EFFECT OF STOCKING DENSITY OF VIETNAMESE KOI (*Anabas testudineus*) IN SEASONAL MINIPONDS

A Thesis

By

MD. IMAMUL MURSALIN

Examination Roll No. 1605549

Session: 2016-2017

Semester: July- December, 2017

MASTER OF SCIENCE (MS)

IN

AQUACULTURE

DEPARTMENT OF AQUACULTURE

HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY UNIVERSITY
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DECEMBER 2017

DECLARATION

I declare that this MS thesis entitled Effect of stocking density of Vietnamese Koi (*Anabas testudineus*) in seasonal miniponds, which I submit to Department of Aquaculture, was conducted by me for the degree of Masters in Aquaculture under the guidance and supervision of (Prof. Dr. Mst. Nahid Akter), Department of Aquaculture, Hajee Mohammad Danesh Science and Technology University, Dinajpur.

In addition, I took reasonable care to ensure that the work is original and has not been taken from other sources except where such work has been cited and acknowledged within the text.

The Author

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The Author

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ABBREVIATIONS

A.M.	Ante meridiem
ANOVA	Analysis of Variance
BAU	Bangladesh Agricultural University
BBS	Bangladesh Bureau of Statistics
BCR	Benefit Cost Ratio
BFRI	Bangladesh Fisheries Research Institute
CM	Centimeter
DEC	Decimal
DMRT	Duncan’s Multiple Range Test
DO	Dissolved Oxygen
DoF	Department of Fisheries
Dr	Doctor
et al.	et alia (and others)
etc.	et cetera
FCR	Food Conversion Ratio
FRSS	Fisheries Research Survey System
g	Gram
GDP	Gross Domestic Product
GIFT	Genetically Improved Farmed Tilapia
Ha	Hectare
HSP	Heavily Shaded Ponds
IFCAS	Integrated Floating Cage Aqua-geoponics System
IUCN	International Union of Conservation of Nature
Kg	Kilogram
L	Liter
M	Meter
mg	Milligram
ml	Milliliter
MoFL	Ministry of Fisheries and Livestock
MS	Master of Science

MSP	Moderately Shaded Ponds
MT	Metric Ton
NFP	National Fisheries Policy
NH_3	Ammonia
No	Number
P	Probability
P.M.	Post meridiem
ppm	Parts per million
Prof	Professor
R_1	Replication-1
R_2	Replication-2
R_3	Replication-3
SE	Standard Error
SGR	Specific Growth Rate
SIS	Small Indigenous Species
SPSS	Statistical Package for Social Science
T	Ton
T_1	Treatment-1
T_2	Treatment-2
T_3	Treatment-3
TSP	Triple Supper Phosphate
UFO	Upazila Fisheries Officer
W	Weight

ABSTRACT

The current study was conducted to evaluate the effect of stocking density of Vietnamese koi (*Anabas testudineus*) in farmer's seasonal mini ponds of Nilphamari and Rangpur district for a period of 120 days. There were three treatments each having three replications. Around 1.3 g weight of Vietnamese koi was stocked at the rate of 300, 400 and 500 individuals per decimal in T_1, T_2 and T_3 respectively. All the experimental fish were fed with a commercial fish feed containing 30% crude protein two times daily at a rate of 5-15% body weight. During the experimental period the water quality parameters were suitable for fish culture. The water temperatures were ranged from 29.33 to 31.81°C; transparency 24.37 to 28.70 cm, pH 7.40 to 7.96, dissolved oxygen 5.17 to 6.28 mg l^{-1} and total ammonia 0.13 to 0.28 mg l^{-1}. Higher temperature and NH_3 content were detected in the higher stocking density compared to those treatments where the stocking densities were lower. The weight gain (197.36±3.28 g) and specific growth rate (SGR 2.28±0.01% per day) were significantly increased ($P<0.05$) after 120 days feeding trial in the treatment T_1 compared with the other treatment groups. While, a significantly lowest ($P<0.05$) feed conversion ratio (FCR) was observed in the treatment T_1 followed by T_2 and T_3. Significantly highest ($P<0.05$) survival was observed in the treatment T_1 compared with the remaining two treatments. At the end of the experiment the benefit-cost ratio (BCR) result revealed that best BCR was 1.64±0.03 in T_1 which did not differed significantly ($P>0.05$) when compared with T_2. Consequently, the results of this study designated that the lowest stocking density (300 individuals dec^{-1}) seems to have a more progressive influence on the development of the growth performance and feed utilization of Vietnamese koi (*A. testudineus*) in the seasonal mini ponds.

KEY WORDS: Stocking density, growth, survival, *Anabas testudineus and* seasonal mini ponds.

CHAPTER 1

INTRODUCTION

Bangladesh is a riverine country. It contains numerous freshwater, brackish water and saline water body including haor, baor, beel, marsh, swamp, canal, pond, lake, river, estuary and sea. In inland fishery production Bangladesh is occupied fourth place just after China, India, and Myanmar and fifth place in closed waters (FRSS, 2016). Fisheries sector are indivisible from the life and lifecycle of the people of Bangladesh. It contributes 3.65% to the national GDP and almost one-fourth (23.81%) to the agricultural GDP and 2.01% of the total protein supply in the diet of the people of Bangladesh (DoF, 2016). About 1.5 million people are directly employed by this sector (DoF, 2016).

About 260 fish and 24 prawn species are comprehend to inhabit in the freshwaters of Bangladesh with 12 exotic fish species have been documented (IUCN, 2000, pp. 1–116; Rahman, 1989, pp. 1–352). There are approximately 40–50 small indigenous fish species which grow to a highest length of 25 cm (Felts *et al.* 1996, pp. 1–41). About 511 marine species together exist within Bangladeshi waters (Murshed-E-Jahan *et al.* 2014). Marine fisheries outgrowth is only 16.28% of the national fish production (FRSS, 2016).

Total area of inland water bodies is 4700795 ha where closed and open water resources comprises 794361 ha and 3906434 ha respectively (DoF, 2016). Fisheries are undoubtedly the most important company in the budget and about 16.5 million individuals are indirectly reliant on fisheries related actions for their living (Ministry of Finance, 2013; DoF, 2011 and 2013).
In the fiscal year 2014-15, foreign income from the fisheries sector is 1.92%. About 11% inhabitants of our country directly or indirectly depend for their employment on fisheries and aquaculture sectors (DoF, 2016). From 2000 and 2016, aquaculture production increased from

712,640 and 2,060,408 mt, a much larger quantity than wild capture production (1.023 million ton) in 2016 (Shamsuzzaman *et al.* 2017).

The highly productive aquatic biodiversity of the Bangladesh has been associated to the world's one of the biggest wetlands and three large river systems (Ganges, Brahmaputra and Jamuna) that rise from the Himalayan Mountains into the Bay of Bengal. Huge inland fisheries resources provide fish and other aquatic flora and fauna to the millions of living people in the Delta (Hossain, 2014).

Leading Rangpur district is a temper liable area. Water retaining capability of the pond is lessening day to day. As a consequence, the number of seasonal ponds (60%) is arising (BBS, 2000). About 55% ponds are seasonal in the northern districts of Bangladesh of which 60% contained water for 4-6 months (Rahman *et al*. 2013) while 40% absorbed for 6-9 months in a year and even more in some areas. Villagers normally, make use of these little water bodies for their domestic purposes while some are still banished due to their neglectful and vagrant mentality.

Nilphamari region is such an area where this culture strategy is the most wanted and adequate for all types and kinds of fish farmers for fish production and economic return. This analysis will be desired to give financial backup for marginal fish farmers and a source of quick return of money also. From the aquaculture point of view, those ponds have a high value for practicing fish culture including the species which have concise life cycle, hurried growth and require minimum inputs, such as Vietnamese koi (*A. testudineus*), Shing (*Heteropneustes fossilis*), genetically improved farmed tilapia (GIFT), Silver barb (*Barbodes gonionotus)*, Magur (*Clarias batrachus*) and sharputi (*Puntius sarana*).

To assure the maximum utilization of those ponds, above species were chosen for ensuring the highest production. The demands for those fishes are owing to their taste and medicinal values.

The belief is that there are beneficial and medicinal criteria in those fishes which are healthful for patients and convalescents by Mookerjee and Mazumder (1950). But absence of knowledge on proper culture methods and scarcity of good fish fry of suitable species at required time are found to be one of the chief constrains to propagate the culture methods of these species in Northern region of Bangladesh.

In the past we were relayed for fish utilization only upon the natural open water reserves but this dependency is out of control due to gradual fragmentation of open water bodies as well as booming our population density (Haque *et al.* 2017). Since the expansion of fish production from our beels, canals, lakes, river and estuaries are generally complicated; we need to relay on aquaculture especially on pond aquaculture for enhancing of our population demand, employment generation and poverty reduction of Bangladesh. Although the people of Bangladesh are increasing geometrically but our reserves are not increasing in the same manner (Haque *et al.* 2017).

The inspiring assumption is that the production of fish in pond may be maximized by culturing a mixture of species with different food and feeding habits. The combination of fish allows better utilization of sufficient natural food grown in a pond. It is the system where fast growing species with different feeding habit are cultured in the same pond (Jhingran, 1975).

The probability of increasing fish production per unit area, via culture, is considerable, when compared with monoculture systems of fish. Different species aggregations in Culture system effectively furnish also to improve the pond environment. Some of the fish species grown in carp ponds include paddlefish, tilapia and bighead carp. In considering pond Culture, certain aspects such as feeding, harvest and marketing should be emphasized first. Statistics have shown that there was no information on culture practice of Vietnamese koi (*A. testudineus*) in Bangladesh.

In Bangladesh aquaculture, there are approximately 92 species of exotic fishes are cultured (Bijukumar, 2000) and provides about 17% to global diet aquaculture production (Shelton and Rothbard, 2006). Koi fishery contributed about 2.8% of total pond catch since the 1980s (DoF, 1992) and in present years it reduces to 0.85% to the total pond catch (DoF, 1999).

In our country Koi is a small native indigenous species. It looks like greenish in appearance. Earlier the native koi was very much plentiful in almost all the freshwater body in our country (Mahmood, 2003). Now a day this species has been reducing day by day due to degradation of natural water body and its growth rate in the farmer's culture pond are not expectable as per need. For this reason Thai koi (*A. testudineus*) had been introduced in Bangladesh from Thailand in 2002 due to higher taste, growth, nutritious value and a high market price (Alam *et al*. 2006). But it has lost its higher growth rate capability features due to inbreeding during fry production period.

