CHAPTER 4

GENERAL DISCUSSION

4.1 Water parameters at cage and non-cage sites

The water parameters measured at both the cage and non-cage areas did not vary much during the 5 months of study from December 1999 to April 2000. Rainfall varied from 0.0 mm to 116.4 mm during the study period. The month of April (2000) exhibited a higher rainfall as it coincided with the onset of the South-West monsoons.

The pH, salinity and temperature were quite similar oscillating within a narrow range. The pH and salinity of the inside and away stations in SSB were almost similar. However, the pH in SSB was generally lower than in SSK. The difference might be due to high bacterial activity in the aquaculture waters (SSB) which could have reduced the water quality and pH level. In SSK, the temperature increased towards offshore. This was because the river banks at the upstream of SSK were covered with mangrove foliage and shade. The downstream of SSK was exposed to open skies which increased the temperature of the water. The salinity was the lowest in the month of April (16‰ – 18‰). This was due to higher rainfall in April. Chong et al. (1998) reported that the rainfall apparently had a depressive effect on salinity only but not on other water parameters. In SSB, salinity ranged from 16.1‰ - 26.2‰. The salinity was relatively higher in SSK (25.4‰ – 26.1‰). This could be due to its smaller size and hence receiving less freshwater input, whereas the main water flow from upstream goes through SSB (see Fig. 3.2).

The dissolved oxygen levels in the inside (IN) stations varied from 0.03mg/l- 8.43mg/l and in away (AWAY) stations, 3.89mg/l-7.89mg/l. The DO concentration was low (< 3.0mg/l) in some of the stations in SSB especially in the inside stations on Transect 2 (early March) and on Transect 3 (late March). This could be due to several reasons acting in unison, which are: (1) the increased zooplankton population or activity at the top layer, (ii) increased bacterial activity associated with the aquacultural activity, and (iii) reduced water circulation in the cage area due to the presence of the floating net cages. Madin (per. comm.) recorded water velocities that ranged from 0.0001 m/s to 0.025 m/s inside the fish cages as opposed to 0.0005 m/s to 0.1 m/s outside the fish cages. The turbidity recorded in this study ranged from 2 to 88 NTU. The readings were only taken until late March. The great difference in the turbidity readings is again due to the influence of tidal flow, with consequent turbulence resulting in the water column being well mixed and turbid. The deeper non-aquaculture river of SSK (compared to SSB) indicates less sedimentation occurring in the river, and this river is now the preferred transit route for boats from Kuala Sepetang.

4.2 Sampling Methodology

Several recent papers have pointed out that biases in sampling methodology may have potentially disastrous effects on testing hypotheses about the distribution and abundance of zooplankton (Hamner and Carleton, 1979; Hillman-Kitalong and Birkeland, 1987). Because of the difference in mesh sizes of nets and methods of capturing zooplankton, caution is required when comparing the results of different studies.

The design features and sampling procedures may affect the density and diversity of zooplankton (Youngbluth, 1982). The biomass and density of zooplankton collected from Matang in the present study was lower than that reported by other studies (see Table 1.1). However, these previous studies were based on towed plankton net catches whereas the present study used a suction pump. In the samples, there were torn pieces of hydromedusae and chaetognaths possibly due to high pressure during passage through the sampler as a result of using the pump. This might cause the lower estimation of zooplankton density in the present study.

Although pumps have been used mainly for sampling plankton, there are objections to its use as there is a well defined gradient of velocity around the pump intake and this stimulates negatively rheotropic organisms to avoid capture (Tonolli, 1971). Herbst (1957) and Elster (1958) have shown that large planktonic organisms especially copepods react by flight when they enter the 'suction' area. This might be caused by the ability of the zooplankton to avoid the inlet hose as they were sensitive to currents and able to resist those of moderate velocities (Youngbluth, 1982). The ability to escape depends upon the speeding and neuromuscular co-ordination and hence is related to the developmental stage of a given spesies; usually older larvae are more efficient than the younger ones (Youngbluth, 1982). On the other hand, Armitage (1978) indicated that animals tend to avoid and escape the net more efficiently than the pump. The turbulence at the net mouth may result in deflection and / or washing–out of organisms and drifting solids. The pump may be harder to escape since water is entering the intake nozzle at a rate faster than water entering the net.

