Ovarian development in *Labeo dyocheilus* (McClelland) during active reproductive phase under captive and wild conditions

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Abstract: Ovarian development in *Labeo dyocheilus* was assessed during active reproductive phase under ambient environmental conditions in captivity and wild. Increasing day length and water temperature seemed favourable for ovarian development in female *L. dyocheilus* under both conditions. Gonadosomatic index (GSI) was lower in May and higher in July in captivity (6.168 and 13.366) and wild (5.798 and 16.166) respectively. Ovarian development started little bit in advance in captivity with late yolk vesicle stage oocyte in May while yolk globule stage oocytes were prominent in wild. Fully grown oocytes with germinal vesicle migration (GVM) and germinal vesicle breakdown (GVBD) stages were observed in July in both the conditions. Degree of transformation of developing oogonials into advance stage oocytes was observed to be better in wild fish compared to captivity reared ones. The histophysiological changes in liver corresponded well to the progression of ovarian development. Successive granulation and vacuolization of hepatocyte cytoplasm were indicative of augmented synthetic activity and probably mobilization of energy content for oocyte growth. These observations indicated that normal ovarian development of *L. dyocheilus* under captivity in Tarai region of Uttarakhand would be useful for success of its seed production in captivity for stock augmentation in wild or species diversification in aquaculture.

Key words: Ovary, Liver, Day length, Temperature, *Labeo dyocheilus*, Hepatosomatic Index, Gonadosomatic index

PDF of full length paper is available with author (*singhij2@gmail.com*)

Introduction

Success of reproduction dependents upon normal gonadal development stimulated by favorable environmental conditions. Photoperiod and temperature are most important factors for controlling the reproductive activities in most of the seasonally breeding teleosts (Lam, 1983; Shankar et al., 2007) including cyprinids (Hontela and Stacey, 1990). Gonadosomatic index (GSI) is generally used as a reliable criterion for expression of gonadal development and reproductive effort in fishes (Calow, 1979; Saksera, 1987). Hepatosomatic index (HSI) has been often used as indicator of energy status in relation to gonadal development and growth of fish (Wootten et al., 1978; Campbell and Love, 1978; Shankar and Kulkami, 2007).

Oogenesis is the process of transformation of primordial germ cells (PGCs) into ova, ready to be fertilized followed by embryonic development. Normally six oogenesis stages i.e. formation of PGCs, transformation of PGCs into oogonia and transformation of oogonia into oocytes (onset of meiosis), vitellogenic growth of oocytes while under meiotic arrest, resumption of meiosis (maturation) and expulsion of the ovum from its follicle (ovulation) are associated with gonadal development and maturation in several teleosts (Patino and Sullivan, 2002), whereas nine maturity stages were described for gonadal development in Mediterranean amberjack (Marino et al., 1995). Captive reared females failed to not only spawn, but also to complete vitellogenesis and oocyte maturation (Micale and Perdichizzi, 1994; Micale et al., 1996, 1999).

Failure of spawning in captivity reared fish was much related with the arrest of oocyte development at tertiary yolk stage as compared to wild fish (Lee and Yang, 2002).

Histological changes in liver (Olivereau and Olivereau, 1979; Medda et al., 1980; Tam et al., 1983; Singh et al., 2005) and HSI (Singh and Singh, 1983,1984 ; Singh et al., 2004) have been correlated with gonadal development and maturation in several female fishes. Similar information is not available for *L. dyocheilus*, hence, the present investigation was undertaken to assess its ovarian development under captive and wild conditions.

Materials and Methods

Adult live specimens of *L. dyocheilus*, collected during 2002 from Domunda and Marchula (river Ramganga) and Tiger top and Kosi barrage (river Kosi) of Uttarakhand hills, were brought to the College of Fisheries, Pantnagar and stocked in earthen ponds (captivity). Fish were fed @ 5% body weight twice daily (morning and evening) with conventional carp feed prepared by mixing rice bran and oil cake in the ratio of 1:1. Fish from wild were collected during May to August 2003 for study of gonadal development. For histological studies and assessing GSI and HSI levels in relation to ovarian development after recording body weight, ovary and liver samples from female specimens of *L. dyocheilus* were collected each month during May to August, 2003 from wild or reared in captivity. GSI and HSI were calculated as percentage of weight of ovary and liver in relation to body weight. Small portions of ovary
between 22.2 and 38.0°C. Total rainfall was recorded maximum in the month of August (573 mm) while it was minimum in May (46.2 mm). Average water temperature ranged between 21.1 and 27.9°C and no marked change was recorded in pH and dissolved oxygen of pond water. Meteorological data and water quality parameters like temperature, pH and dissolved oxygen in pond water are summarized in Table 1.