To overcome from this problem Sharnalata Agro Fisheries Limited has introduced Vietnamese Koi (*A. testudineus*) in 2010 which is very faster growth rate compare to Thai Koi(Ahmed *et al*., 2014). The color of Vietnamese Koi (*A. testudineus*) is whitish. This koi can be culture in pond as Thai Koi. Vietnamese koi eat commercial feed as Thai Koi. As Vietnamese Koi (*A. testudineus*) is a newly opened species in aquaculture of Bangladesh, no record yet been sufficient to us about its culture technique, stocking density, food and feeding behavior, breeding and disease occurrence. It can tolerate harsh environmental conditions such as wide range of temperature, low oxygen and other poor water conditions (Habib and Hasan, 1994).

It has some special faces such as quicker growth rate, shorter culture period (within 3-4 months grown to marketable size) and advanced survival rate (Kohinoor and Zaher, 2006; Jannat *et al.* 2012). It contains great amount of iron, copper and easily edible poly-unsaturated fats and many indispensable amino acids (Saha, 1971; Kohinoor *et al*. 1991).

Islam (2007) carried out an experiment on physico-chemical condition and occurrence of some zooplankton in a pond of Rajshahi University. In our country sustainable development of aquaculture is necessary in order to enhance the production. In these cases, Vietnamese koi can play a vital role to have more production considering with little capital investment but maximum economic returns.

The causes behind the greater extension of Vietnamese koi culture in Bangladesh are: (a) The fish can bear our atmosphere easily; (b) It is cultivable under high stocking density; (c) It is cultivable in shallow depth of water; (d) They can continue in minimum oxygen level; (e) It can be promoted at alive condition; (f) Crop can be achieved within 3-4 months; (g) Since it is beneficial and delicious so, it can be used as patient diet.

It is necessary to examine the growth potential of Vietnamese koi and to compare with the other koi fish accessible in Bangladesh. This technology is a good knowledge in aquaculture to meet up the protein shortage and socio-economic position of the common people of Bangladesh (Chakraborty *et al*. 2014).

As Vietnamese koi (*A. testudineus*) is a freshly introduced species in aquaculture of Bangladesh, no information yet been sufficient to us about its culture practice, stocking density, food habit and feeding habit, breeding and disease prevalence. Considering the above fact, the present trial was conducted to evaluate the effect of stocking density on growth performances of Vietnamese koi in seasonal miniponds with comparing the production at different stocking densities under the culture system and measuring the water quality parameters. Hence, this study has been pictured and proposed to make out these research findings at fishermen’s field as well as to corroborate the technologies.

OBJECTIVES OF THE STUDY

- To determine the effects of stocking density on the growth performances of Vietnamese Koi (*A. testudineus*) in farmers seasonal miniponds;
- To evaluate the water quality parameters of the cultured ponds and
- To assess the benefit cost ratio (BCR) of the cultured patterns.

CHAPTER 2

LITERATURE REVIEW

Before carrying out a research, it is crucial to acquire knowledge on the earlier related research workings. This part identifies the method used in the preceding studies, result and also made justification of conducting the current study. Nevertheless, the research works done in various zone of the world are reviewed beneath for clear and improved understanding of the current investigation. Research activity on culture practices of Vietnamese koi, GIFT tilapia, sharputi, in this area is insufficient. There are a few works regarding the culture of Vietnamese koi, BFRI super tilapia and silver barb in the northern area of Bangladesh and in overseas. Some of the workings done in the earlier period are highlighted now:

2.1 Stocking Density

Haque *et al*., (2017) conducted a thesis to determine the growth performance of Vietnamese koi (*A. testudineus*) in a commercial farm from 01 September, 2013 to 02 April, 2014 at Rahmatpur, in Sadar Upazilla in the district of Mymensingh, Bangladesh, having three treatments designed as treatments T_1, T_2 and T_3 respectively with stocking density of 976, 1000 and 1055 fry dec^{-1}. At last, they suggested that the growth performance of Vietnamese koi is higher in low stocking density. In future more study should be done to know the growth performance of Vietnamese koi.

Kohinoor *et al*., (2017) evaluated the production performances of Koi (*A. testudineus*), with Shing (*H. fossilis*) and GIFT Tilapia (*O. niloticus*) in semi-intensive culture management. They maintained the same stocking density in all treatments in case of Koi but Shing were stocked 37500, 32500 and 27500 fry ha^{-1} in treatments-I, II and III respectively and monosex GIF Tilapia was also stocked 5000, 10000 and 15000 fry ha^{-1} in treatments-I, II and III, respectively. The highest gross production was obtained in treatment-I and the lowest was noted in the treatment-II.

Ali *et al.*, (2016) conducted an experiment to assess the suitability of different fish species for cultivation in integrated floating cage aqua geoponics system (IFCAS) in Bangladesh. Three treatments, namely T_1 (tilapia), T_2 (Vietnamese perch) and T_3 (tilapia and Vietnamese perch = 1:1) in moderately shaded ponds (MSP) and 3 treatments, namely T_4 (tilapia), T_5 (Vietnamese perch) and T_6 (tilapia and Vietnamese perch = 1:1), in heavily shaded ponds (HSP) were used each with 3 replicates and fish were stocked at a rate of 56 m^{-2} cage. The individual growth rate and productivity of Vietnamese perch was higher in T_2 found by authors.

Kohinoor *et al.,* (2016) conducted a research to assess the growth and production performance of climbing perch Thai Koi and Vietnamese Koi Strain (*A. testudineus*) in Bangladesh. The author tested two treatments each with three duplications, containing three ponds under treatment-1 (T_1) were stocked with fry of Vietnamese koi of climbing perch, at the same time as other three ponds which selected as treatment-2 (T_2) were stocked with a Thai species of climbing perch and establish that the advanced growth performance and production were prominent in the case of Vietnamese strain in relation to the Thai strain of climb perch.

Uddin *et al.,* (2016) conducted an experiment to assess the culture potential of Thai climbing perch (*A. testudineus*) in experimental cages at different stocking densities in Kaptai Lake Bangladesh. They stocked Thai climbing perch in cages at 40 fish m^{-3}, 60 fish m^{-3} and 80 fish $m^{-3}$and 100 fish m^{-3} denoted as T_1, T_2, T_3 and T_4 respectively in triplicates. The results represented that T_2 (60 fish m^{-3}) could be suggested based on bulk gain and SGR of Thai climbing perch in cages of lake environment.

Ahmed *et al.,* (2015) carried out an experiment to evaluate the Impact of stocking density on growth and production performance of Vietnamese koi (*A. testudineus*) in semi-intensive culture system at Muktaghasa region of Mymensingh district. They used three different stocking densities as 150, 250 and 350 fry's dec^{-1} and designated as treatment T_1, T_2 and T_3

respectively each having two replicates. From their analysis they found that culture of Vietnamese koi at stocking density (150 fish dec^{-1}) showed higher benefit in short period of time and benefit cost ratio was 1.7, 1.63 and 1.56 in T_1, T_2 and T_3 respectively. After completing research they suggested that individual fish growth rate was increased with the decrease of stocking density.

Ali *et al*., (2015) conducted an experiment to evaluate the optimization of stocking density of Vietnamese climbing perch in cage at coastal region. The stocking density of different treatments T_1, T_2 and T_3 were 50, 75 and 100 fry m^{-3} respectively. The significantly ($p<0.05$) higher fish production was found in T_3 by the authors.

Habib *et al.,* (2015) carried out an experiment to assess the effects of stocking density on growth and production performance of cage reared climbing perch (*A. testudineus*) of high yielding Vietnamese stock. They started the trial in nine cages where they maintained the stocking densities of 100 fish m^{-3}, 150 fish m^{-3} and 200 fish m^{-3}which marked as T_1,T_2 and T_3 respectively having three replications for each. They found that the cage with 150 fishes (T_2) indicated auspicious growth, yield and survival rate which could be suggested to adopt. Nevertheless, more trials are recommended by them to elevate the stocking density and feeding system for better production performance and return.

Haque *et al.,* (2015) carried out a research to evaluate the production and growth performances of carps in different stocking densities of Culture . They established three treatments of different carps i.e. Rohu (*L. rohita*), catla (*G. catla*), mrigal (*C. cirrhosus*) and silver carp (*H. molitrix*) at a stocking densities were 40, 80 and 160 individuals dec^{-1}in T_1, T_2 and T_3 respectively and reported that the stocking density of 80 fingerling dec^{-1} (T_2) was the most suitable to ensure highest production in Culture .

Ahmed *et al.,* (2014) conducted a research to estimate the comparative growth study of Vietnamese koi (*A. testudineus*) and Thai koi (*A. testudineus*) for a period of 90 days. They choose four treatments having two duplication named for Vietnam koi treatments were VT_1 and VT_2 and for Thai koi were TT_1 and TT_2. The growth performances of the both species were higher at the lower stocking densities. Their research findings suggested that Vietnam koi has high growth potential in comparison to Thai koi under mini pond culture condition.

Chakraborty and Haque (2014) conducted an experiment to estimate the growth, yield and returns of koi, *A. testudineus* under semi-intensive aquaculture system using different seed types in Bangladesh. They took three treatments of same stocking density of fry of normal koi, hormone treated koi and Vietnam koi having three replicates each and termed as $T_{1,}$ T_2 and T_3 respectively. The highest growth performances were observed for the treatment of $T_{3.}$

Sarker *et al.* (2014) organized a trial to price out the economics of Thai Koi (Climbing Perch, *A. testudineus*) farming in pond. The authors found that the production of Thai koi for the sampled farms varied among the three feed products and this was the uppermost (17,761 kg ha^{-1}) for floating feeds followed by 15,972 and 15,695 kg ha^{-1} for mixed and sinking feeds groups respectively with an typical of 16,551 kg ha^{-1}.

Shoko *et al.* (2014) designated that the outcome of stocking density on growth, production and economic benefits of mixed sex Nile tilapia (*O. niloticus*) and African sharp tooth catfish (*C. gariepinus*) in Culture and monoculture. They examined the effect of stocking density on growth, yield and economic benefits of Nile tilapia (*O. niloticus*) in monoculture and Culture with African Sharp tooth catfish (*C. gariepinus*) and confirmed that farmers were achieved the maximum net yield and economic benefits by culturing (*O. niloticus*) and (*C. gariepinus*) in Culture .

Roy *et al*. (2013) manipulated a trial to evaluate the culture practice of Thai Koi (*A. testudineus*) with different stocking densities. The experiment was started with three conducts (T_1, T_2 and T_3) each having two duplications (R_1 and R_2). Three different stocking densities planned as treatments viz., 550 in T_1, 400 in T_2 and 350 fry dec^{-1} in T_3.They measured the growth performance by measuring the length (cm) and weight (g). Lower stocking density showed the best growth performance.

Rahman and Marimuthu (2010) evaluated a trial to appraise the effect of different stocking density on development, survival and production of local fish climbing perch (*A. testudineus*, Bloch) fingerlings in nursery ponds. The authors conducted the trial at three different stocking densities viz., 1.0 million ha^{-1}, 1.2 million ha^{-1} and 1.4 million ha^{-1} and the peak growth, survival and production of *A. testudineus* fingerling were noted at 1.0 millionha^{-1} seems to be the most excellent stocking density.

