TOPIC AREA

CLIMATE CHANGE ADAPTATION: INDIGENOUS SPECIES DEVELOPMENT

❖

SUSTAINABLE SNAKEHEAD AQUACULTURE IN CAMBODIA

ASIA PROJECT: CAMBODIA & VIETNAM
US Project PI: Robert Pomeroy, University of Connecticut – Avery Point
HC Project PI: So Nam, Inland Fisheries Research and Development Institute

Climate Change Adaptation: Indigenous Species Development/Experiment/16IND01UC

Collaborating Institutions and Lead Investigators

University of Connecticut-Avery Point (USA)	Robert Pomeroy
Inland Fisheries Research and Development Institute (Cambodia)	So Nam
	Nen Phanna
	Chheng Phen
Can Tho University (Vietnam)	Tran Thi Thanh Hien

Objectives

1. To compare growth performance and survival rate of different snakehead strains regarding to weaning and grow-out experiments.
2. To compare economic efficiency of grow-out experiments of the different snakehead strains.

Significance

In Cambodia wild snakehead were traditionally cultured in small cages and ponds prior to the snakehead aquaculture ban (see below). Feed represented more than 70% of the total operational costs and the main type of feed for wild snakehead culture was small-sized fish (SSF). Importantly, the snakehead production contributed more than 70% of total aquaculture production in Cambodia due to its popularity as food. Snakehead still has high market and trade demand in Cambodia as well as in Vietnam. Snakehead is found in most Cambodian and Vietnamese dishes at all wealth class levels (i.e., from poor, medium to rich people). During the first phase of AquaFish CRSP (2007-2009), the study revealed that 33 freshwater SSF in the Vietnam Mekong Delta (Hien et al., 2015) and nearly 200 SSF in the Cambodia Mekong basin (So Nam et al., 2009) were detected in the supply of SSF for snakehead culture in Vietnam and Cambodia, respectively, including juveniles of commercially important fish species.

The government of Cambodia put a ban on snakehead farming in September 2004 by Announcement No. 4004. After the ban on snakehead culture in Cambodia, snakehead have illegally been imported from the neighboring countries, particularly from Vietnam, to supply high local market demand in Cambodia. Furthermore, freshwater SSF have illegally been exported to Vietnam for feeding the significant and commercial snakehead aquaculture industry (So Nam et al., 2009). The incentives for choosing snakehead over other fish species by tens of thousands of fish farmers are strong as it generates more than 10 times higher profits than other fish species. These snakehead fish farmers have lost their important livelihoods and household income. Moreover, the ban also did not provide positive impacts on snakehead wild stocks as fishing pressure on wild snakehead using illegal and destructive fishing gears, particularly electro-shockers, has increased in recent years in order to supply local and external markets. In April 2016, the Government of Cambodia lifted the decade-old ban on snakehead fish farming following a request from

the Ministry of Agriculture to allow farmers to fish. Signed by Bun Uy, a secretary of state at the Council of Ministers, the statement said the decision to legalize snakehead fish farming again would be accompanied by forthcoming conditions and guidance for farmers. This investigation will provide for this research based guidance on weaning and grow-out of snakehead.

During the second phase of AquaFish CRSP (2009-2011), wild striped snakehead *Channa striata* broodstocks were successfully developed, matured and semi-artificially induced to spawn using the hormone HCG on-station in Cambodia. The *C. striata* aged 30 days old after hatching could gradually and successfully accept AquaFish CRSP Snakehead Formulated Feed developed by the AquaFish CRSP project as a replacement for SSF at the rate of 10% every three days for a period of 30 days of feeding. During the third phase of AquaFish Innovation Lab (2013-2015), the wild snakehead larvae aged 17 days old after hatching could be gradually weaned on formulated feed (using the protocol adopted from Hien and Bengtson 2009; 2015. These fish were grown from larvae to adult fish (as F_1 generation) at FARDeC by feeding them with pelleted feed developed by AquaFish CRSP project (Nen et al., 2015). F_1 mature adult snakehead will be developed into F_2, F_3 and F_4 generations to optimize the growth and survival rate while feeding on formulated or pelleted feed to ensure the efficient production and economic benefits to sustain this aquaculture industry in Cambodia without negatively affecting the wild SSF populations.

