

OCCURRENCE OF FOLLICULAR ATRESIA IN OVARY OF FRESHWATER CATFISH, *HETEROPNEUSTES FOSSILIS* (BLOCH)

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Atresia is a highly regulated process in the vertebrate ovary which appears to be essential for the maintenance of ovarian homeostasis. The oocytes in different stages of growth are lost through atresia (or degeneration) affecting the fecundity/reproductive potential of the fish. The atretic oocytes have been identified in the ovary of the catfish during immature, pre-spawning, spawning and post-spawning periods. An attempt has been made to record the different stages of follicular atresia in *Heteropneustes fossilis*. Possible causes of the phenomenon have also been discussed.

INTRODUCTION

The corpora atretica (atretic follicles or pre-ovulatory corpora lutea) and post-ovulatory follicles (ruptured follicles, discharged follicles or post-ovulatory corpora lutea) are of wide occurrence in the ovaries of vertebrates including fish (Saidapur, 1978, 1982; Guraya, 1993; Wood and Kraak, 2001). It has been generally observed that all the developing follicles (oocytes) do not reach to maturity and ovulate successfully. Many of them become atretic (degenerated) at some stage during their development. Follicular atresia has been recorded in a number of teleostean species such as *Fundulus heteroclitus* (Mathews, 1938), *Heterendria formosa* (Fraser and Renton, 1940), *Neotoca bilineata* (Mendoza, 1943), *Gadus merlangus* and *G. esmarki* (Gokhale, 1957), *Wallagonia attu* (Dixit, 1960), *Satipinna phasa* (Jhingran, 1960), *Mystus seenghala* (Dixit, 1960; Sathyanesan, 1961, 1962), *Ophiocephalus punctatus* (Belsare, 1962, 1975), *Heteropneustes fossilis* (Nair, 1963), *Tor (Barbus) tor* (Rai, 1966), *Xenentodon cancila* (Rastogi, 1966), *Glossogobius giuris* (Rajalakshmi, 1966; Saksena and Bhargava, 1972), *Glyptosternum pectinopterum* (Khanna and Pant, 1967), *Monopterus albus* (Chan et al., 1967), *Clarias batrachus* (Lehri, 1968), *Mystus tengara* (Rastogi, 1968a), *Amphipnous cuchia* (Rastogi, 1968b), *Schizothorax niger* (Malhotra, 1971), *Cyprinus carpio communis* (Guraya et al., 1977), *Mystus cavasius* (Saidapur, 1978), *Channa marulius* (Srivastava, 1980), *Tilapia leucostica* (Kling, 1981; *Schizothorax plagiostomus* (Agarawal and Singh, 1990), *Macodon ancylodon* (Vizziano and Berois, 1990) and *Poecilia reticulata* (Rajkumar and Hemalatha, 2005). Since oocytes in different stages of growth and differentiation are lost through atresia (or degeneration) affecting the fecundity or reproductive potential of the fish (Guraya, 1993, 1994), an attempt has been made to record the follicular atresia in ovary of the commercially important freshwater catfish, *H.*

fossilis and the observations discussed in the light of the various stages of atresia propounded by Belsare (1975) and Saidapur (1978).

MATERIAL AND METHODS

Live specimens of *H. fossilis* (body weight range 62-86 g) were procured from the fields adjoining Bhubaneswar (Orissa) during middle of every month. Ovaries of the catfish were surgically removed and fixed immediately in freshly prepared Bouin's solution. After 24 h, tissues were washed thoroughly in running tap water, dehydrated in ascending series of alcohol, cleared in xylene and embedded in paraffin wax at 60°C. Serial sections were cut at 6 µm and stained with hematoxylin-eosin (H&E), toluidine blue (1%) and bromophenol blue (0.5%) (Pearse, 1968; Bancroft and Stevens, 1977).

