Monitoring Spatial and Temporal Patterns Structuring Eelgrass (*Zostera marina* L.) Fish Diversity in Clayoquot Sound

by

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Abstract

The linking of temporal variability in biodiversity with spatial scale is an integral component of marine conservation. In this study, I evaluated the interplay between the temporal variability in fish communities and local-to-regional environmental properties measured for eelgrass beds (*Zostera marina*) found in southern Clayoquot Sound, British Columbia. Effective monitoring of the temporal variability in eelgrass fish diversity required: 1) triplicate beach seine sets on a rising low tide, 2) sampling in early spring, early summer and late summer, and 3) diel sampling. The analysis of eelgrass fish data required alpha diversity measures (e.g. species richness and taxonomic distinctness), beta diversity measures (e.g. taxonomic similarity), and multivariate methods to detect subtle temporal and between-site differences. Additional temporal changes in eelgrass fish diversity were evaluated in response to changes in local eelgrass bed properties and to regional changes in hydrodynamic energy. I found, in late spring, fish diversity differed between sites but these differences were not explained by eelgrass properties or hydrodynamic energy. However, in late summer, fish diversity appeared to be related to regional hydrodynamic energy but the explanatory power of local environmental and eelgrass variables was not high. Further research addressing additional abiotic variables and the influence of biotic processes is recommended.

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Chapter 1. Introduction

Biogeography is the spatial and temporal study of living organisms (Cox and Moore 1998). Central concepts of biogeography involve determining patterns of biological communities and the processes driving these patterns and how these concepts can be applied to understanding and conserving ecosystems (Brown et al. 1996). Consequently, conservation and monitoring of marine biodiversity and marine reserve establishment requires predicting the consequences and causes of spatial and temporal pattern in ecosystems (Garcia-Charton and Perez-Ruzafa 1999, Yoccoz et al. 2001).

The heterogeneity and patchiness of ecosystems over a wide range of scales is fundamental to the organization of ecological communities (Levin 1992, Garcia-Charton and Perez-Ruzafa 1999). For example, biogeographical scales can range from large, landscape scales to small, local scales (Menge and Olson 1990). Gustafson (1998) identified a number of different methods to measure heterogeneity which included both spatial and nonspatial components. Nonspatial components represent community composition, and are evaluated using diversity measures; spatial components represent configurations, and are measured by patch-based indices including size, shape, and connectivity. Since natural systems exhibit high variability, as communities respond individually to spatial and temporal changes, it is important to measure ecological communities over a diverse range of scales to determine the causes and consequences of ecological patterns (Levin 1992).

The identification of the scale and pattern of ecological processes driving marine communities is essential to evaluate management tools such as marine reserves and monitoring programs (Garcia-Charton and Perez-Ruzafa 1999, Yoccoz et al. 2001). The effect of spatial and temporal variation has been studied on a diverse array of marine communities including soft-sediment invertebrates (Ysabaert and Herman 2002), rocky intertidal algae and invertebrates (Underwood and Skilleter 1996), surf zone fish (Clark et al. 1996), seagrass macroinvertebrates (Hovel 2003) and seagrass epiphytic algal communities (Lavery and Vanderklift 2002). However, research has not demonstrated similar patterns across marine habitats, over time or even within habitats (Hovel et al. 2002). Therefore, due to the complexity of interactions occurring at a variety of scales within marine ecosystems, it is essential to further evaluate processes that drive the high variability existing in marine landscapes.

Seagrass landscapes have been identified as ideal marine models to examine concepts such as scale and pattern (Robbins and Bell 1994, Turner et al. 1999). In addition, from a conservation perspective, eelgrass beds are given high priority as one of the most productive benthic ecosystems in estuarine and coastal areas (McAllister 1995, Gray 1997). Eelgrass beds (*Zostera marina* L.) also cover thousands of hectares of nearshore habitat in the Pacific Northwest (Bulthuis 1995), and coastal development coupled with inadequate scientific information has generated the need to understand eelgrass ecosystems better (Wyllie-Echeverria and Thom 1994). Consequently, the effects of spatial and temporal heterogeneity on eelgrass ecological communities are important components to understand and conserve such productive nearshore ecosystems.

The influence of seagrass bed spatial structure has been examined for macrofaunal communities (Frost et al. 1999, Turner et al. 1999, Hovel 2003), but few studies have examined the influence on fish communities (Bell et al. 2001). This study evaluates the spatial and temporal patterns structuring eelgrass fish communities within and outside the national park boundaries of Pacific Rim National Park Reserve of Canada in Clayoquot Sound, British Columbia, through three components:

- Evaluation of the accuracy and efficiency of the sampling methodology, beach seining, for sampling eelgrass fish diversity;
- Assessment of temporal and spatial patterns of eelgrass fish diversity through the application of a wide array of diversity measures; and
- Investigation of the influence of hydrodynamic regime on fish community properties.

By evaluating these three components, a diverse range of scales and patterns can be used to assess potential factors influencing eelgrass fish community structure. The results of such evaluation will aid in understanding the complex variation in eelgrass ecological communities, and can be applied to national park monitoring programs, marine reserve development and nearshore habitat conservation.

Chapter 2: Evaluation of the Beach Seine Sampling Methodology

2.1. Introduction

The recent global decline of habitats and species has generated the need to develop scientific monitoring programs to conserve biological diversity (Wilson 1988, Yoccoz et al. 2001). In aquatic ecosystems, fish diversity has been used to address ecosystem integrity and biological conservation (Deegan et al. 1997, Angermeier and Winston 1999). Fish diversity in coastal marine ecosystems, such as seagrass beds, has been well documented in Australia, the eastern United States, and southeast Alaska (Sogard et al. 1989a, Edgar and Shaw 1995a, Robards et al. 1999). However, little research has examined fish diversity variability between seagrass beds (Robinson and Yakimishyn unpublished manuscript). Additionally, maintenance of ecological integrity is a primary mandate of Parks Canada and requires developing programs to monitor biological diversity (Parks Canada 1995).

In this study, fish diversity was assessed in eelgrass beds (*Zostera marina*) in Clayoquot Sound on the west coast of Vancouver Island. Eelgrass fish diversity was examined at the species level, by measuring the number and relative abundance of species within an ecosystem (Legendre and Legendre 1983). A true measurement of species diversity would include every species inhabiting the ecosystem (Gauch 1982). However, obtaining the true number of species utilizing the habitat is limited by the sampling methodology, as no single sampling method can adequately sample the entire diversity of fish assemblages (Edgar et al. 2001). Fish diversity can vary depending on numerous factors including the type of sampling gear, habitat heterogeneity, sampling sufficiency, and spatio-temporal variation (Rozas and Minello 1997, Garcia-Charton and Perez-Ruzafa 1999, Cao et al. 2001, Jackson and Jones 1999).

I was interested in developing a method to sample eelgrass fish diversity that was inexpensive, effective at sampling a broad range of fish species, and was easily deployed multiple times within a short time window. Previous work has identified beach seining as a sampling methodology that meets these criteria (Connolly 1994a, Edgar et al. 2001). However, there are several issues that arise in the literature concerning the efficiency of beach seining. In particular:

- Studies using beach seining have found diel and tidal variation to influence fish species caught (Gibson et al. 1996, Morrison et al. 2002). Conflicting results among previous studies have shown inconsistent effects of diel and tidal variability on fish diversity catches (Table 2.1). As a result, I evaluated beach seine sampling sufficiency with respect to diel and tidal effects on eelgrass fish assemblages.
- Beach seining also exhibits high catch variability due to sampling a limited area and aggregations of schooling species (Rosaz and Minello 1997). Therefore, I evaluated how effectively triplicate seines estimated the total number of fish species within eelgrass beds.
- Finally, beach seining can be potentially detrimental to seagrass beds through activities such as boat propellers cutting seagrass, boat hulls dragging through seagrass, and physical dragging of the seine through the beds (Zieman 1976,Walker et al. 1989, Pasqualini et al. 2000). Therefore, I evaluated those potential impacts and their effects on fish assemblages by examining repetitive hauling effects.

By exploring the potential effect of diel and tidal variation, repetitive hauling and catch efficiency, the effectiveness of beach seining methodology was addressed for eelgrass fish biodiversity in Clayoquot Sound.

Table 2.1. Summary of previous work that examined diurnal and tidal effects on fish catch. Studies examined differences in fish species richness and abundance in day versus night (diurnal variation) and low versus high tide (tidal variation), with both significant differences and general trends presented.
(Abbreviations: >=more caught, NA=not applicable to study, NSD=no significant difference found)

				Diurnal Variation		**Tidal Variation**	
Source	**Study Location**	**Habitat Type**	**Sampling Gear**	**Fish Species Richness**	**Fish Abundance**	**Fish Species Richness**	**Fish Abundance**
Heck and Orth 1980	Chesapeake Bay, Virginia	Seagrass and bare sand	Otter trawl, gill net	NSD, trend > at night	NSD, trend > at night	NA	NA
Lasiak 1984	South Africa coast	Surf-zone	Beach seine	NSD, trend > at night	NSD, trend > in the day	NSD, trend > at low tide	NSD, trend > at low tide
Hargreaves et al. 1987	Masset Inlet, British Columbia	Seagrass	Beach seine	NA	NA	Significantly > at low tide on June 7 but NSD on June 5	Significantly > at low tide on June 7 but NSD on June 5
Sogard et al. 1989a	Florida coast	Seagrass	Gill net	NA	Significantly > at dawn, dusk and midnight	NA	NSD
Lubbers et al. 1990	Chesapeake Bay, Virginia	Seagrass and bare sand	Beach seine	NSD	NSD, trend >at dusk and midnight	NA	NA
Edgar and Shaw 1995a	West Port, Australia	Seagrass and bare sand	Beach seine	Significantly > at night	NSD	NA	NA
Gibson et al. 1996	West coast of Scotland	Sandy beach	Beach seine	Significantly > at night	NSD	Significantly > at low tide	Significantly > at low tide
Gray et al. 1998	South-eastern Australia	Seagrass and sand	Beach seine	Significantly > at night in sand	NSD	NA	NA

Table 2.1. Continued

				Diurnal Variation		**Tidal Variation**	
Source	**Study Location**	**Habitat Type**	**Sampling Gear**	**Fish Species Richness**	**Fish Abundance**	**Fish Species Richness**	**Fish Abundance**
Nash and Santos 1998	Azores coast	Sandy beach	Beach seine	Significantly > at night	Significantly > in the day	NA	NA
Mattila et al. 1999	Damariscotta River, Maine	Seagrass and bare sand	Beach seine	Significantly > at night	Significantly > at night	NA	NA
Robards et al. 1999	Cook Inlet, Alaska	Sand and mud	Beach seine	NA	NA	NSD, trend > at low tide	Significantly > at high tide
Hindell et al. 2000	Port Phillip Bay, Australia	Seagrass	Beach seine, gill net	NA	Significantly > small fish at night low tide	NA	Significantly > small fish at night low tide
Layman 2000	Virginia coast	Surf-zone	Beach seine	Significantly > at night	NSD	Significantly > at low tide	NSD
Griffiths 2001	New South Wales coast	Estuarine lagoons	Beach seine	Significantly > at night	NSD	NA	NA
Methven et al. 2001	East coast of Newfoundland	Rock and gravel bay	Beach seine	NSD, trend > at night	NSD	NA	NA
Morrison et al. 2002	New Zealand coast	Tidal mudflat	Beach seine	Significantly > at night during low tide	Significantly > at night during low tide	Significantly > at low tide	Significantly > at low tide
Suda et al. 2002	Japan coast	Sandy beach	Beach seine	Significantly > at night	Significantly > at night	Significantly > at low tide	Significantly > at low tide
This thesis, 2003	Clayoquot Sound, British Columbia	Seagrass	Beach seine	NSD	Significantly > fish caught in the day	Significantly > species caught at low tide	NSD, trend > at low tide

2.2. Methods

2.2.1. Study Location

2.2.1.1. Diel, Tidal, Hauling Effects and Replication Sites

The study site, Grice Bay (49° 06'N 125° 47'W), is located in Clayoquot Sound on the west coast of Vancouver Island, and is a component of the Pacific Rim National Park Reserve of Canada (Figure 2.1). The bay is approximately 7 km^2 in size, and is sufficiently shallow so that most of the bay is exposed to air at a mean low tide of 0m, and has a maximum depth of approximately 6m at high tide (DFO 1995). The substrate consists of mud and fine sand, and the vegetation is dominated by a single seagrass species, the eelgrass *Zostera marina* (LUCO 1999). The eelgrass beds studied were an extensive bed on the southern side of Indian Island (Indian, N 49° 112' 00" W 125° 775' 52") and a second bed at the mouth of Grice Bay (South Bay, N 49° 07' 564" W 125° 48' 645"), adjacent to the southern tip of Meares Island (Figure 2.1). At a low tide of 0.6m or less, both eelgrass beds have a large, flat area of intertidal eelgrass exposed and a gentle sloping subtidal component (5 to 25% subtidal slope). These two eelgrass beds were chosen because the extensive intertidal eelgrass components of the sites allowed for ease of night and variable tidal height sampling. The Indian bed was sampled within a channel approximately 6m at maximum depth with strong tidal currents. At South Bay, a small freshwater creek, approximately 1m in width, flowed down the centre of the bay. The subtidal portion of the eelgrass bed was sampled on both sides of the creek.

2.2.1.2. Sampling Sufficiency Sites

Sampling sufficiency was examined for seven eelgrass beds, including three within Pacific Rim National Park Reserve of Canada in Grice Bay, Auseth, Indian and Kootowis, one in Browning Passage, Ducking, and two in Lemmens Inlet, Sharp and Arakun and one in Van Nevel Channel, Felice. (Figure 2.1). The characteristics of each bed are summarised in Table 2.2.

2.2.1.3. Masset Inlet Sites

Fish assemblage data collected by Hargreaves et al. (1987) in Masset Inlet, Queen Charlotte Islands was also analysed for low and high tide comparisons. Eelgrass fish diversity data from June 5th and 7th in 1985 were examined from this study.

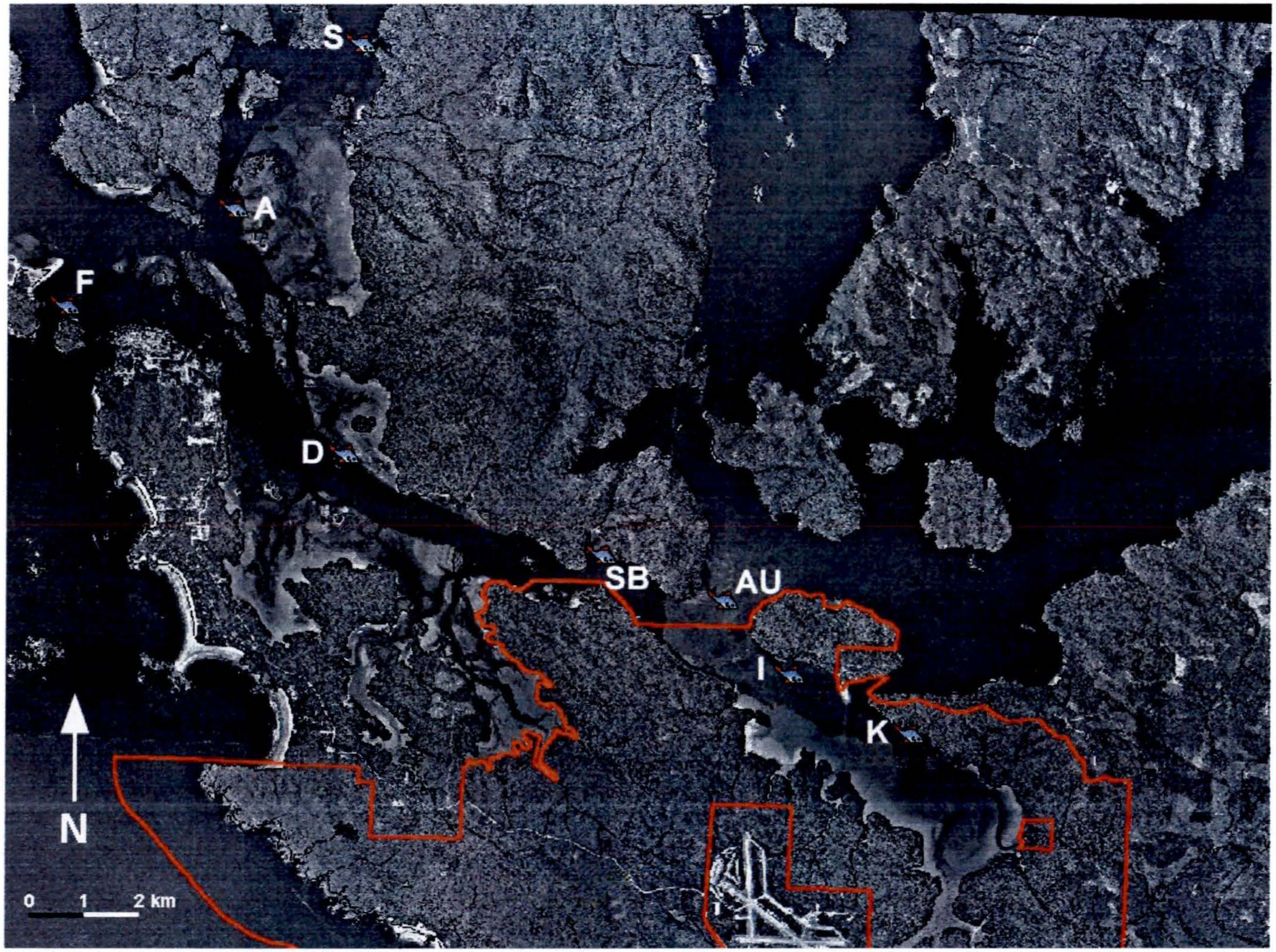

Figure 2.1. Map of study area in Clayoquot Sound where the letters and fish symbols indicate the eelgrass beds sampled for fish diversity and the red lines indicate the boundaries of Pacific Rim National Park Reserve of Canada. (Sites: A = Arakun, AU = Auseth, D = Ducking, F = Felice, I = Indian, K = Kootowis, S = Sharp, SB = South Bay).

Table 2.2. Summary of site characteristics at each eelgrass bed.

Site	Locality	GPS coordinates	Site description
Arakun	Lemmens Inlet	N 49 169' 74" W 125 888' 78"	Located on the southeast side of a sand bar, with a steep sloping subtidal component (30 to 50% subtidal slope)
Auseth	Grice Bay	N 49° 120' 81" W 125° 786' 17"	Located on the northwest side of a shallow sand bar, at the mouth of Grice Bay, with a steep sloping subtidal component (20 to 50% subtidal slope)
Ducking	Browning Passage	N 49° 139' 10" W 125° 865' 08"	An extensive intertidal bed, and the subtidal component is within a large channel with strong tidal currents and a gentle slope (5 to 10% subtidal slope)
Felice	Van Nevel Channel	N 49° 16' 623" W 125° 93' 397"	Located on the southeast side of Van Nevel Channel on the northern side of Felice Island with a gentle subtidal slope (5 to 10% subtidal slope)
Indian	Grice Bay	N 49° 112' 00" W 125° 775' 52"	An extensive bed, on the southern side of Indian Island, sampled within a channel with strong tidal currents and gentle sloping subtidal component (5 to 25% subtidal slope)
Kootowis	Grice Bay	N 49° 104' 35" W 125° 748' 62"	Located in a small protected bay in the north western part of Grice Bay with a gentle subtidal slope (0 to 5% subtidal slope)
Sharp	Lemmens Inlet	N 49° 193' 00" W 125° 862' 48"	Located between two small rocks in Lemmens Inlet with a gentle subtidal (5 to 10% subtidal slope)

2.2.2. Sampling Gear

A 9.2 m long beach seine, with 4mm stretch mesh, having a 3.1 m drop in the centre, and tapering to 1.1 m at the wings, with two 10 m long ropes, one attached to the bottom lead line and the other to the top float line at either end of the net, was used to sample fish assemblages. To prevent the net from rolling, a weight attached to 1 m of rope was tied to the centre of the net on the lead line. Two individuals, using a 24 ft aluminium skiff with a 115 hp outboard engine, deployed the seine. One individual would be dropped off on the bed, holding two lines from one end of the seine. The net and 10 m of rope was stretched perpendicular to shore. When fully extended, the seine was stretched parallel to shore, then the second individual was dropped off on shore, and the two individuals pulled the seine to shore, so that approximately a 9 m X 10 m area (90 m^2) of the eelgrass bed was sampled. Triplicate sets were done for each sampling period, with a minimum 10 metre distance between each haul to avoid the physical disturbance effect caused by pulling the beach through the eelgrass. All fish were placed in buckets of fresh seawater aboard the boat, to be counted, and identified using field guides (Hart 1988, Lamb and Edgell 1986). Due sampling time constraints and the high number of individuals caught, fork lengths, to the nearest millimetre, were only taken of the first 25 individuals of each species and returned to the ocean.

2.2.3. Sampling Design

2.2.3.1. Replication

By relating species richness to sampling effort catch variability between seines was assessed. It is commonly observed that the number of new species caught increases rapidly in the first set of replicates and then drops off (Cao et al. 2001). Previous research has suggested that one or two beach seine hauls is adequate to achieve a reliable estimate of species richness (Allen et al. 1992, Robards et al .1999). This is unrealistic due to the high catch variability of beach seining (Rozas and Minello 1997). Therefore, the number of replicates needed to achieve an accurate estimate of species richness was examined through five consecutive beach seine hauls at the Indian eelgrass bed in July 2001.

2.2.3.2. Diel Variation

Sampling was done in August 2002 at the Indian and South Bay beds to address how fish assemblages vary between day and night. August was chosen as the ideal month because it had tidal heights low enough to beach seine during both the day (0900-1200h) and night (2100-2300h). Triplicate sets, without replacement, were conducted for each sampling event. Day and night sampling was done on the same day with hauling location slightly offset to avoid the effect that earlier hauling may have on fish assemblages. To ensure complete darkness, night time seining occurred one hour after sunset. Tidal height at the Indian bed ranged from 0.8-0.9 m in the day and 0.9-1.0 m at night, whereas tidal height at the South Bay bed ranged from 0.7-0.8 m in the day and 1.3-1.5 m at night.

2.2.3.3. Tidal Variation

In June 2002, the Indian and South Bay beds were seined to address fish assemblage differences at high versus low tidal heights. At each site the beds were sampled at low tide (0.2-0.5 m) in the morning (0800-1100h) and at high tide (1.8-2.2 m) in the afternoon (1200-1400h) on the same day. Triplicate sets, without replacement, were conducted for each sampling event. High tide sampling was done as close to the peak tidal height that permitted seining eelgrass habitat rather than the mud and rock substrate found at higher tidal levels on the beach.

The effect sampling at different tidal stages has on fish diversity was also explored on a rising tide at the South Bay eelgrass bed in June 2002. Replicate sets were made at four tidal heights (0.5, 0.8, 1.3 and 2.1 m) without overlapping sets. At each tidal sampling stage fish were identified, counted and returned to the ocean.

Masset Inlet fish data was compared to Clayoquot Sound fish data to determine if similar tidal trends in species richness and abundance occurred across eelgrass beds in different areas. Fish assemblage data from two days in June 1985 from Masset Inlet (Hargreaves et al. 1987) was also analyzed for low and high tidal comparisons Those sites were beach seined at low tide (0.0-0.5 m) and high tide (1.1-2.6 m). All fish species identified and counted. To avoid seasonal variation in fish assemblage data, and to allow for comparisons to be made between Clayoquot Sound and Masset Inlet, only data from June was used from the Hargreaves et al. (1987) data set.

In addition, fish assemblage data from four days sampled in Masset Inlet within the same week in 1985 (May 27 - June 7) were also analyzed for rising and falling tidal effects. Five tidal stages were analyzed: falling high tide (2.8 to 2.4 m), falling mid tide (2.0 to 1.4 m), falling low tide (1.2 to 0.0 m), rising low tide (0.3 to 1.2 m), and rising high tide (1.9 to 3.0 m). Finally, exploration of effect tidal height has on fish species richness was examined in the entire Masset Inlet data set, from 1983 to 1985, to determine if there were a relationship between tidal height and the number of fish species caught.

2.2.3.4. Hauling Effects

To address the effect previous beach seine hauls may have on eelgrass fish assemblages, repetitive seines were made at the Indian eelgrass bed. Repetitive seining was assessed using triplicate beach seine hauls during three separate sampling periods: June 26th, July 11th and July 24th. All three sets during the first seining event (June 26th) had not been previously sampled. The second sampling event (July 11th) had two areas resampled from June 26th, and one area had never been previously sampled. Lastly, the third sampling event (July 24th) had one set area previously sampled on both June 26th and July 11th, one area was sampled only once before on July 11th,and the third area had never been previously sampled. Each haul was marked with PVC pipe and flagging tape. GPS coordinates were recorded, which allowed the set locations from previous sampling events to be found. Fish assemblages could then be compared between sets to determine if catch differences exist between replicates depending on the number of times each area had been seined previously.

2.2.4. Data Analysis

2.2.4.1. Diel and Tidal Variation

Fish were sampled at diel and tidal variations for species richness and abundance. Data from three replicate hauls were analyzed using a one-factor analysis of variance (ANOVA), where day/night and tidal height were the independent factors. If means differed significantly, Tukey's test was performed to determine where the differences occurred. To account for equal variance all fish abundance data was Log(1+x) transformed (Clarke and Green 1988). Heterogeneity of variance was tested for each data set using Levene's test. If the data failed equal variance, the nonparametric Kruskal-Wallis test was applied (Zar 1999). Finally, regression analysis was applied to determine the effect tidal height has on Masset Inlet fish species richness collected in 1983, 1984 and 1985. SPSS statistical package version 10.0 was used for all statistical analysis.

Diel and tidal comparisons of the abundance of individual fish species groups were also explored. Fish species that contained more than five individuals in the same taxonomic family were grouped into fish family groups such as, shiner perch (*Cymatogaster aggregata*), pile perch (*Rhacochilus vacca*), striped seaperch (*Embiotoca lateralis*) and kelp perch (*Brachyistius frenatus*) were recognized as the seaperch group, the Embiotocidae. However, some fish family groups had insufficient numbers, and fish species were then grouped into functional guilds. For example, Mathieson et al. (2000) used functional guilds to describe fish assemblages in tidal marshes by grouping fish according to vertical distribution, habitat, substrate and dietary preferences. Thereby, families with few individuals were grouped into functional guilds, such as pipefish (*Syngnathus leptorhynchus*), and tube-snouts (*Aulorhynchus flavidus*) were grouped together because both species are found midwater in the eelgrass beds and exhibit similar foraging behaviour. Fish group abundance data from South Bay and Indian were combined for comparison, with the data from two sampling days combined for the Masset Inlet, allowing for ease of interpretation. Additional comparisons of fish species mean fork lengths were done to assess whether different fish size classes utilize eelgrass beds variably depending on diel (day/night) and tidal (low/high) stages. However, due to low sample sizes, lengths of fish species were not compared for the four rising tidal stages.

2.2.4.2. Sampling Sufficiency

The effectiveness of beach seining as a method to sample fish diversity was also determined for Clayoquot Sound eelgrass beds. Cao et al (2001) proposed a method for establishing an empirical relationship between the proportion of total species richness and the similarity among replicate samples. The authors suggest using the autosimilarity measure known as the Jaccard Coefficient (JC). The percent of true total species richness ($\%TSR_{tru}$) for a region is calculated as $100 * JC$, where $JC = c/(a+b+c)$, and where c is the number of species common to both replicates, a is the number of species unique to replicate 1, and b is the number of species unique to replicate 2. Seven eelgrass beds (see section 2.2.1.2) were sampled monthly from May to September in 2001 and bimonthly from April to September in 2002. Sampling sufficiency was addressed by exploring the fish diversity data in two ways. First, a monthly sampling sufficiency was determined by combining data from all sites sampled in 2001 and 2002 for each month. Second, a site-month sampling sufficiency was determined for each site in 2002. Monthly sampling sufficiency was calculated by drawing two equal replicates from the total number of beach seine sets taken monthly in 2001 and 2002 separately. A site-month sampling sufficiency was calculated by drawing two equal replicates from the total number of sets taken for each site-month combination. The JC was then calculated from the species list information from each replicate. For example, if 40 beach seine sets were collected in Clayoquot Sound in July, then 20 sets were randomly drawn, with replacement, from the original 40, and a list of captured fish species was established. Likewise, a second group of 20 sets was randomly drawn from the original list of 40 sets, and a second species list developed. The JC was then calculated from these two species lists. That process was repeated 100 times and an average JC (and variance) was calculated for each month and site-month combination. This information provides an estimated measure of the expected total species richness (TSR_{tru}) that would be caught in that month and site-month combination. Those expected values were then compared to the total species richness actually sampled in each month and site-month combination. Lastly, the percentage of species underestimated was calculated by dividing the observed number of species by the total species richness.

2.3. Results

2.3.1. Replication Analysis

A species accumulation curve did not stabilize after 5 consecutive beach seine hauls, suggesting that more than 5 replicate hauls would be required to acquire the true number of fish species in the eelgrass beds (Figure 2.2). However, there is a limited time window to sample the eelgrass beds because of changes in tidal height. Triplicate hauls were, therefore, chosen to minimize catch variability and avoid sampling artefacts due to tidal height variation.

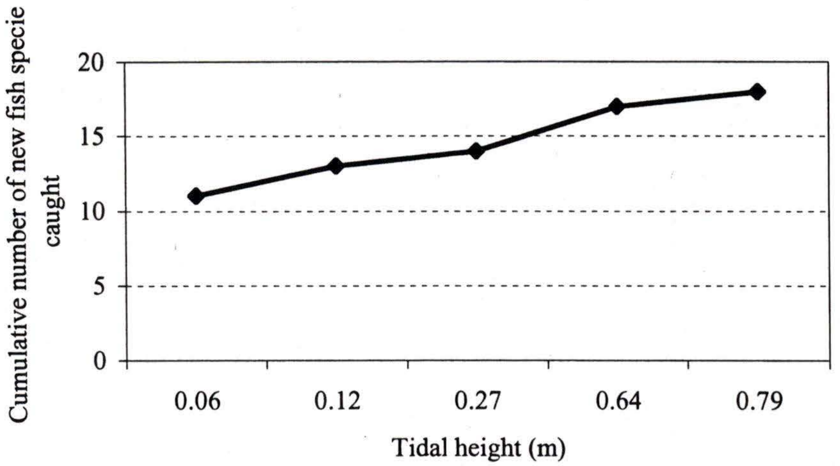

Figure 2.2. Species accumulation curve for Indian in July 2001 after 5 consecutive beach seine hauls.

2.3.2. Diel Analysis

A total of 24 fish species were beach seined in the Indian and South Bay sites in day and night samples combined, with a total of 21 species caught in the day and 18 at night (Appendix 2.1). Indian and South Bay had a suite of 11 common species caught in both in the day and night sampling events. Indian had one fish species unique to day sampling. South Bay had six species unique to day sampling. Both Indian and South Bay had only three unique species caught at night. All fish species caught at night were previously beach seined during other sampling periods in eelgrass beds: therefore, no species were caught only at night. However, observations of approximately 20 spiny dogfish (*Squalus acanthias*) between 0.5 and 1.0m in length were made at the Indian site, but not caught in the beach seine. Overall, fish species richness did not differ significantly between day and night sampling for both sites (Table 2.3).

Mean fish abundance and individual species groups abundance exhibited heterogeneity of variance: therefore, all fish abundance data was Log(x+1) transformed.

Daytime total fish abundance was significantly higher than night catches ($P>0.05$) for both sites (Table 2.3). Combined fish species group abundance data for Indian and South Bay showed only three species groups (perch, sticklebacks, rockfish/greenlings) differing significantly between the day and night (Figure 2.3). Perch and sticklebacks demonstrated significantly higher abundance in the day catches (perch, ANOVA, $F_{1,46} = 4.776$; stickleback, Kruskal-Wallis, df=1, $H = 5.026$; $P<0.05$ for both cases), whereas rockfish and greenlings demonstrated significantly higher abundance in the night catches (Kruskal-Wallis, df=1, $H = 4.555$; $P<0.05$). Mean fork length comparisons showed only the pile perch (*Rhacochilus vacca*) caught at night to be significantly larger. However, other species such as the rockfish and greenlings were consistently larger; but these differences were not significant (Table 2.4).

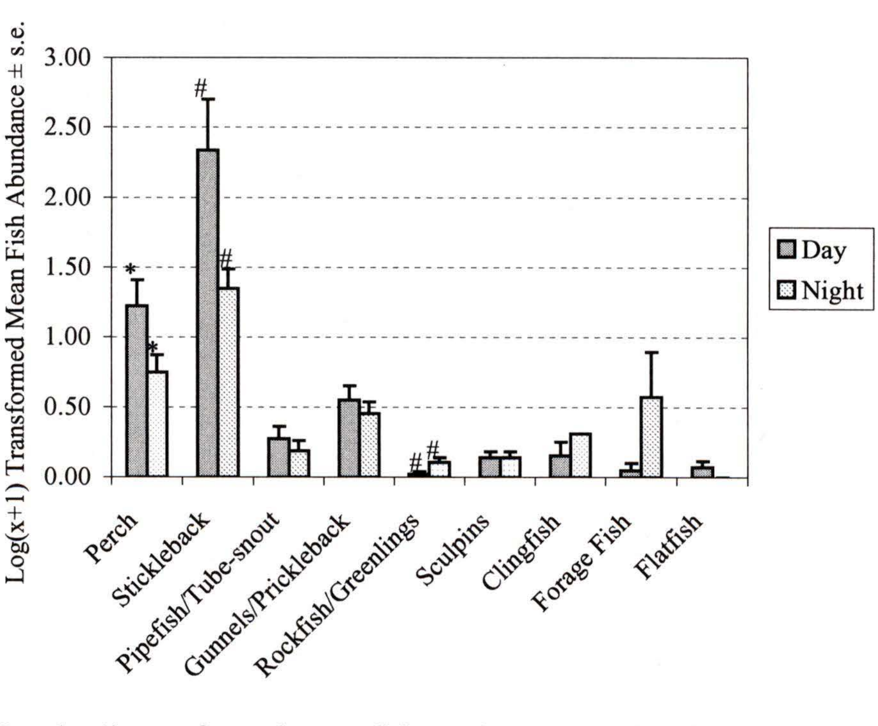

Figure 2.3. Log(x+1) transformed mean fish species groups abundance caught in day/night samples at Indian and South Bay eelgrass beds combined (ANOVA, *$P<0.05$; Kruskal-Wallis test, #$P<0.05$).

Table 2.3. ANOVA results for diel (day/night) and tidal (low/high tide) beach seine samples, the *F* tests had 1 and 4 degrees of freedom. (NS=not significant; **P*<0.05, ***P*<0.001)

Site	Sampling Comparison	Data Type	Levene's Test	ANOVA *F* statistic	Higher diurnal or tidal stage result
Indian	Day/night	Fish species richness	NS	0.469	Night (NS)
Indian	Day/night	Transformed mean fish abundance	NS	0.017*	Day*
South Bay	Day/night	Fish species richness	NS	0.548	Day (NS)
South Bay	Day/night	Transformed mean fish abundance	NS	0.04*	Day*
Indian	Low/high tide	Fish species richness	NS	0.001**	Low**
Indian	Low/high tide	Transformed mean fish abundance	NS	0.284	Low (NS)
South Bay	Low/high tide	Fish species richness	NS	0.053*	Low*
South Bay	Low/high tide	Transformed mean fish abundance	NS	0.221	Low (NS)
Masset Inlet June 5th	Low/high tide	Fish species richness	NS	0.42	Low (NS)
Masset Inlet June 5th	Low/high tide	Transformed mean fish abundance	NS	0.57	Low (NS)
Masset Inlet June 7th	Low/high tide	Fish species richness	NS	0.042*	Low*
Masset Inlet June 7th	Low/high tide	Transformed mean fish abundance	NS	0.047*	Low*

Table 2.4. Mean fork length of fish species caught during day/night sampling from both Indian and South Bay eelgrass beds with corresponding ANOVA and Kruskal-Wallis results. (NS=not significant, NC=no catch; NA=not applicable, **$P<0.001$)

		Day		Night			
Fish species	Common Name	Mean length (mm) ± sd	Sample size	Mean length (mm) ± sd	Sample size	ANOVA *F* statistic	Kruskwal-Wallis x^2 result
Cymatogaster aggregata	Shiner Perch	73.0 ± 22.6	83	68.4 ± 22.9	65	NS	NA
Micrometrus frenatus	Kelp Perch	71.6 ± 38.2	82	74.2 ± 29.6	93	NS	NA
Embiotoca lateralis	Striped Seaperch	80.3 ± 5.9	3	NC	NC	NA	NA
Rhacochilus vacca	Pile Perch	82.5 ± 30.3	43	165.1 ± 83.2	17	NA	23.789**
Gasterosteus aculeatus	Threespine Stickleback	39.5 ± 16.5	66	35.6 ± 12.1	62	NS	NA
Syngnathus leptorhynchus	Bay Pipefish	200.0 ± 31.8	14	191.7 ± 29.6	10	NS	NA
Aulorhynchus flavidus	Tube-snout	85.5 ± 2.1	2	NC	NC	NA	NA
Lumpenus sagitta	Pacific Snake Prickleback	172.7 ± 45.8	38	182.4 ± 56.6	16	NS	NA
Apodichthys flavidus	Penpoint Gunnel	148.9 ± 56.2	7	161.8 ± 62.9	15	NS	NA
Pholis laeta	Crescent Gunnel	134.7 ± 47.5	54	136.6 ± 49.6	44	NS	NA
Pholis ornata	Saddleback Gunnel	146.0 ± 25.5	2	123.0 ± 41.8	3	NS	NA
Sebastes caurinus	Copper Rockfish	47.0	1	156.8 ± 8.11	5	NA	NA
Sebastes flavidus	Yellowtail Rockfish	NC	NC	211.5 ± 29.0	2	NA	NA
Hexagrammos decagrammus	Kelp Greenling	88.0	1	105.5	1	NA	NA
Hexagrammos stelleri	Whitespotted Greenling	NC	NC	189	1	NA	NA
Blepsias cirrhosus	Silverspotted Sculpin	110.0 ± 12.7	2	NC	NC	NA	NA
Leptocottus armatus	Staghorn Sculpin	171.6 ± 39.6	20	142.3 ± 62.7	20	NS	NA
Artedius fenestralis	Padded Sculpin	106.5 ± 50.2	2	94.0 ± 38.3	4	NS	NA
Artedius lateralis	Smoothhead Sculpin	119	1	NC	NC	NA	NA
Myoxocephalus polyacanthocephalus	Great Sculpin	NC	NC	70.3 ± 7.1	3	NA	NA
Rimicola muscarum	Kelp Clingfish	42.3 ± 6.7	3	40.1 ± 3.7	10	NS	NA
Clupea pallasi	Herring	60.0	1	81.7 ± 9.7	30	NA	NA
Platichthys stellatus	Starry Flounder	63.0	1	NC	NC	NA	NA
Parophrys vetula	English Sole	142.5	2	NC	NC	NA	NA

2.3.3. Tidal Analysis

2.3.3.1. High/Low Tide Analysis

A total of 22 fish species were beach seined at the Indian and South Bay sites in the low and high tide samples combined, with a total of 20 species caught at low tide and 13 at high tide (Appendix 2.1). A common suite of seven species were caught at both low and high tide sampling at the Indian site, compared to nine common species caught at South Bay. An additional eight fish species were caught only at low tide at Indian and one unique species was caught at high tide. By comparison, at South Bay, 10 species were caught only at low tide and two at high tide. Mean fish species richness at Indian, South Bay, and Masset Inlet eelgrass beds demonstrated significantly higher species richness at low tide ($P<0.05$), with the exception of the June 7th data collected in Masset Inlet (Table 2.3).

Fish abundance data from Clayoquot Sound (Indian and South Bay), and Masset Inlet (June 5th and 7th) demonstrated heterogeneous variance; therefore, all fish abundance data was Log(x+1) transformed. No significant difference was found between low and high tide samples, with the exception of the samples taken from Masset Inlet on June 7th, which had a higher catch at low tide ($P<0.05$) (Table 2.3). However, there was an overall trend at all four sites of higher catch at low tide versus high tide. Separate comparisons of fish species group abundances at Clayoquot Sound sites, and at Masset Inlet sites showed significantly higher abundance of some species groups caught at low tide (Figures 2.4 and 2.5). In Clayoquot Sound, four species groups (pipefish/tube-snout, gunnels/prickleback, rockfish/greenling, clingfish) showed significantly higher abundance (pipefish/tube-snout, Kruskal-Wallis, df=1, $H = 10.873$; gunnels/prickleback, Kruskal-Wallis, df=1, $H = 11.822$; rockfish/greenling, Kruskal-Wallis, df=1, $H = 7.482$; $P<0.001$ for three previous cases; clingfish, Kruskal-Wallis, df=1, $H = 5.228$; $P<0.05$) versus one species group (flatfish) showing significantly higher abundance in Masset Inlet (flatfish, ANOVA, $F_{1,10} = 9.244$; $P<0.05$). Mean fork length comparisons showed the penpoint gunnel (*Apodichthys flavidus*) and staghorn sculpin (*Leptocottus armatus*) caught during high tide to be significantly larger (Table 2.5).

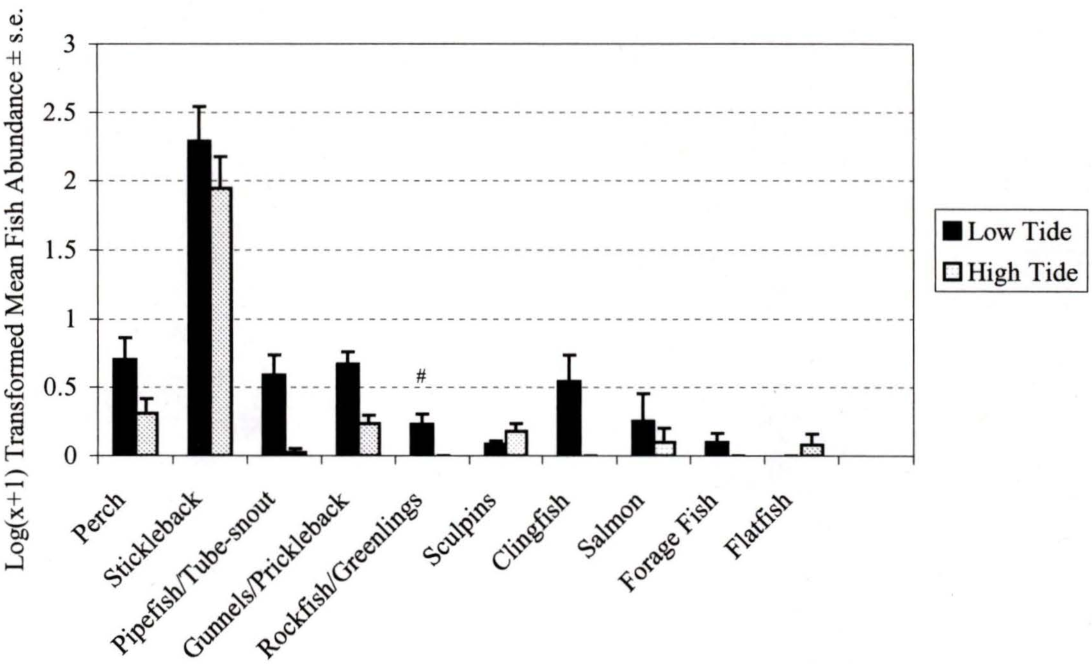

Figure 2.4. Log(x+1) transformed mean fish species groups abundance caught in low/high tide samples at Indian and South Bay eelgrass beds combined (Kruskal-Wallis test, #$P<0.05$).

