CHAPTER I

Introduction
INTRODUCTION

An understanding of the physiological mechanisms of an animal pre-supposes knowledge of the chemical constituents of its body, as well as transformations these chemical constituents undergo within the body. Such an investigation is also essential for the understanding of the balance that exists between the animal and its environment.

Cyclical changes in biochemical composition of animal tissue are mainly studied to assess the nutritive status of an organism. This Information may however, be used in supplementing other studies like assessment of the course of reproductive cycle. In some cases, overall change in the chemical composition during an annual cycle has been specially correlated to the events of gonadal changes of marine organisms (Lubet, 1957; Wilson and Seed, 1974; Pieters et al., 1979; Barkati and Ahmed, 1990).

Studies on seasonal variation in the biochemical composition of bivalve species have been initiated since last two decades. Emphasis was given to the economically important species, such as oysters (Spondylus sp., Khan et al. 1984; Crassostrea rivularis, C. madrasensis and C. gryphoides; Aftab. 1986), mussels (Perna viridis; Fatima et al, 1985), Clams (Marcia cor, Perveen, 1990). These investigations were
related to the analysis of the animal tissues with reference to seasonal changes in protein, lipid, carbohydrate, mineral contents of the soft tissues.

Marine and estuarine bivalves form the food of many coastal people throughout the world. Oysters and Mussels being a good protein food, a comprehensive knowledge of their biochemical constituents during the different seasons of the year would be very valuable.

Despite the rich variety of commercially valuable molluscs in India, their contribution to the yearly catch is insignificant. Various estimates show that the contribution of marine molluscs to the overall annual fishery output varies from 0.85 to 1.0% (Nair and Mahadevan, 1983). At present there is no reliable production data of edible inland molluscs such as the freshwater mussels and other gastropods known to be consumed in various parts of the country. Apart from the tremendous potential to mitigate the protein deficiency in the country, some of the molluscan resources command a lucrative overseas market.
Contribution of Seafood in Human diet

Seafood can also be an important contribution to the diets of many individuals because of their unique nutritional composition. Molluscs are known to be highly palatable and nutritious food. They constitute a valuable fishery resource in various sectors of coastal India. Apart from its food value, it is one of the main sources of lime. The exploitation of bivalves has been observed to be only as a subsistence occupation, but the growing demand for protein food and multiple uses of the molluscan shell in lime-based chemical industries have created tremendous awareness of the benefits of exploiting and developing molluscan resources. Bivalves, cephalopods and gastropods are the principal groups of molluscs exploited from coastal, estuarine and backwater environments. The variety of seafood continually available encourages consumption. Seafood can also be an important contribution to the diets of many individuals because of their unique nutritional composition.

The role of food from the sea in meeting our nutritional requirements can be an interesting subject to study. Nutritional labeling provides a standardized format to appear on all foods giving such information and on a uniform basis for comparison.
Although in India the majority of the population is meat eating by tradition, the intake of animal protein is markedly low. Protein consumption in India is estimated at 52 g per person per day of which animal protein constitutes a meager 6 g, compared to 92 g and 65 g in USA and 92 g and 61 g in Australia. If population growth projections are of any guide, India should, in addition to conventional sources of animal protein (fish, beef, pork, chicken, etc.) explore unconventional areas to increase the present protein level intake as well as to ensure alternative foods at reasonable prices. Among the unconventional sources of animal proteins, molluscs as a group have great prospects. India has a rich variety of edible marine and freshwater molluscs such as oysters, mussels, clams, squid, cuttle fish, octopuses and land snails, apart from the commercially harvested species such as the pearl oysters and sacred conchs.