A study done by Dixon and Robertson (1986) showed that there was no significant difference in the mean abundance of copepods, nauplii and zoea caught using the pump system compared to the boat-towed net method. While the net samples yielded significantly greater numbers of chaetognaths, larvaceans and polychaete larvae, pump samples held more gastropods and bivalve larvae. The Waite-O'Grady filter pump collected greater numbers of rotifers and immature copepods per unit volume and fewer numbers of mature copepods (Waite and O'Grady, 1979).

Armitage (1978) compared the catches of invertebrate drift by pump and surface net. He found that the faunal composition was similar in the two methods but pump catches of drifting benthos, microcrustaceans and suspended solids were significantly greater than those by nets. Pumps have the advantage of providing continuous sampling without clogging. Despite the inefficiency or disadvantage of the pump method, the same method was used for 'control' and aquaculture sites in the present study. Thus, unbiased comparisons could be made in the study.

4.3 Impact of cage culture on biomass and density of zooplankton

This study showed relatively low density of zooplankton compared to the study done by Oka (2000) in the waterways of Matang Mangrove Forest. Using a towed net, he recorded that the density of zooplankton ranged from 4,473 ind. m^{-3} to 212,381 ind. m^{-3} in February, 1998. Copepods were consistently the most abundant component comprising 66.0% to 99.4% of all the zooplankton. For the present study, the density ranged from 116 ind. m^{-3} to 26,280 ind. m^{-3} and was found to be the highest in December.

The zooplankton of SSB and SSK comprised of fifteen broad taxonomic groups; Copepoda, Cirripedia, Heteropoda, Brachyura, Tintinnida, Chaetognatha, Bivalvia, Amphipoda, Polychaeta, Foraminifera, Decapoda, Ostracoda, Mysidacea, Pisces and Stomatopoda. Copepods were the dominant group in both SSB and SSK. The zooplankton density observed fluctuated from one station to another station. This was also observed by Omori and Hamner (1982) where the zooplankton, micronekton and epipelagic animals were usually distributed unevenly both horizontally and vertically. It is likely that a combination of factors was responsible for the observed seasonality of copepods, with different factors operating together or at different times of the year to produce density fluctuation (Miller, 1983). Other factors include predation by other members of the plankton (Miller, 1983) which may have influenced copepod densities.

In SSB, it was found that the differences in zooplankton biomass and density were influenced by months, transects and stations (i.e. whether inside or away from fish cages) based on the two way ANOVA. Both the ANOVA results show that the zooplankton density were influenced by month of sampling, transect and station. Zooplankton biomass was influenced by month of sampling and station as indicated by the first ANOVA. The second ANOVA results show that zooplankton biomass was influenced by transect and station.

Both the ANOVA results show that the zooplankton biomass and density were significantly higher inside the fish cage areas than away from the cage areas. The results therefore support the hypothesis that zooplankton abundance inside cage areas were higher than away from cage areas. This is likely to result from the chain of events that follow from trash fish feeding and cultured fish defecation and excretion which are the major sources of nutrients release into the water column. Bacteria degrade the organic detritus in fish farms and release dissolved inorganic nutrients into the water. The phytoplankton assimilate nutrients to support their growth while some bacteria absorb inorganic nutrients. Zooplankton may be feeding directly on the suspended food particles from the trash fish and fecal particles.

Significant differences in zooplankton abundance were observed amongst transects 2, 3 and 4. Zooplankton biomass and density were significantly higher in Transect 4 as compared to Transect 3 and 2 where these parameters increased in the offshore direction. These results indicate that the zooplankton abundance varied along with the river length, becoming increasingly higher in the offshore direction. This is true as higher diversity and abundance of zooplankton are found in coastal and oceanic systems.