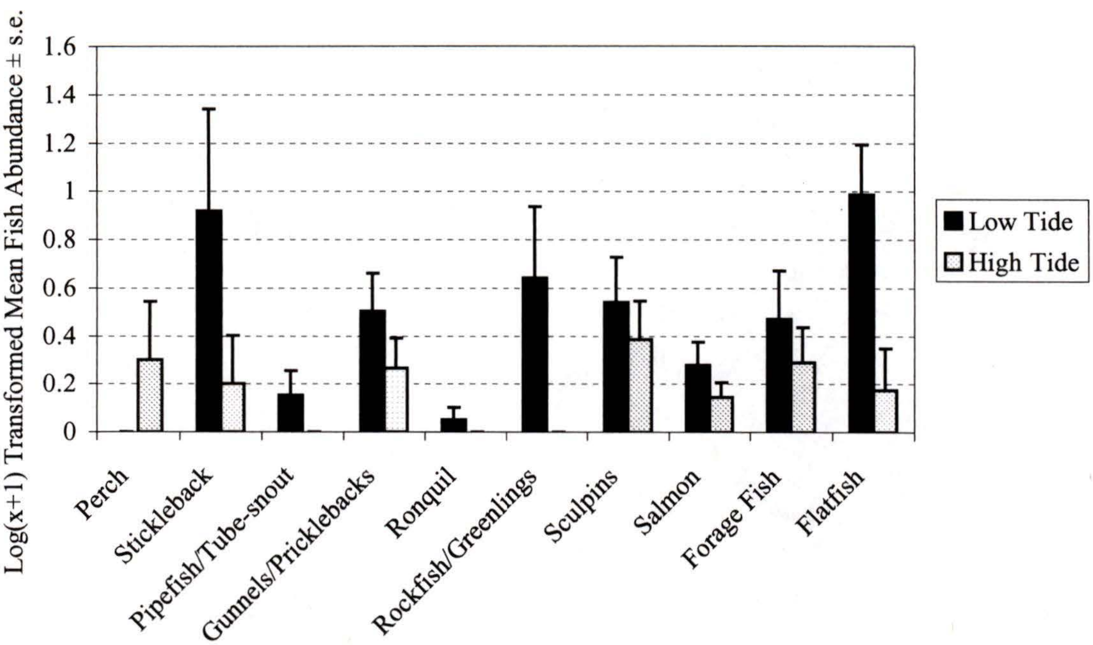

Figure 2.5. Log(x+1) transformed mean fish species groups abundance caught in low/high tide samples at Masset Inlet eelgrass beds (June 5th and 7th samples combined).

Table 2.5. Mean fork length of fish species caught during low/high tide sampling from both Indian and South Bay eelgrass beds with corresponding ANOVA results. (NS=not significant, NC=no catch; NA=not applicable, *$P<0.05$)

		Low Tide		High Tide		
Fish species	Common Name	Mean length (mm) ± sd	Sample size	Mean length (mm) ± sd	Sample size	ANOVA F statistic
Cymatogaster aggregata	Shiner Perch	89.8 ± 20.0	56	89.2 ± 15.7	31	NS
Micrometrus frenatus	Kelp Perch	89.9 ± 5.6	28	98.0 ± 16.2	4	NS
Rhacochilus vacca	Pile Perch	104	1	NC	NC	NA
Gasterosteus aculeatus	Threespine Stickleback	64.0 ± 5.8	51	65.0 ± 4.5	50	NS
Syngnathus leptorhynchus	Bay Pipefish	160.2 ± 35.1	36	165.0	1	NA
Aulorhynchus flavidus	Tube-snout	66.2 ± 26.4	17	NC	NC	NA
Lumpenus sagitta	Pacific Snake Prickleback	155. 8 ± 62.5	58	102.6 ± 69.7	5	NS
Apodichthys flavidus	Penpoint Gunnel	153.1 ± 53.6	14	218.4 ± 29.6	5	6.533*
Pholis laeta	Crescent Gunnel	152.1 ± 34.3	52	145.5 ± 33.6	15	NS
Pholis ornata	Saddleback Gunnel	124.0 ± 28.4	4	118.3 ± 35,2	4	NS
Sebastes caurinus	Copper Rockfish	145.3 ± 67.0	4	NC	NC	NA
Hexagrammos decagrammus	Kelp Greenling	68.9 ± 10.4	8	NC	NC	NA
Blepsias cirrhosus	Silverspotted Sculpin	98.0 ± 67.9	2	NC	NC	NA
Leptocottus armatus	Staghorn Sculpin	168.0 ± 42.6	6	100.5 ± 50.1	45	9.884*
Oligocottus maculosus	Tidepool sculpin	82.0	1	35.5 ± 0.7	2	NA
Clinocottus acuticeps	Sharpnose sculpin	27.0	1	NC	NC	NA
Artedius fenestralis	Padded Sculpin	89.0	1	NC	NC	NA
Myoxocephalus polyacanthocephalus	Great Sculpin	NC	NC	53.0	1	NA
Rimicola muscarum	Kelp Clingfish	40.9 ± 3.5	25	NC	NC	NA
Oncorhynchus kisutch	Coho	79.2 ± 15.4	17	71.3 ± 12.3	3	NS
Theragra chalcogramma	Walleye Pollock	52.0 ± 1.4	2	NC	NC	NA
Platichthys stellatus	Starry Flounder	NC	NC	122.5 ± 3.5	2	NA

2.3.3.2. Tidal Stage Analysis

A total of 23 fish species were caught on a rising tide sampling event at South Bay. Four tidal stages were sampled: 0.5, 0.8, 1.3 and 2.1 metres, where 13, 15, 20 and 12 fish species were caught at each respective tidal height (Appendix 2.2). Six fish species were caught at all four tidal heights: 0.5m, 7 species; 0.8m, 9 species; 1.3m, 14 species; 2.1m, 6 species. Unique species were caught at each tidal height. No significant difference between fish species richness at the four tidal stages was found, although trends show a decrease in species richness at the highest tidal height (Table 2.6). Fish species richness in Masset Inlet did not differ significantly between tidal heights, but trends showed more species caught at a rising low tide compared to that on a falling mid tidal height (Table 2.6).

Total fish abundance did not differ significantly between tidal heights in South Bay; however, a decrease in fish abundance was observed at the highest tidal height was observed (Table 2.6). In Masset Inlet, the low rising tidal stage demonstrated significantly higher fish abundance than falling mid and rising high tidal stages ($P<0.05$) (Table 2.6). In the South Bay eelgrass bed, no fish species groups differed significantly in their abundance between the four tidal heights (Figure 2.6). Additionally, no significant relationship between tidal height and the number of fish species caught set (April, $n = 40$, $r^2 = 0.0101$, $p = 0.54$; May, $n = 90$, $r^2 = 0.0035$, $p = 0.58$; June, $n = 80$, $r^2 = 0.0055$, $p = 0.51$)(Figure 2.7) was found in the Masset Inlet data.

Table 2.6. ANOVA and Kruskal-Wallis test results for tidal stage beach seine samples. For South Bay *F* tests had 3 and 4 degrees of freedom, *H* tests had 3 degrees of freedom. For Masset Inlet *F* tests had 4 and 36 degrees of freedom. (NA=not applicable, NS=not significant, **P*<0.05, ***P*<0.001)

Site	Data Type	Levene Test	ANOVA *F* statistic	Kruskal-Wallis x^2 result	Samples Compared	Tidal stage with higher value
South Bay	Fish species richness	0.000*	NA	NS	NA	1.3m (NS)
South Bay	Transformed mean fish abundance	NP	NA	NS	2.1m and 0.5m	0.5m (NS)
					2.1m and 0.8m	0.8m (NS)
					2.1m and 1.3m	1.3m (NS)
Masset Inlet	Fish species richness	NS	NS	NA	low rising and mid falling	low rising (NS)
Masset Inlet	Transformed mean fish abundance	NS	4.005*	NA	low rising and mid falling	low rising*
					low rising and high rising	low rising*

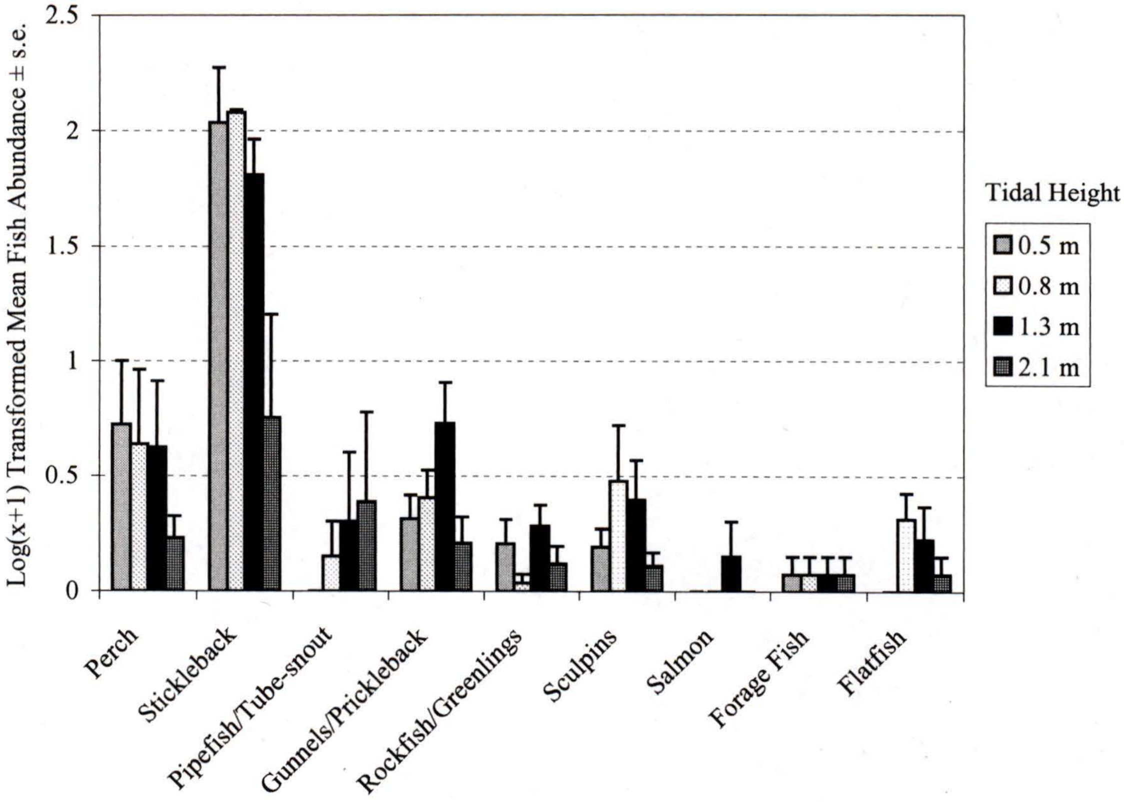

Figure 2.6. Log(x+1) transformed mean fish species groups abundance caught at four tidal heights at South Bay.

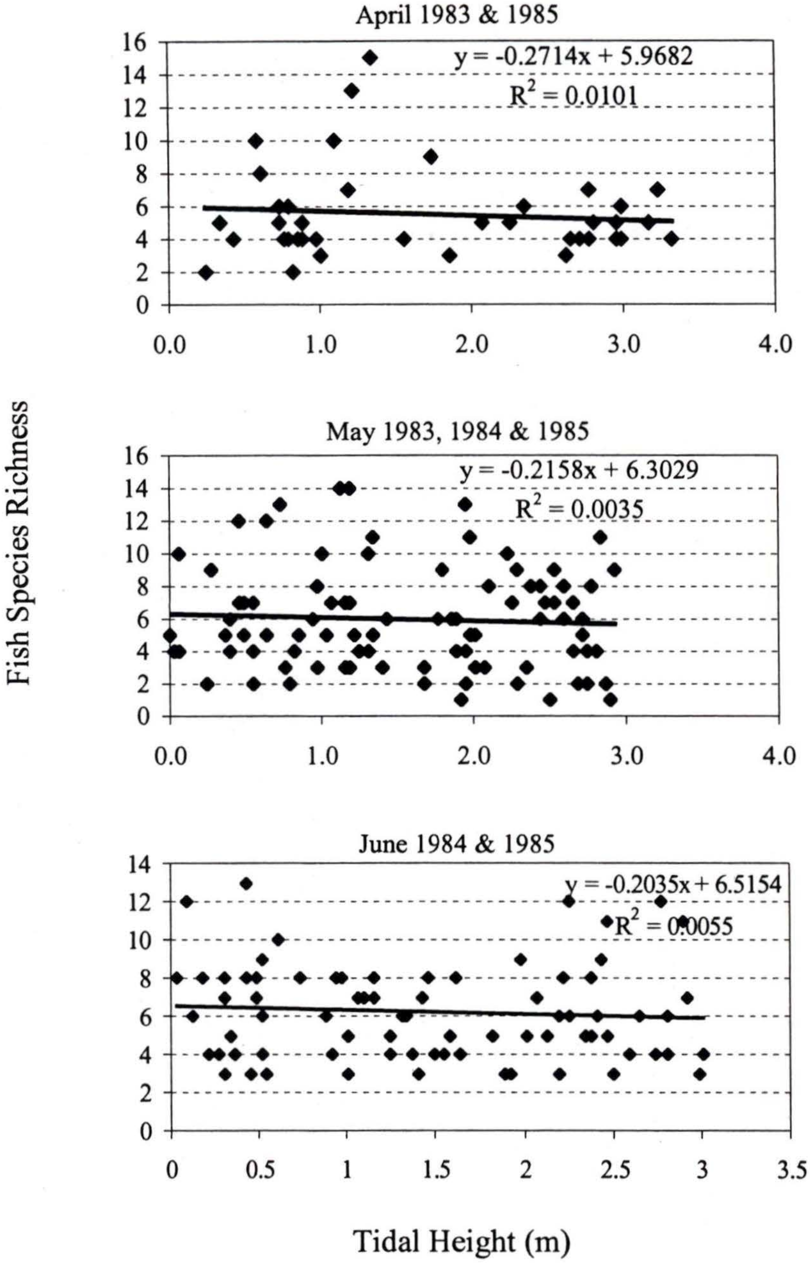

Figure 2.7. Fish species richness versus tidal height regressions for April, May and June in 1983, 1984 and 1985 in Masset Inlet eelgrass beds.

2.3.4. Hauling Effect Analysis

A graphical comparison of fish species richness and number of hauls did not demonstrate any relationship between the number of species caught and number of times an area had been seined (Figure 2.8). In addition, mean fish abundance in each set did not vary according to the number of times the area was sampled (Figure 2.9). Therefore, we are confident that the repetitive beach seine hauling does not impact the fish assemblage catches in this study.

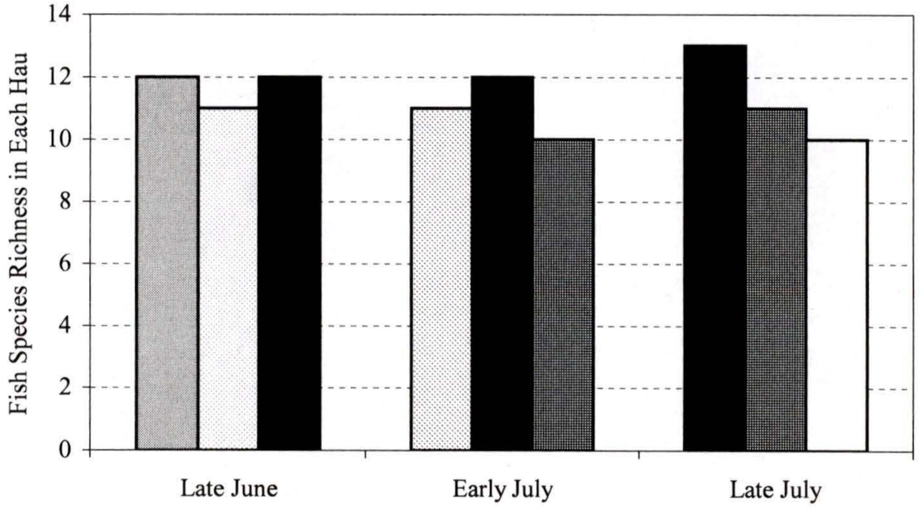

Figure 2.8. Fish species richness in each haul at Indian, from late June to late July, where the bars with the same patterns represent the same area sampled previously.

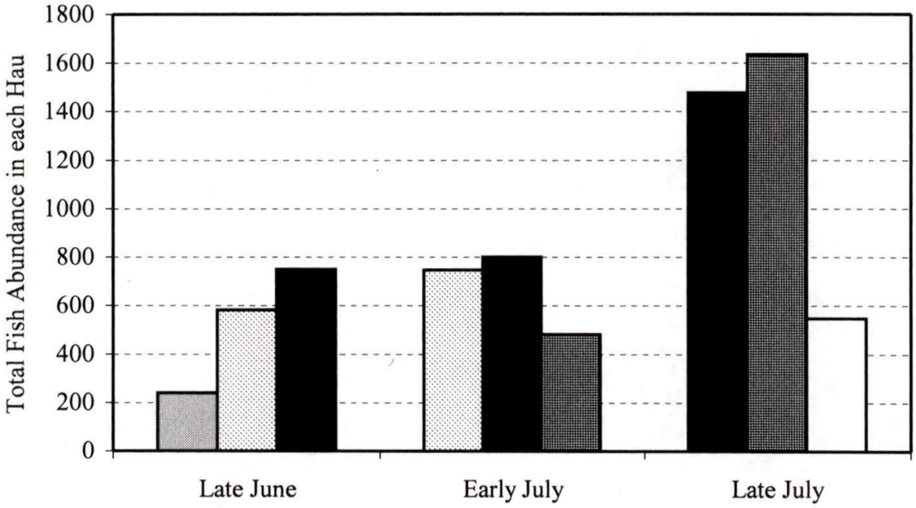

Figure 2.9. Total fish abundance in each haul at Indian, from late June to late July, where the bars with the same patterns represent the same area sampled previously.

2.3.5. Sampling Sufficiency

The effectiveness of beach seining as a sampling method to estimate the number of fish species data was explored from 277 beach seine sets at seven eelgrass beds in Clayoquot Sound during 2001 and 2002. A list of all fish species caught was compiled for each site-month combination and used as the representative fish species list for sampling sufficiency calculation. On a monthly comparison, the average of 100 simulated autosimilarity measures (Jaccard coefficient) was calculated, and showed a 1 to 7% underestimation of fish species in each month, where June 2002 had the lowest underestimation and the highest was in June 2001 (Figure 2.10) A second analysis explored species richness on a site-month basis in 2002, indicating a 1 to 11% underestimation of fish species in each bed monthly, where Ducking in April had the lowest underestimation and Kootowis in April had the highest (Figures 2.11 and 2.12) These results indicate that the beach seining sampling methodology used in this study sampled at least 90% of the fish diversity in Clayoquot Sound eelgrass beds from the representative fish species list.

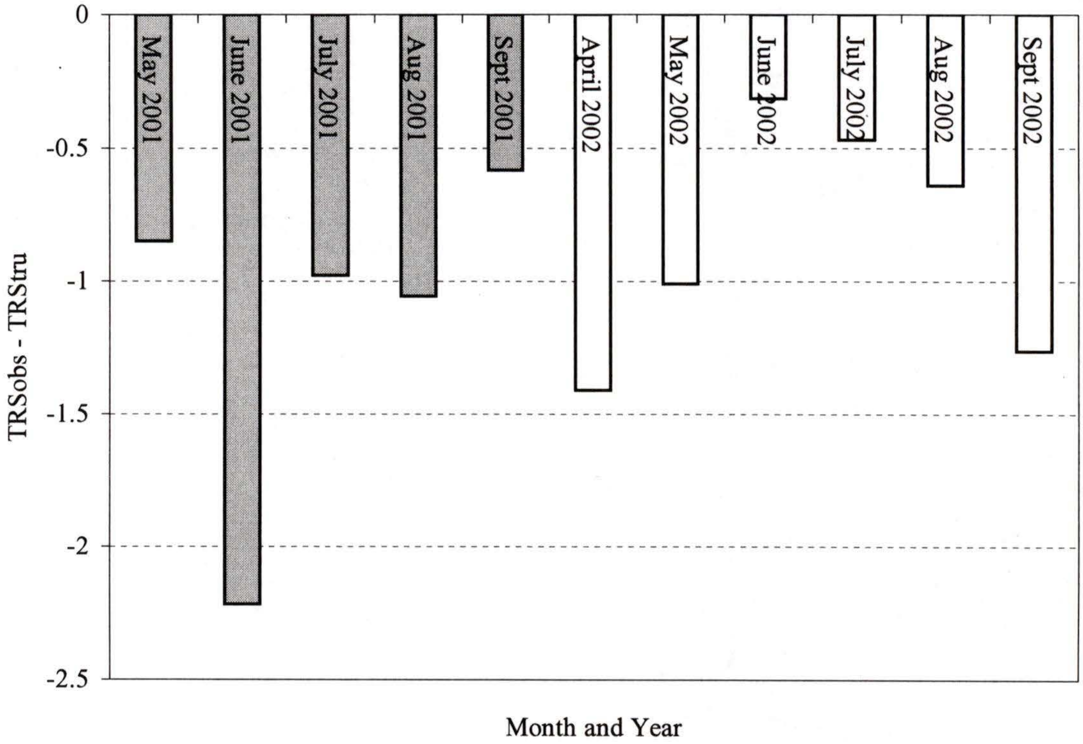

Figure 2.10. Jaccard coefficient calculated for each month in seven eelgrass beds sampled in Clayoquot Sound in 2001 and 2002.

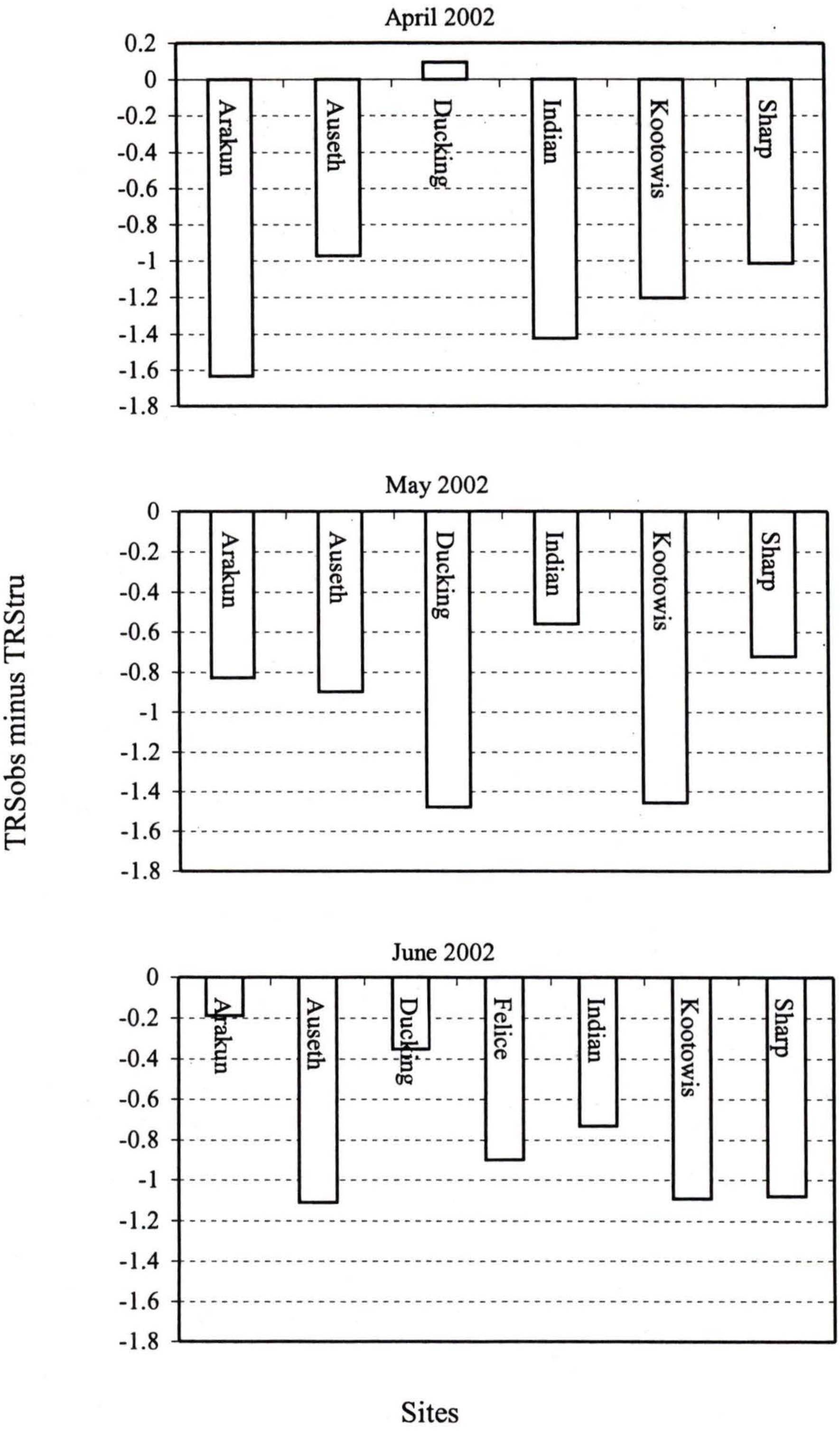

Figure 2.11. Jaccard coefficient calculated for each month in seven eelgrass beds in Clayoquot Sound April to June 2002.

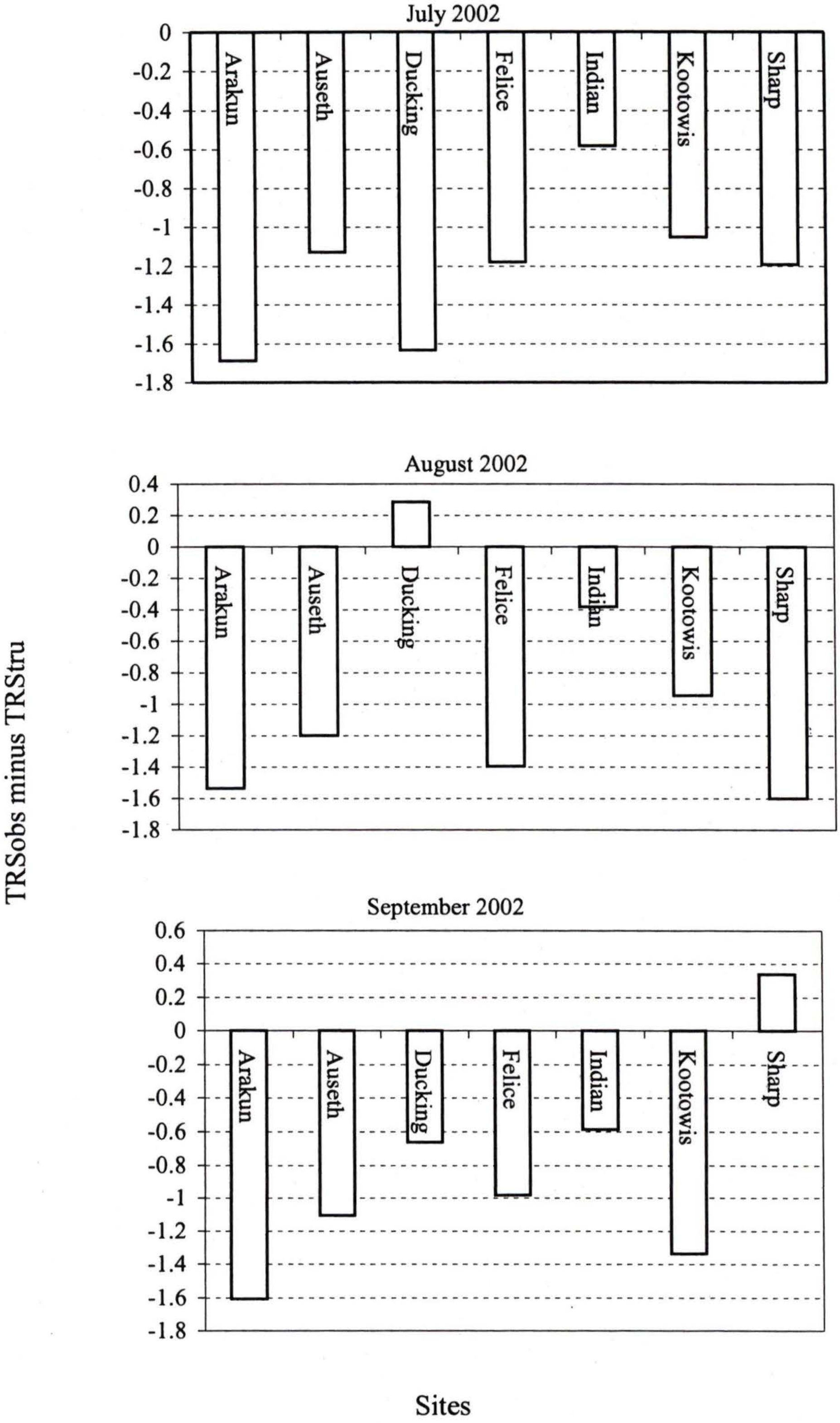

Figure 2.12. Jaccard coefficient calculated for each month in seven eelgrass beds in Clayoquot Sound from July to September 2002.

Table 2.7. Summary of sampling results.

Topic	Conclusions
Replication	Species accumulation curve does not stabilize after 5 replicate hauls
Diel Variation (Day/night)	Unique species: no consistent trends
	Species richness: no consistent trends
	Abundance: significantly more fish caught in the day
	Perch: significantly more fish caught in the day
	Sticklebacks: significantly more fish caught in the day
	Rockfish/Greenlings: significantly more fish caught at night
	Pile perch significantly greater lengths at night
Tidal Variation (Low/high)	Unique species: 5 times more unique fish species caught at low tide
	Species richness: significantly more fish species caught at low tide
	Abundance: trend indicates more fish caught at low tide but only Masset Inlet sampled on June 7th was significant
	Rockfish/Greenling: significantly more fish caught at low tide
	Penpoint gunnel: significantly greater length at high tide
	Staghorn sculpin: significantly greater length at high tide
Tidal Stage	Unique species: highest number of unique fish species caught on a rising 1.3 m tide
	Species richness: no consistent trends in Clayoquot, significantly more species caught on rising low tide than falling mid tide in Masset
	Abundance: trends show fewer fish caught at 2.1 m tide than all other tidal heights in Clayoquot, significantly more fish caught on rising low tide than rising high and falling mid tides in Masset
	Fish groups: no significant differences
	Regression: no relationship found between the number of fish species caught and tidal height in Masset
Hauling Effects	Species richness: no consistent trends
	Abundance: no consistent trends
Sampling Sufficiency	Monthly comparison (2001/2002): 1 to 7% underestimation of fish species richness, June 2002 had the lowest underestimation, June 2001 had the highest underestimation;
	Site-month comparison (2002): 1 to 11% underestimation of fish species richness, Ducking in April had the lowest underestimation, Kootowis in April had the highest underestimation

2.4. Discussion

2.4.1. Previous Research

Biological diversity can be managed and conserved through the development of monitoring programs. One approach to the assessment of biological diversity is an accurate assessment of the species diversity present in the ecosystem (Yoccoz et al .2001). In this study, I evaluated the effectiveness of beach seining as a sampling methodology to address fish diversity in *Zostera marina* beds in Clayoquot Sound (Table 2.7). Previous studies have evaluated beach seining and the effect diel and tidal variation has on the fish diversity in the catch (Gibson et al. 1996, Mattila et al. 1999, Morrison et al. 2001). However, no study has compared research results to determine whether diel and tidal variation effects are consistent across studies. I compared the results of 18 studies, with respect to beach seine catches in relation to diel and tidal variation (Table 2.1). Sixteen studies examined diel effects and 14 of these studies caught a more species at night, with two studies showing no diel effect. Fish abundance results were inconsistent between studies, six having no significant differences, seven being significantly higher at night, and three significantly higher in the day. These results highlight that diel fish diversity in nearshore habitats may vary depending on geographic and habitat variability (Rozas and Minello 1997). The potential for diel fish diversity to vary emphasizes the importance of diel sampling as a crucial component of estuarine fish assemblage assessment (Griffiths 2001). Additionally, the inconsistent fish abundance may be related to variable beach seine size and methodology between studies. Allen et al. (1992) found that beach seine catches have high variability between hauls, and beach seine length can also influence catches (Vanderklift et al. 1998). In comparison, the low and high tide sampling results were consistent between all studies, showing significantly higher species richness and abundance for the low tide catches. This indicates that sampling at low tide is more effective to quantify nearshore fish diversity than sampling at high tide.

2.4.2. Diel and High/Low Tidal Variation

The effect of diel variation on fish diversity was inconsistent with other studies, but the effect of tidal variation was consistent with other studies (Table 2.1). The results of this study were limited as I only sampled on two occasions. Further samples would be required to acquire a more robust assessment of the effect of diel and tidal variation. Results showed fish species richness and abundance higher at low tide. Similar results have been found in other nearshore habitats. At each site, up to five times more fish species were caught during low tide sampling compared to high tide. Therefore, low tide sampling is essential to achieve an accurate description of fish diversity in Clayoquot Sound eelgrass beds. Additionally, penpoint gunnel and staghorn sculpin mean lengths were greater at high tide, suggesting an increase in predation pressure at high tide in higher intertidal areas of eelgrass beds. Armstrong et al. (1994) found the major food source of 1 + and older staghorn sculpins to be the ghost shrimp (*Neotrypaea californiensis*). Larger sculpins caught at high tide may be foraging in adjacent high tidal mudflat areas, ghost shrimp habitat. Consequently, it is importance to assess fish diversity at a species level to understand the role of predation in nearshore habitats.

Fish species richness was not significantly different between day and night catches; however, abundance was lower at night, an opposite trend to that found in other studies. Lower abundance was due to the daytime catch of schools of perch and sticklebacks, and their numbers decreasing at night (Figure 2.3). The decreasing number of perch and stickleback indicated that these fish species were primarily diurnally active in eelgrass beds (Hobson et al. 1981), and may move out of eelgrass at night. In addition, the number of new species caught at night were similar to those caught at in the day, suggesting that the fish communities in eelgrass beds are similar in the day and night in Clayoquot Sound. However, the abundance of large predatory fish such as adult rockfish and kelp greenlings did increase at night (Figure 2.3, Table 2.3). Spiny dogfish were also observed in the Indian eelgrass beds at night though they were not caught during sampling. Therefore, the increase in larger predators at night may be driving the low fish abundance nocturnally. In comparison, Hindell et al. (2000) found an increase in smaller fish abundance at night in shallow areas, suggesting avoidance of migrating nocturnal predators in shallow areas. In this study, the diel movement of the dominant species (perch and sticklebacks) appears to be driving the decrease in abundance at night. The nocturnal predator avoidance behaviour of these species may utilize different habitat than shallow areas in the eelgrass bed. Overall, it is important to examine differences in fish diversity at a species level because diel utilization of eelgrass beds was not consistent among fish species.

2.4.3. Tidal Stage, Hauling Effects, Replication, and Sampling Sufficiency

In addition to incorporating diel and high/low tide sampling, this study also examined the effect of tidal stage, hauling effects, set replication and sampling sufficiency to evaluate beach seining. Such a combination of aspects, not been previously examined by other studies, allows a more complete evaluation of the effectiveness of my sampling methodology in assessing eelgrass fish diversity. Tidal stages influenced fish diversity, such that a peak in fish species richness and abundance occurred during a rising low tide (Table 2.6). Peak fish activity levels have previously been found to occur on falling and rising low tidal phases to avoid predators and being stranded (Sogard et al. 1989a). In this study, peak fish species richness and catches occurred at low rising tidal heights of 0m to1.2m (Table 2.6). In a similar activity pattern, as fish move back into the beds with the tide, the catch increases. However, no correlation between tidal height and fish species richness was found in the Masset Inlet data (Figure 2.7). Such lack of correlation may be due to the objectives of the Hargreaves et al. (1987) study, which was to assess salmonid habitat. Sampling at high tide for instance, will catch a high diversity of salmonid species, but may no longer be sampling the eelgrass habitat. Rather sampling would occur in mud and shell habitat found in high intertidal areas adjacent to eelgrass beds, given that *Zostera marina* is restricted to mid and low intertidal and shallow subtidal regions due to its limited desiccation tolerance and subtidal light requirements (Koch and Beer 1996, Krause-Jensen et al. 2000). Therefore, the beach seines conducted at 2.1 metres and higher may no longer be sampling *Zostera* beds, and a species to tidal height correlation for the Masset data set may not be valid because of the variability of the habitat sampled. In addition, the number of unidentified species was high in the Masset data; consequently, low species richness at lower tidal heights may be an artefact of lack of identification.

Hauling effects due to bimonthly beach seining activities did not appear to affect fish diversity caught in eelgrass beds (Figures 2.8 and 2.9) and may be due to two causes.

First, in the summer months, *Zostera marina*, exhibits peak biomass levels (Davidson and Hughes 1998). Therefore, the hauling effect may be negligible in June and July because eelgrass can readily recover on a bimonthly sampling basis. Second, the Indian *Zostera* bed sampled covered an extensive area, approximately 0.4km^2, indicating that large beds may be able to recover more readily than smaller beds. My study results suggest that bimonthly beach seining during the summer months, in sufficiently large eelgrass beds in Clayoquot Sound provides accurate information on fish diversity without being affected by previous hauling activities.

Previous studies have indicated that single or two consecutive beach seine sets will provide a reliable estimation of species richness (Allen et al. 1992, Robards et al. 1999). Contrastingly, in this study, I found that, after five consecutive beach seine hauls, the species accumulation curve did not stabilize (Figure 2.2), indicating that more than five replicates would be required to accurately estimate fish species richness. However, the limited sampling tidal window, two hours before and after spring and neap low tides, over five replicates would be impossible without encountering tidal changes affecting results. Additionally, high replication within an eelgrass bed over a short time period may not allow the eelgrass to recover from repetitive hauling effects (Gibson et al. 1996). Therefore, to minimize variability and hauling impacts we chose triplicate sets at each site, which was the maximum number of sets allowable in the short tidal window available.

I also evaluated the sampling sufficiency of beach seining by utilizing the comparison method developed by Cao et al. (2001) that examines the total species richness and the similarity among replicate samples. Findings indicate at least 90% of eelgrass fish diversity in Clayoquot Sound was sampled, which demonstrates that triplicate monthly beach seines in each site provided a reliable estimation of species richness.

2.4.4. Sampling Gear Limitations

A beach seine was used in this study because of the ease of use and cost-effectiveness, both important components of effective monitoring programs (Edgar et al. 2001). Beach seine sampling can accurately estimate fish species richness and dominance (Allen et al. 1992, Robards et al. 1999); however, this study indicated that 10% of fish diversity was not sampled. Additionally, beach seines have low catch efficiency and highly variable fish abundance catches between sets (Rozas and Minello 1997). Therefore, the use of different sampling gear is important to increase catch efficiency (Hindell et al. 2000). Gibson et al. (1996) found a strong positive relationship in sandy habitats between species richness and depth, which suggests that a trawl could be used to sample the deeper edges of eelgrass beds, inaccessible to a beach seine, to determine if a similar correlation is present in eelgrass beds. Other studies have suggested using scuba diving visual census to examine fish diversity to assess the fish species that may avoid sampling gear (Harmelin-Vivien and Francour 1992). However, in the summer, eelgrass beds in Clayoquot Sound have extremely low visibility, less than one metre at times. The additional costs and special training required was not possible for this project and many other potential monitoring programs. Finally, large predators in this study, such as the dogfish shark, were able to avoid beach seine capture. Further sampling using different sampling gear such as trawls and gillnets, may capture these larger species and provide important ecological data on the role of eelgrass beds as foraging habitats (Hindell et al. 2000).

2.4.5. Sampling Regime Recommendations

The results of this study expanded the knowledge of assessing eelgrass fish diversity, by comparing results with other studies and incorporating new aspects, such as sampling sufficiency, tidal stage sampling and fish species length and abundance comparisons, to evaluate the effectiveness of beach seining as a sampling technique. Recommendations to ensure an accurate assessment of eelgrass fish communities should include:

- Diel sampling to determine how community structure changes as larger predatory species may move into the beds at night. In addition, to accurately assess diel changes, beds should be sampled using trawls and gillnets to catch more mobile species (Edgar et al. 2001).
- Sampling should occur during a rising tidal phase because the highest number of fish species and abundance was caught during this tidal phase.
- Using triplicate beach seine sets to sufficiently sample fish species richness because this number of replicates only underestimated 10% of fish species.
- Repetitive hauling during summer months in large eelgrass beds because it did not affect fish diversity and is necessary to address temporal shift in eelgrass beds.
- Assessing individual fish species abundances and lengths to determine how eelgrass beds are utilized differently as nursery and foraging habitat by different fish species.

- Use different sampling gear, such as enclosure nets, and compare these results with beach seine catches to evaluate beach seine catch efficiency.

2.4.6. Conclusion

In coastal marine ecosystems, assessment of fish diversity has been used to address ecosystem integrity, an important issue for agencies such as Parks Canada, whose mandate includes conservation of marine ecosystems and species within national park boundaries. Beach seining was identified as an effective method to sample fish diversity that was cost-effective, caught a broad range of fish species, was easily deployed and proved repeatable within a short time window. The effectiveness of a beach seine was evaluated by examining the potential effects of diel and tidal variation, hauling effects and sampling sufficiency to sample eelgrass fish diversity in southern Clayoquot Sound. Overall, diel catches demonstrated significantly higher catches in the day, but no significant difference in species richness. However, large predatory species, including *Squalus acanthias*, *Sebastes caurinus*, *Sebastes flavidus*, *Hexagrammos decagrammus*, and *Hexagrammos stelleri*, occurred in night samples only indicating a diel change in community structure. Tidally, species richness and abundance was highest during a low, rising tidal stage. Repetitive seine hauling did not affect the subsequent fish diversity caught. Sampling sufficiency evaluation indicated triplicate seines underestimated only 10% of the fish species. Sampling regime recommendations for eelgrass fish diversity includes diel samples to address night time shifts in community structure, seining at low tide to provide a wider scope of fish diversity, and triplicate seines to sufficiently sample species richness.

Chapter 3: Spatial and Temporal Analysis of Eelgrass Fish Diversity

3.1. Introduction

Coastal marine biodiversity is under threat from the increasing concentration of human populations and the consequential intensive activities in those regions (Gray 1997). Marine reserves are tools to conserve biodiversity, but it is essential that those reserves effectively protect biodiversity from human impacts and contain a regional representation of that biodiversity (Margules and Pressey 2000). However, to effectively conserve coastal marine biodiversity, it is critical to understand the multidimensional scale of biodiversity (Purvis and Hector 2000). In addition, the establishment of marine reserves requires determination of the temporal and spatial heterogeneity that influence biodiversity within marine ecosystems (Garcia-Charton and Perez-Ruzafa 1999). Therefore, monitoring and measuring spatial and temporal changes in biodiversity is critical to marine conservation planning (Yoccoz et al. 2001, Gray 2000). However, limited knowledge exists regarding how to measure and monitor biodiversity in the marine environment (Gray 1997).

