In crustaceans, body growth and reproduction is interrelated phenomenon which gaining more attention in view of the likely use of such information in aquaculture. The literature survey revealed the striking differences in designs of growth and reproduction among several closely allied Brachyuran species (Knudson, 1964; Erdman and Blake, 1988) and even among members of the same (Brachyuran) species occupying different latitudes and ecological niche (Krouse, 1980; Anilkumar and Adiyodi, 1983; Conan, 1985)

The survival of each species of animals requires its individual members reproduce the new individuals to replace the one killed by natural death in old age or by predators, parasites, environmental pollution or other ecological hazards such as shortage of water, food, oxygen or unavailability of optimum temperature and light. Therefore, to retain the existence of a species on this earth, reproduction is a more vital life process.

Gonad maturation can be explained in terms of morphological, molecular, cellular, physiological, neurohormonal and behavioral adaptations to its environment. The knowledge gained by elucidating the proximate factors controlling reproduction can provide a better understanding of the evolution of life history and reproductive strategies in different environment. In crustaceans, some external factors such as light, photoperiod, rainfall, pH, salinity as well as internal factors, hormones, play significant role in realization of primary, secondary and behavioral sex characters of the individuals (Chariniaux-cotton, 1960)
Reproductive activity in crabs involves behavioral patterns or physiological processes, which has an impact on the overall ‘performance’ of an individual involved. These processes fall into two primary categories – first, those which result in slower growth or in a reduced ‘scope for growth’ this may be either a reduction in energy intake, or a diversion of available energy to reproductive processes and second, there may be an increased risk of mortality, either due to an increased availability to predators, or by a reduced ability to escape predators. These various reproduction cost are associated with the development of crab.

Reproduction is seasonal cyclic and course of gametogenic cycle includes accumulation of ripe gametes i.e. spawning and an egg laying and reproductively spent or quiescent period. Buttler (1960) investigated maturity and breeding of the pacific edible crab, _Cancer magister_ of Northern British Columbia. Subramonium (1963) and Rahman (1967) studied the reproductive and nutritionals cycles of the crab, _Portunus pelagicus_. Reproductive cycle of crustaceans has been studied by Diwan, 1978; Mirajkar, 1980; Victor, 1980; Nagabhushanam, 1987; Araki, 1997; Khapte, 1995. Vernberg and Vernberg (1972) have reviewed extensively the available data on reproductive cycles of marine invertebrates and complexity of controlling factors. The annual reproductive cycle can be subdivided into pre-reproductive, reproductive, and post-reproductive stages in accordance with sequence of happening of each events and this in turn determines pattern of breeding cycle i.e. either semi-annual, annual, biannual or continuous breeding.
Pyromia tuberculate are found to be continuous breeder collected from Tokyo bay, Japan. Murthy (1998) studied breeding and improved hatchway technology in freshwater prawn, Macrobrachium rosenbergii in Karnataka. Tsuchida and Watanabe (1997) reported that grasped crab, Plaguria dentipes ovigerous females occurred from October to December at Banda bay, Japan. Breeding and improved hatchery technology has been studied by Murthy et al (1998) in freshwater prawn Macrobrachium rosenbergii in Karnataka. According to Pinheiro and Fransozo (2002), the reproductive pattern of crustacean decapods can be divided into three distinct groups. 1) Continuous reproduction would be implied when ovigerous individuals and/or females bearing mature gonads are present year round with similar frequencies throughout the months. 2) If breeding is still recorded throughout the year, but monthly frequency of reproducing females varies with recognizable periods of higher reproductive activity, then seasonal-continuous reproduction would apply. 3) Finally if the presences of ovigerous specimens and or females mature gonads are restricted to a determined period, or a season, the seasonal reproduction may be inferred.