Zooplankton biomass and density in SSK were relatively higher than in SSB based on the ANOVA results. This implies a 'less disturbed' habitat in SSK. The same trend as observed in SSB in the present study was also observed in SSK where zooplankton increased in the offshore direction. Interestingly, a study of the macrobenthos in both rivers by Natin (2001) also showed comparatively more animals in SSK than in SSB, in terms of density and diversity.

There are also temporal differences in zooplankton abundance. ANOVA results showed that the zooplankton biomass and density were influenced by month of sampling. The zooplankton biomass and density were significantly higher in December as compared to January, early March. Zooplankton density was however significantly higher in early

March than in April. Temporal changes within a plankton community itself are largely determined by the growth, mortality, sinking, and migration rates of the individual plankters and their predators (Parsons and Takahashi, 1973).

4.4 Impact of fish cage culture on zooplankton community

The more common copepods occurring in SSB were *Oithona* spp., *Acartia* spp. and *Pseudocalanus* spp. Juvenile copepods were also abundant there. Robertson et al. (1988) also recorded the dominant group of zooplankton in mangrove waters as holoplankton copepods, in particular species of *Oithona* spp., *Parvocalanus crassirostris*, *Paracalanus* spp. and *Euterpina acutifrons* as they are found in many tropical and subtropical nearshore regions of the world (Chua, 1973; Sarkar et al., 1984). In the present study, the abundance of these zooplankton might be caused by aggregations of the taxa involved. Omori and Hamner (1982) observed that the most common aggregation pattern for *Oithona aculata* and *Acartia australis* was the formation of swarms of a single species during the day at densities of 500,000 m^{-3} to 1,500,000 m^{-3}. Therefore, aggregation behaviour could explain the numerical dominance of *Oithona* spp. and *Acartia* spp.. Aggregations can result from responses to temperature and salinity gradients or discontinuities, water motion, light intensity or organic matter on which the species feed, and predators as well as from complex social behaviour (Omori and Hamner, 1982).

Fulton (1984) found that small copepods such as *Oithona* spp. were 'avoided' by fish predators which preferred larger bodied *Acartia* and *Euterpina* species. Visual predators could be important mediators of community composition in mangroves, since most newly recruited fish in mangrove systems are zooplanktivores (Robertson and Duke, 1990). However some factors other than body size, such as competition from the mangrove resident spesies, could prevent the intrusion of *Oithona* spp. into the upper reaches of these estuaries since these species are known to tolerate salinities ranging from 25‰ - 40‰ (Grindley, 1981). All these factors contributed to the uneven distribution of zooplankton.

There are some differences in the structure of the zooplankton community between cage and non-cage areas. The zooplankton which dominated inside the fish cage were chaetognaths, juvenile copepods, *Pseudocalanus* spp., *Acartia* spp. *Oithona* spp., nauplii of cirripedes, amphipods and zoeae of *Portunus* spp. Most of these zooplankton are omnivorous and probably fed on the phytoplankton and suspended feed or fecal particles inside the fish cages. Copepods, in particular serve as a principal avenue of energy flow in the grazing food web of estuarine ecosystems, consuming phytoplankton and other zooplankton (Conley and Turner, 1985). For example, *Centropages hamatus*, a coastal marine copepod, not only ingests phytoplankton, but also copepod nauplii. *Labidocera aestiva*, primarily a carnivorous zooplankton also grazes on phytoplankton (Conley and Turner, 1985). Some copepods (e.g. *Anomalocera ornata* and *C. typicus*) effectively eat fish larvae. Detritus may be important in the nutrition of estuarine copepods (Heinle et al., 1977) and the detritus-associated bacterial cells may enhance the nutritional value of detrital particles. Thus, some detritivorous zooplankton could contribute to the higher biomass and density of zooplankton inside the fish cage areas.

