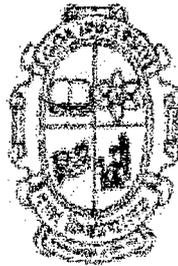


**A COMPARATIVE STUDY ON THE DEVELOPMENT
OF BOTTOM FAUNA IN DIFFERENT SHRIMP CULTURE SYSTEMS**

THESIS SUBMITTED TO

GOA UNIVERSITY



FOR THE DEGREE

OF

DOCTOR OF PHILOSOPHY

IN

MARINE SCIENCES

BY

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OCTOBER, 2007



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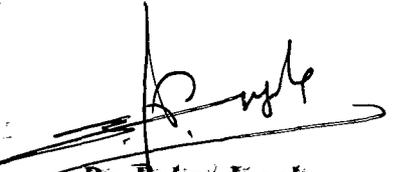
This is to certify that, thesis entitled "A Comparative Study On The Development of Bottom Fauna In Different Shrimp Culture Systems" submitted by Mr. Shantanu S. Kulkarni for the award of the degree of Doctorate of Philosophy in Marine Science is based on original studies carried by him under my supervision.

The thesis or any part thereof has not been previously submitted for any degree or diploma in any university or institute.

Place: Dona Paula, Goa

Date: 12/10/2007




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All the suggestions given
by the referees are incorporated
in the thesis


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DECLARATION

As required under the University ordinance 0.19.8 (vi), I state that the present thesis entitled "A Comparative Study On The Development Of Bottom Fauna In Different Shrimp Culture Systems " is my original contribution and that the same has not been submitted elsewhere for award of any degree to any other University on any previous occasion. To the best of my knowledge the present study is the first comprehensive work of its kind from the area mentioned.

The literature related to the problem investigated has been cited. Due acknowledgements have been made wherever facilities and suggestions have been availed of.



(Shantanu S Kulkarni)

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(Shantanu S. Kulkarni)

DEDICATED TO MY PARENTS ...

PREFACE

FAO defines aquaculture as “the farming of aquatic organisms, including fish, mollusks, crustaceans, and aquatic plants”. Farming here implies some form of intervention in the rearing process to enhance production, such as regular stocking, feeding and protection from predators. Aquaculture is the only alternative to mitigate the threats to the world’s fisheries by taking the pressure off from wild fish stocks while supporting livelihoods and food production. Aquaculture production has increased tremendously over the past few decades and today accounts for almost a half of global fish production by weight, while production from wild fisheries has largely slowed or stagnated.

With the advent of ‘Factory Farming’ concept, farmers have shifted from traditional to more intensive aqua-farming practices. The intensification of culture practices requires careful feed management and monitoring of water and soil quality, particularly in shrimp culture ponds. Oxygen is one of the most critical water quality parameter that limits the production. Further, cultured shrimp species also depend on benthic in-faunal invertebrates such as macro- meio-and micro benthos, and phytoplankton in the water, particularly during the early stages of their growth. They require oxygen for respiration along with cultured species. Dissolved oxygen is also required for oxidation of organic matter of uneaten feed and feces. Sustained reduction of dissolved oxygen in such cases can lead to hypoxic to anoxic conditions, which is harmful not only to cultured species but also to the benthos which form important food source for the cultured species.

In anoxic environments, denitrifying bacteria leads to formation of ammonia and hydrogen sulphide (H₂S) formation takes place through bacterial degradation of accumulated organic matter. These toxicants are known to cause mortality of cultured shrimps. To avoid anoxic environment and formation of such toxicants causing mortality and to achieve optimum production, various types of aeration devices are therefore used in aquaculture. Aeration devices improve the oxygen in the water and help ultimately in maintaining the natural equilibrium of nitrogen, carbon, sulfur and phosphorus cycles in the aquaculture systems. Thus it ultimately

keeps the culture environment healthy by artificially or mechanically mixing atmospheric DO in the water. With the advent of new aeration system developed by HOBAS Norway, its performance, in maintaining dissolved oxygen and subsequently the nutrient variations and possibilities of their effect on the benthos development in shrimp culture ponds was thus monitored. The present study in this view, is an account of the research carried out, describing the changes occurring in water quality and ecobiological parameters over two shrimp culture cycles, in aerated and non-aerated shrimp ponds, over two years in a commercial shrimp farm located in South Western coastal India.

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FEED CONSUMPTION AND GROWTH IN SHRIMP

- Figure P1 Fortnightly average growth in shrimp (gm) during EC-1
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Figure P3 Fortnightly average feed consumption (Kg) during EC-1
Figure P4 Fortnightly average feed consumption (Kg) during EC-2
Figure P5 Correlation Feed consumption vs gut biota in aerated pond EC-1
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CHAPTER 1

Introduction

1.1 Current status of world aquaculture

The proverb “dig the well before you feel thirsty” is used in context when one needs to take the measures before it’s too late. The very proverb seems to be a need of an hour when world’s natural ocean resources for fish are on the verge of decline and at the same time demand for aquaculture products is ever growing. Aquaculture practices can be the “well” to fulfill this, “thirst” of ever-increasing demand of fish food in the world market. Today, there is little doubt about the depleting natural resources. The world community has viewed aquaculture only during last decade and half, as a potential solution to the dilemma of depleted oceans. Furthermore, aquaculture practices have advanced through better management and technological advancement only recently. During this technological advancement, aeration is the most important technique used to maintain the dissolved oxygen concentration of the water. Use of different technological advancement has been studied by many authors (Boyd, 1998; Lee, 1999; Dey *et al.* 2000). Use of remote water quality logger units for monitoring water quality has been studied by Sreepada *et al.* (2005).

Mechanized feeding methods have been described by Goddard (1996), and are used mostly in developed countries with intensive practices. It includes demand feeders and automatic feeders, mainly used in Rainbow trout farming. Today we know such advanced farming technologies, as they are in use, mostly, for intensive cultures, however traditionally aquaculture practices are carried out since ancient times. The advent of aquaculture dates back to (2000 B.C). During this period, common freshwater carp *Cyprinus carpio* in China was cultured most commonly. This history of ancient aquaculture practices has been reviewed in detail by Rabanal (1988). The reason aquaculture practices most likely grew in ancient times lies in necessity. In ancient times, foraging and hunting were not sufficient to provide a stable source of food to local communities. Today, however the necessities of past

has changed in to urgencies, in the form of demands, to maximize production and economic profitability it offers. Subsequently, it has raised distressing concerns regarding environment due to intensification of aquaculture practices. According to Hampel (1993) from the mid of the 19th century the global population has almost doubled. Since 1960, it has grown from 3 to 5.9 billion until 1999, and it has been forecasted by Wurts (2000) that, it will attain the growth of 9.3 billion by 2050. Thus, this increase in population has raised and will continue to raise per capita fish consumption along with other aquatic products.

While catering the needs of this ever growing population, global fisheries production has also reached 130.2 million tons in 2001, and has doubled over the last forty years (FAO, 2002). According to Wurts (2000) food fish demand in all countries is expected to increase to 137 million MT by the year 2015. Asia is the major contributor to this production. Of the world total, the China produced 71.2% of the total volume in 2005 and 54.7% of the total value of aquaculture production. According to Crespi (2005), the other nine top producers besides China were India, Indonesia, Japan, Bangladesh, Thailand, Norway, Chile, Vietnam and the United States of America. In 2002, total world aquaculture production was reported to be 51.4 million tones by volume and US \$ 60 billion by value (Subasinghe, 2005; Nierentz, 2006).

Apart from the economic aspect, nutritionally fishes and crustaceans are the best source of animal protein we can get. They offer advantage over other animal meat that, they are relatively low in fat. Fish and crustaceans form an important part of the diet for large portion of the people living in developing countries. Many species of fish have heart-protective omega-3 poly unsaturated fatty acids, fat soluble vitamins A, D and E and water soluble vitamin B-complex, minerals such as Ca, P, Fe, I, Se (Subasinghe, 2005), and those species which don't have these nutritional properties still possess an edge over fast food delicacies. These nutritional qualities and our insatiable appetite for delicacies, has contributed for the need to either

exploit fishes from seas or grow them at farms. The demand for top predators such as Swordfish, Tuna has put severe pressure on the existing stocks (Tidwell *et al.* 2001). USDA statistics show that, the world commercial fishery landings have decreased from 101 million metric tons to 98 MMT in the last three years. Wild catches of shrimp from the world oceans are estimated to have reached a maximum sustainable yield of 1.6 to 2.2 MMT, and future demands for shrimp can be satisfied only through aquaculture production (Agriculture & Rural Development Department, World Bank Report, 2004).

In shrimp culture practices over 30 species are cultured all over the world and amongst all *Penaeus mondon* are the most important cultured species (Paez-Osuna *et al.* 2001). As many as 230 different species altogether, of algae, mollusks, fish and crustaceans are cultured all over the world indicating the success, this sector has achieved through continuous research and improvement in practices. These practices have shaped up in big industry, providing employment to millions of people (Agriculture & Rural Development Department, World Bank Report, 2004). FAO defines aquaculture as “the farming of aquatic organisms, including fish, mollusks, crustaceans, and aquatic plants.” Farming here implies the intervention in the rearing process, to enhance the production, such as regular stocking, feeding and protection from predators. In 2000, FAO carried out a study on global aquaculture production outlook to evaluate its potential, to meet projected demand for food fish in 2020 and beyond. According to this report, while output from capture fisheries grew at annual average rate of 1.2%, output from aquaculture (excluding aquatic plants) grew at a rate of 9.1% reaching 39.8 million tones in 2002. This rate is also higher than for other animal food producing systems such as terrestrial farmed meat. Much of this aquaculture expansion, as discussed above, has been due to China, whose reported output growth far exceeded the global average production being 69.6% of the total quantity and 51.2% of the total value of aquaculture production (FAO, 2006). According to latest reports, on the current seafood consumption, aquaculture production by 2000 was 32.2% globally, out of which India contributed 35.3%

(Kutty, 2006). Hence, aquaculture must continue and accelerate the current trend of supplying the increasing need for fish and seafood products.

1.2 Trends and status of shrimp aquaculture in India

With global demand driving the economies of the fishery and aquaculture dependent population, shrimp aquaculture has emerged as a sunrise sector in India. It started gaining roots only during the late eighties (Krishnan & Brithal, 2002). During the last decade and half, there has been a tremendous increase in annual shrimp production from 35,500 (1990-91) to 97,096 metric tons (1998-99), with an increase in farm area from 65,100 ha to 145,906 ha (Kumaran *et al.* 2003). India is the fourth largest farmed shrimp-producing nation in the world. The nine maritime states cover about 7000 km of India's coastline. About 70% of the shrimps are produced on India's east coast while remaining 30% are produced on the west coast (FAO, 2000). At present, in India, shrimp farming is carried out in about 154,600 ha of brackish water area out of potential 1,190,000 ha (Krishnan & Birthal, 2002).

According to Krishnan and Birthal, (2002) traditional aquaculture has been carried out along the coastal states of India for several centuries. About 50,000 ha of low-lying impoundment (1-10 ha) along bays and tidal rivers are under traditional cultivation and contributes about 25% of the total coastal aquaculture production. Yields from these traditional systems have been reported, to be very low (300-500 kg/ha/y) due to low input levels, limited management and tidally dependent stocking and water exchange. But with global demand luring the traditional farmers to go for more intensive practices, the culture operations quickly progressed from extensive mixed species to intensive mono-species culture of tiger shrimp. Initially it succeeded, but the fact that, most of the coastal states in India were new to commercial-scale shrimp farming at the start of the 1990, the general ignorance of good farming practices, and lack of sustainable extension services, led to host of the problems. The golden period of commercial-scale shrimp culture in India started in 1990 and the bust came in 1994-96 (Hein, 2002), where, an average yield of around

1000 Kg/ha during 1990-94 came crashing down by half to about 400 to 550 Kg/ ha in 1995-96 (Kumaran, *et al.* 2003). Within span of four to five years outbreak of bacterial, viral disease were spread all over. The trend was very much similar, where aquaculture practices got momentum before India; the countries include China, Tiwan, and Malaysia. Poor management practices for water, feed and nutrient use to enhance indigenous productivity during the culture practices in order to obtain better yield were blamed by scientific community and policy makers all over the world.

1.3 Feeding and environmental issues in shrimp culture practices.

New age aquaculture as such started in western world quite early. During 1960 and late 1970s intensive aquaculture practices of Rainbow trout, Atlantic Salmon farming in Europe, Channel Catfish farming in the United States and Kuruma shrimp in Japan stimulated considerable R&D activity in fields of shrimp nutrition (Goddard,1996). Before 1968, for *Penaeus monodon* culture, no feed was given other than naturally occurring pond biota. It was in 1979, when nutritional data on *Marsupenaeus japonicus* was prepared, based on the formulations of feed for channel catfish in USA. With worldwide culture of *P. monodon* for its fast growing and robust qualities nutrient requirements were studied only in 1990s (Shiau, 2001).

Penaeid shrimp are known to ingest a variety of food items i.e. detrital aggregates, animal and plant matter in their natural habitat, and have been described to be opportunistic omnivorous scavengers by many authors (Dall, 1968; Chong & Sasekumar, 1981). The detail shrimp feeding habits and feeding behavior is reviewed in Chapter 2; Section 2.3. In commercial hatcheries, nurseries and grow out ponds, penaeid shrimp larvae require a series of live microalgal species such as *Skeletonema*, *Chaetoceros*, *Tetraselmis* and brine shrimp naupli (*Artemia*) and artificial feeds. However, in grow out systems dry formulated feed is more important. With the advent of 'Factory Farming' practices especially, intensive aquaculture practices, use of the fertilizers and liming to enhance the indigenous productivity is carried out. Natural productivity is utilized by shrimp initially,

thereafter farmers use artificial feeds. These commercial formulated feeds are nutritionally complete. With multinational companies having great desire to capture the rising market of shrimp culture, endowed extensive research, and came up with the feeds, having higher nutrient manifestations to fulfill the nutritional and energy needs of the farmed animals were proliferated, in the last decade in India. Main reason to adapt these feeds by farmers is the growth achieved by shrimp in the stipulated time period.

In the market driven culture practices with cut throat competition for selling their products in time, and with exporters being munificent by offering competitive prices, due to great global demand to export farmed shrimps, intensification of these practices was inevitable. Another reason in this success story of the filleted feed lies in its maneuverability, as these feeds were easy to handle, apply and store. However, the unscientific ways and irrational feed applications were found to critically burden the aquatic systems, and contributed to high organic loads. Craig *et al.* (2002) has stated that, even with careful management of feed, about 40 to 60% of feed ends up in the waste. Out of 100 units of feed fed, typically about 10 units of feed are uneaten, and 10 units of solid and 30 units of liquid waste constitute 50%, and are produced by cultured species. Of the remaining 50% feed, only 25 % is used for the growth, and another 25 % is used for metabolism, and these percentages, vary with species, size, activity and other environmental conditions. Ultimately through experiences, observations and experimentations, it was noticed that only 40% of the feed is consumed and 60% is accumulated at the bottom and contributed to form organic matter. The water with high organic matter and nutrient load can pose great threat to the adjacent natural environments, and may alter the eco-biology of the natural habitat, such as creeks, and inhabitant mangrove foliage. Trott *et al.* (2004) has shown that, rate of supply of C and N from the shrimp farm exceeds the rates of in situ removal by biological uptake and transformation processes within the creek water and sediment. It is the natural biota right from phytoplankton to fish, which consumes these sources of C and N. This shows the complicated interdependence,

and role played by the organisms residing in these natural environments to utilize these nutrients. The poor water quality of effluent from artificial environments such as the aquaculture pond thus, may alter their functioning.

The harvesting practices discharges large volume of eutrophicated bottom water with highest pollutant loads, and thus pose threat to the residing biota in these environments. Due to higher fish, crustacean densities and due to their constant mobility brings about the sediment disturbances in natural environment. The accumulated food and feces from these animals, lead to high organic carbon accumulation, which necessitate higher oxygen demands for its degradation leading to anoxic to eutrophicated waters (Lin *et al.* 2001). In presence of insufficient oxygen in reducing environment, toxicants like ammonia, nitrite and sulphide concentrations were also found to be lethal to the aquatic organisms not only to cultured organisms, which depend on the natural water, but also to the one who resides in the natural environment such as creek. Beardmore (1997) has stressed the need to maintain the reasonable effective population sizes in natural populations and the relative scale of population sizes of farmed and wild species.

Further, Beardmore (1997) has cited reports on environmental stress, which indicates that, environmental stress affects the organisms residing in the natural habitat. He has also indicated through his study that, hydrocarbons and insecticides are mutagenic, and these substances, bring about mutation in the sensitive species. The effluents increase the biodiversity of pathogenic bacteria and pesticides accumulate in diatoms, phaeophytes and copepods. They become carriers for diseases and cause bacterial infections. Toxic diatomaceous and crustaceans as food organisms for cultured species reach via water intake to culture practices again. These, and other such kind of practices forming nutrient rich effluents, ultimately are known to critically overload systems and brought hyper productivity of the culture systems and led to sequence of problems like eutrophication discussed earlier. With lack of in depth knowledge in most of the cases by non learned farmers, as well as

limited area available for farm with almost all the ponds used for culture, the farmers were found lack the approach and vision for proper disposal of the aquaculture effluents (Latt, 2002). This ultimately led to deterioration in water quality of pond as well as source water. As mentioned above, the same receiving waters often serve as intake water for neighboring farms and thus, provide the means to spread water-borne disease agents from one farm to another (Paez-Osuna, 2001). This leads to spread of disease, frequent outbreaks of viral, bacterial, fungal, disease, which caused huge losses to the farmers in the 1990 to 1995 (Tacon and Forster, 2003). Therefore, with many flaws in management of aquaculture practices, feeding of shrimp culture practices were mainly blamed for the deterioration of the water quality, and subsequently environmental quality degradation. Such consequences raised serious concerns amongst farmers, scientific community and social activist for the better management of the aquaculture practices and rose to very concept of, "sustainable aquaculture". World Health Organization (WHO) defines the sustainability as, "the development that meets the needs of the present without compromising the ability to meet their own needs." This relates to continuity of economic, social, institutional and environmental aspects of human society as well as the non-human environment. The guidelines for sustainable aquaculture and aquaculture effluent management at farm level have been described by Boyd (2003).

In 1996, Honorable Supreme Court of India (SCI) in response to petition (No. 561/1994) as based on the reports of Algarswamy (1995) and on the basis of National Environmental Engineering Research Institute (NEERI, 1995) reports, described the development and the external impacts of shrimp aquaculture. The judgment indicated that, the social and economic costs related to the development of aquaculture sector were substantially higher than the revenues generated. For small farmers in the trade, the ratio of investments and revenues generated were inadequate to improve the economic status of the farmer. Thus, socio-economical and simultaneous concerns regarding environment and its social implications ultimately resulted in framing some stringent norms and direction by Supreme Court of India

(SCI). In its December 1996 judgment, SCI extended enforcement to early 1990 CRZ act, and restrictions were applied to extensive, semi-intensive and intensive shrimp farming to carry out aquaculture practices in sustainable manner. The ordains for the same has been reviewed by Hein (2002). Further, the Hon. SCI (1999), while assessing the environmental impacts caused by pond-based shrimp culture, recommended that, traditional aquaculture should be upgraded using appropriate technology that could result in minimum environmental changes.

The Government of India has also been promoting traditional aquaculture with necessary improvements. However, very little information is available on the environment and production capabilities of modified extensive aquaculture systems in India. As, site-specific reports are available, no serious efforts have been made to study the population structure, recruitment patterns, growth dynamics and sustainability of the systems, nor has been, considered the practicalities of involving rural coastal communities in enhanced production approaches. India as against other countries struggles against major constraints, prohibiting a sustainable development of the shrimp farming industry. It was therefore most important to develop code of best management practices and strategies for enhancement of the utilization of the coastal belt around India for sustainable aquaculture. And in this view present study is important.

1.4 Need for present research

After the consequences of aquaculture practices on the environment many research initiatives until now have focused on the general health management of the shrimp, which aimed to improve disease resistance capacity of shrimp. The introduction of modern tools such as polymerase chain reaction (PCR) for detection of WSSV and other protocols relating to chlorination, pond treatment, cleanliness and modern management techniques such as bio-remediation through various enzymes and probiotics proved to be useful and their use became widespread. However, not much effort have been made to understand the culture systems in an integrated way, in

view, to utilize the indigenous productivity to utilize resources in better manner and reduce the dependencies on external sources for feed and nutrient for cultured species.

It has been documented by Goddard (1996) that, there is very less distinction between semi intensive and intensive production methods in shrimp farming than in fish farming, as natural food organisms play less significant role in both the systems. Overall feeding dynamics are poorly understood especially in shrimp culture practices. It is known that, growth and health of shrimp are primarily dependent upon an adequate supply of nutrient, which generally includes protein, lipid, amino acid and water-soluble vitamins both in terms of quantity and quality (Goddard, 1996). Information concerning the nutritional requirements of many of the cultivated species is well established, however, most has been generated from laboratory based controlled feeding trials, and hence, such information is useful mainly for the formulation or production of nutritionally complete feeds for culture systems (Hasan, 2000). A Large portion of aquaculture in many developing countries is carried out in rural areas, where farmers practice extensive or modified extensive farming methods, as described previously in Section 1.2. In these cases such data on nutritionally complete formulated diets may be less applicable to farming condition where nutrition is from natural food supplies or supplemental artificial feeds.

Experiments involving the use of stable carbon isotopes have shown that 60-70% of shrimp growth in even semi-intensive systems resulted from the consumption of natural food organisms, while formulated feeds accounted for the remaining 30-40% (Anderson *et al.* 1987). This still holds true for the modified extensive prawn farming practiced in developing countries like India and East Asian countries. Hence, it can be speculated that, when natural fauna play such an important role in intensive and semi-intensive farming practices, which completely dependent on artificial feeds, certainly modified extensive farming practices are more dependent on natural feed. Further, this method involves lower stoking densities (8-10 PL.m⁻²)

and use of artificial feed with typical FCRs ranging from 1:1.2 to 1:5. With two-crop system per year, such practices over the couple of production cycles can deplete natural food resources. From previous reports (Ruello, 1973; Bell & Coull, 1978; Hedqvist-Johnson & Andre, 1991; Nuns & Parsons, 2000) it is evident that prawns do feed on the natural benthic fauna. Studies by Geoff *et al.* (1995) indicate that, faunal composition of prepared and controlled ponds were high and prawns also grew faster in such environments. Further, from direct observations (Ingole, 1992) it is speculated that, natural benthic fauna may play an important role in providing diverse food source compared to formulated feed. This natural food may have role in enhancing the overall health and improved immunity with the enhanced disease resistance capacity. In this view, it is important to study the role of natural food in shrimp nutrition.

Comprehensive information about benthic communities from different ecosystems is available, ranging from mangrove ecosystem (Alongi, 1990), Seagrass beds (Walters & Bell, 1986), sandy beaches (Ansari & Ingole, 1983), coralline beaches (Ingole *et al.* 1998), estuarine mudflats (Ansari *et al.* 2001) continental shelves and continental slopes (Gremare, 2002), deep sea meio and macrofauna (Ingole *et al.* 2001;2002). Importance of meiofauna in food chain of benthic environment has been studied by (Gerlach, 1971). Organic matter, bacteria and their extra-cellular products, protozoa and diatoms form a good source of food for benthic in-fauna (Bott, 1999). Influence of physico-chemical properties of soil and water alters distribution of benthic infauna and has been documented (Harkantra & Parulekar, 1989; Smith, 1996). More extensive review of macro-, meio- and microbenthos however has been made in Chapter 2, Section 2.2.

Dissolved oxygen content of the water is one of the most important parameters of water quality. It is a vital factor for all the aquatic organisms having an aerobic type of respiration. To improve the dissolved oxygen concentration and overall water quality, different types of artificial aerators are in use and are reviewed

by Boyd (2003). The artificial aerators help in increasing the oxygen content of all or part of the pond water to certain extent, which ensure the oxygen supply to the shrimp without limiting production at a given management level. Paddle wheel aerators in improving the shrimp growth, and production has been studied earlier by Wyban *et al.* (1989). The oxygen-transfer rate by using aerator in brackish water aquaculture pond was studied by Rogers *et al.* (1991). In all 15 important key water and soil quality parameters such as temperature, salinity, pH, E_h , Biochemical Oxygen Demand (BOD), Chemical Oxygen Demand (COD), Nutrients ($\text{NO}_3\text{-N}$, $\text{NO}_2\text{-N}$, $\text{PO}_4\text{-P}$ and $\text{NH}_3\text{-N}$), Particulate (POC) and sediment organic carbon (OC), H_2S , Chlorophyll-*a* and phaeophytin and their roles has been reviewed in Chapter 2, Section 2.1. However, there is paucity of information on the role, played by artificial aeration in development of bottom fauna from shrimp culture ponds as well as their role in the modified extensive shrimp culture systems, which is practiced by most shrimp farmers in India. With advent of new aeration technology (HOBAS) it was worthwhile to study the effect of artificial aeration on the development of benthic biota (macro- meio- and microbenthos) which can be the main source of natural diet for candidate shrimp species under culture system. Present study in the view, was carried out at the NIO Field Station at Kumta (Uttar Kannad District), Karnataka, India, under the Indo-Norwegian joint research Project entitled "Environmental Management Strategy for Prawn Aquaculture" during 2004-2006. Description of study area and sampling strategies in detail are described in Chapter 3, Section 3.1.

CHAPTER 2
LITERATURE REVIEW

SECTION 2.1

IMPORTANCE OF WATER QUALITY PARAMETERS AND ROLE OF AERATION IN SHRIMP CULTURE

CHAPTER 2

INTRODUCTION

Section 2.1 Importance of Water Quality Parameters and Role of Aeration in Shrimp Culture

Environment has been always remained a concern as it affects the residing flora and fauna on land as well as in water. In respective 'environments', both on land or in aquatic environment, flora and fauna are challenged every moment. In case of fishes and crustaceans, due to shrinking natural fish resources, we started to grow them artificially, and thus, we confine the 'environment', which ultimately have an effect on residing flora and fauna. In case of aquaculture practices, they are fishes, shrimp, mollusk or algae. Identifying and understanding the associations between these species and their physico-chemical environment as well as biotic factors is important (Yimin, 1999). For pond environment, unlike land there are two significant components, water and soil which interact continuously and thus make it more dynamic than the land based systems. The interactions between water and soil influence the culture environment (Fungsmith & Briggs, 1998). Water quality is critical for survival, health and growth of shrimps for all types of farming, especially in semi-intensive and intensive shrimp culture systems, and for the production of quality shrimp seed in the hatchery. To maintain good water quality, management activities include feeding, use of aerators, water exchange and liming. All these practices are very important, and help in regulating the physical and chemical properties of water and soil within certain optimum levels (Table ST13). In shrimp culture practices especially, there are numerous concerns regarding water quality and discharge of wastewater from shrimp culture practices, the most obvious concern is about the nutrient and their toxic byproducts. The need for reducing the nutrient loads and its mitigation through proper management has been discussed by different authors (Lin *et al.* 2001; Paez-Osuna, 2001; Hein, 2002; Tacon, 2003). To overcome the concerns and to use our resources at its best, and to use them in sustainable manner, we must have the knowledge of the environment, that is water and soil in which the cultured species like shrimp inhabit. Hence, for

profitable and environmentally as well as ecologically sustainable coastal aquaculture, it is important to understand the ecological role played by each measured environmental variables. In this regard following are the critical physico-chemical variables which influence the growth and health of the shrimp. Further, management of these critical water quality parameters within optimum levels finally determines the shrimp production.

Temperature

Shrimp and fishes are considered as the *obligate poikilotherms*, which means that they possess little or no ability to internally regulate their body temperatures independently of their environment (Dall, 1986; Goddard, 1996). Water temperature plays an important role in regulating the activities of cultured animals. In natural and pond environment water temperature is influenced by various physical factors such as, air temperature, water depth, groundwater inflow, stream flows, mixing due to water currents and amount of sunlight. In natural water bodies it limits migration, spawning and egg maturation (Addy & Gee, 1997). Effect of temperature has been found (Ocampo, *et al.* 2000) to be more detrimental to general behavior and growth of the shrimp.

In brackish water and shallow ponds, where regular exchange between the tidal water and the pond water is not maintained during the hot dry months, the temperature of pond water may shoot up beyond the tolerance limit causing mortality of reared shrimps. The high rate of evaporation will also occur increasing the water salinity beyond the tolerance level. Similarly, during the winter season, lower water temperatures may have a chilling effect reducing metabolic and growth rates of cultured shrimps, and higher temperatures increase the metabolic rate (Chen & Lai, 1993). Thermal classification for ponds and lakes has been given by Hutchinson and Loffer (1956) as, holomictic, monomictic, dimictic and polymeric stratification, which has been cited by Addy and Gee (1997). Thermal stratification is known to cause extreme DO conditions within a water body. In this regard, the effect of temperature in consumption of dissolved oxygen (DO) has been studied by Tian *et al.* (2004) in Chinese shrimp. In some ponds, the surface water temperature reaches beyond 35 °C in such cases; partial shading may be of help, so that shrimps can take shelter under such extreme

conditions. Other management techniques adopted during winter is to reduce the fertilization and feeding, as growth rate of organisms will be low during the period. The temperature can be adjusted to optimum levels in controlled systems such as hatcheries but it is extremely difficult to do so at affordable cost in large farms. Monitoring water temperature at least twice a daily is most easy and an important practice in shrimp aquaculture. Temperature variations in coastal areas though are small, even small temperature changes in such areas have direct effect on the metabolism. Also, with increase in temperature, the oxygen requirements of shrimp also increases.

Salinity

Salinity is one of the significant parameter which affects the shrimp and as a single factor plays an important role in shrimp farming. In natural waters it determines the distribution of the organisms. Salinity is responsible for several important metabolic processes such as osmoregulation, growth and reproduction *etc.* Shrimps have optimal range of salinity for better growth and survival depending on the species. If the increase in salinity is beyond the optimal limit, the shrimps refrain from taking normal food and hence are emaciated and become susceptible to disease. Most shrimp species are euryhaline and tolerate wide range of salinities (Decampo *et al.* 2003). Many shrimp species grow best at salinities of 15 - 30 ppt has been described by different authors (Rosas *et al.* 1999; Chiba *et al.* 2004; Zang *et al.* 2006). In pond conditions, the tiger shrimp *Penaeus monodon* can tolerate and acclimatize to wide range of salinity from as low as 5 ppt to a high of 40 ppt, but white shrimp *Penaeus indicus* and banana shrimp *P. merguensis* generally prefer brackish water with Salinity 5 to 25 ppt, and salinities above 45 to 60 ppt can be lethal, as it can stress the animal and make them susceptible to diseases. The role of acute salinity changes in disease outbreaks due to viruses like WSSV, has been described by Liu *et al.* (2005) and IHNN by Bray *et al.* (1994). These authors have stated that, large variations in the salinity result in higher mortality. Salinity changes may result in the physiological and biochemical adaptive changes in shrimp with them and expand corresponding amounts of energy in osmotic regulation. Therefore disease

resistance of shrimp in reduced salinities is lower than those in the original salinities.

In the case of brackish water shrimp farming, the maintenance of proper salinity without fluctuation is of great importance. Salinity should be measured at the source before pumping. If there is inadequate flow of freshwater in the estuary, salinity will start rising during spring tide and will decrease during ebb tide. The time for pumping should be decided after measuring the salinity frequently.

Hydrogen ion concentration (pH)

The pH of water is a measure of the concentration of hydrogen ions in the water. The pH gives an idea whether the water is acidic ($\text{pH} < 7$) or alkaline ($\text{pH} > 7$). The impact of variations in the pH has been studied in the fishes by (Randall, 1991). Both extreme acidic (< 6) and alkaline (> 8) pH are known to affect the physiology of shrimp. The most optimum range of pH should be 6.5 to 9.0. Pond bottom soil pH is particularly critical for shrimp since they spend most of their time on the bottom; they burrow in to soil and even ingest it. The influence of sediment characteristics on shrimp physiology with pH as principal effect has been studied by Lemonnier *et al.* (2004).

The growth of shrimp gets retarded if pH falls below 5.0. The causes of low pH are mostly attributed to the iron-sulphate nature of the soil (Boyd, 1995). On exposing pond soils to air due to tilling practices or due to fissures in the compacted hard bottom, or during construction of pond pyrites in the deeper soil layers are exposed, such soils are commonly known as acid sulfate soils and impart acidic pH to the soil on drying. Further, accumulated FeS_2 reacts with O_2 which produces Fe^+ and SO_4 thus, imparting acidity to the soil (Senarath, 2001). To neutralize this acidity application of lime is widely in use, in aquaculture practices. Common liming materials used for treating acidic soils has been described by Boyd (1995), which include agriculture limestone (CaCO_3), burnt lime (CaO) and hydrated lime [$\text{Ca}(\text{OH})_2$]. For the application of lime during the culture period, doses greater than 50 or 100 kg ha^{-1} causes a high pH. However the recommended use of lime for disinfecting the bottoms of pond or pond waters is 1000-2000 kg ha^{-1} during initial pond preparation (Boyd and Tucker, 1998).

On the other hand very high $\text{pH} > 9$ are also harmful, and with increasing level of

H^+ results in gradual breakdown of the gill and epidermis, which results in increasing loss of body salts and difficulties in uptake of oxygen. With higher pH sub lethal effects can be seen which includes gill damage and damage to the lens and cornea of the eye in fish. pH also controls metabolic functions in terms of excretion of ammonia and governs the toxicity of un-ionized ammonia (Samocha *et al.* 2004). Agricultural gypsum ($CaSO_4$) can be applied to correct alkaline soil pH. Water with low pH (acidic condition) can be corrected by the controlled application of lime. Excessive lime application should be avoided as it may reduce the availability of native and added phosphate (fertilizer) and some trace elements like iron and manganese needed for aquatic productivity. Brackish water bodies are well buffered against pH change and pH will seldom fall below 6.5 or rise above 9. Therefore, adverse effects of pH on shrimps are uncommon. However, there are instances where acidic soils are problematic in shrimp farming. This is corrected by the application of lime as described above.

A decrease in the pH increases the sensitivity to low concentrations of oxygen in *P. monodon*. The low pH of 5.9 to 6 causes decreased growth in *P. monodon* has been reported by, Allan and Maguire (1992). Further, Wickins (1984) showed that, a decreasing pH from 7.9 to 6.7 induces carapace weight loss. Martinez *et al.* (1998) reported that, the tolerance to hypoxic conditions of *Penaeus setiferus* post larvae diminished in pH 6, as compared with that obtained in pH 8. The high pH level due to application of lime increases the precipitation of heavy metal. Heavy metals in aquaculture ponds may be naturally present or they intrude via source water through anthropogenic sources. The heavy metals are known to be toxic and are dependent on pH for their toxicity on the cultured species.

Redox potential (E_h)

Measurement of E_h is useful as it reveals the source of oxygen used in the mineralization of the sediment organic matter. In ponds, when the input of organic matter exceeds the supply of oxygen, needed for decomposition of organic matter, anaerobic condition can develop. These reducing conditions can be measured as the redox potential (E_h). Redox potential indicates, whether the water or soil is in reduced condition (E_h with a '-' value) or oxidized condition (with '+' values) by Boyd (1995). Reduced or an aerobic sediment may occur at

the pond bottom of heavily stocked ponds with heavy organic load and poor water circulation. Under anaerobic condition of pond bottom, reduced substances such as H_2S , NH_3 , CH_4 etc. is formed which are toxic to benthic organisms.

In shrimp ponds, development of highly reducing conditions at the surface of the pond mud is highly undesirable. Redox potential, ($E_h = -200mV$) is the critical measure at which sulphides starts to form (Lovley & Goodwin, 1988). Water circulation caused by water exchange, wind or aeration tends to move water across the mud surface and prevent the development of highly reduced conditions. Further, bottoms should be smoothed and sloped to facilitate draining of organic waste and toxic substances (Boyd, 1995). Draining at the centre of pond, as is being practiced by some farmers, is an ideal remedy for the prevention of formation of highly reducing condition during the last phase of culture period.

Dissolved oxygen (DO)

Dissolved Oxygen (DO) is the most critical water quality variable in aquaculture. The availability of dissolved oxygen is a most critical limiting factor. It is vital as shrimp obtain oxygen directly from water. It is required for production of energy, metabolism, growth etc. Oxygen requirement for shrimp vary according to species, age, stage of maturity, and size.

Oxygen concentration in rearing water depends on the physical factors such as temperature, salinity, and altitude, chemical processes as well as bacterial and phytoplankton, algal respiration at the bottom as well as in the water column. Thus oxygen in shrimp culture is considered to be dynamic and ever-changing critical entity. If DO concentration is low, the culture organisms become inactive and they are susceptible to diseases. Furthermore, in ponds where DO concentrations are very low, shrimp may die from lack of oxygen. The lower activity and random swing moments has noted by many authors. To ensure lower feed conversion efficiency, high survival and adequate profits, an aquaculturist must maintain greater level of DO in pond waters.

The ability to detect and actively avoid hypoxic water has been reported in many aquatic species, including the European brown shrimp, *Crangon crangon*

(Hagerman & Uglow, 1979), Kumura shrimp, *Penaeus japonicus* (Egusa and Yamaoto, 1961), Mantis shrimp, *S. empusa* (Pihl & Rosenberg, 1994), freshwater prawn, *Macrobrachium nipponense* (Kang *et al.*, 1995), and several other crustacean and fish species (Jones, 1952; Loesch, 1960; Pihl and Rosenberg, 1994).

Stress caused by unsuitable oxygen concentrations and behavioural responses of white shrimp *P. aztecus* and brown shrimp *P. setiferus* to hypoxic water 2.0 ppm dissolved oxygen were studied by Renaud *et al.* (1986). Study showed, an initial increase in their general level of activity, retreat from hypoxia by walking or swimming, rapid eye-stalk movements, flexing of their antennal scales, and abdominal flexures followed often by "exhaustion". The major behavioural difference between brown shrimp and white shrimp was the absence of abdominal flexures by brown shrimp exposed to hypoxic water as low as 1.5 ppm. White shrimp exhibited abdominal flexing in all oxygen concentrations tested but more frequently in the lower concentrations.

According to Taylor and Spicer (1987) the sensitivity of the decapods at low concentrations of oxygen is due to the limited capacity of their anaerobic metabolism. They have showed that, phytoplankton abundance is controlled by nutrient supply, and that, the DO concentrations are regulated to larger extent by phytoplankton abundance. Feed applied results in pollution of pond waters by organic and inorganic metabolic wastes from shrimps. Uneaten feed also decomposes, releasing nutrients into the water. Phytoplankton abundance increases due to increase in the nutrient levels and consequently as a function of increased phytoplankton feeding rate of shrimp increases. As phytoplankton abundance increases, the diurnal cycle in DO at dawn show lower concentration, while, DO concentrations in the afternoon are higher. In addition, DO declines more rapidly with depth as phytoplankton abundance increases in response to higher feeding rates. If phytoplankton blooms are extremely dense, DO may be low on pond bottoms, even in ponds where the water depth does not exceed 1 m. The probability of DO depletion during cloudy weather and the likelihood of phytoplankton die-offs are greater in ponds with high feeding rates and abundant phytoplankton.

Higher feeding rates may be used in ponds if aeration is applied. Feeding is a proven technique for increasing shrimp production. However excess feeding results in deterioration of water quality, leading to lowering of dissolved oxygen during the night. If feeds are applied in excessive quantity, depletion of DO can result in mortality of shrimp due to higher oxygen demand by bacteria to degrade it. As feeding rate increases, the DO concentration during the night declines further. Chronically lower DO concentrations can have an adverse effect on the appetite and metabolism of shrimp, and conversely, shrimp lose feeding efficiently, where dissolved oxygen concentrations fall below 2 or 3 mg.l⁻¹ during night (Goddard,1996).The existence of a desirable level of DO of 3-10 mg.l⁻¹ in the pond indicates that, the pond system is functioning efficiently. DO concentrations below the minimum range is reported for maximum growth rate is 6 mg.l⁻¹ but at the level of 4 mg.l⁻¹, the feeding rate was found to be reduced (Goddard, 1996). Prolonged exposure to the stress due to, low concentration of DO lowers their resistance to diseases also. In the ponds, the principal cause of oxygen depletion is attributed to sudden change in weather condition and development of plankton bloom. DO concentration of less than 1.5 ppm can be lethal to shrimps depending on the exposure time and other conditions.

The first indication of possible oxygen stress may be reflected in the behavior of shrimps *e.g.*, crowding near the inflow, gasping for oxygen at the water surface by jumping out of water etc (Wu, *et al.* 2002). Supplemental aeration during the night increases the diffusion of oxygen from atmosphere to the water and increase the DO level, conversely, aeration during the afternoon helps to incorporate excess of oxygen from the surface water layer by mixing oxygen rich surface water with sub-surface water. When water is super saturated with DO, the oxygen diffuses from water to atmosphere (Boyd, 1998). Water exchange is best solution to prevent low DO problems in the pond water where aeration is not practical.

To maintain DO at optimum level on continuous cloudy days and during phytoplankton die-off, providing additional aerators for extra hours may be necessary. Water exchange and flushing out of decaying phytoplankton also helps to maintain optimum DO levels.

Role of aeration in aquaculture systems

Aeration increases the DO level of the water and prevents O₂ depletion during night. Different type of aerators such as paddle wheel aeration, vertical diffused aeration systems, pump sprayers, propeller aspirator pumps etc has been described by Boyd (1998), which work on different working principles. However, all of them are aimed towards improving the dissolved oxygen concentrations and only differ in the rates of transfer of oxygen at unit time. It accelerates the diffusion effect of not only O₂ but also enables the capture or release of CO₂, which is important in the process of photosynthesis. Aeration also, facilitates the volatilization of undesirable gases such as, NH₃, CH₄ and H₂S. It helps in reducing the daily fluctuations of pH by maintaining the adequate DO levels in the pond. According to Peterson *et al.* (2001) arrangement of aerators is important, as proper arrangement of aerators helps in less sediment erosion, off the bottom, and from the sides of the pond. According to Peterson *et al.* (2001), aeration arranged to circulate water creates a secondary flow that diverges at the surface and converges at the bottom, which mixes water efficiently. Thus, it diminishes the possible stratification of important physico-chemical parameters of water like temperature salinity, pH and most importantly DO. The aeration helps in evaporation rate where low salinity is observed. According to Fast *et al.* (1999) and Boyd (1998), during aeration the bubbles are created which increases the water-gas exchange area and thus help in diffusion of dissolved oxygen from supersaturated air above water in to the water. Delgado *et al.* (2003) has showed that, paddle wheel aerators are useful in high density low water exchange ponds as it helps in the oxidation of accumulated organic matter and sludge at the bottom of the pond, as water exchanges are limited, which do not allow the sludge to be released out frequently through water exchange practices. Cordava *et al.* (1997) showed that, a water exchange rate of 5% or lower affect the water quality as well as growth of shrimp adversely, and in this view, where water exchange rates are low, aerators help in improving the water quality.

Moulick *et al.* (2002) has showed that, in the biological treatment of wastewater, aeration is an important process employed to raise the dissolved oxygen level to allow aerobic bacteria to reduce biochemical oxygen demand of the effluent and thus to improve the water quality. According to Moulick *et al.* (2002), the oxygen

supplied must be at a rate sufficient to at least balance the rate of removal of the active biomass and hence aerators are the only devices used to supply the oxygen to meet such demands.

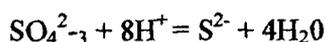
Organic Carbon (OC)

The natural organic material occurring in the terrestrial environment as well as in aquatic systems is defined as, "all weathering material from plants and animals and their degradation products" by Kordel *et al.* (1997). The natural organic material in the aquatic system mainly originates from the dead and decaying algae and phytoplankton, microorganisms, benthic infauna, epifauna and artificial feed in case of shrimp ponds, in and above the sediment. The organic carbon occurs in different forms, as dissolved organic, particulate and accumulated in sediment. The classification of the organic carbon on the basis of sizes has been given by Sharp *et al.* (1973). The high organic matter formation is therefore an indication of high biological productivity, which is considered as an ecological indicator for assessing the organic pollution of the environments (Hyland, 2005). Organic matter in the sediment and in the particulate form in water forms a source of food for the macrobenthic invertebrates and fishes. Organic matter as a source of food for benthic fauna has been studied by Snelgrove *et al.* (1994). Middelburg *et al.* (2006), has showed that, the rivers are the major source of the particulate organic carbon. The rivers transports about 4.0 Gt ($Gt=10^5$ g.) of organic carbon from the continents to the ocean, of which 0.15 to 0.17 Gt is in the particulate form. Further, according to Middelburg *et al.* (2006), OC values remain constant in turbid systems such as estuaries, due to extensive mixing and dilution. However, aquaculture systems behaves differently, and the organic load formed most of the times especially in the intensive and semi-intensive practices are attributed to the high amount of feed, which on disintegration contribute to either forms of organic carbon as described above. Role of microbial communities and phytoplankton in formation of live particulate organic carbon has been shown by Ribs *et al.* (1999).

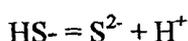
Hydrogen sulphide (H₂S)

Hydrogen sulphide is produced by bacteria in anoxic waters. There are two forms of hydrogen sulphide existing in water HS⁻ (ionized sulphide ion) and H₂S (unionized hydrogen sulphide gas). Unionized hydrogen sulphide gas is toxic and affects gills by causing severe damage (Gopakumar & Kutayama, 1996).

Under anaerobic condition heterotrophic bacteria can use sulphate and other oxidized sulphur compounds as terminal electron acceptors in metabolism and excrete sulphide as illustrated below:



Sulphide is an ionization product of hydrogen sulphide and participates in the following equilibriums:



The pH regulates the distribution of total sulphide among its forms (H₂S, HS⁻ and S²⁻). Un-ionized hydrogen sulphide is toxic to aquatic organisms; the ionic forms, however, have no appreciable toxicity. Concentration of 0.01 to 0.05 mg.l⁻¹ of H₂S may be lethal to aquatic organisms (Gopakumar & kuttayama, 1996). Any detectable concentration of hydrogen sulphide is considered undesirable in culture systems.

Removal of the organic waste accumulated at the pond bottom and application of health stone powder (500 kg/ha) and BNIO (10-20 kg/ha) to absorb H₂S are being practiced. These 'bioaugmentation' materials are said to remove H₂S, NH₃ and methane and speed up process of organic decomposition. Admissible levels of relevant water quality parameters for shrimp culture as recommended by Chanratchakool *et al.* (1994); Paulraj *et al.* (1998) is presented in the Table ST13.

Nutrients – Nitrate, Nitrite, Ammonia and Phosphate

A nutrient element is defined as, "one that is functionally involved in the processes of living organisms", (Parsons, 1975). Thus anything besides water and carbon dioxide that is required by plants in the synthesis of organic matter or skeletal materials is regarded as a nutrient. Nitrogen and phosphorus are considered to be the main limiting nutrients.

The major forms of nitrogen in the marine environment are interrelated by the biogeochemical reactions known as the 'nitrogen cycle'. Primary productivity is main limiting influential factor in nitrogen cycle where temperature, solar irradiance influence the rate of primary productivity which in turn by uptake or release affect the nitrogen cycle (Boyd, 1995; Sundback & Snoeijs, 1991). Nitrogenous compounds like ammonia (NH_3) and nitrite (NO_2) contribute as major factors causing toxicity to the cultured organisms. The $\text{NO}_3\text{-N}$ and $\text{PO}_4\text{-P}$ however are taken up as main nutrient source by phytoplanktons. High levels of nutrients (N and P) can cause eutrophication in fresh waters and coastal zones. Especially in shrimp culture ponds high nutrient accumulation leading to eutrophication is lethal to the cultured species. Hence, applying manure in shrimp culture ponds for optimal primary productivity needs careful monitoring of nutrient concentrations, and understanding local environmental conditions leading to excessive bloom formation. The pond bottom soil contributes to the accumulation of nutrients like phosphorus and nitrogen in the sediment of the pond (Fung-Smith, 1998). These nutrients are taken up by microphytobenthos and algae at the bottom providing important source of natural food for the cultured species.

Nitrate-N

Nitrate in the culture system have various sources like rainfall, fertilizer used to enhance primary productivity and through the process of nitrification where ammonia is converted to nitrates by oxidative processes carried out by chemoautotrophic bacteria of genus *Nitrosomonas* and *Nitrobacter* (Boyd, 1995). It is taken up by the phytoplankton readily. Uriarte *et al.* (2006) has showed in his studies that, microalgal species are widely used in bivalve hatcheries, increasing the nitrogen content of their growth medium from 6.3 to 16.5 mg $\text{NO}_3\text{-N l}^{-1}$ and from 16.5 to 49.4 mg $\text{NO}_3\text{-N l}^{-1}$ increased the protein content of the microalgae after a period of acclimation, and this further resulted in superior growth and survival of bivalve larvae and post-larvae. Collos *et al.* (1982) has showed the use of nitrate reserves by *Skeletonema costatum* and *Phaeodactylum trkomutum* and found that, internal nitrate accumulation of these species could reach to 15% of cell nitrogen compared with 1 to 2% under steady-state conditions. This capacity

for nitrate storage allows these species to maintain high rates of nitrate reduction at low external nitrate levels and grew better. However, Cheng and Chen (2002) has sited that nitrate can be toxic. When fish and crustaceans are subjected to nitrite or nitrate, entry of nitrite and nitrate is considered mainly via diffusion and via active transport to fish and crustaceans.

Nitrite-N

Nitrite (NO_2) is an intermediate product in the bacterial oxidation of ammonia to nitrate (NO_3^{2-}) a process called nitrification is carried out by chemoautotrophic bacteria of genus *Nitrosomonas* and *Nitrobacter*. When oxygen is depleted denitrification occurs, with nitrate as electron acceptor. The toxicity of nitrite is known to be dependent on water pH, and the presence of chloride and calcium ions. Nitrite toxicity increases with increasing pH. It decreases with increasing calcium and chloride concentrations. Further the toxicity of nitrite is expressed through the competitive binding of nitrite to hemoglobin forming methenoglobin, which does not have the capacity to carry oxygen (Hargraves, 1998). Thus, nitrite is toxic to shrimp and exposure to high concentrations may cause retarded growth and mortalities.

Ammonia-N

The main source of ammonia in shrimp aquatic system occurs in the form of shrimp excretion, decomposition of organic matter, uneaten feed pellets, disintegrated supplementary feed particles; these constituents contribute to form organic matter. Oxidation of this organic matter by heterotrophic bacteria produces ammonia NH_3 or ammonium NH_4^+ (Moriarty, 1997). In the aquatic system, a dynamic equilibrium between the toxic ammonia form NH_3 and non-toxic ammonium ion NH_4 gets established and sum of these is known as total ammonia nitrogen (TAN). The equilibrium of these ions is dependent on the temperature and pH (Montoya *et al.* 2002). Very low levels of ammonia are known to be toxic to most of the cultured animals. At alkaline pH, about 95% of the total ammonia nitrogen (TAN) is in the form of ammonium ion. According to (Shan & Obbard, 2003) removal of TAN can be facilitated by providing and maintaining the optimal environment for proliferation of nitrifying bacteria which

is achieved by maintaining water pH between 7 to 9, and temperature of 24°C to 30°C. Ammonia mainly originates in the aquatic environments through the process of denitrification, where denitrifying bacteria reduce the nitrate in suboxic to anoxic condition to ammonia. The accumulation of ammonia can cause mortality of organisms reared in closed culture systems (Chen *et al.* 1990a). In an aqueous ammonia solution, total ammonia comprises un-ionized ammonia (NH₃) and ionized ammonia (NH₄⁺) in equilibrium (Bower & Bidwell, 1978). The former is usually most toxic, as it has high lipid solubility and is able to diffuse readily across cell membranes; NH₄⁺ is also toxic, especially at low pH levels (Allan *et al.* 1990).

According to Hargraves (1998), in the fishes ammonia is excreted as the end product of protein catabolism, and may be toxic if allowed to accumulate. Ammonia toxicity is visible through behavioral changes such as hyperactivity, convulsions, and loss of equilibrium, lethargy and coma. However, ammonia toxicity in aquaculture ponds is most likely expressed as the sub lethal reduction of fish growth or suppression of immunocompetence, rather than as acute toxicity leading to mortality. Ammonia exists as a component of a pH and temperature dependent equilibrium in natural waters. Across the range of pH most commonly encountered in natural waters 6.5 to 9.0, the equilibrium favors the aqueous, ionized form NH₄⁺, or 'ammonium'. Elevated pH >9.3 favor the gaseous, unionized form NH₃, or 'ammonia'.

Phosphate -P

Sources of sedimentary phosphate are microbial breakdown of buried organic matter and redox-driven phosphate desorption from iron and manganese oxyhydroxides. According to Miller *et al.* (2001) soil phosphorus pools vary due to changes in the stability of organic matter and secondary Fe and Al oxyhydroxides and non-crystalline aluminosilicate minerals. In their study on the basis of soil morphology and E_h recording they found that as annual rainfall increased the intensity and duration of reducing conditions in the soil also increased and further, with increased reduction, total phosphorus in the soil declined by nearly two thirds.

According to Follomi (1996) phosphate is an essential nutrient and, phosphorus (in the form of phosphate) represents an important driving and regulating force behind biological productivity. Further he asserted that rates of productivity in turn, determine the rate with which atmospheric CO₂, is converted into organic matter, which is subsequently prone to storage in soils and sediments. Hence, this links the phosphorus cycle closely to the cycles of carbon and other biophile elements, such as oxygen, sulfur, nitrogen and iron, and provides phosphorus with extensive regulating capacities in processes such as climate, environmental and ecological change. The anaerobic conditions cause a large phosphorus release and an increase in exchangeable phosphorus especially below pH 8. The exchangeable phosphorus under anaerobic conditions was found to be ranged from 27 to 49% of the total phosphorus in the sediments of lake by Furumai and Ohgaki (1989).

Thus, these nutrients are taken up by the phytoplankton in the aquatic environment and help in the growth of the phytoplankton in the presence of light. However, their increased concentrations may misbalance the nutrient uptake of primary producers and high nutrient uptake may lead to eutrophication and thus want of oxygen turns out to be lethal to the aquatic organisms. Further their elevated concentrations also affect the metabolic functioning of the aquatic organisms and stress them reducing their growth. Hence monitoring nutrient levels and their maintenance through proper measures is extremely important.

SECTION 2.2

BENTHIC ECOSYSTEM AND ROLE OF MACROFAUNA, MEIOFAUNA AND MICROBENTHOS

CHAPTER 2

Section 2.2 Benthic Ecosystem and Role of Macrofauna, Meiofauna and Microbenthos.

2.2.1 Ecosystem and Benthos

Odum defined ecosystem in (1959) as “the living organisms and their nonliving (abiotic) environment, which inseparably interrelate and interact upon each other. Any area of, nature that includes living organisms and nonliving substances interacting to produce an exchange of materials between the living and non living parts is an ecological system or ecosystem”. He further defined ecosystem as the basic functional unit in ecology, as it includes both organisms (biotic communities) and abiotic environment, where, each influences the properties of the other and both are necessary for maintenance of life. The ecologies on the planet have main two types, first terrestrial ecology and second aquatic ecology. The aquatic ecology especially is unique as well as diverse in the view that, the flora and fauna from this ecosystem requires adaptations for aquatic, benthic as well as, up to some extent terrestrial mode of life, for some species of flora as well as fauna. Therefore, on the basis of their peculiar abiotic and biotic environmental interactions, aquatic ecosystem has been further divided to form many unique ‘subsidiary ecosystems’ such as marine ecosystem, mangrove ecosystem, coralline ecosystem, pelagic ecosystem and benthic ecosystem. These ecosystems are unique because of their flora and fauna. Flora and fauna is a biotic system, and the abiotic system is a system to which they have adapted, or, are the integral parts of the system, known as habitat, making it archetypal as they are. Aquaculture practices with its restrained water bodies thus can be considered as a unique ecosystem, which involves both aquatic environment and benthic ecosystem and their interactions, along with artificially introduced aquatic species, in high density.

In an ecosystem an organism is called as benthic when the organism resides primarily in or on the substrate and doesn't swim or drift for extended periods. Benthos burrows, crawls, walks, and are motile, sessile or permanently affixed to the

substrate or to each other (Gire, 1993). Thus, the term "benthos" refers to anything associated with or occurring on the bottom of a body of water. The animals and plants that live on or in the bottom are known as the benthos.

Benthic habitats are also defined as, the bottom environments with distinct physical, geochemical, and biological characteristics. Marine benthic habitats are divided into zones based upon how deep they are in the water. From deepest to shallowest these include, hadal zone (over 6,000 meters deep), abyssal zone (2,000 to 6,000 meters), bathyal zone (200 to 2,000 meters), and near-shore and estuarine zones (less than 200 meters). Estuarine and near-shore benthic habitats can be highly diverse, including shallow submerged mudflats, rippled sand-flats, rocky hard-bottom habitats, sea-grass beds, kelp forests, shellfish beds, and coral reefs (Gire, 1993).

Categorizing benthos is necessary since the organisms have different modes of living and rates of reproduction and associated energetics. Benthos is divided on the basis of their sizes. Firstly they are divided in 'epifauna' and 'infauna'. Epifaunal organisms are those, which are found above the sediment and infaunal organisms are those, which stay in the interstices of the sediment (Gire, 1993). On the basis of their sizes the benthos are classified as Macrofauna 500 (μm), Meiofauna (63 μm), and Microfauna (42 μm). Epifauna is best collected in nets dragged along the surface while, infauna is collected with the help of a corer, either box or cylindrical hand corer in the benthic studies. In general macrofauna includes larger members of invertebrate phyla consisting of insects, Oligochaeta, Polychaeta, Amphipods, Gastropoda, and Bivalves. Meiofauna includes largely of nematodes, ostracodes, and herpacticoid copepods while microfauna include protozoa consisting of the larval phases of meiofauna, diatoms, cyanobacteria and dinoflagellates (Thiel & Higgins, 1988).

Benthos especially macro meio and microbenthos form an important link in food chain. Feeding on primary production, bacterial biomass and organic matter that they form is the biomass, which is of prime importance to many fishes and crustaceans.

It was understood that many authors have focused only on single factor at a time considering phytobenthos, microalgae, and zooplankton, meiofauna or macrofauna in

shrimp culture systems. However, there is paucity of information pertaining to overall composition of benthic biota, available in terms of macro, meio and microbenthos, and their abundance as natural food organisms for the shrimp, in modified extensive shrimp culture systems. Further, role of aeration in the development of benthic faunal organisms in shrimp culture systems is also lacking. Following is a review of literature in this regard to the above mentioned concerns, on different types of benthos (macro-, meio-, microbenthos) which have been studied earlier.

2.2.2 Macrofauna Review of Literature

The biomass in the marine sediments is dominated by macrofauna, a group of invertebrate polychaetes, molluscs, crustaceans and other phyla. Based on size, macrofaunal organisms are classified as those organisms which are retained on the 500 μm sieve (Reise, 1979). Macrofaunal species number has been shown to range from 5×10^5 to 10×10^6 by Snelgrove (1998). Macrofauna in marine sediments play important roles in ecosystem processes, such as nutrient recycling, pollutant metabolism, dispersion and burial and secondary production (Snelgrove, 1998). Their community composition, biomass and productivity are controlled by multiple interactive physical and biotic factors. Major physical factors include salinity, temperature, current speed, oxygen availability, sediments and water-sediment exchange phenomena. Influence of environmental variables has been studied earlier by Ingole *et al.* (1987) for effect of salinity chlorophyll *a*. Mud content, bed level height and tidal current velocity, has been studied by Ysebare and Herman (2002). They, has further asserted that, on larger spatial scales, 10^3 - 10^4 m assemblages and individual species abundances correlate significantly with the environmental variables. Further, their distribution and abundance is considered to be dominated by local abiotic factors such as grain-size; wave exposure and tidal range (McLachlan, 1996). Physical and effect of anthropogenic activities on macrofauna has been studied by Ansari *et al.* (1986) found that, excavation of sand gravel increases the turbidity and sediment precipitation leading to the changes in the distribution

patterns of macrobenthos. Ingole and Parulekar (1998), Ingole *et al.* (2002) has showed the importance of salinity as controlling factor for the distribution of benthic organisms in tropical environments. According to Austen *et al.* (1998) low salinities contribute to the decline of macrofaunal densities by reducing larval recruitment. Further, according to Lui *et al.* (2002), the benthic macrofaunal activity inside soft sediments affects physico-chemical characteristics of their habitat through their activities such as bioturbation and which in turn bring about changes in the associated meiofauna community structure. Lui *et al.* (2002) has reported weak correlation between macrofauna and dissolved oxygen and salinity and have found weak negative correlation between macrofauna and organic carbon from tidal impoundment of marshy areas. Macrofauna are important in terms of secondary production (i.e. converting plant material to meat) as direct food sources for bottom-feeding species, that are commercially fished (Carlson *et al.*, 1997). The food of macrobenthic animals however, is mainly aerobic, heterotrophic micro-organisms or small meiobenthic animals such as nematodes, ciliates, etc. which depend trophically on microorganisms (Wildish, 1977). Warwick and Price (1975) studied the mud flats and measured the biomass as well as production of the macrofauna and found mean biomass of macrofauna to be 13.2 g.m^{-2} ash free dry weight, and production of 13.3 g.m^{-2} . Gerlach (1978) showed that, productivity of the subtidal meiobenthos is maintained in same order of magnitude as that of macrobenthos by consuming meiobenthic production in the sediments by the macrobenthic invertebrate population, thus controls meiofauna population. According to Montagna and Yoon (1991) abundance and biomass of benthic infauna increased when nutrient loading from river input is transformed into food, for benthic animals. According to him, this occurs because nutrients introduced by a river stimulate primary production and thus macrobenthos feed on the microphytobenthos. Macrofaunal crustaceans, are highly mobile, and normally occur in well oxygenated environments, generally appear to be highly sensitive to hypoxic conditions, surviving only a few hours in oxygen-deficient water (Jorgensen, 1980). Less mobile, sediment dwelling animals are normally more tolerant of low oxygen levels. According to Theede *et al.* (1973)

generally, the most tolerant benthic macrofaunal taxa seem to be a few selected bivalves and soft-bodied taxa such as polychaetes and sea anemones (Jorgensen, 1980). Some of the bivalves by closing their shells and reducing metabolism, survive up to 2 months in anoxic water (Theede, 1973). In macroinvertebrates the bulk of species are either suspension or deposit feeders. Suspension feeding species occur in areas of higher current speeds while deposit-feeding macrofauna can be found where there are low water currents (Wildsh, 1977). The macrofauna play an important role in the supply as well as mineralization of organic matter. Suspension-feeders link the pelagic and the benthic environment, while benthic deposit-feeders redistribute organic matter deposited on the sediment surface by sediment reworking, and oxidize the sediment by ventilation.

Reise (1977) was among the first, who proposed that, predation should be considered as the dominant structuring factor for benthic infaunal communities. Allan and Maguire (1992) found that, macrofauna forms an important source of food for cultured shrimps. They found that, macrobenthic density decreased as the feeding pressure increased, and hence the impact of shrimp predation has been considered as main controlling factor. Sarda *et al.* (1998) studied the impact of epifaunal predation on the structure of macroinfaunal invertebrate communities of tidal salt marshes and found that, the absence of epibenthic predators favors growth of larger organisms and macrofaunal biomass as high as 22.2g dry weight m⁻². They further showed that, macroinfauna in the sediments can support the energy requirements of the predators of the marshes and thus contribute in the food web. Macrobenthic crustacean in the diet of the fish in relation to environmental parameters has been studied by Serrano *et al.* (2003). Six major groups like, Polychaeta, Tanaidacea, Isopoda, Gastropoda, Amphipoda and Bivalvia has been reported by Soares, (2004) from *Farfantepenaeus paulensis* culture pens, and found that the abundance inside pens dropped by 86% due to predation. Hena *et al.* (2004) reported that Gastropoda, Foraminifera, Polychaeta, Bivalvia and Insecta are preyed upon by shrimp. Rejection of *Telescopium telescopium* has been shown in their study, and has mentioned that, they are rejected by shrimps due to their larger (1.0-2.0 cm) size. Shishehchian *et al.*

(2001) showed that, in intensive culture ponds the density of Polychaets, Harpacticoid copepods and insect larvae normally decrease to low level after eight weeks, probably due to the natural fluctuations in benthic populations, grazing by shrimp, or deterioration of pond bottom. Van der Veer (1998) showed that, high shrimp densities $10 \text{ individuals.m}^{-2}$ predate on bivalves up to an extent that, their recruitment process is affected by the rate of predation by shrimps. Polychaets as the predominant macrofauna has been reported by Nunes and Parsons (1999), Nunes, *et al.* (1997) have stated that, polychaeta abundance in commercial penaeid aquaculture operations reflects pond productivity and availability of natural food. Nunes and Parsons (2000) has reported six polychaeta families and found that, their density varied from 956 to 11,921 polychaetes. m^{-2} contributing the biomass of 1.17 to 2.58 g.m^{-2} . From these studies it is evident that, macrofauna makes available a rich source of food to shrimps, which will ultimately help farmer in the optimization of shrimp stocking, feeding and harvesting.

2.2.3 Meiofauna Review of Literature

The term meiofauna is derived from the Greek word meio meaning "small" (Heip, 1992, Gire, 1993). It was first used and defined by Mare (1942), as an assemblage of mobile or hapto-sessile benthic invertebrates distinguished from macrobenthos by their small body size. According to Coull and Bell (1979) meiofauna size refers to the fauna smaller than, what has been defined as the lower size limit of macrofauna. The size of meiofauna ranges in between $50\mu\text{m}$ and $500\mu\text{m}$. Meiofauna occur in both freshwater and marine habitats, from high on the beach to the deepest parts of the sea. They are universal in their distribution and inhabit the sediments of all kinds, from the soft fine muds to the coarsest shell gravels, (Coull, 1999). Usually, larvae of the macrofauna, are a part of the meiobenthos during their juvenile stages and is known as "temporary meiofauna", and many taxaon which are meiobenthic throughout their life cycle are known as "permanent meiofauna" McIntyre (1969). Permanent meiobenthos includes mainly Nematoda, Polychaeta, Copepoda, Ostracoda, Turbellaria, Mystacocarida and many representatives of Rotifera (Thiel

and Higinns, 1988). Meiofauna in the marine environment play an important role, as trophic link between bacteria and larger fauna, it enhances the rate of carbon mineralization by stimulating microbial activity through predation, and most importantly consumption of detritus (Heip, 1992, Schmid and Schmid, 2000). Distribution of meiofauna is affected by various biotic and abiotic factors. A range of abiotic factors includes tidal pressure and wave actions in intertidal sandy beaches and estuarine mudflats (Armonies, 1988). Meiofauna are affected by light intensities, currents and shear caused due to the strong currents has been studied by Palmer and Gust (1985). These fluctuating environments provide a challenge to benthic organisms and force them to adapt to an unstable environment, or to migrate to deeper sediment layers (Steyaert *et al.* 2001; Armonies, 1988). Hence, meiobenthos often show an aggregated vertical distribution within the sediment Ingole *et al.* (2005) and (2006). Kulkarni *et al.* (2003) in their study, on meiofauna from estuarine mud flats found that, most of the meiofauna has been restricted to the upper 2cm and it decreases vertically due to oxygen limitation even though food in the form of organic carbon is persistent with decreasing depth. Hence oxygen and not the food is limiting to meiobenthos in mud flats.

Their patchy horizontal distribution has been studied by Gerlach (1977), who suggested that, dead fish and crustaceans remains at the bottom can provide the source of organic matter which is patchily distributed. This in-turn can limit the horizontal distribution of meiobenthos. The high temporal variations in mud and low variations in sand have been attributed to predation and physical activity due to wave action by Coull (1986). Further, Coull (1988) has showed the importance of grain size where certain taxa were restricted to particular sediment types. He found that, sediments where the median particle diameter is below 125 μm tend to be dominated by burrowing meiofauna. The sand fauna tends to be slender, as it must maneuver through the narrow interstitial openings, whereas the mud fauna is not restricted to a particular morphology but is generally larger.

Schratzberger *et al.* (2000) studied the structure and taxonomic composition of meiofaunal assemblages as an indicator of the status of marine environments found

that abundance and diversity of meiobenthos were found to be higher in offshore sediments than the inshore habitats. This difference was further attributed to the natural as well as anthropogenic activities. Thus, meiofauna density and diversity are basically dependent not only on the biotic, abiotic environment but also are affected by the anthropogenic activities. Gerlach (1971) has compared macrobenthos and meiobenthos and found that, meiobenthos are important in terms of food consumption and in terms of biomass provide 15 % to the food chain in sublittoral silty sand. According to Coull (1999) meiofauna densities in uncontaminated natural estuarine regions can reach up to $10^6 \cdot m^{-2}$ world wide with dry biomass of 0.75 to 2 $g \cdot m^{-2}$. Gerlach in (1978) showed food chain relationship in subtidal silty sands of marine sediments and role of meiofauna for stimulating bacterial productivity, where he showed the importance of bacteria as food for deposit feeding macro, large sized meiofauna and microfauna. In food chain they offer a live or dead food, for many benthic fishes and crustaceans important in the fishery point of view. They are high in protein, fats, cellulose, lignin, starch, waxes and oils that supplemental feed cannot provide (Hena *et al.*, 2004). Nematodes regularly dominate the meiofauna in sediment biotopes comprising >50% the total meiofauna. Harpacticoid copepods are usually second in abundance but may dominate in some coarse grained sediments. Meiofauna generally has highest species diversity than macrofauna. According to Heip *et al.* (1988) and Warwick *et al.* (1988) the time consuming identification of meiofauna species seems to be unnecessary in many instances and higher taxonomic levels may provide sufficient information. This implies to shrimp culture practices also. Considering the omnivorous indiscriminate feeding nature of benthic fishes and crustaceans their species level identification can be unimportant. Smith and Coull (1987) found that juvenile spot given a choice of sediment type prefer to feed in muddy substrates consisting of meiofauna and were responsible for the removal of seven of nine taxa and in contrast grass shrimp removed only two taxa. Nelson and Coull (1989) showed that, harpacticoid copepods account for 70% of the prey items even though sediment composition comprised of meiofaunal nematodes 70% and only 17% of copepods. Consumption rate of pink shrimp *F. duorarum* has been

studied by Corona *et al.* (2000) indicates that, consumption rate of pink shrimp intensified as the prey density of amphipod increases and varies from 1.20 to 2.07 individual.h⁻¹. Similar observations has been reported by Gregg and Fleeger (1998) on Grass shrimp *Palaemonetes pugio* fed on the suspended meiobenthic copepods, where feeding rate increased with increasing suspended harpacticoid copepods in the sediment and reported rate of consumption on meiofauna to be 35 individual.h⁻¹. According to Warwick *et al.* (1979) meiofauna contribute about 65% of the total benthic metazoan production, however only 13% is predated upon by other benthic infauna and rest 83% is available to epibenthic predators such as crab, shrimps, fish and birds. Predatory effects of *Crangon crangon* on meiofauna in Sweden has been studied by Pihl and Rosenberg (1984) and suggested that, infaunal communities are heavily influenced by the epibenthic predators. Gee *et al.* (1984) has shown that, when caged, high densities of brown shrimp *Crangon Crangon*, significantly depleted infaunal communities inside the cages. Jensen and Jensen (1985) found that, *Crangon Crangon* juveniles mainly feed on meiofauna especially harpacticoid copepods, nematodes and oligochaets. Focken *et al.* (1998) studied the contribution of natural food in *Penaeus monodon* found that, crustacean diet comprised only 1.8% and food preference after six weeks was more at night than during day time. He further found that, gut content reduced when dissolved oxygen depleted below 4 mg.l⁻¹. Nematodes were found to be most dominant meiofaunal taxa followed by copepods and naupli by Neira *et al.* (2001), they, also found that, increasing bottom water oxygen concentration and decreasing organic matter availability influenced the meiobenthos abundance. Heip *et al.* (1992) have mentioned as general observation that, copepods are more strongly influenced by the environmental factors such as organic enrichment leading to rise in redox-potential discontinuity layer than nematodes and that, copepods are intolerant to hypoxia and their density decreases well before nematode density when oxygen is depleted in the sediment.