RESULTS AND DISCUSSION

Follicular atresia in the fish ovary is of common occurrence during pre-spawning, spawning and post-spawning periods (Saidapur, 1978, 1982; Guraya, 1993). During the course of maturation process, some of the ova that fail to attain maturity or spawn undergo resorption and are called atretic follicles (Saidapur, 1978, 1982; Guraya, 1993, 1994; Khanna, 2006). Atresia is a highly regulated process in the vertebrate ovary which appears to be essential for the maintenance of ovarian homeostasis (Wood and Kraak, 2001). All the four stages of follicular atresia, described in the teleosts (Belsare, 1962, 1975; Saidapur, 1978), have been encountered during the present study in the catfish. In *H. fossilis*, remnants of atretic follicles in the form of nodule of stroma tissue were observed even in the immature ovaries during December-January (Figs. 1, 2). Previtellogenic atretic follicles in ovary of the catfish depicted excessive vacuolation of ooplasm towards periphery, flocculent appearance of ooplasm and hypertrophied granulosa cells penetrating the zona pellucida or oolema (Fig. 3). Some previtellogenic atretic follicles of *H. fossilis* during March-April exhibited prominent granulosa cells, separation of ooplasm from zona pellucida and disorganization of ooplasm (Fig. 4). Vitellogenic ovarian follicles of *H. fossilis* at the early stage of atresia (May-June) showed prominent granulosa cells, vacuolation of the ooplasm at periphery and ooplasm giving flocculent appearance (Figs. 5, 6). Thickened zona pellucida and hypertrophied granulosa cells were recorded as the atresia advanced in the vitellogenic follicle of *H. fossilis* (Fig. 7). During the advanced stage of atresia (September-October), the oocytes of the catfish depicted disorganized ooplasm, obscure germinal vesicle and hypertrophied granulosa cells. Phagocytic invading of granulosa cells in zona pellucida and ooplasm were also prominent (Fig. 8).

