Technical Manual for Artificial Propagation of the Indonesian Catfish, *Pangasius djambal*
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Chapter VII

Larval Rearing

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The large number of fish farms and the variety of their size, working context and cultivated species have resulted in a great diversity of fry production systems in Indonesia. Although larval rearing could be done successfully in ponds for different fish species, as recently developed for *Pangasianodon hypophthalmus* in Vietnam, this technique has not been used to a large extent for *P. djambal* so far. Some investigations on rearing larvae of this species in ponds led to high variability in terms of survival rates but they remained preliminary. More work is needed to improve and adapt this way of producing *P. djambal* fry.

In Indonesia, fish larvae are generally reared in tanks or aquaria using either stagnant or recycling water.

In this country, mainly for economical reasons, small and middle scale fish producers use the stagnant water aquarium option for rearing larvae. Recycling water systems, synonymous with high technology, generally require a higher investment than stagnant water rearing systems. However, in some situations, using a recycling water system may become a necessity for farmers. This may result either from long term or recurrent shortage of water or from the poor quality (turbidity, oxygen, temperature, ammonia, etc.) of water available in the area, requiring previous treatments.

As the choice of the level of technology used for seed production depends on farmers' needs and possibilities, larval rearing for *P. djambal* larvae has been developed successfully using either stagnant water (Day *et al.*, 2000) or recycling water systems (Legendre *et al.,* 2000). Both technologies will be described in the present chapter together with their associated routine activities.

DESCRIPTION OF REARING STRUCTURE

Although, the sizing of rearing structure is not the main purpose of this guide, some practical indications in this field are given in Appendix I for the recycling water system technology. Indeed it is important to note that the rearing volume needed for seed production either in stagnant or recycling water systems is directly related to the stocking density of fish and the way they are fed. Whatever the technology used, rearing structures are designed for a given maximal stocking density of larvae. Exceeding this number or biomass of fish may lead to a lowering of water quality and the failure of rearing.

In order to make the larval rearing management easier to understand, the following sections describe the working principles of each system.

Recycling water system

Recycling water systems allow rearing larvae in running water, as an open water system. Thanks to mechanical and biological filters, the recycling water is continuously rid of its impurities and toxic dissolved compounds (ammonia particularly) resulting from food waste, fish urine and feces. As the amount of food waste and toxic substances depend directly on the number of reared larvae, the volume of filters must be increased with increasing larval stocking density. This technology also permits decreasing quantity of water supply, controling temperature variations more easily, increasing the stocking density and treating eventual infestation by parasites or bacteria without changing water. The monitoring of the rearing environment has an obvious impact on the performance of the larvae particularly for growth.

A recycling water system is a chain of water treatments and each link corresponds to a specific function. Numerous pieces of equipment are available for each function, but in this practical guide we describe the main steps via the presentation of an original system already used in Indonesia (Appendix I).

Stagnant water system

Rearing in stagnant water means that there is no permanent water flow and water is regularly changed by siphoning. The maintenance of the level of dissolved oxygen in water is ensured by airstones placed in the rearing structure.

Generally carried out in aquaria in order to keep an eye on larvae, the rearing in stagnant water does not require expensive equipment and is well adapted for small and medium scale production. This simple and cheap technology is the most widespread for fry production in Indonesia.

The routine work must be meticulous. Uneaten feed and feces of larvae should be remove manually every day. Moreover, in order to reduce the amount of dissolved toxics, between 50 and 75% rearing water must be changed daily by siphoning. This technique requires a good management of water throughout the larval rearing period.

PREPARATION OF REARING STRUCTURES

Before stocking larvae, all structures have to be ready in order to avoid pathology, stress and escape of newly hatched larvae from rearing structure. Whatever the technology used, all rearing structures should be cleaned and disinfected before filling for a new rearing cycle.

Initial use of recycling water system

Recycling water systems need a special procedure for their very first use in order to prepare the biological filter in good condition. The biological filter can purify ammonia and nitrogen wastes only after the development of nitrobacteria (*Nitrosomas* and *Nitrobacter*). This process needs around 10 to 15 days after the system has been filled with water and started.