Biodiversity can be measured at a range of levels, from genes, to species, to ecosystems (Harper and Hawksworth 1994). The species level has been an integral and common level at which to address biodiversity conservation (Margurran 1988). However, the multifaceted nature of biodiversity requires measuring three major components of biodiversity: the number of species (richness), the distribution of abundance among species (evenness), and the taxonomic relatedness of the species (taxonomic distinctness/diversity) (Purvis and Hector 2000).

Many studies have limited the measurement of biodiversity to the number of species in an area, or species richness (Reid 1998). An overview of 38 nearshore fish community studies demonstrated that over half of these studies examined fish diversity by only evaluating species richness and abundance (Table 3.1). Additionally, only two studies (Hall and Greenstreet 1998, Rogers et al. 1999) applied measures of taxonomic relatedness. Lastly, no study examined fish species richness, evenness and taxonomic relatedness in conjunction with multivariate community measures to evaluate fish diversity.

Species richness, evenness and taxonomic diversity/distinctness can also be measured at spatial scales ranging from species diversity within a sample or habitat (alpha diversity) to species diversity across a seascape (gamma diversity) (Whittaker 1960). Another biodiversity aspect, turnover or beta diversity, has been neglected in the marine environment, and is defined as species composition changing across a gradient (Whittaker 1960, Clarke and Lidgard 1999). Beta diversity is not a scale of diversity but is related to the types of species contained in two or more habitats (Gray 2000). Izsak and Price (2001) developed a taxonomic similarity index to measure beta diversity by comparing species presence/absence data at different taxonomic levels (species, genera, families, orders, etc.) between sites. Such an index provides a wider scope of beta diversity than previous research by examining taxonomic diversity beyond the species level.

In conservation planning, reduction of biodiversity to one measurement, such as species richness is attractive. It is, however, unrealistic due to the multifaceted nature of biodiversity (Margules and Pressey 2000, Purvis and Hector 2000). Few studies have explored the relationship between different biodiversity measures and the insights they provide for understanding ecosystems (Price 2002, Lande 1996). This chapter examines the spatial and temporal changes in eelgrass (*Zostera marina*) fish diversity by utilizing a range of diversity measures, including richness, evenness and taxonomic diversity/distinctness. In addition to examining these alpha diversity measures I examined beta diversity across spatial and temporal gradients. By examining both alpha and beta diversity measures, the heterogeneity of eelgrass fish diversity can be determined (Wilson and Shmida 1984) and applied to managing and monitoring eelgrass ecosystems.

Table 3.1. Summary of previous work that examined nearshore fish diversity with the univariate and multivariate analysis applied in each study.
(Abbreviation: NA=not applicable to study)

Source	Nearshore Habitat	Univariate Diversity and Community Measures	Multivariate Community Analysis
Orth and Heck 1980	Seagrass and adjacent unvegetated areas	Species richness Fish abundance	NA
Sogard et al. 1987	Seagrass	Species richness Fish abundance	NA
Nash 1988	Fjord	Species richness Fish abundance Dominance Shannon-Wiener index Pielou's evenness	NA
a et al. 1989	Seagrass	Species richness Fish abundance	NA
Lubbers et al. 1990	Seagrass and adjacent unvegetated areas	Species richness Fish abundance Fish biomass	NA
Ferrell and Bell 1991	Seagrass and adjacent unvegetated areas	Species richness Fish abundance	Cluster analysis
Bell et al. 1992	Seagrass	Species richness Fish abundance	Cluster analysis Principle components analysis
Gibson et al. 1993	Sandy beach	Species richness Fish abundance Jaccard's coefficient	Two-way indicator species analysis Detrended correspondence analysis
Bell et al. 1994	Seagrass	Species richness Fish abundance	Cluster analysis

Table 3.1. Continued

Source	Habitat	Univariate Diversity and Community Measures	Multivariate Community Analysis
Edgar and Shaw 1995a	Seagrass and adjacent unvegetated areas	Species richness Fish abundance Fish biomass	NA
Santos and Nash 1995	Sandy bay	Species richness Fish abundance Fish biomass Shannon-Wiener index Pielou's evenness	NA
Gibson et al. 1996	Sandy exposed beach	Species richness Fish abundance Fish biomass Dominance	NA
Clark 1997	Sandy exposed beach	Species richness Fish abundance Dominance Shannon-Wiener index	Cluster analysis Non-metric MDS scaling ordination
Gray et al. 1998	Seagrass and adjacent unvegetated areas	Species richness Fish abundance	Non-metric MDS scaling ordination
Jenkins et al. 1997	Seagrass and adjacent unvegetated areas	Species richness Fish abundance Fish biomass	Cluster analysis
Young et al. 1997	Unvegetated estuarine areas	Species richness Fish abundance Fish biomass	Cluster analysis Non-metric MDS scaling ordination Analysis of similarity (ANOSIM)

Table 3.1. Continued

Source	Habitat	Univariate Diversity and Community Measures	Multivariate Community Analysis
Hall and Greenstreet 1998	Demersal trawl surveys	Hill's N1 (exponential of the Shannon-Wiener index) Hill's N2 (reciprocal of Simpson's index) Taxonomic diversity Taxonomic distinctness	NA
Jenkins and Wheatley 1998	Seagrass, reef-algal, and adjacent unvegetated areas	Species richness Fish abundance	Non-metric MDS scaling ordination
Marshall and Elliott 1998	Unvegetated estuarine areas	Fish abundance Dominance	Two-way indicator species analysis Canonical correspondence analysis Principle components analysis
Nash and Santos 1998	Sandy bay	Species richness Fish abundance Fish biomass Dominance	NA
Arrivillaga and Baltz 1999	Seagrass and adjacent unvegetated areas	Fish abundance Shannon-Wiener index Margalef's diversity Pielou's evenness	NA
Jackson and Jones 1999	Unvegetated estuarine areas	Species richness Fish abundance Dominance	Cluster analysis Non-metric MDS scaling ordination Analysis of similarity
Lekve et al. 1999	Fjords	Species richness Fish abundance Shannon-Wiener index	Principle components analysis

Table 3.1. Continued

Source	**Habitat**	**Univariate Diversity and Community Measures**	**Multivariate Community Analysis**
Mattila et al. 1999	Seagrass and adjacent unvegetated areas	Fish abundance Shannon-Wiener index	NA
Robards et al. 1999	Estuarine mud flats	Species richness Fish abundance Pielou's evenness Margalef's index Shannon-Wiener index	NA
Rogers et al. 1999	Bottom habitat trawled off coastal waters	Taxonomic diversity Taxonomic distinctness	NA
Wagner and Austin 1999	Unvegetated estuarine areas	Fish abundance	Two-way indicator species analysis Detrended correspondence analysis Detrended canonical correspondence analysis
Abookire et al. 2000	Unvegetated estuarine areas	Species richness Fish abundance Shannon-Wiener index	NA
Dean et al. 2000	Seagrass and kelp	Fish abundance	Cluster analysis Non-metric MDS scaling ordination
Layman 2000	Sandy exposed beach	Species richness Fish abundance Simpson's diversity index Shannon-Wiener index Pielou's evenness	Non-metric MDS scaling ordination

Table 3.1. Continued

Source	**Habitat**	**Univariate Diversity and Community Measures**	**Multivariate Community Analysis**
Guidetti 2000	Seagrass, rocky-algal reefs, and adjacent unvegetated areas	Species richness Fish abundance	Cluster analysis
Rozas and Zimmerman 2000	Vegetated and nonvegetated marsh areas	Species richness Fish abundance	Canonical analysis
Griffiths et al. 2001	Seagrass and adjacent unvegetated areas	Species richness Fish abundance	Cluster analysis Analysis of similarity (ANOSIM)
Methven et al. 2001	Small rock and gravel	Species richness Fish abundance	Cluster analysis
Desmond et al. 2002	Unvegetated estuarine areas	Species richness Fish abundance Shannon-Wiener index	Non-metric MDS scaling ordination
Hovel et al. 2002	Seagrass	Species richness Fish abundance	Principle components analysis
Morrison et al. 2002	Mud and sand intertidal flats	Species richness Fish abundance	Cluster analysis Correspondence analysis
Suda et al. 2002	Sandy exposed beach	Species richness Fish abundance Fish biomass Index of diversity Shannon-Wiener index Smith & Wilson's evenness measure	NA
Yakimishyn 2003, this study	Seagrass	Species richness Fish abundance Pielou's evenness Taxonomic diversity Taxonomic distinctness Taxonomic similarity	Cluster analysis Non-metric MDS scaling ordination Analysis of similarity (ANOSIM)

3.2. Methods

3.2.1. Study Location

The study area is located in Clayoquot Sound, on the west coast of Vancouver Island, and consists of seven eelgrass beds, dominated by *Zostera marina*. Eelgrass beds were selected according to the estimated hydrodynamic energy these beds receive from waves, wind and tidal currents (see chapter 4). The eelgrass beds studied include three within Pacific Rim National Park Reserve of Canada in Grice Bay, Auseth, Indian and Kootowis, one in Browning Passage, Ducking, and two in Lemmens Inlet, Sharp and Arakun and one in Van Nevel Channel, Felice (chapter 2, Figure 2.1). The characteristics of each bed are summarised in chapter 2 (Table 2.2).

3.2.2. Sampling Method

Each site was sampled using triplicate beach seine sets (see chapter 2, section 2.2.2), in the 2 hour window before and after a daytime mean low of 0.6m or less, from early spring to early fall, monthly in 2001, and bimonthly in 2002 (Appendix 3.1). However, there was greater variability in sampling times in 2001, as not all sites were selected, and storms in August prevented sampling at some sites. Additionally, Felice was only sampled once in August 2001, and bimonthly from June to September in 2002.

3.2.3. Univariate Analysis

3.2.3.1. Diversity Measures

Fish diversity was compared both between months and spatially between the seven sites using species richness, abundance and Pielou's evenness index. All diversity measures were analysed separately for each year (2001 and 2002) and sampling period. The indices used were compared between months and between sites, and are defined as follows:

- Species richness (s) = the total number of species
- Number of individuals (N) = abundance
- Pielou's evenness index (J') = how evenly individuals are distributed among different species (Appendix 3.2)

Each index was examined graphically, in line plots, to compare trends between sites and months. All diversity measures were tested for and failed homogeneity of variance using Levene's test (Zar 1999). Therefore, abundance data was Log(1+x) transformed, which put the data on a common scale and accounted for zeros found in species data (Clarke and Green 1988). Although the transformed fish abundance data continued to fail equal variance, it was kept in the transformed format, because the non-parametric Kruskal-Wallis test is still slightly sensitive to unequal variance (Zar 1999). Species richness, abundance, and Pielou's evenness index were analyzed using a nonparametric ANOVA, the Kruskal-Wallis test. Though 5% less powerful than a parametric ANOVA, the Kruskal-Wallis test is less sensitive to heterogeneous variances (Zar 1999). If significant differences were found between each site and/or month, pair-wise comparisons, using the Mann-Whitney test, were used to determine which sites and months were significantly different (Sokal and Rohlf 1995). Since multiple, pair-wise comparisons increase a type I error, rejecting a true null hypothesis, a Bonferroni correction was applied to the data by dividing the significance level of 0.05 by the number of comparisons, thereby decreasing the significance level and reducing the chance of a type I error (Sokal and Rohlf 1995). All indices were calculated using the statistical package PRIMER version 5.2.8, and subsequent statistical tests were carried out using the SPSS version 10.0 statistical package.

3.2.3.2. Taxonomic Diversity and Distinctness

Warwick and Clarke (1995) introduce the concept of taxonomic diversity (Δ, Δ) that not only takes into account species richness and abundance, but also incorporates the degree to which species in an assemblage are related to each other. Taxonomic diversity is the average taxonomic relatedness or distance between any two randomly drawn individuals (Appendix 3.2).

A second index, taxonomic distinctness (Δ^*), measures the relatedness of individuals, by taking Δ and removing its dependence on species abundance distribution (Appendix 3.2.). Thereby, the average taxonomic path length between two randomly chosen individuals from two different species is taxonomic distinctness (Clarke and Warwick 1998).

In this study, a regional species list was compiled for fish caught in *Zostera marina* beds in Clayoquot Sound (Appendix 3.3). All listed fish species were than taxonomically classified into appropriate hierarchical levels according to Nelson (1994): species, genus, family and order. Each step length between each successive taxonomic level was weighted equally (weighting=1), as Clarke and Warwick (1999) found Δ and Δ^* robust to any changes in path length weighting. Taxonomic diversity and distinctness were calculated using the PRIMER 5 version 5.2.9 statistical package. The resulting Δ and Δ^* were graphically compared by site and month. Heterogeneity of variance was tested using Levene's test and both Δ and Δ^* failed equal variance. Therefore, differences between sites and months were analyzed using the Kruskal-Wallis test and subsequent pair-wise comparisons, using the Mann-Whitney test, to determine which sites and months were significantly different (Sokal and Rohlf 1995, Zar 1999). The SPSS version 10.0 statistical package was used for statistical analysis.

Fish diversity was also examined using average taxonomic distinctness (AvTD, Δ^+), a convergence of Δ and Δ^* to the same statistic, defined as the mean taxonomic distance connecting every species pair on the previously mentioned taxonomically classified eelgrass fish list for Clayoquot Sound (Clarke and Warwick 1998). Average taxonomic distinctness relies on presence/absence data and is therefore, independent of differing sampling effort. The data in this study had a lower sampling effort in 2001; therefore, AvTD may detect sites and seasons that were lower or higher than expected in taxonomic distinctness. Ninety-five percent confidence funnels produced from a randomization test of AvTD were examined to detect if any site-month combination fell outside of the expected values.

Variation in taxonomic distinctness (VarTD, Λ^+), is related to AvTD, and is the unevenness of taxonomic tree, achieved by measuring variance in pair wise taxonomic path lengths (Clarke and Warwick 2001). VarTD relies on presence/absence data and is therefore, independent of sample effort. The combination of AvTD and VarTD by plotting 2 d simulation envelopes can pull out sites with either AvTD or VarTD being higher or lower than expected while the counterpart index remains within expected values (Clarke and Warwick 2001). AvTD and VarTD were analyzed using the PRIMER version 5.2.8 statistical package.

3.2.3.3. Beta Diversity

Izsak and Price (2001) developed a taxonomic similarity index (Δ_s) that uses species presence/absence data to measure beta diversity. Δ_s is derived from taxonomic distance, which calculates the average taxonomic path length between any two species in different sites/areas (Appendix 3.2). The species were classified into species, genus, family and order (Nelson 1994) and each taxonomic level was given a weight of 1 (Clarke and Warwick 1999). Thereby, if the two individuals were the same species the path length was 0, different species within the same genus were weighted 2, etc., where path length compares two species from different sites/areas. Taxonomic similarity was then calculated by subtracting 1 from taxonomic distance, divided by the total number of taxonomic levels minus 1 (Appendix 3.2).

Taxonomic similarity was calculated for fish species data, bimonthly, in 2002. Pair-wise comparisons of fish species were made between each site and sampling period to generate a similarity matrix. The average Δ_s values in the similarity matrix are beta diversity, where Δ_s values are inversely related to beta diversity (e.g. high Δ_s values = low beta diversity) (Iszak and Price 2001). Two different similarity matrices were computed. First, Δ_s values were compared over a spatial gradient, by comparing fish species, between sites, in the same the sampling period. A second comparison of Δ_s values across a temporal gradient was calculated by comparing fish species, over each sampling, within that same site. Descriptive statistics were used to compare beta diversity because of the lack of independence of similarity matrix values.

A Bray-Curtis similarity matrix from the 2002 Log(x+1) fish species abundance data was also used to examine the relationship between distances among sites and fish assemblage similarity. The Bray-Curtis similarity values were reorganized so that the similarity value between each sample was compared over distance. For example, all similarity values within a site over each sampling period were given a distance value of 0 km, and the distance between each site was used to compare all other sites by month similarity values. Bray-Curtis similarity values were also evaluated by examining the relationship between time between each sampling period and fish assemblage similarity. The relationship between distance/time between samples and Bray-Curtis similarity of Log(x+1) fish species abundance data was calculated using a Spearman rank correlation coefficient (Sokal and Rohlf 1995).

3.2.3.4. Fish Assemblage Analysis

Graphical comparisons of the 2002 fish family groups were also compared between each site-month combination to determine which fish families might be driving patterns observed in univariate diversity measures. Additional juvenile and adult abundance comparisons were explored for common fish species, including shiner perch (*Cymatogaster aggregata*), threespine stickleback (*Gasterosteus aculeatus*), tube-snout (*Aulorhynchus flavidus*), Pacific snake prickleback (*Lumpenus sagitta*), and members of the seaperch fish family Embiotocidae.

3.2.4. Multivariate Analysis

Cluster analysis and non-metric multidimensional scaling (MDS) were calculated to determine if site-month trends and distinct fish assemblages were present in the eelgrass fish diversity. For site-month comparisons, data from both 2001 and 2002 were combined for ease of interpretation, as similar trends were found when years and sampling periods were kept separate. Sites were compared on a monthly basis by exploring the fish species data in two forms: Log(x+1) transformed total fish species abundance, and fish species presence/absence data. A Log(x+1) transformation of fish species abundance was used because it put the data on a common scale and accounted for zeros found in species data (Clarke and Green 1988). Separate Bray-Curtis similarity matrices, that included all fish species, were calculated for each data type. Between site-month similarity calculations were ordinated using 2-dimensional non-metric MDS procedure and clustered using group-average cluster analysis on ranked similarity. The fewest number of distinctive groups formed from the cluster analysis were overlaid onto the MDS plots. By comparing these two procedures the formation of discrete site or month clusters was determined (Clarke 1993).

Fish diversity was further compared on a site-month basis by clustering and ordinating the data on a genus, family and order level, to determine if similar patterns in fish diversity were formed at higher taxonomic levels. A second stage MDS compared the similarity matrices at each taxonomic level to determine how patterns of eelgrass fish diversity were similar at decreasing taxonomic resolution (Clarke and Warwick 1994).

Fish assemblage clustering and MDS analysis was done only for the 2002 data, because the data set was more complete and consistent than the 2001 data. To ensure interpretable cluster and ordination analysis, rarer fish species in the species similarity matrices, whose overall abundance consisted of ≥0.1% the total fish abundance, were eliminated leaving ≥50% of the fish species in the matrix (Field et al. 1982). Therefore Bray-Curtis similarity matrices were calculated for a total of 23 common fish species from the 2002 data set. Species similarity matrices were then ordinated, using a 2-dimensional non-metric MDS procedure, and clustered using group-average cluster on ranked similarity. A comparison between the two procedures determined if discrete assemblages of fish occur in the eelgrass beds sampled.

A 2-way crossed analysis of similarity (ANOSIM) was also used to separately analyse 2001 and 2002 fish diversity data by determining whether differences between fish assemblages occurred between each site/month combination. A 2-way crossed ANOSIM is a non-parametric procedure on rank similarity matrix that is not restricted to a balanced number of replicates and compares fixed sites sampled over time (Clarke and Warwick 1994). Bray-Curtis similarity matrices were calculated for Log(x+1) transformed fish species abundance, keeping each replicate beach seine sample separate and including all the fish species collected (Clarke and Warwick 1994). The 2-way crossed ANOSIM was applied to the similarity matrix to identify if significant variation occurred between sites and months. All multivariate analysis was done using the PRIMER version 5.2.8 statistical package.

3.3. Results

3.3.1. Univariate Diversity Measures

Overall, similar trends were found in 2001 and 2002 for each univariate diversity measure (Table 3.2). However, 2002 data had more statistically significant differences between site and month comparisons than that in 2001, perhaps due to more consistent and intensive sampling in 2002.

Table 3.2. Summary of univariate diversity measures and beta diversity results. All significance levels at $P<0.05$.

Indices	**Year**	**Site**	**Month**
Species Richness (s)	2001	Arakun & Auseth: significantly < Ducking, Sharp	Sept: significantly < May, Jun, Aug
	2002	Sharp: significantly > Arakun, Indian	L Apr: significantly < L May
$\text{Log}_{10}(x+1)$ Transformed Abundance	2001	Arakun: significantly < Auseth, Ducking, Indian, Kootowis, Sharp	No significant differences
	2002	Arakun: significantly < Ducking, Felice, Indian, Kootowis, Sharp Indian: significantly > Auseth, Ducking Sharp: significantly > Auseth, Ducking	E Aug, L Aug, E Sept: significantly < L Apr to E Jly
Pielou's Evenness (J')	2001	No significant differences	May: significantly > Aug, Sept
	2002	Arakun: significantly > Indian, Kootowis, Sharp	E Aug, L Aug, E Sept: significantly > L Apr to E Jly L Jly: significantly < L May, E Jun, L Jun, and significantly > E Aug, L Aug, E Sept
Taxonomic Diversity (Δ)	2001	No significant differences	Jun: significantly > Jly, Aug, Sept Sept: significantly < Jly
	2002	Arakun: significantly < Indian	L Jly: significantly > L May, E Jun, L Aug E Aug, E Sept: significantly < E May to E Jly L Aug: significantly < L Apr to L Jly
Taxonomic Distinctness (Δ^*)	2001	Kootowis: significantly > Arakun, Sharp	No significant differences
	2002	Kootowis: significantly > Arakun, Auseth, Ducking, Felice, Indian Sharp: significantly > Arakun	No significant differences
Beta Diversity	2002	Taxonomic similarity (Δ_s) decreases as distance between samples increases Bray-Curtis similarity values decrease as distance between samples increases Kootowis & Felice had the highest mean Δ_s	Taxonomic similarity decreases as time between samples increases All Δ_s values are similar across bimonthly samples

3.3.1.1. Species Richness

In 2001 and 2002, a total of 58 fish species were caught in all the eelgrass beds (Appendix 3.3). Fish species richness differed significantly, by both site and month, in 2001 and 2002 (2001 month, Kruskal-Wallis, df=4, $H = 16.448$; 2002 month, Kruskal-Wallis, df=9, $H = 23.335$; $P<0.01$ for two previous cases; 2001 site, Kruskal-Wallis, df=6, $H = 27.037$; 2002 site, Kruskal-Wallis, df=6, $H = 25.578$; $P<0.001$). Overall trends showed all sites to have similar species richness in April and May, variable richness from June to August and similar species richness again in late August and September (Figure 3.1). Seasonally, richness was significantly less in September than May, June and August in 2001, but differing significantly between late April and late May in 2002 (2001, Mann-Whitney, $P<0.005$; 2002, Mann-Whitney, $P<0.001$). Site comparisons showed Sharp to have significantly higher species richness Arakun and Auseth in 2001 and Arakun and Indian in 2002, and in 2001 species richness at Ducking was also significantly higher than at Arakun and Auseth (Mann-Whitney, $P<0.002$).

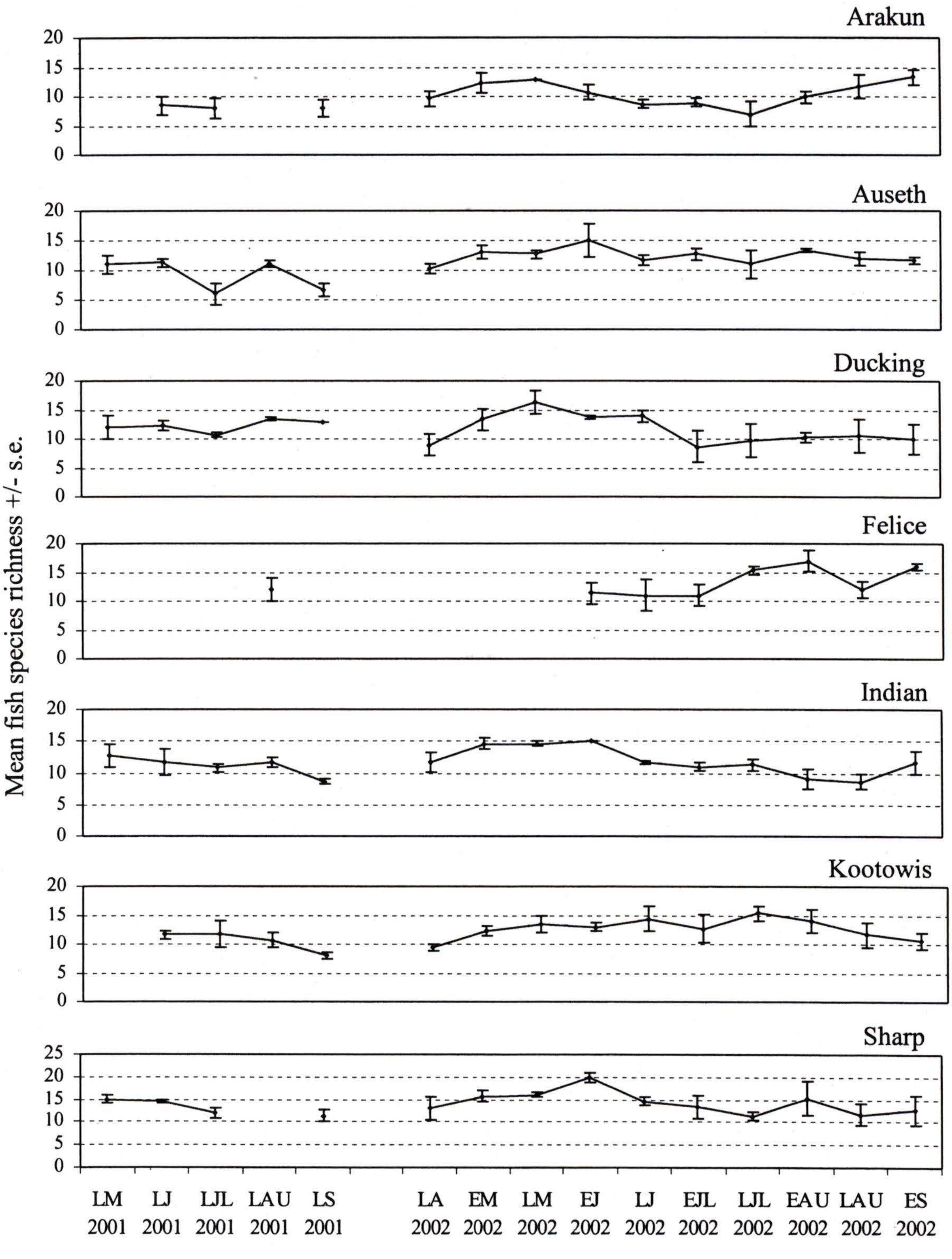

Figure 3.1. Mean fish species richness at each site monthly in 2001 and bimonthly in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

3.3.1.2. Abundance

Log(x+1) transformed total fish abundance differed significantly by both month and site in 2002, but only by site in 2001 (2002 month, Kruskal-Wallis, df=9, H = 60.432; 2001 site, Kruskal-Wallis, df=6, H = 23.036; 2002 site, Kruskal-Wallis, df=6, H = 58.709; P<0.001 for all cases). Overall fish abundance was lower in the late spring and early summer, increased in late July and decreased in September in both 2001 and 2002 (Figure 3.2). Fish abundance in August and September 2002, was significantly higher than all previously sampled months (Mann-Whitney, P<0.001). This trend was driven by an increase in young of the year (YOY) shiner perch (*Cymatogaster aggregata*) and threespine stickleback (*Gasterosteus aculeatus*) in early and late July (Figures 3.3 and 3.4). All the sites in 2002 follow similar trends seasonally until late August and early September (Figure 3.2). Ducking, Felice and Indian all decreased in abundance in late August and this was driven by a decrease in YOY *C. aggregata* in Ducking , fewer YOY *G. aculeatus* at Indian and fewer forage fish (*Ammodytes hexapterus, Hypomesus pretiosus,* and *Clupea pallasi*) caught at Felice (Figures 3.3, 3.4 and 3.5). In September 2002, Arakun and Auseth abundance decreased because of fewer YOY *C. aggregata* at Arakun and fewer YOY *G. aculeatus* at Auseth (Figures 3.3 and 3.4). In comparison, abundance at Kootowis and Sharp did not decrease in September because YOY *G. aculeatus* remained high (Figure 3.4). Fish abundance at Arakun was significantly lower than all sites in 2001, except Felice and 2002, except Auseth (Mann-Whitney, P<0.002). In 2002, fish abundance was lower at Auseth and Ducking than Indian and Sharp (Mann-Whitney, P<0.002).

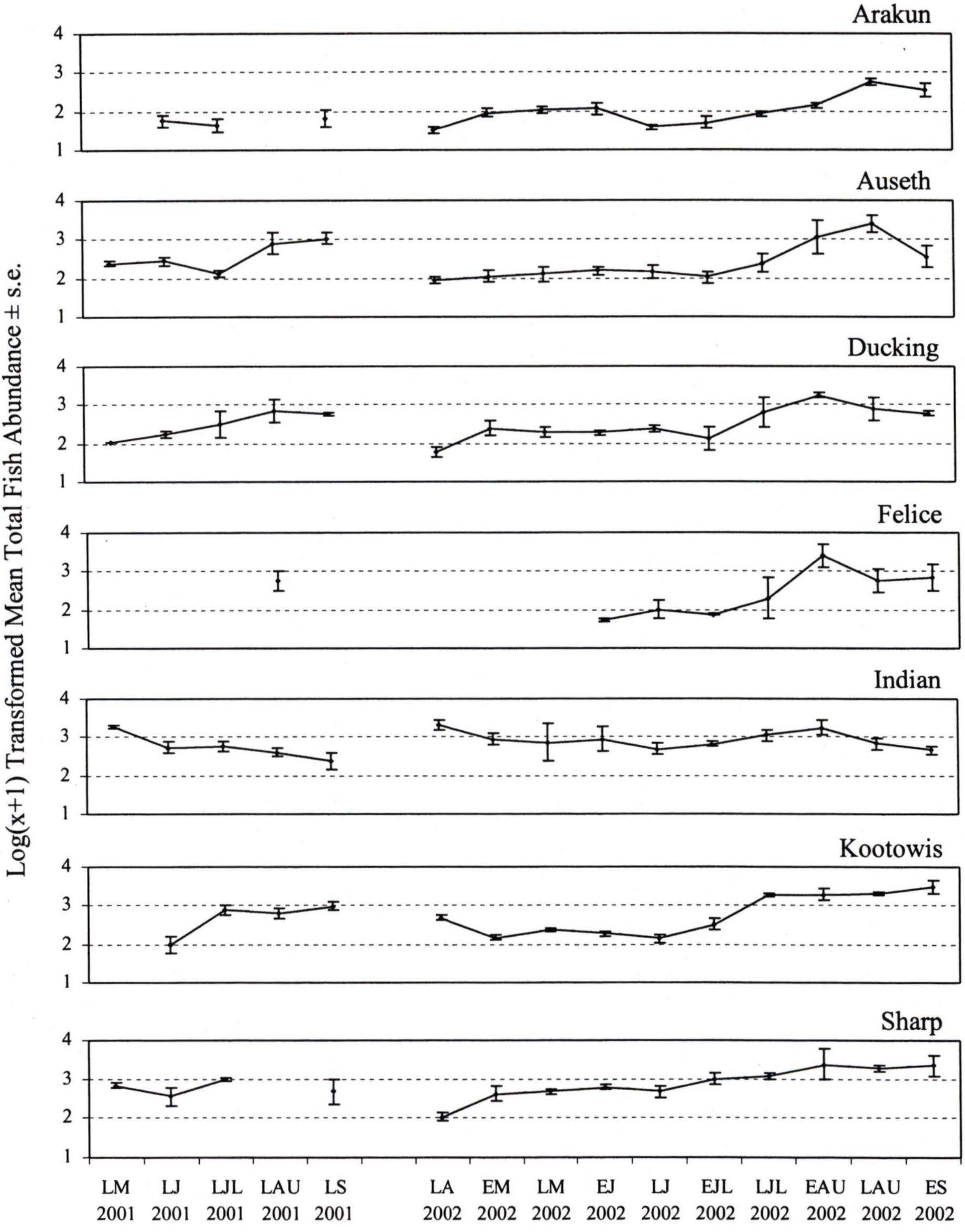

Figure 3.2. Log(x+1) transformed mean total fish abundance at each site monthly in 2001 and bimonthly in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

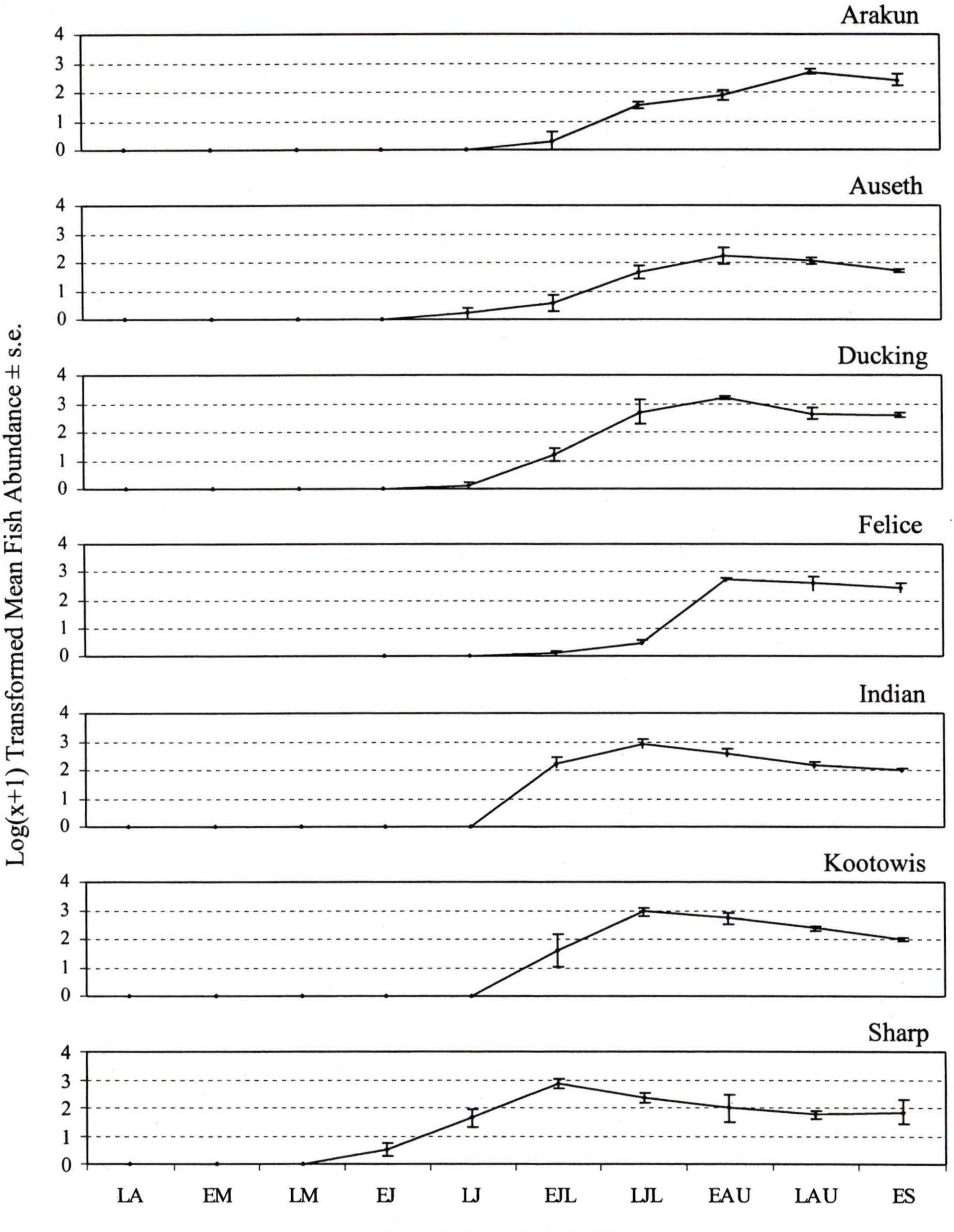

Figure 3.3. Log(x+1) transformed abundance of young of the year (YOY) shiner perch (*Cymatogaster aggregata*) caught bimonthly at each site in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

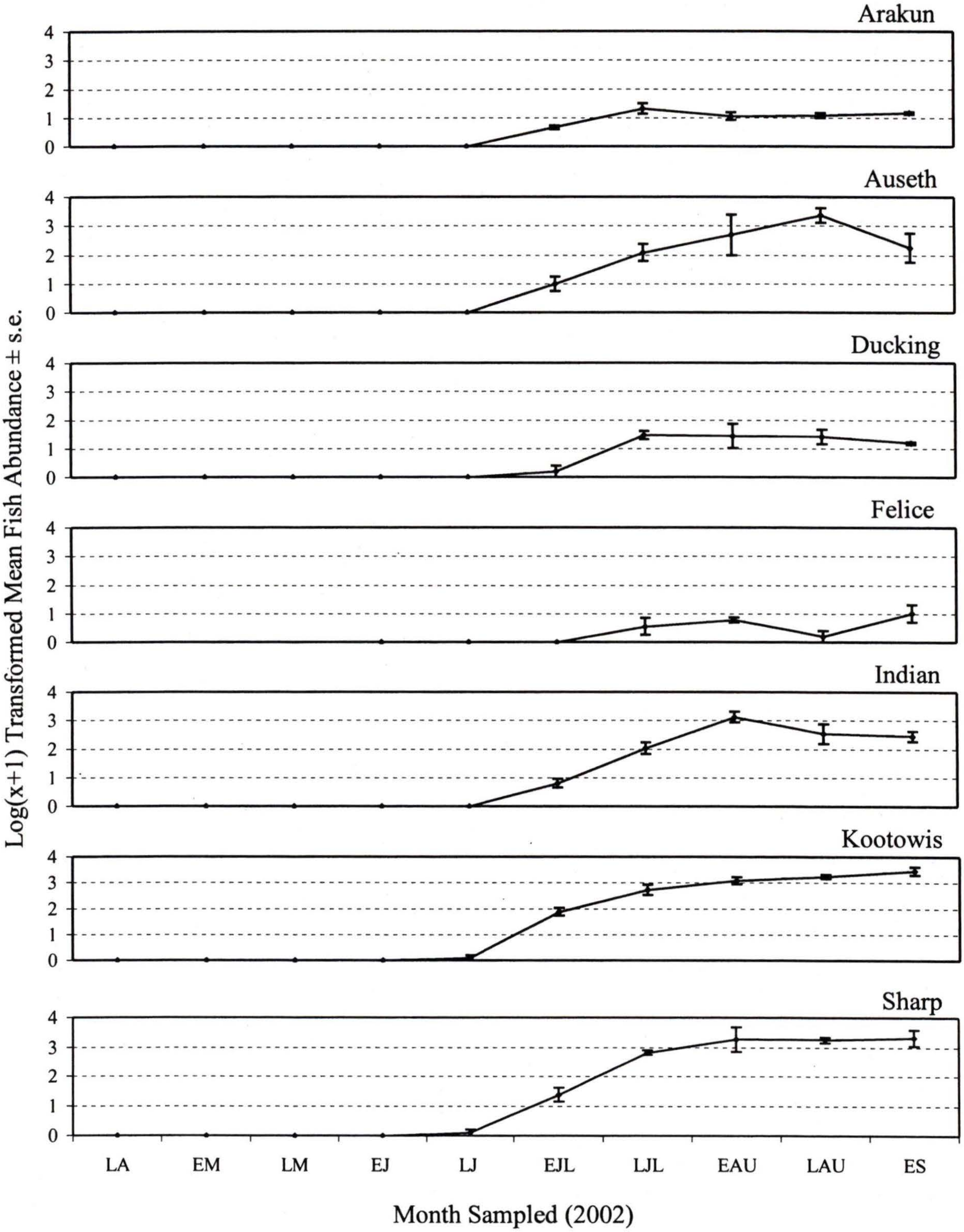

Figure 3.4. Log(x+1) transformed abundance of young of the year (YOY) threespine stickleback (*Gasterosteus aculeatus*) caught bimonthly at each site in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

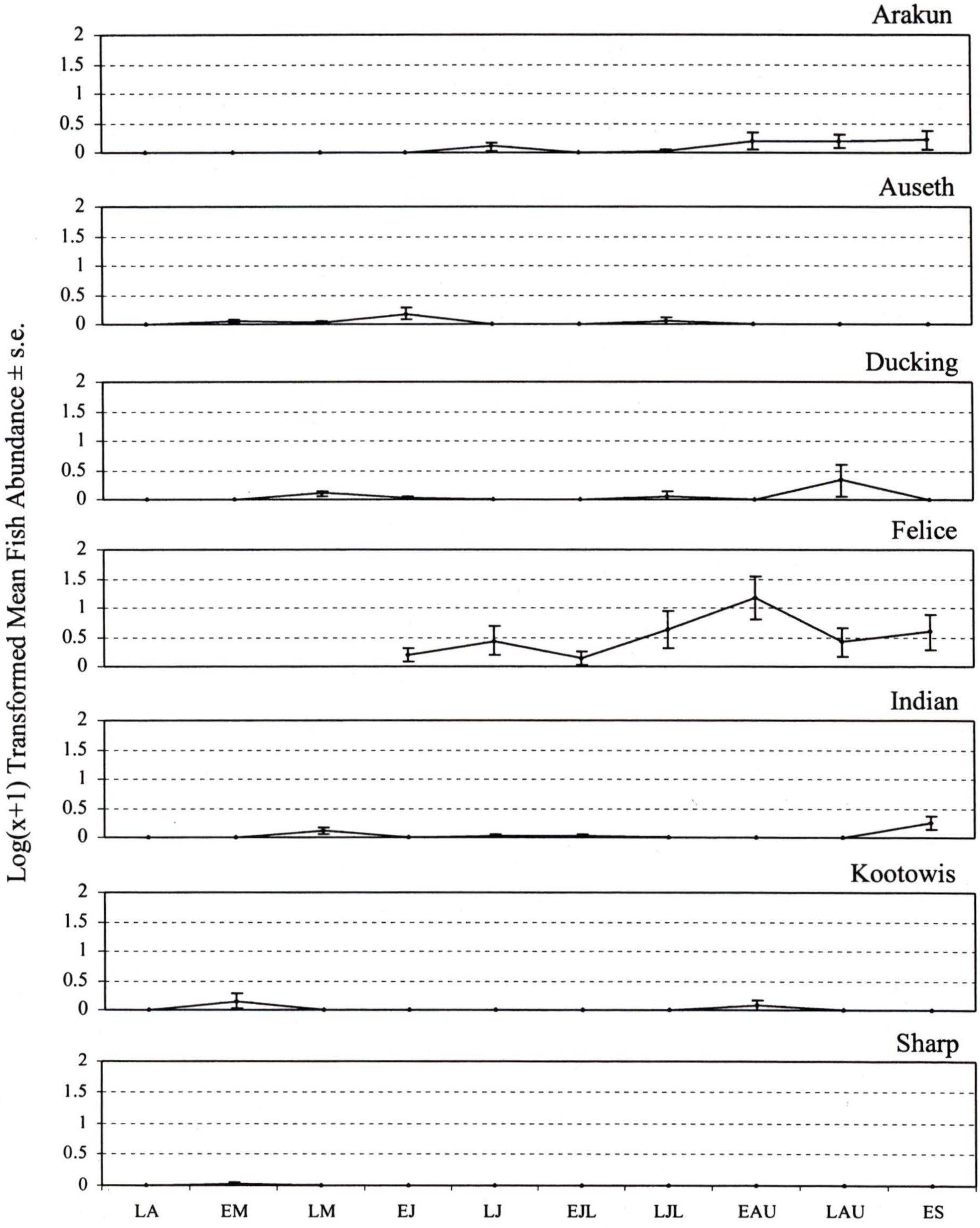

Figure 3.5. Log(x+1) transformed forage fish abundance caught bimonthly at each site in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

3.3.1.3. Evenness

Pielou's evenness differed significantly by both month and site in 2002, but only by month in 2001 (2001 month, Kruskal-Wallis, df=4, $H = 15.670$; $P<0.01$; 2002 month, Kruskal-Wallis, df=9, $H = 84.392$; 2002 site, Kruskal-Wallis, df=6, $H = 25.065$; $P<0.001$ for previous two cases). The overall seasonal trends in 2001 and 2002 showed evenness increasing to June and decreasing from July onwards (Figure 3.6). In 2001, only June data had significantly higher evenness than that for August and September (Mann-Whitney, $P<0.001$). However, in 2002, August and September data sets both demonstrated significantly lower evenness from April to July (Mann-Whitney, $P<0.001$). Evenness in late July was also significantly lower than in late May and June (Mann-Whitney, $P<0.001$). In July, evenness decreased because both YOY *Cymatogaster aggregata* and *Gasterosteus aculeatus* increased in abundance in July (Figures 3.3 and 3.4). In 2002, Arakun data showed significantly higher evenness than that for Indian, Kootowis and Sharp (Mann-Whitney, $P<0.002$), driven by the overall lower numbers of YOY *C. aggregata* and *G. aculeatus* caught at Arakun (Figures 3.3 and 3.4). In April 2002, Kootowis data exhibited low evenness because of the high number of juvenile salmonids (Figure 3.7), whereas evenness at Indian was lower from April to July because there was a high number of plus one-year-old threespine sticklebacks (Figure 3.8).

