12) RAPID # -409276

BORROWER AUM

System Date/Time: 03/24/2004 12:52:42

CALL #: QH301 .R43
LOCATION: Biology

TYPE: Article
USER JOURNAL TITLE: Revista Brasileira de biologia
OCLC JOURNAL TITLE: "Revista Brasileira de biologia"
LDL CATALOG TITLE: Revista Brasileira de biologia
ARTICLE TITLE: Alves: Gonadal differentiation of Schizodon knerii Steindachner, 1875 (Pisces, Anostomidae).
VOLUME: 47
NO: 1-2
YEAR: 1987(1987)
COPYRIGHT: CCL
PAGES: 237-241
ISSN: 0034-7108
OCLC #: 1645004
VERIFIED:

PATRON: Godinho,Alexandre
PATRON ID: 0
PATRON PHONE:
PATRON DEPT: Natural Resources Conservation
PATRON STATUS: DoctoralCandidate
PATRON FAX:
PATRON ADDRESS:
PATRON E-MAIL:
PATRON NOTES:

Rev. Brasil. Biol., 47(1/2):237-241
Fevereiro/Maio, 1987 - Rio de Janeiro, RJ

GONADAL DIFFERENTIATION OF *SCHIZODON KNERII* STEINDACHNER, 1875 (PISCES, ANOSTOMIDAE)

MARLENE D. ALVES and HUGO P. GODINHO
Department of Morphology, Institute of Biological Sciences,
Federal University of Minas Gerais, 31270 Belo Horizonte, MG

(With 5 figures)

RESUMO

Diferenciação gonádica de *Schizodon knerii* Steindachner, 1875 (Pisces, Anostomidae)

A diferenciação gonádica do peixe leporinídeo *Schizodon knerii,* proveniente da represa de Três Marias no rio São Francisco, foi estudada em exemplares medindo 3,0 a 14,0 cm de comprimento total (C_t). As gônadas deste peixe diferenciam-se diretamente em ovários ou testículos. A diferenciação ovárica ocorre em exemplares com 6,5 – 8,9 cm, C_t. Ovários juvenis estão presentes naqueles que medem $\geqslant$12,8 cm C_t. A diferenciação testicular inicia-se em peixes com 8,1 – 10,5 cm, C_t, e testítulo adulto aparece em peixes $\geqslant$13,2 cm, C_t.

Palavras-chave: Peixe, *Schizodon knerii,* diferenciação gonádica.

ABSTRACT

The gonadal differentiation of the leporinid fish *Schizodon knerii* from the Três Marias Reservoir, in the San Francisco river, was studied in specimens measuring from 3.0 to 14.0 cm, total lenght (L_t). The gonads of this fish differentiate directly towards either ovaries or testes. Ovarian differentiation occurs in fish measuring 6.5 – 8.9 cm, L_t, and juvenil ovaries are present in those measuring $\geqslant$12.8 cm, L_t. Testicular differentiation starts in fish measuring 8.1 – 10.5 cm, L_t, and adult testis appears in 13.2 cm, L_t.

Key-words: Fish, *Schizodon knerii,* gonadal differentiation.

INTRODUCTION

The Três Marias Reservoir resulted from the dam built in the San Francisco River, southeastern Brazil, in 1961, with a surface area of 114,000 ha. Despite the impacts that the new lacustrine water body have certainly provoked on the San Francisco potamodromic fish, only recently studies were initiated on the reproductive biology of those species (Godinho, 1984).

In this paper we present data on the gonadal differentiation and sexual maturity of the *Schizodon knerii,* an indigenous leporinid fish from the San Francisco River and one of the most important in the reservoir fishery (Sato and Barbieri, 1983).

Received March 3, 1986
Accepted October 3, 1986
Distributed May 30, 1987

MATERIAL AND METHODS

Thirty-eight especimens of *Schizodon knerii,* measuring 3.0 – 14.0 cm, total lenght (L_t), were used

in the present study. They were collected in the Três Marias Reservoir at the San Francisco River, in the State of Minas Gerais, Brazil (between 18° 45' S and 18° 30' S), during the period of March to June, 1983.