It is known that excretion of ammonia and nitrogen wastes at a level exceeding the filter treatment ability can lead to lethal concentrations for the larvae. In order to avoid a risky situation, the biological filter needs progressive conditioning.

In practice, the recycling water system must be filled with water about 2 weeks before starting the first larval rearing. During this period, in order to create favorable conditions for good development of “nitrobacteria”, the following procedure should be followed.

- First, the concentration of ammonia is increased by adding a food bag of about 100 g of ground pellets per m^3 of water. This bag has to be placed before or directly on the mechanical filter in order to prevent pollution of the biological filter with suspended particles. From desegregating pellets, ammonia and other nitrogen-waste compounds are freed, dissolved in water and stimulate a natural development of the “nitrobacteria”;
- after one week, remove the waste pellets if any and place another bag with ground pellets (100 $g.m^{-3}$);
- two weeks after having filled the rearing structure, rinse with clean water in order to change the total volume. The biological filter has already been colonized by nitrobacteria and the rearing structure is now ready to receive larvae.

Once the biological filter works and the first cycle of rearing has been achieved, the second cycle of larval rearing can be started without any special procedure, except for cleaning and disinfecting the structures before introducing new larvae (see Appendix I).

Stagnant water system

Rearing structures in stagnant water do not need special procedures for the initial use. Except for very new containers which need a period of washing to avoid toxic elution. However this technique needs a sufficient stock of clean water and requires very good water management during the entire rearing time. Aquaria or tanks must be filled with water well before receiving larvae in order to allow equilibration of oxygen and temperature, and to avoid thermal shock and stress for the larvae.

STOCKING DENSITY IN REARING STRUCTURES

The stocking density is the number of larvae reared per litre of water. Unknown and too high density may engender an imbalanced water quality, slow growth, low survival rate and great size heterogeneity of remaining fish. As noted previously, each rearing structure is designed for a maximum fish stocking density. Whatever the technology used, this maximal value should not be exceeded.

Using a low stocking density does not allow operation of the farming system at its full capacity and results in reduced production.

The initial number of larvae transferred to the rearing structures has to be known by counting, or accurately estimated, for a proper subsequent management of feeding and good water quality maintenance in each tank.

To optimize larval production, the stocking densities recommended for *P. djambal* during larval rearing are the following:

- in stagnant water: 15 fish per litre until 8-day of age, then 5 larvae per litre from the age of 8 to 18-day;
- in recycling water: 30 fish per litre until the 15th day of age.

Age (days)	Average weight in recycling water system (mg)	Average weight in stagnant water (mg)
2	5.4	5.7
3		
4	16	11
5		
6	47	20
7		
8	97	30
9		
10	130	65
11		
12	210	100
13		
14	380	190

Table VII.1.

Growth of *P. djambal* larvae in recycling and in stagnant water systems.

GROWTH OF LARVAE

Mean growth performance of *P. djambal* larvae reared in recycling water (RIFA) and in stagnant water (JFADC) are compared in Table VII.1. These data were obtained by individual weighing of larvae every two days using an accurate balance. Although every farmer could not invest in such an expensive measuring device, accurate follow up of growth rate allows one to calculate and readjust the daily feeding rate.

Table VII.1 shows a slower growth in stagnant water illustrating the limits of this rearing technique in comparison to recycling water systems. Nevertheless, the survival rates obtained in both situation (stagnant or recirculated water) are similar and greater than 75%. These results demonstrate that, even if the growth is not optimal in stagnant water, this technique developed by the JFADC remains well adapted for this species and could be used by small scale farmers in Indonesia.

FEEDING PROCEDURES

Artemia nauplii

For each cultured species, optimal feeding levels have generally been established and are specified as a number of nauplii per larva and per distribution or per day. It has been demonstrated that larval rearing success is considerably impaired when optimal feeding levels are not respected. As larvae need a given number of *Artemia* nauplii per distribution, it is recommended to estimate every day the harvested quantity of *Artemia* nauplii produced in the hatchery, in order to manage properly the ration of the day. Incubating, harvesting and counting methods for *Artemia* are described in Appendix II.

P. djambal larvae have to be fed with *Artemia* nauplii from 48-h post-hatching (see Table VII.2) until:

- 5-day of age, i.e. 4-day of feeding, in recycling water;
- 8-day of age, i.e. 7-day of feeding, in stagnant water.