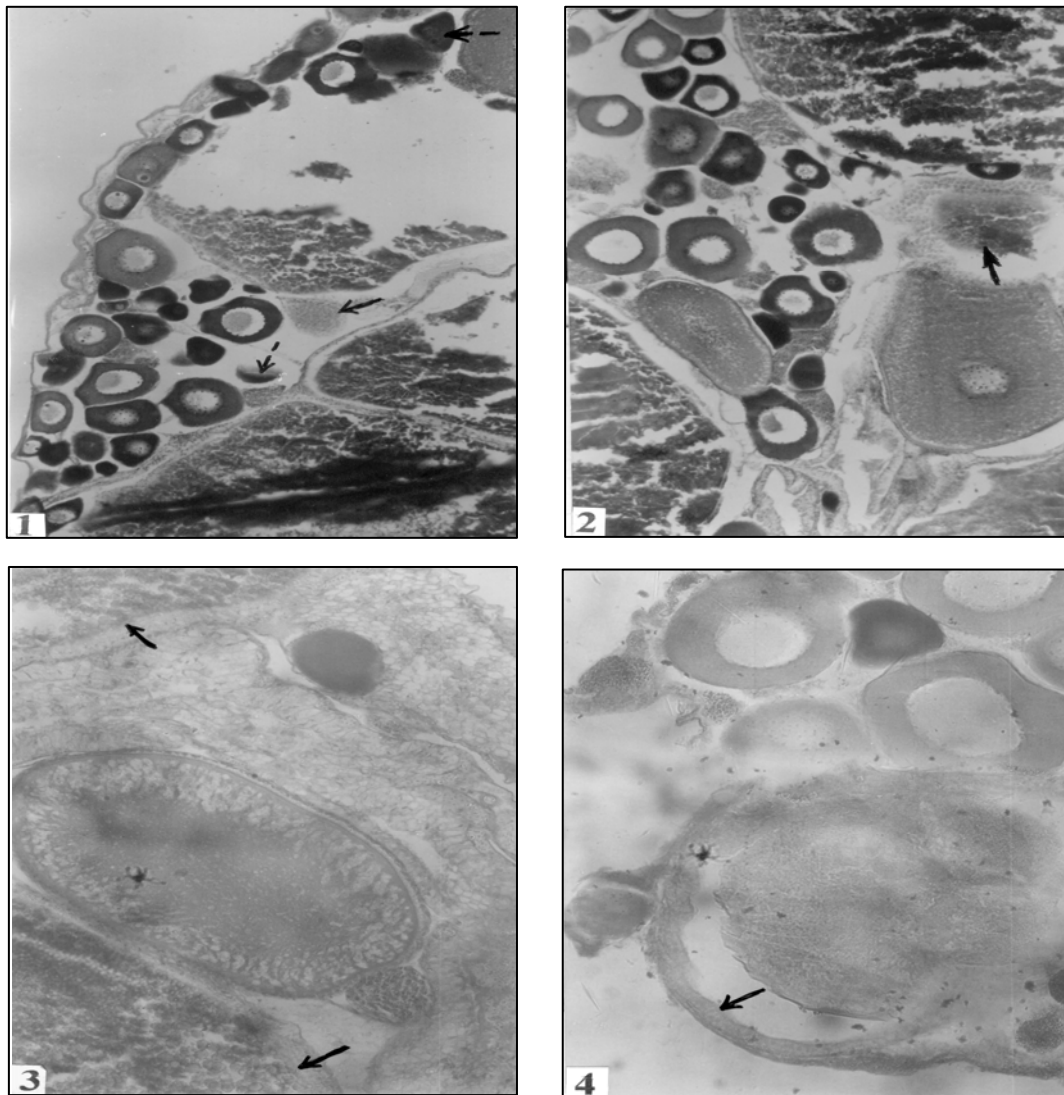


Fig. 1. Ovary of immature *H. fossilis* showing previtellogenic oocytes. Mark the nodule of stroma tissue (arrow) and atretic follicle (broken arrow). H&E. x 100; Fig. 2. Ovary of immature the catfish exhibiting prominent nodule of stroma (arrow) and oocytes in early stages of development. H&E. x 100; Fig. 3. Previtellogenic atretic follicles in ovary of *H. fossilis* depicting excessive vacuolation of ooplasm towards periphery. Mark the flocculent ooplasm and hypertrophied granulosa cells (arrow) penetrating the ooplasm. H&E. x 250; Fig. 4. Previtellogenic ovarian atretic follicle of the catfish showing prominent granulosa cells, separation of ooplasm from zona pellucida (arrow) and disorganization of ooplasm. H&E. x 250.

