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# Hatchery techniques for egg and fry production of Clarias batrachus (Linnaeus)

Christopher F. Knud-Hansen<sup>a,1</sup>, Ted R. Batterson<sup>a</sup>, Clarence D. McNabb<sup>a</sup>, Yani Hadiroseyani<sup>b</sup>, Darnas Dana<sup>b</sup> and H. Muhammed Eidman<sup>b</sup>

<sup>a</sup>Department of Fisheries and Wildlife, Michigan State University, East Lansing, MI 48824 (U.S.A.)

<sup>b</sup>Institut Pertanian Bogor, Facultas Perikanen, Jalan Raya Pajajaran, Bogor (Indonesia)

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#### ABSTRACT

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Egg hatching, and fry growth and survival of the walking catfish, Clarias batrachus (Linnaeus), were investigated under hatchery conditions in West Java, Indonesia. Spawning was environmentally induced in a specialized breeding pond. Gravid females utilized nests containing kakaban, a fibrous matting from local palm trees (Arenga sp.), which facilitated egg collection. Newly hatched fry fed with Artemia nauplii though day 8 (after hatching), an Artemia/cladoceran mix from days 9 to 16, and cladocerans only from days 17 to 23 resulted in over 90% survival of young from hatched eggs. Other diets examined (rotifers, cladocerans, ground fish meal, and ground Nile tilapia flesh) proved inadequate for fry through day 16. Fry reared in hatchery aquaria for 16 days versus 23 days before introduction into nursery ponds showed no significant differences in mean length, mean weight, or percent survival when harvested at day 58. Suggested guidelines are given for hatchery production of C. batrachus fry and fingerlings.

#### INTRODUCTION

Commercial pond aquaculture of the catfish, Clarias batrachus (Linnaeus), commonly known as "pla duk dan" in Thailand, "ikan lele" in Indonesia, and walking catfish in the U.S.A., first developed in Thailand in the late 1950s (Tarnchalanukit et al., 1982; Areerat, 1987). In southeast Asia C. batrachus generally spawn during the rainy season, when rivers rise and fish are able to excavate nests in submerged mud banks and dikes of flooded rice fields. Culture practice in Thailand consists of collecting by hand from 2000 to 15 000 1-week-old fry per nest, which are then transported to nursery ponds.

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<sup>&</sup>lt;sup>1</sup>Correspondence address: Dr. Christopher F. Knud-Hansen, Asian Institute of Technology, Agricultural & Food Engineering Division, G.P.O. Box 2754, Bangkok 10501 (Thailand).

When fingerlings reach 5-7 cm in length, they are transferred to growout ponds. Fed primarily on a diet of ground trash fish and rice bran, the omnivorous air-breathing *C. batrachus*, well suited to waters often anoxic from high organic loading, reach marketable size (150-300 g) in 4-6 months (Sidthimunka, 1982, Areerat, 1987).

Development of "ikan lele" pond culture in Indonesia is more recent. Similar to commercial production in Thailand, fry are generally collected from nests in flooded rice paddies and reared in growout ponds. Some Indonesian Clarias farmers line nests with kakaban, a fibrous matting obtained from palm trees (Arenga sp.). Eggs are naturally covered with a gelatinous matrix, which allows them to stick to the kakaban, thus facilitating egg retrieval from nests. Immediately after spawning the kakaban is removed from nests, and attached eggs are gently shaken into aquaria filled with water to a depth of about 10 cm. Fry hatch within 24 h of spawning and are reared 2–3 weeks in aquaria before transfer to nursery ponds.

C. batrachus is a prized food species in Indonesia, with high economic value. Brood pairs are not readily available, and may cost as much as U.S.\$ 6/kg. The supply of this fish from wild populations has diminished over recent years. Attempts at intensive culture to meet market demands have been limited by the high, commonly over 80%, mortality of eggs and young fry. Although Zonneveld et al. (1988) demonstrated successful induced spawning using carp pituitary extract, this technology is not yet practical for small-scale farmers. The objective of the research reported here was to develop a hatchery strategy for egg handling and fry production without high mortality.

