

SPERMATOGENESIS IN MARINE CRAB, *PORTUNUS SANGUIOLENTUS*

Zoology

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ABSTRACT

India is well known for its megadiversity of biological wealth harbouring over 12% of shell and fin fishes known. Aquatic biodiversity has always been playing an important role in the food and nutritional security of people especially in the rural areas. Dahanu is the rural area along the coast of Arabian Sea. Decapod crustaceans such as crab, prawn, lobster are largely exploited in this area. Rapid urban and industrial development results in deforestation, industrial and domestic water pollution and heavy use of pesticides in agriculture declines crustaceans in this habitat. In crustaceans, the carapace length, fecundity is parameters to ascertain their reproductive maturity. Study of gonad is essential for the determination of breeding season. The present study was undertaken for the histological study of testis of the marine crab, *Portunus sanguinolentus*. Histologically, the testis of *Portunus sanguinolentus* showed small seminiferous tubules with various stages of spermatogenesis. Nutritive cells are present in between the seminiferous tubules. The process of spermatogenesis includes five distinct stages such as spermatogonia, spermatocytes, spermatids, spermatozoa and spermatophore formation.

KEYWORDS

Spermatogenesis, Testis, Crab, *Portunus sanguinolentus*

Introduction:

Araki and Matsura (1995) observed the relation between growth and gonadal maturation in *Geotelphusa dehaani*. Histological observations of gonads also supports to determine reproductive cycle in crustaceans. Cronin (1947) gave a comprehensive study of anatomy and histology of the reproductive system of the male crab, *Callinectes sapidus*. Many earlier workers did a note worthy work in crustaceans (Parmeswaran, 1953; Suco, 1954; Nishioka, 1959; Ryan, 1967; Cheung, 1968; Lee and Lee, 1970; Jyoti, 1974). Deshmukh (1968) observed the morphology and histology of the gonads in marine crab, *Scylla serrata*. Minagawa et.al (1994) reported the histological study of the reproductive system and breeding cycle of male red frog crab, *Ranina ranina*. A systematic description of the morphology and histology structures of the reproductive system of *Eriocheir sinensis* has been given by Hu-Ziqiang and Hu-Yunjin (1997). Araki et al. (1997 a, b) studied the spermatogenesis of freshwater crab, *Geotelphusa dehaani*. Goshima-Seiji et. al. (2000) made histological examination of gonads in stone crab, *Hepalogaster dentata*. The survey of literature revealed that there is little information available on spermatogenesis. Hence the present study was undertaken to ascertain the information of on above aspects.

Material and methods: The marine crab, *Portunus sanguinolentus* was selected for the present study. The histological changes in gonads were studied to assess the reproductive cycle of *Portunus sanguinolentus*. To confirm the reproductive phase and detailed changes followed in the testis, the histological observations were done for the gonad. This method gives an idea about initiation of spermatogenesis, size of spermatocytes, frequency distribution of the spermatocytes and other details of spermatogenesis. For this purpose the testes were fixed in Bouin's fluid. After 24 hours of fixation the tissue were dehydrated in alcohol grade series and were paraffin embedded (58-60°C). The sections were cut at 6-7 and stained with Harri's Haematoxyline eosin.

Results: *Portunus sanguinolentus* is a dark grey coloured marine crab. The carapace is distinctly marked with three large blood red coloured spots. The anterolateral border of the carapace has nine teeth and the posterior most tooth is largest. Spines are absent on the posterior border of the merus of the chelipedes. The male reproductive system of *Portunus sanguinolentus* is bilaterally symmetrical and located in the anterolaterally. It consists of testes, vas deferens and ejaculatory ducts. Each testis is whitish and made up of intermingled tubules located dorsally on the hepatopancreas. The testes are tubular organs formed by lobules. The process of spermatogenesis takes place in the testis. Testis of *Portunus sanguinolentus* showed many small seminiferous tubules with various stages of spermatogenesis. The lumen of each seminiferous tubule was continuous with vas deferens. The nutritive cells are basal in position and have centrally placed nucleus. In *Portunus sanguinolentus* the process of spermatogenesis was completed in distinct five phases such as spermatogonia, spermatocytes, spermatids, spermatozoa and spermatophore

formation. Spermatogonia are differentiated from wall of the seminiferous tubules. They were clearly distinguished by their large nucleus and great amount of cytoplasm. These spermatogonia, undergo two mitotic divisions and transformed into spermatocytes. Spermatocytes have large nuclei and great amount of cytoplasm. The most distinct change that take place during the growth of the spermatocytes was an increase in volume and size of the nucleus. The nutritive cells supply nourishment to the growing spermatocytes. (Plate I, Fig. 1). Spermatids are differentiated spermatocytes by reorganization of the nucleus. Spermatids were spherical in shape and stained darkly with haematoxylin. Spermatids are transformed into spermatozoa. (Plate I, Fig.2). Spermatozoa are formed by the process of cell differentiation. Seminiferous tubules completely filled with spermatozoa after completion of spermatogenesis.

