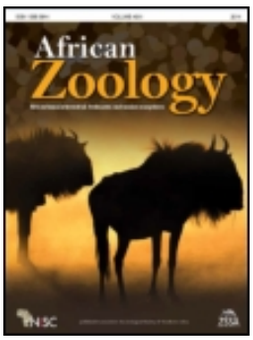

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Histological validation of gonadal macroscopic staging criteria for *Labeo cylindricus* (Pisces: Cyprinidae)

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Histological examination of gametogenesis revealed that the current staging criteria used to assess gonadal recrudescence of the redeye labeo, *Labeo cylindricus,* were adequate. Gametogenesis was qualitatively similar to that of freshwater teleosts with a clearly defined seasonal reproductive cycle. *L. cylindricus* undergoes seasonal gross morphological and cytological gonadal changes with previtellogenesis dominating during the winter, vitellogenic development during spring and summer culminating in large-scale spawning at the end of summer. Post-spawning mass atresia of oocytes was evident in autumn. The histological data presented support macroscopical evidence that *L. cylindricus* is a synchronous iteroparous spawner, reproducing over a short period each year throughout its life-span.

Key words: **gametogenesis, synchronous spawning, potadromesis, gonadal recrudescence, vitellogenesis.**

INTRODUCTION

The genus *Labeo* consists of at least 80 species, comprising 16.4 % of the African cyprinid ichthyofauna (Reid 1985). Labeines are widely distributed throughout the continent, contributing significantly to various commercial and subsistence fisheries (Skelton *et al.* 1991). One common and widely distributed labeine species is the redeye labeo, *Labeo cylindricus* Peters, 1852. Its distribution extends from the Zaire River basin in the north, southwards through the Zambezian and east African coastal drainage systems to the Phongolo system in northern KwaZulu-Natal, South Africa. It is a relatively small labeine species with a maximum length of 25 cm (standard length) and mass of 0.9 kg (Skelton 1993). Various aspects of the life history of *L. cylindricus,* including growth and mortality rates, reproductive seasonality and its potadrometic spawning behaviour, have been described by Weyl & Booth (1999).

Few investigations on reproduction of labeines have included histological studies (van der Merwe *et al.* 1988), most using a combination of macroscopic staging criteria and a gonadosomatic index in the assessment of gonadal recrudescence (Siddiqui *et al.* 1976; Gaigher 1983; van Zyl *et al.* 1995). However, macroscopic staging criteria must be histologically validated if errors in the determination of maturation patterns and reproductive seasonality are to be reduced (West 1990). Histological validation of macroscopical staging criteria and descriptions of gametogenesis also enable past and future reproductive studies to be compared (Booth & Hecht 1997). In this paper, oogenesis and spermatogenesis in *L. cylindricus* are described, previously used macroscopical staging criteria are validated and histological evidence is presented for synchronous iteroparous spawning.

MATERIALS & METHODS

Samples of *Labeo cylindricus* were collected between December 1995 and December 1996 from Lake Chicamba (19°08'S, 33°08'E), a man-made hydroelectric dam situated in central Mozambique. Samples were collected monthly from gill (25, 50, 70, 90 mm stretched mesh) and seine nets (10, 25 mm stretched mesh) with all fish being measured for fork length (mm FL) and weighed whole. The fish were then dissected and sexed. Gonadal recrudescence was assessed, based on the visual characteristics of the gonad (Table 1). The gonads were subsequently removed and weighed (mg) and the eviscerated body mass of the fish (g) recorded. A gonadosomatic index was calculated by expressing the gonad mass as a percentage of the eviscerated mass. Samples of gonadal tissue, from 15 fish in each macroscopic stage, were collected for later histological examination of gametogenesis. Tissue samples were

Table 1. Macroscopic appearance and equivalent histological characteristics of *Labeo cylindricus* gonads at various stages during gonadal recrudescence (modified from Weyl & Booth 1999).

