REPRODUCTIVE CYCLE OF MALE SIGANUS RIVULATUS FORSK. WITH INDICATION TO GONOSOMATIC AND HEPATOSOMATIC INDICES.

E.M. AMIN Institute of Oceanography and Fisheries, Alexandria, Egypt.

ABSTRACT

Studied samples of Siganus rivulatus were obtained from the commercial catch of Red Sea (Jeddah, saudi Arabia). The testicular development of male S. rivulatus started early in this subtropical area and firmly affected by the significant elevation of water temperature. Histologically examined testes showed an active spermatogenses in February, and spawning season extended from March to September. Spent males still possessed remnants of not shedded spermatozoa distributed in the empty testicular tubules and sperm duct. A gradual resorption of such being left spermatozoa took place as a result of phagocytic activities. Determined values of GSI gave a further confirmation to the time of spawning. The increase was towards the more developed testes, whence the beginning of spawning. Values of HSI were extremely decreased as the testes became matured, and significantly increased after spawning and before the onset of a new sexual cycle.

INTRODUCTION

Siganids are tropical and subtropical fishes and represented in the Red Sea (Jeddah region) by five species (Amin and Hussein, 1985). Recently, the induction of Siganids and its application in fish culture has been reviewed (Soh and Lam, 1973; Popper and Gundermann, 1975, Pitt et al, 1979).

In recent years, the aquaculture of Siganus rivulatus in cages has greatly succeeded in Jeddah region, and gives from 200 to 300 gms after 9 months feeding in cages (Thebaity et al, 1984).

The purpose of our study was therefore to examine the growth and testicular development in male **S. rivulatus** to evaluate its suitability for commercial aquaculture.

MATERIAL and METHODS

A total number of 500 specimens were collected monthly from the commercial catch in Red Sea (Jeddah region) in the period from September 1980 to August 1981. The total body length and weight, gutted weight,

testes length, testes and liver weights, and sexual maturation of male S. rivulatus were recorded monthly. Gonadosomatic index (GSI) was computed by using the formula

GSI = gonad weight / gutted body weight X 100

and Haepatosomatic index from the formula

HSI = liver weigh / gutted body weight X 100

Fish age was determined by Peterson's method (1895), testicular maturation was classified into stages according to Hjort (1910). The testes were fixed in Bouin's solution, histological mounts examined and stained with Heidenhain's iron Haematoxylin and eosin.

RESULTS

1- Age and Size at Maturity Stages

.....

Monthly distributions of different stages of testicular maturation of male S. rivulatus were represented in Fig. 1. Fishes with testes in stage I-II were found all the year round with total fish length varying from 12.0 to 16.0 cm corresponding to 2 and 3 years of life, respectively. Males with testes in stage III of maturation entered the catch in December and still represented to May. The average length of right and left testes of stage III varied from 4.2 to 5.5 cm, and total fish length 15.0 to 19.0 cm, aging 3 to 4 years of life, respectively. Nearly ripe males with testes attaining stage IV, were obtained in the catch of January constituting 9.09 % and of June with 6.25 % of the total catch. Fishes with testes in stage IV of maturation varied in total length from 17.0 to 23.0 cm, testes had been greatly increased in size, the left one (average length 7.0 cm) was longer than the right (5.4 cm), the average ages at this maturity stage was 4.5.

Ripe fishes possessed gonads in stage V of maturation filling all the body cavity, they have been found in the catch of February constituting 42.86 % and extended to September where they formed 22.22 % of the total catch. The minimum recorded length of ripe male was 14.5 cm and the maximum length measured 31.0 cm. The average fish length at ripe maturation was 24.0 cm corresponding to a fish of 5 years old. This revealed that male S. rivulatus attained its first maturation after the completion of the 3rd year of its life. On attaining 5 years old, the fish was considered as spawned to the third time. Spent males (stage VI) were firstly noticed in March constituting 21.05 % and still found up to November with 50.0 % of the total catch. Maximum percentage (90.0) of spent males was noted in August. This reveals that the spawning season of S. rivulatus in the southern part of Red Sea starts in March and extends to September, the peak of spawning takes place in August.



Fig. 1. Monthly distribution of different stages of testicular maturation of male S. rivulatus.

