pulling back toward the cage on the transect rope.

Control transects were positioned in the same manner as video transects. To define distances the transect rope, with an anchor at one end, was used to hold the boat. The normal boat anchor was used to keep the line as straight as possible in the desired direction. Distances were let out along this line as required.

There were no differences in the methods for infauna sampling between summer and winter except that, in summer, the 0 m samples were not taken between the cage and predator nets. Instead the sampler was dropped as close to the outside of the predator net as possible.

Sorting of the sediment samples was a labor intensive process. The fixed, stained samples were drained and washed before being weighed. The sediment was then washed into a deep tray with a white base. A brightly lit magnifying glass (Magilamp) was used to sort through the sample with a pair of fine forceps. All of the stained macro-invertebrates (or parts thereof) were removed from the sediment and sorted into groups which were classified as far as possible (Appendix 2). These groups were counted and then placed into labelled vials in a solution of alcohol, glycerol and distilled water. The reference collection for infauna animals is located at the University of Adelaide, Department of Botany.

3.1.3. Photorespirometer measurements

One of the key indicators of the condition of any biological system is a measurement of the amount of available oxygen. With regard to tuna farming, highly oxygenated cages are capable of supporting, healthier populations. Oxygen depletion could result in more disease prone and less productive stock. Understanding the sources and sinks for oxygen in tuna cages is important if we wish to understand the tropho-dynamics of these systems.

A subjective assessment was made of the oxygen producers and consumers in and around the tuna cages. Observation of firstly the degree of fouling on the cages, which was extreme in some cases, and secondly the composition of this fouling, which was predominantly heterotrophic prompted a suspicion that the cages could become oxygen depleted.

Based on these observations it was decided that an investigation of the oxygen content of tuna cages was warranted. A fully automated photorespirometer was used to obtain information on the oxygen content in the lower portion of different cages (Table 2; Fig. 1).

Table 2. Cages in which the photorespirometer was deployed.

Farm/Cage	August 1994	October 1994	February 1995
Research Farm Cage 8	Yes	Yes	Yes
Research Farm Pilchard Test Feed	Yes	No	No
Commercial Farm 1	Yes	Yes	Yes
Commercial Farm 2	Yes	Yes	Yes*

* indicates that the position of the cage was the same as for the previous deployment but the cage itself was different.

The system employed in this study was designed and built by the Botany Department at Adelaide University (see description in Cheshire *et al.*, in press). The device measures the ambient status of both light and temperature as well as the oxygen content at five electrodes every twenty seconds for periods of up to twenty four hours.

Usually a photorespirometer will measure the oxygen exchange of samples isolated in sealed chambers. For this study the five electrodes were mounted in open water at intervals of 0.5 m above the bottom of the tuna cage. A small stirrer was used to maintain water flow over each electrode to reduce boundary layer effects. At the end of twenty four hours the system was retrieved and the data downloaded. The oxygen content at each electrode (converted to $\mu M\ O_2$) was then plotted against time.

A number of problems were encountered in the course of this study. Firstly the deployment depth (16 - 18 m) caused problems for the electrodes, many of which had to be replaced. Secondly the stirrer units were prone to becoming clogged with strands of macro-algal detritus. This reduced the effectiveness of the stirrers and the accuracy of the electrodes. Finally the calibration for the electrodes was conducted at the depth of the deepest electrode which meant that once mounted at the correct depth, most electrodes were slightly off the correct calibration.

In February a system was developed which prevented the stirrers from clogging and allowed for correct calibration at the right depth. Electrode failure continued to be a problem.

The investigation of oxygen content in tuna cages with the above methods was exploratory. A more thorough investigation of the sources and sinks for oxygen in tuna cages was completed by E. Cronin (Honours student, The University of Adelaide). This work has been written as a thesis (copy attached).

3.1.4. Sediment chemistry

Current feeding methods involve hand shovelling thawed pilchards (*Sardinops neopilchardus*) into cages, and/or direct placement of frozen pilchard blocks in small floating enclosures within cages in which pilchards slowly thaw and sink to the tuna below.

The feed conversion ratio for tuna fed on this diet was found to be 13:1 in an experimental feed trial but is higher for commercial operations (17 - 20:1; Smart and Clarke, unpublished). In particular the frozen block method relies heavily on the judgement of feed staff and the observations of divers in order to prevent overfeeding. In addition the mesh in the base of the floating enclosure may become blocked with pilchards that need to be discarded before new blocks are added.

Most of this solid waste (uneaten food and faecal material) settles to the sediment under the cages causing changes in sediment chemistry and the ecology of macrobenthos (Brown *et al.*, 1987). This is due to enhanced aerobic and anaerobic microbial activity in the enriched sediment and results in oxygen depletion, and low oxidation-reduction (E_h) potentials characteristic of anaerobic marine sedimentary environments (Hargrave *et al.*, 1993). In extreme cases the production of toxic substances such as hydrogen sulphide (H_2S) and methane (CH_4) could occur. Reduced dissolved oxygen and the presence of toxins cause undesirable impacts on the local environment and the finfish species in aquaculture (see thesis by E. Cronin for detailed review of this issue).

3.1.4.1.Redox potentials

A rapid method for assessing the redox levels of soft-mud marine sediments was developed by Pearson and Stanley (1979) as a means of assessing the effect of organic pollution with measurements from 'undisturbed' core samples removed using a Craib corer. Such an approach is not without problems. Core compaction and other physico-chemical changes experienced by the sample as it is transferred to the surface can cause changes in the E_h measurement.

This study measured the E_h of sediments under a recently destocked tuna sea-cage. *In situ*, measurements were made by waterproofing a commercially available redox probe and meter, thereby negating the problems inherent with measurement from core samples. In addition, the effects of, harrowing, and adding gram negative bacteria, on sediment recovery rates were investigated using E_h levels as an index of sediment status.

The Research Farm (Fig. 1) was destocked 1 week prior to experiments. The cage had been stocked at 0.8 kg/m^3 for 9 months prior which is about one third of commercial stocking rates. Quadrats (1 m square) were marked with pegs hammered into the sediment under the cage (depth = 17 m). These were arranged as 3 quadrat clusters for each of the treatments of control, harrowing, and bacteria, spread uniformly over the bottom at 5 different locations. Following initial E_h readings, the 5 harrowing quadrats were harrowed with a garden rake, and 1 kg bags of bacteria + substrate were added to each of the bacteria quadrats (bacterial applications were added once-weekly over the 4 week period). The bacteria used was gram positive DMS-1000 Series or 'sludge doctor®', supplied by Admac Agencies.

3.1.4.2.Methanogenesis

In enriched marine sediments it is likely that sulphate reduction, resulting in the formation of hydrogen sulphide (H_2S) is quantitatively the most important process. At some salmonid marine cage farms the level of sulphate reduction has been reported to be sufficient to allow some H_2S to escape from the sediment along with other gases. Capone and Kiene (1988) found that, in marine systems with high deposition rates of organic matter, sulphate can be depleted to the extent that methanogenesis takes on quantitative significance. Samuelson *et al.* (1988) established the composition of gas bubbles released from enriched sediments to be 70% CH_4, 28% CO_2, and 2% H_2S. It is not known whether H_2S in gas bubbles or dissolved in the water is the source of H_2S which can affect fish health, however, the release of gas bubbles from enriched sediments would also act as a mechanism for transporting pathogenic bacteria living in the sediment to fish (Gowen *et al.*, 1991).

On the 9th November 1994, 3 in situ E_h measurements were taken per quadrat at a depth of 50 mm. The electrode used was a combined platinum cap electrode for sample changer with plug-in-head and SGJ, supplied by John Morris Scientific (Type no. 6.0418.120). Two *in situ* E_h measurements per quadrat were taken at the end of the 4 week period. E_h readings were cross checked with test core samples, using 40 mm PVC pipe and end caps.

(a) Gas composition - An upturned funnel with a rubber-stoppered glass vial placed over the spout was used to collect gas bubbles. The funnel was placed over enriched sediments under a tuna cage and a rod was pushed into the sediments and agitated to liberate trapped bubbles. The gas bubbles were collected in the vial which was stoppered and brought to the surface for analysis.

(b) Gas quantity - 5 upturned funnels with legs inserted into the sediments were placed randomly under a tuna cage. A 25 mL syringe housing was placed over the spout of each funnel and held in place by a weighted collar. The funnels were checked at regular intervals to establish the quantity of gas liberated over time as the amount of water displaced out of the syringe housings.

3.2. Experimental approach to assess the impact of tuna farming

The preferred methodology for the assessment of any environmental impact is to undertake a Before and After Control and Impact (or BACI design) survey. Such an approach allows for the direct comparison of the impacted area with the same location prior to disturbance and in comparison with control (unimpacted) sites. The nature of this study has meant that there was no possibility of surveying prior to the impact of tuna cages. The *post hoc* assessment of the effect of tuna cages through a comparison to areas without cages has two main limitations:

i) Differences may exist between the cage sites and the undisturbed sites which are not the result of tuna farming.

ii) The high degree of variability in natural systems may mask impacts except when subjected to very detailed (and hence costly) investigations.

The effects of human activities on the marine environment is inevitable but it is important to distinguish between putative and real impacts. The ultimate goal of any impact assessment is to determine whether or not there has been a disruptive influence on the system. Attention should be directed to the sampling design of a survey and the conclusions reached, as the predictive ability of any study is circumscribed by the limitations inherent in the design. There is ongoing debate over many (if not most) aspects of the assessment process which suggests that there is no standardised approach to environmental monitoring (Underwood, 1991; Fairweather, 1991; Smith, 1991; Keough and Quinn, 1991).

This study has attempted to account for these limitations but it remains imperative that management decisions are made with recognition of these limitations. As a corollary, the principle that the best available information should be employed in guiding management decisions should also be followed.

Research on souring of the seafloor around sea-cages has been undertaken elsewhere in the world (Brown *et al.*, 1987; Frid and Mercer, 1989; Gowen and Bradbury, 1987; Kaspar *et al.*, 1988; Lumb, 1989; Tsutsumi, 1995) but this has pertained primarily to salmonid farming. Within Australia most data relates to the farming of Atlantic salmon in Tasmania (eg Hodson and Burke, 1994) although Bond (1993) does deal with a number of water quality issues in the vicinity of tuna cages and the dispersal of organic nutrients. While there is little doubt as to the polluting effect of sea-cages on the localised surrounding environments for comparison purposes there are a number of critical

differences (not the least of which include the farming of different species of fish, the use of different feed types, and the warmer location) that make this study unique.

In most natural systems there is a high degree of spatial heterogeneity (or patchiness) in the distribution and abundance of organisms. Such patchiness gives rise to high variability in the estimation of environmental parameters such as the average abundance or richness of taxa. In order to establish difference in systems which are highly variable it is necessary to either take a large number of individual samples or the samples need to cover a large enough area so as to effectively integrate across the patches.

Data collection in any project must be circumscribed by pragmatic considerations not the least of which is the availability of funds. This means that conclusions that are drawn are based on the best available information with a clear recognition of the limitations of the sampling design. This, unfortunately, does not mean that they can't be wrong. In such cases it is important to understand the nature and implications of such errors.

3.2.1. Implications of errors

The assumption is made in any survey that the impact sites are no different to the unimpacted sites (ie. there is no impact). This is commonly called the Null Hypothesis (H_0). The alternative hypothesis is that an impact does exist.

In statistical terms we would say:

H_0: The sites are not different.

H_1: The sites are different.

The data collected in the study is used to test the hypotheses; one will be accepted, the other will be rejected.

There is a real risk that we may accept the wrong hypothesis. This is known as a type 1 or type 2 error depending on the circumstances as follows:

	If in reality:	
	H_0 is correct	H_1 is correct
Accept H_1	Type 1 Error	Correct Decision
Accept H_0	Correct Decision	Type 2 Error

A type 1 error arises because the data (by chance) makes the sites appear different when in reality they are not (Zar, 1984). In this case we may act upon the assumption of an impact that does not exist. Our response will be conservative in environmental terms and this error is hence less of a problem.

A type 2 error arises because the study was not powerful enough to detect a difference that was present (Zar, 1984). This is the most serious form of error as it incorrectly concludes that the impact has had no effect on the system when in reality it has. It is also the most likely form of error given a patchy system for which we only have *post hoc* data (such as the system in Boston Bay).

Managers should be aware of the possibility of error, particularly as it relates to *post hoc* surveys of impacts, in which impact assessment is based solely on a comparison with surrounding areas.

A major issue in the design of environmental impact assessments is therefore to determine the likelihood of making a type 2 error. The probability of not making such an error is referred to as the "power" of the study and is discussed in the following.

4. Results - Power of sampling methods and cost estimates

4.1. Power analysis - benefits

A major objective in using surveys to assess environmental impacts is to determine whether the communities in a putatively impacted area differ from those in a non-impacted control area. Generally speaking it is hoped that there will be no significant difference between the two areas. Such a result would be used to conclude that there is "no impact". The major problem with this approach is that a poorly designed experiment, with too few replicates, may not have the power to detect a difference in the systems. In such a circumstance it is possible that, in concluding that no impact has occurred, a type 2 error will be made. Such an error will have significant repercussions for the ongoing management of the system.

Power analyses are used to determine whether the null hypothesis ("that there is no impact") can be accepted or whether it results from insufficient replication. In such cases, if no significant differences are detected and the power is not high, then nothing can be concluded.

Power is generally defined as a measure of the confidence we have in accepting the null hypothesis. In environmental impact assessments we would generally expect a power greater than 0.85 (85% chance that acceptance of the null hypothesis will not create a type 2 error; 15% chance that acceptance of the null hypothesis will be incorrect).

An alternative approach is to define the least significant number (LSN) which is the minimum number of replicates needed to detect a change of a given magnitude whilst providing an appropriate level of power. For this project it was agreed that for most parameters the surveys should be able to detect a change of 20% in any of a range of parameters with a power of 0.85.

The mathematical calculation of power is specific to the design of the experiments and includes the number of comparisons (groups) and the variance associated with given parameters. In order to standardise this we have defined the power analyses in terms a single parameter, this being the distance from the cage with close (< 20 m), distant (≥ 20 m) but still on the cage transect and the control (≥ 1000 m). This allowed for a one way comparison with three levels.

Three parameters were analysed: taxa richness, Shannon diversity (of taxa) and equitibility (taxa evenness).

The cost-benefit analysis can therefore be defined in terms of the cost of undertaking a survey which provides the desired benefit (ie. has a power of 0.85 to detect a Δ of 20%). This is defined as:

$$\text{LSN} \times \text{CostPerReplicate} = \text{CostOfPower}$$

The following provides a discussion of power with respect to the various survey methods employed in this study.

Table 3. Taxa richness, Shannon diversity and taxa equitibility for each of the survey approaches used in this study. Each index was considered across three distance classes (close, distant and control). Where N is the number of samples collected in this study across all locations, LSN is the least number of samples required to determine a 20% change in the index with a power of 0.85.

Survey Method	Index	N	Mean across all distances	Δ = 20% of the mean	Power	Number of samples to achieve a power of 0.85	LSN	P (ANOVA)
Diver Epibenthic	Taxa Richness	71	3.90	0.78	0.98	40	24	0.01
	Shannon Diversity	71	0.43	0.09	0.97	45	26	0.28
	Equitibility	71	0.31	0.06	0.99	25	16	0.05
Video Epibenthic	Taxa Richness	194	3.10	0.62	1.00	45	25	0.8
	Shannon Diversity	194	0.40	0.08	1.00	50	31	0.96
	Equitibility	194	0.32	0.06	1.00	40	25	0.93
Infauna	Taxa Richness	161	8.94	1.79	1.00	43	26	0.27
	Shannon Diversity	161	0.49	0.09	1.00	30	19	0.00
	Equitibility	161	0.22	0.05	1.00	19	12	0.00

4.1.1. Power analysis of diver collected epibenthic data.

Both taxa richness and equability changed significantly depending on the distance from the tuna cage. Taxa richness at both short and intermediate distances was significantly higher than the control (Table 3; Fig. 3A) and equitibility was significantly different between all distance classes (Table 3; Fig. 3B). There were no significant differences in Shannon diversity (Table 3; Fig. 3C).

Using the most conservative index - Shannon diversity - a 20% change with a minium power of 0.85 should be detected after 45 samples. Our data set, which is comprised of 71 samples is more than adequate to the task and gave a power of 0.97. The least significant number (LSN) was 26.

4.1.2. Power analysis of the video data

There were no significant differences detected between any distance classes for taxa richness, Shannon diversity or equitibility (Table 3; Fig. 4A-C). This is probably a reflection of the comparatively low taxonomic resolution in the data which has resulted in fewer taxa to characterise each site (diver survey had 20 taxa, infauna survey had 34 taxa while the video survey had 16 taxa) and the difficulty in counting individuals of these taxa compared to the diver survey.

The Shannon diversity indicated that the minimum number of samples required to detect a difference between the means would be 31. Our power, with 194 samples, was 1.00 (Table 3).

4.1.3. Power analysis of the infauna data

Taxa richness was not significantly different at any distance class (Table 3; Fig. 5A). There were significant differences between sites for both the equitibility and Shannon diversity. Equitibility of

taxa was lower close to cages than on the controls (Table 3; Fig. 5B). At increased distance from the cage the diversity was slightly higher than the controls although this was not significant. Shannon diversity close to the cages was significantly lower than controls (Table 3; Fig. 5C). Further from the cages the diversity was much the same as for the controls.

The most conservative index in this instance was taxa richness. A total of 43 samples was required to obtain a power of 0.85 with a 20% change. The number of samples collected (161) was far in excess of the number required and gave a power of 1.00. The least significant number (LSN) was 26.

4.1.4. Photorespirometry deployments and REDOX measurements

Neither the photorespirometer work nor measurements of Eh potential have been considered in terms of power.