The zooplankton taxa in SSK were different from SSB. Multidimensional scaling (MDS) which was based on species similarities of samples, revealed the clustering of samples in SSK that was apart from samples in SSB. The PCA plot shows that the group that dominated in SSK were *Microsetella* spp., Cirripedia nauplii and cypris, and copepod nauplii in late March. The downstream of SSK was mainly dominated by *Sapphirina* spp. as these zooplankton are known stenohaline marine component which only penetrates into the mouths of estuaries (Grindley, 1984). Other zooplankton encountered were *Calanus* spp. and Foraminifera which are euryhaline component penetrating further up the estuary (Grindley, 1984).

There were more nauplii of cirripedes and copepods in all transects in SSK. The densities increased towards the offshore direction. The densities of the cirripede nauplii were greater than the copepod nauplii. The higher abundance of these nauplii in SSK was due to recent spawnings and probably a higher survival rate since the river is less polluted. There was no aquaculture activity in the river, as compared to SSB. The difference in zooplankton taxa between SSB and SSK may indicate changes in the structure of zooplankton community in SSB by aquaculture activity.

Based on the results from MDS and PCA, there appears to be a seasonal effect on the zooplankton community structure. From the MDS plot, samples taken from SSB in late March and April formed a separate cluster from those taken in December, January and early March. This shows that there were differences in the community structure of zooplankton during these months. The PCA plot exhibits a coherent group in December, January and early March comprising of brachyuran zoea larvae, heteropods, amphipods, fish larvae, mysids, chaetognaths, foraminiferans, juvenile copepods and adult copepods like *Centropages* spp., *Acartia* spp, *Pseudocalanus* spp., *Oithona* spp, *Sapphirina* spp. Another group which was found important in late March and April consisted of bivalve larvae, cirripedes, polychaete larvae, copepod and cirripede larvae and harpacticoid copepods like *Microsetella* spp. The presence of these two different groups indicates marked temporal variations in the community structure. This might be due to the different breeding seasons of the zooplankton or macrobenthos. For example reproduction by benthic populations adds to the meroplankton larvae at certain months.

4.5 Zooplankton in relation to phytoplankton and nutrients

The estuarine habitat is complex, with large spatial and temporal changes in nutrients, light availability and salinity. Both zooplankton and phytoplankton exhibit a patchy distribution (Steele, 1978; Bennett and Denman, 1985). The patchiness of phytoplankton and zooplankton is especially noteworthy in estuaries and coastal waters characterized by abrupt changes in environmental conditions (e.g. areas of wind-induced turbulence and river run-off).

In seasonal plankton cycles of estuaries, phytoplankton blooms are commonly superceded by a peak in zooplankton abundance, although strong oscillations in both phytoplankton and zooplankton abundance tend to be suppressed (Levinton, 1982). This phenomenon is observed in the 12-hour study in SSB. In Figure 3.19, the peak of the standing crop of phytoplankton productivity in terms of chlorophyll-a concentrations occurred in late afternoon from 1500hr-1700hr. This is likely due to the maximum of light intensity received in SSB between this time. Furthermore, the nutrients in SSB appeared to be not limiting as the source came from the aquaculture activity along that river (Fig. 3.20). The density of the zooplankton also peaked concurrently with the phytoplankton productivity during this period. In the fish farm, zooplankton were attracted to the abundance of phytoplankton probably as a food source. They were also likely to utilize other sources of food in the cage farm such as left-overs of trash fish and fish fecal pellets. After 1800hr, the concentration of measured chlorophyll-*a* decreased due to the reduction of phytoplankton production (light-limited). The zooplankton density did not significantly decrease (Fig. 3.21) as zooplankton continued to graze on remaining phytoplankton as well as the other food sources, while nocturnal species are likely to ascend toward the surface during night to feed. Such vertical night movements were also observed away from the cages, and in both stations the zooplankton populations were almost similar in numbers and about 3 times their numbers after dawn (see Fig. 3.21).