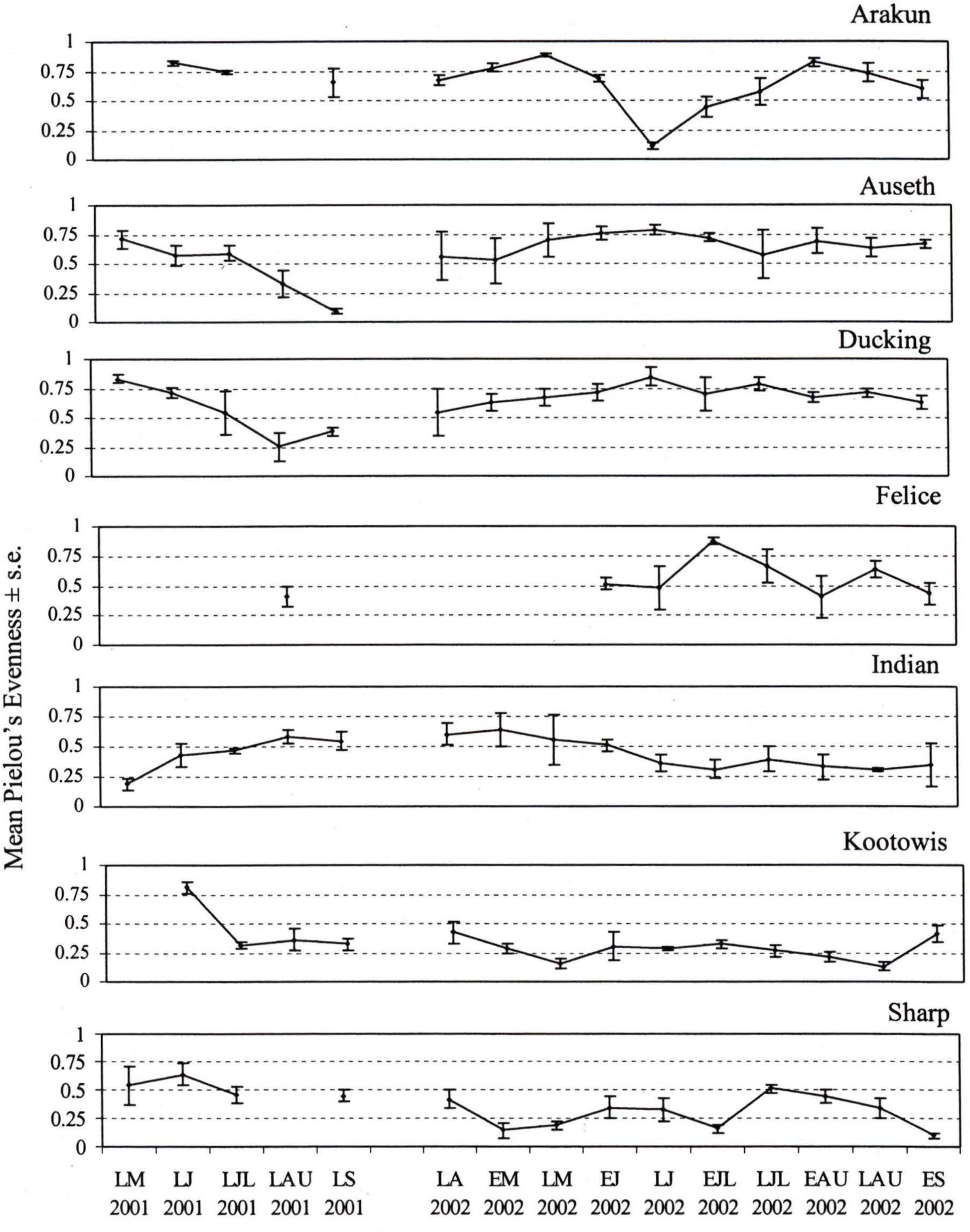

Figure 3.6. Mean Pielou's evenness values at each site monthly in 2001 and bimonthly in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

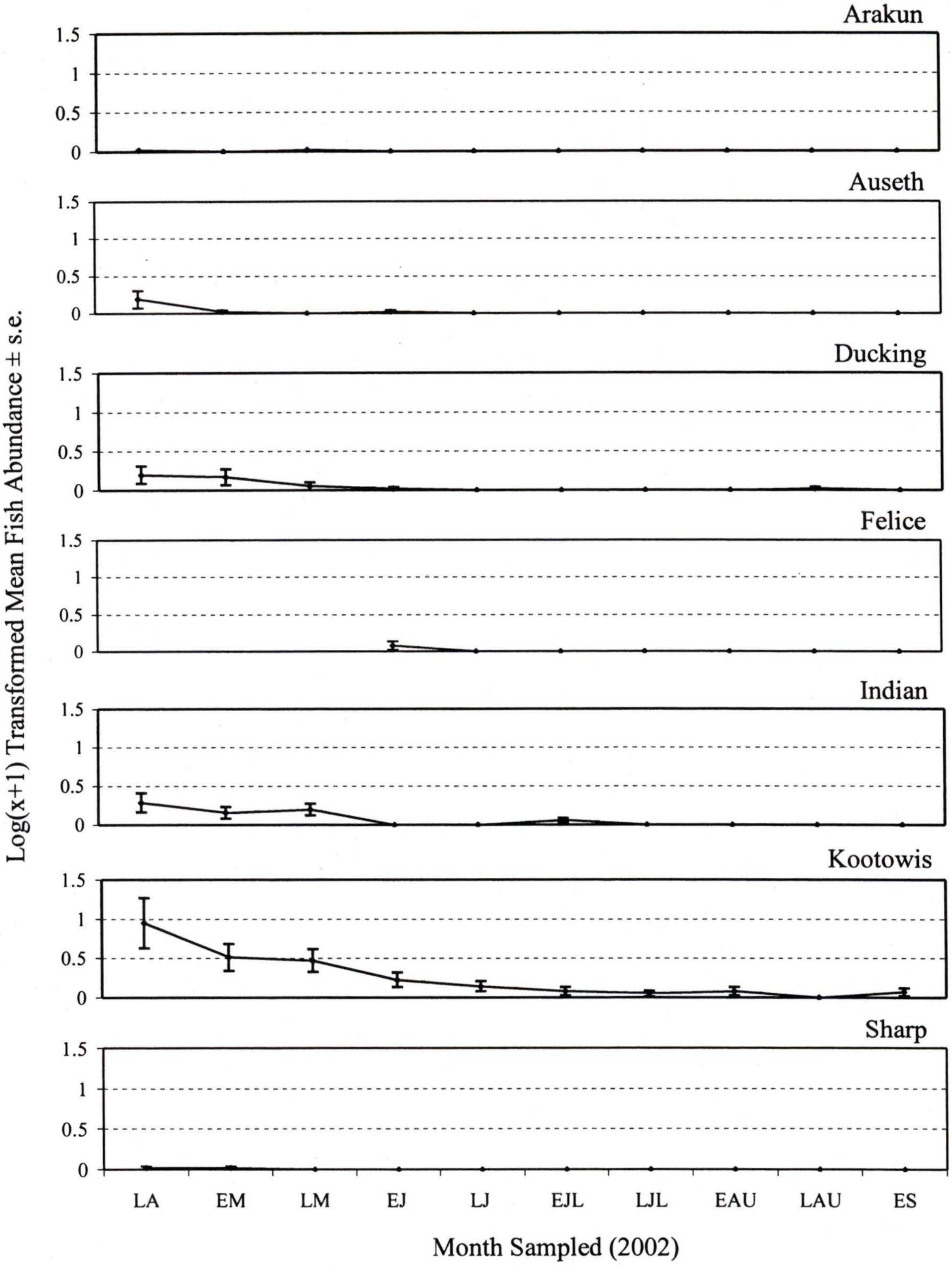

Figure 3.7. Log(x+1) transformed salmonid abundance caught bimonthly at each site in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

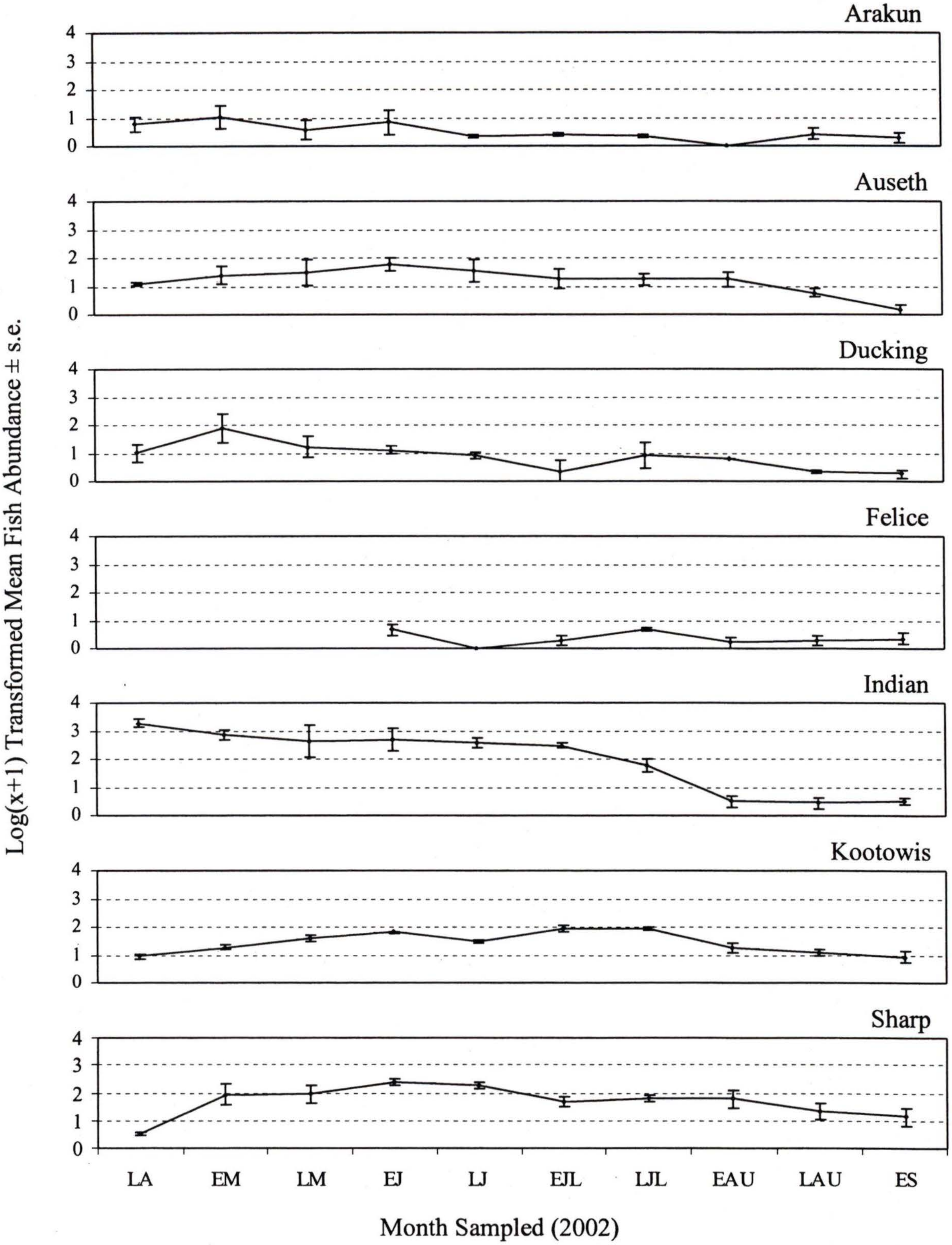

Figure 3.8. Log(x+1) transformed abundance of 1+ year old threespine stickleback (*Gasterosteus aculeatus*) caught bimonthly at each site in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

3.3.2. Taxonomic Diversity and Distinctness

Taxonomic diversity (Δ) differed significantly by both month and site in 2002, but only by month in 2001 (2001 month, Kruskal-Wallis, df=4, $H = 23.310$; 2002 month, Kruskal-Wallis, df=9, $H = 89.184$; $P<0.001$ for previous two cases; 2002 site, Kruskal-Wallis, df=6, $H = 15.638$; $P<0.05$). The overall seasonal trend in 2001 and 2002 showed an increase in Δ in June, decreasing in July and August and increasing slightly in September (Figure 3.9). In 2001, September data displayed significantly lower Δ than that for June and July (Mann-Whitney, $P<0.001$). June also had significantly higher Δ than July and August in 2001 (Mann-Whitney, $P<0.005$). Similarly, in 2002 August and September data were significantly lower in Δ than that for May, June and early July, whereas Δ in April was only significantly higher than that in late August (Mann-Whitney, $P<0.001$). In late July Δ was less than late May, early June and late August (Mann-Whitney, $P<0.001$). The seasonal differences in Δ between May, June and early July, and August and September were driven by the increased abundance of YOY *Cymatogaster aggregata* and *Gasterosteus aculeatus* in late July (Figures 3.3 and 3.4). In 2002, significant differences in Δ were not seen between April, and August and September because Kootowis and Indian both had low Δ in April. Kootowis data had low Δ because of the high number of juvenile salmonids (Figure 3.7), whereas the Δ at Indian was lower from April to July because there was a high number of adult *G. aculeatus* (Figure 3.8). However, site comparisons showed only Arakun data to have significantly higher Δ than that at Indian in 2002 (Mann-Whitney, $P<0.001$).

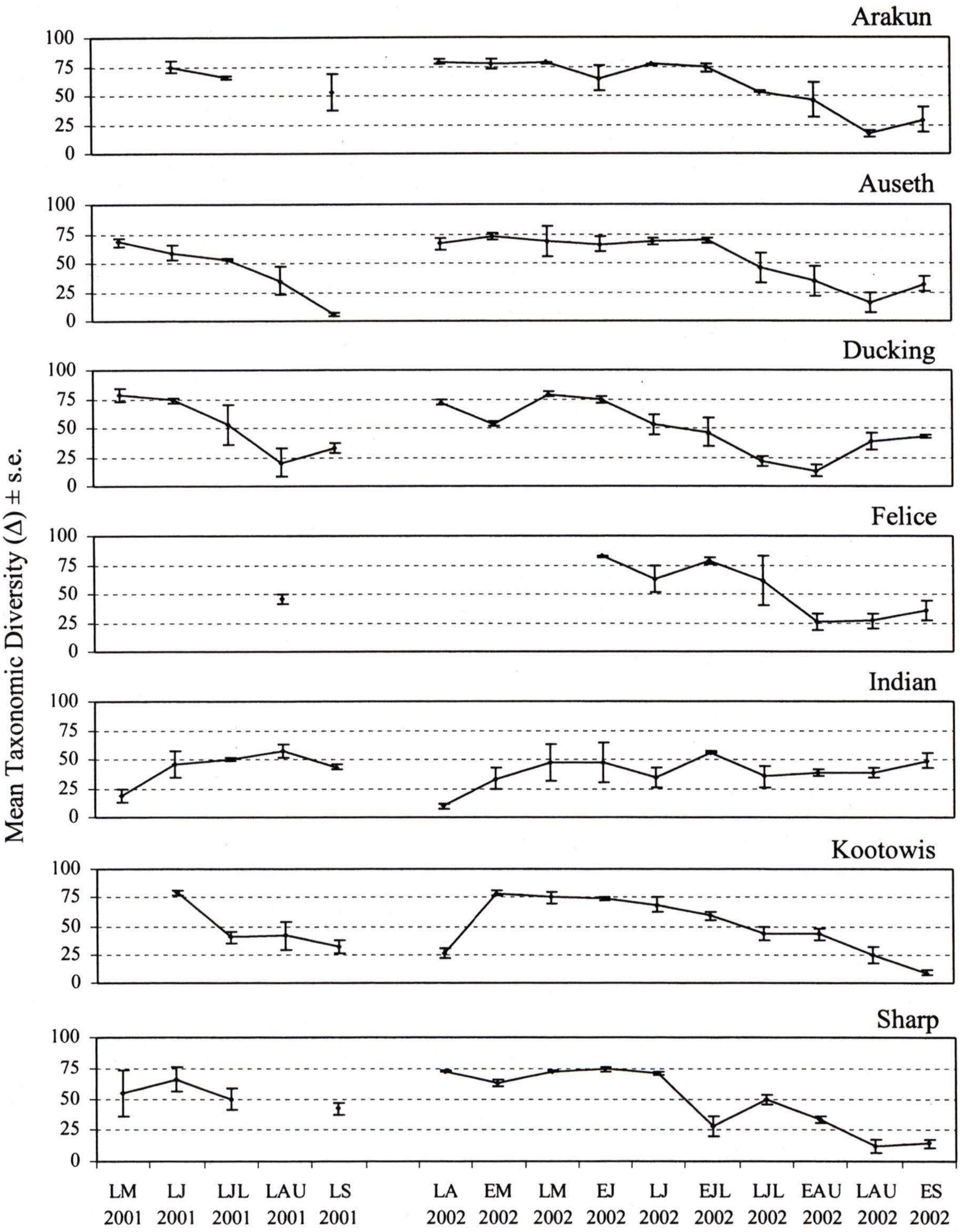

Figure 3.9. Mean taxonomic diversity (Δ) at each site monthly in 2001 and bimonthly in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

Taxonomic distinctness (Δ^*) differed significantly only by site in 2001 and 2002 (2001 site, Kruskal-Wallis, df=6, $H = 20.672$; $P<0.002$; 2002 site, Kruskal-Wallis, df=6, $H = 33.883$; $P<0.001$). Seasonally, all sites followed similar trends from May to July, and in early August, taxonomic distinctness varied between sites (Figure 3.10). In April 2002, Kootowis data demonstrated much lower Δ^* than all other sites due to the high number of salmonids species caught (Figure 3.10). In early August 2002, Δ^* decreased at Arakun and Felice because YOY *Cymatogaster aggregata* decreased, and in late August and early September, Δ^* at Ducking, Auseth and Indian decreased because both YOY *C. aggregata* and *Gasterosteus aculeatus* decreased (Figures 3.3 and 3.4). Finally, Kootowis and Sharp Δ^* data did not decrease in September because the number of YOY *G. aculeatus* remained high (Figure 3.4). However, only Kootowis data had significantly higher Δ^* than that collected at Sharp and Arakun in 2001 and all six sites in 2002 (Mann-Whitney, $P<0.002$).

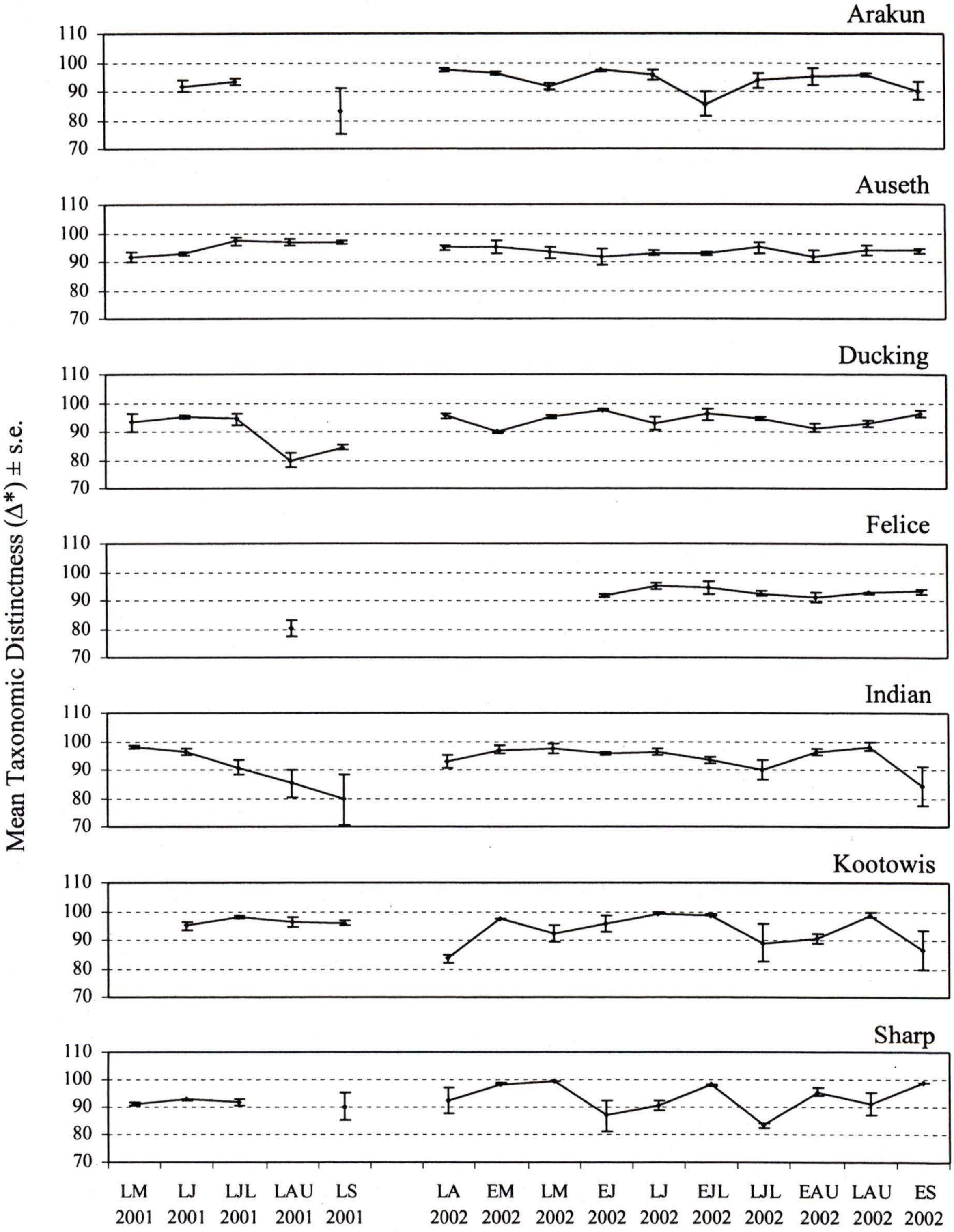

Figure 3.10. Mean taxonomic distinctness (Δ*) at each site monthly in 2001 and bimonthly in 2002. (Abbreviations: LA=late April, EM=early May, LM=late May, EJ=early June, LJ=late June, EJL=early July, LJL=late July, EAU=early August, LAU=late August, ES= early September, LS=late September).

Average taxonomic distinctness (AvTD) and variation in taxonomic distinctness (VarTD) were examined for 2001 and 2002 separately using, 2 dimensional simulation envelope plots. In 2001, only samples for Ducking in late May and Kootowis in late June fall outside the 95% confidence envelope due to higher AvTD than expected (Figure 3.11). In 2002, no samples fall outside the expected 95% confidence envelope (Figure 3.12). However, samples taken at Kootowis in 2002, in early April had a higher VarTD value and a higher AvTD value in early May (Figure 3.12).

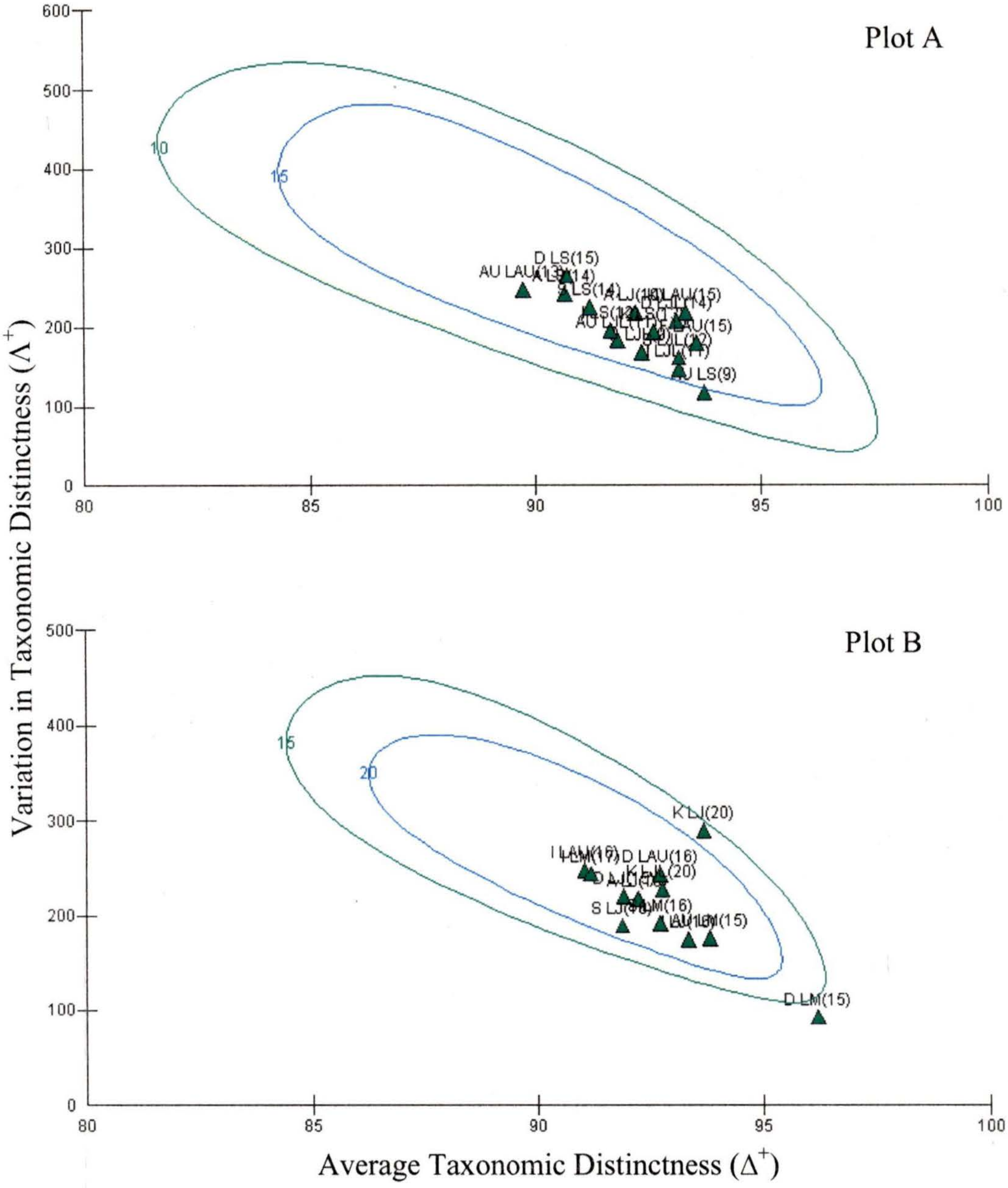

Figure 3.11. Fitted 95% probability contours of the average and variance of taxonomic distinctness from 1000 simulation for species ranges in 2001. (Plot A: < 15 species. Plot B: 15-20 species). On each sub plot, are the observed taxonomic distinctness values for each site-month. Only for Kootowis in late June (KLJ) and Ducking in late May (DLM) have points (Plot B) that fell outside the expected 95% confidence envelope.

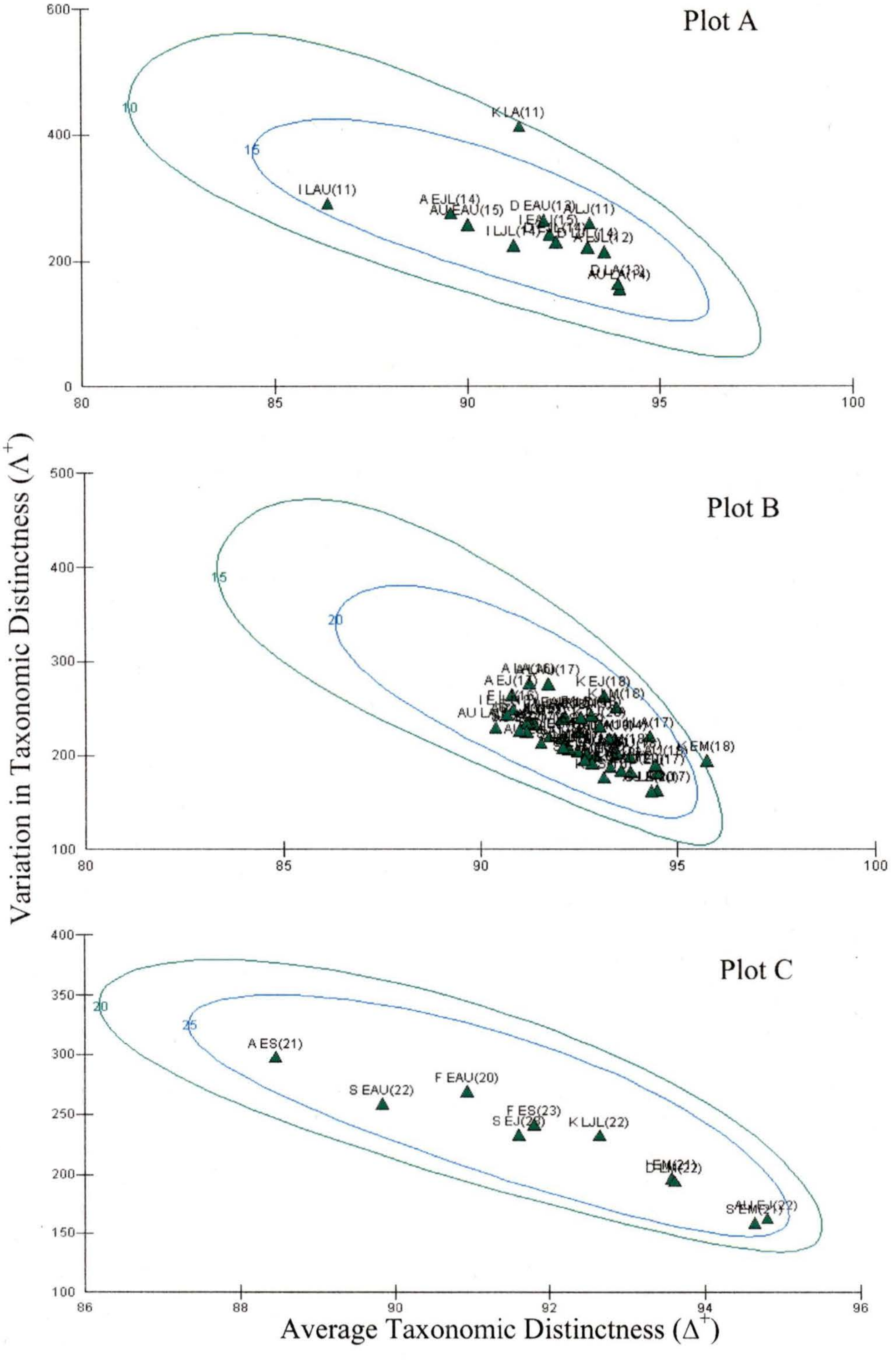

Figure 3.12. Fitted 95% probability contours of the average and variance of taxonomic distinctness from 1000 simulation for species ranges in 2002. (Plot A: < 15 species; Plot B: 15-20 species; Plot C: 20-25 species). On each sub plot, are the observed taxonomic distinctness values for each site-month.

3.3.3. Beta Diversity

Taxonomic similarity (Δ_s) calculated for each site in 2002 was lower for data from Felice and Kootowis than for data from the other sites, suggesting greater taxonomic turnover and hence higher beta diversity at these two sites (Table 3.3). Bimonthly comparisons of Δ_s at all sites combined showed similar monthly Δ_s values, suggesting no real difference in taxonomic turnover and therefore beta diversity by month (Table 3.4).

Using Δ_s, beta diversity was examined along a gradient of increasing distance (Figure 3.13). The trend showed Δ_s decreasing with increasing distance, and hence greater beta diversity the greater the distance between two sites. Taxonomic similarity was also compared over an increasing time gradient, where Δ_s decreased as the separation between sampling days increased (Figure 3.14). Therefore, higher beta diversity, greater taxonomic turnover, occurs over an increasing time gradient.

Bray-Curtis similarity values calculated for fish assemblages across sites were compared for each sample over distance (Figure 3.15). A significantly negative correlation was found between Bray-Curtis similarity values across increasing distance between samples (Spearman rank correlation coefficient, $r_s = -0.33$, $P<0.01$). Therefore, similarity of Log(x+1) fish species abundance decreased and beta diversity increased, as distance increased between the samples.

Bray-Curtis similarity values for fish assemblages were also compared for each sample over time (Figure 3.16). A significantly negative correlation was found between Bray-Curtis similarity values across increasing time between samples (Spearman rank correlation coefficient, $r_s = -0.30$, $P<0.001$). Consequently, Log(x+1) fish species abundance decreased and beta diversity, as time increased between samples.

Table 3.3. Summary statistics for taxonomic similarity (Δ_s) calculated for each site in 2002 using fish species presence/absence data.

	Arakun	Auseth	Ducking	Felice	Indian	Kootowis	Sharp
Mean	0.648	0.687	0.655	0.574	0.621	0.570	0.652
Standard error	0.016	0.013	0.015	0.019	0.016	0.016	0.013
Range	0.498	0.432	0.459	0.515	0.647	0.731	0.556

Table 3.4. Summary statistics for taxonomic similarity (Δ_s) calculated bimonthly in 2002 using fish species presence/absence data.

	L April	E May	L May	E June	L June	E July	L July	E Aug	L Aug	E Sept
Mean	0.637	0.688	0.718	0.673	0.592	0.635	0.600	0.572	0.608	0.638
Standard error	0.031	0.020	0.017	0.014	0.022	0.018	0.020	0.021	0.022	0.020
Range	0.624	0.348	0.360	0.309	0.433	0.354	0.446	0.463	0.428	0.386

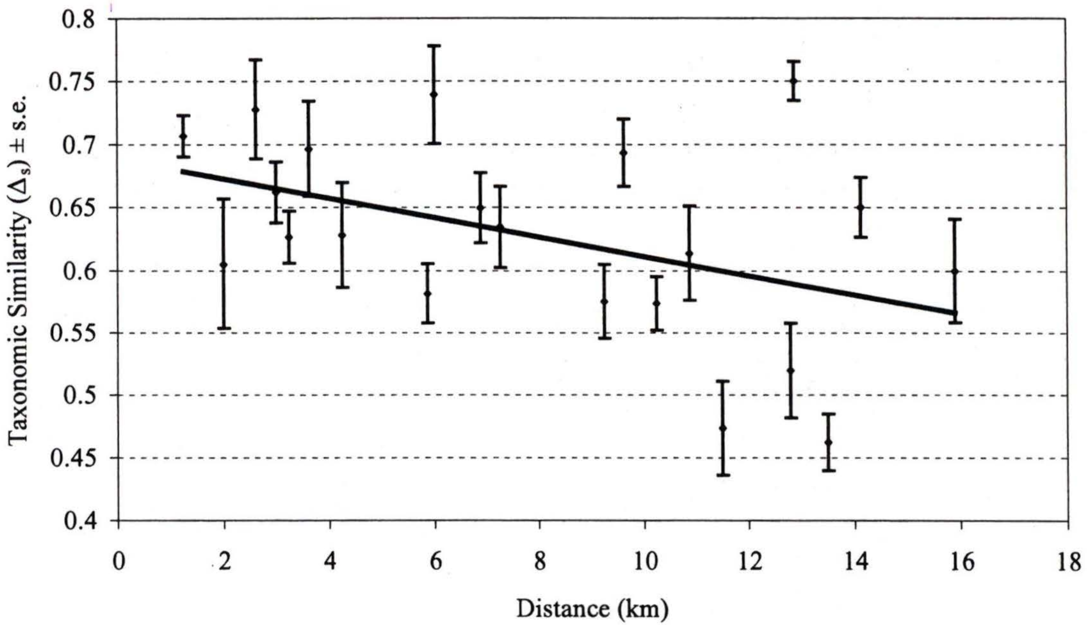

Figure 3.13. Plot comparing taxonomic similarity against distance between each site in 2002 with a trend line overlaid.

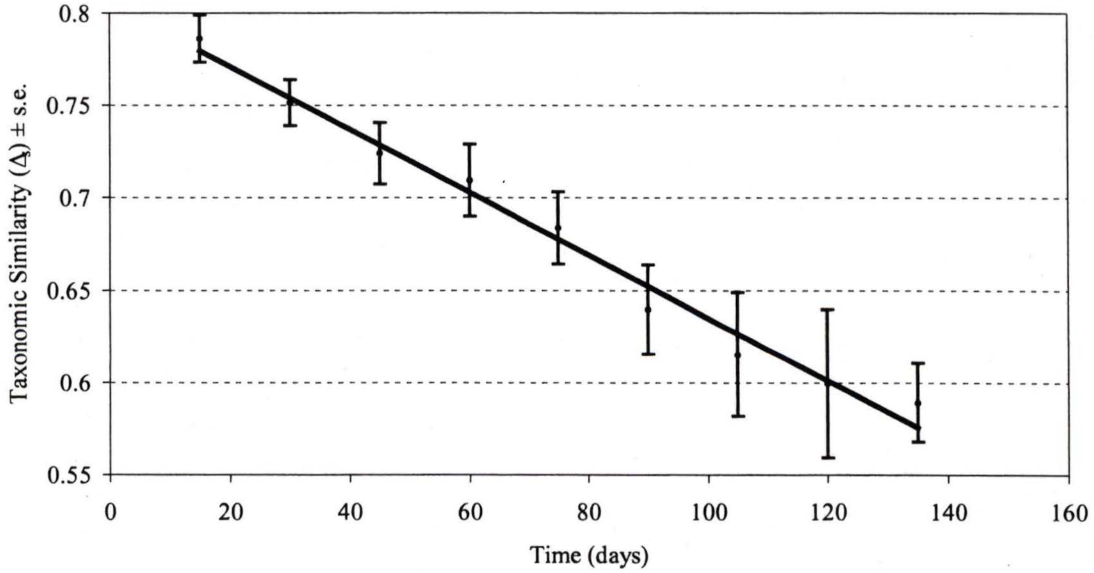

Figure 3.14. Plot comparing taxonomic similarity against time between each sampling period site in 2002 with a trend line overlaid.

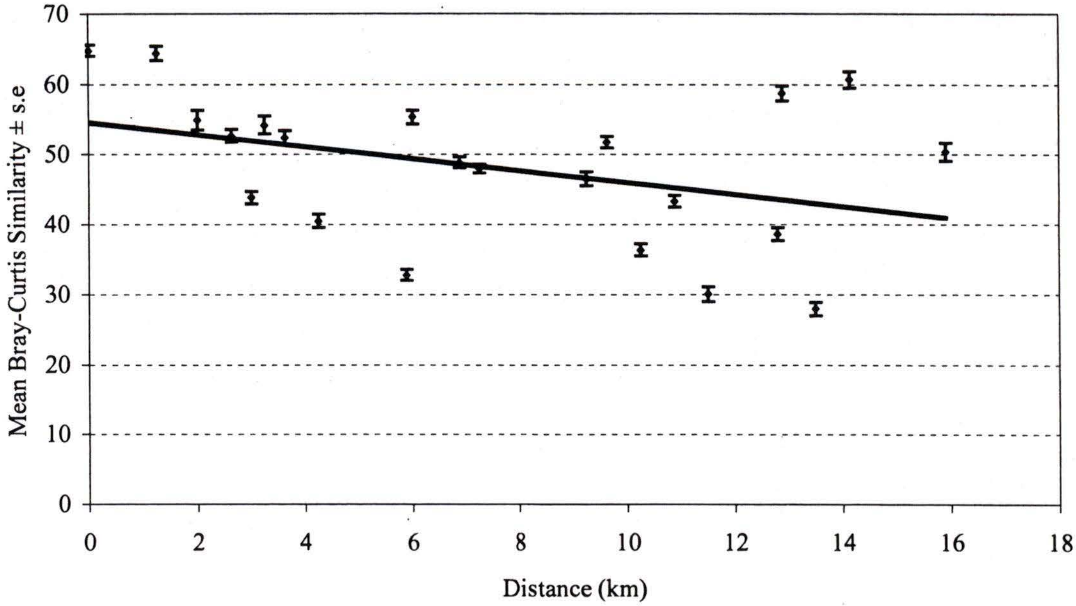

Figure 3.15. Plot of Bray-Curtis similarity of 2002 Log(x+1) transformed fish species abundance data against distance between each similarity value with a trend line overlaid.

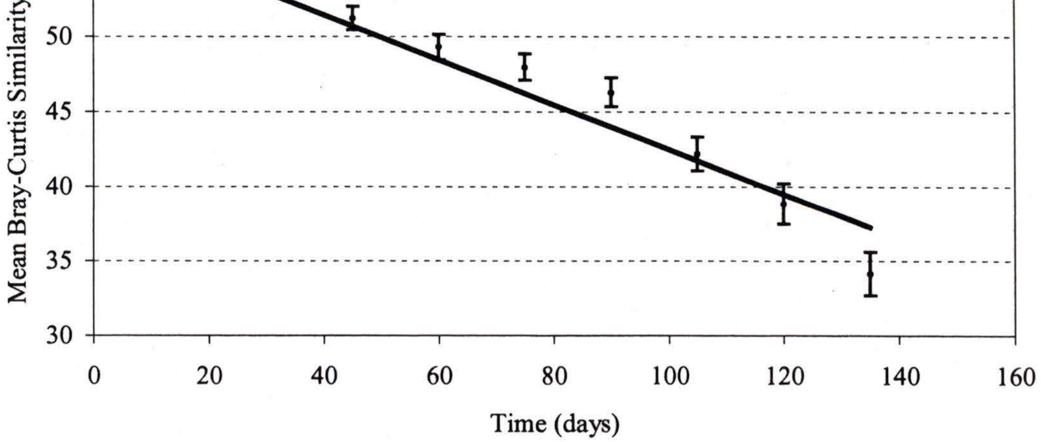

Figure 3.16. Plot of Bray-Curtis similarity of 2002 Log(x+1) transformed fish species abundance data against time between each similarity value with a trend line overlaid.

3.3.4. Multivariate Analysis

Overall, the multivariate techniques detected more subtle temporal and spatial differences that were not found using univariate techniques (Table 3.5). The site by month MDS plot of fish diversity data, including both species and abundance, showed a stronger relationship between sites than between months (stress = 0.14) (Figure 3.17). The superimposed ranked cluster groups on the MDS plot produce 4 clusters at a rank of 500 and 7 clusters at a rank of 350 (Figures 3.17 and 3.18). The five major clusters formed were:

- Kootowis in April
- Felice from all four months sampled
- Kootowis in May and June
- April samples from Arakun, Auseth, Ducking, Indian and Sharp
- All other site-month samples.

