## Nepalese Journal of

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# Nepalese Journal of 

# Aquaculture and Fisheries 

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## Status of Aquaculture in Nepal

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#### Abstract

Aquaculture is an important and blooming food sector throughout the world. Similarly, Nepalese aquaculture is fast growing. Eventhough, the amount of fish produced in Nepal is negligible compared to the world aquaculture production; however, the progress achieved in recent years is highly encouraging. Nepal is rich in natural water resources. Rivers, lakes, reservoirs, ghols and irrigated paddy fields are the major source of fresh water in Nepal. Various types of aquaculture practices are being adopted in Nepal which has all together produced 36020 mt fish in fiscal year 2012/13. Pond aquaculture is the major contributor which alone generated $86.6 \%$ ( 31221 mt ) of the total aquaculture production. Capture fisheries is very important in Nepal because of its role in fish production as well as employment generation. Capture fisheries production in Nepal is 21500 mt . Like in most of the other developing countries, employment is a serious issue in Nepal. Fisheries sub sector can provide better employment within country and minimize youth migration. Currently, there are 462,070 people engaged in capture fisheries among them $60 \%$ are female. The aquaculture development trend in Nepal is quite encouraging in recent years. Among the various culture practices, pond culture is the dominant fish rearing practice in Nepal while other aquaculture activities remained more or less constant. Seed is the most important input in aquaculture. In Nepal carp seeds are produced in sufficient quantity and distributed in three forms: hatchlings, fry and fingerlings. Both public and private sectors are responsible for seed supply. There are 14 government and 67 private hatcheries, 222 Nursery and 20 fish seed traders working in Nepal. However, import of fish


and fisheries products is in increasing trend. Fresh fish import in 2004/05 was 2547.38 mt which increased to 9963.06 mt in $2012 / 13$ with $391.1 \%$ increment. Export of fresh fish from Nepal was only 0.2 mt in 2012/13. The best export situation was recorded in 2009/10 which was 850 mt . Having tremendous scope of aquaculture development in Nepal appropriate policy, plan and programs are needed to drive this sector in right path.

Keywords : aquaculture, capture fisheries, employment, migration, export, import

## Introduction

Aquaculture is an important and blooming food sector throughout the world. Having landlocked nature of the country, only freshwater aquaculture is possible in Nepal which is basically focused on finfish culture including both warm and cold water species and most of the species under cultivation are exotic. Nepalese aquaculture is in growing stage and the amount of fish production is too low compared to the world aquaculture production; however the progress achieved in recent years is highly encouraging.

Fish is a gift of nature for health, wealth and prosperity. In Hindu religion, it is considered as an incarnation of the God and the symbol of success. It is a good source of animal protein; however its consumption in Nepal is rather low compared to poultry, buff or mutton. Its consumption in Terai belt is higher than in other regions of Nepal. Increasing health awareness among people has led to rise in fish consumption throughout the nation which is a positive sign for aquaculture industry. Fisheries and aquaculture is a reliable source of income. Thousands of people are directly or indirectly engaged in this sector that has contributed in reduction of youth migration to some extent.

This paper will discuss present status of aquaculture and fisheries, its contribution in economic development and employment generation that provides sufficient information to all national and international readers about
aquaculture and its development trend in Nepal. This paper will also be useful to planners and policy makers in identifying intervention areas and developing appropriate fisheries and aquaculture policies, plans and programs in future.

## History of Aquaculture

History of Nepalese aquaculture is very short however catching fish from nature is being practiced since ancient time. In Nepal, aquaculture was institutionalized in 2003 BS (1946/47) by establishing fisheries unit under Nepal Agricultural Research Council. This fisheries unit faced organizational modification time to time passing through the golden era of fisheries, in terms of organizational strength, when department of fisheries was established. At present, Directorate of Fisheries Development (DOFD) under Department of Agriculture (DOA), Ministry of Agriculture Development (MOAD) is the focal government organization for aquaculture development whereas fisheries research is being carried out by the Fisheries Research Division (FRD) under Nepal Agricultural Research Council (NARC), Ministry of Agriculture Development.

The first ever fisheries development program was launched in 2004 BS (1947/48 AD) but modern aquaculture started from late 1950s by introducing Common carp (Cyprinus carpio) whose successful breeding took place in mid 1960s. Three cultivable species of Chinese carps (Ctenopharyngodon idella, Hypophthalmicthys molitrix, Aristichthys nobilis) were introduced in the early 1970s followed by their successful induced breeding in mid 1970s. In the late 1970s breeding techniques of indigenous major carps (Labeo rohita, Cirrhinus mrigala and Catla catla) were established (Singh and Yadav 1996) which was significant achievement in aquaculture history that provided momentum to polyculture system in Nepal.

## Natural Water Resources

Nepal is rich in natural water resources. Rivers, lakes, reservoirs, ghols and irrigated paddy fields are the major source of fresh water in Nepal. Among them rivers and irrigated paddy fields are the most dominant water resources within the country.


Figure 1. Natural Water resources (ha) in Nepal

## Present Status of Aquaculture and Fisheries

Various types of aquaculture practices are being adopted in Nepal which all together produced 36020 mt fish in fiscal year 2012/13 (DOFD 2013). Pond aquaculture is the major contributor to aquaculture production contributing $86.6 \%$ ( 31221 mt ).

In pond aquaculture, Chinese carps and Indian major carps are the dominant species with average productivity of $3.89 \mathrm{mt} / \mathrm{ha}$. These species are generally stocked under polyculture system. However, monoculture of Common carp, Tilapia and Pangasius has also been reported.

Table 1. Status of aquaculture and fisheries in 2012/13

| Particulars | Pond <br> (No.) | Total <br> Area (ha) | Fish <br> Production <br> $(\mathrm{mt})$ | Yield <br> $(\mathrm{kg} / \mathrm{ha})$ |
| :--- | :---: | :---: | :---: | :---: |
| A. Fish Production from Aquaculture Practices |  | 36,020 |  |  |
| A1 Pond fish culture | 32,020 | 8,020 | 31,221 | 3,893 |
| Mountain | 85 | 5 | 9 | 1,800 |
| Hill | 1,940 | 210 | 440 | 2,095 |
| Terai | 29,995 | 7,805 | 30,772 | 3,943 |
| A2 Other area (ghols) |  | 2,700 | 4,050 | 1,500 |
| A3 Paddycum fish culture (ha) |  | 100 | 45 | 450 |
| A4 Cage fish culture (m${ }^{3}$ ) |  | 60,000 | 360 |  |
| A5 Enclosure fish culture (ha) |  | 100 | 140 | 1,400 |
| A6 Trout culture in raceway (m²) |  | 10,000 | 180 |  |
| A7 Fish production in public <br> sector (Mt.) |  |  | 24 |  |
| B. Fish production from capture fisheries |  | 21,500 |  |  |
| B1 Rivers |  | 395,000 | 7,110 | 18 |
| B2 Lakes |  | 5,000 | 850 | 170 |
| B3 Reservoirs |  | 1,500 | 385 | 257 |
| B4 Marginal swamps/ghols |  | 11,100 | 5,990 | 540 |
| B5 Irrigated low land paddy <br> fields |  | 398,000 | 7,165 | 18 |
| Total Fish Production (mt) |  |  | 57,520 |  |

Source: (DOFD 2013)

In Nepal, pond aquaculture has been categorized into extensive, semi-intensive and intensive farming. Intensive farming of Cirrhinus mrigala under single stocking and multiple harvesting to produce smaller size fish, called Chhadi, is also a successful farming system in Nepal. Now it is popular in eastern Terai
and gaining popularity in other regions too. Farmers have reported productivity of Chhadi system up to $12-15 \mathrm{mt} / \mathrm{ha}$. Such fish are demanded in hotels and restaurants mainly along highways.

Second contributor in fish production is Ghol. There are 2700 ha Ghols used in aquaculture with 4050 mt production in 2012/13. Fish culture in cage and enclosure together produced 500 mt fish. In Nepal, cage technology was used for the first time in 1962 in Lake Phewa to raise brood fish of Common carp (DOFD 2055). Current data shows that in Nepal, cages occupy $60,000 \mathrm{~m}^{3}$ with average productivity of $6 \mathrm{~kg} / \mathrm{m}^{3}$. This is proven as the best method of income generation for land less fishermen communities. In Lake Phewa, about 90 Jalari families have landed their cages which are a reliable source of income for them to sustain their family. However, cage culture is confined to only few lakes of Pokhara Valley and Kulekhani Reservoir that needs to be extended in other potential water bodies as well.

Rice cum fish culture is a famous farming technique in many parts of world but this could not get much attention in Nepal. Though it is promoted by various sectors, it is still limited to 100 ha with only 45 mt production.

In Nepal, Rainbow trout, a coldwater species, was introduced in 1969 from India (Rai 2010). However, the first attempt failed to expand it. Second attempt was made by introducing rainbow trout from Japan in 1988 and its commercial farming started in 2002 under one village one product program in Rasuwa and Nuwakot district. Trout farms integrated with restaurants is a common and successful practice in Nepal. By the end of fiscal year 2012/13 trout production reached to 180 mt . At present it has expanded to 18 districts and there is a potential for further expansion. Trout is a unique and the most expensive fish species in Nepalese market because of its taste and high nutritional value. National Inland Fisheries and Aquaculture Development Program is the focal government organization that implements and monitors trout development programs within the country.

Capture fisheries is very important in Nepal because of its role in fish production as well as employment generation. Capture fisheries production in Nepal is 21500 mt . Irrigated paddy fields, rivers and ghols have significant contribution in fish production whereas reservoirs and lakes have least contribution this is because lakes and reservoirs occupies small water surface areas. Fish production from ghols seems high because of its higher productivity compared to others natural water bodies.


Figure 2. Fish capture (mt) from various water bodies

## Employment Generation by Fisheries Subsector

Like in most of the other developing countries, employment is a serious issue in Nepal. There is an increasing trend of youth migration to other countries in search of better employment. Fisheries sub sector can provide better employment within country and minimize youth migration.

## Aquaculture in Employment Generation

Aquaculture is a small agriculture practice in Nepal; however it plays a significant role in employment generation. Different ages of people are involved in aquaculture from equipment preparation, fish husbandry to marketing of fish and fisheries items. There are about hundred thousand people working directly or indirectly in this sector among them male covers $67 \%$ while female occupies only $33 \%$.


Figure 3. Employment opportunity in aquaculture

## Capture Fisheries in Employment Generation

Natural water especially rivers and lakes are the source of economy to many fisheries communities such as Majhi, Jalari, Mallah in Nepal. These communities live close to water source and are dependent on its resources from generation to generation. There are 462,070 people engaged in capture fisheries among them $60 \%$ are female. Females are not only engaged in capturing fish but also in preparing fishing gears, equipments, fishing and selling fish in the market.


Figure 4. Employment in capture fisheries

## Development Trend

The aquaculture development trend in Nepal is quite encouraging in recent years. Among the various culture practices, pond culture is the dominant fish rearing practice in Nepal while other aquaculture activities remained more or less constant.

## Expansion of Pond Area

In last fifteen years 2636 ha of ponds have been constructed for aquaculture. There is slow growth of pond construction in the beginning which improved after implementation of fish mission program in the fiscal year 2007/08. The highest pond construction ( 662 ha ) was achieved in the fiscal year 2011/12. The drop in pond area in fiscal year 2008/09 was the consequence of natural disaster (flood) that damaged many ponds in Terai. Again the slow growth of pond construction in 2012/13 (only 81 ha ) is due to limited budget allocation caused by certain technical problems of then government. This clearly shows the importance of political stability for overall development of the nation.