L_t and body weight of the specimens were recorded. After opening their abdominal cavity, they were dipped into Bouin's liquid during 3 to 24 h. In specimens measuring up to 5.2 cm, L_t, the gonads were removed with the corresponding segment of the body wall, gaseous bladder and kidney. In larger specimens the gonads were easily dissected out after Bouin's fixation. Gonad fragments were dehydrated in ethyl alcohol, embbeded in 2-hydroxyethyl methacrylate, cut at 3 μm of thickness and stained with 1% toluidine blue. The germinal cells were identified and measured. Observations about the distribution of their nuclear material and nucleolar appearance were also made.

RESULTS

S. knerii measuring 3.0 – 5.2 cm, L_t, showed undifferentiated gonads situated dorsolaterally in the peritoneal cavity along with the gaseous bladder. These gonads contained few primordial germ cells (Fig. 1, PG) which were gradually surrounded by flat somatic cells (Fig. 1, S; Fig. 2, S) during fish growth. The PG cells were round or ovoid in shape. Their cytoplasm was scanty and faintly stained, and the nucleus showed one or more nucleoli and chromatin material scattered in the nucleoplasm or concentrated near the nuclear envelope. The nucleus of the PG cells measured 8.5 ± 0.6 μm in diameter.

Near differentiation (fish measuring 5.3 – 7.1 cm, L_t), granules of dense material appeared in the cytoplasm of the PG cells (fig. 2, arrows). Their nucleus showed the chromatin material close to the nuclear envelope and the nucleoli with central vacuolization.

Female gonadal differentiation appeared in fish measuring 6.5 – 8.9 cm, L_t. Their gonads exhibited oocytes at different phases of the meiotic prophase, corresponding to the chromatin-nucleolus and early perinucleolar stages. The morphological characteristics of the oogonia (Fig. 3,0) in these fish were similar to those of the PG cells near differentiation. The peripheral nuclear material of the oogonia seemed to flow towards the cytoplasm where it could not be distinguished from the dense cytoplasmic particles. The chromatin-nucleolus stage encompassed zygotene and pachytene phases. During zygotene (Fig. 3, Z), the chromosomes were arranged at one of the nuclear poles, and at pachytene (Fig. 3, P), they were dispersed in the nucleoplasm. The cell limits at zygotene were not distinguishable. During this stage the nucleus of the germ cells increased sligthly. Groups of oogonia and young oocytes (zygotene and pachytene), bounded by flat cells, were seen in clusters at a same stage of development.

In the early perinucleolar stage (diplotene phase), the oocyte nucleoli increased in number and occupied a peripheral position under the nuclear envelope. They were initially halfmoon shaped and gradually became rounded. The size of the oocyte increased during this stage (Fig. 3, D_1, D_2) at more pronunced rate than that at the chromatin-nucleolus stage. The oocyte cytoplasm became gradually basophyl and showed an yolk nucleus situated close to the nuclear envelope. Lump-brush chromosomes were seen in the largest oocytes of this stage. Juvenil ovaries occurred in specimens ⩾12.8 cm, L_t (Fig. 4).