It has been mentioned by Coull (1999) that, predators depend on the meiofauna, however meiofauna prey populations tend to be large and predatory removal in nature is very small, and thus predation is unlikely to drastically reduce prey

populations. Hence, unique systems such as shrimp culture ponds; meiofaunal abundance can be limited not only by predation but also by the physical and chemical constraints. From literature reviewed, it is clear that meiofauna is affected by physico-chemical environment, they also form secondary link in food chain thereby, controlling microbenthic population and providing food for higher trophic levels. However, there is paucity of information and lack of understanding for the meiofaunal abundance and diversity and their role as prey organisms to cultured shrimp in modified extensive shrimp cultures. During this study an attempt in this view has been made to study the meiobenthos from modified extensive shrimp culture systems with aeration systems to check whether aeration play any role in development of meiobenthos.

2.2.4 Microbenthos Review of Literature

Microbenthos includes phyto and zoobenthos and is classified on the basis of their size, as those groups of organisms which are retained on 42 μm sieve. The benthos in the euphotic zones comprises vegetated habitats colonized by photosynthetically active microorganisms. Microphytobenthos is a term which refers to the microscopic, unicellular eukaryotic algae (Baccilariophyceae, Chlorophyceae and Dinophyceae) and the prokaryotic Cyanobacteria which live on sediment surfaces (McIntyre, 1996). They grow in habitats ranging from intertidal sand and mud flats, salt marshes, submerged aquatic vegetation beds to subtidal sediments. On sandy and muddy substrate, edaphic microalgae living on a variety of benthic surfaces are often dominated by diatoms (Colijn and De Jonge, 1984, Admiraal *et al.* 1984, De Jonge & Colijn, 1994, Agatz *et al.* 1999) whereas, coccal and filamentous green algae and cyanobacteria are usually known to occur at seasonal stages (Yallop *et al.*, 1994, Taylor and Paterson, 1998, Hillebrand & Kahlert, 2001, Nozaki, *et al.* 2003). Many diatom genera have representatives in groups such as *Nitzschia sp.*, *Navicula sp.*, *Amphora sp.* (Wolff, 1979). The typical characteristic of a microphytobenthic community shows that, benthic algae may not be strictly edaphic and that planktonic forms can temporarily dwell on sediments (Drebes, 1974, De Jong & De Jonge,

1995). Microphytobenthos can be resuspended by water currents and wave action and thus, they dwell in the water column and contribute to the planktonic community (Drebes, 1974; De Jonge & van Beusekom, 1995; De Jong & De Jonge, 1995). On the other hand, phytoplanktonic organisms can sink to the bottom under calm conditions and settle on the sediment in considerable amounts where they live on and photosynthesize and become incorporated into the microphytobenthos (Potter *et al.*, 1975; Blomqvist, 1996). Thus, these benthopelagic forms must be taken into consideration.

Microphytobenthos contribute significantly to primary production in littoral zones than macroalgae or vascular plants (Daehnick *et al.*, 1992, Pinckney & Zingmark, 1993, Colijn & De Jonge, 1984) and in many shallow aquatic systems the biomass of benthic microalgae exceeds that of the phytoplankton in the overlying waters. They alter sediment properties like erodibility, either directly by forming a mat or scum on the sediment surface, and indirectly by modifying the activities of benthic infauna (e.g., pelletization, burrowing, tube building, and sediment tracking). Carbon dioxide fixed by the microphytobenthos supports higher, grazing trophic levels. These include deposit-feeding and suspension-feeding macrofauna as well as meiofauna and microfauna. Quantitative relations between the feeding and growth rates of macrofauna and the abundance of microphytobenthos and suspended organic matter (i.e., functional responses) are reviewed.

The main limiting factors of microphytobenthos are the availability of nutrients and light. Hence, microphytobenthic assemblages are found at the uppermost surface layers of the sediments, right at the sediment-water interface. As the penetration of light is largely confined to the upper 0.2-2 mm, the distribution of benthic microalgae is restricted to this relatively thin surface layer (MacIntyre *et al.*, 1996). The layers in which light is good enough allowing the microphytobenthos to photosynthesize vary both with the granulometry of the sediment and its organic content (MacIntyre *et al.*, 1996). Many benthic microalgae are known for their mobility and they show diel rhythms of vertical migration, moving to and away from the surface in response to a multitude of factors e.g. light, tide cycles, desiccation,

predation or resuspension (Admiraal *et al.*, 1984, Pinckney & Zingmark, 1991, Paterson *et al.*, 1998).

Within a few millimeters of depth, the sediment properties go from fully oxygenated to anoxic conditions and pH, sulphide, irradiance, and nutrients are known to show strong vertical variability (Joergensen *et al.*, 1983, Wiltshire, 1992, Wiltshire, 1993). Despite their variability over vertical scales, there also appear to be considerable spatial fluctuations on a horizontal scale (centimeters to meters). Possible causes for the patchy distribution are variations in the texture (Joergensen & Revsbech, 1983; Jumars & Nowell, 1984) or microscale nutrient, irradiance, water content and salinity gradients (Wolff, 1979). Because of their location at the sediment surface, the microphytobenthos play an important role in modulating nutrient fluxes at the sediment-water interface and this is particularly important with regard to oxygen and nitrogen budgets of the sediments (Sundbaeck *et al.*, 1991, Wiltshire, 1993, Wiltshire *et al.*, 1996). In general it is assumed that, growth of benthic microalgae is not limited by nutrients, since nutrient concentrations in the pore water are generally high (Cadée & Hegemann, 1974, Admiraal, 1984). However, in the thin layer diatoms at the sediment surface biomass may be highly concentrated, and thus nutrients may temporarily become depleted (Admiraal, 1977). When abundant, the microphytobenthos can stabilize the sediment surface against resuspension and erosion by secreting mucilaginous films and forming thin, brownish mats or carpets (Paterson *et al.*, 1990, Delgado *et al.*, 1991, De Brouwer & Stal, 2001). These biofilms are mainly formed by diatoms excreting Extracellular Polymeric Substances (EPS), whereas the amount of excretion is directly related to the rate of primary production (Cadée & Hegemann, 1974).

The life cycles of benthic microalgae are complex and past research shows that some broad generalizations can be made. According to many authors, the biomass of microphytobenthos in sheltered, muddy habitats is higher than in exposed sandy habitats (Cadée & Hegemann, 1974, Colijn & Dijkema, 1981, Delgado, 1989; Sundbaeck *et al.* 1991). Further, variations in the biomass of microphytobenthos in adjacent but distinct habitats have been shown to be as high as those over large

geographic distances (Sullivan & Moncreiff, 1990; Pinckney & Zingmark, 1993; Moncreiff & Sullivan, 2001). In temperate regions microphytobenthic biomass, primary production and chlorophyll contents show a spring or summer maxima similar to biomass peaks of planktonic algae (Admiral & Peletier, 1980, Khondker & Dokulil, 1988, De Jonge & Colijn, 1994, Sundbaeck *et al.*, 2000, Nozaki *et al.*, 2003). Due to seasonal variations in light intensities in the northern hemisphere, these blooms are of relatively short duration and with increasing latitude they occur later in the year. Many studies on primary productivity and chlorophyll contents of microphytobenthic assemblages have been conducted over the last 30 years. The dominance of particular algal groups at different times of the year have been shown by several authors and it was found that, despite a general dominance of diatoms, green algae and cyanobacteria are known to occur in high abundances during the summer period (Khodker & Dokulil, 1988, Kann, 1993, Yallop *et al.*, 1994, Taylor & Paterson, 1998, Hillebrand & Kahlert, 2002, Nozaki *et al.*, 2003). Seasonal succession has also been demonstrated on genera and species levels and inter-annual taxonomic fluctuations are known to occur (De Jonge & Colijn, 1994; Khodker & Dokuli, 1988).

Microphytobenthos are most important link in food chain as all meiobenthos, macrobenthos as well as megabenthos, fishes, and crustaceans either actively or passively graze on the microphytobenthos. Fenchel (1975) has stated that, different elements of microbenthos which includes protozoa and micrometazoa forms intricate and complex food chains and the base of these food chain is constituted by microalgae and bacteria. They constitute a standing stock of about $100\text{mg C.m}^{-2}\cdot\text{day}^{-1}$. The importance of microphytobenthos as meiobenthic food has been studied by Montegna (1984; 1995) found that, meiofauna grazing rates increases due to increase in microphytobenthic production. Further he found that, when chlorophyll *a* concentration in sediment increased from 31.7 mg.m^{-2} to $124.\text{mg.m}^{-2}$ grazing rates of meiofauna increased from $32.9\text{ ng chlorophyll a.h}^{-1}$ to $176\text{ ng chlorophyll a h}^{-1}$. Effect of meiobenthic copepode grazing on microphytobenthic diatoms has been studied by Decho *et al.* (1986;1988). Bund *et al.* (2004) found decrease in total

biovolume of diatoms with fertilization, but diatom increased with higher densities of planktivorous fish and thus, showed that predation by fishes in fact enhances the production of phytoplankton. Kang *et al.* (2006) studied seasonal development of microphytobenthos and found that, they serve as food source and are responsible for growth and reproductive activities of bivalves in mudflats. Thus, microbenthic diatoms play an important role in nutrition of many benthic organisms and are at the base of the food chain. Low chlorophyll *a* concentrations in natural ponds has been studied to show less concentration than semi-intensive ponds (Budford, 1997). Johnston *et al.* (2002) studied the water quality and plankton densities in shrimp mangrove forestry farming systems and found that, zooplankton and macrobenthos though were sufficient only for lower stocking densities; however, they were insufficient along with phytoplankton in higher density shrimp ponds. According to Johnson *et al.* (2002) low densities of phytoplankton can be the factor, which limits the grazers like macrobenthos in the shrimp culture ponds. Thus, microphytobenthos is an important link to higher trophic levels. Microalgae and microbenthos thus have been studied extensively, however their diversity and abundance from modified extensive shrimp culture system with effect of aeration has been lacking. In this view, present study focuses on the development of microbenthos and their contribution in shrimp nutrition.

SECTION 2.3

***FEEDING BEHAVIOR OF SHRIMP
AND IMPORTANCE OF
NATURAL FOOD
IN SHRIMP NUTRITION***

CHAPTER 2

Section 2.3 Feeding Behavior of Shrimp and Importance of Natural food in shrimp nutrition

Behavior in an organism is learnt, by adapting to outside conditions. The behaviorism theory applies in its basic sense in terms of adaptation of an organism to the ambient environment, and thus, behavior emerges as the response to the ambient environment. According to Ruben *et al* (2005) behavior reflects the condition in which the organism resides, which is also known as its habitat. Shrimp is a benthic burrower and spends its adult life on or near to the bottom hovering, resting or burrowing. The burrowing behavior is a characteristic. Earlier, Ruello (1973), Joshi *et al.* (1979) has studied the behavior of *Metapenaeus macleayi*. This, shrimp buries itself by fanning the sediment particles backwards with the outstretched pleopodes. It simultaneously moves downwards and forwards in to the substratum with the periopods and tailfan provides the further thrust. After this, shrimp lies with the abdomen flat below surface and only antennules, eyes and distal part of the rostrum is visible.

During present study, shrimp feeding behavior was observed from the modified extensive shrimp culture systems. These are unique environments and are different, in view that, they are confined, and thus differ from its natural habit and habitat. However, the borrowing behavior reported for different species of shrimp and rather crustaceans in general is similar. Different authors have studied shrimp behavior in terms of their environment and its ultimate effect on their feeding and growth in shrimp culture practices. Martin *et al.* (1998) has shown that conditions at the water and sediment interface may turn deleterious during low oxygen conditions. He has speculated that, feed and excreta accumulating at the bottom increases the oxygen demand at the sediment water interface and thus creates anoxic conditions. In such condition shrimp may come under stress and cease feeding. Reduction in living space on feeding, growth and survival has been documented by Allen and Maguire (1992), Rasheed and Bull (1992). They have shown that, due to space constraint,

shrimp behavior changes and ultimately feeding and growth is affected in shrimps. Shrimp behavioral studies further shows that, shrimp are active during the day time and they rest through the day. At day time as described above, they burrow themselves under the sediment and at night they become active (Vance, 1992, Cartes, 1993). The burrowing and sheltering behavior of juvenile tiger prawns during light and dark periods has been studied by Kenyon (1995). He has asserted that, they show this behavior so as to seek protection from the predators in the natural environment. In shrimp culture systems however, this behavior seems to be more natural instinctive, as there are no predators as such. Saying that, shrimp also show cannibalism, but only weak and small or freshly dead shrimps are predated upon. The role of overcrowding and competition for the food leading to cannibalistic feeding behavior has been described by Abdussamand and Thampy (1994). They found that, deteriorated water quality also, exert its toxicity to the organisms and change their behavior for feeding, and show stunted growth due to underfeeding. In shrimps the different stages from naupli to zoea, mysis, post larvae and adult stage show different feeding preferences Rothlisberg (1998). Penaeid larvae hatch as a non-feeding stage, called nauplis (ranges from 5-6 stages) and pass through three protozoal (PZ1-3) stages and three mysis (M) stages before reaching the postlarval stage (PL). Penaeid hatcheries conventionally rear penaeid shrimp larvae on microalgae (diatoms, flagellates etc.) during the zoeal stage, and zooplankton *Artemia*, rotifers during later stages 2, 3, & 4. Kumlu (1996) has studied the role of live unicellular algal feed in penaeid shrimp nutrition during the protozoa stages and animal prey are added along with algal feeds during the mysis and early postlarval stages. He has reported that, several species of algae, e.g. the phytoflagellates, *Tetraselmis* and *Isochrysis*, and the diatoms *Skeletonema*, *Thalassiosira* and *Chaetoceros* are adequate food for penaeid larvae. The mysis and postlarval stages in the hatcheries are fed on the carnivorous diet such as artemia (Rothlisberg 1998). The adult shrimp has been described as the omnivorous predator. Studies by Wassenberg and Hill, in early (1987), had shown the predatory behavior of shrimp. They have asserted that, in natural environment adult shrimp

predominately feeds on carnivorous diet, such as small mollusca, crustaceans and ophurides. Marte (1980) has shown that, female *Penaeus monodon* do feed more actively than the males as seen from the higher mean gut index of females, in all samples. He has shown that, in shrimps feeding activity differs from season to season and has shown the feeding behavior of the male and female shrimp from September to December being more active than other periods. Food selection and consumption of the shrimp *Crangon crangon* in shallow marine areas has been studied by Pihl and Rosenberg (1984). They found that, food was selected according to size, that is, larger shrimp ate large pray and that, the consumption changes according to seasons due to the changes in infaunal organisms. According to Solis (1988), *Penaeus monodon* diet preferences includes crustaceans and mollusks (85% ingested food), and the remaining 15% consisting of vegetable matter, polychaetes, fish, debris and sand, indicating that the giant tiger prawn is more of a predator rather than a scavenger or detritus feeder.

According to Marte (1980), shrimp increases feeding activity during ebb tide. Further, according to Taylor and Peck *et al.* (2004) the metabolic rates of crustaceans are known to be highly variable. They have sited that, the metabolic rates of crustaceans are influenced by several environmental (exogenous) and biological (endogenous) factors. Exogenous factors include salinity, light regime, dissolved oxygen and temperature, while endogenous factors include such as elevated activity levels and body size. Shrimp feeding has been studied as early as (1952) by Gopalkrishna, for different size groups of *Penaeus indicus*. Hall (1962) has shown that, crustacean were the most dominant prey, while polychaeta ranked next in the abundance in the shrimp feeding. The classical experiment carried out by Bell and Coull (1978) shows that, shrimp feed on wide infaunal benthic organisms which mostly includes copepods, nematodes, polychaetes and oligochaetes. In an experiment they found that, the meiobenthic species differed significantly after introducing shrimp to the experimental tank, and thus concluded that, shrimp predation regulates meiofaunal organisms. Wayban *et al.* (1987) showed that small scale shrimp farming including extensive and modified extensive shrimp farming,

the quality and quantity of the natural food resources is very important. Further, Cano *et al.* (2004) has showed that in shrimp culture natural productivity of the water generate 100–300 kg/ha/yr and fertilization generate 600–1 000 kg/ha/yr. According to him in intensive farming the use of feeds can raise productivity up to 20,000 have been reported in Taiwan or even 30,000 kg/ha/yr in Japan; however it can bring about the nutrient rich high BOD waters. The intensive studies carried out by Jory (1995) indicates that use of high protein feeds proves to be unnecessary when high abundance of natural food is present in the culture system. However, D' Abramo and Conklin (1995) showed that, as the shrimp biomass increases the contribution of natural food becomes insufficient to cover the protein requirements of the farmed organisms like shrimp.

Aquatic species such as shrimp and fish, when reared in high densities, require a high-quality, nutritionally complete, balanced diet to grow rapidly and remain healthy. When they are reared in environments with high density indoor systems or confined in cages, they cannot forage freely on natural feeds, and hence they are provided with a complete diet. Prepared or artificial diets may be either complete or supplemental. Complete diets supply all the ingredients (protein, carbohydrates, fats, vitamins, and minerals) necessary for the optimal growth and health of the aquatic species (Aquacop and G. Cuzon, 1987. Al-ameeri *et al.* 2006). The artificial diets consisting of ingredients such as squid meal, fish meal, yeast has been studied by Deshimaru as early as (1974), Goddard (1998). Craig and Helfrich (2002), showed that, most fish farmers use complete diets, those containing all the required protein (18-50%), lipid (10-25%), carbohydrate (15-20%), ash (< 8.5%), phosphorus (< 1.5%), water (<10%), and trace amounts of vitamins, and minerals. In contrast, supplemental (incomplete, partial) diets are intended, only to help support the natural food (insects, algae, and small fish) normally available to fish in ponds or outdoor raceways. Supplemental diets do not contain a full complement of vitamins or minerals, but are used to help fortify the naturally available diet with extra protein, carbo-hydrate and lipid. In case of purified diets some protein sources like casein, albumin, gelatine, have been proved successful (Kanazawa, 1972, Deshimaru, 1975)

and more recently crab protein (Boghen *et al.* 1982; Castell, 1987) which is about to be used as a reference protein source. According to Dall *et al.* (1991) the development of artificial diets for penaeid prawns is an empirical process which is driven by the need to provide a diet that gives good growth at minimum costs and further to rationalize the formulation of artificial diets by mimicking the naturally occurring food. In this regard in their study Dall *et al.* (1991) studied the biochemical composition of prey species of *Penaeus esculentus*. In this study it was found that, combined soft tissues of all the natural organisms in diet provide high protein diets, with relatively low mean lipid (12.3%) and carbohydrates (10.6%). Thus, the natural diet provides proteins at least equal to or in the excess of that in successful empirically derived artificial diets.

Gregg and Fleeger (1998) in their experimental study showed that, grass shrimp predate on meiofaunal organisms with rates up to 35 individuals.ha⁻¹. Abundance of benthic organisms in *Penaeus vennamei* has been described by Cordava *et al.* (1998) where they showed that, naupli of different crustaceans, juvenile and adult copepods, larval and adult polychaets, and insects were most dominant organisms. They also found that, shrimps in aeration ponds with 0 to 6 hrs.day⁻¹ showed higher abundance of organisms in their gut. Further, the benthic organism abundance has been reported to be highest, from week 3 to 8, while gut faunal abundance was showed to be higher at week 6 and lowest at week 17. Thus, indicating active predation by shrimp on the benthic organisms. Moss *et al.* (2006) has shown that, in extensive and even in semi intensive shrimp farming systems natural pond biota play an important role and these food sources resemble the food sources taken up in the natural systems. Further, in the same study Moss *et al.* (2006) has showed the deleterious effect of over fortified artificial diets containing vitamins and minerals. Over fortification of the artificial feeds with vitamins such as riboflavin, niacin and vitamin B6 as well as minerals is practiced to avoid loss and leaching of these substances in the water; however this, according to Moss *et al.* (2006) increases the cost and adversely affects the growth of the shrimp. Further, he has concluded that, by endogenously produced microbes and associated detritus,

shrimp farmers and feed manufacturers can reduce vitamin levels in shrimp feeds, resulting in reduced feed costs without compromising shrimp growth, survival, or FCR. Cordava *et al.* (2003) has cited that, diets with high protein levels have negative impact on water quality and natural food contributes about 70% of the nutritional needs of the shrimp and hence their management is important. Thus, the vast information is available on the shrimp feeding behavior on natural fauna as well as artificial feed for different species of shrimp and in different culture systems. According to Cordava, *et al.* (2003) however, the environmental stress imparted due to the artificial feed accumulation and their high nutrient levels leading to production of metabolites especially ammonia occurs, when shrimps are overfed or ponds are over fertilized. Hence, the information available indicates that, shrimp feed on the natural food organisms, and do undergo behavioral changes according to which feeding rate, intensity or selection criterion varies in different shrimp culture systems. From this review it was understood that, there is paucity of information on the contribution of natural biota from modified extensive shrimp culture systems. Further, during present study aeration is used, hence, it is fascinating to see if aeration has any effect on the development of bottom fauna and whether gut organisms differs in composition and quantity in aerated and non aerated shrimp culture ponds.

OBJECTIVES

According to the information obtained from literature review and in view of past and present aquaculture trend and culture practices carried out, it is understood that, there is paucity of information, regarding the distribution and role of benthos collectively, in terms of macro-, meio- and microbenthos in modified extensive shrimp aquaculture system.

With the advent of new aeration system, developed by HOBAS engineering (Norway), it is worthwhile to study the effect of this aeration system in improving water quality and in turn, its effect on the development of bottom fauna. In this view, it was hypothesized that, as aeration is known to improve the water quality, it will also help in the development of bottom fauna due to better oxygen enrichment to macro-, meio-, and microbenthos, which are important source of food for cultured shrimp. In this regard, present research was carried out over complete two culture cycles during 2004- 2006 with following objectives in consideration.

Objectives :

1. To study the effect of artificial aeration on the distribution of benthos and its environment in shrimp culture systems.
2. To study the quantitative and qualitative distribution of benthic fauna in shrimp culture systems.
3. To study the contribution of natural biota in shrimp nutrition, through shrimp gut content analysis.

CHAPTER 3

MATERIALS AND METHODS

CHAPTER 3

Materials and Methods

3.1: Description of Study area.

The area is located on the North-West Coast of Karnataka State of India (Figure 1). Since, the shrimp farming activities on the west coast are concentrated along the banks of Kali and Agnashini creeks, in Karwar and Kumta talukas, respectively, few shrimp at Alvekodi, near Kumta town in North Kanara District of Karnataka, were selected. Beside, the experimental ponds were ideally located from about 10-12.kms. from the local hatchery (Skyline Hatcheries Pvt. Ltd.) providing constant supply of *P.monodon* larvae. The farm also had an easy access for the National Highway No.17, which provided better transport option and good power supply, which was important in present study, as the application of aeration systems were to be studied, which required continuous and quality power supply.

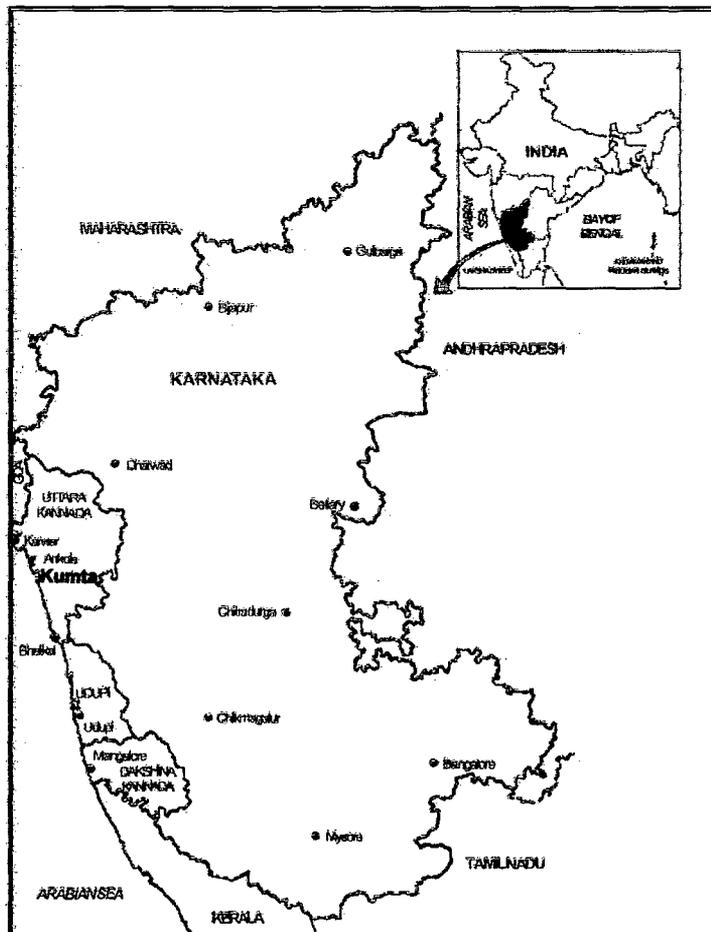


Figure: 1 Map of study area

Geographical location of study area			
Aerated Pond	Non-aerated Pond	Creek	
		Creek1	Creek2
14 ^o 24'42.24"N	14 ^o 24'40.03"N	14 ^o 24'49.20"N	14 ^o 25'09.20"N
74 ^o 24'27.45"E	74 ^o 24'27.78"E	74 ^o 24'28.17"E	74 ^o 24'23.41"E

Experimental Ponds

The farm has a total water spread area of ~6.0 ha of which, two ponds of 0.65 and 0.60 ha adjacent to the feeding creek were selected (Figure.2) for this study. The creek adjacent to the experimental ponds, was tidal fed and ran about 3 to 5-km away from the sea harbouring about 40 ponds with an approximate water spread area of 0.5 to 0.6 ha. Two experimental ponds, namely Aerated Pond and Non-aerated Pond having an area of 0.65 and 0.60 ha respectively and having a average depth of 1.20 m were selected. Two culture cycles were studied i.e., Experimental Cycle -1 (EC-1, 6 June 2004- 2 September 2004) and Experimental Cycle-2 (EC-2, 5 April 2005- 3 August 2005).

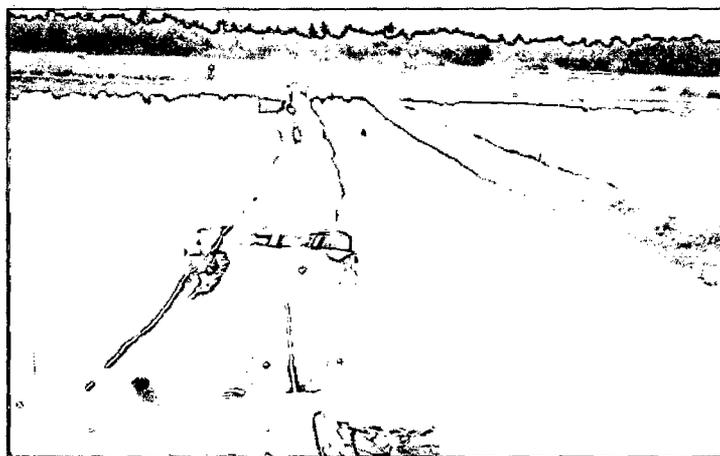


Figure 2: Experimental ponds (LHS: Non-aerated & RHS: Aerated) with Inlet/Outlet canal



3.2: Sampling Program

In each experimental pond, three sampling locations designated as P₁A, P₁B, P₁C and P₂A, P₂B, P₂C were fixed. In the adjacent creek, two sampling stations, namely CR₁ and CR₂, were selected, as it was the only source of water for the culture ponds. All sampling locations in aerated and non-aerated pond and creek are depicted in (Figure 3).

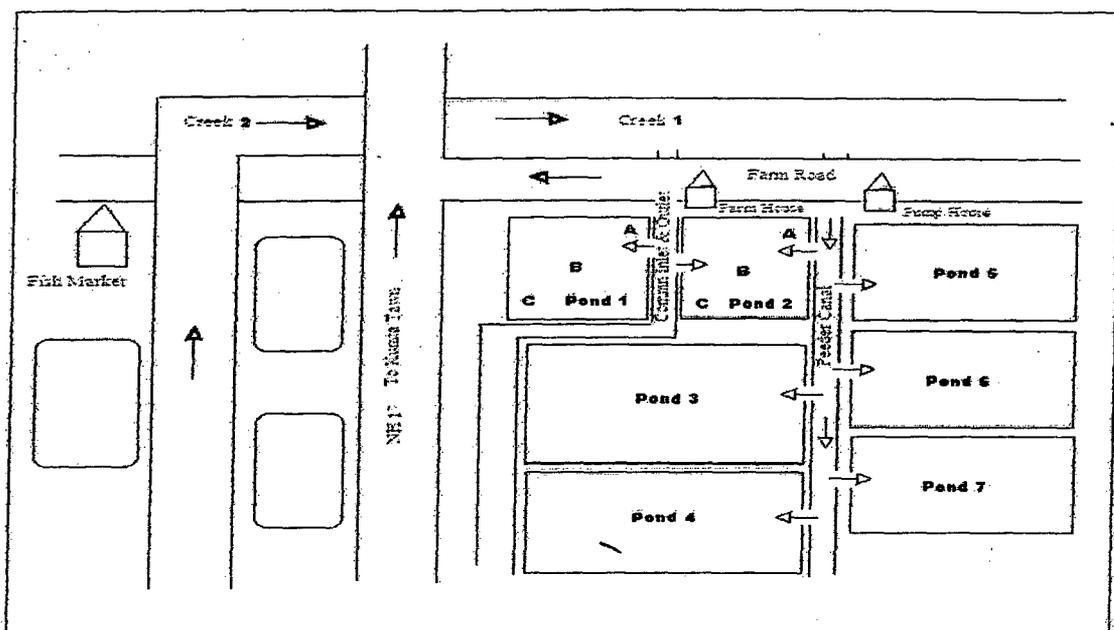


Figure 3: Schematic diagram of Aerated and Non-aerated pond and sampling locations in pond and adjacent creek.

Sampling was done at fortnightly intervals in the morning hours, one hour prior to the high tide at pre-determined sampling locations. Samples were collected for water and soil, for studying physico-chemical parameters, benthos (macro-, meio- and microbenthos). Ten shrimps were collected from aerated and non aerated pond at morning (10.00 hrs.) and night (22.00hrs.) for studying the gut content, to understand its foraging preferences and behavior. Records for same samples were maintained for studying the growth in culture shrimp over two production cycles from May, 2004 to August, 2004 and May, 2005 to August, 2005. Analysis for physico-chemical parameters were done immediately at the field laboratory situated at the experimentation-cum-demonstration farm, wherein benthos and shrimp gut content analysis was carried out at NIO, Goa.

3.3 HOBAS Aeration System

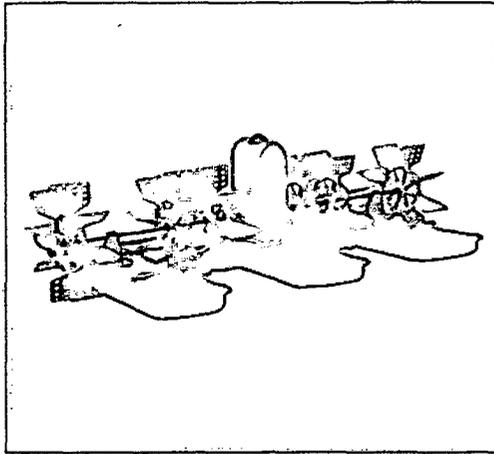


Figure 4: Paddle wheel aerator

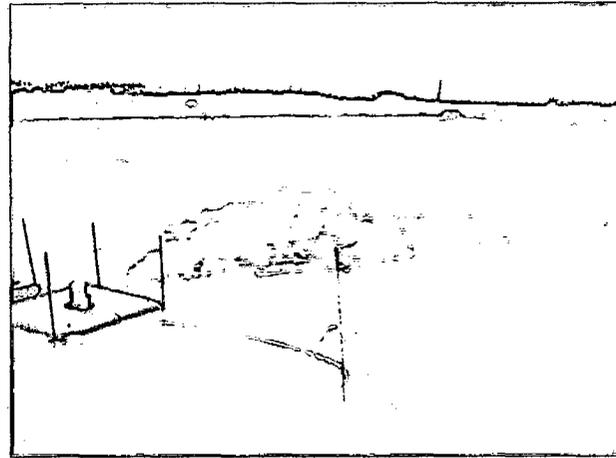


Figure 5: HOBAS aeration unit in operation (Aeration pond) with onshore air regulator.

3.3.1 Description of new aeration (HOBAS) technology

Most conventional aerators employed in aquaculture mixes water with air, either after the pump (e.g. ejector base aeration) or with a propeller in open water, e.g. Aire O₂, Paddle-wheels etc (Figure 4). The HOBAS technology (Figure 5-6) mechanically mixes the air and water, in order to replace oxygen deficits, and to flush the excreted and potentially toxic, carbon dioxide and ammonia. The pump creates horizontal physical water current in the pond when sufficient numbers of aerators are employed and implementation is done correctly. The core unit consists of a pump, a valve, an air inlet hose and a specially adjusted raft for vertical implementation in the pond. While, the pump is running, the partial vacuum in the pump house (submerged in water at the lower part of the *Venturi*) cause air to be pulled into the intake port where air is dispersed into the water (as shown below). Onshore, a valve is used to regulate the amount of air being introduced into the impeller. Moreover, compared to other aeration technologies, the HOBAS technology also introduces several other advantages in practical farm management.

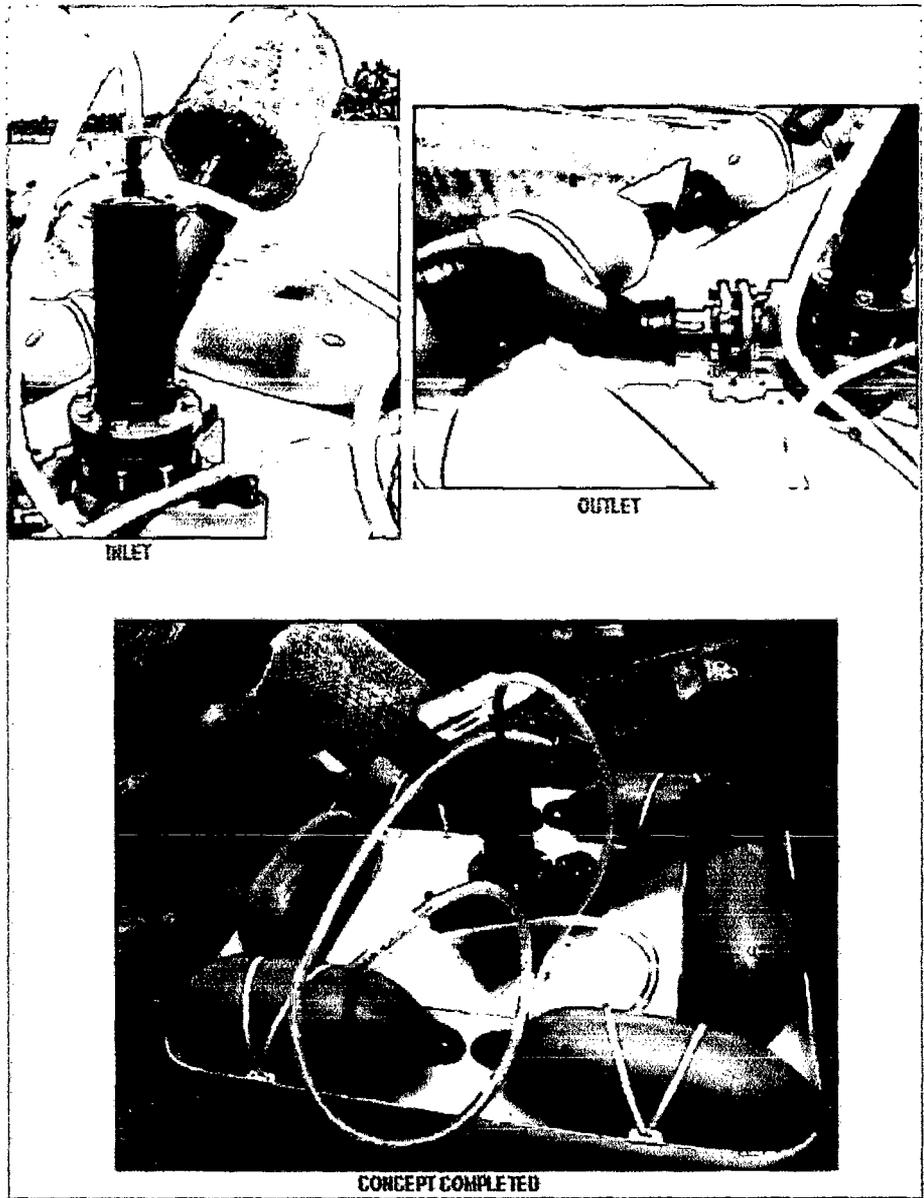


Figure 6: Ventral view of complete HOBAS aeration unit with Inlet, Outlet and floats.

3.3.2: Arrangement of aerators

Arrangement of aerators in experimental ponds was done in such a way, by taking in consideration the wind flow directions and currents generated through wind driven water circulation to achieve better circulation, mixing and to accumulate sludge at the centre of the pond (Figure 7). Four numbers of aerators of 1HP each as showed below were fixed at four corners of experimental ponds.

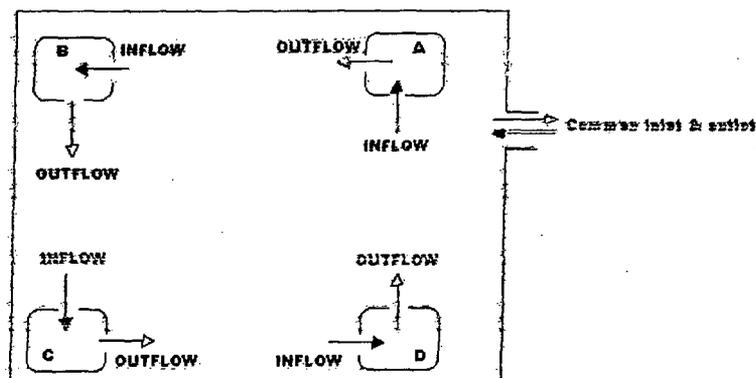


Figure 7: Arrangement of aerators and direction of current flow in experimental pond

3.3.3 Advantages of the HOBAS aeration technology

- Farmers can freely regulate the angle of the outlet from the pump and thus avoid agitation of the pond bottom independent of pond water depth in addition to controlling the direction of physical water current according to the pond shape (individually adjustable).
- Farmers can control and regulate the amount of aeration and circulation according to the DO levels in culture water (e.g. no aeration during the day and only circulation and full aeration during the night) with onshore regulator (Figure 3).
- The pump prevents any kind of stratification and creates uniform conditions in the water column.
- The HOBAS technology is easy to use, has low maintenance, runs smoothly and makes less noise than conventional technology.

3.4: Pond preparation, prestocking activities and stocking of ponds

3.4.1 Pond Preparation

Pond preparation and management was carried out according to the standard shrimp culture practices following the guidelines provided by Aquaculture Authority of India (1999). Before stocking, two ponds were dried for a period of one month and standard liming procedure was followed by applying lime 100kg/ ha to the pond bottom for maintaining pH of soil. Algal mats, dead gastropod shells and organisms were removed manually. The ponds were then filled with water five days prior to the stocking during three high tides with the help of sluice gates (Figure 8) leading to common channel acting as common inlet/outlet channel for adjacent ponds as shown in figure 2 and 8 and electric pump of 10HP through feeder canal.

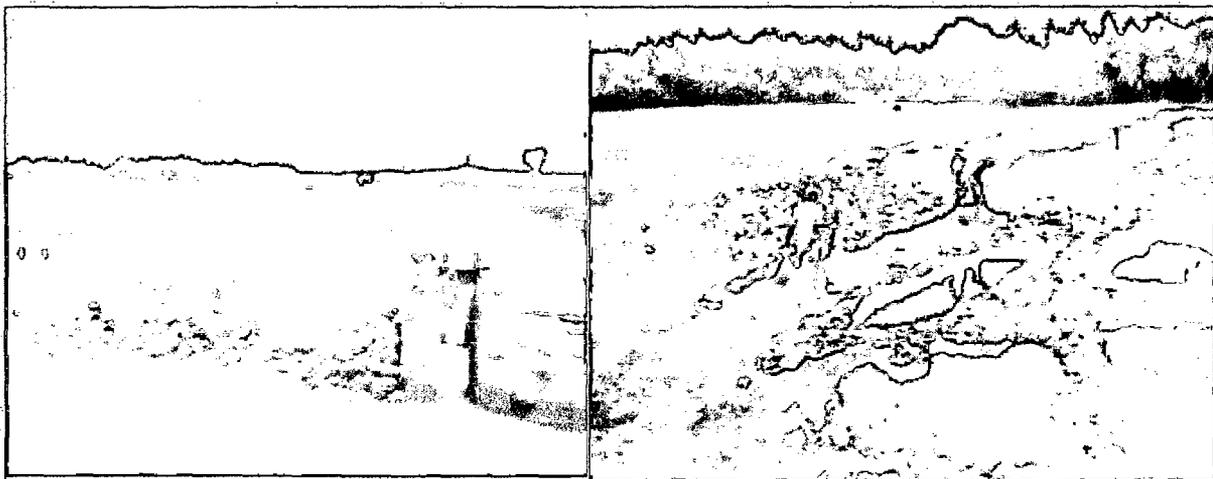


Figure 8: Preparation of ponds before stocking of Prawn Seed

3.4.2 Stocking of ponds

Stocking of ponds was usually carried out in late evenings for which seed was transported from nearby hatchery, 10 km from the experimental ponds. Bags consisting of the shrimp larvae (PL-15) were kept in pond water for acclimatization for ≈ 02 hr and were then released, in previously marked areas.

3.4.2. Shrimp feeding

Shrimp were fed with commercial supplementary feed having stability of 4 hr in water. Feed was administered manually from the periphery of the pond, four times a day, at regular intervals at 6.00, 12.00, 18.00 and 00.00 hr. During administration of the feed, aeration was stopped before and after 1 to ½ hr. Utilization of the feed was monitored with the help of check trays in respective ponds and daily dosages were adjusted according to the check tray monitoring and shrimp body weight.

3.5: Methods physico-chemical parameters of water and soil quality analysis

All physico-chemical analysis of water and soil parameters were carried out using standard analytical methods as described by Strickland and Parsons (1972); Grasshoff *et al.* (1983); Parson *et al.* (1984). All standard, calibrated analytical instruments such as electronic weighing balance (Afcoset ER-200A), pH meter (Lab India μ p controlled PHAN pH analyzer), visible spectrophotometer (Shimadzu UV-1201 V), and Eltek centrifuge (TC-4100 D) were used for the chemical analysis.

3.5.1: Temperature

On the sampling day, water and air temperatures were measured *in situ* with hand held Hg centigrade thermometer (Make-GH-Zeal-London).

3.5.2: pH & E_h

The pH was measured with Orion- combo pH electrode having an accuracy of ± 0.01 . The instrument was first calibrated using standard pH buffers of pH 4.2, 7.0 and 9.2. The reducing or oxidizing potential (E_h) of soil and water were measured (Orion-combo redox platinum electrode) with a reference electrode silver/silver chloride. Positive and negative readings recorded in milli- volts (mv) shows the general oxygen regime of the sediment.

Titrimetric Analysis

3.5.3: Salinity

Salinity of water was measured by Mohr-Knudsen argentometric titration method (Strickland & Parsons, 1972) where, halogen ions of chloride, bromide and iodide form a precipitate with a low solubility product with silver ions.

$$S=1.80655 \times Cl \text{ (PSU)}$$

3.5.4: Dissolved oxygen (DO)

DO was measured by the Winkler's method as described in (Strickland & Parsons, 1972). The dissolved oxygen was initially fixed by adding the Winkler's reagent (WA-Manganous Chloride and WB-Alkaline Potassium Iodide) to the measured volume of water sample immediately after collection. The precipitate so formed was allowed to settle and the DO was analyzed by the titrations after dissolving the precipitate with concentrated HCL.

3.5.5: Biochemical Oxygen Demand (BOD)

The BOD samples were collected by the same method as DO. Immediately after collection the sample bottles (without the addition of any reagent) were kept in dark for 5 days at 20°C. Dissolved oxygen in the samples was determined by Winkler's method after fixing the samples soon after the completion of 5 days of incubation. The BOD was computed from the difference between the initial and final concentrations.

3.5.6: Chemical Oxygen Demand (COD)

COD is a measure of the total amount of oxygen which is required to oxidize all organic matter in sample to $\bar{C}\bar{O}_2$ and $\bar{H}_2\bar{O}$. In this method potassium dichromate ($K_2Cr_2O_7$) is used for carrying out the oxidation. The excess $K_2Cr_2O_7$ is back titrated by standard ferrous ammonium sulphate ($Fe(NH_4)_2(SO_4)_2$) to know actual amount of standard $K_2Cr_2O_7$ required for oxidation of organic matter in water sample.

3.5.7: Sediment Organic Carbon (OC)

Determination of sediment organic carbon was done using wet oxidation method of El Wakeel and Riley (1956). In this method, sediment sample was heated to 175°C . The sample were treated with chromic acid, were heated in boiling water bath for 15 minutes and was cooled and titrated with ferrous ammonium sulphate by using ferrous phenanthroline indicator. OC values obtained were expressed in parent dry weight.

Spectrophotometric Analysis

Spectrophotometric analysis of nutrients as well as chlorophyll-*a* and phaeophytins were carried out using standard analytical methods as described by Strickland and Parsons (1972).

3.5.8: Nitrate-nitrogen ($\text{NO}_3\text{-N}$)

Nitrate in the water sample was first quantitatively reduced to nitrite by heterogeneous reduction by passing buffered sample through amalgamated cadmium column and the resultant nitrite was analyzed as above. The measured absorbance (Shimadzu; UV-1201) at 543nm was due to the initial nitrite in the sample and the nitrite obtained after the reduction of nitrate. Necessary correction was then made for any nitrite initially present in the sample. Nitrate nitrogen obtained so was expressed as $\mu\text{mol.l}^{-1}$.

3.5.9: Nitrite-nitrogen ($\text{NO}_2\text{-N}$)

Nitrite in the water sample was measured by diazotization reaction. A known quantity of sample was reacted with sulfanilamide and then coupled with N-1-naphthylethylenediamine-dihydrochloride. The absorbance of the resultant 'azo dye' was measured at 543 nm on a spectrophotometer. Nitrite nitrogen obtained so was expressed as $\mu\text{mol.l}^{-1}$

3.5.10: Ammonia-nitrogen ($\text{NH}_3\text{-N}$)

Ammonia nitrogen was determined by the indophenol blue method based on the principle that in a moderately alkaline medium, ammonia reacts with hypochlorite to

form monochloramine which in the presence of phenol and an excess of hypochlorite and catalytic amounts of nitroprusside forms indophenol blue. The resultant blue complex was measured at 630 nm on a spectrophotometer. Ammonia-nitrogen obtained so was expressed as $\mu\text{mol.l}^{-1}$

3.5.11: Phosphate-phosphorous ($\text{PO}_4\text{-P}$)

The dissolved reactive phosphate was measured by the method of Murphy and Riley. A known quantity of the sample was made to react with acidified molybdate reagent. The resultant compound so formed was then reduced to molybdenum blue using ascorbic acid. The absorbance of the colored complex was measured at 885 nm on a spectrophotometer. Phosphate-phosphorus obtained so was expressed as $\mu\text{mol.l}^{-1}$

3.5.12: Chlorophyll *a* and phaeophytin

For the estimation of chlorophyll- *a*, a known volume of water sample was filtered through GF/F glass fiber filter paper (47 mm diameter; 1.2 μm pore size) and extracted in 90% acetone overnight. The extracts were used to measure extinction spectrophotometrically at 645nm and 750nm. Chlorophyll *a* and phaeophytin in water obtained so, was expressed as mg.m^{-3} in water and $\mu\text{g.g}^{-1}$ in sediment.

3.5.13: Particulate Organic Carbon (POC)

A known volume of water sample was filtered under vacuum 1/3 atmospheric pressure through Glass Fiber Whatman GF/F filter (47 mm diameter; 1.2 μm pore size) previously ignited in a muffle furnace (450°C for 3 hours). Two ml of sodium sulphate solution (4.5% v/v) was added separately to remove interference of chloride ions. Filters were dried and stored in an aluminium foil until further analysis. Material retained on the filter paper was oxidized by acid-chromate solution and the reduction in Optic Density (OD) was measured spectrophotometrically at 440nm. Standardization was done using glucose as the source of carbon and factor-F was found and column values of POC were calculated and expressed as mg C m^{-3} .

3.5.14: Sulphide (H₂S)

The determination of dissolved sulphide in water was done by methylene blue method. Since hydrogen sulphide is volatile, extreme care was taken to avoid bubbling samples were immediately treated with N, N-dimethyl p-phenylenediamine sulphate and ferric chloride in acidic medium, the methylene blue color thus formed is measured spectrophotometrically at 670 nm.

3.6: Benthos preservation and analysis

Benthos, macrofauna, meiofauna and microbenthos sample collection and preservation were carried out according to Holme and McIntyre (1984).

3.6.1: Macrofauna

Hand corer of 10.5 cm inner diameter was used to collect the macrofauna samples. After collection, these samples were washed through a 0.5 mm mesh size stainless steel sieve and the retained samples were preserved in 10% sea water-formalin containing Rose-Bengal stain. The samples were again washed through 0.5 mm mesh sieve in the laboratory to clear the adhering sediments. All the stained animals were picked and preserved in neutralized 5% formaldehyde. Later, all the organisms were sorted and counted group-wise under a stereoscopic zoom binocular microscope the density was then expressed in no.m⁻². Identification of macrofauna was done up to group level following the taxonomic key of Gossner (1971).

3.6.2: Meiofauna

An acrylic hand corer of 4.5 cm inner diameter was used to collect the meiofauna samples. All the samples were immediately preserved in 5 % seawater- formalin containing Rose-Bengal stain. In laboratory, these samples were passed through a set of sieves, where 0.5mm sieve was placed above the 0.062mm sieve. The materials retained on a 62µm mesh were used for analysis of meiofauna. All organisms were counted by using a counting chamber and microscope as used in macrofauna counting and density was expressed as No. 10 cm⁻². Identification of meiofauna was also carried out up to the group level following the key of Thiel and Higgins (1988).

3.6.3: Microbenthos

A plastic syringe (2 cm inner diameter) was used to collect microbenthos samples. These samples were preserved in Lugol's solution and 5% formalin. All the samples were sieved with the help of 42µm sieve and analyzed using Olympus BX 41 stereoscopic binocular microscope. Identification of microbenthos which comprised mainly the diatoms was done following the techniques described by Horner (2002); Ingole and Goltekar (2004).

3.7 Shrimp Gut content Analysis

Shrimps were collected randomly by using cast net. From four to five hauls 10 shrimps were selected with full guts at 10.00 hrs and 22 hrs, two hrs. prior to feeding. Shrimp were anesthetized in ice chilled water and were dissected immediately. Gut content analysis was done by simple percent occurrence method as described by Hyslop (1980) for establishing the role of natural biota from an experimental and control pond. For this study, the shrimp gut was removed carefully and transferred to a clean petri dish. It was cut opened with the help of scalpel and entire content were transferred to plastic vials containing 5% Formalin and Lugol's solution. All the samples were analyzed for the presence of biota and/or undigested biotic material, if any. Entire gut fauna was identified to a nearest group level. Identification was done using Olympus BX 51 compound microscope and Olympus binocular stereoscopic zoom microscope. Sedgwick Rafter was used for identification of remains of meiofaunal soft body parts and for microbenthos. Bogorove counting chamber was used for the identification of relatively larger and hard remains of macrofauna and algae.

3.8: Shrimp Growth Studies

Data on shrimp length and weight and feeding were collected at fortnightly intervals by taking the length and weight of shrimps as described by Goddard (1996). The absolute growth rate was calculated for shrimp by using following formula.

$$\text{Absolute Growth rate} = \frac{(W_2 - W_1)}{T}$$

Where W_2 - Final weight,

W_1 - Initial weight of the shrimp

T- time in days of culture.

The Feed conversion ratio (FCR) and Feed conversion efficiencies (FCE) were calculated by standard formulas as follows.

$$\text{Feed Conversion Ratio (FCR)} = \frac{\text{Total weight of shrimp (Kg.)}}{\text{Total food fed (Kg.)}}$$

$$\text{Feed conversion efficiency (FCE)} = \frac{\text{Total weight of shrimp (Kg.)}}{\text{Total food fed (Kg.)}} \times 100$$

3.9 Data Processing/ Statistical Analysis

Data processing was done with the help of statistical software packages like Statistica 5.0, SPSS 7.5 for windows and MS-Excel. Correlation analysis was carried out using Statistica 5.0 to study the linear relationship between multiple parameters. The correlations thus obtained at $p \leq 0.05$ have been depicted in statistical tables ST1 to ST6 for aerated non aerated ponds and creek over two experimental cycles. Multiple regression analysis was carried out using SPSS 3.0 for studying the linear relationship between one dependent variable and several independent variables for water quality, benthos, gut content analysis and are described on the basis of their significant relationships at $p < 0.05$ $p < 0.01$ and $p < 0.001$).

Descriptive statistics has been presented in Table ST 7 to Table ST12 for aerated, non aerated pond and creek showing average values, standard deviation, minimum and maximum values observed during Experimental Cycle (EC-1) and Experimental Cycle (EC-2) by using SPSS summary statistics option. Principle component analysis was also carried out using Statistica 5.0 to test, which variables vary most between studied parameters divided over the study period. One-Way ANOVA was carried out using MS-Excel to test whether there are any significant differences between the means of several parameters, studied from aerated and non-aerated ponds as well as creek, over two experimental cycles and to test for an interaction between the two factors.

CHAPTER 4

RESULTS AND DISCUSSION

SECTION 4.1

VARIATIONS IN WATER QUALITY

CHAPTER 4

Section 4.1: Variations in Water quality Parameters

Introduction

Shrimp culture practices require a careful management and monitoring of water and soil quality in culture ponds not only to avoid economic losses due to impairment in the water and soil quality but also to reduce the environmental deterioration caused otherwise due to its effluents in receiving water bodies. Physico-chemical parameters of water are influenced by the seasons. Different seasons such as summer, summer monsoon, winter and winter monsoon reciprocate their traits and alter the hydrology. These hydrological traits are diverse in different physical environments be it open oceans, coastal waters, lagoons, estuaries and rivers and their role in influencing the entire physico-chemical regime is well known. In lagoons and pond water ecosystems changes in physico-chemical parameters turn out to be critical due to the confined nature of these systems. Similarly, aquaculture ponds are artificially confined units and are greatly affected by the seasonality and pose great challenges to shrimp farmer's right from stocking till the harvest. Its understanding therefore is at most important and critical in the decision making in high gain high risk shrimp culture industry.

The importance of maintaining water quality in shrimp ponds has been reviewed in Chapter 2, Section 2.1, and their permissible levels in the water to avoid toxicity are given in table ST13. Experimental ponds during study period were under the influence of summer and summer monsoon. Therefore, one-way ANOVA was carried out to see whether there were any differences in the water quality variables in between two production cycles. It was observed that most of the water quality parameters studied during the **Experimental Cycle (EC)** abbreviated as **(EC-1)** and **Experimental Cycle (EC-2)** did not show any significant differences $p > 0.05$. The variations in individual water and sediment quality during EC-1 and EC-2 has been described prior, followed by discussions either combined or individually on the basis

of better significance of either correlation or regression models carried out by Pearson's product moment correlation in Statistica 5.0 and stepwise and backward multiple regression analysis by using SPSS 7.5 respectively to understand the governing processes by analyzing the linear relationship between water and soil quality parameters.

4.1.1 Results and Discussion

4.1.1 Temperature ($^{\circ}\text{C}$)

Experimental Cycle (EC-1)

Water temperature during the culture period ranged from minimum of, 27.9°C to 35.4°C in aerated pond on 105 and 45 DoC. There were no significant difference between aerated pond and non aerated pond, thus, showed similar temperature values, wherein creek showed slightly lower temperatures ranging from 27.20 to 34.80°C (Figure, EC1.1.), with overall mean of $31.27 \pm 2.21^{\circ}\text{C}$ (Table ST9).

Experimental Cycle (EC -2)

During second experimental study cycle temperature showed decreasing trend, with the progressing culture, it decreased gradually till the end of the culture period. Minimum temperature during culture period was observed to be 28.8°C on 105 DoC for aerated and non-aerated pond. Maximum temperature of 32.6 for aerated pond and 32.8 for non-aerated pond was observed on 0 DoC and 15 DoC for both the ponds as well as for creek. Temperature showed strong positive co-relationship with that of salinity $r=0.87; p \leq 0.05$ (Table, ST4).

Temperature is an important variable as it affects the salinity and subsequently dissolved oxygen in the marine environment. Temperature was high initially up to 30 Day of Culture (DoC) and thereafter it decreased till the end of the cycle during EC-1 (Figure EC1.1), wherein for EC-2 the fall in the temperature was more gradual and started right from next fortnight (Figure EC2.1). The fall in water

temperature can be attributed to the cloud cover during the study period due to which there were less temperature variations. Fortnightly average temperature variations for EC-1 and EC-2 are depicted in the Table and Figure EC1.1 and Table and Figure EC2.1 respectively.

4.1.2 Salinity

Fortnightly average salinity variations are presented in Table and Figure EC1.2 as well as Table and Figure EC2 respectively during EC-1 and EC-2.

Experimental Cycle (EC -1)

Salinity of pond water was as high as 43.18 ± 0.02 PSU on 15 DoC for aerated pond. From 45 DoC it reduced gradually till the end of the cycle to 7.20 ± 0.02 PSU on 120 DoC (Table & Figure EC1.2). For non-aerated pond it ranged from 41.89 ± 0.03 PSU to 8.10 ± 0.01 PSU (Table & Figure EC1.2). The lowest salinity of 1.05 ± 0.07 PSU was recorded on the 60 DoC in the creek (Table & Figure EC1.2).

Experimental Cycle (EC -2)

Salinity showed decreasing trend (Figure EC2.2). On 0 DoC, it was found to be 27.38 ± 0.09 to 27.5 ± 0.05 PSU for aerated and non-aerated ponds. In the creek salinity was found to be 27.64 PSU. Temperatures of the pond and creek showed very minor variations off 1°C (Table & Figure EC2.2). Salinity decrease was observed in aerated pond, being 25.90 ± 0.09 and 26.14 ± 0.04 PSU for non-aerated for pond that is only 1 PSU from 60 DoC. Thereafter it reduced sharply till the end of the cycle. For aerated, pond it was observed to be 4.44 ± 0.08 on 105 DoC and non-aerated pond 5.39 ± 0.07 on 105 DoC. For creek it was found to be 0.28 ± 0.04 PSU on the 105 DoC, is depicted in (Figure, EC2.2).

In the open ocean, salinity is dependent on the temperature and wind. Mohan Kumar *et al.* (1995) has studied the effect of summer monsoon forcing on coastal waters of Karwar, the nearest reported area approximates the hydrological conditions of the coastal waters of Kumta and location of study area. A characteristic

monsoonal gradual decrease in salinity was also observed during the present study for both the experimental cycles. The reason behind the drop in salinity is certainly the fresh water runoff from the creek due to the moderate to heavy raining as it was a south west monsoon season. The salinity of the creek water remained lower than the pond water throughout the culture period and showed fortnightly highly significant differences ($p < 0.001$) with that of pond water during both the experimental cycles. Salinity though decreased and showed drastic variations in the creek it remained slightly higher in the ponds, due to its confined nature as well as limited water exchanges and showed no significant differences ($p > 0.05$) in between experimental ponds. Salinity in confined areas is known to be higher due to more evaporation and lack of water exchanges. In present case therefore, due to the cloud cover temperature variations were not drastic during the day and incessant raining which brought about fresh water input is the main cause of decreasing salinities of water in the pond as well as in the creek. Similar trends of low temperatures and salinity due to wet season have been shown by Cowan *et al.* (1999) in Thai commercial shrimp ponds. Present study area was under the influence of summer monsoon season and role of monsoon in bringing about the changes in water quality and subsequently on planktonic abundance and standing stocks has been studied recently by Madhu *et al.* (2007) in Cochin backwaters.

4.1.3 Hydrogen ion concentration (pH)

Hydrogen ion concentration is an important factor and its importance has been described earlier in Chapter 2. It is influenced by the photosynthetic activity which includes uptake of CO_2 , which increases the pH, while during night time it decreases the pH due to decrease in carbonic acid due to uptake of O_2 . According to Al-Busaidi *et al.* (2003), when calcium ions are present in sufficient quantity, CaCO_3 precipitates. As long as calcium dominates the cation exchange complex, rather than sodium, the soil pH is buffered and unlikely to rise above 8.5. The one way ANOVA was carried out to see whether pH differed in experimental ponds. The analysis showed no significant differences $p > 0.05$ in the average pH values of water and

sediment during EC-1 and EC-2 or in between aerated and non-aerated pond in respective culture cycles and remained fairly stable. Fortnight average values of pH in water are depicted in (Table EC 1.3 & Figure EC 1.3) for EC-1 and (Table & Figure EC 2.3) for EC-2. Sediment pH is depicted in (Table & Figure EC1.4) during EC-1 and (Table & Figure EC 2.4) for EC-2.

Experimental Cycle (EC -1)

Mean water pH remained to be 7.67 ± 0.41 with minimum of 7.09 ± 0.06 on 45 DoC and maximum 8.32 ± 0.04 on 105 DoC for aerated pond (Table ST7). In non aerated pond pH was 7.72 ± 0.07 on 0 DoC and highest 8.50 ± 0.02 on 105 DoC (Table ST8). For creek at the start of the culture period pH of 7.74 ± 0.21 and lowest 7.11 ± 0.08 on 105 DoC was observed.

The sediment pH in aerated pond was found to be better than non-aerated pond with mean value of 7.01 ± 0.58 and 6.50 ± 1.13 respectively during entire culture period (Table ST8). pH in the sediment ranged from lowest 7.02 ± 0.85 on 0 DoC to 7.62 ± 0.07 on 90 DoC for aerated pond (Table ST7). For non-aerated pond it ranged from 6.48 ± 0.16 on the 0 DoC to 7.47 ± 0.29 on 90 DoC. Creek also recorded lower sediment pH values initially ranging from 4.74 ± 0.98 to highest of 7.61 ± 0.08 during the culture period with more profound variations (Table ST9).

Regression analysis for Aerated pond (EC-1)

Backward multiple regression analysis carried out, showed that, during EC-1, in aerated pond, pH was linearly related with that of independent variables, such as temperature ($B=-0.969$), $\text{NO}_2\text{-N}$ ($B=-2.61$), $\text{NH}_3\text{-N}$ ($B=1.64$), $\text{NO}_3\text{-N}$ ($B=1.39$) and chlorophyll *a* ($B=0.94$) in water ($B=-1.63$) E_h of water explained 92.2% variations $F_{6,2}=16.76; p=0.05$. Dalkiran *et al.* (2006) showed the interdependence of pH and the nitrification processes, where, he has ascertained that, at lower pH nitrification rate is slower, and it increases with the increasing or alkaline pH.

In sediment, pH showed highly significant linear relationship, between temperature (B=-0.81) pH water (B=-0.32), E_h in water (B=-0.55), chlorophyll *a* in sediment (B=0.089), phaeophytin in the sediment (B=0.37), $\text{NO}_3\text{-N}$ (B=-0.97), POC and $\text{NH}_3\text{-N}$ (B=0.28). The model explained 99.9 % variance due to variations in these parameters, at $F_{7,1}=9979.2$; $p=0.002$. From beta values, nitrate reduction and temperature seems to have more profound effect on the variations of pH in the sediment.

Regression analysis for Non-aerated pond (EC-1)

For non-aerated pond, stepwise multiple regression analysis showed that, phaeophytin in the water alone (B=0.89) was significantly responsible for the changes in the pH in the water, accounting 77.6%, with $F_{1,7}=28.76$; $p=0.001$. The decrease in phaeophytin in the water on 60 DoC (Figure EC1.19) and simultaneous decrease in the pH of water (EC1.3) on same DoC can be seen. pH is known to affect the phytoplankton growth. Chein and Durbin (1994) has showed that, the changes in the pH levels in marine systems correlates with the changes in the temperature, DO concentration and thus phytoplankton production is affected.

In the sediment, pH seems to have been affected by the changes in pH water (B=0.115), E_h of water (B=-1.00), E_h in the sediment (B=-0.17), H_2S in the water (B=0.19), $\text{NO}_2\text{-N}$ (B=0.33), POC (B=0.65), and temperature (B=-1.22). On backward regression analysis, these variables explained 99.9% variance in sediment E_h with overall significant model $F_{7,1}=2378.19$; $p=0.016$.

Regression analysis for Creek (EC-1)

For the creek, backward multiple regression analysis showed significant linear relationship between, pH of water and salinity (B=1.23), BOD (B=-0.36), E_h of water (B=-0.38), $\text{NO}_3\text{-N}$ (B=-0.97), $\text{NH}_3\text{-N}$ (B=0.057) and $\text{PO}_4\text{-P}$ (B=-0.75). The model explained 99.9% variability in water pH due to changes in these parameters.

The relationship was found to be significant $F_{6,2}=1762.6;p=0.001$. Among these parameters $PO_4\text{-P}$, $NO_3\text{-N}$ and salinity respectively seems to have more profound effect on the pH of water in the creek. From this, it can be said that, in the creek nitrification and denitrification through oxidative and reductive pathways have altered E_h , which have further played role for changes in the pH in creek waters. Nitrification rates as the function of pH for seawater samples taken from both the euphotic and aphotic zone are studied by, Huesemann *et al.* (1999). They have sited that, in euphotic and aphotic zones, the nitrification rates drop drastically with decreasing pH, and it was completely inhibited at pH 6. Further, they showed that, nitrification inhibition occurs at low pH values, and in marine environments ammonium oxidation rates reduces as the pH drops from 7.8 to 7.3. In case of ammonia (NH_3) to ammonium (NH_4) is drastically reduced with decreasing pH. In the sediment stepwise regression analysis for pH, showed sediment organic carbon ($B=0.84$) accumulation or oxidative processes, explained 66.1% variance in sediment pH $F_{1,6}=14.63;p=0.009$.

Experimental Cycle (EC -2)

At the start of the culture period water pH was 7.91 ± 0.07 and 7.73 ± 0.05 for aerated and non-aerated pond respectively (Table EC2.3 & Figure EC2.3). The pH of water decreased to 7.2 ± 0.10 for aerated pond and 7.2 ± 0.07 for non-aerated pond on 30 and 45 Day of Culture (DoC) respectively, from initial 7.91 ± 0.07 and 7.73 ± 0.05 on 0 DoC. From 60 DoC it increased till the 105 DoC with highest values of 8.30 ± 0.03 and 8.48 ± 0.30 on 105 DoC for aerated and non-aerated pond respectively (Figure EC2.3). For creek, it showed decreasing trend from 7.63 ± 0.10 till the end of culture period with a minimum value of 6.88 ± 0.04 (Figure EC2.3).

In sediment, pH was observed to be 7.32 ± 0.03 and 7.28 ± 0.03 for aerated and non-aerated pond respectively on 0 DoC. pH of sediment decreased to its lowest on 30 DoC for non-aerated pond, being 6.42 and for aerated pond being 7.10 ± 0.05 on 30 DoC. Variations in sediment pH are depicted in Table (EC2.4) & Figure (EC2.4).

In aerated pond pH fluctuations in the sediment were considerably less, than non-aerated pond. Creek showed less pH values than both the ponds on 30 and 45 DoC being 6.95 ± 0.14 and 6.80 ± 1.33 (Table EC2.4 and Figure EC2.4). In present study the creek showed mean pH of 7.37 ± 0.33 for water. pH was found to be in the range of and 6.88 to 7.72 for water and 6.80 to 7.85 in the sediment (Table ST12).

For the sediment pH it can be seen from the Table and Figure (EC1.4) as well as, Table and Figure (EC2.4) that, it was low only on the first fortnight. In shrimp culture practices, in between two cycles, ponds are drained and dried. The pH value in the sediment of aerated pond being 6.48 ± 0.16 (Table & Figure EC1.4) was possibly due to the slightly acid sulfate nature of the dry compact soil with more Fe^+ , due to intercrop drying practice, which invariably came to its optimal, as the culture period progressed. According to Boyd (1995), dried and compacted grayish streaks or crusts shows a typical, acid-sulphate nature of the soils. The low pH of 3.5 and 4.5 have been reported by Smith *et al.* (1996) in the dry sediments and at the same time adjacent mangrove soils have been reported to show neutral pH 7.0. Overall, the mean pH values for aerated pond, non-aerated pond were 7.6 ± 0.4 and 7.7 ± 0.4 . These optimal pH values (Table ST13) indicate well buffered and well managed pond practices. According to Gire *et al.* (1993), alkaline values of pH 7.5 to 8.5 are well buffered against the sudden large fluctuations. The water quality management practice during current study period included the application of dolomite at the rate of 25-30 kg for 0.6 ha. / Pond every week, and seems to be the reason for optimal alkaline pH levels being maintained in water and sediment during the culture period.