To further break the groups into 7 clusters included (Figures 3.17 and 3.18):

- Kootowis in April
- Felice all months
- Arakun in April
- Auseth, Ducking, and Indian in April
- Kootowis in May and June with Sharp in April
- All remaining Arakun and Ducking samples
- All remaining Auseth, Indian, Kootowis and Sharp samples

Table 3.5. Multivariate diversity analysis results summary.

Analysis	**Year**	**Site**	**Month**
MDS of Log(x+1) transformed fish species abundance	2001 and 2002	Felice sampled from June to September forms group Arakun and Ducking forms a group Auseth, Indian, Kootowis and Sharp form a single large group	Kootowis in May and June and Sharp in April form a group April only month forming a distinct group that includes Arakun, Auseth, Ducking, and Indian Kootowis in April forms a single distinct group
MDS of fish species presence/ absence	2001 and 2002	Felice forms a group that also includes Arakun and Ducking in August Kootowis sampled from May to September forms a group Arakun, Auseth, Ducking, Indian and Sharp form a single large group	April only month forming a distinct group that includes Arakun, Auseth, Ducking, Indian and Sharp Kootowis in April forms a single distinct group
2nd level MDS of taxonomic resolution	2001 and 2002	Similar site grouping at the species, genus, and family levels and site groups lost at the order level	Similar month grouping at the species, genus, and family levels and site groups lost at the order level
ANOSIM	2001	Significantly higher difference in fish diversity between sites than within ($P<0.05$) Felice is the only site not significantly different than all other sites, due to a much smaller sample size	All months differ significantly with the differences increasing as time between samples increases ($P<0.05$)
ANOSIM	2002	Significantly higher difference in fish diversity between sites than within ($P<0.05$) Fish diversity between each site differs significantly ($P<0.05$)	All months differ significantly with the differences increasing as time between samples increases ($P<0.05$)

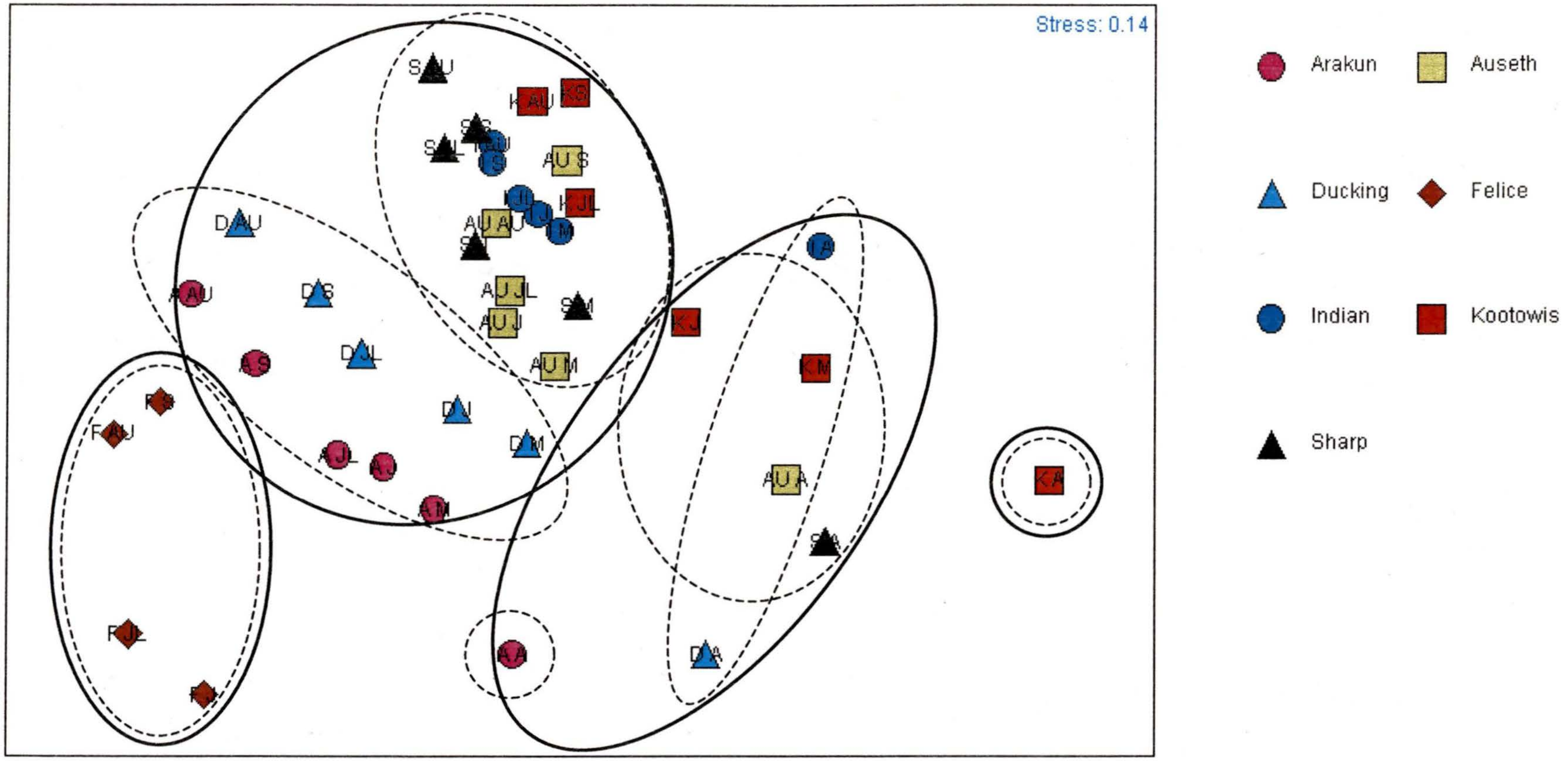

Figure 3.17. MDS ordination for Log(x+1) transformed fish species abundance at seven eelgrass beds, with data combined from both 2001 and 2002 by month with the superimposed ranked cluster groups. Solid lines indicating 4 clusters formed at a rank of 500 and the dotted lines indicating the 7 clusters formed at a rank of 350 (Site labels: A = Arakun, AU = Auseth, D = Ducking, I = Indian, K = Kootowis, S = Sharp; Month labels: A = April, M = May, J = June, JL = July, AU = August, S = September).

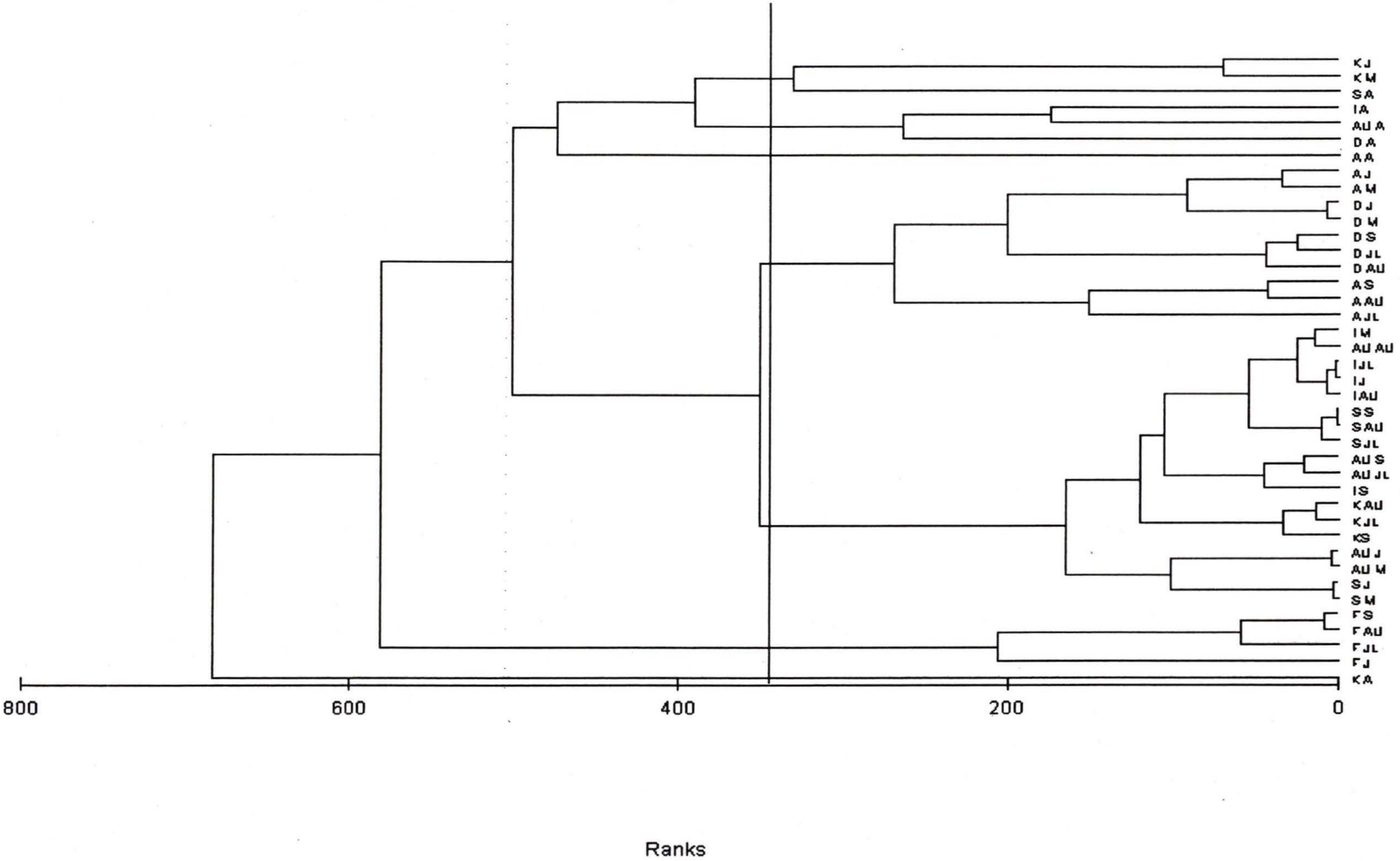

Figure 3.18. Ranked clustered groups for Log(x+1) transformed fish species abundance at seven eelgrass beds, with data combined from both 2001 and 2002 by month. Solid line indicates the 4 clusters formed at a rank of 500 and the dotted line indicates the 7 clusters formed at a rank of 350 (Site labels: A = Arakun, AU = Auseth, D = Ducking, I = Indian, K = Kootowis, S = Sharp; Month labels: A = April, M = May, J = June, JL = July, AU = August, S = September).

In comparison, a MDS plot of fish presence/absence data showed a loss of grouping of Ducking and Arakun. However, Felice, Kootowis and April groupings were formed; but, the relationship was weaker (stress = 0.18)(Figure 3.19).

Similar patterns in the MDS plots were found when the fish diversity data was compared at species, genus and family levels; but, the pattern was no longer apparent at the order level. This trend was also seen in the second MDS plot where the similarity matrices of species and genus data were most closely related, the family level was further removed, and the order level was the most different (Figure 3.20).

The MDS plot of the 23 most frequently caught fish species in 2002 superimposed with the ranked cluster groups at a rank of 125 produce 5 clusters (Figures 3.21 and 3.22). The five groups include:

- Sockeye (*Oncorhynchus nerka*)
- Plainfin midshipman (*Porichthys notatus*)
- Three pelagic schooling species, the Pacific sandlance (*Ammodytes hexapterus*), *surf smelt* (*Hypomesus pretiosus*) and Pacific herring (*Clupea pallasi*)
- Coho (*Oncorhynchus kisutch*), Chum (*Oncorhynchus keta*),starry flounder (*Platichthys stellatus*) and English sole(*Parophrys vetulus*)
- All remaining nine fish species

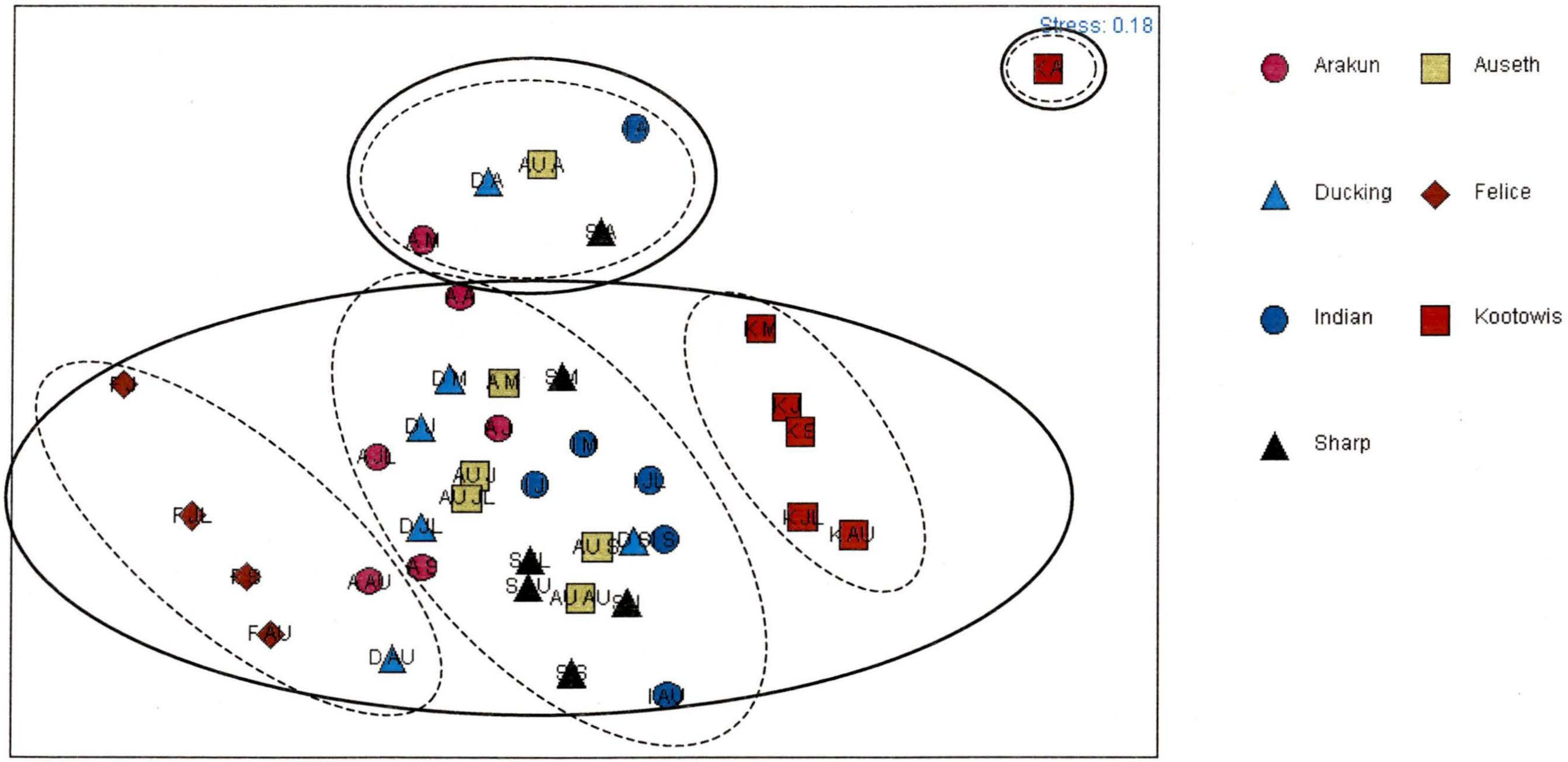

Figure 3.19. MDS ordination for fish species presence/absence at seven eelgrass beds, with data combined from both 2001 and 2002 by month with the superimposed ranked cluster groups. Solid lines indicating 4 clusters formed at a rank of 500 and the dotted lines indicating the 7 clusters formed at a rank of 350 (Site labels: A = Arakun, AU = Auseth, D = Ducking, I = Indian, K = Kootowis, S = Sharp; Month labels: A = April, M = May, J = June, JL = July, AU = August, S = September)..

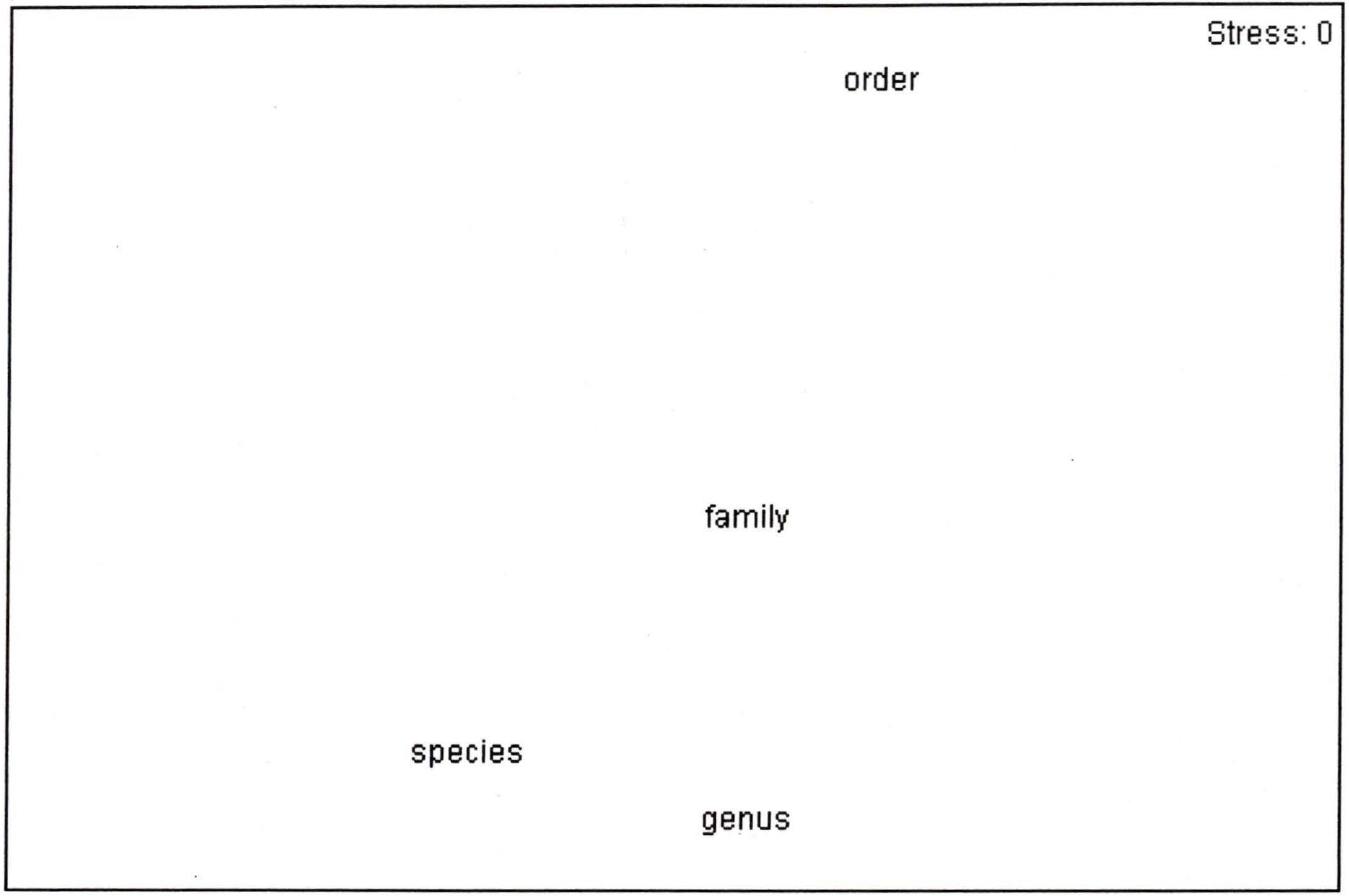

Figure 3.20. Second stage MDS plot of Log(x+1) transformed fish abundance data showing similarity between similarity matrices calculated for decreasing taxonomic resolution: species, genus, family, and order.

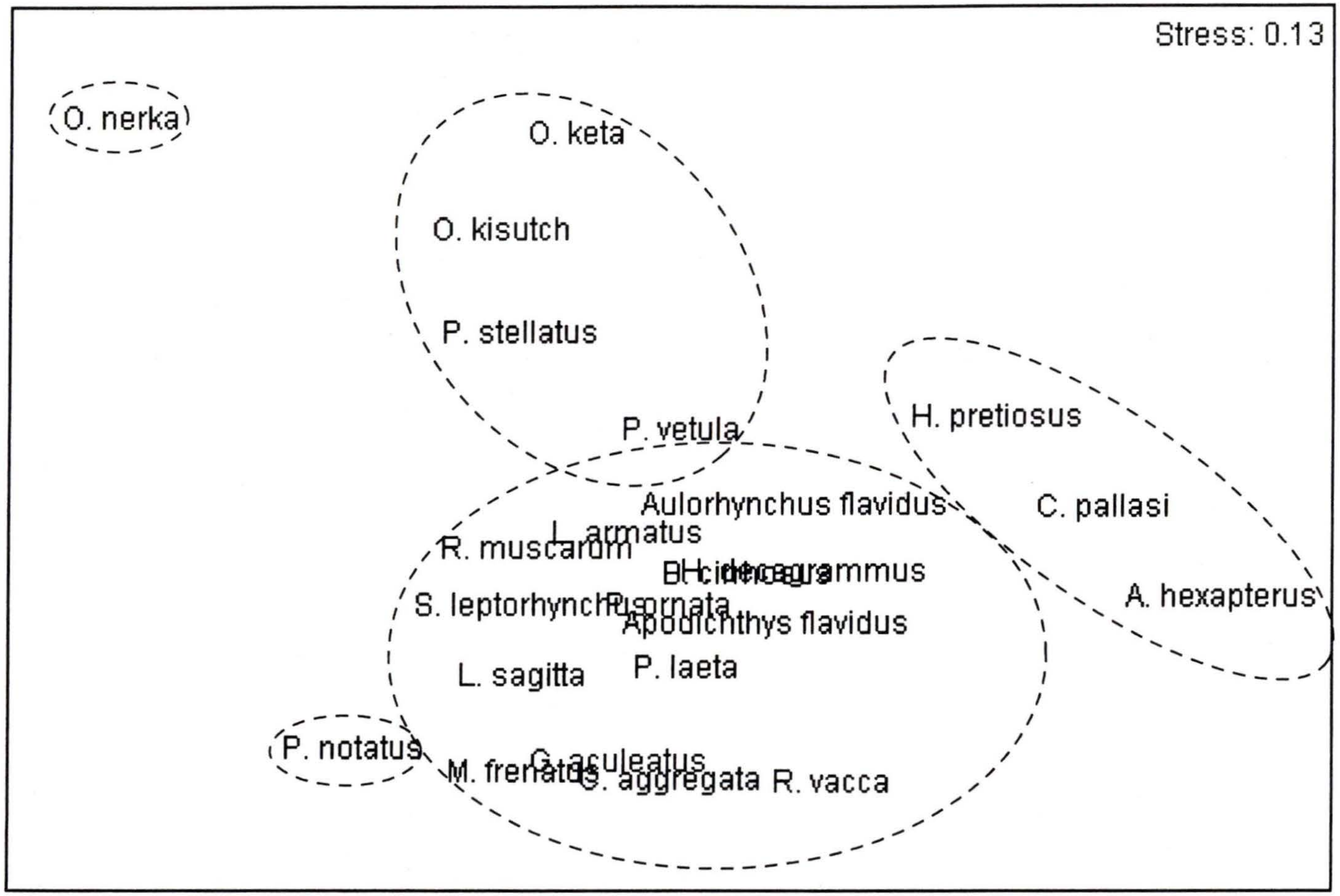

Figure 3.21. MDS plot of the Log(x+1) transformed abundance of the 23 most frequently caught fish species in 2002 superimposed with the ranked cluster groups at a rank of 125 to form 5 species groups.

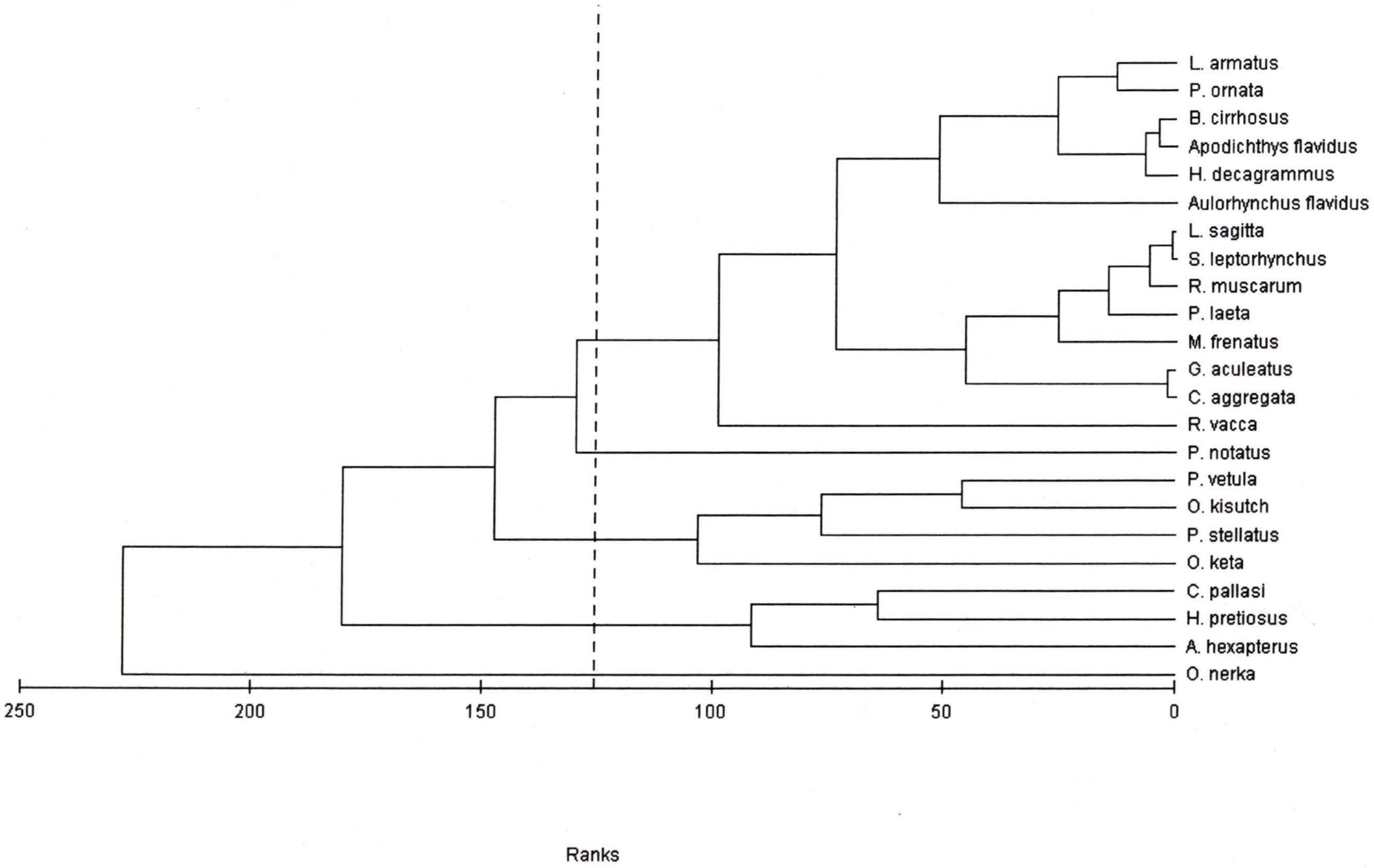

Figure 3.22. Ranked clustered groups for the Log(x+1) transformed abundance of the 23 most frequently caught fish species in 2002. The dotted line indicates the 5 clusters formed at a rank of 125.

A 2-way crossed analysis of similarity (ANOSIM) on Log(x+1) transformed fish species abundance compared site and month differences in 2001 and 2002 (Tables 3.5 and 3.6). The Global *R* values showed that there was a significantly strong difference between data for each site ($R = 0.78$ and $R = 0.78$, $P<0.001$ in 2001 and 2002 respectively) and a significant though slightly lower difference monthly ($R = 0.53$ and $R = 0.56$, $P<0.001$ in 2001 and 2002 respectively). Samples for all sites differed significantly in 2002; however, in 2001, samples for all sites differed significantly except for Felice, and this may be due to lower sample sizes (Appendix 3.1). In both 2001 and 2002, the pair-wise *R* values between sites were high, close to 1, indicating that strong differences existed between sites (Table 3.6). In comparison, monthly differences in both 2001 and 2002 increased significantly in strength, and *R* values were higher, as differences between sampling times increased (Table 3.7).

Table 3.6. ANOSIM analysis of similarity of Log(x+1) transformed fish diversity between each site for 2001 and 2002. Global R = the overall difference between sites, A = Arakun, AU = Auseth, D = Ducking, I = Indian, K = Kootowis, S = Sharp, NS = not significant, NA = not applicable

	2001		2002	
Groups	R statistic	*P*	R statistic	*P*
Global	0.78	<0.001	0.78	<0.001
A, AU	0.67	<0.01	0.65	<0.001
A, D	0.64	<0.001	0.67	<0.001
A, F	NA	NA	0.55	<0.001
A, I	0.83	<0.001	0.95	<0.001
A, K	0.91	<0.001	1.00	<0.001
A, S	0.79	<0.01	0.95	<0.001
AU, D	0.96	<0.001	0.65	<0.001
AU, F	1.00	NS	0.97	<0.001
AU, I	0.72	<0.001	0.57	<0.001
AU, K	0.73	<0.001	0.75	<0.001
AU, S	0.88	<0.001	0.63	<0.001
D, F	1.00	NS	0.90	<0.001
D, I	0.92	<0.001	0.93	<0.001
D, K	0.86	<0.001	0.94	<0.001
D, S	0.93	<0.01	0.90	<0.001
F, I	1.00	NS	1.00	<0.001
F, K	1.00	NS	1.00	<0.001
F, S	NA	NA	1.00	<0.001
I, K	0.78	<0.001	0.86	<0.001
I, S	0.79	<0.001	0.86	<0.001
K, S	0.81	<0.01	0.84	<0.001

Table 3.7. ANOSIM analysis of similarity of Log(x+1) transformed fish diversity between each month for 2001 and 2002. Global = the overall difference between sites, LA = late April, EM = early May, LM = late May, EJ = early June, LJ = late June, EJL = early July, LJL = late July, EAU = early August, LAU = late August, ES = early September, LS = late September, NS = not significant, NA = not applicable

	2001		2002	
Groups	R statistic	*P*	R statistic	*P*
Global	0.530	<0.001	0.56	<0.001
LA, EM	NA	NA	0.53	<0.001
LA, LM	NA	NA	0.85	<0.001
LA, EJ	NA	NA	0.90	<0.001
LA, LJ	NA	NA	0.90	<0.001
LA, EJL	NA	NA	0.88	<0.001
LA, LJL	NA	NA	0.95	<0.001
LA, EAU	NA	NA	0.98	<0.001
LA, LAU	NA	NA	0.98	<0.001
LA, ES	NA	NA	0.97	<0.001
EM, LM	NA	NA	0.38	<0.001
EM, EJ	NA	NA	0.58	<0.001
EM, LJ	NA	NA	0.69	<0.001
EM, EJL	NA	NA	0.70	<0.001
EM, LJL	NA	NA	0.79	<0.001
EM, EAU	NA	NA	0.93	<0.001
EM, LAU	NA	NA	0.98	<0.001
EM, ES	NA	NA	0.96	<0.001
LM, EJ	NA	NA	0.09	NS
LM, LJ	0.33	<0.01	0.42	<0.001
LM, EJL	NA	NA	0.45	<0.001
LM, LJL	0.68	<0.001	0.67	<0.001
LM, EAU	NA	NA	0.76	<0.001
LM, LAU	0.88	<0.05	0.87	<0.001
LM, ES (LS)	0.78	<0.001	0.92	<0.001
EJ, LJ	NA	NA	0.15	NS
EJ, EJL	NA	NA	0.27	<0.01
EJ, LJL	NA	NA	0.57	<0.001
EJ, EAU	NA	NA	0.68	<0.001
EJ, LAU	NA	NA	0.77	<0.001
EJ, LS	NA	NA	0.76	<0.001
LJ, EJL	NA	NA	0.17	<0.05
LJ, LJL	0.34	<0.05	0.56	<0.001
LJ, EAU	NA	NA	0.63	<0.001
LJ, LAU	0.66	<0.001	0.84	<0.001
LJ, ES (LS)	0.62	<0.001	0.75	<0.001
EJL, LJL	NA	NA	0.28	<0.001
EJL, EAU	NA	NA	0.50	<0.001
EJL, LAU	NA	NA	0.72	<0.001
EJL, ES	NA	NA	0.57	<0.001
LJL, EAU	NA	NA	0.31	<0.001
LJL, LAU	0.55	<0.01	0.42	<0.001
LJL, ES (LS)	0.51	<0.01	0.35	<0.001
EAU, LAU	NA	NA	0.09	NS
EAU, ES	NA	NA	0.21	<0.05
LAU, ES (LS)	0.42	<0.01	0.06	NS

3.4. Discussion

An integral component of the management of marine protected areas is the assessment of biodiversity (Yoccoz et al. 2001, Gray 2000). However, the multidimensional nature of biodiversity dictates that a single number or measurement cannot accurately define biodiversity (Purvis and Hector 2000). Therefore, in this study, I evaluated eelgrass fish diversity in Clayoquot Sound by using a variety of measurements to assess biodiversity.

3.4.1. Univariate Diversity Measures

Many studies use the variety of species, richness, as the main measurement for assessing biodiversity (Magurran 1988). Analysis of differences in fish species richness between different types of habitats has provided interpretable trends. Comparisons between vegetated and non-vegetated habitats have demonstrated consistently higher fish species richness in vegetated habitats (Lubbers et al. 1990, Edgar and Shaw 1995a, Gary et al. 1998). In contrast, clear differences between fish species richness within the same habitat are not consistent across studies (Sogard et al. 1989a, Bell et al. 1994, Hovel et al. 2002). In this study, fish species richness followed no apparent trend across eelgrass beds nor were monthly trends interpretable (Figure 3.1). This lack of an interpretable trend in species richness within eelgrass beds emphasizes that differences in eelgrass fish diversity cannot be explained species richness alone.

By also examining fish abundance and evenness, spatial and temporal differences between fish assemblages were made apparent. However, spatial differences in fish diversity between eelgrass beds were difficult to interpret by examining only univariate measurements. Arakun data, for example, had lower species richness and abundance, and higher evenness than at many of the other sites. Determining the processes structuring fish diversity is critical for interpreting spatial patterns in eelgrass fish communities (see chapter 4).

Temporal trends in richness, abundance and evenness were highly variable between sites from April to July, and all sites followed similar trends in August and September. Those trends were driven by an increase in young of the year (YOY) *Cymatogaster aggregatta* and *Gasterosteus aculeatus* that were recruited into the beds in July (Figures 3.3 and 3.4). The stabilization of fish communities across sites in August and September is an important component in developing monitoring programs, such as indices of biotic integrity (Deegan et al. 1997). Overall, univariate measures of diversity demonstrated interpretable temporal patterns in fish communities, but spatial patterns in fish diversity require further investigation.

3.4.2. Taxonomic Diversity and Distinctness

Examining the taxonomic relatedness within a community is an important component to assess biodiversity (Purvis and Hector 2000, Clarke and Warwick 2001). Vane-Wright et al. (1991) have suggested that conservation of taxonomically distinct groups should be a higher priority than conserving species. In this study, taxonomic diversity did not provide any new insight into explaining eelgrass fish diversity, as similar trends were found in both evenness and abundance measures (Table 3.2). Hall and Greenstreet (1998) also found taxonomic diversity to follow similar trends as evenness, and suggested that perturbations structuring fish communities were large enough to be detected by both conventional and taxonomic diversity measures. Similarly, temporal shifts in fish diversity, in this study, may also be large enough to be detected by conventional diversity measures, and evaluation of taxonomic diversity is not necessary.

In contrast, measurement of taxonomic distinctness (Δ^*) did provide new insight into interpretation of biodiversity. Unlike other univariate diversity measures, Δ^* was highly variable between sites in April, August and September, and the fish communities were more similar between sites from May to July (Figure 3.10). In April 2002, the Kootowis fish community displayed significantly lower Δ^*, driven by the higher abundance of salmonid species at this site than all the other sites (Figure 3.7), a difference not apparent using the other diversity measures. In addition, the high variability in Δ^* between sites in August and September indicate differential use of eelgrass beds by fish species. For example, at Kootowis and Sharp YOY *Cymatogaster aggregatta* and *Gasterosteus aculeatus* remained in those beds longer than other beds, suggesting juveniles preferentially selecting environmental features found only within those beds. Taxonomic distinctness provided insight into local scale, spatial differences in eelgrass fish communities, by interpreting the fish diversity at a higher degree of resolution (Rogers et al. 1999).

Variation in taxonomic distinctness, another biodiversity index that explores the evenness of the taxonomic tree, was also used to assess eelgrass fish diversity (Clarke and Warwick 2001). In this study, fish communities sampled in 2001 at Ducking in May and Kootowis in June fell outside of the expected values for the Clayoquot Sound fish species list (Figure 3.11). At Ducking only one surfperch species, the shiner perch, was caught which influenced the lower than expected variation in taxonomic distinctness. In comparison, the four salmonids species caught in June at Kootowis were responsible for the higher than expected variation in taxonomic distinctness values. Overall, all other site-month samples fell within the expected 95% confidence envelopes, indicating that the fish diversity of each bed fell within the expected average taxonomic distinctness for the Clayoquot Sound region. In addition, fish diversity sampled at Kootowis in April and early May of 2002 did have slightly higher variation in taxonomic distinctness than other sites, suggesting a trend driven by preferential use of this bed by migrating salmonids, *Oncorhynchus nerka* and *Oncorhynchus keta* (Figure 3.7).

3.4.3. Beta Diversity

Species turnover was also examined across temporal and spatial gradients, using both taxonomic similarity and percent similarity among beds to determine the heterogeneity of eelgrass habitat supporting fish diversity (Hull 1999, Kendall and Widdicombe 1999, Izsak and Price 2001). Taxonomic similarity and percent similarity were found to decrease as distance between samples increased and a similar trend was found temporally, where beta diversity increased overtime. The exploration of beta diversity demonstrated the heterogeneous nature of eelgrass beds in supporting fish diversity in Clayoquot Sound.

3.4.4. Multivariate Diversity Measures

Site-month ordination and clustering techniques showed fish species abundance to be more similar within sites than seasonally, with the exception of data for April (Figure 3.17). Unlike the univariate diversity measures, these results demonstrate the higher sensitivity of multivariate techniques in detecting spatial change in eelgrass fish assemblages. Additionally, ordination plots provided a clear visualization of which eelgrass fish communities were more similar among sites. Similarly, other studies (Clark 1997, Young et al. 1999) found that using multivariate ordination techniques also provided additional interpretation of the entire fish community, that univariate measures alone could not provide.

Temporal ordination patterns demonstrated that April supported a distinct fish assemblage across sites indicates a seasonal shift in eelgrass fish community structure from April to May, supporting the observation of seasonal migration of fish species out of eelgrass habitat in the winter and back in the spring (Jackson et al. 2001). Further sampling into the fall may show a similar, distinct, seasonal shift in community structure as fish species migrate out of the beds. In this study, late September was not late enough to detect such emigration. Spatial and temporal patterns present in ordination and cluster plots were also confirmed by the ANOSIM results. Overall, fish species abundance was found to be more dissimilar among eelgrass beds than within, and temporal fish species abundance similarity decreased as time between samples increased.

By examining fish diversity at different levels of taxonomic resolution, including species presence/absence, genus, family and order, I determined the effect increasing taxonomic level has on the temporal and spatial fish diversity patterns in eelgrass beds in

Clayoquot Sound. Analysis of fish species presence/absence produced similar seasonal trends; however, spatial patterns in fish diversity differed, and new trends emerged, such as all Kootowis samples forming a group (Figure 3.19). These results suggest that presence-absence data reflected the taxonomic distinctness of the Kootowis fish assemblage; however, subtle site differences were lost, demonstrating the importance of collecting both species and their numbers to assess differences between eelgrass fish assemblages. Ordination of different taxonomic levels fish data produced similar results to Vanderklift et al. (1998), where similar temporal and spatial patterns occurred up to the family level, but were lost at the level of order. Many of the fish species patterns were still represented at a family level, which reflects the important role fish family groups play in structuring eelgrass fish communities in Clayoquot Sound.

Five discrete fish groups were formed from the 23 most abundant eelgrass fish species caught in 2002:

- *Portichthys notatus* and *Oncorhynchus nerka* each formed a single species group. *P. notatus* was the only species caught consistently, in relatively high numbers in the two eelgrass beds Sharp and Kootowis, the substrate at both these sites consists of fine mud substrate (see chapter 4), a preferred habitat for this burrowing species (Hart 1973). In comparison, *O. nerka* was caught in high numbers, approximately 300 individuals in each seine haul, in April at the Kootowis eelgrass bed. The sockeye salmon (*O. nerka*) spends a very short time in nearshore habitats in early spring on its seaward migration compared to other salmonids species (Groot and Margolis 1991). In addition, Kootowis, in this study, is the closest eelgrass bed to the outflow of the lower Kennedy River, where that population of sockeye salmon originates.

- Another fish group formed consisted of three forage fish species: *Ammodytes hexapterus, Hypomesus pretiosus* and *Clupea pallasi*. These schooling species do use eelgrass beds exclusively but as temporary foraging habitat and protection from predation (Murphy et al. 2000).
- Another group consisted of four fish species: *Oncorhynchus kisutch, Oncorhynchus keta, Platichthys stellatus* and *Parophrys vetulus*. Each of these species were caught in most of the eelgrass beds sampled in relatively lower numbers. However, *O. keta* clustered out first from the group as this salmon species was only caught in late spring and migrates out to the ocean by the end of May, whereas the other three species continued to be caught consistently throughout the remaining summer months.
- The remaining 14 species form a final large cluster. All 14 fish species were caught in all 7 eelgrass beds, with the exception of three species not caught at Felice (*Syngnathus leptorhynchus, Lumpenus sagitta* and *Rimicola muscarum*) and two species not caught at Kootowis (*Apodichthys flavidus* and *Blepsias cirrhosus*).

The fish assemblage structure also provides potential explanation for some of the temporal and spatial patterns; for example, the presence of large numbers of salmonids driving temporal patterns in April, and the high abundance of the forage fish, *A. hexapterus* contributing to the grouping of all the Felice samples. Further exploration of preference of these common fish species for different habitat qualities may determine which environmental factors are driving the spatial differences seen in eelgrass fish assemblages.

3.4.5. Recommendations and Conclusions

This is the first study examining eelgrass fish diversity by calculating both conventional univariate measures of biodiversity, such as species richness, abundance and evenness, as well as looking at taxonomic relatedness and multivariate measurements. Overall, univariate and multivariate diversity measures used in unison provided a clearer depiction of spatial and temporal patterns in eelgrass fish communities. Some measures, however, were more useful than others at detecting spatial and/or temporal patterns (Tables 3.8).

Table 3.8. Summary of biodiversity indicators and the significant spatial and temporal differences in eelgrass fish diversity measured by these indicators.