4.1.5. Conclusions

We have only considered taxa richness, Shannon diversity and taxa equitibility across three distance classes (close, distant and control) for the diver epibenthic, video epibenthic and infauna data. There are a large number of other indices that can be used as descriptors of the data and there is considerable debate as to the validity or usefulness of any of them (including those employed here). The indices we have used are probably the most common but the efficacy of a variety of measures should be tested to fully appreciate which are best for this system.

The number of samples required by each of the survey methods were all very similar (40-50) as were the least significant numbers (LSN's = 26-31.). This similarity probably reflects the fact that each of the methods is applied in essentially the same system and thus are operating on the same scales and/or gradients. In all cases the number of samples collected in the survey was considerably in excess of the actual number required (with the power considered at 0.85 for a 20% change).

Figure 3A. Taxa Richness By Distance Class

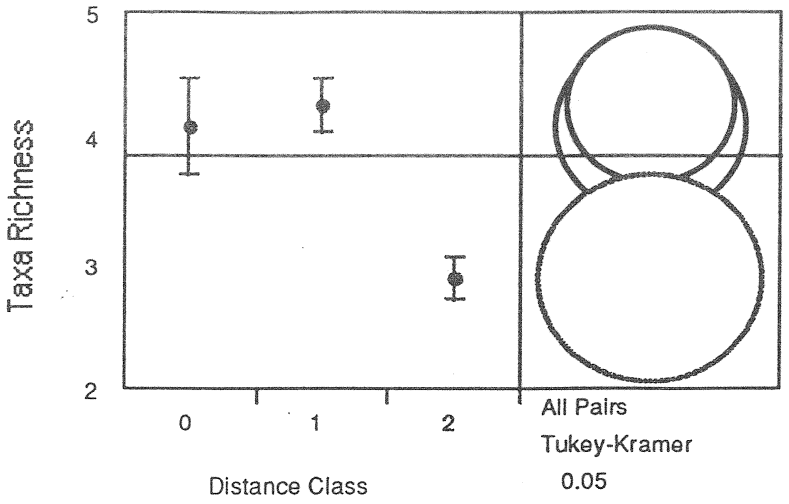


Figure 3B. Shannon Diversity By Distance Class

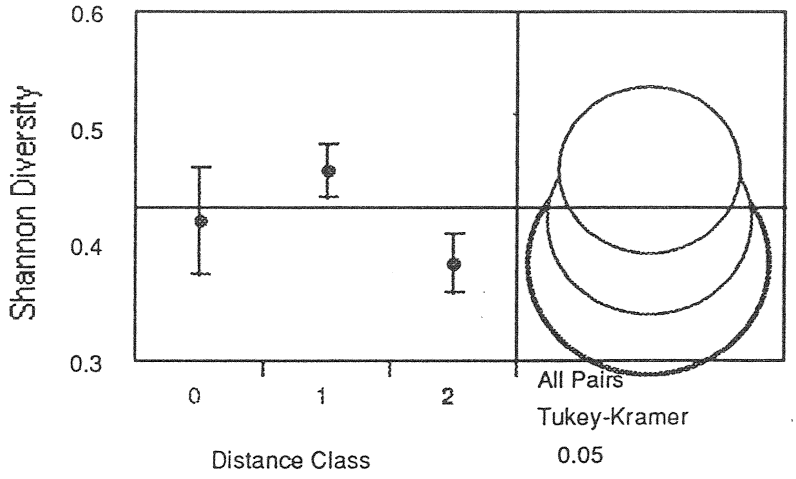


Figure 3C. Equitability By Distance Class

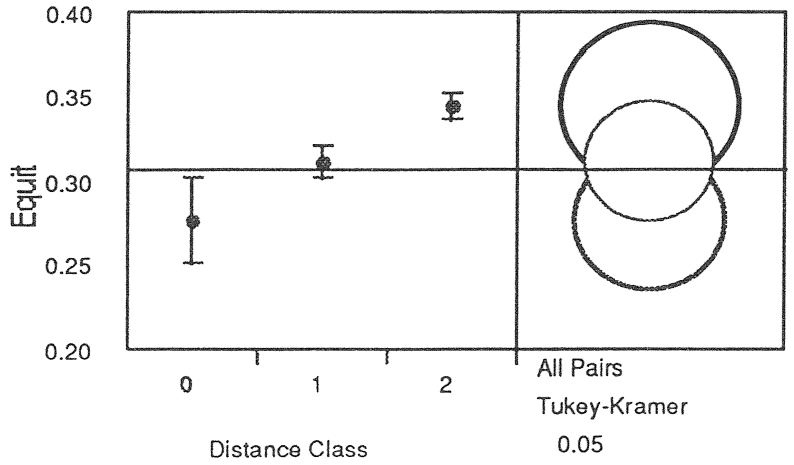


Figure 4A. Taxa Richness By Distance Class

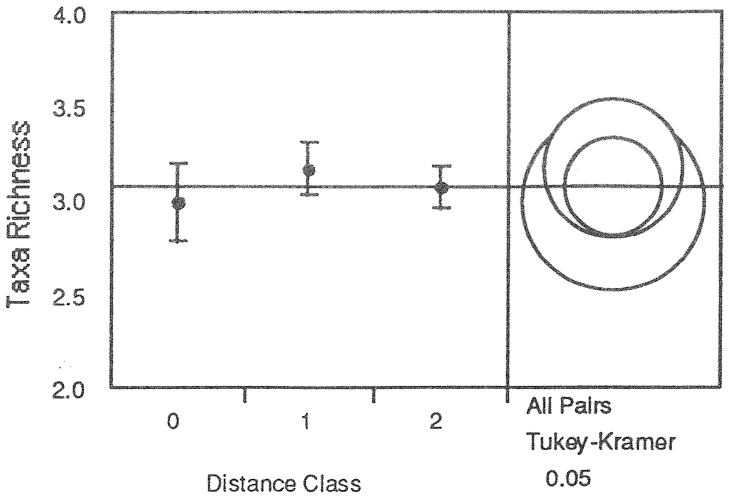


Figure 4B. Shannon Diversity By Distance Class

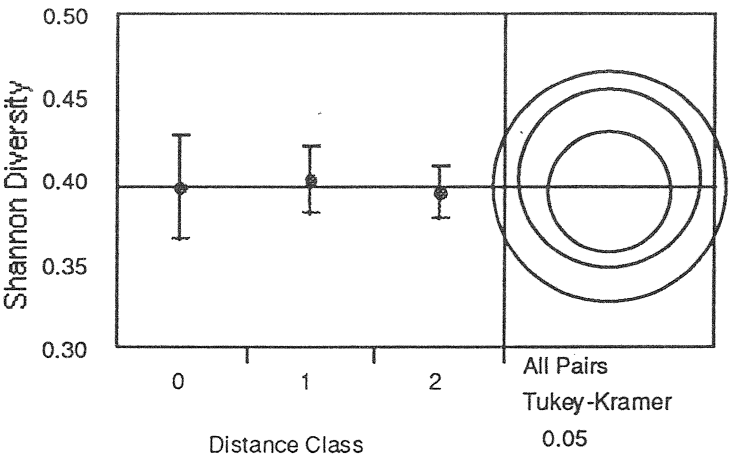


Figure 4C. Equitability By Distance Class

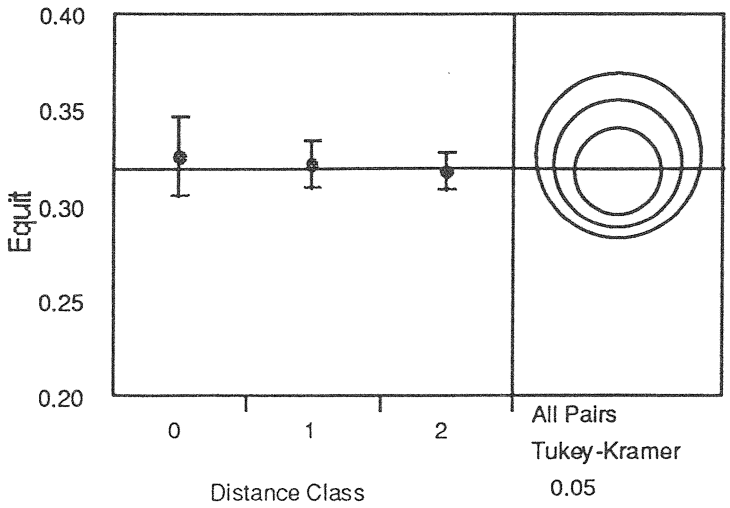


Figure 5A. Taxa Richness By Distance Class

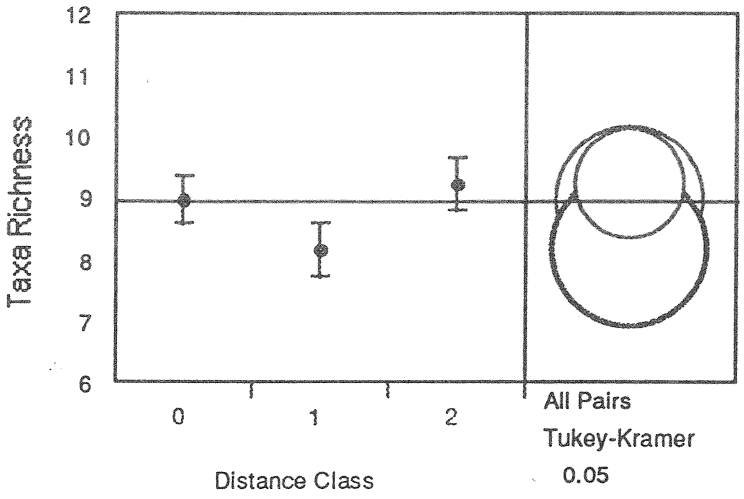


Figure 5B. Shannon Diversity By Distance Class

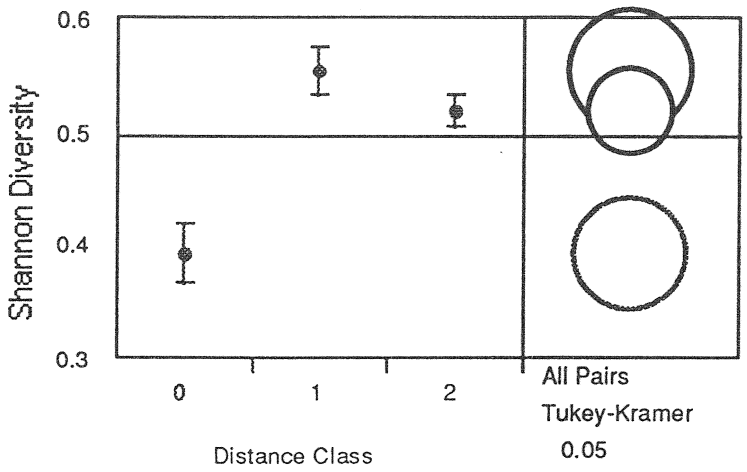
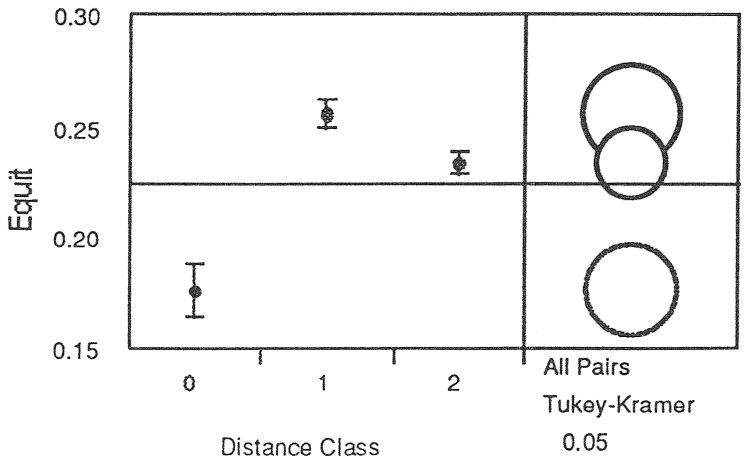


Figure 5C. Equitability By Distance Class



4.2. Cost-benefit analysis of survey protocols

In the following analysis the data will be assessed at the rate of cost per transects worth of information. The organisation of the field teams will be discussed and it will be assumed that the procedure will provide for the most cost effective application of each method.

4.2.1. Epibenthic surveys - data collected by diver.

The typical costing is based on a dive team of 4 which undertakes 4 dives (transects) per day. It is assumed that each diver undertakes two dives and that each dive requires two divers who collect replicate data sets for the same transect. The transect length is 200 m with quadrats at 0, 5, 10, 15, 20, 50, 100, 150 and 200 m.

Item	Number of units	Units	Unit cost $	Marginal cost $	Totals $
Personnel:					
Divers with biological training	4	hrs	20	80	
Boat persons (alternate with divers)	4	hrs	20	80	
Transcribe data to computer	1	hrs	20	20	
Analyse and interpret data	4	hrs	20	80	
					260
Equipment/Facilities:					
Boat time	2	hrs	15	30	
Measuring tapes and transect lines	1	depr.	10	10	
					40
Consumables:					
Scuba fills	2	fills	6	12	
Fixatives/collection bottles	1	misc.	20	20	
					32
Sub-total - marginal cost per transect					**332**
Provisional costs:					
Delay due to bad weather[1]	30% of field personnel			78	
Diver training (in biology)[2]	10%			35	
Insurance for hazardous conditions[3]	15%			50	
					163
Total cost per transect					**495**
Total cost per quadrat					**55**

4.2.1.1. Discussion of provisional costs:

[1]Bad weather is a major problem for diver surveys. Diver safety and seafloor visibility are both compromised under adverse conditions.

[2]In order to ensure data quality divers need to be trained in biology (either formally or through training programs).

[3]Diver safety is of major concern. Repetitive dive schedules need to be rigidly followed. There is also a potential risk from shark attack which cannot be costed in dollar terms.

4.2.1.2. Data quality:

High quality in terms of both taxonomic resolution and accuracy of abundance estimates.

From the power analysis the maximum number of quadrats required to detect a 20% change in either of the taxa richness, Shannon diversity or equitibility of taxa was 26. On the basis of cost per quadrat this gives:

$$26 \times \$55 = \$1,430$$

4.2.2. Epibenthic surveys - data collected by video

The typical costing based on a field team of 2 which undertakes 4 transects per day. The transect length is 150 m with quadrats at 0, 2, 10, 20, 40, 60, 80, 100, 120, 140 and 150 m.

Item	Number of units	Units	Unit cost $	Marginal cost $	Totals $
Personnel:					
Boat persons	4	hrs	15	60	
Transcribe data to computer	1	hrs	20	20	
Analyse and interpret data	3	hrs	20	60	
					140
Equipment/Facilities:					
Boat time	2	hrs	15	30	
Video camera, sled and VCR	1	depr.	20	20	
					50
Consumables:					
Video Tapes	1	1	15	15	
					15
Sub-total - marginal cost per transect					**237**
Provisional costs:					
Delay due to bad weather[1]	30% of field personnel			42	
					42
Total cost per transect					**279**
Total cost per quadrat					**26**

4.2.2.1. Discussion of provisional costs:

[1]Bad weather is less of a problem for video surveys. The commitment to a smaller boat team means that the cost of delays and rescheduling is proportionally less.

4.2.2.2. Data quality:

Low resolution in taxonomic terms, with many abundance estimates difficult to make due to overlapping fields of view. The permanent record that is obtained from these surveys does allow re-analysis/interpretation of the tapes and immediate discussion of gross results with the tuna farm operator.

From the power analysis the maximum number of quadrats required to detect a 20% change in either of the taxa richness, Shannon diversity or equitibility of taxa was 31. On the basis of cost per quadrat this gives:

$$31 \times \$26 = \$806$$

4.2.3. Infauna surveys - data collected by remote sampler

The typical costing based on a field team of 2 which undertakes 3 transects per day. The transect length is 200 m with 3 replicate samples collected at each of the following distances along the transect 0, 2, 10, 20, 50, 100, 150 m.

Item	Number of units	Units	Unit cost $	Marginal cost $	Totals $
Personnel:					
Boat persons	6	hrs	15	90	
Sort and identify infauna	72	hrs	15	1080	
Transcribe data to computer	1	hrs	20	20	
Analyse and interpret data	6	hrs	20	120	
					1310
Equipment/Facilities:					
Boat time	2	hrs	15	30	
Suction sampler/compressor	1	depr.	20	20	
Sorting facilities[1]	24	daily bench fee	50	1200	
					1250
Consumables:					
Fixatives/collection bottles	1	misc.	100	100	
Specimen jars	1	misc.	25	25	
					125
Sub-total - marginal cost per transect					**2685**
Provisional costs:					
Delay due to bad weather[2]	30% of field personnel			27	
					27
Total cost per transect					**2712**
Total cost per sample					**129**

4.2.3.1. Discussion of costings:

[1]Sorting facilities are costed in and account for the need to have a well setup laboratory with adequate fume-cupboards and microscopes. They have been accounted for with a nominal bench fee of $50 per day.

[2]Bad weather is less of a problem for remote infauna surveys. This costing is based on 30% of boat time with totals $140 per transect. The commitment to a smaller boat team means that the cost of delays and rescheduling is proportionally less.

4.2.3.2. Data quality:

Very high resolution in taxonomic terms, and abundance estimates are very accurate.

From the power analysis the maximum number of quadrats required to detect a 20% change in either of the taxa richness, Shannon diversity or equitibility of taxa was 26. On the basis of cost per quadrat this gives:

$$26 \times \$129 = \$3,354$$

4.2.4. Redox Measurements

Th typical costing is based on a minimum field team of three, at least two of which must be experienced divers. Costing is based on the collection of E_h samples and reading gas collection devices for one day at one cage. Cost of the bacterial treatment has not been included.