As snakehead culture in Vietnam becomes more domesticated, there is concern about the possibility of inbreeding in hatchery broodstocks. A method is needed to try to minimize inbreeding. We will conduct a breeding experiment at CTU between wild and domesticated snakehead collected in Vietnam. If the offspring from this back-cross perform as well in hatchery conditions as the same domesticated parent, back-crossing could be a good strategy to reduce potential inbreeding resulting from long-term domestication.

With the above initial results, the Ministry of Agriculture, Forestry and Fisheries announced the release of the ban on snakehead aquaculture in Cambodia in April 2016. At the same time, the Fisheries Administration formed a Technical Working Group to discuss an Action Plan and Technical Guidelines for sustainable development of snakehead aquaculture in Cambodia.

This research will provide technical support to Cambodia's Fisheries Administration for the formulation of an appropriate action plan and technical guidelines to adequately develop a sustainable snakehead aquaculture industry in Cambodia without doing harm to the wild fisheries on which so many people depend. In addition, this research will also contribute to the goal of the overall project, which is to enhance trade and investment for global fishery markets and improved nutrition and food security through sustainable aquaculture development in Cambodia, especially in the context of the recently released ban on snakehead aquaculture. There is a great potential for enhanced trade in snakehead fresh and processed forms and investment for snakehead in Cambodia after the ban released. Moreover, if the wild snakehead fisheries are sustainably managed after releasing the ban, there is also good potential to increase the trade and marketing of fresh and processed forms of wild snakehead with other Mekong riparian countries such as Thailand, Lao PDR and Vietnam, and other countries in Asia, Europe, America and Australia. As a result, household food security, nutrition, and income of tens of thousands of snakehead farmers who depend on snakehead aquaculture in Cambodia will be improved through sustainable aquaculture development and aquatic resources management.

The objective of the study will focus on optimization of the domestication and development of F_1 into F_2 snakehead in Cambodia as comparing to wild snakeheads collected from five different natural water bodies in Cambodia and domesticated-hatchery snakehead collected from Vietnam in regard to weaning and grow-out on formulated or pelleted feed. Furthermore, the assessment of economic efficiency during grow-out will also be conducted in a purpose to provide technical and policy recommendations for sustainable snakehead farming in Cambodia.

Quantified Anticipated Benefits

This research will provide information on domesticated breeding, weaning and grow-out of snakehead fish, especially development of Cambodia's snakehead aquaculture technologies, in order to support the lifting of the ban on snakehead culture in Cambodia. The following are quantifiable anticipated benefits:

- At least 20,000 farmers in Cambodia will benefit from this Investigation by restarting their snakehead culture leading to increased household income and improved snakehead fish market and trade.
- 250 scientists, researchers, government fisheries officers/managers and policy makers, extension workers, NGO staff, and private sector working on the issues of snakehead aquaculture in Cambodia as well as in other Mekong riparian countries will be better informed about research methods and findings, and have better recommended policies and strategies for sustainable snakehead aquaculture.
- Two (under) graduate students will be supported and trained by this investigation through their B.Sc./M.Sc. thesis research.
- At least 1,000,000 people who consume snakehead fish in Cambodia and other Mekong riparian countries will receive indirect benefits from this research.
- Benefits to the US include improved knowledge and technologies on domestication breeding, weaning and growth out of snakehead and this aquaculture is considered as a climate change adaptation measure. It will also allow Dr. Pomeroy to continue and expand his research program on economics and policy of fisheries and aquaculture in Southeast Asia.

Research Design and Activity Plan

Location

All weaning and grow-out trials will be conducted at the Freshwater Aquaculture Research and Development Center (FARDeC), Prey Veng province, Cambodia under the direct supervision of the Inland Fisheries Research and Development Institute (IFReDI), which has many broodstocks, breeding and weaning earthen ponds, hapa-nets and cement tanks, a small fish feed mill for fish pellet production and laboratories.