In crustaceans reproductive phase is determined with the help of carapace length and gonadosomatic index, Haley (1972) reported that Ocypoda quadrata obtained sexual maturity at 25-34.9 mm carapace length. Diwan (1978) reported that carapace length is a parameter to ascertain reproduction in Barytelphusa cunicularis. Mirajkar et al (1982) studied reproductive cycle in Macrobrachium
*kistnensis* and reported that carapace length is related to a reproductive phase in prawn i.e. maximum carapace length was attained at maturity. In Natantian prawns, growth and ovarian maturation are synergistic events (Wilder and Aida, 1991). Diesel and Horst (1995) in, *Sesurma jarvisi* observed that the juveniles develop inside the shell until they reach 8 mm carapace width. Carapace length is an important indication of reproduction in male and female *Cytograpsus angulates* too. Santos et al (1996) in, *Mertunus spinimahus* described the size and age of crab are important for determining the reproduction output. The relationship between carapace length chela heights was estimated by Goshima – Sejii (2000). Nakasone-yukio (2001) observed carapace length of ovigerous females of three land hermit crabs.

Size at the onset of physiological maturity is one of the most important parameters of the life cycles of crustaceans to know, and its estimation has been attempted by many different methodologies (Gonadal development, presence of spermatophore) by Conan and Comeau, 1986; Paul, 1992; Sainte-Marie et al 1995). Megumi and Satoru (1997) reported that male and females of spiny lobster, *Panulirus japonicas* attained physiological maturity at 54 mm CL and 45 mm CL. Mantel and Dudgeon (2005) in *Macrobrachium hainanense* observed that gonad maturation in male and female occurred at 12-18 mm CL and 15-17 mm CL respectively. Maria and Leme (2005) reported that female, *Sesarma rectum* attains physiological maturity at 18 CW. Veronica and Lopez-Greco
(2006) in *Aegla uruguayana* stated that gonad maturation, in male and female occurred at 19 mm CL and 17 mm CL respectively. Gonad index is a function of breeding cycle in marine and estuarine crustaceans (Giese, 1955). Giese (1959) formulated a corelationship between gonad maturity and body weight. Same methodology was applied by Farooqui (1980), Reddy (1982), Mirajkar (1985), Sarojini (1986). The index that determines the reproductive status of marine invertebrates is based on the observation of gonad maturation at the morphological and microscopic levels, by doing observations on their form, weight and gonad colour (Lopez-Greco and Rodriguez, 1999). The reproductive period of hermit crabs can be analyzed based on the frequency of ovigerous females during the twelve months of the year (Lancaster, 1998; Imazu and Askara, 1994; Manjon-Cabeza and Garcia-Raso, 1995) and by means of gonadal studies (Lancaster, 1990; Bertini and Fransozo, 2002). Gonadosomatic index value in female *Mictyris brevidactylus* started increasing in August and reached at peak in November and December; when gonadosomatic index dropped, crab entered in their reproductive season (Shih, 1993). Koga (1995) in *Scorpimera globasa* observed that the gonadal index decrease with time advancement in early reproductive season. In *Palinurid lobsters*, the gonadosomatic index or appearance of gonad such as colour, size and shape have been adopted as index of maturation (Grey, 1979; Plaut, 1993). In freshwater prawn, *Macrobrachium rosenbergii* the gonadosomatic index increased during vitellogenesis (Wilder et al 1991). Ovary
indices and percentage of ovigerous females was studied in *Uca lactra* by Yamaguchi (2001).