Stage	Macroscopic appearance	Histological appearance
Juvenile	Not possible to visibly distinguish sex Gonad appears a translucent gelatinous strip	Oogonia, chromatin-nucleolus and perinuclear oocytes within the ovary Spermatogonia predominate within the testis.
Resting	Sex distinguishable. Ovary small band of orange-red tissue	Ovary dominated by oogonia, chromatin-nuclear and perinuclear oocytes within the ovary Few primary vesicle oocytes present
	Testis discernible as a thin white band	Primary and secondary spermatocytes predominate throughout the testis
Developing	Ovary increases in size, is flattened dorsoventrally and is orange-red in colour Oocytes visible	Characterized by all oocyte stages up to the primary vesicle stage. More primary vesicle oocytes are noticeable than during the 'resting' stage
	Testis increases in size and is white in colour Sperm is not extrudable from the testis	Testis shows all stages of spermatogenesis dominated by primary and secondary spermatocytes
Ripe	Ovary turgid with oocytes and fills the entire abdominal cavity Oocytes are olive-green to brown and loosely attached to ovigerous lamellae	All stages of vitellogenesis present including secondary and tertiary yolk vesicle oocytes
	Testis creamy white, showing constrictions Sperm can be extruded from the testis	Testis shows all stages of spermatogenesis dominated by spermatozoa production
Spent	Ovary flaccid and sac like with few vitellogenic oocytes visible	All oocyte stages present in the ovary together with atresia of yolk vesicle oocytes
	Testis reduced in size and dirty grey in colour	Testis with all stages of spermatogenesis yet dominated by spermatocytes

fixed in 10 % formalin for three to four days after which they were stored in 50 % propyl alcohol. Tissue samples were embedded in Paraplast using routine methods, sectioned between 3 and 7 μm and stained using Gill's haematoxylin and eosin and examined using standard light microscopy. Classification of oogenesis was based on criteria used by Wallace & Selman (1981) and Tyler & Sumpter (1996). Spermatogenesis is described using terminology from de Vlamming (1972) and Van der Horst & Erasmus (1978).

Stained and mounted gonadal sections from at least five fish per microscopic stage were digitally scanned and measurements of cell and nucleus diameter, zona radiata thickness, and the number of nucleoli per nucleus digitally recorded using imaging software (Sigma-Scan, Jandel Scientific). All cell and nucleus diameters were calculated as an average of two measurements per cell, across the nucleus and perpendicular to one another. Cytoplasmic changes were noted by calculating nucleus diameter/cell diameter and nucleus area/cytoplasm ratios.

RESULTS

Macroscopic appearance

Visual inspection of the gonads of *Labeo cylindricus* revealed five macroscopic stages within the ovaries and testes. The various stages differed predominantly with regard to colour, size and general appearance of the gonad (Table 1).

Ovarian recrudescence based on macroscopic staging criteria, and the use of a gonadosomatic index, followed a distinct seasonal pattern (Fig. 1). Over 85 % of the ovaries were in a 'resting' condi-

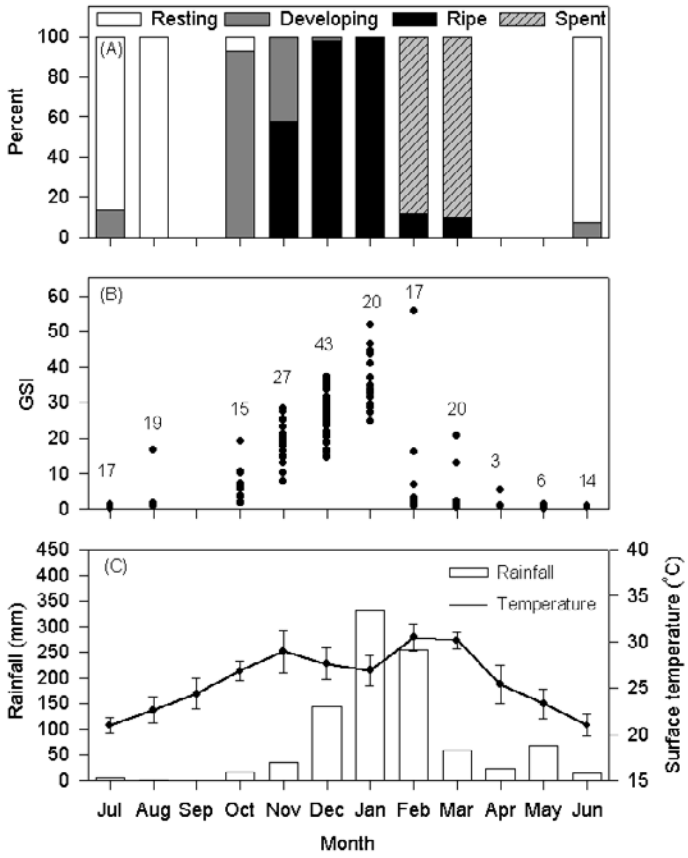

Fig. 1. (**A**) female monthly maturity stages and (**B**) individual gonadosomatic indices (GSI) for female *Labeo cylindricus*. (**C**) rainfall and mean surface water temperatures (± standard deviation) from Lake Chicamba, Mozambique, between December 1995 and December 1996. Modified from Weyl & Booth (1999).

tion during winter from June to August. In October, over 92 % of the sampled population had 'developing' ovaries. By November, over 57 % of the females were in ripe condition and in December and January all mature females were 'ripe'. Spawning activity had ceased by February, with 88 % of ovaries sampled in a 'spent' condition. This trend was mirrored by the rapid rise in the gonadosomatic index from a mean winter low of 0.6 % in June and July, a steady rise from October and peaking in December and January at means of 27.1 % and 35.2 %, respectively. By February the mean gonadosomatic index had dropped to 6 %.