Item	Number of units	Units	Unit cost $	Marginal cost $	Totals $
Personnel:					
Boat person	6	hrs	15	90	
Dive team	12	hrs	15	180	
Preparation of equipment	2	hrs	15	30	
Download and analyse data	4	hrs	15	60	
					360
Equipment/Facilities:					
Boat time	6	hrs	15	90	
					90
Consumables:					
Batteries	4	misc.	4	16	
Computer disks	1	misc.	5	5	
Air fills	3	misc.	6	18	
Bottles	5	misc.	1	5	
Chemicals	1	misc.	10	10	
					59
Sub-total - marginal cost per transect					**504**
Provisional costs:					
Delay due to bad weather[3]	30%			152	
Down time for essential repairs	10%			51	
					202
Total cost per transect					**706**
Total cost per replicate (gas collectors - 25 %)					**12**
Total cost per replicate (E_h measurements - 75 %)					**36**

4.2.4.1. Discussion of costings:

Costs assume that all measurements can be made within the span of a single dive. Costs per replicate are based on the proportion of the total cost in time and resources allocated to either E_h or gas release measurements. In this instance the E_h has been placed at 75 % of the total cost. The remaining 25 % is allocated to gas collection.

Bad weather is less of a problem for sediment chemistry surveys although it is not certain that sediment churned up by rough conditions would give a meaningful result.

4.2.4.2. Data quality:

Redox values obtained from this study were much lower than those observed in other studies. While this does not invalidate the method, further development and testing may be required. No gas was detected evolving from the sediment (possible because the E_h was so low). This method requires more development before it can be used routinely.

4.2.5. Photorespirometer Deployments

The typical costing is based on a field team of 3 for a total work time of 1 day. This time is spread over a period of 2 days as each deployment requires twenty four hours to complete.

Item	Number of units	Units	Unit cost $	Marginal cost $	Totals $
Personnel:					
Boat person	6	hrs	15	90	
Dive team	12	hrs	15	180	
Preparation of equipment	2	hrs	15	30	
Download and analyse data	1	hrs	15	15	
					315
Equipment/Facilities:					
Photorespirometer[1]	1	daily rate	150	150	
Lab space[2]	1	daily bench fee	50	50	
Boat time	6	hrs	15	90	
					290
Consumables:					
Batteries	6	misc.	4	24	
Computer disks	1	misc.	5	5	
Air fills	4	misc.	6	24	
Bags	5	misc.	1	5	
Fixative	2	litres	1	2	
Bottles	5	misc.	1	5	
					65
Sub-total - marginal cost per transect					**670**
Provisional costs:					
Delay due to bad weather[3]	30%			205	
Down time for essential repairs	10%			71	
					276
Total cost per deployment					**946**
Total cost per replicate					**190**

4.2.5.1. Discussion of costings:

[1]The daily rate for the use of the photorespirometer covers the eventual cost of replacement (conservatively placed at $20,000) and is the standard rate for users of the system.

[2]Laboratory facilities are costed to account for the need to have a well setup laboratory with adequate fume-cupboards and microscopes. They have been accounted for with a nominal bench fee of $50 per day.

[3]Delays due to bad weather have a two-fold threat as a delay in the deployment of the system can mean the loss of a day. A delay in the retrieval of the system can cause the degradation of the

photorespirometers batteries. As a result the provisional cost covers expenses for the whole deployment (not just the field component).

4.2.5.2. Data quality:

For a detailed synthesis refer to Cronin (1995).

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Appendix 1: Database design

This section provides details of the optimal design for a relational database to store and summarise survey data. Such a database will provide an invaluable tool for managers for keeping track of information on environmental quality.

Relational databases employ a number of constructs for the storage and manipulation of data. Data are stored in **Tables**. Manipulation of data occurs through **Queries**. Tables store data in columns (or fields), the format of which is defined by the user. The philosophy of design for the database developed for this study is based on that employed for The ASEAN-Australia Living Coastal Resources Database (Bujang *et al.*, 1993).

The majority of biological/environmental databases require three tables as the most efficient means of storing information. These tables consist of:

Sample Information - A table in which all of the site information for the samples are located. The types of data stored in this table are the Date, Location of sampling, Comments on the weather conditions and any other ambient details that might be relevant to the batch of data collected at that site and at that time. A SAMPLE_ID field within this table is used as a common link to other tables.

Taxonomic Information - In this table the field TAXCODE is used to link the common names for organisms in the Abundance Data table to biologically informative names. A field generally called ANALYSIS_ID is included. This field contains a label which indicates the level at which the data will be aggregated and analysed. The Taxonomic table is essential where a number of different persons have collected the data as there are inevitable differences in the common names between different observers/observations.

Abundance Data - The table in which all of the names and numbers of organisms for the basic sampling units (ie quadrats) are located. Records within this table can be linked, via the TAXCODE field, to the Taxonomic Information Table to allow for summaries at different taxonomic levels or to the Sample Information Table (via the SAMPLE_ID field) to allow for summaries at different spatial scales.

The structures of all three survey databases (Diver Epibenthic, Video Epibenthic and Infauna) are highly similar in their basic form, however, there are some specific differences that are particular to each survey. For this reason the basic design of all three databases is depicted. These designs can be constructed under a number of different database packages, we have used Microsoft ACCESS.

Diver Transects

Diver - Sample information

Column	Data Type	Length	Description
Year	Number (Integer)	4	Year that the transect was run
Site	Text	50	Location of the transect
Date	Date	10	Date that the transect was run
SAMPLE_ID	Number (Integer)	4	Unique number for each transect
Diver_id	Text	20	Unique label for each diver
Dive time	Number (Integer)	3	Time (in minutes) for the dive time from when the divers leave the surface to when they return.
Bottom time	Number (Integer)	3	Time (in minutes) spent on the bottom reading the transect
Depth	Number (Single)	3	Depth of the transect (m)
Weather condition/ Comments	Memo	-	Short summary of weather conditions during the transect and any comments other than weather that might be relevant

Diver - Abundance data

Field Name	Data Type	Length	Description
SAMPLE_ID	Number (Integer)	4	Unique number for each divers data within each transect (ie there will be two for each transect)
Quadrat	Number (Integer)	8	Quadrat distance 0,5,10,15,20,50,100,150 and 200 m
TAXCODE	Text	20	Label used to describe the organism observed in the quadrat
Abundance	Number (Integer)	8	Abundance value for the number of organisms seen in the quadrat. *Botryocladia* sp. is considered on a scale from 1-5.

Diver - Taxonomic information

Column	Data Type	Length	Description
TAXCODE	Text	20	TAXCODE in the abundance table collection (See above)
Description	Text	50	Short description of the taxa
Phylum	Text	20	Phylum of the TAXCODE
Class	Text	20	Class of the TAXCODE
Order	Text	20	Order of the TAXCODE
Family	Text	20	Family of the TAXCODE
Genus	Text	20	Genus of the TAXCODE
Species	Text	20	Species of the TAXCODE (includes genus name)
ANALYSIS_ID	Text	20	Label given to the TAXCODE for analysis

Infauna transects

Infauna - Transect information

Column	Data Type	Length	Description
Year	Number (Integer)	4	Year of the survey
Site	Text	12	Location of the transect
Date	Date/Time	8	Date that the transect was undertaken
Method	Text	12	The manner in which the suction sample was collected (eg Remote, Diver Operated, etc.)
SAMPLE_ID	Number (Integer)	4	Unique number for each transect
Comments	Memo	-	Description of the conditions for each particular transect

Infauna - Abundance data

Field	Data Type	Length	Description
SAMPLE_ID	Number (Integer)	4	Unique number for each transect
Distance	Number (Integer)	4	Distance along the transect (0, 2, 20, 100 and 200 m)
Replicate	Text	2	Label of the replicate for each distance (A, B, C, D, E)
Weight	Number (Double)	8	Wet weight (in grams) of the sample
TAXCODE	Text	20	Label of the taxa found in each sample
Abundance	Number (Double)	4	Absolute number of each taxa in each sample

Infauna - Taxonomic information

Column	Data Type	Length	Description
TAXCODE	Text	20	TAXCODE from the Abundance Table
Phylum	Text	20	Phylum of the TAXCODE
Class	Text	20	Class of the TAXCODE
Order	Text	20	Order of the TAXCODE
Family	Text	20	Family of the TAXCODE
Genus	Text	20	Genus of the TAXCODE
Species	Text	20	Species of the TAXCODE (includes the genus name)
ANALYSIS_ID	Text	20	Taxa label used in Analysis

Video transects

Video - Transect information

Column	Data Type	Length	Description
Year	Number (Integer)	2	Year in which the sample was collected
Location	Text	20	Place in which the video was taken
Date	Date/Time	8	Date the sample was taken
TRANSECT_ID	Number (Integer)	2	Unique number of the transect
Viewer	Text	10	Person who viewed the transect
Comments	Memo	-	Notes on the location conditions etc

Video - Abundance data

Column	Data Type	Length	Description
TRANSECT_ID	Number (Integer)	4	Unique number of the video transect
Distance	Number (Double)	8	Location along the transect (0,5,10,15,20,50,100, 150 and 200 m)
TAXCODE	Text	20	Taxa found at each location along the transect
Abundance	Number (Double)	4	Number of organisms in each screen view, with macro-algae considered as an estimate of percentage cover of the image

Video - Taxonomic information

Column	Data Type	Length	Description
TAXCODE	Text	20	Reference label for each TAXCODE in the abundance table
Description	Text	50	Short description of the organism
Phylum	Text	15	Phylum of the TAXCODE
Class	Text	20	Class of the TAXCODE
Order	Text	15	Order of the TAXCODE
Family	Text	15	Family of the TAXCODE
Genus	Text	15	Genus of the TAXCODE
Species	Text	25	Species of the TAXCODE (includes genus name)
ANALYSIS_ID	Text	20	Label used in analysis

Appendix 2: Taxa lists from each of the survey methods

List of diver survey taxa

List of ANALYSIS_ID, descriptions of data types of the lifeforms found from diver oriented epibenthic surveys.

Standardised classification used for data analysis	**Specific description of epibenthic lifeforms found**	**Record of abundance**
Anthozoa	Sea Anemone	Count
Ascideacea	Ascidian	Count
Asteroidea	Sea Star	Count
Asteroidea	Star Fish	Count
Bivalvia	Bivalve	Count
Bivalvia	Cockle	Count
Bivalvia	Mussel	Count
Bivalvia	Oyster	Count
Bivalvia	Scallop	Count
Botryocladia	*Botryocladia obovata* (red macro-algae)	Count
Bryozoan	Bryozoan	Count
Cephalopoda	Octopus	Count
Chlorophyta	*Caulerpa* sp.(green macro-algae)	Count
Chlorophyta	*Ulva* sp.(green macro-algae)	Count
Fish	Blenny	Count
Fish	Cling Fish	Count
Fish	Dragonet	Count
Fish	Fish	Count
Fish	Gudgeon	Count
Fish	Stink Fish	Count
Gastropoda	Gastropod	Count
Gastropoda	Snail	Count
Holothuroidea	Sea Cucumber	Count
Malocostraca	Crab	Count
Malocostraca	Hermit Crab	Count
Mollusca	Mollusc	Count
Mollusca Egg	Mollusc Egg	Presence
Ophiuroidea	Brittle Star	Count
Phaeophyta	*Ectocarpus* sp.(brown macro-algae)	Count
Porifera	Sponge	Count
Posidonia Drift	*Posidonia* drift (seagrass fragments)	Presence
Rhodophyta	*Gracilaria* sp.(red macro-algae)	Count
Rhodophyta	Red Macro-algae	Count
Seagrass	*Halophilla* sp.(seagrass)	Count
Seaurchin	Sea Urchin	Count

List of video survey taxa

Taxa defined from the video transects for the October 1994 and February 1995 field trips. Note that, while the taxa described here and in the diver transects are very similar, the level of analysis is different in may cases. This is due to the sparsity of data in some cases. The Crustacea and Drift groups were excluded from the analysis for similar reasons.

Standardised classification used for data analysis	**Specific description of epibenthic lifeforms found**	**Record of abundance**
Annelida	Polycheat head	Count
Anthophyta	Seagrass	Count
Anthozoa	Anthozoan	Count
Ascideacea	Sand Ascidian	Count
Ascidiacea	Ascidian	Count
Ascidiacea	Clearhead Ascidian	Count
Ascidiacea	Colonial Ascidian	Count
Ascidiacea	Large yellow Ascidian	Count
Ascidiacea	Small yellow Ascidian	Count
Ascidiacea	White Ascidian	Count
Ascidiacea	White Colonial Ascidian	Count
Ascidiacea	Yellow Ascidian	Count
Bryozoa	Bryozoan	Count Clumps
Chlorophyta	*Caulerpa cactoides* (green macro-algae)	Count
Chlorophyta	*Ulva* sp.(green gacro-algae)	Count
Crustacea	Hermit Crab	Count
Drift	Drift Algae	Presence
Drift	Drift seagrass (not necessarily *Posidonia*)	Presence
Echinodermata	Sea Star	Count
Echinodermata	Sea Urchin	Count
Echinodermata	Holothurian	Count
Mollusca	Pinna sp.	Count
Mollusca	Queen Scallop	Count
Mollusca	Octopus	Count
Mollusca	Gastropod	Count
Mollusca	Whelk	Count
No Data	Used to indicate that a quadrat was surveyed but nothing was found in it.	-
Osteichthyes	Fish	Count
Osteichthyes	Puffer Fish	Count
Phaeophyta	Brown Algae	Percentage Cover
Phaeophyta	Brown filamentous Algae (growing on the sand)	Percentage Cover

List of video survey taxa cont.

Standardised classification used for data analysis	Specific description of epibenthic lifeform	Record of abundance
Phaeophyta	Filamentous Brown Algae	Presence
Phaeophyta	*Sargassum* sp.(brown macro-algae)	Percentage Cover
Porifera	Black Sponge	Count
Porifera	Grey Sponge (non-erect)	Count
Porifera	Orange coloured encrusting sponge	Count
Porifera	Upright grey sponge	Count
Porifera	White Sponge	Count
Porifera	White sponge with lots of pores	Count
Porifera	Yellow Encrusting Sponge	Count
Rhodophyta	*Botryocladia* sp. Drift	Presence
Rhodophyta	*Botryocladia obovata* (red macro-algae)	Count
Rhodophyta	Red Algae type A	Percentage Cover
Worm Hole	Worm Hole	Presence
Worm Tube	Worm Tube	Presence

List of infaunal survey taxa

List of taxa found in the infauna samples from Boston Bay from both the October 1994 and February 1995 collections

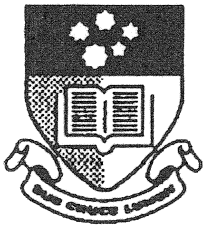
Standardised classification used for data analysis	Common names for the organisms	Record of abundance
Amphipoda	Amphipod	Count
Anthozoa	Anemone	Count
Annelida	Annelid	Count
Ascidiacea	Ascidian	Count
Bivalvia	Bivalve	Count
Ophiuroidea	Brittle Star	Count
Bryozoa	Bryozoa	Count taxa *
Polyplacophora	Chiton	Count
Brachyura	Crab	Count
Cumacea	Cumacea	Count
Echiura	Echiura	Count
Eggs	Eggs - unidentified	Count
Osteichthyes	Fish - unidentified	Count
Osteichthyes	Fishlarv - unidentified	Count
Turbellaria	Flatworm	Count
Paguridae	Hermit Crab	Count
Hydrozoa	Hydroid	Count taxa *
Isopoda	Isopod	Count
Mysidacea	Mysid Shrimp	Count
Nematoda	Nematode	Count
Opistobranchia	Nudibranch	Count
Ostracoda	Ostracod	Count
Polycheata	Polycheat Worm	Count
Holothuroidea	Sea Cucumber	Count
Asteroidea	Sea Star	Count
Echinoidea	Sea Urchin	Count
Decapoda	Shrimps	Count
Sipuncula	Sipunculins	Count
Astacillidae	Skeleton Louse	Count
Caprellidae	Skeleton Shrimp	Count
Neballidae	Slender Legged Sea Flea	Count
Gastropoda	Snail	Count
Porifera	Sponge	Count
Cephalopoda	Squid	Count
Tanaidacea	Tanaidacea	Count

* For colonial organisms such as bryozoans and hydroids the sample is usually highly fragmented - making a count of individuals impossible. Data for these organisms is collected in terms of the number of different taxa in a sample.

INVESTIGATING THE ENVIRONMENTAL EFFECTS OF SEA-CAGE TUNA FARMING.

II. THE EFFECT OF SEA-CAGES.

Anthony Cheshire[1], Grant Westphalen[1], Alastair Smart[2] and Steven Clarke[2].



Project 94/091

INVESTIGATING THE ENVIRONMENTAL EFFECTS OF SEA-CAGE TUNA FARMING.

II. THE EFFECT OF SEA-CAGES.

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Table of contents

1. Executive overview - Non-technical summary

1.1. Background

In this paper we present the results from a comprehensive series of studies designed to assess the extent of souring of the seafloor associated with sea-cage farming of southern bluefin tuna (*Thunnus maccoyii*) in Boston Bay (Port Lincoln, South Australia) during 1994 and 1995.

The environmental effects of sea-cage farming have been documented for a variety of other finfish species (mostly salmonoids, eg Gowen and Bradbury, 1987; Frid and Mercer, 1989; Lumb, 1989; Hall *et al.*, 1990; Tsutsumi, 1995) but little is known about the effects of farming southern bluefin tuna. Sea-cage farming requires much higher stocking densities than those found in natural populations and results in a constant rain of organic and non-organic waste onto the surrounding benthos. Uneaten food, ammonia and faeces are the major components of this waste (Wildish *et al.*, 1990) the accumulation of which may be exacerbated by inefficient assimilation by the stock and the need for sheltered locations, which limits dispersal (Braatan *et al.*, 1983).

