NGUYỄN ĐÌNH VINH

RESEARCH ON BIOLOGICAL CHARACTERISTICS AND SEED PRODUCTION TECHNIQUES OF THE HELMET CATFISH - Cranoglanis bouderius (Richardson, 1846) IN CULTURE CONDITION OF NGHE AN PROVINCE

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SUMMARY OF BIOLOGY DOCTORAL THESIS

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This doctoral thesis was completed at: Graduate University of Science and Technology, Vietnam Academy of Science and Technology.

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Reviewer 1: …
Reviewer 2: …
Reviewer 3: …

This doctoral thesis will be defended at the graduate university committee of doctoral thesis evaluation at Graduate University of Science and Technology, Vietnam Academy of Science and Technology on ……………..

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PREAMBLE

1. The necessity of the thesis

Helmet Catfish - *Cranoglanis bouderius* (Richardson, 1846) is a species of order Siluriformes, family Cranoglanididae. In the world, Helmet Catfish are distributed in China (the areas sharing the same border with Vietnam are Hainan, Guangdong, Guangxi, Yunnan). In Vietnam, the Helmet Catfish is commonly found in all river systems from the North (Red River, Ma River, Lam River) to the South Central. The lowest limit toward the South of this species is Tra Khuc River - Quang Ngai. Helmet Catfish is distributed in the bottom level and bottom nearing level, preferring to live in places where water runs regularly or slowly, muldy sandy bottom. Fishes usually live in shoals, mainly in the downstream areas more than middle and upstream rivers in the Northern provinces.

Helmet Catfish has high nutritional value. Zhang et al. (2009), conducted an analysis of fatty acid to evaluate the nutritional value of Helmet Catfish meat and indicated that there was total of 11 fatty acids including 4 saturated fatty acids and 7 unsaturated fatty acids. Quality point of saturated fatty acids was 33.9% and unsaturated fatty acids was 66.03%; in which, 50.49% of monounsaturated fatty acids and 15.54% of polyunsaturated fatty acids. The fatty acids in Cranoglanis meat contain the three main types: C18:1, C16:0 and C18: 2n-6, all of which have a quality point of 80.44% in total. In addition, the fatty acid contents in Cranoglanis meat differs significantly from other economically important fish species.

However, the wild Helmet castfish resources in Viet Nam are severely decreased due to overexploitation, human activities that causes changing of the flow, residence and breeding grounds. Recently, Red List of Threatened Species of International Union for Conservation of Nature (IUCN, 2017) rate Helmet castfish under Vulnerable (VU) and belong to the list of valuable, extinct risk species that need to be protected, restored and developed according to the Decision No. 82/2008-QD-BNN of the Ministry of Agriculture and Rural Development (the species is risk in vulnerable - VU).

Based on the above scientific and practical requirements, with the consent of the Institute of Ecology and Biological Resources, Vietnam Academy of Science and Technology, we have selected and implemented the thesis: “*Study on biological characteristics and seed production techniques of the Helmet Catfish - Cranoglanis bouderius* (Richardson, 1846) in culture conditions of Nghe An province”. The
results of the study will become the scientific basis for the conservation and resources development of this species. Also, it can be the premise for research on seed production in order to diversify the cultured freshwater species in Nghe An and Northern Central in Vietnam.

2. The objectives of the thesis

To provide scientific data on biological characteristics, breeding techniques, nursing and grow-out of Helmet Catfish - *Cranoglanis boudierius* (Richardson, 1846) in culture condition of Nghe An province. The success of the thesis will greatly contribute the basic knowledge to develop the seed production technique that can help to support the seed resources for commercial culture models and gene resources conservation for this species.

3. The contents of the thesis

3.1. Research on biological characteristics of wild Helmet Catfish collected in Nghe An province

- Research on classification characteristics of the Helmet Catfish;
- Research on morphology, surgery characteristics of the Helmet Catfish;
- Research on distribution characteristics of the Helmet Catfish in the wild;
- Research on nutrition characteristics of the Helmet Catfish in the wild;
- Research on growth characteristics of the Helmet Catfish in the wild;
- Research on reproductive characteristics of the Helmet Catfish in the wild.

3.2. Research on reproduction technique of Helmet Catfish in culture conditions

- Research on reproductive techniques of the Helmet Catfish;
- Research on egg incubation techniques of the Helmet Catfish;
- Research on nursing and grow-out techniques of the Helmet Catfish.

Chapter 1. OVERVIEW OF DOCUMENTS

1.1. Scientific classification

- Kingdom: Animalia
- Phylum: Chordata
- Class: Actinopterygii
- Order: Siluriformes
- Family: Cranoglanididae
- Genus: Cranoglanis
- Species: *C. boudierius*
1.2. Research situation on Helmet Catfish

1.2.1. The research situation on Helmet Catfish in the world

In 1846, Richardson have based on a Chinese oil painting in which the Helmet Catfish - *Cranoglanis bouderius* was firstly described.

Up to 1880, Peter have described Helmet Catfish with a new species, *Cranoglanis sinensis*. In 1839, Vaillant have described *Cranoglanis henrici*, although not much attention was paid.

Jayaram (1955) have concluded that two species of *C. multiradiatus* and *C. sinensis* was only one species of *C. bouderius*. Recently, *C. bouderius* and *C. multiradiatus* have been described as two different species.

Zhang et al. (2009) have resulted that the meat content and nutritional composition of Helmet Catfish were 69.92% of meat, 17.89% of protein, 5.20% of fatty axit and 1.10% of ash.

Zhou et al. (2012) have experimented the artificial breeding of Helmet Catfish using LHRH-A, DOM and carp pituitary. After that, the embryo development also have been checked.

1.2.2. The situation on Helmet Catfish research in Vietnam

Cao Xuan Dung et al. (2010) have studied on the reproductive biological characteristics and experimented on artificial breeding of *C. henrici*. Nguyen Dinh Vinh et al. (2013) have conducted a research on some reproductive biological characteristics and spawning under artificial conditions for *C. bouderius* in North Central Vietnam.

Chapter 2. MATERIALS, CONTENTS AND METHODS

2.1. Materials

Helmet Catfish - *Cranoglanis bouderius* (Richardson, 1846) collected in Nghe An province.