Histological appearance

Oogenesis

The ovaries of *L. cylindricus* are large, paired, sac-like organs suspended within the body cavity by mesovaria. Histologically, oogonia and six discrete stages of oocyte development (chromatin-nucleolus oocytes, perinuclear oocytes, primary yolk vesicle oocytes, secondary yolk vesicle oocytes, tertiary yolk vesicle oocytes, atretic oocytes) were discernible based on nuclear and cytoplasmic characteristics.

Oogonia were most frequently observed at the periphery of the ovigerous lamellae and were embedded in nests within the germinal epithelium. Oogonia were characterized by their small size, large nucleus to cytoplasm ratio and lightly basophilic (blue-staining) cytoplasm (Fig. 2a, Table 2).

Cromatin nucleolus oocytes were slightly larger than oogonia, characterized by a centrally located nucleus within a lightly basophilic cytoplasm (Fig. 2a,b, Table 2). These oocytes were restricted to the oogonial nests and contained a primary chromatin-rich nucleus with densely basophilic chromatin around the periphery of the nucleus.

Table 2. Mean (± standard deviation) of various stages of oogenesis in *Labeo cylindricus*. Measurements are only representative of the fixed material and have not been corrected for shrinkage or swelling attributed to histological preparation. All diameters are an average of two measurements per cell, across the nucleus and perpendicular to one another. *n* = number of cells measured.

Stage	Cell diameter (μm)	Nucleus diameter (μm)	Number of nucleoli	Zona radiata thickness (μm)	*n*
Oogonia	11.25 ± 1.77	8.50 ± 1.29	–	–	20
Chromatin-nucleolus oocytes	16.25 ± 3.77	9.65 ± 0.82	–	–	20
Perinuclear oocytes					
a) Pre-	50.58 ± 15.39	21.97 ± 6.75	2.90 ± 1.25	–	20
b) Early-	98.73 ± 26.56	41.88 ± 12.48	12.50 ± 2.64	–	20
c) Late-	176.42 ± 21.09	75.5 ± 21.09	26.60 ± 5.77	–	20
Primary yolk vesicle oocytes	187.75 ± 25.68	82.00 ± 18.75	24.90 ± 9.50	1.71 ± 0.38	20
Secondary yolk vesicle oocytes	381.67 ± 42.98	109.33 ± 10.83	38.43 ± 7.49	1.70 ± 0.41	15
Tertiary yolk vesicle oocytes	818.75 ± 55.25	329.23 ± 71.76	>60	1.72 ± 0.38	20

With the initiation of the first meiotic division and further growth, perinuclear oocytes developed. They were strongly basophilic, containing numerous nucleoli and surrounded by a well-defined follicular layer (Fig. 2a,b,c, Table 2). Pre-perinuclear oocytes were polygonal in shape with an intensely basophilic cytoplasm and were found closest to the germinal epithelium. The nucleus contained two or three large nucleoli and a number of smaller nucleoli. Early-perinuclear and late-perinuclear oocytes increased in diameter, became more ovoid in shape and less basophilic with a proliferation of nucleoli in the nucleus. The formation of the zona granulosa occurred within late-perinuclear oocytes.

The formation of the zona radiata between the follicular layer (zona granulosa and theca) and the developing oocyte, marked the end of the primary growth phase and was followed by the appearance of primary yolk vesicles (cortical alveoli) in the cytoplasm. Primary yolk vesicle oocytes had a uniformly stained basophilic cytoplasm (Fig. 2c, Table 2).

With the onset of exogenous yolk sequestration, acidophilic (red-staining) yolk globules were visible in the region of the cortical alveoli. Later this extravesicular yolk was sequestered throughout the cytoplasm together with the cortical alveoli (Fig. 2d, Table 2). During vitellogenesis the nucleus contained chromatin granules and peripheral nucleoli. The cortical alveoli surrounding the nucleus became enlarged, the zona radiata and zona granulosa increased in thickness and the zona radiata became striated.

With continued exogenous yolk sequestration, the acidophilic yolk globules dominated the cytoplasm (Fig. 2e, Table 2). Sectioning of the vitellogenic oocytes proved difficult due to their large size and yolk globule-dominated cytoplasm.