Pond fish culture is dominant in Terai belt but its expansion in hill regions has also accelerated after implementation of pond expansion program in mid-hill districts from fiscal year 2011/12 by Nepal government.


Figure 5. Expansion trend of pond area

## Pond Production and Productivity

There is slow and steady increment of fish production annually. Significant improvement of fish production was recorded in the fiscal year 2011/12 which was natural because at the same fiscal year highest area of pond construction was achieved.


Figure 6. Production trend of pond culture

Pond productivity in 1981/82 was only $0.8 \mathrm{mt} / \mathrm{ha}$ whereas it was $3.89 \mathrm{mt} / \mathrm{ha}$ in 2012/13. This increased productivity has significant impact in increasing national fish production. Introduction of aerators, pellet machines and good management practices are the reasons for increased productivity. Mechanization of aquaculture is must to improve overall fish production. Realizing this fact, fish mission program has also given priority in mechanization.


Figure 7. Productivity trend
National Production


Figure 8. Aquaculture and capture fisheries share in national production

According to FAO country profile of Nepal, national production of fish was 500 mt in 1950. This production was entirely contributed by capture fisheries. Aquaculture production was recorded only from 1966 with only 3 mt of fish production. Aquaculture production kept increasing slowly and steadily because of growing aquaculture education and technologies. Capture fisheries shows increasing trend in the beginning but remained constant at 21500 mt since 2007/08. Keeping this capture level at stand still is a big challenge for all aquaculture and fisheries workers. Production status of fiscal year 2012/13
shows that out of 57520 mt fish production, $37.38 \%$ comes from capture fisheries whereas $62.62 \%$ from aquaculture.
Per capita fish availability is also in increasing trend. From 1981/82 to $2012 / 13$, it has significantly increased from 330 g to 2138 g due to improved national production.


Figure 9. Fish availability trend

## Fish Seed Production

Seed is the most important input in aquaculture. In Nepal fish seed are distributed in three forms: hatchlings, fry and fingerlings. Both public and private sectors are responsible for seed supply. There are 14 government and 67 private hatcheries, 222 Nursery and 20 fish seed traders working in Nepal. In last decade seed supply by public sector remained more or less constant while private sector has jumped from 5.7 million in 2001/02 to 93 million in 2012/13 this is because government has given priority to private sector in seed supply. To empower private sector, various supportive programs have been launched in the past. In fiscal year 2013/14, government has established five fish seed resource centers under private ownership one in each developmental regions of Nepal.

Table 2. Status of fish seed production in 2012/13

| C. Fish seed production/distribution (No. in '000) | 116,190 |
| :---: | ---: |
| C1 Public Sector | 22,409 |
| a. Fry | 16,612 |
| b. Fingerling | 5,797 |
| C2 Private Sector | 93,781 |
| a. Fry | 93,781 |
| C3 Public Sector | 127,645 |
| a. Hatchling* |  |

* Hatchling of public sector is distributed for fry production in private sector Source: DOFD 2012/13

Because of increasing demand, seed supply is challenging not only in terms of quantity but also in terms of quality. Government is responsible in quality control by providing financial and technical supports as well as monitoring their activities. In 2012, Food and Agriculture Organization of the United Nations (FAO) supported national fisheries program in quality seed production and regulation through a project entitled "Improving the National Carp Seed production System in Nepal (TCP/NEP/3303)". This project has drafted an act "Nepal Fish Seed Act and Carp Hatchery Accreditation \& Seed Certification" (FAO 2012) which is on the process for approval. This act will be a milestone in assuring quality seed supply in the country.


Figure 10. Fish seed production trend

## Fish Market

Fish marketing system in Nepal varies from place to place. Farmers themselves either sell their fish from the production site or send it to local markets. In case of huge production, fish is generally marketed through contractors. There are also farmers' organizations that produce fish and sell them through cooperatives. Harpan Phewa Matsya Sahakari working in Kaski district is a successful example practicing such fish marketing system. There are also such cooperatives in Nawalparasi, Rupandehi and Kanchanpur districts (ACEPP 2010). Recently concept of live fish marketing system has emerged and the number of live fish shop is increasing day by day. Government has also provided financial support in establishing fish marketing stalls and collection centers.

In last ten years, price of fish throughout the nation has increased. In 2001/02 price of fresh fish was reported to be Rs. 100 per kg which is now Rs. 250 per kg on average but this price is still lower than price of other animal meats in current Nepali market. Fish price varies from place to place. Fish are more expensive in big cities like Pokhara and Kathmandu. Study report has shown higher fish demand in winter. The least fish consumption was reported to be in Asadh, Shrawan and Bhadra (ACEPP 2010).


Figure 1 1. Market price of fresh fish

## Export/Import Scenario

Import of fish and fisheries products is in increasing trend. Fresh fish import in 2004/05 was 2547.38 mt which increased to 9963.06 mt in 2012/13 with $391.1 \%$ increment. Similarly, fish seed import is also increasing this is due to increasing caffish farmers in Nepal. These seeds are imported from India through fish seed traders and agents. Import of bone less fresh fish, dried fish and fish meal was decreased in 2012/13 compared to 2011/12.

Export of fresh fish from Nepal was only 0.2 mt in 2012/13. The best export situation was recorded in 2009/10 which was 850 mt . No record of fish seed export was shown in recent years however, 233475 fish seeds were reported in 2004/05 export record.

Table 3. Trade of fish and fisheries products

| $s$ | Year | Import |  |  |  |  |  | Export |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  | Fresh fish (mt) | Boneless fresh fish (mt) | Fish seed (No.) | Dried fish <br> (mt) | Fish meal (mt) | Aquarium fish (kg) | Fresh fish <br> (mt) | Fish seed (No.) |
| 1 | 2004/05 | 2547.38 | - | 949235 | 74,75 | 166.43 | 1950 | 1.56 | 233475 |
| 2 | 2005/06 | 2058,11 | - | 1884200 | 246.07 | 1602.95 | 3666 | 6.42 | 113000 |
| 3 | 2006/07 | 2261.23 | - | 849270 | 2510.83 | 30.02 | 549764 | 2.86 | - |
| 4 | 2007/08 | 2034.77 | - | 172590 | 277.12 | 351.2 | 2611884 | 4.15 | 22300 |
| 5 | 2008/09 | 3469.94 | - | 14212 | 313.68 | 1097.75 | - | 134.65 | 25100 |
| 6 | 2009/10 | 4334.86 | 253.2 | 7493 | 294.89 | 432.2 | - | 850.0 | - |
| 7 | 2010/11 | 5370.2 | 18.0 | 3287834 | 334.11 | 481.0 | 11158 | 0.36 | - |
| 8 | 2011/12 | 7424.94 | 381.82 | 8975129 | 580.81 | 272.33 | 28972 | 0.095 | - |
| 9 | 2012/13 | 9963.06 | 270.8 | 14564100 | 514.64 | 214.12 | 104548 | 0.2 | - |

Source: CAQO 2013.

## Conclusion and Recommendation

- Aquaculture is a blooming sector in Nepal. Realizing its importance and potential, it is receiving attention from all concerned. The yearly increment in budget allocation in recent years shows government's priority towards the sector unfortunately it did not get similar attention in the past.
- Demand of fish in Nepal is increasing because of its nutritional quality and taste. Numerous commercial fish farms are being established. Such attraction of aquaculture is only because it is highly profitable business.
- The technical knowledge of our manpower is inadequate to represent aquaculture of 21 st century. Specialization trainings and studies in specific field like breeding, disease, nutrition, genetics and water quality is required.
- Weak coordination between development, research and education has been realized. Therefore, strong interaction and bonding between these organizations is needed to promote aquaculture and fisheries program effectively and efficiently.
- Ghols are the second contributor in fish production but only $21 \%$ of them are used in aquaculture, therefore, proper planning and management is required for their optimal utilization in fish production.
- Nepal has great potential for trout therefore, identifying cost effective construction of raceways is required to attract more and more farmers towards trout culture in future.
- Mechanization, feed and seed supply, post harvest and fish market are the main area of intervention from government for considerable progress of aquaculture. Establishment of Pangasius hatchery is urgent to address today's national demand.
- Aquaculture and fisheries is a potential sector for employment generation, so, youth focused aquaculture programs should be implemented and continued.
- Highly profitable, locally initiated Chhadi and Pangasius farming should be institutionalized by the government extension system.
- National policy is needed to guide and utilize vast and diversified water resources in fish production. Any provision in wetland act that contradicts with aquaculture development should be discarded. Having tremendous scope for aquaculture development, policy makers should be very careful while drafting acts that prohibit aquaculture development in Nepal.


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Genetic variation of Mahseer (Tor putitora) populations from hatchery, lake and major rivers of Nepal

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#### Abstract

This study was carried out to assess genetic variation within and among natural and hatchery populations of Mahseer (Tor putitora). Four natural populations from Kali Gandaki River, Koshi River, Trishuli River and Phewa Lake of Nepal and two hatchery populations derived from wild population of Phewa Lake, of Mahseer were collected for genetic variation study. Analysis of five polymorphic allozyme loci in populations of Mahseer from a mid hill lake, three glacial fed rivers and two consecutive generations in a hatchery showed that all populations confirmed to be in Hardy-Weinberg equilibrium ( $P>0.01$ ), except the Trishuli River population. Observed heterozygosity (average $H_{0}=0.119 \pm$ 0.143 ) and the average number of alleles per locus (average $A=1.45 \pm$ 0.181) in the hatchery populations was less than that in lake and river populations $\left(H_{0}=0.239 \pm 0.063 ; A=1.79 \pm 0.90\right)$. The alleles EST-2*74, $I \mathrm{DH}^{*} 70$ and $\mathrm{GDH}^{*} 33$ were present in the wild populations and were absent in both hatchery generations suggested that the founders (20-30 individuals) of the hatchery populations represented bottlenecks of very small $N_{e}$. These results indicated that larger numbers of breeders and controlled mating are needed to ensure retaining large genetic variation.


Keywords: allozyme variation, heterozygosity, Mahseer

## Introduction

Fish of a genus Tor, commonly referred to as the Sahar or Mahseer, are important to trans-himalayan mid hill regions for biodiversity reasons, and are also sought affer as high-valued food and game fish (Bista et al. 2002). Among Mahseers, T. putitora is the most prevalent. T. putitora lives in the headwaters and forms a substantial natural fishery in the major riverine and lacustrine ecosystems of Nepal (Gurung et al. 2003). Despite their importance, their biological diversity is being threatened by various anthropogenic factors. These has resulted in depletion of natural stocks of $T$. putitora to such an extent that they have been identified as critically endangered species in the region (Islam 2002). Recognizing the importance of $T$. putitora, the Nepal Agricultural Research Council (NARC), made a concerted attempt to evaluate its aquaculture potential, including captive breeding using long term pond reared brood stock commencing in the late 1980s. Recent hatchery productions of $T$. putitora are coming from the second generation of hatchery bred brood stock. Hatchery produced offspring of this species are being used for stock enhancement in several natural water and for an evaluation of aquaculture potential in ponds.

The successful hatchery production of $T$. putitora brought to the most problematic questions regarding genetic variation of the hatchery stocks (brood stock). Long-term management of aquaculture production and conservation of this species would require information on levels of genetic variability within and among stocks which permits fish breeders to avoid potential detrimental effects of inbreeding and other genetic changes from one generation to another (Gjedrem 1992; Nguyen et al. 2005). There have been documented cases in many fish species of genetic changes and loss of genetic variability in hatcheryreared stocks, such as in rainbow trout, Oncorhynchus mykiss (Koljonen 1986), guppy, Paecillia reticulata (Barinova and Nakajima 1999), siamese fighting fish, Betta splendens (Meejui et al. 2005), Atlantic salmon, Salmo salar (Koljenon et al. 2002), chum salmon, Oncorhynchus keta (Hindar 2001), Pacific abalone, Haliotis discus hannai (Li et al. 2004). Alteration of genetic diversity of wild counterparts would result due to interbreeding with escapees of
hatchery-reared stocks (Cliford et al. 1998) or those used for restocking (Hinder et al. 1991). Genetic variation of hatchery populations of these species decreased due to small effective numbers of founders (Allendorf and Phelps 1980; Norris et al. 1999).