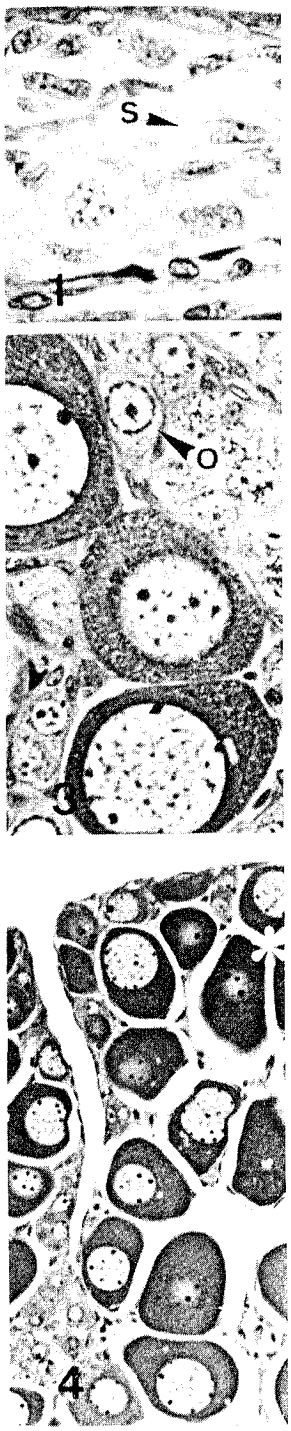
Testicular differentiation was observed at 8.1-10.5 cm, L_t. In these fish, the spermatogonia were arranged in clumps along the main axis of the testis. They were surrounded by somatic (Sertoli) cells and had similar morphological characteristics of the PG cells near differentiation. Mitotic figures of spermatogonia were rare although these cells were more abundant than the PG cells in undifferentiated gonads. Testes of specimens measuring ⩾13.2 cm, L_t, presented adult morphology (Fig. 5).

DISCUSSION

The morphology of the PG cells of *Schizodon knerii* corresponds to that of other teleost fish (Satoh and Egami, 1972; Hurk and Slof, 1981; Lebrun *et alii*, 1982; Roblin and Bruslé, 1983). The arrangement of the somatic cells around the germ cells is similar to that found by Yamamoto and Onozato (1965), Combs (1969) and Forberg (1982). Morphological distinction among PG cells, spermatogonia and oogonia (Satoh and Egami, 1972; Bruslé and Bruslé, 1978; Hurk and Slof, 1981; Lebrun *et alii*, 1982) could not be made in our fish similarly to the findings of Roblin and Bruslé (1983) in the sea bass.

The gonads of *S. knerii* differentiate directly towards either ovary or testis, with ovarian differentiation preceeding the testicular one. Ovaries were found in specimens measuring from 6.5 cm, L_t, whereas testes were only observed in fish 8.0 cm, L_t. Earlier ovarian differentiation has also been reported in other teleosts (Satoh and Egami, 1972; Bruslé and Bruslé, 1978; Hurk and Slof, 1981; Lebrun *et alii*, 1982).

The initial oocyte growth is commonly divided into chromatin-nucleolus and perinucleolar stages (Yamamoto and Yamazaki, 1961; Yamamoto and Onozato, 1965; Rai, 1967; Lehri, 1968; Tokarz, 1978; Guraya *et alii*, 1975; Wallace and Selman,



Figs. 1-5 – Are lightmicrographs primordial germ (PG) and somatic (S) cells cytoplasmic granules (thick arrows) and su (O), oocytes at chromatin-nucleolus stage (D_1 and D_2 at different growth developmer perinucleolar stage within an ovigerous la development. (x250)