Atretic oocytes were characterized by phagocytic cells invading the vitelline envelope. Phagocytosis was initiated on the periphery of the oocyte, invading the cytoplasm towards the nucleus (Fig. 2f, Table 2). Only secondary and tertiary yolk vesicle oocytes were found to be atretic.

Spermatogenesis

The testes of *L. cylindricus* are paired structures suspended in the body cavity by mesorchia with a tunica albuginea composed of connective tissue surrounding each testis. A number of seminiferous tubules lead into secondary sperm ducts, joining posteriorly to form the main sperm duct. Four discrete stages of spermatogenesis (spermatogonia, spermatocytes, spermatids, spermatozoa) were observed.

Spermatogonia were characterized by their large size, prominent cytoplasm and lightly basophilic nuclear chromatin (Fig. 3a, Table 2). The spermatocytes, which had smaller nuclei than the spermatogonia, occurred towards the lumen of the seminiferous tubule (Fig. 3a,b,c, Table 2). With the rupturing of the secondary spermatocytic cysts, spermatids were released into the tubule lumen and formed small nests on the edge of the lumen (Fig. 3c,d, Table 2). Spermatozoa, characterized by their small size and intensely basophilic heads, accumulated within the tubules and moved towards and accumulated within the sperm ducts (Fig. 3c,d, Table 2).

Staging validation

Minor qualitative differences between the macroscopic and histological stages were evident.

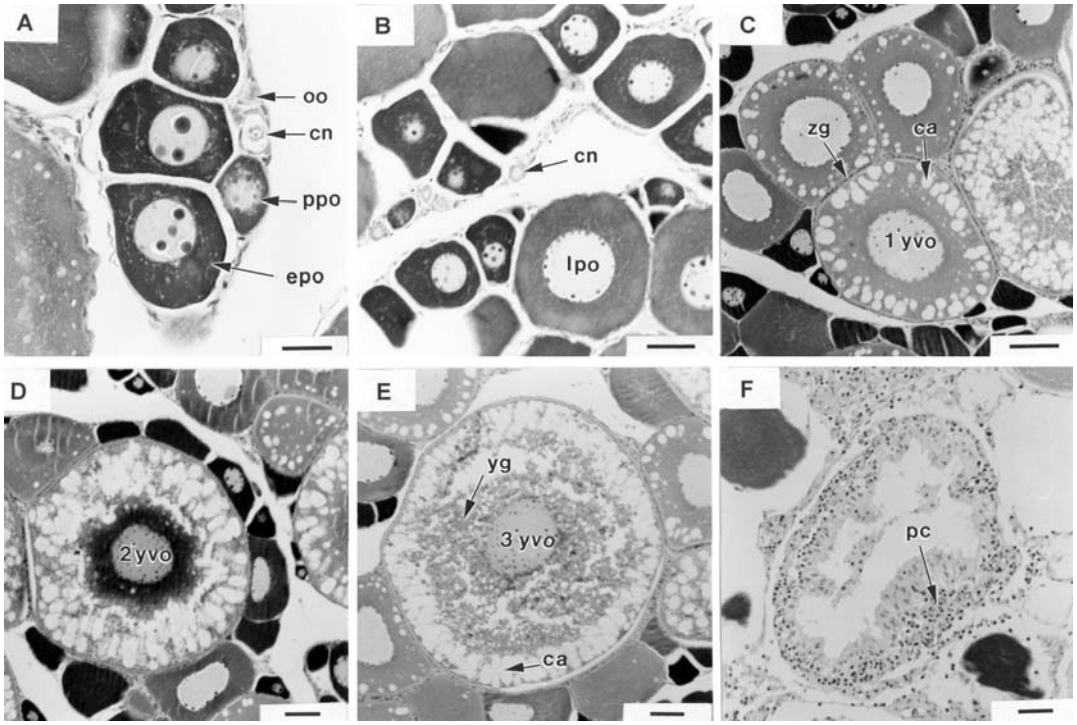

Fig. 2. Transverse sections through ovaries of *Labeo cylindricus* illustrating oogenesis. **A**, immature ovary containing oogonia (oo), chromatin-nucleolus oocytes (cn), pre- (ppo) and early- (epo) perinuclear oocytes; **B**, immature ovary containing chromatin nucleolar oocytes and all stages of perinuclear oocytes. lpo = late perinuclear oocyte; **C**, the onset of maturation begins with the appearance of primary yolk vesicle oocytes (1°yvo) with cortical alveoli forming in the periphery of the cytoplasm, the zona radiata and zona granulosa (zg) are developed; **D**, secondary yolk vesicle oocytes (2°yvo) appear with the sequestration of vitellogenic yolk, with the yolk globules and cortical alveoli in similar proportions; **E**, tertiary yolk vesicle oocytes (3°yvo) are characterized by cortical alveoli (ca) migrating to the cytoplasm periphery with the zona radiata (zr) and zona granulosa well-developed, the cytoplasm was dominated by yolk globules (yg); **F**, atretic oocytes are characterized by phagocytosis by invading phagocytes (pc). All sections stained using haematoxylin and eosin. Scale bars: A, 7 µm; B–C, 50 µm; D–F, 100 µm.