Adhikary *et al.* (2009) carried out an experiment on culture of Thai koi (*A. testudineus*) in earthen ponds with dissimilar feeding regimes to judge the effect of poultry droppings on growth performance in monoculture in the absence of fertilization. The research was carried out with two treatments T_1 (normal pond) and T_2 (integrated pond). Though growth performance of koi under T_1 was better but T_2 might be a probable technique of koi culture as it reduces the feeding charge.

Kohinoor *et al*. (2009) organized a research to assess the culture potentials of climbing perch, *A. testudineus* (Bloch) under different stocking densities at semi-intensive management. They tested three treatments of climbing perch with three replication at a stocking density of 50000, 56250 and 62250 fry ha^{-1} which designated as T_1, T_2 and T_3 respectively. They observed the highest production in T_1.

Gokcek and Akyurt (2007) studied an experiment of the effect of stocking density on yield, growth and feed efficiency of Himri barbel (*Barbus luteus*) nursed in cages of Turkey. The authors were shown four different stocking densities at 40, 60, 80 and 100 fish per cage in $1m^3$ and observed a highest mean weight was obtained in the lowest stocking density.

Rahman *et al.* (2005) carried out an experiment on the effect of stocking density on survival and growth of critically endangered masher, *T. putitora* (Hamilton), in nursery ponds. The authors were reported three different stocking density at 0.6 million ha^{-1} (T_1), 0.8 million ha^{-1} (T_2), 1.0 million ha^{-1} (T_3) and showed that 0.6 million ha^{-1}appears to be the most suitable stocking density for rearing of masher fingerlings in single- stage nursery system.

Azad *et al.* (2004) designated the Culture of carps, tilapia and pangas using low cost inputs. They evaluated three treatments in Culture using tilapia (*O. niloticus*), pangas *(P. hypophthalmus*), mrigal (*C. cirrhosis*) and silver carp (*H. molitrix*) at the stocking density was 100 fish dec^{-1} at different species compositions and reported that pangas showed the lowest growth performance and production in all treatments relate with other species.

Rahman *et al*. (2004) supervised an experiment on the effects of stocking densities on growth, survival and production of calbasu (*L. calbasu*) in secondary treatment, they confirmed that the survival of fingerlings were 91.1±1.29, 79.4±1.82 and 69.2±2.52 at 0.6, 0.8, 1.0 million fry ha^{-1} stocking densities correspondingly. The final average length of 8.1 ±0.01, 7.4 ±0.03 a and 6.3 ±0.01 cm and final average mass of 3.7 ±0.03, 3.1 ±0.02 and 2.3 ±0.02 g were monitored at stocking densities of 0.6, 0.8 and 1.0 million fry ha^{-1} correspondingly. After 8 weeks of nursing, the mean net production were found 2007 ±13.79, 1869.1 ±31.69 and 1418 ±71.43 kg ha^{-1} at 0 .6, 0.8 and 1.0 million fry ha^{-1} densities correspondingly.

Sayed (2002) carried out a trial on consequence of stocking bulk and feeding levels on growth and feed effectiveness of rohu (*L. rohita*) fry. The authors calculated that fish survival percentage, weight gain and specific growth rate (% SGR) were harmfully interrelated with stocking density. The most excellent performance was gained at 3 fry liter^{-1}. Nevertheless no significant dissimilarity in growth parameter was originated between 3 and 5 fry liter^{-1}. Body composition was not considerably affected by stocking density.

Haque and Razzaque (1998) carried out a Culture test maintaining the stocking density and ratio of rohu, catla, mrigal, common carp and Thai sharputi at the rate of 8000-10000 fish ha^{-1} in the ratio of 16:12:15:12:15:30 and reported that the yield found at 18-20 kg dec^{-1} in six months (4500-5000 kg of fish ha^{-1} in six months).

Kohinoor *et al.* (1998) conducted an experiment to evaluate the effect of mola (*A. mola*) on the growth and production of carps in Culture . The authors found the growth of rohu and mirror carp are severely affected when stocked with mola and suggested that mola may not be cultured with carps.

BFRI (1997) carried out a trial on the culture of koi (*A. testudineus*) at a stocking density of 20,000 ha^{-1} to found the production potential of koi in mud ponds. In the trial of T_1, the gross production was 425 kgha^{-1}in which rice bran 50%, mustard oil cake 30% and fish meal 20% were used as a complementary feed. On the other hand, marginal production value of 286 kg ha^{-1} was gained where rice bran 50% and mustard oil cake 50% were used as an additional feed (T_2). From that experiment it was found that the gross production of koi was considerably higher ($P<0.05$) in T_1 than of T_2.

Backiel and Le Cren (1992) estimated that stocking bulk has straight effects on growth and survival and therefore production of fish fry in nursery fishpond.

Middendrop and Hussain (1990) designed an experiment on Florida red tilapia (*O. aureus*)for a period of 84 days to assess the growth, survival and feed exchange rate. The fish were stocked at an early weight of 8.78 g and densities of 100, 200 and 300 fingerlings m^{-3}. Fish were fed with commercially prepared feed containing 28% and 32% protein. Final mean weight was higher for fish fed the diet with 28% protein (average 176.8 g) than those fed with 32% protein (mean 166.4 g). Individual growth rate was increased with decreasing stocking density. The mean daily gain was 1.94 g and SGR was 3.54% per day.

Rouse *et al.* (1987) stated the effects of stocking size and density of tilapia on *M. rosenbergii* in Culture . They tested two treatments which one was monoculture of prawn and another was Culture of prawn with tilapia and reported that survival was highest in Culture ponds.

Dev (2009) conducted a trial for 165 days to calculate the growth and production of fishes at different species combination in culture using *O. niloticus*, *L. rohita* and *H. molitrix*. Three stocking densities 268, 288 and 312 fish dec^{-1} were given in T_1, T_2 and T_3 respectively. Amongst the three treatments the maximum production was found in T_1 which were significantly higher than other treatments at lower stocking densities.

2.2 water quality parameter

Haque *et al*. (2017) conducted a thesis to determine the growth performance of Vietnamese koi (*A. testudineus*) in a commercial farm. The authors examined the water quality parameters in the experimental period, and found the average temperature 22.53±0.60, 21.24±.1.20 and 22.62±0.45°C in treatment T_1, T_2 and T_3 respectively. The average values of pH content of the water body in T_1, T_2 and T_3 were 7.45±0.36, 7.60±0.26 and 7.62±0.23, respectively. The average values of dissolved oxygen of the water in the treatment of T_1, T_2 and T_3 were 4.77±0.18, 4.77±0.18 and 4.79±0.14 mg l^{-1} respectively.

Kohinoor *et al.* (2017) manipulated an experiment to evaluate the growth performances of Koi (*A. testudineus*), with Shing (*H. fossilis*) and GIFT Tilapia (*O. niloticus*) in semi-intensive culture management. The authors observed the suitable range of water temperature from 26.06 to 31.97°C for fish culture. They found that transparency between 15-40 cm appropriate for fish culture. The observed pH values of water ranging from 7.1-8.9 indicated that the experimental ponds were suitable for fish culture. Most suitable range of DO in a water body for fish culture was suggested from 5.0-8.0 mg l^{-1}. They observed that the suitable range of ammonia-nitrogen in fish culture less than 0.1 mg l^{-1} and ammonia nitrogen content were higher that might be due to higher stocking density in all the treatments.

Uddin *et al.* (2016) supervised an experiment to assess the culture potential of Thai climbing perch (*A. testudineus*) in experimental cages at different stocking densities in Kaptai Lake Bangladesh. They found the following optimum water quality parameters: Water temperature (°C) 26.67±1.03 (23-29), DO (mg l^{-1}) 6.73±0.90 (5.3-7.3), Free CO_2 (mg l^{-1}) 2.50±1.4 (1.8-3.2), pH 7.42±0.29 (7.0-7.8), Total hardness (mg l^{-1}) 45.45±6.8 (34.2-51.3), Total alkalinity (mg l^{-1}) 48.60±8.3 (38.3-56.5), Ammonia (NH_3) Nill.

Habib *et al.* (2015) carried out an analysis to assess the effects of stocking density on growth and production performance of cage reared climbing perch (*A. testudineus*) of high yielding Vietnamese stock. The calculated optimum water quality parameters were as follows, mean water temperature was 29.49± 0.33℃, average dissolved oxygen of cage water was 5.86± 0.12 mg l^{-1}, average transparency was 38.66±0.79 cm, pH was 7.70±0.02 and ammomia- nitrogen was 0.14± 0.01 mg l^{-1}.

Ahmed *et al.* (2014) conducted a research to estimate the comparative growth study of Vietnam koi (*A. testudineus*) and Thai koi (*A. testudineus*). The authors examined the water quality parameter in the experimental period, and found the average temperature were 29.78 in VT_1, 30.42 in VT_2, 30.50 in TT_1 and 29.99°C in TT_2, dissolved oxygen were 7.21, 7.42 , 7.14 and 7.34 mg l^{-1} in VT_1, VT_2, TT_1 and TT_2 respectively, mean pH values were 7.42 in VT_1, 7.29 in VT_2, 7.50 in TT_1 and 7.45 in TT_2, ammonia were 0.21, 0.24, 0.28 and 0.23 mg l^{-1} in VT_1, VT_2,

TT_1 and TT_2 respectively and alkalinity were 200.00, 205.00, 210.00 and 205.00 ppm in VT_1, VT_2, TT_1 and TT_2 respectively.

Chakraborty *et al.* (2014) supervised an experiment to estimate the growth, yield and returns to koi, *A. testudineus* (Bloch, 1792) under semi-intensive aquaculture system using different seed types in Bangladesh. Authors figured out that water quality parameters in all the experimental ponds were within the normal range for fish culture for water recycling method.

Rahman and Monir (2013) calculated the physico-chemical conditions of nine nursery ponds located at the Freshwater sub-station, Bangladesh Fisheries Research Institute, Saidpur, and Nilphamari. The researchers logged the average values of different water quality parameters were, water temperature 29 to 33°C, transparency 31 to 58 cm, dissolved oxygen 3.4 to 6.2 mg l^{-1}, pH 6.8 to 8.5, alkalinity 95 to 185 mg l^{-1} and NH_4-N 0 to 1 mg l^{-1}.

Roy *et al.* (2013) manipulated a trial to evaluate the culture practice of Thai Koi (*A. testudineus*) with different stocking densities. Authors figured out those water quality parameters during the trial period which was measured fortnightly. The typical temperature was found 28.21±0.870, 28.78±0.510 and 28.76±0.78^{0}C in conducts T_1, T_2 and T_3respectively. They also found the pH ranges were diverse between 7.70 to 9.10 among the three treatments. The average values of dissolved oxygen were 5.33±0.34, 4.23±0.13, and 4.16±0.14 mg l^{-1} respectively in the three successive treatments.