All marine molluscs store large quantities of glycogen and fat which make them highly nutritious as human food (Young, 1928). Among shellfishes, oysters are considered as a valuable food item because they can provide many of the elements, which are essential for balanced diet. Shellfish like oysters and mussels are known to be high in protein, low in fat and low in calories.
Sea mussel is not a very popular item of food except among the poorer sections of the coastal population and that too only when fish is scarce. On the other hand in Europe, particularly in west Germany, Sweden, France, Belgium, Turkey, Yugoslavia, Ireland and Holland, it is a popular item of food and since natural supply is unable to meet the large demand.

Catch statistics

As per the available information, the total annual production of molluscs is 1.4 lakh tons, which is 10% of, total marine fish landing. The yearly landing of cephalopods, bivalves and gastropods are 43,000; 96,300 and 1,256 MT, respectively. The exploited cephalopods include cuttlefish (60%), squid (39%) and octopus (1%). Bivalves comprise clams (70%), cockles (9.5%), mussels (3.6%), oysters (1.0%) and windowpane oysters (14.5%).

Growth and production are directly affected by ecological conditions of the oyster and mussel bed. The main objective of the study is to develop a background for subsequent development of a site suitability index for culture of *C. madrasensis* or *Perna viridis* along the Indian coast and to spread awareness of nutritional importance of these species in human diet.
Despite the fact that Kali estuary has rich molluscan resources and suitable sites for their cultivation, the extent of their exploitation and utilization has been limited. It is necessary to popularise the culture methods in rural areas. A large section of the population is either unaware of the value of molluscan as a food or prejudiced against their use due to conservative food habit. The role that edible molluscs are going to play along with fish and crustaceans in meeting the country's need for balanced diet has to be more widely recognized.

**Medicinal values of bivalves**

Many proteins produced by aquatic organisms can be utilized in diagnostic applications, either as biomarkers in response to a specific environmental factor or as binding proteins that could be used in assaying toxic compounds (Butler, 2005). Extracts prepared from marine bivalves such as green mussel (*Perna viridies*), estuarine oyster (*Crassostrea madrasensis*), giant oyster (*Crassostrea gryphoides*), estuarine clam (*Meretrix casta*), black clam (*Villorita cyprinoides*), and mud crab (*Polymesoda erosa*) were found to possess high antiviral activity when tested with influenza virus strains type-A (Chatterji *et al.*, 2002).
Pacific Oyster Extract (Once the Oysters are harvested, they are immediately freeze-dried. Pacific Oyster Extract typically contains: 38% protein, 4% L-Taurine, 40% carbohydrate, 7% minerals) is supplemented with 250 mg of pure L-Taurine. Controlled clinical studies show the Pacific Oyster Extract relieves angina, normalizes blood pressure, corrects cardiac arrhythmia, normalizes blood sugar levels in diabetics, and alleviates liver disturbances caused by poor fat metabolism and alcohol consumption. L-Taurine is vital to adrenal gland function, liver and gallbladder function, nerve impulse transmission and eye physiology (Source - internet).

Bursatellanin-P, a 60 kDa protein was purified from the purple ink of the sea hare *Bursatella leachii* which exhibited anti-HIV activity (Rajaganapathi *et al.*, 2002)

Oysters are a valuable source of protein, vitamins and minerals needed for good health. A 3-ounce serving, or about 6 oysters, is a good source of protein and low in fat. That same serving has only 58 calories and 46 milligrams of cholesterol.
These shellfish are an excellent source of the minerals iron and zinc, as well as vitamin B12. A serving of oysters supplies 31 percent of the Daily Value for iron, 268 percent for vitamin B12 and 509 percent for zinc.

Iron functions primarily as a carrier of oxygen in the blood and muscles. Vitamin B12 assists in forming red blood cells and in building genetic material. It helps the nervous system to function and protein and fat to metabolize in the body. Vitamin B12 is found only in animal products. Zinc helps form protein in the body. It assists with wound healing, blood formation and general tissue growth and maintenance. Because zinc is a component of many enzymes, it is involved in most metabolic processes.