The artificial propagation implies an obvious risk of reducing the total amount of genetic diversity within the species (Ryman and Stahl 1981; Allendrof and Phelps 1980, 1981; Cross and King 1983). In hatchery for aquaculture some selection pressure is exerted for improving the quality of fish and thus such selection process change the genetic composition of the base population by reducing its genetic variability due to small population size (Hallerman et al. 1986). In contrast, programs that produce fish for release into the wild strive to maintain the genetic variation of the original wild population which requires sufficient number of founding stock to accurately reflect the genetic composition of the natural population from which it was derived (Allendorf and Ryman 1987). Unfortunately the differences in the genetic goal of aquaculture and producing fish for release in natural water have not been kept clear with regard to Sahar (Tor putitora), although hatchery production of this species with inadequate founding stock is acclaimed for release in natural water. In the long run, release of such hatchery population into the wild would produce harmful effect on a wide variety of important characteristics e.g. fecundity, survival, growth, fitness and local adaptation.

Direct genetic interactions between wild and hatchery fish have been demonstrated in many studies. Hatchery-propagated Atlantic salmon were found to compete directly with native salmon for resources such as space, food or mates, alter predation regimes and transfer disease and parasites (Fleming et al. 2000). Arrificial propagation of Steelhead (Oncorhynchus mykiss), stream-type chinook salmon (O. tshawytscha; Healey 1991), Coho salmon (O. kisutch), and probably other Pacific salmon results in significant genetic change which lowers fitness (Reisenbichler and Rubin 1999). When such fish spawn naturally, the productivity and viability of natural spawning population declines
substantially. Such cases demonstrated problems that may rise in other species as well (Utter 2003).

Genetic variability is pivotal in maintaining the capability of re-stocked fish to adapt to a new or changing environment (Avise 1994). For conservation purpose, a successful restocking program depends largely on a broodstock management strategy that ensures maintenance of a wide gene pool (Nguyen et al. 2006). This minimize adverse effects on the genetic diversity of wild populations once stock enhancement commences, thereby helping to maintain the genetic integrity of the species under consideration (Vrijenhoek et al. 1985).

An understanding of the genetic structure of wild and hatchery populations is required for effective genetic and long-term management of populations involved in reintroduction, translocations or enhancements of natural populations. The lack of genetic study to determine the genetic variability within and between hatchery stocks and their wild counterparts of $T$. putitora promoted the present study, the results of which may help in design of hatchery stock management strategy and subsequent application to future aquaculture production and genetic conservation strategies. Allozyme markers have been used to quantify genetic variation within and among populations of Mahseer from hatchery and different water drainage system of Nepal.

## Materials and methods

## Sample collection

Liver and muscle tissues were collected during 2004 to 2005 from a total of 144 samples of T. putitora, comprising four wild populations and two hatchery populations. The wild populations were from three glacial fed rivers, 1) Kali Gandaki River system ( $27^{\circ} 58^{\prime} \mathrm{N}$ and $83^{\circ} 35^{\prime} \mathrm{E}$ ), 2) Trishuli River ( $27^{\circ} 58^{\prime} \mathrm{N}$ and $84^{\circ} 52^{\prime} \mathrm{E}$ ), 3) Koshi River ( $26^{\circ} 43^{\prime} \mathrm{N}$ and $87^{\circ} 20^{\prime} \mathrm{E}$ ), and one from Phewa Lake ( $28^{\circ} 13^{\prime} \mathrm{N}$ and $84^{\circ} 00^{\prime} \mathrm{E}$ ) (Figure 1). The hatchery populations comprised
of the first and second generations of mahseer derived from a wild population from Phewa Lake.


Figure 1. Map of Nepal showing natural habitat of T. putitora sample collection: (A) Phewa Lake, (B) Kali Gandaki River, (C) Trishuli River, and (D) Koshi River

## Isozyme analysis

Seven enzyme systems were analyzed following methods described by Morizot and Schmidt (1990), and Hara and Na-Nakorn (1996). Horizontal starch gel $(11 \% \mathrm{w} / \mathrm{v}$ hydrolyzed potato starch) electrophoresis was operated in appropriate buffer systems. Details of enzymes, E.C. numbers, tissue, buffer used, and resolved are shown in Table 1. A voltage of 40 mA (constant current) and 220-280 V (voltage) were applied for approximately 6 hours at $4^{\circ} \mathrm{C}$. Chemical visualization was done following Morizot and Schmidt (1990) and gene nomenclature following Shaklee et al. (1990).

Table 1. Names of enzyme systems, their E.C. number, tissue and buffer used, and the resolved loci.

| Names of enzyme | E.C. number | Tissue | Loci |
| :---: | :---: | :---: | :---: |
| Isocitrate dehydrogenase (IDH) | E.C.1.1.1.42 | L | $1 \mathrm{DH}^{*}$ |
| Esterase (EST) | E.C.3.1.1.1 | L | EST-1* |
|  |  |  | EST-2* |
| Malate dehydrogenase (MDH) | E.C.1.1.1.37 | M | sMDH-7* |
|  |  |  | sMDH-2* |
| Malic enzyme (ME) | E.C.1.ו.ו. 40 | M | ME-7* |
|  |  |  | ME-2* |
| Sorbitol dehydrogenase (SDH) | E.C.1.1.1. 14 | M | SDH-1* |
|  |  |  | SDH-2* |
| Glucose-6-phosphate dehydrogenase (G6PD ) | E.C.1.1.1.49 | M | G6PD* |
| Glucose dehydrogenase (GDH) | E.C.1.1.1.47 | L | GDH* |

$\mathrm{L}=$ liver, $\mathrm{M}=$ muscle
Buffer: Tris-citrate pH 8.0

## Data analysis

The presumed genotypes were used for calculation of allele frequencies. Populations were tested for departure from Hardy-Weinberg equilibrium using Markov chain method. Genetic variation within populations, percentage of polymorphic loci-P (a locus was considered polymorphic if the highest allele frequency did not exceed 0.95), number of allele per locus, observed and expected heterozygosity ( $H_{o}$ and $H_{e}$, respectively) were calculated following Hedrick (1985).

The chance of drawing no individuals with given alleles in a sample were calculated as the frequency of alternate alleles ${ }^{(2 \times \text { number of individual) }}$ (Dillon and Manzi 1987).

Locus-wise $F_{\text {is }}$ (Weir and Cockerham 1984) were calculated within each of the populations. Wright's F-statistic approach (Wright 1951, 1978) and its exact
test were calculated to test for genetic population structure. Population differentiation and genotypic disequilibrium were tested using the Markov chain method. The calculations including genetic distances and UPGMA dendrogram (Nei 1978) were performed by the POPGENE version 1.32 (Yeh and Boyle 1997) and software TFPGA (Miller 1997). Independent sample comparison (Archie 1985) was used to test for differences between observed and expected heterozygosities. Multiple simultaneous tests used the Bonferroni correction for expected probabilities (Rice 1989) of heterozygosities and Locus-wise $F_{\text {is }}$.

## Results

Genetic diversity of natural and hatchery populations of Tor putitora
Analysis of seven enzyme systems resulted in 11 loci being resolved. Five loci were polymorphic (a locus was considered polymorphic if a frequency of most common allele did not exceed 0.95-P. ${ }_{.95}$ ) (EST-2*, sMDH-1*, IDH*, GDH* and MEP-1 ${ }^{1}$. Allele frequencies of the polymorphic loci are shown in Table 2.

Table 2. Allele frequencies of five polymorphic allozyme loci in four wild populations of Tor putitora from lake and river systems, and two hatchery populations in Nepal.

| Loci | Allele Frequencies |  |  |  |  |  |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Allele | Kali Gandaki | Trishuli River | Koshi <br> River | Phewa Lake | Hatchery pop. I | Hatchery pop. II |
| EST-2* | 116 | 0.430 | 0.426 | 0.733 | 0.371 | 0.325 | 0.150 |
|  | 100 | 0.558 | 0.574 | 0.267 | 0.611 | 0.675 | 0.850 |
|  | 74 | 0.012 | 0 | 0 | 0.018 | 0 | 0 |
| IDH* | 100 | 0.569 | 0.833 | 0.700 | 0.656 | 0.875 | 0.900 |
|  | 83 | 0.431 | 0.167 | 0.300 | 0.281 | 0.125 | 0.100 |
|  | 70 | 0 | 0 | 0 | 0.063 | 0 | 0 |
| GDH* | 174 | 0.333 | 0.416 | 0.687 | 0.250 | 0.525 | 0.235 |
|  | 100 | 0.633 | 0.500 | 0.313 | 0.694 | 0.475 | 0.765 |
|  | 33 | 0.034 | 0.084 | 0 | 0.056 | 0 | 0 |
| sMDH-7* | 100 | 0.659 | 0.583 | 0.553 | 0.333 | 0.625 | 0.792 |
|  | 51 | 0.341 | 0.417 | 0.447 | 0.667 | 0.375 | 0.208 |
| MEP-7* | 100 | 0.800 | 0.621 | 0.588 | 0.750 | 0.800 | 0.607 |
|  | 39 | 0.200 | 0.379 | 0.412 | 0.250 | 0.200 | 0.393 |

Departure from HW equilibrium and linkage disequilibrium
Significant departure from the Hardy-Weinberg equilibrium ( $P<0.05$ ) was observed in one (Trishuli River) out of six populations after Bonferroni correction as indicated by significant $F_{\text {is }}$ value (Table 3). Linkage disequilibrium was significant ( $P<0.05$ ) between $I D H^{*}$ and $M E P-1{ }^{*}$ in Koshi River population.

Table 3. Measures of genetic variability, average observed $\left(H_{0}\right)$ and expected $\left(H_{e}\right)$ heterozygosity, fixation index $\left.\left[F_{i s}=\left(\mathrm{H}_{\mathrm{e}}-\mathrm{H}_{\mathrm{o}}\right) / \mathrm{H}_{\mathrm{e}}\right)\right]$, probability for the Hardy-Weinberg exact test (Marcov chain method; Bonferroni corrected $P=0.003$ ) within hatchery and natural populations of Tor putitora in Nepal. Means in the same column superscripted by different letters were significant difference ( $P<0.01$ )

| Populations | No. samples per Locus | No. alleles per locus (SE) | Average heterozygosity |  | $F_{\text {is }}{ }^{\text {a }}$ | $p$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  | $H_{0}(S D)$ | $H_{e}(S D)^{+}$ |  |  |
| Kali Gandaki | 41 | $\begin{aligned} & 1.63 \\ & (0.81)^{b} \end{aligned}$ | $\begin{aligned} & 0.203 \\ & (0.246) \end{aligned}$ | $\begin{aligned} & 0.204 \\ & (0.239) \end{aligned}$ | 0.003 | 0.0382 |
| Trishuli River | 29 | $\begin{aligned} & 1.54 \\ & (0.68)^{c} \end{aligned}$ | $\begin{aligned} & 0.250 \\ & (0.329)^{a} \end{aligned}$ | $\begin{aligned} & 0.208 \\ & (0.249) \end{aligned}$ | -0.083 | 0.0001 |
| Koshi River | 17 | $\begin{aligned} & 2.27 \\ & (0.64)^{a} \end{aligned}$ | $\begin{aligned} & 0.323 \\ & (0.239)^{a} \end{aligned}$ | $\begin{aligned} & 0.372 \\ & (0.189) \end{aligned}$ | 0.131 | 0.0189 |
| Lake Phewa | 25 | $\begin{aligned} & 1.72 \\ & (0.90)^{b} \end{aligned}$ | $\begin{aligned} & 0.179 \\ & (0.221)^{a} \end{aligned}$ | $\begin{aligned} & 0.184 \\ & (0.196) \end{aligned}$ | 0.02 | 0.4316 |
| Hatchery I | 17 | $\begin{aligned} & 1.45 \\ & (0.52)^{c} \end{aligned}$ | $\begin{aligned} & 0.181 \\ & (0.233)^{a} \end{aligned}$ | $\begin{aligned} & 0.176 \\ & (0.216) \end{aligned}$ | 0.02 | 0.6414 |
| Hatchery II | 15 | $\begin{aligned} & 1.45 \\ & (0.52)^{c} \end{aligned}$ | $\begin{aligned} & 0.119 \\ & (0.143)^{b} \end{aligned}$ | $\begin{aligned} & 0.145 \\ & (0.181) \end{aligned}$ | 0.17 | 0.0111 |

Genetic variation within natural and hatchery populations
Based on 5 polymorphic loci, all populations had 45.45 percentage polymorphic loci. The number of alleles per locus ranged between 1.45 to 2.27, with the hatchery populations (average $A=1.45 \pm 0.18$ ) showing less variation in average than the wild populations $(A=1.79 \pm 0.90)$, although the difference in value between the hatchery and Trishuli River population was not significant (Table 3).