1.2. Scope

The aims of our research were to apply the methodologies developed through the first part of this research program (Cheshire *et al.*, 1996) in order to assess the extent of the impact on the benthic environments of Boston Bay. In summary our objectives were:

1) To briefly summarise the literature with respect to what is known about sea-cage farming, the effects this has on the local environment, and the sorts of environmental monitoring that have been conducted.

2) To assess the environmental impact of tuna sea-cages on the epibenthic community, infaunal community and sediment chemistry.

3) To formulate proposals for the management of future environmental monitoring programs and to suggest areas for future research.

1.3. Approach

Surveys of epibenthic and infaunal communities were conducted by collecting samples along transects. These transects ran 200 m (diver epibenthic and infaunal) and 150 m (video epibenthic) from the edge of the cages. Abundances of various taxa were recorded at intervals along these transects and compared with abundances recorded from control transects which were run at distances greater than 1 km from the cages. The choice of transect length and the intensity of sampling along transects was based upon the assumption that impacts would decline exponentially away from the cages. Preliminary information from prior research (Bond, 1993) and our own observations supported this proposal in that they suggested that most changes in community structure took place within the first 20 m (infaunal communities) and 150 m (epibenthic communities) from the cage.

1.4. Summary of results

In general the epibenthic communities were impacted up to 150 m from the cages. Surveys at 200 m indicated that epibenthic communities were not different to those on the control transect. Effects on

infaunal communities were significant within 20 m of the cage but beyond this communities were not significantly different to those on the control transects.

In summary, four zones similar to those described by Brown *et al.* (1987) for salmonid farms, were associated with the tuna cages investigated in Boston Bay.

1) An area of high impact which extends from directly below the cages to a distance of roughly 5 m from the cage margin. The dominant taxa in this region include elevated numbers of polychaetes, nebalids, brachyurans and anthozoans with intermediate numbers of ascidians, holothurians and sea urchins.

2) A zone from 5 m to 20 m from the cage which is characterised by moderate levels of organic detritus. Dominant taxa in this zone include elevated numbers of ascidians and holothurians but there is a reduction in both polychaetes and sea urchins relative to the inner zone.

3) A zone extends from 20 m to around 120 - 150 m from the cage. Although there is little evidence of a build up of organic detritus in this area there is an increased abundance of epibenthic filter and deposit feeding organisms which rely upon organic inputs for their nutrition (eg. ascidians and holothurians). The infaunal communities in this region show no significant differences from those on the control transects.

4) The final zone comprises the area beyond 150 m from the cage including the areas in which control transects were run.

1.5. Recommendations

1.5.1. More detailed study of cage rotation strategies on seafloor souring

Further studies should be undertaken to assess the effect of differences in farm management on the processes of seafloor souring and recovery.

1.5.2. Refinement of video survey techniques

Methods for using video surveys should be refined and developed to enable their use as a routine monitoring tool.

1.5.3. Detailed investigations of the use of harrowing and bacterial applications

Further studies should be undertaken to investigate the use of bacterial applications, harrowing and other techniques on maintaining the health or accelerating the recovery of sediments under sea-cages.

1.5.4. Taxonomic resolution of infaunal data sets

The relative benefit of using an increased taxonomic resolution vs. decreased sampling intensity in infaunal studies should be undertaken in order to evaluate alternative sampling strategies for this system.

1.5.5. More detailed study of summer vs winter using appropriate replication

Further studies should be undertaken to assess the effect of changes in the physical environment between summer and winter on the processes of seafloor souring and recovery.

1.5.6. Use of BACI designs

Future studies should use a Before and After Control and Impact design to clearly document the impact associated with aquaculture development.

1.5.7. Use of bay-level controls

Future studies should address the issue of controls for bay-level responses.

2. Background

Farming of the southern bluefin tuna (*Thunnus maccoyii*) is a new industry that only occurs in coastal waters adjacent to Pt Lincoln, South Australia. Farming methods used in the industry have been adapted from those used elsewhere in Australia and around the world, mostly for the farming of a variety of salmonid species (eg. Hall *et al.*, 1990). This technique uses large open water sea-cages into which wild caught fish are on grown to commercial size. These cages consist of two concentric rings of netting slung from a circular pontoon 30 - 50 m in diameter. The inner net is closed at the bottom and entraps the stock while the outer net drapes to the seafloor and acts as a barrier to predators. Cages float in 15-18 m deep water around Boston Island (Fig. 1).

2.1. Need

The environmental effects of this form of aquaculture have been documented for a number of other species (mostly salmonids, eg Gowen and Bradbury, 1987; Frid and Mercer, 1989; Lumb, 1989; Hall *et al.*, 1990; Tsutsumi, 1995). Sea-cage farming results in a constant rain of organic and non-organic waste onto the surrounding benthos. Uneaten food, ammonia and faeces are the major components of this waste (Wildish *et al.*, 1990) the accumulation of which is exacerbated by low water movement that characterises these sheltered locations (Braatan *et al.*, 1983). Gowen and Bradbury (1987) estimated a deposition rate up to 10 $kg.m^{-2}.year^{-1}$ immediately under a salmon cage and up to 3 $kg.m^{-2}.year^{-1}$ within the local area.

There is growing concern over the effect of sea-cage waste both on the stock in the cage and the local environment (Druff, 1987; Hammond, 1987; Frid and Mercer, 1989; Hall *et al.*, 1990). These relate to changes in the sediment chemistry, water chemistry, and the local biota. To avoid possible threats to the stock, farm managers are inclined to regularly move their cages between sites (Frid and Mercer, 1989).

Enrichment of the seafloor beneath cages results in changes in water chemistry including reductions in the dissolved oxygen content of the water body and the presence of H_2S (Brown *et al.*, 1987; Lumb, 1989; Wildish *et al.*, 1990) and methane (CH_4). These changes result from alteration of the sediment chemistry (Brown *et al.*, 1987) and the formation of anoxic sediments (Pearson and Rosenburg, 1978). Brown *et al.* (1987) found highly reducing conditions up to 3 m from the edge of a salmon cage and suggested that H_2S was being produced continuously with the possibility of some CH_4 in a few months of the year. They also found that oxygen levels in bottom water under the cage were reduced relative to ambient levels. Changes in the chemistry are a particular threat to the stock as water quality is a key issue in the maintenance of fish health (Poxton, 1991).

The benthic flora and fauna in the immediate vicinity of sea cages is generally altered quite significantly in terms of community composition, species richness, diversity, abundance of taxa and biomass (Brown *et al.*, 1987; Gowen *et al.*, 1988). Brown *et al.* (1987) describe four zones of effect on the benthic community around a salmon cage in a Scottish sea loch. A zone completely devoid of life up to 3 m from the cage, a second area with low species diversity dominated by opportunistic polychaetes and a third zone from 15 m, that was highly diverse and contained not only taxa common to the zones on either side but also taxa specific to that area. The fourth and last zone, 150 m away from the cage, was indistinguishable from the "normal" benthic environment.

Brown *et al.* (1987) concluded that the effects of salmon farming can be as severe as other forms of organic pollution but the extent of degradation is generally confined to a small area directly underneath and adjacent to the cages. Frid and Mercer (1989) recommended the sitting of sea-cages

in areas of high tidal flow as this would disperse the sediment rain over a broader area and reduce the more localised environmental impact. They note, however, that nutrient enrichment of the water body for a longer period could stimulate the growth of phytoplankton.

An alternative approach advocated by some resource managers and used for the farming of tuna in South Australia, is to accept that the accumulation of wastes will exceed the natural assimilative capacity of the seafloor community. In response farmers are issued with a larger lease area so as to allow the practice of cage rotation and seafloor fallowing (Bond 1993). In South Australia, farms are 20 ha in size, which is around three times the area required for the number of cages farmed at any time. Therefore, if cages are moved every 2 years this will allow 4 years of fallowing before an area is reused. Other research in Tasmania and elsewhere has shown that this provides sufficient time for the physical and chemical properties of the seafloor to return to a state such that the site can be re-used. Such data are lacking for Boston Bay.

Environmental monitoring strategies to measure the impact of sea-cage aquaculture have been suggested (eg. Frid and Mercer, 1986; Wildish *et al.*, 1990). These vary in the types of data required and the complexity of design. Frid and Mercer (1989) proposed that apart from monitoring of the benthos, as in other studies (Brown *et al.*, 1987; Gowen *et al.*, 1988), that water chemistry, sediment chemistry (in particular redox potentials) as well as biological monitoring should be undertaken. A very detailed monitoring program devised by Wildish *et al.* (1990) proposed a broadly based sampling program dealing with water quality issues as well as a benthic sampling component. This included regular measurements of temperature, salinity, dissolved oxygen, ammonia, nitrogen, phosphates, microbial biomass, chlorophyll a, adenosine triphosphate, phytoplankton, benthic ecology and sediment redox potential.

Such a program, while comprehensive, is costly and difficult to coordinate. Simpler, more defined, survey methods may give much the same answers without high cost and logistic demands.

Bond (1993) investigated a number of features of tuna cage systems, including sedimentation rates, water chemistry and nutrient dispersion, and the epibenthic flora and fauna. This information, in conjunction with research from other aquaculture studies formed the basis for the "Port Lincoln Aquaculture Management Plan 1993". That plan places a strong emphasis on the need for environmental monitoring and more detailed research upon this system.

Our research has considered the benthic ecology in the vicinity of tuna cages, dissolved oxygen and the redox potential of sediment.

2.2. Objectives

The aims of our research were to apply the methodologies developed through the first part of this research program (Cheshire *et al.*, 1996) in order to assess the extent of the impact on the benthic environments of Boston Bay. In summary our objectives were:

1) To briefly summarise the literature with respect to what is known about sea-cage farming, the effects this has on the local environment, and the sorts of environmental monitoring that have been conducted.

2) To assess the environmental impact of tuna sea-cages on the epibenthic community, infaunal community and sediment chemistry.

3) To formulate proposals for the management of future environmental monitoring programs and to suggest areas for future research.

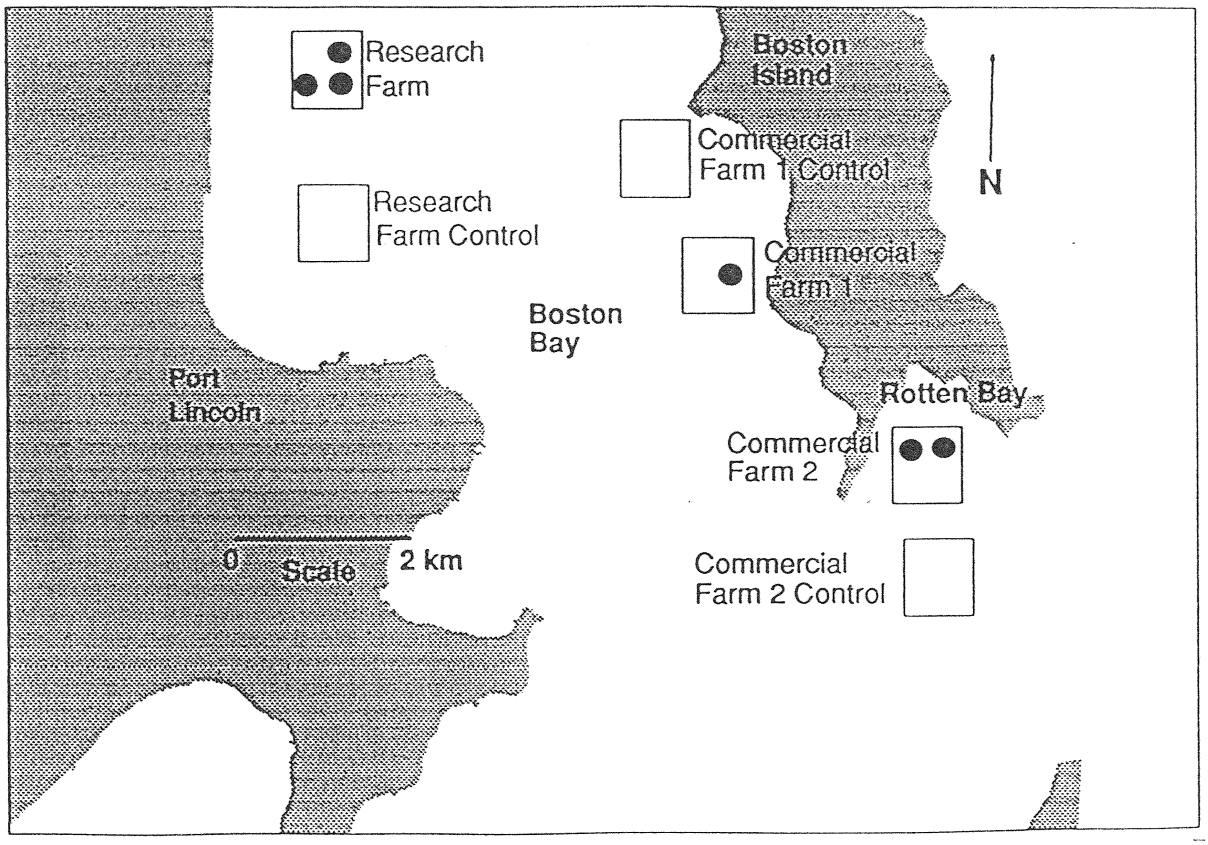
3. Methods

3.1. Description of the study area

The study area was contained within Boston Bay, Pt Lincoln (Fig. 1). Although this bay has two entrances water movement is limited with much of the bay experiencing current speeds which are typically less than 2 $cm.s^{-1}$. The bay experiences moderate sea conditions under some circumstances although the biota of the harbour in locations suitable for sea-cages is probably influenced more by tidal flow than wave action (Petrusivics; in Bond 1993).

A number of anthropogenic influences other than the effects of tuna sea-cages are experienced within the bay. The Billy Lights Point Engineering and Water Supply sewage treatment plant inputs primary treated (as of 1994) effluent from the whole of the Pt Lincoln township into the bay. A few small creeks run into the bay and a number of small industries including an oil terminal, fish factories and an abattoir also discharges waste. There is some heavy shipping (for which there has been associated dredging) for the loading of grain and fertiliser from which a lot of dust is produced. Prawn fishermen have been known to regularly test their trawling equipment in the harbour.

Figure 1. Map of Boston Bay, Pt Lincoln showing the cages and area where survey work was undertaken.



Six locations were selected for the survey work, three of these were associated with stocked cages and three were control sites. Sites were widely dispersed throughout the bay so that the variability within the study area could be considered (Fig. 1). The sites chosen were;

Experimental Farm (**ExperFarm**) situated on the north west side of the harbour in an exposed position. Cages in this farm had been in place for the longest and may have represented an extreme in terms of environmental impact.

Commercial Farm 1 (**ComFarm1**) located on the north east side of the harbour. The exposure of this farm was considered to be intermediate to that of the Experimental and Commercial Farm 2 sites. Cages in this farm were cleaned and moved regularly (in line with current methods).

Commercial Farm 2 (**ComFarm2**) situated in Rotten Bay at the south end of Boston Island. This farm was considered to be in the most wave sheltered location although currents in the southern channel between the sea and bay are higher (<12 cm. s^{-1} for 75% of this time in the channel c/f < 5 cm. s^{-1} offshore; Petrusivics in Bond, 1993). As with the other commercial farm the cages were cleaned and moved regularly, however there was a higher density of cages within this small bay. Two other cages within Rotten Bay, (also labelled Commercial Farm 2 for convenience) were used in the survey.

Control Sites - all control sites were located so that they experienced roughly the same degree of exposure to water movement as their associated cage sites while at the same time being a considerable distance (1-2 km) from any farming activity. This was not always possible, and the Rotten Bay control sites were in locations with higher water movement than the cage transects (Fig. 1).

3.2. Approach

For full details on the nature and resolution of data recorded for the following studies refer to Cheshire *et al.* (1996).

3.2.1. Field work

Field work at Pt Lincoln was carried out in three trips.

August 1994 (winter)	Diver transects and photorespirometer deployments.
October 1994 (winter)	Video transects, remote suction samples and photorespirometer deployments.
February 1995 (summer)	Video transects, remote suction samples and photorespirometer deployments.

The timing for these trips was chosen to enable a comparison of the system in the winter (August-October 1994) and the summer (February 1995). These trips coincide with high stocking densities and feed inputs (winter) and the lowest (summer) when the cages are virtually destocked. It should be recognised however, that because there is no replication of winter and summer seasons general conclusions about summer vs. winter are not valid. Instead, we will refer to differences in the context of "the winter" and "the summer" and where possible indicate where such differences may in fact be attributable to general seasonal effects or stage stocking differences as opposed to simple changes through time. Such changes are likely to be further confounded by the seasonal timing of sampling as well as the varying residency times for the tuna cages at each location. Throughout both the winter and the summer surveys, the same cages were examined and controls were collected from sites distant from the cages. Control transects were always run in the same direction as the cage transects to ensure that the influence of incidental environmental gradients (other than cage effects)

was approximately the same in both cases. Redox sampling was ongoing (mostly in spring 1994) at the Experimental Farm.

3.2.2. Diver surveys

Divers using SCUBA surveyed a series of four 200 m transects in August 1994. Cage transects (two at ExperFarm and one at ComFarm1) were placed so that they began as close as possible to the outside of the predator net. Quadrats were located at distances of 0, 5, 10, 15, 20, 50, 100, 150 and 200 m along the transect line.

3.2.3. Video surveys

A series of six transects were run in October 1994 with a further twelve in February 1995. Transects were run from the edge of the cages for a distance of 150 m. Stops of 30 to 60 seconds were made at 0, 2, 10, 20, 40, 60, 80, 100, 120, 140 and 150 m using 2 m markings on the rope as a measure of distance. Control transects (three controls in 1994, six in 1995) were run in a similar manner.