Oysters are also a good source of the mineral phosphorus, providing 12 percent of the Daily Value. Phosphorus helps build strong bones and teeth. It is also involved in releasing energy from fat, protein and carbohydrates during metabolism, and in forming genetic material, cell membranes and many enzymes.
Culture aspects of bivalves

Seasonal variation in the composition of the animal in association with the seasonal variation in the environment should be studied in order to arrive at the efficient means of culture and conservations. The bivalves are largely influenced by the environmental changes such as rainfall, amount of freshwater discharge of rivers, tidal amplitude, temperature, changes in salinity and pH, and occurrence of obnoxious blooms of phytoplankton. On top of these are the domestic, agricultural and industrial pollutants. At present, bivalves such as clams are processed and marketed without much consideration for the product's sanitary quality. Although aquaculture practices on cultivable molluscs have not been initiated on a commercial scale, experimental trials on the farming methods of a number of species have been carried out and suitable farming techniques evolved (Mahadevan et al., 1980; Kuriakose and Appukuttan, 1980; Narasimham, 1980; Rao, 1980). Further, successful production of seed of a number of bivalves through hatchery has been achieved (Nayer et al., 1984, Appukuttan et al., 1987; Narasimham et al., 1980).

There is vast potential for culturing edible oysters in India, especially the large *C. madrasensis* and *C. gryphoides*. Techniques for their spat collection and culture have been developed and demonstrated feasible in various parts of the country (CMFRI, 1980;
Joseph and Joseph, 1983; Nair and Mahadevan, 1983; Reuben et al., 1983). Oyster seed production through hatchery techniques have also been developed at the Central Marine Fisheries Research Institute (CMFRI) in Cochin (Anon, 1982).

The Crassostrea-type oysters are the most important commercial species and have the greatest development potential due to their tolerance of estuarine conditions and usually abundant spat fall. Many of them have been experimentally and commercially cultured. C. gigas and C. virginica have been the object of numerous attempts at introduction into tropical waters. Potential exists for transplantation of several species within the tropical zone.

In Mexico oyster production by culture practices is slowly increasing and its production comes mainly from one species, Crassostrea virginica. Culture technology in Mexico, utilizes low-cost materials and is adaptable to rural development.

The CMFRI has developed a technique for rope culture of mussels. The green mussels are cultured on 5–10 m long ropes suspended from wooden rafts of 36 to 100 sq. m. in area. The ropes are seeded by embedding the spat on the rope with the help of cloth firmly stitched to it. Mussel seeds in the size range of 20–35 mm are
considered suitable for rearing. They reach the marketable size of 55–60 mm during a culture period of about 5 months. The average production is 10–12 kg of mussels per metre of rope in the bay in 7 months and 15 kg in the open sea in 5 months. On a raft of 30 sq. m. in area, 50 ropes (each 6 m in length) can be suspended for culturing mussels.

According to the composition of aquaculture products, molluscs are second to finfish in the list with an estimate of over 2.67 million tonnes (Pillay, 1990). Oysters account for about 12% of the total world production from aquaculture. In India the picture is not different from global level. Though fish provides the largest single source of animal protein, its demand outstrips supply owing to the ever-increasing human population. The annual per capita fish consumption in India is only 4kg against the recommended 31Kg by Nutrition Advisory Committee on human nutrition. This shows that our protein demand is so great that it is important to increase fish production, and the only alternative available is aquaculture (Santhanam et al., 1990). India has a coastal line running to 7517 km and the total spread of continental shelf is about 40 million hectares. The east and west coasts of India are productive and are suitable for undertaking mariculture practices. It has been suggested by many that while the coastal areas offer much scope for large scale culture of organisms such as oysters, mussels, seaweed
etc. the open sea could be utilised for suspended cage, raft and long line culture of fish and shellfish. Mariculture is of recent origin in India, although Hornell visualized the vital importance of oyster farming in this country as early as 1917 and carried out the culture of *Crassostrea madrasensis* in the Pulicat lake. India produces about 0.14 million tonnes of fish and shellfish through aquaculture and the contribution through coastal aquaculture is only 7% (Santhanam et al., 1990).