2.2. Methods

2.2.1. Theoretical bases
2.2.2. Method on biological characteristics

2.2.2.1. Sample collection method: referenced from "Fish Study Guide" of Pravdin (1963);

2.2.2.2. Species identification based on morphology and molecular biology methods


c. Species identification method by molecular markers: extracting total DNA from the fin-clip, amplifying the COI gene by PCR, sequencing, aligning, analysing, calculating genetic distance and species identification.

2.2.2.3. Research method of distribution characteristics

2.2.2.4. Research method of nutritional characteristics of the Helmet Catfish

a. Determination of the digestive organs


2.2.2.5. Research method of growth characteristics: references from Laurencen (1951)

2.2.2.6. Research methods of reproductive characteristics

* Determining maturity coefficient: Determining the reproductive of the fish.

2.2.3. **Research methods of artificial reproduction techniques**

2.2.3.1. **Research methods of domestication:**

* Selection of domesticated fish: Domesticated fishes have a good appearance, no deformities; no signs of disease and > 0.5 kg/fish.

* Determination of culture model: The experiment includes 3 trials on culture system, repeated twice and 10 fish for each trial. Trial 1: using hapa size at 6 x 4 x 1.5m placed in 800 m2 earthen pond; 1.3 - 1.8 m in depth; Trial 2: using composite tank with a volume of 30 m$^3$; Trial 3: using 500 m2 ponds; 1.3 - 1.8 m in depth.

* Determination of feed: The experiment includes 3 trials on feed formulas, repeated 3 times and using 50 m2 earthen pond with 1.3 - 1.8 m in depth. The fish density is 1 fish/m2: Trial 1: (TA1): Using 100% trash fish; Trial 2: (TA2): using 95% of trash fish and 5% of bloodworm; Trial 3: (TA3): using commercial feed (40% protein).

2.2.3.2. **Research method of Helmet Catfish production technique**

*a. Research methods of broodstock at pre-spawning stage*

**Experiment 1: Experiment on broodstock pre-spawning stage from different feed sources**

Experimental design: The experiment includes 3 trials on feed formulas, repeated 3 times and using hapa in earthen pond. The fish density is 10 pairs of broodstock/hapa (1 male: 1 female). Trial 1 (TA1): using 100% trash fish; Trial 2 (TA2): using 50% trash fish and 50% commercial feed (40% protein); Trial 3 (TA3): 100% commercial feed (40% protein).

*b. Research methods of spawning technique*

**Experiment 2: Research on types and doses of gonadotropin on reproductive of the Helmet Catfish**

* Broodstock standard: size: 1.02-1.2 kg
* Experimental design: 6 trials for male fish and 6 trials for female fish, each trial is repeated four times.

Trial 1: 30 μg LRHa + 9 mg DOM/kg female; Trial 2: 40 μg LRHa + 9 mg DOM/kg female; Trial 3: 50 μg LRHa + 9 mg DOM/kg female; Trial 4: 2500 IU
HCG/kg female; Trial 5 (CT5): 3000 IU HCG/kg female; Trial 6 (CT6): 3500 IU HCG/kg female. The dose injected to the male is 1/3 the dose for the female fish.

**Experiment 3: Determine the appropriate fertilization method to achieve high fertilization rates**

This experiment includes 2 trials, repeated 4 times and randomly designed. Trial 1: dry fertilization method (TT1) and Trial 2: natural fertilization method (TT2).

**Experiment 4. Egg incubation with different incubators**

Eggs are incubated with different types of devices: AT1: Incubated in a spongy tank (0.4mx0.3mx0.3m) with aeration; AT2: Incubated on tilapia egg incubation tray: (0.37m × 0.23m × 0.05m), the egg are submerged in water 3-4 cm.

c. **Research method of Helmet Catfish nursing technique**

**Experiment 5: Nursing of fry in different models**

* Experiment design: Newly hatched fish are cultured in two trial of culturing systems: CT1: using hapa (2 x 2 x 1.2 m) placed in ponds; CT2: using composite tank (3m³). The culture time is 28 days at density of 500 fry/m³. Each trial is repeated randomly 3 times.

**Experiment 6: Effect of densities on survival and growth rate of Helmet Catfish from fry to fingerling stage**

The experiment is conducted with 4 different densities (MD): MD1: 40 fish/m²; MD2: 50 fish/m²; MD3: 60 fish/m² and MD4: 70 fish/m², arranged randomly in 12 hapas. Each trial is repeated 3 times. The fish is fed fully the bloodworm with the frequency twice daily, at 7 am and 4pm for the period of 60 days.

**Experiment 7: Effect of feed on survival rate and growth rate of the Helmet Catfish from fry to fingerling stage**

The experiment is conducted with 3 trials (TA): TA1: using 100% of grinding trash fish; TA2: using 100% of blood worms; TA3: using homemade feed consisting of 50% fish meal and 50% soy meal. Each trial is repeated 3 times and randomly arranged in 9 hapas.

2.2.3.3. **Research method of Helmet Catfish diseases**


* Infection method: For each bacterial strain, two trials will be designed. For each trial, 5 healthy fish are kept in the tank and injected by 0.5 ml of bacterial suspension with concentrations of $10^4; 10^5, 10^6, 10^7, 10^8$ (cfu/ml). For the trial 2, the control fish is injected using physiological saline.
*Antibiogram test method*

According to Bauer (1997), the results were recorded using diffusion method on agar plate with standard antibiotic paper plates (CODE 1334-OXOID). Antibiotics method include Ampicillin (10μg), Erythromycin (30μg), Tetracyclin (30μg), Allium sativum L. (150 μl), Psidium guajava L. (150 μl). The results were recorded using diffusion method on agar plate with standard disposable paper plates (CODE 1334-OXOID).

2.2.3.4. Monitoring criteria: Environmental factors; fish reproduction characteritics; growth rate and survival rate.

2.2.4. Data analysis

The data are analysed using statistical analysis in biological research by SPSS 16.0 software. The differences between experimental trials are compared by applying DUNCAN test at $\alpha = 0.05$.

2.3. Research location and time: Research location: Nghe An province; Research time: From January 2014 to September 2017.