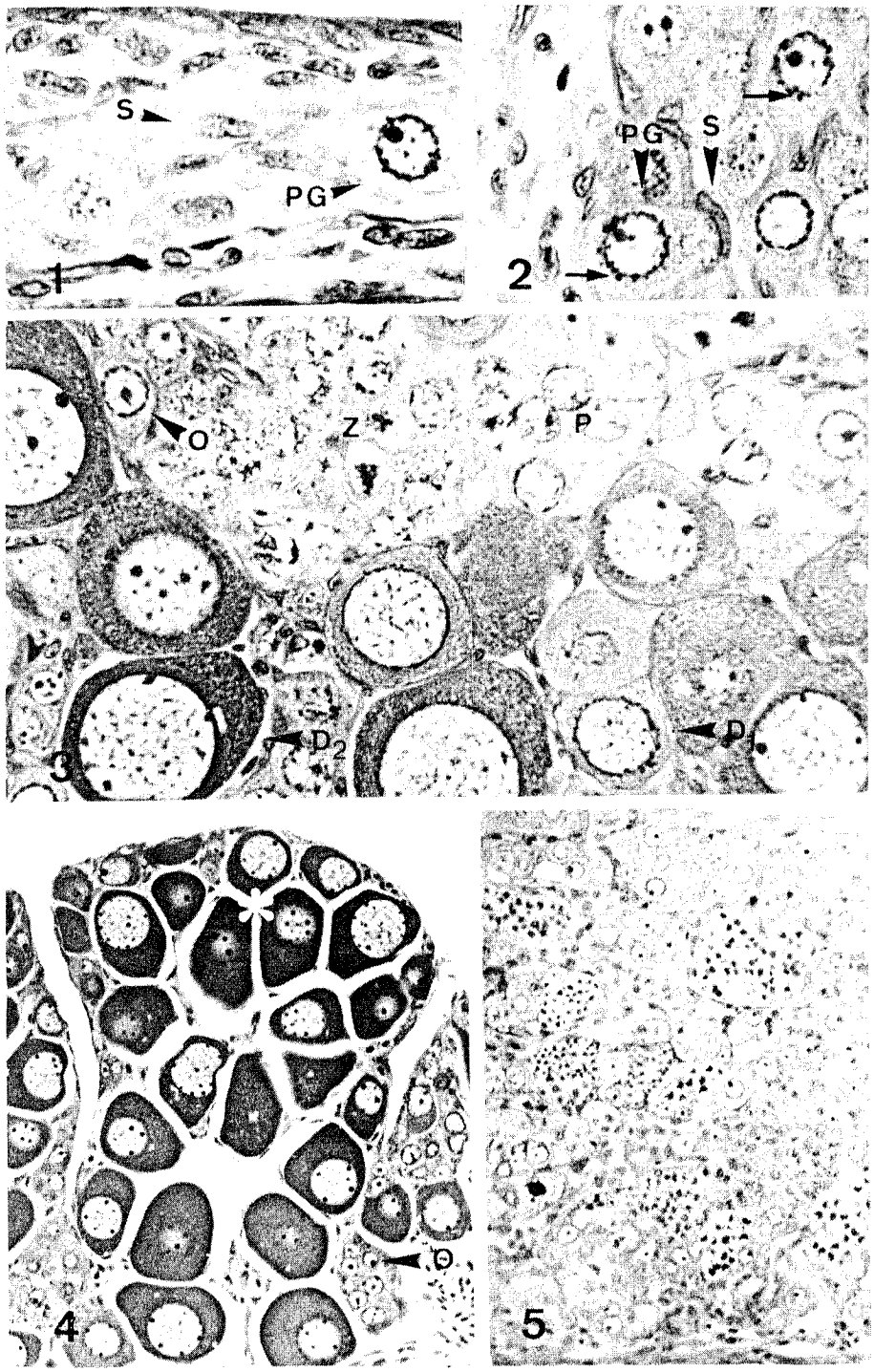
)uring this stage the nucleus of the
ied sligthly. Groups of oogonia and
ygotene and pachytene), bounded
e seen in clusters at a same stage of

ly perinucleolar stage (diplotene
e nucleoli increased in number and
pheral position under the nuclear
were initially halfmoon shaped and
e rounded. The size of the oocyte
this stage (Fig. 3, D_1, D_2) at more
han that at the chromatin-nucleolus
yte cytoplasm became gradually
owed an yolk nucleus situated close
nvelope. Lump-brush chromosomes
largest oocytes of this stage. Juvenil
in specimens ⩾12.8 cm, L_t (Fig. 4).
differentiation was observed at
t. In these fish, the spermatogonia
n clumps along the main axis of the
re surrounded by somatic (Sertoli)
nilar morphological characteristics of
ar differentiation. Mitotic figures of
were rare although these cells were
than the PG cells in undifferentiated
of specimens measuring ⩾13.2 cm,
lult morphology (Fig. 5).

