CHAPTER 8
EMBRYONIC AND LARVAL DEVELOPMENT

The embryonic development commences with the penetration of sperm into the ova. In teleost, fertilization is external followed by absorption of water by the egg and hardening of the chorion. Hatchling is the tiny free swimming larva. The larval development begins with the external feeding of fry till the formation of juvenile. In the present treatise the entire development process has been grouped into three phases (a) Embryonic phase (b) Hatchling phase and (c) Larval phase.

8.1 Embryonic phase

(a) Fertilized egg: The fertilized egg of *Nandus nandus* (Ham) is spherical, transparent and non-adhesive. The size of fertilized egg varies from 1.00 – 1.05 mm. The embryo proper lies between 0.75 – 0.8 mm (x. 0.775 mm) and is elucidated in Figure 12-1.

(b) Cleavage:

(i). The 1st cleavage plane is noticeable 15 mins after fertilization in the cap of cytoplasm on the animal pole, dividing it into two equal blastomeres. The second cleavage is perpendicular to the first and
divides into 4-celled blastomeres at 30 mins after fertilization, depicted in Figure 12-2 and it forms 8 blastomeres at 45 mins after fertilization. The 16 blastomeres stage is reached at 1 hr after fertilization and 32 blastomeres stage at 1 hr 15 mins.

(ii) Morula: At 1 hr 30min after fertilization cleavage consists of cohering, sticky and cluster of blastomeres above the yolk mass, thus attaining morula stage (Figure 12-3).

(iii) Blastula formation:
Early blastula: The blastoderm is seen to expand over the yolk mass at 1 hr 45 mins after fertilization.
Blastula: The central area of blastoderm is continuously seen to expand over the yolk after 2 hr as in Figure 12-4.

(c) Gastrulation:

(i) Early gastrula: The expanding blastoderm covers three-fourth of the yolk sac 3 hr after fertilization. The central germ ring is thickened as soon as gastrulation begins.

(ii) Gastrula: The yolk is nearly covered by blastodermal layer in gastrula stage at 3 hr after fertilization. The embryonic shield is developed to form embryo, generally known as yolk plug stage.

(iii) Elongation of yolk mass: The earliest indication of the embryo with antero-posterior axis is distinct at 5 hr after fertilization (Figure 12-5).

(d) Organogenesis:

(i) At 6.55 hrs after fertilization, the cephalic region becomes distinguished. Four somites are formed due to mesodermal segmentation.

(ii) 7 hrs, after fertilization, 6 somites are noticed.
(iii) Optic cup and notochord with 12 somites are noticed at 9 hr 55 mins after fertilization.

(iv) 22 somites are noticed at 10 hr 55 mins after fertilization.

(v) 12 hr 10 mins after fertilization, the caudal region getting free from embryo is recorded.

(vi) 14 hr 55 mins, twitching movement of the embryo is noticed (figure 12 Abbr. 6 & 7). Twitching movement is first noticed in the caudal region. The caudal region becomes free from the yolk mass and most of the yolk is covered by the embryo.

(vii) Embryonic fin fold is noticeable from mid-dorsal region to anal region 16 hr 35 mins after fertilization.

(viii) At 19 hr 35 mins, gill organization is noticed. Eyes prominently developed.

(ix) 20 hr 45 mins after fertilization, the embryo moves vigorously inside the egg capsule, as a result the vitelline membrane burst and the embryo wriggles out with tail first as depicted in figure 12 Abbr. 8. 28 somites are noticed along with two dark pigmented band in between 6th and 7th somite and 14th and 15th somite towards the caudal region.

8.2. Hatchling phase

(a) Free embryo stage

(i) Newly hatched pro-larva: The newly hatched pro-larva is transparent and measures about 2.6 – 3.0 mm (× 2.8 mm) in length. Yolk sac, granulated and elliptical in shape, slightly broader anteriorly and narrow posteriorly extends behind the head to the middle of the body. The primordial fin fold covers the whole of dorsal side along with the tail and the ventral side up to the posterior end of yolk sac. The cephalic portion is broader with prominent optic lobes. The larva remains motionless but shows uncoordinated movement (spiral movement with head region pointing downwards and the caudal region is spirally rotated keeping at an angle of 90°) for a
FIGURE 12. IN-HOUSE DEVELOPMENTAL STAGES OF NANDUS NANDUS

(Abbreviations: 1, Fertilized egg; 2, 4-cell stage; 3, morula; 4, blastula; 5, gastrula; 6&7, twitching stage; 8, hatching with tail first; 9&10, pro-larva; 11 & 12, post larva; 13, clinging stage; 14, fry stage.)
short period. Two dark bands are noticed at 6th and 12th somite region. Pectoral fin with rudimentary fin rays is observed. Photographic representation of Pro-larvae is depicted in Figure 12-9&10

(ii) 12 hrs after hatching: The total length is 3.1 – 3.2 mm ($\bar{x}$ 3.14 mm). The dark bands present at 6th and 12th somites become broader. Four aggregation of star shaped dark pigmentation are observed in the area of the primordial fin fold of the dorsal side. Caudal fin is slightly demarcated with rudimentary fin rays. Yolk sac still persists. Dark pigmentation is observed in the cephalic region.