Ovaries within the 'juvenile', 'resting' and 'developing' stages (Fig. 4a–c) contained only previtellogenic oocytes (from oogonia to primary vesicle oocytes). The 'resting' and 'developing' macroscopic stages were qualitatively similar, both containing oocyte stages up to the primary vesicle oocyte stage (Figs 4b–c). Ovaries in the 'developing' stage contained more primary vesicle oocytes, giving them a coarser and grainy visual appearance. Ovaries that were considered 'ripe' contained all oocyte stages, being dominated by vitellogenic oocytes (Fig. 4d) with final maturation characterized by the presence tertiary yolk vesicle oocytes similar in size to migratory nuclei (Fig. 4e). Only 'spent' ovaries contained evidence of atresia, with atretic vitellogenic oocytes found throughout the ovary (Fig. 4f). Greater differences existed between the macroscopic and microscopic testicular stages. Testes of 'juvenile' fish were predominantly composed of spermatogonia, 'resting', 'developing' and 'spent' testes (Fig. 3b) were dominated by spermatogenesis with spermatozoa abundant in the 'ripe' testicular stage (Fig. 3d).

During early stages of oogenesis, a marked change in cytoplasmic growth was noticeable, with the nucleus/cytoplasm area and nucleus/cell diameter ratios decreasing rapidly until the perinuclear stages (Fig. 5). Little change in the two ratios investigated was evident until the initiation of exogenous yolk sequestration in the secondary yolk vesicle stage after which there was continued nuclear growth in tertiary yolk vesicle oocytes.

DISCUSSION

An important component of many studies concerning fish reproductive biology is the assessment of gonadal development of individual fish.

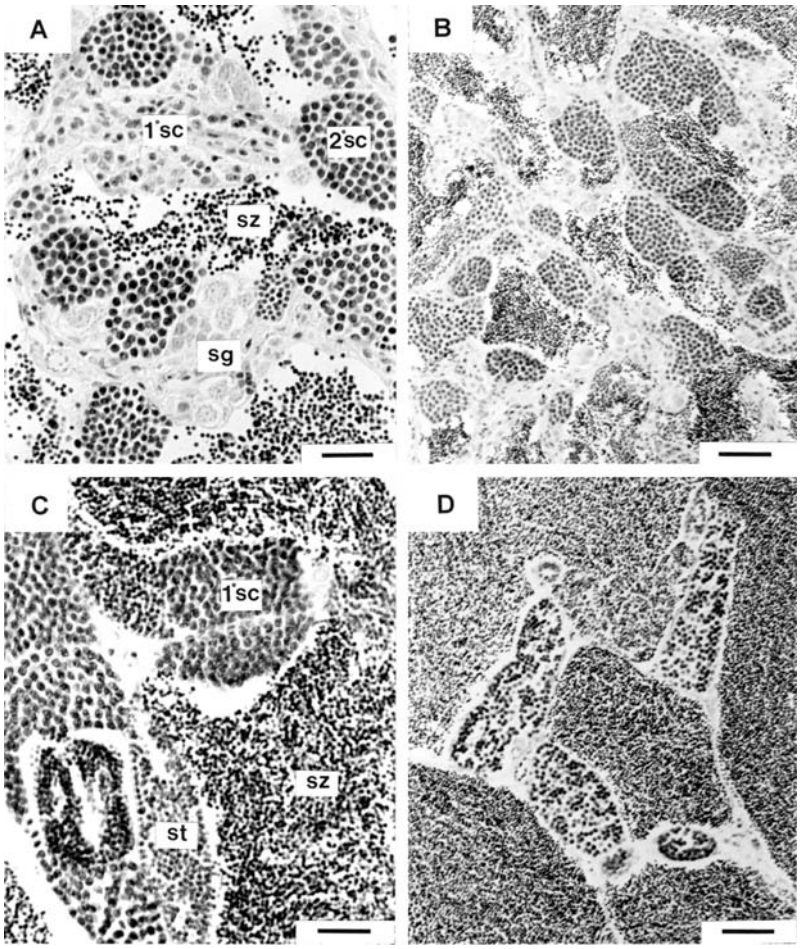

Fig. 3. Transverse sections through testes of *Labeo cylindricus* illustrating spermatogenesis. **A, B**, spermatogonia (sg) and primary (1°sc) and secondary spermatocytes (2°sc) dominate immature testes; **C, D**, during the spawning season, spermatozoa (sz) fill the lumen in large quantities with spermatids (st) visible in nests surrounding the tubules. All sections stained using haematoxylin and eosin. Scale bars: A, 25 µm; B, 50 µm; C, 25 µm; D, 50 µm.