DISCUSSION

hology of the PG cells of *Schizodon*
ids to that of other teleost fish (Satoh
72; Hurk and Slof, 1981; Lebrun *et*
Roblin and Bruslé, 1983). The
f the somatic cells around the germ
to that found by Yamamoto and
), Combs (1969) and Forberg (1982).
distinction among PG cells,
and oogonia (Satoh and Egami, 1972;
ruslé, 1978; Hurk and Slof, 1981;
1982) could not be made in our fish
findings of Roblin and Bruslé (1983)

ads of *S. knerii* differentiate directly
r ovary or testis, with ovarian
preceeding the testicular one. Ovaries
specimens measuring from 6.5 cm, L_t
were only observed in fish 8.0 cm.
arian differentiation has also been
her teleosts (Satoh and Egami, 1972;
ruslé, 1978; Hurk and Slof, 1981;
1982).
ial oocyte growth is commonly divided
in-nucleolus and perinucleolar stages
nd Yamazaki, 1961; Yamamoto and
55; Rai, 1967; Lehri, 1968; Tokarz
a *et alii*, 1975; Wallace and Selman



Figs. 1-5 – Are lightmicrographs of *S. knerii* gonads stained with 1% toluidine blue; 1 – Undifferentiated gonad: primordial germ (PG) and somatic (S) cells. (x940); 2 – Gonad near differentiation: primordial germ cell (PG) showing dense cytoplasmic granules (thick arrows) and surrounded by somatic cell (S). (x940); 3 – Ovary just after differentiation: oogonia (0), oocytes at chromatin-nucleolus stage (zygotene, Z; pachytenes in cluster, P) and at early perinucleolar stage (diplotenes, D_1 and D_2 at different growth development). (x625); 4 – Juvenil ovary showing clusters of oogonia (0) and oocytes at early perinucleolar stage within an ovigerous lamella . (x375); 5 – Testis with cysts of germ cells at different stages of germ cell development. (x250)

1981; Forberg, 1982). The chromatin-nucleolus stage may still be subdivided into presynaptic, synaptic and postsynaptic substages (Lehri, 1968; Tokarz, 1978; Forberg, 1982). Zygotene and pachytene figures corresponding respectively to the synaptic and postsynaptic substages were registered in *S. knerii* (present study). However, the presynaptic substage, which marks the transition between oogonia and the initial phase of the meiotic prophase, did not exhibit evident morphological arrangement in this species. The appearance of small nucleoli close to the nuclear envelope (Rai, 1967; Lehri, 1968; Guraya *et alii*, 1975) and lump-brush chromosomes (Rai, 1967; Tokarz, 1978; Lehri, 1968; Wallace and Selman, 1981) were the elements used to identify the perinucleolar stage in *S. knerii*. At this stage its oocytes increased in size, mainly in females >7.5 cm, L_t.

At the beginning of the meiotic prophase, the cytoplasm of *S. knerii* oocytes built up aggregations of dense basophylic material. According to Toury *et alii* (1979) and Wallace and Selman (1981), such a material ("nuage") reaches the cytoplasm through the pores of the nuclear envelope and is constituted by ribonucleoprotein particles. The organization of cytoplasmic organelles coincides with the passage of material from the nucleus to the cytoplasm suggesting that the nucleus actively participates in this process (Zahnd and Porte, 1966). The number of mitochondria increases considerably during the meiotic prophase and are organized in clusters near the nuclear envelope amongst the dense intermitochondrial cement, forming the yolk nucleus or Balbiani body (Toury *et alii*, 1979; Wallace and Selman, 1981). The yolk nucleus of *S. knerii* was present at the perinucleolar stage as observed in other fish (Yamamoto and Onozato, 1965; Rai, 1967; Lehri, 1968; Guraya *et alii*, 1975; Forberg, 1982). The dense cytoplasmic granules observed in the PG cells near differentiation possibly originated in the nucleus and were associated with the presence of vacuolated nucleoli. This vacuolization may reflect an increase in the synthesis and exportation of nucleolar material mainly when the vacuole is centrally located in the nucleolus (Azevedo and Coimbra, 1980). The dense material found at the nuclear membrane and at the perinuclear region of the cytoplasm might constitute the germinal plasm (germinal factor), characteristic of the vertebrate germinal cells (Kerr and Dixon, 1974).