(iii) Cling stage: 24 hrs (1 day) after hatching. The length of the larva is 3.9 – 4.1 mm ($\bar{x}$ 4.0 mm) and is observed to cling to the wall of the aquaria or to the bottom of the aquaria (Figure 12-13) with sporadic vibratory movement of the caudal region while the hatchling remains clinging to the wall of the aquaria.

(b) Mouth formation stage

Mouth formation of the larva is noticed at 48 hrs (2 days) after fertilization. The length of larva is 4.35 – 4.7 mm ($\bar{x}$ 4.52 mm). Absorption of yolk mass is observed. Movement of lower lip is observed. Mouth is protrusible. Eyes become movable. Eye pigment is brick red in colour.

8.3. Larval phase

(a) Free swimming stage

The larva mostly remains at the bottom of the aquarium but can swim freely. Gut formation is visible in between yolk sac and fin fold. The length of larva is 4.7 mm – 5.0 mm ($\bar{x}$ 4.85 mm).
(b) **Fin formation stage**

(i) **Dorsal fin:** The first indication of dorsal fin formation is observed at 4 days. 7 crests are observed at the primordial fin fold region. Two dorsal spines are observed on 4th day and 4 dorsal spines on 6th day after hatching. Four distinct dark bands are also observed 23 dorsal fin rays are observed at 15th day after hatching and by 20th day after hatching the dorsal fin formation is completed with the formation of 12 dorsal spines and 11 rays. The soft portion of the dorsal fin is transparent.

(ii) **Anal fin:** The anal fin bud is observed at 3rd day after hatching. On 7th day after hatching 4 anal rays are noticed. 15 days after hatching, anal fin formation is completed with the formation of 3 spines and 8 rays. Anal fin is transparent with dark pigmentation at the base of 1st spine.

(iii) **Pectoral fin:** The pectoral fin is developed about 12 hrs after hatching with rudimentary fin rays. 5 fin rays – 2 unbranched and 3 branched are observed on 3rd day after hatching. Pectoral fin formation is completed on 5th day after hatching with the formation of 15 fin rays. Pectoral fin is transparent with pigmentation at the base of the pectoral fin.

(iv) **Pelvic fin:** The pelvic fin bud is observed at 3rd day after hatching and is completed by 12th day after hatching with the formation of 1 spine and 5 rays. Pelvic fin is transparent with dark pigmentation on the 1st pelvic spine.

(v) **Caudal fin:** At 1st day, the caudal fin starts differentiating from the primordial fin fold by caudal fin constriction. The caudal fin formation is completed on 3rd day after hatching with 15 rays. The caudal fin is transparent.
with 3 rows of dark pigmentation. Advance fry stage is depicted (Figure 12-14)

(c) **Colour formation stage:**

The colour pigments are observed at 20 hr 45 min after fertilization. 24hrs after hatching aggregation of star-shaped melanophores are observed in the cephalic region. 3 dark bands alternating with lemon yellow bands are observed on the 6th day after hatching. On 15th day after hatching, 3 distinct dark vertical bands and a dark blotch at the base of caudal fin is noticed. Pigmentation is also noticed at the head region. All the different embryonic and larval stages are schematically shown in figure 13 & 14.
FIGURE 13. SCHEMATIC DIAGRAM SHOWING EMBRYONIC DEVELOPMENT IN NANDUS NANDUS
Explanation of Figure 13

(i) Fertilized egg; (ii) 15 minutes after fertilization; (iii) 30 minutes after fertilization; (iv) 3 hours after fertilization; (v) 5 hours after fertilization; (vi) 6 hours after fertilization; (vii) 7 hours after fertilization; (viii) 9 hours 55 minutes after fertilization; (ix) 12 hours 50 minutes after fertilization; (x), (xi) and (xii) 14 hours 55 minutes after fertilization (twitching movement starts); (xiii) 20 hours 45 minutes after fertilization; (xiv) Pro-larva.
FIGURE 14. SCHEMATIC DIAGRAM SHOWING LARVAL DEVELOPMENT IN NANDUS NANDUS
Explanation of Figure 14

(1) Pro-larva; (2) 1st day after hatching; (3) 2nd day after hatching; (4) 3rd day after hatching; (5) 4th day after hatching; (6) 5th day after hatching; (7) 12th day after hatching; (8) 15th day after hatching;