Biodiversity indicator	**Spatial differences in biodiversity (Site)**	**Temporal differences in biodiversity (Month)**	**Both spatial and temporal differences in biodiversity**
Species richness			X
Fish abundance			X
Pielou's evennes			X
Taxonomic diversity			X
Taxonomic distinctness	X		
Taxonomic similarity			X
MDS of Log(x+1) transformed fish species abundance			X
MDS of fish species presence/ absence	X		

The conventional use of species richness as measurement of biodiversity in this study provided no interpretable comparison of spatial and temporal patterns in eelgrass fish, thereby highlighting the importance of exploring additional measures of biodiversity. Nonetheless, I recommend continuing to use species richness in conjunction with other diversity measures because it is frequently used, and can be compared to other studies (Table 3.1).

Abundance and evenness measurements emphasized temporal changes in fish community structure and the importance of eelgrass beds as nursery habitat. Additionally, taxonomic diversity followed similar trends as fish abundance and evenness (Table 3.2). Therefore, it is important to collect both species and abundance data. I also recommend the use of a single measure, total fish abundance, rather than also evaluating evenness and taxonomic. Total fish abundance alone would adequately detect temporal changes in eelgrass fish communities.

Taxonomic distinctness was also important to examine how fish communities differentially utilize eelgrass beds. Spatial variability of taxonomic distinctness demonstrated that Kootowis fish community data was more taxonomically distinct than all other sites. Consequently, evaluating taxonomic distinctness is essential to detect subtle spatial differences in fish communities that are not apparent by only using conventional univariate measures.

In addition to alpha diversity measurements, beta diversity was also measured. Beta diversity measures demonstrated how fish species and taxonomic similarity decreased as distance increases. Taxonomic similarity measures over time also demonstrated how fish community differences are greater as the time between sampling periods increases. Spatial and temporal heterogeneity of eelgrass beds contributing to fish diversity was determined by comparing taxonomic similarity among beds. Subsequently, it is important to assess beta diversity measures, in addition to alpha diversity measures, to determine the heterogeneity of eelgrass beds as it relates to fish diversity.

Finally multivariate measures highlight spatial patterns in fish diversity not detected by univariate measures, emphasizing the need to use multivariate techniques to detect subtle changes in community structure. MDS ordination plots provided interpretable visualization of how fish communities are similar among eelgrass beds. In addition, fish species abundance similarity matrices provided clearer site distinction than just using species presence/absence data.

In conclusion, detecting and interpreting spatial and temporal patterns in marine biodiversity are critical components of conservation (Levin 1992). To adequately assess patterns in biodiversity a suite of measures that accurately evaluate diversity must be determined. This chapter evaluated a multitude of biodiversity measures and recommended a suite of measures to assess eelgrass fish biodiversity. Application of those recommended univariate and multivariate diversity measures is essential for conservation planning to examine and understand the complex nature of biodiversity.

Chapter 4: Effect of Hydrodynamic Energy on Fish Community Structure

4.1. Introduction

Biogeography involves determining patterns of biological communities, the processes driving these patterns and applying these concepts to understanding and conserving ecosystems (Brown et al. 1996). In this study fish community patterns exhibited variability across eelgrass beds (see chapter 3). Consequently, determining the processes driving these differences in eelgrass fish communities is critical for conservation and management of eelgrass ecosystems.

Hydrodynamic setting influences the spatial arrangement of seagrass habitats (Fonseca and Bell 1998). Seagrass beds distributed in high energy areas are more patchy and low energy beds are more continuous (Murphey and Fonseca 1995). This variability in seagrass landscapes increases habitat heterogeneity and directly influences the faunal communities it supports (Fonseca et al. 1983).

Changing spatial configurations of seagrass habitat effects patterns in faunal community structure and abundance (Frost et al. 1999, Hovel et al. 2002). The influence of bed spatial structure on macrofaunal invertebrates have shown decreased survival in smaller seagrass patches and areas of low seagrass cover (Irlandi et al. 1996, Irlandi 1997). Additionally, higher hydrodynamic energy within seagrass beds has been directly correlated with decreasing faunal abundance (Hovel et al. 2002). However, few studies have examined the influence of hydrodynamic energy on seagrass fish communities (Bell et al. 2001, Hovel et al. 2002). Bell et al. (2001) found both fish species richness and abundance to increase in lower energy seagrass beds. A similar trend was found in sandy habitats, where fish abundance was negatively correlated with higher wave exposure

(Clark 1997). Finally, seagrass beds have been well documented as valuable nursery habitat (Jackson et al. 2001), but no study has examined the influence of hydrodynamic energy on structuring fish length and age class.

The objective of this chapter was to determine the influence of hydrodynamic setting on fish community properties. Variables previously used to assess hydrodynamic energy in seagrass beds include sediment silt-clay fraction, tidal current, wind and wave exposure, temperature and eelgrass percent cover (Irlandi 1996, Fonseca and Bell 1998, Hovel et al. 2002). Similarly in this study the hydrodynamic setting of the eelgrass beds was evaluated using physical and eelgrass variables that directly and indirectly measure hydrodynamic energy. Previous studies have examined hydrodynamic effects on seagrass fish by applying univariate diversity measures (Bell et al. 2001), but few studies have also applied multivariate techniques (Hovel et al. 2002). Hence, in this study fish community properties were examined among eelgrass beds by using univariate and multivariate diversity measures.

Fish community properties examined included fish lengths, age class, species richness, abundance, taxonomic distinctness and multivariate measures. Expected fish community trends among beds included, greater length fish would be caught in higher energy eelgrass beds and a higher number of young of the year fish would be caught in lower energy beds. Fish species richness, abundance and taxonomic distinctness were expected to be greater at the lower energy beds. Lastly, multivariate analysis of fish communities would demonstrate beds with similar hydrodynamic influences would have similar fish communities.

4.2. Methods

4.2.1. Study Location

In southern Clayoquot Sound six eelgrass beds were selected according to their exposure to hydrodynamic energy. Hydrodynamic energy was evaluated by examining the potential exposure these beds received from tidal currents, wind and waves. Therefore eelgrass beds exposed to higher hydrodynamic energy had higher currents and wind-wave exposure compared to lower energy beds.

Eelgrass beds were initially selected by examining direct fetch working from the Canadian Hydrographic Service nautical chart 3673. Paired high and low energy beds were chosen in three areas in southern Clayoquot Sound: Lemmens Inlet, Grice Bay and Browning Passage. In Lemmens Inlet, Arakun was categorized as high energy and Sharp was low. In Grice Bay, Auseth was categorized as high and Kootowis was categorized as low energy bed. Field surveys determined that no adequate low energy bed was available to pair with Ducking in Browning Passage, but this site was kept as an intermediate bed between the two other areas. In addition, the Indian site was selected as it is one of the largest beds within Grice Bay and its channel locality complimented the channel component of Ducking (Chapter 2, Figure 2.1). The characteristics of these six eelgrass beds are summarised in chapter 2 (Table 2.2).

4.2.2. Sampling Dates

Each eelgrass bed was sampled in August 2001, May 2002 and August 2002 for fish diversity and hydrodynamic variables (Appendix 4.1). Hydrodynamic variables measured included a relative exposure index, sediment silt-clay fraction, maximum tidal current, water temperature, and eelgrass percent cover. Due to equipment and time limitations sediment samples were only taken in August 2001, and current was only measured in May 2002.

4.2.3. Hydrodynamic Regime

The hydrodynamic energy of each site was determined through the examination of both direct and indirect measures. Direct measures of hydrodynamic energy used in this study included tidal current and a relative exposure index calculation. Previous research has shown both tidal currents and wind-wave dynamics to play important roles structure seagrass beds (Fonseca and Bell 1998, Turner et al. 1999). Indirect measures of hydrodynamic energy included sediment silt-clay fraction, water temperature, and eelgrass percent cover. Sediment composition indirectly measured fluid energy, where course, sandy sediment indicated high energy, and fine silty sediment indicated low energy (Fonseca et al. 1983). In addition, areas with high water turnover usually have lower water temperatures (Koch and Gust 1999). Lastly, hydrodynamic energy also effects the spatial arrangement of seagrass habitats (Fonseca and Bell 1998). Murphey and Fonseca (1995) found high energy beds increased in patchiness and low energy beds were more continuous, therefore eelgrass percent cover was also calculated.

4.2.3.1. Direct Hydrodynamic Measures

4.2.3.1.1. Tidal Current

To acquire a maximum tidal current measurement at each site, tidal currents were measured using a Sensa RC2© electro-magnetic water velocity meter at each site during the peak spring falling tide in May 2002. Current measurements were taken over three days, as the distance between each site was too great to measure all sites on the same day. Four replicate current measurements were taken every half an hour from slack low tide to slack high tide. However, fewer measurements were taken at the Lemmens Inlet and Grice Bay beds because of the increased travel distance between sites. Each measurement was taken approximately 50cm above the eelgrass canopy to avoid any influence of surface friction caused by the eelgrass (Irlandi 1996). Mean current velocity taken over one minute was used as a single measurement. Maximum tidal current was determined for each site as the peak tidal current measured during the 6 hour sampling window.

4.2.3.1.2. Relative Exposure Index

The relative exposure index (REI) used in this study, incorporated maximum wind speed, direction and effective fetch. Keddy (1982) developed this method to measure the effect of wind-wave dynamics have on aquatic macrophytes. Murphey and Fonseca (1995) applied this REI calculation to seagrass beds and their methodology was used in this study:

$$\mathrm{REI} = \sum_{i=1}^{8} (V_i \times P_i \times F_i)$$

where i = ith compass bearing (1 to 8: N, NE, E, SE, S, SW, W, NW), V = mean monthly maximum wind velocity in m/s, P = percent frequency wind occurred in the ith direction and F = effective fetch (m).

Hourly daytime wind velocity and directional data was acquired from Environment Canada's weather station archives for Tofino airport, located within 15km of all study sites. Only exceedance winds (>7m/s), wind speeds that exceeded 95% of wind data for the study period, were used in the relative exposure index (REI) calculation (Keddy 1982). Therefore the maximum wind speed (V) was the grand mean of all exceedance wind velocities for the month of interest. Wind velocities were used from the same month and year that the fish communities were sampled in.

Fetch was defined as the distance over water along a compass bearing from each site to land (U.S. Army Coastal Engineering Research Center 1984). Effective fetch takes into account shoreline irregularities by weighting multiple fetch measurements (Keddy 1982). Therefore effective fetch was calculated for each site by overlaying a transparency with lines radiating from the centre at 11.25° increments on 1:40,000 scale nautical chart. For each of the eight compass bearings five measurements at 11.25° increments on either side of the compass bearing were recorded in metres. The effective fetch for that compass bearing was the average of these five fetch values.

Water depth also influences the effect of wind-wave interactions (U.S. Army Coastal Engineering Research Center 1984). Therefore the maximum tidal depth at a 0m tidal height was recorded for each site on the same day as tidal current was measured. Little variation was found between the six sites and therefore was not included as a variable in this study (Arakun = 3.66m, Auseth = 3.45m, Ducking = 2.90m, Indian = 3.75m, Kootowis = 3.41m, Sharp = 3.18m).

4.2.3.2. Indirect Hydrodynamic Measures

4.2.3.2.1. Sediment Analysis

In August 2001, four replicate surface cores, 5.08cm in diameter and 5cm deep were taken at each site. Each core was taken within the eelgrass bed with approximately 10m separating each sample. The samples were placed in Ziplock bags and put immediately on ice and later frozen. Particle size of the sediment was analysed within two weeks of collection.

The silt-clay fraction was determined by wet sieving the sample through a 63µm mesh sieve. Wet sieving was used to avoid amalgamation of the clay particles (Buchanan 1984). The silt/clay fraction and the sediment remaining in the 63µm mesh sieve were dried separately for 24 hours at 100°C. The sediment was removed from the oven after 24 hours of drying and the dried sediment was again sieved through a 63µm mesh sieve to ensure removal of the entire silt-clay fraction.

4.2.3.2.2. Water Temperature

Temperature was measured 0.5m from the water's surface using an YS1 handheld meter in the eelgrass sites at the same time as it was beach seined. In June 2001 a temperature logger was also placed subtidally at each of the six sites and recorded hourly temperature readings. For each fish sampling period temperature values from a two hour window around the mean daytime high tide were used from the same day the fish were sampled. However, temperature logger data was not available for Ducking in August 2001.Therefore, point samples taken with the probe were used. In addition, there were no values for early May for Arakun, Auseth, Ducking, Indian and Sharp because the loggers were removed, therefore temperature values from late May were used (Appendix 4.1).

4.2.3.2.3. Eelgrass Percent Cover

The methodology used to determine bed patchiness at each site was modelled after research conducted by Murphey and Fonseca (1995). For each site three 10 X 10m plots were sampled for presence and absence of eelgrass, with each replicate separated by ten metres. A $100m^2$ plot area was chosen because the area the beach seine was pulled through was approximately $100m^2$. All plots began at approximately 0m tidal height and therefore all plots were subtidal. The 0m tidal height was marked at each site by setting an anchor adjacent to the permanent temperature loggers on a previous day at low tide. This ensured that the depth of the bed being sampled was the same as that which was beach seined for fish diversity. Compass bearings were used to ensure direction and area of plots. One hundred $1.0m^2$ sections were visually inspected for presence or absence of eelgrass by flipping a $1.0m^2$ quadrat in the 10 X 10 m subsample plot in August 2001. In May and August 2002 percent cover was recorded in each $1.0m^2$ quadrat by ranking the area on a scale of 0 to 4, where 0 = eelgrass absent, 1 = 25% eelgrass cover, 2 = 50% eelgrass cover, 3 = 75% eelgrass cover, and 4 = 100% eelgrass cover (Figure 4.1). By recording a range of percent cover in 2002 more fine scale differences between sites could be explored.

Shore (Anchor)

1.0 m	4	4	4	4	4	4	4	4	4	4
2.0 m	4	4	4	4	4	4	4	4	4	4
3.0 m	4	4	4	4	4	4	4	4	4	4
4.0 m	4	4	4	4	4	4	4	4	4	4
5.0 m	3	4	3	3	3	3	3	3	3	3
6.0 m	3	4	3	3	3	4	2	2	2	2
7.0 m	1	4	3	3	3	4	2	4	4	4
8.0 m	1	1	3	3	3	4	2	4	4	4
9.0 m	1	1	1	1	0	0	2	2	2	2
10.0 m	1	1	0	0	0	0	2	1	1	1

Subtidal

Legend
0 = eelgrass absent
1 = 25% eelgrass cover
2 = 50% eelgrass cover
3 = 75% eelgrass cover
4 = 100% eelgrass cover

Figure 4.1. Example of a 10 X 10m plot taken in May and August 2002, where each square represents a 1.0m^2 area, and the shore is at the 0 m tidal height.

4.2.4. Fish Sampling Methodology

Six eelgrass beds were sampled using triplicate beach seine sets (Section 2.2.2, chapter 2) in two hour window before and after a daytime mean low tide of 0.6m or less. Fish were sampled from early spring to early fall, monthly in 2001 and bimonthly in 2002. There was greater variability in sampling times in 2001, as not all sites were selected and storms in August prevented sampling at some sites. Chapter 3 results indicate that temporally fish diversity was more similar between adjacent sampling periods. Therefore, in 2001 sets done in August and September were used for analysis with August 2001 data. Whereas, in 2002 sets from early May to late June were used for May 2002 and early August to early September were used for August 2002 analysis with eelgrass and environmental parameters (Appendix 4.1).

4.2.5. Site Ranking Along a Hydrodynamic Energy Gradient

Each site was positioned along a hydrodynamic gradient by ranking both direct and indirect hydrodynamic measures. Relative exposure index, tidal current, silt-clay fraction, water temperature, and eelgrass percent cover were ranked from one to six; one indicated high energy and six indicated low energy. If two sites ranked equally a mean rank value was given to each of the sites. A total rank value for each site was calculated as the mean of the rank values of the five hydrodynamic variables. The total rank value was then used to place each site along the hydrodynamic gradient.

4.2.6. Data Analysis

4.2.6.1. Univariate Analysis

Each site was characterized according to a hydrodynamic energy regime by examining direct (maximum tidal current, relative exposure index) and indirect measures (sediment silt-clay fraction, water temperature, eelgrass percent cover) of hydrodynamic energy. Among site differences for each parameter were analyzed separately using a one-factor analysis of variance (ANOVA), where location was the independent factor. If means differed significantly Tukey's test was performed to determine where the differences occurred. If homogeneity of variance was violated among site differences were analyzed using the Kruskal-Wallis test (Zar 1999). Significant among site comparisons were then made using the Mann-Whitney test and the Bonferroni correction was applied for multiple comparisons (Sokal and Rohlf 1995).

In May and August 2002, separate among site analysis compared the mean fork length of the four most common fish species: shiner perch (*Cymatogaster aggregata*), threespine stickleback (*Gasterosteus aculeatus*), crescent gunnel (*Pholis laeta*), and staghorn sculpin (*Leptocottus armatus*). These four species were examined because they were the only species caught consistently at the six eelgrass beds.

Age class evaluation was only examined for shiner perch and threespine sticklebacks because young of the year (YOY) and one plus year old stages were identifiable and consistently caught. Shiner perch age classes were compared in May and August 2002. In comparison, threespine stickleback age classes were only examined in August because YOY sticklebacks were not caught until July (see chapter 3, Figure 3.3). An ANOVA or a non-parametric Kruskal-Wallis test was performed to test for significant differences among sites. All univariate analysis was done using SPSS version 10.0.

4.2.6.2. Multivariate Analysis

The effect hydrodynamic variables have on structuring fish communities were examined using the BIOENV procedure (Figure 4.2). BIOENV is a non-parametric procedure that generates a subset of environmental variables by finding the best match of similarities between separate biotic and abiotic matrices (Clarke and Warwick 1994). In this study the biotic matrices calculated used two types of fish community data: fish: species presence/absence and Log(x+1) transformed fish species abundance. The abiotic matrix was composed of hydrodynamic variables from three separate sampling periods: August 2001, May 2002 and August 2002. All correlations between hydrodynamic variables were also less than 0.7 and therefore all variables were included in the analysis (Clarke and Warwick 1994). Additionally, different hydrodynamic variables were analysed for each sampling period due to discrepancies between data collection

(Appendix 4.1). Analysis for the three sampling dates was also kept separate. In August 2001, variables analysed included sediment silt-clay fraction, relative exposure index, water temperature and eelgrass percent cover. Whereas in May 2002,variables included tidal current, relative exposure index, water temperature and eelgrass percent cover. Lastly, in August 2002, temperature, relative exposure index, water temperature and eelgrass percent cover were analysed.

Overall, separate Bray-Curtis similarity matrices were calculated for the two fish data forms and Euclidean distance was used to compare hydrodynamic variables. Fish community and hydrodynamic variable similarity matrices were compared using weighted Spearman rank correlation coefficients (ρ_w) to determine which hydrodynamic variables best correlated with the fish data. The fewest hydrodynamic variables that generated the highest ρ_w value were ordinated using 2-D multidimensional scaling (MDS). Hydrodynamic variable ordinations were then compared for concordance with the fish community ordinations. Similarity matrices were also clustered using group-average cluster on ranked similarity and clusters were overlaid onto the MDS plots (Clarke and Warwick 1994). A further confirmatory analysis, a RELATE test was performed to compare the fish data and the reduced set of hydrodynamic variables. This procedure tests for significant similarity between the fish similarity matrix and the hydrodynamic variables similarity matrix. BIOENV, MDS, and RELATE calculations were implemented in the PRIMER program version 5.2.8.

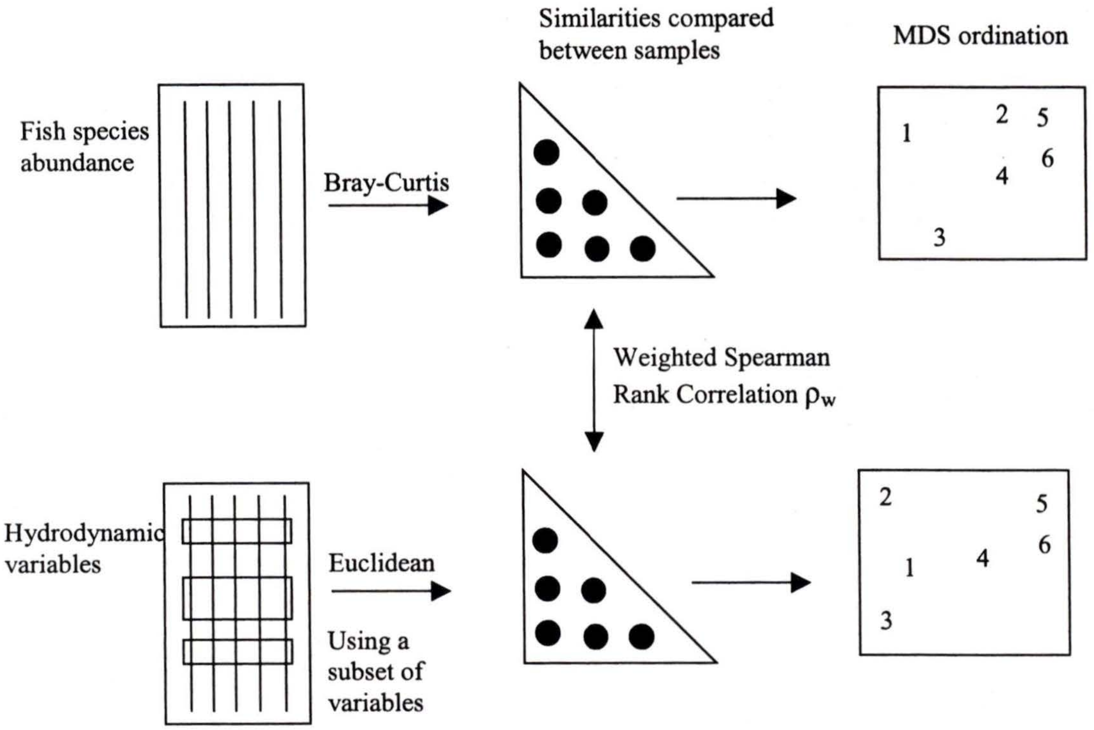

Figure 4.2. BIOENV procedure demonstrating the selection of a subset of hydrodynamic variables that gives the highest weighted rank correlation coefficient between the fish species and hydrodynamic variables similarity matrices (adapted from Clark and Warwick 1994).

4.3. Results

4.3.1. Direct Hydrodynamic Measures

The relative exposure index (REI) did not differ significantly between sites for each of the three sampling periods. However, all sites followed similar trends between sampling periods (Figure 4.3). Ducking and Indian ranked highest indicating high energy. Auseth and Arakun had intermediate rank values and Kootowis and Sharp ranked lowest, indicating low energy (Table 4.1).

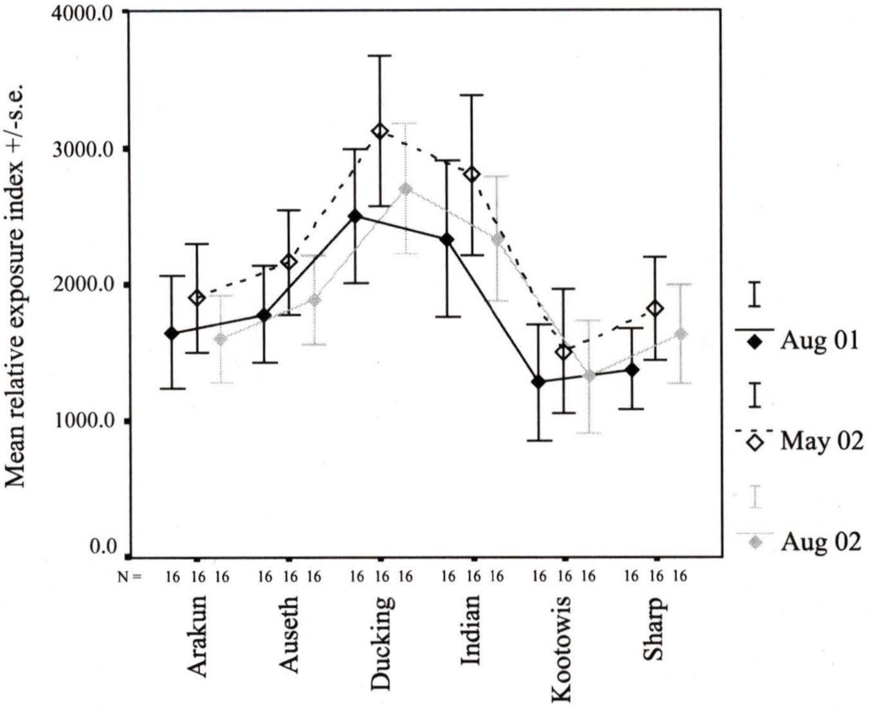

Figure 4.3. Mean relative exposure index (REI) at six eelgrass sites over three sampling periods.

Table 4.1. Rank values of each hydrodynamic variable for each site (1=high energy, 6=low energy). The total rank value is the average of the five hydrodynamic variables rank values for each site.

Site	REI	Current	Silt-clay fraction	Temperature	% Eelgrass cover	Total rank
Arakun	4	1	1	1	2	1.8
Auseth	3	4	2	3.5	3	3.1
Ducking	1	3	3	3.5	1	2.3
Indian	2	2	5	5.5	6	4.1
Kootowis	6	6	4	2	4	4.4
Sharp	5	5	6	5.5	5	5.1

Maximum tidal current differed significantly between sites in May 2002 (Kruskal-Wallis, df=5, $H = 115.490$, $P<0.001$) (Figure 4.4, Appendix 4.2). Overall Arakun had the highest tidal current, and thereby received the highest rank value. The next highest ranking was Indian, followed by Ducking, Auseth, and Sharp. Finally, the lowest rank value indicating the bed with the lowest energy was Kootowis (Table 4.1).

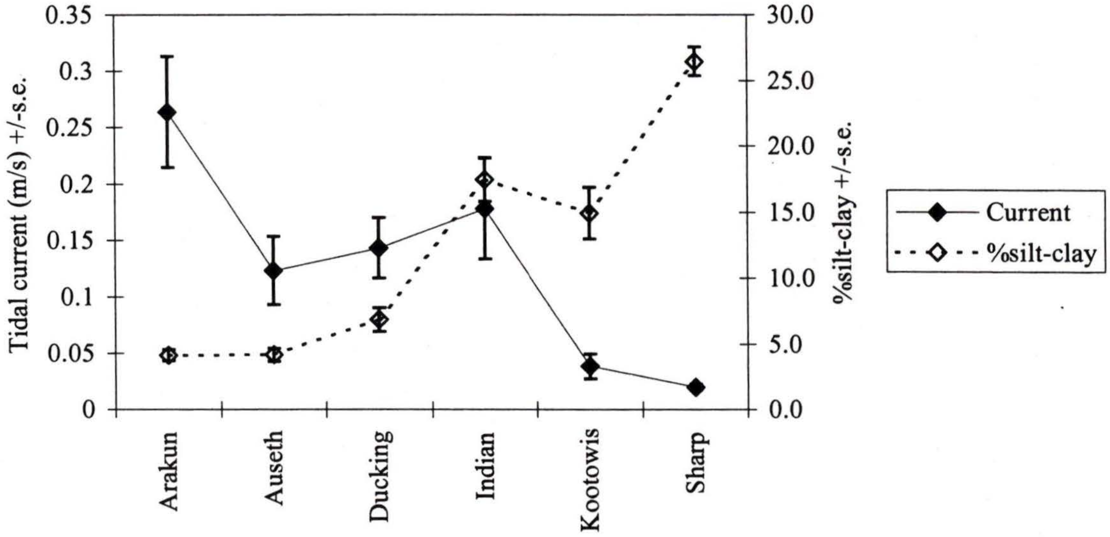

Figure 4.4. Mean tidal current and sediment silt-clay fraction at six eelgrass sites, where current was measured in May 2002 and %silt-clay in August 2001.

4.3.2. Indirect Hydrodynamic Measures

Sediment silt-clay fractions differed significantly between sites in August 2001 (ANOVA, $F_{5,18} = 52.940$, $P<0.001$) (Figure 4.4). Site comparisons ranked Arakun as high energy, followed by Auseth, Ducking, Kootowis, Indian and lastly Sharp being the lowest energy bed (Table 4.1, Appendix 4.2).

Mean water temperature differed significantly between sites for all three sampling periods (August 2001, Kruskal-Wallis, df=5, H=16.300, P<0.01; May 2002, ANOVA, $F_{5,72}$ = 4.499, P<0.001; August 2002, Kruskal-Wallis, df=5, H=35.277, P<0.001) (Figure 4.5). Overall, Arakun ranked as having the highest energy, followed by Kootowis. Next Auseth and Ducking ranked equally as beds of intermediate energy, and Indian and Sharp ranked equally as low energy beds (Table 4.1, Appendix 4.2).

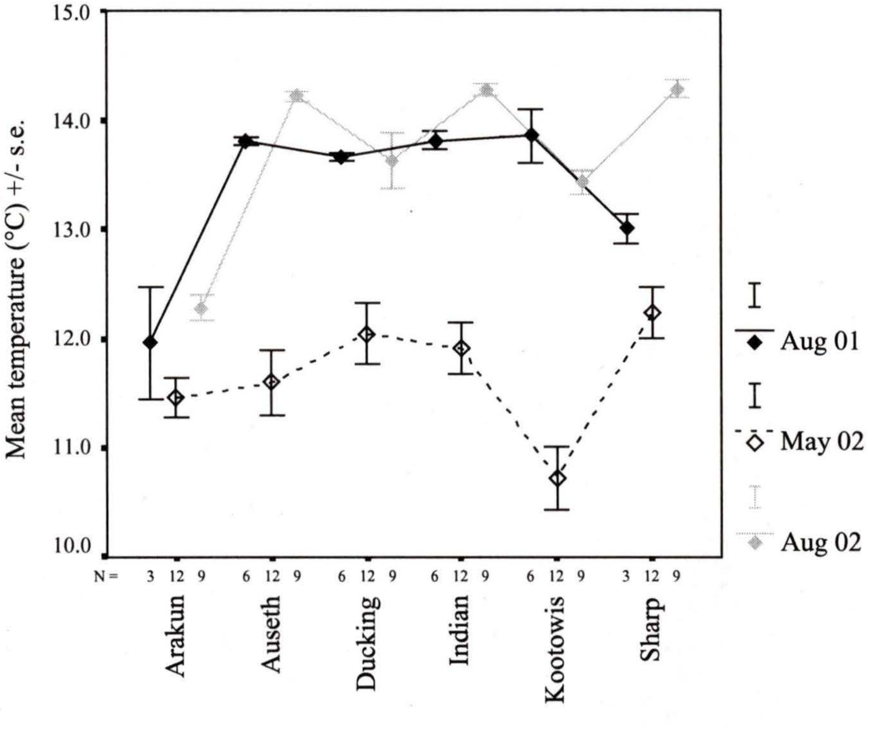

Figure 4.5. Mean temperature at six eelgrass sites over three sampling periods.

Eelgrass percent cover differed significantly between sites for all three sampling periods (August 2001, Kruskal-Wallis, df=5, *H*=20.877; May 2002, Kruskal-Wallis, df=5, *H*=51.200; August 2002, Kruskal-Wallis, df=5, *H*=39.630, *P*<0.001 for all cases) (Figure 4.6). Overall, Ducking was ranked as the highest energy bed, followed by Arakun, Auseth, Kootowis, Indian and lastly Sharp was ranked as the lowest energy bed (Table 4.1, Appendix 4.2).

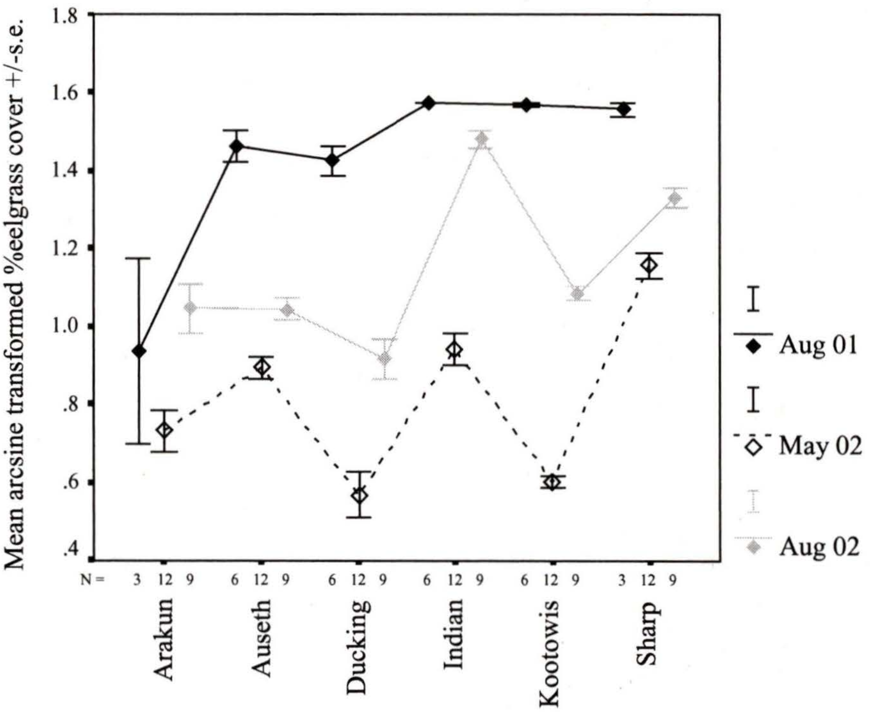

Figure 4.6. Arcsine transformed mean percent eelgrass cover at six eelgrass sites over three sampling periods.

4.3.3. Hydrodynamic Ranking of Sampling Sites

By ranking both direct and indirect measures of hydrodynamic energy eelgrass bed sites were placed along a hydrodynamic gradient (Table 4.1). Overall results indicate that Arakun and Ducking were most frequently characterized as high energy eelgrass beds, Auseth, Indian and Kootowis were intermediate and Sharp was the lowest energy bed (Figure 4.7).

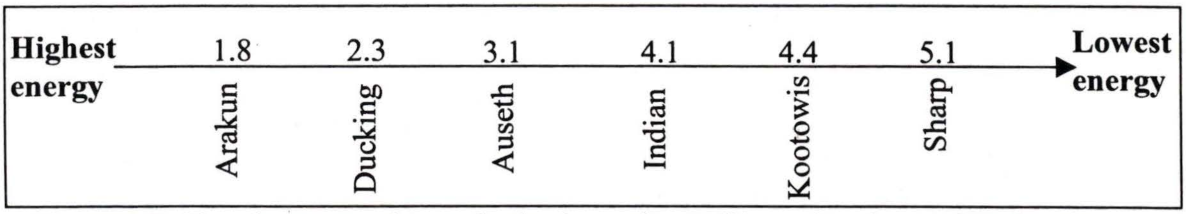

Figure 4.7. Site placement along a hydrodynamic gradient using the total rank values indicated above each site.

4.3.4. Fish Size and Hydrodynamic Energy Gradient

The four fish species examined differed significantly among beds. However fish length differences were not significant when examined along an energy gradient, nor were these differences consistent across time from May to August (Figures 4.8 and 4.9). Shiner perch lengths differed significantly between sites in both May and August 2002 (May 2002, Kruskal-Wallis, df=5, $H = 50.726$, $P<0.001$; August 2002, ANOVA, $F_{5,277} = 3.963$, $P<0.01$) (Figures 4.8 and 4.9). Pair wise comparisons in May and August showed no consistent trend in adult shiner perch lengths across an energy gradient. In May, the low energy site Sharp had significantly shorter fish, but in May and August the low energy site Kootowis had significantly larger shiner perch.

Threespine stickleback lengths also differed significantly between sites in both May and August 2002 (May 2002, ANOVA, $F_{5,527} = 7.490$, $P<0.001$; August 2002, ANOVA, $F_{5,241} = 2.814$, $P<0.05$) (Figures 4.8 and 4.9). In May, threespine stickleback caught at the high energy Arakun had significantly greater lengths than those caught at the low energy beds Kootowis and Sharp, but also the high energy bed Ducking. Additionally, sticklebacks caught at the low energy bed Sharp were also significantly smaller than those caught at intermediate energy beds Auseth and Indian. In comparison in August, no significant trends were demonstrated along the energy gradient.

Crescent gunnel lengths also differed significantly between sites both in May and August (May 2002, Kruskal-Wallis, df=5, $H = 86.023$, $P<0.001$; August 2002, Kruskal-Wallis, df=5, $H = 99.567$, $P<0.001$) (Figures 4.8 and 4.9). However, length differences among beds did not demonstrate a significant difference along an energy gradient in May or August.

Finally, staghorn sculpins differed significantly between sites in May but not in August (May 2002, Kruskal-Wallis, df=5, $H = 104.380$, $P<0.001$) (Figures 4.8 and 4.9). Overall length differences among beds did not differ significantly along an energy gradient. Among bed comparisons in May demonstrated larger staghorn sculpins caught at the intermediate energy beds Auseth and Indian than those caught at the high energy beds, Arakun and Ducking, and the low energy bed Kootowis. The low energy bed also had shorter staghorn sculpins than those caught at Auseth.

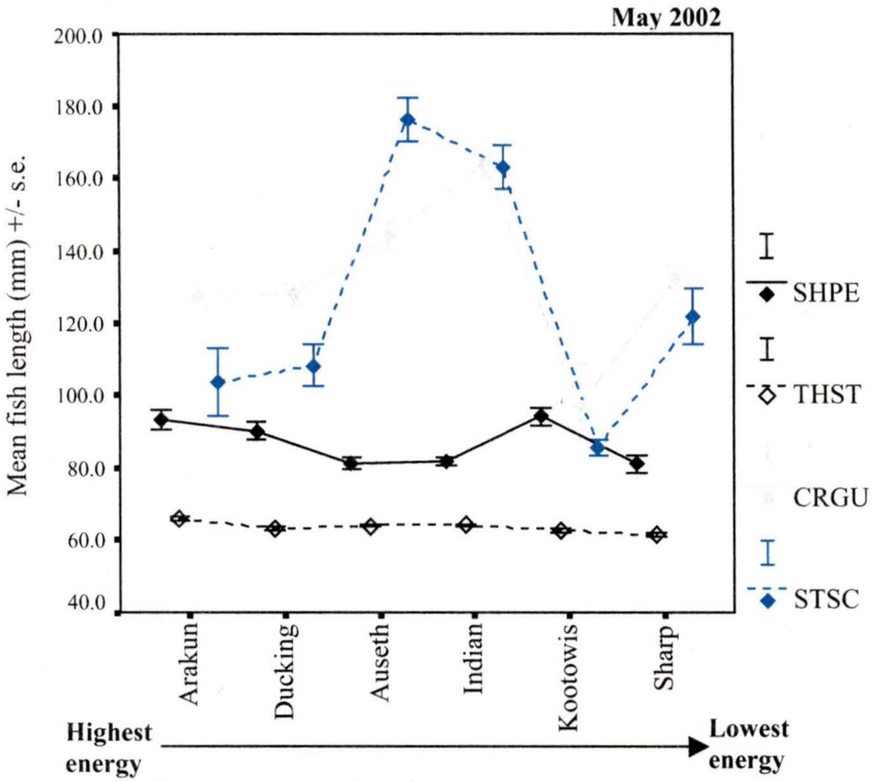

Figure 4.8. Mean fish length in millimetres for four common fish species at each site along a hydrodynamic gradient ranging from high to low energy in May 2002. (Fish abbreviations: SHPE=shiner perch, THST=threespine stickleback, CRGU=crescent gunnel, STSC=staghorn sculpin)

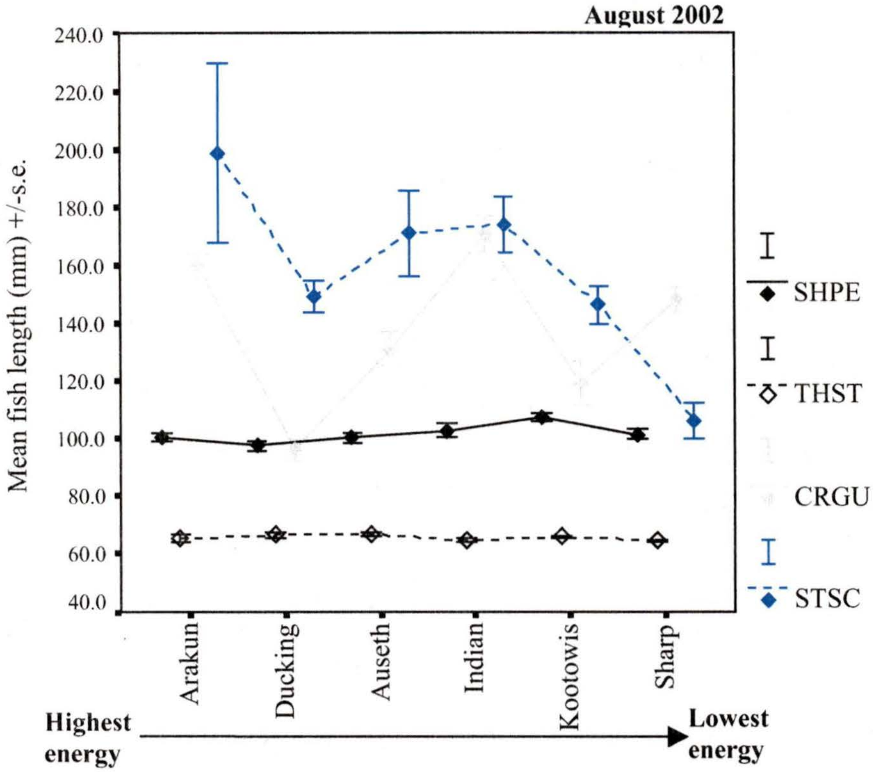

Figure 4.9. Mean fish length in millimetres for four common fish species in August 2002 with sites ordered from high to low hydrodynamic energy. (Fish abbreviations: SHPE=shiner perch, THST=threespine stickleback, CRGU=crescent gunnel, STSC=staghorn sculpin)

4.3.5. Fish Age Classes and Hydrodynamic Energy Gradient

The abundance of 1+ year old shiner perch did not differ significantly between sites in May or August 2002 (Figure 4.10). However, there were significant between site differences in both May and August for young of the year (YOY) shiner perch (May 2002, Kruskal-Wallis, df=5, $H = 23.344$, $P<0.001$; August 2002, ANOVA, $F_{5,48} = 5.804$, $P<0.001$) (Figure 4.10). In May, the lower energy bed Sharp had significantly more YOY shiner perch than Arakun (high energy), Indian (intermediate energy) and Kootowis (low energy) ($P<0.05$). Contrastingly, in August significantly more YOY shiner perch were caught at the higher energy bed Ducking than Auseth (intermediate energy) and Sharp (low energy) ($P<0.05$).