## MATERIALS AND METHODS

Site description

A breeding pond and hatchery building were constructed at the Institut Pertanian Bogor Babakan Fisheries Station, Bogor, West Java, Indonesia. A description of the Station is given in Egna et al. (1987). The cement-walled breeding pond ( $20 \text{ m} \times 10 \text{ m}$ ) was characterized by an earthen trough 3 m wide and 0.75 m deep running the length of the pond. On one side of the trough, the pond bottom sloped gradually upward from the channel's edge to 20 contiguous concrete nesting boxes ( $0.4 \text{ m} \times 0.4 \text{ m} \times 0.4 \text{ m}$ ). Removable wooden lids provided access to the nesting boxes. A 20-cm PVC tube (10 cm i.d.) connected each box to the pond below maximum water level. At maximum pond volume, pond depth was 1.5 m in the trough and 0.3 m in the nesting boxes. Source water was channelled surface drainage passed through a conditioning system (sedimentation basin followed by sand, crushed limestone, and charcoal filtration; McNabb et al., 1985).

The partially enclosed hatchery was equipped with electricity, a compressed air supply, and 24 glass aquaria ( $60 \,\mathrm{cm} \times 40 \,\mathrm{cm} \times 40 \,\mathrm{cm}$ ). The aquaria,

used for egg hatching and initial fry rearing, were wrapped in black plastic and covered with plastic mesh to minimize light and more closely reflect natural nest conditions. Twelve concrete nursery ponds  $(5 \text{ m} \times 2 \text{ m} \times 1 \text{ m})$  were constructed adjacent to the hatchery for fry growout experiments. A headtank connected to the water conditioning system provided water for aquaria and nursery pond experiments.

## Egg production

One hundred mature C. batrachus (1:1 sex ratio), each weighing between 150 and 250 g, were placed in the breeding pond, which was filled only to the top of the trough (0.75 m). Female conditioning and ovulation were induced by doubling the normal feeding rate of fish meal (35% protein) from 5% to 10% body wt/day for 10 days. Kakaban was spread over the bottom of the spawning boxes and the pond water level was raised to 1.5 m, filling entry tubes and providing breeders with access to flooded spawning boxes. This process simulated natural spawning events in riverine habitats at the start of the rainy season. Nests were monitored frequently for egg deposition during the next 7 days.

Because an estimated 10–20% of the deposited eggs fell off the fibrous kakaban when removed from the nest, initial experiments focused on alternative methods of using kakaban. To facilitate handling, plastic buckets and 10-l plastic jars (presoaked 3 weeks in pond water) were placed horizontally in random nests with the openings facing the PVC tube connection to the breeding pond. Over two spawning cycles a total of 19 nests was tested with kakaban alone, six with kakaban in buckets, and 12 with kakaban in jars.

# Egg handling

Eggs were collected at around 08.00 h, and carefully transferred in closed containers to the hatchery. Initial observations showed poor embryo survival, possibly due to fungal infections of the gelatinous material surrounding the eggs. Sodium sulphite, which effectively dissolves the organic matrix around channel catfish eggs (Dorman, 1986), was used to remove this gelatinous material. Immediately upon collection, *C. batrachus* eggs were soaked in a 1% sodium sulphite solution for 5 min, then rinsed three times with tap water before being placed in aquaria.

To reduce the time and excessive handling for individual egg counts, the linear regression equation relating egg number to volume of eggs was determined ( $r^2 = 0.98$ , Fig. 1). Volumes of up to 12 000 sodium sulphite treated eggs were measured by water displacement in graduated cylinders, yielding approximately 800 eggs/ml. Microscopic measurements of egg diameters (0.89 mm  $\pm 0.04$  ( $\pm 1$  s.e., n = 90)) suggested little variability within or between nests. The linear relationship between egg number and weight of eggrindicated that 1.0 g wet wt. approximately equals 918 eggs; however, the pro

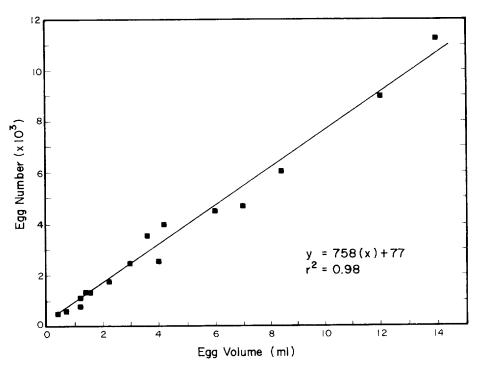


Fig. 1. Relationship between number and volume (ml) of *C. batrachus* eggs treated with 1.0% sodium sulphite (see text).

cedure was more tedious and the relationship ( $r^2 = 0.63$ ) not as good. Therefore, all hatchery experiments used volumes to estimate egg number.