The demand for mussels of good quality exceeds the supply (Manson, 1991). Since mussels are positioned low in the food web feeding directly on phytoplankton, they are considered most suitable for cultivation producing greater yields per unit area compared to species at higher trophic levels (Ryther and Bardach, 1968).

Presently more than 101 million tonnes of mussels are harvested annually throughout the world (FAO, 1990). China and Spain are two prominent producers of mussels, contributing 40 and 20 per cent respectively, of the world production (Hickman, 1992).

Mussels prefer the wave exposed flat or gently shelving shores where they emerge as dominant community in temperate latitudes (Lewis, 1964). Although mussels are more abundant in the intertidal zone, can grow equally well sub tidally if provided refuge from sea-star predation.
Uncertainties in the availability of mussel spat and the seasonal nature of their availability are major constraints in developing mussel culture as a viable industry. It appears necessary to develop a technology to artificially induce mussels to spawn and techniques to rear larvae indoors up to the transplantable stage (young mussels). Green mussels have been successfully spawned at the Kovalam field laboratory of the CMFRI and over 20,000 juvenile spat have been produced (Sreenivasan et al., 1988). Structure of the rafts need to be suitably designed to withstand the rough weather allowing year round operation in the open sea.

The status of major molluscan resources and its economic importance in India particularly Oysters and Mussels

OYSTERS

Although edible oysters are considered as prized delicacies in the west and other advanced countries, they still remain more or less alien to the Indian palates. Despite the wide distribution of this mollusc species, there is very little demand in the domestic market. Oysters collected from the wild satisfy the local demand in Karwar. The present fishery of edible oysters is highly localized and it caters mainly to the
western style hotels. The oyster capture fishery from wild stocks is under-exploited and, there are no commercial oyster culture ventures in uttar kannada district to supply domestic and export markets.

Resource distribution

Oyster

Thirteen species of edible oysters have been reported from India (Awati and Rai, 1931; Rao, 1974) among which, four species viz., Crassostrea gryphoides, C. discoides, C. madrasensis and Saccostrea cucullata are of commercial value. C. gryphoides and C. discoides are mostly found in the North-Western coast of India; the former being more important and occurring in the coastal areas of North Canara and Maharashtra (Nair and Mahadevan, 1983). According to Alagarswamy and Narasimhan (1973), C. discoides is distributed in the muddy creeks of Kutch, the Arama creek and off Poshetra point, Port Okha, Dwaraka and Porbunder. Along the Maharashtra coast, C. gryphoides is the prevalent species although the other species also occur at a few places. Crassostrea madrasensis enjoys a wider distribution along the Southwest and East coasts of India and it is found in almost all the estuaries and backwaters.
Malad, Boiser, Satpuri, Palghar, Kelva, Navapur, Utsali, Dahisar and Mahim creeks are all important fishing grounds around Bombay, Palghar being the most important one. Oyster spats are collected from the natural beds and reared to the market size for almost 12 months in suitable fattening sites.

In the State of Maharashtra, oyster fishing is practiced in Alibag, Ratnagiri, Jayatpur and Malwan. Oyster beds are often 4 to 5 fathoms deep and the divers use chisels and hammers to detach the molluscs from the substrate. In Goa, oysters are exploited at Ribander, Siolim and Curca. The oyster beds of Karkal Island and Ladies Beach near Kanwar are not exploited to any appreciable extent. Along the coast of Cochin, in Mulki, Udayavara, Coondapur and along the Kali river, oysters are collected for domestic consumption.