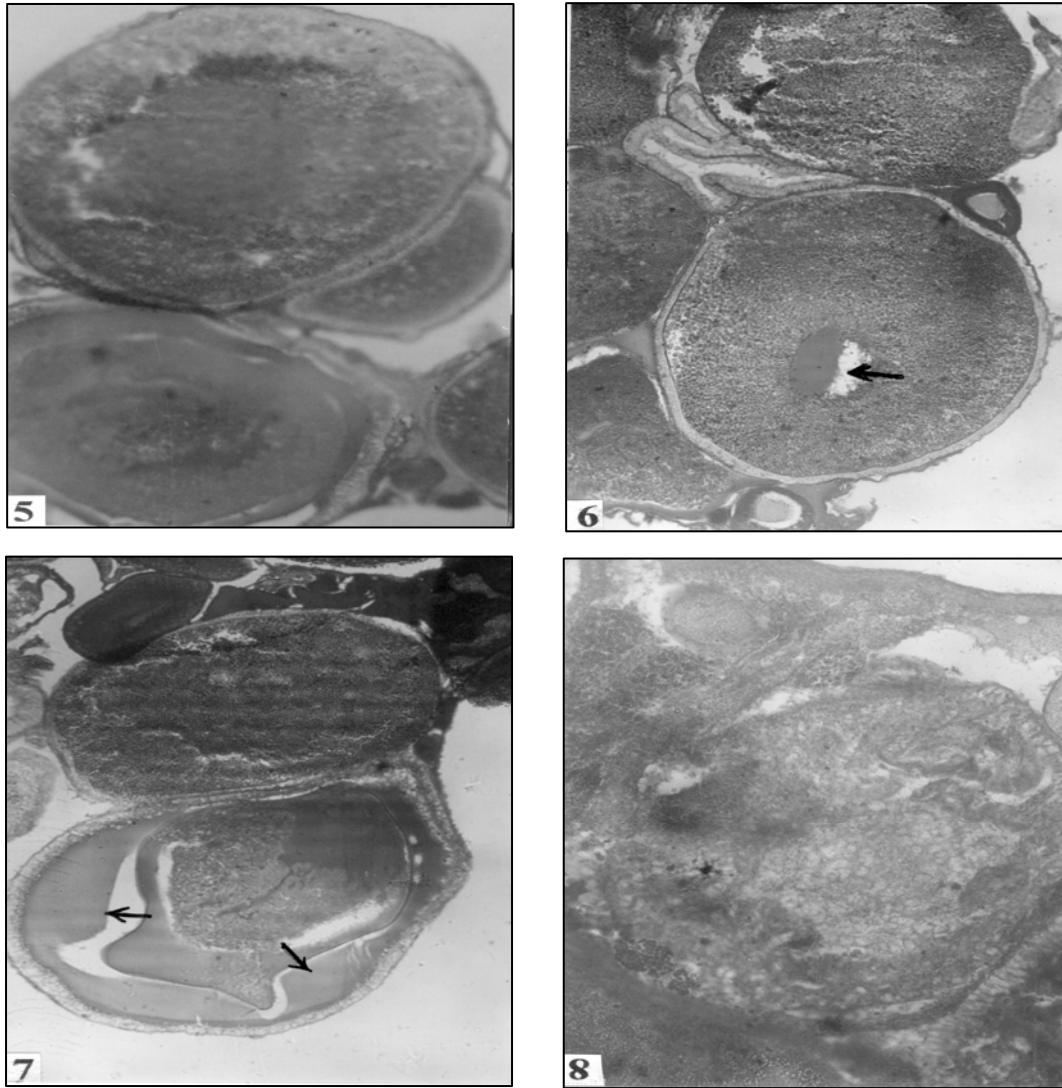


Fig. 5. Vitellogenic ovarian follicles of *H. fossilis* at the early stage of atresia with prominent granulosa cells and vacuolation of the ooplasm at periphery. Mark the flocculent appearance of ooplasm in the oocytes. H&E. x 250; Fig. 6. Vitellogenic follicles in ovary of *H. fossilis* at the early stage of atresia showing vacuolated germinal vesicle (arrow) and vacuolation of ooplasm at periphery. H&E. x 250; Fig. 7. Atretic vitellogenic follicle in ovary of *H. fossilis* with vacuolated cytoplasm, thickened zona pellucida (arrow) and hypertrophied granulosa cells. H&E. x 250; Fig. 8. Vitellogenic follicle in ovary of *H. fossilis* with advanced stage of atresia depicting disorganized ooplasm, obscured germinal vesicle and hypertrophied granulosa cells. Mark the phagocytic granulosa cells invading zona pellucida and ooplasm. H&E. x 250.

Though the precise causes of follicular atresia in teleosts have not yet been clearly defined, several exogenous (photoperiod, temperature, rainfall, crowding, captivity, nutrition, physico-chemical characteristics of ambient water, pollutants/biocides etc) as well as endogenous (insufficient gonadotrophic hormone, imbalance of hormones and steroids) have been implicated in the process (Sundararaj and Goswami, 1968; Saidapur, 1978, 1982; Kling, 1981; Saksena and Raizada, 1984; Guraya, 1993, 1994; Rodriguez *et al.*, 1995; Miranda *et al.*, 1999; Wood and Kraak, 2001; Khanna, 2006). Further studies are required to resolve the causes and functions of follicular atresia for management of broodstocks to realize optimum fecundity of the fish.