The diurnal pattern of chlorophyll *a* concentration observed in SSK was different from that in SSB. As the chlorophyll-a concentrations increased from 1600 hr to 1800 hr in SSB, it already decreased in SSK. Although there was no significant difference in chlorophyll-a concentration between SSB and SSK, the chlorophyll-a concentration in SSK was generally higher than in SSB (Fig. 3.19). It peaked earlier at 1400 hr and dropped beyond that. This might be due to the lower concentration of dissolved nutrients in SSK as nutrients appeared to be limited here (Fig. 3.20) where for instance, ammonia was not detected. Interestingly, the zooplankton density in SSK was relatively higher than that in SSB. It peaked around 1800 hr before it decreased. The biomass of the zooplankton seemed to be lower in SSB (Fig. 3.21) suggesting that the zooplankton in SSK comprised mainly of young zooplankton which might occur in swarms. However, at 2000 hr, the biomass in SSK was at its highest while the density was not which indicates the presence of larger animals found then; these zooplankton were *Acetes* spp, and mysids which were apparently nocturnal. Beyond 1800 hr, the zooplankton abundance in SSK dropped three-folds, significantly more than that observed in SSB (Fig. 3.21).

Fish feeds are the major sources of inorganic nutrient input in fish cultures and they introduce nutrients into the water column through rapid dissolution of the liquid components and by the release of nutrients from the feed leftovers. Qian et al. (2001) reported that the nutrient-release coefficients indicated the fast inorganic nutrient input into the water through the rapid dissolution of inorganic ions originating from the trash fishes' surface content, especially the orthophosphate and ammonia. The aquaculture operator in SSB used minced trash fish as feed for the cultured fish. Minced trash fish has a higher instant pollution level, and using it as fish feed may lead to serious inorganic pollution of the water (Qian et al., 2001). The minced trash fish shows the highest orthophosphate, nitrite, nitrate and ammonia release rates. In general, the excretion of nitrogenous wastes of fishes depends on the size of the fishes, feeding condition, how the fish is processed and the protein content of their diets (Handy and Poxton, 1993). Alongi et al. (in press) indicated that the input of trash fish by cage farms in Matang equates to a small proportion of carbon derived from phytoplankton and mangrove. However, trash fish inputs contribute to a moderate proportion of nitrogen and more phosphorus to the estuary than do phytoplankton and mangroves. This contribution is reflected in the often higher dissolved inorganic nutrient concentrations (especially ammonium and phosphate) in cages than in non-cage waters, and the lower concentrations in the control river (SSK).

The nutrient concentrations of phosphate, nitrate, nitrite and ammonia were low from 1500 to 1900 hr in SSB and SSK (Fig. 3.20). The low nutrient concentrations probably resulted from their utilization by phytoplankton for growth and reproduction, concomitant with the photosynthesis process. Nutrient concentrations were comparatively higher in the morning between 800hr to 1400hr. However, phytoplankton density or production was not high during this period, apparently being limited by light availability. When nutrients are abundant, phytoplankton primary production is directly proportional to light availability in the euphotic zone of the water column (Cole et al., 1986)

The density and biomass of zooplankton were apparently not influenced by either light changes (diurnal effect) or tide (tidal phase effect). However Fig. 3.21 shows that the lack of significant effect by light may be due to a lack of night catches as compared to day catches, where the latter also comprises very high zooplankton catches due to peak phytoplankton abundance. In fact if these catches were eliminated from the ANOVA analysis the night catches were significantly (3 times) higher in zooplankton abundance than the day catches (i.e. those collected from 0800 hr – 1200 hr), in both inside and outside the farm area ($p < 0.05$) (see Fig. 3.21). As for tidal effect, no clear reason can be offered given the scope of the study except that during ebb tide there was no significant impact by river water on the salinity of the estuary.