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**Chapter 3. RESULTS AND DISCUSSION**

3.1. Results on the research of biological characteristics of the Helmet Catfish

3.1.1. Morphology identification and molecular biology

3.1.1.1. Results of morphological classification by measurement and counting based

Based on the results of X-ray analysis of 26 samples collected at Tuong Duong, Con Cuong, Thanh Chuong and Nam Dan district (Nghe An Province, two species: C. bouderius and C. henrici (Figure 3.1) have been identified.

![Figure 3.1. X-ray image of fish sample](image)

3.1.1.2. Classification results by molecular biology: DNA analysis results show that the length of gene is from 634 bp to 689 bp, coverage ranged from 83 to 97% and the identity reach over 98% compare to the samples registered on the genebank with code JTA292338.1. The results have shown the scientific name of all investigated samples which are the C. bouderius species.

3.1.2. Research on morphological characteristics of Helmet Catfish

3.1.2.1. Descriptions: Helmet Catfish has elongated body, flat side toward the tail. The
distance from the top of the snout to the back of the dorsal fin look like a straight line. The end of tail shrinks. The head is vertical flat, with a pyramid shape. Distance between two orbits is wide, with a deep groove running from the occipital to the end of eye and in the middle of the head. The fish body is white silver, the upper back is light gray. The body is smooth with no scales.

3.1.2.2. Measurement criteria: The results above are consistent with the description about Helmet Catfish following Nguyen Van Hao (2005) and Ng. and Kottelat (2000).

3.1.3. Results of survey and distribution area

Helmet Catfish are distributed in four sampling collection sites along the Lam River system.

Helmet Catfish fingerlings appeared at all four sample collection sites; they presented more at Nam Dan and Thanh Chuong, while growth fish appeared more at Tuong Duong and Con Cuong (Table 3.5).

Table 3.5. Distribution of Helmet Catfish

<table>
<thead>
<tr>
<th>Location</th>
<th>March</th>
<th>April</th>
<th>May</th>
<th>June</th>
<th>July</th>
<th>August</th>
<th>September</th>
<th>October</th>
</tr>
</thead>
<tbody>
<tr>
<td>TD</td>
<td>0</td>
<td>47</td>
<td>21</td>
<td>35</td>
<td>48</td>
<td>27</td>
<td>29</td>
<td>37</td>
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<tr>
<td>CC</td>
<td>0</td>
<td>55</td>
<td>27</td>
<td>42</td>
<td>57</td>
<td>24</td>
<td>25</td>
<td>46</td>
</tr>
<tr>
<td>TC</td>
<td>0</td>
<td>25</td>
<td>52</td>
<td>12</td>
<td>27</td>
<td>54</td>
<td>58</td>
<td>21</td>
</tr>
<tr>
<td>ND</td>
<td>0</td>
<td>22</td>
<td>57</td>
<td>20</td>
<td>20</td>
<td>57</td>
<td>55</td>
<td>18</td>
</tr>
<tr>
<td>Total</td>
<td>0</td>
<td>149</td>
<td>157</td>
<td>109</td>
<td>152</td>
<td>162</td>
<td>167</td>
<td>122</td>
</tr>
</tbody>
</table>


3.1.4. Nutritional characteristics of Helmet Catfish

* Digestive organ structure of the Helmet Catfish

Figure 3.24. Mouth shape

Figure 3.25. Gill shape

Figure 3.26. Oesophagus

Figure 3.27. Oesophagus cross-section

A: Round muscle, B: Longitudinal muscle, C: mucous membrane, D: muscularis
**Appearance Frequency of feed for Helmet Catfish**

Analysis of feed in the stomach and intestines of 87 Helmet Catfish samples found that common feed species such as small fishes, crustacean (shrimps, crabs), insects (*worms, white ant*), *organic dust*, ... in which organic dust appears with highest frequency (54.02%).

**The diversity of feed of Helmet Catfish**

Crustacean, fish, organic dust are the main feed ingredients which are found in the peptogaster of Helmet Catfish.

**Correlation of intestinal length and standard length of the Helmet Catfish**

The survey results on the intestinal length and the body length of the Helmet Catfish on 95 samples showed that: RLG index = 1.23.

Conclusions: From external morphological characteristics and structure of some organs inside peptogaster of the Helmet Catfish such as: Location of mouth, teeth, gill, esophagus, structural size of stomach and intestine shown that eating behavior of the Helmet Catfish is an omnivorous prefer to animal.

**3.1.5. Growth characteristics of the Helmet Catfish**

The result of the regression equation between the length and the body weight of the Helmet Catfish is: \( W = 0.00492L^{2.90718} \).

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*Figure 3.28. Stomach*

*Figure 3.29. Stomach cross-section (A: Wall, B: Under mucosa, C: Mucosa, D: sinus vessels)*

*Figure 3.30. Intestine*

*Figure 3.31. Intestinal cut (A: Outer membrane, B: smooth muscle, C: branch of folds, D: subcutaneous layer, E: sinus capillary with erythrocyte)*

*Figure 3.34. Feed appearance frequency (n = 87)*

*Figure 3.35. Feed spectrum*

*Figure 3.36. Graph the relationship between length and volume*
The results show that this correlation is a positive correlation between length and weight with growth coefficient $b=2.90718 \pm 0.01$ and condition coefficient $0.0049\pm0.0003$.