Regression analysis for Aerated pond (EC-2)

The backward multiple regression analysis showed significant linearity between pH of water in aerated pond with that of DO ($B=1.16$), BOD ($b=-0.80$) and COD ($B=-0.49$) variations and model signifies 95% variances, $F_{3,4}=45.46; p=0.02$ in the pH of water due to these variables. Stepwise and backward multiple regression analysis

showed that, pH of water ($B=0.79$) had significant effect of 57.6% on the sediment pH with $F_{1,6}=10.5$; $p=0.018$.

Regression analysis for Non-aerated pond (EC-2)

In non-aerated pond, stepwise multiple regression analysis showed that, pH variation were significantly related with variations in DO ($B=0.30$), BOD ($B=-0.52$), COD ($B=-0.84$), chlorophyll *a* ($B=-0.30$) and phaeophytin in the water ($B=0.83$) and explained 99.2% variations with overall significance of $F_{5,2}=180.76$; $p=0.006$. Amongst these parameters, phaeophytin in the water in terms of degradation product of phytoplankton seems to have more profound effect, on the pH of water. None of the water quality parameters showed to have any significant linear relationship with that of sediment pH in non-aerated pond.

Regression analysis for Creek (EC-2)

Amongst all variables, only temperature ($B=0.99$; $p<0.001$) and $\text{NO}_2\text{-N}$ ($B=0.32$; $p=0.031$) explained the variance of, 92.3%, and was significant at $F_{2,5}=42.8$; $p=0.001$ carried out by stepwise multiple regression analysis. In the sediment OC ($B=0.84$) showed significant relationship $F_{1,6}=14.63$; $p=0.009$ with that of pH in the sediment and explained 66.1% variance in pH sediment alone, due to changes in the sediment organic carbon. Thus, in both the ponds overall, oxidation of organic matter and, diurnal DO changes due to photosynthetic activities, have brought about the changes in the pH. It is known that, degradation of organic matter is better facilitated in alkaline environments than the acidic environments, Avnimelech *et al.* (2003). Therefore, adequate liming application at appropriate intervals in the pond ensures better oxidation of organic matter. During present analysis, overall, no significant linear relationship were observed for changes in pH of the sediment, with any of the water and soil quality parameters during EC-1 and EC-2, with an exception for aerated pond during EC-2, for both the experimental ponds. However, DO in the aerated pond and pH of water seems to be the reason for variations in the sediment pH in the aeration pond.

4.1.4 Oxidation Reduction Potential– E_h (mv.)

Experimental Cycle (EC-1)

The fortnight values of average E_h in water and sediment are depicted in Table & Figure EC1.5 and EC1.6 respectively. The mean E_h of the aerated pond water was 86.29 ± 16.97 SD and for sediment it was found to be 68.20 ± 21.47 mv, respectively (Table ST7). For aerated pond maximum E_h was observed on 105 DoC being 112.77 ± 9.79 mv and lowest 62.8 ± 4.12 mv on 60 DoC (Table EC1.5). The mean E_h of water and sediment for non-aerated pond during EC-1 was observed to be 72.95 ± 16.39 and 59.92 ± 16.30 mv respectively (Table ST8). For non aerated pond, the maximum value of 94.6 ± 4.37 mv and minimum of 54.6 ± 4.63 mv was observed on 0 and 60 DoC, respectively (Table ST8). In sediment, E_h was slightly lower than water for both experimental ponds. In the creek, E_h showed more stable values for both water and sediment with mean 66.72 ± 14.49 for water and 58.16 ± 15.15 mv for sediment (Table ST9). The mean values of water and sediment E_h in experimental ponds and creek for EC-1 are depicted in Table, ST7, ST8 and ST9.

Regression analysis for Aerated Pond (EC-1)

For aerated pond, backward multiple regression analysis carried out showed that, E_h of water is linearly related with that of temperature ($B=-0.26$), DO($B=-0.78$), BOD ($B=-0.51$), pH water ($B=0.18$), $\text{NO}_3\text{-N}$ ($B=0.65$), chlorophyll *a* in the water ($B=-0.25$) H_2S ($B=0.017$) with overall significant model with $F_{7,1};p=0.003;p<0.01$. These, variables in the water seem to perfectly limit E_h values (99.9%) in the aerated pond. In the sediment, E_h was linearly related to the DO ($B=-0.63$), BOD ($B=-0.60$), COD ($B=0.40$), OC ($B=-0.32$) and sediment pH ($B=-0.58$) and this model showed 99.5% variability in E_h of sediment due to changes in these sets of parameter at $F_{5,3}=322.41;p<0.001$.

Regression analysis for Non-aerated Pond (EC-1)

For non-aerated pond, backward multiple regression analysis revealed the highly significant linear relationship, with 92.5% variability, in E_h of water, which is explained by temperature ($B=-1.0$), pH of sediment ($B=-0.82$), POC ($B=0.56$), NO_2-N ($B=0.29$) with significant model at $F_{4,4}=25.62;p=0.004$. For sediment, E_h values were found to show linearity with that of temperature ($B=-2.5$), DO ($B=-2.4$), E_h of water ($B=-0.53$), phaeophytin in sediment ($B=-1.87$) and OC ($B=-1.75$). This model accounted for 98.5% variance in the E_h of sediment, with overall significance of model being $F_{6,2}=86.93;p=0.011$. The possible process here seems to be, the decomposition of organic matter, by aerobic micro-organisms which depletes the oxygen supply, and lowers the redox potential. Once the soil becomes anaerobic, decomposition of organic matter by anaerobes drives the redox potential even more downward (Boyd, 1995). Hence, processes of denitrification in the suboxic conditions might have also prevailed, in the water and in the soil at the bottom of the pond. Also, the oxidation of accumulated organic matter might have been the reason for low E_h values in the soil

Regression analysis for Creek (EC-1)

The E_h in the creek water, as well as sediment was found to show low values in the creek than in ponds. Mean E_h was found to be in the range of 86.29 ± 16.97 and 99.0 ± 29.35 mV. in the water for EC-1 and EC-2 respectively. For soil, mean E_h was found to be 68.2 ± 21.47 and 103.04 ± 16.84 mv during EC-1 and EC-2 respectively. These values are, indicative of slightly reducing environments. The moderately low E_h values shows potential shift to reducing environments, due to suboxic conditions prevailing mostly at the bottom of the pond. This can be due to the sampling time or can be due to the sustained sub-oxic conditions prevailing during the night time, as the sampling was carried out in the morning hours, extending from dawn till after, the sunrise.

During both the experimental cycles, E_h values overall ranging from 24.97 to 145.07 mv with DO ranging from 3 to 5 mg.l^{-1} were observed. Similar results are reported by Shimabukuro *et al.* (2004). According to Smolders *et al.* (2006), in anaerobic oxidation processes, the consumption sequence of oxidants depends largely on their relative oxidation strengths and the anaerobic respiration pathways are not entirely mutually exclusive, further, different pathways of oxidation normally show considerable overlap. Hence, during the anoxic conditions denitrification is the first oxidative process where available NO_3 is reduced to form NO . This is followed by the sequence of reactions which involve Fe, Mn, SO_4 and CO_2 as an electron acceptor. This gives rise to reducing processes which impart acidity to such environments. Approximate redox potential indicating oxidative and reductive systems measured in milli-volts (mv.) have been described by Avnimelech *et al.* (2003).

Experimental cycle (EC-2)

The fortnight values of average E_h in water and sediment are depicted in Table & Figure (EC2.5) and (EC2.6). The E_h of water showed increasing trend with the progressing culture period. Overall, aerated pond showed higher E_h values than non-aerated pond and creek (Figure EC2.5 & Figure EC2.6). It showed cyclic variations with sharp decrease on 15 & 45 DoC followed by increase on 105 DoC, depicted in Table & Figure (EC2.5). It was found to be highest in water on 75 DoC for aerated pond being 145.07 ± 6.93 mv and for non-aerated pond 140.47 ± 11.62 mv and 133.45 ± 5.96 mv for creek (Figure EC2.5 & Figure EC2.6).

Similar trend of cyclic minimum and maximum E_h was observed for sediment with minimum values on 15 DoC and 45 DoC till 75 DoC showed in the Figure EC2.6. The E_h values for aerated pond in water were low as compared to sediment. For aerated pond minimum E_h was found to be 78.20 ± 2.06 on 45 DoC.

24.97±5.48 mv for non-aerated pond and 42.45±0.79 mv for creek on 15 DoC. Maximum E_h for Aerated pond was 128.87±2.05 and for non-aerated was 126.60±3.95. Further, for creek it was found to be 124.05±4.24 on 75 DoC in the sediment. The mean values of water and sediment E_h in experimental ponds and creek for EC-2 are depicted in Tables ST10, ST11 and ST12. The one way ANOVA carried out showed, no significant differences $p>0.05$ in the average E_h values during EC-1 and EC-2 in aerated and non aerated pond.

Regression analysis for Aerated Pond (EC-2)

The stepwise and backward multiple regression analysis showed that E_h in the water fluctuated due to variations in BOD ($B=0.75$) alone 49.1 %, $F_{1,6};p=0.032$. The backward model explained 73.7 % of variations due to parameters such as temperature ($B=-1.57$), pH of water ($B=-0.97$) and H_2S ($B=-0.88$) concentration in the water $p\leq 0.040$. Thus, higher E_h values in the aeration pond indicate better oxygen availability for the oxidative processes in the pond. In the sediment stepwise multiple regression analysis showed linearity in between E_h of sediment and E_h of water, pH of water, POC and OC $F_{4,3}=240.6;p<0.001$. The adjusted R^2 of 99.3 % indicates best model.

Regression analysis for Non-aerated Pond (EC-2)

Backward multiple regression analysis showed no significant linear relationship between any of the water quality parameters. However, the effect of water quality variables like temperature, E_h of water and H_2S dissolution in the water in turn, seems to have significant effect on E_h of the sediment. The regression model showed 98 % variance explained with overall significant model at $F_{3,4}=117;p<0.001$.

Regression analysis for Creek (EC-2)

In the creek E_h of water and sediment were found to influence each other. Stepwise multiple regression analysis for E_h in water and soil was carried out. The analysis showed 99.3% variability due to chlorophyll a in sediment, E_h of sediment and pH of

water at $F_{3,4}=319.1$; $p<0.001$. Wherein, for sediment the regression analysis showed, 95.9% variance $F=_{1,6};p<0.001$ with similar parameters. This indicates that, both the oxidation and reduction process prevailed, possibly due to the turbulent nature of the creek, in rainy season.

4.1.5 Dissolved Oxygen (DO.mg.l⁻¹)

The fortnight values of average DO concentrations in water are depicted in Table & Figure (EC1.7) as well as Table & Figure (EC2.7).

Experimental Cycle (EC-1)

During EC-1 highest DO value of 4.06 ± 0.23 and lowest of 2.93 ± 0.23 mg.l⁻¹ was registered on 60 and 105 DoC respectively (Table & Figure, EC1.7). For Non aerated pond DO concentrations were found to be in the range of 2.86 ± 0.06 to 4.81 ± 0.13 mg.l⁻¹ (Table & Figure, EC2.7). Conspicuously, maximum DO concentrations were observed for both the ponds as well as for creek from 30 DoC to 75 D0C.

Regression analysis for Aerated pond (EC-1)

In aerated pond among all parameters variations in DO showed highly significant relationship with that of phaeophytin in water, pH of water and POC at $F_{1,7}=28.54$ $p\leq 0.01$. These variables found to explain 91.2% of variability in the DO concentration in aerated pond. This indicates that, phytoplankton degradation was possibly prevalent in aerated pond, which further contributed to increase the POC (Table & Figure, EC1.10). The degradation of chlorophyll to from phaeophytin and ultimately, degradation and oxidative processes in phtoplankton might gave rise to form higher POC ($B=0.31;p=0.042;p<0.05$), phaeophytin ($B=-1.2;p<0.001$) concentrations towards the end of the culture period and can be attributed to the, changes of DO concentration in aerated pond.

Regression analysis for Non-aeration pond (EC-1)

In non-aeration pond, stepwise multiple regression analysis carried out showed best linear relationship between DO and chlorophyll *a* in the water at $F_{1,7}=14.87;p<0.01$. The adjusted R^2 explained the 63.4 % variability of DO is due to the changes in the chlorophyll *a*. The backward multiple regression carried out however, showed an adjusted R^2 , that explained 99.0% variations in DO were due to temperature, $\text{NO}_2\text{-N}$, phaeophytin in water and sediment as well as meiofauna respiration at $F_{5,3}=155.59;p<0.001$.

The lower chlorophyll *a* values of water were observed on 30 DoC to 75 DoC (Table & Figure EC1.17) and at same time slightly elevated DO increase during same period (Table & Figure EC1.7) as well as trend line pattern observation indicates, stable DO and chlorophyll *a* concentrations overall. Therefore, in non-aerated pond the DO changes can be explained in view of the natural diffusion of oxygen through the atmosphere, whenever lower DO concentration occurred. According to Boyd (1998), the net transfer of dissolved oxygen to the water from air through natural gaseous exchanges occurs, when, DO concentration of waters falls below atmospheric DO concentration.

Regression analysis for Creek (EC-1)

For the creek during EC-1 the backward multiple regression analysis showed that, BOD and salinity together were significantly linear to that of DO at $F=2.6;p=0.005;p<0.01$. The analysis explained 76.7% variability due to changes in BOD and salinity. The low salinity values as explained earlier are due to the influx of fresh water due to rain (Figure EC1.2). The rain water flushing the creeks, did not lead to critical overloading of organic matter, which was transported to sea, and decreasing salinity might have induced the stress on the microbial activity. Therefore with less accumulation of organic matter, and salinity stress might have hindered the microbial activity thereby reducing the BOD values, seems to have influenced the DO regime in the creek.

Experimental Cycle (EC-2)

DO concentrations were marginally higher in aerated pond than non-aerated pond. It ranged from $2.67 \pm 0.07 \text{ mg.l}^{-1}$ to $4.64 \pm 0.03 \text{ mg.l}^{-1}$ in aerated pond and 2.7 ± 0.10 to 4.21 ± 0.03 for non-aerated pond (Table ST7 and Table ST8). In the creek concentration of DO were found to be 2.29 ± 0.09 and $4.68 \pm 0.04 \text{ mg.l}^{-1}$ (Table ST9). One-way ANOVA showed that, there were no significant differences in the average dissolved oxygen concentration ($p > 0.05$) in between two cycles EC-1 and EC-2 at fortnight intervals. The minimum and maximum mean DO values overall, during EC-1 and EC-2 for both the ponds were in the range of 3.36 ± 0.45 to $3.8 \pm 0.63 \text{ mg.l}^{-1}$, wherein, for creek it was in the range of 3.73 ± 0.55 to $3.66 \pm 0.84 \text{ mg.l}^{-1}$. One-way ANOVA carried out confirmed that, overall, there were no significant differences in the DO concentrations in aerated and non aerated ponds, but different process responsible for DO variations were understood during both the experimental cycles and are analyzed as described below in the form of regression analysis.

Regression analysis for Aerated Pond (EC-1)

In aeration pond stepwise multiple regression analysis showed highly significant relationship of DO with $\text{NO}_3\text{-N}$, and temperature $F_{2,5}=50.80; p < 0.001$. This model accounted for 93.4 % variance in the DO. The temperature showed decreasing trend (Table & Figure EC2.1). $\text{NO}_3\text{-N}$ was almost stable initially, and increased after 45 DoC (Table & Figure EC2.13) The backward regression analysis, showed better model at $F_{3,4}=64.40; p = 0.001$ explaining 96.5% variability of DO due to changes in BOD, COD and pH of water. From this, it can be understood that, in the aeration pond nitrogenous BOD and degradation of suspended organic matter at higher pH seems to have controlled the DO regime in the aerated pond.

Regression analysis for Non-aerated Pond (EC-2)

Stepwise multiple regression analysis showed highly significant relationship between DO and salinity, $\text{NO}_3\text{-N}$, chlorophyll *a* water and H_2S in the water ($F_{4,3}=1617.3; p < 0.001$). Comparatively initial lower chlorophyll *a* (Figure EC2.17)

and higher $\text{NO}_3\text{-N}$ (Table & Figure EC2.13) indicates similar processes, as that of aeration pond. Here, $\text{NO}_3\text{-N}$ probably was not utilized by the phytoplankton as source of nutrient entirely, further, possibly non utilization of $\text{NO}_3\text{-N}$ have showed higher ambient concentrations of $\text{NO}_3\text{-N}$ in the water. The parameters considered in regression analysis of non-aerated pond during EC-2 thus, explained 99.9% variability in the DO.

Regression analysis for Creek Data (EC-2)

In the creek during EC-2 the backward multiple regression analysis revealed significant linear relationship between DO with that of BOD, E_h of water, $\text{NH}_3\text{-N}$ and H_2S in the water $F_{4,3}=12.72;p<0.05$. Both DO and BOD concentration showed increasing trend during study period. Despite increasing BOD (Table & Figure EC2.10) DO (Table & Figure EC2.9) did not depleted. From this it can be understood that, DO replenishment was better in the creek, due to the turbulent nature of the creek, due to fresh water runoff during rainy season. The lack of fortnight and seasonal differences during the study period can be attributed to diurnal changes in DO and time of sampling, varying from morning 6:30 am to 10:00 am. This type of behavior of DO has also been showed by Guerrero *et al.* (1999) for the tendency of dissolved oxygen decrease in the morning hours during the culture period. The tendencies of DO concentrations can be explained by two factors, first is the level of dissolved oxygen at dawn decline as the growing season progresses, due to the DO requirement, by cultured species, further, feeding rate also increases, which also demands higher DO due to decomposition of uneaten accumulated feed at the bottom and periphery of the pond. Second reason is the seasonality, and water temperature, which affects the dissolution of oxygen. In present scenario however, cloud cover in rainy season leading to lower photosynthesis, and this in turn, lowers the DO concentration in the water. Variation in DO during the culture periods with the lowest concentration were observed towards the end of the culture period and have been reported by Cordava *et al.* (1998), in low water exchange pond farming *Penaeus vennamei*. According to Cordava *et al.* (1998), ponds with aeration rates

being 0 to 6 hrs.day⁻¹, morning DO at 6 am were recorded to be very low and ranged from 1.76 to 2.22 mg.l⁻¹. Even with, 0 to 24 hrs of aeration rates per day, values of DO towards the end of culture period at morning as low as 1 mg.l⁻¹ has been reported by him. During present study conspicuously, dissolved oxygen concentrations never fell below 2.0 mg.l⁻¹, which is considered as the hypoxic and critical concentration for shrimp culture practices (Allan & Maguire, 1991). Along with this, there were no signs of distress, such as, shrimp coming to surface or at the banks for lack of DO, was not observed, and there were no mortalities recorded at least due to lack of oxygen. The reason behind this possibly is that, these low DO levels remained so, only for short period of time and showed diurnal DO fluctuations, which compensated for low DO time periods. Thus, shrimp were not subjected to continuous low DO levels. In the same study diurnal variations of DO measured by continuous logger probes for DO pH etc has been described by Sreepada *et.al* (2005) indicate clear cut diurnal variations in DO.

As described above DO showed no significant fortnight variations, but ANOVA analysis showed overall highly significant effect $p < 0.01$ of dissolved oxygen on the growth of the shrimp. DO along with temperature is known to alter the metabolic rates of the shrimp. Temperature is one of the major physical factors influencing the metabolic rate in 'ectotherms' like shrimp and these metabolic rates generally increases with the increase in temperature (Chen & Lai, 1993; Chain & Nan, 1993; Villareal & Ocampo, 1993; Villeral & Rivera, 1993). Most of the penaid shrimp are also able to tolerate the salinities between 1 to 40 PSU but 15-25 PSU is considered as the optimal salinity for culture (Ponce-Palafore *et al.* 1997). According to Rosas *et al.* (1999) salinity and DO in combination affects the metabolic rate and observed that at 15 PSU, ammonia excretion was greater than higher in 35 PSU. In present study therefore, though DO levels in the range described above seems to be sufficient at least in the modified extensive shrimp culture practices in the following evidence to the DO requirement of intensive shrimp culture practices.

DO value higher than 5mg.l^{-1} have been generally recommended for intensive shrimp culture practices (Cheng *et al.* 2003). However, low dissolved oxygen has been reported in the extensive shrimp culture ponds by Alongi *et al.* (1999). Thus, during present study oxygen was never depleted to lethal levels during the culture period. Further, according to Herried *et al.* (1980), the mechanism involved in response to hypoxia depends on the environment and the physiological state of the animal. Also, during the study there was difference in the stocking densities being higher 12 and 10 pl.m^{-2} in aerated pond and 8 and 5 pl.m^{-2} in non-aerated pond during EC-1 and EC-2 respectively. Considering this difference in stocking density aeration seems to have helped in sustaining higher stocking densities, thereby keeping same oxygen levels as that of the non-aerated pond and natural environment that is creek.

4.1.6 Biochemical oxygen demand (mg.l^{-1})

The fortnight variations of average BOD in water are depicted in Table and Figure EC1.8, for EC-1 and Table and Figure EC2.8 for EC-2. The mean BOD values for respective experimental cycles EC-1 and EC-2 and associated aeration and non-aeration pond and creeks are given in Table ST7 to ST12.

Experimental Cycle (EC-1)

During EC-1 BOD was found to be the maximum of $3.05\pm 0.23\text{ mg.l}^{-1}$ on 0 DoC and minimum 0.30 ± 0.06 on 75 DoC in aerated pond. In non-aerated pond initially values were found to be much constant till 60 DoC (Figure EC1.8). The highest BOD value of 3.73 ± 0.11 and lowest of $1.16\pm 0.37\text{ mg.l}^{-1}$ on 120 DoC. After 60 DoC more cyclic variations were observed in aerated pond till the end of the culture period.

Regression analysis for Aerated Pond

The backward multiple regression analysis for BOD showed that, POC ($B=-3.86$) $\text{NO}_2\text{-N}$ ($B=0.94$), DO ($B=-0.19$) and $\text{NO}_3\text{-N}$ ($B=-0.29$) all together accounted for 95.3% variance in the BOD concentrations and model was significant at

$F_{4,4}=41.60;p=0.002$. This indicates that, intermediate $\text{NO}_2\text{-N}$ oxidation and bacterial degradation of suspended particulate matter was prevalent.

Regression analysis for Non-aerated Pond

In non aerated pond when backward multiple regression analysis was carried out, the linear relationship model accounted for 69.7% of variance in the BOD due to the $\text{NH}_3\text{-N}$ ($B=-0.85$), $F_{1,7}=19.43;p=0.003$. Thus, the oxidation of $\text{NH}_3\text{-N}$ by nitrifying bacteria might have contributed to the higher BOD values (Table & Figure. EC1.8).

Regression analysis for Creek

For the creek, backward multiple regression analysis showed significant linear relationship between BOD and DO, OC, salinity, pH water, E_H water and $\text{NO}_2\text{-N}$, explained 99.9 % variations, with overall significant model at $F_{7,1}=1538.83;p=0.020$. Amongst all variables DO ($B=1.49$), OC ($B=1.19$), salinity ($B=0.9$), pH ($B=0.7$). From this it can be said that, oxidation of organic carbon along with decreasing salinities contributed for the variations in BOD. Hence it can be said that, decreasing salinities might have exerted stress on the normal microbial flora in the creek, which may increase the oxygen demand.

Experimental cycle (EC-2)

Overall, BOD showed an increasing trend from start till the end of the culture period. BOD on day 0 DOC was 0.98 ± 0.03 and $1.20\pm 0.07 \text{ mg.l}^{-1}$ in aerated and non-aerated pond, respectively (Table & Figure EC2.8). It showed gradual increase till 75 DoC and on 90 DoC and decreased to $0.88\pm 0.16 \text{ mg.l}^{-1}$ for aerated pond (Table & Figure EC2.8). In the creek, BOD values showed more cyclic variations with higher BOD values on 30 and 75 DoC being 2.31 ± 0.08 and $2.93\pm 0.17 \text{ mg.l}^{-1}$ respectively (Table & Figure EC2.8).

Regression analysis for Aerated pond

In the aerated pond, stepwise multiple regression analysis model accounted for 98.5% variability in BOD, due to temperature ($B=0.571$), E_h water ($B=0.711$), NO_2-N ($B=0.644$), and NH_3-N ($B=0.439$) at $F_{4,3}=113.84$; $p=0.001$. This signifies that, nitrogenous biological oxygen demand by nitrifying bacteria to convert ammonia to more stable nitrates contributed for the variations in BOD.

Regression analysis for Non-aerated Pond

In the aerated pond backward multiple regression analysis model accounted for 97.2% variability in BOD due to temperature ($B=-0.362$), COD ($B=-0.725$), E_h sediment ($B=-0.242$), NH_3-N ($B=-0.281$), DO ($B=-0.368$) and overall significance of $F_{5,2}=48.77$; $p=0.020$. The, oxidation of NH_3-N by *nitrobacter* sp. might have been responsible for the oxygen demand in the water in non aerated pond (Boyd,1995).

Regression analysis for Creek

In the creek, backward multiple regression analysis, where model accounted for 100% variability due to changes in POC ($B=1.12$), OC ($B=-0.80$), NO_3-N ($B=-0.55$), E_h of sediment ($B=0.60$). Amongst these, oxidation of suspended and particulate organic matter possibly dominated the scenario, for variations in BOD in the creek.

4.2.7 Chemical Oxygen Demand ($mg.l^{-1}$)

The fortnight average variations of COD in water are showed, in Table and Figure, EC1.9, and Table and Figure EC2.9 for EC-1 and EC-2 respectively.

Experimental Cycle (EC-1)

In aerated pond, COD ranged from minimum of 10.01 ± 1.53 to maximum 87.00 ± 7.72 $mg.l^{-1}$, wherein, in non aerated pond it ranged from, 14.01 ± 6.51 to 93.01 ± 12.89 $mg.l^{-1}$ variations in COD are presented in Table ST7 and ST8. In aerated pond, initially it was high but on 30 DoC, it reduced drastically to 10.01 ± 1.53 $mg.l^{-1}$ (Table & Figure EC1.9). From the 45 DoC, COD reduced gradually till the end of the culture (Table & Figure EC1.9). Similarly, non aerated pond also showed decrease in COD values

but with more variations, and in general showed higher COD values than aerated pond (Table & Figure EC1.9).

Regression analysis for Aerated Pond

Backward multiple regression analysis showed highly significant relationship explaining 99.5% variance in COD due to $\text{NH}_3\text{-N}$ ($B=-0.83;p=0.001$), $\text{NO}_2\text{-N}$ ($B=0.17;p=0.003$), $\text{NO}_3\text{-N}$ ($B=0.20;p=0.002$), $\text{PO}_4\text{-P}$ ($B=1.43;p<0.001$), POC ($B=0.038;p=0.011$), OC ($B=-0.883;p=0.001$) and H_2S ($B=-0.068;p=0.005$). These relationships indicate that, oxidation of the particulate and sediment organic matter and ammonia was carried out in the water and sediment effectively in presence of DO.

Regression analysis for Non-aerated Pond

For non aerated pond stepwise multiple regression analysis model accounted for 66.4% variability in COD due to $\text{PO}_4\text{-P}$ ($B=0.69;p=0.015$) and OC ($B=-0.506;p=0.048$) with overall significant linear relationship at $F_{2,6}=8.89;p=0.016$.

Regression analysis for Creek

In the creek, stepwise multiple regression analysis model accounted for 97.9% variance in COD $F_{5,3}=75.63;p=0.002$ due to POC ($B=0.398;p=0.06$), DO ($B=-0.441;p=0.006$), $\text{NH}_3\text{-N}$ ($B=-0.408;p=0.039$), $\text{NO}_2\text{-N}$ ($B=0.953;p=0.004$) and $\text{PO}_4\text{-P}$ ($B=0.285;p=0.19$). Here, oxidation of ammonia to nitrite possibly is explained by the utilization of DO and thus COD variations in the creek.

Experimental Cycle (EC-2)

COD increased gradually till 30 DoC and values were highest 137.45 ± 12.82 for aerated pond (Table ST10), and $166.04\pm 8.66 \text{ mg.l}^{-1}$ for non-aerated pond (Table ST11) and $169.60\pm 2.76 \text{ mg.l}^{-1}$, for creek (Table ST12). Thereafter, it decreased to its minimum value of 16.01 ± 1.54 and 14.22 ± 1.30 on 75 DoC for aerated, non-aerated pond respectively (Table & Figure EC2.9). On the last 105 DoC it again increased to

110.76±2.57 and 114.20±2.09 for aerated and non-aerated pond respectively (Table & Figure EC2.9).

Regression analysis for Aerated Pond

In the aerated pond backward multiple regression analysis model accounted for 93.7% variance in COD due to oxidation of NH₃-N, degradation products of phytoplankton (B=1.04;p=0.003) and suspended organic matter, mineralized as sulfide and then oxidized to form sulfate in the water at F_{4,3}=26.85;p=0.011. Oxidation of H₂S (B=0.564;p=0.21) and NH₃-N (B=-0.784;p=0.006) form sulfuric acids (Boyd,1995) and further, NO₂-N (B=0.730;p=0.007) consumed oxygen in aerated pond and can be the main reason for the variations in the COD, in aeration pond.

Regression analysis for Non-Aerated Pond

For non aeration pond chlorophyll *a* in the water (B=-0.616), phaeophytin in the sediment (B=0.531) and microphytobenthos (B=-0.692) showed significant linear relationship with that of COD F_{3,4}=6.92;p=0.046 explaining 71.7% variance due to these variables on the COD. Thus, variations in the COD points towards the probable process that, degradation of microphytobenthic diatoms and settling phytoplankton might have prevailed at the bottom of the non-aerated pond.

Regression analysis for Creek

In the creek stepwise multiple regression analysis model accounted for 99.9% variance in COD due to chlorophyll *a* (B=0.067) and phaeophytin in the sediment (B=0.652;p=0.002), NO₃-N (B=-0.352) and NH₃-N, as well as temperature and macrofauna (B=0.665;p=0.002) with overall significance of F_{1,6}=74649.33;p=0.003. This analysis points to the possible process that, dead macrobenthic biomass, phytoplankton and diatoms may have contributed to the organic matter and trend is much similar to non aeration pond. The degradation of this organic matter possibly leads to the variations in the COD in creek.

Comparison of BOD and COD

Mean BOD values during EC-1 for aerated pond were 2.00 ± 1.09 (Table ST7) and 2.64 ± 0.73 mg.l^{-1} for non-aerated pond (Table ST8). During EC-2 for aerated pond it showed mean value of 1.35 ± 0.43 (ST10) and for non-aerated pond it was 1.48 ± 0.54 mg.l^{-1} (Table ST11). The mean COD values during EC-1 were 41.15 ± 26.62 mg.l^{-1} for aerated pond (Table ST7) wherein, for non-aerated pond it was 43.09 ± 31.87 mg.l^{-1} (Table ST8). The mean COD values during EC-2 aerated pond were found to be 88.13 ± 43.44 (Table ST10) and for non-aerated pond it was 98.58 ± 52.49 (ST11). From the mean BOD and COD values it can be said that, BOD and COD values were in the optimum limits during both culture periods in aerated, non-aerated ponds and creek as well. Their permissible limits for aquaculture practices are given in Table ST13. Further, there were significant differences ($p < 0.05$) in the mean COD concentrations in aerated and non-aerated pond during EC-1 and EC-2. However, overall, BOD values unlike COD values did not differ for EC-1 and EC-2, neither it significantly differed $p > 0.05$ fortnightly during EC-1, except for EC-2 $p < 0.05$. Overall, it was observed that BOD (Table & Figure EC1.8) and COD (Table & Figure EC1.9) values of aerated ponds were marginally lower than non-aerated pond with an exception of COD values, overall, slightly higher after 60 DoC in aerated pond than non-aerated pond during EC-1 (Figure EC1.8). During EC-1 COD values were lower than EC-2. Further, contradictory to the COD values the BOD values were higher during EC-1 and lower during EC-2. From this it can be said that during EC-1 biological oxidation, while during EC-2 chemical oxidation were prevalent. Therefore optimal BOD and COD is an indication that there were no high organic loads from these systems.

4.1.7 Particulate Organic Carbon (POC. mg C.l^{-1})

Variations in POC in experimental ponds and creek for EC-1 are showed in Table and Figure EC1.10. For EC-2 variations in POC are presented in Table and Figure EC2.10. An approach to the POC composition with that of C:H:N analysis has been

given by Ribs et.al.(1999) shows that POC comprise of live and dead organic carbon in detrital form and live organic carbon in the form of chlorophyll *a*.

Experimental Cycle (EC-1)

During EC-1, POC from initial low $3.41 \pm 0.55 \text{ mg.C.l}^{-1}$ showed increasing trend in aerated pond till 90 DoC with maximum value of $18.63 \pm 17.32 \text{ mg.C.l}^{-1}$ was observed on 105 DoC (Table & Figure EC1.10). In non-aerated pond more fluctuations in the POC values were observed than in aerated pond (Table & Figure EC1.10) with higher values on 30 and 90 DoC being 11.72 ± 2.28 and $12.56 \pm 3.47 \text{ mg.C.l}^{-1}$ respectively. In creek POC values were marginally lower than aerated and non aerated pond and are depicted in Table & Figure EC1.10. POC, ranged from 3.51 ± 1.36 on 0 DoC to $9.15 \pm 4.01 \text{ mg.C.l}^{-1}$ on next fortnight (Table ST9).

Regression analysis for Aerated Pond

As per the study by Danovaro *et al.* (1997), organic carbon of detrital origin represented the main fraction of POC. Hence, backward multiple regression analysis was used to determine the possible source of particulate matter by observing linear relationship. Chlorophyll-*a* ($B=1.12$) in water and chlorophyll *a* ($B=-0.750$) in sediment was regressed with respect to POC. The model generated, explained 87.8% variance in POC due to chlorophyll *a* in the water and sediment at ($F_{2,6}=29.80; p=0.01$).

Regression analysis for Non-aerated Pond

Backward multiple regression analysis carried out explained 99.9% variance due to temperature ($B=1.67$), chlorophyll *a* in water ($B=0.267$) and sediment ($B=-1.08$), phaeophytin in water ($B=0.63$) and sediment ($B=0.62$), as well as salinity ($B=-1.02$) and pilleted feed ($B=0.53$), With the model being significant at $F_{7,1}=781.11; p=0.028$.

Regression analysis for Creek

In the creek, parameters like chlorophyll *a* in water ($B=1.33$) and sediment ($B=-1.95$), phaeophytin ($B=0.73$) in water, DO ($B=1.21$), and E_h of water ($B=-1.12$)

explained 97.2% variability in POC, and was significant at $p \leq 0.01$, $F_{5,3} = 55.91$; $p = 0.004$. This explains that, apart from chlorophyll *a* and phaeopigment in water, and the turbulent nature of creek might have suspended the sediment bound phytoplankton in the water which on oxidative degradation contributed for the POC variations.

Experimental Cycle (EC-2)

In aerated pond POC ranged from 4.14 ± 0.07 to 11.81 ± 0.32 mg.C.l⁻¹ (Table ST10). In non-aerated pond it was found to be in the range of 2.21 ± 0.08 to 10.79 ± 0.17 mg.C.l⁻¹ (Table ST11). Overall aerated pond showed slightly higher values than non-aerated pond. Highest POC values were observed on 45 DoC 11.81 ± 0.32 and 10.79 ± 0.17 mg.C.l⁻¹ for aerated and non-aerated pond, respectively (Table & Figure EC2.10).

Regression analysis for Aerated Pond

Stepwise regression analysis did not revealed any significant relationships of POC with mainly chlorophyll *a* of either water or sediment, which are considered to have contributed to POC in the form of live or dead phytoplankton. Backward regression analysis however, showed significant relationship, with parameters other than chlorophyll *a* and feed applied in the pond. Backward regression analysis was also not indicative of direct relationship of other parameters to form POC in aerated pond.

Regression analysis for Non-aerated Pond

Stepwise multiple regression analysis carried out showed that, phaeophytin in the water and sediment NO₃-N, NO₂-N, feed applied and BOD showed overall significant linear relationship and explained 99.9% variance due to these parameters in the formation of particulate organic carbon. Thus amongst these parameters NO₂-N ($B = -1.73$; $p = 0.001$), BOD ($B = 0.440$; $p = 0.02$) and feed ($B = 0.061$) point out towards the process that, the accumulated feed and dead phytoplankton contributed to form the particulate organic matter. In the presence of oxygen the suspended organic matter was oxidized and thus by mineralization of this organic matter lead to form

the NH_3 in the water, which immediately got oxidized to form the $\text{NO}_3\text{-N}$ by nitrifying bacteria. The process is explained by Boyd (1995) here indicates that natural cycling of nutrient and degradative processes were perfectly balanced.

Regression analysis for Creek

In the creek backward multiple regression analysis explained 99.9% variance due phaeophytin in water ($B=0.67$), OC ($B=1.09$), COD($B=-0.57$), E_h of water ($B=-0.21$) and DO($B=0.49$), $\text{NO}_2\text{-N}$ ($B=-0.63$) showed significant linear relationship with that of POC $p<0.05$. Thus, POC could have been originated through degradation processes, through biological or chemical oxidation of dead phytoplankton in the water. Sreepada *et al.* (1996) has studied the particulate organic matter and its constituent protein and carbohydrates. They have sited that, in general, there is high degree of co-relation between phytoplankton biomass and concentration of particulate compounds also during persistent blooming, the contribution of phytoplankton carbon do not exceed 50% of the total particulate organic carbon as large quantities of detrital matter are associated with phytoplankton.

4.1.8 Sediment organic carbon (OC %)

Experimental Cycle (EC-1)

In pond sediment, OC was low initially, being $2.26\pm 1.25\%$ and $1.01\pm 1.28\%$ in aerated and non aerated pond respectively with more high values in the creek $3.05\pm 1.27\%$ (Table & Figure EC1.11). On 30 DoC highest average OC values were observed for both the ponds and creek as well, with values 9.24 ± 0.28 , 8.46 ± 0.40 and $9.72\pm 0.59\%$ for aerated pond, non-aerated pond and creek respectively, depicted in Table & Figure EC1.11. Thereafter, there was sharp decrease in OC at next fortnight and remained almost constant with range of values in between 1 to 2 % till 105 DoC in both the ponds. At 120 DoC it was found to be higher again (Table & Figure EC1.11).

Regression analysis for Aerated Pond

In aeration pond backward multiple regression analysis explained 99.9 % variance due to changes in chlorophyll *a* in water ($B=0.334$) and sediment ($B=-0.114$), phaeophytin ($B=-0.47$) in sediment, DO ($B=-0.27$), COD ($B=-0.81$) and temperature ($B=0.99$) for OC. The overall linear relationship amongst these parameters was significant at $F_{6,2}=1083.4;p=0.001$. Amongst these parameters, chlorophyll-*a* in sediment and COD might have formed OC in the aerated pond.

Regression analysis for Non-aerated Pond

In non aerated pond, feed ($B=-1.18;p=0.024$) and COD ($B=-2.15;p=0.57$) were found to explain variations in OC. The linear relationship between feed, chlorophyll *a* in water ($B=0.69$), phaeophytin in sediment ($B=-0.43$), pH of sediment ($B=1.12$) and temperature ($B=-0.59$) explained overall 94.4% variance due to these all parameters $F_{6,2}=23.58;p=0.041$ in OC. From this, it can be said that accumulated feed at the bottom of the pond might have contributed to form the organic matter, further, its oxidation possibly was responsible for the variations in the OC in the sediment.

Regression analysis for Creek

In the creek chlorophyll *a* ($B=-0.18$) and phaeophytin ($B=-0.21$), E_h of water ($B=-0.20$), POC ($B=0.61$), phaeophytin of sediment ($B=1.45$), BOD ($B=0.13$) and temperature ($B=-0.92$) were significantly linearly correlated with that of OC $p<0.01$. Amongst these parameters, phaeophytin in the sediment and E_h of water may have amounted for more significant variations in OC.

Experimental Cycle (EC-2)

The fortnight variations in the OC are presented in Table & Figure EC2.11. During this cycle organic carbon showed an increasing trend towards the end of the culture period for both the ponds depicted in Table & figure (EC2.11). Minimum of $1.57\pm 0.36\%$ and maximum $5.22\pm 0.24\%$ OC was observed for aerated pond (Table ST10). For non-aerated pond minimum of $0.93\pm 0.11\%$ on 15 DoC and maximum

6.04±0.12% on 105 DoC (Table ST11) were observed. For creek it was found to be minimum 1.10±0.04 and maximum 6.67±0.11 % OC (Table ST12).

Regression analysis for Aerated Pond

In aerated pond Chlorophyll *a* of water (B=0.52) and sediment (B=1.63), phaeophytin of water (B=-0.50) and sediment (B=-0.97), DO (B=0.51) and COD (B=-1.36) were found to explain 99.9% relationship between these parameters and formation of organic carbon in the sediment. The relation was significant at $F_{6,1}=3598.0, p=0.013$. Thus indicating that, the source of the OC was mainly of phytal origin.

Regression analysis for Non-aerated pond

In the non aerated pond backward regression model explained 99.9% variability of OC due to chlorophyll *a* in the water as well as sediment, phaeophytin and E_h of the sediment, feed applied and DO at $F_{6,1}=1226.12; p=0.022$. Amongst these variables, chlorophyll *a* in water (B=9.79; $p=0.022$), phaeophytin in the sediment (B=-6.67; $p=0.036$) and E_h of the sediment (B=1.06; $p=0.043$) had highly significant effect for the variations, in the sediment OC. Thus, settling phytoplankton as well as accumulation of feed and its degradation on the itinerary, contributed to the variations in OC.

Regression analysis for Creek

In the creek, stepwise multiple regression model explained 96.2% variance in OC due to chlorophyll *a* in water (B=0.42) and sediment (B=0.74) as well as COD (B=-0.26) with overall significant model $F_{3,4}=60.70; p=0.001$. From these analysis overall it can be said that, during both the culture periods, sediment organic matter mostly originated from algal or phytal degradation. The low OC values during both the culture cycles are evident and are depicted in figure EC1.11 for EC-1 and table & figure EC2.11. The relatively low amount of organic matter accumulated in the sediments has been explained by Gonzalez *et al.* (1996). According to them, oxic

conditions existing in the ponds as well as low depths and higher temperature leads to a poor preservation of the organic matter and simultaneously stimulate the bacterial activity of the microbial biomass. Thus, from above analysis it is clear that, experimental ponds accumulated the organic loads which were rework-able through naturally occurring interdependent degradative processes in the water and sediment.

4.1.9 Hydrogen sulphide (H_2S $\mu\text{g.l}^{-1}$)

Experimental Cycle (EC-1)

During EC-1 H_2S concentration in water showed fluctuating concentrations (Table & Figure EC1.12), it was initially low and was found to be maximum on 120 DoC being $1.26 \pm 0.08 \mu\text{g.l}^{-1}$ and minimum of $0.43 \pm 0.20 \mu\text{g.l}^{-1}$ for aerated pond (Table ST7) on 30 DoC. Initially H_2S concentration was more in non-aerated pond than aerated pond and in later half aerated pond showed higher concentrations than non aerated pond (Table & Figure EC1.12).

In non-aerated pond highest H_2S concentration was observed on 45 DoC being $1.37 \pm 0.17 \mu\text{g.l}^{-1}$ and lowest $0.38 \pm 0.23 \mu\text{g.l}^{-1}$ on 0 DOC (Table ST8). In the creek H_2S concentration ranged from 0.11 ± 0.12 and highest $1.37 \pm 0.28 \mu\text{g.l}^{-1}$ (Table ST 9). The fortnight average variations of H_2S are depicted in table & figure EC1.12.

In the water H_2S was significantly correlated with that of phaeophytin in the water, chlorophyll *a* in water and sediment, COD, BOD as well as with pH of the water and explained 99.5% variations in H_2S , $F=287.70$; $p=0.003$.

Experimental Cycle (EC-2)

The fortnight average variations in H_2S are depicted in Table & Figure (EC2.12). From figure it can be seen that, during EC-2, H_2S concentration showed increasing trend with the progressing culture period. On 0 DoC it was $0.03 \pm 0.07 \mu\text{g.l}^{-1}$ for aerated pond and 0.03 ± 0.02 for non-aerated pond. In the creek it was found to be $0.01 \pm 0.01 \mu\text{g.l}^{-1}$. At the end of the culture period, on 105 DoC it was found to be

0.80±0.03 for aeration pond, 0.75±0.06 for non-aerated and for creek it was found to be 0.69±0.11 µg.l⁻¹(Table &Figure EC2.12).

Organic matter in the soil and water on decomposition releases sulfur (mineralized) to form sulphide. The oxidation of sulfide to sulphate occurs through simple chemical reactions (Boyd, 1995). Sulphate being highly soluble is taken up by plants and microbes. In anaerobic environments however, sulfate is used as a hydrogen acceptor in microbial metabolism to ultimately form H₂S. H₂S is known to be toxic to aquatic organisms. Hydrogen sulfide in the water during both the culture period showed an increasing trend with the progress in the culture period (Table & Figure EC 1.12; Table & Figure EC2.12). One way ANOVA carried out, showed no significant differences $p>0.05$ in aerated and non aerated pond during both the cycles EC-1 and EC-2. It can be seen from Table & Figures EC1.12 and EC2.12, that during both culture periods the H₂S in aerated pond was marginally higher than non aerated pond. This can be attributed to comparatively higher particulate organic carbon (Table & Figure EC1.10; Table & Figure EC2.10) and sediment organic carbon (Table & Figure EC1.11 and EC2.11) accumulation in the aerated pond due to higher feed input requirement due to higher stocking densities. The mean H₂S concentrations during EC-1 were 0.84±0.31 and 0.77±0.30 µg.l⁻¹ in aerated pond and non-aerated pond and are depicted in Table ST7 and ST8, respectively. During EC-2 the concentrations of H₂S in the water were 0.56±0.24 and 0.50±0.21 µg.l⁻¹ for aerated and non-aerated pond and are depicted in Table ST10 and ST11, respectively.

During both the culture periods adjacent creek also showed no significant differences $p>0.05$ in the average H₂S concentrations between EC-1 and EC-2. The concentrations of H₂S during EC-1 and EC-2 in the creek water were clearly less than both the ponds. During EC-1 H₂S concentrations were high initially till 60 DoC but thereafter concentrations in the creek decreased and showed similar trend to that during EC-1 being less than the ponds. The lower concentrations of dissolved H₂S in

the creek water can be attributed to effective degradation of organic matter as discussed above in POC and OC regression analysis (Sections 4.1.7 and 4.1.8). Besides, tidal incursion and excursion along with the rain water might have resulted in the dilution of H₂S thereby decreasing the concentration of H₂S in the creek water.

4.1.10 Variations in Nutrients (NO₃-N, NO₂-N, NH₄-N and PO₄-P μmol.l⁻¹)

According to Gopinath *et al.* (2002) anything beside water and carbon dioxide that is required by plants in the synthesis of organic matter or skeletal materials is regarded as nutrient. Nitrogen and phosphorus are considered as the main limiting nutrients. Nitrogen is reviewed to be limiting factor in marine environments, while phosphorus is limiting in the fresh water environments (Follomi, 1996). Concentrations based on the classification report by different workers in the coastal waters of red sea has been sited by Fahmy (2003), for determining the oligotrophic to eutrophic nature on the basis of NO₃-N and NH₃-N concentrations in the red sea. The oligotrophic water has been shown to have the concentration of NH₃-N and NO₃-N being 0.5 μmol.l⁻¹ and eutrophic water in order of 2.0 μmol.l⁻¹ for NH₃-N and 4.0 μmol.l⁻¹ for NO₃-N Dalkiran *et al.*(2006). Further, Dalkiran *et al.* (2006) have concluded that, pH and DO affects the internal nutrient loadings in the eutrophic shallow lake. Thus, indicating the complex relationship of nutrients with other chemical variable.

4.1.11 A) Nitrate-Nitrogen (NO₃-N)

Experimental Cycle (EC-1)

Overall NO₃-N concentrations were found to be slightly higher in aerated pond than in non-aerated pond (Table & Figure EC1.13). In aerated pond, it ranged from 2.72±0.23 on 0 DoC to 0.30±0.03 μmol.l⁻¹ on 120 DoC. In non-aerated pond it was lowest on 90 DoC being 0.33±0.06 and highest on 30 DoC 0.98±0.51 μmol.l⁻¹. In the creek concentrations of NO₃-N were highest on 30 and 45 DoC being 2.94±0.53 and 3.57±0.44 μmol.l⁻¹. The average fortnight variations in NO₃-N are presented in Figure EC1.13. Thus, during EC-1, NO₃-N remained in the mean range of 0.86±0.73,

0.59±0.22 μmol.l⁻¹ and 1.27±1.16 μmol.l⁻¹ for aerated and non-aerated pond as well as in the creek during EC-1.

Experimental Cycle (EC-2)

The fortnightly average variations of NO₃-N are depicted in Figure EC2.13. During EC-2 NO₃-N concentrations were low initially and remained fairly constant till 45 DoC (Table & Figure EC2.13) with an average values of 0.08 μmol l⁻¹ and 0.15 μmol l⁻¹ in aerated and non-aerated pond respectively, depicted in Table ST10 and ST11, respectively. Thereafter, there was an increase in the concentration of NO₃-N till the end of the culture period, with highest concentrations on 105 DoC 8.48±0.04 for aerated pond and 4.88±0.21 μmol l⁻¹ for non-aerated pond, but at the same time in the creek; NO₃-N concentrations were observed to be low with 0.56±0.01 μmol l⁻¹ (Table & Figure EC2.13). During EC-2, NO₃-N in this regard remained in the mean range of 1.13±1.45 for aerated pond, 1.26±1.78 μmol.l⁻¹ for non-aerated pond and 2.11±1.92 μmol.l⁻¹ for creek.

4.1.12 B) Nitrite-Nitrogen (NO₂-N)

Experimental Cycle (EC-1)

Overall NO₂-N values were almost constant throughout the culture period, however, found to be marginally higher in non-aerated pond than in aerated pond are depicted in Figure (EC1.14). In aerated pond, NO₂-N concentrations ranged from 0.06±0.01 to 0.53±0.11 μmol.l⁻¹ and in non-aerated pond from 0.08±0.02 to 3.42±2.66 μmol.l⁻¹. NO₂-N concentrations were higher initially and were found to drop marginally showing decreasing trend with an exceptionally higher concentration of 3.42±2.66 μmol.l⁻¹ for non aerated pond at end of the culture period that is on 120 DoC, (Figure EC1.14). In the creek NO₂-N ranged from 0.19±0.04 to 1.38±0.09 μmol.l⁻¹ during culture period.

Experimental Cycle (EC-2)

The fortnightly average variations of NO₂-N are depicted in Table & Figure. EC2.14. In aerated pond NO₂-N in aerated pond was marginally higher than non-

aerated pond. It showed an increasing trend with progress in culture period (Figure EC 2.14). Minimum concentration of $0.30 \pm 0.02 \mu\text{mol l}^{-1}$ and maximum concentration of $4.12 \pm 0.06 \mu\text{mol l}^{-1}$ was registered on 0 and 105 DoC respectively for aerated pond. For non-aerated pond also, it was found to be highest $2.04 \pm 0.04 \mu\text{mol l}^{-1}$ on 105 DoC and lowest 0.38 ± 0.01 on 15 DoC. In the creek, $\text{NO}_2\text{-N}$ concentrations were lower than both the ponds, with an exceptional high concentration of $4.34 \pm 0.01 \mu\text{mol l}^{-1}$ on 60 DoC, the variations of $\text{NO}_2\text{-N}$ have been depicted in Table & Figure (EC2.14).

4.1.13 C) Ammonia-Nitrogen ($\text{NH}_3\text{-N}$)

Experimental Cycle (EC-1)

The fortnightly average variations of $\text{NH}_3\text{-N}$ are depicted in Table & Figure (EC1.15). For aerated pond $\text{NH}_3\text{-N}$ concentrations were observed to be minimum $0.10 \pm 0.06 \mu\text{mol.l}^{-1}$ and maximum 1.93 ± 0.24 on 90 and 30 DoC respectively. For non-aerated pond minimum concentrations of $\text{NH}_3\text{-N}$ were observed to be 0.31 ± 0.04 and maximum of $4.76 \pm 0.13 \mu\text{mol.l}^{-1}$ were observed on 15 and 120 DoC. The $\text{NH}_3\text{-N}$ concentrations peaked on 30 DoC for both ponds as well as for creek (Figure EC1.15). In the Creek slightly higher concentrations of $\text{NH}_3\text{-N}$ than pond water were evident. The cyclic variations for $\text{NH}_3\text{-N}$ in the creek are depicted in Table & Figure EC1.15.

Experimental Cycle (EC-2)

The fortnightly average variations of $\text{NH}_3\text{-N}$ are depicted in Table & Figure (EC2.15). During EC-2, $\text{NH}_3\text{-N}$ concentrations in aerated pond were higher than non-aerated-pond and creek. It increased gradually, and was found to be highest on 75 DoC $0.60 \pm 0.04 \mu\text{mol l}^{-1}$ for aerated pond. In non-aerated pond it was found to be highest on 60 DoC being $0.33 \pm 0.02 \mu\text{mol l}^{-1}$. In the creek $\text{NH}_3\text{-N}$ minimum concentrations of $0.12 \pm 0.01 \mu\text{mol l}^{-1}$ and maximum of 0.40 ± 0.01 were observed on 0 and 105 DoC, respectively.

Combined correlation and regression analysis of NO₃-N, NO₂-N and NH₃-N for EC-1 and EC-2

Experimental Cycle (EC-1)

Correlation analysis for Aeration Pond

Correlations described in this section for aeration pond are depicted in Table ST1. Nitrate, nitrite and ammonia nitrogen all showed decreasing trend with the progressing culture period in aerated pond (Table & Figure EC1.13, EC1.14 and EC1.15). During EC-1 in aeration pond NO₂-N was strongly correlated to both NO₃-N and NH₃-N ($r=0.7; p\leq 0.05$), the correlation between NO₃-N, NO₂-N, NH₃-N with that of chlorophyll *a* in water and sediment were weak $p\leq 0.05$ (Table ST1). As can be seen from the Table & Figure EC1.17, chlorophyll *a* in the water from 30 DoC to 75 DoC dropped from about 5mg.l⁻¹ to 1mg.l⁻¹. Along with this, low nutrient and cloud cover with heavy raining and reduced light might have been the reason for lack of correlation or low uptake of the nutrients by phytoplankton. Conspicuously, with low photosynthesis, oxygen regeneration was not possible. It can be seen during the same time that oxygen concentration was highest of 4 mg.l⁻¹ on 60 DoC and shows that aeration possibly helped in maintaining the dissolved oxygen concentrations.

Correlation analysis for Non-aerated Pond

Correlations described in this section for non-aeration pond are depicted in Table ST2. During EC-1 in non-aerated pond more or less similar trend was observed, to that of aeration pond, where NO₃-N and NO₂-N were weakly correlated ($r=0.33; p\leq 0.05$) and are depicted in Table ST2. At the same time NO₂-N was strongly correlated with that of NH₃-N ($r= 0.85; p\leq 0.05$). Further, NH₃-N was also correlated with that of chlorophyll *a* in water ($r=0.51$) and sediment ($r=0.85; p\leq 0.05$). These trends indicate that, NH₃-N might have been taken up as a source of nutrient in the non-aerated pond during EC-1. It has been sited by Hargreaves (1998) that, ammonia is the preferred N substrate for phytoplankton, and only after this has been depleted (<0.03 mg N.l⁻¹) nitrates in significant quantities will be assimilated by the

phytoplankton. Dham *et al.* (2002) studied seasonal variations in uptake, and in situ regeneration of nitrogen in mangrove waters have also showed that, NO₃-N followed by NO₂-N uptake by phytoplankton was dominant during post monsoon season and NH₃-N and urea uptake by phytoplankton dominated during the pre monsoon season. Hence, these processes signifies the uptake of the nutrients by phytoplankton, however, the fluctuating nature of NO₃-N, NO₂-N and NH₃-N without any particular behavior can be attributed to the monsoonal rain, where fresh water influx might have played role in either decreasing or increasing nutrient concentration due to lower photosynthetic rates or from runoff water in to the pond.

Correlation analysis for Creek

Correlations described in this section for creek are depicted in Table ST3. In creek multiple regression analysis carried out explained 92.9% variation in NO₃-N due to changes in the chlorophyll *a* in the water and sediment, BOD, temperature and salinity with overall significant model at $F_{5,3}=21.81;p=0.015$. For NO₂-N backward multiple regression analysis carried out explained 92.1% variation in NO₂-N due to changes in the chlorophyll *a* of water and sediment, E_h of water and sediment with overall significant model at $F_{4,4}=24.24;p=0.005$. For NH₃-N backward multiple regression analysis carried out explained 99.6% variation in NH₃-N due to changes in the chlorophyll *a* water and sediment, phaeophytin in the water, E_h and pH of water and temperature with overall significant model at $F_{6,2}=321.92;p=0.003$.

Experimental Cycle-2

Correlation analysis for Aerated pond

Correlations described in this section for aeration pond are depicted in Table ST4. In aeration pond during EC-2 mean NO₃-N values were higher 1.12 ± 1.45 than mean NH₃-N 0.36 ± 0.11 with very stable NO₂-N (Table & Figure EC2.14) At the same time one way ANOVA for NH₃-N showed no significant differences in the fortnightly as well as overall mean concentrations $p>0.05$. It can be seen from the Table & Figure (EC2.15) that, NH₃-N remained very low. There were strong positive correlations in the NO₃-N and NO₂-N ($r=0.96$). NO₃-N also showed good co-

relations with chlorophyll *a* in the water and sediment with $r=0.74$ and $r=0.56$; $p \leq 0.05$. The lack of strong correlations with ammonia suggests that both nutrients $\text{NO}_3\text{-N}$ and $\text{NO}_2\text{-N}$ were not limiting and were used in the primary productivity. This is again, reflected in good co-relationship of Chlorophyll *a* in water with that of dissolved oxygen ($r=0.61$; $p \leq 0.05$) shows that, oxygen was generated through photosynthetic processes. Further, DO was strongly correlated with $\text{NO}_3\text{-N}$, $\text{NO}_2\text{-N}$, $\text{PO}_4\text{-P}$ and $\text{NH}_3\text{-N}$ with ($r= 0.9$) for the first three variables and ($r=0.43$) for $\text{NH}_3\text{-N}$ shows that oxygen was not deficient and oxidation of $\text{NH}_3\text{-N}$ to $\text{NO}_3\text{-N}$ was best carried out in the pond environment.

Correlation analysis for Non-aerated Pond

Correlations described in this section for aeration pond are depicted in Table ST5. As can be seen from the Table & Figures EC2.13; EC2.14; EC2.15, all nutrients showed lower concentrations. These nutrients when correlated with chlorophyll *a* in water and sediment showed weak correlations (Table ST5).

Correlation analysis Creek

Correlations described in this section for creek are depicted in Table ST6. A sharp increase in $\text{NO}_3\text{-N}$ can be seen on 30 and 45 DoC during EC-1 and 60 to 90 DoC during EC-2 for creek. The sharp rise of nitrate after initial low values can be attributed to the incessant monsoon rains, which might have brought about the $\text{NO}_3\text{-N}$ from land water runoff and flooded creek due to fresh water runoff also from upstream. It was observed that, when $\text{NO}_3\text{-N}$ increased on 30 DoC during EC-1 and 60 DoC and 90 DoC during EC-2. Subsequently, $\text{NO}_2\text{-N}$ and $\text{NH}_3\text{-N}$ values had shoot up for the creek. Overall positive correlation was observed amongst $\text{NO}_3\text{-N}$, $\text{NO}_2\text{-N}$ and $\text{NH}_3\text{-N}$. With ($r=0.3, r=0.5, p \leq 0.05$) during EC-1 and EC-2 creek values indicate that, with simultaneous increase or decrease in the values of $\text{NO}_3\text{-N}$ there was increase or decrease in the $\text{NO}_2\text{-N}$ and $\text{NH}_3\text{-N}$. Thus creek showed more natural variations and nitrogen cycling with prevalent nitrification and denitrification processes.

4.1.15 D) Phosphate-Phosphorus (PO₄-P)

Experimental Cycle (EC-1)

For PO₄-P more variations were observed in non aerated pond than in aerated pond. PO₄-P concentrations were high initially on 30 DoC $3.97 \pm 0.47 \mu\text{mol.l}^{-1}$ in aerated pond and showed a sharp decrease in concentration $2.00 \pm 0.32 \mu\text{mol.l}^{-1}$ on 45 DoC (Table & Figure EC1.16). Thereafter, it decreased gradually to its minimum $0.13 \pm 0.04 \mu\text{mol.l}^{-1}$ on 90 DoC. From Table & Figure EC1.16, it can be seen that in non aerated pond PO₄-P concentrations varied cyclically.

Correlation analysis for Aerated pond

Correlations described in this section for aeration pond are depicted in Table ST4. Temperature and BOD were correlated being $r=0.77$ and $r=0.69; p \leq 0.05$ respectively with PO₄-P. The lack of strong correlations amongst chlorophyll *a* and PO₄-P, OC and BOD indicates that decreasing temperature and redox reactions might have been responsible for decrease in the PO₄-P in aerated pond.

Correlation analysis for Non-aerated pond

In non-aerated pond there were large cyclic variations, Table & Figure (EC1.16). When backward multiple regression analysis was carried out, PO₄-P showed linearity with that of many environmental factors such as temperature, chlorophyll *a* of water, organic carbon, BOD, COD and pH of sediment $p < 0.05$. Thus cyclic variations in PO₄-P can be attributed to the changes in these physico-chemical parameters in non-aerated pond rather than uptake by the phytoplankton.

Correlation analysis for Creek

For PO₄-P backward multiple regression analysis carried out explained 99.9% variation in PO₄-P due to changes in the chlorophyll *a* of sediment, E_h of water, pH of water, pH of sediment and DO, as well as temperature with overall significant model at $F_{7,1}=1894.4; p=0.018$. Amongst these variables, DO ($B=2.21; p=0.014$), pH

of water ($B=-3.8;p=0.012$) and E_h of water ($B=-9.2;p=0.008$) were responsible, mostly for changes in the $PO_4\text{-P}$ in the creek water.

Experimental Cycle (EC-2)

During EC-2 $PO_4\text{-P}$ showed large variations as well as cyclic trends of high and low concentrations during the culture period for both ponds (aeration and non aeration pond) as well as creek. $PO_4\text{-P}$ concentrations were highest 2.46 ± 0.15 and 2.75 ± 0.07 $\mu\text{mol.l}^{-1}$ on 60 DoC for aerated and non-aerated pond respectively is depicted in Table & Figure EC2.16. At the end of the culture period on 105 DoC $PO_4\text{-P}$ was found to be 1.43 ± 0.05 and 0.87 ± 0.09 $\mu\text{mol.l}^{-1}$ in aerated and non-aerated respectively. For creek it varied from 0.92 ± 0.0 to 2.00 ± 0.01 $\mu\text{mol.l}^{-1}$ on 0 DoC and 60 DoC.

As it is known, bio-chemical weathering on continents represents the most significant source of bio available phosphorus. As essential nutrient phosphorus is taken up as inorganic phosphate and represents an important driving and regulating force behind biological productivity. High temperatures, low pH and presence of sulphate promote the mobilization of phosphate (Follomi, 1996). Transformations of particulate to dissolved $PO_4\text{-P}$ and vice versa are observed during the river discharges, due to redox changes and oxidation due to temporary burial in riverine sediment and consequent reworking especially in case of ferric phosphate adsorbed on to redox sensitive iron oxyhydroxides (Follmi, 1996). Fortnightly average $PO_4\text{-P}$ concentrations are depicted in Table & Figure EC1.16, for EC-1 and Table & Figure EC2.16, for EC-2.

Correlation analysis for Aerated Pond

During EC-2 in aerated pond $PO_4\text{-P}$ showed strong positive correlation-ship with that of chlorophyll a in the water ($r=0.72;p<0.05$) and is depicted in Table ST4. It can be seen from Table & Figure EC2.16 that, there was an increase in $PO_4\text{-P}$ on 60 DoC and it was characteristic, when compared with that of $NO_3\text{-N}$. This sudden surge in

the nutrients can only be attributed to the rainy season which might have brought about an increase in these nutrients. Again, from these observation it can be seen that, PO₄-P in aerated pond was more or less stable. During present study, overall mean concentration in aerated pond for PO₄-P was found to be 1.33±1.23 μmol.⁻¹ and are depicted in Table ST10.

Correlation analysis for Non-aerated Pond

Similar trends of PO₄-P were observed for non-aerated pond as that of aerated pond with increase in PO₄-P on 60 DoC (Table & Figure EC2.16). Further, PO₄-P showed significant correlation with that of chlorophyll *a* in the water ($r=0.47;p<0.05$) and is depicted in Table ST11.

Correlation analysis for Creek

In creek, PO₄-P when subjected to multiple regression analysis showed its linearity to temperature and BOD. There were cyclic variations in creek and PO₄-P showed decreasing trend during the culture period (Table & Figure EC 2.16). For the creek the decreasing trend can be attributed to changes in temperature and salinity while the cyclic changes can be attributed to the land water runoff during monsoon bringing additional PO₄-P in the creek. In cyclic variations decreasing PO₄-P can be attributed to, utilization by phytoplankton.

Furumai and Ohjaki (1989) has sited that PO₄-P is dependent on the pH and redox of overlying and interstitial waters. He has shown that, the drop in E_h leads to the release of phosphates from the sediment and both physico-chemical adsorption desorption of phosphorus and biological uptake-release takes part in phosphorus exchange when no inhibitor for bacteria was added. The role of anaerobic and aerobic sequences in phosphorus removal and increase due to bacterial action has been shown by Hood and Randall *et al.* (2001).

4.1.16 Chlorophyll *a* and Phaeophytin (mg.m^{-3})

Experimental Cycle (EC-1)

Water chlorophyll *a* and phaeophytin

Fortnightly averages for water chlorophyll *a* are presented in Table & Figure (EC1.17) and phaeophytin in the water are depicted in Table & Figure EC1.19. During EC-1, chlorophyll *a* concentrations of water in the beginning were higher for both aerated pond and non aerated pond with concentration $6.41 \pm 1.07 \text{ mg.m}^{-3}$ and $4.98 \pm 2.83 \text{ mg.m}^{-3}$ respectively on 15 and 120 DoC (Table EC1.17; Figure EC1.17). Thereafter, concentration of chlorophyll *a* in water decreased from 30 DoC till 75 DoC and value of 1.42 mg.m^{-3} was in aerated pond. Chlorophyll *a* again increased from 60 DoC onwards till the end of the culture period with values of 4.98 ± 0.62 on 15 DoC and $5.46 \pm 3.02 \text{ mg.m}^{-3}$ on 120 DoC for aerated pond and non aerated pond respectively. Similar trends were observed in the creek water, for chlorophyll *a* concentrations in the water with lowest $1.14 \pm 0.08 \text{ mg.m}^{-3}$ concentration on 60 DoC and $5.93 \pm 0.25 \text{ mg.m}^{-3}$ on 120 DoC.

Variations in phaeophytin concentrations followed similar trend (Table & Figure EC1.19) being high at the start of the culture period, followed by decrease, till 60 DoC to 75 DoC and again gradual increase till 120 DoC. In aerated pond phaeophytin concentration in water ranged from 1.59 ± 0.73 to $5.26 \pm 0.16 \text{ mg.m}^{-3}$ on 60 and 105 DoC. In non-aerated pond it ranged from 1.02 ± 0.10 on 60 DoC to $5.36 \pm 0.22 \text{ mg.m}^{-3}$ on 105 DoC, and in creek it ranged from 1.06 ± 0.04 to $4.28 \pm 0.38 \text{ mg.m}^{-3}$ on 60 and 105 DoC respectively.

Sediment chlorophyll *a* and phaeophytin

The fortnightly average concentrations of chlorophyll *a* in sediment are presented in Table & Figure EC1.18 and phaeophytin in sediment are presented in Table & Figure EC1.20 during EC-1. Chlorophyll *a* and phaeophytin concentrations in sediment also showed same trend as that of water (Table & Figure EC1.18), with conspicuous decrease on 45 DoC to 60 DoC (Figure EC1.19). Minimum chlorophyll *a* in

sediment for aerated pond during EC-1 was $1.38 \pm 0.33 \mu\text{g.g}^{-1}$ on 60 DoC and maximum on 120 DoC $14.24 \pm 5.56 \mu\text{g.g}^{-1}$. Minimum and maximum concentrations of chlorophyll *a* in sediment for non-aerated pond were 2.06 ± 0.53 and $10.68 \pm 12.24 \mu\text{g.g}^{-1}$ on 60 DoC and 120 DoC and are depicted in Table and Figure EC1.18. For creek chlorophyll *a* concentrations were found to be 1.24 ± 0.08 and $10.68 \pm 3.78 \mu\text{g.g}^{-1}$. Phaeophytin pigment concentration showed wide variations in the sediment (Table & Figure EC1.20) as compared to water (Table & Figure EC 1.17). Phaeophytin in sediment, in both the ponds was low on 45 DoC and 60 DoC (Table & Figure EC.1.19). Overall it ranged from 1.05 ± 0.23 on 105 DoC to 3.78 ± 2.63 on 90 DoC $\mu\text{g.g}^{-1}$ in the sediment. In the creek phaeophytin in sediment was found to be highest 3.22 ± 0.52 on 0 DoC and lowest on 90 DoC being $1.28 \pm 1.12 \mu\text{g.g}^{-1}$.

Experimental Cycle (EC-2)

Water chlorophyll *a* and phaeophytin

The fortnightly average concentrations of chlorophyll *a* in water are presented in Table & Figure (EC2.17) and phaeophytin in water are depicted in Table & Figure (EC2.19). During EC-2 chlorophyll *a* values showed increasing trend in aerated pond, while in non-aerated pond there were large variations in the chlorophyll *a* concentrations with lowest values on 30 DoC and 75 DoC being 3.20 ± 0.06 and $4.27 \pm 0.22 \text{mg.m}^{-3}$ respectively (Table & Figure EC2.17). For aerated pond till 45 DoC it averaged 10.50mg.m^{-3} and was increased to $30.97 \pm 1.26 \text{mg.m}^{-3}$ and $55.54 \pm 3.31 \text{mg.m}^{-3}$ on 60 DoC and 105 DoC (Table & Figure EC2.17). In the creek, chlorophyll *a* concentrations were highest being $8.54 \pm 0.0 \text{mg.m}^{-3}$ on 0 DoC and $1.07 \pm 0.01 \text{mg.m}^{-3}$ on 75 DoC.

Phaeophytin concentrations of water were marginally higher in aerated pond than non-aerated pond (Table & Figure EC2.19) and, were stable till 60 DoC. Overall mean value of 3.33 ± 3.48 and $2.59 \pm 0.99 \text{mg.m}^{-3}$ for aerated pond and non-aerated pond were observed respectively. A thereafter phaeophytin concentration increased till the end of the culture period and was found to be highest on 90 DoC 9.93 ± 0.06 and $6.56 \pm 0.29 \text{mg.m}^{-3}$ in aerated and non-aerated pond. In creek overall phaeophytin

concentrations were higher than both the ponds and showed large variations with highest values on 30 DoC and 75 DoC being 18.16 ± 0.38 and 12.39 ± 0.08 $\text{mg} \cdot \text{m}^{-3}$ respectively.