## Reduction of fry mortality

Initial attempts to raise fry on *Moina* spp. resulted in nearly 100% mortality within the first 7 days after hatching. Water quality measurements suggested that mortality was not related to pH (7.5–7.8), diel fluctuations in water temperature (22–30°C), dissolved oxygen (92–100% saturation), or unionized ammonia-N concentrations (<0.01 mg/l) calculated from total ammonia measurements (phenol-hypochlorite method; Solorzano, 1969).

To examine the relationship between food type and fry survival, three feeding experiments were conducted in aquaria filled to a depth of 10 cm (=241). The shallow depth reduces energy costs for the air-breathing fry to reach the surface. Rocks and PVC tubes placed in aquaria were often used by fry as refuges.

The first experiment tested Artemia nauplii, Artemia nauplii in pond water, rotifers, cladocerans (principally Daphnia spp. and Moina spp. passed through a 1-mm sieve), and ground fish meal (35% protein). Artemia cysts (San

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Francisco Bay, CA, U.S.A.) were hatched in aerated cylinders (Sorgeloos et al., 1976). Three aquaria/treatment were stocked with approximately 1600 (2.0 ml) *C. batrachus* eggs. Food was introduced in a slurry with source water, except for the *Artemia* nauplii in pond water which contained pond biota and detritus. Fry were fed ad libitum twice per day. Accumulated particulate matter was siphoned daily from each aquarium to avoid reduction of water quality.

Although high fry survival through day 7 was obtained when fry were fed a diet of *Artemia* nauplii (see results), subsequent mortality before day 23 prompted feeding experiments 2 and 3. In both experiments three aquaria/treatment were stocked with 500 day-7 fry reared on *Artemia* nauplii. The second feeding experiment tested *Artemia* nauplii, cladocerans, and freshly ground Nile tilapia. The third examined a variable food-type regime. Fry were fed only *Artemia* nauplii through day 9, a 1:1 by weight mixture of *Artemia* nauplii and cladocerans from days 10 to 16, and only cladocerans from days 17 to 23. *Artemia* nauplii, *Artemia* nauplii and ground fish meal, and ground fish meal alone also were tested as fry food during days 10 to 16. Fry were fed ad libitum twice per day with accumulated particulate matter removed daily.

A growout experiment examined minimum hatchery residence time by comparing growth and survival of fry stocked in triplicate nursery ponds at day 16 after hatching versus day 23. Ponds were fertilized with fresh chicken manure (0.5 kg wet wt./m²) 10 days before stocking, and inoculated with a mixture of *Daphnia* spp. and *Moina* spp. 3 days after adding the manure. Beginning 2 days after stocking (50 fish/m²), fry were fed ad libitum pelleted fish meal (35% protein) twice per day. This provided a smooth transition in diet from cladocerans to pelleted feed, since zooplankton may not be utilized by larger *C. batrachus* fingerlings (Varghese et al., 1973). Harvesting from nursery ponds took place on day 58 after hatching. Treatment comparisons included percent survival, and mean lengths and weights (n=90 fish/treatment).

Treatments were analyzed for significant differences (P<0.05) using Student's t-test (Steel and Torrie, 1980). Means are given  $\pm 1$  standard error (s.e.).

#### **RESULTS AND DISCUSSION**

## Egg production and handling

Eleven successful environmentally induced spawnings were obtained using kakaban as the nesting material. Egg deposition and fertilization most frequently occurred during the night following nest flooding, with the remaining spawnings observed 2 and 4 days later. All spawnings occurred in nests with kakaban only; breeders avoided nests with kakaban in buckets or plastic jars.

In a typical spawning event we collected about 3000-12 000 eggs per nest. Percent live eggs ranged from 85 to 96%, of which an average of 49.7% suc-

TABLE 1

Clarias batrachus egg number and hatching rates from four different kakaban nests

Nest no.	Total no. of eggs	% Eggs with live embryos	% Hatching of live eggs	
1	11 687	96.4	47.7	
2	9416	95.9	60.2	
3	5459	85.4	58.8	
4	2906	85.9	2.4	
Total	29 468	93.2	49.7	

cessfully hatched (Table 1). Both the timing of spawning and number of eggs per nest were consistent with observations made by Sidthimunka (1972). Whether the same or different females were spawning each time the pond was raised was not determined. Under natural conditions in subtropical India (Thakur, 1978) and Bangladesh (Barua et al., 1986), C. batrachus spawn just once per year during the warmest months of the monsoon season. Zonneveld et al. (1988) observed no clear reproductive season in tropical East Java, but they suggested an inverse relationship between annual temperature variations and length of the reproductive cycle. In Thailand, manipulation of pond depth may increase spawning frequency of pond-reared C. batrachus to once per month (Areerat, 1987).