3.1.6. Reproductive characteristics

* Sexual characterization

![Female fish](Figure 3.37) ![Male fish](Figure 3.38) ![Ovary](Figure 3.40) ![Oophorogenesis](Figure 3.42)

* Gonadogenesis stages

- **a / The development stages of the oocyte:**
  - Stage I: The gonad is thin, transparent, sometimes yellowish or pinky, which cannot distinguish by the eye.
  - Stage II: The size of gonad increases and divides clearly the lobe, accounting for $1/3 \div 1/4$ volume of abdominal cavity. Observations on the specimen found oocytes with large, round nucleus, located in the middle.
  - Stage III: Gonad grow rapidly with increasing size apprettly and occupy $1/3 \div 1/2$ volume of abdominal cavity, light yellow oocyte, blood vessels are distributed on the oocyte. Observations on specimen shown that the egg cells have transferred to the nutritious stage, also known as growth on nutrition, this lead to increases quickly the size of the oocytes by the number of granular yolks and vacuoles.
  - Stage IV: The gonad size is larger which occupied most of the abdominal cavity. The blood vessels were distributed much more in the ovaries, and the yellowis slighter and darker than the ovaries in stage III. The large eggs, the binding force between the egg cells decreases which lead to the egg have tend to separate.
  - Stage V: Spawning stage, fish have a big abdomen, abdominal walls are soft and go down in two sides, genital holes are large, slightly convex. The ovaries are circular stretch, yellow-brown or red-brown, and have large blood vesels on the membranes. When stroking lightly on the belly, the egg will flow from the genital holes. Observation on the specimen, the eggs are round and easy to separate.
  - Stage VI: Stage after spawning. At that time, the shape of ovary flushed down, pastry and bluish red due to hemorrhage when the egg follicle break down.
b/ The development stages of the oophortestis

- Stage I: At this stage, there is the presence of spermatogonium, the number of large spermatogonium lie in the cysts.

- Stage II: The oophortestis are long, small, white or opalescent, the blood vessels are unclear. The number of spermatogonium increases rapidly and fold into the clusters shapes which form small and solid spermiducts, among these vases have the connective tissue to detach.

- Stage III: The oophortestis have larger size, opalescent, on the surface appear more pink tracks, this marks the development of blood vessel. Observation on the specimen, mainly secondary spermatogonium is in the period of separation into the andro spernum.

- Stage IV: The size of oophortestis are larger than those in the previous stage with milky white, the blood vessels grow strongly, forming the spermatozoa chambers in the oophortestis, in the middle of the spermatozoa chambers are dense spermatozoa.

- Stage V: Spermatozoa become ripe with light or yellowish-white, blood vessels grow clearly. Observations on the sections in the spermatocyst indicate a lot of sperm.

- Stage VI: After spawning stage, the semen has run out, the oophortestis shrink like a thin strip. Blood vessels are wider so the oophortestis are pink or brown.
* Reproductive season of the Helmet Catfish

a/ Mature coefficient fluctuation:

Figure 3.62 shows that GSI of female and male Helmet Catfish vary continuously for 12 months. The GSI coefficient of females and males increases from January and reach the highest peak in June (Females 4.00%, males 3.01%) before decrease to the lowest in December (Females 1.56%, males 0.44%).

b/ Fluctuation of mature stages of the gonadal: The gonadal mature rate of female and male fishs increases from April to June. During this period, the gonads of Helmet Catfish in stages III, IV and V occupy over 90%.

c/ Fluctuation of Fulton Fatness Factor and Clark Fatness Factor:

The Fulton and Clark fatness values of the Helmet Catfish vary according to the months, ranging from 2.10 to 3.01% (Fulton fatness) and 1.87 to 2.74% (Clark fatness).

The highest fatness of Helmet Catfish in January are 3.01% (Fulton fatness) and 2.74% (Clark fatness), after that the fatness reduces slowly and lowest in June (2.10% Fulton fatness and 1.87% Clark fatness).
Reproductive season: Reproductive season of the Helmet Catfish in the North Central is concentrated in April to June every year. Therefore, in the artificial reproductive process, it is necessary to base on the reproductive season to create appropriate conditions such as environmental factors, spawning grounds, rate of flow... to stimulate fish reproduction which can get the highest spawning rate.

3.1.6.6. Reproductive fertility

The results of analysis 85 female Helmet Catfish collected from January to December 2014 showed that the absolute reproductive fertility of the this species ranged from 5348 to 14867 eggs/female, the relative reproductive fertility ranged from 25 to 32 eggs/g female with weight varies from 465.95 to 1131.5 g/fish.

3.1.6.7. The stages of embryo development of the Helmet Catfish

The egg segment process of the Helmet Catfish is uncompleted.

Conclusion: The incubated period of the Helmet Catfish egg prolongs from 26 - 27 hours. It is divided into some stages: Segment: about 3 - 4 hours; Embryo development: about 5-6 hours; Morula: about 2 hours; Differentiation of embryonic stem consists of formation of chordate, neural tube, eye nerve, ear sac and other organs prolonged from 14 to 17 hours.

3.2. Results of reproduction technique of Helmet Catfish in culture condition of Nghe An Province

3.2.1. Research on Helmet Catfish production techniques

3.2.1.1. Testing results some kind of feeds for broodstock pre-spawning
Table 3.13. Testing results some kind of feeds for broodstock pre-spawning (TB ± SD)

<table>
<thead>
<tr>
<th>Criteria</th>
<th>TA1</th>
<th>TA2</th>
<th>TA3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Males mature rate (%)</td>
<td>82.26(^a) ± 8.7</td>
<td>94.76(^a) ± 8.5</td>
<td>95.76(^a) ± 7.2</td>
</tr>
<tr>
<td>Females mature rate (%)</td>
<td>82.25(^a) ± 8.4</td>
<td>94.76(^a) ± 8.6</td>
<td>100(^a) ± 00</td>
</tr>
<tr>
<td>Rate of fish with eggs stage III (%)</td>
<td>8.2</td>
<td>10.3</td>
<td>11.4</td>
</tr>
<tr>
<td>Rate of fish with eggs do not grow (%)</td>
<td>11.2</td>
<td>7.4</td>
<td>0.0</td>
</tr>
</tbody>
</table>

From the results obtained in Table 3.13, the quality of reproductive performance of craw brooders formulas by experimental diets generally showed a high mature rate.

Using different feed sources to pre-spawning of the brooders, the results were shown in Table 3.14.