Sediment chlorophyll a and phaeophytin

The fortnightly average concentrations of chlorophyll *a* in sediment are presented in Table and Figure EC2.18, and phaeophytin in sediment are depicted in Table and Figure EC2.20. It was clearly observed that, in aerated pond chlorophyll *a* in sediment showed higher concentrations than non-aerated pond. Chlorophyll *a* values were found to be lowest on 75 DoC with an average value of 1.18 ± 0.03 $\mu\text{g} \cdot \text{g}^{-1}$ in aerated pond, in non-aerated pond it was found to be 1.12 ± 0.06 $\mu\text{g} \cdot \text{g}^{-1}$, in the creek it was 1.07 ± 0.06 depicted in Table & Figure EC2.18. For aerated pond chlorophyll *a* in sediment was observed to be 4.27 ± 0.13 $\mu\text{g} \cdot \text{g}^{-1}$ on 0 DoC and 11.75 ± 1.76 $\mu\text{g} \cdot \text{g}^{-1}$ on 105 DoC. For non-aerated pond it was observed to be 3.69 ± 0.51 $\mu\text{g} \cdot \text{g}^{-1}$ and 7.94 ± 0.07 $\mu\text{g} \cdot \text{g}^{-1}$ on the 0 and 105 DoC respectively. In creek sediment chlorophyll *a* values were lower and showed similar trends to that of non-aeration pond with lowest value 1.07 ± 0.06 on 75 DoC and highest 7.48 ± 0.11 $\mu\text{g} \cdot \text{g}^{-1}$ on 105 DoC.

Phaeophytin concentrations in sediment showed an increasing trend with progressing culture period for both the ponds (Figure EC2.20) and showed cyclic high and low values, with highest on 15, 60 and 105 DoC for both the ponds and are depicted in Table & Figure. (EC2.20). For creek variations were more profound and registered highest concentrations of phaeophytin in the sediment 56.60 ± 1.12 and 60.66 ± 0.79 $\mu\text{g} \cdot \text{g}^{-1}$ on 30 DoC and 60 DoC respectively (Table & Figure. EC2.20).

Chlorophyll *a* concentrations showed interesting results in view that, both the cycles were influenced by monsoon period with cloud cover. One way ANOVA single factor was carried out to see if there were any differences in the concentrations of chlorophyll *a* in between aerated and non-aerated pond in EC-1 and EC-2, as well as to see the difference if any, in between two cycles EC-1 and EC-2 carried out over two production cycles for the respective aeration and non-aeration ponds (Inter-annual). This analysis showed that, there were no significant differences in between

aerated and non aerated pond ($p>0.05$) during respective cycles EC-1 and EC-2. Interestingly however, chlorophyll *a* differed in between two cycles EC-1 and EC-2 when aerated ponds and non-aerated ponds from respective cycles were analyzed. This analysis showed that, there were significant to highly significant differences in between chlorophyll *a* concentrations for aerated pond ($p<0.05$) and non aerated pond ($p<0.001$) of EC-1 and EC-2.

Chlorophyll *a* concentrations during EC-2 were found to be higher than EC-1 in both the ponds. During EC-2 abrupt photoperiods or micronutrient availability right from the start of the culture period might have been the reason to sustain the higher chlorophyll *a* in the pond water. The mean concentrations of chlorophyll *a* in water in aerated pond were 3.68 ± 1.79 in non-aerated pond it was found to be 3.84 ± 1.42 $\text{mg}\cdot\text{m}^{-3}$ during EC-1. During EC-2 mean chlorophyll *a* concentrations in water for aerated pond were 18.49 ± 17.40 and for non-aerated pond 16.36 ± 10.63 $\text{mg}\cdot\text{m}^{-3}$.

Good correlation of chlorophyll *a* in water with that of DO ($r=0.61$, $\text{PO}_4\text{-P}$ $r=0.72$, $\text{NO}_3\text{-N}$ $r=0.74$, $\text{NO}_2\text{-N}$ $r=0.72$, and chlorophyll *a* in sediment $r=0.56$, $r=0.60$, $r=0.60$ with that of $\text{NO}_3\text{-N}$, $\text{NO}_2\text{-N}$ and $\text{PO}_4\text{-P}$ in water during EC-2 in aerated pond (Table ST4) indicates that, dissolved oxygen was released in the photosynthetic process and nutrient were not limiting for primary productivity in aeration pond. This can be seen from the overall average values of chlorophyll *a* in water and sediment. The average values of chlorophyll *a* in water were 18.49 ± 17.40 $\text{mg}\cdot\text{m}^{-3}$ and for sediment it was 5.94 ± 3.12 $\text{mg}\cdot\text{m}^{-3}$. Low concentration of chlorophyll *a* in sediment can be attributed to lower light intensities reaching to the bottom. Again it can be confirmed from Table & Figure EC2.2 for salinity and chlorophyll *a* (Table & Figure EC2.17) on 60 DoC, during EC-2 in pond water there was possible break in the rain which might have triggered the sharp increase in the chlorophyll *a* also on 60 DoC. Thus possibly light intensities were higher during the day time for EC-2 than EC-1 and that, may have sustained the higher chlorophyll *a* in the water. The role of

photosynthetically active radiation and light intensities and its role in plant growth have been shown by Nathan *et al.* (1984).

Chlorophyll *a* in the Creek

Chlorophyll *a* concentration in creek water during EC-2, as can be seen from Table & Figure EC2.17 were comparatively low than the experimental ponds. All the nutrients in the creek $\text{NO}_3\text{-N}$, $\text{NO}_2\text{-N}$, $\text{NH}_3\text{-N}$ and $\text{PO}_4\text{-P}$ were negatively correlated with that of chlorophyll *a* in the creek water during EC-2, and are depicted in Table ST6. Higher phaeophytin was observed in the creek water from 45 to 90 DoC with an exception on 60 DoC (Table & Figure EC2.19). It can be derived from salinity drop that (Table & Figure EC2.2) from 45 DoC, initial low rains followed by heavy rains might have affected the nutrient concentrations in the creek and though nutrients were available, the rate of phytoplankton decay dominated over the production.

Table EC1.1:Fortnightly average variations in the temperature (°C)

	0	15	30	45	60	75	90	105	120
Aerated Pond	32.2	32.7	35.4	34.3	32.5	31.5	29.9	27.9	32.2
Non Aerated Pond	32.1	32.8	35.3	34.3	32.5	31.5	29.8	27.8	31.2
Creek	32.1	31.4	34.8	33.4	32.0	30.8	29.3	27.2	30.7

Table EC1.2:Fortnightly average variations in the salinity (PSU)

	0	15	30	45	60	75	90	105	120
Aerated Pond	38.00 ± 0.0	43.18 ± 0.02	41.89 ± 0.04	33.64 ± 0.04	18.53 ± 0.03	18.09 ± 0.02	13.20 ± 0.03	7.50 ± 0.04	7.20 ± 0.02
Non aerated Pond	39.54 ± 0.03	40.59 ± 0.02	41.89 ± 0.03	31.70 ± 0.02	17.47 ± 0.02	18.75 ± 0.02	14.00 ± 0.05	9.40 ± 0.02	8.10 ± 0.01
Creek	36.78 ± 0.01	39.46 ± 0.05	38.02 ± 0.02	22.55 ± 0.21	1.05 ± 0.07	2.10 ± 0.14	4.00 ± 0.0	3.25 ± 0.35	6.50 ± 0.71

Table EC1.3:Fortnightly average pH variations in the water

	0	15	30	45	60	75	90	105	120
Aerated Pond	7.64 ± 0.07	8.00 ± 0.04	7.96 ± 0.03	7.09 ± 0.06	7.27 ± 0.04	7.96 ± 0.10	7.23 ± 0.09	8.32 ± 0.04	7.59 ± 0.07
Non aerated Pond	7.72 ± 0.07	7.88 ± 0.06	7.94 ± 0.02	7.32 ± 0.08	7.16 ± 0.04	7.46 ± 0.11	7.46 ± 0.11	8.50 ± 0.02	7.90 ± 0.10
Creek	7.74 ± 0.21	8.03 ± 0.03	7.83 ± 0.07	7.11 ± 0.08	7.28 ± 0.05	7.41 ± 0.03	7.60 ± 0.06	7.69 ± 0.11	7.53 ± 0.21

Table EC1.4:Fortnightly average pH variations in the sediment

	0	15	30	45	60	75	90	105	120
Aerated Pond	7.02 ± 0.85	7.29 ± 0.48	6.98 ± 0.24	6.86 ± 0.12	7.13 ± 0.05	7.21 ± 0.17	7.62 ± 0.07	6.99 ± 0.31	7.39 ± 0.06
Non aerated Pond	6.48 ± 0.16	7.17 ± 0.45	6.06 ± 0.10	6.17 ± 0.15	7.03 ± 0.05	7.04 ± 0.06	7.47 ± 0.29	6.66 ± 0.08	7.11 ± 0.07
Creek	6.19 ± 0.98	7.00 ± 0.09	6.15 ± 0.36	6.01 ± 0.01	7.21 ± 0.04	6.72 ± 0.15	7.61 ± 0.08	6.05 ± 0.14	6.84 ± 0.08

Figure EC1.1:Fortnightly average variations in temperature⁰C.

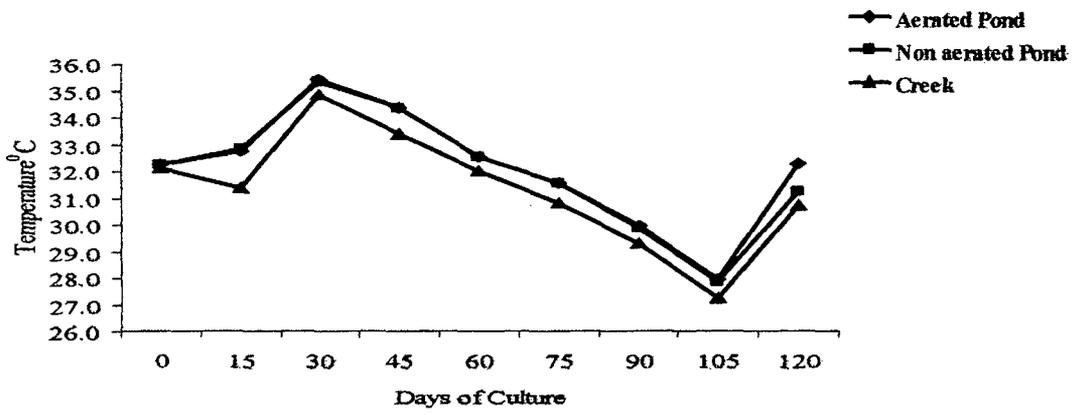


Figure EC1.2:Fortnightly average variations in salinity (PSU).

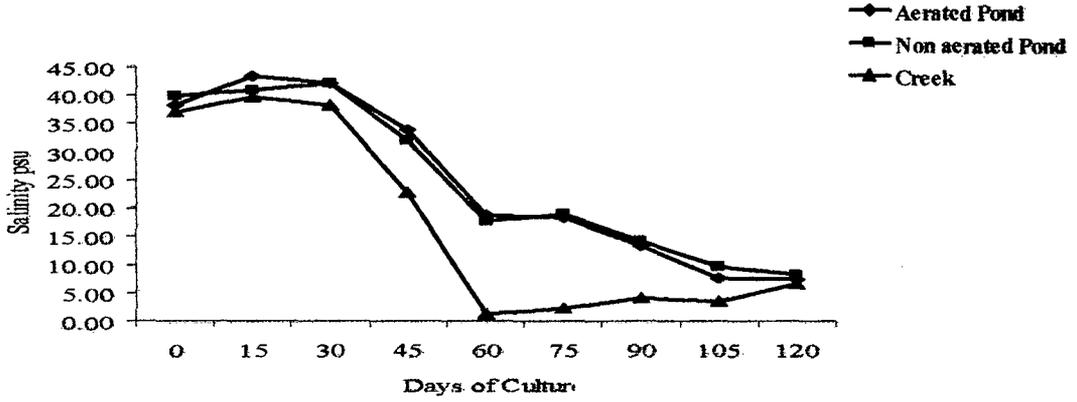


Figure EC1.3:Fortnightly average variations in pH of water:

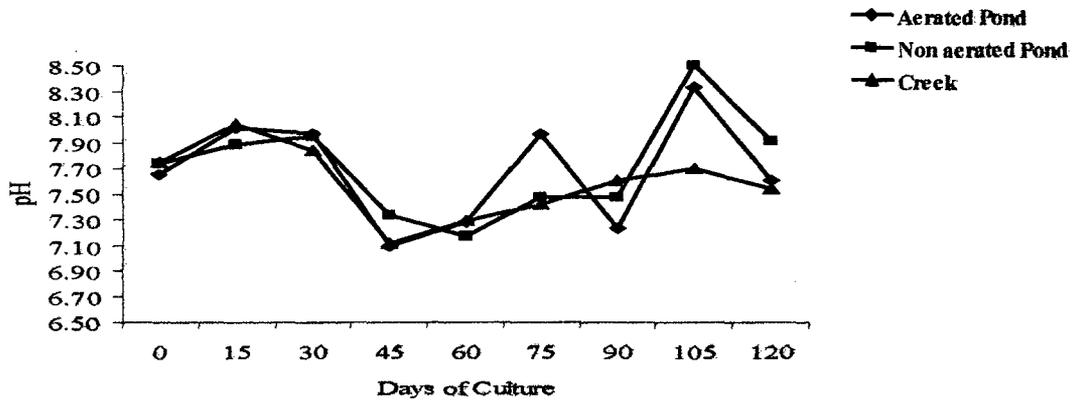


Figure EC1.4:Fortnightly average variations in pH of sediment.

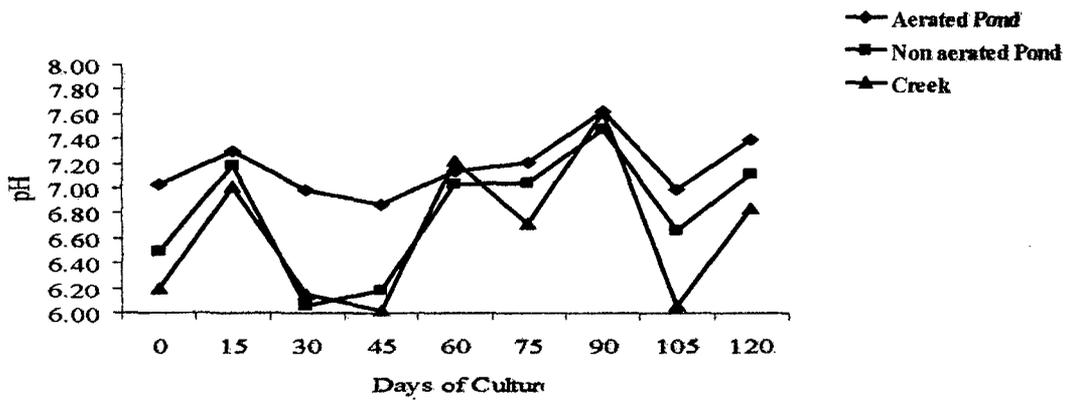


Table EC1.5:Fortnightly average variations in of Eh in the water (mv.)

	0	15	30	45	60	75	90	105	120
Aerated Pond	101.07 ± 4.62	72.00 ± 4.36	72.10 ± 1.61	77.03 ± 2.57	62.80 ± 4.12	84.23 ± 6.74	104.17 ± 6.26	112.77 ± 9.79	90.47 ± 4.50
Non-aerated Pond	94.60 ± 4.37	55.40 ± 8.65	64.60 ± 1.21	69.00 ± 2.55	54.60 ± 4.63	57.83 ± 2.16	88.93 ± 2.12	94.03 ± 4.18	77.57 ± 0.97
Creek	89.17 ± 1.91	49.77 ± 1.34	59.83 ± 1.20	58.57 ± 4.17	65.07 ± 0.99	48.14 ± 3.68	82.18 ± 1.84	80.41 ± 8.77	67.36 ± 2.05

Table EC1.6:Fortnightly average variations of Eh in the sediment (mv.)

	0	15	30	45	60	75	90	105	120
Aerated Pond	82.30 ± 0.13	44.67 ± 0.33	53.00 ± 0.17	54.77 ± 0.06	36.53 ± 0.23	72.83 ± 0.07	84.97 ± 0.11	97.43 ± 0.23	87.30 ± 0.06
Non-aerated Pond	72.03 ± 0.06	35.03 ± 0.57	48.73 ± 0.17	47.33 ± 0.17	48.87 ± 0.13	67.50 ± 0.17	57.90 ± 0.23	83.20 ± 0.11	78.67 ± 0.06
Creek	80.94 ± 0.08	38.54 ± 0.80	42.58 ± 0.32	43.41 ± 0.16	56.86 ± 0.24	60.43 ± 0.06	55.00 ± 0.16	76.47 ± 0.16	69.22 ± 0.08

Table EC1.7:Fortnightly average variations of dissolve oxygen in the water. (mg.l⁻¹).

	0	15	30	45	60	75	90	105	120
Aerated Pond	3.53 ± 0.13	3.53 ± 0.33	3.57 ± 0.17	3.53 ± 0.06	4.06 ± 0.23	4.02 ± 0.07	3.04 ± 0.11	2.93 ± 0.23	3.45 ± 0.06
Non-aerated Pond	3.42 ± 0.06	3.30 ± 0.57	3.56 ± 0.17	4.43 ± 0.17	4.81 ± 0.13	4.13 ± 0.17	3.60 ± 0.23	4.39 ± 0.11	2.86 ± 0.06
Creek	3.44 ± 0.08	3.38 ± 0.80	3.27 ± 0.32	3.04 ± 0.16	4.34 ± 0.24	3.76 ± 0.06	4.17 ± 0.16	3.49 ± 0.16	4.68 ± 0.08

Table EC1.8:Fortnightly average variations of BOD in the water (mg.l⁻¹)

	0	15	30	45	60	75	90	105	120
Aerated Pond	3.05 ± 0.23	2.74 ± 0.13	2.59 ± 0.12	2.41 ± 0.35	3.05 ± 0.30	0.30 ± 0.06	1.39 ± 0.13	0.88 ± 0.03	0.62 ± 0.16
Non-aerated Pond	2.63 ± 0.28	2.97 ± 0.17	2.14 ± 0.23	3.20 ± 0.37	3.73 ± 0.11	2.41 ± 0.17	3.01 ± 0.47	2.48 ± 0.52	1.16 ± 0.37
Creek	3.22 ± 0.08	2.71 ± 0.64	2.77 ± 0.08	2.09 ± 0.08	3.17 ± 0.16	2.83 ± 0.80	3.73 ± 0.16	2.49 ± 0.32	3.44 ± 0.23

Figure EC1.5:Fortnightly average variations in Eh of water(mv).

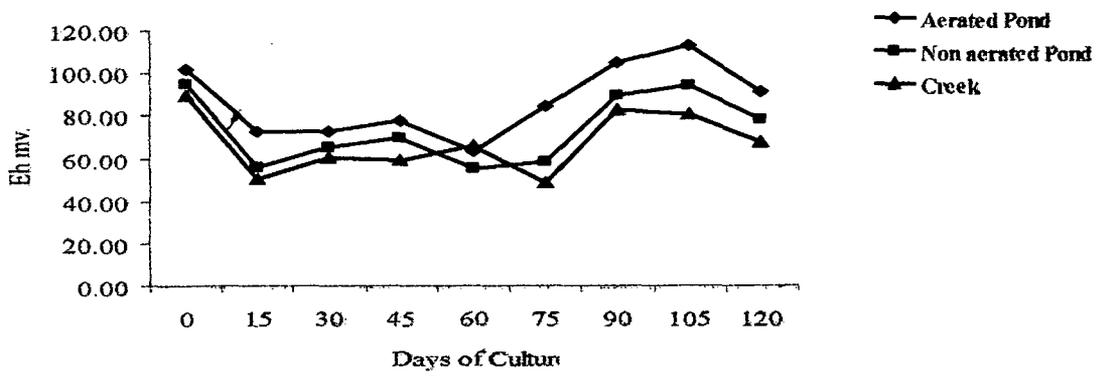


Figure EC1.6:Fortnightly average variations in Eh of sediment(mv).

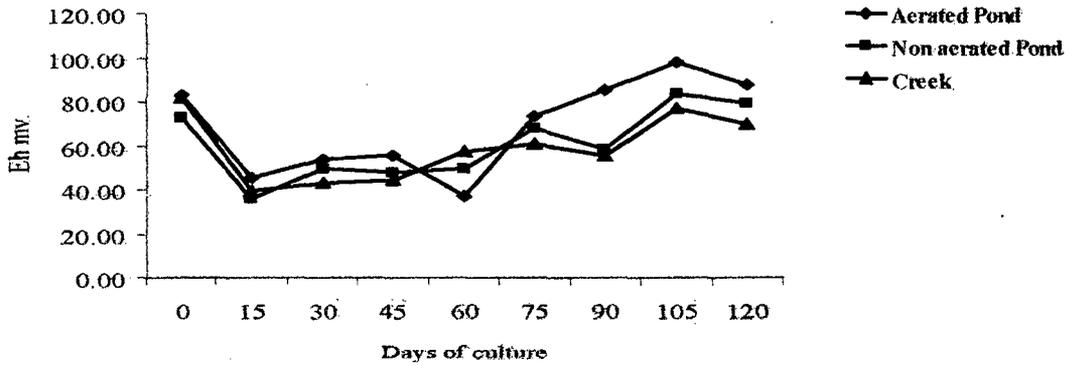


Figure EC1.7:Fortnightly average variations in dissolved oxygen (mg.l⁻¹).

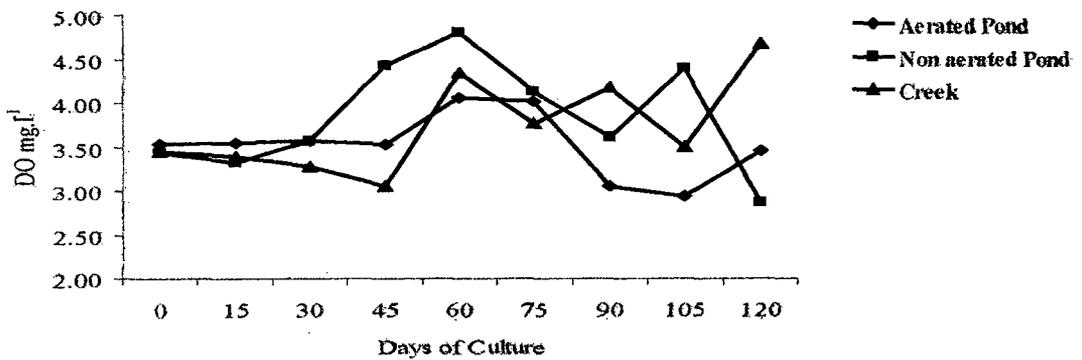


Figure EC1.8:Fortnightly average variations in biochemical oxygen demand(mg.l⁻¹).

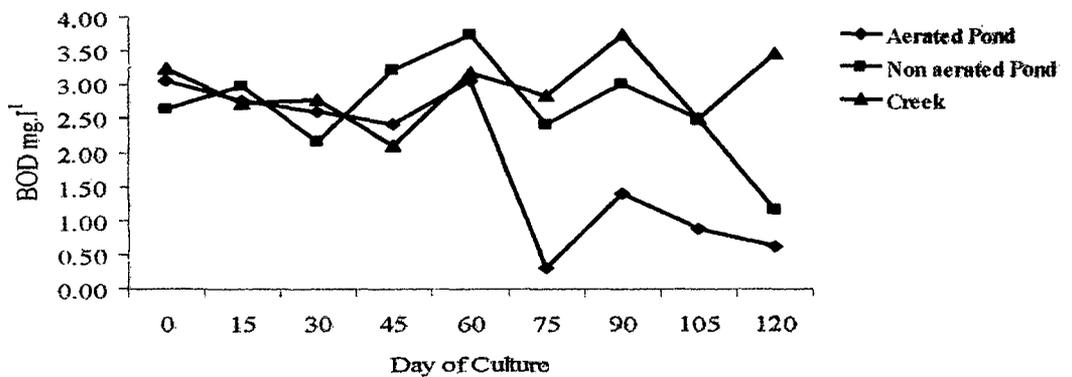


Table EC1.9: Fortnightly average COD variations in the water (mg.l⁻¹).

	0	15	30	45	60	75	90	105	120
Aerated Pond	61.00 ± 3.20	68.00 ± 3.76	10.01 ± 1.53	87.00 ± 7.72	46.03 ± 6.58	32.00 ± 4.60	36.02 ± 6.07	16.07 ± 4.97	14.28 ± 5.02
Non-aerated Pond	50.00 ± 9.86	54.00 ± 9.56	93.01 ± 12.89	89.00 ± 8.55	14.01 ± 6.51	45.00 ± 6.78	16.00 ± 2.04	10.72 ± 3.23	16.07 ± 2.51
Creek	14.50 ± 1.98	45.50 ± 3.68	71.50 ± 3.21	45.00 ± 3.61	19.50 ± 4.55	14.50 ± 2.40	9.50 ± 2.40	5.35 ± 1.06	13.39 ± 1.15

Table EC1.10: Fortnightly average particulate organic carbon variations in the water(mgC.l⁻¹).

	0	15	30	45	60	75	90	105	120
Aerated Pond	3.41 ± 0.55	3.20 ± 1.82	7.36 ± 1.45	7.67 ± 2.57	6.34 ± 0.76	8.96 ± 2.87	9.22 ± 3.31	3.75 ± 1.33	18.63 ± 17.32
Non-aerated Pond	3.85 ± 0.73	4.75 ± 1.66	11.72 ± 2.28	9.31 ± 3.84	5.55 ± 0.85	5.52 ± 2.04	12.56 ± 3.47	3.87 ± 0.87	4.89 ± 2.36
Creek	3.51 ± 1.36	9.15 ± 4.01	7.59 ± 1.61	8.51 ± 2.38	6.57 ± 0.06	4.90 ± 0.82	8.88 ± 2.02	4.50 ± 0.35	6.80 ± 2.33

Table EC1.11: Fortnightly average variations in the sediment organic carbon (%).

	0	15	30	45	60	75	90	105	120
Aerated Pond	2.26 ± 1.25	2.90 ± 1.46	9.24 ± 0.28	1.42 ± 0.49	1.73 ± 1.30	1.24 ± 0.73	1.93 ± 0.83	2.07 ± 2.15	5.43 ± 1.51
Non-aerated Pond	1.01 ± 1.28	3.54 ± 0.44	8.46 ± 0.40	1.19 ± 0.52	1.70 ± 0.35	1.33 ± 0.92	1.97 ± 1.38	2.94 ± 1.18	3.22 ± 1.04
Creek	3.05 ± 1.27	6.01 ± 0.68	9.72 ± 0.59	1.18 ± 1.07	1.52 ± 1.88	1.91 ± 1.36	1.53 ± 1.07	2.68 ± 0.69	4.80 ± 0.49

Table EC1.12: Fortnightly average hydrogen sulphide variations in the water(µg.l⁻¹).

	0	15	30	45	60	75	90	105	120
Aerated Pond	0.60 ± 0.38	0.67 ± 0.36	0.43 ± 0.20	1.29 ± 0.34	0.74 ± 0.26	0.78 ± 0.38	0.62 ± 0.22	1.15 ± 0.41	1.26 ± 0.08
Non-aerated Pond	0.38 ± 0.23	1.03 ± 0.28	0.65 ± 0.17	1.37 ± 0.28	0.81 ± 0.25	0.66 ± 0.28	0.50 ± 0.13	0.63 ± 0.36	0.92 ± 0.18
Creek	0.47 ± 0.18	1.11 ± 0.35	0.68 ± 0.14	1.37 ± 0.17	0.68 ± 0.14	0.66 ± 0.29	0.11 ± 0.12	0.32 ± 0.15	0.28 ± 0.09

Figure EC1.9:Fortnightly average variations in chemical oxygen demand(mg.l^{-1}).

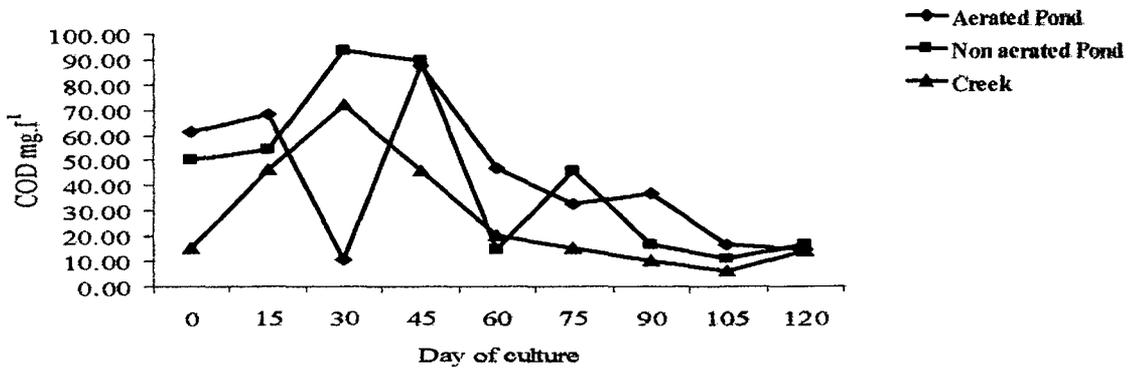


Figure EC1.10:Fortnightly average variations in particulate organic carbon (mgC.l^{-1}).

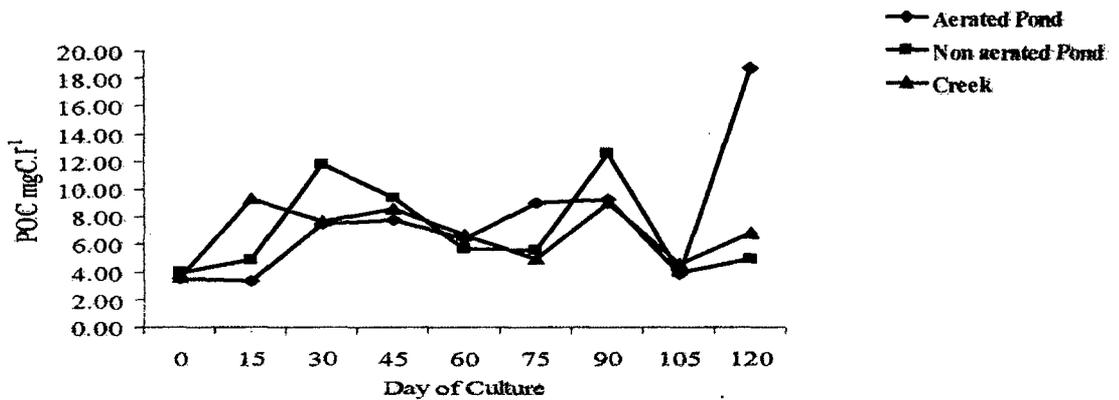


Figure EC1.11:Fortnightly average variations in sediment organic carbon(%).

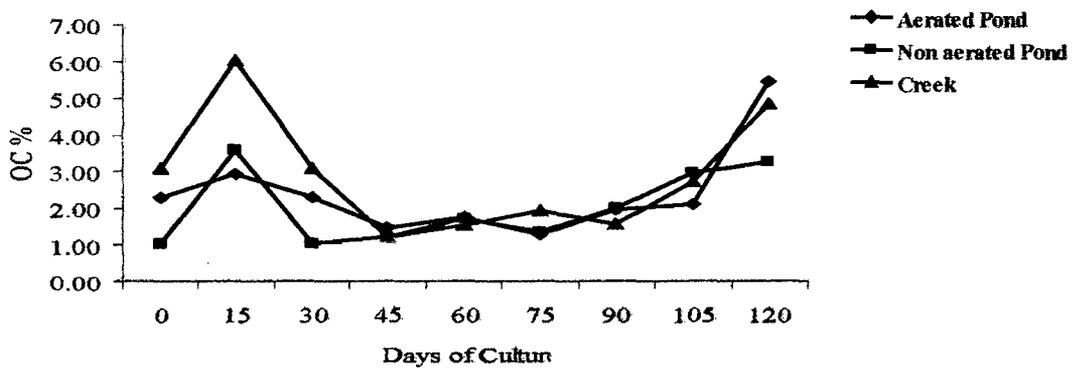


Figure EC1.12:Fortnightly average variations in hydrogen sulphide ($\mu\text{g.l}^{-1}$).

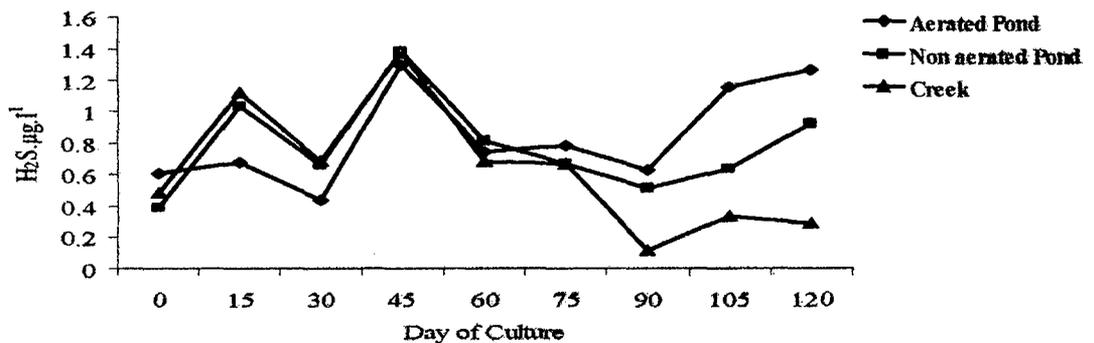


Table EC1.13 Fortnightly average nitrate-nitrogen variations in the water ($\mu\text{mol.l}^{-1}$)

	0	15	30	45	60	75	90	105	120
Aerated Pond	2.72	0.70	0.61	0.75	1.07	0.44	0.55	0.61	0.30
	\pm 0.23	\pm 0.33	\pm 0.27	\pm 0.40	\pm 0.09	\pm 0.11	\pm 0.08	\pm 0.11	\pm 0.03
Non-aerated Pond	0.45	0.72	0.98	0.43	0.76	0.53	0.33	0.36	0.75
	\pm 0.26	\pm 0.34	\pm 0.51	\pm 0.26	\pm 0.13	\pm 0.23	\pm 0.06	\pm 0.07	\pm 0.26
Creek	1.00	0.97	2.94	3.57	0.91	0.68	0.51	0.31	0.53
	\pm 0.16	\pm 0.17	\pm 0.53	\pm 0.44	\pm 0.13	\pm 0.08	\pm 0.09	\pm 0.02	\pm 0.17

Table EC1.14 Fortnightly average nitrite-nitrogen variations in the water ($\mu\text{mol.l}^{-1}$)

	0	15	30	45	60	75	90	105	120
Aerated Pond	0.53	0.35	0.39	0.34	0.44	0.06	0.14	0.16	0.11
	\pm 0.11	\pm 0.03	\pm 0.13	\pm 0.04	\pm 0.06	\pm 0.01	\pm 0.09	\pm 0.08	\pm 0.01
Non-aerated Pond	0.74	0.46	0.44	0.39	0.54	0.08	0.12	0.37	3.42
	\pm 0.06	\pm 0.07	\pm 0.13	\pm 0.13	\pm 0.09	\pm 0.02	\pm 0.03	\pm 0.04	\pm 2.66
Creek	0.58	0.28	2.09	0.48	0.75	0.29	0.19	0.37	1.38
	\pm 0.04	\pm 0.06	\pm 0.25	\pm 0.14	\pm 0.14	\pm 0.06	\pm 0.04	\pm 0.05	\pm 0.09

Table EC1.15 Fortnightly average ammonia-nitrogen variations in the water ($\mu\text{mol.l}^{-1}$)

	0	15	30	45	60	75	90	105	120
Aerated Pond	1.10	0.59	1.93	1.02	0.88	0.31	0.10	0.90	0.15
	\pm 0.03	\pm 0.46	\pm 0.24	\pm 0.07	\pm 0.04	\pm 0.21	\pm 0.06	\pm 0.53	\pm 0.06
Non-aerated Pond	1.21	0.31	2.00	0.31	0.48	0.39	0.38	2.55	4.76
	\pm 0.01	\pm 0.04	\pm 0.17	\pm 0.10	\pm 0.02	\pm 0.12	\pm 0.06	\pm 0.31	\pm 0.13
Creek	1.23	0.69	1.77	1.01	0.81	0.86	0.50	0.95	2.11
	\pm 0.01	\pm 0.14	\pm 0.04	\pm 0.01	\pm 0.13	\pm 0.26	\pm 0.01	\pm 0.10	\pm 0.11

Table EC1.16 Fortnightly average Phosphate-phosphorus variations in the water ($\mu\text{mol.l}^{-1}$)

	0	15	30	45	60	75	90	105	120
Aerated Pond	3.28	2.56	3.97	2.00	1.73	0.57	0.13	0.76	0.99
	\pm 1.06	\pm 0.56	\pm 0.47	\pm 0.32	\pm 0.22	\pm 0.35	\pm 0.04	\pm 0.39	\pm 0.91
Non-aerated Pond	3.06	2.26	3.52	0.72	1.42	3.91	0.74	2.15	1.22
	\pm 0.41	\pm 1.81	\pm 0.37	\pm 0.22	\pm 0.08	\pm 0.18	\pm 0.02	\pm 0.56	\pm 0.60
Creek	2.77	3.50	1.90	3.63	2.44	3.08	0.36	1.49	2.10
	\pm 0.68	\pm 1.85	\pm 0.08	\pm 0.84	\pm 0.16	\pm 0.0	\pm 0.07	\pm 0.21	\pm 2.34

Figure EC1.13:Fortnightly average variations in nitrate-nitrogen($\mu\text{mol.l}^{-1}$).

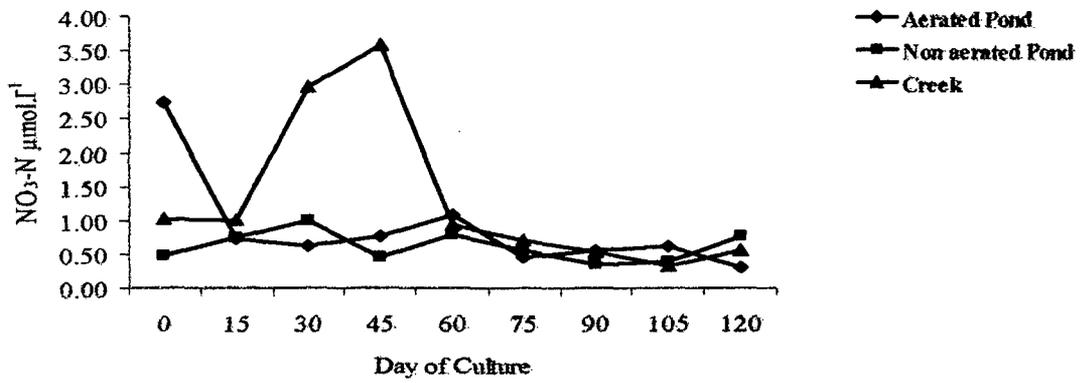


Figure EC1.14:Fortnightly average variations in nitrite-nitrogen ($\mu\text{mol.l}^{-1}$).

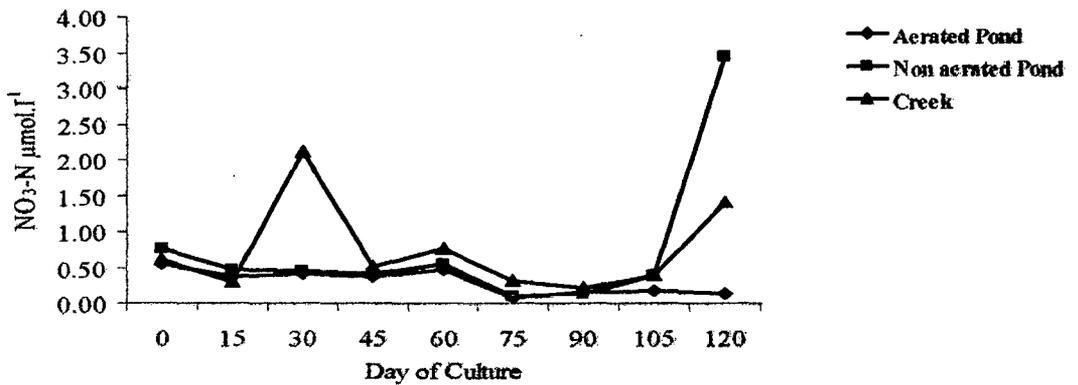


Figure EC1.15:Fortnightly average variations in ammonia-nitrogen($\mu\text{mol.l}^{-1}$).

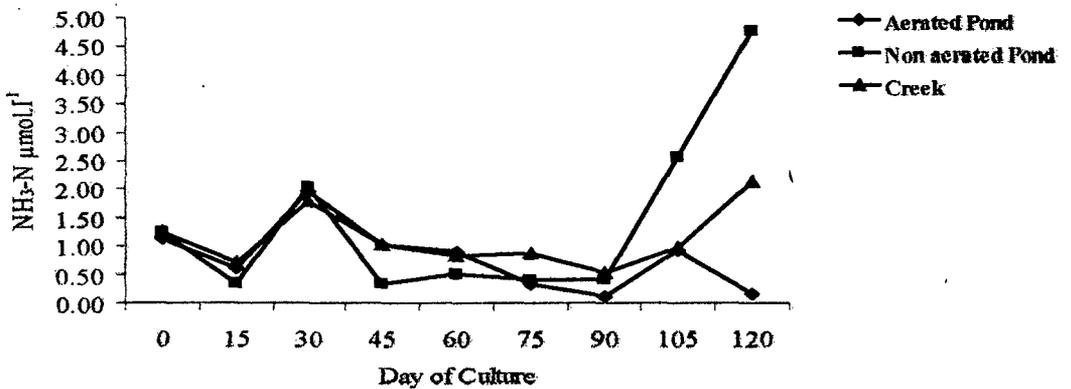


Figure EC1.16:Fortnightly average variations in phosphate-phosphorus($\mu\text{mol.l}^{-1}$).

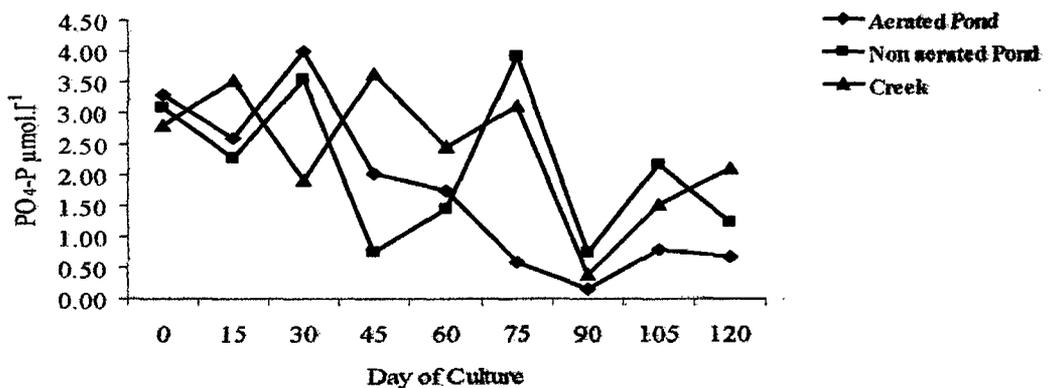


Table EC1.17 Fortnightly average chlorophyll *a* variations in the water (mg.m⁻³)

	0	15	30	45	60	75	90	105	120
Aerated Pond	5.35 ± 0.47	6.41 ± 1.07	3.20 ± 0.0	1.78 ± 0.62	1.69 ± 0.21	1.42 ± 0.62	3.72 ± 0.03	4.63 ± 1.63	4.98 ± 0.62
Non-aerated Pond	5.03 ± 2.55	4.98 ± 2.83	3.20 ± 1.85	1.90 ± 0.79	1.31 ± 0.53	2.02 ± 1.32	4.00 ± 2.37	3.80 ± 1.60	5.46 ± 3.02
Creek	4.83 ± 0.76	4.51 ± 0.50	3.68 ± 1.26	4.16 ± 0.27	1.14 ± 0.08	2.25 ± 0.25	4.52 ± 0.50	3.56 ± 0.0	5.93 ± 0.25

Table EC1.18 Fortnightly average chlorophyll *a* variations in the sediment (µg.g⁻¹)

	0	15	30	45	60	75	90	105	120
Aerated Pond	5.45 ± 0.19	5.16 ± 0.39	3.66 ± 0.14	2.45 ± 0.38	1.38 ± 0.33	2.81 ± 0.18	3.56 ± 0.62	3.92 ± 1.23	14.24 ± 5.56
Non-aerated Pond	4.00 ± 0.04	4.30 ± 1.02	2.94 ± 0.71	2.62 ± 0.15	2.06 ± 0.53	4.26 ± 0.38	3.08 ± 0.82	4.51 ± 0.74	10.68 ± 12.24
Creek	4.63 ± 0.10	4.18 ± 0.35	3.27 ± 0.34	2.54 ± 0.23	1.24 ± 0.08	4.12 ± 0.04	3.64 ± 1.01	4.47 ± 0.25	10.68 ± 3.78

Table EC1.19 Fortnightly average phaeiohytin variations in the water (mg.m⁻³)

	0	15	30	45	60	75	90	105	120
Aerated Pond	3.28 ± 0.34	3.31 ± 0.18	3.30 ± 0.10	2.74 ± 0.55	1.59 ± 0.73	2.86 ± 0.79	3.63 ± 0.34	5.26 ± 0.16	4.02 ± 0.34
Non-aerated Pond	3.17 ± 0.82	3.49 ± 0.39	3.72 ± 0.06	2.83 ± 0.23	1.02 ± 0.10	2.09 ± 0.88	3.57 ± 0.39	5.36 ± 0.22	3.72 ± 0.52
Creek	3.31 ± 0.22	3.11 ± 0.09	3.53 ± 0.01	2.90 ± 0.24	1.06 ± 0.04	1.38 ± 0.25	3.21 ± 0.06	4.28 ± 0.38	4.12 ± 0.24

Table EC1.20 Fortnightly average phaeophytin variations in the sediment (µg.g⁻¹)

	0	15	30	45	60	75	90	105	120
Aerated Pond	3.59 ± 0.18	3.56 ± 0.41	2.72 ± 0.34	1.51 ± 0.16	1.51 ± 0.47	2.60 ± 0.52	3.78 ± 2.63	1.05 ± 0.23	2.89 ± 0.92
Non-aerated Pond	2.91 ± 0.82	2.58 ± 0.67	2.39 ± 0.29	1.72 ± 0.04	1.68 ± 0.22	2.63 ± 0.19	3.29 ± 5.27	2.08 ± 0.04	2.77 ± 0.68
Creek	3.22 ± 0.52	2.88 ± 0.65	2.98 ± 0.33	2.04 ± 0.23	1.65 ± 0.03	1.80 ± 1.20	1.28 ± 18.12	2.05 ± 0.01	2.95 ± 1.28

Figure EC1.17: Fortnightly average variations in chlorophyll *a* in water ($\text{mg}\cdot\text{m}^{-3}$).

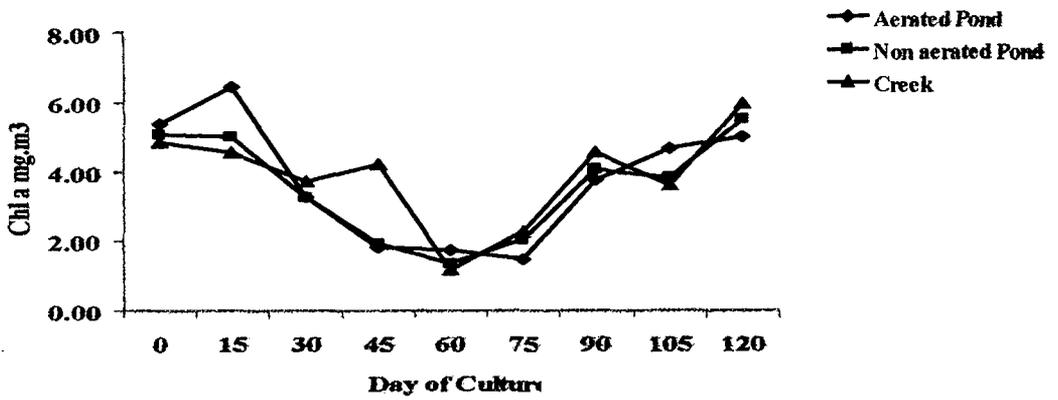


Figure EC1.18: Fortnightly average variations in chlorophyll *a* in sediment ($\mu\text{g}\cdot\text{g}^{-1}$).

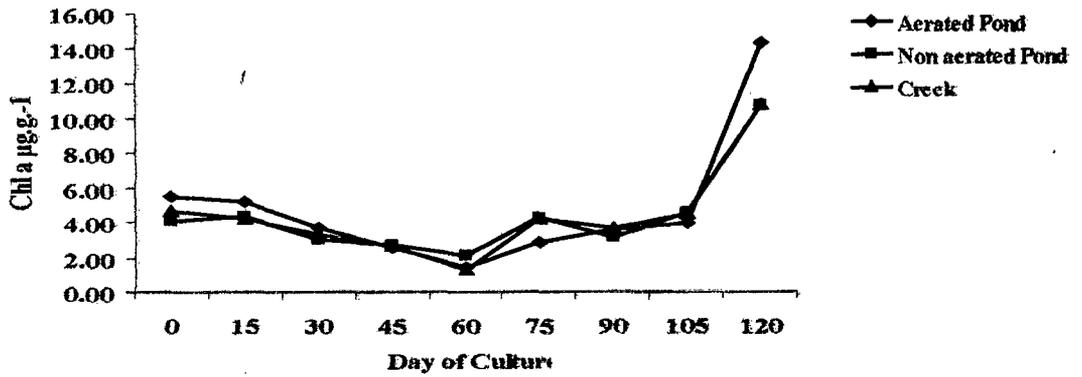


Figure EC1.19: Fortnightly average variations in phaeophytin in water ($\text{mg}\cdot\text{m}^{-3}$).

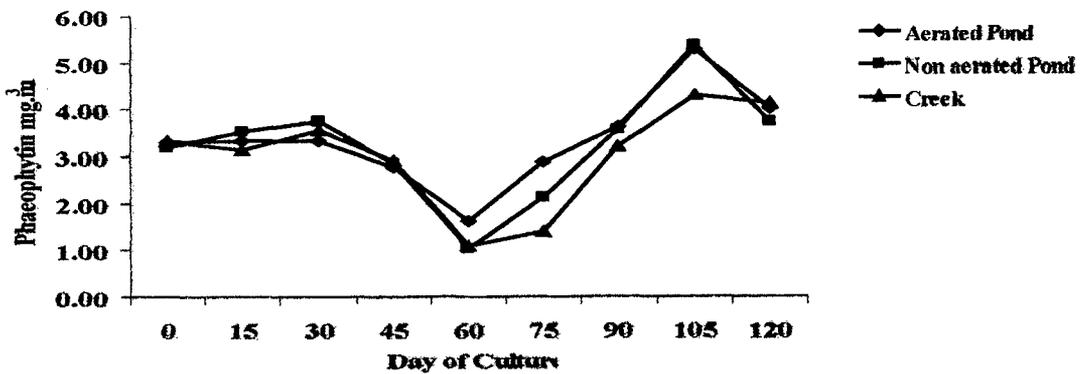


Figure EC1.20: Fortnightly average variations in phaeophytin in sediment ($\mu\text{g}\cdot\text{g}^{-1}$).

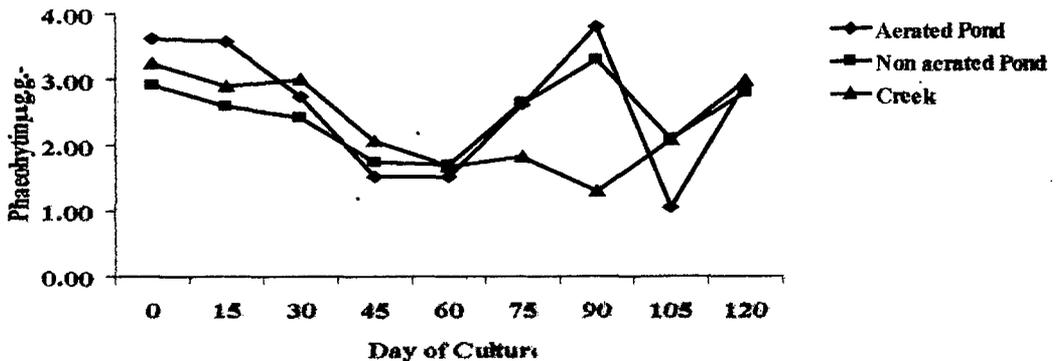


Table EC2.1 Fortnightly average temperature variations in the water ($^{\circ}\text{C}$)

	0	15	30	45	60	75	90	105
Aerated Pond	32.2	32.6	31.6	31.4	30.5	30.4	30.1	28.8
Non-aerated Pond	32.8	32.4	31.6	31.5	30.5	30.3	30.1	28.8
Creek	32.4	32.2	31.9	31.4	30.4	30.1	29.7	28.9

Table EC2.2 Fortnightly average salinity variations in the water (PSU)

	0	15	30	45	60	75	90	105
Aerated Pond	27.38 ± 0.09	27.37 ± 0.08	26.98 ± 0.04	25.12 ± 0.07	25.90 ± 0.09	12.31 ± 0.09	7.24 ± 0.03	4.44 ± 0.08
Non-aerated Pond	27.50 ± 0.05	26.90 ± 0.08	25.78 ± 0.09	25.37 ± 0.04	26.14 ± 0.04	11.20 ± 0.15	8.24 ± 0.05	5.39 ± 0.07
Creek	27.64 ± 0.07	25.50 ± 0.07	22.78 ± 0.03	22.50 ± 0.01	21.57 ± 0.04	0.88 ± 0.00	0.50 ± 0.04	0.28 ± 0.04

Table EC2.3 Fortnightly average pH variations in the water

	0	15	30	45	60	75	90	105
Aerated Pond	7.9 ± 0.07	7.65 ± 0.05	7.20 ± 0.04	7.20 ± 0.10	7.32 ± 0.04	7.48 ± 0.05	7.60 ± 0.02	8.30 ± 0.03
Non-aerated Pond	7.73 ± 0.05	7.65 ± 0.06	7.42 ± 0.04	7.20 ± 0.07	7.20 ± 0.07	8.55 ± 0.03	8.50 ± 0.01	8.48 ± 0.30
Creek	7.63 ± 0.10	7.50 ± 0.10	7.67 ± 0.06	7.72 ± 0.09	7.42 ± 0.14	7.22 ± 0.04	6.90 ± 0.30	6.88 ± 0.04

Table EC2.4 Fortnightly average pH variations in the sediment

	0	15	30	45	60	75	90	105
Aerated Pond	7.32 ± 0.03	7.40 ± 0.09	7.10 ± 0.05	7.30 ± 0.08	7.20 ± 0.04	7.15 ± 0.06	7.11 ± 0.02	7.76 ± 0.07
Non-aerated Pond	7.28 ± 0.03	7.30 ± 0.08	6.42 ± 0.15	7.30 ± 0.12	7.63 ± 0.07	7.21 ± 0.04	7.10 ± 0.01	7.23 ± 0.04
Creek	7.76 ± 0.13	7.65 ± 0.10	6.95 ± 0.14	6.80 ± 1.33	-7.10 ± 0.17	7.20 ± 0.06	7.63 ± 0.11	7.85 ± 0.07

Figure EC2.1: Fortnightly average variations in temperature ($^{\circ}\text{C}$).

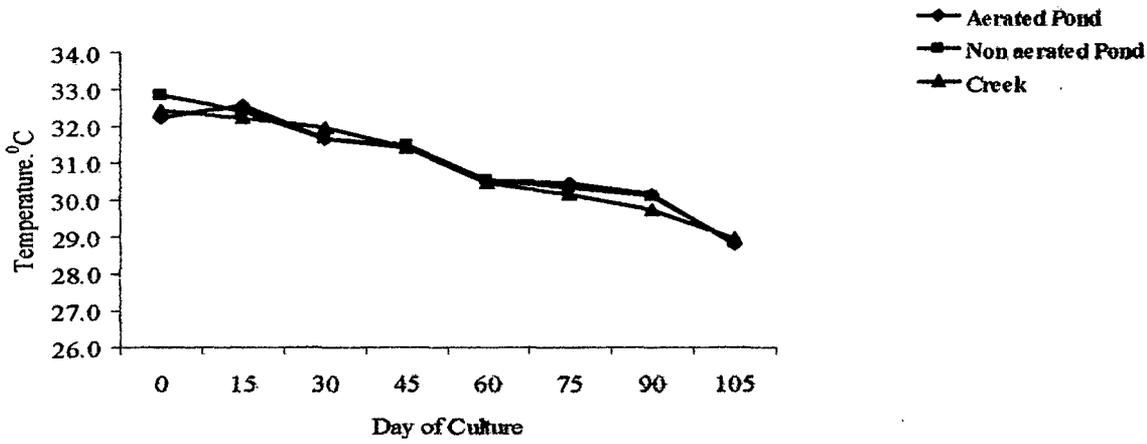


Figure EC2.2: Fortnightly average variations in salinity (PSU).

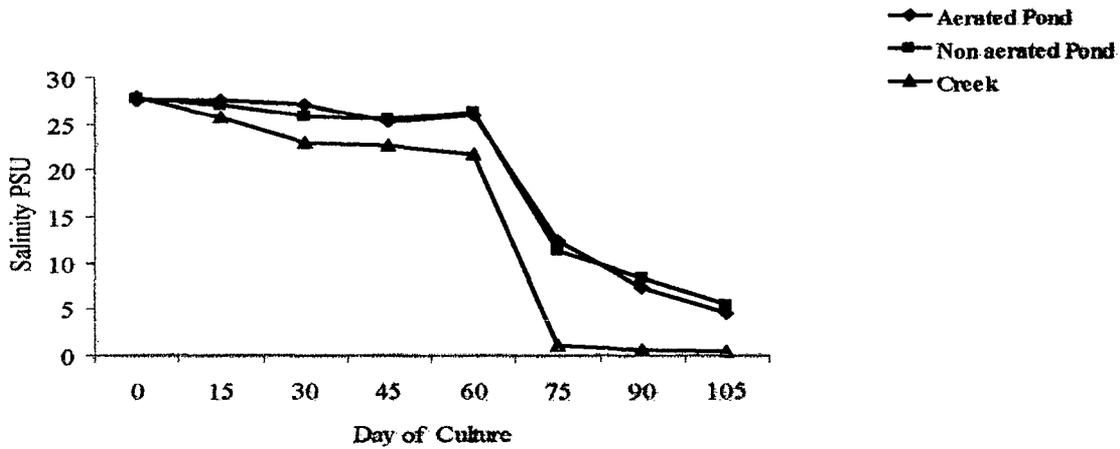


Figure EC2.3: Fortnightly average variations in pH of water.

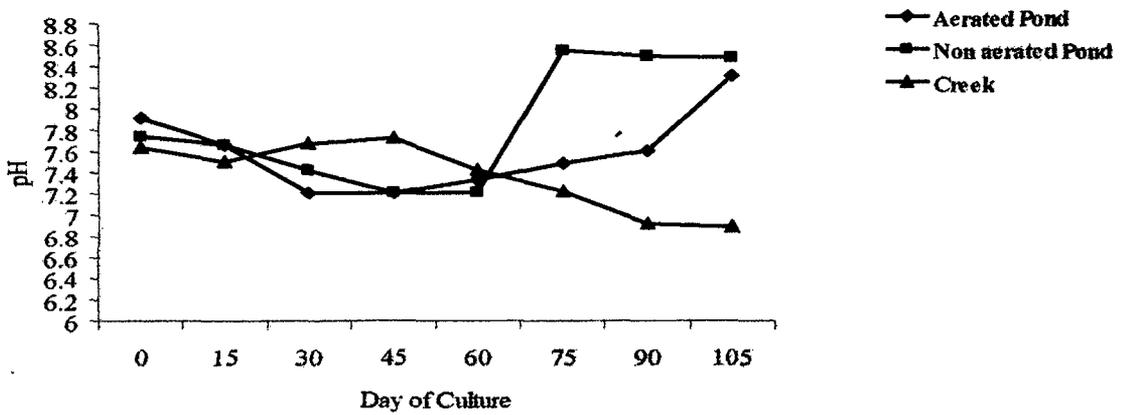


Figure EC2.4: Fortnightly average variations in pH of sediment.

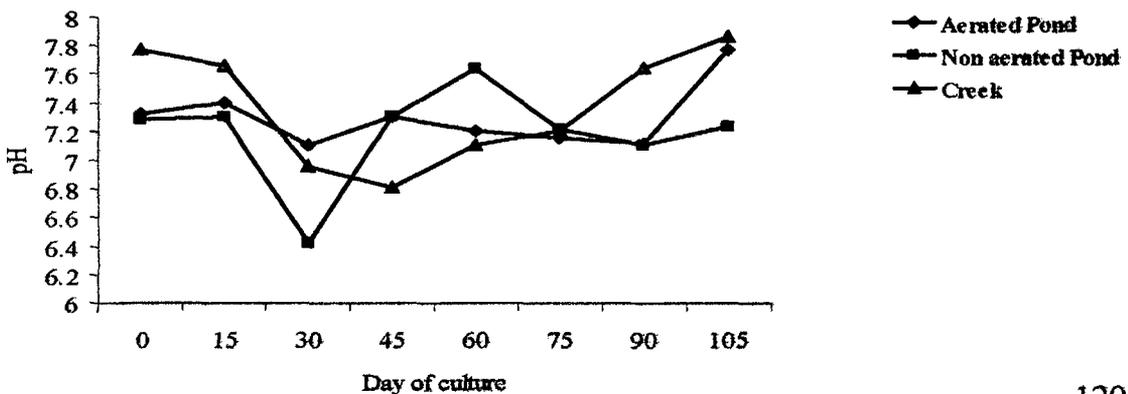


Table EC2.5 Fortnightly average Eh in the water.(mv.)

	0	15	30	45	60	75	90	105
Aerated Pond	70.43 ± 0.82	51.83 ± 0.09	104.60 ± 0.90	88.24 ± 0.77	122.47 ± 1.18	145.07 ± 6.93	111.87 ± 1.75	98.30 ± 0.44
Non-aerated Pond	67.43 ± 0.91	32.50 ± 1.31	104.37 ± 1.35	27.77 ± 3.29	116.40 ± 6.95	140.47 ± 11.62	106.00 ± 3.37	99.00 ± 3.55
Creek	62.90 ± 1.98	28.80 ± 0.57	99.90 ± 3.30	30.25 ± 0.04	113.90 ± 4.04	133.45 ± 5.96	97.35 ± 1.06	96.95 ± 0.0

Table EC2.6 Fortnightly average Eh in the sediment.(mv.)

	0	15	30	45	60	75	90	105
Aerated Pond	96.27 ± 4.88	88.60 ± 4.46	95.00 ± 2.02	78.20 ± 2.06	118.07 ± 1.72	128.87 ± 2.05	103.77 ± 1.39	115.57 ± 1.49
Non-aerated Pond	94.77 ± 1.68	24.97 ± 5.48	106.47 ± 3.19	35.03 ± 1.38	115.30 ± 2.21	126.60 ± 3.95	96.53 ± 4.22	100.40 ± 2.14
Creek	74.35 ± 1.06	42.45 ± 0.79	106.15 ± 0.57	53.20 ± 2.04	107.35 ± 2.50	124.05 ± 4.24	86.15 ± 1.34	96.85 ± 3.37

Table EC2.7 Fortnightly average dissolved oxygen in the water. (mg.l⁻¹)

	0	15	30	45	60	75	90	105
Aerated Pond	3.40 ± 0.03	3.42 ± 0.07	3.40 ± 0.03	3.31 ± 0.06	3.14 ± 0.03	3.63 ± 0.03	2.67 ± 0.07	4.64 ± 0.03
Non-aerated Pond	3.08 ± 0.03	3.03 ± 0.12	3.21 ± 0.06	3.48 ± 0.06	3.34 ± 0.09	3.81 ± 0.09	2.70 ± 0.10	4.21 ± 0.03
Creek	2.54 ± 0.09	4.25 ± 0.11	4.17 ± 0.02	3.78 ± 0.18	2.29 ± 0.09	4.11 ± 0.03	3.55 ± 0.04	4.68 ± 0.04

Table EC2.8 Fortnightly average biochemical oxygen demand in the water. (mg.l⁻¹)

	0	15	30	45	60	75	90	105
Aerated Pond	0.98 ± 0.03	1.02 ± 0.01	1.56 ± 0.03	1.37 ± 0.02	1.45 ± 0.03	2.12 ± 0.03	0.88 ± 0.16	1.60 ± 0.12
Non-aerated Pond	1.20 ± 0.07	1.06 ± 0.01	0.90 ± 0.06	1.39 ± 0.01	1.58 ± 0.17	2.46 ± 0.07	1.67 ± 0.07	1.90 ± 0.07
Creek	0.78 ± 0.06	0.56 ± 0.04	2.31 ± 0.08	1.25 ± 0.10	0.48 ± 0.05	2.93 ± 0.17	1.35 ± 0.00	2.48 ± 0.20

Figure EC2.7: Fortnightly average variations in Eh of water (mv).

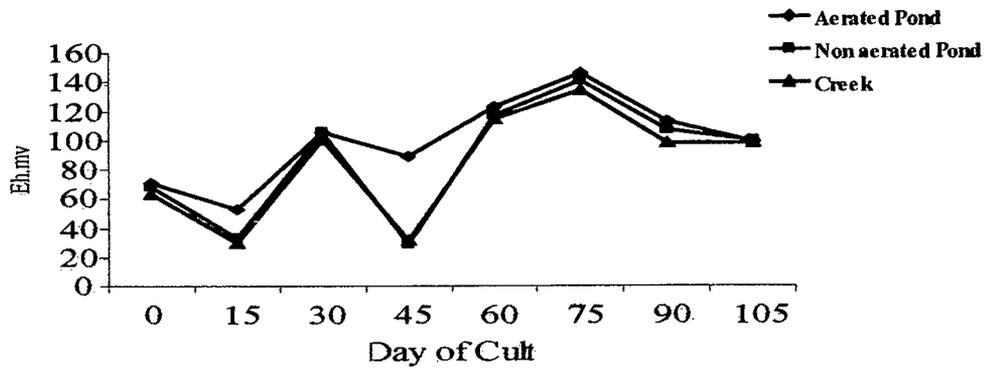


Figure EC2.8: Fortnightly average variations in Eh of sediment (mv).

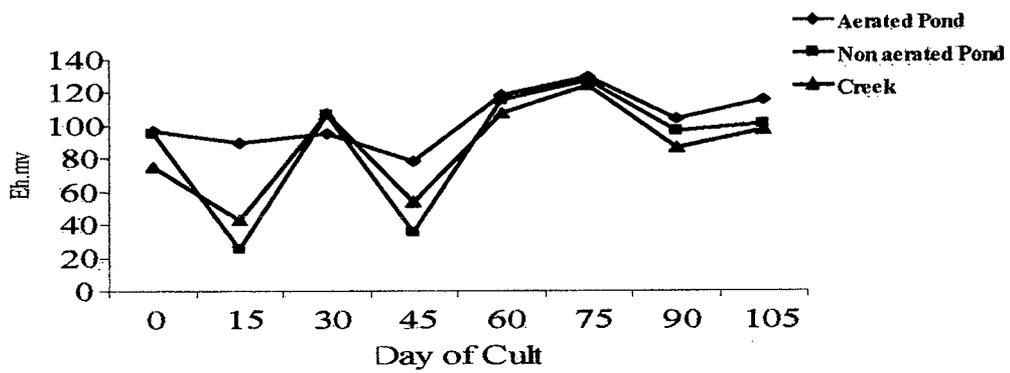


Figure EC2.9: Fortnightly average variations in dissolved oxygen (mg.l⁻¹).

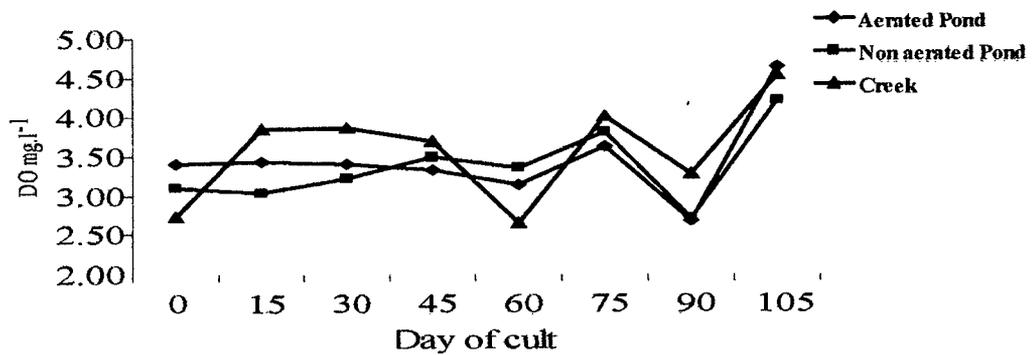


Figure EC2.10: Fortnightly average variations in BOD (mg.l⁻¹).