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REFERENCES

- Agarawal, N. K. and H. R. Singh, 1990. Preovulatory follicular atresia (corpora atretica) in snowtrout, *Schizothorax plagiostomus*. *J. Anim. Morphol. Physiol.*, **37**: 29-33.
- Bancroft, J. D. and A. Stevens, 1977. Theory and Practice of Histological Techniques. Churchill Livingstone, New York.
- Belsare, D. K., 1962. Seasonal changes in the ovary of *Ophiocephalus punctatus* (Bloch). *Indian J. Fish.*, **8**: 140-156.
- Belsare, D. K., 1975. Component of the ovarian endocrine tissue in teleost species. *Zool. Pol.*, **25**: 5-11.
- Chan, S. T. H., A. Wright and G. Phillips, 1967. The atretic structures in the gonads of the rice field eel (*Monopterus albus*) during natural sex reversal. *J. Zool.*, **153**: 527-539.
- Dixit, R. K., 1960. Seasonal growth of oocytes of *Mystus seenghala* (Sykes) and *Wallagonia attu* (Bloch), with an inference about their spawning habits. *Proc. Natl. Acad. Sci. India*, **30B**: 241-245.
- Fraser, E. A. and R. M. Renton, 1940. Observations on the breeding and development of the viviparous fish, *Heterandria formosa*. *Quart. J. Microsc. Sci.*, **81**: 479-490.
- Gokhale, S. V., 1957. Seasonal histological changes in the Whitting (*Gadus merlangus*) and Norway Pout (*Gadus esmarki*). *Indian J. Fish.*, **4**: 311-317.
- Guraya, S. S., 1993. Follicular (or oocyte) atresia and its causes and functional significance in the fish ovary. In: *Adances in Fish Research* (Ed. B. R. Singh). Narendra Pub. House, New Delhi. pp. 313-332.
- Guraya, S. S., 1994. Gonadal development and production of gametes in fish. *Proc. Indian Natl. Sci. Acad.*, **60B**: 15-32.