2- Gonadosomatic Index GSI

Data representing the average values of GSI were plotted against months of collection (Fig. 2). A noticeable increase was shown in such values from January to September . In April and August a characteristic drop in GSI values, counted 2.32 and 0.18 respectively. A remarkable decrease was also noticed during October, November and December (0.17, 0.04 and 0.43 respectively). Three high values, on the other hand were determined for males obtained in February (8.87), May (8.08) and July (6.13). This may be due to the presence of males with ripe gonads which affected the calculation of GSI. The mean calculated value of GSI was 3.80 ± 0.35 ; in spring and summer (March-September) the mean GSI was 4.74 ± 0.41 , whereas in autumn and winter (October-February) it was 2.52 ± 0.14 .



Fig. 2. Monthly fluctuations of gonadosomatic index and hepatosomatic index with relation to water temperature.

ï

3- Hepatosomatic Index HSI

The average calculated values of HSI were plotted against months of collection of male S. rivulatus (Fig. 2). Significant variations in such values were obvious in different months. The calculated values of HSI were shown to increase with increasing fish length as a result of consequent increase of liver weight. The average value of HSI was 1.71 \pm 0.05. In spring and summer the mean HSI was 1.0 \pm 0.04, in autumn and winter it was 1.92 \pm 0.15.

Maximum determined average values of HSI were registered in December 2.79, then a significant decrease occured up to August and September. From October, a noticeable increase occured to 1.71; in November, male have increased their HSI to 1.99.

4- Spermatogenesis

The testis of **S** rivulatus is a small elongated, paired organ, joined posteriorly to form a Y-shaped structure, and lies ventrolateraly to the air bladder. It is triangular in shape with acute angles, the base pointing dorsally. In immature and active conditions, it is composed of seminiferous regions in radial type from the periphery to the centre and is penetrated by connective tissues as shown in Fig. 3. The testis is covered with two thin layers of testicular membranes, the external layer coats the testis and is made up of small elongated cells with central nuclei. The inner layer is formed of fibrous connective tissue, spindle-shaped and forms the greatest part of the thickness of tunica propria (Fig. 4).

In seminiferous regions, active spermatogenesis which appears ansynchronous could be observed. In the periphery, the mitotic divisions of germ cells were taking place. These divisions developed synchronously to form spermatogonia in their first stage of maturation near the margin of testis, each spermatogonium was spherical, surrounded by a thin layer of connective tissue. On further growth (stage II of maturation), the cysts of spermatogonia were formed and contained large number of small spermatogonia, the cysts increased in size to form spermatogonial clumps (Fig. 5).

On reaching stage III of maturation, small sacs were formed as a result of the extension of tissues (Fig. 6). The formed spermatogonia developed to primary spermatocytes then to secondary spermatocytes by reducetion division, and abundant numbers of spermatids were then developed (Fig. 7).

On further maturation, the main sperm duct gave rise to smaller ducts (vasa efferentia) which penetrated ventrally, and laterally to form tubules. As the sacs became elongated, these tubules formed an extensive system of seminiferous tubules which could be followed almost to the periphery of the testis, these tubules were shown to be filled with the spermatids (Fig. 8).



Fig. 3. A transverse section in the testis showing its triangular shape, S: seminiferous region; C.T. : connective tissue. $(X \ 100)$.



7

Fig. 4. Tunica propria made up of two layers, E.L.: external layer; I.L. internal layer. (X 600).



Fig. 5. The miotic division and development of germ cells, SG.: spermatogonium; C: cyst cell; SG.C: spermatogonial clump. (X 400).



Fig. 6. Small sacs developed in stage III of testicular maturation. (X 100).



7

Fig. 7. Fine structure of the testis in stage III showing the active spermatogenesis, P.SC: primary spermatocysts; S.SC.: secondary spermatocysts; ST.: spermatids. (X 400).



Fig. 8. Seminiferous tubules, ST: spermatid. (X 1200).

In stage IV of maturation the spermatids were developed to give spermatozoa in a process of spermiogenesis. "The Sertoli cells which are supporting cells prominent in the basement membrane of the seminiferous tubules, believed to play a nutritive role during spermiogenesis" (Lofts 1966). As a result of spermiogenesis the metamorphosed spermatozoa filled the cavities of tubules, and the basement membrane of these tubules became very thin. In a more advanced maturing testis, the lumina was shown to have considerable amount of spermatozoa, packed together at the blind ends of tubules near the periphery, the formed spermatozoa possessed intensively stained heads and visible thread-like tails, groups of heads were directed to one direction and tails to the other forming parachute-like structures (Fig. 9), large amounts of formed spermatozoa were distributed in the sperm duct (Fig. 10). The sexual cells became at about the same level of development (Stage V Fig. 11), and a testicular fluid is secreted to facilitate the process of spermiation.