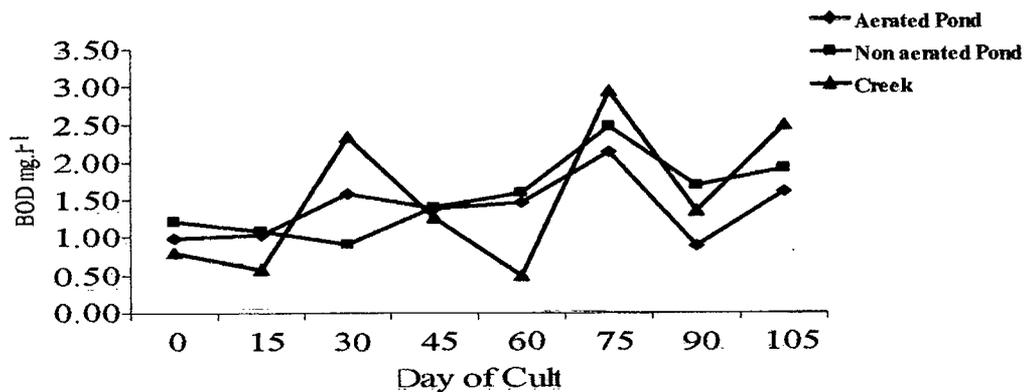


Table EC2.9: Fortnightly average chemical oxygen demand variations in the water (mg.l^{-1})

	0	15	30	45	60	75	90	105
Aerated Pond	67.80	105.40	137.40	121.30	110.70	16.01	35.70	110.70
	±	±	±	±	±	±	±	±
	6.54	4.73	12.82	8.96	4.23	1.54	2.15	2.57
Non-aerated Pond	71.41	123.40	166.04	146.01	114.24	14.22	39.23	114.20
	±	±	±	±	±	±	±	±
	3.00	2.08	8.66	4.67	2.07	1.30	1.87	2.09
Creek	73.22	131.08	169.61	121.62	114.23	8.92	44.61	114.20
	±	±	±	±	±	±	±	±
	2.37	3.30	2.76	2.75	3.14	1.00	3.20	1.41

Table EC2.10: Fortnightly average particulate organic carbon variations in the water (mg.C.l^{-1})

	0	15	30	45	60	75	90	105
Aerated Pond	8.21	5.71	6.38	11.81	4.14	8.34	5.53	6.53
	±	±	±	±	±	±	±	±
	0.05	0.08	0.04	0.32	0.07	0.10	0.06	0.12
Non-aerated Pond	9.06	5.68	6.15	10.79	4.64	7.82	6.15	2.21
	±	±	±	±	±	±	±	±
	0.03	0.04	0.07	0.17	0.14	0.12	0.17	0.08
Creek	7.42	6.58	7.12	7.87	5.96	8.61	9.65	10.55
	±	±	±	±	±	±	±	±
	0.07	0.11	0.05	0.06	0.10	0.08	0.09	0.21

Table EC2.11: Fortnightly average organic carbon variations in the sediment (%)

	0	15	30	45	60	75	90	105
Aerated Pond	3.31	1.56	1.84	1.84	1.57	1.75	3.77	5.22
	±	±	±	±	±	±	±	±
	0.85	0.36	0.12	0.29	0.24	0.18	0.51	0.24
Non-aerated Pond	2.99	0.93	1.15	1.15	2.53	2.48	4.38	6.04
	±	±	±	±	±	±	±	±
	0.19	0.11	0.09	0.04	0.24	0.25	0.16	0.12
Creek	4.43	2.35	1.72	1.38	1.10	1.79	4.83	6.67
	±	±	±	±	±	±	±	±
	0.25	0.07	0.14	0.04	0.04	0.08	0.06	0.11

Table EC2.12: Fortnightly average hydrogen sulphide variations in the water ($\mu\text{g.l}^{-1}$)

	0	15	30	45	60	75	90	105
Aerated Pond	0.03	0.63	0.49	0.64	0.60	0.54	0.75	0.80
	±	±	±	±	±	±	±	±
	0.07	0.07	0.08	0.10	0.05	0.05	0.09	0.03
Non-aerated Pond	0.03	0.54	0.43	0.47	0.52	0.59	0.65	0.75
	±	±	±	±	±	±	±	±
	0.02	0.05	0.06	0.04	0.04	0.08	0.06	0.06
Creek	0.01	0.32	0.30	0.34	0.49	0.49	0.60	0.69
	±	±	±	±	±	±	±	±
	0.01	0.13	0.01	0.03	0.07	0.01	0.06	0.11

Figure EC2.9: Fortnightly average variations in COD (mg.l^{-1}).

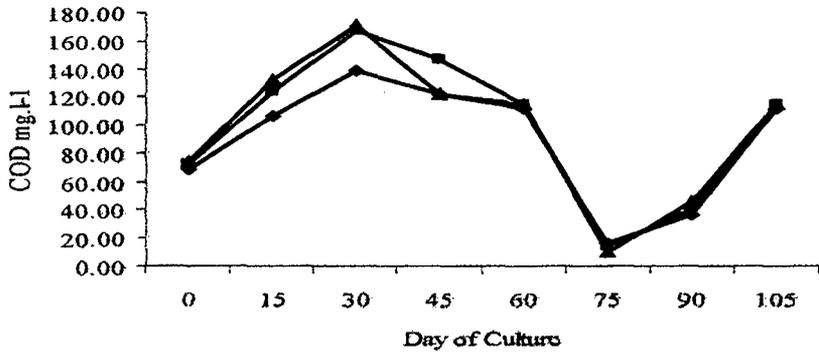


Figure EC2.10: Fortnightly average variations in POC (mg.C.l^{-1}).

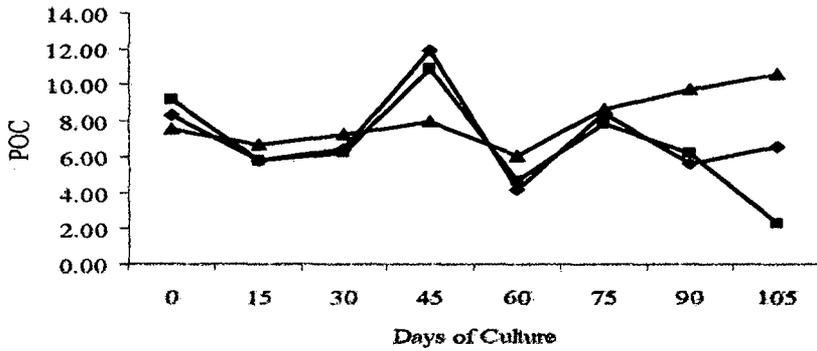


Figure EC2.11: Fortnightly average variations in sediment organic carbon(%).

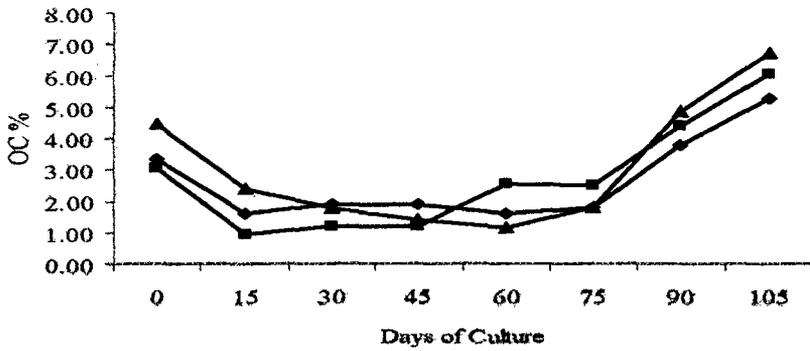


Figure EC2.12: Fortnightly average variations in hydrogen-sulphide ($\mu\text{g.l}^{-1}$).

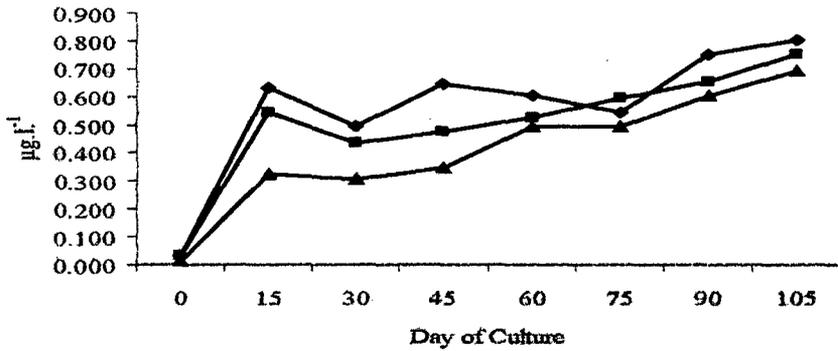


Table EC2.13: Fortnightly average nitrate-nitrogen variations in the water. ($\mu\text{mol.l}^{-1}$)

	0	15	30	45	60	75	90	105
Aerated Pond	0.05	0.04	0.14	0.11	0.72	1.59	0.27	8.48
	±	±	±	±	±	±	±	±
	0.01	0.01	0.10	0.04	0.04	1.18	0.02	0.04
Non-aerated Pond	0.08	0.15	0.15	0.24	0.64	3.19	0.33	4.88
	±	±	±	±	±	±	±	±
	0.01	0.04	0.01	0.04	0.03	0.05	0.02	0.21
Creek	0.38	0.22	0.47	0.32	4.64	3.46	4.80	0.56
	±	±	±	±	±	±	±	±
	0.03	0.00	0.01	0.00	0.01	0.00	0.01	0.01

Table EC2.14: Fortnightly average nitrite-nitrogen variations in the water. ($\mu\text{mol.l}^{-1}$)

	0	15	30	45	60	75	90	105
Aerated Pond	0.30	0.34	1.75	1.27	1.03	1.20	0.26	4.12
	±	±	±	±	±	±	±	±
	0.02	0.01	2.44	0.15	0.04	0.03	0.02	0.06
Non-aerated Pond	0.47	0.38	0.44	0.83	0.83	1.53	0.83	2.04
	±	±	±	±	±	±	±	±
	0.05	0.01	0.02	0.15	0.02	0.05	0.05	0.04
Creek	0.23	0.40	0.25	0.53	4.34	0.61	0.70	1.15
	±	±	±	±	±	±	±	±
	0.00	0.00	0.00	0.00	0.01	0.01	0.00	0.00

Table EC2.15: Fortnightly average ammonia-nitrogen variations in the water. ($\mu\text{mol.l}^{-1}$)

	0	15	30	45	60	75	90	105
Aerated Pond	0.24	0.26	0.30	0.31	0.39	0.60	0.23	0.41
	±	±	±	±	±	±	±	±
	0.02	0.01	0.02	0.02	0.03	0.04	0.03	0.05
Non-aerated Pond	0.29	0.24	0.30	0.26	0.33	0.18	0.20	0.17
	±	±	±	±	±	±	±	±
	0.01	0.01	0.02	0.02	0.02	0.03	0.02	0.05
Creek	0.12	0.22	0.14	0.22	0.33	0.28	0.31	0.40
	±	±	±	±	±	±	±	±
	0.01	0.00	0.01	0.01	0.01	0.01	0.01	0.01

Table EC2.16: Fortnightly average phosphate-phosphorus variations in the water. ($\mu\text{mol.l}^{-1}$)

	0	15	30	45	60	75	90	105
Aerated Pond	1.08	1.23	0.84	0.85	2.46	1.55	0.30	1.43
	±	±	±	±	±	±	±	±
	0.02	0.09	0.04	0.03	0.15	0.22	0.05	0.05
Non-aerated Pond	1.92	1.49	0.94	0.94	2.75	0.92	1.49	0.87
	±	±	±	±	±	±	±	±
	0.05	0.08	0.03	0.03	0.07	0.08	0.09	0.09
Creek	0.92	1.65	1.02	0.99	2.00	0.94	1.26	0.94
	±	±	±	±	±	±	±	±
	0.00	0.01	0.01	0.00	0.01	0.00	0.02	0.01

Figure EC2.13: Fortnightly average variations in nitrate-nitrogen ($\mu\text{mol.l}^{-1}$).

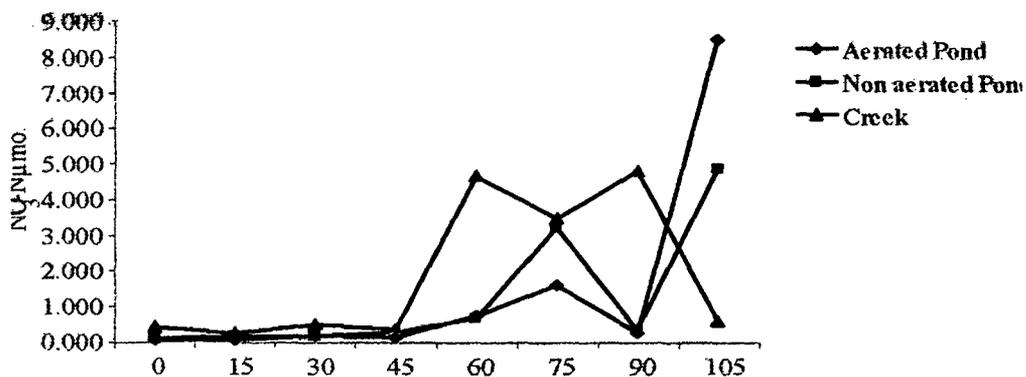


Figure EC2.13: Fortnightly average variations in nitrite-nitrogen ($\mu\text{mol.l}^{-1}$).

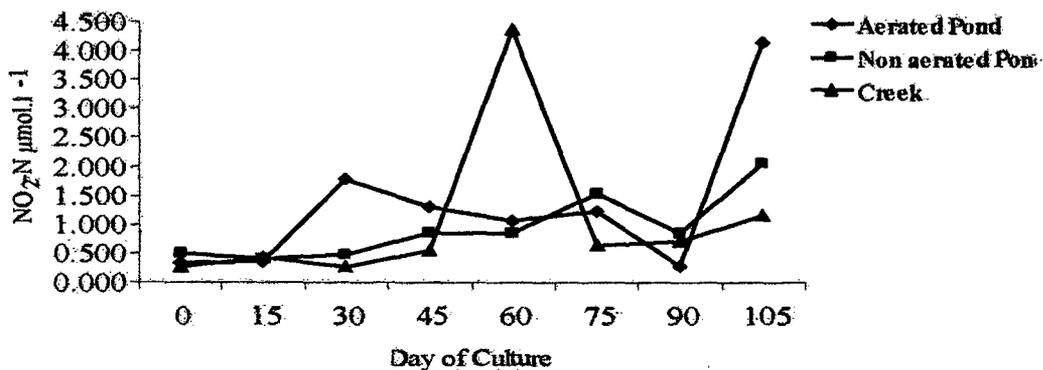


Figure EC2.14: Fortnightly average variations in ammonia-nitrogen ($\mu\text{mol.l}^{-1}$).

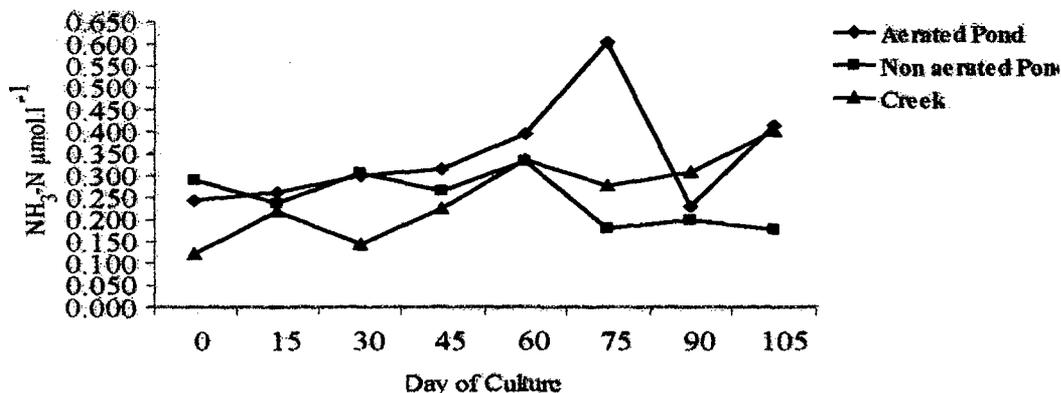


Figure EC2.15: Fortnightly average variations in phosphate-phosphorus ($\mu\text{mol.l}^{-1}$).

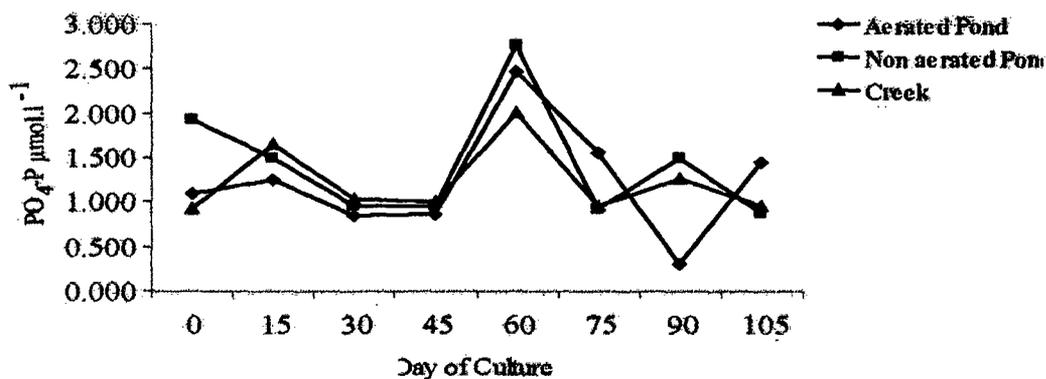


Table EC2.17: Fortnightly average chlorophyll *a* variations in the water. ($\text{mg}\cdot\text{m}^{-3}$)

	0	15	30	45	60	75	90	105
Aerated Pond	14.95	8.70	9.61	6.80	30.97	2.14	19.22	55.54
	±	±	±	±	±	±	±	±
	0.29	0.31	0.21	0.15	1.26	0.04	1.43	3.31
Non-aerated Pond	24.56	11.50	3.20	10.50	23.50	4.27	33.11	20.29
	±	±	±	±	±	±	±	±
	0.38	0.35	0.06	1.00	1.13	0.22	0.81	1.10
Creek	8.54	6.25	4.27	3.25	1.07	1.07	4.27	5.34
	±	±	±	±	±	±	±	±
	0.00	0.04	0.08	0.04	0.03	0.01	0.05	0.00

Table EC2.18: Fortnightly average chlorophyll *a* variations in the sediment. ($\mu\text{g}\cdot\text{g}^{-1}$)

	0	15	30	45	60	75	90	105
Aerated Pond	4.27	5.27	5.34	5.98	5.34	1.18	8.54	11.75
	±	±	±	±	±	±	±	±
	0.13	0.03	0.05	0.17	0.08	0.03	0.08	1.76
Non-aerated Pond	3.69	4.16	3.46	3.51	3.23	1.12	7.61	7.94
	±	±	±	±	±	±	±	±
	0.51	0.06	0.05	0.10	0.08	0.06	0.26	0.07
Creek	3.20	4.11	3.20	3.40	3.20	1.07	7.48	7.48
	±	±	±	±	±	±	±	±
	0.07	0.00	0.00	0.01	0.03	0.06	0.10	0.11

Table EC2.19: Fortnightly average phaeophytin variations in the water. ($\text{mg}\cdot\text{m}^{-3}$)

	0	15	30	45	60	75	90	105
Aerated Pond	1.50	1.20	1.30	1.40	1.30	3.10	9.93	8.01
	±	±	±	±	±	±	±	±
	0.08	0.02	0.04	0.05	0.03	0.04	0.06	0.07
Non-aerated Pond	0.98	0.85	0.98	0.73	0.98	3.10	6.56	6.56
	±	±	±	±	±	±	±	±
	0.09	0.06	0.07	0.04	0.11	0.14	0.29	0.10
Creek	1.92	3.64	18.16	8.28	2.67	12.39	4.27	0.85
	±	±	±	±	±	±	±	±
	0.00	0.00	0.38	0.05	0.05	0.08	0.11	0.11

Table EC2.20: Fortnightly average phaeophytin variations in the sediment. ($\mu\text{g}\cdot\text{g}^{-1}$)

	0	15	30	45	60	75	90	105
Aerated Pond	16.66	20.60	8.66	15.50	26.08	25.10	30.33	47.31
	±	±	±	±	±	±	±	±
	0.31	0.41	0.24	0.38	0.41	0.24	0.65	1.96
Non-aerated Pond	6.51	18.42	10.25	17.66	25.95	6.41	22.30	22.43
	±	±	±	±	±	±	±	±
	0.53	0.71	0.83	0.76	0.77	0.45	0.66	0.79
Creek	16.98	14.28	56.60	42.30	60.66	19.97	28.20	32.36
	±	±	±	±	±	±	±	±
	0.04	0.04	1.12	0.00	0.79	0.06	0.21	0.00

Figure EC2.17: Fortnightly average variations in chlorophyll a in water (mg.m^{-3})

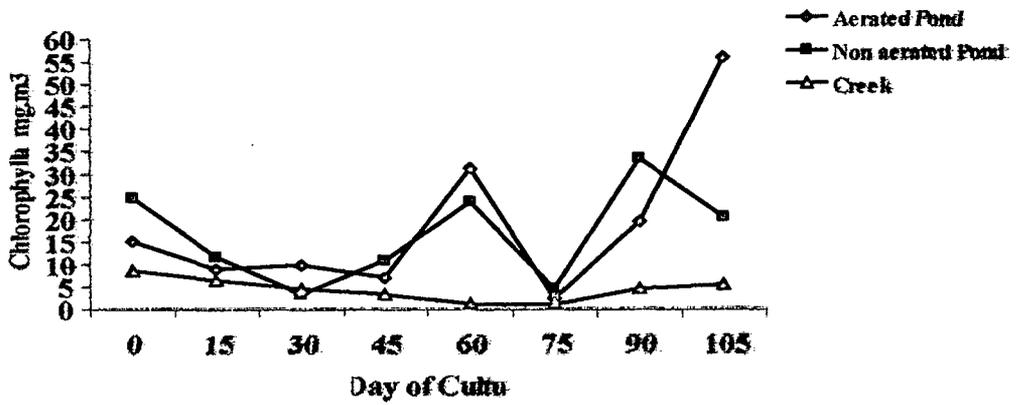


Figure 2.18: Fortnightly average variations in chlorophyll a sediment ($\mu\text{g.g}^{-1}$)

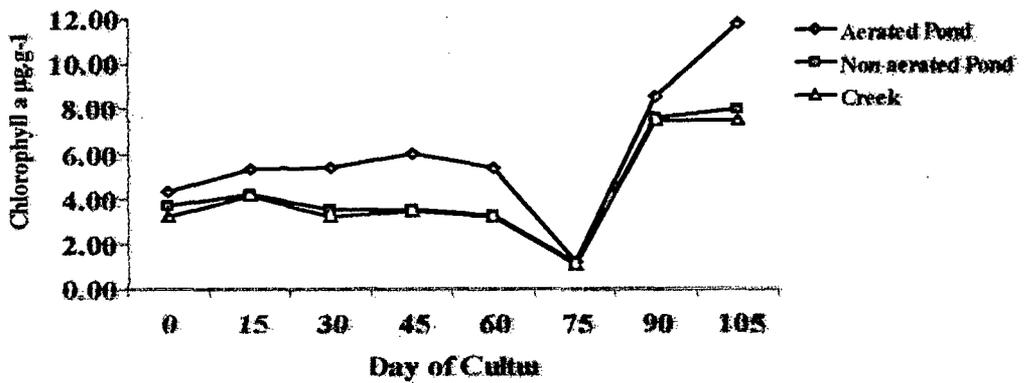


Figure EC2.19: Fortnightly average variations in phaeophytin in water (mg.m^{-3})

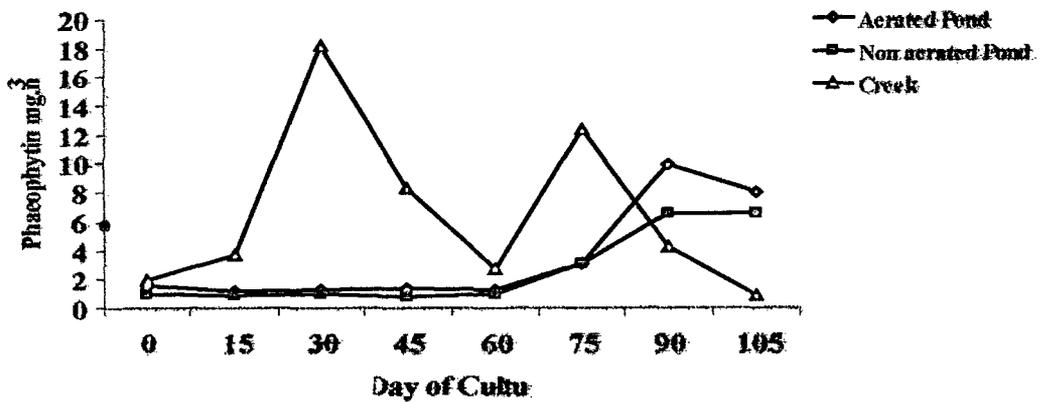
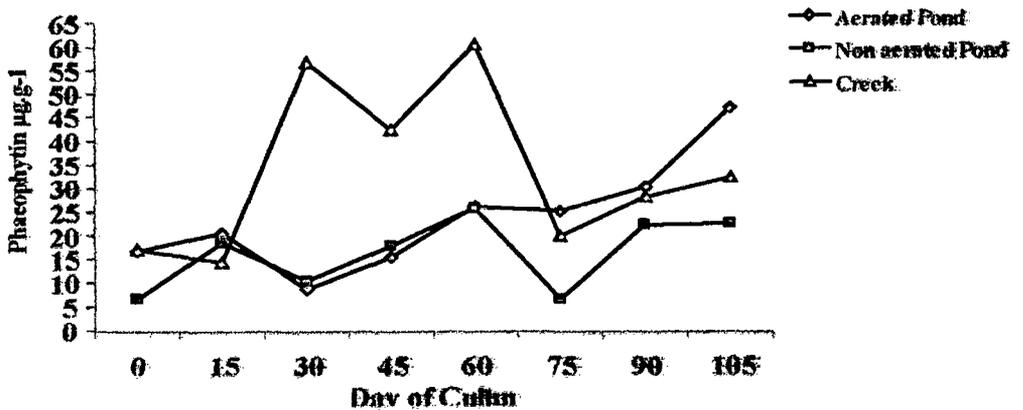


Figure EC2.20: Fortnightly average variations in phaeophytin in sediment ($\mu\text{g.g}^{-1}$)



4.1.2 Factor Analysis

Having considered, many individual relationships, multiple regression analysis could show only which parameters are linearly related and which amongst those affected the criterion variable. Though these relationships gives an idea about the behavior of each parameter during the culture period, overall, which parameters varied most, in aerated non-aerated pond as well as creek during two culture periods would give the more clear understanding of the situation and to understand the process as a whole. All together out of 20 physico-chemical parameters, 16 water quality and 4 soil quality parameters along with the macro, meio and microfauna were subjected to factor analysis using Statistic 5.0 to understand overall process.

Experimental Cycle (EC-1)

Aerated Pond

The first three factors captured major portion (65.02%) of the variance present in the data. It was observed that, first two factors Factor1 and Factor2 (Table EC1.21) explained the variance of 52.24% with 34.29% for Factor1 and 17.95% for Factor2. Factor1 has high loading values for temperature (0.78) pH of sediment (-0.90), E_h of sediment (-0.83), DO (0.79), phaeopigments in the water (-0.84) and macrofauna (-0.79). For Factor 2, high loading values were observed for, pH of the sediment (-0.86), BOD (0.74), NO_3-N (0.79), NO_2-N (0.85), PO_4-P (0.79). The remaining 3 Factors all together explained (29.34 %) variability due to OC (0.82), phaeophytin in the sediment (0.91) and chlorophyll *a* in the sediment (-0.81) in Factor 3,4 and 5 respectively. Therefore it can be said that oxidation reduction processes and degradation of phytoplankton to form phaeophytin in the water and sediment ultimately leading to formation of organic carbon, seems to have been the overall process prevailing in the aeration pond. The changes in these parameters might have been responsible for the changes in the other water quality parameters.

Non-aeration Pond

In non-aerated pond, similar to aerated pond, first three Factors captured the major portion of the variance (63.24%), and the data is given in Table (EC1.22). First two

Factors cumulatively contributed to 50.93% variance, and 30.92% by Factor1 and 20.01% for Factor2 respectively. The examination of loading vector associated with Factor1 showed high values for BOD (-0.80), NO₂-N, NH₃-N, Chlorophyll *a* sediment (0.92), macrofauna (0.74).

For Factor2 high values were observed for pH sediment, microfauna (-0.86). Factor3, Factor4, and Factor5 together found to show 29.91% variance and together almost is equivalent to the variance explained by the Factor1. Factor 3 showed high values for temperature(0.82), E_h of water(-0.81), E_h sediment(-0.71), NO₃-N(0.80), Factor4 showed higher values for pH water (0.84), PO₄-P (0.76), phaeophytin in the water (0.83), Factor5 showed higher values for DO (-0.80), and OC (0.86). The factor loading on the basis of their values indicates that, nutrients from overlying water in the form of NH₃-N, NO₃-N, and PO₄-P were taken up by the benthic microphytobenthos which mainly consisted of the diatoms. The uptake of nutrients and subsequently dead benthic diatoms led to form the organic matter.

Creek

In the creek the highly interrelated process were observed and these interdependencies amongst variable seems to have evolved due to DO changes and BOD in the water. The Factor1 captured the major portion of the variations accounting 28.84% (TableEC1.23). In the Factor1, highest values were shown by DO (0.82), BOD (0.82) indicates that, DO and BOD were perfectly co-related. The subsequent factors are shown in the Table 1.23. From these factors it can be said that, the oxygen consumption by phytoplankton, benthos in the sediment and nitrification by nitrifying bacteria in presence of DO may have prevailed.

Table EC1.21 Factor Loadings (Varimax normalized) for aerated pond EC-1
Extraction: Principal factors (comm.=multiple R-square)
(Marked loadings are > .70)

	Factor	Factor	Factor	Factor	Factor
	1	2	3	4	5
Temperature	0.78	0.40	0.20	0.15	-0.30
Salinity	0.40	0.67	0.16	0.40	0.14
pH water	-0.39	-0.03	0.63	0.00	0.22
pH Sediment	0.14	-0.86	0.17	0.02	0.05
Eh water	-0.90	-0.06	-0.11	-0.04	-0.08
Eh sediment	-0.83	-0.18	-0.04	-0.10	-0.32
DO	0.79	0.02	-0.17	0.02	-0.02
BOD	0.13	0.74	0.05	0.02	0.54
COD	0.25	0.44	-0.64	0.06	0.18
NO3-N	-0.15	0.79	-0.35	0.20	0.05
NO2-N	0.30	0.85	-0.04	0.14	0.21
NH3-N	0.30	0.67	0.53	-0.15	0.21
PO4-P	0.39	0.79	0.31	0.04	0.01
Chl <i>a</i> water	-0.64	0.28	0.22	0.40	-0.13
Chl <i>a</i> sed	-0.33	-0.10	0.16	0.06	-0.81
phaeo water	-0.84	-0.10	0.39	-0.13	-0.11
Phaeo sed	-0.13	0.03	-0.07	0.91	-0.27
OC	0.19	0.13	0.82	0.18	-0.37
POC	0.10	-0.50	0.02	-0.09	-0.80
H2S Water	-0.21	-0.16	-0.21	-0.75	-0.36
Macrofauna	-0.79	-0.31	-0.16	0.12	0.31
Meiofauna	-0.52	-0.01	-0.02	0.03	0.61
Microfauna	-0.59	-0.17	0.27	-0.56	-0.33
Expl. Var	6.06	5.04	2.50	2.23	2.93
Prp. Totl	0.26	0.22	0.11	0.10	0.13

Eigenvalues

Extraction: Principal factors (comm.=multiple R-square)

	Eigenval	% total	Cumul.	Cumul.
		Variance	Eigenval	%
1	7.89	34.29	7.89	34.29
2	4.13	17.95	12.02	52.24
3	2.94	12.78	14.95	65.02
4	2.01	8.72	16.96	73.74
5	1.80	7.84	18.76	81.58

Table EC1.22 Factor Loadings (Varimax normalized) non aerated pond
Extraction: Principal factors (comm.=multiple R-square)
(Marked loadings are > .70)

	Factor	Factor	Factor	Factor	Factor
	1	2	3	4	5
Temperature	-0.20	-0.47	0.82	-0.09	0.04
Salinity	-0.46	-0.58	0.48	0.20	0.26
pH water	0.33	0.04	-0.27	0.84	0.02
pH Sediment	0.09	0.91	0.21	-0.05	0.01
Eh water	0.16	-0.30	-0.81	0.28	0.16
Eh sediment	0.59	-0.10	-0.71	0.07	-0.16
DO	-0.48	0.16	-0.09	-0.18	-0.80
BOD	-0.81	0.10	0.02	-0.30	-0.26
COD	-0.33	-0.55	0.58	0.23	0.07
NO3-N	0.26	0.00	0.80	0.12	0.00
NO2-N	0.92	-0.06	0.12	-0.07	0.12
NH3-N	0.90	-0.05	-0.07	0.34	-0.03
PO4-P	-0.09	-0.43	0.38	0.76	0.06
Chl <i>a</i> water	0.44	-0.29	-0.11	0.30	0.62
Chl <i>a</i> sed	0.92	0.08	-0.05	0.04	0.23
phaeo water	0.22	0.02	-0.37	0.83	0.20
Phaeo sed	0.15	0.01	-0.35	-0.05	0.86
OC	0.09	0.01	0.50	0.66	0.14
POC	-0.37	0.04	0.23	0.09	0.30
H2S Water	0.05	0.08	0.59	-0.03	-0.24
Macrofauna	0.74	0.38	-0.13	0.24	0.41
Meiofauna	-0.23	0.34	0.23	0.30	0.39
Microfauna	0.42	-0.86	0.01	-0.01	0.10
Expl. Var	5.51	3.10	4.32	3.09	2.57
Prp. Totl	0.24	0.13	0.19	0.13	0.11

Eigenvalues

Extraction: Principal factors (comm.=multiple R-square)

		% total	Cumul.	Cumul.
	Eigenval	Variance	Eigenval	%
1	7.11	30.92	7.11	30.92
2	4.60	20.01	11.71	50.93
3	2.83	12.31	14.54	63.24
4	2.30	10.00	16.85	73.24
5	1.75	7.60	18.59	80.84

Table EC1.23 Factor Loadings (Varimax normalized) creek EC-1
Extraction: Principal factors (comm.=multiple R-square)
(Marked loadings are > .70)

	Factor	Factor	Factor	Factor	Factor
	1	2	3	4	5
Temperature	-0.58	0.61	0.18	-0.22	0.06
Salinity	-0.60	0.36	0.14	0.29	0.52
pH water	0.12	0.25	0.18	0.42	0.57
pH Sediment	0.51	-0.14	0.71	-0.17	-0.37
Eh water	0.43	-0.19	-0.49	0.15	0.56
Eh sediment	0.42	-0.11	-0.86	0.15	0.06
DO	0.82	0.13	-0.01	-0.02	-0.42
BOD	0.82	0.15	0.00	0.00	0.14
COD	-0.62	0.51	0.51	-0.02	0.18
NO ₃ -N	-0.76	0.26	0.23	-0.10	0.07
NO ₂ -N	-0.01	0.91	0.05	0.08	0.02
NH ₃ -N	0.00	0.77	-0.26	0.42	-0.19
PO ₄ -P	-0.78	0.04	-0.09	-0.01	-0.37
Chl <i>a</i> water	0.03	0.04	0.01	0.89	0.18
Chl <i>a</i> sed	0.36	0.24	-0.22	0.75	-0.33
phaeo water	0.08	0.08	-0.07	0.82	0.33
Phaeo sed	-0.31	0.61	-0.23	0.56	0.19
OC	-0.12	0.76	0.30	0.30	0.27
POC	-0.10	-0.04	0.91	0.13	-0.05
H ₂ S Water	-0.88	-0.04	0.29	-0.10	-0.22
Macrofauna	0.04	0.12	-0.35	-0.02	0.84
Meiofauna	-0.50	-0.71	0.08	0.04	-0.10
Microfauna	-0.27	0.16	0.39	0.67	-0.38
Expl. Var	5.59	3.99	3.33	3.50	2.73
Prp. Totl	0.24	0.17	0.14	0.15	0.12

Eigenvalues

Extraction: Principal factors (comm.=multiple R-square)

	Eigenval	% total Variance	Cumul. Eigenval	Cumul. %
1	6.63	28.84	6.63	28.84
2	5.24	22.76	11.87	51.60
3	3.13	13.60	15.00	65.20
4	2.25	9.77	17.24	74.97
5	1.91	8.30	19.15	83.28

Experimental Cycle (EC-2)

Aerated Pond

According to the factor analysis carried out, first three factors captured major variations (75.59%) in the data, which is depicted in (Table EC2.21). Factor1 showed high loading value for chlorophyll *a* in the sediment (0.73). The Factor2 showed high value for microbenthos (0.83), this indicates that, benthic microphytobenthos may have played important role in formation of the OC (0.84) in the sediment. Further, in the process of oxidation of organic matter and oxidation of $\text{NH}_3\text{-N}$ by nitrifiers, was found to be responsible for the variations in the DO. The high chlorophyll *a* during EC-2 and its governing parameters like light and nutrient has been discussed earlier with the help of correlation and regression analysis. During this cycle, high microbenthos and phytoplankton abundance is evident and justifies the processes dependent on its abundance at the bottom of the pond.

Non-aerated Pond

According to factor analysis, first three factors amounted for cumulative variance of 68.75% (Table EC2.22). Where, Factor1 explained 41.60% variances due to chlorophyll *a* (0.87), phaeophytin in the water (0.84) and organic carbon (0.79) is an indicative of the process that, high chlorophyll *a* in the form of benthic diatoms on degradation, had contribute to form phaeopigments, and thus, ultimately led to the organic matter formation at the bottom of the pond. Factor2 showed higher values for BOD (0.81), E_h water (0.80), E_h sediment (0.79) and, Factor3 showed pH sediment (0.74) point outs that, degradation of organic matter at the bottom of the pond may have led to changes in the E_h and ultimately changes in the pH of the pond soil.

Creek

In the creek sediment chlorophyll *a* and the nutrient uptake with high values of $\text{NH}_3\text{-N}$ from Factor1 and $\text{NO}_2\text{-N}$ (0.84) and $\text{PO}_4\text{-P}$ (0.84) from the water column were responsible for higher productivity. Further, degradation of chlorophyll *a* to form phaeophytin and the resulting ORP changes shows the interdependence of these parameters in the creek. It was found that, all five factors together explained 88.53% variance (Table EC2.23).

Table EC 2.21 Factor Loadings (Varimax normalized) aerated pond EC-2
Extraction: Principal factors (comm.=multiple R-square)
(Marked loadings are > .70)

	Factor	Factor	Factor	Factor	Factor
	1	2	3	4	5
Temperature	-0.65	-0.33	-0.37	-0.45	-0.24
Salinity	-0.43	-0.65	-0.20	-0.54	-0.08
DO	0.16	-0.30	0.53	0.73	-0.05
BOD	0.07	0.09	0.95	-0.02	-0.07
COD	0.41	-0.85	-0.09	-0.14	-0.06
pH water	-0.03	0.01	-0.10	0.95	0.17
pH sed	0.27	-0.40	0.05	0.80	-0.04
eh water	0.16	0.55	0.66	-0.22	0.25
eh sediment	-0.06	0.40	0.64	0.23	0.57
NO ₃ -N	0.47	-0.09	0.53	0.65	0.05
NO ₂ -N	0.55	-0.30	0.47	0.56	-0.02
PO ₄ -P	0.55	-0.30	0.47	0.56	-0.02
NH ₃ -N	0.01	0.12	0.89	-0.02	-0.13
chl a water	0.49	-0.17	0.05	0.63	0.48
chl a sediment	0.73	-0.08	-0.33	0.53	0.13
Phaeo water	0.49	0.63	-0.18	0.49	0.15
Phaeo sed.	0.44	0.25	0.17	0.68	0.36
POC	-0.13	-0.01	0.13	0.01	-0.84
OC	0.29	0.22	-0.19	0.84	0.08
H ₂ S water	0.85	0.20	0.13	-0.02	0.08
Macrofauna	0.11	-0.20	0.01	-0.83	0.42
Meiofauna	0.08	0.83	0.24	-0.42	-0.04
Microfauna	0.21	0.95	0.06	-0.05	-0.04
Expl. Var	3.84	4.38	4.04	6.70	1.81
Prp. Totl	0.17	0.19	0.18	0.29	0.08

Eigenvalues

Extraction: Principal factors (comm.=multiple R-square)

		% total	Cumul.	Cumul.
	Eigenval	Variance	Eigenval	%
1	9.27	40.29	9.27	40.29
2	4.93	21.43	14.19	61.71
3	3.19	13.87	17.38	75.59
4	2.04	8.86	19.42	84.45
5	1.34	5.83	20.76	90.28

Table EC-2.22 Factor Loadings (Varimax normalized) non aerated pond EC-2
 Extraction: Principal factors (comm.=multiple R-square)
 (Marked loadings are > .70)

	Factor	Factor	Factor	Factor	Factor
	1	2	3	4	5
Temperature	-0.47	-0.16	-0.24	-0.77	0.10
Salinity	-0.69	-0.46	0.11	-0.46	-0.07
DO	-0.02	0.15	-0.15	0.84	-0.09
BOD	0.22	0.81	0.11	0.45	-0.14
COD	-0.27	-0.81	-0.01	0.18	-0.14
pH water	0.62	0.56	-0.30	0.23	0.24
pH sed	-0.04	0.11	0.74	0.10	0.02
Eh water	0.02	0.80	-0.01	0.23	0.23
Eh sediment	0.04	0.79	0.00	0.09	0.12
NO ₃ -N	0.31	0.35	-0.15	0.79	-0.03
NO ₂ -N	0.36	0.48	-0.07	0.67	0.28
PO ₄ -P	-0.32	-0.36	0.31	-0.19	-0.75
NH ₃ -N	-0.20	0.05	0.82	-0.33	-0.27
chl a water	0.63	0.08	0.60	-0.35	0.21
chl a sediment	0.87	-0.28	0.16	0.13	0.23
Phaeo water	0.84	0.31	-0.01	0.32	0.16
Phaeo sed.	0.31	-0.34	0.71	0.35	0.22
POC	-0.34	-0.02	-0.26	-0.54	-0.65
OC	0.79	0.33	0.16	0.28	0.14
H ₂ S water	0.30	0.04	0.10	0.75	0.20
Macrofauna	-0.83	-0.07	0.29	-0.26	0.18
Meiofauna	0.05	-0.40	-0.08	0.31	-0.80
Microfauna	0.02	0.84	0.03	0.24	0.11
Expl. Var	5.04	4.91	2.63	4.62	2.25
Prp. Totl	0.22	0.21	0.11	0.20	0.10

Eigenvalues (avgcy4p2.sta)

Extraction: Principal factors (comm.=multiple R-square)

		% total	Cumul.	Cumul.
	Eigenval	Variance	Eigenval	%
1	9.57	41.60	9.57	41.60
2	3.43	14.89	12.99	56.49
3	2.82	12.26	15.81	68.75
4	2.13	9.25	17.94	78.00
5	1.51	6.57	19.45	84.57

Table EC2.23 Factor Loadings (Varimax normalized) creek EC-2
 Extraction: Principal factors (comm =multiple R-square)
 (Marked loadings are > .70)

	Factor	Factor	Factor	Factor	Factor
	1	2	3	4	5
Temperature	-0.89	-0.07	0.10	-0.28	0.20
Salinity	-0.74	0.12	-0.32	-0.32	0.42
DO	0.44	-0.23	0.71	-0.16	0.32
BOD	0.32	-0.15	0.75	0.45	0.01
COD	-0.17	0.31	0.14	-0.16	0.86
pH water	-0.84	0.21	0.02	-0.26	0.30
pH sed	0.41	-0.77	-0.42	0.00	0.00
Eh water	0.23	0.13	0.13	0.90	-0.22
Eh sediment	0.14	0.16	0.20	0.92	-0.13
NO3-N	0.18	0.51	-0.16	0.24	-0.70
NO2-N	0.12	0.84	-0.42	0.02	-0.04
PO4-P	0.85	0.41	-0.07	-0.01	-0.17
NH3-N	-0.05	0.84	-0.40	-0.24	-0.08
chl a water	-0.13	-0.78	-0.26	-0.34	0.27
chl a sediment	0.89	-0.32	0.01	0.02	0.12
Phaeo water	-0.42	0.20	0.80	0.25	0.08
Phaeo sed.	-0.03	0.76	0.01	0.28	0.30
POC	0.76	-0.37	0.32	0.03	-0.27
OC	0.64	-0.65	-0.18	0.02	-0.01
H2S water	0.85	0.30	0.16	0.19	-0.12
Macrofauna	-0.44	-0.05	0.14	-0.41	0.67
Meiofauna	-0.24	0.62	0.03	-0.63	0.11
Microfauna	0.26	0.39	0.33	-0.31	-0.48
Expl Var	6.40	5.24	2.80	3.22	2.71
Prp. Totl	0.28	0.23	0.12	0.14	0.12

Eigenvalues (avgcy4crk.sta)

Extraction: Principal factors (comm =multiple R-square)

	Eigenval	% total Variance	Cumul. Eigenval	Cumul. %
1	8.12	35.29	8.12	35.29
2	5.33	23.17	13.45	58.46
3	3.19	13.89	16.64	72.35
4	2.32	10.08	18.96	82.43
5	1.40	6.10	20.36	88.53

SECTION 4.2

***MACRO-, MEIO-
AND MICROBENTHOS
ABUNDANCE AND VARIATIONS***

CHAPTER 4

Section 4.2 Macro-Meiofauna and microbenthic abundance and variations

4.2.1 Introduction

In culture systems, shrimp productions relies ultimately on factors maximizing primary productivity and minimizing nutrient losses (Alongi *et al.*1999). Use of supplemental food for growth of shrimp is a common practice (Guillaume *et al.*, 1989, Nunes *et al.*1999, Craig *et al.*, 2002). Despite the higher use of supplemental feed, shrimp are known to feed on the natural food in earthen ponds. Studies by different workers (Kneib *et al.*1985; Raymond *et al.*1990; Mctigue *et al.*1998; Shishehchian *et al.*1999) have focused on the natural food contribution either by macro-, meio- and microbenthic populations at a time, or by their different stages feeding on nanoplakton in natural, or laboratory conditions. Contribution of natural food and supplemental feed to the gut of the shrimp, *Penaeus monodon* in semi-intensive shrimp culture systems earlier has been studied by Focken, (1998). Recently, benthic faunal composition of *Penaeus monodon* has also been studied by (Hena *et al.* 2004). In the above studies, it has been showed that, benthos form an important part of shrimp diet. The detailed reviews of macro-, meio- and microbenthos have been given in Chapter 2, Section 2.2. For the role of natural food, in shrimp nutrition, review has been given in Chapter 2, Section 2.3. It was clear from the literature survey that, there is lack of information on the natural diet of culture species, especially the modified extensive shrimp culture practices with holistic approach. Consequently, during present study, all natural benthic food components such as macro- meio-and microbethos were considered, in aerated and non aerated modified extensive shrimp culture system and adjacent creek, to understand their qualitative and quantitative distribution.

Understanding of variations in benthic fauna in relation to changes in the physico-chemical parameters of water and sediment is necessary. This will help to predict the variations in the benthic densities caused due to the changes in physico-

chemical parameters on the seasonal basis. This in turn will be helpful to manage supplemental feed and avoid the wastage of feed. The results obtained from the experimental study are described below for two production cycles. According to the objective, it was intended to study the effect of aeration on the development of bottom biota. Aeration is done primarily to enhance the dissolved oxygen concentration (DO). Therefore, DO is considered as the index parameter, to see whether, aeration in terms of DO has any effect on the development of bottom biota. The results were tested with one way ANOVA to see, whether, average macro, meio, and microbenthic densities in aerated and non-aerated ponds differed inter and intra seasonally. This was followed by backward multiple regression analysis, to see, which environmental parameters explain the linearity. It also helps in understanding the role of each parameter in building the natural benthic biota as well as maintaining the water quality.

4.2.1.2 Results on Macrofauna

For macrofauna one way ANOVA analysis single factor was carried out to see whether average densities in aerated and non aerated pond differed significantly between two cycles EC-1 and EC-2. It was observed that, there were no significant differences ($p>0.05$) in the average macrobenthic density between aerated (Table MaST1) and non aerated (Table MaST2) pond for respective EC-1 and EC-2, however, there were significant differences ($p<0.02$; Table MaST3) in average macrofauna densities between EC-1 and EC-2. There were no significant differences observed in aerated and non-aerated pond during respective EC-1 (MaST4) and EC-2 (Table, MaST5).

4.2.1.2.1 Experimental Cycle (EC-1)

During EC-1 shelled gastropode *Cerithedium* sp. were the apparently dominant group observed both in the aerated and non-aerated pond. Average macrofauna density (no. \pm sd.m⁻²) varied from 886 \pm 67 on 30 DoC to 5777 \pm 6910 no.m⁻² on 90 DoC and 693 \pm 504 on 45 DoC to 3312 \pm 3169 no.m⁻² on 120 DoC for aerated and non-

aerated pond respectively. Gastropoda contributed 100% to the macrofaunal density during EC-1 (Table MaC1.1 and Table MaC 1.2, Figure MF1.1 and MF1.2) However, in the creek along with Gastropoda, Polychaeta were observed, but only on 75 and 90 DoC and contributed 50% and 74% respectively (Table MaC 1.3 & Figure MF1.3). Overall, in experimental ponds, macrofauna density showed an increasing trend, while in creek, it showed slightly decreasing densities with the progressing culture period. Fortnightly average density in experimental ponds and creek is presented in Table MaC1.4, Figure MF1.4

4.2.1.2.1 A) statistical analysis for Aerated pond

In present study, the density of macrobenthos comprised only gastropodes in aerated pond (Figure MF1.1). To study the effect of aeration by considering DO as indicator factor of aeration for macrofauna, one way ANOVA was carried out. The analysis showed no significant effect of DO ($F_{6,2}=12.41;p=0.76$;Table AB1) on the development of macrofauna. Therefore, backward multiple regression analysis was carried out to see if, other parameters play any role on the development of macrofauna in aerated ponds. The analysis showed best linearity between chlorophyll *a* in water ($B=1.32$), chlorophyll *a* in sediment ($B=-2.0$) and phaeopigments ($B=0.71$) in the water OC ($B=-0.47$) and POC ($B=1.54$) which, explained 86.4 % variance. The model was significant at ($F_{5,3}=11.12;p=0.038$).

4.2.1.2.1 B) Statistical analysis Non-aerated pond

To study the effect of DO on macrofauna in non aeration pond, one way ANOVA was carried out. The analysis showed no significant effect of DO ($F_{6,2}=12.41;p=0.76$; Table AB1) on the development of macrofauna. On backward regression analysis for non-aerated-pond, macrofauna showed best linear relationship with sediment temperature ($B=0.73$), pH ($B=0.51$) and E_h ($B=0.91$) as well as phaeopigments ($B=0.47$) and organic carbon in sediment ($B=0.58$), altogether explained 92.5% variability. The overall relationship was significant ($F_{5,3}=20.81;p=0.016$). Also, stepwise regression analysis was carried showed linearity between macrofauna and

chlorophyll *a* in the sediment ($F_{1,7}=20.44, p=0.003$) and explained 70.9% variance. It suggests that, 70.9% variability in the macrofaunal density actually was due to the fluctuations in the chlorophyll *a* content in the sediment. This, indicate that macrobenthos in non-aerated pond was dependent largely on chlorophyll *a* production and foraged on the phytoplankton at the bottom of the pond.

4.2.1.2.1 C) Statistical analysis Creek

In the creek, Gastropoda contributed 86% while, remaining 14% comprised of Polychaeta (Figure MF1.3). For creek macrofauna showed linear relationship with that of temperature ($B=-1.0$), OC ($B=-1.39$), dissolved oxygen ($B=1.16$) and salinity ($B=2.57$) at $F_{4,4}=7.57; p=0.038$ and explained 76.7% variance.

Table MaC 1.1 : Fortnightly groupwise macrofauna (%) in aerated pond

	0	15	30	45	60	75	90	105	120
Gastropoda	100	100	100	100	100	100	100	100	100

Table MaC 1.2 : Fortnightly groupwise macrofauna (%) in non-aerated pond

	0	15	30	45	60	75	90	105	120
Gastropoda	100	100	100	100	100	100	100	100	100

Table MaC 1.3 : Fortnightly groupwise macrofauna (%) in creek.

	0	15	30	45	60	75	90	105	120
Gastropoda	100	100	100	100	100	50	26	100	100
Polycheata	0	0	0	0	0	50	74	0	0

Table MaC 1.4 : Fortnightly average macrofauna variations in aerated, non-aerated pond & creek.

	0	15	30	45	60	75	90	105	120
Aerated Pond	2503 ± 176	2657 ± 1312	886 ± 67	1618 ± 1155	1040 ± 306	2118 ± 1472	5777 ± 6910	5199 ± 3126	1810 ± 1203
Nonaerated pond	1040 ± 917	1733 ± 1040	1579 ± 1750	693 ± 504	732 ± 291	1656 ± 1163	2041 ± 2435	1964 ± 462	3312 ± 3169
Creek	1675 ± 408	693 ± 654	1098 ± 1062	693 ± 163	809 ± 163	462 ± 327	1098 ± 572	867 ± 572	578 ± 163

Figure: MF1.1 Fortnightly group-wise macrofauna variations (%) in aerated pond.

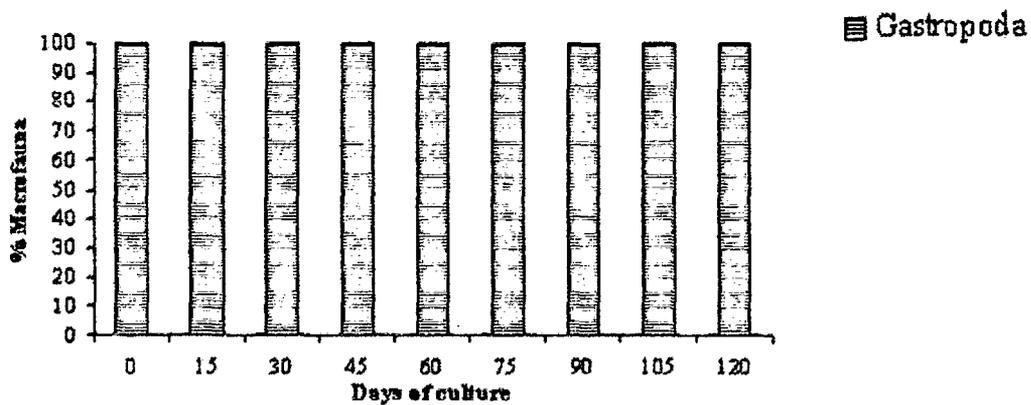


Figure: MF1.2 Fortnightly group-wise macrofauna variations (%) in non-aerated pond.

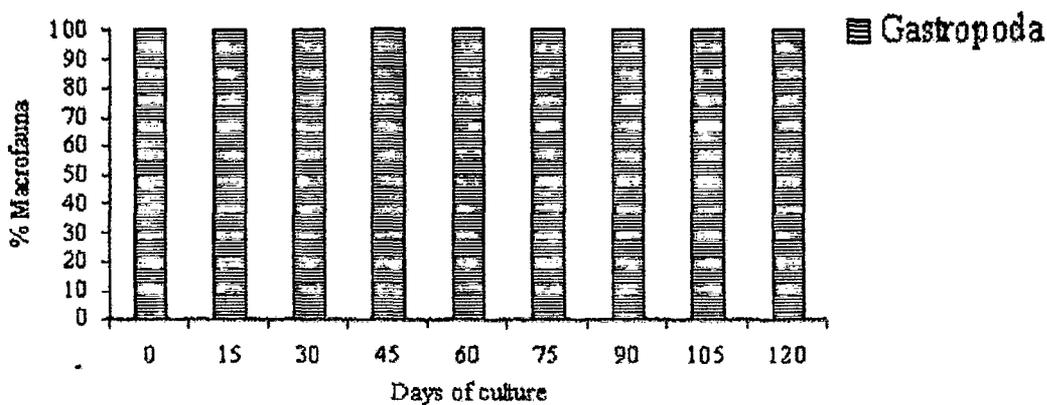


Figure: MF1.3 Fortnightly group-wise percent (%) macrofauna variations in creek.

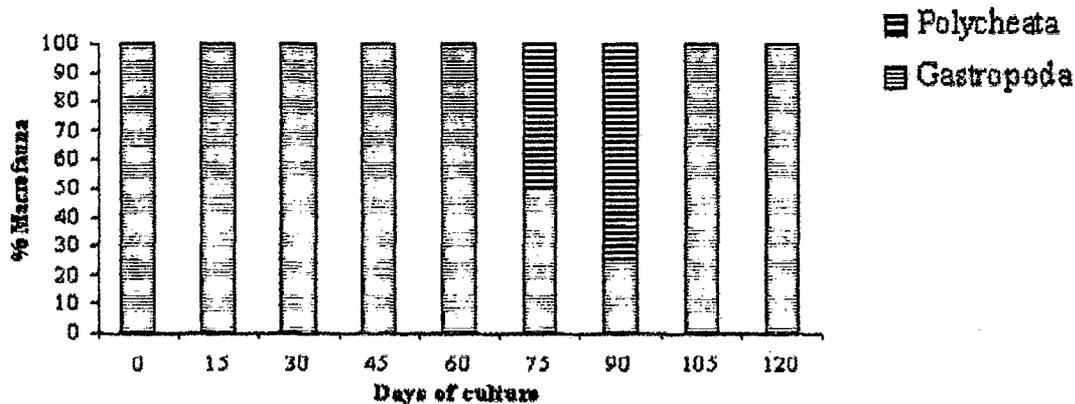
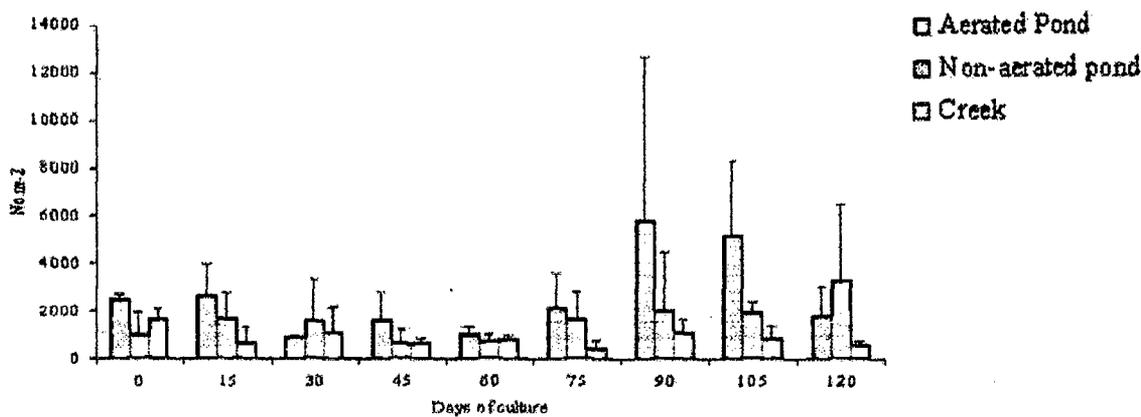


Figure: MF1.4 Fortnightly average macrofauna variations in aerated, non-aerated pond & creek.



4.2.1.2.2 Experimental Cycle (EC-2).

During EC-2 Ploychaeta and Gastropoda were the only two groups recorded, in the macrobenthos. Overall, the density of Polychaeta was higher than Gastropoda in both the experimental ponds and contributed 70 % and 60% respectively in aerated and non-aerated pond. Groupwise macrofauna composition is depicted in Table MaC2.1-3 and Figure MF2.1-3). In aerated pond, average macrofauna density was lowest being 1386 ± 809 on 105 DoC to highest 7318 ± 1273 no.m⁻² on 60 DoC. In non-aerated pond, the density varied from 693 ± 231 on 105 DOC to 3043 ± 1110 no.m⁻² on 60 DoC, wherein for creek the values varied from 522 ± 635 on 90 to 4102 ± 1378 no.m⁻² on 15 DoC. Thus, overall aeration pond showed highest macrofauna density, followed by creek, and non-aerated pond. The results showed that, the artificial aeration was capable of sustaining higher percent density of macrofauna compared to the natural creek habitat.

ANOVA between aerated and non-aerated pond for macrobenthos showed significant differences ($F=7.39$; $p=0.016$; Table MaST5). Unlike, EC-1 during EC-2 Ploychaeta dominated over Gastropoda population in aerated pond. For non-aerated pond, overall macrofauna comprised of 60% Polychaeta and 40% Gastropoda during EC-2. For creek, however, reverse trend was observed, with Gastropoda constituting 81% and Polycheta 19% of total macrofauna density. Gastropoda population in aerated and non-aerated pond showed increasing trend and Ploychaeta slightly decreasing trend (Figure MF2.2), wherein, in the creek, it showed exactly reverse trend during the culture period. This indicates that, pond environment was conducive for the Polychaeta due to their ability to utilize the accumulated fauna, organic matter coming from the artificial feed, Kirstensen, (2001), has showed the role of Polychaeta in effectively utilizing the organic matter. Further, Polychaeta as an alternative protein source has been studied for juvenile shrimp *Penaeus monodon* by Sunderyono *et al.* (1995). Thus, signifies the importance of Polychaeta in shrimp culture as food.

4.2.1.2.2 A Statistical analysis Aerated Pond

To study the effect of aeration by considering DO as indicator factor, one way ANOVA was carried out. The analysis showed no significant effect of DO ($F_{6,1}=1.111$; $p=0.621$; Table AB3) on the development of macrofauna. Stepwise regression analysis showed, linearity between macrofauna and organic carbon and explained 43.4% variations due to changes in the organic carbon ($B=-0.71$, $F_{1,6}=6.37$, $p=0.045$). Backward multiple regression analysis carried out, showed linearity between macrofauna and temperature ($B=1.72$), pH of water ($B=-1.9$), chlorophyll *a* in water ($B=0.82$), phaeophytin in sediment ($B=1.75$), and E_h of sediment ($B=0.20$) and explained 98.4% variance. The overall model was significant at $F_{5,2}=85.45$; $p=0.012$. Thus, water pH on stepwise regression analysis, accounted for 51.4% variance in macrofauna density ($F=8.39$; $p=0.027$). The role of pH to affect the benthic faunal distribution has been described by Ingole et al. (2006).

4.2.1.2.2 B Statistical analysis Non-aerated Pond

To study the effect of DO on macrofauna one way ANOVA was carried out. The analysis showed no significant effect of DO (Table AB4) on the development of macrofauna. The stepwise regression analysis showed 57.1% variations in macrofauna due to changes in the sediment organic carbon ($B=-0.79$) alone $F_{1,6}=10.32$, $p=0.018$ OC. The inverse relationship between OC and macrofauna indicates that, macrofauna specifically, Polychaeta effectively utilized the accumulated organic matter. The backward regression analysis however, showed that, macrofauna was also, linearly related with that of chlorophyll *a* ($B=-1.0$) and phaeophytin in the sediment ($B=0.67$; $F_{2,5}=6.32$, $p=0.043$) and explained 60.3% variations. This indicates that, macrofauna decrease was not exactly limited by presence of food in terms of OC and POC. Further, stepwise regression showed that, salinity of pond water ($B=0.73$) has more profound effect and model explained 46.7% variability, alone due to changes in salinity ($F_{1,6}=7.10$; $p=0.037$).

4.2.1.2.2 C Statistical analysis Creek

For creek, DO ($B=0.98$), temperature ($B=0.52$), phaeophytin, the degradation product of phytoplankton, algal material ($B=-0.42$) and POC ($B=-0.81$) showed significant linearity ($F_{4,3}=42.11; p=0.006$) with that of macrofauna. These parameters together explained 95.5% variance in macrofauna. Amongst three variables, DO and temperature of water seems to have more effect on the macrofauna density in the creek during EC-2. Thus, indicating that, DO play a decisive role in the creek for development of bottom fauna as bottom fauna get exposed during low tides. The role oxygen limiting the distribution of macrofauna due to oxygen deficiency has been studied by Theede (1969).

Table MaC 2.1 : Fortnightly groupwise macrofauna (%) in aerated pond

	0	15	30	60	75	90	105
Gastropods	5	31	45	14	53	47	33
Polycheata	95	69	55	86	47	53	67

Table MaC 2.2 : Fortnightly groupwise macrofauna (%) in non-aerated pond

	0	15	30	60	75	90	105
Gastropoda	28	46	50	47	23	50	44
Polycheata	72	54	50	53	77	50	56

Table MaC 2.3 : Fortnightly groupwise macrofauna (%) in creek

	0	15	30	60	75	90	105
Gastropoda	67	53	78	81	81	35	8
Polycheata	33	46	18	19	19	52	84
Bivalve	0	0	4	0	0	0	0

Table MaC 2.4 : Fortnightly average macrofauna variations in aerated, non-aerated pond & creek

	0	15	30	45	60	75	90	105
Aerated Pond	2311 ± 872	4159 ± 693	4699 ± 667	4236 ± 1163	7318 ± 1273	3197 ± 1738	3736 ± 1935	1386 ± 809
Non-aerated Pond	1887 ± 467	2657 ± 809	2272 ± 812	2080 ± 1312	3043 ± 1110	2157 ± 467	1618 ± 1138	693 ± 231
Creek	1733 ± 358	4102 ± 1378	3120 ± 489	2195 ± 927	1386 ± 963	1040 ± 655	522 ± 635	1276 ± 336

Figure: MF2.1 Fortnightly group-wise macrofauna variations (%) in aerated pond.

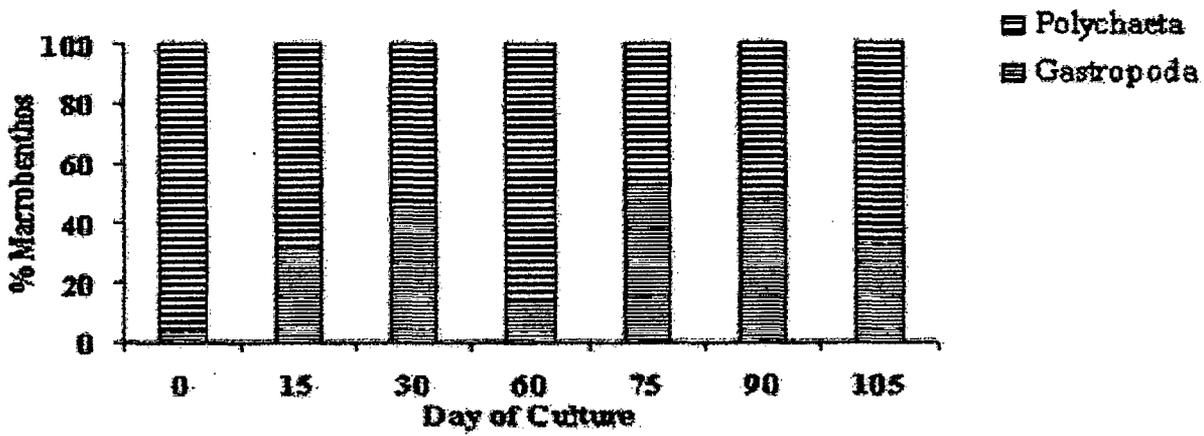


Figure: MF2.2 Fortnightly group-wise macrofauna variations (%) in non-aerated pond.

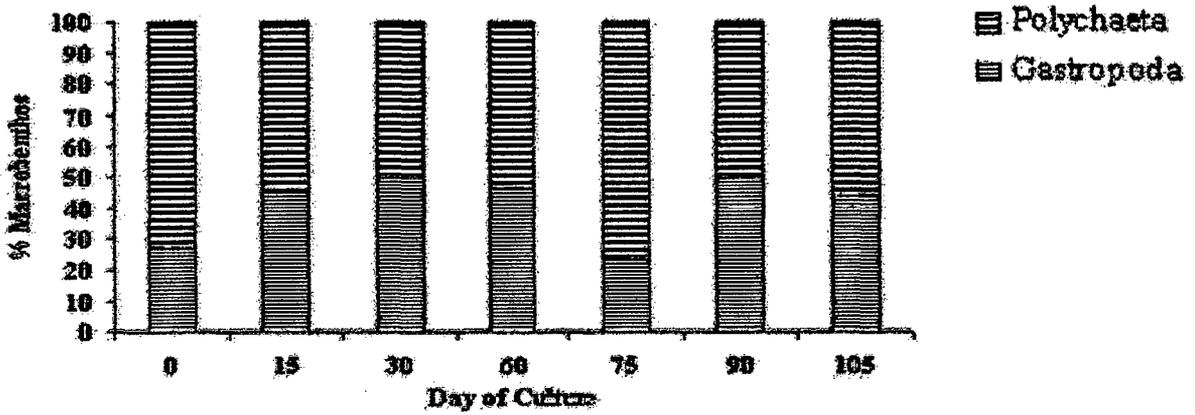


Figure: MF2.3 Fortnightly group-wise macrofauna variations (%) in creek.

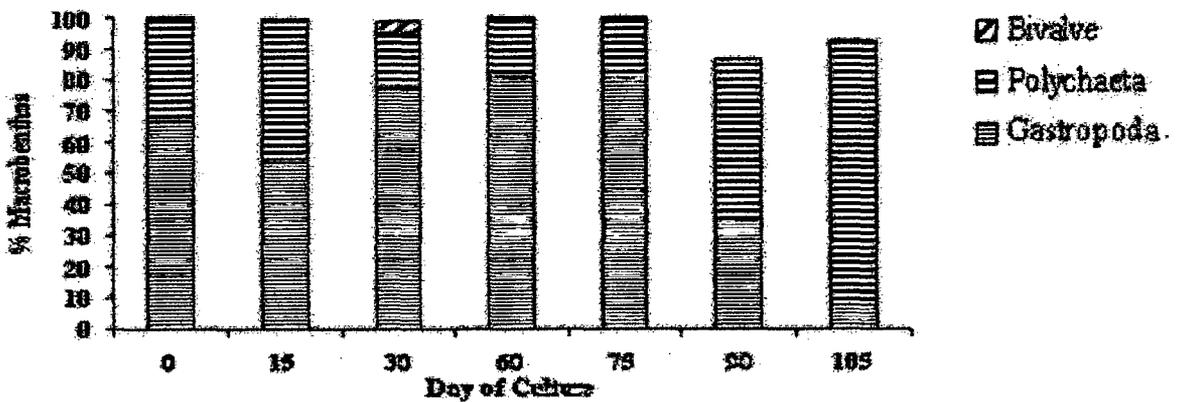
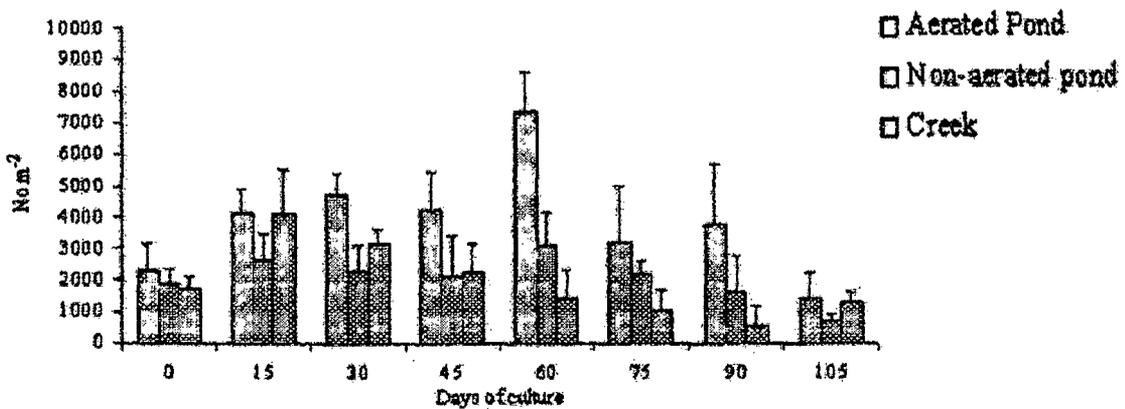


Figure: MF2.4 Fortnightly average macrofauna variations in aerated, non-aerated pond & creek.