3.2.4. Infauna surveys

In October 1994 samples were collected from six transects comprising 3 cage transects and 3 controls. On each cage transect samples were collected at distances of 0, 2, 20, 100 and 200 m from the edge of the cages. For control transects samples were collected at distances of 0, 2, 20, 100 and 200 m from the beginning of the transect. In February 1995 a further six transects were run (3 control and 3 cage) but only the first two distances for each transect (0 and 2 m) were sorted due to the excessive time required to sort the samples and the need to complete the study by its due date.

3.2.5. Sediment chemistry

Redox potentials (E_h) were measured *in situ* under a recently destocked tuna cage (ExperFarm). This approach was used rather than the standard coring method where compaction and other physico-chemical changes experienced by the samples (as they are transferred to the surface) can cause changes in the E_h measurement.

Five groups of 1 m^2 quadrats were selected with three measurements taken per quadrat. Following these initial measurements, quadrats in each group were selected for, harrowing, bacterial application or control. Harrowing was achieved with a garden rake (used once) while the bacteria was a commercial gram positive DMS-1000 Series (or "Sludge Doctor®") supplied by Admac Agencies that was applied weekly in 1 kg bags over a four week period. Two redox measurements per quadrat were taken following this treatment.

In addition to the measurement of sediment E_h the release of gasses from the sediment was also assessed. The formation of hydrogen sulphide (H_2S) is quantitatively the most important of these gasses but the possible formation of CH_4 is also important. Upturned funnels on stilts driven into the sediment under the cage were used to collect any gasses that might form.

3.3. Data analysis

3.3.1. Infauna and epibenthic data

All survey data was analysed at a coarse taxonomic level (Phylum, Class, Order or Family). Analysis of this kind has been undertaken before with little or no loss of information (Warwick, 1988a,b; Ferraro and Cole, 1990). Rather, the use of higher order taxa in analyses of environmental impact can give clearer results as the data are not cluttered with "nuisance" information (Warwick, 1988a). Such an approach requires a less exhaustive understanding of the taxonomy of the system which is often poorly documented (Agard *et al.*, 1993).

Diver collected epibenthic data, video epibenthic data and infauna data were all considered in the same manner. Taxa with few individuals for the combined surveys were excluded from the univariate analyses. Data from samples collected during the winter were analysed using a two-way Analysis of Variance (ANOVA) (location and distance). Where the ANOVA identified differences between samples these were assessed *post-hoc* using Tukey's HSD test to indicate where significant differences occurred.

Comparisons between winter and summer were made using a three-way ANOVA (location, distance and season). For the infauna data only 2 distances (0 and 2 m on both cage and control transects) were considered because sorting of the summer samples was not completed due to logistic constraints. No diver epibenthic data were collected during the summer sampling period.

Data were also analysed using the multivariate ordination technique *Semi-Strong Hybrid Non-Metric Multi-Dimensional Scaling* (SSH). This approach enables ordination plots (in 2 or 3 dimensions), to be created from a multi-dimensional (in the context of this study - multi-taxa) data set. An ordination into 2 or 3 dimensions enables an easier visualisation of the relationship between samples but the faithfulness of the ordination, in terms of its capacity to truly represent relationships between samples, is a function of the associated stress value. Stress is a measure of the extent to which the ordination of inter-sample distances, in the reduced dimensional space, reflects the multi-variate association value for the samples. A high stress value indicates that little faith can be placed in interpretations of relationships between samples. In this report only ordinations with a stress less than 0.2 have been reported. Such ordinations will provide a reasonable interpretation of the relationships between samples in the data set.

3.3.2. Sediment chemistry

A one-way Analysis of Variance (ANOVA) was performed to examine differences in redox values between treatments for the change between the mean final and mean initial E_h readings. In addition, a split plot ANOVA was performed to examine the relationship between treatments and time. No gasses were detected and therefore no analysis of these data was possible.

4. Results and discussion

4.1. Benthic communities

4.1.1. Patterns in the winter (1994)

During the winter four zones, similar to those described by Brown *et al.* (1987), were identified associated with the tuna cages in Boston Bay.

1) An area of high impact extended from directly below the cages to a distance of roughly 5 m from the cage margin. This zone was characterised by high levels of organic detritus which had settled beneath the cages and drifted small distances outwards. The dominant biota included intermediate numbers of ascidians and holothurians (Fig. 2) and elevated numbers of sea urchins (Fig. 2) in the macro benthos. Infaunal communities in this zone were characterised by elevated numbers of anthozoans, brachyurans (crabs), gastropods, nebalids and polychaetes (Fig. 3). Further, there was a significant reduction in the number of shrimps in this region relative to more distant locations or the control transects. Brown and red macro-algae (Phaeophyta and Rhodophyta) are also reduced in this zone (Fig. 2; Fig. 4).

2) A second zone extended from 5 to 20 m from the cage and was characterised by intermediate levels of organic detritus. The biota in this region typically comprised moderate numbers of ascidians and holothurians (Fig. 2) but with a reduced number of sea urchins relative to the inner zone (Fig. 2). Infaunal taxa that were in large numbers close to the cage (see above) were all much less abundant (anthozoans, brachyurans, nebalids and polychaetes were significantly lower; Appendix 1; Fig. 3) whilst the numbers of decapods (shrimps) increased significantly (Appendix 1; Fig. 3).

3) The third zone extended from 20 to 150 m from the cage. This zone was largely indistinguishable from the 200 m region on the cage transects or the control areas which were at least 1 km distant from any cages. In this area there was little superficial evidence of organic detritus associated with the sea-cages. There was however, clear evidence that this region was affected by increased organic loadings; the number of ascidians and holothurians in the epibenthos reached maximal levels between 50 and 150 m from the cage (Fig. 2). At greater distances the number of these organisms dropped off significantly (Appendix 1). Sea urchins were virtually absent from this region (Fig. 2) and most infaunal taxa tended toward background levels (eg amphipods and polychaetes; Fig. 3).

4) The final zone comprised the area from the 200 m mark on the cage transects and the entirety of the control transects.

Figure 2A-E. Diver epibenthic abundance data plotted against distance. Distances labelled 1000 m include all locations on control transects.

Figure 2A. Ascidians

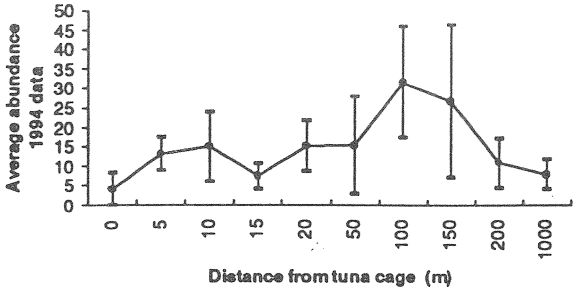


Figure 2D. Holothurians

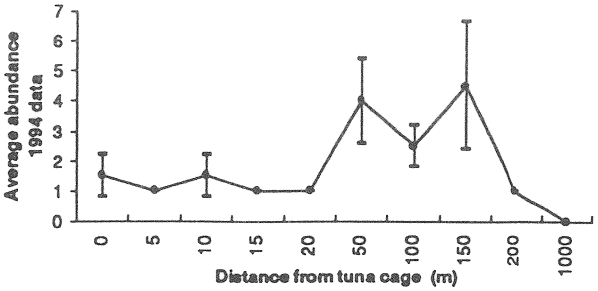


Figure 2B. *Botryocladia* sp,

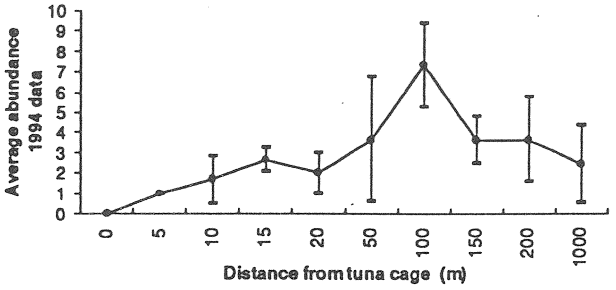


Figure 2E. Sea Urchins

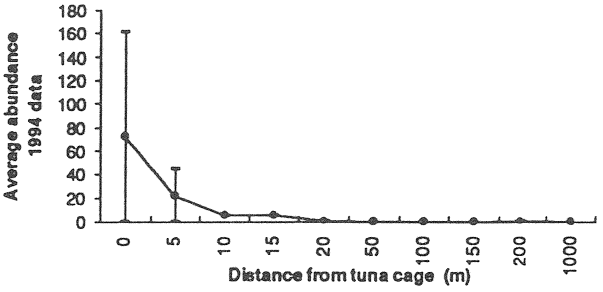


Figure 2C. Chlorophyta (Green algae)

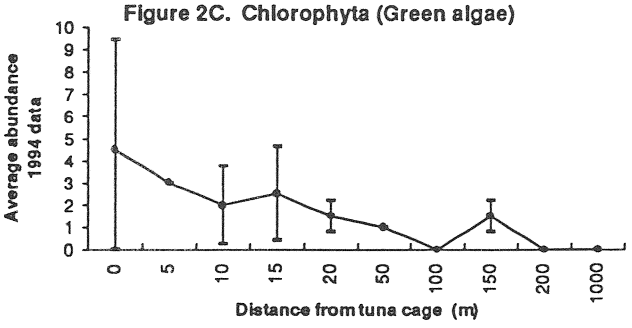


Figure 3A-Q. Infauna abundance data plotted against distance. Distances labelled >=1000 m represent samples collected from control transects.

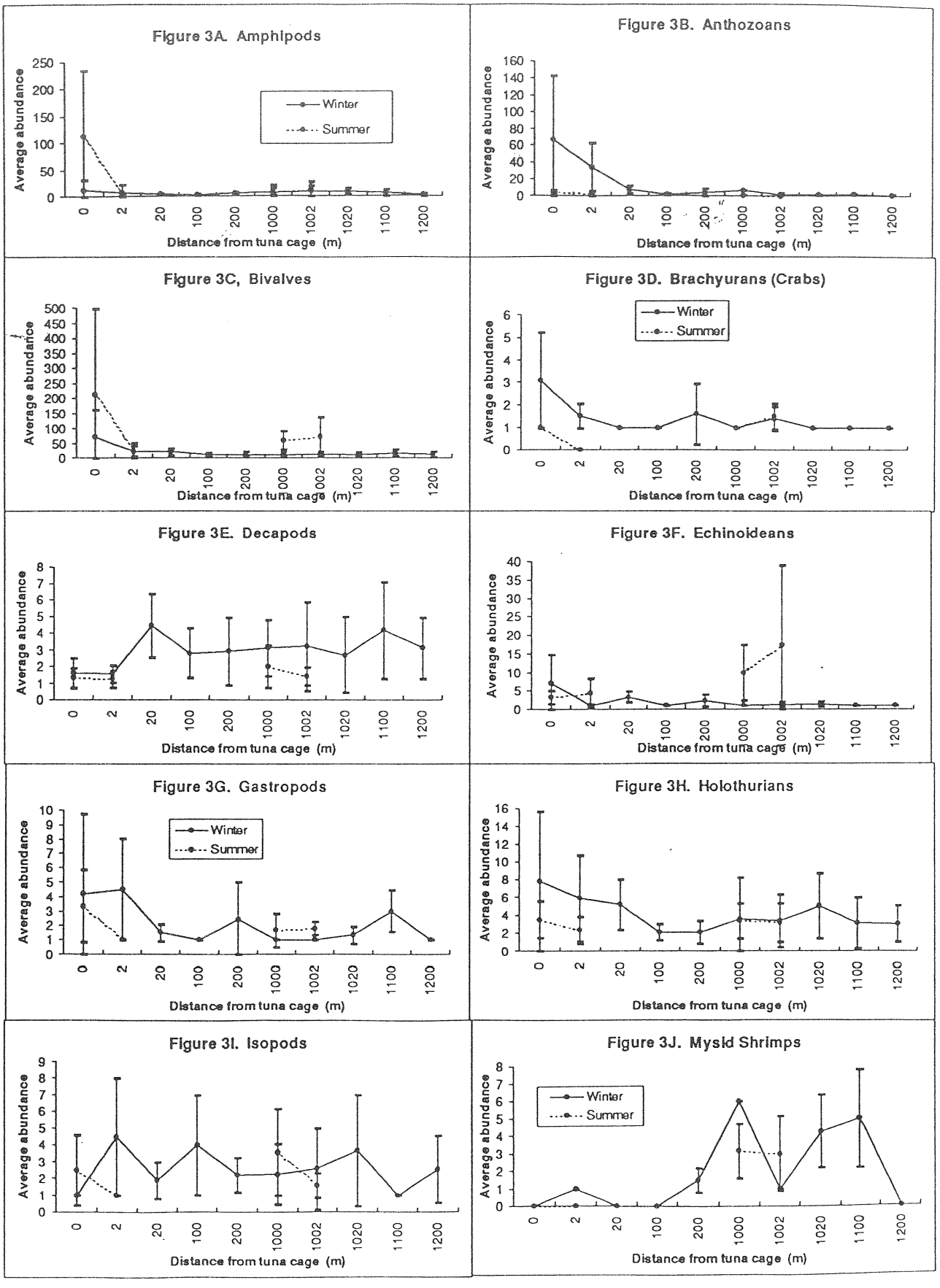


Figure 3K. Nebalids

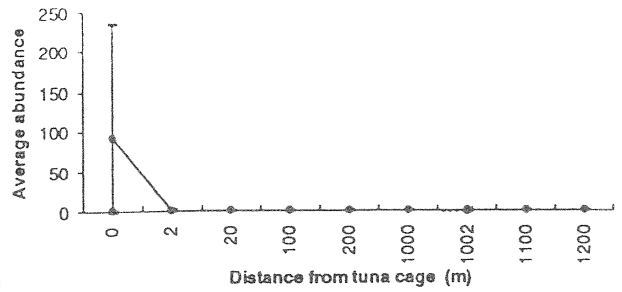


Figure 3L. Ophiuroideans

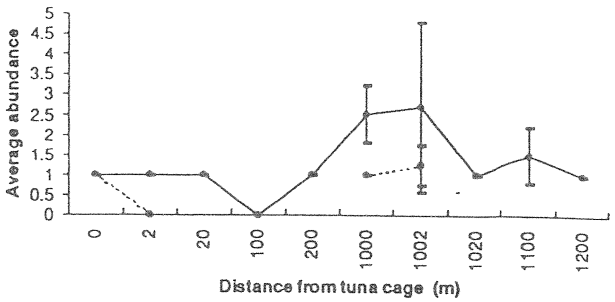


Figure 3M. Opistobranchia

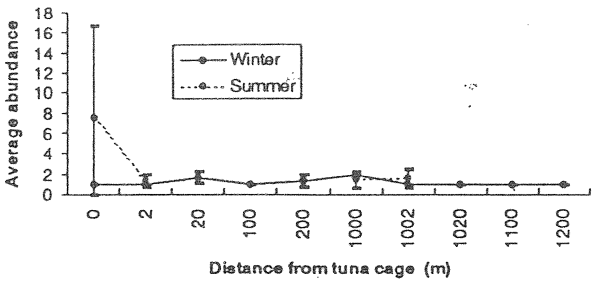


Figure 3N. Ostracods

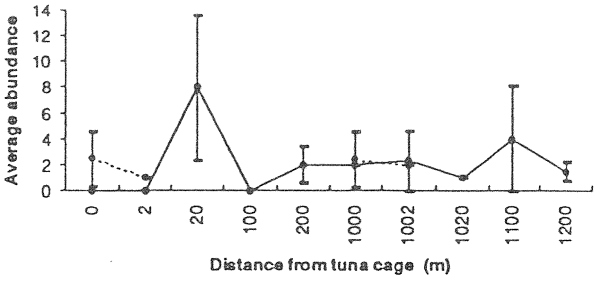


Figure 3O. Polychaetes

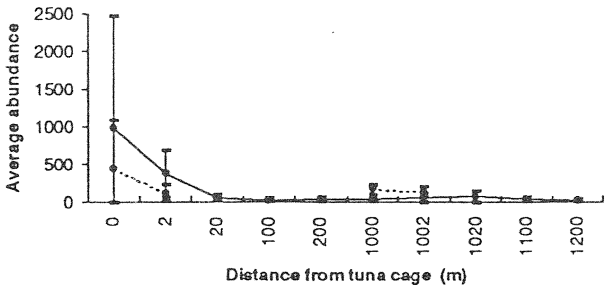


Figure 3P. Sipunculins

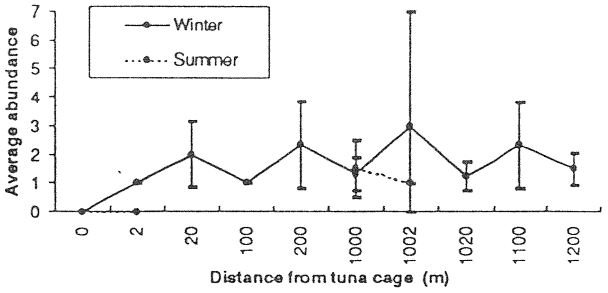
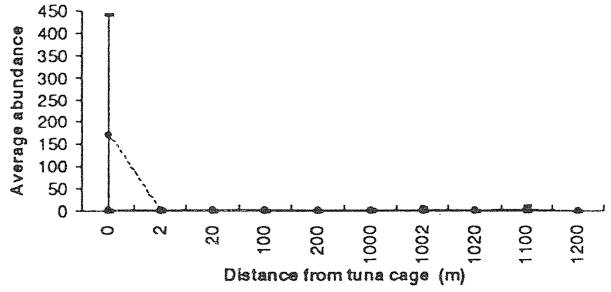


Figure 3Q. Tanaidaceans



In general terms the biota in each of these four zones was consistent with what would be expected based on the level of organic loading experienced at the seabed. Organisms in the inner zone were smothered by organic detritus typically comprising large particles such as uneaten fish, pilchards or synthetic feeds. In addition, there was a steady rain of fouling material which was dislodged from the nets forming the cage. This fouling material included numerous bivalves, soft bodied animals and a variety of algal fronds particularly the green alga *Ulva* (Fig. 2). Organisms inhabiting this zone would need to avoid smothering, be tolerant of low light conditions (shading from the nets) and be able to benefit from the high organic loading. Such conditions would suit taxa such as holothurians, sea urchins, brachyurans (crabs) and polychaetes. It is believed that the elevated numbers of bivalves and green algae found in this region originated from the fouling community and that they did not colonise this zone but rather were dislodged and accumulated on the seabed where they presumably died and contributed to the overall organic loading in this zone.