In recycling water

From the very first meal, the quantity of *Artemia* nauplii distributed to the larvae should be respected and increased daily as shown in Table VII.2. This table gives the recommended number of nauplii for one meal and for one larva according to age. The evaluation of the volume of water containing the *Artemia* nauplii to use for feeding the larvae is also explained in detail in Appendix II.

In a recycling water system, it is recommended to stop the water flow for 30 minutes at each feeding time in order to maintain the living prey in the tanks.

The *Artemia* nauplii number recommended per larva has to be multiplied by the total number of larvae grown in each tank. An example is given in Table VII.2 for one tank of a recycling water system stocked with 7500 larvae.

The quantity of *Artemia* nauplii per distribution given in Table VII.2 was evaluated for a feeding schedule of 7 meals per day. The recommended feeding frequency is to feed larvae every 3 hours from 6:00 to 00:00.

Table VII.2.

Calculation of the *Artemia* nauplii quantity for one meal.

Age (hours)	Age (days)	Number of *Artemia* nauplii per distribution and per larva	Quantity of dried diet	Number of *Artemia* nauplii per distribution for 7500 larvae	
				Calculation	Number
48 - 73	2	20	0	20 x 7500	150 000
72 - 96	3	50	0	50 x 7500	375 000
96 - 120	4	70	0	70 x 7500	725 000
120 - 144	5	100	trace	100 x 7500	750 000

In stagnant water

In order to preserve water quality, the number of *Artemia* nauplii is distributed as required (*ad libitum*). As stated above, the number of *Artemia* nauplii per larva should be increased daily until the weaning time. Therefore, for evaluating the right number of prey to distribute farmers should observe accurately the behavior of larvae to see if they are filled up or not.

To obtain optimal performance, the following feeding frequency is recommended:

- between the 2nd and 8th days of age, fish are fed 5 times per day, every 4 hours between 7:00 and 23:00 with a fasting period between 23:00 and 7:00.

Weaning time

In many fish species weaning time is a very delicate period because the anatomical and functional development of the gut is not totally achieved yet. In *P. djambal*, substituting *Artemia* nauplii with a new artificial diet does not cause any problem. However, it is recommended to get the larvae used to their new diet before stopping distribution of *Artemia* nauplii.

In recycling water

From the 4th day of feeding, when using the recycling water technique, it is advisable to distribute a small quantity of artificial diet half an hour before feeding the larvae with *Artemia* nauplii. As stated previously, total substitution of the *Artemia* nauplii with artificial diet from the 5th day of feeding significantly decreases operating cost of larval rearing (Chapter VI).

In stagnant water

Nowadays, artificial diet is not commonly used to feed larvae in stagnant water to avoid water pollution. Therefore, *Artemia* nauplii are substituted by *Tubifex* sp. (blood worm) after the 8th day after hatching.

Artificial diet

The following comments concern only larvae reared in recycling water because artificial diet is generally used in this system.

An appropriate protein content and size of particles are required for successful weaning. Particle diameter of the initial dried diet should be well adapted to the mouth of larvae to permit or facilitate ingestion and also to aid digestion. The size of the initial dried diet recommended for *P. djambal* weaning should be between 270 and 410 µm.

Since a deficiency in essential nutrients can affect growth, it is recommended to rear larvae on a balanced protein-rich diet (40 – 45% of crude protein). The best way to feed larvae is to distribute artificial diet as required by the fish (*ad libitum*). However sometimes the limit between *ad libitum* distribution and over-distribution is very thin. An excess of food in a rearing container can reduce water quality, resulting in disease or fish mortality.

Therefore, we recommend to give a daily ration of 20% of the biomass from the 6th to the 9th day; 15% from the 10th to 13th day and then 10% from the 14th day (Table VII.3). After this period, the feeding rate will be decreased step by step according to age.

Calculation of the feed quantity (Table VII.3)

- Mean body weight of larvae must be known and multiplied by the total number of larvae to obtain the total biomass.
- Total biomass is multiplied by the daily ration in % to obtain the daily quantity of artificial diet to distribute (ration).
- Daily quantity of artificial diet calculated above must be divided by the number of distribution done during the day to obtain the quantity of feed per distribution.