4.2.1.2.3 Discussion Macrofauna

During present study it was found that, overall, average macrofauna densities in experimental ponds were higher during EC-2 than the EC-1. The Gastropoda were dominant during EC-1 and included *Telescopium* and *Cerithedium* which together contributed 100% to the macrofaunal density in both aerated and non-aerated pond (Figure MF1.1 and Figure MF1.2). Macrofauna seems to consume, phytoplankton and benthic diatoms, as it showed direct relationship with sediment chlorophyll *a* (Figure EC1.18). The source of the chlorophyll *a* was perceptibly from the water column. It was the diatoms and phytoplankton getting settled in the sediment, in addition to their growth within sediment on which the benthic Gastropoda fed during the EC-1 (Smith *et al.* 1996; Montani *et al.* 2003). The negative beta value for phaeophytin indicates that, degraded phytoplankton and diatomic biomass were consumed by macrofauna. It is known that, benthic gastropod *Cerithium* sp. predominantly feeds on benthic micro algae (Smith *et al.* 1996; Denadai *et al.* 2004), hence the availability of benthic diatoms in the pond sediment ensures the supply of food material. This in turn, helps in developing the Gastropoda population due to availability of diatomaceous or algal food.

During EC-2 unlike EC-1, macrofauna consisted of two prominent groups, Polychaeta and Gastropoda. Further, it was observed that, Polychaeta contributed higher percentages and dominated over the Gastropoda population (Figure MF2.1 and Figure MF2.2). The feeding behavior of both these groups is different. Gastropoda are known as the grazers mainly feeding on the microphytobenthos. However, Polychaets are known to prefer organic matter as their food. Ingole *et al.* (2002) has showed that, macrobenthic Polychaeta is a predominant group in the coastal water of the west coast of India, where organic carbon percentage is also high being over > 4 %. During present study, the organic carbon values were found to be higher in the non-aerated pond than aerated pond during EC-2 (Figure EC2.11). Chlorophyll *a* was higher in the water (Figure EC2.17) during EC-2 than in the sediment (Figure EC2.18), which indicates that, chlorophyll *a* in water of aerated pond could not settle on the bottom leading to less contribution to the macrobenthic food especially

Gastropoda compared to Polychaeta during EC-2. Thus, it can be concluded from stepwise and backward regression for aerated and non-aerated pond during EC-2 that, chlorophyll *a* and phaeophytin was consumed by Gastropoda while Polychaeta consumed organic matter in the sediment. The temporal, spatial and total standing stock of macrofauna in relation to food gradient has been studied by (Josefson, 1998) and showed that, under the abundant food conditions the distribution of macrobenthos is limited by space. However, during present study no spatial constraint were noted for macrobenthos in the shrimp ponds and macrofauna were found to be fairly distributed in the pond. For creek DO, salinity and temperature seems to have more profound effect on the macrofaunal distribution. This is very well understood in the view that, macrofauna in the creek is subjected to the tidal variations (Frost *et al.* 2004). Thus, during low tide the organisms are exposed to desiccation and limit their distribution. Temperatures and salinity, during present study showed decreasing trend during both cycles EC-1 (Figure EC1.1 & EC2.1) and EC-2 (Figure EC1.2 & EC2.2), respectively, might have affected the distribution of microbenthos in the creek. Conspicuously, macrofauna were found to be higher in the pond than the creek. Salinity in the pond were comparatively higher than the creeks and considering the pond habitat of macrofauna, it remained submerged throughout study period, thereby, provided prevention from desiccations otherwise caused in the creek during low tide exposures. Thus, mainly food availability in the pond environment, and in the creek, it was, physico-chemical parameters like temperature and salinity limited the distribution of macrofauna.

Table MaST 1 One Way ANOVA for macrofaunal abundance during two cycles EC-1 and EC-2 of aerated pond.

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
EC-1	9	23608.67	2623.186	3006396
EC-2	8	31041.75	3880.218	3122827

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	6692317	1	6692317	2.18651	0.159916	4.54306
Within Groups	45910962	15	3060731			
Total	52603279	16				

TableMaST2 One way ANOVA for macrofauna abundance in two cycles EC-1 and EC-2 of Non-aerated Pond.

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
EC-1	9	14750.61	1638.956	644525
EC-2	8	16406.68	2050.835	495732.3

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	718492.5	1	718492.5	1.24936	0.281258	4.543068
Within Groups	8626326	15	575088.4			
Total	9344819	16				

TableMaST3: One way ANOVA for macrofauna abundance in two cycles EC-1 and EC-2 of creek.

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
EC-1	9	7972.26	885.8067	133494.9
EC-2	8	15373.37	1921.671	1391298

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	4544532	1	4544532	6.307736	0.023955	4.543068
Within Groups	10807045	15	720469.7			
Total	15351577	16				

Table MaST4 Variation in macrofauna density between aerated and non-aerated pond during EC-1

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
Aerated Pond	9	23608.67	2623.186	3006396
Non-aerated Pond	9	14750.61	1638.956	644525

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	4359186	1	4359186	2.387992	0.141819	4.493998
Within Groups	29207369	16	1825461			
Total	33566555	17				

Table MaST5 Variation in macrofauna density between aerated and non-aerated pond during EC-2

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
Aerated Pond	8	31041.75	3880.218	3122827
Non-aerated Pond	8	16406.68	2050.835	495732.3

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	13386574	1	13386574	7.398841	0.016591	4.600111
Within Groups	25329919	14	1809280			
Total	38716492	15				

CHAPTER 4

Section 4.2.1.3 Results of Meiofauna

To study the variations in meiofaunal abundance between aerated and non-aerated pond, one way ANOVA was carried out for two culture cycles EC-1 and EC-2. It was observed that, there were no significant differences in the meiofauna densities between EC-1 and EC-2 in aerated (Table MeST1) and non-aerated ponds (Table MeST2). There were no significant differences (Table MeST3) in average meiofauna densities in creek either. The average meiofauna density observed in aerated pond and non-aerated pond during EC-1 (MeST4) and EC-2 (Table, MeST5) respectively, also did not differ significantly ($P>0.05$).

4.2.1.3.1 Experimental Cycle (EC-1)

During EC-1, the dominant meiofaunal groups observed consisted of Nematoda, Turbellaria, Copepoda and other groups with comparatively small proportion, consisted of eggs, nauplii, insect larvae, Nemertina and Polychaeta. Meiofaunal density was found to be very low throughout the culture period. The values varied from 0 to 252 ± 274 and 0 to 985 ± 419 no.10cm⁻² for aerated and non-aerated pond, respectively. The most dominant group was Nematoda which contributed overall, 89%, 46% and 89% for aerated, non-aerated pond and creek, respectively. The average Nematode density was lowest on 75 DoC being 21 ± 36 and highest on 105 DoC 252 ± 274 no.10cm⁻² for aerated pond. Copepod and Eggs probably of harpacticoid copepoda were found only during first two fortnights for aerated pond and on 0 and 15 DoC being 20% and 18%. For non aerated pond with overall mean density was 482 ± 508 no.10cm⁻² (Figure MeC1.4). In the creek, similar groups were observed but the density was much higher than both the ponds and values ranged from 0 to 2517 ± 445 no.10cm⁻². Compared to the non-aerated pond (Table MeC1.2 & Figure MF1.6), the percent occurrence of nematode was slightly higher in aerated pond (Table MeC 1.1 & Figure MF1.5) but much lower than the creek.

4.2.1.3.1 A Statistical analysis for Aerated Pond meiofauna

To study the effect of aeration on meiofauna, one way ANOVA was conducted. The analysis showed no significant effect of DO ($F_{6,2}=0.601;p=0.734$;Table AB1) on the development of meiofauna. Meiofauna in the aerated pond was nil on 30 and 45 DoC (Figure MF1.8). Overall average density during EC-1 was found to be 105 ± 97 no.10cm⁻² (Table ST7). When backward multiple regression analysis was carried out, amongst all the water and sediment quality parameters, temperature ($B=-0.63$), pH sediment ($B=0.24$), chlorophyll *a* in water ($B=1.28$), and in sediment ($B=-0.44$), phaeophytin in water ($B=-0.64$) and sediment ($B=-0.51$) were found to explain 95.9% variance ($F_{6,2}=32.57;p=0.030$).

4.2.1.3.1 B Statistical analysis for Non-Aerated Pond

For non-aerated pond one way ANOVA showed no significant effect of DO ($F_{6,2}=0.601;p=0.734$;Table AB2) on the development of meiofauna. In non-aerated pond meiofauna density was lower than aerated pond. From Figure (MF1.8) it can be seen that, only on 15 DoC average meiofauna density was 985 ± 419 no.10cm⁻². Excluding average density value on 15 DoC, the values for 0 DoC and from 30 DOC averaged only 46 ± 64 no.10cm⁻². On 45 and 75 DoC, meiofauna was totally absent in the samples (Figure MF1.8). Backward multiple regression analysis carried out showed, highly significant relationship between the meiofauna and temperature ($B=-0.87$), E_h of water ($B=-0.39$), POC ($B=0.17$), salinity ($B=1.19$), OC ($B=0.87$). These parameters explained 99.1% variance in meiobenthic density. The relationship was significant at ($F_{6,2}=145.49;p=0.007$).

4.2.1.3.1 C Statistical analysis for Creek meiofauna

In the creek, average meiofaunal density was higher being 766 ± 808 (Table ST9) no.10cm⁻² than aerated pond (105 ± 97 ;Table ST7) and non aerated pond (150 ± 319 ; Table ST8). Noticeably, when average meiofaunal density on 45 DoC was lowest in

both the experimental ponds (Figure MF1.8), creek showed higher average faunal densities $2517 \pm 445 \text{ no.10cm}^{-2}$ (Figure MF1.8). This indicates that, due to free connection of creek, with adjacent sea (Arabian) water, recruitment of meiofauna may have played role in maintaining the higher density of meiofauna in the creek, due to tidal incursion and excursion. Further, backward regression analysis carried out explained 91.7% variance in meiofauna density, due to chlorophyll *a* in water ($B=0.92$), E_h of water ($B=-0.77$), phaeophytin in the sediment ($B=-1.63$) and pH of sediment ($B=-1.17$) the relationship was significant at ($F_{4,4}=23.12; p=0.005$).

Table MeC 1.1 : Fortnightly groupwise meiofauna (%) in aerated pond

	0	15	30	45	60	75	90	105	120
Nematode	80	64	0	0	100	100	100	100	100
Copepod	20	18	0	0	0	0	0	0	0
Eggs	0	18	0	0	0	0	0	0	0

Table MeC 1.2 : Fortnightly groupwise meiofauna (%) in non-aerated pond

	0	15	30	45	60	75	90	105	120
Nematode	100	30	100	0	100	0	67	100	100
Copepod	0	49	0	0	0	0	33	0	0
Eggs	0	19	0	0	0	0	0	0	0
Larvae	0	2	0	0	0	0	0	0	0

Table MeC 1.3 : Fortnightly groupwise meiofauna (%) in creek

	0	15	30	45	60	75	90	105	120
Nematode	67	100	0	86	100	100	66	96	100
Copepod	14	0	0	5	0	0	8	4	0
Eggs	14	0	0	5	0	0	13	0	0
Larvae	5	0	0	4	0	0	13	0	0

Table MeC 1.4 : Fortnightly average meiofauna variations in aerated, non-aerated pond & creek.

	0	15	30	45	60	75	90	105	120
Aerated Pond	105	231	0	0	168	21	126	252	42
	±	±	±	±	±	±	±	±	±
	73	346	0	0	192	36	63	274	73
Non Aerated Pond	27	985	21	0	2	0	126	168	21
	±	±	±	±	±	±	±	±	±
	12	419	36	0	3	0	126	96	36
Creek	660	346	0	2517	32	1258	1199	755	126
	±	±	±	±	±	±	±	±	±
	756	222	0	445	45	1512	539	445	178

Figure: MF1.5 Fortnightly group-wise meiofauna variations(%) in aerated pond.

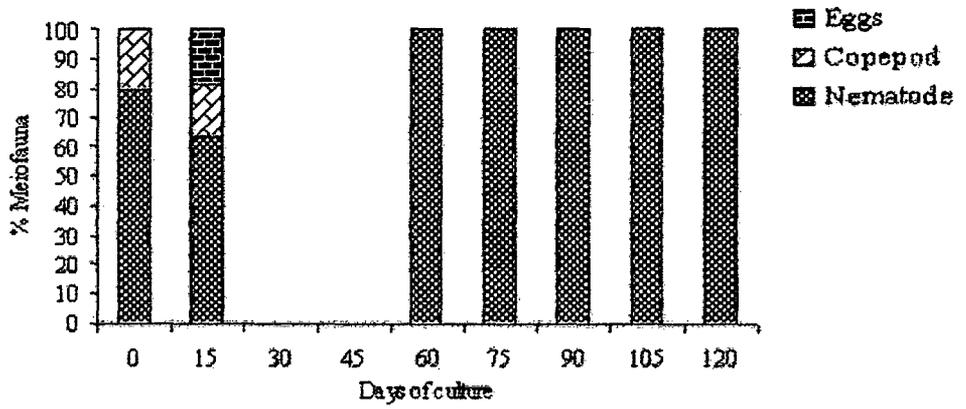


Figure: MF1.6 Fortnightly group-wise meiofauna variations (%) in non-aerated pond.

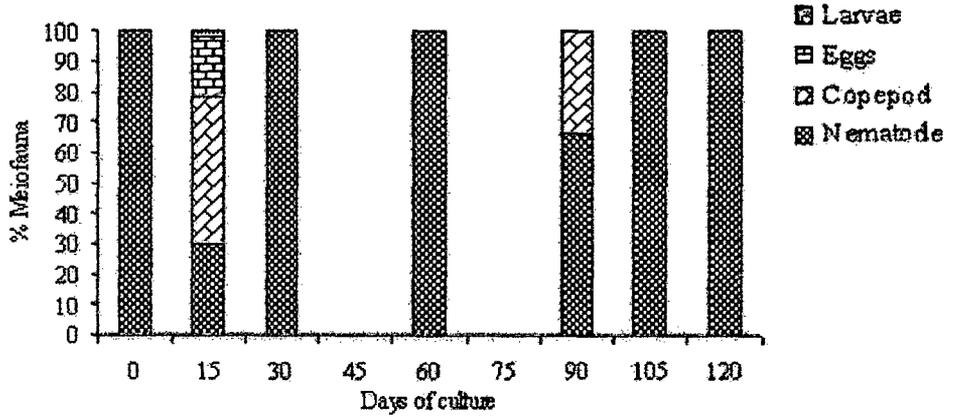


Figure: MF1.7 Fortnightly group-wise meiofauna variations (%) in creek.

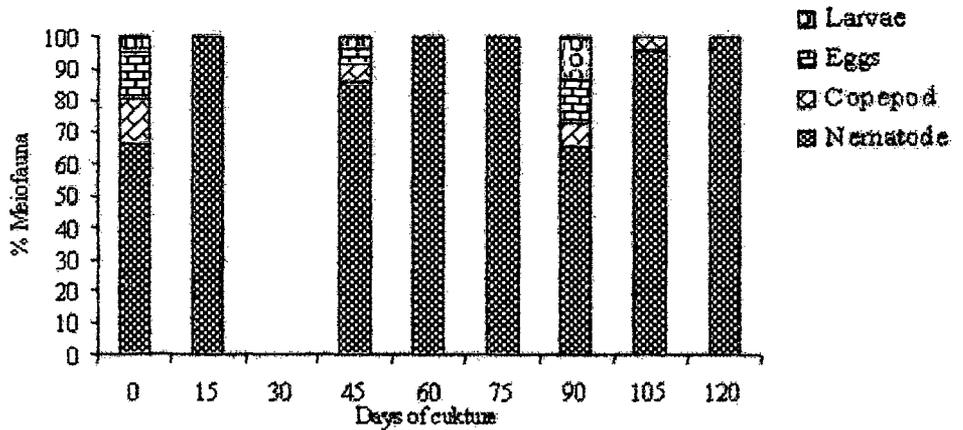
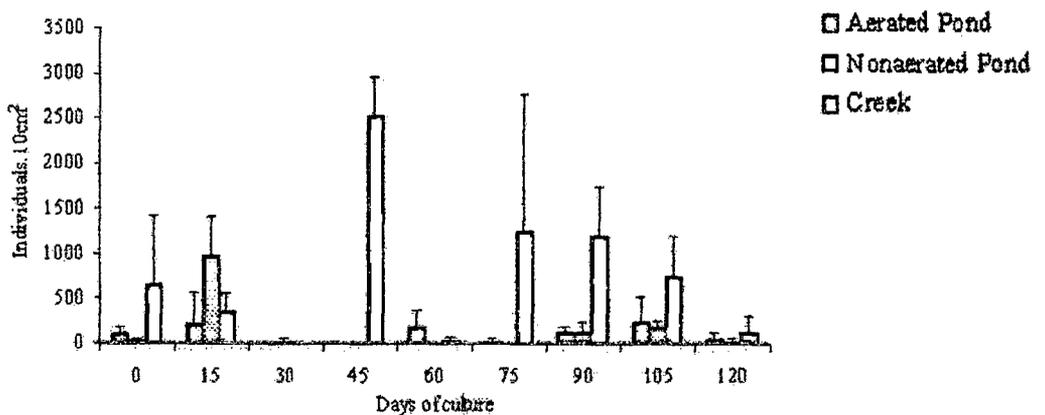


Figure: MF1.8 Fortnightly average meiofauna variations in aerated, non-aerated pond & creek.



4.2.1.3.2 Experimental Cycle (EC-2)

During EC-2 meiofauna was more diverse, with wide variations in densities. In aerated pond average density was lowest being 33 ± 32 on 105 DoC to and highest 1260 ± 1635 no.m⁻² on previous fortnight i.e. on 90 DoC. Percent contribution wise, overall average naupli contributed 36%, nematode 28% and bryozoans 18%, while copepods contributed only 7% to the total population density in aeration pond (Figure MF2.5). In non-aerated pond, overall percent contribution calculated from total density of groups were in decreasing order, where nematode contributed 44%, bryozoans 25%, Copepod 15% and Eggs 11% (Figure MF2.6). The average density in non-aerated pond varied from 68 ± 50 on 15 DoC to 653 ± 561 on 105 DoC no.m⁻² (Table MeC 2.4 and Figure MF2.8).

For creek average density varied from lowest of 131 ± 53 on 90 DoC to 1545 ± 199 no.m⁻² on 15 DoC and was dominated by nematode 71% followed by copepod 12% and bryozoa 10% (Figure MF2.7). Overall it was observed that, aeration pond showed increasing trend in meiofauna density as compared to non-aerated pond (Figure MF 2.8), wherein the meiofaunal density showed decreasing trend in the creek (Figure MF2.8).

4.2.1.3.2 A Statistical analysis for Aerated pond

For aerated pond the average meiofauna values were found to be 534 ± 512 no.10cm⁻² in aerated pond (Table ST10). The one way ANOVA analysis showed no significant effect of DO ($F_{6,1}=1.51$; $p=0.552$; Table AB3) on the development of meiofauna. Therefore, the backward multiple regression analysis was performed to check for linear relationship between meiofauna and other environmental parameters. It showed highly significant linear relationship between various parameters. The model explained 99.9 % variability in meiofauna due to the changes in pH of pond water ($B=-0.58$), E_h of the sediment ($B=0.58$), chlorophyll *a* in surface water ($B=-0.49$),

POC (B=0.12), phaeophytin in pond water (B=0.87) and sediment (B=-0.17; p=0.003).

4.2.1.3.2 B Statistical analysis for Non-aerated pond

For non-aerated pond overall average meiofauna was found to be 422 ± 477 no.10cm⁻² (Table ST11). The average meiofauna density overall was found to be slightly lower than aerated pond (Table ST10). To study the effect of DO on meiofauna one way ANOVA was carried out. The analysis showed no significant effect of DO (p>0.05) (Table AB4) on the development of meiofauna. When multiple regression analysis was carried out, none of the water quality parameter showed significant relationship with meiofauna density. With an exception the highest density of 1449 ± 1523 no.10cm⁻² on 45 DoC was observed (Table MeC2.4 Figure MF2.8). Meiofauna densities excluding this point were found to be; below than overall average as showed in Table ST11 and for rest of the days it averaged 275 ± 254 no.10cm⁻². Regression analysis did not show any significant linear relationship between meiofauna and water or soil quality parameters.

4.2.1.3.2 C Statistical analysis for Creek

Creek showed higher average meiofauna density than, both the culture ponds (Table ST12). The overall average value for creek was found to be 959 ± 875 no.10cm⁻² from Table MeC 2.4). For creek, backward multiple regression analysis showed highly significant relationship between meiofauna and temperature (B=-1.21), pH of water (B=1.29), E_h of sediment (B=-0.66), OC (B=-0.76), POC (B=0.44) and explained 96.3% variances with overall significant relationship ($F_{5,2}=37.71$; p=0.026). Average meiofauna showed overall decreasing trend (MF 2.8). The regression model shows that, temperature and food availability in terms of organic carbon probably did not have any effect on the development of meiofauna in creek. However, pH (Figure EC2.3) seems to have affected meiofauna in the creek (Figure MF2.8.) as meiofauna in the creek decreased with decreasing pH (Figure, EC2.3).

Table MeC 2.1 : Fortnightly groupwise meiofauna (%) in aerated pond

	0	15	30	45	60	75	90	105
Bryozoa	0	0	0	1	17	23	29	26
Cnidaria	0	0	3	23	12	1	0	0
Copepoda	29	0	15	3	16	3	4	5
Eggs	18	17	17	5	2	0	3	32
Foraminifera	0	0	0	0	2	0	0	0
Naupli	0	0	4	66	51	72	0	0
Nematoda	53	83	50	0	0	0	63	37
Polycheata	0	0	1	0	0	0	0	0
Sarcomastigophora	0	0	4	0	0	0	0	0
Turbellaria	0	0	5	0	0	0	0	0

Table MeC 2.2 : Fortnightly groupwise meiofauna (%) in non-aerated pond

	0	15	30	45	60	75	90	105
Bryozoa	0	0	0	26	0	37	3	60
Kinorhina	0	0	0	0	0	0	0	0
Cnidaria	0	0	0	0	0	0	0	0
Copepoda	19	0	46	11	9	4	10	1
Eggs	52	87	25	4	29	0	2	3
Foraminifera	0	0	1	0	0	0	0	0
Larvae	0	0	0	2	2	0	0	0
Naupli	0	0	10	0	0	0	0	0
Nematoda	29	13	15	55	57	58	85	36
Oligocheata	0	0	0	0	0	1	0	0
Polycheata	0	0	0	0	2	0	0	0
Sarcomastigophora	0	0	2	0	2	1	0	0
Turbellaria	0	0	1	0	0	0	0	0

Table MeC 2.3 : Fortnightly groupwise meiofauna (%) in creek

	0	15	30	45	60	75	90	105
Bryozoa	0	0	0	29	0	0	0	0
Copepoda	28	1	31	17	2	13	4	7
Eggs	7	1	1	2	0	0	0	0
Foraminifera	0	0	3	0	0	0	0	0
Larvae	0	0	12	0	1	0	0	6
Naupli	7	3	0	0	0	0	0	0
Nematoda	45	87	51	51	94	84	96	86
Oligocheata	7	7	0	0	3	2	0	2
Polycheata	3	0	0	0	0	0	0	0
Sarcomastigophora	3	1	2	0	0	0	0	0
Turbellaria	1	1	0	0	0	0	0	0

Table MeC 2.4 : Fortnightly average meiofauna variations in aerated, non-aerated pond & creek

	0	15	30	45	60	75	90	105
Aerated Pond	39 ± 16	63 ± 36	642 ± 546	492 ± 738	453 ± 92	1290 ± 1146	1260 ± 1635	33 ± 32
Non-aerated Pond	82 ± 12	68 ± 50	625 ± 221	1449 ± 1523	116 ± 59	215 ± 305	165 ± 35	653 ± 561
Creek	333 ± 19	1545 ± 199	614 ± 104	2713 ± 567	1393 ± 160	670 ± 84	131 ± 53	272 ± 62

Figure: MF2.5 Fortnightly group-wise percent meiofauna variations in aerated pond.

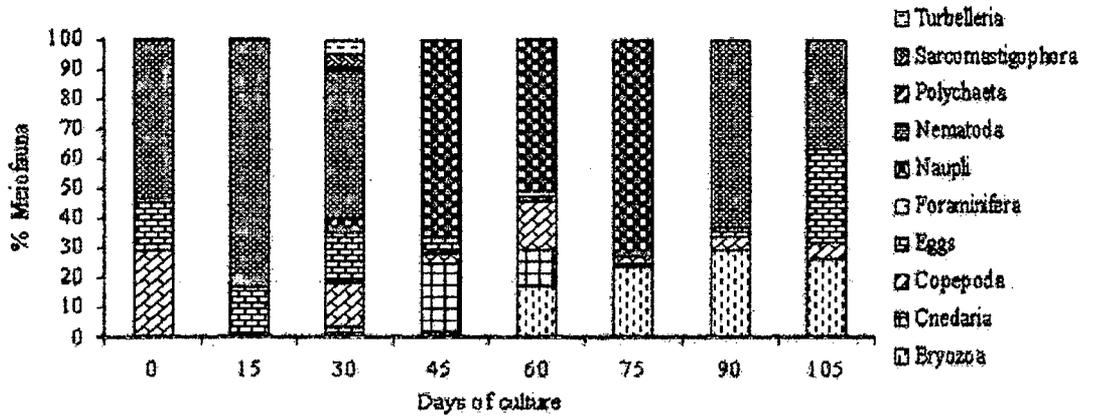


Figure: MF2.6 Fortnightly group-wise percent meiofauna variations in non-aerated pond.

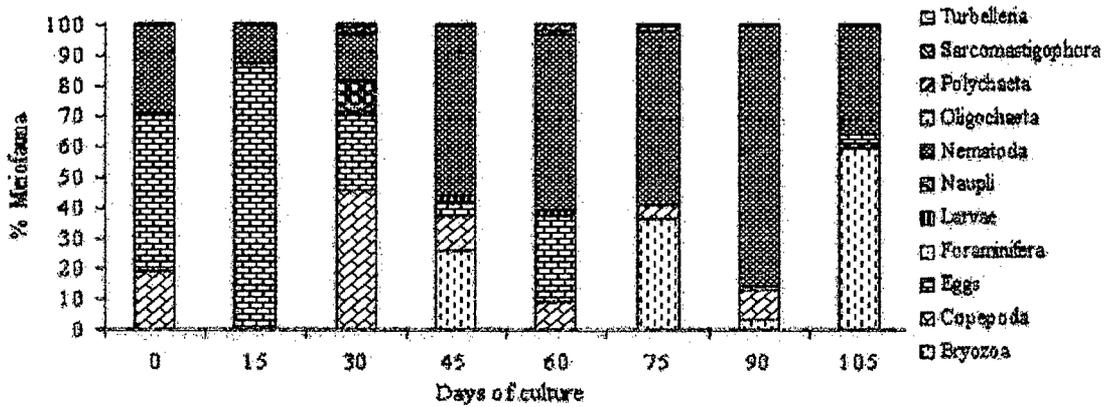


Figure: MF2.7 Fortnightly group-wise percent meiofauna variations in creek.

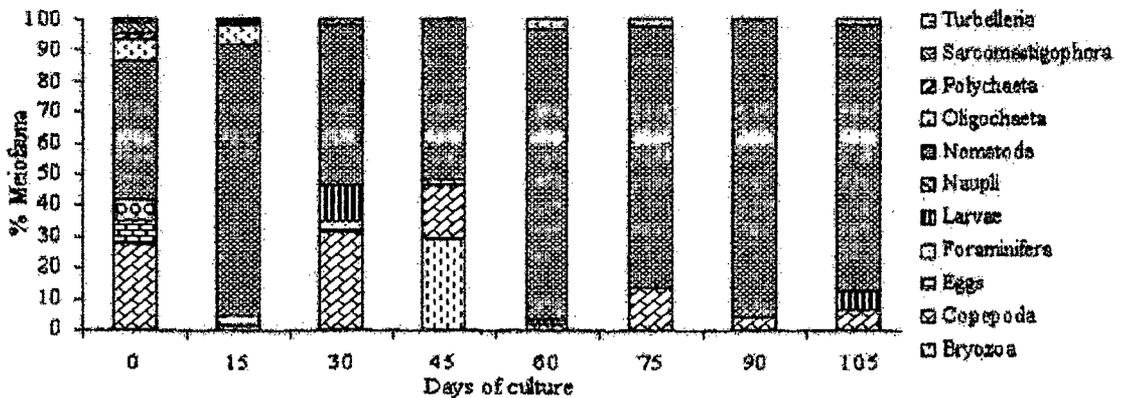
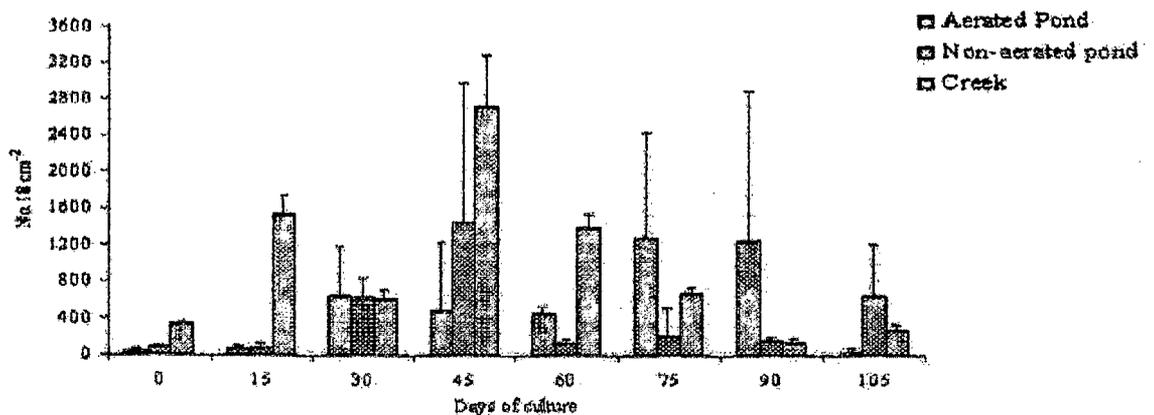


Figure: MF2.8 Fortnightly average meiofauna variations in aerated, non-aerated pond & creek.



4.2.1.3.3 Discussion Meiofauna

From results it was found that, overall meiofauna showed higher average densities during EC-2 (Figure MF1.8) than EC-1 (Figure MF2.8) in aerated as well as non-aerated pond. During EC-1, phytoplankton from water as well as sediment was consumed by meiobenthos, as there was significant weak correlation between sediment chlorophyll *a* and meiofauna ($r=0.30$). With higher ($B=1.28$) chlorophyll *a* in water, the phytoplankton biomass settling on the sediment was higher. One-Way ANOVA did not show any significant effect of DO on the development of bottom fauna (Table.AB1), indicating that, DO was not limiting for the meiofauna in aerated pond during EC-1, and higher density was maintained due to the higher chlorophyll *a* i.e. phytoplankton, acting as food for meiofauna in the water column which probably was facilitated due to the artificial aeration.

In non-aerated pond, chlorophyll *a* (Figure EC1.17) and phaeophytin (Figure EC1.19) in the water were found to be marginally less during EC-1, than the aerated pond. Therefore, organic carbon availability (Figure EC1.11) seems to have more profound effect on the development of bottom fauna in non-aerated pond. Also, decreasing salinity (Figure EC1.2) showed profound effect on the development of meiofauna ($B=1.19$). During EC-2, increased pH (Figure EC2.3) and E_h (Figure EC2.6) in aerated pond seems to have more profound effect on the development on the meiofauna. The E_h values varied between 50 mv to 200 mv (Figure EC2.6) indicates potential reducing environments. Similar E_h values are reported by Mata *et al.* (1999) in culture ponds suggested that, these values indicate sufficient oxygen, for survival of benthos. The DO during study period was maintained by the phytoplankton in the water, as the chlorophyll *a* concentration was found to be high (Figure EC2.17) and showed similar trend to that of aeration pond during EC-1. From these observations, main governing factor for settled meiofauna in both the pond seems to be the food availability, in terms of live and dead phytoplankton biomass, as well as organic carbon, in particulate form that accumulated in sediment. In case of diversity, as general feature over two studied cycles, it was found that,

nematode followed by copepod, naupli, eggs of copepod and polychaeta were most abundant groups. Cordava *et al.* (2002) has also reported, the abundance of these groups from low water exchange ponds, farming blue shrimp *L. stylirostris*. However, during present study, the most abundant group was found to be the nematode for both the experimental cycles in the ponds as well as in the creeks. Nematode is known to feed on the wide assortment of food, and on the basis of their feeding behavior they are described as, selective, non-selective deposit feeders, epistrate feeders, omnivores to predator (Nanajkar & Ingole, 2007). Their feeding on wide assortment of food available has been studied by Kito *et al.* (1998) in the shrimp culture ponds in Thailand. Three species belonging to genera *Diplolaimella*, *Thalassomonhystera* and *Theristus* has been described. Since shrimp is non-selective indiscriminate feeder, it seems that, nematode were ingested passively whenever shrimp foraged on the sediment. With different feeding types of nematode and food in terms of bacteria, phytoplankton or particulate matter may correlate and explain the nature of food available to them in the shrimp culture system.

The Chlorophyll-*a* and OC and bacteria seems to have been main food for nematode in the shrimp culture ponds and thus indicates that, they were mostly grazers. Mones *et al.* (1999) has stated lack of correlation between chlorophyll *a* and nematode density. There was negative relation between the meiobenthic population and phaeopytin in water and sediment. According to, Pinckey and Sanduli (1990) for nematode and copepod negative relationship between them and phaeophytin concentrations was an indication of chlorophyll *a* consumption by these groups. Most of the meiofaunal groups do feed on the bacteria and microalgae (Ripper, 1978). Nematode further specifically has been sited to feed on the diatoms by Mones *et al.* (1999). Meiofaunal groups like nematodes and oligochaeta have short life cycles, and sustain in low salinities and hypoxic conditions (Bouwman *et al.* 1984). They selectively feed on the microbenthic diatoms. Diatoms were also found during the present study and their abundance varied approximately from 189 to 9000 no.3.2 cm⁻², their average variations in the sediment has been described in Figure (MF1.12)

and Figure (2.12). Significant ($p < 0.05$), but weak correlation was observed between microphytobenthos and meiofauna, were mainly consisted of diatoms and nematods respectively. This confirms the dependence of meiofaunal groups on the microbenthic diatom for food.

The second most abundant group was found to be Copepoda. The percent contribution was low for copepod population during both the EC-1 (Figure MF1.5, MF1.6) and EC-2. (Figure MF2.5, MF2.6). From this study it was understood that, both nematode and copepod are affected by the physico-chemical factors and between them nematode are more resistant to the fluctuations in environment than the copepod. Amongst these two, copepods are more sensitive to the changes in dissolved oxygen but can withstand all salinity ranges from brackish water to hypersaline pools and salt pans. During this study, harpacticoid copepod naupli also contributed significantly and was the third most abundant category of meiofauna groups. According to Dahms and Qian (2004), harpacticoida is virtually the only meiofaunal taxon whose, development includes naupli larvae. From this, it can be confirmed that, the naupli found during present study were most probably of harpacticoid copepods. However, their percentage contribution was less during EC-1 (Figure MF 1.5; MF1.6) and EC-2 (MF2.5; MF2.6). According to Dahms and Qian (2004) the life cycles of the harpacticoid copepods are smaller than season cycles; hence they show short generation times. This can be the reason why, their density in pond seems to be less. Dahms and Qian (2004) further have described the copepod as a particle feeder. This indicates that, they prefer the particulate or accumulated organic matter on the bottom. The effect of food and temperature has been studied by Escribano and McLaren (1992) on the length and weights of marine copepod and showed that, low temperatures and food availability results in small individual copepod body size. During present study, also large variations in copepod sizes were observed.

During EC-2 in non-aerated pond, there was lack of significant relationship of meiofauna with that of physico-chemical quality of water and sediment, and DO also showed no significant effect on their development (Table AB4). This indicates that, neither DO nor food availability was responsible for decreased meiofaunal densities in the non-aerated pond during EC-2. Such scenario indicates that, meiofauna abundance was limited by the physical processes. The physical process here, imply the process of recruitment. Recruitment of meiofaunal organisms from the source water (creek) may have been responsible for the less density of meiofauna in non-aerated pond. During EC-1 and EC-2 the higher meiofauna density and diversity in the creek was observed. This phenomenon is attributed to the recruitment due to free connection with the sea water. Thus, in shrimp culture ponds meiofaunal densities can be invariably less due to the limited water exchanges from the source water. Even-though water exchanges are carried out, the quantification of suspended meiofauna congregating in the shrimp ponds needs further study.

Table MeCST1 One way ANOVA for meiofauna abundance in two cycles EC-1 and EC-2 of aerated pond.

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
EC-1	9	943.8667	104.8741	9342.228
EC-2	8	4272.583	534.0729	262220.6

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	780190.5	1	780190.5	6.126245	0.025738	4.543068
Within Groups	1910282	15	127352.1			
Total	2690473	16				

Table MeCST2 One way ANOVA for meiofauna abundance in two cycles EC-1 and EC-2 of non-aerated pond.

Anova: Single Factor

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
EC-1	9	1350.21	150.0233	101785.3
EC-2	8	3372	421.5	227544.8

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	312139.4	1	312139.4	1.94512	0.183426	4.543068
Within Groups	2407096	15	160473.1			
Total	2719235	16				

Table MeCST3: One way ANOVA for meiofauna abundance in two cycles EC-1 and EC-2 of creek.

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
EC-1	9	5044.603	560.5115	279228.5
EC-2	8	7669.625	958.7031	765787.3

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	671533.8	1	671533.8	1.326383	0.267476	4.543068
Within Groups	7594340	15	506289.3			
Total	8265873	16				

Table MeCST4 Variation in meiofauna density between aerated and non-aerated pond during EC-1

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
Aerated Pond	9	943.8667	104.8741	9342.228
Non-aerated Pond	9	1350.21	150.0233	101785.3

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	9173.057	1	9173.057	0.165091	0.689895	4.493998
Within Groups	889020.5	16	55563.78			
Total	898193.6	17				

Table MeCST5 Variation in meiofauna density between aerated and non-aerated pond during EC-2

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
Aerated Pond	8	4272.583	534.0729	262220.6
Non-aerated Pond	8	3372	421.5	227544.8

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	50690.65	1	50690.65	0.207	0.656105	4.600111
Within Groups	3428358	14	244882.7			
Total	3479048	15				

CHAPTER 4

Section 4.2.1.4 Microbenthos Results

For determination of significant differences in average microbenthos density one way ANOVA was carried out in aerated and non-aerated pond, showed significant difference between two cycles EC-1 and EC-2. In aerated pond it was ($F_{1,15}=4.49$; $p=0.051$; Table MiST1) and in non-aerated ponds it was ($F_{1,15}=4.15$; $p=0.059$; Table MiST2). There were highly significant differences ($F_{1,15}=9.37$; $p=0.007$; Table MiST3) in average microbenthic densities between EC-1 and EC-2 in creek also. Further, there were no significant differences in average microbenthic density in aerated pond and non-aerated pond during respective EC-1 (MiST4) and EC-2 (Table MiST5).

4.2.1.4.1 Experimental Cycle (EC-1)

During EC-1, the microbenthos was comprised mostly of diatoms *Coscinodiscus*, *Pleurosigma* and *Navicula* in both the experimental ponds. Overall average density ranged from 53 ± 70 to 1858 ± 2053 and 35 ± 41 to 1345 ± 1006 no. 3.2cm^{-2} for aerated pond (Table MiC 1.4) and non-aerated pond, respectively (Table MiC1.4). An overall average calculated for 120 day cycle in aeration pond, was found to contribute *Cosinodiscus* (46%), *Navicula* (34%) and *Pleurosigma* (18%) Table (MiC 1.1). In non-aeration pond, *Pleurosigma* contributed higher densities (47%), followed by *Cosinodiscus* (38%) and *Navicula* (10%) Table (MiC1.2). In the creek higher density as well as diversity of microbenthos as compared to culture ponds was observed. The values varied from 3715 ± 4466 to 17728 ± 4767 no. 3.2cm^{-2} . In creek overall *Coscinodiscus* contributed (54%) followed by *Navicula* (30%) and *Pleurosigma* (13%) Table (MiC1.3). Overall increasing trend for microbenthic densities was observed in aerated pond, wherein for non-aerated pond the reverse trend was observed, with an exception on last fortnight, where average density was found to be higher (Figure MF 1.12).

4.2.1.4.1 A) Statistical analysis Aerated Pond

During EC-1 in aerated pond the average density of microbenthos was found to be $552 \pm 712 \text{ no.} \cdot 3.2 \text{ cm}^{-2}$ (Table ST7) Microbenthos consisted of mainly diatoms, showed an increasing trend till the end of culture period with highest percent contributions. *Coscinodiscus* percentage was found to be lowest on 105 DoC; being 23% and highest percent contribution was 88% on 75 DoC (Table MiC1.1; Figure MF1.9). To study the effect of aeration on microbenthos, one way ANOVA was carried out. It showed significant effect of DO ($F_{6,2}=118.75; p=0.008$; Table AB1) on the development of microbenthos.

4.2.1.4.1 B) Statistical analysis Non-aerated Pond

For non-aerated pond overall average microbenthic density for 120 days, was found to be $506 \pm 444 \text{ no.} \cdot 3.2 \text{ cm}^{-2}$ (Table ST8), with decreasing trend. Alongwith *coscinodiscus* *Pleurosigma* were the most dominant microbenthic species. *Coscinodiscus* percentage was highest on 75 DoC being 88% and lowest being 22% on 105 DoC (Table MiC1.2; Figure MF1.10). For *Pleurosigma* highest percent contribution was 61% on 0 DoC and lowest on 105 DoC (Table MiC1.2; Figure MF1.10). Thus, *Pleurosigma* showed decreasing trend as the culture progressed. Stepwise multiple regression analysis showed that, the significant linear relationship $p < 0.05$ between microbenthos and sediment chlorophyll *a* ($B=0.58$), pH sediment ($B=-0.80$), and temperature ($B=0.28$) and explained 92.7% variations due to these parameters and the relationship is found to be significant at $F_{3,5}=35.02; p=0.001$.

4.2.1.4.1 C) Statistical analysis Creek

In the creek, average microbenthic density was $9413 \pm 5641 \text{ no.} \cdot 3.2 \text{ cm}^{-2}$ (Table ST9) and values were found to be highest than both the culture ponds (Figure MF1.12). Overall, it showed stable average percent contribution due to cyclic increase and decrease in the microbenthic densities (Table MiC1.3; Figure MF1.11). The creek showed significant linear relationship between Chlorophyll *a* in water and sediment ($p < 0.05$). Chlorophyll *a* in water ($r=0.54$) and in sediment ($r=0.48; p < 0.05$) thus indicate that, increase in microbenthic diatomaceous population was a result of diatoms settling on the bottom of the creek from water column.

Table MiC 1.1 : Fortnightly groupwise microbenthos (%) in aerated pond

	0	15	30	45	60	75	90	105	120
Coscinodiscus	33	80	58	68	76	88	50	23	49
Foraminifera	3	0	11	14	0	0	17	2	1
Navicula	7	0	26	8	12	12	33	71	12
Nematoda	0	0	5	0	0	0	0	0	2
Pleurosigma	57	20	0	11	12	0	0	0	37
Prorocentrum	0	0	0	0	0	0	0	3	0

Table MiC 1.2 : Fortnightly groupwise microbenthos (%) in non-aerated pond

	0	15	30	45	60	75	90	105	120
Coscinodiscus	34	44	37	51	32	82	75	42	22
Foraminifera	1	0	6	12	5	0	0	0	0
Navicula	4	0	3	0	16	5	0	31	29
Nematoda	0	4	0	0	0	0	0	0	0
Pleurosigma	61	52	54	38	42	14	25	0	49
Prorocentrum	0	0	0	0	0	0	0	27	0
Ostracoda	0	0	0	0	5	0	0	0	0

Table MiC 1.3 : Fortnightly groupwise microbenthos (%) in aerated pond

	0	15	30	45	60	75	90	105	120
Biddulphia	0	0	0	0	0	0	0	0	0
Coscinodiscus	41	29	61	73	45	42	57	68	63
Dictyocha	0	1	2	0	0	1	1	0	0
Foraminifera	0	0	1	1	3	2	1	14	1
Navicula	41	58	35	20	31	50	25	11	8
Nematoda	1	0	0	0	0	0	0	0	0
Pleurosigma	15	12	1	6	21	5	15	7	26
Prorocentrum	1	0	0	0	0	0	0	0	0
Thalassiothrix	0	0	0	0	0	0	0	0	1

Table MiC 1.4 : Fortnightly average microbenthos variations in aerated, non-aerated pond & creek

	0	15	30	45	60	75	90	105	120
Aerated Pond	265	177	168	327	150	230	53	1858	1743
	± 0	± 306	± 123	± 100	± 131	± 125	± 70	± 2053	± 984
Non-aerated Pond	1345	239	619	752	168	195	35	230	973
	± 1006	± 414	± 692	± 332	± 206	± 177	± 41	± 81	± 637
Creek	4432	17834	8599	13960	6237	3715	4684	7532	17728
	± 525	± 5255	± 188	± 2552	± 638	± 4466	± 94	± 871	± 4767

Figure: MF1.9 Fortnightly group-wise microbenthos variations (%) in aerated pond.

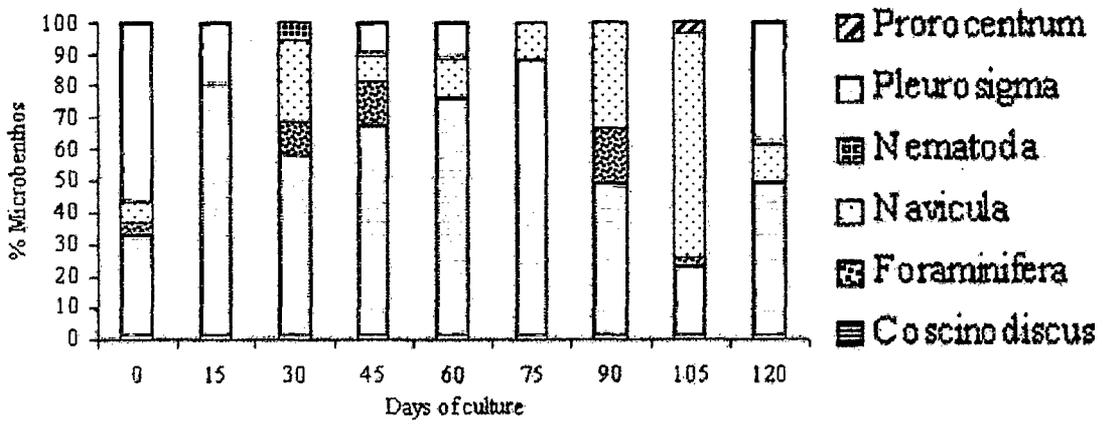


Figure: MF1.10 Fortnightly group-wise microbenthos variations (%) in non-aerated pond.

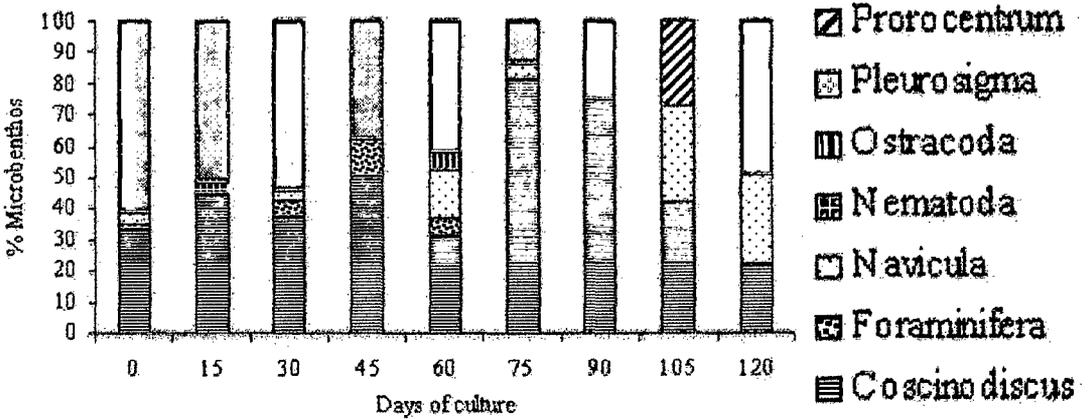


Figure: MF1.11 Fortnightly group-wise microbenthos (%) variations in creek.

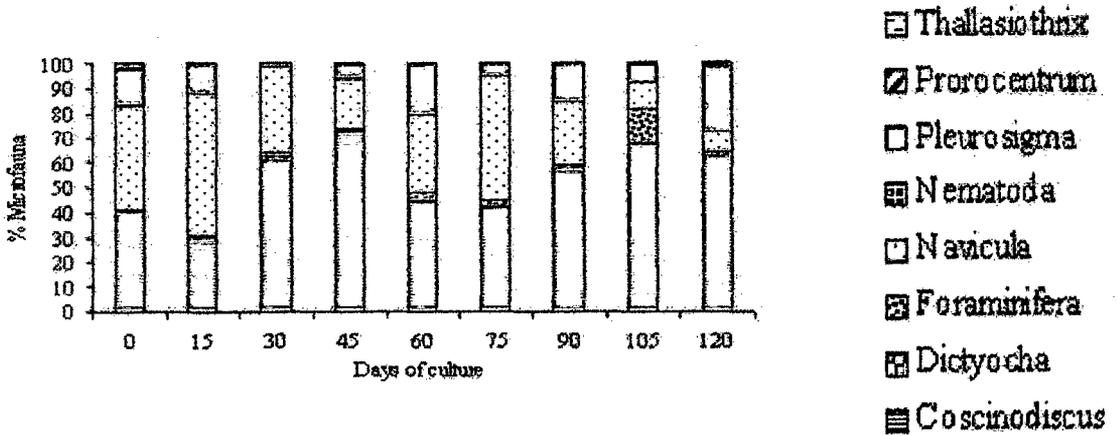
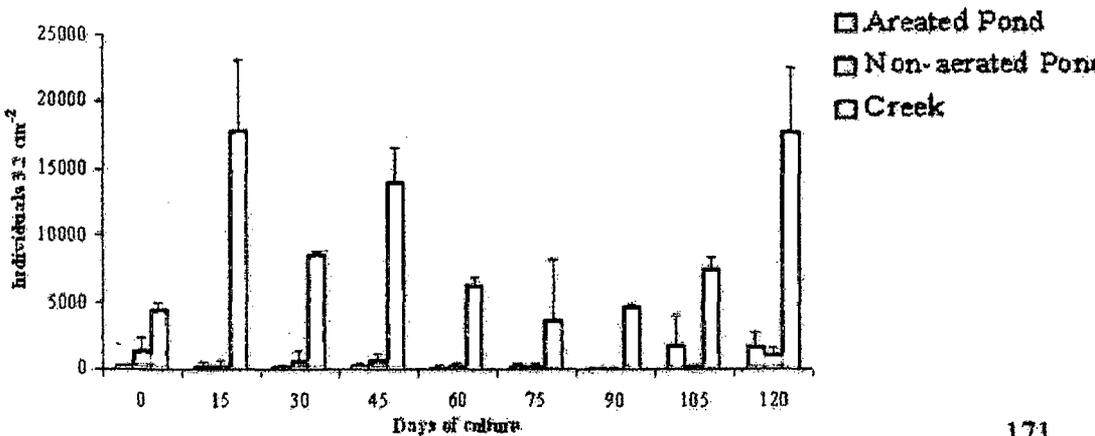


Figure: MF1.12 Fortnightly average microbenthos variations in aerated, non-aerated pond & creek.



4.2.1.4.2 Experimental cycle (EC-2)

It was observed during EC-2 that, two diatomic species, *Pleurosigma* and *Cosinodiscus* dominated the microbenthic population in both the experimental ponds. *Pleurosigma* sp. was clearly the dominant species amongst all diatoms, in the culture ponds. Overall average over 120 day cycle showed that, in aerated pond, *Pleurosigma* contributed (81%) followed by *Cosinodiscus* (17%) (Figure MF2.9). In non-aerated pond *Pleurosigma* contributed (91%) and *Cosinodiscus* only (5%) of the total microbenthic density over 102 DoC (MF2.10). In the aerated pond, average microbenthic density varied from 189 ± 205 on 0 DOC to 9404 ± 9223 no. 3.2cm^{-2} on 90 DOC (Figure MF2.12). For no-aerated pond, the density varied from 154 ± 12 to 4653 ± 1354 no. 3.2cm^{-2} on 75 DoC (Figure MF2.12). The microbenthic density in creek varied from 548 ± 27 on 0 DoC to 6503 ± 656 no. 3.2cm^{-2} on 90 DoC (Figure MF2.12). Unlike culture ponds, the creek showed less diversity but highest density over the period of 102 days culture period of *Navicula* (48%) followed by *Pleurosigma* (29%) and *Cosinodiscus* (22%) Figure (MF2.11). Overall, an increasing trend of microbenthic density was observed for the culture pond as well as for creek. The average microbenthic densities were found to be highest in the aerated pond (Table ST10) followed by creek (Table ST12) and non-aerated pond (Table ST11).

4.2.1.4.2 A) Statistical analysis for Aerated pond

In aerated pond, microbenthos showed an increasing trend till the end of the culture period (Figure MF2.12). Overall, average density was found to be 2982 ± 3369 no. 3.2cm^{-2} (Table ST10). Stepwise-regression analysis carried out showed significant relationship between microbenthos and salinity ($B=-0.93$), pH sediment ($B=-0.67$) and explained 90.1% variance. The model was significant at $F_{2,5}=32.94; p=0.001$.

4.2.1.4.2 B) Statistical analysis for Non-aerated Pond.

For Non-aerated pond also, similar increasing trend, as that of aerated pond was observed (Figure MF 2.12). However, the average density values were lower

(1637±1603) No.3.2cm⁻² (Table ST11), than the aerated pond (Table ST10; Figure MF2.12). The backward multiple regression analysis for microphytobenthos showed significant linear relationship with that of sediment chlorophyll *a* (B=-0.69) and salinity (B=-1.0), it explained 60.8% variance at F_{2,5}=6.43;p=0.041.

4.2.1.4.2 C) Statistical analysis for Creek

The average density of microbenthos in creek was found to be 2861±2271 no.3.2cm⁻² (Table ST 12). Similar to ponds in creek also percent contribution of *Coscinodiscus* and *Pleurosigma* was higher (Table MiC2.3; Figure MF2.11). In the creek, an increasing trend was observed for microbenthic density (Figure MF2.11), but the density values did not show any significant linear relationship with that of water or soil parameters. This indicates that, the variations in the density of microbethos in the creek can be attributed to the turbulent nature of creek due to tidal incursion and excursions.

Table MiC 2.1 : Fortnightly groupwise microbenthos (%) in aerated pond

	0	15	30	45	60	75	90	105
Ceratium	0	0	0	0	0	0	3	0
Coscinodiscus	72	72	5	81	88	51	9	1
Foraminifera	0	0	0	0	2	0	0	0
Navicula	0	0	12	13	5	26	1	0
Nematoda	0	0	4	6	0	23	3	0
Nitzschia closterium	0	0	0	0	5	0	0	0
Nitzschia seriata	0	0	31	0	0	0	0	0
Pleurosigma	28	28	47	0	0	0	83	99

Table MiC 2.2 : Fortnightly groupwise microbenthos (%) in non-aerated pond

	0	15	30	45	60	75	90	105
Coscinodiscus	48	48	4	18	5	4	2	7
Foraminifera	0	0	0	0	0	0	1	0
Navicula	0	0	0	0	1	0	1	0
Nematoda	0	0	0	0	2	3	6	3
Pleurosigma	52	52	96	82	92	93	90	90

Table MiC 2.3 : Fortnightly groupwise microbenthos (%) in creek

	0	15	30	45	60	75	90	105
Biddulphia	0	0	0	0	2	0	0	0
Coscinodiscus	47	47	0	13	10	60	23	35
Dictyocha fibula	0	0	0	0	2	0	0	0
Navicula	19	19	95	87	68	18	1	45
Pleurosigma	34	34	5	0	18	22	76	20

Table MiC 2.4 : Fortnightly average microbenthos variations in aerated, non-aerated pond & creek

	0	15	30	45	60	75	90	105
Aerated Pond	189 ± 205	189 ± 205	1327 ± 748	2459 ± 2574	1106 ± 841	6927 ± 7441	9404 ± 9223	2256 ± 2938
Non-aerated Pond	154 ± 12	154 ± 12	1194 ± 1036	292 ± 391	2256 ± 2095	4653 ± 1354	3061 ± 1367	1336 ± 226
Creek	548 ± 27	548 ± 27	2892 ± 564	6092 ± 459	2128 ± 368	2047 ± 4	6503 ± 656	2135 ± 209

Figure: MF2.9 Fortnightly group-wise percent microbenthos variations in aerated pond.

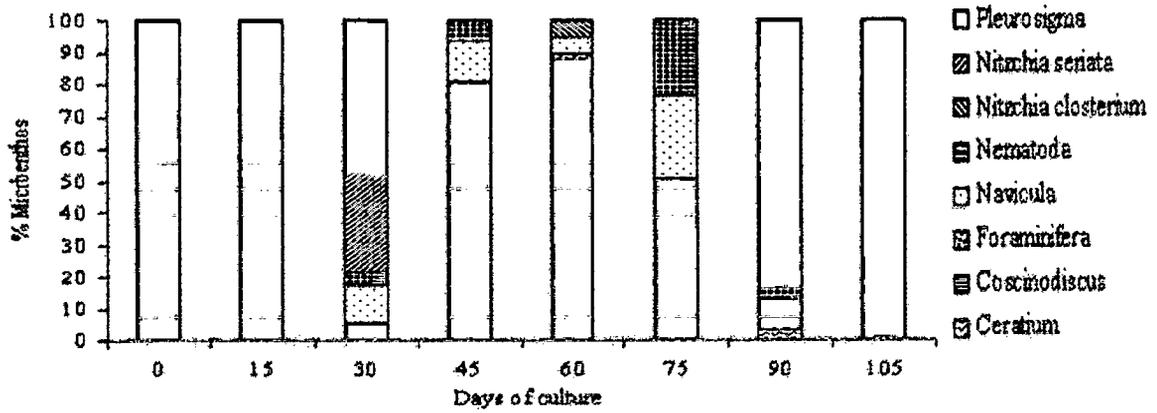


Figure: MF2.10 Fortnightly group-wise percent microbenthos variations in non-aerated pond.

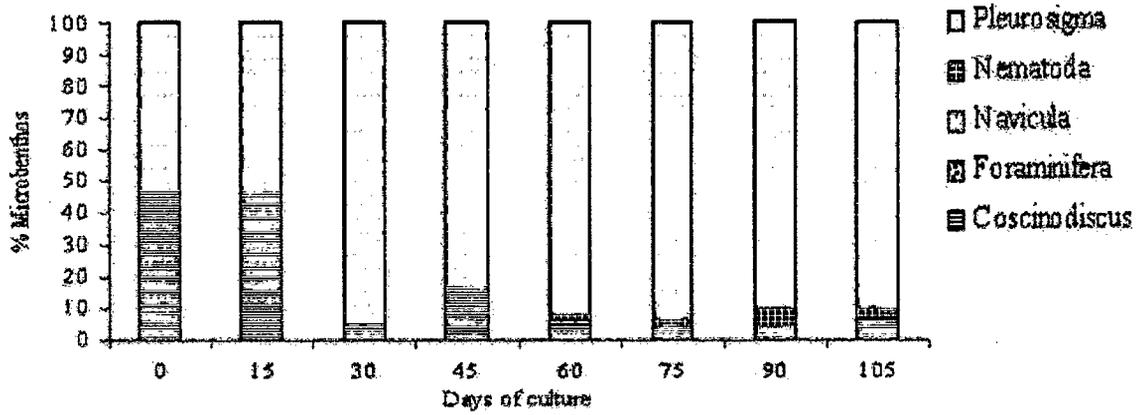


Figure: MF2.11 Fortnightly group-wise percent microbenthos variations in creek.

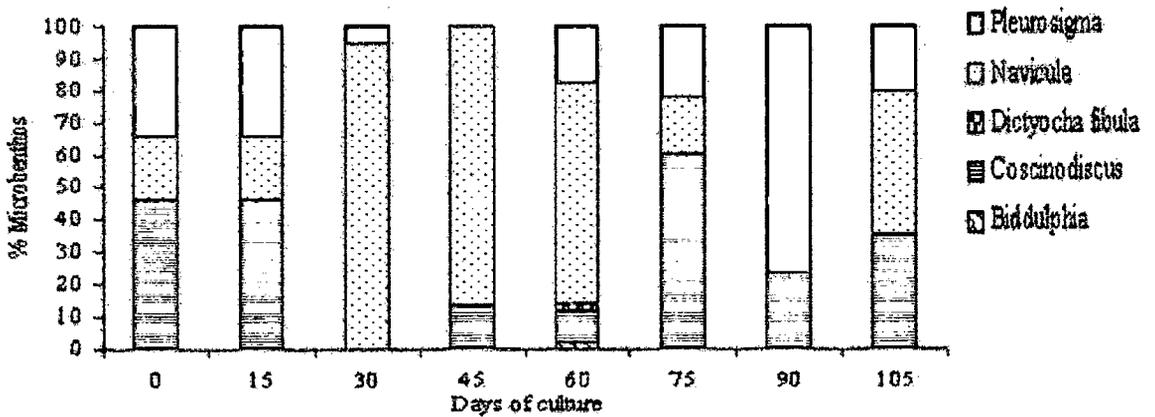
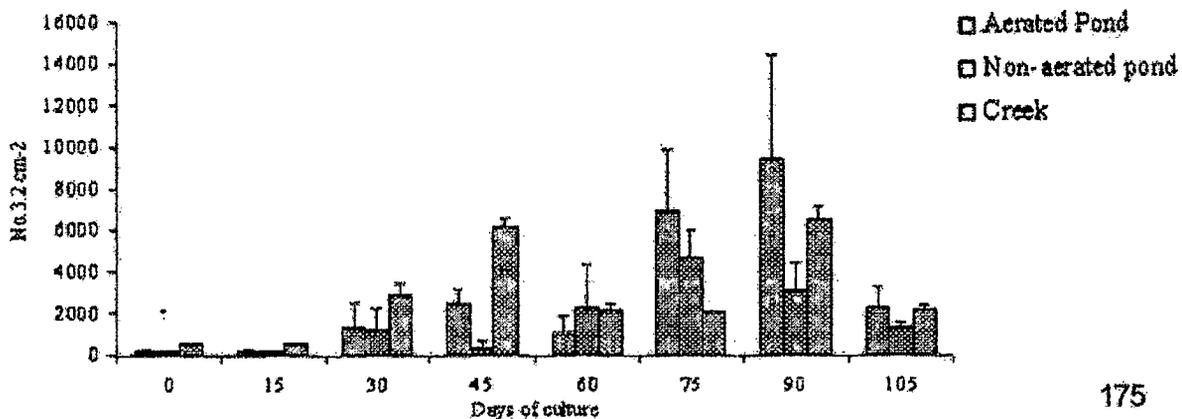


Figure: MF2.12 Fortnightly average microbenthos variations in aerated, non-aerated pond & creek.



4.2.1.4.3 Discussion Microbenthos

During present study microbenthos showed significant correlation with sediment chlorophyll *a* ($r=0.62;p<0.05$) and a weak relationship with that of pond water chlorophyll *a* ($r=0.33;p<0.05$). The respiration by microphytobenthos seems to be an important process to maintain the normal functioning of the cells. The respiration by microphytobenthos in the dark and deeper areas of pond during early morning hours do not show any photosynthesis due to less light penetration and thus lower light intensities may lead to the higher respiration than photosynthetic process. In such conditions less DO concentration may hamper the respiration processes. This seems to be the main reason for observed significant effect of DO on the microbenthos ($p<0.05$). In low light intensities the rate of photosynthesis decreases, and therefore respiration may have overtaken the photosynthesis process, which requires oxygen. Thus decreasing DO may have affected the density of the microphytobenthos. Further, the microbial population in culture ponds may also contribute to the oxygen uptake and lower the oxygen concentration which is deleterious for maintaining the adequate bloom during dark periods (Perkins *et al.* 2001).

During EC-1 sediment pH showed profound effect on the development of microbenthos. The values of pH in the sediment remained low (Figure EC1.4) for most of the times below 7 pH (Figure EC1.4). There were values pH recorded with sudden rise of pH, from 6 to 7 between 60 to 90 DoC (Figure EC1.4) during EC-1. Due to this fluctuations, the low pH levels could have been responsible for the lower abundance of microbenthos observed during this time (Figure MF1.12) during EC-1. During EC-2, however, salinity seems to have affected the microbenthic density both in aerated and non aerated ponds, as was observed on the basis of regression analysis which has been, described in section (4.2.1.4.2 A) and (4.2.1.4.2 B) Rasmuseen *et al.* (1983) has studied the possible causes of temporal fluctuations in primary production of the microphytobenthos due to changes in salinity and showed that, higher salinities, greater than 30 ppt and more than 9 pH, affects the primary productivity negatively.

The significant relationships between chlorophyll *a* in water and sediment and microbenthos as well as simultaneous dominance of diatoms in the microbenthic population asserts that, chlorophyll *a* in the sediment and water can be attributed to have originated from diatomaceous population in the experimental ponds. MacIntyre *et al.* (1996) has defined phytoplankton on the basis of their operational behavior. Phytoplankton operationally has been defined by MacIntyre *et al.* (1996) as “microautotrophs found within the water column and the microphytobenthos are those which are found in the sediment”. According to MacIntyre (1996) the distinction of microautotrophs and microbenthos is artificial in the shallow water systems where there can be large exchange of living and non-living matter between the sediment and the water column. Under calm conditions, the phytoplankton can sink to the bottom. At bottom they continue to photosynthesize given sufficient light and get incorporated as microphytobenthos. These microphytobenthos again become part of the water column and remain in suspension due to waves, currents and any kind of turbulence in the form of movements caused due to benthic dwellers. During present study in the shrimp culture ponds with water depths of 1.4 to 1.6 meters indicates their shallow nature.

Due to shallow nature, water exchange practices and turbulences caused due to wind action may suspend the microbenthos in the water column and similarly may also settle quickly at the bottom of the pond. According to the present results, diatoms were the most abundant group of phytoplankton found and was dominated by *Coscinodiscus*, *Navicula*, *Nitzschia* and *Pleurosigma* sp. These are known as pinnate diatoms, and are most common type of phytoplankton observed in varying temperature, salinity and withstand wide range of light intensities. Solar radiation is a fundamental ecological factor in aquatic and terrestrial ecosystems. Blachard *et al.* (1994) has studied the use of photosynthesis as mechanism of adaptation of microphytobenthic algae to changing environmental conditions by photo acclimation. It was observed during present study, that, microphytobenthic density was higher during EC-2 than EC-1. Underwood *et al.* (2001) studied the primary

production by ^{14}C radio tracer and have showed higher maximum production rates in biofilms exposed to ambient light at mid day, than in biofilms exposed to shaded or ambient light towards the emersion period. Thus, indicating that, light intensities of higher and lower magnitude may have played role in maximum and minimum net primary production by benthic diatoms. Barranguet (1998) has also reported that, in shallow environments with 0.6 to 0.7 m depths, photosynthesis is regulated by both light and temperature, and, temperature has been showed by him to be dominant factor. During present study, however, temperature during both the cycles showed similar range from 28 to 32 $^{\circ}\text{C}$, with no significant differences ($p > 0.05$), neither it, showed any significant linearity with microphytobenthic density. Nutrients like $\text{NO}_3\text{-N}$ and $\text{NH}_3\text{-N}$ concentration showed significant correlation with that of chlorophyll α concentrations in water ($p < 0.05$). This indicates that, neither nutrient nor temperature and salinity were limiting and probably high and low light intensities may have played role in the net production of microphytobenthos. During present study it was observed that, during EC-2 there were abrupt and comparatively longer breaks in the monsoon compared to the EC-1. This would have provided maximum ambient light intensities for the higher production of phyto-benthic diatoms and thus, the average microbenthic abundance was higher during EC-2 than EC-1.

4.2.1.4.4 Discussion for Macro, meio and Microbenthos

During present study it was also, observed that, due to free connection with that of sea water and tidal influences macro, meiofaunal densities were found to be higher in the creek. Macro meio and microbenthos showed weak correlations in between, without any definite pattern. Their correlations are significant ($P < 0.05$) and are depicted in Table ST1-6. These relations however, are not indicative of predation either by macrofauna on meiofauna, as their presence in respective systems is governed by different physico-chemical processes. Further, from the results of macrofauna, meiofauna and microbenthos, various physico-chemical parameters like temperature, pH, E_h and salinity as well as food availability in terms of either microbenthos or organic carbon was found to be affecting reciprocally with that of the other environmental parameters on the macro- and meiofauna. Role of physical and chemical parameters affecting benthic fauna has been studied by Harkantra and Parulekar (1991), Ingole and Parulekar (1998). Further, while studying the effect of salinity on meiofauna Ingole and Parulekar (1998) showed that increasing salinities were favorable for harpacticoid copepod, whereas decreasing salinities favor the production of Turbellaria and *Macrostomus* sp. The dominant groups during present study in macrofauna were Gastropoda and Polychaeta, wherein, for meiofauna, they were, Nematoda and Copepoda, and diatoms dominated in microbenthos population. Though diatoms were dominant, their density was low in the culture ponds during EC-1.