The second zone also experienced high levels of organic detritus. However, in this region the organic material comprised small particles typically faeces or particulate remains of non-ingested food. Sedimentation rates in this region were high and there was a build up of material which was still sufficient to smother non-mobile organisms which could not prevent themselves from being buried. As with the first zone there were elevated numbers of holothurians, sea urchins, and polychaetes in this zone. The numbers of sea urchins and polychaetes was, however, significantly reduced relative to the inner zone.

The third zone showed little evidence of accumulated organic material. The biota in this region did, however, indicate that elevated organic inputs were experienced. The rise in the number of ascidians was indicative of this elevated level of particulate organic material (Fig. 2 and 4). This zone was characteristically different from areas more distant from the cages. Numbers of ascidians reached an intermediate maxima in this region which suggested that sedimentation rates were too high close to the cage to support large populations whereas the availability of organic particles for food was reduced at more distant locations. Similar arguments may be proposed to explain the changes in the abundance of holothurians which also reached an intermediate maxima in this zone.

Locations more distant from the cages (greater than 150 m) were typical of soft bottomed communities in this region. There was a wide diversity of taxa, although few taxa were found in any great abundance (relative to abundances found close to the cages). Significantly, holothurians and sea urchins were either absent or had very low abundances. Only one taxa, mysid shrimps, was found almost exclusively in this region while they were not found closer to the cages (Fig. 3).

Overall, these zones were more or less distinct depending upon which survey method was used to sample the system. Diver epibenthic surveys provided data which clearly differentiated these zones. Samples collected between 0 and 5 m from the cage were seen to be quite different from those collected in the 10 to 20 m zone (Fig. 5). Whereas there were differences between the commercial farm and the experimental farm, there was still clear evidence for the existence of at least three zones. The Video surveys provided less clear results (Fig. 6). Overall, there was evidence to suggest that the 0 to 2 m zone differed from the remaining area of the cage transect. This was not, however, as clear cut as the results from the diver surveys. In general, the reduction in the number of taxa which could be identified using the Video technique meant that this technique was less able to distinguish changes in community structure. Infaunal surveys clearly differentiated the 0 to 2 m zone (Fig. 7). There was however, no evidence from infaunal surveys for the division of the remaining areas into separate zones even though there was some evidence of the impact gradient in both epibenthic surveys (Fig. 5 and 6).

Figure 4. Video epibenthic abundance data plotted against distance from cage margin.

Figure 4A. Ascidians

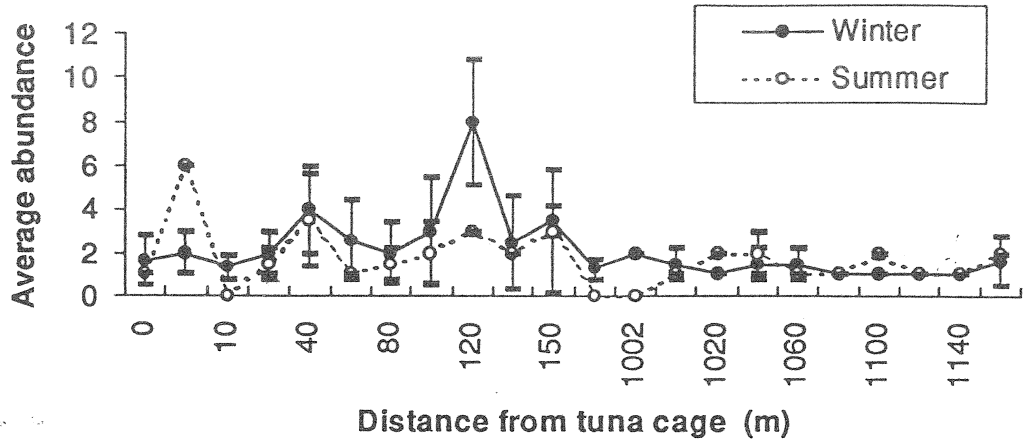


Figure 4B. Rhodophyta (Red algae)

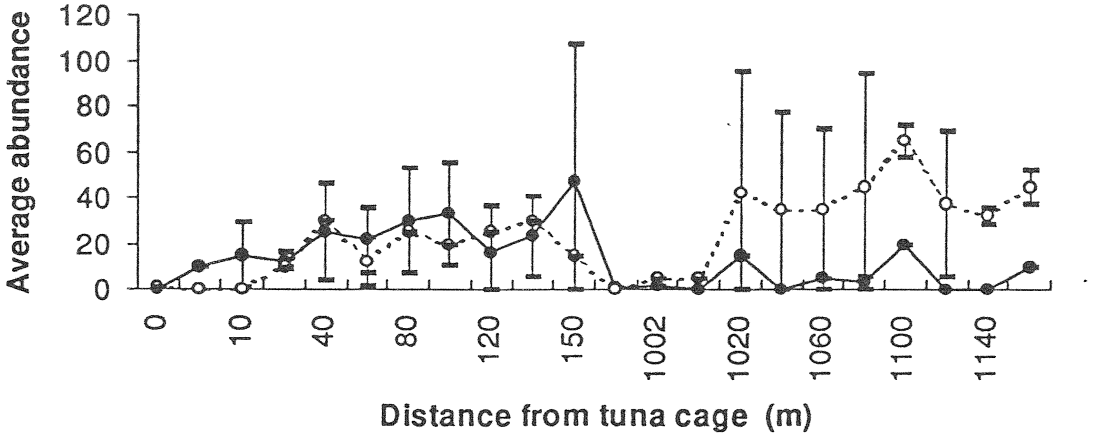
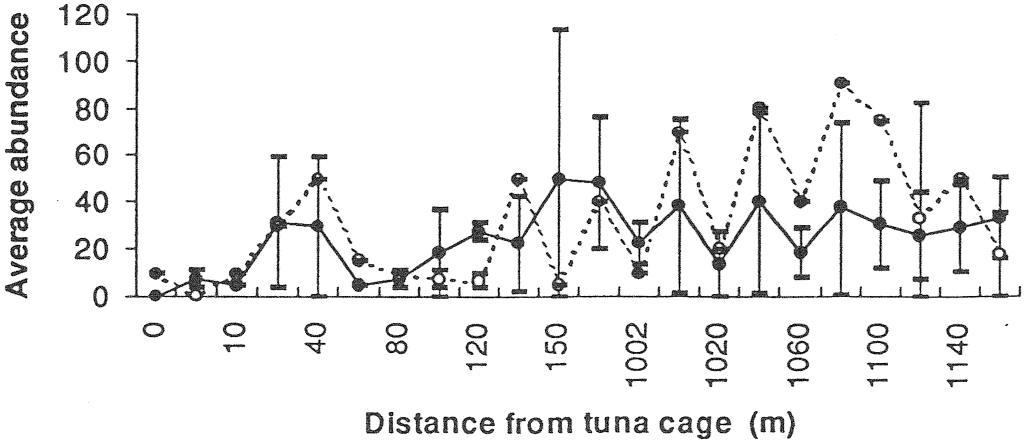


Figure 4C. Phaeophyta (Brown algae)



Pearson and Rosenburg (1978) described the differences in distribution of deposit feeders, suspension feeders and carnivores in response to inputs of detritus. They suggested that suspension feeders were often disadvantaged especially in areas with high sedimentation rates because organic matter may clog the cilia and siphons of filtering mechanisms. Deposit feeders on the other hand, are favoured by detrital input, and are able to create and maintain a level of instability on the surface of fine sediments which can exclude the settlement and development of suspension feeders. Extrapolation of their results to this study would support our finding that suspension feeders such as ascidians tend to be found in maximal numbers at intermediate distances from the cages. Polychaetes, which may frequently be deposit feeders, would tend to be found in larger numbers close to the cages. Caution is required in taking this interpretation to far; there are likely to be a wide range of feeding methods within each of the taxonomic groups that we identified. Polychaetes, for example, exhibit a wide range of feeding strategies, that are closely correlated with the lifestyles of each group. In such a diverse class of organisms it is difficult to make generalisations about feeding modes.

Figure 5. Diver epibenthic abundance data. MDS ordination in two dimensions (Stress=0.2166). A gradient of sites can be observed with samples close to cages (0-5 m) forming a group in the upper right corner of the plot. Intermediate distances (5-20 m and 20-150 m) and controls (>= 200 m) occur along a roughly diagonal gradient from the upper right to lower left.

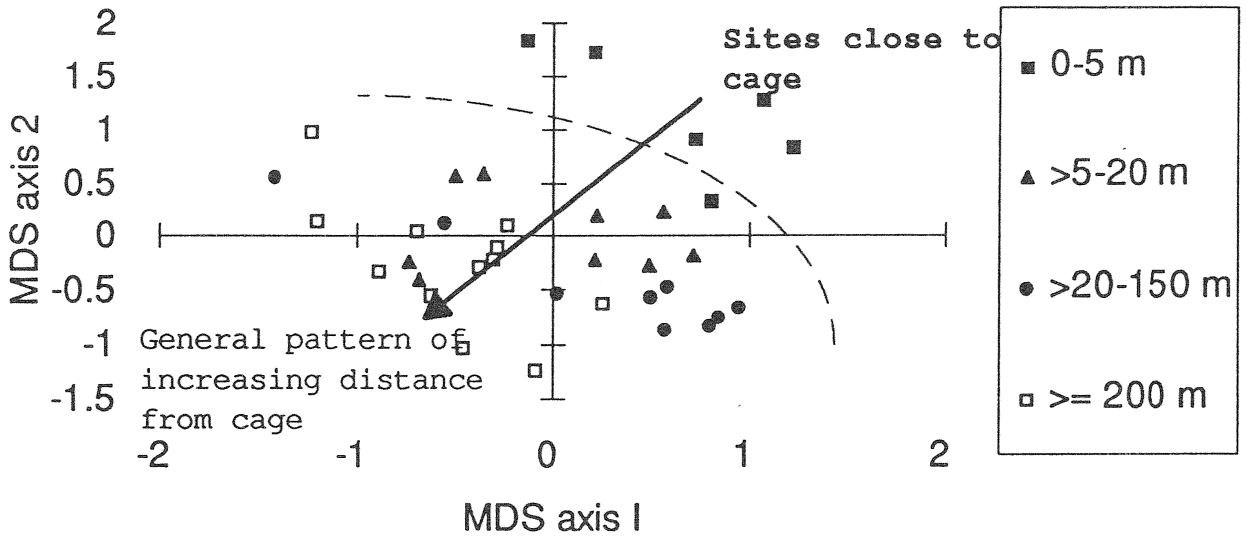


Figure 6. Video epibenthic abundance data. MDS in two dimensions (Stress=0.1497). A gradient of sites is also apparent in this ordination. With sites close to cages (0-5 m) forming a broad band across the lower half of the plot. Other distances form a similarly broad but more homogeneous group in the upper half.

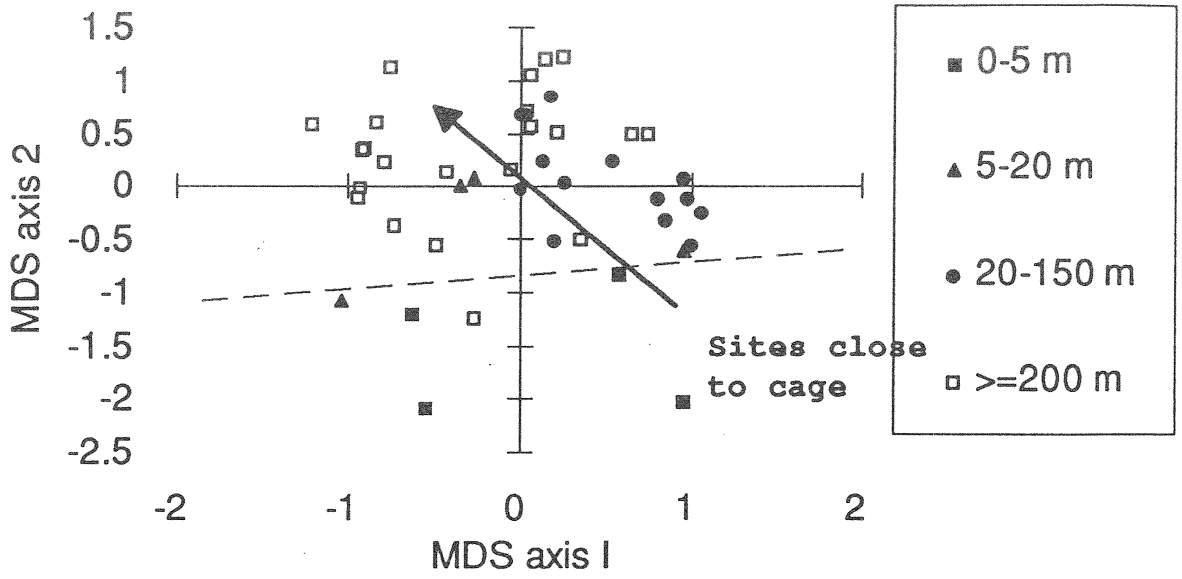
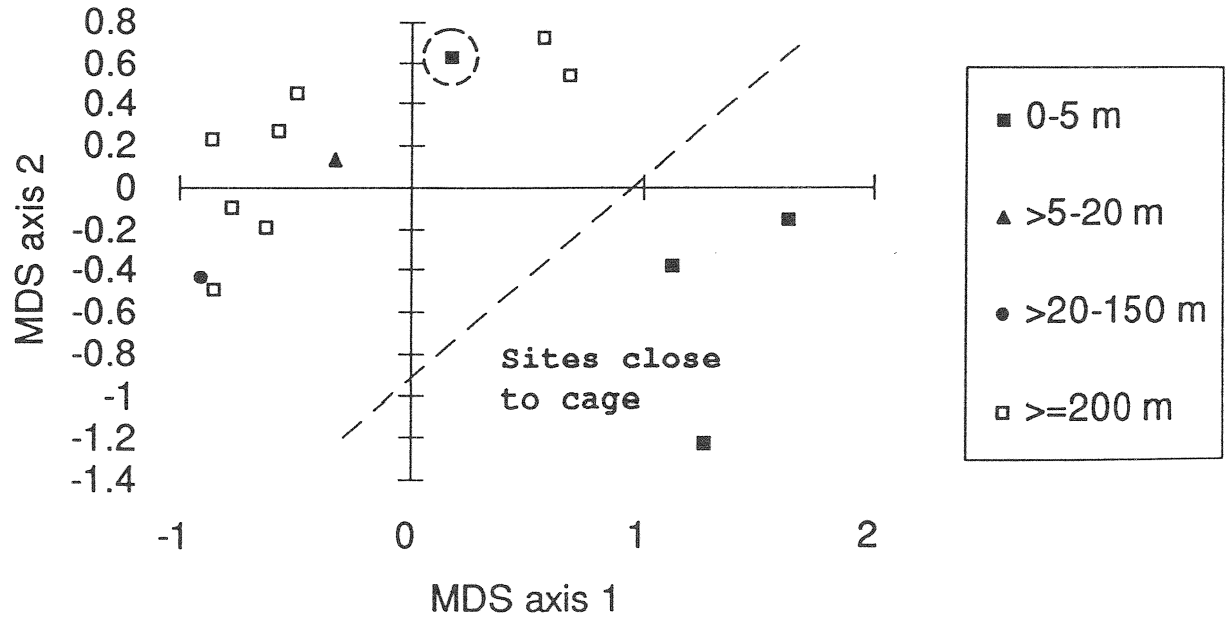


Figure 7. Infauna abundance data. MDS ordination of average abundance by distance and site in two dimensions (Stress=0.095). Samples close to cages are very different (note circled outlier) from those at distances greater than 5 m. Beyond 5m there is a higher level of variability.



4.1.2. Summer winter comparisons

There are a number of significant differences in the abundance of infaunal taxa during the winter and the summer. Twelve of the fourteen infaunal taxa show a significant change between the winter and the summer sampling periods (Appendix 2).

It is important to note however, that the responses are not simple. There are generally significant interaction terms which indicate that changes are not consistent either across locations (cages) or in terms of distance from the cages. Polychaetes, for example (Fig 8) illustrate a different trend for each of the 3 farms considered. On ComFarm1 there is no consistent pattern in the distribution of polychaetes at either sampling period (intermediate numbers both close to and distant from the cage). At ComFarm2 there is a pattern of decreasing numbers with distance in the winter (1994) but the reverse pattern in the summer (1995). Whilst at ExperFarm the pattern is the same (decreasing numbers with distance from the cage) during both sampling periods. Nebalids, on the other hand, show a more consistent pattern (Fig. 8) with an overall reduction in numbers close to the cage from the winter (1994) to the summer (1995).

With the Video data there were no consistent patterns of change between the summer and the winter. In many cases (e.g. Red algae - Table 1) distributions were highly variable both between sampling periods (time of year) and across sites. In the case of brown and red algae there was a predominant cage effect with few algae being found close to the cages (within 20 m). This was not unexpected given the shading effects of the cage and the higher sedimentation rates. At greater distances red algae may or may not have been found but this did not illustrate any consistent relationship to location or time of year (Table 1). Overall, interpretations are complicated with different cages and distances showing different changes through time.