Table 3.14. Results for artificial reproduction at pre-spawning stage

<table>
<thead>
<tr>
<th>Criteria</th>
<th>TA1</th>
<th>TA2</th>
<th>TA3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of female can be reproduction (fish)</td>
<td>5</td>
<td>7</td>
<td>9</td>
</tr>
<tr>
<td>Fish size in the reproduction group (kg/fish)</td>
<td>1-1.2</td>
<td>1-1.2</td>
<td>1-1.2</td>
</tr>
<tr>
<td>Total weight of female (kg)</td>
<td>6.00</td>
<td>8.40</td>
<td>10.98</td>
</tr>
<tr>
<td>Ovulation rate (%)</td>
<td>88.4</td>
<td>90.2</td>
<td>91.3</td>
</tr>
<tr>
<td>Realistic reproductive capacity (egg/kg)</td>
<td>2548 ± 221</td>
<td>2554 ± 301</td>
<td>2586 ± 287</td>
</tr>
<tr>
<td>Fertilization rate (%)</td>
<td>50.13±18.34</td>
<td>65.04±17.36</td>
<td>66.18±16.54</td>
</tr>
<tr>
<td>Hatching rate (%)</td>
<td>11.3±11.21</td>
<td>16.21±9.51</td>
<td>29.5±13.62</td>
</tr>
<tr>
<td>Deformation rate (%)</td>
<td>2.51 ± 0.21</td>
<td>1.71 ± 0.01</td>
<td>1.49 ± 0.16</td>
</tr>
<tr>
<td>Number of fry (fish)</td>
<td>866</td>
<td>2261</td>
<td>5543</td>
</tr>
<tr>
<td>Fry capacity (fry/kg female)</td>
<td>144</td>
<td>269</td>
<td>504</td>
</tr>
</tbody>
</table>

Table 3.14 reveals that the results of artificial reproduction in the broodstock pre-spawning stage used 100% commercial feed showed the highest results such as number of reproductive females (9 fish) , the ovulation rate (91.3%), real reproductive capacity (2586 eggs/kg), fertilization rate (66.18%), hatching rate (29.5%), fry capacity (504 fry/kg female). Thus, it may be advisable that pre-spawning the brooders should use commercial feed with a protein content of 40%.
3.2.1.2. Results of technical for reproduction of Helmet Catfish

a. Stimulation Helmet Catfish reproductive by using different gonadotropin dosages

Testing results show that using HCG with a dose of 2,500 IU to 3,500 IU or combination with 9 mg DOM along with amount LRHa of 30μg to 50μg reach the rate of females that can stroke eggs to the absolute ratio (100%).

**Table 3.15.** The number, rate of mature females and reproductive rate of the Helmet Catfish when using different gonadotropin types and doses

<table>
<thead>
<tr>
<th>Experimental formula</th>
<th>Number of fish can release eggs</th>
<th>Rate of fish release eggs (%)</th>
<th>Female mature rate (%)</th>
<th>Male mature rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CT1</td>
<td>5</td>
<td>100</td>
<td>2.16</td>
<td>0.19</td>
</tr>
<tr>
<td>CT2</td>
<td>5</td>
<td>100</td>
<td>2.18</td>
<td>0.21</td>
</tr>
<tr>
<td>CT3</td>
<td>5</td>
<td>100</td>
<td>2.14</td>
<td>0.22</td>
</tr>
<tr>
<td>CT4</td>
<td>5</td>
<td>100</td>
<td>2.21</td>
<td>0.20</td>
</tr>
<tr>
<td>CT5</td>
<td>5</td>
<td>100</td>
<td>2.19</td>
<td>0.23</td>
</tr>
<tr>
<td>CT6</td>
<td>5</td>
<td>100</td>
<td>2.17</td>
<td>0.22</td>
</tr>
</tbody>
</table>

**Table 3.16.** Effective time and egg capacity of female fish using different types and doses of gonadotropin (TB ± SD)

<table>
<thead>
<tr>
<th>Experimental formula</th>
<th>Effective Time (min)</th>
<th>Absolute reproduction (thousand eggs/female)</th>
<th>Relative reproduction (1000 eggs/kg female)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CT1</td>
<td>542.00 ± 82.25&lt;sup&gt;ab&lt;/sup&gt; 420 ÷ 600</td>
<td>3.21 ± 14.91&lt;sup&gt;a&lt;/sup&gt; 2.20 ± 4.04</td>
<td>1.84 ± 2.11&lt;sup&gt;a&lt;/sup&gt; 0.71 ± 2.01</td>
</tr>
<tr>
<td>CT2</td>
<td>593.75 ± 88.63&lt;sup&gt;b&lt;/sup&gt; 490 ÷ 670</td>
<td>3.06 ± 18.28&lt;sup&gt;a&lt;/sup&gt; 1.70 ± 7.72</td>
<td>1.04 ± 4.06&lt;sup&gt;a&lt;/sup&gt; 0.67 ± 1.74</td>
</tr>
<tr>
<td>CT3</td>
<td>627.50 ± 57.37&lt;sup&gt;b&lt;/sup&gt; 560 ÷ 680</td>
<td>4.04 ± 12.57&lt;sup&gt;a&lt;/sup&gt; 2.30 ± 6.75</td>
<td>1.61 ± 2.66&lt;sup&gt;a&lt;/sup&gt; 0.16 ± 2.30</td>
</tr>
<tr>
<td>CT4</td>
<td>483.00 ± 78.63&lt;sup&gt;a&lt;/sup&gt; 430 ÷ 600</td>
<td>5.61 ± 16.46&lt;sup&gt;a&lt;/sup&gt; 3.20 ± 9.27</td>
<td>2.41 ± 3.49&lt;sup&gt;a&lt;/sup&gt; 1.23 ± 3.62</td>
</tr>
<tr>
<td>CT5</td>
<td>519.00 ± 74.17&lt;sup&gt;ab&lt;/sup&gt; 460 ÷ 615</td>
<td>2.14 ± 1.37&lt;sup&gt;a&lt;/sup&gt; 1.60 ± 2.90</td>
<td>2.01 ± 0.42&lt;sup&gt;a&lt;/sup&gt; 1.63 ± 3.50</td>
</tr>
<tr>
<td>CT6</td>
<td>566.75 ± 84.16&lt;sup&gt;ab&lt;/sup&gt; 465 ÷ 662</td>
<td>2.37 ± 10.39&lt;sup&gt;a&lt;/sup&gt; 1.30 ± 3.70</td>
<td>1.18 ± 4.06&lt;sup&gt;a&lt;/sup&gt; 1.0 ± 2.27</td>
</tr>
</tbody>
</table>

The results in Table 3.16 show that the average effective time of reproduction in each trial ranged from 483.00 minutes to 627.50 minutes (i.e. from 7 hours to 10.5...
hours). Absolute reproductive ability of Helmet Catfish ranged from 2.14 to 5.61 (thousand eggs/female) and relative reproductive ability of this species ranged from 1.04 to 2.61 (thousand eggs/kg female), the difference among the trials have no statistic significance (p> 0.05).