Oogenesis is an energy consuming process that can be divided into several phases. Pillai (1960) suggested that breeding cycle is determined by carapace length, gonad index and histological data. Ryan (1967) and Cheung (1968) has been studied the oogenesis, spermatogenesis and histology or reproductive system of *Portonus sanguinolentus*. The process of oogenesis has been studied in marsh crab, *Paratelphusa hydrodromous* by Adiyodi (1969). Lee and Lee (1970) reported the ovarian cycle and oogenesis of *Penaeus japonics*. Differentiation of oocytes and vitellogenesis progresses reported in *Uca pugilator, Cambarus clarkia* and *Libinia emerginata* (Wolin et al 1973). The simultaneous observations on carapace length, gonad index and histological data leads to conclusion about breeding cycle (Jyoti, 1974). The phases of oogenesis is characterized by the accumulation of yolk proteins in the growing oocytes and by significant increase in oocyte diameter, are referred as primary and secondary vitellogenesis (Meusy and Charniaux-Cotton, 1984; Meusy and Payen, 1988). Jayalakshmi (1986) made some observations on cyclic changes in ovary of *Macrobrachium lamerrui*. Amato and Payen (1978) studied the spermatogenesis in *Pontastacus leptodactylus*. Joshi and Khanna (1986) reported oogenesis and spermatogenesis in freshwater crab, *Potamon koolooense*. In freshwater crab, *Gecarcinus steniops*, the oogenesis and folliculogenesis were studied by Santhama and Adiyodi (1991). Trinadha Babu et al
Ronquillo et al (2006) observed early embryonic and larval stages of green tiger prawn, *Penaeus semisulcatus.* Sexual dimorphism is the existence of physical difference between the sexes, other than difference in the sex organs. Darwin contended that sexual dimorphism evolved by means of sexual selection. Morphometric maturity is related to change in the relative growth of secondary sexual characters (relative to a somatic character of reference, such as carapace length) at the onset of sexual maturity. It has been widely used in population biology and fisheries (Hartnoll, 1974; Mingawa and Huguchi, 1997; Amaro pinheiro and Fransozo, 1998; Goshima et al 2000; Conan et al 2001; Flores et al 2002).

Crustaceans are particularly suited for relative growth due to their hard skeleton that facilitate precise body measurements. The type of growth allows unequal division of their ontogeny into distinct phases and exhibits great difference between the growth rate of males and females as well as in juveniles and adults (Hartnoll, 1978). For brachyuran crustaceans, changes are conspicuous in the male cheliped, female abdomen and pleopod of both sexes during the transition from juvenile to the adult stage (Castiglioni and Negreiros-Fransozo, 2004b).

Crustaceans generally show sexual dimorphism in their external morphology, particularly in relation to positioning of opening of gonoduct. Sexual difference observed in the growth of several body parts relative to carapace size have often been used to examine the relationship between morphometrics and sexual
activity, in addition to morphometric difference among populations or species (Kanno, 1972; Huber, 1985; Aiken and Waddy, 1989) Determining the size of sexual maturity of fish and crabs has been a constant by researchers focusing, principally, the management of fishery resources. However, sexual dimorphism is useful also for purpose of conservation of biodiversity by characterizing population aspects concerned with abundance, since that amount of mature individuals in a given population is responsible by the future generation. That kind of study gives substitute for the management of natural stock of animal populations which also must be monitored for ends of habitat conservation. Size and form are closely related to feeding, and habitat occupation, among others ecological features, influencing the fitness of an organism, being the result of evolutionary processes (Peresneto, 1995). Allometric growth occurs when some parts of animal’s body grow at a different rate in relation to a reference dimension, generating changes in body proportions. Some changes are abrupt, marking crucial ontogenic stages, such as prepuberal or pubertal molting (Hartnoll, 1978 and 1982). The allometric relationship among various parts of the body is functionally important in the biology of the organism and particularly it serves as a predictive tool for ecological investigations.

In female brachyuran crabs, a change in allometric growth of abdomen, which plays a role in covering, protecting and eggs-carrying, occurs at start of sexual maturity (Hartnoll, 1974). This change is prominent in some species and is termed as puberty molt,
but it does not always coincide with gonadal maturation (Hartnoll, 1982). Maria (2005) in *Sesarma rectum* studied growth pattern of abdomen and reported that it grew faster and larger at the onset of puberty, and all morphological changes of abdomen are associated with gonadal development. In crustaceans, sex can be readily distinguished by size of leaf like exopods of pleopods in adults. Spawned eggs are attached to the endpods of the pleopods and are protected by exopods; therefore description of relative growth of pleopods has been examined in decapods species. Mingawa (1997) studied growth of pleopods in *P. japonicas* and reported that pleopods in males (>28 mm CL) and females (>43 mm CL) grew at different rate.