The gonadal development was almost similar in female of L. dycoelius reared in captivity or collected from wild during May to August as reflected by changes in GSI values (Table 2). GSI increased gradually from May to July and decreased in August in females of both groups. GSI was higher in captive reared female in May but from June to August it was significantly higher (p<0.01) in wild collected females compared to captive reared ones. GSI ranged from 6.168 to 13.366 in captive reared and 5.798 to 16.166 in wild groups. Monthly variations in GSI were also significant (p<0.01) in both the groups but these changes exhibited insignificant correlation with rearing conditions (Table 3).

The HSI showed negative relationship with GSI in female of both groups (Table 2). HSI was lowest in May (0.620) in captive reared fish and highest in June (0.760) and in wild collected females it was lowest (0.576) in July and highest (0.868) in August. A gradual decrease in HSI was observed from May to July in wild collected females which increased sharply in August. HSI differed significantly (p<0.01) between wild and captive reared females and also varied significantly (p<0.01) in relation to months in both the groups. It was higher in June and July in captive stock and in May and August in wild ones. The difference in HSI of both groups showed significantly (p<0.05) correlation between captive and natural habitat conditions (Table 3).

In the month of May, ovary in captive reared female was dominated by late yolk vesicle stage oocyte along with perinucleolar

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### Table 1: Meteorological parameters and physico-chemical characteristics of pond water during May to August, 2003

<table>
<thead>
<tr>
<th>Month</th>
<th>Meteorological parameters at Pantnagar</th>
<th>Physico-chemical characteristics of pond water</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Range and average of max. temp. (°C)</td>
<td>Temperature (°C)</td>
</tr>
<tr>
<td></td>
<td>Range and average of min. temp. (°C)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Total rainfall (mm)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Day length (hr)</td>
<td></td>
</tr>
<tr>
<td>May</td>
<td>36.0-41.1 (38.0)</td>
<td>26.8</td>
</tr>
<tr>
<td>June</td>
<td>32.3-37.3 (34.3)</td>
<td>27.2</td>
</tr>
<tr>
<td>July</td>
<td>29.3-37.5 (33.0)</td>
<td>27.9</td>
</tr>
<tr>
<td>Aug.</td>
<td>30.9-33.1 (31.8)</td>
<td>26.8</td>
</tr>
</tbody>
</table>

Source: Department of Agro-Meteorology, College of Agriculture, GBPUA and T, Pantnagar

### Table 2: Correlative changes in GSI and HSI in adult female of L. dycoelius reared in captivity and collected from wild

<table>
<thead>
<tr>
<th>Group</th>
<th>Parameters</th>
<th>May</th>
<th>June</th>
<th>July</th>
<th>August</th>
</tr>
</thead>
<tbody>
<tr>
<td>Captive</td>
<td>GSI</td>
<td>6.168 ±0.420</td>
<td>11.192 ±1.892</td>
<td>13.366 ±0.919</td>
<td>10.156 ±1.166</td>
</tr>
<tr>
<td></td>
<td>HSI</td>
<td>0.620 ±0.054</td>
<td>0.760 ±0.101</td>
<td>0.675 ±0.094</td>
<td>0.733 ±0.016</td>
</tr>
<tr>
<td>Wild</td>
<td>GSI</td>
<td>5.798 ±0.264</td>
<td>12.102 ±2.341</td>
<td>16.166 ±1.821</td>
<td>13.573 ±1.432</td>
</tr>
<tr>
<td></td>
<td>HSI</td>
<td>0.696 ±0.006</td>
<td>0.638 ±0.061</td>
<td>0.576 ±0.010</td>
<td>0.868 ±0.043</td>
</tr>
</tbody>
</table>

Values are mean ± SD

### Table 3: ANOVA-matrix for GSI and HSI of captivity reared and wild collected specimens of L. dycoelius

<table>
<thead>
<tr>
<th>Source / Index</th>
<th>GSI</th>
<th>HSI</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>DF</td>
<td>SS</td>
</tr>
<tr>
<td>Treatment (a)</td>
<td>1.0</td>
<td>17.115</td>
</tr>
<tr>
<td>Factors (b)</td>
<td>3.0</td>
<td>243.016</td>
</tr>
<tr>
<td>(a*b)</td>
<td>3.0</td>
<td>13.598</td>
</tr>
</tbody>
</table>

a = Captive and wild conditions, b = Different months, a*b = Correlation between a and b
ns = Non-significant, * = Significant at 5%, ** = Significant at 1%
and early yolk vesicle stage oocytes. Oogonials and immature oocytes were also present in moderate number (Fig. 1) while the ovary of female collected from wild possessed significant number of yolk globule stage oocytes along with a few oogonials (Fig. 2). In June ovaries were full of late yolk vesicle and yolk globule stage oocytes with randomly distributed cortical alveoli in cytoplasm (Fig. 3) in female reared in captivity while yolk globule stage oocytes with GVM and GVBD were observed in female of wild group ovary (Fig. 4). Peak ovarian development with dominance of fully grown/ maturation stage oocytes with well developed follicular layer were recorded in ovary of female of captive group in July (Fig. 5). Oocytes exhibited similar developmental events in wild group also in July (Fig. 6). In August, ovaries had persistent dominance of fully grown oocytes packed with yolk mass and disappearance of cortical alveoli in females of captive group (Fig. 7) while in wild group fully grown oocytes advanced towards ovulatory stage (Fig. 8).