Observed heterozygosities showed a similar pattern, with those for hatchery populations being less ( $H_{0}=0.119 \pm 0.143$ ) than the wild populations ( $H_{0}=0.239 \pm 0.063$ ) on average. It is pertinent that the values for Lake Phewa were less than that of the wild rivers and similar to those for hatchery generation I, and that heterozygosity decreased from hatchery generation I to generation II. Although the error range was large in all estimates, there were no significant differences between the values for observed and expected heterozygosity within populations.

Paired sample $t$-test revealed that both hatchery populations exhibit significantly lower ( $P<0.05$ ) number of alleles per locus (mean $A=1.45$ ) compared to that of natural population (mean $A=1.79$ ). $A$ between hatchery populations and the source population - Lake Phewa were significantly different ( $\mathrm{P}<0.05$ ). Alleles EST-2*74, IDH*TO and $G D H^{*} 33$ were not found in either hatchery generation although they were all present in their wild source population - Lake Phewa, albeit at frequencies less than 0.06 (Table 2). The chance of completely missing allele $I \mathrm{DH}^{*} 70$ and $G D H^{*} 33$ in the sample of hatchery populations was just 0.017 and 0.035 , respectively.

Except for the Trishuli River, each population had a significant deficit of heterozygotes at only one locus. In three natural populations this was at $G D H^{*}$, in hatchery population I at $s M D H-1^{*}$ and hatchery population II was at MEP-1*. Two loci EST-2* and MEP-1* showed significant excess of heterozygotes in the Trishuli River population (Table 4).

Table 4. Locus-wise $F_{\text {is }}$ (Weir and Cockerham 1984) within each of 6 populations of Tor putitora in Nepal

| Locus | Kali <br> Gandaki | Trishuli <br> River | Koshi <br> River | Phewa <br> Lake | Hatchery <br> Pop. I | Hatchery <br> Pop. II |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| EST-2* | -0.097 | $-0.733^{*}$ | -0.333 | -0.117 | -0.000 | -0.152 |
| $I D H^{*}$ | -0.178 | 0.227 | 0.548 | 0.385 | -0.077 | -0.059 |
| GDH $^{*}$ | $0.460^{* *}$ | 0.288 | $0.725^{* *}$ | $0.410^{*}$ | -0.280 | 0.050 |
| sMDH-7 $^{*}$ | 0.090 | -0.012 | 0.376 | 0.270 | $0.486^{*}$ | 0.283 |
| MEP- $^{*}$ | -0.236 | $-0.556^{*}$ | -0.185 | -0.318 | -0.200 | $0.576^{*}$ |

'Statistically significant ( $\mathrm{P}<0.002$ - Bonferroni correction)

## Genetic variation between populations

The average unbiased estimated of $F_{\text {ST }}$ across the five loci was 0.0844 (EST-2* $=0.1239, I D H^{*}=0.0766, G D H^{*}=0.0957, s M D H-7^{*}=0.0786$, MEP-1* $=0.0390$ ). $F_{\mathrm{st}}$ values for $E S T-2^{*}$ implied that it contributed the most for population differentiation.

The pairwise analysis of $F_{\text {ST }}$ revealed that population from Koshi River was moderately differentiated ( $F_{\mathrm{ST}}=0.0467$ to 0.1186 ) from rest of the wild populations. Pairwise $F_{\text {ST }}$ value estimated between populations hatchery II and its source population Lake Phewa was 0.1145 indicating strong population differentiation.

## Genetic distance

Nei's genetic distance was generally greatest (ranging between $0.0133-0.0642$ ) between the Koshi River population and all others (Table 5). Differences between the other rivers, Lake Phewa and Hatchery population I were usually less than 0.01 although that between Lake Phewa and Hatchery population I was 0.0149 . In contrast Hatchery population II tended to show greater differentiation from all wild populations than Hatchery I.

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Table 5. Genetic distance (Nei 1978) for the six Tor putitora populations of Nepal below diagonal, pairwise Fst above diagonal (after Bonferroni correction)

| Population | Kali <br> Gandaki | Trishuli <br> River | Koshi <br> River | Phewa <br> Lake | Hatchery <br> Pop I | Hatchery <br> Pop II |
| :--- | :---: | :---: | :---: | :---: | :---: | :---: |
| Kali Gandaki | ${ }^{* * *}$ | 0.0306 | $0.0803^{*}$ | 0.0298 | 0.0340 | $0.0931^{*}$ |
| Trishuli River | 0.0083 | ${ }^{* * *}$ | 0.0467 | 0.0345 | -0.0014 | $0.0570^{*}$ |
| Koshi River | 0.0239 | 0.0133 | ${ }^{* * *}$ | $0.1186^{*}$ | $0.0777^{*}$ | $0.2128^{*}$ |
| Phewa Lake | 0.0086 | 0.0099 | 0.0379 | ${ }^{* * *}$ | $0.0512^{*}$ | $0.1145^{*}$ |
| Hatchery Pop I | 0.0093 | 0.0010 | 0.0230 | 0.0149 | ${ }^{* * *}$ | $0.0529^{*}$ |
| Hatchery Pop II | 0.0252 | 0.0136 | 0.0642 | 0.0308 | 0.0127 | ${ }^{* * *}$ |

*Statistically significant at 0.003 after Bonferroni correction

A UPGMA clustering confirmed the relative genetic distance of Lake Phewa from all other populations and demonstrated that the genetic distance between the Hatchery populations and their wild progenitor (Lake Phewa) was greater than that between the hatchery populations and some of the other wild sites (Figure 2).


Figure 2. UPGMA Cluster drawn from Nei (1978) unbiased genetic distances.

## Discussion

Levels of intrapopulation variation in natural populations of $T$. putitora as measured by $H_{0}$ (mean $=0.239$; ranged $0.179-0.323$ ) in this study were similar to that reported in a populations in India ( $H_{0}=0.194-0.236$ ) by Haniffa et al. (2007). $H_{0}$ of natural populations of $T$. putitora were high as compared to population of siamese fighting fish ( $H_{0}=0.065$ ) and hatchery populations of other species, based on the same allozyme marker, such as black sea bream, Acanthopagrus schlegeli ( $H_{0}=0.048-0.052$ ), Oreochromis mosssambicus in Japan and Philippines ( $H_{0}=0.022$ ), $O$. niloticus in Japan, Philippines, Taiwan and Thailand $\left(H_{0}=0.073\right)$; Cyprinus carpio in Japan ( $H_{0}=0.074$ ) (Macaranas and Fujio 1990).

The lower genetic variability of the domesticated stocks compared to wild populations reflect a phenomenon found in other fish species too, i.e., masu salmon (Nakajima et al. 1986), rainbow trout ( Paaver 1986), Atlantic salmon (Stahl 1983; Verspoor 1988) and brown trout (Vuorinen 1984b). The loss of genetic variability can be explained by genetic drift due to small population sizes and/or by inbreeding due to high selection intensities (e.g. in ornamental Koi carp). Bottleneck effects at the beginning or during the culture of populations as well as genetic adaptations to captive and local environmental conditions could be additional contributing factors.

Allendroff and Phelps (1980) detected a significant reduction in electrophorectically detectable genetic variation in a hatchery stock of Salmo clarki in comparison to the wild stock from which it was derived. Allendrof and Utter (1979) and Ryman and Stahl (1980) reported similar results for other salmonid species. Genetic differences between wild and hatchery stock can be atributed to a number of factors, e.g. size of the founder parents stock, differential survival of larvae under hatchery conditions, degree of inbreeding (Wilkins 1975).

The range in genetic distance between the Nepalese populations also fell within that considered to reflect variation within a species of 0.002 to 0.07 (mean 0.05 ) (Shaklee et al. 1982). The mean $F_{\text {sT }}$ value ( 0.0844 ) indicated moderate genetic differentiation among populations of $T$. putitora under study.

The extent to which the patterns of variation in the wild and hatchery populations of T. putitora observed in this study to provide a basis for management decisions is limited by the small number of allozyme loci used, and by the relatively small sample sizes analyzed. Nevertheless this first study of fish populations in Nepal has provided evidence for a reduction of genetic variability in the hatchery populations.

Allele numbers were less and heterozygosity slightly less in the hatchery populations relative to their wild source - Lake Phewa. It seems likely that these alleles were not simply missed in founding the hatchery population but may have been lost since. Genetic drift and /or selection could have lead to loss of alleles in the hatchery stocks. Once alleles are lost through drift, new variation can only be introduced through mutation or through the introduction of new stock into the hatchery.

However, the comparison of the wild populations showed that Lake Phewa had lower allelic diversity and heterozygosity than some other river populations, in particular Koshi River. Regular restocking of hatchery produced fish to Lake Phewa might have reduced the genetic diversity of lake population. Stock enhancement may cause extinction if the stocking program is carried out for such a long time that the natural population gradually is replaced by the released stock (Laikre et al. 1999). In such case the result is that the gene pools of indigenous and possibly locally adapted wild populations are replaced by the gene pools of non-native domesticated fish. Cultured fish may differ genetically from natural populations by origin or through selection or inbreeding. Such selection and inbreeding may be unintended but may nevertheless result in genetic changes (Ruzzante 1994). Hence, the artificial propagation implies an obvious risk of reducing the total amount of genetic diversity within the species (Ryman and Stahl 1980).

This suggests that caution should be exercised in enhancing stock from different catchments with material only from Lake Phewa. The magnitude of reduction was $18.6 \%$ in the average number of alleles per locus in hatchery populations and $33.3 \%$ in the observed heterozygosity in hatchery population II relative to the average for wild populations. Generally, heterozygosity of hatchery populations tend to decline due to increased inbreeding rate as the result of small effective population size (Falconer 1983). The detrimental effects of inbreeding in Crassostrea virginica (Longwell and Stiles 1973) and in salmonids (Kincaid 1976a; Kincaid 1976b) have been well documented. The reduction in general measures of genetic diversity and the decreasing heterozygosity from hatchery generation I to II provide evidence of loss of genetic variation not just in founding the population, but as a result of inbreeding and genetic drift over time. It is likely that effective population size were much smaller than the numbers of fish used to produce hatchery population II. It seems essential to make efforts to increase the number of fish which contribute to reproduction of subsequent generation. Kapuscinski and Jacobson (1979) recommended that inbreeding could be minimized if effective population size $\left(N_{e}\right)$ exceeded 50 or by using a number of brooders at between 263 and 344 fish per generation.