Females *S. knerii* reach sexual maturity later than males despite presenting earlier gonadal differentiation. This statement is based on unpublished work about the reproductive cycle of this species, conducted also at Três Marias Reservoir, in which we exammined 35 females measuring 14.2 – 22.0 cm L_t. Ovarian maturation occurred in 5 specimens which measured from 18.6 to 21.7 cm, L_t.

Acknowledgments – We thank "CNPq" and "FINEP" for their financial support and to Mrs. A.A.L. Friche for her skilfull technical assistance. Thanks are also due to the staff of Três Marias Fishery Station of CODEVASF, at Três Marias, MG, for the fish capture.

REFERENCES

AZEVEDO, C. and COIMBRA, A., 1980, Evolution of nucleoli in the course of oogenesis in a viviparous teleost (*Xiphophorus helleri*). *Biol. Cell.*, 38: 43-48.

BRUSLÉ, S. and BRUSLÉ, J., 1978, Early sex differentiation in *Mugil (Liza) auratus* Risso, 1810 (Teleost, Mugilidae). An ultrastructural study. *Ann. Biol. Anim. Bioch. Biophys.*, 18: 871-875.

COMBS, R.M., 1969, Embryogenesis, histology and organology of the ovary of *Brevoortia patronus*. *Gulf Res. Rep.*, 2(4): 335-429.

FORBERG, K.G., 1982, A histological study of development of oocytes in capelin, *Mallotus villosus villosus* (Müller). *J. Fish Biol.*, 20: 143-154.

GODINHO, H.P., 1984, Reprodução dos peixes da represa de Três Marias. *Inf. Agropec. Belo Horizonte*, 10(110): 29-34.

GURAYA, S.S., KAUR, R. and SAXENA, P.K., 1975, Morphology of ovarian changes during the reproductive cycle of the fish, *Mystus tengara* (Ham.). *Acta. Anat.*, 91: 222-260.

HURK, R. v.d. and SLOF, G.A., 1981, A morphological and experimental study of gonadal sex differentiation in the rainbow trout, *Salmo gairdneri*. *Cell Tiss. Res.*, 218: 487-497.

KERR, J.B. and DIXON, K.E., 1974, An ultrastructural study of germ plasm in spermatogenesis of *Xenopus laevis*. *J. Embryol. Exp. Morphol.*, 32: 573-592.

LEHRI, G.K., 1968, Cyclical changes in the ovary of the catfish *Clarias batrachus* (Linn.). *Acta Anat.*, 69: 105-124.

LEBRUN, C., BILLARD, R. and JALABERT, B., 1982, Changes in the number of germ cells in the gonads of the rainbow trout (*Salmo gairdneri*) during the first 10 post-hatching weeks. *Reprod. Nutr. Dévelop.*, 22(2): 405-412.

RAI, B.P., 1967, Cyclical changes in the ovary of *Tor (Barbus) tor* (Ham.). *Acta Zool.*, 48: 289-307.

ROBLIN, C. and BRUSLÉ, J., 1983, Ontogenèse gonadique et différenciation sexuelle du Loup *Dicentrarchus labrax*, en conditions d'élevage. *Reprod. Nutr. Dévelop.*, 23(1): 115-127.

SATO, Y. and BARBIERI, G., 1983, Crescimento de *Schizodon knerii* Steindachner, 1875 (Pisces, Anostomidae) na represa de Três Marias, Minas Gerais. *III Seminário Regional de Ecologia, Universidade Federal de São Carlos*, São Carlos, S.P., Anais, p. 201-221.

SATOH, N. and EGAMI, N., 1972, Sex differentiation of germ cells in the teleost, *Oryzias latipes*, during normal embryonic development. *J. Embryol. Exp. Morph.*, 28: 385-395.

TOKARZ, R.R., 1978, The vertebrate ovary (R.E. Jones, ed.). Plenun Press, New York, p. 145.

TOURY, R., CLEROT, J.-C. and ANDRE, J., 1979, Les groupements mitochondriaux des cellules germinales des poissons téléostéens cyprinidés. *Biochimie*, 61: 405-413.