5- Resorption of Spermatozoa

In October, the obtained samples of male S. rivulatus appeared with extremely reduced shrinkaged and kidney shaped testes (Fig. 12). The tubules contained few amounts of remnant spermatozoa, besides, few numbers of comparatively large spermatogonial cells were distributed in the tubular periphery (Fig. 13). In November, there was an obvious loss of spermatozoa in both tubules and sperm ducts. All tubules were devoid of any spermatozoa (Fig. 14). The epithelium with a sort of granules contained some few cellular debris, "granules seemed to be originated from the epithelial cells, but their significance is not known" (Billard and Takashima, 1983), the remnant of spermatozoa as well as the degenerated and residual bodies could be detected. Distributed spermatozoa in the lumen of tubules were passing different degrees of resorption (Fig. 15).

The epithelium lining the lumen of sperm duct showed a high phagocytic activity indicated by the appearance of many granules surrounded by cellular debris and a wide distribution of blood vessels, epithelial cells appeared in the form of squamous or columnar cells (Fig. 16).

The empty tubules became filled with fluid which appeared in a faint yellowish colour when stained with H.E. Accumulations of cellular debris and degenerated spermatozoa as a result of phagocytic activity could be detected in the tubules (Fig. 17). Fishes with such testes shedded out the yellowish fluid through the genital opening, whence the fish belly was stripped.



Fig. 9. The spermatozoa forming a parachute-like structure, SZ : spermatozoa. (X 1200).



Fig. 10. Spermatozoa in sperm duct. (X 400).



Fig. 11. Testis in a ripe condition. (X 1200).



Fig. 12. A transverse section of the testis (kidney shape) in a spent condition. (X 100).

а



3

۲

ç

Fig. 13. Fine structure of the tubule in the testis obtained in October, SZ: spermatozoa; SG: spermatogonium. (X 1200).



Fig. 14. Empty tubules in the testis obtained in November. (X 600).



Fig. 15. Fine structure of the tubule containing variou residual bodies, BM: basement membrane; C.D. cellular debris; G.: granules; SZ: spermatozoa in resorption stage. (X 1200).



Fig. 16. Epithelial cells of sperm duct of male S. revulatus in October, various residual bodies with blood vessel (B.V.); C.E.: columnar epithelial cell; S.E.: equamous epithelial cell. (X 1200).



Fig. 17. The accumulation of cellular debris and degeneration of spermatozoa in the testis. (X 400).

DISCUSSION

Siganus rivulatus has a rapid rate of growth in the Red Sea. The wide distribution of green algae which constitute the main food of Siganids ensures a sufficient and permanent food base for such fishes. The high rate of growth virtually affects the sexual maturation, besides the high water temperature influences the spawning season which was found to extend for seven months. Ripe males firstly appear in the catch in February while the appearance of ripe females is delayed to March (Amin, 1985 a). This phenomenon was noticed by Popper et al., (1973) in S. rivulatus obtained from the Gulf of Agaba.

The testicular structure of male S. rivulatus is similar to many other teleosts (Stenger, 1959; Sakun and Butskaia, 1968).

The functional structure of testis is firmly correlated with the endocrine balance, important hormones are secreted through the process of spermatogenesis (Hoar, 1969).

Many consequent changes take place during the testicular development of S. rivulatus, mainly in the formed cysts, tubules, and walls forming these structures. Active spermatogenesis starts with the intermediate stage III-IV of gonad maturation. This activation continues in different male individuals of the population to October. The early testicular development, which is fairly stimulated by water temperature makes it possible to get spermatozoa early in February.

The synchronization of spermiation and ovulation is a very important process in reproduction. Female **S. rivulatus** is a monocyclic fish lays eggs once a year. The secretion of sexual hormones at the beginning and the end of spermiation regulates such synchronization (Liley 1969).

After the completion of spawning, the testes of S. rivulatus still contain remnants of spermatozoa. At the next spermatogenic cycle, these remnants have completely disappeared, indicating that a type of resorption has taken place. This resorption was believed by Lofts (1966), as due to a phagocytic activity carried out by the Sertoli cells detached from the tubular wall. Billard and Takashima (1983), have reported that phagocytosis which occurs in the sperm duct is mainly done by the epithelial cells.