Mondal *et al.* (2010) paralleled the aquaculture of Thai climbing perch between cage and pond under three management systems in Bangladesh. In this experiment, water temperature ranged from 15.3°C to 28.5°C mean water transparency in Caged Tilapia ponds was lower (25.98cm). The measured pH in the present study in different treatments was slightly alkaline (7.0mgl^{-1} to 8.9mgl^{-1}).

Adhikary *et al.* (2009) carried out an experiment to justify the growth Performance of Thai Koi (*A. testudineus*) integrated culture system. They found the following water quality parameters: water temperature of the integrated pond was 27.85±1.21 (°C), and normal pond 28.45±1.32°C, Dissolved oxygen was found to be within 5.5 to 8.5 mg l^{-1}, pH values were almost neutral (6.8-7.5). Alkalinity of water was found 81 to 127 mg l^{-1} and total alkalinity was measured in T_1 (107.25±16.58 mg $CaCO_3l^{-1}$).

Rahman *et al.* (2009) calculated some physico-chemical property of water in nine mud nursery ponds containing critically endanger Reba Carp (*C. ariza*) at the Freshwater Station, Bangladesh Fisheries Research Institute, Mymensingh, Bangladesh. The researchers measured the ranges of physico-chemical parameters as temperature 29.10 to 32.40°C, pH 7.20 to 8.10, dissolved oxygen 3.10 to 6.00 mg l^{-1}, transparency 30.50 to 58.50 cm, total alkalinity 65.50 to 171.50 mg l^{-1} and Ammonia-nitrogen 0.01 to 1.00 mg l^{-1}.

Ali *et al.* (2008) supervised a test at the south-east corner of the Fisheries Faculty Building, Bangladesh Agricultural University, Mymensingh. He originated that the temperature varied from 27.53 to 28.46°C, pH from 7.86 to 8.49; dissolved oxygen from 5.16 to 5.63 mg l^{-1} and transparency from 30.65 to 35.71 cm.

Mukhopadhyay *et al.* (2003) affirmed that the freshwater prawn *M. rosenbergii* was the most significant cultural freshwater species of prawn with a quite high growth rate and broad range of temperature (15 – 35)°C tolerance.

Chowdhury (2000) studied the comparative abundance of benthos in fertilized and unfertilized ponds and establish that the different groups of benthic organisms were maximum in fertilized ponds and minimum in unfertilized ponds. The ideals of water quality parameters such as temperature, transparency and pH and dissolved oxygen were within the productivity range during the study age.

Haque *et al.* (2000) established that the water temperature diverse from 29 to 32°C, transparency from 30 to 56 cm, dissolved oxygen from 3.5 to 7.8 mg l^{-1}, pH from 6.8 to 7.8, nitrate nitrogen from 1.64 to 2.31 mg l^{-1} and the phosphate-phosphorus from 0.31 to 1.07 mg l^{-1} during the examination of water quality parameter in six pond which were located at the southeast place of the Fisheries Faculty Building, Bangladesh Agricultural University, Mymensingh.

Hossain (2000) calculated the effect of organic and inorganic fertilizers on water quality parameter of fish pond. He finished that the water quality parameter assorted considerably among the treatments and the temperature varied with a range of 22-33°C, transparency 26.52 cm, dissolved oxygen 3.8-6.9 mg l^{-1} and pH 6.8-8.2 total alkalinity 40- 80 mg l^{-1} amongst three treatments.

Kohinoor (2000) calculated a number of water quality parameters in the field laboratory of Bangladesh Agricultural University, Mymensingh and logged water temperature ranged from 18.5 to 32.9°C, transparency ranged from 15 to 58 cm, dissolved oxygen ranged from 20 to 7.4 mg l^{-1} and pH ranged from 6.5 to 8.0.

Kohinoor (2000) calculated some physico-chemical parameter in carp Culture with small indigenous fish species. He logged that the range of water temperature was 24.2 to 33.3°C, transparency 12-15 cm, DO 2.0-7.5 mg l^{-1} and pH was 6.9-8.6 among the treatments.

Sarker (2000) accompanied a test in six ponds on the effect of periphyton to GIFT Tilapia monoculture located at the south-east corner of the Fisheries Faculty Building, Bangladesh Agricultural University, Mymensingh. He established that the temperature varied from 19.8 to 22.82°C, pH from 6.8 to 8.3, dissolved oxygen from 7.5 to 8.84 mg l^{-1}, nitrate-nitrogen from 0.7 to 4.2 mg l^{-1}, and phosphate-phosphorus from 0.49 to 4.07 mg l^{-1}.

Kohinoor *et al.* (1998) monitored that the water quality parameters in six research ponds. The authors declared the average values of water quality parameters such as, water temperature (°C), transparency (cm), pH, dissolved oxygen (mg l^{-1}), and ammonia nitrogen (mg l^{-1}) were27.72±0.01, 32.58±2.401, 7.18±0.06, 4.20±0.12, and 0.12±0.02 respectively.

Wahab *et al.* (1995) calculated a large number of water quality parameters in nine fishponds of Bangladesh Agricultural University campus, Mymensingh. They noticed that the water temperature diverse from 27.2 to 32.4°C, transparency from 26 to 50 cm, pH was all the time around 6.0, dissolved oxygen from 2.2 to 7.1 mg l^{-1}, total hardness from 45 to 104 mg l^{-1} and total ammonia from 0.0 to 0.43 mg l^{-1}.

Wahab *et al*. (1995) calculated a number of water quality parameters in nine trial ponds at Bangladesh Agricultural University campus. They logged that the water temperature vary between 22 to 32.4°C, Transparency 50 cm, pH was all times at around 6.0 and DO was always low and assorted between 2.2 to 7.5 mg l^{-1}.

Akiyama *et al*. (1991) calculated that temperatures below 14°C or above 35°C are generally fatal29 - 3l°C being the optimal.

2.3 Growth performance

Haque *et al*. (2017) conducted a thesis to determine the growth performance of Vietnamese koi (*A. testudineus*) in a commercial farm. The authors examined the growth performance of Vietnamese koi and found the mean length 13.7±0.03 cm, 13.64±0.02 cm and 13.58±0.01 cm in T_1, T_2 and T_3 respectively and mean weight gain 90.0±2.0 gm, 88.0± 1.0 g and 86.02±1.0 g for T_1, T_2 and T_3 respectively. The production was found to be 83.57 kg dec^{-1} in T_1 followed by T_2 (83.60 kg dec^{-1}) and T_3 (84.01 kgdec^{-1}) respectively.

Habib *et al.* (2015) carried out an analysis to assess the effects of stocking density on growth and production performance of cage reared climbing perch (*A. testudineus*) of high yielding

Vietnamese stock. They found SGR and FCR that were best for fish in T_2.The highest harvesting weight and survival rate was observed in T_2.

Ahmed *et al.* (2014) conducted a research to estimate the comparative growth study of Vietnam koi (*A. testudineus*) and Thai koi (*A. testudineus*). The authors examined the growth performance and found the best weight gain of 80.00 g was observed in VT_1, mean weight gain (g) were 0.83, 0.88, 0.51 and 0.61; SGR (% day^{-1}) were 3.93, 3.95, 3.65 and 3.71%; FCR were 1.50, 1.67, 1.70 and 1.90; survival rate were 78.50, 76.50, 90.00 and 82.00% and fish production were 7,839; 15,923; 5,519 and 11,820 kg ha^{-1} for 3 months in VT_1, VT_2, TT_1 and TT_2 respectively.

Sarker *et al.* (2014) organized a trial to price out the economics of Thai Koi (Climbing Perch, *A. testudineus*) farming in ponds in Bangladesh. The authors found that the weight per day gain of fish was slower for sinking or mixed feed than the floating feed users and the best FCR value was also gained for the floating feed. The study also exposed that the usual production charge of producing kg^{-1} Thai koi was lower for floating feed compared to other feed technologies.

Adhikary *et al*. (2009) carried out an analysis to assess the growth performance of Thai Koi (*A. testudineus*) in integrated culture system. They found the following growth performance: T_1 (59.73±4.22 g) showed weight gain followed by T_2 (48.22±10.74 g), Mean length gain for T_1 was (12.75±1.50 cm) and for T_2 (12.22±1.73 cm) and SGR (% day^{-1}) in T_1 (3.51) and T_2 (3.31).

CHAPTER 3

MATERIALS AND METHODS

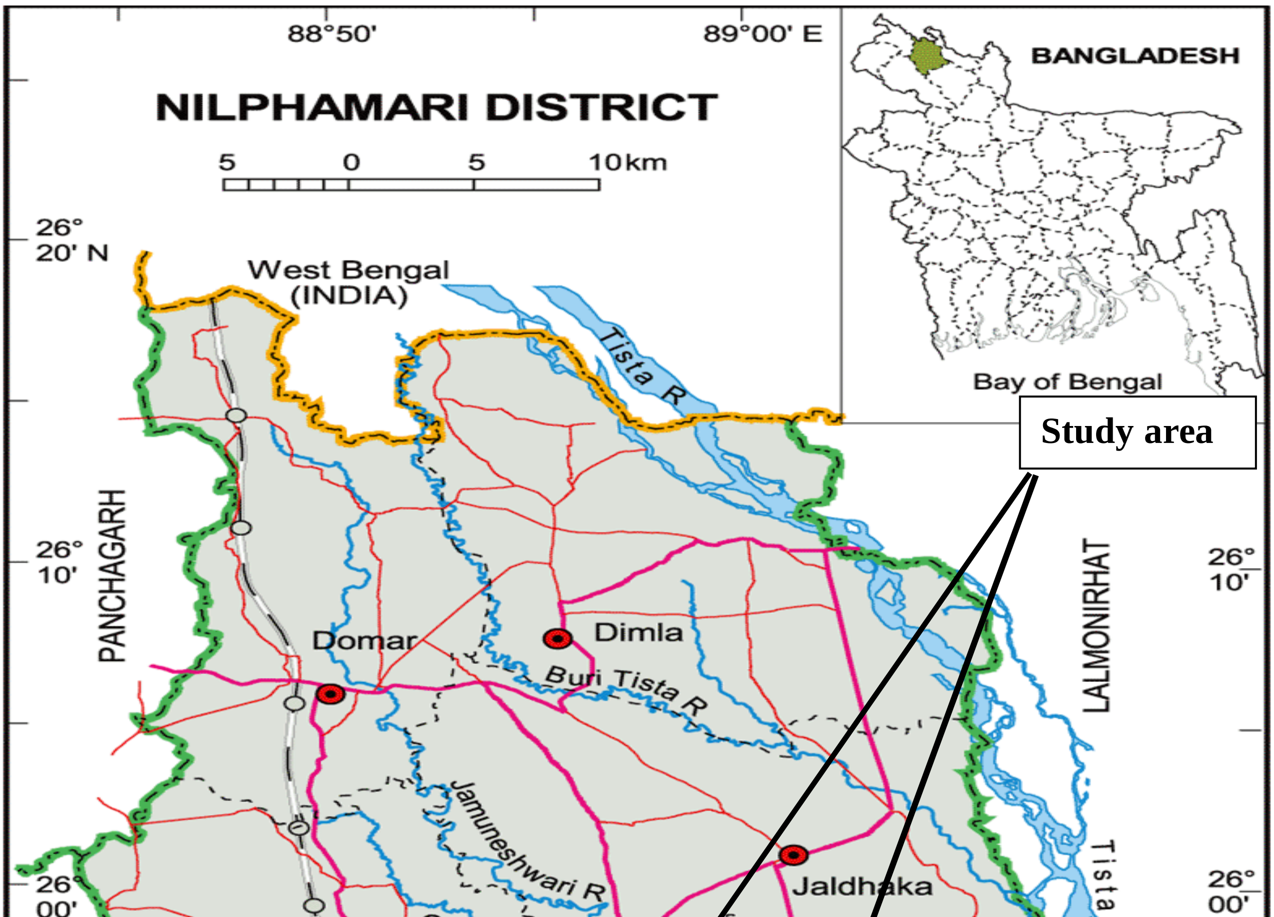
3.1 Introduction

This research was planned to achieve the objectives of this present study. For this trial, study was scheduled to determine the effect of stocking density on growth, survival and production of Vietnamese koi in seasonal miniponds. All work plans were listed according to the activity chart of Bangladesh Fisheries Research Institute, Freshwater Sub-station, Saidpur, Nilphamari.