**Figure 3.73.** Fertility and hatching rate of the Helmet Catfish when using different types and dosages of gonadotropin

Figure 3.73 shows that using CT5 to stimulate broodstocks reproduction with different types and doses of gonadotropin shows the highest fertilization and hatching rate (fertilization rate is 58% and hatching rate is 23%) and the lowest results belong to CT3 (fertilization rate is 35.1% and hatching rate is 12%).

From this results we suggest that 30μg LRHa should combinewith 9mg DOM per one kg of female or HCG at dose of 2500 IU HCG/kg female to stimuate Helmet Catfish reproduction.

**b. Results of fertilization test for Helmet Catfish caviar**

The results showed that Helmet Catfish egg was fertilized by dry fertilization method gain the rate of 50.34% that was higher than by natural fertilization method (23,14).

**Figure 3.74.** The fertilization rate of Helmet Catfish egg with different methods
c. Testing results of the Helmet Catfish egg incubation

**Table 3.17.** Effects of egg incubation methods on hatching and
derformation rate of Helmet Catfish

<table>
<thead>
<tr>
<th>Trial</th>
<th>Number of incubating eggs (psc)</th>
<th>Incubation density (eggs/cm²)</th>
<th>Hatching rate (%)</th>
<th>Deformation rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>AT1</td>
<td>5000</td>
<td>10</td>
<td>22.51 ± 7.10</td>
<td>1.72 ± 0.01</td>
</tr>
<tr>
<td>AT2</td>
<td>5000</td>
<td>10</td>
<td>13.69 ± 6.50</td>
<td>1.84 ± 0.03</td>
</tr>
</tbody>
</table>

The results in Table 3.17 show that the hatching rate was higher when eggs incubated in spongy tank using air bubbling (AT1) with 22.51% and lower when they are incubated in the tilapia incubator tray (AT2) which only reached 13.69%.

3.2.2.3. Results of Helmet Catfish nursing

a. Experiment of nursing Helmet Catfish from fry to fingerling by using different methods.
a1) Survival rate of fry
The obtained results in Figure 3.75 show the survival rate of nursing process. Survival rate of fish grow in tanks after 28 days was 84%, higher than those in the hapa (69%).

![Figure 3.75. Survival rate of fish growth seedlings in different forms](image)

a2) Growth of body length and weight of the Helmet Catfish
The results of experiments of Helmet Catfish nursing in 28 days (4 weeks) were presented in Table 3.18.

**Table 3.18.** The growth of fish using different methods

<table>
<thead>
<tr>
<th>Targets</th>
<th>Unit</th>
<th>Fish growth in the nets</th>
<th>Fish growth in the tank</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fish size (Start)</td>
<td>P (g)</td>
<td>0.03 ± 0.0096&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.03 ± 0.009&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>L (mm)</td>
<td>3.97 ± 0.632&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.96 ± 0.705&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Fish size (Finish)</td>
<td>P (g)</td>
<td>0.12 ± 0.010&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.10 ± 0.016&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>L (mm)</td>
<td>13.09 ± 1.025&lt;sup&gt;b&lt;/sup&gt;</td>
<td>11.52 ± 0.96&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>ADG</td>
<td>P (g/day)</td>
<td>0.003 ± 0.000&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.003 ± 0.000&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>L (mm/day)</td>
<td>0.33 ± 0.004&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.270 ± 0.008&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>
The results in Table 3.18 show that the size of fish at beginning is the same, after the fish are nursed in the hapa, the results of weight, length, and average daily growth speed as follow (P: 0 12 g/individual; L: 13.09 mm; P: 0.003 g/individual; L: 0.33 mm/individual) that are higher than those cultured in the tank.

*b. Testing results of Helmet Catfish fry growing with different feed sources*

**Table 3.19.** Growth (quantity, g) of the Helmet Catfish using feed

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Experimental feed</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Trash fish</td>
</tr>
<tr>
<td>$W_0$ (g)</td>
<td>1.05 ± 0.01&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>$W_{fl}$ (g)</td>
<td>4.80 ± 0.23&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>AG (g)</td>
<td>3.75 ± 0.23&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR (%/day)</td>
<td>2.53 ± 0.08&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>Blood worms</td>
</tr>
<tr>
<td>$W_0$ (g)</td>
<td>1.06 ± 0.02&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>$W_{fl}$ (g)</td>
<td>6.82 ± 0.30&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>AG (g)</td>
<td>5.76 ± 0.28&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR (%/day)</td>
<td>3.11 ± 0.05&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>Pale fish meal + Soy meal waste (rate 1:1)</td>
</tr>
<tr>
<td>$W_0$ (g)</td>
<td>1.06 ± 0.01&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>$W_{fl}$ (g)</td>
<td>4.95 ± 0.28&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>AG (g)</td>
<td>3.89 ± 0.28&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR (%/day)</td>
<td>2.57 ± 0.10&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

**Table 3.20.** Growth (size, cm) of the Helmet Catfish by experimental feed

<table>
<thead>
<tr>
<th>Size criteria</th>
<th>Experimental feed</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Trashfish</td>
</tr>
<tr>
<td>$TL_0$ (cm)</td>
<td>2.25±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>$TL_{fl}$ (cm)</td>
<td>12.94±0.28&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>AG (cm)</td>
<td>10.70±0.30&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR (%/day)</td>
<td>2.92±0.05&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>CV60 (%)</td>
<td>3.44±0.09&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>Bloodworm</td>
</tr>
<tr>
<td>$TL_0$ (cm)</td>
<td>2.23±0.05&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>$TL_{fl}$ (cm)</td>
<td>16.47±0.45&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>AG (cm)</td>
<td>14.24±0.42&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR (%/day)</td>
<td>3.33±0.03&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>CV60 (%)</td>
<td>2.74±0.33&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>Pale fish meal + Soy meal waste (rate 1:1)</td>
</tr>
<tr>
<td>$TL_0$ (cm)</td>
<td>2.25±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>$TL_{fl}$ (cm)</td>
<td>13.27±0.87&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>AG (cm)</td>
<td>11.03±0.86&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR (%/day)</td>
<td>2.96±0.10&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>CV60 (%)</td>
<td>3.32±0.48&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Results in Table 3.19, 3.20 and Figure 3.76 show that: Bloodworm is the best diet in the test, which should be used as a optimal feed for the Helmet Catfish nursing period of fry to nursing in order to increase the growth rate of fish and reduce rearing time.