- Guraya, S. S., H. S. Toor and S. Kumar, 1977. Morphology of ovarian changes during the reproductive cycle of the fish, *Cyprinus carpio communis* (Linn.). *Zool. Beitr.*, **23**: 405-437.
- Jhingran, A. G., 1961. Studies on maturity and fecundity of the Gangetic anchovy, *Satipinna phasa* (Hamilton). *Indian J. Fish.*, **8**: 281-311.
- Khanna, S. S., 2006. An Introduction to Fishes. 5th Edn. Silver Line Publications, Faridabad.
- Khanna, S. S. and M. C. Pant, 1967. Seasonal changes in the ovaery of a sisorid catfish, *Glyptosternum pectinopterum*. *Copeia*, **1**: 83-88.
- Kling, D., 1981. Total atresia of the ovaries of *Tilapia leucosticta* (Cichlidae) after intoxication with the insecticide, Lebaycid. *Experientia*, **37**: 73-74.
- Lehri, G. K., 1968. Cyclical changes in the ovary of the catfish, *Clarias batrachus*. *Acta Anat.*, **69**: 105-124.
- Malhotra, P., 1971. Studies on the seasonal changes in ovary of *Schizothorax niger* (Heckel) from Dal Lake in Kashmir. *Jap. J. Ichthyol.*, **17**: 110-116.
- Mathews, S. A., 1938. The seasonal cycle in the gonads of *Fundulus*. *Biol. Bull. (USA)*, **75**: 67-74.
- Mendoza, G., 1943. The reproductive cycle of the viviparous teleost, *Neotoca bilineata*, a member of the Family Goodeidae. IV. The germinal tissue. *Biol. Bull. (USA)*, **84**: 87-98.
- Miranda, A. C. L., N. Bazzoli and Y. Sato, 1999. Ovarian follicular atresia in two teleost species: a histological and ultrastructural study. *Tissue & Cell.*, **31**: 480-488.
- Nair, P. V., 1963. Ovular atresia and the formation of so-called corpus inteum in the ovary of the Indian catfish, *Heteropneustes fossilis*. *Proc. Zool. Soc. (Bengal)*, **16**: 51-65.
- Pearse, A. G. E., 1968. Histochemistry: Theoretical and Applied. Vol. 1. Churchill Livingstone, London & Edinburgh.
- Rai, B. P., 1966. Cyclical changes in the ovary of *Tor (Barbus) tor*. *Acta Zool.*, **43**: 289-307.
- Rajalakshmi, M., 1966. Atresia of oocytes and ruptured follicles in *Gobio giuris*. *Gen. Comp. Endocrinol.*, **6**: 378-385.
- Rajkumar, R. and K. K. Hemlatha, 2005. Observations on follicular atrition in *Poecilia reticulata* (Peters) (Atheriniformes: Teleostei). *J. Inland Fish Soc. India*, **37(2)**: 21-25.
- Rastogi, R. K., 1966. A study of the follicular atresia and evacuated follicles in the Indian teleost, *Xenotodon cancila*. *Acta Biol. Acad. Sci. Hung.*, **17**: 52-63.
- Rastogi, R. K., 1968a. The occurrence and significance of follicular atresia in the catfish, *Mystus tengara*. *Acta Zool.*, **49**: 309-319.
- Rastogi, R. K., 1968b. The occurrence and significance of ovular atresia in the mud eel, *Amphipnous cuchia*. *Acta Anat.*, **73**: 148-160.
- Rodriguez, J. N., Z. J. Oteme and S. Hem, 1995. Comparative study of vitellogenesis of two African catfish species, *Chrysichthys nigrodigitatus* (Claroteidae) and *Heterobranchus longifilis* (Clariidae). *Aquat. Living Resour.*, **8**: 291-296.
- Saidapur, S. K., 1978. Follicular atresia in ovaries of non-mammalian vertebrates. *Intern. Rev. Cytol.*, **54**: 225-244.
- Saidapur, S. K., 1982. Structure and function of post-ovulatory follicles (corpora lutea) in the ovaries of non-mammalian veretebrates. *Intern. Rev. Cytol.*, **58**: 243-285.

- Saksena, D. N. and H. N. Bhargava, 1972. The corpora atretica, post-ovulatory follicles and spawning periodicity of Indian freshwater goby, *Glossogobius giuris* (Ham.). *Zool. Jb. Anat.*, **89**: 611-620.
- Saksena, D. N. and A. K. Raizada, 1984. On the corpora atretica and post-ovulatory follicles and spawning periodicity in some freshwater Indian teleosts. *Intl. J. Acad. Ichthyol.*, **5**: 11-22.
- Sathyanesan, A. G., 1961. A histological study of the ovular atresia in the catfish, *Mystus seenghala* (Sykes). *Rec. Indian Mus.*, **59**: 75-82.
- Sathyanesan, A. G., 1962. The ovarian cycle in the catfish, *Mystus seenghala* (Sykes). *Proc. Natl. Inst. Sci. India*, **28B**: 497-506.
- Srivastava, S. J., 1980. Seasonal histological changes in ovary of a freshwater large murrel, *Channa marulius* (Ham.). *Zool. Jb. Anat.*, **104**: 492-499.
- Sundararaj, B. I. and S.V. Goswami, 1968. Effect of short- and long-term hypophysectomy on the ovary and interrenal of catfish, *Heteropneustes fossilis* (Bl.). *J. Exp. Zool.*, **168**: 85-104.
- Vizziano, D. and N. Berois, 1990. Histology of the ovary of *Macrodon ancylodon* (Bloch & Schneider, 1801) (Teleostei: Sciaenidae). Oogenesis. Post-ovulatory follicles. Atresia. *Rev. Bras. Biol.*, **50**: 523-536.
- Wood, A.W. and G. J. van der Kraak, 2001. Apoptosis and ovarian function: novel perspectives from the teleosts. *Biol. Reprod.*, **64**: 264-271.