There was moderate genetic differentiation between Koshi River and other wild sites, and significant divergence of the hatchery populations from its source stock. Divergence of Hatchery generation II from all populations was greater than Hatchery generation I, suggesting that drift or hatchery selection was altering allele frequencies in the hatchery population. Another observation indicating significant excess heterozygosity at Trishuli River suggests the possibility of selective differences between populations, and/or of hybridization between populations fixed at different loci. The occurrence of heterozygote deficits at single loci in other populations could arise from mixing of populations differing in allele frequencies. Together, these data indicate geographical genetic variation and also mean that more consideration needs to be given as to the sources used for given enhancement operations, as much as to the hatchery management processes used for the present hatchery population.

The preliminary data obtained in this study have shown clear evidence of differentiation of wild populations and loss of variation in the present hatchery processes used for enhancement of Tor putitora in Nepal. The basic aquaculture processes needed to sustain fishery enhancement programs have been achieved, but these new data suggests that translocation of this species among rivers and lake systems, while feasible, requires further planning to obtain long term genetic sustainability.

## Conclusions

The benefits (production) from a stock enhancement programme may be less, perhaps much less, than those expected without consideration of genetic changes. Future brood stock management, breeding designs and equalization of family contribution of the offspring to be released will be required for this endangered species to avoid unintended genetic differentiation between the wild population and the hatchery populations. $\mathrm{N}_{\mathrm{e}}$ of 500 will prevent inbreeding - and genetic drift-related problems in farmed populations and that $\mathrm{N}_{\mathrm{e}}$ of 1,000 can do a fairly effective job of conserving genetic variance and minimizing inbreeding in populations that are used for stocking programmes. In the process of developing recovery plans, managers of threatened or endangered species must establish recovery criteria and goals for management of critical populations. Managers may also prioritize limited resources for habitat conservation and restoration, based on some measure of risk (Allendorf et al. 1997). If the general guidance of the 50/500 criteria (Allendorf and Ryman 1987) is to be accepted a cautious interpretation of the results would be that approximately 100 (100 30.5550 ) adults spawning each year would be required to minimize the risks of inbreeding in any population. An average of 1,000 (1000 30.55500 ) adults spawning annually would be necessary to maintain genetic variation indefinitely. Those criteria might be relaxed if there were clear evidence that the adult population is larger than the number of fish spawning in any year (because all females do not spawn in all years), or if more precise estimates

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Genetic changes after two generations of divergent mass selection for body weight in Thai walking caffish (Clarias macrocephalus günther, assessed by allozyme variation

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#### Abstract

Divergent (bidirectional) mass selection for body weight was performed in Thai walking catfish Clarias macrocephalus. Response to selection after two generations was $3.19 \mathrm{~g}(3.4 \%)$ at 33 weeks of age. Significant divergence in body weight of the low and high line occurred at 29 and 33 weeks of age. Change in genetic variation was measured by comparing allozyme variation of the population before second selection $\left(F_{1}\right)$ and offspring of the selected individuals $\left(F_{2}\right)$. Results showed that mean numbers of alleles per locus significantly reduced ( $\mathrm{P}<0.05$ ) in the successive generation in both lines. The values of the mean observed heterozygosity $\left(\mathrm{H}_{0}\right)$ were 0.0804 and 0.0803 for the low line and 0.0788 and 0.1003 for the high line, respectively, in generation $F_{1}$ and $F_{2}$; and were not significantly different ( $P>0.05$ ) between generations of the respective line. Allele frequencies were changed in the successive generation of the low line. Distributions of transformed allele frequency variance and ratio of observed and expected allele frequency variance indicated that change in allelic frequencies between generations in low line was caused by random genetic drift at some variable loci. Selection for


body weight caused allele frequency change, although not significant, between generations in high line at most of the variable loci.

Keywords: Thai walking caffish, isozyme, mass selection, generation, allele frequency

## Introduction

Aquaculture production of Clarias comprises significant portion of freshwater fish culture production in many countries such as India, Thailand and Indonesia (SEAFDEC 1993). The hybrid caffish (Clarias macrocephalus X C. gariepinus) is one of the most preferred species because it grows faster than the native ones. The annual production of $C$. macrocephalus, including its hybrid was 86,475 tones in Thailand (DOF 2004). However, the production per unit area of the hybrid caffish is decreasing which might be due to the quality of the male African caffish which was introduced into Thailand a long time ago (FITC 2004). Production per unit area of $C$. macrocephalus is low due to slow growth rate and high disease susceptibility (Na-Nakorn et al. 1993). Superior strain of C. macrocephalus is required to promote the expansion of culture of this species as well as to avoid a threat on biodiversity caused by introgressive hybridization and/or exclusive competition, which may have occurred due to escape of the hybrid into natural water (Na-Nakorn 1999).

Selection is an effective method used for fish genetic improvement, provided that a target trait is mainly controlled by additive genetic variance (Falconer 1989). Selection methods have been efficiently used to improve economical important traits of aquatic animals (Hershberger et al. 1984; Gjedrem 1997) but selection programs aiming to improve traits such as growth and disease resistance of C. macrocephalus were not successful (Na-Nakorn 1993; NaNakorn et al. 1994; Chamnankuruvet 1996).

A selection program has often entailed the collection of a limited number of superior individuals to be used as broodstock. The limitation of population size during selection may lead to a reduction in genetic variation of the selected
population (Kincaid 1983). The potential harmful consequences of directional artificial selection are random genetic drift and inbreeding. It has been observed that the reduction in genetic variability in selected populations was mainly caused by random genetic drift, leading to the fixation of minor alleles at a few loci and the change in the alleles of intermediate frequency (Taniguchi et al. 1983). Combined effect of genetic drift and inbreeding caused loss of allelic diversity and reduction of heterozygosity in selected populations of Japanese pearl oysters (Wada 1986) and kuruma prawn (Sbordoni et al. 1987). Loss of genetic variation in selected population in the face of genetic drift and inbreeding correlated with declines in performance traits (Pante et al. 2001) and physiological traits (Chiyokubo et al. 1998). Therefore, it is necessary to investigate the consequences of selection breeding practices on their genetic structure as well as on their performance. There had been no studies on the quantitative estimation of genetic variation at isozyme level for the identification of source and amount of variation among generations of divergent lines of $C$. macrocephalus selected for body weight.

This study considers response to mass selection and expression of 14 biochemical loci in divergent lines of C. macrocephalus which had been selected for two generations for body weight. The objectives of this study were to measure the response of selection from divergent lines of $C$. macrocephalus and to determine levels of genetic variability in terms of change in allele frequencies among generations and the consequence of selection or genetic drift in observed allele frequency changes between generations of divergent lines.

## Materials and methods

## Experimental fish

Divergent selection on body weight was performed by mass selection on 10 months old $C$. macrocephalus, derived from crossing of 6 natural populations, for two generations at Kasetsart University, Bangkok, Thailand. Selection intensity (i) was 1.50 and 1.46 , respectively, for the low and high lines in the
first selection (May 2000), and it was 1.48 and 1.61 , respectively, for the low and high line in the second generation of selection (March 2001).

Fifty pairs of $F_{2}$ selected brooders from each line of $C$. macrocephalus were randomly taken for reproduction. Ovulation was induced by administration of LHRH-a and domperidone at rates of $35 \mu \mathrm{~g} / \mathrm{kg}$ and $10 \mathrm{mg} / \mathrm{kg}$ of ripe females, respectively, while males received LHRH-a and domperidone at rates of 10 $\mu \mathrm{g} / \mathrm{kg}$ and $5 \mathrm{mg} / \mathrm{kg}$, respectively. A single pair mating was followed. Nursing of fish larvae was carried out in separate tanks for each line. First feeding to larvae started swimming was started with freshwater cladoceran (Moina sp.) at $0.5-1.0 \mathrm{~kg}$ per 100,000 fry for the first $7-10$ days of feeding. Then artificial powder feed of $40 \%$ protein (Charoenphokphan Feed Company) was used until they reached $6-7 \mathrm{~cm}$ body length at 7 weeks of age. The fingerlings were subsequently transferred to net-cages for a growth trial.

## Study of selection response

Growth performance of high and low line was compared using a completely randomized design (CRD). The fingerlings of 7 week old were reared in nylon net cages of $2.7 \mathrm{~m}^{3}$ each, placed in a 800 -square meters earthen pond. A stocking rate was 100 fry per $\mathrm{m}^{3}$ of cage and the initial average weight of fry was $2.7 \pm 0.2$ and $2.8 \pm 0.2 \mathrm{~g}$, respectively, for low line and high line. Fry were fed to satiation on pellete containing $41 \%$ protein for 14 weeks daily. Later diet was changed to $30 \%$ protein content. Monthly growth check of fry was done by weighing 30 fish per cage. The fish were raised until they were 33 weeks old and reached a marketable size. At harvesting, survival rates were calculated. Response to selection as one half of the divergence between selected lines was evaluated during each monthly growth check.

## Assessment of genetic change

Seventy-seven fish of the low line and 115 fish from the high line were randomly taken from the $F_{1}$ population. Fifty individuals from $F_{2}$ generation of each selected line were obtained alive and the tissue (muscle and liver) dissected. The tissue samples were immediately frozen and stored at $-30^{\circ} \mathrm{C}$ in the laboratory until isozyme analysis was performed.

Horizontal starch-gel ( $11 \% \mathrm{w} / \mathrm{v}$ hydrolyzed potato starch) electrophoresis of tissues (liver or muscle) of sampled fish were carried out following Hara and Na-Nakorn (1996) and chemical visualization by using methods described by Morizot and Schmidt (1990). Details of enzyme systems, E.C. numbers and tissues used are given in Table 1. Shaklee et al. (1990) was followed for genotype nomenclature. Loci are identified by italicized lettered abbreviations reflecting the name of enzyme or protein assigned by the International Union of Biochemistry's Nomenclature Committee (IUBNC).

Table 1. List of protein or enzyme systems, E.C. number and tissues used in electrophoresis study of selected lines of C. macrocephalus

| Enzyme or protein | E. C. No. | Tissue used $^{1}$ |
| :--- | :---: | :---: |
| Glucose-6 phosphate isomerase (GPI) | 5.3 .1 .9 | L |
| Malate dehydrogenase (MDH) | 1.1 .1 .37 | M |
| Malic enzyme-NADP (MEP) | 1.1 .1 .40 | M |
| Mannose-6-phosphate isomerse (MPI) | 5.3 .1 .8 | L |
| Phosphoglucomutase (PGM) | 5.4 .2 .2 | L |
| Phosphogluconate dehydrogenase (PGDH) | 1.1 .1 .44 | M |
| Alcohol dehydrogenase (ADH) | 1.1 .1 .1 | L |
| Glycerol-3-phosphate dehydrogenase (G3PDH) | 1.1 .1 .8 | M |
| Esterase (EST) | $3.1 .1 .-$ | L |
| Leucine aminopeptidase (LAP) | 3.4 .11 .1 | M |
| Isocitrate dehydrogenase (IDHP) | 1.1 .1 .42 | L |
| General protein (PROT) |  | M |

Buffer: TC pH 8; Tris 16.35 g ; Citric acid 9.04 g ; Distilled water to 1 L .
${ }^{1}$ Tissue: $M=$ muscle; L=liver

## Data analysis

Individual body weight of both selected lines at various growth stages and survival rates at harvesting were analyzed using analysis of variance (ANOVA). Mean body weight was compared using $t$-test. Due to a concern
that variation in survival rates between cages might have affected variation in growth performance, correlation between survival rates and mean body weight was computed with Pearson correlation method. Differences in coefficient of variation of body weight among and within selected lines were compared by $\dagger$ test.