While studying the harpacticoid population, Mirto *et al.* (2000) showed a decrease in the densities of macro and meio-benthos with decreasing phytoplankton pigments. Further, they stated that, Nematodes are more resistant to the organic loading and resulting hypoxic to anoxic condition at the bottom of the fish ponds. During present study, DO did not found to affect macro- meio- or microbenthos significantly (Table AB1 to Table AB5) and DO in the water never depleted below 3 mg.l⁻¹. Jayaraj *et al.* (2007) have shown that, for benthos a single environmental factor is not sufficient for predicting the spatial variability. Further, they have shown that, for spatial benthic variability salinity, temperature and particle size, are together important. The high correlation of salinity and low correlation of DO, with that of

macrobenthos has been shown from tidal impoundment, by Lui *et al.* (2002). Thus, apart from food availability and water quality, there can be many physical factors like space, particle size, turbulence caused due hydrodynamic processes including currents, waves, near the bed (Eckman, 1983) affect the benthos in the creek and sea. Dahms *et al.* (2004) has stated that, macro- and meiofaunal larvae choose settlement sites based on a variety of selection criteria which influences post-larval settlement and development. According to Dahms *et al.* (2004) upon contact with benthic substrata, larvae discriminately screen the substratum for the cues that indicate substratum suitability and complete their pelagic lifecycle, and if site is not suitable larvae re-enter the water column. They further stated that, extrinsic factors such as, food availability, predation competition and chemical cues also limit the benthic density and diversity.

During present study, lower densities of benthos in shrimp culture ponds were observed and probably were due to the parameters like recruitment and pre-stocking activities like pond drying, application of lime etc also, seems to have limited the development of the benthos in the shrimp culture ponds. Another noteworthy practice of water pumping, where the suction of pump placed very near to the bottom advantageously to exploit minimum tidal height sucks water only beneath it and thus the areas below suction inlet can be devoid of any fauna. Besides these, suction inlets are protected by the netting to avoid the large chunks of wood and phytal or domestic debris such as plastic and paper entering the pump to avoid damage to the pump. Thus with this the only option remains that, the flora and fauna only in suspension form in the water during high tide with comparatively stronger current and that also if water is taken up through sluice gates will enter the farms. These practices further can be speculated to diminish the chance of the recruitment of flora and fauna in the culture ponds. The research in this direction in enhancing recruitment and promotion of pond biota and sustaining the available pond biota in shrimp culture practices is needed.

Table MiCST 1: One way ANOVA for microbenthos abundance in two cycles
EC-1 and EC-2 of aerated pond.

SUMMARY				
Groups	Count	Sum	Average	Variance
EC-1	9	4971.691	552.4102	507180.8
EC-2	8	23856.33	2982.042	11351697

ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	25001405.75	1	25001406	4.490231	0.051188	4.543068
Within Groups	83519322.45	15	5567955			
Total	108520728.2	16				

Table MiCST 2 One way ANOVA for microbenthos abundance in two cycles
EC-1 and EC-2 of aerated pond of non-aerated pond.

SUMMARY				
Groups	Count	Sum	Average	Variance
EC-1	9	4555.909	506.2122	196876.4
EC-2	8	13099.79	1637.473	2571839

ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
EC-1	5420126.7	1	5420127	4.152742	0.059606	4.543068
EC-2	19577882.39	15	1305192			
Total	24998009.08	16				

Table MiCST 3 One way ANOVA for microbenthos abundance in two cycles
EC-1 and EC-2 of aerated pond of non-aerated pond of Creek.

SUMMARY				
Groups	Count	Sum	Average	Variance
EC-1	9	84721.34	9413.482	31832333
EC-2	8	22893.13	2861.641	5162303

ANOVA						
Source of Variation	SS	df	MS	F	P-value	F crit
Between Groups	181806838.714	1	1.82E+08	9.3781	0.007902	4.543068
Within Groups	290794785.034	15	19386319			
Total	472601623.7	16				

Table MiCST 4 : Variation in microbenthos density between aerated and non-aerated pond during EC-1

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
Aerated Pond	9	4971.691	552.4102	507180.8
Non-aerated Pond	9	4555.909	506.2122	196876.4

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	9604.15	1	9604.15	0.027282	0.870876	4.493998
Within Groups	5632458	16	352028.6			
Total	5642062	17				

Table MiCST 5 : Variation in microbenthos density between aerated and non-aerated pond during EC-2

SUMMARY

<i>Groups</i>	<i>Count</i>	<i>Sum</i>	<i>Average</i>	<i>Variance</i>
Aerated Pond	8	23856.33	2982.042	11351697
Non-aerated Pond	8	13099.79	1637.473	2571839

ANOVA

<i>Source of Variation</i>	<i>SS</i>	<i>df</i>	<i>MS</i>	<i>F</i>	<i>P-value</i>	<i>F crit</i>
Between Groups	7231455.593	1	7231456	1.038738	0.325405	4.600111
Within Groups	97464747.21	14	6961768			
Total	104696202.805	15				

Table AB1 Effect of DO on macrofauna, meiofauna and microbenthos from aerated pond EC-1

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Macrofauna	Between Groups	23419304	6	3903217	12.414	.076
	Within Groups	628820.67	2	314410.3		
	Total	24048125	8			
Meiofauna	Between Groups	48216.000	6	8036.000	.601	.734
	Within Groups	26754.000	2	13377.000		
	Total	74970.000	8			
Microfauna	Between Groups	4048117.3	6	674686.2	118.755	.008
	Within Groups	11362.667	2	5681.333		
	Total	4059480.0	8			

Table AB2 Effect of DO on macrofauna, meiofauna and microbenthos from non-aerated pond EC-1

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Macrofauna	Between Groups	5155249	8	644406.1		
	Within Groups	.000	0			
	Total	5155249	8			
Meiofauna	Between Groups	813440.0	8	101680.0		
	Within Groups	.000	0			
	Total	813440.0	8			
Microfauna	Between Groups	1575566	8	196945.7		
	Within Groups	.000	0			
	Total	1575566	8			

SECTION 4.3

SHRIMP GUT CONTENT ANALYSIS

Table AB4 Effect of DO on macrofauna, meiofauna and microbenthos from aerated pond EC-2

ANOVA

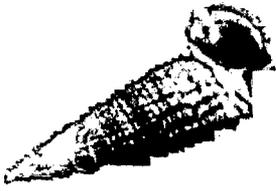
		Sum of Squares	df	Mean Square	F	Sig.
Macrofauna	Between Groups	1.9E+07	6	3168822	1.111	.621
	Within Groups	2851272	1	2851272		
	Total	2.2E+07	7			
Meiofauna	Between Groups	1654664	6	275777.3	1.517	.552
	Within Groups	181804.5	1	181804.5		
	Total	1836468	7			
Microfauna	Between Groups	7.9E+07	6	1.3E+07	20.287	.168
	Within Groups	647522.0	1	647522.0		
	Total	7.9E+07	7			

Table AB4 Effect of DO on macrofauna, meiofauna and microbenthos from non-aerated pond EC-2

ANOVA

		Sum of Squares	df	Mean Square	F	Sig.
Macrofauna	Between Groups	3470767	7	495823.8		
	Within Groups	.000	0			
	Total	3470767	7			
Meiofauna	Between Groups	1592748	7	227535.4		
	Within Groups	.000	0			
	Total	1592748	7			
Microfauna	Between Groups	1.8E+07	7	2571663		
	Within Groups	.000	0			
	Total	1.8E+07	7			

Plate 1 Photographs showing macrofauna, meiofauna and microbenthos



Cerithidium

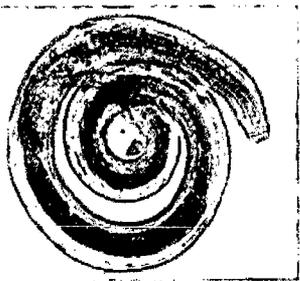


Telescopium

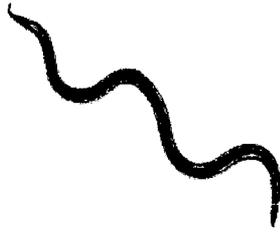


Polychaeta

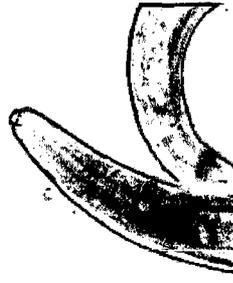
Nematoda



Oligochaeta



Copepoda



Polychaeta



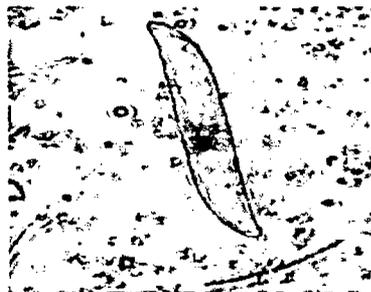
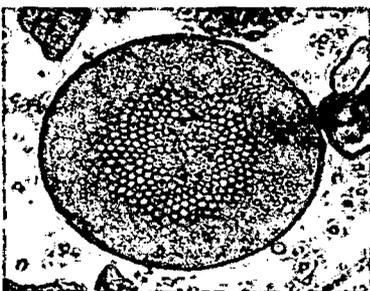
Coscinodiscus



Pleurosigma



Nitzschia



Chapter 4

Section 4.3 Shrimp gut content analysis

4.3.1 Introduction

According to Ruben *et al.* (2005) behavior of the animal depends on the ecological circumstance in which foraging takes place. Roach (1998) showed that, starvation and predation risk determines the ultimate transformations in the populations of predator as well as prey species and has substantial effect on the ecology of the prey species, however, the significance of the predation varies differently in different aquatic systems. According to Roach (1998) the effectiveness of prey predation phenomenon can be modified by various factors such as habitat complexity (Roach, 1998). Shrimp aquaculture system in this point of view can be considered as a unique habitat, where shrimp predate on the natural flora and fauna, and at the same time also compete for prey. These culture systems differ from the natural habitat due to their confined nature. Due to the habitat constraint in shrimp culture ponds the natural food structure of flora and fauna is altered, and has been discussed in Chapter 4; Section 4.2.1.4.4. Therefore the supplemental feed administration is a general and effective means to cater their nutritional needs. According to the stocking method, with low to high supplementary food input varying from modified to intensive shrimp culture method is a general practice for application of supplementary feed in shrimp culture pond since more than last two decades. However, naturally shrimp has been described Chapter 2, Section 2.3 to feed on diverse flora, fauna and detritus, and has been categorized as omnivorous benthic predator. Therefore, according to prey predator relationship it is apparent that, there can be possible changes in the shrimp behavior for selection of the prey or food in general due to limited resources.

According to Jiang and Morin (2005) various biotic and abiotic factors affect the prey predator relationship, and productivity, by far determines the patterns of species abundance and distribution on which predator is dependent. Thus understanding bottom up (productivity based) and top-down (predation based) approaches are important in commercial shrimp culture practices for effective

management of resources in terms of natural food and application of artificial feed. Therefore, it was hypothesized during this study that, shrimp totally is dependent on the supplementary food. Further, it was also hypothesized that shrimp are unable to detect supplementary food in the grow out earthen ponds, which is possible, due to the feed application patterns, site, quantity, impaired water and sediment quality, leading to offensive odors, may suppress the detecting abilities of shrimp and shrimp may remain undernourished. In such scenario absence of natural food may hamper its growth further, which is highly possible due to confined nature of shrimp culture practices with limited water exchanges. Hence, it was decided to study the natural fauna available and its contribution and prey predator relationship if any is prevalent in the shrimp culture systems. For this shrimp gut content were observed for the presence of natural fauna collected at two different time periods and simultaneous supplementary feed utilization was studied. Described here are the results and discussion below.

4.3.2 Results and Discussion

4.3.2.1 Experimental Cycle (EC-1)

4.3.2.1. A) Aerated Pond 10.00 Hrs.

During the EC-1 *Pleurosigma* and *Navicula* were most dominant items recorded on 75 DoC constituted (76%) and (13%) respectively of the total fauna. Thereafter the occurrence of *Pleurosigma* and *Navicula* in gut of the shrimp declined from 90 DoC to 120 DoC, (Figure G1.1). The least number of 19 organisms.individuals⁻¹ were recorded on 105 DoC for aerated pond, and just 36 organisms.individuals⁻¹ in non-aeration pond Table G1.5; Figure G1.5. Fortnightly percent contribution of natural fauna in the gut has been depicted in Table G1.1 and Figure G1.1 at 10.00 am and Table G1.2 and Figure G1.2 at 10.00pm.

4.3.2.1. B) Aerated Pond 22.00 Hrs.

In aerated pond at night from 45 DoC till 75 DoC crustacean body remain dominated and contribution for crustacean body parts was highest to be (72%) on 60 DoC. Thereafter conspicuously phytal material was higher and was highest (55 %), (36%),

and (22%) on 90, 105 and 120 DoC, respectively. This feature was even observed at the start of the culture period on very first fortnight with (80%) of algal phytal material observed in the shrimp gut. This trend indicates that, in absence of diatoms and also other fauna like crustaceans shrimp do the surviving act on the available phytal or algal material. Fortnightly percent contribution of natural fauna in the gut has been depicted in Table G1.2 and Figure G1.2. For aerated pond backward multiple regression analysis was performed to see whether there is any significant linear relationship between gut fauna and macro, meio and microbenthos, Chlorophyll *a* water and sediment, as independent variables. This analysis explained 75.7% variance due to macrofauna ($B=-0.695;p=0.001$) and chlorophyll *a* ($B=-0.621;p=0.012$) in the sediment $F_{2,6}=13.43;p=0.006$.

4.3.2.1. C) Non-aerated Pond 10.00 Hrs

During morning time in non-aerated pond crustacean body remains were observed in par with the algal phytal materials and diatoms. This observation is different in view that, previous observations followed day night patterns in the occurrence being algal material higher during day time and crustacean material at night. This may be an indication of the preference by shrimp when alternate resources are present at plenty. Fortnightly percent contribution of natural fauna in the gut has been depicted in Table G1.3 and Figure G1.3. Crustacean body parts were observed to be highest on 45 and 90 DoC being (83%) and (93%) respectively. At morning initially i.e. on 30 DoC *Navicula* (55%), *Pleurosigma* (12), and algal strands (21%) dominated as gut biota of shrimp which eventually decreased. With an exception for algal material which was highest being 48 and 70 % on 105 and 120 DoC. (Table G1.3; Figure G1.3). The trend indicates that algal and diatomaceous food intake was higher during day time.

4.3.2.1. D) Non-aerated Pond 22.00 Hrs

On all culture days crustacean body remains were highest observed item in the gut content with 100% occurrence on 105 and 120 DoC. Fortnightly percent contribution

of natural fauna in the gut has been depicted in Table (G1.4) and Figure (G1.4). With an exception on 75 and 90 DoC the % occurrence of gut fauna for algal material was intake was higher (Table G1.4; Figure G1.4) much similar to the aerated pond at 22.00 hrs (Table G1.2; Figure G1.2).

One way ANOVA with single factor showed that, there were significant difference in the means of average densities of gut contents in between morning and at night ($p=0.048; p \leq 0.05$) in the non-aerated pond. Stepwise multiple regression analysis was performed to see whether there is any significant linear relationship between gut fauna as dependent and macro, meio and microbenthos, Chlorophyll *a* water and sediment, phaeopigments in the water and sediment, OC and POC as independent variables. The stepwise regression analysis explained (55.2 %) variability in gut fauna due to macrofauna alone ($B=-0.78; F_{1,7}=10.71; p=0.013$). The backward multiple regression analysis however explained 73.1% variability of gut fauna due to chlorophyll *a* from water ($B=0.533; p=0.055$) and macrobenthos ($B=-1.087; p=0.046$) with overall relationship significant at $F_{2,6}=5.69; p=0.041$.

Table G1.1 : Fortnightly natural biota contribution (%) in the gut of the shrimp in aerated pond, 10.00am.

	30	45	60	75	90	105	120
Algal Strands	24	42	4	3	66	63	47
<i>Coscinodiscus</i>	21	0	6	3	6	0	0
Crustacian body parts	17	42	8	4	25	0	13
Eggs and larva	14	0	9	0	0	0	0
Fish scales	11	1	11	0	3	0	0
Gastropod shell remains	8	7	13	0	0	0	0
<i>Navicula</i>	4	5	15	13	0	0	0
<i>Pleurosigma</i>	1	3	17	76	0	38	33
Polychaeta remains	0	0	18	0	0	0	7

Table G1.2 : Fortnightly natural biota contribution (%) in the gut of the shrimp in aerated pond, 10.00pm.

	30	45	60	75	90	105	120
Algal strands	34	15	3	16	55	36	22
<i>Coscinodiscus</i>	26	0	0	0	16	0	11
Crustacian body parts	14	63	72	67	29	0	11
Eggs and larva	3	0	0	0	0	0	0
Fish scales	1	1	0	0	0	0	0
Gastropod shell remains	3	0	9	2	0	0	11
<i>Navicula</i>	2	8	0	2	0	0	0
Nematoda	5	0	2	2	0	0	0
<i>Pleurosigma</i>	2	2	2	6	0	64	44
Polychaeta remains	8	12	12	4	0	0	0

Table G1.3 : Fortnightly percent biota contribution (%) in the gut of the shrimp in non-aerated pond, 10.00am.

	30	45	60	75	90	105	120
Algal material	21	0	10	0	0	48	70
<i>Coscinodiscus</i>	1	0	1	0	7	0	0
Crustacian body parts	2	83	19	16	93	52	30
Eggs and larva	7	2	0	0	0	0	0
Fish scales	0	1	0	0	0	0	0
<i>Navicula</i>	55	4	29	0	0	0	0
<i>Pleurosigma</i>	12	6	39	84	0	0	0
Polychaeta remains	1	5	3	0	0	0	0

Table G1.4 : Fortnightly natural biota contribution (%) in the gut of the shrimp in non-aerated pond, 10.00pm.

	30	45	60	75	90	105	120
Algal material	8	0	5	76	37	0	0
<i>Coscinodiscus</i>	0	0	0	0	5	0	0
Crustacian body parts	56	73	56	21	51	100	100
Eggs and larva	2	0	5	0	0	0	0
Fish scales	0	21	5	3	7	0	0
Gastropod shell remains	5	4	15	0	0	0	0
<i>Navicula</i>	20	0	3	0	0	0	0
<i>Pleurosigma</i>	1	0	3	0	0	0	0
Polychaeta remains	7	1	8	0	0	0	0

Table G1.5 : Fortnightly total natural biota contribution in the gut of the shrimp.

	30	45	60	75	90	105	120
Aerated Pond	337	338	395	497	63	19	24
Non - aerated Pond	183	354	144	108	55	36	18

Figure G1.1 Fortnightly percent contribution of natural biota in shrimp gut in aerated pond at 10.00 am

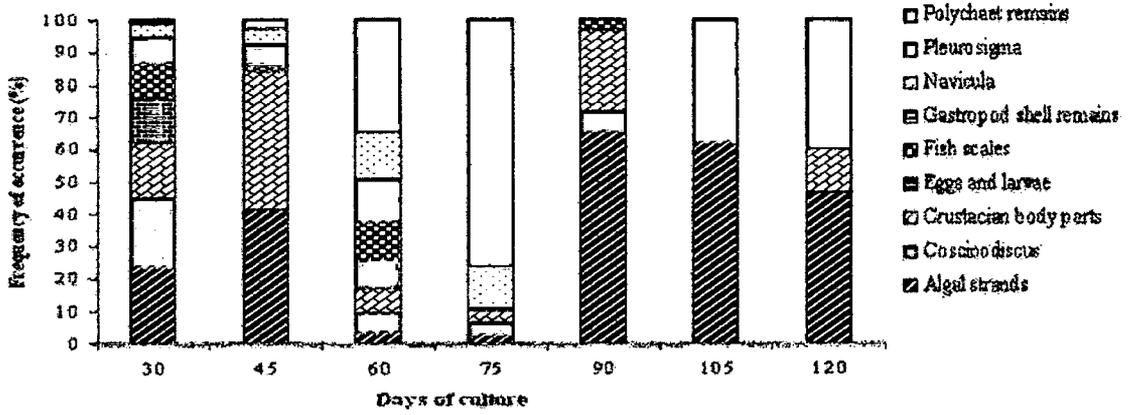


Figure G1.2 Fortnightly percent contribution of natural biota in shrimp gut in aerated pond at 10.00 pm

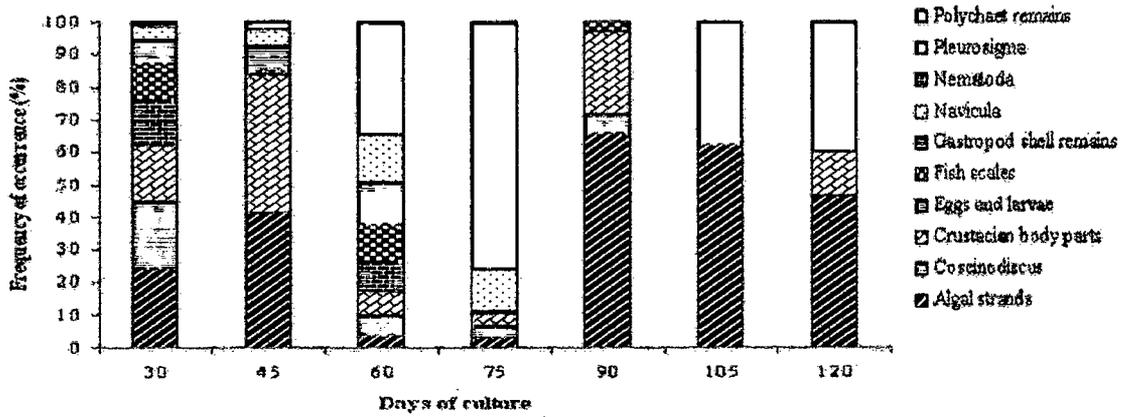


Figure G1.3 Fortnightly percent contribution of natural biota in shrimp gut in non-aerated pond at 10.00 am

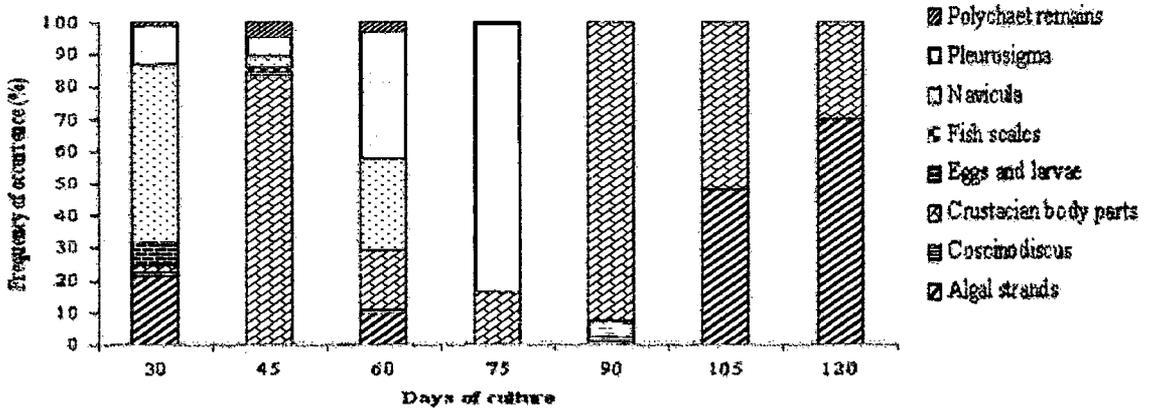


Figure G1.4 Fortnightly percent contribution of natural biota in shrimp gut in non-aerated pond at 10.00 pm

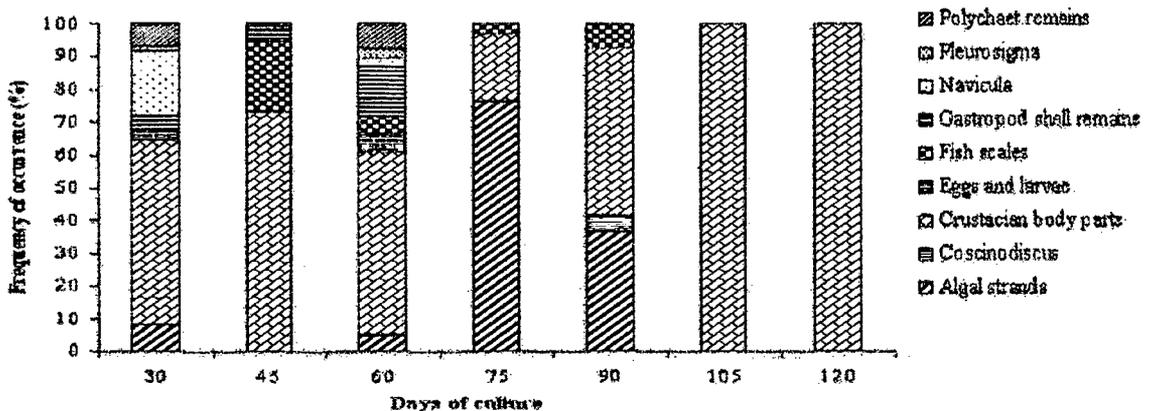
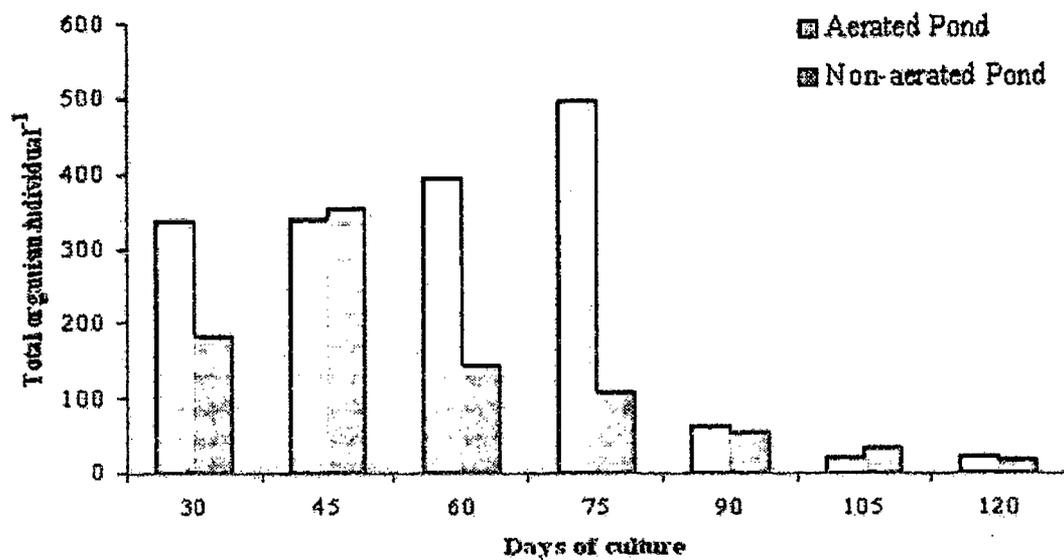
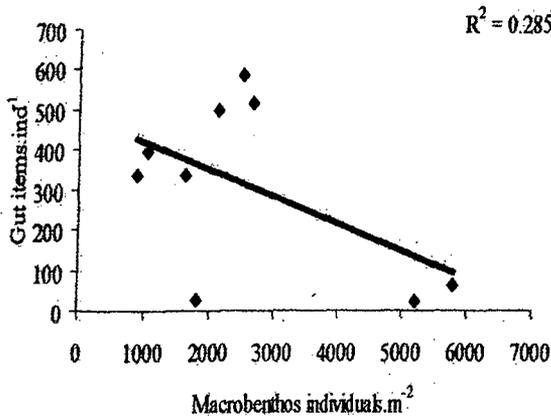


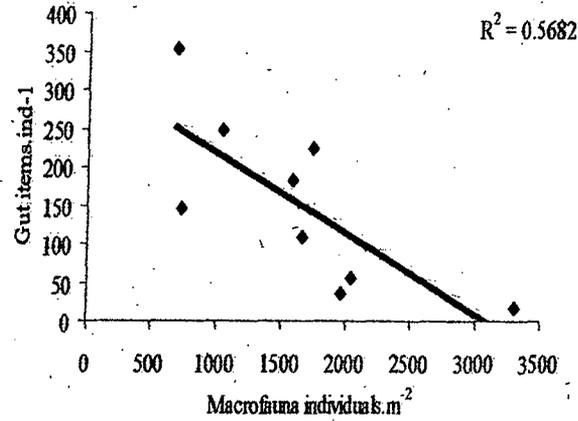
Figure G1.5: Fortnightly total gut biota of shrimp



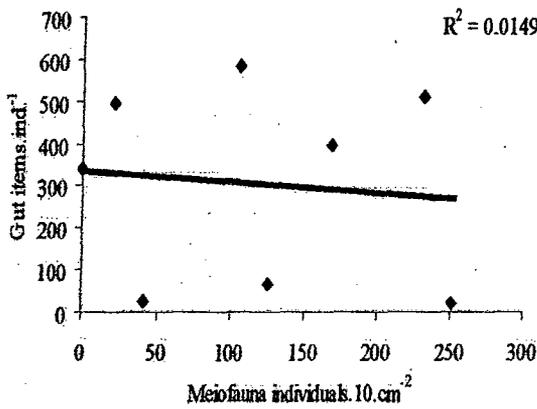
G1.6 Co-relation Gut biota Vs macrofauna aerated pond



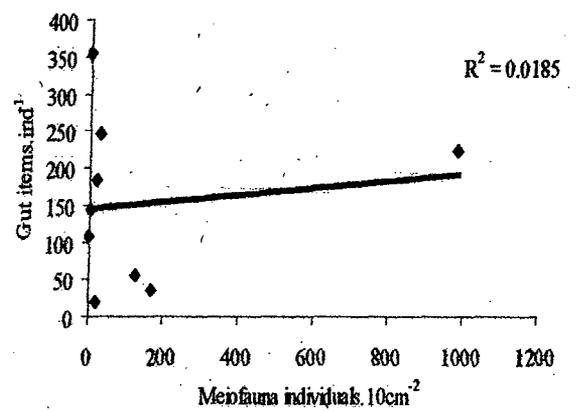
G1.7 Co-relation Gut biota Vs macrofauna non-aerated pond



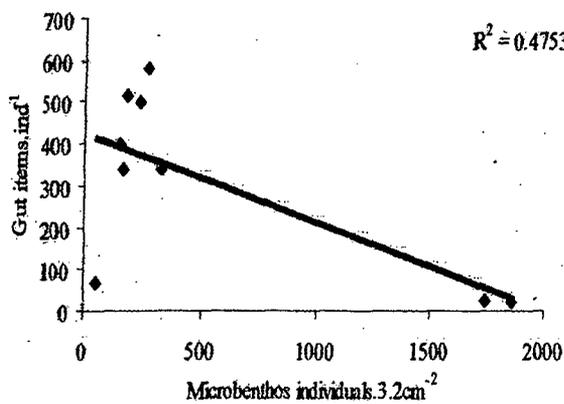
G1.8 Co-relation gut biota Vs meiofauna aerated pond



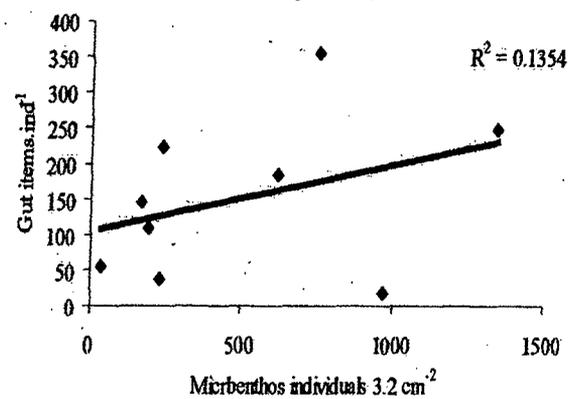
G1.9 Co-relation gut biota Vs meiofauna non-aerated pond



G1.10 Co-relation gut biota Vs microbenthos aerated pond



G1.11 Co-relation gut biota Vs microbenthos non-aerated pond



4.3.2.2 Experimental Cycle (EC-2)

4.3.2.2 A) Aerated Pond 10.00 Hrs.

The total gut content items showed decreasing trend from 45 DoC till the end of the culture period (Figure G2.5) especially for aerated pond than non aerated pond. Highest percent was observed on 30 and 45 DoC for *Navicula* (61%) which reduced to (37%) on 60 DoC. From 60 DoC onwards percent contribution of *Navicula* was decreased and was found to be replaced by phytal material with highest percentage of (83%) on 75 DoC and the crustacean body parts (98%) on 105 DoC at the end of the culture period. Fortnightly percent contribution of total natural biota in the gut has been depicted in Table G2.1 and Figure G2.1. The analysis indicates that, when diatomaceous food decreased shrimp switched over to the algal material as its food and at the end it feed exclusively on crustacean diet.

4.3.2.2 B) Aerated Pond 22.00 Hrs

Fortnightly percent contribution of natural fauna at night in the gut of the shrimp has been depicted in Table G2.2 and Figure G2.2. It can be seen from table and figure that, the crustacean body remains in the gut showed definite pattern with higher values on 30 and 45 DoC being (86%) and (67%). The percent gut content of crustacean body remains decreased on 60 DoC with just (16. %) occurrence of crustacean body parts, but at same time the variety of food items taken also increased with gastropod (19%) and polychaeta contributing (13%) remains till the end of the culture period. Along with it algal strand *Navicula* and *Pleurosigma* intake was also higher (Table G2.2 and Figure G2.2). One way ANOVA with single factor showed that, there were significant difference in the average densities in between morning and at night $p < 0.05$. Backward multiple regression analysis explained (99.5%) variability in gut content due to macrofauna ($B=0.392$), meiofauna ($B=1.58$), microfauna ($B=-4.93$) chlorophyll *a* water ($B=-3.0$), phaeophytin in the water ($B=2.71$) and phaeophytin in the sediment ($B=1.87$) with overall relation significant at $F_{6,1}=249.49; p=0.048$. Thus in aerated pond the degradation products of

phytoplankton from water and sediment followed by meiofauna and probably macrofauna were also taken up.

4.3.2.2 C) Non-aerated Pond 10.00 Hrs

Fortnightly percent contribution of natural fauna in the gut of the shrimp has been depicted in Table (G2.3) and Figure (G2.3). *Pleurosigma* sp. showed its presence in the gut of the shrimp till 60 DoC with (74%) occurrence on 45 DoC. Thereafter it was the *Navicula* sp. which was more dominant than *pleurosigma* sp. on at least 60 and 90 DoC being 49 and 28 % respectively. Also, crustacean body parts dominated at the later half being 84% 30 % and 100% on 75 , 90 and 105 DoC respectively, and interestingly *Navicula*, *Pleurosigma*, *Cosinodiscus* and algal strands were less preferred (Table G2.3; Figure G2.3). It was observed that highest gut biota contribution of (74%) on 45 DoC is consistent with the day time observations, implies that, *Pleurosigma* sp. was abundant enough in water and shrimp might have fed in the water column due to its abundance than the crustaceans which up-till now was found to be dominant food observed during the night time.

4.3.2.2 D) Non-aerated Pond 22.00 Hrs

Fortnightly percent contribution of natural fauna in the gut at night has been depicted in Table G2.4 and Figure G2.4. Apart from crustacean body part contribution of 49% on 30 DoC at night, *Pleurosigma* and *Navicula* sp. were also found to be preferred on 30 DoC. Thereafter, crustacean body remains were found to be dominant and showed increasing occurrence in the gut content of the shrimp with an average value of 70% from 45 to 100% on 105 DoC. Overall occurrences of biota in the gut was more stable, however, it showed fortnightly cyclic variations. The One way ANOVA with single factor showed that, there were significant difference in the means of the average densities in between morning and at night ($p < 0.05$) in the non-aerated pond.

Table G 2.1 : Fortnightly natural biota contribution (%) in the gut of the shrimp in aerated pond, 10.00am.

	30	45	60	75	90	105
Algal material	9	9	17	83	28	0
<i>Coscinodiscus</i>	1	4	14	0	0	0
Crustacian body parts	1	2	8	15	33	98
Gastropod shell remains	0	1	2	2	6	0
<i>Navicula</i>	61	61	37	0	0	0
Nematoda	0	0	0	0	0	0
<i>Pleurosigma</i>	28	22	21	0	0	0
Polychaeta remains	0	0	1	0	33	2

Table G2.2 : Fortnightly natural biota contribution (%) in the gut of the shrimp in aerated pond, 10.00am.

	30	45	60	75	90	105
Algal strands	9	7	26	47	18	0
Crustacian body parts	86	67	16	35	36	100
Gastropod shell remains	0	10	19	6	14	0
<i>Navicula</i>	3	4	19	0	0	0
<i>Pleurosigma</i>	0	3	6	0	9	0
Polychaeta remains	2	7	13	12	23	0

Table G2.3 : Fortnightly natural biota contribution (%) in the gut of the shrimp in aerated pond, 10.00am.

	30	45	60	75	90	105
Algal strands	6	5	2	6	30	0
<i>Coscinodiscus</i>	1	1	0	0	0	0
Crustacian body parts	8	8	2	84	30	100
<i>Fragellaria</i>	32	0	0	0	0	0
Gastropod shell remains	1	1	1	1	0	0
<i>Navicula</i>	6	11	49	0	28	0
<i>Pleurosigma</i>	45	74	45	0	11	0
Polychaeta remains	0	0	1	8	0	0

Table G2.4 : Fortnightly natural biota contribution (%) in the gut of the shrimp in aerated pond, 10.00am.

	30	45	60	75	90	105
Algal strands	12	10	32	5	0	0
<i>Coscinodiscus</i>	1	2	0	0	0	0
Crustacian body parts	49	70	58	67	63	100
Fish scales	0	1	0	0	3	0
<i>Fragellaria</i>	0	0	0	0	0	0
Gastropod shell remains	4	3	0	1	0	0
<i>Navicula</i>	21	6	3	11	20	0
Nematoda	0	1	0	0	0	0
<i>Pleurosigma</i>	9	2	1	15	14	0
Polychaeta remains	4	5	6	1	0	0

Table G2.5 : Fortnightly total natural biota contribution in the gut of the shrimp.

	30	45	60	75	90	105
Aerated Pond	838	580	536	235	40	582
Non Aerated Pond	345	583	558	404	88	443

Figure G2.1: Fortnightly percent contribution of natural biota in shrimp gut in aerated pond at 10.00 am

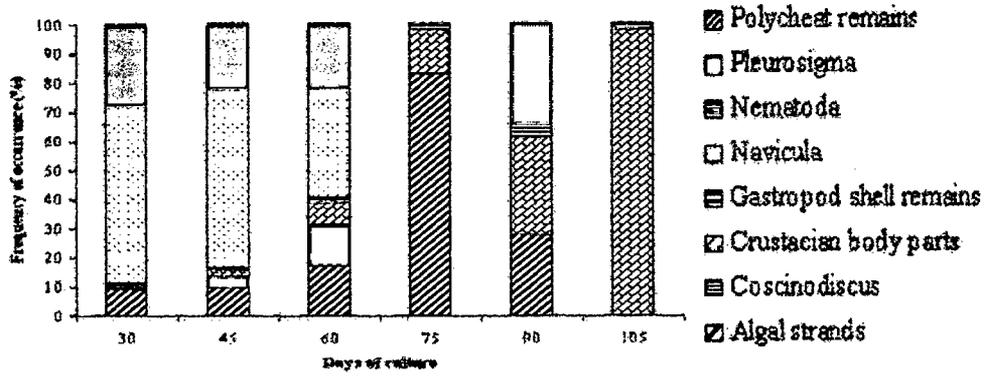


Figure G2.2: Fortnightly percent contribution of natural biota in shrimp gut in aerated pond at 10.00 pm

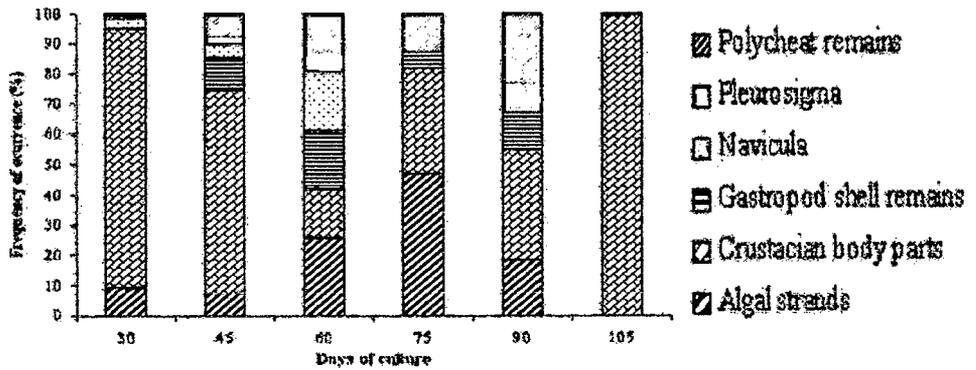


Figure G2.3: Fortnightly percent contribution of natural biota in shrimp gut in non-aerated pond at 10.00 am

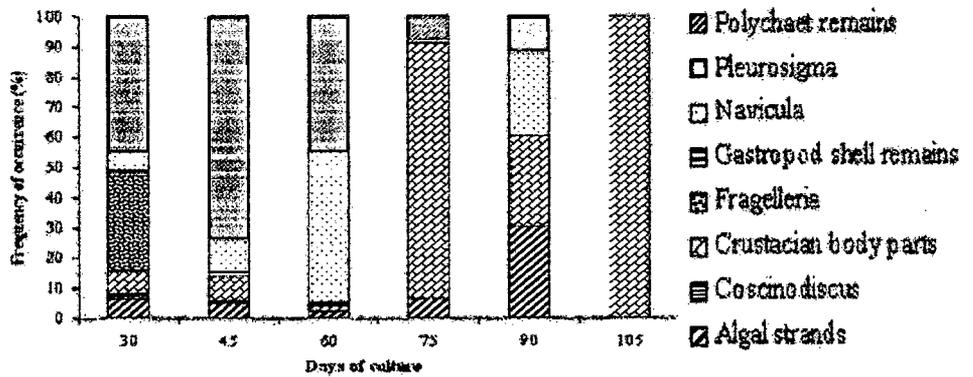


Figure G2.4: Fortnightly percent contribution of natural biota in shrimp gut in non-aerated pond at 10.00 pm

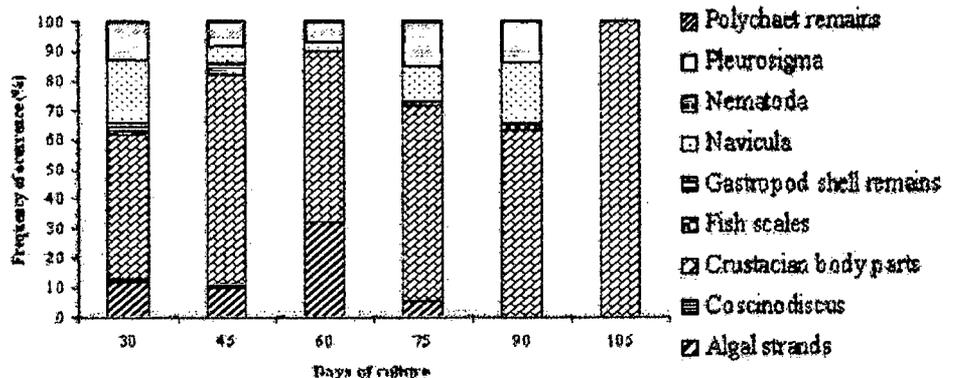
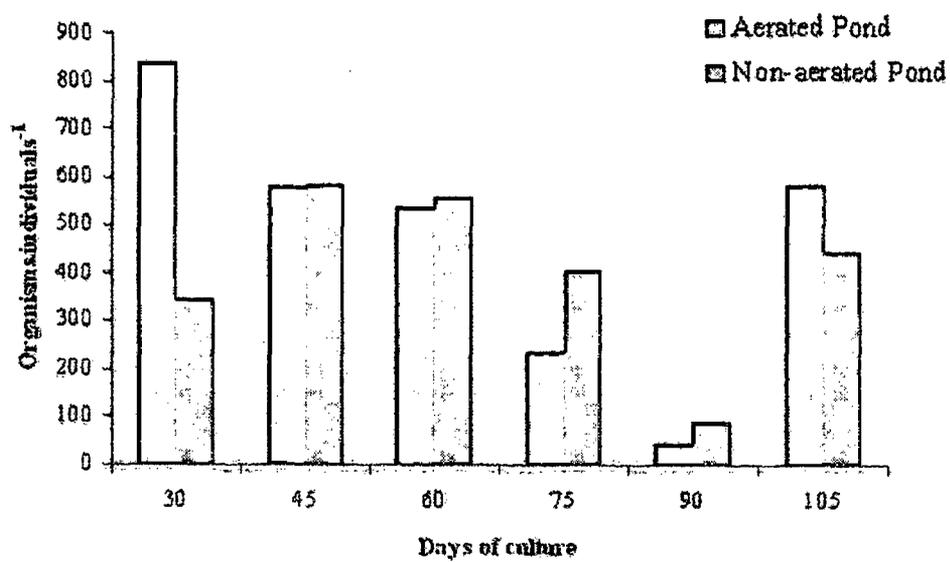
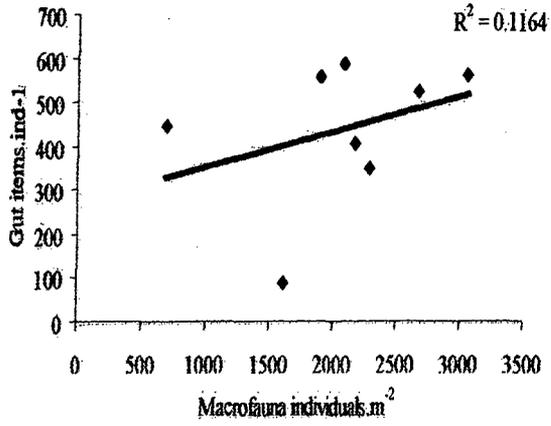


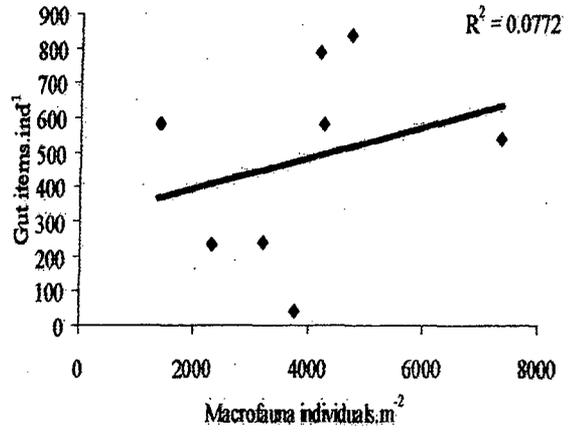
Figure G2.5: Fortnightly total gut biota of shrimp



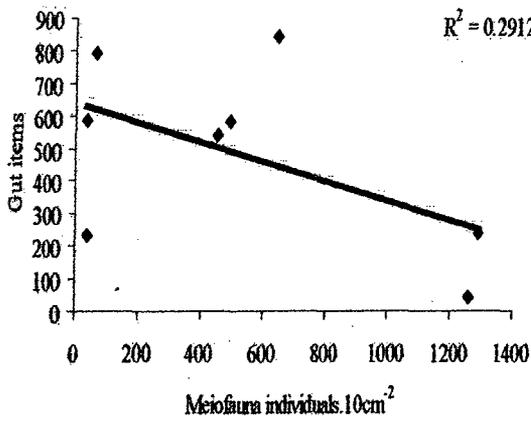
G2.6 Co-relation gut biota Vs macrofauna aerated pond



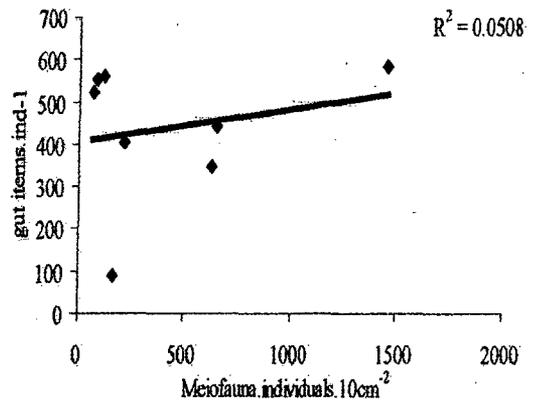
G2.7 Co-relation gut biota Vs macrofauna non-aerated pond



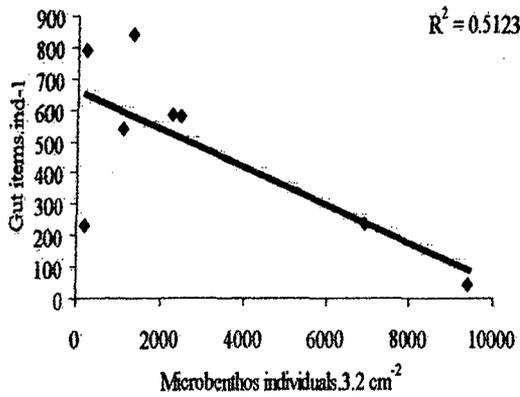
G2.8 Co-relation gut biota Vs meiofauna aerated pond



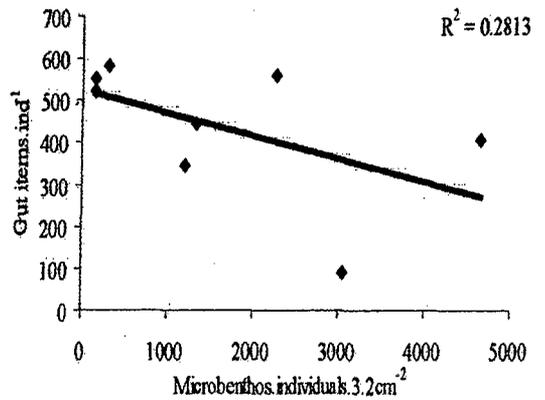
G2.9 Co-relation gut biota Vs meiofauna non-aerated pond



G2.10 Co-relation gut biota Vs microbenthos aerated pond



G2.11 Co-relation Gut biota Vs microbenthos non-aerated pond



4.3.2.3 Shrimp Gut Content Analysis Discussion

From this study during EC-1 it was understood that, shrimp in both aerated and non-aerated pond did feed on the macrobenthos and crustacean body parts, and along with it, shrimp also fed on the phytoplankton or diatoms suspended in the water column. Further, during EC-2 it preferred phytoplankton diet. It can be seen that, there was significant but weak correlation amongst gut biota and macrofauna ($R^2=0.28$) in aerated pond and ($R^2 = 0.56$) during EC-1 depicted in Figure G1.6-1.7. During EC-2 macrofauna showed weak correlation ($R^2=0.11$) in aerated and non aerated pond ($R^2= 0.077$) depicted in Figure G2.6, & G2.7.

For meiofauna during both cycles EC-1 and EC-2 weak correlations were noted. During EC-1 gut biota and meiofauna showed correlation $R^2=0.014$ and $R^2=0.018$ and are depicted in Figure G1.8-1.9 for aerated pond and non aerated pond respectively. Further during EC-2 non aeration pond also showed weak co-relations for gut biota and meiofauna, were reported, being ($R^2=0.29$ and $R^2=0.050$) depicted in Figure G2.8-2.9 for aerated and non aerated pond, respectively.

Gut biota showed significant strong correlations with microbenthos both in aerated pond being ($R^2=0.47$ and $R^2=0.51$) depicted in Figure G1.10 and G2.10 during EC-1 and EC-2 respectively. In non aerated pond microbenthos showed weak correlations compared to aeration pond. The correlations were found to be ($R^2=0.13$) and ($R^2=0.28$) during EC-1 and EC-2 respectively for non aerated pond. During EC-2 for aerated pond macro, meio as well as microbenthos showed significant relationship ($P<0.05$) only in aerated pond as per Figure G2.6;2.8;2.10, respectively. For non-aerated pond however, there was weak co relationship with the gut biota occurrence and the benthos. From correlation analysis as shown in Figure G2.10-11 it was observed that during EC-2 microbenthos were taken up more effectively in both aerated as well as non-aerated pond than, other two benthic components and showed even higher degree of correlations than EC-1. This again is in context with the results obtained already discussed earlier (Chapter 4 Section 4.2.1.4.3) that during EC-2 microphytobenthos density was higher compared to EC-1.

Thus, from direct gut content observations it implies that, phytoplankton and diatoms in the sediment formed an important part in the diet of the shrimp along with artificial feed. Overall from Figure G1.5 & G2.5, it can be seen that, total natural biota decreased in the gut of the shrimp and this especially was observed toward the end of the culture period. Again from same Figures G1.5 and G2.5, it can be seen that, total gut biota was higher in aerated pond than non aerated pond indicating better predation by shrimp. Further, it is clear from the percent occurrence of gut fauna that, clear food preference was existent for day and night time (Figure G1.1 to Figure G1.4.) during EC-1 and (Figure G2.1 to Figure G2.4) during EC-2. Shrimp towards the end of the culture period at night time preferred the crustacean diet (Figure G1.2; G1.4 during EC-1) and (Figure G2.2; Figure 2.4 during EC-2) due to absence of the diatoms during night and again preferred diatoms during daytime (Figure G1.1; Figure G1.3 during EC-1) and (Figure G2.1; Figure G2.3). Further, it can be seen from the meiofauna percentage occurrence (Figure MF1.5 & Figure MF1.6) that, copepod and nauplii were the second most dominant group, after the nematode, during EC-1. Their low densities and cyclic existence especially during EC-2 (Figure MF2.5 & Figure 2.6) indicates that, they were predated upon by the shrimp during night time. It is known that, diatoms migrate deeper in to the sediment during night time, and they again appear during the day time on the surface. The vertical migratory rhythms of benthic diatoms in tropical intertidal sand flat and influence of radiance and tides has been studied by Mitbavkar and Anil (2004). It is evident from the percent microbenthic densities (Figure G1.2 and G1.4 & Figure G2.2 & G2.4) that, microbenthic diatoms were abundant during both the culture periods in the day hours.

The significant linear relationship of shrimp weight and length with that of sediment chlorophyll *a* and amount of artificial feed indicates its importance in the shrimp diet. While discussing the use of microalgae in aquaculture Feuga (2000) has stated that, the requirement of micro-algae for different organisms like shrimp or filter feeding mollusks is different, and it depends largely on the bio-chemical composition of the algal species. Biochemical compositions of algae based on their amino acid composition has been studied by Brown and Jeffery (1995;1997) and

have mentioned that, diatom species consists of higher proteins, and knowledge of difference in the protein content of different diatoms is necessary for their usefulness in the shrimp diet, as shrimp requires high protein rich diets. Kelly *et al.* (2001) has sited that, microphytobenthos can supply up to (33%) of the organic budget in the estuarine coastal system and thus provide important energy source for the estuarine food web. Duerr *et al.* (1998) has showed the use of cultured microalgae as aquaculture feeds.

The use of *Skeletonema* sp with high protein content is in use mostly for larval rearing in shrimp hatcheries and role of other diatoms and phytoplankton in adult shrimp is necessary. The nutritional properties and gross chemical composition of 40 microalgae has showed that, microalgae as primary producers are important as the food in shrimp diet due to their variations in biochemical composition (Durr *et al.*, 1998). Lebeau and Robert (2003) has sited that, diatomic species of *C.gracilis* and *S.costatum* are in use for penaeid shrimp culture in Japan. In present study however, the pinnate diatoms like *Nitzschia* and *Navicula* dominated in the gut fauna of adult shrimp. Moorthy and Altaf (2002) had showed similar results for the gut content and have shown presence of phytoplankton and pinnate diatoms like *Nitzschia* in *P.monodon* in modified extensive shrimp culture ponds. The direct evidence of diatom availability in the gut of the adult shrimp and their role in shrimp nutrition however up till now has been lacking. Hence, the role of these two diatoms in shrimp nutrition needs further study.

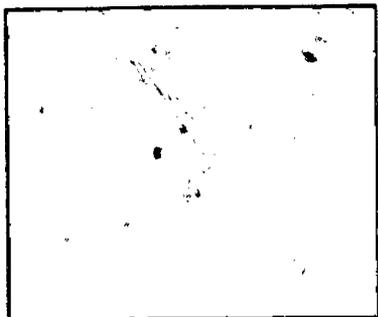
During present study, second abundant fauna in the shrimp gut was found to be crustacean body parts. Wassenberg and Hill (1987) studied the natural diets of the tiger prawns, where he found a wide variety of food organism in shrimp gut, which included bivalves, gastropoda, various crustaceans, ophiuroids, nematodes and polychaeta. Costantini and Rossi (2001) in their laboratory study on grass shrimp feeding preferences have reported that, isopoda in the shrimp diet increased exponentially with the shrimp biomass. During present study also, high crustacean diet was evident and this can be attributed to either predation on harpacticoid copepod or on the vestiges of molted shrimp or the relatively weaker or dead

shrimps. Crustacians are known to be cannibalistic, and in their study Costantini and Rossi (2001) has sited that, shrimp depend heavily on the remains of conspecifics when profitable prey was not available. Allen *et al.* (1995) has sited that, in the lower stocking density shrimp ponds, natural food is capable of sustaining prawn growth for several weeks with little or no inputs of the supplementary feed. In their study he has further showed that, after four weeks shrimp showed decreased growth rates has been attributed to the less abundance of natural food organisms.

Mohanty *et al.* (2001) has shown that, scientific shrimp farming depends largely upon the commercial formulated feed, which constitutes nearly (55%) of the total operational costs. These supplemental feeds are provided especially in the commercial shrimp farming due to the insufficient availability of the natural food. Cordava *et al.* (2002) has shown to have promoted biota by application of organic fertilizers, zooplankton promoters, farming *Litopenaeus stylirostris*, and have sited that, natural food organisms contribute about (75%) of the requirement of the farmed organisms. During present study, though shrimp has shown reliance on the artificial feed and decrease of uptake of fauna towards the end of the culture period. This can not be taken as a reliance on the artificial diet completely due to observed presence of natural biota in the shrimp gut simultaneously. In the present study the major portion of natural shrimp diet consisted of the chlorophyll *a* correlated in terms of benthic diatoms. At night these diatoms could not sustain the biomass they offer during the day time. It was again observed that, in the pond invertebrate benthic biota especially meiofauna was less dense and diverse compared to creek. During present study the application of artificial fertilizers in the pond to promote natural biota was not done. Hence, farming practices such as drying and liming of pond soil during inter-crop period also might have affected the faunal composition and has been discussed earlier in Chapter 4, Section 4.2.1.4.4.

Plate 2 Photographs showing gut biota of shrimp

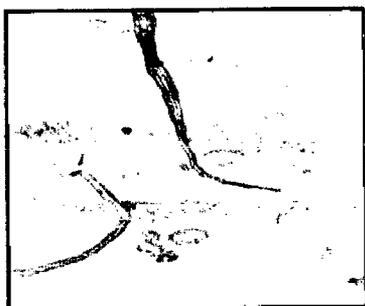
Crustacian body parts



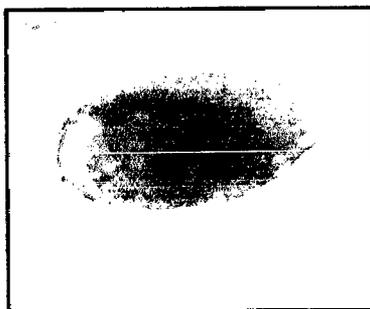
Fish scale



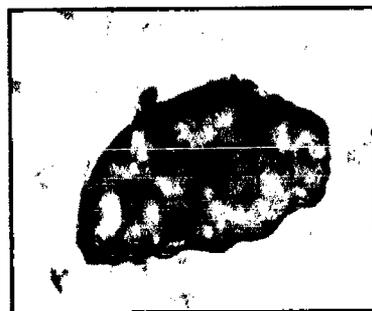
Algal strands



Egg



Sand particle



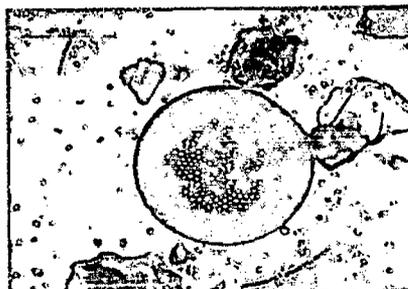
Polychaeta body parts



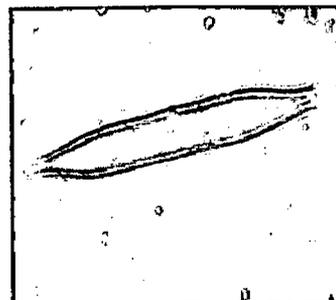
Pleurosigma



Coscinodiscus



Nitzschia



SECTION 4.4

SHRIMP GROWTH AND FEEDING

Chapter 4

4.4 Shrimp Growth and Feeding

4.4.1 Introduction

Achieving growth in stipulated time limits is the most important aspect in shrimp farming. From the traditional to intensive shrimp farming practices, task turn out to be most important, as profitability becomes vital. Hence, attaining best feed conversion ratio (FCR) and feed conversion efficiency (FCE) is considered most important in aquaculture practices. This, in turn is important as of today in the point of view of environment also. According to Biao *et al.* (2004) aquaculture effluents mainly consist of suspended organic matter, carbon, nitrogen and phosphorus, and most of it has origin from the accumulated feed in the pond. The high nutrient laden effluent water has implications for the receiving water which is nearby creek. The nutrient rich effluent from shrimp culture practices may turn the receiving water eutrophic and thereby can alter the ecology of these habitats by decreasing or totally eliminating natural residing biota and important inhabitant fishery resources (Elliot & Jonge, 2002). These situations further not only lead to socio-economic conflicts but also turn as self-destructive. Hence, management of feeding practices through careful monitoring, for attaining low FCR and high FCE, not only assures the better profitability, but also are important scales to map, and keep tab on the nutrient loading of the effluents from aquaculture practices. During present study, therefore, growth related parameters such as absolute growth rate; feed conversion ratio and feed conversion efficiency were observed over two experimental cycles EC-1 & EC-2, for monitoring pond environment by better feed management as well as its effectiveness in shrimp nutrition, along with the natural diet of the shrimp. The formulae for these parameters have been described in the (Chapter 3 Section 3.8). The stocking densities, average growth over fortnight period and absolute growth rate along with percent survival and FCR as well as FCE are depicted in Table P5-6 for EC-1 and EC-2, respectively.

4.4.2 Results

4.4.2.1 Experimental cycle (EC-1)

During EC-1 fortnightly average shrimp growth for aerated and non-aerated pond has been depicted in the Table P1 and Figure P1. Further, fortnightly average feed consumption for aerated and non-aerated pond has been depicted in the Table P3 and Figure P3.

4.4.2.1 A Regression analysis Aerated Pond

The absolute growth rate in terms of weight of the cultured shrimps from aerated pond was found to be 0.23 g.day^{-1} . In aerated pond FCR was found to be 1.30 and FCE of (77.11%) for EC-1 and are depicted in Table P5. On the basis of these observations, to check the linearity between shrimp growth, natural diet and supplementary feed, backward regression analysis was carried out which explained 91.3% variance in shrimp weight due to artificial feed ($B=3.62; p=0.001$) and chlorophyll a ($B=0.75; p=0.041$) in the sediment at ($F_{2,6}=42.8, p=0.001$). This indicates that, artificial feed had more significant effect on the growth of the shrimp in aerated pond.

4.4.2.1 B Regression analysis Non-aerated Pond

For non aerated pond stepwise multiple regression analysis explained the 90.7% changes in shrimp weight due to artificial feed ($B=6.79; p=0.001$) and chlorophyll a ($B=1.17; p=0.022$) in the sediment, with overall significant relationship at ($F_{2,6}=40.20; p=0.001$) thus was found to be similar to aerated pond. In non-aerated pond beta values suggested that, artificial feed played more significant role in the shrimp growth. The significant contribution of microbenthos in shrimp diet was evident from the presence of higher chlorophyll a in the sediment and the higher diatom counts in shrimp diet.

4.4.2.2 Experimental cycle (EC-2)

Fortnightly average shrimp growth for aerated and non-aerated pond has been depicted in the Table P2 and Figure P2. Fortnightly average feed consumption for aerated and non-aerated pond has been depicted in the Table P4 and Figure P4.

4.4.2.2 A Regression analysis Aerated Pond

In aerated pond stepwise multiple regression analysis explained 96.1% shrimp growth due to artificial feed and chlorophyll *a* in the sediment with overall significant relationship at $F_{2,5}=86.16;p=0.000$. Artificial feed ($B=4.49;p=0.001$) and chlorophyll *a* in the sediment in terms of microbenthic diatoms ($B=0.59;p=0.038$) played less significant role compared to artificial feed. This means, feed utilization was better in aerated pond and gut fauna only was taken up by opportunity or passively.

4.4.2.2 B Regression analysis Non-aerated Pond

In non aerated pond, gut biota was not significantly related with either of the parameters. Therefore, stepwise multiple regressions were carried out to check how significantly artificial feed contributed to the shrimp growth. The model explained 78.2% of the growth due to artificial feed ($F_{1,6}=26.15;p=0.002$). This particular trend on comparison with the trends for growth and contribution of artificial feed in aerated pond and non-aerated pond during EC-1 and non-aerated pond EC-2 indicates that approximately about 22% growth was due to the natural food biota available in the pond although feed was less utilized. The absolute growth rate of the shrimp during EC-2 was found to be almost similar to that of EC-1. For aerated pond, it was 0.23 g.day^{-1} and for non-aerated ponds it was 0.24 g.day^{-1} .

Table P1 Fortnightly average growth (gms) of shrimp during EC-1

	0	15	30	45	60	75	90	105	120
Aerated Pond	0.05	1.23	2.98	4.72	6.46	11.78	15.81	20.37	27.10
	±	±	±	±	±	±	±	±	±
	0.03	0.70	0.58	1.43	3.03	2.90	3.78	1.90	4.91
Non-aerated Pond	0.05	2.22	4.33	8.30	12.28	9.99	17.06	21.92	25.47
	±	±	±	±	±	±	±	±	±
	0.02	1.20	0.69	1.69	3.22	1.83	3.96	3.85	3.18

Table P2 Fortnightly average growth (gms) of shrimp during EC-2

	0	15	30	45	60	75	90	105
Aerated Pond	0.024	5.31	8.93	11.43	13.94	17.37	21.83	23.81
	±	±	±	±	±	±	±	±
	0.02	2.10	3.27	2.71	3.75	2.98	6.19	3.88
Non-aerated Pond	0.024	5.13	7.86	12.50	16.77	18.16	23.32	25.52
	±	±	±	±	±	±	±	±
	0.02	1.60	0.25	2.66	3.19	2.69	3.79	3.50

Figure P1 Fortnightly average shrimp growth (gm) during EC-1

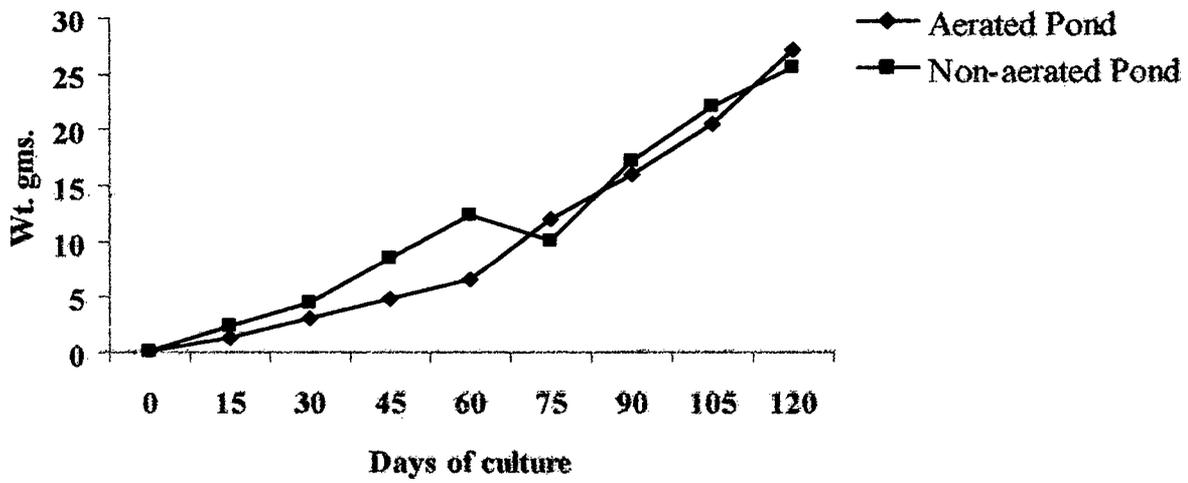


Figure P2- Fortnightly average shrimp growth (gm) during EC-2

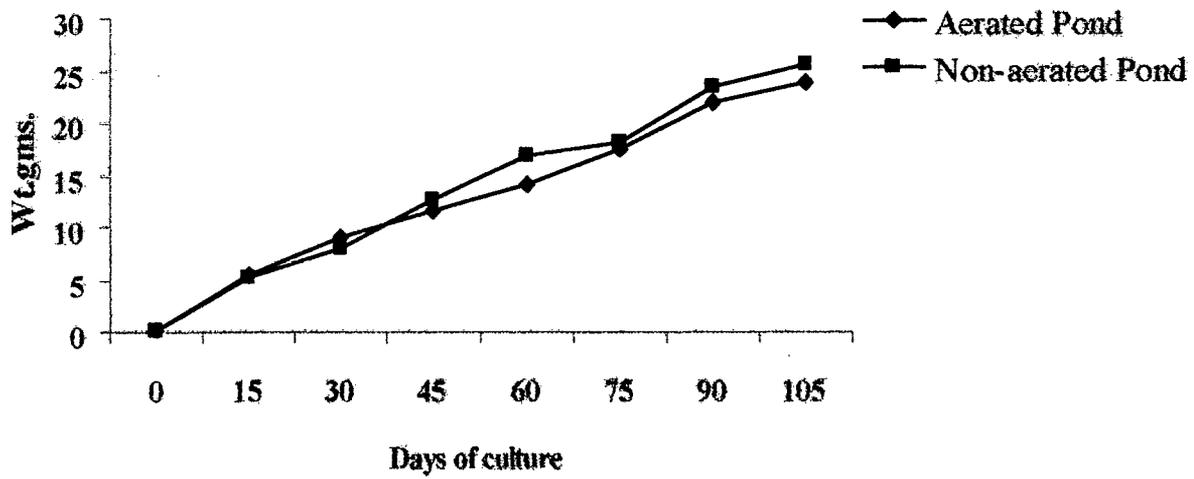


Table P3 Fortnightly average feed consumption (Kg) by shrimp during EC-1

	0	15	30	45	60	75	90	105	120
Aerated Pond	0.08 ± 0.15	1.03 ± 0.13	1.38 ± 0.21	2.30 ± 0.27	2.83 ± 0.24	4.33 ± 0.28	5.73 ± 0.66	4.45 ± 0.77	6.03 ± 0.46
Non-aerated Pond	0.09 ± 0.10	0.68 ± 0.13	0.60 ± 0.08	1.23 ± 0.21	2.15 ± 0.31	2.05 ± 0.13	3.03 ± 0.68	2.70 ± 0.45	2.55 ± 0.42

Table P4 Fortnightly average feed consumption (Kg) by shrimp during EC-2

	0	15	30	45	60	75	90	105
Aerated Pond	0.00 ± 0.00	0.45 ± 0.53	0.95 ± 0.64	1.43 ± 0.17	2.13 ± 0.17	3.70 ± 0.35	4.05 ± 0.48	3.83 ± 0.13
Non-aerated Pond	0.00 ± 0.00	0.30 ± 0.36	0.43 ± 0.29	0.98 ± 0.19	0.78 ± 0.13	2.13 ± 0.43	1.95 ± 0.21	1.80 ± 0.45



Figure P3 Fortnightly average feed consumption (Kg) during EC-1 from aerated and non-aerated pond

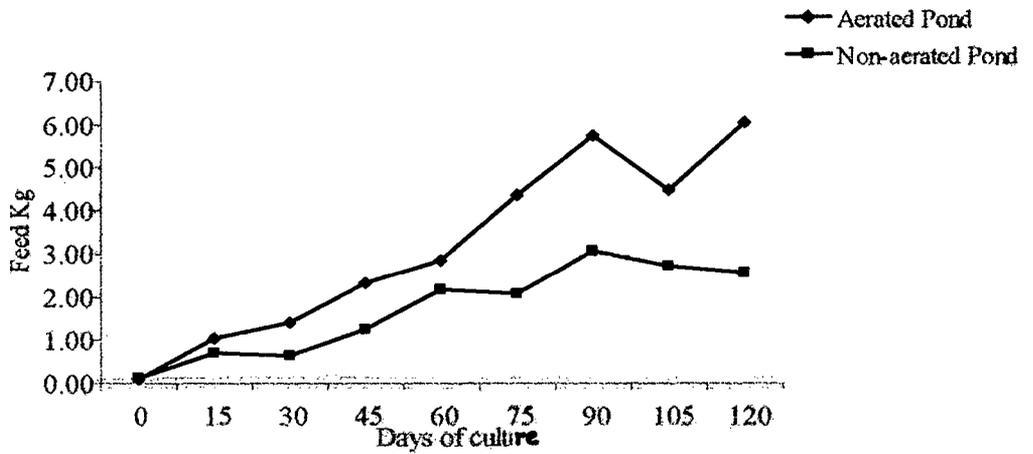
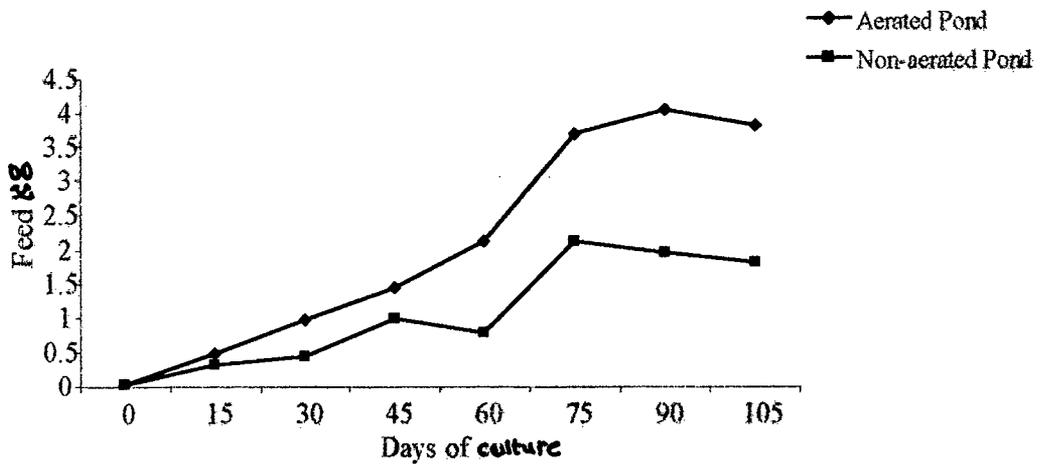


Figure P4 Fortnightly average feed consumption (Kg) during EC-2 from aerated and non-aerated pond



4.4.3 Shrimp Growth and Feeding Discussion

From the results of present study it is understood that, the natural fauna uptake was decreased with progressing culture period (Figure G1.5 & Figure G2.5), while artificial feed showed increasing trend is depicted in Figure P3-4 for EC-1 and EC-2 respectively. There was significant negative co-relationship ($p < 0.05$), between the natural and artificial feed uptake by the shrimp, during both the culture periods depicted in table ST1-2 and table ST4-5, respectively for EC-1 and EC-2. During EC-1, significant correlation ($p \leq 0.05$) between feed consumption and available total gut biota was observed ($R^2 = 0.64$) and ($R^2 = 0.61$) for aerated pond (Figure P5) and non aerated ponds (Figure P6) respectively. During EC-2 also, significant but comparatively weak correlation between artificial feed consumption and total gut fauna was observed ($R^2 = 0.23$) and ($R^2 = 0.30$) for aerated pond (Figure P7) and non aerated pond (Figure P8). From this correlation analysis it can be concluded that, during EC-1 feed utilization was higher than the natural biota compared to the EC-2. Earlier in (Chapter 4, Section 4.3.2.3) it has been discussed that, the total gut fauna from aerated pond was slightly higher than the non- aerated pond (Figure G1.5 and Figure G2.5).