*Figure 3.76.** Survival rate of the Helmet Catfish by experimental feed
c. Results of the Helmet Catfish nursing test in the fingerlings period at the different densities

*Effect of nursing density on growth rate*

**Table 3.21.** Growth (quantities, g) of the Helmet Catfish by density

<table>
<thead>
<tr>
<th>Criteria</th>
<th>Experimental density (fish/m²)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>40</td>
</tr>
<tr>
<td>$W_0$ (g)</td>
<td>1.03±0.04&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>$W_{fl}$ (g)</td>
<td>7.51±0.77&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>AG (g)</td>
<td>6.48±0.74&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR (%/day)</td>
<td>3.30±0.12&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

**Table 3.22.** Growth (size, cm) of the Helmet Catfish by density

<table>
<thead>
<tr>
<th>Size criteria</th>
<th>Experimental density (fish/m²)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>40</td>
</tr>
<tr>
<td>TL₀ (cm)</td>
<td>2.20±0.02&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>TL₈ (cm)</td>
<td>16.11±0.83&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>AG (cm)</td>
<td>13.91±0.83&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGR (%/day)</td>
<td>3.32±0.09&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>CV (60, %)</td>
<td>2.46±0.11&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

The results in Table 3.21, and 3.22 show that Helmet Catfish fry were selected for nursing with density of 40 individuals/m2 achieving the best of weight and length.

In terms of distinguished size level, the fish should be nursed at the density of 40 to 50 individuals/m2.

*2) Effect of nursing density on survival rate*

The testing results showed that the density of 50 individuals/m2 was suitable for nursing the Helmet Catfish from fry to fingerling, which can be used to supplement in the fingerling production process.

**Figure 3.77.** Survival rate of rotifers according to experimental density
3.2.3. Results of the Helmet Catfish disease

* Results of bacterial separation in the Helmet Catfish

Figure 3.78. Some signal of abnormal Helmet Catfish

- Fish has meteorism
- Coin sores
- Sore in tail

Isolation and identification by Nicky B. Buller (2004), the results are as follow:

- *Aeromonas hydrophila* (Chester, 1901)
- *Streptococcus* sp.

* Prevalence of bacterial species in the Helmet Catfish

With 120 Helmet Catfish samples collected from natural zones and the fish were collected at domesticated Freshwater Aquaculture Farms show that the prevalence of bacterial species varied among months of the year.

In which *Aeromonas hydrophila* is present in whole months of year, but the increasing from July to September, and the highest in August with a prevalence of 60%. For *Streptococcus* sp., prevalence is lower, some months in the year such as February, November there is not present of this bacteria in the collected samples. However, the rate of bacterial prevalence is high in May, June and August, with a rate of 30%, the highest rate is in September with a prevalence of 40%.

* Testing results of antibiotic susceptibility of bacterial species

Table 3.24. Results on antibiotic susceptibility of bacterial species

<table>
<thead>
<tr>
<th>Bacteria</th>
<th>Ampicillin (10µg)</th>
<th>Erythromycin (30µg)</th>
<th>Tetracycline (30µg)</th>
<th>Psidium guajava L (150µl)</th>
<th>Allium sativum L (150µl)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>A. hydrophila</em></td>
<td>22.7 ± 1.36</td>
<td>21.5 ± 0.82</td>
<td>22.8 ± 0.54</td>
<td>15.5 ± 1.12</td>
<td>18.7 ± 0.45</td>
</tr>
<tr>
<td><em>Streptococcus</em> sp.</td>
<td>20.5 ± 1.43</td>
<td>21.0 ± 0.92</td>
<td>20.3 ± 0.6</td>
<td>15.5 ± 1.2</td>
<td>20.9 ± 1.01</td>
</tr>
</tbody>
</table>

(The data in the same row with different exponents sign appearing the difference in statistical significance P <0.05).
The results showed that *A. hydrophila* bacteria are well susceptible to antibiotics Ampicillin, Erythromycin and Tetracyclin, average for *Allium sativum* L. and *Psidium guajava* L., *Streptococcus* sp. are well susceptible to Ampicillin, Erythromycin, Tetracyclin and *Psidium guajava* L., average for *Allium sativum* L.

### 3.2.4. Proposed solutions to the breeding techniques of the Helmet Catfish

Based on the research results, it is possible to propose technical solutions to produce the Helmet Catfish fry in culture conditions of Nghe An province with the following basic technical parameters:

**Table 3.27.** The main technical parameters of the process

<table>
<thead>
<tr>
<th>No.</th>
<th>Criteria</th>
<th>Unit</th>
<th>Parameters</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Mature rate of female fish</td>
<td>%</td>
<td>&gt; 95</td>
</tr>
<tr>
<td>2</td>
<td>Mature rate of male fish</td>
<td>%</td>
<td>&gt; 90</td>
</tr>
<tr>
<td>3</td>
<td>Natality rate of female fish (compared to mature fish)</td>
<td>%</td>
<td>&gt; 80</td>
</tr>
<tr>
<td>4</td>
<td>Hatching rate of fertilized eggs</td>
<td>%</td>
<td>&gt; 20</td>
</tr>
<tr>
<td>5</td>
<td>Survival rate from hatching to fry</td>
<td>%</td>
<td>&gt; 60</td>
</tr>
<tr>
<td>6</td>
<td>Survival rate from fry to fingerling</td>
<td>%</td>
<td>&gt; 70</td>
</tr>
</tbody>
</table>

**CONCLUSION AND SUGGESTIONS**

**CONCLUSION**

1. **Research on Helmet Catfish biological characteristics in Nghe An**

1.1. **For identification and distribution characteristics:** By molecular biology method, 26/26 samples collected in Nghe An province are classified into *Cranoglanis bouderius*, although the traditional classification and the Renghen method pointed out that 4/26 samples are *Cranoglanis henrici*. Small Helmet Catfish appear from April to September at the investigating sites and easy to find in Nam Dan and Thanh Chuong districts.