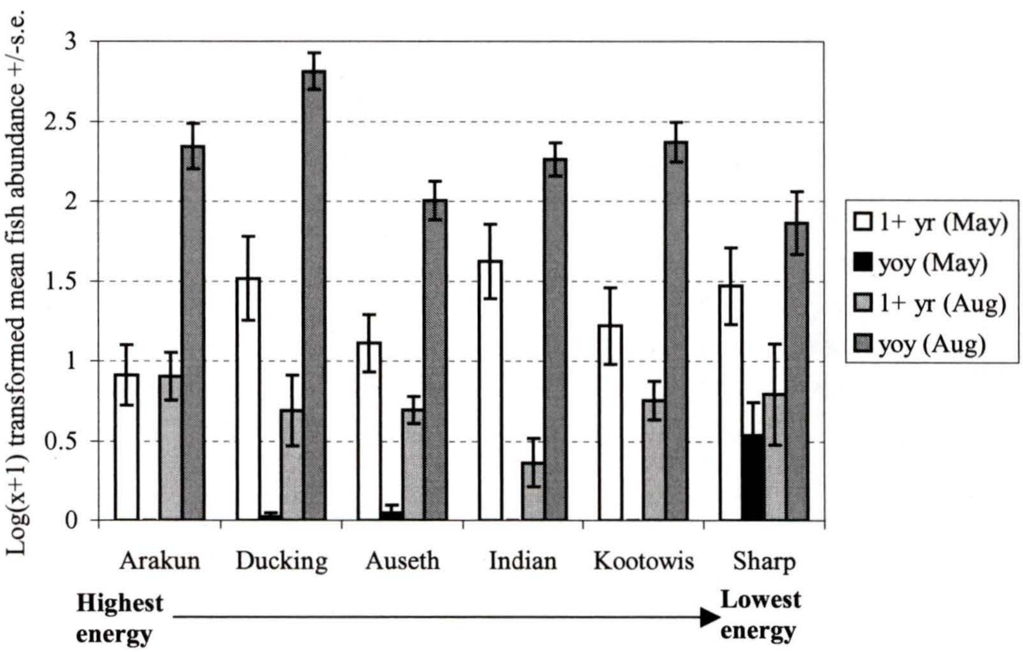

Figure 4.10. Log(x+1) transformed mean shiner perch abundance for +1 year fish and young of the year (YOY) in May and August 2002 with sites ordered from high to low hydrodynamic energy.

In comparison, both 1+ year old and YOY threespine stickleback abundances demonstrated significant differences among sites (+1 year, ANOVA, $F_{5,48} = 11.505$, $P<0.001$; YOY, Kruskal-Wallis, df=5, $H = 37.297$, $P<0.001$) (Figure 4.11). Among bed comparisons demonstrated the lower energy beds Kootowis and Sharp to have significantly more +1 year old threespine stickleback than all other sites ($P<0.05$). Additionally, the lower energy beds Kootowis and Sharp also had a significantly higher number of YOY stickleback than the higher energy beds Arakun and Ducking, as well as the intermediate energy bed Indian. The higher energy beds Arakun and Ducking also had significantly lower numbers of YOY fish than all other sites.

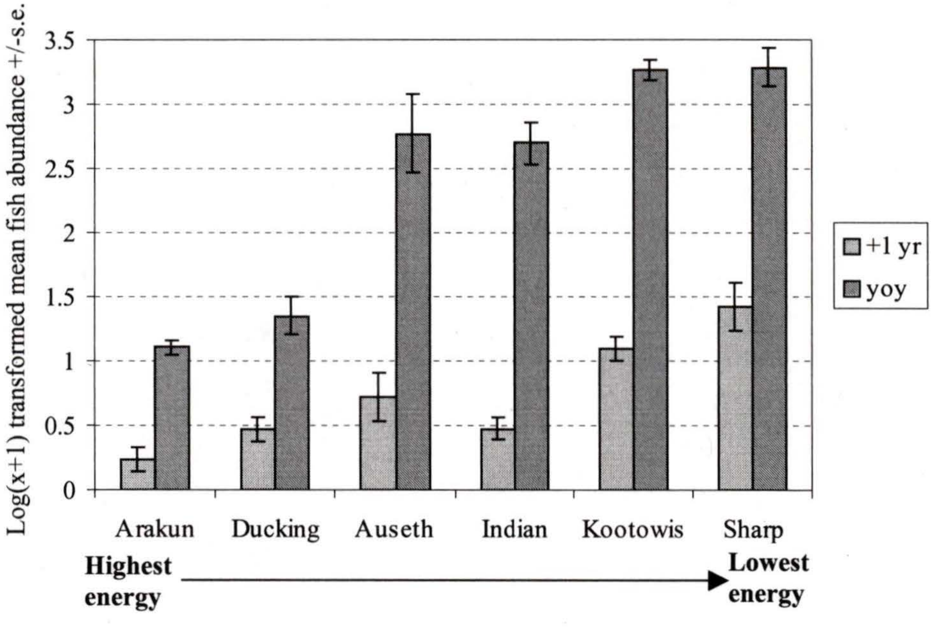

Figure 4.11. Log(x+1) transformed mean threespine stickleback abundance for +1 year fish and young of the year (YOY) in August 2002 with sites ordered from high to low hydrodynamic energy.

4.3.6. Fish Diversity and Hydrodynamic Energy Gradient

Previous analysis of univariate fish diversity measures in chapter 3 found significant differences among sites in species richness, abundance, and taxonomic distinctness (chapter 3, section 3.3.1). Evenness and taxonomic diversity are not included in this analysis because in chapter 3 abundance, evenness, and taxonomic diversity followed similar trends.

Fish species richness was significantly higher in the lower energy bed Sharp than the higher energy bed Arakun and the intermediate energy Indian (chapter 3, section 3.3.1.1). Overall, species richness was lowest at the higher energy bed Arakun and highest at the low energy bed Sharp, but all other sites were not significantly different along a hydrodynamic gradient (Figure 4.12).

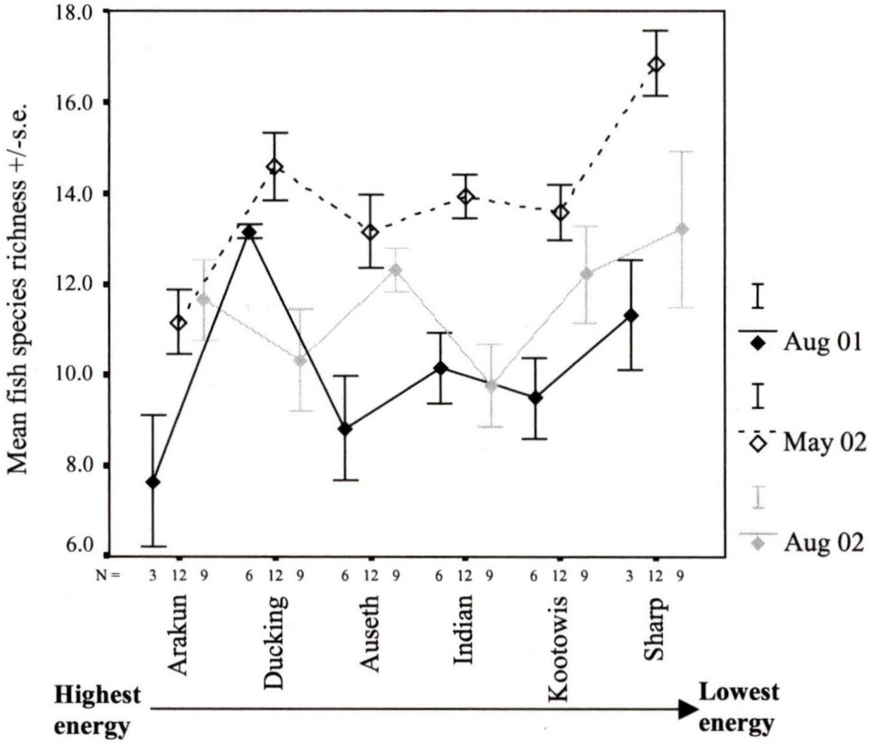

Figure 4.12. Mean fish species richness at six eelgrass sites over three sampling periods, with the sites ordered from high to low hydrodynamic energy.

In comparison fish abundance was significantly lower at the highest energy site Arakun than all other sites. The high energy bed Ducking and the intermediate energy bed Auseth also had significantly fewer fish than the intermediate energy bed Indian and the lowest energy bed Sharp. Overall trends demonstrated fish abundance to increase along a hydrodynamic gradient from high to low energy (Figure 4.13).

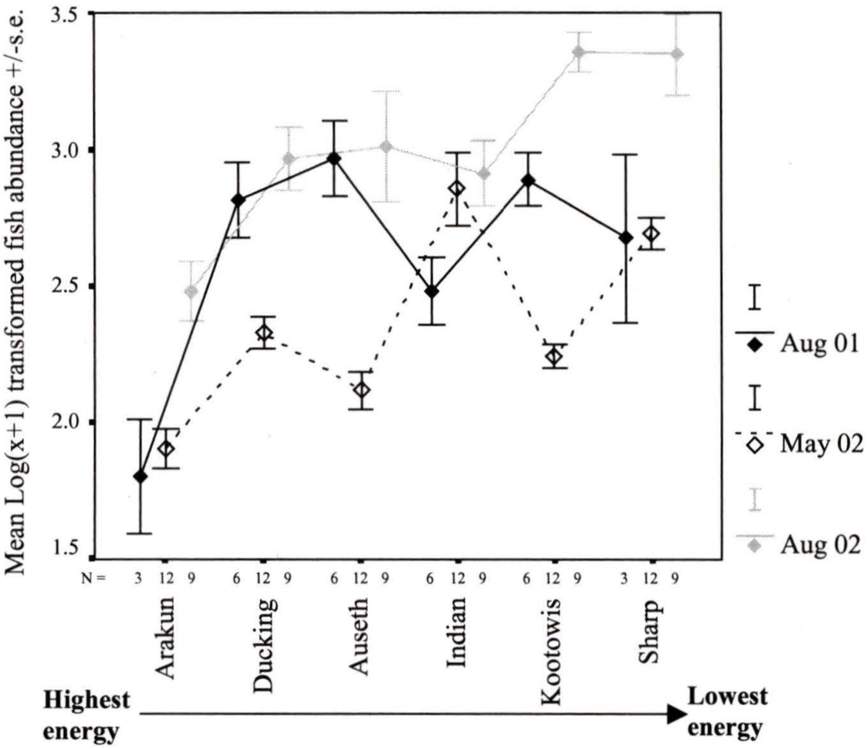

Figure 4.13. Mean Log(x+1) transformed fish abundance at six eelgrass sites over three sampling periods, with the sites ordered from high to low hydrodynamic energy.

Finally taxonomic distinctness was significantly higher at the low energy bed Kootowis than all other sites, except the lower energy bed Sharp. In addition, Sharp had significantly higher taxonomic distinctness than the high energy bed Arakun. Overall trends showed taxonomic distinctness to increase from high to low energy along a hydrodynamic gradient (Figure 4.14).

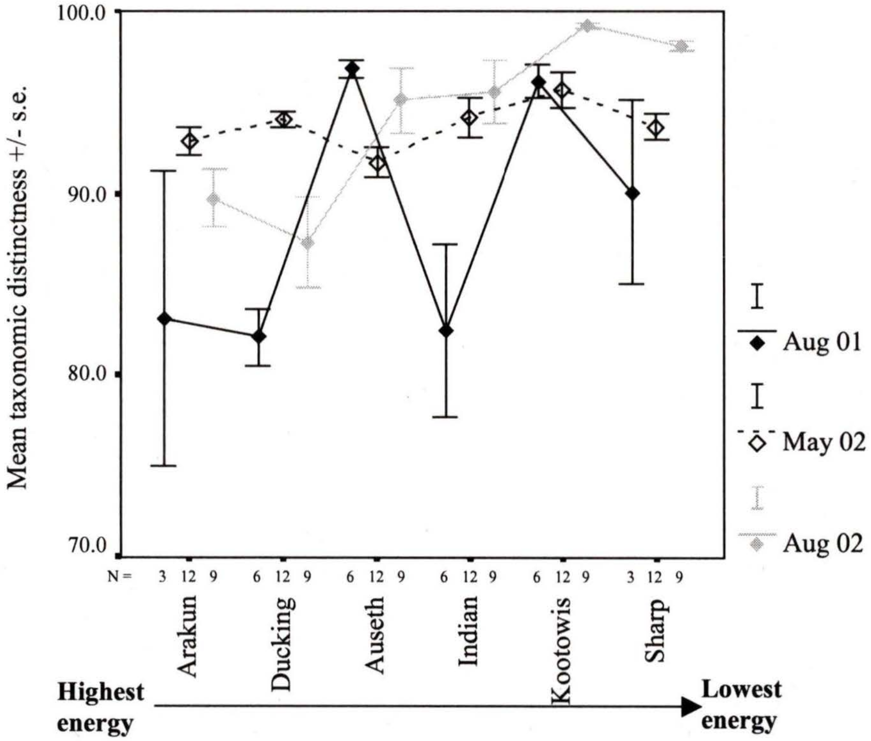

Figure 4.14. Mean taxonomic distinctness at six eelgrass sites over three sampling periods, with the sites ordered from high to low hydrodynamic energy.

4.3.7. Multivariate Analysis Examining Fish Diversity and Hydrodynamic Energy Gradient

The highest weighted Spearman rank correlation coefficients (ρ_w) values yielded a different subset of hydrodynamic variables that best explained the fish diversity for each of the three sampling periods (Table 4.2). In August 2001 using Log(x+1) transformed fish species abundance a single variable, eelgrass percent cover, produced a ρ_w of 0.49. In comparison, species presence/absence data was also best explained by eelgrass percent cover but generated a lower ρ_w of 0.27. In May 2002, using Log(x+1) transformed fish species abundance, current gave the highest ρ_w of 0.31. Fish species presence/absence data was also best explained by current, but generated a lower ρ_w of 0.20. Lastly, in August 2002 using Log(x+1) transformed fish species abundance, two variables, water temperature and eelgrass percent cover yielded the highest ρ_w of 0.28. Species presence/absence was also best explained by temperature and eelgrass percent cover, but produced a lower ρ_w of 0.22. A separate analysis using the RELATE procedure compared all the matrices and found each comparison to be significantly correlated at *P*<0.001.

Table 4.2. Summary of the BIOENV results with the weighted spearman rank correlation (ρ_w) values for each fish community property and the hydrodynamic variables that generated the highest ρ_w values. The RELATE procedure confirmed all correlations were significant at *P*<0.001.

	August 2001		**May 2002**		**August 2002**	
Fish Community Property	**Variables**	ρ_w	**Variables**	ρ_w	**Variables**	ρ_w
Log(x+1) fish species abundance	Eelgrass %cover	0.49	Current	0.31	Temperature, Eelgrass %cover	0.28
Fish species presence/absence	Eelgrass %cover	0.29	Current	0.20	Temperature, Eelgrass %cover	0.33

A spatial configuration comparison of the fish community data and the reduced set of hydrodynamic variables were done separately for each sampling time using 2-D MDS plots. Only the Log(x+1) transformed fish species abundance data were plotted because the ρ_w values generated from the BIOENV procedure were higher than those generated from the species presence/absence data. In August 2001, very little concordance was found between the fish communities and hydrodynamic variables (Figure 4.15). Although, Arakun (high energy) did form separate groupings in both plots a strong relationship was not apparent. In May 2002, the overall pattern of site distribution was similar for both plots. The Kootowis (low energy) and Arakun (high energy) sites exhibited the highest differences in both their fish communities and hydrodynamic variables. However, the placement of Sharp (low energy) was not complimentary between the two plots (Figure 4.16). Lastly in August 2002, Arakun formed a distinct cluster in both plots, but there was no concordance between the two plots for the other sites (Figure 4.17).

Figure 4.15. Separate MDS ordination for Log(x+1) transformed fish species abundance and hydrodynamic variables at six eelgrass beds in August 2001, with the superimposed ranked cluster groups. Solid lines indicating 3 clusters formed at a rank of 300 for fish species data and 4 clusters formed at a rank of 300 for hydrodynamic variables. Weighted Spearman rank similarity value of 0.49 between fish diversity and hydrodynamic variable matrices.

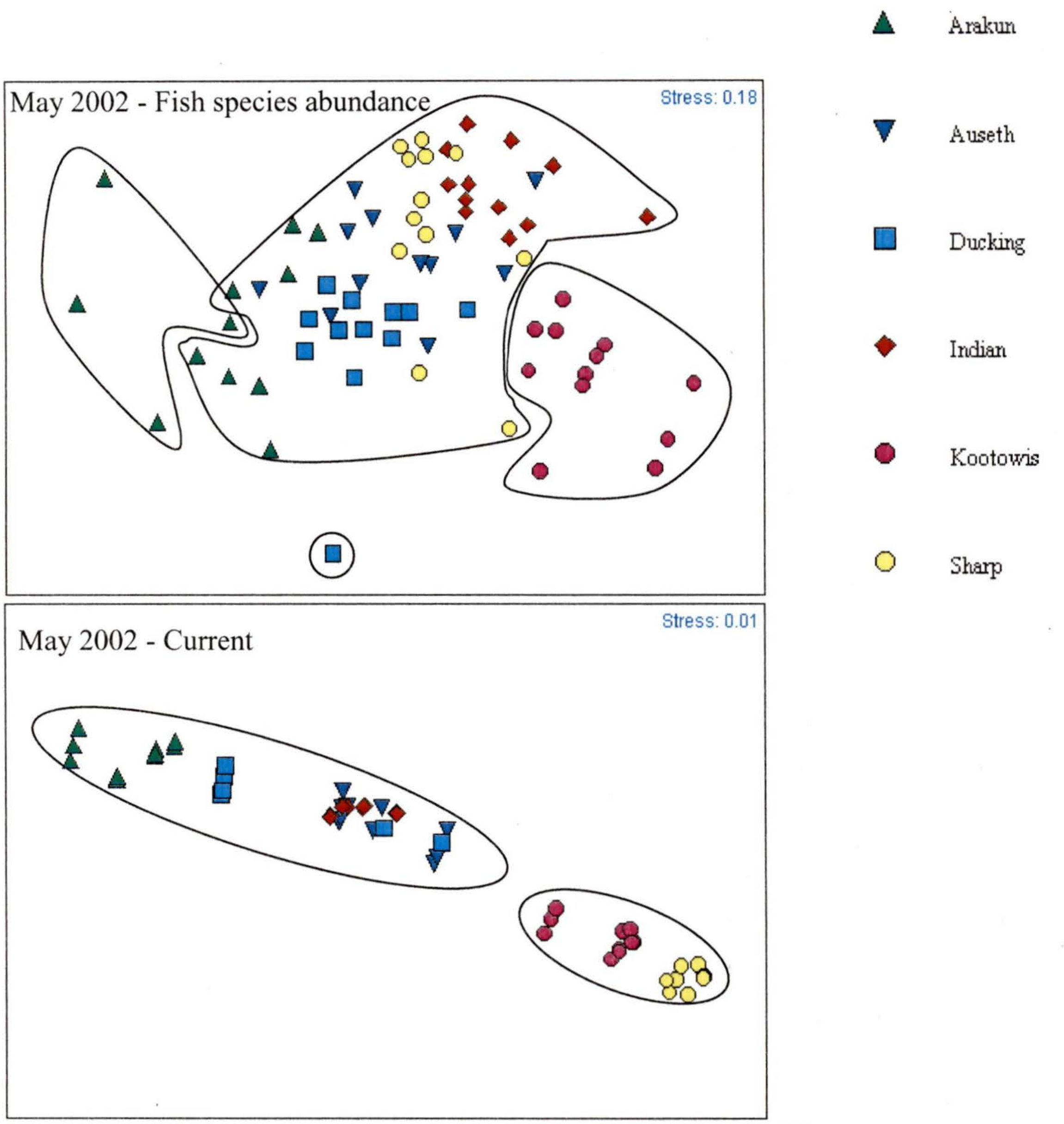

Figure 4.16. Separate MDS ordination for Log(x+1) transformed fish species abundance and hydrodynamic variables at six eelgrass beds in May 2002, with the superimposed ranked cluster groups. Solid lines indicating 4 clusters formed at a rank of 1500 for fish species data and 2 clusters formed at a rank of 1500 for the hydrodynamic variables. Weighted Spearman rank similarity value of 0.31 between fish diversity and hydrodynamic variable matrices.

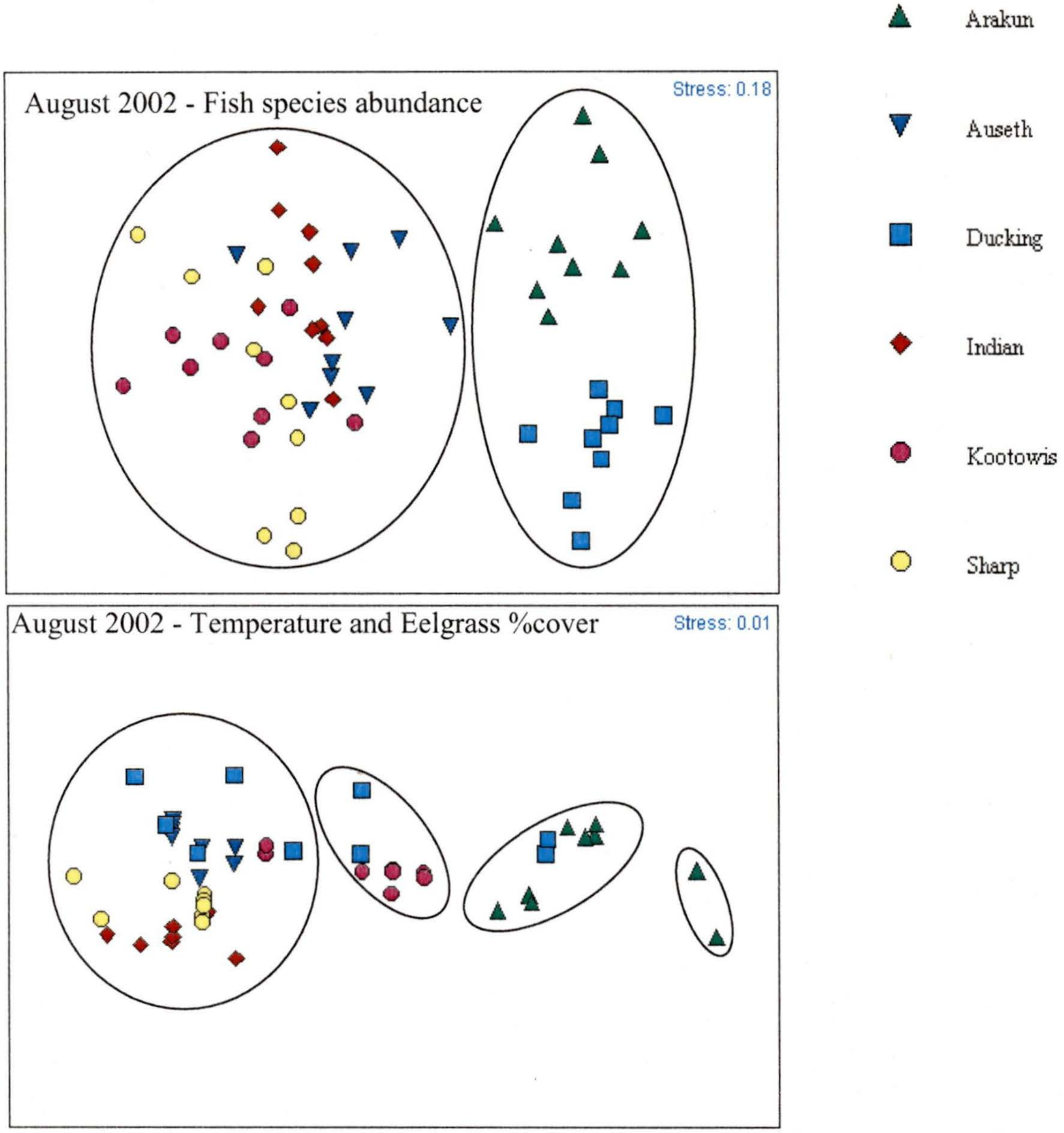

Figure 4.17. Separate MDS ordination for Log(x+1) transformed fish species abundance and the best hydrodynamic variables at six eelgrass beds in August 2002, with the superimposed ranked cluster groups. Solid lines indicating 2 clusters formed at a rank of 750 for fish species data and 4 clusters formed at a rank of 750 for the hydrodynamic variables. Weighted Spearman rank similarity value of 0.28 between fish diversity and hydrodynamic variable matrices.

4.4. Discussion

Nearshore fish communities vary across habitats with some habitats supporting higher diversity than others (Arrivillaga and Baltz 1999, Murphy et al. 2000). Seagrass beds support high fish diversity (Jackson et al. 2001), however few studies have examined the spatial variability of fish diversity among seagrass beds (Bell et al. 1994, Hovel et al. 2002). Hydrodynamic energy has been shown to structure both seagrass structure and macrofaunal communities within seagrass beds (Murphey and Fonseca 1995, Fonseca and Bell 1998). Therefore, this study examined hydrodynamic energy as the primary process driving the variability in fish communities among eelgrass beds in southern Clayoquot Sound.

4.4.1. Hydrodynamic Gradient of Eelgrass Beds

In this study, I found that Arakun and Ducking ranked highest along the hydrodynamic gradient, Auseth and Indian were intermediate, and Kootowis and Sharp ranked low (Table 4.1). This ranking was also consistent across seasons from May and August. The hydrodynamic gradient of these six eelgrass beds was calculated using direct and indirect measures. However, each hydrodynamic measure was weighted equally in the final site ranking. Since some measures did not differ significantly among sites (i.e. relative exposure index), whereas other measures did (i.e. current) an equal weighting of these measures may not be accurate. Therefore, a higher weighting of hydrodynamic measures that differed significantly among beds may generate a more accurate hydrodynamic gradient.

Additionally, indirect measures of hydrodynamic energy may also be influenced by other environmental factors. For example, eelgrass percent cover can also decrease with lower salinities and higher temperatures (Davidson and Hughes 1998, Kamermans et al. 1999). Therefore, the higher energy ranking of Kootowis for this variable (Table 4.1) may be related to environmental factors other than hydrodynamic energy. At Kootowis the freshwater inflow from Kootowis Creek and the more estuarine conditions of this site may directly influence the lower eelgrass cover. Consequently, weighting indirect hydrodynamic measures lower than direct measures could overcome these discrepancies. Overall, sites ranking highest and lowest on the hydrodynamic gradient, such as Arakun and Sharp, were consistently reflected in the direct and indirect measures. Thus, choosing sites that rank constantly high or low for direct and indirect measures may provide a clearer depiction of how hydrodynamic energy influences fish community properties.

4.4.2. Fish Size and Age Class Comparisons and Hydrodynamic Gradient

Previous research has shown that fish size differs according to hydrodynamic energy. Clark (1997) caught smaller fish, usually less than 30mm at lower energy sites and larger fish, greater than 120mm at higher energy sites. This may be related to food availability and feeding abilities of the fish (Clark 1997). In areas of higher water energy the abundance of smaller invertebrates is lower (Bell et al. 1994), and swimming and prey capture abilities of small fish also decreases with higher exposures (Clark 1997). In this study, among site differences in the fish size did not differ significantly along an energy gradient. However, the intermediate energy sites, Auseth and Indian did have larger crescent gunnels and staghorn sculpins than the higher and lower energy beds

(Figures 4.8 and 4.9). Additionally, fish length trends among sites were not the same for all four species. Clark (1997) also found only two out of the four fish species examined were larger as exposure increased. This highlights the important of examining among site differences of individual fish species along a hydrodynamic gradient. In addition, further research that evaluates both fish length and weight may demonstrate different condition factors for site along a hydrodynamic gradient.

Seagrass beds have been cited as important nursery habitat (Jackson et al. 2001). Few studies have looked at juvenile fish selection of different seagrass bed characteristics (Horinouchi and Sano 1999), but no studies have examined seagrass habitat selection by juvenile fish in relation to hydrodynamic energy. In this study, the abundance of +1 year old shiner perch was not significantly different among eelgrass beds along a hydrodynamic gradient. However the young of the year (YOY) were caught in higher abundances in different energy beds (Figure 4.10). In May, higher numbers of YOY shiner perch were caught in the low energy bed, Sharp than all other sites. Additional examination of the number of gravid females may also show differences along a hydrodynamic gradient in +1 year fish distributions. In comparison, in August a higher number of YOY shiner perch were caught in the high energy site, Ducking. Shiner perch are known to migrate to deeper waters in the winter (Hart 1988). Since the Ducking bed was in a larger channel, these YOY shiner perch may have moved through this area as they migrated to deeper waters in the fall.

In contrast, both +1 year old and YOY threespine sticklebacks were caught in higher numbers at the low energy beds Kootowis and Sharp. Both of these eelgrass beds have low tidal currents and are protected from wind and wave exposure (Figures 4.3 and

4.4). Sticklebacks are a relatively small fish species reaching a maximum length of 10cm (Hart 1988). Therefore, stickleback may prefer low energy eelgrass beds because their swimming abilities are limited in areas of higher currents and wave exposure (Clark 1997). Overall, this suggests that lower energy beds may be better rearing habitat for both shiner perch and threespine sticklebacks.

4.4.3. Fish Diversity and Hydrodynamic Gradient

Hydrodynamic energy has been shown to play an important role in structuring seagrass landscapes and the faunal communities within these habitats (Irlandi 1996, Townsend and Fonseca 1998). In this study, sites categorized along a hydrodynamic energy gradient (Table 4.1) were expected to explain some of the among site differences in the univariate fish diversity measures. Sharp, the low energy bed, had higher species richness in 2001 and 2002 compared to the other sites. Higher food availability for smaller fish species in lower energy seagrass beds may influence this increase in the number of fish species (Bell et al. 1994).

Fish abundance, in this study, was also found to be lower in the higher energy beds, Arakun and Ducking. A consistent trend was also found with fish biomass being negatively correlated with wave exposure in seagrass habitats (Edgar and Shaw 1995b). It has also been suggested that water movement influences fish immigration and emigration in seagrass beds (Sogard et al. 1989b). Therefore areas of higher energy may have more fish migrating through this habitat more quickly, rather than remaining in the area. Additionally, fish abundance patterns among beds were often a reflection of the number of shiner perch and threespine sticklebacks. Higher energy beds had fewer schooling shiner perch and threespine stickleback indicating that habitat selection by these species influence overall fish abundance differences among eelgrass beds (see chapter 3, Figure 3.3).

Taxonomic distinctness also increased in lower energy beds in this study (Figure 4.14). A more taxonomically diverse fish community may utilize eelgrass beds in areas with lower currents and wind and wave exposure. An increase in epifaunal communities in higher energy beds (Bell et al. 1994) may also increase food availability for a wider diversity of fish species.

Multivariate analysis indicated low correlation between fish diversity and the hydrodynamic variables, explaining very little of the variation in fish communities among beds. However, the fish community MDS plots did show a clustering of fish communities in August at the higher energy beds, Arakun and Ducking (Figure 4.15 and 4.17). Hydrodynamic energy may have a greater effect on fish communities in higher energy beds. Whereas, dissimilarities between fish communities in intermediate and low energy beds may not be detectable. Therefore, examining the two extreme ends of hydrodynamic energy, high and low, rather than a gradient of exposures, may provide a clearer depiction of the role hydrodynamic energy plays in structuring eelgrass fish communities.

The influence of hydrodynamic energy may also vary seasonally. Fish communities formed distinctive site groupings in May but these patterns were not explained by hydrodynamic energy (Figure 4.16). In comparison, the fish communities caught in August at the high energy beds Arakun and Ducking formed a distinctive group. Whereas, fish communities caught in the intermediate and lower energy beds formed a single cluster (Figures 4.15 and 4.17). This may reflect a seasonal shift in fish preferentially choosing or avoiding higher energy beds later in the summer. In addition, habitat complexity within seagrass beds has been linked to settlement preferences of juvenile fish species (Rooker et al. 1998, Horinouchi and Sano 1999). Consequently, in May many fish species including threespine sticklebacks, shiner perch, and kelp perch may be searching for suitable rearing habitat for their young (Hart 1988). These fish species may be selecting specific habitat features related to hydrodynamic energy. Hovel et al. (2002) also found inconsistent patterns in factors influencing seagrass macrofaunal densities both seasonally, between spring and fall, and between years. He suggested this may be due to covariance between seagrass structural complexity and hydrodynamic influences acting at different spatial scales. These differences between August and May fish communities and hydrodynamic regimes highlights the importance of studying communities over various temporal scales (Jackson and Jones 1999).

4.4.4. Additional Explanatory Variables

It is unlikely that hydrodynamic energy alone would be responsible for shaping fish community structure and function. Predation is an integral component in structuring seagrass communities (Irlandi 1996, Hindell et al. 2000). Throughout the sampling period avian predators, such as the great blue heron (*Ardea herodias*), were observed foraging at all sites and on adjacent eelgrass habitat. Large predatory piscivores were also observed foraging in eelgrass beds. Rockfish were caught at all sites and spiny dogfish (*Squalus acanthius*) were also observed at night in the Indian bed. Further research is required to assess the effect these predators have on structuring eelgrass fish communities.

Assessment of how prey availability varies between beds and how this relates to differences in fish communities is also required (Connolly 1994b). Fish abundance and biomass is positively correlated with an increase in epifaunal communities (Bell et al. 1994, Edgar and Shaw 1995b). Epifaunal communities also increase with higher seagrass densities (Attrill et al. 2002). Therefore, the importance of epifaunal communities in structuring eelgrass fish communities needs to be determined.

Habitat linkages and configurations can also strongly influence community structure (Micheli and Peterson 1999, Goodsell and Connell 2002). Movement between habitats through vegetated corridors provide essential linkages for predators and production transfer between habitats (Irlandi and Crawford 1997, Micheli and Peterson 1999). Determining the connectivity between eelgrass beds and other adjacent habitats would also provide insight into the complexity of the nearshore ecosystems. Additionally, predation pressure increases in seagrass edge habitat (Peterson et al. 2001, Laurel et al. 2003). Therefore, fish communities on the edge of eelgrass beds versus within, needs to be compared.

Finally, the fish sampling methodology, beach seining, was limiting in the fish species caught and the area sampled (Rozas and Minello 1997). Additionally, fish caught in higher energy areas and foraging piscivores are larger and presumably more mobile (Clark 1997, Hindell et al. 2002). Since more mobile fish species can avoid the beach seine, the use of more intensive sampling methods, such as gill nets and trawls could increase catch efficiency (Hindell et al. 2000). Increased catch efficiency could provide a more accurate measure of all fish species utilizing specific eelgrass habitat, and a stronger link between fish communities and hydrodynamic energy regimes may be established.

4.4.5. Conclusion

Hydrodynamic energy did explain some of the variability observed in eelgrass fish communities in southern Clayoquot Sound. Young of the year (YOY) shiner perch and threespine stickleback were caught in higher numbers in low energy eelgrass beds. Additional trends indicated fish species richness, abundance and taxonomic distinctness to increase as hydrodynamic energy decreased. Higher food availability in lower energy beds (Bell et al. 1994) or fish mobility decreasing in higher exposures (Clark 1997) may influence these changes fish community properties. Lastly, trends were not consistent between spring and summer months, as May fish communities were not clearly linked to hydrodynamic energy. Inconsistencies over time suggest a variety of processes may be acting at different spatial scales (Hovel et al. 2002). Therefore, the influence of hydrodynamic energy over a larger temporal scale, and at the high versus low energy ranges, rather than along gradient may provide a clearer depiction of how hydrodynamic regime influences fish communities.

In addition, processes structuring fish communities may also be related to hydrodynamic energy. Increasing hydrodynamic energy is negatively correlated with epifaunal abundance (Attrill et al. 2002) and positively correlated increasing fish size (Clark 1997). Therefore, biotic processes, such as foraging ability and predation are linked to changes in hydrodynamic regime. Hydrodynamic setting also influences seagrass bed spatial arrangement and within bed structural complexity (Fonseca and Bell 1998, Turner et al. 1999). These changes in seagrass habitats create edges, habitat corridors and alter seagrass bed structural complexity, all processes that can potentially structure fish communities (Rooker et al. 1998, Micheli and Peterson 1999, Laurel et al. 2003).

In summary, fish community patterns were found to be heterogeneous across eelgrass beds in southern Clayoquot Sound. Determining the causes and consequences of these community patterns is critical for understanding and conserving ecosystems (Levin 1992). This study provided incites into the complex processes driving eelgrass fish communities though the examination of hydrodynamic energy. Further assessment of how abiotic and biotic processes are linked to hydrodynamic regimes and their influence on eelgrass fish communities is required.

Chapter 5: Conclusion

Understanding the link between spatial and temporal biogeographical patterns in ecological communities and the processes structuring these patterns are integral components of marine conservation (Levin 1992, Brown et al. 1996, Gracia-Charton and Perez-Ruzafa 1999). In this study, spatial and temporal patterns of eelgrass fish communities and the potential process structuring those patterns was evaluated in southern Clayoquot Sound. The study entailed three main components; first, evaluating beach seining as a monitoring method, second, addressing spatial and temporal patterns in fish diversity by applying a broad array of diversity measures, and third, determining the influence of hydrodynamic setting on fish diversity patterns. By evaluating each of these components, eelgrass fish community patterns and processes structuring those patterns could be assessed and applied to understanding complex interactions influencing eelgrass fish communities.

First, the adequacy of beach seining as a monitoring method was evaluated by looking at the effect of diel and tidal patterns on fish communities and the overall catch efficiency of seining. Diel patterns demonstrated a shift in community structure, where larger, predatory fish species were caught at night, as well as an overall decrease in fish abundance at night. The highest fish diversity caught occurred during a rising low tide. Monthly triplicate beach seines underestimated only 10% of the eelgrass fish diversity. Overall, beach seining caught a broad range of fish species, was easily deployed, and an inexpensive method to monitor eelgrass fish diversity.

Second, fish diversity was investigated over different spatial and temporal scales, using a variety of diversity measures. The conventional univariate diversity measures applied included species richness, abundance, and evenness. Spatial and temporal patterns in fish species richness were not interpretable, but should be evaluated to compare with other studies. Total fish abundance and evenness showed seasonal shifts in fish communities; however, spatial differences among sites were not interpretable. Additional assessment of taxonomic diversity of eelgrass fish communities demonstrated similar patterns to that found for abundance and evenness. Therefore, a single measure, total fish abundance would be adequate. In comparison, taxonomic distinctness highlighted among site differences and seasonal shifts in fish communities not apparent by only using traditional univariate diversity measures. In addition to these alpha diversity measures, beta diversity measures were also evaluated, and highlighted how community differences increased over time and geographical distances. Finally, multivariate diversity measures drew attention to spatial patterns in fish communities not detected by univariate measures, emphasizing the need to use multivariate techniques to describe community structure. By exploring a multitude of diversity measures at a variety of scales subtle changes in community structure could be detected over space and time, and in combination aided in understanding patterns in eelgrass fish diversity.

Third, hydrodynamic energy was evaluated as the potential process structuring patterns in eelgrass fish diversity. Each eelgrass bed was placed along a hydrodynamic gradient from high to low energy by evaluating both indirect and direct measures of hydrodynamic energy. Hydrodynamic energy explained some of the fish community patterns. Young of the year shiner perch and threespine stickleback were caught in higher numbers in lower energy beds. Overall trends demonstrated fish species richness, total abundance, and taxonomic distinctness to decrease as hydrodynamic energy increased.

Lastly, trends were not consistent between spring and summer months, as May fish communities were not clearly linked to hydrodynamic energy. Additional examination of the influence of hydrodynamic energy over larger temporal scales, and at the high versus low energy ranges, rather than along gradient may provide a clearer depiction of how hydrodynamic regime influences fish communities. Furthermore, research should evaluate biological factors potentially linked to hydrodynamic energy, such as predation and foraging strategies. Understanding how abiotic and biotic parameters are linked is key to determine the processes influencing eelgrass fish community patterns.

Overall, my study provided insight into patterns of eelgrass fish communities and the potential processes structuring those patterns in southern Clayoquot Sound. Through this research I have developed the following research recommendations for monitoring eelgrass fish diversity within Pacific Rim National Park Reserve of Canada:

- Beach seining eelgrass fish diversity should include triplicate sets at low tide, diel sampling, and a minimum of three sampling periods from spring to late fall to address shifts in community structure.
- Additional sampling methods such as, otter trawls and gill nets are required to sample areas of the eelgrass beds inaccessible to beach seining, and to catch more mobile fish species.
- Analysis of fish diversity should include the traditional univariate diversity measures, species richness and total abundance, but should also include taxonomic distinctness. Beta diversity measures, such as taxonomic similarity, should also be evaluated to

assess if the heterogeneity of eelgrass beds contributes to fish diversity. Lastly, using multivariate community analysis is essential to detect subtle among site differences.

- Length data of fish species caught should be assessed to determine if fish age class and size differ among eelgrass beds.
- Future collection of physical and eelgrass variables must be in concurrence with fish sampling to address the influence environmental and habitat factors have on eelgrass fish diversity.
- The influence of hydrodynamic energy on fish community patterns should be further evaluated in high versus low energy beds rather than over a hydrodynamic gradient. By exploring the two ends of the gradient a clearer depiction of how hydrodynamic energy influences fish diversity patterns could be determined.
- Finally, evaluating the influence of biotic processes such as predation, competition, and foraging have on fish diversity patterns is crucial to understand the processes driving eelgrass ecosystems.

In summary, this study emphasized the importance of examining spatial and temporal variation of marine communities by applying a diverse array of univariate and multivariate measures. Once marine community patterns are determined, evaluating the potential process structuring these communities is critical. Marine ecological communities are complex and highly variable over time; therefore, it is unlikely that a single process shapes those differences. Quantifying the influence of a process on community structure, and determining how it may be linked to other processes is crucial for conserving marine biodiversity. Determining marine community patterns and the causal processes of those patterns are critical components to develop monitoring programs within national park boundaries and establishing a network of marine reserves (Garcia-Charton and Perez-Ruzafa 1999, Yoccoz et al. 2001).

The current multitude of threats to marine diversity is a driving force behind the need to understand marine ecosystems (Gray 1997). Understanding pattern and scale are fundamental components in comprehending ecological communities. In addition, discerning heterogeneous spatial and temporal patterns in ecosystems is essential in the planning and development of marine reserves. By addressing these complex issues the challenge of conserving marine ecological communities will become an achievable goal and a less daunting task.

References

Abookire, A. A., J. F. Piatt, and M. D. Robards. 2000. Nearshore fish distributions in an Alaskan estuary in relation to stratification, temperature and salinity. Estuarine, Coastal and Shelf Science **51**:45-59.

Allen, D. M., S. F. Service, and M. V. Ogburn-Matthews. 1992. Factors influencing the collection efficiency of estuarine fishes. Transactions of the American Fisheries Society **121**:234-244.

Angermeier, P. L., and M. R. Winston. 1999. Characterizing fish community diversity across Virginia landscapes: prerequisite for conservation. Ecological Applications **9**:335-349.

Armstrong, J. L., D. A. Armstrong, and S. B. Mathews. 1994. Food habits of estuarine staghorn sculpin, *Leptocottus armatus*, with focus on consumption of juvenile Dungeness crab, *Cancer magister*. Fishery Bulletin **93**:456-470.

Arrivillaga, A., and D. M. Baltz. 1999. Comparison of fishes and macroinvertebrates on seagrass and bare-sand sites on Guatemala's Atlantic coast. Bulletin of Marine Science **65**:301-319.

Attrill, M. J., J. A. Strong, and A. A. Rowden. 2000. Are macroinvertebrate communities influenced by seagrass structural complexity? Ecography **23**:114-121.

Bell, J. D., D. J. Ferrell, S. E. McNeill, and D. G. Worthington. 1992. Variation in assemblages of fish associated with deep and shallow margins of the seagrass *Posidonia australis*. Marine Biology **114**:667-676.

Bell, S. S., M. O. Hall, and M. S. Fonseca. 1994. Evaluation of faunal and floral attributes of seagrass beds in high and low energy regimes: a geographic comparison. Pages 267-272 in K. R. a. D. E. C. F. Dyer, editor. Changes in fluxes in estuaries: implications from science to management. Olsen and Olsen Press, Fredensborg.

Bell, S. S., R. A. Brooks, B. D. Robbins, M. S. Fonseca, and M. O. Hall. 2001. Faunal response to fragmentation in seagrass habitats: implications for seagrass conservation. Biological Conservation **100**:115-123.