Brachyuran males and some other decapod species develop the pereiopods especially the chelipeds, for combat, display and courtship (Hartnoll, 1974, 1982). Relative growth pattern of pereiopods change at a certain carapace size and after that pereiopod size generally grow larger in male than in female of the same size. This change has been considered to occur in relation to sexual maturity (Hartnoll, 1982). Mariappan (2004) studied developments of individual segments of pereiopods in *Macrobrachium nobilii* and reported that ratio between cheliped length and carapace length is species specific. Recently, Suzuki and Kasamura (1997) differentiated two prawn species on the basis of length and width of dactyl of second pereiopod. Kubo (1988) studied the sexual difference in pereiopods of *P. japonics* and reported that males tended to have longer pereiopods than females.
of the same carapace size. In *Anomuran* families, relative growth has been studied by many workers in the *Diogenide* and *Paguridae* (Bertini and Fransozo, 1999; Mantelatto and Martinelli, 2001; Among others). Hence study of these changes may provide insight into evolutionary (sexual selection), ecological (life history strategy) and biological (reproductive behavior and strategy) constraints and patterns in the population size and structure of species, *Barytelphusa cunicularis*. The study of above parameters is utilized to differentiate the mature and immature population in many decapods, which are not sexually dimorphic (Pinheiro and Fransozo, 1998; Munio et al 1999).

In crustacean, the allometric relationship between body size and various organs have been used to estimate the size at maturity, assuming that secondary sexual traits appear and grow at different rates in the immature and mature phases. The transitional phase in brachyurans involves morphological and morphometric changes which can be detected by inflection or discontinuities in the series of linear or curvilinear relationship using bivariate analysis (Haefner, 1990). Several methods have been employed to study allometric relationship between body’s parts, such as the MATURE proposed by Somerton (1980) or von Bertalanffy’s equations. These later methods have been criticized (Day and Taylor 1997) because they employ personal choice of the determinant component in analysis and graphical perception. Cleveland and McGill (1985) discussed the matter of judging the
angle rather than slopes by personal choice, which leads to distortions in judgments of line segments (Maria H., 2005)
The study of fecundity of any species is important to have a full understanding of its reproductive biology and population dynamics. Fecundity can be defined as the number of ova shed during a particular spawning season (Pillay, 1964) or number of ripening egg in the female brood pouch prior to the next spawning season (Bagenal, 1968) and fecundity determines the reproductive potential of species and stock size of its population (Emerson, 1999). Fecundity studies may provide important information on the rate of replacement in natural population. This information allows a better understanding of reproductive strategies, dynamics and evolution of given population (Garcia-Montes et al 1987).

Fecundity of crabs varies from species to species and also varies within same species due to various factors such as age, size, nourishment, ecological conditions of the water body etc. Variation in the fecundity was primarily a reflection of variation in the size of crab at maturity. Henmi capital (1989), a lack of relationship between body size and fecundity may have several causes, such as individual variation in egg production, seasonal food availability, and multiple spawning, in addition to natural egg loss. The season or batch number could influence egg size and the bigger eggs have a better chance of survival (Hadden, 1994). Koga et al (2000) analyzed Ocypodidae fecundity and stated that the production of big progenies could not be continuous, while females with small progenies reproduce continuously.
Brachyurans show a great diversity of embryogenic development, especially owing to a significant variation in egg size (Hines, 1982). In crab fecundity is traditionally measured as the number of eggs produced in each clutch and it is described as function of body size (Corey and Reid, 1991). Shield (1991) reported that some species of cancer produce many small eggs (higher fecundity) that quickly develop into small larvae and some species produce relatively larger eggs (lower fecundity) that slowly develop into large larvae. The peripheral layers of eggs may be vulnerable to damage from various sources i.e. pathogenic and parasitic agents (Shields, 1991), and even active removal during grooming or under stressful conditions (Horman and Jones, 1993) the impact of egg mortality during incubation period may acts as a causal factor, thus contributing to the dynamics of crabs population cycles. Shield and Wood (1993) assessed the impact of parasites on the reproductive biology and fecundity of the blue sand crab, *Portunus pelagicus* from Australia. Comeau et al (1999) examined the reproductive cycle and fecundity of multiparous female snow crabs. Poulin (2002) observed that the number of eggs produced by females was strongly correlated with carapace width in three species of New Zealand shore crabs, *Macrophthalmus hirtipes*, *Hemigrapsus edwardsii* and *H. crenulatus*. In freshwater crabs, female incubate their eggs, which remain attached to the pleopods, from spawning to hatching. The incubation varies with species and temperature (Guerrcro and Hendrickx, 2004). Tania Costa (2006) defined reproductive pattern similarities and differences of most