Two important things such as farmers training and field day were conducted by authority of the Bangladesh Fisheries Research Institute, Freshwater Sub-station, Saidpur, Nilphamari.

3.2 Depiction of the study area

This research was conducted in Saidpur and Kishoreganj upazila of Nilphamari district and Taraganj Upazila of Rangpur district. The selection of ponds was done with the help of respective Senior Scientific Officer (SSO) and Scientific Officer (SO), Bangladesh Fisheries Research Institute, Freshwater Sub-station, Saidpur, Nilphamari and relevant Upazila Fisheries Officer (UFO).



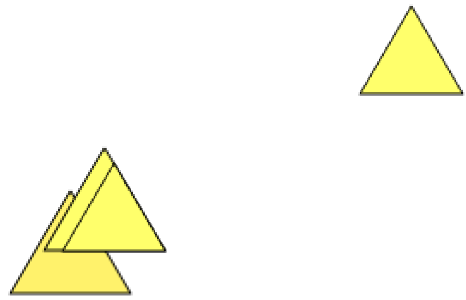


Figure 3.1 Map presenting the study zone of Saidpur and Kishoreganj upazila of Nilphamari district.

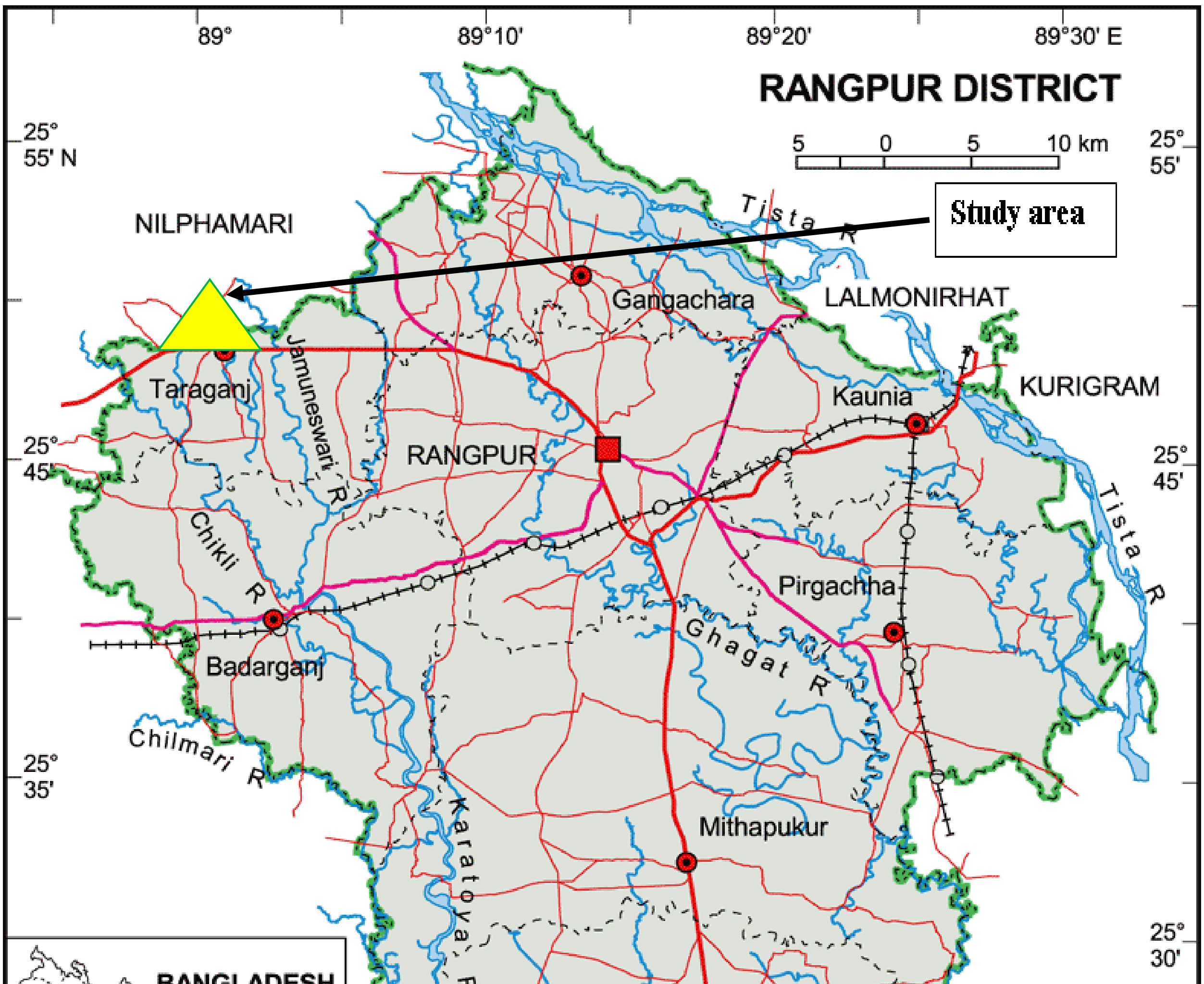


Figure 3.2 Map presenting the study zone of Taraganj Upazila of Rangpur district.

3.3 Time duration for the study

The research was conducted in farmers seasonal mini ponds cited in the Northern area of Bangladesh including Nilphamari and Rangpur district for a period of 120 days from 01 May to 30 August, 2017.

3.4 Depiction of the Trial units

Nine seasonal earthen fishponds were selected for the trial purposes at Saidpur and Kishoreganj upazila of Nilphamari district and Taraganj Upazila of Rangpur district to detect the growing performance of Vietnamese koi. The fishponds were rectangular in size and about 10 decimal in magnitude in which mean water depth of ponds existed 4 feet. Ponds were identical in basin configuration, bottom types and contour. For the appropriate supervision and protection from thieving, ponds were selected adjacent to the farmer household. The ponds were fully exposed to prevailing sunlight. Leading source of water was the rainwater, but occasionally water was provided from a deep tube-well via a flexible plastic pipeline at ever needed. The bank of the pond was well sheltered and concealed with grassland.

3.5 Experimental design

The research was started with three treatments (T_1, T_2 and T_3) each having three repetitions (R_1, R_2 and R_3). Three diverse stocking densities of Vietnamese koi fingerling such as 300, 400 and 500 were released $dec^{-1}$of pond water and intended as T_1, T_2 and T_3 respectively while the other species persist in fixed densities mentioned in Table 3.1.

The design of Culture patterns are showing in Table 3.1.

Table 3.1 Culture of Vietnamese koi under diverse stocking densities in the field level supervision

Treatments	Replication	Species	Stock. density (individual dec^{-1})
T_1	R_1 R_2 R_3	Vietnamese koi+ GIFT+Sharputi	300+10+5
T_2	R_1 R_2 R_3	Vietnamese koi+ GIFT+Sharputi	400+10+5
T_3	R_1 R_2 R_3	Vietnamese koi+ GIFT+Sharputi	500+10+5

3.6 Pond preparation

3.6.1 Control of unwanted species and aquatic weeds

Pond preparation is a pre-requisite for successful fish culture. Before starting the experiment all fishponds were reconditioned and aquatic floras were aloof physically. Ponds were drained to remove all fishes, later the dried ponds were left visible to sunlight for several days and then pond bottom were treated with bleaching powder at the level of 20 g dec^{-1} and left for five days. The unwanted plant of bank was removed manually and pond banks were fixed by using the burrowed bottom mud's.

Undesirable fishes and other aquatic creatures were removed by the use of rotenone at 25-35g $dec^{-1}ft^{-1}$. After that, the research ponds were well enclosed by nylon net with the support of bamboo sticks (Plate 3.1).

Plate 3.1 Preparation of the experimental pond

3.6.2 Liming

Lime ($CaCO_3$) was used at a degree of 1 kg dec^{-1} after one week of rotenone application. Lime was melted into an earthen container and then used by scattering evenly in the fishponds.

3.6.3 Fertilization

After 7 days of liming, ponds were nourished using cow- dung, urea and TSP each at a rate of 5-6 kg dec^{-1}, 100g dec^{-1} and 75 g dec^{-1} respectively. TSP was scattered all over the fishponds next 9 to 11 hours of liquefying in flexible buckets.

3.7 Fry collection and stocking

The fry of Vietnamese koi (*A. testudineus*) was selected for the trial purpose. The Vietnamese koi fry of an average weight of 1.3g and length 3.8 g were brought from authorized commercial fish hatchery which have a friendly relationship with BFRI sub- station, Saidpur, Nilphamari and carried to the BFRI sub-station within oxygenated plastic bags occupied with freshwater. Before the beginning of the study, the trial fish were adjusted for 7 days in the pond of BFRI sub-station, Saidpur. Then, fish were stocked at the densities of 300, 400 and 500 fry $dec^{-1}$of pond which represented as T_1, T_2 and T_3 respectively. For GIFT and Sharputi, stocking density was maintained at 10 and 5 per decimal of pond water respectively (Plate 3.2).

Plate 3.2 Stocking of fish in the experimental pond.

3.8 Feed selection and feeding strategy

Feeding frequency and rate was continued according to the body weight of the trial fish. All the fish were nourished with a marketable feed comprising 30% crude protein (commercial available fish feed) two times a day with two equal split of the ration at a rate of 5-15 % body weight. The feed was sprinkled uniformly all over the pond’s surface in morning and evening (Plate 3.3).