1.2. **For nutritional characteristics:** Helmet Catfish is an omnivorous fish, feed components in stomach consist of: tiny fish, shrimp, mussels, snails, worms, cold rice, organic dust and other feeds. It appears all levels of satiety (from level 0 to level 4) in the collected samples. The rate of intestinal length and body length $L_r/L_t = 1.23 \pm 0.01$ indicates that the Helmet Catfish is an omnivorous fish mainly on the animal.
1.3. For growth characteristics: Correlation equation between the length and quantity of the Helmet Catfish has a very closed regression relation based on the equation \( W = 0.00492L^{2.90718} \), with the determination coefficient \( R^2 = 0.9483 \). It shows that the correlation between length and quantity is positive with the growth system \( b = 2.90718 \pm 0.01 \) and the condition coefficient \( 0.0049 \pm 0.0003 \).

1.4. For reproductive biological characteristics: GSI mature coefficient and gonadal mature rate for both the Helmet Catfish females and the males increase from April to June, the Helmet Catfish fatness is highest in January and lowest in June, The reproductive season of the Helmet Catfish in Nghe An is concentrated mainly in April ÷ June. Absolute reproductive capacity of the Helmet Catfish ranged from 5348 ÷ 14867 eggs/female, the relative reproductive capacity ranged from 25 ÷ 32 eggs/g female.

2. Research on Helmet Catfish fry production techniques in Nghe An

2.1. Research on domestication techniques:

- Domesticated Helmet Catfish culture in hapa and ponds show higher survival rate of 100% and growth rate (0.83 ± 0.03 g/day in hapa and 0.77 ± 0.05 g/day in ponds) than the domesticated fish culture in the tank (survival rate is 90% and growth rate is 0.50 ± 0.04 g/day).

- The Helmet Catfish show highest growth rate when they are feed by commercial feed contains 40% protein. Following is 95% of trash fish + 5% bloodworm formular and the lowest is 100% of trash fish. The survival rate in domesticated of Helmet Catfish is relatively high, 90-100%.

2.2. Research on seed production techniques:

- Culturing the mature broodstocks use 100% commercial feed with 40% protein show the highest efficiency.

- LRHa combinate to DOM at the dose of \((30\mu g \text{ LRHa} + 9\text{mg DOM})/\text{kg female or HCG with 2500 IU HCG/kg female} \) can show the best results for stimulate Helmet Catfish reproduction. Applying dry fertilization method for Helmet Catfish egg can get the highest fertilization rate.

- Helmet Catfish in the stage from hatching to fry should be nourished in the hapa. The nourishing period from to fry to fingerling, the growth rate can reach highest at density of 50 fish/m2 (0.11 ± 0.01 g/day and 0.24 ± 0.01 cm/day) and survival rate (68.67 ± 2.67%). Therefore, it can be applied into seed production process, nursing and growing out the Helmet Catfish. Using bloodworm to feed the fry show the fastest growth rate (0.096 ± 0.005 g/day and 0.24 ± 0.01 cm/day) and the highest survival rate (70.67 ± 2.67%).
- Two pathogenic bacteria strains (*Aeromonas hydrophila* and *Streptococcus* sp.) (Richardson, 1846) were isolated for growing out stage of *Cranoglanis bouderius*. One-year monitoring results showed that high prevalence of fish was from July to September, the peak was in August with prevalence of *A. hydrophila* and September with prevalence of *Streptococcus* sp.

- *A. hydrophila* is highly susceptible to antibiotics such as Ampicillin, Erythromycin and Tetracyclin, average for Allium sativum L and *Psidium guajava* L. *Streptococcus* sp. is highly susceptible to Ampicillin, Erythromycin, Tetracyclin and Allium sativum L, average for *Psidium guajava* L.

- Based on the reproductive experiments, the study have suggested the technical solutions for seed production of *Cranoglanis bouderius* (Richardson, 1846) under artificial conditions. The results can be achieved: mature rate for female > 95%; mature rate for male > 90%; spawning rate > 80%; hatching rate > 20%; survival rate from hatching to fry > 60%; Survival rate from fry to fingerling > 70%.

**SUGGESTIONS**

- Based on the results of research on biological characteristics, domestication and reproduction of Helmet Catfish in artificial conditions in Nghe An, the production model can be expanded for this species.

- It is possible to produce fingerling of Helmet Catfish in Nghe An in particular and North Central Vietnam in general by applying all above technical solutions.

- It is necessary to further research on nutritional demands, disease prevention/treatment, technical solutions and marketing to improve the seed production efficiency of Helmet Catfish in artificial conditions.

**THE CONTRIBUTION OF THE THESIS**

This is a systematic and comprehensive research on the biological characteristics and seed production techniques of the *Cranoglanis bouderius* (Richardson, 1846), first published in Vietnam. The contents of the thesis include the following news points:

- The thesis highly contributes some important biological characteristics of *Cranoglanis bouderius* (Richardson, 1846) such as the classification exactly of this species by molecular biological and morphological methods, providing data on characteristics of growth, nutritional and reproductive of Helmet Catfish.
- In particular, the thesis has successfully researched on the production techniques of Helmet Catfish under artificial conditions such as: domestication techniques; feed mature; stimulating reproduction; fertilization and incubation; from hatching to fry and fry to fingerling stage.

- For the first time it is provided the scientific information on pathogens and initially given the suggestions on some prevention/treatment methods in Helmet Catfish.

- Based on the basis scientific experiment on artificial reproduction of the Helmet Catfish, the thesis have suggested the technical solutions for seed production of this species under conditions Nghe An province.
THE AUTHOR’S PUBLICATION LIST
REGARDING TO THE DOCTORAL THESIS