Individual data on presumed genotypes were used to calculate mean and effective number of alleles per locus, the percentage of polymorphic loci, and the mean observed and expected heterozygosity (Nei 1978) by using soffware TFPGA 1.3 (Miller 1997). An independent sample comparison of heterozygosity data was done following Archie (1985). Changes of allele frequencies in $F_{2}$ selected generation of both low and high lines from the previous generation was tested using likelihood ratio $\left(G^{2}\right)$ tests.

Indirect estimate of effective population size for the divergent lines was made based on the theory of selectively neutral alleles in a finite population (Pollak 1983). Allele frequencies of $F_{1}$ and $F_{2}$ generations of each line were considered at time $t=0$ and $t=1$, respectively. Temporal variance of allelic frequencies $\left(F_{k}\right)$ for each lines over the interval from the $t=0$ to $t=1$ was calculated according to Nei and Tajima's (1981 cited by Hedgecock and Sly 1990). From these estimates of variances in allelic frequencies between $F_{1}$ and $F_{2}$ generation, effective population size ( $\mathrm{N}_{\mathrm{e}}$ ) of the divergent lines of $C$. macrocephalus was estimated, using a method based on the inverse relationship between the magnitude of temporal shift in the frequencies of selective neutral genes and $\mathrm{N}_{e}$ (Pollak 1983).

For drift assessment, temporal allele frequency variances $\left(F_{\mathrm{k}}\right)$ of each selected lines over successive generations ( $F_{1}$ vs $F_{2}$ ) were transformed to standard normal variates ( x ) (Hedgecock and Sly 1990). These transformed variates were plotted against standard normal variates to observe whether the resulting distribution was normal. Normality was assessed by the Kolmogorov-Smirnov goodness-of-fit test (plots and test done with SYSTAT ver. 5.04). Measurement of genetic drift was made by comparing expected (under drift alone) and
observed variances of the change in gene frequencies (Crow 1954 cited by Rich et al. 1979). Expected variance of gene frequency (Rich et al. 1979) and observed variance of gene frequency in succeeding generation (Weir 1996) was estimated for the divergent lines. F-ratio was used to detect significant differences between expected and observed gene frequency variances. Bonferroni procedure of multiple test of significance was employed to controls the probability of rejecting one or more true null hypothesis (Hochberg 1988).

## Results

Selection response
Body weight of fish from the selected low line (LL) and high line (HL) from 7 to 33 weeks of age and response to selection are shown in Table 2. The number of fish per cage varied slightly during grow-out. However, the correlation between survival and growth rate was not statistically significant ( $\mathrm{P}>0.05$ ) in both LL and HL. This indicated that variation of density of fish between cages was small and did not influence growth between cages.

Table 2. Mean body weight (g), percentage coefficient of variation (\% CV) for weight, response to selection as half of the total divergence and percent response of divergent selected lines of Clarias macrocephalus in second generation of mass selection ${ }^{1}$

| Age <br> (week) | Low line |  | High Line |  | Response to selection |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
|  | Weight <br> $(\mathrm{g})$ | \% CV | Weight <br> $(\mathrm{g})$ | \% CV | Response <br> $(\mathrm{g})$ | $\%$ <br> Response |
| 7 | $2.7^{a}$ | $79.6^{a}$ | $2.8^{a}$ | $66.3^{a}$ | 0.036 | 1.30 |
| 13 | $14.0^{a}$ | $41.2^{a}$ | $14.7^{a}$ | $37.3^{a}$ | 0.351 | 2.45 |
| 18 | $31.8^{a}$ | $28.8^{a}$ | $32.2^{a}$ | $37.3^{a}$ | 0.184 | 0.57 |
| 25 | $47.3^{a}$ | $36.9^{a}$ | $49.6^{a}$ | $38.4^{a}$ | 1.123 | 2.30 |
| 29 | $79.8^{a}$ | $28.3^{a}$ | $86.7^{b}$ | $29.2^{a}$ | 3.467 | 4.16 |
| 33 | $90.7^{a}$ | $25.8^{a}$ | $97.2^{b}$ | $24.6^{a}$ | 3.190 | 3.40 |

${ }^{1}$ Mean body weight and $\% \mathrm{CV}$ of fish at the same age superscripted with the same letter in a row are not statistically different between lines.

Significant divergence ( $\mathrm{P}<0.05$ ) in weight of high and low line occurred at 29 weeks and 33 weeks of age. Weight difference between the LL and HL did not increase proportionately with age. Response to selection for weight measured as the half of the divergence between LL and HL was $3.467 \mathrm{~g}(4.16 \%)$ and $3.190 \mathrm{~g}(3.4 \%)$ at 29 and 33 weeks of age, respectively (Table 2). Response increased continuously with age except at 33 weeks of age ( $\mathrm{P}>0.05$ ). Percent coefficient of variation (\% CV) in body weight indicated that C. macrocephalus became more homogenous ( $\mathrm{P}<0.05$ ) when age increased and their weight distribution was normal.

## Genetic variation in selected lines

Allele frequencies differed substantially between generations of each selected lines (Table 3). Change in allele frequencies was observed in the low line. A few alleles with low frequencies observed in $F_{1}$ were missing in $F_{2}$ (Table 3). While a few low frequency alleles tended to increase their frequencies. Likelihood ratio $\left(G^{2}\right)$ test conformed the significance changes in allele frequencies at loci $M P I^{*}, P G M^{*}$ and $A D H^{*}$ in the low line. No significant difference ( $\mathrm{P}>0.05$ ) was observed in changes in allele frequencies between generations of high line.

Table 3. Allele frequencies of polymorphic loci in generations of selected low and high line of Clarias macrocephalus.

| Locus | Allele frequencies |  |  |  |  |
| :--- | :---: | :---: | :---: | :---: | :---: |
|  | Allele | LL (F1) | LL (F2) | HL (F1) | HL (F2) |
| ADH $^{*}$ | -100 | 0.9714 | 1 | 0.9943 | 1 |
|  | -29 | 0.0286 | 0 | 0.0057 | 0 |
|  | N | 77 | 46 | 88 | 47 |
| G3PDH* $^{*}$ | 100 | 1 | 1 | 0.9903 | 0.9565 |
|  | 63 | 0 | 0 | 0.0097 | 0.0435 |
|  | N | 77 | 42 | 103 | 46 |
| GPI* | 100 | 0.9805 | 0.96 | 0.9911 | 0.9694 |
|  | 77 | 0.0195 | 0.04 | 0.0089 | 0.0306 |
|  | N | 77 | 50 | 112 | 49 |
|  | sMDH-1* | 260 | 0.28 | 0.36 | 0.3174 |
|  |  |  |  |  | 0.39 |

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|  | 100 | 0.72 | 0.64 | 0.6826 | 0.61 |
| :--- | :---: | :---: | :---: | :---: | :---: |
|  | N | 75 | 50 | 115 | 50 |
| MPI* $^{*}$ | 100 | 0.9722 | 1 | 0.9851 | 1 |
|  | 88 | 0.0278 | 0 | 0.0149 | 0 |
|  | N | 72 | 47 | 101 | 46 |
| PGDH $^{*}$ | 100 | 0.9914 | 1 | 0.9875 | 0.9783 |
|  | 77 | 0.0086 | 0 | 0.0125 | 0.0217 |
|  | N | 58 | 48 | 80 | 46 |
| PGM $^{*}$ | 100 | 0.7297 | 0.59 | 0.6408 | 0.5638 |
|  | 57 | 0.2703 | 0.41 | 0.3592 | 0.4362 |
|  | N | 74 | 50 | 103 | 47 |
|  | 115 | 0.1364 | 0.0918 | 0.1609 | 0.1633 |
| EST* $^{*}$ | 100 | 0.8636 | 0.9082 | 0.8391 | 0.8367 |
|  | N | 77 | 49 | 115 | 49 |
|  | 100 | 0.9803 | 1 | 0.9954 | 1 |
|  | 83 | 0.0197 | 0 | 0.0046 | 0 |
|  |  | N | 76 | 50 | 108 |
|  |  |  |  | 49 |  |

Both the low and high line exhibited lower mean number of allele ( $n_{a}$ ) in $F_{2}$ generation compared to their previous generation ( $\mathrm{P}<0.05$ ). On contrary, estimated effective number of allele per locus ( $n_{e}$ ) significantly increased $(P<0.05)$ in $F_{2}$ compared to the previous generation of the high line. Such difference was not significant ( $\mathrm{P}>0.05$ ) in the low line. Significant differences ( $\mathrm{P}<0.05$ ) were also observed in $\mathrm{n}_{\mathrm{e}}$ between divergent lines of $C$. macrocephalus in $F_{2}$ generation. Observed heterozygosity was high and remained unchanged over generations ( $\mathrm{P}>0.05$, Archie 1985) in both the high and low lines (Table 4). Estimates of allele frequency variance, $\mathrm{F}_{\mathrm{K}}$, in $\mathrm{F}_{1}-\mathrm{F}_{2}$ comparison of low line range from 0.0073 to 0.0426 (Table 5) and effective population size for the low line was 28.206 . Estimates of $F_{k}$, in $F_{1}-F_{2}$ comparison of the high line ranged from 0.0002 to 0.0218 and effective population size estimated was 64.23.

Table 4. Indices of genetic variability at 14 loci in $F_{1}$ and $F_{2}$ of selected lines of Clarias macrocephalus

| Lines | $\begin{gathered} \text { Mean number } \\ \text { of } \\ \text { alleles/locus } \\ \pm S D^{\prime} \end{gathered}$ | Effective number of alleles/locus $\pm$ $S^{2}$ | Percentage of polymorphic loci (no criterion) ${ }^{3}$ | Mean heterozygosity ${ }^{4}$ |  |
| :---: | :---: | :---: | :---: | :---: | :---: |
|  |  |  |  | Direct count $\left(H_{e}\right)$ | Hardy -Weinberg expectations $\left(\mathrm{H}_{\mathrm{e}}\right)$ |
| LL-F ${ }_{1}$ | $1.47 \pm 0.51$ | $1.13 \pm 0.23$ | 57.14 | 0.0804 | 0.0881 |
| LL-F | $1.28 \pm 0.46$ | $1.15 \pm 0.33$ | 28.57 | 0.0803 | 0.0857 |
| HL-F | $1.64 \pm 0.49$ | $1.15 \pm 0.30$ | 64.28 | 0.0788 | 0.0917 |
| HL-F | $1.43 \pm 0.51$ | $1.17 \pm 0.33$ | 42.85 | 0.1003 | 0.1031 |

${ }^{1}$ Significant difference ( $\mathrm{P}<0.05$ ) between generations within lines.
${ }^{2}$ Significant difference ( $\mathrm{P}<0.05$ ) between generations within high line.
${ }^{3}$ Regardless of allelic frequencies.
${ }^{4}$ No significant difference ( $P>0.05$ ) in mean heterozygosity between generations, lines, and between expected and observed values.