In the East coast of India, *C. madrasensis* fishery used to be a lucrative activity, particularly during the 1920's. This species is also abundant in Sonapur backwaters of the Bahudi River and lake Chilka. In Pulicat Lake small-sized oysters were traditionally collected and then reared in shallow waters for the market in Madras City. This practice no longer exists and the sporadic local demand is now met by collecting the oysters from natural environment.
At present, the exploitation of wild oyster stocks is mainly carried out to cater to the needs of western-style hotels in major Indian cities. In coastal villages and communities, this edible mollusc forms a supplementary food for the poor who consume the shellfish with very little concern regarding the quality and hygiene of the product.

Mussels

The mussels belong to the family Mytilidae. Three or four species have been recorded from India. Two species of edible mussels, *Perna viridis* and *Perna indica* are widely distributed along the entire coast of India. *Perna indica* (brown mussel) occurs in southern parts of Indian peninsula between Quilon on the west coast and Rameswaram on the east coast. The *Perna viridis* (green mussel) has wider distribution on both east and west coasts particularly in north Kerala, Karnataka, parts of Mumbai, Chennai and Orissa and also some bays and backwaters. They are found on hard substrata in clusters to a depth of 10 m and are usually chiselled out by the fishermen at low tide. The green mussel, *P. viridis* enjoys a wide distribution along the west coast (Nair and Rao, 1985). In the west coast thick beds exist at Quilon, Alleppey, Cochin, Malabar, Karwar, Goa, Malvan, Ratnagiri and Gulf of Kutch, where they are regularly exploited for their meat. This mussel is particularly abundant on the rocky coasts from Calicut to Tellicherry, where there is an active fishery. In the Coromandel Coast, *P. viridis* occurs at very few
places such as Visakhapatnam, Kakinada and Madras. It attains a length of 92 mm at the end of one year at Kakinada and its breeding period is prolonged, extending from December to July (Narasimhan, 1980). The brown mussel has a limited distribution from Varkalai near Quilon on the southwest coast to Cape Comorin with rich beds at Varkalai, Kovalam, Vizhinjam, Poovar, Muttom and Colachel. The brown mussel grows to a size of 35–36 mm in a year at Vizhinjam coast and breeds from May to September (Appukuttan and Nair, 1980). The existing sustenance fishery of brown mussel at Vizhinjam is estimated to be 50–150 MT a year (CMFRI).

Fouling organisms and parasites

A large number of fouling organisms settle on the spat collectors and cultured spat. The most common fouling organisms are several species of barnacles (Balanus amphitrite communis, B. amphitrite denticulata and B. amphitrite cocqiensis). Tube-dwelling polychaetes, Hydroides norwegica, Pomatoceros triquetor, Spirobis sp. and Polydora ciliata are also found to compete with the oysters for space. Heavy fouling by barnacles and polychaetes impairs the growth of oyster spats. The problem is compounded by the fact that the breeding seasons of all these unwanted associates coincide (Joseph and Joseph, 1983). Oysters cultured by the CMFRI along the Tuticorin coast
also suffered mortalities up to 15% by the predatory gastropod *Cymatium cingulatum*. Fouling is a serious problem in the temperate species and culture systems must be evolved which can economically overcome it.

Studies on the control of biological competitors, parasites and predators need attention in India. This would stimulate the development of oyster farming as an industry.

**Canning of edible oysters**

Successful experiments have been conducted in India to can oyster meat (Balachandran *et al.*, 1984). The canning yield of oyster was 15% higher from farmed oysters than from wild specimens.

According to the CMFRI, an area that demands immediate attention is the marketing of oysters in the domestic and overseas markets. Extension work on market development, product marketing on relatively low rates, and ascertaining the consumer preferences of quality of flesh are needed in the domestic front. Proper technology for oyster meat processing and preservation to satisfy the consumer taste need to be developed to foster the overseas markets.
Muraleedharan et al. (1982) have developed processing methods for dried and pickled mussels, fried and pickled mussels and smoked and pickled mussels. India has exported 2 MT of canned mussels worth Rs. 0.48 million in 1979. More work on processing and preservation of mussel meat is needed.