Methods

Null hypothesis: No differences among larvae are produced by the following snakehead broodstocks: current F1strain, Tonle Sap wild strain, Mekong River wild strain and Vietnamese domesticated striped snakehead.

(a) **Collection of brood snakehead fish:** In addition to available breeders developed at FARDeC, adult/mature wild snakehead (*C. striata*) will be collected from six different natural water bodies in six provinces of Cambodia (i.e. Kampong Chnang, Pursat, Siem Reap, Kampong Cham, Kandal and Prey Veng provinces) and conditioned at FARDeC hatchery, Cambodia in order to find the best strain for the highest growth rate or disease resistance. This program will contribute to the broodstock development in the near future and also preserve a genetic pool of hatchery broodstock native to Cambodia. It is doable and there is enough time to achieve this task because the first maturation age of the striped snakehead *Channa striata* is about one year. Frequent use of imported domesticated stocks of snakehead from Vietnam carry pathogens that could spread to the Cambodian wild snakehead and other fish stocks according to the study on snakehead disease and water quality conducted in the second phase of AquaFish project (2009-2011) (Pham Minh Duc et al., 2011). Domesticated snakehead (having already adapted to pellet feed) will be purchased from three different hatcheries in three provinces of the Mekong Delta in Viet Nam: Dong Thap, An Giang and Can Tho/Vinh Long provinces, quarantined to assure that pathogens are not being introduced,

and also conditioned at FARDeC for induced spawning to produce larvae to serve as the control for the experiment.

(b) **Induced spawning using HCG hormone:** All snakehead broodfish will be checked monthly for egg maturation based on the method of Nikolsky (1963). Broodfish with egg maturation at the same stage will be selected for induced spawning in order to simultaneously produce batches of larvae for the weaning experiment. HCG hormone will be used for induced spawning using the best dose obtained from So et al., (2011) in the final technical report during AquaFish CRSP phase 2.

(c) **Weaning experiment:** Comparison of survival and growth rate of larvae/fingerling from several broodstocks: current F1strain, Tonle Sap wild strain, Mekong River wild strain and Vietnamese domesticated striped snakehead. The larvae of all above brooders will be weaned at the same time using the same weaning protocol for *C. striata* adapted from Nen Phanna et al., (2015), So Nam et al., (2011) and Hien and Bengtson (2009; 2011) from live *Moinasp.* to formulated feed.

c.1) T he protocol: After yolk absorption at 3 days after hatching (dah), larvae will be fed with live *Moina* for 7 consecutive days till 10 day-old larvae (dah), and then larvae will be fed with a mixture of dead *Moina* and ground freshwater small-sized fish (replacing *Moina* by 20% per day) for 7 days more till 17 day-old larvae (dah). We will start weaning *Channa striata* larvae on formulated feed at 17day with replacement of trash fish by 10% per day until freshwater small-sized fish will be completely replaced by formulated feed. Formulated feed contains 40-50% crude protein.

c.2) Experimental set-up for weaning: Each of the four treatments will have five replicates. Larvae will be stocked in 50-L tanks with stocking density of 5 fish/L in recirculating water system. The fish will be fed to satiation by hand twice daily. Any remaining feed and feces will be siphoned out before feeding. Fish mortality, food consumption, and water quality (temperature, pH, and dissolved oxygen) will be recorded daily. Larvae will be sampled, weighed and measured at biweekly intervals. At the end of the experiment, final body weight (FBW, mg) and wet weight gain (WWG, mg) including survival rate will be determined. The experiment will last for 30 days.

c.3) Statistical analysis: Difference among the treatments on growth and survival rate and feed intake of snakehead larvae will be determined by one- way ANOVA. Tukey's HSD test is used to determine specific differences among means where appropriate.