During EC-1, for aerated pond overall gut biota averaged 239 organisms.individual⁻¹ for aerated pond and 128 organisms.individual⁻¹ non-aerated respectively. For EC-2 the overall average was almost double being 469 organisms.individual⁻¹ for aerated pond and 404 organisms.individual⁻¹ for non-aerated pond. Thus, from correlation analysis, total gut biota (Figure G1.5 and Figure G2.5) and average gut biota values, it can be said that, during EC-1 due to less availability of natural biota in pond, was responsible for higher uptake of the feed, by the shrimp. During EC-2, however, natural biota was comparatively double and feed uptake by shrimp thus was lower. This is evident again from the FCR values being higher during EC-1 being 1.30 and 1.56 (Table P5). During both the culture periods, though the decrease in the amount of natural gut biota was gradual, it was characteristic in a view that, its presence in the gut decreased drastically from 75

DoC onwards, till the end of the culture period (Figure G1.5 & Figure G2.5). The correlation of shrimp growth and contribution of natural biota, in shrimp gut showed significant ($p \leq 0.05$) correlation-ship. These relationships are depicted in Figure P9 and Figure P10 during EC-1 and in Figure P11 and Figure P12 during EC-2 for aerated and non-aerated ponds respectively. According to the established possibilities as well as numerous studies on juvenile shrimp nutritional analysis indicate that, the larval stages of shrimp like naupli exclusively feed on the algal diet like *Chaetoceros* and *Skeletonema*, and is a well known practices at the shrimp hatcheries.

Their role in shrimp nutrition has been studied by Nunez *et al.* (2002), Kumlu (1998); Su *et al.* (1997). However, in the grow-out ponds, once the post larvae are released in the water, the general practice is that, shrimp are not fed on for first few days or are fed only twice instead of four times a day. This practice is prevalent in view that, the post larvae utilize the available phytoplankton in the water column. The adult prawn previously has been revived and discussed in (Chapter 4, Section 4.3.23.) to feed on variety of food in the natural as well as in the artificial environment. Unlike natural environment, however, the availability of the natural fauna in the artificially constructed pond is certainly low. During present study also, all components of benthos which consisted of macro, meio and microbenthos are also known to provide good source of food to adult shrimps in the natural environment, and are reviewed in (Chapter 2 Section 2.2).

In present study, the average macro, meio and microbenthos (Figure MF1.4, Figure MF1.8, Figure MF1.12, and Figure MF2.4, Figure MF2.8, and Figure, MF2.12) showed higher percent abundance in the adjacent creek than the experimental ponds. It can be seen from the average gut biota for aerated and non aerated pond that, the decrease for aerated and non aerated pond during EC-1 was from 75 DoC and gut biota though decreased thereafter, the counts were marginally higher in aerated pond (Figure G1.5). However, counts for aerated and non-aerated

pond during EC-2 decreased from 60 DoC and non-aerated pond showed slightly higher gut counts than the aerated pond except on 105 DoC (Figure MF2.5). Further from the growth and feeding data the growth rate determination revealed interesting and comparable results with respect to contribution of natural feed. It was observed that, the growth rates from 75 to 120 DoC were higher being 0.34 g.day^{-1} , over 0.18 g.day^{-1} for non-aerated pond. The backward regression analysis during EC-1 showed the dependence of the shrimp growth, on the chlorophyll *a* and the macrofauna. The macrofauna possibly was taken up less by the shrimp as macrofauna showed an increasing trend during EC-1 (Figure MF1.4).

Further, the macrofauna was comprised of only Gastropoda (Figure MF1.1) during EC-1 in aerated and non aerated pond, which is difficult for shrimp to consume. Another important factor, chlorophyll *a* showed significant linearity with that of gut biota. The chlorophyll *a* can be attributed to have originated from the microbenthos which included mostly the pinnate diatoms like *Navicula*, *Nitzschia*, *Pleurosigma* and centric diatoms such as *Coscinodiscus* have been depicted in (Figure MF1.9). During EC-1 in non-aerated pond the microbenthos showed decreasing trend (Figure MF1.12) while in aerated pond it showed increasing trend (Figure MF1.12). Thus, it can be stated that, better growth rates in aerated pond were achieved due to availability of higher biomass of diatoms along with artificial supplementary diet. The FCR remained high and FCE low during EC-1. The FCR was found to be 1.30 and 1.56, while FCE was 77.11% and 63.92% for aerated and non-aerated pond respectively. This further confirms the reliance on the natural fauna in view that, feed was not utilized effectively by shrimp initially, especially in the non-aerated pond thus, shrimp showed higher average weights during EC-1 (Figure P1) than the EC-2 (Figure P2).

During EC-2, the FCR of 1.04 and 0.86 and FCE of 100 % and 86% for aerated pond and non-aerated pond indicate very good utilization of artificial feed. However, there was slight underachievement of shrimp weight (Figure P2).

Specifically, in non-aerated pond the average faunal densities over 105 days were slightly lower than the aerated pond. Further, it was observed that, the growth rate of shrimp differed for non-aerated pond, being 0.28 g.day^{-1} till 60 days and bettered over, the next 45 day growth rate, being 0.16 g.day^{-1} . In aerated pond however, the difference in growth rates was comparably negligible being 0.23 and 0.21 g.day^{-1} for first 60 and last 45 days respectively for aerated pond. Another conspicuous feature was that, average benthic densities over 105 days were higher during EC-2 in ponds as well as in the gut of the shrimp than EC-1 over 120 days. The lack of any linear relationship between benthos and shrimp gut in non aerated pond during EC-2 and significant relationship with all benthic components macro, meio and microfauna for aerated ponds indicates that, fauna was better utilized in aerated pond than non-aerated pond. Overall the FCR and FCE were better in aerated pond than the non-aerated pond (Table P5 and P6). The underachievement in weight however, during EC-2 can be attributed to the lack of better utilization of available benthic fauna or the changes in the quality of artificial feed seems to be the possible reason especially during EC-2 for both the ponds.

Mohanty *et al.* (2001) showed that, scientific shrimp farming depends largely upon the commercial formulated feed, which constitutes nearly 55% of the total operational costs. These supplemental feeds are provided especially in the commercial shrimp farming due to the insufficient availability of the natural food. Cordava *et al.* (2002) has shown to have promoted biota by application of organic fertilizers and further zooplankton, farming *Litopenaeus stylirostris*, and have sited that, natural food organisms contribute about 75% of the requirement of the farmed organisms. During present study though, shrimp has shown reliance on the artificial feed (Figure P3 and Figure P4) for EC-1 and EC-2 respectively and decrease of uptake of fauna towards the end of the culture period (Figure G1.5 and Figure G2.5), and (Figure P9 to Figure P12), as well as from FCR (Table P5 & P6) it indicates that, natural feed by shrimp was consumed amounted approximately, 20-25% of total food requirements of the cultured shrimp. This can be supported by the fact that, the

diversity and abundance of natural benthic biota especially meiofauna was limited in the culture ponds (Figure MF1.8 and Figure MF 2.8). Further, Gastropoda shell remains showed less percent abundance overall in the gut content and higher densities in the pond and have been discussed earlier in (Chapter4, Section4.3.2.3). Microbenthos, however, was found to be the most significantly consumed component of benthos and was also abundant in the shrimp pond as well (Chapter 4, Section 4.2.1.4.3). Hence, the result obtained here shows the dependence of shrimp on the natural fauna during initial culture days. Further, along with the abundance, limitations of natural biota, in the pond water, and its quantification in terms of the caloric value it provides to shrimp, needs further detail evaluation.

Figure P9 Correlation of shrimp growth vs Gut biota from aerated pond EC-1

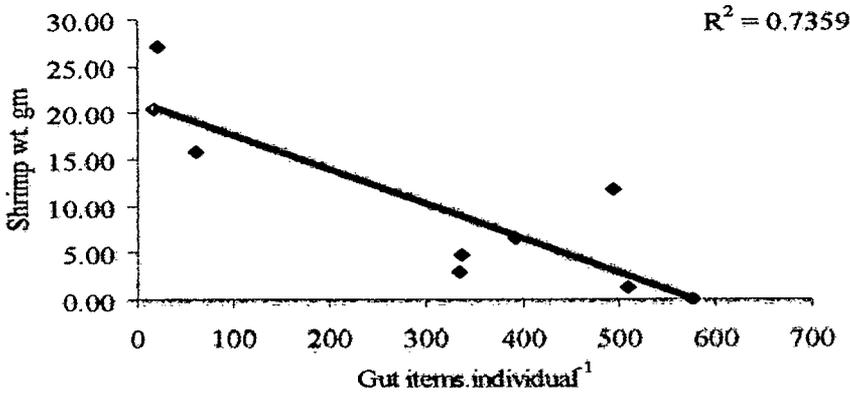


Figure P10 Correlation of shrimp growth vs Gut biota from non-aerated pond EC-1

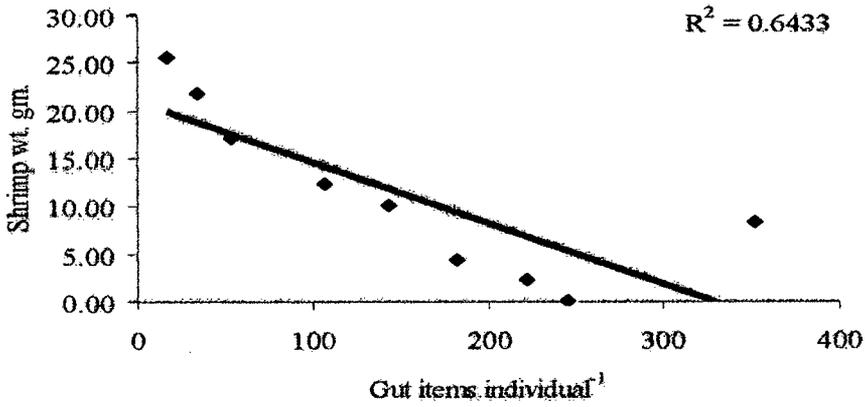


Figure P11 Correlation of shrimp growth vs Gut biota from aerated pond EC-2

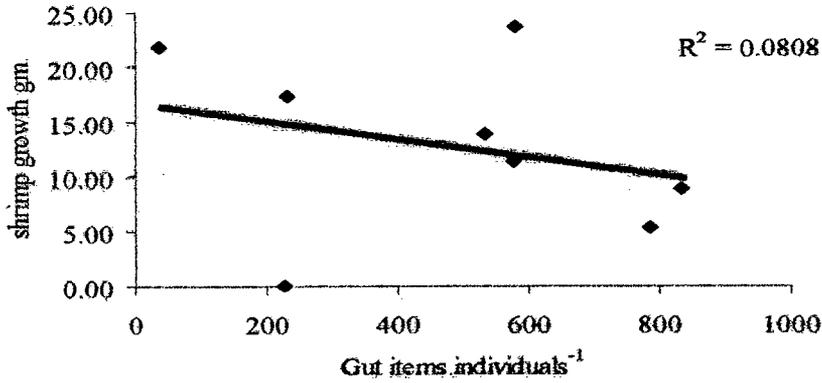


Figure P12 Correlation of shrimp growth vs Gut biota from non-aerated pond EC-2

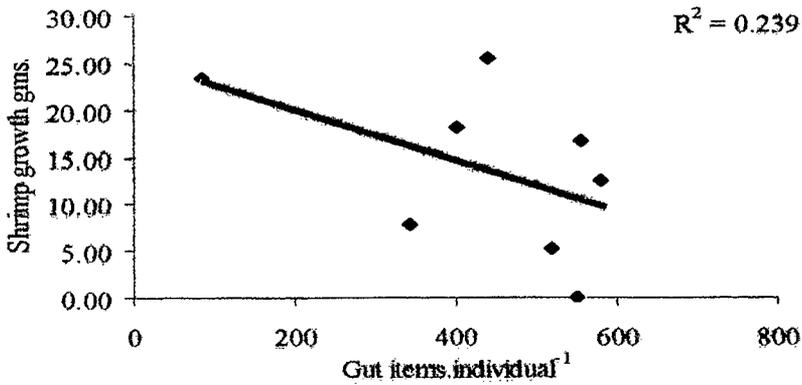


Figure P5 Correlation feed consumption vs Gut fauna aerated pond EC-1

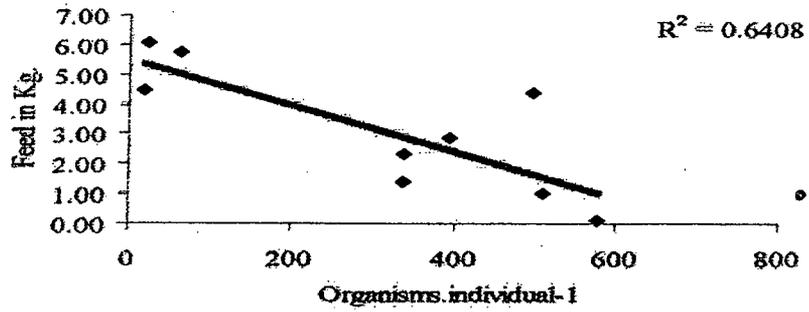


Figure P6 Correlation feed consumption vs Gut fauna non-aerated pond EC-1

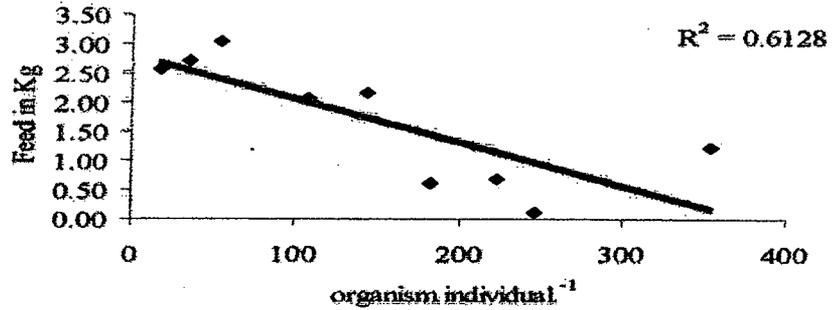


Figure P7 Correlation feed consumption vs Gut fauna aerated pond EC-2

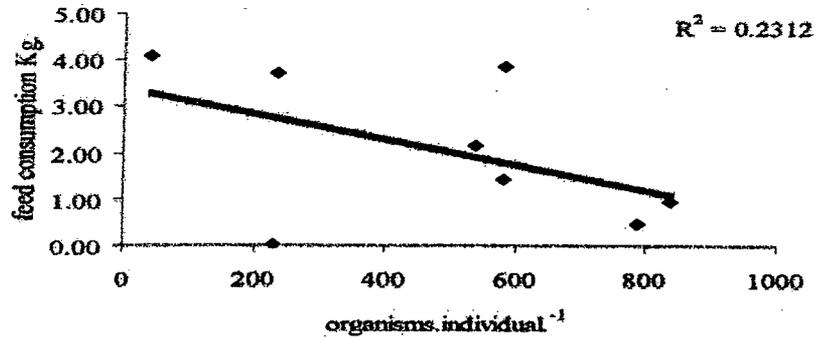


Figure P8 Correlation feed consumption vs Gut fauna non-aerated pond EC-2

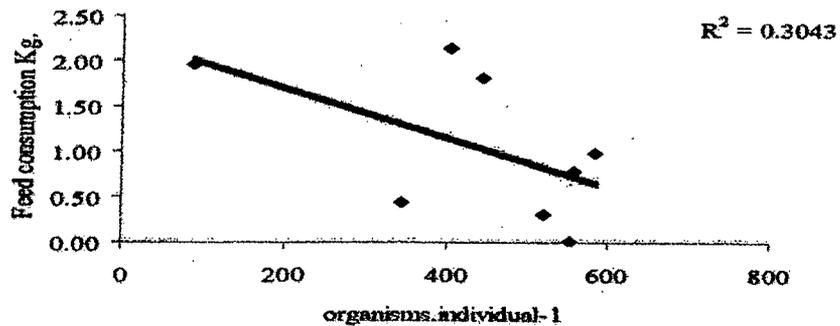


Table P5 Table showing Stocking density, production and FCR during EC-1

Pond Type	Area (ha.)	PL. Stocked No.m ⁻²	Production (Kg.)	% Survival	Mean Length (cm.)	Mean weight (gm)	Total feed used (Kg.)	FCR	FCE
Aerated Pond	0.65	12	1607	54.63	15.74 ± 1.19	29.27 ± 7.16	2084.01	1.30	77.11
Non-aerated Pond	0.6	6	715	63.26	16.30 ± 1.04	33.78 ± 6.96	1118.67	1.56	63.92

Table P6 Table showing Stocking density, production and FCR during EC-2

Pond Type	Area (ha.)	PL. Stocked No.m ⁻²	Production (Kg.)	% Survival	Mean Length (cm.)	Mean weight (gm)	Total feed used (Kg.)	FCR	FCE
Aerated Pond	0.65	10	1121	74.24	15.74 ± 1.19	29.27 ± 7.15	1070.8	1.04	105
Non-aerated Pond	0.6	5	536	68.35	14.78 ± 1.78	25.52 ± 6.33	621.56	0.86	86

CHAPTER 5
CONCLUSIONS

CHAPTER 5

CONCLUSIONS

5.1 Water quality

Physico-chemical parameters governing the individual water quality parameters and thus actual process were found to be different in different treatments. The proven interdependence of the water quality parameters during present and earlier studies, point towards the rather naturally controlled processes in experimental ponds. Water-quality parameters except chlorophyll *a* concentrations did not show statistically significant differences in between to experimental cycles. The non significant differences of DO and other water quality parameters over to production cycles (EC-1 and EC-2) can be attributed to the identical sampling period ranging from summer to monsoonal months form May to August.

There were significant differences in the Chlorophyll *a* concentrations during two experimental cycles studied over two years. With no significant differences in nutrient concentrations the higher chlorophyll *a* during EC-2 than EC-1 as discussed earlier can be attributed to higher light intensities and longer breaks in the monsoon or less dense cloud cover during the study period.

The objective of present study was to see the effect of aeration on the DO concentrations. The insignificant result for the difference in aerated and non-aerated pond here does not mean that aeration had no effect on the water quality. The sampling time and frequency seems to provide inadequate information about DO regime, where DO is known to vary diurnally. The higher and lower DO concentrations during present study thus can be attributed to the sampling time where sampling was done from morning 6 to 9 Hrs. The lower DO further can be due to the carry forward effects from night due to respiration by phytoplankton in addition to the cultured species, and higher DO can be attributed to aeration and primary productivity during day time ultimately increasing the DO concentrations.

Though, lower DO concentrations were observed DO value never decreased below 3 mg.l^{-1} as a general observation and was indicative that aeration systems in aerated pond helped maintaining the DO level during the night time to sustain the dissolved oxygen to its optimal. During day time, the circulated water due to currents produced by circulation kept the bottom water clean and further provided better oxidative environments which brought about effective degradation of organic matter present at the bottom of the pond. The lack of significant differences in DO in aerated and non-aerated pond can be attributed to the diurnal fluctuations in the DO, as well as lower density of shrimp in non-aeration pond. Further, diurnal fluctuations in DO and primary productivity also played role in controlling the micro-scale fluctuations in nutrient and interlinked ORP and pH changes and followed standard fluctuations.

Thus, full growth cycles of 120 days and direct observations of shrimp showing no distress is an indication that water quality was at its optimal best. Water quality despite higher stocking densities in aeration pond remained at optimal best, shows that there was no critical overload on the environment of harmful toxicants with stocking densities of 10 and 12 Pl.m^{-2} . Thus, the aerated pond environment resembled of more natural water with respect to non aeration pond having lower stocking densities of 5 to 6 Pl.m^{-2} . Hence with stocking densities of 10 to 12 PL.m^{-2} and aeration gives farmer an assurance of healthy environment as well as an opportunity to make use of its resources at the best to increase the production per unit area.

5.2 Benthos

For benthos (macro-, meio-and microbenthos) overall, it was found that, macro- and meiobenthos did not differ significantly from aerated and non-aerated pond during inter and intra experiment. Only microbenthos differed significantly ($p < 0.05$) for EC-1 and EC-2, however again there were no significant differences observed in aerated and non aerated ponds in intra experiment. Further, it was observed that, DO

has no significant effect ($p > 0.05$) on the development of bottom fauna. Further, it was found that, their abundance and diversity though was controlled by the availability of food, it was undoubtedly influenced by the practices such as drying of the pond in between two crops, liming at the start of the culture period, and water intake practices and exchanges leading to flushing of the fauna, which eliminates any kind of fauna and flora available previously at the bottom of the pond during such practices.

5.2 A Macrofauna

It was found that, the average macrofauna densities.m⁻² were higher during EC-2 than EC-1. During EC-1, interestingly only gastropod population were observed in the pond as well as in the creek. Overall, it was observed that, in experimental ponds, macrofauna density showed an increasing trend, while in creek, it showed slightly decreasing densities with the progressing culture period during EC-1. Phytoplankton and diatom was prominent food uptake by gastropods during EC-1. During EC-2 unlike EC-1, Polychaeta dominated over gastropod both in culture ponds as well as in the creek. The reason why it was higher in the non-aerated pond seems to be the presence of slightly higher organic carbon available as food for Polychaeta. The higher chlorophyll-*a* in water, probably was not settled in the aerated pond and may have lead to less availability of food for gastropod and thus they were less in number compared to EC-1. Hence, food availability and preference was evident as gastropods fed on the phytoplankton and polychaetes fed on the organic carbon available in the pond. The organic carbon values further were found to be in the optimal limits. Hence, it can be speculated that, in aeration pond, the aeration helped in the effective oxidation of organic matter while in the non-aeration pond, it was macrobenthic polychaeta would have helped in reworking of accumulated organic matter. Macrofauna from the creek was more stable and clearly was observed to be influenced by tidal regime. The higher densities of macrofauna in culture ponds are indicative of the fact that, they privileged the favorable conditions in view that, they

remained submerged during the culture period in the experimental ponds, unlike creek, which is influenced by the tidal regime.

5.2 B Meiofauna

Dominant meiofaunal groups observed consisted of Nematoda, Turbellaria, Copepoda and others with comparatively small proportion consisted of eggs, naupli, insect larvae, Nemertina and Polychaeta. Meiofauna density was found to be very low throughout the culture period during EC-1 than EC-2. From ANOVA (Table.AB1) DO did not show any significant effect on the development of bottom fauna. This indicates that, DO probably was not limiting factor for meiofauna in aerated pond during EC-1, and higher density was maintained due to the food availability in terms of higher chlorophyll-*a* (phytoplankton) in the water column settled on the bottom of the pond. In non-aerated pond however it was organic carbon that was found to have taken up by the meiobenthic organisms. Overall, it can be said that, during EC-1, the meiofauna was limited by food availability and during EC-2, it was limited by the physical factors such as pH and E_h . Thus, indicates that even in unfavorable physical conditions, if food availability is ensured, meiobenthic organisms can resist and maintain their population structure. Further, the reason why they were found to be less in the culture ponds than in the creek lies in fact that, they were recruited from the adjacent coastal waters and probably mangrove patches provided better settlement ground than culture ponds, where sediment is always more compacted compared to the soft sediment in the creek, which is the perfect habitat for the soft bodied meiofaunal organisms.

5.2 C Microbenthos

The significant relationships between chlorophyll-*a* in water and sediment and microbenthos as well as simultaneous dominance of diatoms in the microbenthic population asserts that, chlorophyll-*a* in the sediment and water can be attributed to have originated from diatomaceous population in the experimental ponds. During present study, diatoms were the most abundant group of phytoplankton found in the

water. This was dominated by *Coscinodiscus*, *Navicula*, *Nitzschia* and *Pleurosigma* sp. These are commonly known as pinnate diatoms, and are most common type of phytoplankton observed in varying temperature, salinity and withstand wide range of light intensities. Conspicuously it was observed that, during EC-1 respiration and hence DO availability was responsible for their limitation. During EC-2, the density of microbenthos was higher. During the study period, monsoon was progressive, further nutrients were found to be available at their optimal along with other physico-chemical parameters of water and soil, and thus they were not limiting. Hence, the remaining factor of light, that is increasing and decreasing light intensities due to variable cloud cover influenced the densities of the microphytobenthos. Hence during EC-1, the lower light intensities led to lower DO concentrations during morning which were further sustained due to low respiration as light intensity was low during day and respiration for cell growth during night time. Thus acting in conjunction might have limited the growth of the diatoms at the bottom of the pond. Contrasting to this abrupt photoperiods and higher light intensities were responsible for higher densities of the microphytobenthos.

5.3 Gut content analysis and shrimp growth

From direct gut content observations it implies that, phytoplankton and diatoms in the sediment formed an important part in the diet of the shrimp along with artificial feed. Macro, meio as well as microbenthos showed significant ($p < 0.05$) relationship with gut fauna. There were, clear food preferences existent for day and night time. In present study the pinnate diatoms *Nitzschia* and *Navicula* dominated in the gut of the shrimp, followed by crustacean body parts. During morning hours diatom dominated, while during night feeding crustacean diet was preferred by shrimp. Better correlation of microbenthos with that of gut fauna in aerated pond than non-aerated pond indicates that diatoms in aerated pond were consumed more effectively than non-aerated pond during EC-1 as well as EC-2. Further, shrimp has shown reliance on the artificial feed towards the end of culture period from 45th to 60th DOC onwards and decrease of uptake of fauna towards the end of the culture period.

Further, the growth rates of shrimp attained were higher during the first 45, 60 DOC and were correlated to chlorophyll-*a*. Better growth rates in aerated pond were achieved due to availability of higher diatom biomass along with artificial supplementary diet. During both the studies EC-1 and EC-2, a general trend was that last 45 days the growth was reduced and FCR remained in 0.90 to 1.3. The FCR achieved during present studies thus are indicative of good feed management thereby assuring the better conversion by the shrimp. Physico-chemical parameters were at their optimal best, benthic fauna though comparably less than natural environment, its presence in the shrimp gut confirms its utilization. Further, it can be speculated, to have provided less density per unit area per individual. Therefore, ultimately, the decreased feed quality due to disintegration and leaching in water seems to be the possible reason for the under nourishment and hence underachievement in growth especially during EC-2 than EC-1 during present practices.

STATISTICAL TABLES

Table ST1 Correlation matrix of the various parameters for aerated pond EC-1

	Temperature	Salinity	pH Water	pH sediment	Hywater	Hy sediment	DO	BOD	COD	NO ₃ N	NO ₂ N	PO ₄ N	NH ₃ N	Chl a water	Chl a sediment	Phaeo water	Phaeo sed	OC	POC	HIS water	Den Macrofauna	Den Meiofauna	Den Microfauna	Cat fauna	Shrimp wt	Feed	
Temperature	1.00																										
Salinity	0.82	1.00																									
pH Water	-0.30	-0.01	1.00																								
pH sediment	-0.18	-0.40	-0.03	1.00																							
Hywater	-0.78	-0.50	0.25	-0.21	1.00																						
Hy sediment	-0.75	-0.53	0.31	-0.20	0.98	1.00																					
DO	0.55	0.26	-0.19	-0.07	-0.76	-0.68	1.00																				
BOD	0.34	0.50	-0.13	-0.47	-0.27	-0.41	-0.03	1.00																			
COD	0.39	0.43	-0.55	-0.26	-0.27	-0.36	0.20	0.31	1.00																		
NO ₃ N	0.09	0.34	-0.15	-0.32	0.19	0.14	0.09	0.53	0.34	1.00																	
NO ₂ N	0.58	0.68	-0.28	-0.64	-0.41	-0.50	0.27	0.90	0.41	0.70	1.00																
PO ₄ N	0.82	0.86	-0.08	-0.49	-0.44	-0.46	0.13	0.68	0.28	0.34	0.77	1.00															
NH ₃ N	0.60	0.54	0.18	-0.45	-0.30	-0.28	0.18	0.60	-0.20	0.23	0.61	0.85	1.00														
Chl a water	-0.25	0.33	0.45	-0.26	0.37	0.25	-0.53	0.41	0.07	0.35	0.25	0.15	0.01	1.00													
Chl a sediment	-0.13	0.45	0.48	-0.44	0.44	0.38	-0.46	0.21	0.06	0.45	0.19	0.22	0.06	0.91	1.00												
Phaeo water	-0.63	-0.28	0.63	-0.01	0.80	0.78	-0.86	-0.14	-0.42	-0.13	-0.42	-0.20	-0.02	0.55	0.55	1.00											
Phaeo sed	0.12	0.42	-0.04	-0.09	0.11	0.07	-0.07	-0.40	0.10	0.29	0.10	0.03	-0.25	0.50	0.63	-0.03	1.00										
OC	0.04	0.51	0.40	-0.12	0.01	-0.11	-0.35	0.57	0.00	0.23	0.42	0.34	0.23	0.91	0.76	0.31	0.47	1.00									
POC	0.17	-0.30	-0.47	0.51	-0.15	-0.05	0.21	-0.67	-0.18	-0.51	-0.49	-0.27	-0.24	-0.78	-0.63	-0.34	0.01	-0.71	1.00								
HIS water	-0.28	-0.36	-0.12	0.05	0.19	0.18	-0.23	-0.07	0.35	-0.23	-0.27	-0.14	-0.15	-0.26	-0.33	0.21	-0.73	-0.45	-0.04	1.00							
Den Macrofauna	-0.86	-0.61	0.15	0.26	0.84	0.78	-0.83	-0.32	-0.24	-0.16	-0.57	-0.64	-0.58	0.41	0.32	0.74	0.17	0.14	-0.05	0.17	1.00						
Den Meiofauna	-0.66	-0.32	0.35	0.14	0.29	0.16	-0.39	0.31	-0.06	0.04	-0.02	-0.39	-0.32	0.62	0.30	0.40	-0.06	0.55	-0.68	0.08	0.53	1.00					
Den Microfauna	-0.66	-0.50	0.57	-0.06	0.58	0.58	-0.56	0.02	-0.36	-0.12	-0.32	-0.24	0.08	0.22	0.11	0.77	-0.60	0.03	-0.43	0.58	0.48	0.50	1.00				
Cat fauna	0.59	0.68	-0.02	-0.48	-0.54	-0.50	0.76	0.19	0.47	0.50	0.52	0.40	0.15	0.04	0.19	-0.60	0.33	0.11	-0.22	-0.34	-0.72	-0.28	-0.57	1.00			
Shrimp wt	-0.83	-0.92	0.21	0.49	0.63	0.65	-0.51	-0.57	-0.54	-0.51	-0.82	-0.79	-0.47	-0.18	-0.26	0.56	-0.36	-0.36	0.28	0.38	0.75	0.31	0.63	-0.82	1.00		
Feed	-0.67	-0.89	-0.07	0.68	0.42	0.46	-0.32	-0.71	-0.42	-0.62	-0.86	-0.84	-0.63	-0.38	-0.45	0.27	-0.22	-0.51	0.58	0.28	0.64	0.12	0.28	-0.73	0.92	1.00	

Correlation matrix of the various parameters for non-aerated pond EC-1

	Temperature	Salinity	pH Water	pH sediment	E ₄ water	E ₄ sediment	DO	BOD	COD	NO ₂ -N	NO ₃ -N	PO ₄ -N	NH ₃ -N	Chl a water	Chl a sediment	Phaeo water	Phaeo sed	OC	POC	H ₂ S water	Den Macrofauna	Den Meiofauna	Den Microfauna	Gut fauna	Shrimp wt	Feed	
Temperature	1.00																										
Salinity	0.76	1.00																									
pH Water	-0.42	-0.07	1.00																								
pH sediment	-0.27	-0.56	-0.09	1.00																							
E ₄ water	-0.59	-0.26	0.46	-0.42	1.00																						
E ₄ sediment	-0.69	-0.61	0.47	-0.20	0.69	1.00																					
DO	-0.07	-0.20	-0.33	0.11	-0.22	-0.12	1.00																				
BOD	0.12	0.21	-0.56	0.03	-0.26	-0.59	0.69	1.00																			
COD	0.24	0.02	-0.13	-0.39	-0.33	-0.53	-0.05	0.04	1.00																		
NO ₂ -N	0.63	0.33	0.01	0.11	-0.63	-0.39	-0.25	-0.26	0.31	1.00																	
NO ₃ -N	-0.06	-0.34	0.20	0.06	0.13	0.41	-0.58	-0.72	-0.28	0.33	1.00																
PO ₄ -N	0.48	0.64	0.49	-0.35	0.07	-0.28	-0.24	-0.18	0.70	0.31	-0.03	1.00															
NH ₃ -N	-0.25	-0.40	0.60	0.01	0.35	0.64	-0.49	-0.86	-0.28	0.28	0.85	0.17	1.00														
Chl a water	-0.06	0.10	0.40	-0.21	0.52	0.23	-0.83	-0.59	0.08	-0.12	0.57	0.40	0.51	1.00													
Chl a sediment	-0.28	-0.42	0.39	0.15	0.20	0.55	-0.66	-0.85	-0.35	0.16	0.92	-0.09	0.85	0.62	1.00												
Phaeo water	-0.45	-0.10	0.90	-0.05	0.65	0.42	-0.38	-0.49	-0.07	-0.24	0.15	0.53	0.51	0.61	0.33	1.00											
Phaeo sed	-0.28	-0.02	0.14	-0.05	0.42	0.25	-0.76	-0.42	-0.21	-0.19	0.17	-0.19	0.13	0.56	0.35	0.28	1.00										
OC	-0.49	-0.38	0.51	0.54	0.03	0.11	-0.35	-0.27	-0.52	0.03	0.44	-0.02	0.41	0.36	0.58	0.45	0.12	1.00									
POC	0.34	0.15	-0.30	0.21	-0.03	-0.41	-0.01	0.14	0.40	0.06	-0.30	0.25	-0.25	0.05	-0.39	0.00	0.16	-0.41	1.00								
H ₂ S water	0.42	0.10	-0.24	0.32	-0.51	-0.47	0.17	0.15	0.37	0.16	0.17	0.28	-0.07	0.07	0.07	-0.17	-0.58	0.22	0.03	1.00							
Den Macrofauna	-0.45	-0.51	0.51	0.42	0.26	0.49	-0.68	-0.82	-0.44	0.11	0.70	-0.09	0.76	0.58	0.86	0.53	0.55	0.65	-0.08	-0.13	1.00						
Den Meiofauna	-0.01	0.35	0.27	0.26	-0.27	-0.49	-0.30	0.17	0.03	0.11	-0.13	0.24	-0.23	0.21	0.90	0.22	0.14	0.65	-0.22	0.24	0.10	1.00					
Den Microfauna	0.34	0.36	0.10	-0.79	0.36	0.30	-0.45	-0.44	0.34	0.10	0.51	0.36	0.43	0.56	0.38	0.10	0.12	-0.26	-0.23	-0.01	0.01	-0.30	1.00				
Gut fauna	0.74	0.80	-0.38	-0.51	-0.28	-0.61	0.18	0.47	0.79	0.03	-0.36	0.48	-0.54	-0.02	-0.51	-0.31	-0.38	-0.47	0.11	0.47	-0.75	0.14	0.37	1.00			
Shrimp wt	-0.68	-0.94	0.26	0.56	0.32	0.68	0.02	-0.39	-0.70	-0.20	0.51	-0.36	0.62	0.13	0.57	0.32	0.02	0.49	-0.06	0.02	0.67	-0.29	-0.24	-0.79	1.00		
Feed	-0.72	-0.96	0.01	0.71	0.19	0.43	0.22	-0.09	-0.75	-0.34	0.18	-0.60	0.25	-0.14	0.27	0.11	0.06	0.37	0.08	-0.07	0.48	-0.26	-0.56	-0.78	0.90	1.00	

Table ST 3 Correlation matrix of various parameters for creek EC-1

	Temp	Salinity	ph water	Ph sed	ch water	ch sed	DO	BOD	COD	No3	No2	po4	nh3	chl a water	chl a sed	phaeo water	phaeo sed	OC	POC	h2s water	DenMacro	DenMeio	Den micro	
Temp	1.00																							
Salinity	0.63	1.00																						
ph water	-0.09	0.57	1.00																					
Ph sed	-0.26	-0.52	-0.11	1.00																				
ch water	-0.40	-0.11	0.10	-0.36	1.00																			
ch sed	-0.54	-0.36	-0.02	-0.44	0.71	1.00																		
DO	-0.35	-0.67	-0.20	0.59	0.17	0.32	1.00																	
BOD	-0.23	-0.28	0.17	0.37	0.45	0.33	0.78	1.00																
COD	0.81	0.72	0.23	-0.10	-0.53	-0.78	-0.57	-0.47	1.00															
No3	0.80	0.51	-0.27	-0.31	-0.33	-0.60	-0.63	-0.60	0.81	1.00														
No2	0.58	0.30	0.16	-0.14	-0.11	-0.14	0.07	0.06	0.56	0.41	1.00													
po4	0.48	0.42	-0.17	-0.33	-0.61	-0.28	-0.45	-0.60	0.36	0.39	-0.12	1.00												
nh3	0.36	0.20	0.08	-0.32	0.01	0.19	0.18	0.07	0.26	0.22	0.85	0.01	1.00											
chl a water	-0.06	0.37	0.38	-0.22	0.31	0.13	0.00	0.20	0.02	0.01	0.14	-0.11	0.44	1.00										
chl a sed	-0.27	-0.10	0.20	-0.02	0.12	0.41	0.47	0.35	-0.29	-0.37	0.27	-0.12	0.67	0.71	1.00									
Phaeo water	-0.25	0.28	0.49	-0.31	0.45	0.24	-0.14	-0.01	0.04	0.00	0.27	-0.36	0.46	0.81	0.57	1.00								
phaeo sed	0.42	0.75	0.57	-0.59	0.09	0.10	-0.26	-0.08	0.40	0.18	0.53	0.36	0.68	0.55	0.47	0.49	1.00							
OC	-0.02	0.47	0.74	0.01	-0.21	-0.07	0.05	0.08	0.20	-0.25	0.20	0.22	0.36	0.54	0.59	0.45	0.72	1.00						
POC	0.27	0.18	0.02	0.62	-0.41	-0.06	-0.02	-0.02	0.52	0.39	0.06	-0.02	-0.16	0.19	-0.14	0.83	-0.18	0.14	1.00					
h2s water	0.58	0.48	-0.21	-0.14	-0.67	-0.67	-0.63	-0.76	0.64	0.69	-0.08	0.84	-0.16	-0.17	-0.42	-0.30	0.14	0.04	0.38	1.00				
DenMacro	0.15	0.46	0.36	-0.59	0.75	0.36	-0.24	0.30	-0.01	0.03	0.10	-0.29	0.02	0.23	-0.18	0.27	0.32	-0.10	-0.32	-0.31	1.00			
DenMeio	-0.03	-0.11	-0.58	-0.14	-0.06	-0.15	-0.43	-0.46	-0.06	0.41	-0.53	0.25	-0.39	0.05	-0.27	-0.10	-0.43	-0.55	0.15	0.40	-0.15	1.00		
Den micro	0.16	0.31	0.15	0.15	-0.41	-0.39	-0.01	-0.26	0.37	0.22	0.21	0.39	0.37	0.54	0.48	0.40	0.45	0.68	0.54	0.44	-0.45	-0.06	1.00	

Table ST4 Correlation matrix of various parameters for aerated pond EC-2

	Temp	Salinity	pH water	pH sed.	eh water	eh sed.	DO	BOD	COD	NO3-N	NO2-N	NH3-N	PO4-P	Chl a water	Chl a sed.	Phaeo watr	Phaeo sed.	POC	OC	H2s water	Macrofauna	Meiofauna	Microfauna	Gut fauna	Shrimp wt	Feed	
Temp	1.00																										
Salinity	0.87	1.00																									
pH water	-0.44	-0.53	1.00																								
pH sed.	-0.46	-0.31	0.80	1.00																							
eh water	-0.51	-0.47	-0.28	-0.41	1.00																						
eh sed.	-0.60	-0.56	0.26	0.01	0.76	1.00																					
DO	-0.53	-0.40	0.66	0.79	0.04	0.37	1.00																				
BOD	-0.41	-0.27	-0.15	-0.03	0.75	0.63	0.52	1.00																			
COD	0.14	0.50	-0.18	0.31	-0.43	-0.51	0.20	-0.10	1.00																		
NO3-N	-0.80	-0.65	0.58	0.71	0.27	0.48	0.91	0.55	0.15	1.00																	
NO2-N	-0.70	-0.47	0.48	0.73	0.12	0.28	0.90	0.49	0.40	0.96	1.00																
NH3-N	-0.38	-0.26	-0.10	0.09	0.58	0.57	0.43	0.86	-0.23	0.43	0.34	1.00															
PO4-P	-0.70	-0.47	0.48	0.73	0.12	0.28	0.90	0.49	0.40	0.96	1.00	0.34	1.00														
Chl a water	-0.74	-0.50	0.67	0.71	0.05	0.37	0.61	0.04	0.25	0.74	0.72	-0.07	0.72	1.00													
Chl a sed.	-0.63	-0.51	0.54	0.63	-0.21	-0.10	0.36	-0.29	0.34	0.56	0.60	-0.38	0.60	0.81	1.00												
Phaeo watr	-0.75	-0.91	0.52	0.25	0.26	0.33	0.16	-0.09	-0.41	0.45	0.30	-0.14	0.30	0.53	0.69	1.00											
Phaeo sed.	-0.90	-0.83	0.73	0.66	0.23	0.57	0.56	0.14	-0.22	0.74	0.61	0.23	0.61	0.82	0.66	0.75	1.00										
POC	0.20	0.12	-0.17	0.05	-0.11	-0.43	0.08	0.19	-0.02	-0.05	-0.01	0.27	-0.01	-0.41	-0.23	-0.24	-0.31	1.00									
OC	-0.62	-0.72	0.84	0.62	-0.06	0.20	0.52	-0.14	-0.15	0.61	0.52	-0.26	0.52	0.74	0.78	0.79	0.74	-0.09	1.00								
H2s water	-0.72	-0.57	-0.01	0.23	0.26	0.16	0.12	0.15	0.12	0.45	0.45	0.27	0.45	0.37	0.55	0.54	0.56	-0.21	0.16	1.00							
Macrofauna	0.25	0.53	-0.76	-0.57	0.22	-0.03	-0.59	-0.02	0.31	-0.49	-0.39	-0.01	-0.39	-0.22	-0.30	-0.45	-0.40	-0.36	-0.72	0.12	1.00						
Meiofauna	-0.20	-0.41	-0.46	-0.71	0.76	0.38	-0.43	0.35	-0.64	-0.18	-0.32	0.29	-0.32	-0.41	-0.32	0.34	-0.06	0.02	-0.19	0.26	0.17	1.00					
Microfauna	-0.45	-0.72	-0.07	-0.38	0.62	0.39	-0.27	0.17	-0.75	0.02	-0.17	0.18	-0.17	-0.13	0.03	0.70	0.30	0.01	0.21	0.40	-0.15	0.89	1.00				
Gut fauna	0.52	0.60	0.05	0.31	-0.78	-0.50	0.07	-0.38	0.55	-0.17	0.00	-0.19	0.00	-0.14	-0.07	-0.55	-0.30	-0.13	-0.33	-0.13	0.08	-0.69	-0.75	1.00			
Shrimp wt	-0.94	-0.89	0.22	0.19	0.63	0.56	0.29	0.40	-0.26	0.64	0.52	0.36	0.52	0.53	0.53	0.80	0.77	-0.19	0.48	0.80	-0.14	0.48	0.68	-0.62	1.00		
Feed	-0.89	-0.93	0.23	0.08	0.71	0.68	0.23	0.43	-0.50	0.55	0.37	0.42	0.37	0.41	0.36	0.80	0.75	-0.17	0.46	0.66	-0.20	0.60	0.79	-0.72	0.97	1.00	

Table ST5 Correlation matrix of various parameters for non-aerated pond EC-2

	Temp	Salinity	pH water	pH sed	eh water	eh sed	DO	BOD	COD	NO3-N	NO2-N	NH3-N	PO4-P	Chl a water	chl a sed	phaeo watr	phaeo sed	POC	OC	H2s water	Macrofauna	Meiofauna	Microfauna	Gut fauna	Shrimp wt	Feed	
Temp	1.00																										
Salinity	0.79	1.00																									
pH water	-0.49	-0.91	1.00																								
pH sed.	-0.18	0.02	-0.06	1.00																							
eh water	-0.35	-0.53	0.50	-0.15	1.00																						
eh sed.	-0.19	-0.41	0.40	-0.16	0.95	1.00																					
DO	-0.56	-0.41	0.30	0.15	0.24	0.21	1.00																				
BOD	-0.64	-0.76	0.69	0.30	0.69	0.61	0.52	1.00																			
COD	0.12	0.55	-0.74	-0.28	-0.50	-0.45	0.05	-0.71	1.00																		
NO3-N	-0.74	-0.75	0.64	0.13	0.42	0.36	0.91	0.73	-0.25	1.00																	
NO2-N	-0.69	-0.82	0.76	0.14	0.61	0.53	0.75	0.75	-0.42	0.93	1.00																
NH3-N	0.20	0.59	-0.78	0.08	-0.47	-0.36	-0.23	-0.34	0.52	-0.45	-0.70	1.00															
PO4-P	0.15	0.43	-0.56	0.57	-0.06	0.07	-0.35	-0.05	0.06	-0.42	-0.40	0.61	1.00														
Chl a water	-0.15	-0.26	0.22	0.44	0.05	0.13	-0.38	0.08	-0.34	-0.12	0.09	-0.15	0.47	1.00													
Chl a sed.	-0.53	-0.56	0.41	-0.02	-0.06	-0.10	-0.04	-0.01	0.03	0.23	0.32	-0.32	-0.16	0.65	1.00												
Phaeo watr	-0.74	-0.95	0.85	-0.03	0.43	0.34	0.23	0.58	-0.46	0.61	0.72	-0.59	-0.35	0.48	0.77	1.00											
Phaeo sed.	-0.59	-0.20	-0.10	0.43	-0.08	-0.19	0.00	-0.02	0.25	0.07	0.14	0.04	0.33	0.52	0.64	0.34	1.00										
POC	0.57	0.51	-0.40	-0.24	-0.36	-0.26	-0.43	-0.25	0.04	-0.54	-0.73	0.65	0.18	-0.37	-0.59	-0.58	-0.63	1.00									
OC	-0.62	-0.80	0.71	0.18	0.42	0.46	0.35	0.56	-0.40	0.65	0.76	-0.50	-0.09	0.63	0.72	0.88	0.34	-0.65	1.00								
H2s water	-0.88	-0.70	0.46	0.04	0.31	0.05	0.42	0.46	-0.06	0.57	0.60	-0.37	-0.37	-0.03	0.47	0.64	0.60	-0.62	0.38	1.00							
Macrofauna	0.52	0.74	-0.67	0.18	-0.09	-0.14	-0.40	-0.35	0.18	-0.64	-0.57	0.31	0.43	-0.27	-0.67	-0.78	-0.02	0.29	-0.80	-0.27	1.00						
Meiofauna	-0.31	0.04	-0.31	-0.19	-0.40	-0.37	0.24	-0.08	0.54	0.12	-0.22	0.70	0.01	-0.36	0.03	-0.09	0.07	0.39	-0.16	0.13	-0.26	1.00					
Microfauna	-0.44	-0.62	0.61	0.02	0.85	0.69	0.16	0.83	-0.72	0.38	0.54	-0.43	-0.10	-0.01	-0.13	0.46	-0.06	-0.18	0.28	0.47	-0.01	-0.32	1.00				
Gut fauna	0.30	0.45	-0.30	0.27	-0.62	-0.72	0.03	-0.50	0.31	-0.20	-0.21	-0.11	-0.22	-0.27	-0.15	-0.44	0.10	-0.11	-0.48	-0.01	0.42	-0.18	-0.48	1.00			
Shrimp wt	-0.95	-0.86	0.59	0.15	0.54	0.38	0.39	0.72	-0.31	0.65	0.70	-0.30	-0.09	0.29	0.53	0.82	0.56	-0.57	0.69	0.85	-0.48	0.12	0.63	-0.46	1.00		
Feed	-0.83	-0.92	0.77	0.07	0.57	0.39	0.38	0.86	-0.58	0.68	0.70	-0.38	-0.27	0.11	0.32	0.80	0.21	-0.32	0.58	0.75	-0.49	0.08	0.78	-0.47	0.90	1.00	

Table ST6 Correlation matrix of various parameters for creek EC-2

	Temp	Salinity	Ph water	ph sed	ch water	ch sed	DO	BOD	COD	NO3	NO2	NH3	PO4	Chl a water	chl a sed	phaeo watr	phaeo sed	POC	OC	H2s water	Macrofauna	Meiofauna	Microfauna	Gut fauna	Shrimp wt	Feed	
Temp	1.00																										
Salinity	0.87	1.00																									
Ph water	-0.44	-0.53	1.00																								
ph sed	-0.46	-0.31	0.80	1.00																							
ch water	-0.51	-0.47	-0.28	-0.41	1.00																						
ch sed	-0.60	-0.56	0.26	0.01	0.76	1.00																					
DO	-0.53	-0.40	0.66	0.79	0.04	0.37	1.00																				
BOD	-0.41	-0.27	-0.15	-0.03	0.75	0.63	0.52	1.00																			
COD	0.14	0.50	-0.18	0.31	-0.43	-0.51	0.20	-0.10	1.00																		
NO3	-0.80	-0.65	0.58	0.71	0.27	0.48	0.91	0.55	0.15	1.00																	
NO2	-0.70	-0.47	0.48	0.73	0.12	0.28	0.90	0.49	0.40	0.96	1.00																
NH3	-0.38	-0.26	-0.10	0.09	0.58	0.57	0.43	0.86	-0.23	0.43	0.34	1.00															
PO4	-0.70	-0.47	0.48	0.73	0.12	0.28	0.90	0.49	0.40	0.96	1.00	0.34	1.00														
Chl a water	-0.74	-0.50	0.67	0.71	0.85	0.37	0.61	0.84	0.25	0.74	0.72	-0.07	0.72	1.00													
chl a sed	-0.63	-0.51	0.54	0.63	-0.21	-0.10	0.36	-0.29	0.34	0.56	0.60	-0.38	0.60	0.81	1.00												
phaeo watr	-0.75	-0.91	0.52	0.25	0.26	0.33	0.16	-0.09	-0.41	0.45	0.30	-0.14	0.30	0.53	0.69	1.00											
phaeo sed	-0.90	-0.83	0.73	0.66	0.23	0.57	0.56	0.14	-0.22	0.74	0.61	0.23	0.61	0.82	0.66	0.75	1.00										
POC	0.20	0.12	-0.17	0.05	-0.11	-0.43	0.88	0.19	-0.02	-0.85	-0.01	0.27	-0.01	-0.41	-0.23	-0.24	-0.31	1.00									
OC	-0.62	-0.72	0.84	0.62	-0.06	0.20	0.52	-0.14	-0.15	0.61	0.52	-0.26	0.52	0.74	0.78	0.79	0.74	-0.09	1.00								
H2s water	-0.72	-0.57	-0.01	0.23	0.26	0.16	0.12	0.15	0.12	0.45	0.45	0.27	0.45	0.37	0.55	0.54	0.56	-0.21	0.16	1.00							
Macrofauna	0.25	0.53	-0.76	-0.57	0.22	-0.03	-0.59	-0.02	0.31	-0.49	-0.39	-0.01	-0.39	-0.22	-0.30	-0.45	-0.40	-0.36	-0.72	0.12	1.00						
Meiofauna	-0.20	-0.41	-0.46	-0.71	0.76	0.38	-0.43	0.35	-0.64	-0.18	-0.32	0.29	-0.32	-0.41	-0.32	0.34	-0.06	0.02	-0.19	0.26	0.17	1.00					
Microfauna	-0.45	-0.72	-0.07	-0.38	0.62	0.39	-0.27	0.17	-0.75	0.02	-0.17	0.18	-0.17	-0.13	0.03	0.70	0.30	0.01	0.21	0.40	-0.15	0.89	1.00				
Gut fauna	0.20	0.50	-0.21	0.27	-0.38	-0.38	0.27	0.07	0.88	0.18	0.41	0.04	0.41	0.05	0.10	-0.52	-0.26	-0.07	-0.35	0.18	0.28	-0.54	-0.72	1.00			
Shrimp wt	-0.94	-0.89	0.22	0.19	0.63	0.56	0.29	0.40	-0.24	0.64	0.52	0.36	0.52	0.53	0.53	0.80	0.77	-0.19	0.48	0.80	-0.14	0.48	0.68	-0.28	1.00		
Feed	-0.89	-0.93	0.23	0.08	0.71	0.68	0.23	0.43	-0.50	0.55	0.37	0.42	0.37	0.41	0.36	0.80	0.75	-0.17	0.46	0.66	-0.20	0.60	0.79	-0.48	0.97	1.00	

Table ST7 Descriptive statistics for aerated pond EC-1

Parameter	Mean	Standard Deviation	Minimum	Maximum
Temperature ($^{\circ}\text{C}$)	32.07	2.21	27.90	35.40
Salinity	24.58	14.62	7.20	43.18
pH water	7.67	0.42	7.09	8.32
pH sediment	7.01	0.58	5.58	7.62
E_h water (mv)	86.29	16.97	62.80	112.77
E_h sed. (mv)	68.20	21.47	36.53	97.43
DO (mg.l^{-1})	3.52	0.37	2.93	4.06
BOD (mg.l^{-1})	2.00	1.09	0.26	3.05
COD (mg.l^{-1})	41.15	26.63	10.00	87.00
$\text{NO}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	0.86	0.73	0.30	2.72
$\text{NO}_2\text{-N}$ ($\mu\text{mol.l}^{-1}$)	0.28	0.17	0.06	0.53
$\text{NH}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	0.77	0.57	0.10	1.93
$\text{PO}_4\text{-P}$ ($\mu\text{mol.l}^{-1}$)	1.96	1.68	0.11	4.65
Chl <i>a</i> water (mg.m^{-3})	3.69	1.79	1.42	6.41
Chl <i>a</i> sed. (mg.m^{-3})	4.74	3.78	1.38	14.24
Phaeo water (mg.m^{-3})	3.33	0.99	1.59	5.26
Phaeo sed. (mg.m^{-3})	2.58	1.01	1.05	3.78
OC (%)	2.36	1.25	1.24	5.43
POC (mg.l^{-1})	7.62	4.74	3.20	18.63
H_2S water ($\mu\text{g.l}^{-1}$)	0.84	0.31	0.43	1.29
Macrofauna (no.m^{-2})	2623.19	1733.90	885.81	5777.00
Meiofauna (no.10cm^{-2})	104.87	96.66	0.00	251.67
Microbenthos (no.3.2cm^{-2})	552.41	712.17	53.08	1857.75
Gut biota (organism.ind^{-1})	307.09	219.22	19.00	579.69
Shrimp weight (gms)	10.05	9.37	0.05	27.10
Feed (Kg)	3.13	2.12	0.08	6.03

Table ST8 Descriptive statistics for non-aerated pond EC-1

Parameter	Mean	Standard Deviation	Minimum	Maximum
Temperature (°C)	31.93	2.25	27.80	35.30
Salinity	24.60	13.82	8.10	41.89
pH water	7.71	0.41	7.16	8.50
pH sediment	6.50	1.13	3.75	7.47
E _h water (mv)	72.95	16.39	54.60	94.60
E _h sed. (mv)	59.92	16.30	35.03	
DO (mg.l ⁻¹)	3.83	0.64	2.86	4.81
BOD (mg.l ⁻¹)	2.64	0.73	1.16	3.73
COD (mg.l ⁻¹)	43.09	31.87	10.71	93.00
NO ₃ -N (µmol.l ⁻¹)	0.59	0.22	0.33	0.98
NO ₂ -N (µmol.l ⁻¹)	0.73	1.03	0.08	3.42
NH ₃ -N (µmol.l ⁻¹)	1.38	1.51	0.31	4.76
PO ₄ -P (µmol.l ⁻¹)	1.85	1.04	0.41	3.58
Chl <i>a</i> water (mg.m ⁻³)	3.84	1.43	1.14	5.93
Chl <i>a</i> sed. (mg.m ⁻³)	4.27	2.55	2.06	10.68
Phaeo water (mg.m ⁻³)	3.22	1.20	1.02	5.36
Phaeo sed (mg.m ⁻³)	2.45	0.54	1.68	3.29
OC (%)	1.99	0.99	1.01	3.54
POC (mg.l ⁻¹)	6.89	3.39	3.85	12.56
H ₂ S water (µg.l ⁻¹)	0.77	0.30	0.38	1.37
Macrofauna (no.m ⁻²)	1638.96	802.82	693.24	3312.15
Meiofauna (no.10cm ⁻²)	150.02	319.04	0.00	985.47
Microbenthos (no.3.2cm ⁻²)	506.21	443.71	35.39	1344.66
Gut biota (organism.ind ⁻¹)	151.91	110.97	18.00	354.00
Shrimp weight (gms)	11.29	8.78	0.05	25.47
Feed (Kg)	1.67	1.05	0.09	3.03

Table ST9 Descriptive statistics for creek EC-1

Parameter	Mean	Standard Deviation	Minimum	Maximum
Temperature ($^{\circ}\text{C}$)	31.27	2.21	27.20	34.80
Salinity	17.08	17.00	1.05	39.46
pH water	7.58	0.28	7.11	8.03
pH sediment	6.48	0.85	4.74	7.61
E_h water (mv)	66.72	14.49	48.14	89.17
E_h sed. (mv)	58.16	15.16	38.54	80.94
DO (mg.l^{-1})	3.73	0.55	3.04	4.68
BOD (mg.l^{-1})	2.94	0.50	2.09	3.73
COD (mg.l^{-1})	26.53	22.29	5.36	71.50
$\text{NO}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	1.27	1.16	0.31	3.57
$\text{NO}_2\text{-N}$ ($\mu\text{mol.l}^{-1}$)	0.71	0.63	0.19	2.09
$\text{NH}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	1.10	0.52	0.50	2.11
$\text{PO}_4\text{-P}$ ($\mu\text{mol.l}^{-1}$)	2.36	1.04	0.36	3.63
Chl <i>a</i> water (mg.m^{-3})	3.84	1.43	1.14	5.93
Chl <i>a</i> sed (mg.m^{-3})	4.31	2.62	1.24	10.68
Phaeo water (mg.m^{-3})	2.99	1.10	1.06	4.28
phaeo sed (mg.m^{-3})	2.32	0.70	1.28	3.22
OC (%)	2.86	1.62	1.18	6.01
POC (mg.l^{-1})	6.71	2.03	3.51	9.15
H_2S water ($\mu\text{g.l}^{-1}$)	0.63	0.40	0.11	1.37
Macrofauna (no.m^{-2})	885.81	365.37	462.16	1675.33
Meiofauna (no.10cm^{-2})	765.89	807.65	0.00	2517.00
Microbenthos (no.3.2cm^{-2})	9413.48	5642.02	3715.50	17834.40

Table ST10 Descriptive statistics for aerated pond EC-2

Parameter	Mean	Standard Deviation	Minimum	Maximum
Temperature ($^{\circ}\text{C}$)	30.81	1.12	28.70	32.20
Salinity	19.59	9.87	4.44	27.38
pH water	7.58	0.38	7.20	8.30
pH sediment	7.29	0.22	7.10	7.76
E_h water (mv)	99.08	29.36	51.83	145.07
E_h sed. (mv)	103.04	16.85	78.20	128.87
DO (mg.l^{-1})	3.41	0.57	2.67	4.64
BOD (mg.l^{-1})	1.35	0.43	0.85	2.12
COD (mg.l^{-1})	88.13	43.44	16.00	137.40
$\text{NO}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	1.13	1.45	0.05	4.48
$\text{NO}_2\text{-N}$ ($\mu\text{mol.l}^{-1}$)	1.33	1.23	0.26	4.12
$\text{NH}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	0.36	0.12	0.23	0.60
$\text{PO}_4\text{-P}$ ($\mu\text{mol.l}^{-1}$)	1.33	1.23	0.26	4.12
Chl <i>a</i> water (mg.m^{-3})	18.49	17.40	2.14	55.54
Chl <i>a</i> sed (mg.m^{-3})	5.95	3.12	1.07	11.75
Phaeo water (mg.m^{-3})	3.47	3.49	1.20	9.93
Phaeo sed (mg.m^{-3})	23.78	11.72	8.66	47.31
OC (%)	2.60	1.35	1.56	5.21
POC (mg.l^{-1})	7.08	2.36	4.14	11.81
H_2S water ($\mu\text{g.l}^{-1}$)	0.56	0.24	0.03	0.80
Macrofauna (no.m^{-2})	3880.22	1767.15	1386.48	7317.53
Meiofauna (no.10cm^{-2})	534.07	512.07	33.25	1289.75
Microbenthos (no.3.2cm^{-2})	2982.04	3369.23	188.96	9403.75
Gut biota (organism.ind^{-1})	478.94	283.43	40.00	838.00
Shrimp weight (gms)	12.83	8.11	0.05	23.81
Feed (Kg)	2.07	1.61	0.00	4.05

Table ST11 Descriptive statistics for non-aerated EC-2

Parameter	Mean	Standard Deviation	Minimum	Maximum
Temperature ($^{\circ}\text{C}$)	30.74	1.24	28.70	32.80
Salinity	19.57	9.50	5.39	27.50
pH water	7.84	0.58	7.20	8.55
pH sediment	7.18	0.34	6.42	7.63
E_h water (mv)	86.74	40.34	27.77	140.47
E_h sed. (mv)	87.51	37.08	24.97	126.60
DO (mg.l^{-1})	3.37	0.46	2.70	4.21
BOD (mg.l^{-1})	1.48	0.55	0.80	2.46
COD (mg.l^{-1})	98.58	52.49	14.20	166.00
$\text{NO}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	1.26	1.78	0.08	4.88
$\text{NO}_2\text{-N}$ ($\mu\text{mol.l}^{-1}$)	0.85	0.64	0.10	2.04
$\text{NH}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	0.27	0.11	0.17	0.51
$\text{PO}_4\text{-P}$ ($\mu\text{mol.l}^{-1}$)	1.46	0.70	0.85	2.75
Chl <i>a</i> water (mg.m^{-3})	16.37	10.63	3.20	33.11
Chl <i>a</i> sed. (mg.m^{-3})	4.14	2.23	1.07	7.48
Phaeo water (mg.m^{-3})	2.59	2.56	0.73	6.56
Phaeo sed. (mg.m^{-3})	16.24	7.59	6.41	25.95
OC (%)	2.70	1.77	0.92	6.03
POC (mg.l^{-1})	6.81	2.71	2.21	10.79
H_2S water ($\mu\text{g.l}^{-1}$)	0.50	0.21	0.03	0.75
Macrofauna (no.m^{-2})	2050.84	704.08	693.24	3042.55
Meiofauna (no.10cm^{-2})	421.50	477.02	68.25	1449.00
Microbenthos (no.3.2cm^{-2})	1637.47	1603.70	153.93	4653.22
Gut biota (organism.ind^{-1})	436.83	163.78	88.00	583.00
Shrimp weight (gms)	13.66	8.92	0.05	25.52
Feed (Kg)	1.04	0.82	0.00	2.13

Table ST12 Descriptive statistics for Creek EC-2

Parameter	Mean	Standard Deviation	Minimum	Maximum
Temperature ($^{\circ}\text{C}$)	30.76	1.25	28.70	32.20
Salinity	15.21	12.28	0.28	27.64
pH water	7.37	0.33	6.88	7.72
pH sediment	7.37	0.40	6.80	7.85
E_h water (mv)	82.94	38.38	28.80	133.45
E_h sed. (mv)	86.32	28.12	42.45	124.05
DO (mg.l^{-1})	3.66	0.84	2.29	4.68
BOD (mg.l^{-1})	1.52	0.94	0.48	2.93
COD (mg.l^{-1})	102.16	43.15	44.60	169.60
$\text{NO}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	2.11	1.92	0.38	4.80
$\text{NO}_2\text{-N}$ ($\mu\text{mol.l}^{-1}$)	1.26	1.48	0.20	4.34
$\text{NH}_3\text{-N}$ ($\mu\text{mol.l}^{-1}$)	0.26	0.10	0.12	0.40
$\text{PO}_4\text{-P}$ ($\mu\text{mol.l}^{-1}$)	1.25	0.40	0.92	2.00
Chl <i>a</i> water (mg.m^{-3})	4.26	2.53	1.07	8.54
Chl <i>a</i> sed (mg.m^{-3})	6.11	5.94	2.14	19.22
Phaeo water (mg.m^{-3})	6.52	6.03	0.85	18.16
phaeo sed (mg.m^{-3})	33.92	17.74	14.28	60.66
OC (%)	3.03	2.02	1.10	6.67
POC (mg.l^{-1})	7.97	1.55	5.96	10.55
H_2S water ($\mu\text{g.l}^{-1}$)	0.41	0.21	0.01	0.69
Macrofauna (no.m^{-2})	1921.67	1179.53	521.63	4101.67
Meiofauna (no.10cm^{-2})	958.70	875.09	130.50	2712.63
Microbenthos (no.3.2cm^{-2})	2861.64	2272.07	547.77	6503.03

Table ST13 Permissible limits for important water quality parameters in shrimp culture practices. (Paulraj et al 1998, Ramachandra et al 1999)

Water quality parameter	Recommended levels
Temperature	28 – 33°C
pH	7.5 – 8.5
Salinity	10 – 30 ‰
Dissolved oxygen	4 – 6 mg/L
BOD	<3 mg/L
COD	50 mg/L -100 mg/L
Nitrate (NO ₃ -N)	< 1.1 mg/L
Nitrite (NO ₂ -N)	<1.0 mg/L
Phosphate	5 mg/L
Ammonia (NH ₄ -N)	< 1.0 mg/L
Un-ionised ammonia (NH ₃)	< 0.1 mg/L
Chlorophyll <i>a</i> & phaeophytin	20-250 µg.l ⁻¹
Organic Carbon	0.05 -5 %
H ₂ S	< 0.003 mg/L
Water colour	Light brown or green

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