Methods used range from those that are highly detailed to those that are cursory, each method having inherent advantages and disadvantages (West 1990). While microscopical examination is the most detailed and accurate, it is also the most costly and time-consuming. Macroscopical assessment, on the other hand, is the fastest and the cheapest. Unfortunately it lacks accuracy and needs histological validation. Gonadosomatic indices provide useful insights into the changes in gross gonadal morphology. They must, however, be used to complement results from other methods, as gonadosomatic indices are biased when indices from different-sized fish are compared (de Vlamming *et al.* 1982). Therefore, a comprehensive reproductive study requires a combination of available assessment methods.

The histological results obtained in this study suggest that the visual macroscopic criteria (Table 1) used to stage *Labeo cylindricus* recrudescence were adequate. Despite the slight qualitative differences, principal phases of gamete development were easily discernible, as the macroscopic changes within the gonads were discrete and visually distinct.

Several discrepancies exist regarding the number of phases of gamete development, particularly with regard to oogenesis (reviewed by Tyler & Sumpter 1996). Several staging systems have been proposed which incorporate physiological, biochemical, morphological and histological criteria. The major developmental events that occur can be

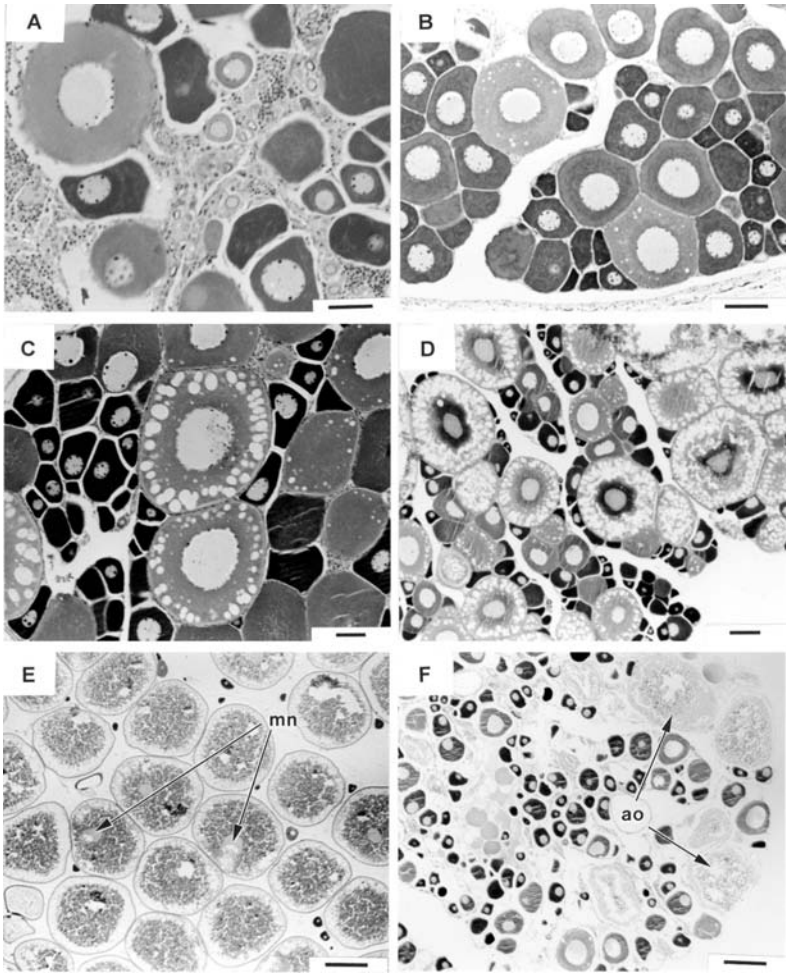

Fig. 4. Transverse sections through gonads of *Labeo cylindricus* illustrating the various microscopic stages of gametogenesis within the various macroscopic ovarian stages. **A**, 'juvenile' dominated by previtellogenic, perinuclear oocytes; **B**, 'resting' few primary vesicle oocytes are visible within a previtellogenic ovary; **C**, 'recovering' larger cortical alveoli are visible within the primary vesicle oocytes, giving the macroscopic gonad a grainy appearance; **D**, **E**, 'ripe' secondary yolk vesicle oocytes mark the beginning of vitellogenesis with the ovaries enlarging considerably in size. Prior