Table 5. Variance of allele frequencies between two generations ( $\mathrm{F}_{1}$ and $\mathrm{F}_{2}$ ), Fk at nine polymorphic loci, averaged Fk and estimated effective population size $\left(N_{k}\right)$ of divergent lines of Clarias macrocephalus

| Locus | Fk (allele frequency variance in the generation comparison |  |
| :---: | :---: | :---: |
|  | Low line (F1-F2) | High line ( $\mathrm{F}_{1}-\mathrm{F}_{2}$ ) |
| ADH* | 0.0286 | 0.0057 |
| G3PDH* | * | 0.0218 |
| GPI* | 0.0073 | 0.0121 |
| MDH-1* | 0.0146 | 0.0115 |
| MPI* | 0.0278 | 0.0149 |
| PGDH* | 0.0086 | 0.0025 |
| PGM** | 0.0426 | 0.0123 |
| EST* | 0.0098 | 0 |
| LAP* | 0.0197 | 0.0046 |
| $\mathrm{F}_{\mathrm{K}}{ }^{1}$ | 0.0199 | 0.0095 |
| ESTIMATED $\mathrm{N}_{\mathrm{K}}$ | 28.206 | 64.238 |

[^0]The distribution of transformed allele-frequency variances in the two generations of the low and the high lines of Clarias macrocephalus were significantly different from a standard normal distribution ( $\mathrm{P}<0.01$ in the Kolmogorov-Smirnov chi-square test). These transformed variances did not form straight line when plotted against a $Y$-axis of expected standard normal variates (Figure 1).


Figure 1. Normal probability plots of transformed allele-frequency variances at 8 and 9 loci examined in the low and high line of Clarias macrocephalus, respectively.

Removal of locus specific transformed allele-frequency case by case revealed that the curvelinerity may be introduced by disparate value for $\mathrm{PGM}^{*}, \mathrm{GPI}^{*}$ and MPI* in the low line. For the high line, deviant values of transformed allele-
frequency variances were introduced by loci MDH-1*, GP1* and G3PDH*. When $\mathrm{F}_{\mathrm{K}}$ for these loci of respective lines were removed and transformed variates were recalculated, the distribution of remaining loci was normal ( $\mathrm{P}>0.05$ ) in the low line. While remaining loci in the high line, nevertheless, did not show normal distribution ( $P<0.05$ ). This indicated that more loci were affected by forces other than drift in high line which could not be detected.
Observed variance of allele frequency change was not significantly higher than the expected variance due to drift alone at loci GP1*, MDH-1*, PGM* and EST* (F ratio $=0.259$ to $0.736, \mathrm{P}>0.05$ ) (Table 6). This indicated that drift alone was responsible to cause allele frequency changes at these loci in successive generation $\left(F_{2}\right)$ of the low line.

Table 6. Expected (due to drift) and observed variance of gene frequencies in derived $\mathrm{F}_{2}$ generation of the low line of Clarias macrocephalus

| Loci | Df $\left(F_{1} / F_{2}\right)$ | Expected Variance | Observed <br> Variance | F-Ratio |
| :--- | :---: | :---: | :---: | :---: |
| GPI $^{*}$ | $76 / 49$ | 0.000339 | 0.000249 | $0.736^{\text {ns }}$ |
| MDH-1 $^{*}$ | $74 / 49$ | 0.003574 | 0.001536 | $0.4300^{\text {ns }}$ |
| PGM $^{*}$ | $73 / 49$ | 0.003496 | 0.001634 | $0.467^{\text {ns }}$ |
| EST $^{*}$ | $76 / 48$ | 0.002088 | 0.000541 | $0.259^{\text {ns }}$ |

${ }^{1}$ Expected variance was estimated based on allele frequencies of Fl generation and effective population size (28.20) used to produce F2 generation.
${ }^{n s}$ denotes no significance ( $P>0.05$ ).

The analyses of variance indicated observed variances of allele frequencies at the G3PDH* ${ }^{*}$ GPI* and PGDH* loci in the high line were significantly greater $(\mathrm{P}<0.01,<0.05)$ than the theoretical expectation of variance due to drift alone. This suggested that allele frequency change from generation to generation at these loci in the high line was resulted from forces other than drift. The F-ratio ( 1.32 to $1.46, \mathrm{P}>0.05$ ) of the observed and expected variance of allele frequencies indicated that drift was a main force operating to alter allele frequencies at PGM*, MDH-1* and EST* in the successive generation of the high line (Table 7).

Table 7. Expected (due to drift) and observed variance of gene frequencies in derived $\mathrm{F}_{2}$ generation of the high line of Clarias macrocephalus

| Loci | Df $\left(\mathrm{F}_{1} / \mathrm{F}_{2}\right)$ | Expected Variance | F-Ratio |  |
| :--- | :---: | :---: | :---: | :---: |
| G3PDH $^{*}$ | $102 / 45$ | 0.000075 | 0.000452 | $6,049^{* *}$ |
| GPI $^{*}$ | $111 / 48$ | 0.000069 | 0.000303 | $4.409^{* *}$ |
| MDH-1 $^{*}$ | $114 / 49$ | 0.001686 | 0.002379 | $1.411^{\text {ns }}$ |
| PGDH $^{*}$ | $79 / 45$ | 0.000096 | 0.000231 | $2.402^{*}$ |
| PGM $^{*}$ | $102 / 46$ | 0.001792 | 0.002616 | $1.460^{\text {ns }}$ |
| EST $^{*}$ | $114 / 48$ | 0.001051 | 0.001394 | $1.327^{\text {ns }}$ |

${ }^{1}$ Expected variance was estimated based on allele frequencies of F 1 generation and effective population size (64.23) used to produce F2 generation.

* and ${ }^{* *}$ denote significant difference ( $\mathrm{P}<0.05, \mathrm{P}<0.01$, respectively).
${ }^{\text {ns }}$ denotes no significant difference ( $P>0.05$ ).


## Discussion

Although difference between the divergent lines in this study was significant, selection response obtained was small compared to the response of $16.4 \%$ obtained by Chamnankuruvet (1996) from second generations of divergent mass selection of $C$. macrocephalus. Relatively high selection responses have also been observed in mass selection program conducted in other aquacultured fish species; per generation response of 12-18\% in channel caffish, Ictalurus punctatus (Dunham 1987) and 23\% in tilapia Oreochromis niloticus (Gjedrem 1997).

Selection response was not significant during early stages because environmental component of the total variance of weight was particularly high at the early phase of life, hence resulted in low heritability (Kirpichnikov 1981). Relatively low heritability at an early stage of life was also found in channel caffish (Reagan et al. 1976), rainbow trout (Gall and Gross 1978) and oysters (Newkirk et al. 1977). The low response obtained in this study could also have been a result of the asymmetry response of the divergent lines (Falconer 1989). It is likely that the response to divergence selection in this study was asymmetric,
probably due to difference in selection differential $(S)$ between the divergent lines ( S for LL and HL was -57.5 g and 107.5 g , respectively).
The genetic variation reported in this study was quite high as compared with other studies because high proportion of polymorphic loci were included in an attempt to verify the genetic change in the successive generation. In this study allele number per locus reduced significantly ( $\mathrm{P}<0.05$ ) in the successive generation for both selected low and high lines of Clarias macrocephalus while heterozygosity was unchanged in both lines. Loss of rare allele was observed in $F_{2}$ of both low and high divergent lines, which might be a result of combined effect of selection and genetic drift (Hallerman et al. 1986). It was also observed in this study that selection significantly affected a few loci, for example, PGM*, G3PDH* and GPI* eventhough isozyme loci were said to be neutral (Myers et al. 2001).

In the present study, two generations of divergent selection did not change mean heterozygosity. Similar trends of maintenance of higher level of heterozygosity and overall genetic variability until after two generations of selection for body weight has been reported for crossbred population of channel catfish (Hallerman et al. 1986) and for Sydney rock oysters, Saccostrea glomerata (English et al. 2000). Generally, reduction of number of alleles per locus is always accompanied by reduction of heterozygosity. The explanation for the retaining high heterozygosity while allele number reduced is that heterozygosity at a locus is affected both by the number of alleles and the evenness of allelic frequencies. Reduction in heterozygosity owing to loss of rare alleles can be compensated by an increase in the evenness of frequencies of the remaining alleles (Watterson 1984).

Estimated $N_{k}$, suggested that the effective parental contribution was quite small in the low line ( $N_{k}=28.20$ ) compared to high line ( $N_{k}=64.23$ ) although the actual number of parents used was 100 . The high $\mathrm{F}_{\mathrm{K}}$ value (0.0199) between the $F_{1}$ and $F_{2}$ generation of the low line suggested that drift occurred when selection was performed in $F_{1}$ generation which underestimated the $N_{k}$ of the $F_{2}$
generation. In the face of relatively low $F_{K}$ value ( 0.0095 ) observed between $F_{1}$ and $F_{2}$ generation and the poor evidence of drift at some of the variable loci in the high line, indicated that the estimated low $N_{k}$ value (64.23) might be the result of differences in family size. In general, the differences in apparent number of parent and estimated $N_{k}$ might be due to differential fertility or progeny viability in both low and high line (Gaffney et al. 1992). Since directional selection generates differences in the reproductive success of individuals which increase the variance in gene frequencies (Enrique and Caballero 1998). Moreover, transferred allele frequency variance revealed that genetic drift and selection were responsible to allele frequency change in both the low and high lines. The greatest opportunity for natural selection had occurred during growout period of Clarias macrocephalus, when disease incident caused more than $60 \%$ mortality. This suggests that drift variance inflated by natural selection in randomly varying environment could have lowered the effective population size ( $\mathrm{N}_{\mathrm{k}}$ ) (Mueller et al. 1985).

In a randomly breeding closed population, the array of allele and genotype frequencies would not be expected to change between generations if there were no selection or drift (Hallerman et al. 1986). In the present study, allele frequency change between $F_{1}$ and $F_{2}$ were statistically significant $\left(G^{2}<0.05\right)$ at three of nine loci. No significant allele frequency change was observed in the high line. The observed changes in allele frequencies might be attributable to (a) artificial and natural selection on the biochemical loci (b) genetic drift, or (c) sampling error which might have occurred during sampling of fish for allozyme analysis. In order to evaluate the cause of the allele frequency changes, distribution of allele-frequency variances were examined by transforming $\mathrm{F}_{\mathrm{K}}$ values to standard normal variates. These variates were not normally distributed in both of the selected lines for their respective generations comparison. This suggests that drift alone was insufficient to explain the pattern observed here.

Comparisons of observed and expected variance of gene frequencies (under drift) suggested that genetic change occurred in the low line was solely due to driff, since there were no significant differences between observed and
expected variance of gene frequencies. On the contrary, deviation from the standard normal distribution of the transformed allele-frequency variance of loci PGM* and GPI* in the low line indicated that the allele frequencies change occurred in these loci was not entirely explained by drift alone. So far, there has been no argument against the transforming $\mathrm{F}_{\mathrm{K}}$ values to standard normal variates to examine distribution of allele-frequency variances but for the observed and expected variance of gene frequencies (under drift), it is obvious that the expected variance of allele frequencies might have been inflated due to the underestimated $N_{k}$. Mueller et al. (1985) demonstrated that $N_{k}$ calculated based on variance of allele frequency change can be easily underestimated. Therefore, the explanation that the selection force (Kolmogorov - Smirnov test) might have affected these loci, seems more likely explanation for the allele frequencies change.

Although allele frequency change in the high line was not statistically significant, both expected and observed variance of allele frequency, and the distribution of transformed allele-frequency agreed that selection acted on the loci G3PDH* and GPI* while only drift was responsible for the change at loci PGM* and EST*. Similar to the case observed in the low line, deviation from the standard normal distribution of the transformed allele-frequency variance of loci sMDH-1* and PGDH* in the high line indicated that the allele frequency change occurred in these loci were not entirely explained by drift or selection. Since the method of observed and expected variance comparison seems to be less sensitive to assess the cause of allele frequency change (Mueller et al. 1985), selection acted upon the locus $\mathrm{sMDH}-1^{*}$ while drift was responsible at the locus PGDH* as indicated by standard normal variates (Kolmogorov - Smirnov test) would be more likely explanation for the allele frequencies changes. Regarding the unchanged heterozygosity in $\mathrm{F}_{2}$ of both low and high line, the simplest explanation would be selection acted to maintain heterozygosity, thereby retarding drift at allozyme loci (Voijenhoek et al. 1990).