The variations of the determined values of GSI and HSI were used to identify the spawning season of **S. rivulatus.** The maximal values of HSI was obtained in Decemeber, at the same time a low value of GSI was calculated. The high value of HSI may be related to the big amount of deposited fat in winter and before the spawning season (Amin, 1985).

The first sign of decrease in HSI value was noted in January, and this reveals the gonadal development at such time. Small fishes were shown to have lower values of HSI than the bigger ones. Generally, the average values of HSI of male S. rivulatus ranged from 0.96 to 2.79. Schmidt-Nielsen (1975), stated that the value of HSI of Osteichthes is usually about 1.0 to 2.0, while Oguri (1978) showed that it varies from 1.0 to 5.65.

The increase in the values of GSI of male **S. rivulatus** inhabiting the Gulf of Aqaba starts in May, i.e. later than that of **S. rivulatus** in southern Red Sea (Jeddah region).

It could be concluded that the elevation of water temperature activates the development of sexual cells in the testes of **S. rivulatus**, and furtherly determines the duration of spawning.

REFERENCES

- Amin, E.M. 1985. Seasonal variation in fat concentration during the gonadal maturation of Siganus rivulatus Forsk. from the Red Sea. Bull. Fac. Sci. Alex. Univ., 24 (4): 154-165.
- Amin, E.M. 1985. Seasonal developmental changes in the ovaries of Siganus rivulatus from the Red Sea. Submitted to Bull. Jap. Soc. Sci. Fish.

- Amin, E.M. and Kh.A. Hussein 1985. Identification of family Siganidae in the Red Sea, Submitted to Bull. Inst. Ocean. Fish., Cairo.
- Billard, R. and F. Takashima 1983. Resorption of spermatozoa in the sperm duct of Rainbow Trout during the post-spawning period. Bull. Jap. Soc. Sci. Fish. 49 (3): 387-392.
- Hjort, J. 1910. Herring investigation untill January 1910 : Reproduction.Publ. Circons. Pern. Internat. Explor. Mer.
- Hoar, W.S. 1969. Reproduction, fish physiology. Acad. Press, New York, Part II, 8-14.
- Liley, N.R. 1969. Reproduction, fish physiology. Acad. Press, New York, Part II, 76-85.
- Lofts, S. 1966. Effects of methyl testosterone on the testes of a hypophysectamized Cyprinodont fish, Fundulus heteroclitus. Gen. Comp. Endocrinol., 6, 74-88.
- Oguri, M. 1978. On the hepatosomatic index of Holocephalian fish. Bull. Jap. Soc. Fish., 44 (2): 131-134.
- Petersen, J. 1895. Einige methoden Zuk Bestimmung des Alters und Wachaes der Fische. Milteil. D. Deutsch. Seefisherelvereins.
- Popper, D.; H. Gordin and G.W. Kissil 1973. Fertilization and hatching of the rabbit fish Siganus rivulatus. Aquaculture, 2 (1): 37-44.
- Popper, D. and N. Gundermann 1975. Some ecological and behavioural aspects of siganid populations in the Red Sea and Mediterranean coasts of Israel in relation to their suitability for aquaculture. **Aquaculture**, 6, 127-141.
- Popper, D.; R. Pitt and Y. Zohar 1979. Experiments on the propagation of Red Sea siganids and some notes on their reproduction in nature. Aquaculture, 16, 177-181.
- Sakun, O.F. and N.A. Butskaia 1968. Determination of maturity stages and studies of sexual cycle in fishes.PINRO, 5-23.
- Schmidt Nielsen, K. 1975. Animal physiology, Adaptation and Environment. Cambridge Univ. Press, London, p. 542.
- Soh, C.L. and T.J. Lam 1973. Induced breeding and early development of the rabbitfish Siganus oramin Schneider. Proc. Symp. Biol. Res. and Nat. Dev., 49-56.
- Stenger, H.N. 1959. A study of the structure and development of certain reproductive tissues of Mugil cephalus L. Zoologica, USA, 44 (2): 118-127.
- Thebaity, S.; F. Bokhary and A. Badawi 1984. Cultivation of siganids in the Red Sea.Min. Agr. and Water Res., S.A. p. 71.

r