Reproductive output per brood is strongly correlated with body size among brachyuran crabs. Successful management of fishery resources is dependent on understanding the exploited species reproductive potential, which in turn requires spatially relevant estimates of the size at maturity and the size fecundity relationship (Shelly, 2007). According to Hines capital (1982) and Brachyuran crabs usually carry their eggs attached to the pleopodal setae, forming partially exposed mass of eggs, as occur with *Stenorhynchus seticornis* (Cabo, 2005). Arshad A. (2006) in blue swimming crab, *P. pelagicus* stated that number of eggs increases linearly with increasing of carapace length, carapace width, body weight and weight of oviposition.

Crabs are commercially important as there is a rapidly expanding demand for crab meat both in local and international markets also there is increase in their sale value. Of several crab species, the
freshwater crab *Barytelphusa cunicularis* has high potential to meet this demand due to its excellent meat quality and rapid growth during culture. Crab culture in the India is an important source of income for small-scale fish farmers in this region and is currently viewed as a viable alternative to fish farming in underutilized and abandoned fish ponds. A major barrier to further develop mud crab aquaculture is an insufficient supply of seed. Over-exploitation, pollution, and Climate change have contributed to this loss. Although spawning of captive crabs are reported to occur year-round, larval survival is very low and inconsistent. Thus, the importance of developing reliable techniques for broodstock and seed production of freshwater crab to answer the problem of lack of seedstock has been recognized. Nutrition of broodstock of fish and crustacean plays a major role in achieving reproductive success and has a considerable influence on gonadal maturation, fecundity, egg hatchability, and larval viability. A freshwater crab, *Barytelphusa cunicularis* has short larval duration and survivorship. Nevertheless, a large number of juveniles must be produced at low cost, allowing aquaculture raised animals to compete with wild caught animals in the trade market, which can be an alternative to minimize the effect of wild harvest of crabs and other animals in fresh water ecosystem.

Many physiological and behavioral processes in crustaceans are regulated by neurohormones. In decapods the nervous system and sense organ were studied by George (1961). The crustacean eyestalk contain group of neurosecretory cells forming x-organ and
sinus gland, which act as a neurohaemal organ for storage and release of hormones (Weatherby, 1981). The sinus gland receives axons from different parts of the central nervous system (Bliss and Welsh, 1952), and serve as site for release of hormones. The relationship between types of neurosecretory cells and reproductive activity has been reported in crustaceans. Matsumoto (1958) while working on the five different species of crabs, observed the secretion of A, A₁ and E cells showed a close relation with reproductive cycle. These cells were found active during the breeding season. The number and neurosecretory cell activity in male crab *Uca lactea annulipes* responsible for increase in the size and the development of the reproductive and associated structures with season.

In crustaceans, neurosecretory cells have been well studied (*Caridina weberi, Metapenaeus monocers, Palaemon serratus, Macrobrachium kistnensis* etc). The presence of NSCs in the various part of the CNS of crustacean has been reported by many workers. These cells are localized in more or less discrete groups. They have been classified into distinct groups on the basis of their size and structures, thus in crayfish, four types of cell have been reported in the brain and eyestalks whereas twelve types of these cell in the CNS of crabs. In *Scylla serrata* (Deshmukh and Rangnekar, 1965) and in *Paratelphusa jacquemontii* five types of cells are identified in the CNS.

In crustaceans many functions like molting, growth, metabolism and gonad maturation have been attributed to the crustacean
eyestalk hormones. Eyestalk ablation in crustacean brings about changes in their physiological functions such as growth (Kamaguchi, 1971; Nakatani and Otsu, 1980) and reproduction (Adiyogi, 1970; Hisch and Benett, 1979). Eyestalk ablation brings about rapid maturation of the ovary in some cases and in others previous vitellogenesis does not take place. Kamaguchi (1971) investigated the effect of eyestalk ablation in prawn, *Palemon paucidens* at various stages of ovary. Mirajkar et al (1980) proved endocrine control over reproduction in *Macrobrachium kistnensis* by performing eyestalk ablation. Wear and Santiago (1977) induced the maturation of ovary and spawning in, *Penaeus monodon* by unilateral eyestalk ablation. The eyestalk of female decapods crustacean inhibit vitellogenesis in the ovaries by producing and secreting ovary inhibiting hormone, however, experiment involving eyestalk removal have reveled that in some phases reproductive cycle, this operation fails to accelerate vitellogenesis (Aoto and Nishida, 1956; Adiyodi and Adiyodi, 1988)