**Tips for successful culture of Bivalves**

Among the important recent suggestions for development of oyster and mussel culture in India, is the need for a comprehensive picture of sites suitable for oyster farming for all maritime states (James et al., 1993) through understanding of environment-species relationship leads to selection and identification of sites which have physical, chemical and biological properties suitable for higher growth, survival and production of candidates species (Brown and Hartwick, 1988a)

Aquaculture of molluscs, especially bivalves, is one of the earliest forms of mariculture. Being sessile and low tropic level filter feeders, bivalves can be raised at relatively low cost. Among several species of bivalves and a few gastropods, which are cultivated, the important ones are the oysters (Family Ostreidae), mussels (Family Mytilidae and Aviculadae), clams (Family Veneridae), scallops (Family Pectenidae), the abalone (Family Haliotidae) and cockles (Family Arcidae). The
group that accounts for the largest production through aquaculture is
the oysters and several species are cultured in many parts of the world.
Cultivated oysters belong to two genera, Crassostrea (cupped oysters)
and Ostrea (flat oysters). The important species cultured are
Crassostrea gigas (Pacific oyster), C. virginica (American oyster), C.
angulata (Portugese oyster), C. commercialis (Sydney rock oyster), C.
glomerata (Auckland rock oyster), C. plicata and C. rivularis (Chinese
oysters), Ostrea edulis (European oyster) and C. chilensis (Chilean
oyster). The mangrove oyster, C. rhizophorae is cultivated on relatively
smaller scale.

Despite favourable climatic conditions, availability of inexpensive
labour and the need to produce protein food at low cost in tropical
countries, most of the bivalve culture has developed in subtropical and
temperate climatic countries (Quayle, 1980).

Considerations of habitat requirements of aquaculture in coastal
management policies have gained great importance worldwide. Long-
term viability of bivalve culture is particularly dependent upon selection
of suitable sites, with conditions necessary to promote rapid growth,
high survival and production of the candidate species.
Location specific variations in growth and survival of bivalves have been demonstrated for *Mytilus edulis* (Incze et al., 1980), *Mya arnaria* (Appeldoorn, 1983), *Crassostrea virginica* (Mallet and Haley, 1983), *C. gigas* (Brown and Hartwick, 1988a) and *Ostrea edulis* (Utting, 1988). Thorough understanding of environment-species relationship leads to selection of optimal sites, which is an essential requirement for attaining higher production through culture.

Habitat variables potentially critical to culture of many bivalve species have been studied and identified. Bayne and Newell (1983) stated that growth in marine bivalves is principally affected by interaction of several environmental variables particularly water temperature and food availability. Seawater temperature is reported to have major seasonal influence on growth and survival of bivalve spat (Spencer and Gough, 1978). In the field experiments on *virginica* (Butler, 1953), *Macoma balthica* (Gilbert, 1973), *Ostrea puelchana* (Fernandez and Boddoy, 1987) and O. *edulis* (Wilson, 1987), water temperature has been reported to be the most important factor resulting in differences in growth among stations and growth rate in different periods. Loosanoff and Nomejko (1949), Quayle (1952), Askew (1972), Dame (1972) and Malouf and Breese (1977) reported that low temperatures arrested oyster growth and it increased with elevated water temperature.
The present study was to determine the biochemical composition of *Crassostrea madrasensis* and *Perna viridis* occurring at Kali estuary, Karwar. Population of Oysters growing on the seabed occurs quite away from the one station to another. Oysters growing on oyster bed (dead shells) are exposed during low tide whereas in Kodibag station oysters scattered all over and mussels found attached to bed, which are always submerged (grow sub tidally all the time). This would provide an opportunity to examine and compare the pattern of biochemical changes in oysters and mussels from different stations of Kali estuary.

These above said investigations and informations were studied, related to the analysis of the molluscan tissues with references to seasonal changes in biochemical analysis. The main intension of the research is to exploit molluscan resource of the Kali estuary and to generate nutritional food for human population.