However, as stomach emptying is totally different after 8 days (Chapter VI), it is recommended that larvae be fed 6 or 7 times a day (as for *Artemia* nauplii) until the age of 7-day and 5 times a day from the 8th day.

Table VII.3.

Calculation of the feed quantity for *P. djambal* larvae.

Age	Mean body weight (mg)	Total biomass for 7500 larvae		Daily quantity for 7500 larvae		Quantity of dried diet per distribution for 7500 larvae	
		Calculation	Biomass (g)	Calculation	Ration (g)	Calculation	Quantity (g)
6	47	7500 x 0.047	353	20% x 353	71	71 / 7	10 – 11
7					71		10 – 11
8	97	7500 x 0.097	728	20% x 728	146	146 / 5	29 – 30
9					146		29 – 30
10	130	7500 x 0.130	975	15% x 975	146	146 / 5	29 – 30
11					146		29 – 30
12	210	7500 x 0.210	1575	15% x 1575	236	236 / 5	47 – 48
13					236		47 – 48
14	380	7500 x 0.380	2850	10% x 2850	327	327 / 5	57 – 58

Blood worms *(Tubifex* sp.*)*

Due to their low price and their abundance in some Indonesian areas, *Tubifex* are the most widespread wild food used for feeding fish larvae or fry in stagnant water. However, *Tubifex* are not available everywhere in Indonesia and their abundance depend on the season. Even if their price remains lower than that of dried diet, it is very variable according to season and locality.

It is known that *Tubifex* can carry parasites for fish such as myxozoa or cestodes (De Kinkelin, 1985) and can trigger disease. Moreover, these worms are often collected in sludge or in pluvial evacuation, and then put in plastic bag for transportation at ambiant temperature sometimes exceeding 30°C. Although, farmers wash *Tubifex* sp. with clean running water and add airstone before feeding to larvae, the risk of parasitic and bacterial contamination exists.

For these reasons, and since in recycling water artificial diet is very efficient, we advise not using *Tubifex* in this structure.

If *Tubifex* is to be used, we strongly recommend disinfecting *Tubifex* with formalin in the following way before distribution:

- put *Tubifex* in a formalin solution at 50 ppm.; i.e. 50 $mL.m^{-3}$;
- mix for 2 minutes;
- rinse at least 3 times with clean water;
- distribute to larvae.

Generally, in stagnant water, farmers prefer *Tubifex* to dried diet. In fact, distributed alive, the worms group together and form a ball at the bottom of the rearing tank and stay alive a long time, which is a great advantage:

- easy to control the ration (as long as some *Tubifex* remain it is not necessary to re-distribute);
- no pollution of the rearing water (live-food);
- larvae eat when they want (self-feeding).

Chopped or ground *Tubifex* could also be used for very young larvae, but in this case rearing water is quickly polluted when stagnant.

In JFADC, for *P. djambal* larvae live *Tubifex* were successfully used to substitute *Artemia* nauplii from the 7th to the 16th day of age.

CLEANING AND WATER MANAGEMENT

In order to preserve water quality and keep larvae in good health, it is strongly recommended, from the 2nd day of rearing, to remove the food wastes and feces daily from the bottom of the tank by siphoning. This cleaning operation should be done before the first feeding in the morning.

As indicated previously (Chapter VI), during larval rearing particular attention must be paid to oxygen concentration because requirement of this species is quite high.

In recycling water

Specific routine activities and water management related to mechanical and biological filters are explained in Appendix I.

To keep good quality of water in the rearing tank, the flow rate must be increased progressively according to the age of larvae. In fact, very young larvae of *P. djambal* habitually swim against the current created by the water flow. This behavior probably makes exchange easier with oxygen. Now, if the current is too strong, larvae wear themselves out to swimming or are squashed on the outlet net.

In fact, without a measuring device to control water quality, the watchfulness of the farmer is important determining when the water flow is:

- too low:
  - larvae are concentrated on aeration or water flow;
  - water becomes whitish.
- too strong:
  - larvae tire themselves swimming;
  - larvae concentrated or squashed on the outlet net;
  - food wastes and feces are totally evacuated by the current.
- well adjusted:
  - larvae are well spread out around the tank;
  - larvae swim slowly against current;
  - water is clear;
  - food wastes and feces are partly concentrated near the outlet.