(d) **Grow-out diet experiments:** Comparison of grow-out performance for the same strains used in the weaning experiments. Successfully weaned-larvae/fingerling in each treatment above will be moved to continue grow-out in hapas allocated in four earthen ponds ($300m^2$) to continue compare their growth performance and survival rate and to see their feeding adaptation on pelleted feed.

d.1) Experimental set-up for grow-out on pelleted feed: There are four treatments (F1 strain, wild Tonle Sap strain, wild Mekong River strain, and Vietnamese domesticated strains). Each treatment will have five replicated hapas (5x3x1.5m), and allocated separately in one individual earthen pond. A stocking density will correspond to the fish remaining in each treatment from the weaning experiment. The fish will be fed with commercial pelleted feed (45% CP) to satiation by hand twice daily at 09:00h and 16:00h. The amount of feed used and fish mortality will be recorded daily. Water quality parameters (Secchi disk transparency, temperature, pH, dissolved oxygen, NH_3, and NO^-_2) will be monitored weekly. Growth will be measured monthly by weighing 30 fish/hapa. The survival rate will be determined at the end of experiment. The experiment will last for 6 months.

d.2) Cost and benefit analysis will also be conducted to determine economic returns and efficiencies of *C. striata* cultured on pelleted feed by the following formulas:

-Total Revenue = Quantity * Price/unit

-Total Cost =Variable Cost + Fixed Cost

-Net return = Total Revenue – Total Cost
-Economic Efficiency = Total Revenue/Total Cost

The analysis will be based on farm-gate prices in Cambodia for harvested *C. striata* and current local market prices for all other items (e.g. cost of the pelleted feed; hapa net) and will be expressed in US dollars (US$).

d.3) Statistical analysis Difference among the treatments on growth and survival rate and feed intake of snakehead larvae including economic return will be determined by one-way ANOVA. Turkey's HSD test is used to determine specific differences among means once the ANOVA indicates that significant differences are present at p= 0.05 using SPSS 16.0.

(e) **Back-crossing experiment in Vietnam:** Wild broodtock collected from Camau conservation park and domesticated broodstock bought from a hatchery in Dong Thap or An Giang province, Vietnam will be held in the same hatchery conditions at CTU for at least one month before we induce reproduction through hormonal injections. Four crosses will then be carried out: (i) wild snakehead male and female (WxW); (ii) domesticated snakehead male and female (DxD); (iii) wild male and domesticated female (WxD); and (iv) domesticated male and wild female (DxW). Fry from these crosses will be separately reared in tanks (for 1-2 month) through weaning under the same conditions. Each cross will have three replicate tanks Fish will be reared using the standard CTU procedures, i.e., live feed until weaning to formulated feed in the hatchery water quality will be monitored for temperature, dissolved oxygen, ammonia and nitrite. Fish performance evaluation will include survival and growth rates in the hatchery Data will be analyzed using one-way analysis of variance followed by Tukey's HSD test where appropriate. The null hypothesis is that no differences exist in fish performance among offspring from the four crosses.

Trainings and Deliverables

Short-term training

On-station training on domestication breeding, weaning, grow-out and feed formulation technologies of the striped snakehead *Channa striata* obtained from first and second phases of AquaFish CRSP and third phase of AquaFish Innovation Lab will be conducted in the fourth quarter of 2017 at FARDeC for Cambodian fish farmers and government fisheries officers who are working at seven provinces where wild snakehead breeders and fin clips are being collected for the domestication and genetic diversity assessment by IFReDI.

Long-term training

One undergraduate (BSc) and one graduate (MSc) students will be partially supported for their thesis work under this investigation.

The deliverables of this investigation will include (1) a final technical report, including policy recommendations, and (2) a factsheet.

Schedule

The duration of implementation of this proposed investigation will be 24 months, starting from 1 March 2016 and ending 28 February 2018.

This investigation is planned to be implemented as below:

Activity	Beginning	Ending
Collection of wild or non-domesticated broodfish	March 2016	April 2016
Collection of domesticated broodfish	March 2016	April 2016
Experimental designs	April 2016	May 2016
Implementation of the experiments	June 2016	July 2017
Analysis of results and report preparation	August 2017	December 2017
Conduct of on-station training on domestication breeding, weaning, grow-out and feed formulation technologies of the striped snakehead *Channa striata*	October 2017	October 2017
Consultation and dissemination workshop on research findings	January 2018	February 2018
Finalization of final technical report	January 2018	February 2018