to spawning most oocytes have sequestrated exogenous yolk globules and have reached the tertiary yolk vesicle stage and are of a similar size. The final oocyte stage before ovulation is when the nucleus migrates (mn) to the periphery of the cytoplasm; **F**, 'spent' – atretic secondary and tertiary yolk vesicle oocytes (ao) are visible throughout the ovary. All sections stained using haematoxylin and eosin. Scale bars: A, 60 µm; B, 100 µm; C, 50 µm; D, 200 µm; E, 500 µm.

classified into six phases or periods according to the state of oocyte growth: oogenesis, primary oocyte growth, cortical alveolus stage, vitellogenesis, maturation and ovulation. The macroscopic staging criteria for *L. cylindricus* allowed for the distinction between the phases to be made. It must be noted that maturation and ovulation are often rapid within synchronous spawners (Wallace & Selman 1981; Tyler & Sumpter 1996), and these stages were not macroscopically or microscopically noted during the study. Greater differences existed between the histological and macroscopical appearance of *L. cylindricus* testes. While the testes exhibited marked microscopical changes, these were less pronounced than the microscopical changes of oogenesis. As a result, little seasonal change was observed macroscopically within the testes throughout the year. It was for these reasons that male maturity and spawning patterns derived from macroscopical

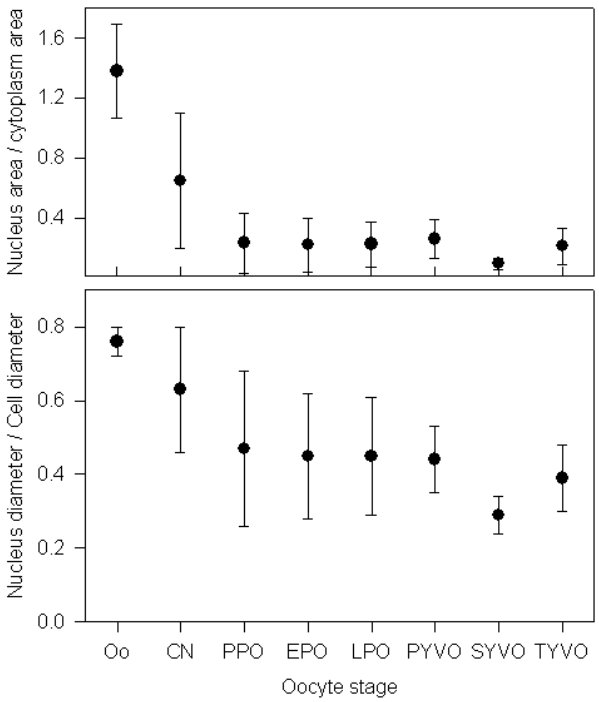

Fig. 5. Oocyte nucleus diameter/cell diameter and nucleus area/cytoplasm area ratios (mean ± standard deviation) during various stages of oogenesis in *Labeo cylindricus.* Oo = oogonia, CN = chromatin nucleolus oocytes, PPO = pre-perinuclear oocytes, EPO = early-perinuclear oocytes, LYVO = late-perinuclear oocytes, PYVO = primary yolk vesicle oocytes, SYVO = secondary yolk vesicle oocytes, TYVO = tertiary yolk vesicle oocytes.

methods need be viewed with caution. Overall, *L. cylindricus* gametogenesis was qualitatively similar to that of other freshwater teleost species (de Vlamming 1972; Wallace & Selman 1981; van der Merwe *et al.* 1988; Tyler & Sumpter 1996).