Plate 3.3 Feeding in the experimental pond

3.9 Observing and data collection

The preliminary data (length and weight) were recorded before releasing fish in to the ponds. Regular observation of the stocked fish was carried out through feed supply. The persistence of fish were recorded throughout grow out period. Sampling was done for the estimation of feeding rate in every 15 days intervals by a net. For observing the development and to regulate the amount of feedstuff to be given, the bulk mass of around 10 experimented fish were taken from each pond. The weight and length of Vietnamese koi were calculated by random sampling using a digital electronic balance (TANITA KD160) and meter scale respectively. During the trial period, all the fishponds were visited frequently for checking the pond conditions. The experimented fish were handled very carefully in order to lessen the handling stress (Plate 3.4).

Plate 3.4 Measuring weight (g) of Vietnamese koi during the study period.

3.10 Water quality assessment

Environmental factors exert a massive effect on the maintenance of a healthy aquatic situation and production of natural food organisms. Physico-chemical and biological environment of a water body typically influenced by water temperature which uniquely of the most important physical factors.

3.10.1 Procedure of the study

Transportable digital equipment's and kits were used for recording data on different water quality parameters. In the current trial, water sample were collected from each ponds. Recording on the spot data and collection of samples were made between 10.30 to 11.30 A.M. Water temperatures, transparency, pH, dissolved oxygen (DO), total alkalinity, and total hardness were recorded every 15 days interval between 9.00 am to 11.30 am.

3.10.1.1 Temperature

Generally relates to air temperature, but decreases with depth. It impacts on biological life of fish and water chemistry. Increase in temperature beyond 30°C increases the activity level and fish metabolism. Temperature (°C) of water was taken from each pond by using a digital thermometer (Digi-thermo WT-2, China) (Plate 3.5).

Plate 3.5 Measurement of water temperature (°C) during the trial period

3.10.1.2 Transparency

Transparency is a measure of the turbidity of water in the pond. The color of the water gives an indication of what sort of turbidity it is. Transparency (cm) of water was recorded from each pond by using a Secchi disc (Plate 3.6).

Plate 3.6 Measurement of transparency (cm) during the experimental period

3.10.1.3 pH

pH –"power of hydrogen". pH determines solubility and biological availability of chemicals in water. It is a measure of acidity or alkalinity in pond water. pH of water was taken from each pond by a digital pH meter (Elico-Li-120) (Plate 3.7).

Plate 3.7 Measurement of pH during the experimental period

3.10.1.4 Dissolved Oxygen

Amount of oxygen in a liter of water is relative to total amount of oxygen water could potentially hold at that temperature. The solubility of oxygen in water decreases as the water temperature increases. Typically, dissolved oxygen is measured either in mg. per liter (mg I^{-1}) or parts per million (ppm). The dissolved oxygen was measured by using digital DO meter (YSL, Model 58, and USA) (Plate 3.8).

Plate 3.8 Measurement of dissolved oxygen during the experimental period

3.10.1.5 Ammonia

Ammonia is the second gas of importance in fish culture; its significance to good fish production is overwhelming. Additionally, ammonia is converted from toxic ammonia (NH_3) to nontoxic ammonium ion (NH_4+) at pH below 8.0.The ammonia content of water was calculated by using an ammonia test kit for freshwater (Hanna, Romania) (Appendix A).

3.11 Harvesting

Fishes were entirely harvested after 120 days of culture period. Harvesting of fishes was completed by first repeated netting, using a seine and then dewatering of the ponds. During harvesting, the weights of all fish and number of fish from each pond were recorded (Plate 3.9).

Plate 3.9 Harvesting of fish at the end of the experiment.

3.12 Growth performance and production of fish

The growth performances and production of fish were calculated using the following formulas:

(i) Growth

a) Weight gain (g)

Weight gain (g) = Final weight (g) –Initial weight (g)

b) % Weight gain

% weight gain was estimated by the following formula:

$$\% \text{ Weight gain} = \frac{\text{Final weight (g)} - \text{Initial weight (g)}}{\text{Initial weight (g)}} \times 100$$

c) Average daily gain (ADG day^{-1})

$$\text{Average daily gain (day}^{-1}\text{)} = \frac{\text{Mean final weight} - \text{mean initial weight}}{\text{Days}}$$

d) Specific growth rate (SGR % day^{-1})

The SGR is the momentary alteration in weight of fish calculated as the percentagewise in body weight per day over a given time interval and is written as:

$$\text{SGR (\% day}^{-1}\text{)} = \frac{\text{In } W_2 - \text{In } W_1}{T_2 - T_1} \times 100$$

Where,

W_1 = the initial live body weight (g) at time T_1 (day).

W_2 = the final live body weight (g) at time T_2 (day).

e) Survival rate (%)

Survival rate was estimated by the following formula:

$$\text{Survival rate (\%)} = \frac{\text{Final number of fish}}{\text{Initial number of fish}} \times 100$$

f) Feed conversion ratio (FCR)

Feed conversion ratio is defined as the amount of dry feed intake per unit live weight gain. It is calculated as:

$$FCR = \frac{\text{Total dry feed intake (g)}}{\text{Total wet weight gain (g)}}$$

To measure FCR, the dry weight of the feedstuff was attained by using a correction for the investigated moisture content of the diet. FCR is a measure of the degree of gross use of feed for growth.

3.13 Economic analysis

Gross margin (BDT treatment -1) = Gross return – Gross variable costs

Benefit cost ratio was estimated by the following formula:

$$BCR = \frac{\text{Gross return}}{\text{Gross variable costs (BDT)}}$$

3.14 Data analysis

Entirely data were confirmed using one-way Analysis of Variance (ANOVA). ANOVA was executed on all the dependent variables to see whether the treatment had any significant effect or not. Significant outcomes (*P<0.05*) were further verified using one-way ANOVA followed by Duncan's Multiple Range Test (DMRT) to identify significant variances between means. The data are stated as average ± SE and statistical analysis was done by using SPSS version 22 and Microsoft Office Excel for Window.

CHAPTER 4

RESULTS

4.1 Water quality Parameters

The water quality parameters in the case of culture system of Vietnamese koi with tilapia and sharputi of all treatments are shown in Table 4.1. Mostly, there were no significant variation in case of temperature, transparency, pH and dissolved oxygen but significant variation was detected in case of ammonia content.

The results showed that the highest temperature of 30.80±0.33°C was recorded for treatments T_3 while the lower temperature of 30.64±0.50°C and 30.74±0.48°C were observed in treatments T_1 and T_2 respectively. However, there was no significant ($P>0.05$) difference in the water temperature recorded among the treatments. Similarly a non-significant ($P>0.05$) highest transparency was observed in the treatments T_2 (27.29±0.60 cm) compared to T_1 (26.54±0.98 cm) and T_3 (26.33±0.54 cm).

In the case of pH, a non-significantly higher ($P>0.05$) amount of water pH was observed in T_2 (7.77±0.08) than T_1 (7.63±0.10) and T_3 (7.73±0.87). In the same way, a non-significantly higher ($P>0.05$) content of dissolved oxygen (mg l^{-1}) was detected in the treatment of T_1compared to those treatments that were stocked at higher stocking densities. The average values of dissolved oxygen content in experimental ponds were found as 5.97±0.14, 5.65±0.11 and 5.55±0.17 mg l^{-1} in T_1, T_2 and T_3 respectively.

Significantly the lowest ($P<0.05$) ammonia content was noted in the treatment of T_1 when compared with remaining two treatments. In the whole trials, ammonia content (unionized) was recorded in three different treatments T_1, T_2 and T_3 as 0.15±0.01, 0.24±0.02 and 0.25±0.01 mg l^{-1} respectively (Table 4.1). The value of ammonia in T_2 (0.24±0.02 mg l^{-1}) and T_3 (0.25±0.01 mg l^{-1}) indicated higher quantity compared to T_1 but T_2 and T_3 did not vary significantly ($P>0.05$).

Table 4.1 Water quality parameters examined in different treatments of Vietnamese Koi (*A. testudineus*) culture of the research over 120 day's culture periods in fishponds

Parameters	Treatments		
	T_1	T_2	T_3
Temperature (˚C)	30.64±0.50	30.74±0.48	30.80±0.33
	(29.52-31.75)	(29.67-31.81)	(29.33-30.81)
Transparency (cm)	26.54±0.98	27.29±0.60	26.33±0.54
	(24.37-28.70)	(25.96-28.61)	(25.53-27.92)
Water pH	7.63±0.10	7.77±0.08	7.73±0.87
	(7.40-7.86)	(7.58-7.96)	(7.54-7.92)
DO (mg l^{-1})	5.97±0.14	5.65±0.11	5.55±0.17
	(5.66-6.28)	(5.40-5.91)	(5.17-5.94)
Ammonia(mg l^{-1})	$0.15±0.01^{a}$	$0.24±0.02^{b}$	$0.25±0.01^{b}$
	(0.13-0.17)	(0.20-0.28)	(0.23-0.28)

Data presented as mean ± SE. Data with different superscripts in the same row indicate significant differences ($P < 0.05$).

4.2 Growth performance

The growth performances, feed utilization and survival of the culture of Vietnamese koi (*A. testudineus*) with tilapia and sharputi under different stocking densities were recorded (after 120 days rearing) are given in table 4.2. Growth of Vietnamese koi in seasonal mini ponds was investigated and the results shown that the growth rates were varied according to their stocking densities.

Though, there is no significant change in initial length and initial weight of fishes in different treatments but the final weight of *A. testudineus* were varied significantly ($P<0.05$). Final weight of *A. testudineus* was gained as 198.66±3.28, 172.33±6.74 and 167.33±3.71 g in T_1, T_2 and T_3 respectively (Table 4.2). Final weight was attained highest in T_1 where the stocking density was lowest and final weight was lowest in T_3 where the stocking density was highest. Final weight of *A. testudineus* in T_1 (198.66±3.28 g) was varied significantly ($P<0.05$) compared with the remaining two treatments T_2 (172.33±6.74 g) and T_3 (167.33±3.71 g) while T_3 was not differed significantly ($P>0.05$) from $T_{2.}$

In the present research, final length of *A. testudineus* was obtained in T_1, T_2 and T_3 as 19.96±0.17, 19.63±0.38 and 18.90±0.20 cm respectively (Table 4.2). Therefore, the results shown that the higher final length was noted in the lowest stocking density. Final length in T_1 was not differed significantly ($P>0.05$) from T_2 but significantly ($P<0.05$) differed from T_3.

Weight gain of *A. testudineus* in three treatments was found as 197.36±3.28, 171.01±6.73 and 165.97±3.71 g in T_1, T_2 and T_3 respectively (Table 4.2). Weight gain of *A. testudineus* in T_1 was varied significantly ($P<0.05$) compared to the remaining two treatments T_2 and T_3while T_3 was not differed significantly ($P>0.05$) from T_2.