To avoid stress to larvae, problems of water quality must be anticipated and water flow increased every 2-day according to the following rates:

- 25% of water volume of the tanks changed **per hour** from the 1st to the 2nd day of rearing;
- 50% from the 3rd to 5th day of rearing;
- 100% from the 6th to 15th day of rearing.

Water flow is considered here by the percentage of rearing water changed in the tank during one hour while it is generally stated in litre per hour or per minute.

Although, the previous expression is currently used by farmer, we have to be careful using this expression, because for the same rate of water exchange, water flow is different depending upon the volume of the tank. The following explanation gives two examples of calculations for converting the percentage of water exchange into flow rate.

Calculations

An experimental tank with a rearing volume of 30 L and water change of 25% per hour, has a water flow of 7.5 $L.h^{-1}$, i.e. 25% x 30 = 7.5 $L.h^{-1}$.

A production tank with a rearing volume of 1000 L and water change of 25% per hour, should have a water flow of 250 $L.h^{-1}$; i.e. 25% x 1000 = 250 $L.h^{-1}$.

In stagnant water

Since the rearing is done in stagnant conditions, the water is removed by siphoning during cleaning wastes. Siphoning should be done slowly with a plastic pipe into a bucket. To avoid sucking up larvae during the cleaning, the end of the siphon should be fitted with an outlet net large enough to avoid larvae squashing against it.

To limit stress, the quantity of water renewed should be adapted to the age of the larvae and can be programmed as follows:

- 50% of water change per day from the 2nd to the 6th days of rearing;
- 75% from the 7th to 16th day of rearing.

EQUIPMENT AND TOOLS

Recycling water system

1. Recycling water system with bio-filter and mechanical filter (equipments are listed in Appendix I).
2. Rearing tanks with adjustable inlet and outlet nets of suitable diameter.
3. Air blower with airstones in each rearing tank.
4. Watch to measure the water flow in each tank.
5. Calculator for calculating water flow.

Stagnant water

1. Aquarium 60 x 50 x 40 cm.
2. Air blower with airstones in each aquarium.
3. Plastic bucket for filling aquarium.
4. Tank for storing clean water.

Water quality control (recommended)

1. Oxygen kit or oxymeter.
2. Ammonia kit.
3. Nitrite kit.
4. pH kit or pH meter.

Handling and stocking newly hatched larvae

1. Aquarium with airstone and clean water.
2. Plankton net (80 µm mesh size) for catching larvae.
3. Plastic bowl for transferring larvae to the aquarium.
4. Flashlight for concentrating normal larvae in the beam of light.
5. Plastic pipe for cleaning the bottom of white eggs and abnormal larvae and then siphoning normal larvae.
6. Plastic bucket for transferring normal larvae in rearing structures.

Feeding

Artemia

1. Equipment described in Appendix II.

Artificial diet

1. Balance (500 g ± 1 g) for weighing daily feeding ration.
2. Plastic bucket for stocking feed for each rearing structure.

Blood worm

1. Small tanks with water flow and airstone for stocking *Tubifex*.
2. Formalin for disinfecting.
3. Syringe for measuring formalin.

Cleaning

1. Plastic pipe with a large outlet net for siphoning the bottom.
2. Plastic bucket for catching the siphoned water.

REFERENCES

Day, D., M. Bahnan, Ediwarman and Maskur, 2000. Pemeliharaan larva patin Jambal *(Pangasius djambal)* selama 18 hari. *Pertemuan Pengembangan teknologi perbenihan Budidaya Air Tawar, Payau dan laut, lintas UPT Direktorat Jenderal Perikanan tanggal 11-14 Juli 2000 di Bandar Lampung.*

De Kinkelin, P., C. Michel and P. Ghittino, 1985. Précis de pathologie des poissons, *INRA-OIE ed.*, 340 p.

Legendre, M., L. Pouyaud, J. Slembrouck, R. Gustiano, A. H. Kristanto, J. Subagja, O. Komarudin and Maskur, 2000. *Pangasius djambal*: A new candidate species for fish culture in Indonesia. *IARD journal*, 22: 1-14.

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