Brown, J. H., G. C. Stevens, and D. M. Kaufman. 1996. The geographic range: Size, shape, boundaries, and internal structures. Annual Review of Ecology and Systematics **27**:597-623.

Buchanan, J. B. 1984. Sediment analysis. Pages 41-65 *in* N. A. Home and A. D. McIntyre, editors. Methods for study of marine benthos. Blackwell Scientific Publications, London.

Bulthuis, D. A. 1995. Distribution of seagrasses in a North Puget Sound estuary: Padilla Bay, Washington, USA. Aquatic Botany **50**:99-105.

Cao, Y., D. P. Larson, and R. M. Hughes. 2001. Evaluating sampling sufficiency in fish assemblage surveys; a similarity-based approach. Canadian Journal of Fisheries and Aquatic Sciences **58**:1782-1793.

Clark, B. M., B. A. Bennett, and S. J. Lamberth. 1996. Factors affecting spatial variability in seine net catches of fish in the surf zone of False Bay, South Africa. Marine Ecology Progress Series **131**:17-34.

Clark, B. M. 1997. Variation in surf-zone fish community structure across a wave-exposure gradient. Estuarine, Coastal and Shelf Science **44**:659-674.

Clarke, K. R., and R. H. Green. 1988. Statistical design and analysis for "biological effects" study. Marine Ecology Progress Series **46**:213-226.

Clarke, K. R. 1993. Non-parametric multivariate analyses of changes in community structure. Australian Journal of Ecology **18**:117-143.

Clarke, K. R., and R. M. Warwick. 1994. Change in marine communities: An approach to statistical analysis and interpretation. Natural Environment Research Council, Plymouth, UK.

Clarke, K. R., and R. M. Warwick. 1998. A taxonomic distinctness index and its statistical properties. Journal of Applied Ecology **35**:523-531.

Clarke, A., and S. M. Lidgard. 1999. Spatial patterns of diversity in the sea: bryozoan species richness in North Atlantic. Journal of Animal Ecology **69**: 799-814.

Clarke, K. R., and R. M. Warwick. 1999. The taxonomic distinctness measure of biodiversity: weighting of step lengths between hierarchical levels. Marine Ecology Progress Series **184**:21-29.

Clarke, K. R., and R. M. Warwick. 2001. A further biodiversity index applicable to species lists: variation in taxonomic distinctness. Marine Ecology Progress Series **216**:265-278.

Connolly, R. M. 1994a. Comparison of fish catches from a buoyant pop net and a beach seine net in a shallow seagrass habitat. Marine Ecology Progress Series **109**:305-309.

Connolly, R. M. 1994b. Removal of seagrass canopy: effects on small fish and their prey. Journal of Experimental Marine Biology and Ecology **184**:99-110.

Cox, C. B., and P. D. Moore. 1998. Biogeography: an ecological and evolutionary approach. Blackwell Scientific Publications, Oxford.

Davidson, D. M., and D. J. Hughes. 1998. *Zostera* Biotopes: An overview of dynamics and sensitivity characteristics for conservation management of marine SACs. Scottish Association for Marine Science (UK Marine SACs Project).

Dean, T. A., L. Haldorson, D. R. Laur, S. C. Jewett, and A. Blanchard. 2000. The distribution of nearshore fishes in kelp and eelgrass communities in Prince William Sound, Alaska: associations with vegetation and physical habitat characteristics. Environmental Biology of Fishes **57**:271-287.

Deegan, L. A., J. T. Finn, and J. Buonaccorsi. 1997. Development and validation of an estuarine biotic integrity index. Estuaries **20**:601-617.

Department of Fisheries and Oceans Canada (DFO). 1995. Clayoquot Sound Chart number 3673. Ottawa: Canadian Hydrographic Service.

Desmond, J. S., D. H. Deutschman, and J. B. Zedler. 2002. Spatial and temporal variation in estuarine fish and invertebrate assemblages of an 11-year data set. Estuaries **25**:552-569.

Edgar, G. J., and C. Shaw. 1995a. The production and trophic ecology of shallow-water fish assemblages in southern Australia I. Species richness, size-structure and production of fishes in Western Port, Australia. Journal of Experimental Marine Biology and Ecology **194**:53-81.

Edgar, G. J., and C. Shaw. 1995b. The production and trophic ecology of shallow-water fish assemblages in southern Australia III. General relationships between sediment, seagrasses, invertebrates and fishes. Journal of Experimental Marine Biology and Ecology **194**:107-131.

Edgar, G. J., H. Mukai, and R. J. Orth. 2001. Fish, Crabs, Shrimps and Other Large Mobile Epibenthos: Measurement Methods for their Biomass and Abundance in Seagrass. Pages 255-270 *in* F. T. Short, R. G. Coles, and C. A. Short, editors. Global Seagrass Research Methods. Elsevier Science, Amsterdam.

Ferrell, D. J., and J. D. Bell. 1991. Differences among assemblages of fish associated with *Zostera capricorni* and bare sand over a large spatial scale. Marine Ecology Progress Series **72**:15-24.

Field, J. G., K. R. Clarke, and R. M. Warwick. 1982. A practical strategy for analysing multispecies distribution patterns. Marine Ecology Progress Series **8**:37-52.

Fonseca, M. S., J. S. Fisher, J. C. Zieman, and G. W. Thayer. 1983. Influence of the seagrass, *Zostera marina* L., on current flow. Estuarine, Coastal and Shelf Science **15**:351-364.

Fonseca, M. S., and S. S. Bell. 1998. Influence of physical setting on seagrass landscapes near Beaufort, North Carolina, USA. Marine Ecology Progress Series **171**:109-121.

Frost, M. T., A. A. Rowden, and M. J. Attrill. 1999. Effect of habitat fragmentation on the macroinvertebrate infaunal communities associated with the seagrass *Zostera marina* L. Aquatic Conservation: Marine and Freshwater Ecosystems **9**:255-263.

Garcia-Charton, J. A., and A. Perez-Ruzafa. 1999. Ecological heterogeneity and the evaluation of the effects of marine reserves. Fisheries Research **42**:1-20.

Gauch, H. G. 1982. Multivariate analysis in community ecology.

Gibson, R. N., A. D. Ansell, and L. Robb. 1993. Seasonal and annual variations in abundance and species composition of fish and macrocrustacean communities in a Scottish sandy beach. Marine Ecology Progress Series **98**:89-105.

Gibson, R. N., L. Robb, M. T. Burrows, and A. D. Ansell. 1996. Tidal, diel and longer term changes in the distribution of fishes on a Scottish sandy beach. Marine Ecology Progress Series **130**:1-17.

Goodsell, P. J., and S. D. Connell. 2002. Can habitat loss be treated independently of habitat configuration? Implications for rare and common taxa in fragmented landscapes. Marine Ecology Progress Series **239**:37-44.

Gray, C. A., R. C. Chick, and D. J. McElligott. 1998. Diel changes in assemblages of fishes associated with shallow seagrass and bare sand. Estuarine, Coastal and Shelf Science **46**:849-859.

Gray, J. S. 1997. Marine biodiversity: patterns, threats and conservation needs. Biodiversity and Conservation **6**:153-175.

Gray, J. S. 2000. The measurement of marine species diversity, with an application to the benthic fauna of the Norwegian continental shelf. Journal of Experimental Marine Biology and Ecology **250**:23-49.

Griffiths, S. P. 2001. Diel variation in the seagrass ichthyofaunas of three intermittently open estuaries in south-eastern Australia: implications for improving fish diversity assessments. Fisheries Management and Ecology **8**:123-140

Groot, C. and L. Margolis. 1991. Pacific salmon life histories. University of British Columbia Press, Vancouver, British Columbia.

Guidetti, P. 2000. Differences among fish assemblages associated with nearshore *Posidonia oceanica* seagrass beds, rocky-algal reefs and unvegetated sand habitats in the Adriatic Sea. Estuarine, Coastal and Shelf Science **50**:515-529.

Gustafson, E. J. 1998. Quantifying landscape spatial pattern: What is the state of the art? Ecosystems **1**:143-156.

Hall, S. J., and S. P. Greenstreet. 1998. Taxonomic distinctness and diversity measures: responses in marine fish communities. Marine Ecology Progress Series **166**:227-229.

Hargreaves, N.B., E.W. Carter, and R.J. LeBrasseur. 1987. Beach seine catches of juvenile salmon and other fish in Massett Inlet and Massett Sound, B.C., in 1984. Canadian Data Report of Fisheries and Aquatic Sciences **640**:78 p.

Harmelin-Vivien, M. L., and P. Francour. 1992. Trawling or visual censuses? Methodological bias in the assessment of fish population in seagrass beds. Marine Ecology**13**:41-51.

Harper, J. L., and D. L. Hawksworth. 1994. Biodiversity: measurement and estimation. Philosophical Transaction of the Royal Society of London Series **345**:5-12.

Hart, J. L. 1988. Pacific Fishes of Canada. Fisheries Research Board of Canada, Ottawa.

Heck, K. L., and R. J. Orth. 1980. Structural components of eelgrass (*Zostera marina*) meadows in the lower Chesapeake Bay - decapod crustacea. Estuaries **3**:289-295.

Hindell, J. S., G. P. Jenkins, and M. J. Keough. 2000. Variability in abundance of fishes associated with seagrass habitats in relation to diets of predatory fishes. Marine Biology **136**:725-737.

Hobson, E. S., W. N. McFarland, and J. R. Chess. 1981. Crepuscular and nocturnal activities of Californian nearshore fishes, with consideration of their scotopic visual pigments and their photic environment. Fishery Bulletin **79**:1-30.

Horinouchi, M., and M. Sano. 1999. Effects of changes in seagrass shoot density and leaf height on abundance and distribution patterns of juveniles of three gobiid fishes in a *Zostera marina* bed. Marine Ecology Progress Series **183**:87-94.

Hovel, K. A., M. S. Fonseca, D. L. Myer, W. J. Kenworthy, and P. E. Whitfield. 2002. Effects of seagrass landscape structure, structural complexity and hydrodynamic regime on macro-faunal densities in North Carolina seagrass beds. Marine Ecology Progress Series **243**:11-24.

Hovel, K. A. 2003. Habitat fragmentation in marine landscapes: relative effects of habitat cover and configuration on juvenile crab survival in California and North Carolina seagrass beds. Biological Conservation **110**:401-412.

Hull, S. L. 1999. Comparison of tidepool phytal ostracod abundance and assemblage structure on three spatial scales. Marine Ecology Progress Series **182**:201-208.

Irlandi, E. A. 1996. Habitat patch size and energy effects on clam growth. Journal of Marine Research **54**:161-185.

Irlandi, E. A., and M. K. Crawford. 1997. Habitat linkages: the effect of intertidal saltmarshes and adjacent subtidal habitats on abundance, movement, and growth of an estuarine fish. Oecologia **110**:222-230.

Izsak, C., and A. R. G. Price. 2001. Measuring beta-diversity using a taxonomic similarity index, and its relation to spatial scale. Marine Ecology Progress Series **215**:69-77.

Jackson, E. L., A. A. Rowden, M. J. Attrill, S. J. Bossey, and M. B. Jones. 2001. The importance of seagrass beds as a habitat for fishery species. Oceanography and Marine Biology: An Annual Review **39**:269-303.

Jackson, G., and G. K. Jones. 1999. Spatial and temporal variation in nearshore fish and macroinvertebrate assemblages from a temperate Australian estuary over a decade. Marine Ecology Progress Series **182**:253-268.

Jenkins, G. P., H. M. A. May, M. J. Wheatley, and M. G. Holloway. 1997. Comparison of fish assemblages associated with seagrass and adjacent unvegetated habitats of Port Phillip Bay and Corner Inlet, Victoria, Australia, with emphasis on commercial species. Estuarine, Coastal and Shelf Science **44**:569-588.

Jenkins, G. P., and M. J. Wheatley. 1998. The influence of habitat structure on nearshore fish assemblages in a southern Australian embayment: comparison of shallow seagrass, reef-algal and unvegetated sand habitats, with emphasis on their importance to recruitment. Journal of Experimental Marine Biology and Ecology **221**:147-172.

Kamermans, P., M. A. Hemminga, and D. J. de Jong. 1999. Significance of salinity and silicon levels for growth of a formerly estuarine eelgrass (*Zostera marina*) population (Lake Grevelingen, the Netherlands). Marine Biology **133**:527-539.

Keddy, P. A. 1982. Quantifying within-lake gradients of wave energy: interrelationships of wave energy, substrate particle size and shoreline plants in Axe Lake, Ontario. Aquatic Botany **14**:41-58.

Kendall, M. A., and S. Widdicombe. 1999. Small scale patterns in the structure of macrofaunal assemblages of shallow soft sediments. Journal of Experimental Marine Biology and Ecology **237**:127-140.

Koch, E. W., and S. Beer. 1996. Tides, light and the distribution of *Zostera marina* in Long Island Sound, USA. Aquatic Botany **53**:97-107.

Koch, E. W., and G. Gust. 1999. Water flow in tide- and wave-dominated beds of the seagrass *Thalassia testudinum*. Marine Ecology Progress Series **184**:63-72.

Krause-Jensen, D., A. L. Middeldoe, K. Sand-Jensen, and P. B. Christensen. 2000. Eelgrass, *Zostera marina*, growth along depth gradients: upper boundaries of the variation as a powerful predictive tool. Oikos **91**:233-244.

Lamb, A., and P. Edgell. 1986. Coastal Fishes of the Pacific Northwest. Harbour Publishing, Madeira Park, BC.

Land Use Coordination Office (LUCO). 1999. Coastal Resource & Oil Spill Response Atlas for the West Coast of Vancouver Island. Victoria: Province of British Columbia.

Lande, R. 1996. Statistics and partitioning of species diversity, and similarity among multiple communities. Oikos **76**:5-13.

Lasiak, T. 1984. Structural aspects of the surf-zone fish assemblage at King's Beach, Algoa Bay, South Africa: short-term fluctuations. Estuarine, Coastal and Shelf Science **18**:347-360.

Laurel, B. J., R. S. Gregory, and J. A. Brown. 2003. Predator distribution and habitat patch area determine predation rates on Age-0 juvenile cod *Gadus* spp. Marine Ecology Progress Series **251**:245-254.

Lavery, P. S., and M. A. Vanderklift. 2002. A comparison of spatial and temporal patterns in epiphytic macroalgal assemblages of the seagrasses *Amphibolis griffithii* and *Posidonia coriacea*. Marine Ecology Progress Series **236**:99-112.

Layman, C. A. 2000. Fish assemblage structure of the shallow ocean surf- zone on the eastern shore of Virginia Barrier Islands. Estuarine, Coastal and Shelf Science **51**:201-213.

Legendre, L., and P. Legendre. 1983. Numerical Ecology. Elsevier Scientific Publishing Company, New York.

Lekve, K., N. C. Stenseth, J. Gjosaeter, J. Fromentin, and J. S. Gray. 1999. Spatio-temporal patterns in diversity of a fish assemblage along the Norwegian Skagerrak coast. Marine Ecology Progress Series **178**:17-27.

Levin, P. 1992. The problem of pattern and scale in ecology. Ecology **73**:1943-1967.

Lubbers, L., W. R. Boynton, and W. M. Kemp. 1990. Variations in structure of estuarine communities in relation to abundance of submerged vascular plants. Marine Ecology Progress Series **65**:1-14.

Magurran, A. E. 1988. Ecological diversity and its measurement. Princeton University Press, Princeton, NJ.

Margules, C. R., and R. L. Pressey. 2000. Systematic conservation planning. Nature **405**:243-253

Marshall, S., and M. Elliot. 1998. Environmental influences on the fish assemblage of the Humber Estuary, U.K. Estuarine, Coastal and Shelf Science **46**:175-184.

Mathieson, S., A. Cattrijsse, M. J. Costa, P. Drake, M. Elliot, J. Gardner, and J. Marchand. 2000. Fish assemblages of European tidal marshes: comparison based on species, families and functional guilds. Marine Ecology Progress Series **204**:225-242.

Mattila, J., G. Chaplin, M. R. Eilers, K. L. H. Jr., J. P. O'Neal, and J. F. Valentine. 1999. Spatial and diel distribution of invertebrate and fish fauna of a *Zostera marina* and nearby unvegetated sediments in Damariscotta River, Maine (USA). Journal of Sea Research **41**:321-332.

McAllister, D. E. 1995. Status of the world ocean and its biodiversity. Sea Wind **5**:28-32.

Methven, D. A., R. L. Haedrich, and G. A. Rose. 2001. The fish assemblage of a Newfoundland estuary: diel, monthly and annual variation. Estuarine, Coastal and Shelf Science **52**:669-687.

Menge, B. A., and A. M. Olson. 1990. Role of scale and environmental factors in regulation of community structure. Trends in Ecology and Evolution **9**:52-57.

Micheli, F., and C. H. Peterson. 1999. Estuarine vegetated habitats as corridors for predator movements. Conservation Biology **13**:869-881.

Morrison, M. A., M. P. Francis, B. W. Hartill, and D. M. Parkinson. 2002. Diel and tidal variation in the abundance of the fish fauna of a temperate tidal mudflat. Estuarine, Coastal and Shelf Science **54**:793-807.

Murphey, P. L., and M. S. Fonseca. 1995. Role of high and low energy seagrass beds as nursery areas for *Penaeus duorarum* in North Carolina. Marine Ecology Progress Series **121**:91-98.

Murphy, M. L., S. W. Johnson, and D. J. Csepp. 2000. A comparison of fish assemblages in eelgrass and adjacent subtidal habitats near Craig, Alaska. Alaska Fishery Research Bulletin **7**:1-21.

Nash, R. D. M., and R. S. Santos. 1998. Seasonality in diel catch rate of small fishes in a shallow-water fish assemblage at Porto Pim Bay, Faial, Azores. Estuarine, Coastal and Shelf Science **47**:319-328.

Nash, R. D. M., and R. S. Santos. 1998. Seasonality in diel catch rate of small fishes in a shallow-water fish assemblage at Porto Pim Bay, Faial, Azores. Estuarine, Coastal and Shelf Science **47**:319-328.

Nelson, J. S. 1994. Fishes of the World, third edition. John Wiley & Sons, Inc., New York, New York.

Parks Canada. 1995. State of the Parks - 1994 Report. Department of Canadian Heritage.

Pasqualini, V., P. Clabaut, G. Pergent, L. Benyoussef, and C. Pergent-Martini. 2000. Contribution of side sonar to the management of Mediterranean littoral ecosystems. International Journal of Remote Sensing **21**:367-378.

Peterson, B. J., K. R. Thompson, J. H. Cowan, and K. L. Heck. 2001. Comparison of predation pressure in temperate and subtropical seagrass habitat based on chronographic tethering. Marine Ecology Progress Series **224**:77-85.

Price, A. R. G. 2002. Simultaneous "hotspots" and "cold" spots of marine biodiversity and implications for global conservation. Marine Ecology Progress Series **241**:23-27.

Purvis, A., and A. Hector. 2000. Getting a measure of biodiversity. Nature **405**:212-219.

Orth, R. J., and K. L. Heck. 1980. Structual components of eelgrass (Zostera marina) meadows in the lower Chesapeake Bay - Fishes. Estuaries **3**:278-288.

Reid, W. V. 1998. Biodiversity hotspots. Trends in Ecology and Evolution **13**:275-280.

Robards, J. P., A. B. Kettle, and A. A. Abookire. 1999. Temporal and geographic variation in fish communities of lower Cook Inlet, Alaska. Fishery Bulletin **97**:962-977.

Robbins, B. D., and S. S. Bell. 1994. Seagrass landscapes: a terrestrial approach to marine subtidal environment. Trends in Ecology and Evolution **9**:301-304.

Robinson, C. L. K., and J. L. Yakimishyn. The diversity of fishes in *Zostera marina* L. beds. Unpublished Manuscript.

Rogers, S. I., K. R. Clarke, and J. D. Reynolds. 1999. The taxonomic distinctness of coastal bottom-dwelling fish communities of the North-east Atlantic. Journal of Animal Ecology **68**:769-782.

Rooker, J. R., G. J. Holt, and S. A. Holt. 1998. Vulnerability of newly settled red drum (*Sciaenops ocellatus*) to predatory fish: is early-life survival enhanced by seagrass meadows? Marine Biology **131**:145-151.

Rozas, L. P., and T. J. Minello. 1997. Estimating densities of small fishes and decapod crustaceans in shallow estuarine habitats: A review of sampling design with a focus on gear selection. Estuaries **20**:199-213.

Rozas, L. P., and R. J. Zimmerman. 2000. Small-scale patterns of nekton use among marsh and adjacent shallow nonvegetated areas of Galveston Bay Estuary, Texas (USA). Marine Ecology Progress Series **193**:217-239.

Santos, R. S., and R. D. M. Nash. 1995. Seasonal changes in a sandy beach fish assemblage at Port Pim, Faial, Azores. Estuarine, Coastal and Shelf Science **41**:579-591.

Sogard, S. M., G. V. N. Powell, and J. G. Holmquist. 1987. Epibenthic fish communities on Florida Bay banks: relations with physical parameters and seagrass cover. Marine Ecology Progress Series **40**:25-39.

Sogard, S. M., G. V. N. Powell, and J. G. Holmquist. 1989a. Utilization by fishes of shallow, seagrass-covered banks in Florida Bay: 1. Species composition and spatial heterogeneity. Environmental Biology of Fishes **24**:53-65.

Sogard, S. M., G. V. N. Powell, and J. G. Holmquist. 1989b. Spatial distribution and trends in abundance of fishes residing in seagrass meadows on Florida Bay mudbanks. Bulletin of Marine Science **44**:179-199.

Sokal, R. R., and F. J. Rohlf. 1995. Biometry: The principles and practice of statistics in biological research, 3rd edition. W. H. Freeman and Company, New York.

Suda, Y., T. Inoue, and H. Uchida. 2002. Fish communities in the surf zone of a protected sandy beach in Doigahama, Yamaguchi Prefecture, Japan. Estuarine, Coastal and Shelf Science **55**:81-96.

Townsend, E. C., and M. S. Fonseca. 1998. Bioturbation as a potential mechanism influencing spatial heterogeneity of North Carolina seagrass beds. Marine Ecology Progress Series **169**:123-132.

Turner, S. J., J. E. Hewitt, M. R. Wilkinson, D. J. Morrisey, S. F. Thrush, V. J. Cummings, and G. Funnell. 1999. Seagrass patches and landscapes: The influence of wind-wave dynamics and hierarchical arrangements of spatial structure on macrofaunal seagrass communities. Estuaries **22**:1016-1032.

Underwood, A. J., and G. A. Skilleter. 1996. Effects of patch-size on the structure of assemblages in rock pools. Journal of Experimental Marine Biology and Ecology **197**:63-90.

US Army Coastal Engineering Research Centre. 1984. Shore Protection Manual, fourth edition.. U.S. Government Printing Office, Washington.

Vanderklift, M. A., T. J. Ward, and J. C. Phillips. 1998. Use of assemblages derived from different taxonomic levels to select areas for conserving marine biodiversity. Biological Conservation **86**:307-315.

Vane-Wright, R. I., C. J. Humphries, and P. H. Williams. 1991. What to protect? Systematics and the agony of choice. Biological Conservation **55**:235-254.

Wagner, C. M., and H. M. Austin. 1999. Correspondence between environmental gradients and summer littoral fish assemblages in low salinity reaches of the Chesapeake Bay, USA. Marine Ecology Progress Series **177**:197-212.

Walker, D. I., R. J. Lukatelich, G. Bastyan, and A. J. McComb. 1989. Effect of boat moorings on seagrass beds near Perth, western Australia. Aquatic Botany **36**:69-77.

Warwick, R. M., and K. R. Clarke. 1995. New "biodiversity" measures reveal a decrease in taxonomic distinctness with increasing stress. Marine Ecology Progress Series **129**:301-305.

Whittaker, R. H. 1960. Vegetation of the Siskiyou Mountains. Oregon and California. Ecological Monographs **30**:279-338.

Wilson, E. O. 1988. The current state of biological diversity. Pages 3-18 *in* E. O. Wilson, editor. Biodiversity. National Academy Press, Washington.

Wilson, M. V., and A. Schmida. 1984. Measuring beta diversity with presence and absence data. Journal of Ecology **72**:1055-1064.

Wyllie-Echeverria, S., and R. M. Thom. 1994. Managing seagrass systems in western North America: research gaps and needs. Alaska Sea Grant Report **94-01**:1-21.

Yoccoz, N. G., J. D. Nichols, and T. Boulinier. 2001. Monitoring of biological diversity in space and time. Trends in Ecology and Evolution **16**:446-453.

Young, G. C., I. C. Potter, G. A. Hyndes, and S. d. Lestang. 1997. The ichthyofauna of an intermittently open estuary: implications of bar breaching and low salinities on faunal composition. Estuarine, Coastal and Shelf Science **45**:53-68.

Ysebaert, T., and P. M. J. Herman. 2002. Spatial and temporal variation in benthic macrofauna and relationships with environmental variables in an estuarine, intertidal soft-sediment environment. Marine Ecology Progress Series **244**:105-124.

Zar, J. H. 1999. Biostatistical Analysis, fourth edition. Prentice Hall, Upper Saddle River, New Jersey.

Zieman, J. C. 1976. The ecological effects of physical damage from motor boats on turtle grass beds in southern Florida. Aquatic Botany **2**:127-139.

Appendix 2.1. Total number of fish species caught in diel (day/night) and tidal (low/high) sampled Indian and South bay sites.

		Indian				South Bay			
Common Name	Species Name	Day	Night	Low	High	Day	Night	Low	High
Bay Pipefish	*Syngnathus leptorhynchus*	8	2	40	1	7	8	10	0
Coho	*Oncorhynchus kisutch*	0	0	0	0	0	0	17	3
Copper Rockfish	*Sebastes caurinus*	0	1	2	0	1	4	2	0
Crescent Gunnel	*Pholis laeta*	24	10	47	0	32	34	25	15
English Sole	*Parophrys vetula*	0	0	0	0	2	0	0	0
Great Sculpin	*Myoxocephalus polyacanthocephalus*	0	0	0	0	0	3	0	1
Herring	*Clupea pallasi*	0	76	0	0	1	1	0	0
Kelp Clingfish	*Rimicola muscarum*	1	10	24	0	2	0	2	0
Kelp Greenling	*Hexagrammos decagrammus*	0	0	0	0	1	1	8	0
Kelp Perch	*Micrometrus frenatus*	45	69	12	1	107	37	21	3
Padded Sculpin	*Artedius fenestralis*	1	1	0	0	1	3	1	0
Pacific Snake Prickleback	*Lumpenus sagitta*	50	15	42	5	9	1	16	0
Penpoint Gunnel	*Apodichthys flavidus*	2	1	9	0	5	14	5	5
Pile Perch	*Rhacochilus vacca*	17	5	1	0	41	13	0	0
Saddleback Gunnel	*Pholis ornata*	0	1	3	2	2	2	1	3
Sharpnose sculpin	*Clinocottus acuticeps*	0	0	0	0	0	0	1	0
Shiner Perch	*Cymatogaster aggregata*	1363	141	88	6	776	30	123	58
Silverspotted Sculpin	*Blepsias cirrhosus*	0	0	1	0	2	0	1	0
Smoothhead Sculpin	*Artedius lateralis*	1	0	0	0	0	0	0	0
Staghorn Sculpin	*Leptocottus armatus*	8	5	2	21	12	15	4	24
Starry Flounder	*Platichthys stellatus*	0	0	0	0	1	0	0	2
Striped Seaperch	*Embiotoca lateralis*	0	0	0	0	4	0	0	0
Tidepool sculpin	*Oligocottus maculosus*	0	0	0	1	0	0	1	1
Threespine Stickleback	*Gasterosteus aculeatus*	4852	85	1283	798	110	77	797	127
Tube-snout	*Aulorhynchus flavidus*	0	0	16	0	2	0	1	0
Walleye Pollock	*Theragra chalcogramma*	0	0	1	0	0	0	1	0
Whitespotted Greenling	*Hexagrammos stelleri*	0	0	0	0	0	1	0	0
Yellowtail Rockfish	*Sebastes flavidus*	0	0	0	0	0	2	0	0

Appendix 2.2. Total number of each fish species caught at four rising tidal heights at South Bay eelgrass bed.

Common Name	Species Name	1.6 ft	2.7 ft	4.4 ft	6.8 ft
Bay Pipefish	*Syngnathus leptorhynchus*	0	1	3	5
Coho	*Oncorhynchus kisutch*	0	0	1	0
Copper Rockfish	*Sebastes caurinus*	0	0	1	0
Crescent Gunnel	*Pholis laeta*	4	11	43	5
English Sole	*Parophrys vetula*	0	3	3	1
Herring	*Clupea pallasi*	0	1	1	0
Kelp Greenling	*Hexagrammos decagrammus*	4	1	3	4
Kelp Perch	*Micrometrus frenatus*	64	2	12	2
Pacific Snake Prickleback	*Lumpenus sagitta*	0	0	1	4
Padded Sculpin	*Artedius fenestralis*	1	1	5	0
Penpoint Gunnel	*Apodichthys flavidus*	6	6	15	0
Pile Perch	*Rhacochilus vacca*	3	0	0	0
Saddleback Gunnel	*Pholis ornata*	2	2	8	0
Shiner Perch	*Cymatogaster aggregata*	71	253	124	5
Silverspotted Sculpin	*Blepsias cirrhosus*	2	2	0	1
Smoothhead sculpin	*Artedius lateralis*	0	0	1	0
Staghorn Sculpin	*Leptocottus armatus*	3	75	21	2
Starry Flounder	*Platichthys stellatus*	0	2	1	0
Striped Seaperch	*Embiotoca lateralis*	0	1	1	1
Surf Smelt	*Hypomesus pretiosus*	1	0	0	1
Threespine Stickleback	*Gasterosteus aculeatus*	247	238	135	16
Whitespotted Greenling	*Hexagrammos stelleri*	0	0	2	0
Yellowtail Rockfish	*Sebastes flavidus*	4	0	4	0

Appendix 3.1. Sampling dates, number of seine hauls on each sampling date, and tidal height sampled at each site.

	Arakun		Auseth		Ducking		Felice		Indian		Kootowis		Sharp	
Sampling Date	# of seines	Tidal ht(m)	# of seines	Tidal ht(m)	# of seines	Tidal ht(m)	# of seines	Tidal ht(m)	# of seines	Tidal ht(m)	# of seines	Tidal ht(m)	# of seines	Tidal ht(m)
L May/01	NA	NA	3	0.20	2	0.55	NA	NA	3	0.36	NA	NA	2	0.58
L June/01	2	0.29	3	0.28	3	0.22	NA	NA	3	0.05	3	0.015	2	0.46
L July/01	3	0.16	3	0.41	3	0.41	NA	NA	5	0.34	3	-0.02	2	0.05
L Aug/01	NA	NA	3	0.43	3	0.16	2	0.17	3	0.05	3	0.47	NA	NA
L Sept/01	3	0.83	3	0.37	3	0.87	NA	NA	3	0.84	3	0.44	3	0.61
L April/02	3	0.53	3	0.45	3	0.46	NA	NA	3	0.55	2	0.14	3	0.24
E May/02	3	0.72	3	0.45	3	0.71	NA	NA	3	0.68	3	0.47	3	0.42
L May/02	3	0.48	3	0.15	3	0.66	NA	NA	3	0.59	3	0.65	3	0.27
E June/02	3	0.60	3	0.34	3	0.68	3	0.44	3	0.38	3	0.41	3	0.29
L June/02	3	0.37	3	0.18	3	0.46	3	0.34	3	0.21	3	0.41	3	0.17
E July/02	3	0.57	3	0.18	3	1.22	3	0.36	3	0.35	3	0.20	3	0.21
L July/02	3	0.44	3	0.33	3	0.65	3	0.37	3	0.37	3	0.73	3	0.30
E. Aug/02	3	0.27	3	0.17	3	1.13	3	0.34	3	0.81	3	0.90	3	0.34
L Aug/02	3	0.49	3	0.54	3	0.58	3	0.56	3	0.74	3	0.98	3	0.67
E Sept/02	3	0.34	3	0.55	3	0.59	3	0.28	3	0.67	3	0.80	3	0.48

Appendix 3.2. Formulas and calculations for all univariate diversity measures.

Pielou's evenness index (J'):

$$J' = H'/\log(s)$$

where $H' = \log s$, and s is the number of species (Brower et al. 1989).

Taxonomic diversity (Δ):

$$\Delta = (\Sigma\Sigma_{i<j}\omega_{ij}x_i x_j)/(n(n-1)/2)$$

where x_i represents the abundance of the ith species, n denotes the total number of individuals in the sample and ω_{ij} is the distinctness weight of the path length linking I and j species in a hierarchical classification (Clarke and Warwick 1998).

Taxonomic distinctness (Δ^*)

$$\Delta^* = (\Sigma\Sigma_{i<j}\omega_{ij}x_i x_j)/((\Sigma\Sigma_{i<j}x_i x_j)$$

where x_i represents the abundance of the ith species and ω_{ij} is the distinctness weight of the path length linking i and j species in a hierarchical classification (Clarke and Warwick 1998).

Average taxonomic distinctness (AveTD, Δ^+)

$$\Delta^+ = 2((\Sigma\Sigma_{i<j}\omega_{ij})/s(s - 1))$$

where ω_{ij} is the distinctive weight between species i and j and s is the number of species present (Clarke and Warwick 1998).

Variation in taxonomic distinctness (VarTD, Λ^{+})

$$\Lambda^{+} = (\Sigma\Sigma_{i \neq j}\omega_{ij}^{2}/s(s - 1)) - \Delta^{+2}$$

where ω_{ij} is the distinctive weight between species i and j, s is the number of species present and Δ^{+} is the average taxonomic distinctness (Clarke and Warwick 2001).

Taxonomic distance (TD)

$$\mathrm{TD} = (\Sigma_{i}\mathrm{w}_{iB} + \Sigma_{j}\mathrm{w}_{jA})/\mathrm{n}_{A} + \mathrm{n}_{B}$$

where w_{iB} is the minimum taxonomic path length between species i at site A and all species at site B, w_{iA} is the minimum taxonomic path length between species j at site B and all species at site A, and n_{A} and n_{B} are the number of species at sites A and B (Izsak and Price 2001).

Taxonomic similarity (Δ_{s})

$$\Delta_{s} = 1 - (\mathrm{TD}/L - 1)$$

Where L is the number of taxonomic levels (Izsak and Price 2001).

Appendix 3.3. List of fish species caught in each eelgrass bed in 2001 and 2002. (+ = caught at least one time, 0 = no catch)

Fish species	Arakun	Auseth	Ducking	Felice	Indian	Kootowis	Sharp
Agonopsis vulsa	0	0	0	0	0	+	0
Ammodytes hexapterus	+	+	+	+	0	+	0
Apodichthys flavidus	+	+	+	+	+	0	+
Artedius fenestralis	0	+	+	0	+	+	+
Artedius lateralis	+	0	0	0	+	+	+
Aulorhynchus flavidus	+	+	+	+	+	+	+
Blepsias cirrhosus	+	+	+	+	+	0	+
Chitonotus pugetensis	0	+	0	0	0	0	0
Citharichthys stigmaeus	0	+	0	+	+	0	+
Clinocottus acuticeps	+	+	+	0	0	+	+
Clupea pallasi	+	+	+	+	+	0	0
Cymatogaster aggregata	+	+	+	+	+	+	+
Embiotoca lateralis	+	0	+	+	+	+	+
Enophrys bison	+	+	+	+	+	+	+
Gasterosteus aculeatus	+	+	+	+	+	+	+
Hemilepidotus spinosus	0	0	0	0	0	+	+
Hemilepidotus hemilepidotus	+	+	+	+	0	+	+
Hexagrammos decagrammus	+	+	+	+	+	+	+
Hexagrammos lagocephalus	0	0	0	+	0	0	0
Hexagrammos stelleri	+	+	+	+	0	0	+
Hyperprosopon ellipticum	0	0	0	0	0	+	0
Hypomesus pretiosus	+	+	+	+	+	0	0
Lepidogobius lepidus	0	0	0	0	0	+	+
Lepidopsetta bilineata	0	0	0	+	0	0	0
Leptocottus armatus	+	+	+	+	+	+	+
Liparis cyclopus	0	0	0	0	+	+	0
Liparis florae	+	0	0	0	0	0	0
Lumpenus sagitta	+	+	+	0	+	+	+
Microgadus proximus	0	0	0	0	+	0	0

Appendix 3.3. Continued.

Fish species	Arakun	Auseth	Ducking	Felice	Indian	Kootowis	Sharp
Micrometrus frenatus	+	+	+	+	+	+	+
Microstomus pacificus	+	0	0	0	0	0	+
Myoxocephalus polyacanthocephalus	+	+	+	+	+	+	+
Nautichthys oculofasciatus	0	+	+	+	0	0	+
Odontopyxis trispinosa	0	0	0	0	0	0	+
Oligocottus maculosus	+	0	+	0	0	+	+
Oncorhynchus clarki clarki	0	0	0	0	+	+	0
Oncorhynchus keta	+	+	+	+	+	+	+
Oncorhynchus kisutch	0	+	+	+	+	+	0
Oncorhynchus nerka	+	0	+	0	+	+	0
Oncorhynchus tshawytscha	0	0	+	+	0	+	0
Ophiodon elongatus	0	+	0	0	+	0	+
Pallasina barbata	0	0	+	+	0	0	0
Parophrys vetula	+	+	+	+	+	+	+
Pholis laeta	+	+	+	+	+	+	+
Pholis ornata	+	+	+	+	+	+	+
Platichthys stellatus	+	+	+	0	+	+	+
Pleuronichthys coenosus	0	0	0	+	0	0	0
Porichthys notatus	0	+	0	0	0	+	+
Rhacochilus vacca	+	+	+	+	+	+	+
Rimicola muscarum	+	+	+	0	+	+	+
Scorpaenichthys marmoratus	+	0	+	+	+	0	+
Sebastes brevispinus	0	0	0	+	0	0	0
Sebastes caurinus	+	+	+	+	+	0	+
Sebastes flavidus	+	+	0	+	+	+	+
Sebastes miniatus	0	0	0	+	0	0	0
Syngnathus leptorhynchus	+	+	+	0	+	+	+
Theragra chalcogramma	0	+	+	0	+	0	+
Xiphister atropurpureus	0	0	0	+	0	0	0

Appendix 4.1. Summary of data collection dates and methods used for the August 2001, May 2002 and August 2002 sampling dates.

Parameter	Sampling date	Arakun	Auseth	Ducking	Indian	Kootowis	Sharp
Sediment	August 2001	Aug 2, 2001	Aug 2, 2001	Aug 2, 2001	Aug 2, 2001	Aug 2, 2001	Aug 2, 2001
Tidal current	May 2002	May 26, 2002	May 27, 2002	May 25, 2002	May 27, 2002	May 27, 2002	May 26, 2002
REI - wind	August 2001	Aug & Sept 2001	Aug & Sept 2001	Aug & Sept 2001	Aug & Sept 2001	Aug & Sept 2001	Aug & Sept 2001
REI - wind	May 2002	May & Jun 2002	May & Jun 2002	May & Jun 2002	May & Jun 2002	May & Jun 2002	May & Jun 2002
REI - wind	August 2002	Aug & Sept 2002	Aug & Sept 2002	Aug & Sept 2002	Aug & Sept 2002	Aug & Sept 2002	Aug & Sept 2002
Temperature	August 2001	Temperature logger: 3 samples during daytime high tide same date as fish	Temperature logger: 3 samples during daytime high tide same date as fish	Temperature probe: point samples same date as fish	Temperature logger: 3 samples during daytime high tide same date as fish	Temperature logger: 3 samples during daytime high tide same date as fish	Temperature logger: 3 samples during daytime high tide same date as fish
Temperature	May 2002	Temperature logger: 3 samples during daytime high tide, early May values taken from late May	Temperature logger: 3 samples during daytime high tide, early May values taken from late May	Temperature logger: 3 samples during daytime high tide, early May values taken from late May	Temperature logger: 3 samples during daytime high tide, early May values taken from late May	Temperature logger: 3 point samples during daytime high tide	Temperature logger: 3 samples during daytime high tide, early May values taken from late May
Temperature	August 2002	Temperature logger: 3 point samples during daytime high tide same date as fish	Temperature logger: 3 point samples during daytime high tide same date as fish	Temperature logger: 3 point samples during daytime high tide same date as fish	Temperature logger: 3 point samples during daytime high tide same date as fish	Temperature logger: 3 point samples during daytime high tide same date as fish	Temperature logger: 3 point samples during daytime high tide same date as fish
Eelgrass % cover	August 2001	July 26 ,2001	August 1, 2001	August 1, 2001	July 30 ,2001	July 28, 2001	July 27, 2001
Eelgrass % cover	May 2002	May 31, 2002	June 1, 2002	June 4, 2002	June 3, 2002	June 1, 2002	May 31, 2002
Eelgrass % cover	August 2002	August 13, 2002	August 14, 2002	August 14, 2002	August 15, 2002	August 15, 2002	August 13, 2002
Fish	August 2001	Sept 19, 2001	August 20, 2001 Sept 17, 2001	August 20, 2001 Sept 17, 2001	Aug 19, 2001 Sept 18, 2001	Aug 19, 2001 Sept 18, 2001	Sept 19, 2001
Fish	May 2002	May 14 & 29/02 June 12 & 24/02	May 15 & 28/02 June 14 & 25/02	May 13 & 30/02 June 13 & 26/02	May 13 & 28/02 June 13 & 26/02	May 15 & 30/02 June 14 & 25/02	May 14 & 29/02 June 12 & 24/02
Fish	August 2002	Aug 8 & 21/02 Sept 7/02	Aug 10 & 23/02 Sept 9/02	Aug 9 & 22/02 Sept 8/02	Aug 11 & 22/02 Sept 8/02	Aug 10 & 23/02 Sept 9/02	Aug 8 & 21/02 Sept 7/02

Appendix 4.2. Summary of the significant differences in hydrodynamic variables between the 6 sites sampled. Tukey test significance levels at $P<0.05$ and Mann-Whitney test significance level at $P<0.003$.

Physical variable	Sampling date	Pair-wise statistical test	Significant site differences
Tidal current	May 2002	Mann-Whitney test	Arakun > Auseth, Ducking, Kootowis, Sharp; Auseth, Ducking, Indian > Kootowis, Sharp; Kootowis > Sharp
Sediment silt-clay fraction	August 2001	Tukey test	Arakun, Auseth, Ducking < Indian, Kootowis, Sharp; Indian, Kootowis < Sharp
Temperature	August 2001	Mann-Whitney test	No significant between site differences
	May 2002	Tukey test	Kootowis < Auseth, Ducking, Indian
	August 2002	Mann-Whitney test	Arakun < Auseth, Ducking, Indian, Kootowis, Sharp; Kootowis < Auseth, Indian, Sharp
Eelgrass %cover	August 2001	Mann-Whitney test	No significant between site differences
	May 2002	Mann-Whitney test	Sharp > Arakun, Auseth, Ducking, Indian, Kootowis; Kootowis < Arakun, Auseth, Indian; Arakun, Ducking < Auseth, Indian
	August 2002	Mann-Whitney test	Sharp > Arakun, Auseth, Ducking, Indian, Kootowis; Indian, Auseth >, Ducking, Kootowis

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Natural Science and Engineering Research Council	2002

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Title of Thesis:

Monitoring Spatial and Temporal Patterns Structuring Eelgrass (*Zostera marina* L.) Fish Diversity in Clayoquot Sound

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