Similar results were found in the case of percent weight gain of *A. testudineus* and this was recorded as (15184.82±319.22), (12953.39±474.08) and (12421.06±316.68) in T_1, $T_2$and T_3respectively (Table 4.2). The % weight gain of *A. testudineus* in T_1 was varied significantly ($P<0.05$) compared to the remaining two treatments T_2 and T_3 while T_3 was not differed significantly ($P>0.05$) from T_2. The % weight gain of *A. testudineus* in T_1 (15184.82±319.22) was highest where the lowest stocking density take place.

The value of average daily gain (ADG) was found as 1.65±0.03, 1.42±0.06 and 1.38±0.03 in T_1, T_2 and T_3 respectively (Table2). During the investigation, the highest value of ADG was obtained in T_1 (1.65) and the lowest was observed in T_3 (1.38). Significantly ($P<0.05$) higher ADG was recorded in T_1 (1.65) compared to the remaining two treatments T_2 (1.42) and T_3 (1.38).

The specific growth rate of fish in different treatments was varied among the treatments. The highest value of specific growth rate (SGR) was observed in T_1 and the lowest was seen in T_3. The SGR in T_1 (2.28±0.01) was significantly ($P<0.05$) higher than T_2 (2.24) and T_3 (2.20).

The FCR value of T_1, T_2 and T_3 were noted as 1.21±0.01, 1.30±0.01 and 1.37±0.01 respectively while the FCR value of T_1 was observed to be significantly ($P<0.05$) lowest which indicates that lower amounts of feed needed to produce one unit fish biomass and the highest was obtained in T_3. Therefore, FCR was best for fish in T_1 where the lowest number of fry was stocked (300 individual's dec^{-1}).

Throughout the analysis of the current experiment, survival of Vietnamese koi was observed as 85.52±0.86, 81.76±0.50 and 80.66±1.76 in T_1, T_2 and T_3 respectively (Table 4.1). Significantly highest ($P<0.05$) survival rate of *A. testudineus* was noted in the T_1 (85.52%) compared to T_2 (81.76%) and T_3 (80.66%) but there was no significant differences between T_2 and T_3.

At the completion of the experiment, the mean production of *A. testudineus* in different treatments was noted as 50.89±0.64, 58.72±2.81 and 70.25±0.89 (kg dec^{-1}) in T_1, T_2 and T_3 respectively (Table 4.2). Significantly ($P<0.05$) highest production was obtained in T_3 (70.25±0.89 kg dec^{-1}) compared with remaining two treatments T_1 (50.89±0.64 kg dec^{-1}) and T_2 (58.72±0.81 kg dec^{-1}) due to higher stocking density.

Table 4.2 Growth performances, feed utilization and production in different treatments of Vietnamese Koi (*A. testudineus*) over 120 day's cultured periods.

Data presented as mean±SE, obtained from three replicate ponds (n=3); Data with different superscripts in the same row indicate significant differences (*P < 0.05*).

Parameters	Treatments		
	T_1	T_2	T_3
Initial Weight (g)	1.30 ± 0.01^{a}	1.32 ± 0.01^{ab}	1.36 ± 0.01^{b}
Final Weight (g)	198.66 ± 3.28^{b}	172.33 ± 6.74^{a}	167.33 ± 3.71^{a}
Initial Length (cm)	3.81 ± 0.01^{a}	3.83 ± 0.01^{b}	3.84 ± 0.00^{b}
Final Length (cm)	19.96 ± 0.17^{b}	19.63 ± 0.38^{ab}	18.90 ± 0.20^{a}
Weight gain (g)	197.36 ± 3.28^{b}	171.01 ± 6.73^{a}	165.97 ± 3.71^{a}
Percent Weight gain (%)	15184.82 ± 319.22^{b}	12953.39 ± 474.08^{a}	12421.06 ± 316.68^{a}
Average daily gain (day^{-1})	1.65 ± 0.03^{b}	1.42 ± 0.06^{a}	1.38 ± 0.03^{a}
SGR (% day^{-1})	2.28 ± 0.01^{c}	2.24 ± 0.01^{b}	2.20 ± 0.00^{a}
FCR	1.21 ± 0.02^{a}	1.30 ± 0.01^{b}	1.37 ± 0.01^{c}
Survival rate (%)	85.52 ± 0.86^{b}	81.76 ± 0.50^{ab}	80.66 ± 1.76^{a}
Production of Vietnamese koi (kg dec^{-1})	50.89 ± 0.64^{a}	58.72 ± 2.81^{b}	70.25 ± 0.89^{c}
Production of Vietnamese koi (kg ha^{-1})	12572.43 ± 158.48^{a}	14284.10 ± 915.06^{a}	17351.75 ± 221.65^{b}

4.3 Economic analysis

A modest economic study was conducted to assess the net profit from these three types of culture operations. The expenses in three different treatments were diverse significantly ($P<0.05$) among themselves. Significantly ($P<0.05$) lowest production costs (BDT ha^{-1}) was observed in T_1 (967734±19572) compared to T_2 (1224365±51255) and T_3 (1527737±3884). The total production of Vietnamese koi as recorded in T_1, T_2 and T_3 were (13279±78), (15237±669) and (17860±230) (kg ha^{-1}) respectively. Significantly ($P<0.05$) lowest production was found in T_1. The production of fish was highest in T_3 and varies significantly with T_2 and T_1. Gross production value (BDT ha^{-1}) was found in T_1, T_2 and T_3 as (1591864±9702), (1869503±13225) and (2142168±27634) respectively which were significantly vary among these three treatments. In addition, net profit (BDT ha^{-1}) was found in T_1, T_2 and T_3 as (624130±9961), (645142±64948) and (614431±2453) respectively together with significantly ($P<0.05$) highest BCR was recorded in T_1 (1.64±0.03) followed by T_2 (1.52±0.03) and T_3 (1.40±0.02) (Table 4.3). The highest BCR was found in the lowest stocking density of Vietnamese koi in culture system.

Table 4.3 Benefit and cost analysis of Vietnamese koi (*A. testudineus*) culture over 120 day's cultured periods.

Item wise expenditures/operational cost	**Treatments**		
	T_1	T_2	T_3
Pond preparation (BDT ha^{-1})	12355	12355	12355
Price of fry (BDT ha^{-1})	92662.5	122314.5	151966.5
Feed cost (BDT ha^{-1})	813296.70	1040274.52	1313995.43
Lime, chemicals, transport, harvest etc. (BDT ha^{-1})	49420	49420	49420
Total production cost (BDT ha^{-1})	967734±19572[a]	1224365±51255[b]	1527737±3884[c]
Incomes and output			
Total production (kg ha^{-1})	13279±78[a]	15237±669[b]	17860±230[c]
Gross production value (BDT ha^{-1})	1591864±9702[a]	1869503±13225[b]	2142168±27634[c]
Net profit (BDT ha^{-1})	624130±9961[a]	645142±64948[b]	614431±2453[c]
Benefit cost ratio (BCR)	1.64±0.03[c]	1.52±0.03[b]	1.40±0.02[a]

Data presented as mean±SE, data with different superscripts in the same row indicate significant differences ($P < 0.05$).

CHAPTER 5

DISCUSSION

Growth, feed efficacy and feed ingestion of fish are typically directed by few environmental factors (Fry, 1971). Environmental aspects employ a massive effect on the maintenance of a vigorous aquatic environment and production of food organism. Amongst the environmental issues, water quality parameters show a significant part in the culture of fish and other aquatic organisms. Good water quality is absolutely a precondition for fish growth and their survival. The water quality parameters were measured in the current study during the experimental period which showed a significant variation among the treatments. However, all of those parameters were within the suitable range for fish culture (Jhingran, 1991).

Temperature shows a vigorous role in respect of fish production. In the existing study, the average temperature was recorded as 30.64±0.50, 30.74±0.48, 30.80±0.33°C in T_1, T_2 and T_3 respectively. From an experiment, Akhteruzzaman (1988) found water temperature 25.5°C to 30.0°C is favorable for fish culture. Similar results were also observed by Rahman *et al*. (1982), Patra (1993), Kohinoor *et al*. (1998) and Wahab *et al*. (2003). Formerly, several studies revealed that 25 to 35°C (Aminul, 1996), 26.93 to 27.41°C (Roy *et al*. 2002), 27.60 to 31.00°C (Rahman and Marimuthu, 2010) and 26.80 to 31.80°C (Kohinoor *et al*. 2016) are appropriate for fish culture. There was no significant variation of temperature among all the treatment groups and those values were within the suitable ranges for fish culture.

Water transparency in the existing study was detected during the experimental period and the average level was measured as 26.54±0.98, 27.29±0.60 and 26.33±0.54 cm in T_1, T_2 and T_3 respectively. Ahmed *et al*. (2009) recorded 22 to 33 cm which is more or less related to the present study. The present experiment was also relevant with Hossein (2009). Kohinoor *et al.* (2016) found that the transparency content lies between 26.75 to 30.44 cm. Thus, it can be believed that the water transparency in the present experimental pond is suitable for fish culture.

In addition, pH in the water body completely is an important factor for positive fish culture. Unexpected change of pH in the culture system may hinder the production alarmingly. In the current study, the normal values of pH in the treatments were 7.63±0.10, 7.77±0.08 and 7.73±0.87 in T_1, T_2 and T_3 correspondingly. According to Boyd (1992) pH 6.5 to 8.5 are suitable for fish culture; 0.03 to 9.03 for carp SIS Culture (Roy *et al*. 2002); 6.5 to 8.1 for benthic fauna (Shariful *et al.* 2009). Rahman and Marimuthu (2010) also noted that the suitable range of pH value is 7.40-8.50 for endangered native fish climbing perch and. Thus, it can be said that the experimental ponds were appropriate for fish culture.

Typically successful fish culture depends on the careful management of dissolved oxygen at optimal level. According to Wahab *et al.* (1995) dissolved oxygen contented of a productive pond should not be less than 4 mg l^{-1}. DoF (1996) stated that the range of dissolved oxygen content for fish culture should be 5.0-8.0 mg l^{-1}. The proper range of dissolved oxygen can vary depending on the species being cultured. Kohinoor *et al.* (2012) stated that the dissolved oxygen content between 4.23-5.32 mg l^{-1} are suitable for indigenous stinging catfish (*H. fossilis*) culture, while 4.13 to 4.71 mg l^{-1} are adequate for nursing of Thai koi (*A. testudineus*) (Rahman *et al*. 2013). The average dissolved oxygen levels in the present study were 5.97±0.14, 5.65±0.11 and 5.55±0.17 mg l^{-1} in T_1, T_2 and T_3 respectively. Therefore, the dissolved oxygen content in the present study was tolerable for fish culture.