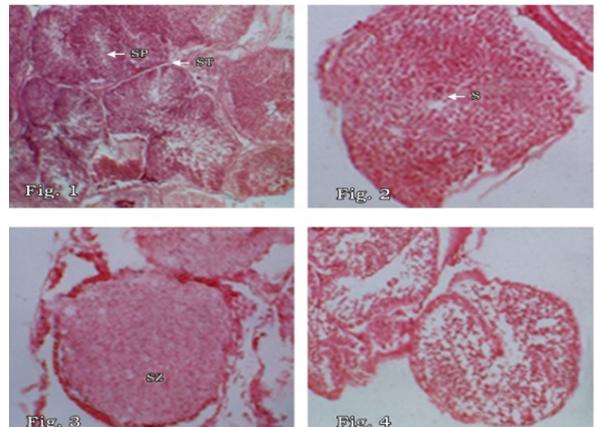
Plate – I Histological section of testis of crab *Portunus sanguinolentus*

Fig. 1 Transverse section of testis of crab, *Portunus sanguinolentus* showing primary spermatocytes. 400X.

Fig. 2 Transverse section of testis of crab, *Portunus sanguinolentus* showing spermatids. 400X.

Fig. 3 Transverse section of testis of crab, *Portunus sanguinolentus* showing spermatozoa. 800X

Fig. 4 Transverse section of spent testis of crab, *Portunus sanguinolentus* showing degenerating seminiferous tubules. 800X

In *P. sanguinolentus* the spermatozoa are a funnel like or conical cup shaped. The cup region is named as the nuclear cup. (Plate I, Fig. 3). In *P. sanguinolentus*, the spermatophore formation took place in the vas

deferens. Spermatophores were grouped together in vas deferens showed various shapes i.e triangular oval or round. Spermatophores were transferred to the vas deferens, where they were stored till their ejaculation. After transfer of spermatophores to vas deferens, the seminiferous tubules look vacuolated.

Discussion: In crustaceans different stages of testes maturation are classified on the basis of colouration. In male *Portunus sanguinolentus* there is no distinct colour change in the testis however immature testes were transparent and mature testes were milky white. These colour changes clearly observed during reproductive phases of *P. sanguinolentus*. The secretory cells helped in movement of spermatozoa. In crustaceans, the process of spermatogenesis has been studied in *Partelphusa masonoiana* (Vasishth and Recten, 1971); *P. hardwickii* and *P. stylifera* (Joshi, 1980). The pattern of spermatogenesis is more or less same in almost all the crustaceans. In *P. sanguinolentus* initiated by proliferation of the spermatogonia from the testis follicles or acini. Later the stages like spermatocytes, spermatids and spermatozoa were found. Formation of spermatophores took place in the vas deferens of *P. sanguinolentus*. Similar fact is also observed in *Libinia emarginata* (Hinsch and Cone, 1969). In freshwater crab, *Geotelphusa dehaani*, spermatogenesis was divided into six phases i.e. the primary spermatocyte, the secondary spermatocyte, the spermatid and spermatozoa (Araki Matsuura, 1997 b). The morphology of spermatozoa in crustaceans varies with groups and show remarkable differentiation compared to the usual organization of the gametes. However, the basic pattern of sperm formation is similar. In crabs, sperms are aflagellate and three to six pseudopodial rays arise from their head. The sperm head is represented by a basophilic nucleus and acrosome while the tail piece is made up of eosinophilic mitochondrial vesicle, basophilic centrosome and axial filament (Ryan, 1967; Langret, 1969). The sperms are rod like with club shaped acrosome in anomomuran crab *Emerita asiatica* (Subramoniam, 1977). Joshi (1980) observed that the sperms had a round head with a short tail, while Farooqui (1980) described the funnel or conical cup shaped spermatozoa in marine crab *Scylla serrata*. In crustaceans there are variations in the morphogenic appearance, location, formation and transfer of spermatophores. In *Portunus sanguinolentus* spermatophore formation observed in vas deferens. Hinsch and Cone (1969) in *Libinia emarginata* have shown that the sperms are transferred from the testis to the vas deferens where spermatophore formation takes place, the spermatophore are lodged there until their ejaculation. In *Parapenaeopsis stylifera* and *Parapenaeopsis hardwickii* (Joshi 1980) reported spermatophore formation inside the testis. In the anomomuran crab *C. asiatica*, the spermatophores are arranged in a row forming a ribbon like structures and inside each spermatophore sperms are packed irregularly (Subramoniam, 1977). **Conclusion:** The male reproductive system of marine crab, *Portunus sanguinolentus* consists of testes, vas deferens and ejaculatory duct. Testes are transparent or milky white. Testes showed small seminiferous tubules with various stages of spermatogenesis. The lumen of each seminiferous tubule was continuous with vas deferens. The nutritive cells are basal in portion and have centrally placed nucleus. In *P. sanguinolentus*, the process of spermatogenesis is completed in distinct five phases such as spermatogonia, spermatocyte, spermatid, spermatozoa and spermatophore formation.

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