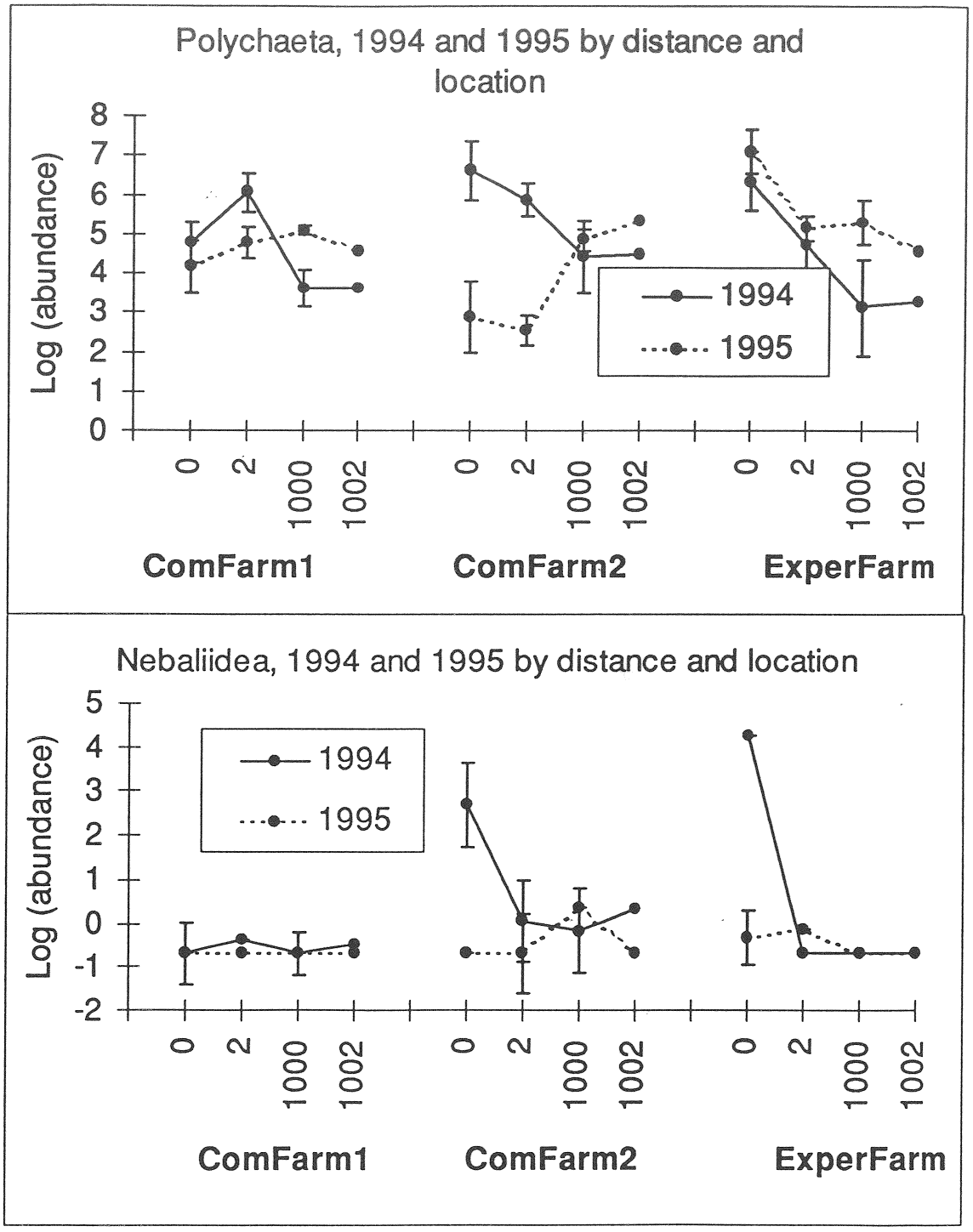
Table 1 Three Way Analysis of Variance of the natural log transformed data for abundance of Rhodophyta found on video transects. Year represents 1994/1995, Cage (ComFarm1, ComFarm2, ExperFarm), Distance (0 - 150 m, 1000 - 1150 m).

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Year	1	1	0.268211	0.5063	0.4794
Location	2	2	0.804633	0.7594	0.4722
Distance	21	21	29.903176	2.6880	0.0013
Year*Location	2	2	0.536422	0.5063	0.6052
Year*Distance	21	21	36.734662	3.3020	0.0001
Location*Distance	42	42	50.088351	2.2512	0.0017
Year*Location*Distance	42	42	84.004266	3.7755	0.0000

Whether any of these differences (in infaunal or epibenthic communities) are in fact attributable to changes between summer and winter or whether they simply reflect changes over a 6 month period cannot be ascertained from these data. In summary, differences over time may reflect seasonal responses to changes in factors such as light, daylength and temperature. These factors will variously interact with a variety of processes including recruitment and growth. Alternatively the differences may simply be associated with the increased period for which the cages have been in any given location. More detailed studies are required that include replication over a number of years

before any conclusions can be drawn about seasonal effects as distinct from those which relate simply to the period during which the cages have been in place.

Figure 8. Changes in the log abundance of polychaetes and nebalids between locations, over distance and between sampling period (winter 1994, summer 1995).



4.1.3. Differences between cages (importance of rotation strategies)

A significant result from this study is the difference in the communities associated with the 3 cages studied. As indicated above the 3 cage locations represent different environments and have been managed differently through time. The Experimental Farm (**ExperFarm**) is situated on the north

west side of the harbour in an exposed position. Cages in this farm had been in place for the longest period of time and generally illustrated the highest level of impact (see for example Fig. 8). Commercial Farm 1 (**ComFarm1**) however, is located on the north east side of the harbour. The exposure of this farm was considered to be intermediate to that of the Experimental and Commercial Farm 2 sites. Cages in this farm were cleaned and moved regularly (in line with current methods) and there were very low levels of impact associated with this cage system. Commercial Farm 2 (**ComFarm2**) was situated in Rotten Bay at the south end of Boston Island. This farm was considered to be in the most wave sheltered location. As with the other commercial farm the cages were cleaned and moved regularly, however there was a higher density of cages within this small bay. This system showed intermediate levels of impact relative to the other two locations.

A greater number of cages are required (enabling replication of management strategies) to definitively conclude that these differences are caused by the local farm management. It is important however, to recognise that the differences in the levels of impact associated with these 3 farms are consistent with what is known about their management history. Importantly, this result illustrates that good management (including regular movement) will result in lower levels of seafloor souring which should have consequential benefits in terms of both environmental management and farm production.

4.2. Photorespirometry

A number of problems were encountered in the course of the photorespirometry study. Firstly the deployment depth (16 - 18 m) caused many of the electrodes to fail. Secondly the stirrer units were prone to becoming clogged with strands of filamentous algae, *Ulva* sp. or other drift. This reduced the effectiveness of the stirrers and the accuracy of the electrodes. Finally the calibration for the electrodes was conducted at the depth of the deepest electrode which meant that, once mounted at the correct depth, most electrodes were slightly off the correct calibration.

In February a system was developed which prevented the stirrers from clogging and allowed for correct calibration at the right depth. Electrode failure continued to be a problem.

Further preliminary work provided the basis for a more detailed study of the sources and sinks for oxygen in tuna cages. This work has since been published separately (see Honours Thesis - E. Cronin, Department of Botany, Adelaide University, 1995).

4.3. Sediment chemistry

4.3.1. Redox

The E_h levels measured *in situ* generally appeared to be very low indicating highly anaerobic conditions (Table 2). Variation between some quadrats was high but variation between repeated E_h measurements within quadrats was low, and only 2 repeated readings per quadrat were taken (Table 2). The E_h readings for core samples were not significantly different to *in situ* values ($P>0.05$). E_h levels became more positive for all treatments over the 4 week period ending on November 9 1994 with the largest increases seen in the bacteria treatments, then harrowing and control (Fig. 9).

Figure 9. Changes in mean redox (E_h) potentials for the different groups of quadrats over the 1 month period. Bars represent the change whilst error bars represent the standard error for differences in replicate quadrats.

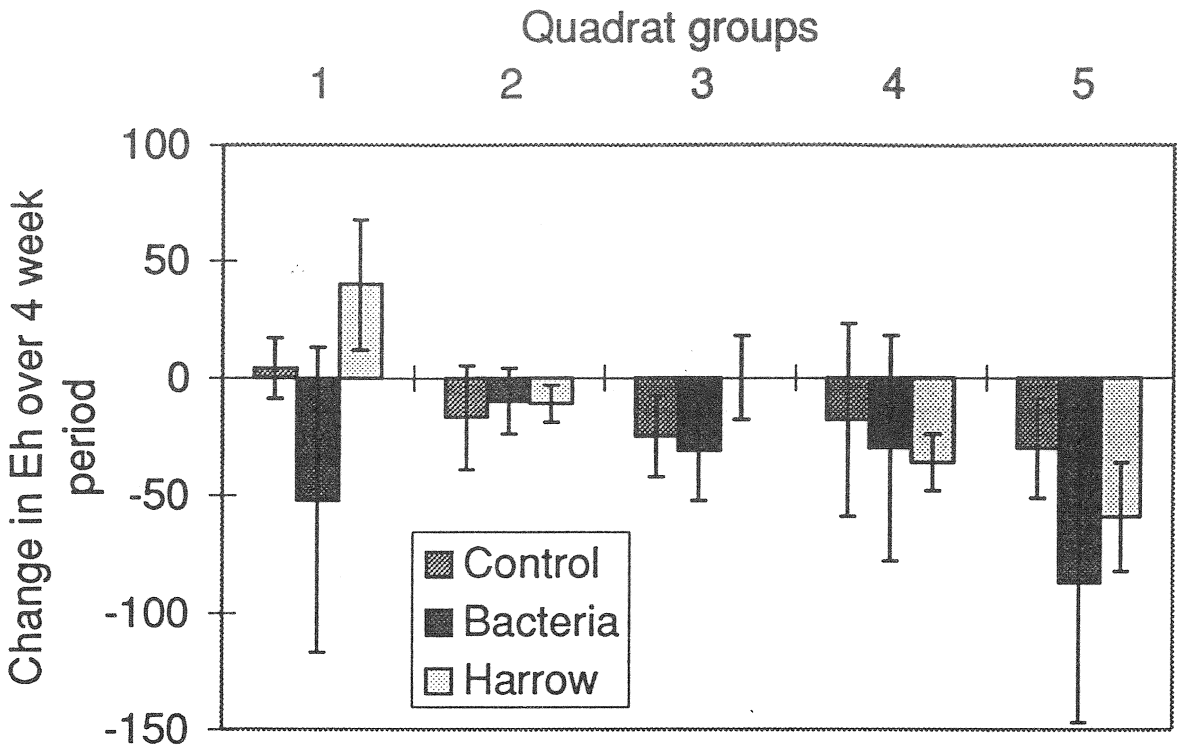


Table 2 Redox (E_h) levels for the 3 treatments of control, bacterial application, and harrowing at times 0 and 4 weeks (mean ± SD).

Treatment	n	Quadrat 1	Quadrat 2	Quadrat 3	Quadrat 4	Quadrat 5
Control 0	3	-387 ± 8	-382 ± 1	-390 ± 4	-428 ± 6	-447 ± 18
Control 4	2	-391 ± 5	-365 ± 21	-365 ± 13	-410 ± 35	-417 ± 3
Bacteria 0	3	-401 ± 18	-386 ± 4	-410 ± 3	-370 ± 6	-408 ± 4
Bacteria 4	2	-349 ± 47	-376 ± 10	-379 ± 18	-340 ± 42	-321 ± 56
Harrow 0	3	-350 ± 4	-387 ± 3	-394 ± 2	-389 ± 4	-409 ± 3
Harrow 4	2	-390 ± 24	-376 ± 5	-394 ± 16	-353 ± 8	-350 ± 20

There were no significant differences between treatments for the change in mean redox values ($P>0.05$), however, there were some differences between quadrats within treatments, especially quadrats 1 and 2 when compared with quadrats 3, 4, and 5 (Fig. 9). The quadrats were therefore clustered into these 2 groups and the ANOVA repeated. This gave a significant difference between bacteria ($F=5.01$, $P=0.05$) and the other 2 treatments of control and harrow.

The results from this study showed that E_h readings under a tuna cage varied considerably without noticeable differences in the appearance of surface sediments. Little variation was experienced between E_h readings taken from cores at the surface and *in situ* readings, however, the nature of the sediments was such that they were well retained in the cores. A less dense substrate could affect the integrity of the core and cause some mixing within the corer.

The increase in E_h levels for all treatments after a month demonstrates that sediment health improves over time after destocking a tuna cage. The bacterial application treatment showed the greatest increase in E_h levels over the experimental period. The addition of bacterial applications such as 'sludge doctor®', has proven to be useful in shrimp pond aquaculture and regular applications may maintain sediment health. Harrowing, may speed the rate of sediment recovery but may not be advisable when fish are present as toxic gasses and pathogenic microorganisms may be stirred up and transported to the fish.

The E_h levels recorded in this study were lower than those previously documented for other marine cage farms (Brown *et al.*, 1987; Gowen *et al.*, 1991; Hargrave *et al.*, 1993). This was unexpected but may explain why no gasses could be liberated from the sediments (see 4.3.2). It has generally been observed that degassing under cages is associated with the presence of anoxic surface sediment, the accumulation of organic waste, and sediment surface E_h between -150 and -200 mV; the sediments in this study had a much lower redox potential.

The management plan for tuna farming in Boston Bay advocates that farmers maintain two thirds of their leases destocked at all times and rotate their cages every 2 years, eventually returning to the original sites after 4 years. This is a preventative strategy as the rates of sediment recovery have not yet been documented for Boston Bay. Underwater measurement of sediment redox potentials in concert with measurement of the rate of sediment degassing and gas composition, offer potential as rapid methods for assessing sediment status under tuna cages. These methods could provide tuna farmers with the basis to make more accurate management decisions concerning cage movement within leases, and fallowing periods.

4.3.2. Methanogenesis

Despite repeated attempts to liberate gasses from sediments under a number of tuna cages, no bubbles were found.

5. General discussion and conclusions

The effect of tuna cages on the benthic flora and fauna of Boston Bay has been shown to be severe within the immediate vicinity of the tuna cages. There is clear evidence for a significant effect up to a distance of 20 metres around each cage with a lesser impact for a further 100 to 150 m from the cages. At a distance of 200 m there is no evidence of an impact relative to control sites situated 1 km distant. These effects are similar to those described for many salmonoid farm sites and, in general, are consistent with those described in the Port Lincoln Aquaculture Management Plan (Bond, 1993) and for which the management strategies were tailored.

In the immediate vicinity of cages (0 - 20 m) the principal impacts can be attributed to the excessive build up of organic detritus which smothered the resident biota. At a slightly greater distance from the cages (20 - 150 m) the rate of input of organic material is still sufficiently high to support elevated populations of some taxa of benthic fauna. In this instance however we presume that the feeding rate of the fauna and the respiration rate of the microbial populations is sufficient to prevent the build-up of detrital material.

A number of factors contribute to the elevated organic loads which result from the tuna farming activities. Within the immediate vicinity of cages non-ingested food contributes to the build-up of detritus. The extent to which these factors contribute to the self pollution of these farms relates by and large to the effective management of the farm. Over feeding, resulting in high wastage, will contribute to the rapid accumulation of organic waste.

The region immediately adjacent to the cages (0 - 5 m), is also impacted by physical disturbance from the predator nets which tend to scrape around on the substrate due to slack in the moorings. Further, if nets are allowed to develop heavy fouling communities this will result in greater amounts of dislodged material building up around the cages.

The build-up of detrital material in the 5 to 20 m zone can be attributed to both rapid sedimentation of larger particles (eg. non-ingested food) as well as accumulation of smaller particles which are derived from feeding tuna. When feeding, tuna may lose a considerable amount of food in the form of fine particulate matter which escapes through their gill slits. The second major source of organic material to this zone comes from faecal material vented by the tuna. In both cases these finer particulates can be distributed away from the cage and will be the primary source of nutrition for the benthic heterotrophs which characterise the seabed up to 150 m from the cage.

The magnitude of impacts will depend largely upon the stocking density of the cage and the nature of the food. It is likely that feeds which break up when taken by the tuna will contribute significantly to detrital loads in the surrounding environments. Further, digestibility will affect the carbon content of faecal material. Stocking rates and choices of feeds will influence the level of organic detritus in the immediate vicinity of the cages but are also central to the commercial viability of the farm. There is therefore a complex interplay between the choice of feeds and stocking densities and the implications these have on detrital levels (and the associated biochemical oxygen demand, methanogenesis, and hydrogen sulphide production) and the level of disease or mortality in the cages.

It is important to recognise that Boston Bay has a number of uses other than tuna farming. The bay has been used as a testing ground for prawn trawling equipment, contains a grain bulk and fertiliser loading facility, is a popular recreational diving and fishing base and is the receiving body for the sewage discharge from Port Lincoln and a number of industries. Accordingly, the management strategies for the harbour need to recognise the multiple uses/impacts (Bond 1993).

Having established the current nature of the disturbance it remains to recommend the future direction of research/monitoring. A variety of questions remain unexamined. Mostly these pertain to the recovery of benthic communities following the destocking and shifting of a cage. The rate of decline and subsequent recovery of the substrate is unknown but can be measured using either or all of redox, epibenthic and infauna measurements. This study has briefly considered recovery through the measurement of sediment E_h. Results of this experiment were however, inconclusive, and accordingly there is broad scope for further refinement of this approach.