The presence of testis inhibiting factors in the eyestalk of male was shown in crabs. In these animal’s eyestalk ablation resulted in the increase of the weight of testis and the diameter of vas deference. However, in large number of crustaceans, the effect of eyestalk ablation is manifested by moulting alone and not by gonadal development (Bauchau, 1961). Diwan (1972) in male *Barytelphusa cunicularis* proposed that eyestalk ablation results in gonadal developments. In *Scyll serrata*, Rangnekar (1971) proved that
eyestalk ablation in the young crab resulted in an increase in the weight of testis accompanied by an increase in the diameter of seminiferous tubules, vas deference and number of cells per seminiferous tubules. Young (1974) investigated the effects of eyestalk ablation on the spermatogenesis in *Uca pugnax*. The effect of eyestalks removal is not uniform in studied species of crab; various factors such as age, season and perhaps sex seem to influence the course of events. It has been confirmed by diversified experiments, such as organ ablation and transplantation, hormone purification and gene clone, that x organ–sinus gland complex is synthesis center of GIH, and secretion of C₃ cells in x-organ reduces as ovary develops, and reaches the minimum when ovary enter the development stage and near-mature stage (Fingerman, 1997; Huberman, 2000).

Adiyodi and Adiyodi (1974) reviewed that the role of the hormones of eyestalk, brain and thoracic ganglion in the crustacean reproduction. The effect of brain and thoracic ganglion in the oogenesis has been worked out by Hinsch and Benett (1979) in *Libinia emerginita*. The hormonal control of spermatogenesis was studied in *Lysmata seticaulata* (Touir and Grasse, 1977) in *Armadillium vulgare* (Juchauet and Legrand, 1978) and *Asellus aquaticus* (Demessieux and Balesdent, 1979). Ovarian development of female crustacean is under the direct regulation of two neurohormones gonad- stimulating hormone (GSH) secreted by brain and thoracic ganglion and gonad inhibiting hormones (GIH) secreted optic ganglion (Fingerman, 1997; Huberman,
2000). In-vitro studies on, *Uca pugilator, Procumbarus clarkia and Scylla serrata* have clarified that GIH and GSH have direct influence on the ovary i.e. ovary is target organ of GIH and GSH (Eastman Recks and Fingerman, 1984; Kulkarni et al 1991; Jin et al 2003). Ye Huihui and Huang haiyang in *Scylla serrata* confirmed that brain and thoracic ganglion regulate testicular development through AG in male crustacean. Eyestalk removal also result in proportionality larger increase in size with each ecdysis in adults (Carlisle and Knowles, 1959; Passano, 1960; Bliss et al 1966) Paul and Robert (1999) stated that animal which underwent bilateral eyestalk ablation earlier in larval life grew to larger overall size to unablated controls.

The gonadal maturity of freshwater crab, *Barytelphusa cunicularis* was studied and its correlation with sexual dimorphism was carried out because less information is available on sexual dimorphism of freshwater crabs and moreover study of individual segments of chelipeds is very rare, so the present work was carried out to report the exact carapace length at which sexual dimorphism occurs.

On fecundity of freshwater crab, *Barytelphusa cunicularis* very little work has been done and besides no information is available on its relation with secondary sexual characters; hence the present work was carried out.

Hormonal control of gonad maturation in fresh water crabs is not studied and so far nobody has studied effect of neuroendocrine hormones on growth of secondary sexual organs, hence it was
decided to find out effect of eyestalk ablation and administration of brain, thoracic ganglion and gonadal hormones on development of gonads and of secondary sexual organs in crab, *Barytelphusa cunicularis.*