In Lake Chicamba, gonadal recrudescence first became evident in October, with increasing water temperatures after the winter months probably being the primary stimulant. The gonads matured rapidly and by January all female fish were ripe with vitellogenic oocytes and a high gonadosomatic index. Although ripe fish were recorded in November, at the start of the rainy season, the high river discharge rates and concomitant flooding of the lakes shoreline occurred in January, by which time all the *L. cylindricus* sampled were in a 'ripe' condition. By February, most of the females had spawned and were in a 'spent' condition, exhibiting evidence of atresia of vitellogenic oocytes. Few 'ripe' fish were recorded in the population after March. Both the macroscopic and histological examination of the ovaries revealed that all oocytes (excluding the perinuclear oocytes) were of a similar size and were in the final stages of vitellogenesis. During the winter period there was only evidence of atresia in vitellogenic oocytes, which is common in synchronously spawning fish (Wallace & Selman 1981; West 1990; Tyler & Sumpter 1996). These data together with a short spawning period and the rapid decrease in gonadosomatic index from *c.* 30 % to *c.* 5 %, provides further evidence of a synchronous spawning pattern, with all oocytes being ovulated and spawned during the annual fish migration up the rivers. The remaining perinuclear oocytes would develop and mature during the following summer, providing another batch of vitellogenic oocytes. The mitotic division of oogonia embedded in the ovigerous lamellae would in turn, replace these oocytes. This pattern of oocyte development is characteristic of iteroparous spawning fish (*cf.* semelparous – West 1990), classifying *L. cylindricus* as a synchronous iteroparous spawner, spawning almost all its matured

gametes over a short period, annually throughout its life-time.

The need for biological monitoring, including the continued assessment of reproductive patterns of fish populations, especially if they are commercially important, in order to understand possible life-history changes is imperative (Booth & Hecht 1997). A thorough understanding of the reproductive patterns of a species, including its reproductive seasonality and patterns of maturation, provide critical information for stock assessment models and other management-related issues such as the setting minimum size limits. For this to be achieved successfully, reproductive studies based on a combination of macroscopic staging, gonadosomatic indices and histological investigation must be applied.

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REFERENCES

BOOTH, A.J. & HECHT, T. 1997. Description of gametogenesis in the panga *Pterogymnus laniarius* (Pisces: Sparidae) with comments on changes in maturity patterns over the past two decades. *South African Journal of Zoology* **32**: 49–53.

DE VLAMMING, V.L. 1972. Reproductive cycling in the estuarine gobiid fish, *Gillichthys mirabilus. Copeia* **1972**: 278–291.

DE VLAMMING, V., GROSSMAN, G. & CHAPMAN, F. 1982. On the use of the gonadosomatic index. *Comparative Biochemistry and Physiology* **73A**: 31–39.

GAIGHER, I.G. 1983. Reproduction of *Labeo umbratus* (Pisces, Cyprinidae) in Wuras Dam, a shallow, turbid impoundment. *South African Journal of Zoology* **19**: 105–108.

REID, G. M. 1985. A revision of African species of *Labeo* (Pisces: Cyprinidae) and a redefinition of the genus. *Thesis Zoologicae,* Vol. 6. J. Cramer Verlag, Braunschweig.

SIDDIQUI, A.Q., CHATTERJEE, A. & KHAN, A.A. 1976. Reproductive biology of the carp, *Labeo bata* (Ham.) from the River Kali, India. *Aquaculture* **7**: 181–191.

SKELTON, P.H. 1993. *A Complete Guide to the Freshwater Fishes of Southern Africa.* Southern Book Publishers, Halfway House.

SKELTON, P.H., TWEDDLE, D. & JACKSON, P.B.N. 1991. Cyprinids of Africa. In: *Cyprinid Fishes: Systematics, Biology and Exploitation,* (eds) I.J. Winfield & J.S. Nelson, pp. 211–239. Chapman & Hall, London.

TYLER, C.R & SUMPTER, J.P. 1996. Oocyte growth and the development in teleosts. *Reviews in Fish Biology and Fisheries* **6**: 287–318.

VAN DER HORST, G. & ERASMUS, T. 1978. The structure of the testis of the mullet *Liza dumerili* (Teleostei: Mugilidae) with special reference to spermatogenesis. *Zoologica Africana* **13**: 233–243.

VAN DER MERWE, W., VAN VUUREN, J.H.J. & VERMAAK, J.F. 1988. Cyclic histomorphological changes in the ovary of mudfish, *Labeo capensis. Aquaculture* **72**: 349–358.

VAN ZYL, B.J., HAY, C.J. & STEYN, G.J. 1995. Some aspects of the reproduction biology of *Labeo capensis* (Smith, 1941) (Pisces, Cyprinidae) in relation to exploitation and extreme environmental conditions in Hardap Dam, Namibia. *South Africa Journal of Aquatic Science* **21**: 88–98.

WALLACE, R.A. & SELMAN, K. 1981. Cellular and dynamic aspects of oocyte growth in teleosts. *American Zoologist* **21**: 325–343.

WEST, G. 1990. Methods of assessing ovarian development in fishes: a review. *Australian Journal of Marine and Freshwater Research* **41**: 199–222.

WEYL, O.L.F & BOOTH, A.J. 1999. On the life history of a cyprinid fish, *Labeo cylindricus. Environmental Biology of Fishes* **55**: 215–225.