Community studies which combine surveys of both the epibenthic and infaunal communities have been shown to illustrate the effect of tuna cages but none of the approaches is suitable in isolation. Monitoring programs should therefore utilise a composite of these approaches. It will remain problematical however, that the costs associated with these approaches, particularly infaunal studies, makes them expensive and the degree of technical expertise required to analyse and interpret the data will make them difficult to apply in a routine manner. We would still maintain that the studies of the community responses are more effective than physico-chemical studies because they directly measure the impact upon the ecosystem. Whereas measurement of a subset of physico-chemical parameters is useful this cannot replace direct measurements of the biota. Infauna and epibenthic studies need not be as intensive as the approach described in these reports; provided that the restrictions of the data are fully recognised a more modest sampling program could be instituted. Such a monitoring program could then be used as a basis for adaptively managing the tuna farming industry.

Use of the remote suction grab, is a less expensive method of data collection in terms of field time, however the cost of sorting infauna samples is restrictive. Alternative methods for sorting this material, can be pursued such as the use of graded sieves and subsampling. Selection of a few significant taxa would also simplify and speed up the sorting process.

The video survey method was also less expensive in terms of field time but did not provide sufficient resolution and its use as a monitoring tool is marginal in its current format. This technique does however, provide the greatest scope for modification and improvement. If developed further this method is likely to be of significant importance in reducing the cost of the biological monitoring programs.

In this analysis we have used video images at set distances to obtain our dataset; this approach fails to make full use of the data resource. An approach that collects data from the transect in strips, called Line Intercept Transecting (LIT; see English *et al.*, 1994), would be worth exploring as it would provide a more complete picture of the change with distance from the cages. This method would require an accurate measurement of the distance that the sled covers (to the nearest 5 cm or better) and would also need to incorporate different ways of mounting the camera.

6. Recommendations

6.1. More detailed study of cage rotation strategies on seafloor souring

Further studies should be undertaken to assess the effect of differences in farm management on the processes of seafloor souring and recovery.

The frequency that cages are moved needs to be considered in more detail using a replicated study which compares degree of souring as a function of the residence time of cages at a given location. This needs to be done within a given region of the Bay to prevent results being confounded by gross differences between locations around Boston Island (e.g. Rotten Bay vs. Boston Bay). Such a study would contribute significantly to providing advice about rotation (and consequently sitting) strategies.

6.2. Refinement of video survey techniques

Methods for using video surveys should be refined and developed to enable their use as a routine monitoring tool.

Video surveys are clearly an ideal technique for a variety of reasons; the lack of a need for diving, easy storage of data, visual effects are easily communicated and it is the least expensive approach to enable large scale ongoing monitoring of the system. Currently this approach is limited by the taxonomic resolution it provides and the inability to accurately gauge distance along the transect. These two factors should be addressed through a detailed development program in order to fully assess the utility of this tool. If, for example, a resolution comparable to diver surveys can be achieved this would provide a very powerful tool for use within this and related industries.

6.3. Detailed investigations of the use of harrowing and bacterial applications

Further studies should be undertaken to investigate the use of bacterial applications and harrowing on maintaining the health or accelerating the recovery of sediments under sea-cages.

Bacterial applications and harrowing may be of benefit in maintaining the health of the seafloor in association with cages (and consequently in reducing mortality in fish) and in accelerating the recovery of sediments during fallowing. The full utility of these approaches needs to be considered in a more detailed investigation which incorporates both physical measurements of redox (as in this report) with investigations of changes in infaunal communities.

6.4. Taxonomic resolution of infaunal data sets

The relative benefit of using an increased taxonomic resolution vs. decreased sampling intensity in infaunal studies should be undertaken in order to evaluate alternative sampling strategies for this system.

Infaunal studies potentially provide a very powerful tool for investigating the level of impact on the ecosystem. The extent to which this potential is realised relates to two problems; the cost of sorting the samples and the taxonomic resolution of the data. The methodology should be further developed to enable a more complete understanding of the extent to which taxonomic resolution improves the power and the cost-benefit of this approach relative to other methods.

6.5. More detailed study of summer vs winter using appropriate replication

Further studies should be undertaken to assess the effect of changes in the physical environment between summer and winter on the processes of seafloor souring and recovery.

A more complete understanding of the process of seafloor souring and its subsequent recovery requires a knowledge of how changes in the physical environment between summer and winter affect these processes. Importantly, changes from summer to winter include changes in temperature, light and oxygen holding capacity of the water. These factors in turn interact with the biota (eg changes in respiration rate vs. oxygen availability) and the outcome of these interactions needs to be understood to evaluate the efficacy of different management options. Importantly, the current study is limited in that it does not contain any replication of seasonal effects and conclusions are therefore limited.

6.6. Use of BACI designs

Future studies should use a Before and After Control and Impact design to clearly document the impact associated with tuna sea cages.

The current study is limited in its capacity to identify impacts from tuna farming because there is no adequate data from before the introduction of farms against which to compare our results. The results do illustrate clear gradient responses from the edge of cages to more distant locations. Given that these responses are consistent with what has been found elsewhere in studies of point source organic pollution and that the changes are not seen on the control transects it is reasonable to conclude that the impacts identified are real. It is frequently the case that environmental monitoring is initiated after a development has begun. In these situations the conclusions drawn from the data must be made with reference to this limitation.

6.7. Use of bay-level controls

Future studies should address the issue of controls for Bay-level responses.

The current study uses control transects which are located within Boston Bay and therefore does not allow us to identify impacts which occur at the level of the entire Bay. Our conclusions assume that such impacts do not occur and that sites distant from the farms (>1,000 m) are appropriate controls: this assumption has not been tested. Until such time as this issue has been resolved one cannot make any claims about the extent to which impacts are limited in their spatial extent. This issue is complicated in that the choice of bay-level controls is highly problematical. Few other bays in this region are in any way comparable with Boston Bay and therefore may not be suitable to use as controls. This issue should however be investigated further with appropriate studies of benthic communities in nearby coastal locations.

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9. Appendix 1 - Changes in abundance of infauna during winter

Two Way Analysis of Variance results for changes in log abundance of infauna (per sample) between cages (ComFarm1, ComFarm2 and ExperFarm) and with increasing distance from the cages in the winter (1994).

Response: Amphipoda

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	13.353356	7.8904	0.0007
Distance	9	9	21.698619	2.8492	0.0052
Location*Distance	18	18	66.202265	4.3465	0.0000

Response: Anthozoa

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	9.59146	6.2480	0.0028
Distance	9	9	203.19683	29.4142	0.0000
Location*Distance	18	18	52.75161	3.8181	0.0000

Response: Astacillidae

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.0679136	1.5868	0.2100
Distance	9	9	0.4450224	2.3106	0.0215
Location*Distance	18	18	1.0191188	2.6457	0.0012

Response: Asteroidea

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.03179966	0.8741	0.4206
Distance	9	9	0.21711720	1.3262	0.2339
Location*Distance	18	18	0.48119774	1.4697	0.1190

Response: Bivalvia

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.495299	0.4235	0.6560
Distance	9	9	14.538616	2.7627	0.0065
Location*Distance	18	18	40.741283	3.8709	0.0000

Response: Brachyura

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	1.540696	2.2882	0.1071
Distance	9	9	21.545237	7.1106	0.0000
Location*Distance	18	18	8.637677	1.4254	0.1380

Response: Bryozoa

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.2890950	3.0865	0.0503
Distance	9	9	0.4587464	1.0884	0.3788
Location*Distance	18	18	1.0399470	1.2336	0.2515

Response: Caprellidae

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.42465604	3.2022	0.0451
Distance	9	9	0.76043405	1.2743	0.2612
Location*Distance	18	18	0.93971728	0.7874	0.7098

Response: Cephalopoda

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.01420023	0.6912	0.5035
Distance	9	9	0.05087480	0.5503	0.8340
Location*Distance	18	18	0.11956628	0.6467	0.8532

Response: Cumacea

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.3794268	0.9501	0.3904
Distance	9	9	0.9233694	0.5138	0.8613
Location*Distance	18	18	2.7608537	0.7681	0.7312

Response: Decapoda

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	2.285493	2.1658	0.1203
Distance	9	9	21.626615	4.5542	0.0001
Location*Distance	18	18	25.412006	2.6757	0.0011

Response: Echiura

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.08053664	0.6893	0.5045
Distance	9	9	0.82936396	1.5774	0.1333
Location*Distance	18	18	0.78175616	0.7434	0.7582

Response: Eggs

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.2524580	0.8804	0.4180
Distance	9	9	5.7467481	4.4533	0.0001
Location*Distance	18	18	1.8669994	0.7234	0.7793

Response: Gastropoda

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	2.827719	3.5890	0.0315
Distance	9	9	5.780387	1.6303	0.1177
Location*Distance	18	18	18.289365	2.5792	0.0016

Response: Holothuroidea

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	1.666833	0.9095	0.4063
Distance	9	9	15.924148	1.9308	0.0567
Location*Distance	18	18	43.077543	2.6115	0.0014

Response: Hydrozoa

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.1021027	0.4311	0.6511
Distance	9	9	0.9306905	0.8732	0.5520
Location*Distance	18	18	1.5665013	0.7349	0.7673

Response: Isopoda

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	2.703532	2.9777	0.0557
Distance	9	9	16.666981	4.0793	0.0002
Location*Distance	18	18	38.112354	4.6641	0.0000

Response: Mysidacea

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	8.527642	29.8660	0.0000
Distance	9	9	6.694688	5.2103	0.0000
Location*Distance	18	18	13.916416	5.4154	0.0000

Response: Nebaliidae

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	8.716139	11.0152	0.0001
Distance	9	9	85.104823	23.9008	0.0000
Location*Distance	18	18	54.392923	7.6378	0.0000

Response: Ophiuroidea

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	6.1507163	24.9327	0.0000
Distance	9	9	3.1613411	2.8477	0.0052
Location*Distance	18	18	7.6586384	3.4495	0.0000

Response: Opistobranchia

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.6644721	1.6319	0.2010
Distance	9	9	1.2434766	0.6787	0.7264
Location*Distance	18	18	3.3813628	0.9227	0.5537

Response: Osteichthyes

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.0035501	0.0337	0.9669
Distance	9	9	0.7198882	1.5194	0.1524
Location*Distance	18	18	1.0811567	1.1410	0.3270

Response: Ostracoda

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	3.328869	6.7030	0.0019
Distance	9	9	3.834126	1.7156	0.0961
Location*Distance	18	18	13.311765	2.9783	0.0003

Response: Paguridae

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.1744224	2.4258	0.0939
Distance	9	9	0.7038768	2.1754	0.0305
Location*Distance	18	18	1.8011854	2.7833	0.0007

Response: Polychaeta

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	5.316343	3.4656	0.0353
Distance	9	9	90.785773	13.1515	0.0000
Location*Distance	18	18	21.086057	1.5273	0.0977

Response: Polyplacophora

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.06732664	1.2101	0.3028
Distance	9	9	0.52466877	2.0955	0.0374
Location*Distance	18	18	0.44537081	0.8894	0.5922

Response: Porifera

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	12.261709	8.6669	0.0004
Distance	9	9	10.557132	1.6582	0.1102
Location*Distance	18	18	21.180374	1.6634	0.0603

Response: Talitridae

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.01915023	0.9943	0.3738
Distance	9	9	0.08706009	1.0045	0.4421
Location*Distance	18	18	0.18054864	1.0416	0.4229

Response: Tanaidacea

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	7.762124	8.5927	0.0004
Distance	9	9	7.754083	1.9075	0.0601
Location*Distance	18	18	12.549311	1.5436	0.0923

Response: Turbellaria

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Location	2	2	0.7606098	5.1866	0.0073
Distance	9	9	2.2724122	3.4434	0.0011
Location*Distance	18	18	5.6614870	4.2895	0.0000

10. Appendix 2 - Changes in abundance of infauna between the summer and the winter

Three Way Analysis of Variance results for changes in log abundance of infauna between cages, with increasing distance from the cages and over time (winter -1994 vs. summer -1995).
Response - Amphipoda.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	24.168809	25.2839	0.0000
Location	2	2	33.226726	17.3799	0.0000
Distance	3	3	38.524956	13.4341	0.0000
Time*Location*Distance	6	6	5.463772	0.9526	0.4647
Time*Location	2	2	5.993004	3.1348	0.0506
Location*Distance	6	6	41.027315	7.1534	0.0000
Time*Distance	3	3	7.223585	2.5190	0.0663

Response - Anthozoa.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	44.508263	49.1001	0.0000
Location	2	2	0.809294	0.4464	0.6420
Distance	3	3	71.674361	26.3563	0.0000
Time*Location	2	2	6.012204	3.3162	0.0429
Time*Distance	3	3	44.428856	16.3375	0.0000
Location*Distance	6	6	24.316777	4.4709	0.0008
Time*Location*Distance	6	6	10.847998	1.9945	0.0802

Response - Bivalvia.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	28.509902	43.2325	0.0000
Location	2	2	2.847060	2.1586	0.1242
Distance	3	3	17.317425	8.7534	0.0001
Time*Location	2	2	3.990553	3.0256	0.0559
Time*Distance	3	3	6.407152	3.2386	0.0281
Location*Distance	6	6	40.394548	10.2091	0.0000
Time*Location*Distance	6	6	6.460067	1.6327	0.1535

Response - Brachyura.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	5.4810629	15.3631	0.0002
Location	2	2	2.0618216	2.8896	0.0632
Distance	3	3	3.6102258	3.3731	0.0240
Time*Location	2	2	0.7273090	1.0193	0.3669
Time*Distance	3	3	6.9033021	6.4499	0.0007
Location*Distance	6	6	6.7021176	3.1309	0.0097
Time*Location*Distance	6	6	0.7845911	0.3665	0.8973

Response - Caprellidea.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	21.407274	42.0460	0.0000
Location	2	2	8.774169	8.6167	0.0005
Distance	3	3	18.220464	11.9289	0.0000
Time*Location	2	2	6.828695	6.7061	0.0023
Time*Distance	3	3	16.861798	11.0394	0.0000
Location*Distance	6	6	9.182779	3.0060	0.0122
Time*Location*Distance	6	6	10.274875	3.3635	0.0063

Response - Decapoda.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	6.002811	16.3002	0.0002
Location	2	2	1.652797	2.2440	0.1147
Distance	3	3	12.473231	11.2901	0.0000
Time*Location	2	2	1.651279	2.2420	0.1149
Time*Distance	3	3	1.887596	1.7085	0.1746
Location*Distance	6	6	8.614942	3.8989	0.0023
Time*Location*Distance	6	6	2.373235	1.0741	0.3880

Response - Echinoidea.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	18.159895	33.6887	0.0000
Location	2	2	8.787180	8.1506	0.0007
Distance	3	3	6.106319	3.7760	0.0149
Time*Location	2	2	1.901708	1.7639	0.1800
Time*Distance	3	3	32.383847	20.0253	0.0000
Location*Distance	6	6	21.089861	6.5207	0.0000
Time*Location*Distance	6	6	22.352063	6.9109	0.0000

Response - Isopoda.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	0.303833	0.6853	0.4110
Location	2	2	1.941947	2.1901	0.1206
Distance	3	3	20.057614	15.0805	0.0000
Time*Location	2	2	5.624088	6.3428	0.0031
Time*Distance	3	3	1.295732	0.9742	0.4108
Location*Distance	6	6	3.783652	1.4224	0.2208
Time*Location*Distance	6	6	4.867207	1.8297	0.1081

Response - Mysidacea.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	6.003028	26.6384	0.0000
Location	2	2	3.203833	7.1085	0.0017
Distance	3	3	11.540842	17.0708	0.0000
Time*Location	2	2	1.449289	3.2156	0.0470
Time*Distance	3	3	5.878324	8.6950	0.0001
Location*Distance	6	6	4.807491	3.5555	0.0044
Time*Location*Distance	6	6	3.472435	2.5681	0.0277

Response - Nebaliidae.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	11.961868	21.1253	0.0000
Location	2	2	10.086739	8.9069	0.0004
Distance	3	3	23.125803	13.6138	0.0000
Time*Location	2	2	4.338151	3.8307	0.0271
Time*Distance	3	3	26.216721	15.4334	0.0000
Location*Distance	6	6	19.642563	5.7816	0.0001
Time*Location*Distance	6	6	18.696267	5.5031	0.0001

Response - Opistobranchia.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	4.1308782	11.1677	0.0014
Location	2	2	2.0780703	2.8090	0.0681
Distance	3	3	1.1211931	1.0104	0.3944
Time*Location	2	2	1.2731719	1.7210	0.1875
Time*Distance	3	3	1.0283508	0.9267	0.4334
Location*Distance	6	6	2.5634677	1.1550	0.3422
Time*Location*Distance	6	6	3.0891201	1.3919	0.2324

Response - Ostracoda.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	3.6125447	14.8835	0.0003
Location	2	2	4.6505402	9.5800	0.0002
Distance	3	3	4.9429702	6.7883	0.0005
Time*Location	2	2	0.6718615	1.3840	0.2583
Time*Distance	3	3	2.8750448	3.9483	0.0122
Location*Distance	6	6	6.6336847	4.5551	0.0007
Time*Location*Distance	6	6	0.7675309	0.5270	0.7856

Response - Polychaeta.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	0.045904	0.0475	0.8281
Location	2	2	2.166760	1.1221	0.3322
Distance	3	3	13.265642	4.5799	0.0059
Time*Location	2	2	22.444970	11.6235	0.0001
Time*Distance	3	3	31.781683	10.9725	0.0000
Location*Distance	6	6	27.654737	4.7738	0.0005
Time*Location*Distance	6	6	8.901724	1.5366	0.1815

Response - Polyplacophora.

Source	Nparm	DF	Sum of Squares	F Ratio	Prob>F
Time	1	1	0.37743876	7.7731	0.0071
Location	2	2	0.18729620	1.9286	0.1541
Distance	3	3	0.59573852	4.0896	0.0104
Time*Location	2	2	0.18729620	1.9286	0.1541
Time*Distance	3	3	0.59573852	4.0896	0.0104
Location*Distance	6	6	0.61041448	2.0952	0.0667
Time*Location*Distance	6	6	0.61041448	2.0952	0.0667