Hepatocytes in liver exhibited active biosynthetic stage packed with cytoplasmic content in both captive reared and wild collected females in May (Fig. 9, 10). In June, hepatocytes in captive group showed cytoplasmic vacuolization attributable to mobilization of cytoplasmic content (Fig. 11) but in wild group mobilization of energy reserve seemed slightly delayed (Fig. 12). In July liver cells of females of both, captive and wild groups were in exhaustive...
condition as indicated by degranulation and decreased nucleolar size (Fig. 13, 14). However, reorganization of liver cells with darkly stained residual synthetic material was evident in females reared in captivity indicating initiation of growth phase (Fig. 15) but exhaustive condition still persisted in liver cells of female collected from wild (Fig. 16).

In most of monsoon breeding teleosts especially cyprinids progressive stages of gonadal development in Indian subtropical regions are well correlated with environmental cues particularly increasing daylength and temperature. In female *L. rohita* from either captive or wild conditions, increasing level of gonadal development appeared positively correlated with increasing daylength and temperature till ovarian maturity but lowering of temperature associated with rainfall seemed to be crucial factor for final oocyte maturation and subsequent processes. A positive relationship between increasing temperature and daylength with gonadal development during preparatory and prespawning phases and fall of temperature due to rainfall associated with upsurge in gonadotropin level during spawning phase has been reported for the Indian major carp, *Cirrhinus mrigala* (Singh and Singh, 1984). Like wise gonadal development and maturation in an other major carp, *Labeo rohita* was found to be influenced positively by increasing day length and temperature (Kumar et al., 2003; Singh et al., 2005).

The gonadosomatic index (GSI) is correlated positively with gonadal development and more often is used as reliable indicator of its maturity (Rae and Calvo, 1995). Hepatosomatic index, used
as indicator of energy status (Wootton et al., 1978), has been observed to exhibit negative correlation with GSI (Singh and Singh, 1983, 1984; Kumar et al., 2001; Singh et al., 2005) suggesting mobilization of energy stored from liver to ovary for egg production (Rae and Calvo, 1995). In L. dyocheilus female also, like other cyprinids of the region, HSI exhibited negative correlation with changes in GSI in specimens of both groups despite having variation in monthwise level of changes between the groups.

Transformation of immature and less developed oocytes into developed and mature stage oocytes is associated with the accumulation of lipid globules in preparatory and prespawning seasons. Failure of completion of vitellogenesis and oocyte maturation (Micale and Perdichizzi, 1994; Micale et al., 1996, 1999) and arrest of oocyte development in tertiary yolk stage (Lee and Yang, 2002) were reported in some captive reared female fishes. Despite slight variation with regard to timing the presence of developmental and maturational oocyte stages particularly yolk vesicle and globule stages, yolk globule stage oocytes with GVM and GVBD in both captive and wild group of specimens of L. dyocheilus clearly demonstrated that ovarian development in this fish was normal under captive conditions. Similar sequences of oocyte development processes were reported in the Indian major carp, L. rohita in captivity under similar ambient environmental conditions (Singh et al., 2005). Conversion of oocyte into ova exhibited nine stages starting from basic oogonial cell type to final mature ovum with post ovulatory follicles in Mediterranean amberjack, Seriola dumerilli (Marino et al., 1995). Oocytes with peripheral cortical alveoli, lobated nucleus and several nucleoli transformed into large hydrated oocytes with increased number and size of yolk granules during pre-spawning and spawning period in common dentex, Dentex dentex (Loir et al., 2001).

Increased cell and nuclear size, cytoplasmic granulation and vacuolization in hepatocytes during pre-spawning and spawning phases in L. dyocheilus exhibited biosynthetic activity and apparent correlation with ovarian development. In eostrogen treated freshwater eel, Anguilla anguilla (Olivereau and Olivereau, 1979) and red grouper, Epinephelus akara (Tam et al., 1983) an increase in the hepatosomatic index, enlargement of hepatocytes and their nuclear size and vacuolization in cytoplasm were correlated with biosynthetic activity.

Correlative changes in GSI and HSI, histological features of ovary and liver in captive reared and wild group of fish, L. dyocheilus, demonstrated that ovarian development and maturation was normal in captivity in this fish. Normal ovarian development and maturation in
female L. dyocheilus, an important food fish, under captive condition in tarai region of Uttarakhand has wide scope for application in its controlled quality seed production for stock supplementation in nature/wild and species diversification under culture system.

References