WALLACE, R.A. and SELMAN, K. dynamic aspects of oocyte gr *Zool.*, 21; 325-343.

YAMAMOTO, K. and YAMAZAKI development in the oocyte of *auratus*. *Bull Fac. Fish. H* 93-110.

YAMAMOTO, D. and ONOZATO

pecimens which measured from 18.6

gments – We thank "CNPq" and "FINEP" support and to Mrs. A.A.L. Friche for her assistance. Thanks are also due to the staff Fishery Station of CODEVASF, at Três he fish capture.

REFERENCES

and COIMBRA, A., 1980, Evolution of n the course of oogenesis in a viviparous phophorus helleri). *Biol. Cell.*, 38: 43-48.

and BRUSLÉ, J., 1978, Early sex ition in *Mugil (Liza) auratus* Risso, 1810 Mugilidae). An ultrastructural study. *Ann. Bioch. Biophys.*, 18: 871-875.

, 1969, Embryogenesis, histology and y of the ovary of *Brevoortia patronus*. *Gulf* 2(4): 335-429.

.G., 1982, A histological study of ent of oocytes in capelin, *Mallotus villosus* üller). *J. Fish Biol.*, 20: 143-154.

., 1984, Reprodução dos peixes da represa arias. *Inf. Agropec. Belo Horizonte*, 10(110):

, KAUR, R. and SAXENA, P.K., 1975, gy of ovarian changes during the ive cycle of the fish, *Mystus tengara* (Ham.). t., 91: 222-260.

id SLOF, G.A., 1981, A morphological and ital study of gonadal sex differentiation in ow trout, *Salmo gairdneri*. *Cell Tiss. Res.*, 497.

DIXON, K.E., 1974, An ultrastructural germ plasm in spermatogenesis of *Xenopus* mbryol. *Exp. Morphol.*, 32: 573-592.

968, Cyclical changes in the ovary of the *larias batrachus* (Linn.). *Acta Anat.*, 69:

ILLARD, R. and JALABERT, B., 1982, n the number of germ cells in the gonads of w trout (*Salmo gairdneri*) during the first 10 ing weeks. *Reprod. Nutr. Dévelop.*, 22(2):

7, Cyclical changes in the ovary of *Tor* or (Ham.). *Acta Zool.*, 48: 289-307.

and BRUSLÉ, J., 1983, Ontogenèse et différenciation sexuelle du Loup *hus labrax*, en conditions d'élevage. *utr. Dévelop.*, 23(1): 115-127.

BARBIERI, G., 1983, Crescimento de *knerii* Steindachner, 1875 (Pisces, dae) na represa de Três Marias, Minas *III Seminário Regional de Ecologia*, de Federal de São Carlos, São Carlos, S.P., 201-221.

EGAMI, N., 1972, Sex differentiation of ls in the teleost, *Oryzias latipes*, during mbryonic development. *J. Embryol. Exp.* 8: 385-395.

, 1978, The vertebrate ovary (R.E. Jones, n Press, New York, p. 145.

EROT, J.-C. and ANDRE, J., 1979, Les nts mitochondriaux des cellules germinales ons téléostéens cyprinidés. *Biochimie*, 61:

WALLACE, R.A. and SELMAN, K., 1981, Cellular and dynamic aspects of oocyte growth in teleosts. *Amer. Zool.*, 21; 325-343.

YAMAMOTO, K. and YAMAZAKI, F., 1961, Rhythm of development in the oocyte of the gold-fish *Carassius auratus*. *Bull Fac. Fish. Hokkaido Univ.*, 12(2). 93-110.

YAMAMOTO, D. and ONOZATO, H., 1965, Electron microscope study on the growing oocyte of the goldfish during the first growth phase. *Mem. Fac. Fish. Hokkaido Univ.*, 13(2): 79-106.

ZAHND, J.-P. et PORTE, A., 1966, Signes morphologiques de transfert de matériel nucléaire dans de citoplasme des ovocytes de certaines espèces de poissons. *C.R. Acad. Sc. Paris*, 262-D: 1977-1978.