Ammonia content was found to diverge from 0.15±0.01, 0.24±0.02 and 0.25±0.01 mg l^{-1} throughout the experimental period in T_1, T_2 and T_3 respectively. Proliferation in stocking density may also reason the deterioration of water quality, resulting in traumatic conditions (Barton and Iwama, 1991 and Pankhurst and Kraak, 1997). The ammonia content may vary from 0.20 to 0.57 mg l^{-1} for Nile tilapia and freshwater prawn culture (Rahman, 2005) and 0.01 to 0.82 mg l^{-1} for freshwater prawn post larvae treatment (Asaduzzaman *et al*. 2006). The maximum amount of ammonia was found in the treatment of T_3, which might be owed to the release of higher amounts of faecal supplies in the ponds of higher number of fish (Ahmed *et al.* 2017). This present study confirmed that the lowest stocking density showed a comparatively minor amount of ammonia, which might be a cause for the best growth performance and feed utilization parameter in the treatment of T_1.

Stocking density is a very vigorous parameter which may affect straight the growth of fish and its production (Backiel and Le Cren, 1978). The outcomes of this study clearly establish that the stocking density significantly affected growth rate, feed utilization and production of *A. testudineus* reared at different stocking density in seasonal grow out ponds.

Growth in terms of weight gain of fingerlings of *A. testudineus* was significantly higher in T_1 where the stocking density (300 individuals dec^{-1}) was low related to those of T_2 (400 individuals dec^{-1}) and T_3 (500 individuals dec^{-1}) though same feed was applied at an identical ratio in all the treatments. This is might be due to competition for feed and habitation as of the higher number of fish (Rahman and Marimuthu, 2010). Similar findings have also been stated in *M. gulio* (Begum *et al.* 2008), *A. testudineus* (Rahman and Marimuthu, 2010) and *O. niloticus* (Mensah *et al.* 2013).

The maximum value of percent weight gain was found in T_1 (15184.82±319.22) whereas the lowermost was found in T_3 (12421.06±316.68). The results designated that the percent weight gain diverse with different stocking densities which coincides with the findings of Rahman and

Marimuthu (2010). From this background, it can be believed that stocking density is known to be one of the significant parameters in fish culture.

In the existing study, the specific growth rate (% day^{-1}) was found greater in the treatment of T_1 (2.28) related to T_2 (2.24) and T_3 (2.20). The peak specific growth rate was found in the lowest stocking density, whereas the lowest specific growth rate was detected in the highest stocking density. Similar outcomes were also noted in Mirror carp, Calbasu, Indian and Chinese carps and Thai koi (Haque *et al.* 1994; Rahman *et al.* 2004, 2005 and Kohinoor *et al.* 2007). The higher specific growth rate in the lowest stocking density is might be owed to availability and superior consumption of food.

Yet, significantly lowermost FCR was detected in T_1 (1.21) followed by T_2 (1.30) and T_3 (1.37). The FCR values of the current study are minor than those stated by many workers (Reddy and Katro, 1979; Das and Ray, 1989; Islam, 2002; Islam *et al.* 2002 and Rahman *et al.* 2005), which might be because of the appropriate consumption of feed for smaller ration size and advanced digestibility. Cumulative trend of FCR values were detected with increasing ration size that were fed to Indian major carp and air-breathing catfish (Das and Ray, 1989) and common carp (*C. carpio*) fed with complementary feed (Ghosh *et al.*1984). De Silva and Davy (1992) stated that digestibility plays a significant role in dropping the FCR value of effective utilization of food. The digestibility in turn depends on many causes such as daily feeding rate, its frequency, and the type of feed used (Chiu *et al.* 1987). Nevertheless, the lower FCR value in the existing study designates better food consumption efficiency, despite the values increased with growing stocking densities.

The significantly higher survival rate was attained in T_1 (85.52) wherever the stocking density was the lowest related to those in T_2 (81.76) and T_3 (80.61). These outcomes are similar to those formerly reported by (Uddin *et al.* 1988; Saha *et al.* 1989; Haque *et al.* 1993, 1994; Kohinoor *et* *al.* 1994; Rahman and Rahman 2003; Rahman *et al.* 2005 and Asadujjaman *et al.* 2013) during the experiments of various indigenous or exotic carp and barb species. The reasons for reducing the survival rates in the upper stocking density were accounted for upper stocking density of fingerlings as well as competition for diet and space in the experimental ponds (Rahman *et al.* 2009).

After completion of the research, the maximum production was attained in T_3 (70.25 kg dec^{-1}) followed by T_2 (58.72 kg dec^{-1}) and T_1 (50.89 kg dec^{-1}). It might be due to relatively lower numbers of fry stocked in T_1 than those of T_2 and T_3 but highest individual growth was obtained in T_1. Hence, the detected poor growth at higher stocking densities could be owing to space limiting effect, stressful situation caused by supplementary feed, some variations in environmental parameters and less availability of natural food (Ahmed *et al.* 2017). This result was consistent with the results of Asadujjaman *et al.* (2013), Monir and Rahman (2015).

Generally, the maximum growth, survival, and production were found in the seasonal grow out ponds on 300 individuals dec^{-1}stocking density compared to the ponds of 400 individuals dec^{-1} and 500 individuals dec^{-1}. Physico-chemical parameters of pond water throughout the study period were within the suitable range for culture management, the growth of Vietnamese koi fry to a superior extent was reliant on the quality and quantity of food available. In the current research, the quantities of feeds provided in different ponds were founded on the number of fries stocked and the amount provided per fries was kept at the same level. Therefore, the perceived low growth at higher stocking densities might be due to less availability of natural food and any negative changes in environmental factors (Kohinoor *et al.* 1997). The existing findings are very close agreement with those stated by (Kohinoor *et al.* 1994; Rahman and Rahman, 2003 and Rahman *et al.* 2004 and 2005).

The cost-effective analysis of the grow out systems was directed to determine the economic return under little input management. The highest adjustable cost (BDT dec^{-1}) was recorded in T_3 (1527736.93±3884.32) followed by T_1 (967734.20±19571.88) and T_2 (1224364.60±51255.32) while the significantly minor gross margin (BDT) was found in the T_3 (614431.39±2452.73) in followed by T_2 (645141.730±64948.08) and T_1 (624129.81±9960.93) due to the lowest individual weight of *A. testudineus* for higher stocking density. Kohinoor *et al.* (1994) detected that monoculture of Rajputi (*P. gonionotus*) provided a net profit of BDT 68135 to 75028 ha^{-1} for 6 months cultured period. In an alternative study, Rahman *et al.* (2013) establish that the net benefit BDT 100784 to 443458 ha^{-1} from 6 months monoculture of Thai koi (*A. testudineus*) in northern Bangladesh.

Nevertheless, the lowest BCR was logged in T_3 (1.40±0.02) followed by T_2 (1.52±0.03) and T_1 (1.64±0.03) which might be due to higher FCR and higher cost necessary for the culture of koi in the treatment T_3 related to T_1 and T_2 (Chakraborty *et al.* 2005; Khan *et al.* 2003 and Usmani *et al.* 2003). Based on the outcomes of growth performances, feed consumption parameters and profit cost proportion it can be assumed that for Vietnamese koi (*A. testudineus*) culture 300 individuals dec^{-1} stocking density would be the most suggested stocking density for the fish farmers of the northern areas.

CHAPTER 6

SUMMARY AND CONCLUSION

This experiment was carried out to assess the effect of stocking density of Vietnamese koi in seasonal mini ponds over a period of 120 days in Saidpur and Kishoreganj upazila of Nilphamari district and Taraganj Upazila of Rangpur district. The water quality parameters and growth performance of Vietnamese koi (*A. testudineus*) was monitored under different stocking densities in the seasonal grow out ponds. For this trial purpose, three treatments were evaluated which each having three replications. The stocking densities were 300, 400, and 500 individuals' dec^{-1} in T_1, T_2 and T_3 correspondingly. From the opening of the research, all the experimental fish were fed with a commercial fish feed which containing 30-35 % protein.

The water quality parameters such as temperature were ranged from 29.33 to 31.81°C; transparency 24.37 to 28.70 cm, pH 7.40 to 7.96; dissolved oxygen 5.17 to 6.28 mg $l^{-1}$and ammonia content 0.13 to 0.28 mg l^{-1}. Among all the water quality parameters only a significant changes were found between the treatments in case of ammonia content. Observed all the water quality parameters of this existing study were proper for successful culture of Vietnamese koi.

For estimating the growth performance, sampling was performed after 15 days interval and the weight (g) of Vietnamese koi were logged. At the conclusion of the experiment, the average weight gain of Vietnamese koi was 197.36±3.28, 171.01±6.73 and 165.97±3.71g correspondingly in T_1, T_2 and T_3. The differences in weight gain under three treatments were significant. The peak growth rate was found in T_1in which stocking density was $300dec^{-1}$. The SGR among the three treatments were 2.28±0.01, 2.24±0.01 and 2.20±0.00 in T_1, T_2 and T_3 respectively. The SGR were significantly different three treatments and the maximum SGR was observed in the T_1 whereas the FCR was lowest in the T_1. The survival rates under the three treatments were 85.52±0.86%, 81.76±0.50% and 80.66±1.76% in T_1, T_2 and T_3 correspondingly. The most excellent survival rate was obtained in T_1 in which the stocking density was the minimum. The Benefit Cost Ratio (BCR) was monitored at the end of the trial. The best benefit was gained from T_1 in which BCR was 1.64±0.03 and the other two treatments BCR was 1.52±0.03 in T_2 and 1.40±0.02 in T_3.

The outcome of the current study indicated that the individual fish growth was lessening according to the rise of stocking density. Based on the outcome of this experiment, farmers could be suggested to culture Vietnamese koi (*A. testudineus*) at lower stocking densities (300 individuals dec^{-1}) in order to get higher production in a short period of time in the seasonal miniponds. The production of Vietnamese koi (*A. testudineus*) through the application of these current results might have important implications for the farmers to increase the quality Vietnamese koi (*A. testudineus*) production in the seasonal miniponds which finally will facilitate to progress their economic and livelihood condition.

In order to authenticate the result of this study more field trials will be needed. As a follow up to this existing research, future researches proposed are as follows:

- Effect of stocking density on the health status of Vietnamese koi in grow-out phage of northern region of Bangladesh.

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APPENDICES

Appendix A: Water Quality Analysis

A. Examination of Ammonia

- Detached the cap from the plastic beaker. Soaked the plastic beaker with water sample before filling it up to the 10 ml mark.
- Added 2 drops of Ammonia Reagent 1 for fresh water replace the cap and mix by cautiously swirling the beaker in tight circles.
- Added 8 drops of Nessler Reagent replace the cap and mix by carefully swirling the beaker.
- Detached the cap and transfer the solution into the color comparator cube. Wait for 5 minutes to allow color to grow.

- Determine which color matches, the solution in the cube, and record the results in mg l^{-1} (ppm).
- It is superior to match the color with a white sheet of around 10 cm behind the comparator.