## Reduction of fry mortality

Initially, C. batrachus hatchlings were fed rotifers and a cladoceran mix of Daphnia spp. and Moina spp. The zooplankton diet, suggested by Hora and Pillay (1982), is commonly used in Indonesia. Fry survival was over 90% after day 1, but by day 7 all fish had died. Since environmental conditions seemed adequate and microscopic examinations for parasites were negative, feeding experiments were conducted to determine possible nutritional deficiencies in newly hatched fry. The observation that yolk sacs generally disappeared about day 4, while high mortality occurred on day 5 supported this hypothesis.

The first feeding experiment showed that Artemia nauplii, widely used for raising fish larvae (Léger et al., 1986), were the best food tested for C. batrachus larvae, with over 86% survival after 7 days (Table 2). Observations included healthy fry swarming behavior during feeding and red nauplii visible through translucent stomachs. Fry raised in pond water and fed Artemia nauplii had close to 100% survival, although not significantly different (P < 0.05) from fry fed Artemia nauplii without the detrital supplement. Rotifers represented a much poorer food source (25% fry survival), but better than the cladoceran mix or ground fish meal in which nearly all fry died by day 7.

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TABLE 2 Percent survival (mean  $\pm 1$  s.e.) of *C. batrachus* 7 days after hatching when fed different food types (first feeding experiment, see text)

Food types	% Survival <sup>a</sup>	Relative survival	
Artemia nauplii	86.2± 8.7	0.83	
Artemia nauplii/pond water	$104.2 \pm 5.5$	1.00	
Rotifers	$25.2 \pm 13.8$	0.24	
Cladocerans	$0.0 \pm 0.0^{\rm b}$	0.00	
Ground fish meal	$1.5 \pm 0.9$	0.01	

<sup>&</sup>lt;sup>a</sup>Based on expected egg hatching rate of 50% (see text).

Polling et al. (1988) obtained different results for *C. gariepinus*. Young fry raised on zooplankton yielded higher growth rates than when fed *Artemia* nauplii, although fry under both treatments had > 90% survival and grew better than when fed dry feed. Low fry survival using dry feeds in this and other studies (e.g. Sitasit and Fedoruk, 1981) discourages their use for larval nutrition. High mortality observed in treatments without *Artemia* nauplii may have been due to food particles being too large to ingest, as well as energy costs in capturing motile cladocerans. It could be significant that fry were fed with San Francisco Bay *Artemia* nauplii, since other commercial strains are often nutritionally inadequate and too large to be swallowed by predators (Sorgeloos et al., 1976).

The second feeding experiment revealed the limited utility of Artemia nauplii as live feed for C. batrachus fry. Fry fed with Artemia nauplii maintained survival rates above 90% through day 17, but by day 21 nearly all had died (Fig. 2). After day 17, Artemia nauplii may either be too small or insufficient to provide adequate nutrition required by developing Clarias fry. Cladocerans and ground Nile tilapia flesh were totally inadequate foods for fry of this age, with almost 100% mortality by day 14.

As zooplankton have been considered the preferred food source for 3–4-week-old fry (Sidthimunka, 1972), the third feeding experiment incorporated a transitional period between using *Artemia* nauplii and cladocerans as principal food sources. This feeding schedule proved to be optimal, with an average of 97.2% ( $\pm 0.6$ ) fry survival from day 9 to day 23 (Fig. 3). Apparently, the transitional period in which both *Artemia* nauplii and cladocerans were available was necessary for fry survival. Using *Artemia* nauplii, *Artemia* nauplii and ground fish meal, and ground fish meal alone resulted in greater fry mortality; by day 16, percent survival averaged 87.0% ( $\pm 3.5$ ), 72.6% ( $\pm 5.8$ ), and 33.3% ( $\pm 12.7$ ), respectively.



<sup>&</sup>lt;sup>b</sup>100% mortality by day 5.

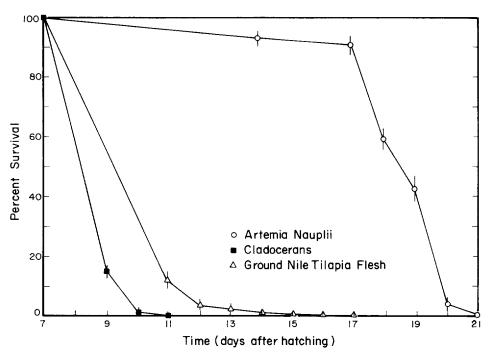


Fig. 2. Percent survival ( $\pm 1$  s.e.) of *C. batrachus* fry raised on *Artemia* nauplii, cladocerans, and ground Nile tilapia flesh, from day 7 to day 21 after hatching (second feeding experiment).

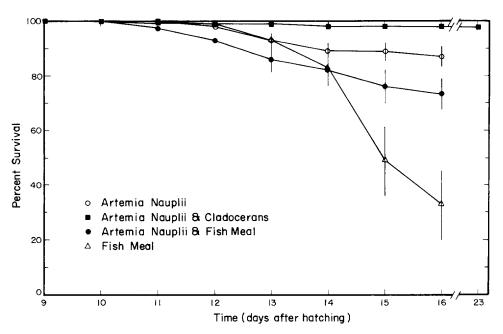


Fig. 3. Percent survival ( $\pm 1$  s.e.) of *C. batrachus* fry raised on fish meal, *Artemia* nauplii, and *Artemia* nauplii in combination with cladocerans and fish meal, from day 9 to day 16 after hatching. Fry which received *Artemia* nauplii and cladocerans from day 9 to day 16 received only cladocerans from day 17 to day 23 (third feeding experiment).

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TABLE 3

Percent survival, mean length and weight ( $\pm 1$  s.e., n=90) of C. batrachus fry when stocked in nursery ponds (n=3) 16 and 23 days after hatching

Measurements	Stocked on day 16	Stocked on day 23	· ·
At stocking			
Number (fish/pond)	500	500	
Mean length (cm)	$1.47 \pm 0.02$		
Mean weight (g)	$0.040 \pm 0.003$	$0.047 \pm 0.003$	
At harvest (day 58 after hate	ching)		
Percent survival	$56.7 \pm 16.5$	$60.2 \pm 16.0$	
Mean length (cm)	$7.2 \pm 0.2$	$7.0 \pm 0.2$	
Length range (cm)	4.4 – 11.6	4.7 – 10.7	
Mean weight (g)	$3.9 \pm 0.2$	$4.3 \pm 0.2$	
Weight range (g)	1.0 - 10.4	2.2 - 9.0	

The last question addressed the timing of fry transfer from aquaria to nursery ponds. Early placement of fry in nursery ponds without increased mortality benefits production efficiency by reducing intensive hatchery feed and labor costs. Fry reared in aquaria for 16 days versus 23 days before the transfer showed no significant differences (P < 0.05) in growth or survival when harvested 58 days after hatching (Table 3). Fry stocked on days 16 and 23 averaged 7.2 cm( $\pm 0.2$ ) and 7.0 cm( $\pm 0.1$ ) in total length, and 3.9 g ( $\pm 0.2$ ) and 4.3 g ( $\pm 0.2$ ) in weight, respectively. These mean weights are approximately twice the reported mean weights for 7-cm C. batrachus fry collected in the wild (2.0 g) or in farm ponds (2.3 g) in Thailand (Srisuwantach and Yingcharoen, 1981). The variable and relatively low fry survival (about 60%, Table 3) observed in this study was probably due to predation from tadpoles, in addition to a large (40 g) C. batrachus retrieved from one of the experimental nursery ponds.

#### **SUMMARY**

The following summarizes suggested guidelines for hatchery production of *C. batrachus* fry and fingerlings based on results described above. Using these procedures, we were able to maintain over 90% survival of hatchlings through transfer to nursery ponds.

- Use kakaban or a similar natural fiber as a nest material to facilitate egg collection and transfer to hatchery.
- Collect eggs from the nests early in the morning after spawning, and immediately treat them with 1.0% sodium sulphite for approximately 5 min.

- If egg enumeration is necessary, use the relationship with volume.
- Egg hatching and initial fry rearing can take place in aquaria filled with aerated water to a depth of about 10 cm. It may be beneficial to maintain reduced light conditions. Rocks and PVC tubes may be placed in aquaria as refuges for fry.
- Fry should be fed *Artemia* nauplii for days 1 to 9, *Artemia* nauplii and cladocerans from day 10 to 16, and cladocerans only from day 17 until transfer to nursery ponds. Hatchery foods, particularly *Artemia* nauplii, are expensive, but may be cost effective relative to market value of *C. batrachus*.
- Fertilize nursery ponds and inoculate with cladocerans prior to transferring fry from the hatchery.
- Transfer fry to nursery ponds as early as day 16 to avoid additional hatchery costs.
- Protect nursery ponds against predation to eliminate losses.

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