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## Conclusion

The study suggested that the response increased perpetually with age of Clarias macrocephalus selected for body weight. The observed average response to mass selection of Clarias macrocephalus, as one half of the divergence between lines, was low in two generations. The divergent selection for body weight resulted in reduction of number alleles per locus. Allele frequency changes and fixation at some loci occurred in both selected lines of Clarias macrocephalus. Genetic drift was the main driving force for such changes in the low line, while selection acted at some of the variable loci to cause observed genetic changes in the high line.

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# Production potential of carp-SIS-prawn polyculture in Chitwan, Nepal 

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#### Abstract

Potential of carp-Small Indigenous Fish Species (SIS)-Prawn polyculture in Nepal was assessed through an experiment in farmers' ponds of $100 \mathrm{~m}^{2}$ each at Chitwan District for 250 days. There were four treatments each with three replicates. Treatments included $\mathrm{T}_{1}$ - carps only (rohu Labeo rohita, mrigal Cirrhinus mrigala, silver carp Hypophthalmichthys molitrix and bighead carp Hypophthalmichthys nobilis), $\mathrm{T}_{2}$ (carps + prawn Macrobrachium rosenbergil), $\mathrm{T}_{3}$ (carps + prawn + dedhuwa Esomus danricus), and $\mathrm{T}_{4}$ (carps + prawn + pothi Puntius sophore). The stocking density of carps, SIS, and prawn were 75 (rohu 30 , mrigal 10 , silver carp 25 and bighead carp 10), 250, and 100 per 100 $\mathrm{m}^{2}$, respectively. Fish were fed daily on dough of rice bran and soybean cake $(2: 1)$ at the rate of $3 \%$ of body weight. Adding prawn and SIS in carp ponds did not affect growth and production of carps except rohu. Mean harvest weight ( $221.3 \pm 43.2 \mathrm{~g}$ ) and growth rate ( $0.87 \pm 0.17 \mathrm{~g} /$ fish $/ \mathrm{d}$ ) of rohu was significantly higher ( $\mathrm{P}<0.05$ ) in $\mathrm{T}_{1}$ than other treatments. Adding prawn in carp ponds $\left(T_{3}\right)$ increased profit ( $\mathrm{P}<0.05$ ) over carp polyculture. Carps, SIS and prawn polyculture appears to be an appropriate fish culture system in Nepal.


Keywords: carp, SIS, prawn, polyculture, production

## Introduction

Semi-intensive carp polyculture is the major and established aquaculture system of Nepal but the system does not promote household fish consumption. Per caput fish consumption rate is 1.85 kg (Directorate of Fisheries Development 2013) which is very low in Nepal where as neighboring countries India, Bangladesh and China have $5 \mathrm{~kg}, 15 \mathrm{~kg}$ and 25 kg fish per caput consumption, respectively (Bhujel 2009). In the existing carp polyculture system, farmers often sell large carp in the market instead of consuming. Selling large fish certainly increases the household income but does not improve the nutritional status of the family. Therefore, it is essential to develop a production system that increases household fish consumption. In this regard, polyculture of SIS with carps and prawn seems to be a possible approach. Incorporating SIS and prawn in carp ponds benefits farmers in two ways (i) improves nutritional status by regular harvesting and consuming of SIS because SIS are self recruiting and are also richer in vitamins, calcium and iron compared to carps (Roos et al. 2006) and (ii) increases household income by selling valuable carps and prawn. Despite the high potential of carp-SIS-prawn polyculture no study has been done on this in Nepal to date. This experiment is therefore, to assess the potential of polyculture of carps (rohu, mrigal, silver carp and bighead carp), SIS (dedhuwa and pothi) and freshwater prawn in Chitwan, Nepal.

## Materials and methods

The experiment was conducted in 12 farmers' pond of approximately $100 \mathrm{~m}^{2}$ (66-150 m${ }^{2}$ ) at Bhandara Village Development Committee of Chitwan for 250 days. The experiment was conducted following Completely Randomized Design (CRD) and included four treatments each with three replicates. Treatments includes $T_{1}$ (carps only), $\mathrm{T}_{2}$ (carps + prawn), $\mathrm{T}_{3}$ (carps + prawn + dedhuwa), and $\mathrm{T}_{4}$ (carps + prawn + pothi).

Ponds were newly constructed and limed at the rate of $500 \mathrm{~kg} / \mathrm{ha}$ with agriculture lime $\left(\mathrm{CaCO}_{3}\right)$. After 15 days of liming, the fertilization was done with cowdung at the rate of $36,000 \mathrm{~kg} / \mathrm{ha}$. After liming and organic
fertilization, all the ponds were filled with clean and fresh water. Water depth was maintained at 1.0 meter deep. Then, the ponds were fertilized with Urea and Double Ammonium Phosphate at the rate of $470 \mathrm{~g} / 100 \mathrm{~m}^{2}$ and 350 $\mathrm{g} / 100 \mathrm{~m}^{2}$ (equivalent to $0.4 \mathrm{~g} \mathrm{~N} / \mathrm{m}^{2} /$ day and $0.1 \mathrm{~g} \mathrm{P} / \mathrm{m}^{2} /$ day), respectively (Knud-Hansen et al.1993). Ponds were stocked with fingerlings of rohu $(3.63 \pm 0.04 \mathrm{~g}$ individual weight), mrigal $(3.60 \pm 0.11 \mathrm{~g})$, silver carp ( $3.75 \pm 0.13$ $\mathrm{g})$, bighead carp $(3.62 \pm 0.06 \mathrm{~g})$, dedhuwa ( $1.35 \pm 0.18 \mathrm{~g}$ ), and pothi $(2.12 \pm 0.02 \mathrm{~g})$ and prawn juvenile $(0.62 \pm 0.05 \mathrm{~g})$ at rates of $3,000,1,000$, $1,000,2,500,25,000,25,000$ and 10,000 per hectare, respectively (Table 1). Fish and Prawn were fed with freshly made dough of rice bran and soybean oil cake ( $20 \%$ Crude protein) every morning at $2: 1$ ratio by weight. Feed was given at the rate of $3 \%$ of carp body weight. Partial harvesting of dedhuwa and pothi was done three months after stocking and continued onwards until final harvest. The harvested dedhuwa and pothi were used for household consumption. Final harvesting of all fish, including dedhuwa, pothi and prawn was done on $30^{\text {th }}$ December 2009 after complete draining of each pond. During harvest, all carps and prawn were counted and weighed specieswise separately to assess survival rate and production performance of each.

Table 1. Stocking density (number of fingerlings/juveniles per hectare) of carps, dedhuwa, pothi and prawn at different treatments

| Species | $\mathrm{T}_{1}$ | $\mathrm{~T}_{2}$ | $\mathrm{~T}_{3}$ | $\mathrm{~T}_{4}$ |
| :--- | :---: | :---: | :---: | :---: |
| Rohu | 3,000 | 3,000 | 3,000 | 3,000 |
| Mrigal | 1,000 | 1,000 | 1,000 | 1,000 |
| Bighead Carp | 1,000 | 1,000 | 1,000 | 1,000 |
| Silver Carp | 2,500 | 2,500 | 2,500 | 2,500 |
| Dedhuwa | - | - | 25,000 | - |
| Pothi | - | - | - | 25,000 |
| Prawn | - | 10,000 | 10,000 | 10,000 |
| Total | 7,500 | 17,500 | 42,500 | 42,500 |

$\mathrm{T}_{1}$ (carps), $\mathrm{T}_{2}$ (carps + prawn), $\mathrm{T}_{3}$ (carps + prawn + dedhuwa) and $\mathrm{T}_{4}$ (carps +prawn+ pothi)

Fish were sampled monthly for growth determination. At least $20 \%$ of each species were netted and weighed to determine growth. Dissolved oxygen (DO), pH , transparency and temperature were measured fortnightly at 7-9 am in situ, whereas total alkalinity, total ammonium nitrogen (TAN), soluble reactive phosphorus (SRP), and chlorophyll-a were analyzed at laboratory of the Aquaculture Department monthly. Economic analysis was done using gross margin analysis. The variable cost included cost of fingerlings, feed, lime, and chemical fertilizer based on current market price. Household labor and cattle manure was not accounted in production cost analysis. Extra time of women farmers and their farm product like cattle manure were sufficient for fish production in small scale.

Data were analyzed using one-way analysis of variance (ANOVA) using MSTAT-C (1990) Microsoft computer program. Mean differences were compared using the least significant difference (LSD) after ANOVA. Significant differences were tested at $5 \%(\mathrm{P}<0.05)$. Means were given with standard error (Mean $\pm$ S.D.).

## Results

## Water quality analysis

All analysed water quality parameters did not fluctuate drastically and were in safe range during the entire experimental period (Table 2).

Table 2. Mean and range of water quality parameters in different treatments during experimental period (Mean $\pm$ S.D.)

| Parameters | Treatment |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
|  | $\mathrm{T}_{1}$ | $\mathrm{~T}_{2}$ | $\mathrm{~T}_{3}$ | $\mathrm{~T}_{4}$ |
| Temperature $\left({ }^{\circ} \mathrm{C}\right)$ | $25.4 \pm 0.8$ | $24.9 \pm 0.7$ | $25.2 \pm 0.5$ | $25.1 \pm 0.7$ |
|  | $(15.5-32.3)$ | $(14.1-31.6)$ | $(15.0-32.0)$ | $(15.0-32.2)$ |
| Dissolved Oxygen | $7.3 \pm 0.9$ | $6.7 \pm 1.3$ | $6.5 \pm 1.2$ | $6.3 \pm 1.4$ |
| $(\mathrm{mg} / \mathrm{L})$ | $(3.8-10.2)$ | $(2.7-9.9)$ | $(1.6-10.1$ | $(2.1-10.0)$ |
| pH | 7.4 | 7.6 | 7.5 | 7.8 |
|  | $(5.8-9.1)$ | $(6.0-9.2)$ | $(5.9-9.2)$ | $(6.5-9.2)$ |
| Secchi disk depth $(\mathrm{cm})$ | $27.9 \pm 5.4$ | $27.3 \pm 4.2$ | $27.3 \pm 4.9$ | $28.5 \pm 5.4$ |
|  | $(12.0-45.0)$ | $(13.0-43.0)$ | $(13.0-48.5)$ | $(12.0-47.0)$ |
| Total alkalinity $(\mathrm{mg} / \mathrm{L}$ | $104.7 \pm 15.1$ | $107.6 \pm 15.0$ | $106.8 \pm 14.0$ | $106.5 \pm 12.6$ |
| Ca CO $\left._{3}\right)$ | $(73.9-135.9)$ | $(75.3-142.2)$ | $(78.5-135.6)$ | $(76.6-116.9)$ |
| Chlorophyll- $a\left(\mathrm{mg} / \mathrm{m}^{3}\right)$ | $13.7 \pm 5.5$ | $13.2 \pm 3.6$ | $16.5 \pm 3.8$ | $13.3 \pm 3.6$ |
|  | $(2.7-34.8)$ | $(3.3-30.3)$ | $(4.1-32.9)$ | $(6.1-25.9)$ |
| Total ammonium | $0.047 \pm 0.017$ | $0.028 \pm 0.019$ | $0.033 \pm 0.014$ | $0.030 \pm 0.018$ |
| nitrogen $(\mathrm{mg} / \mathrm{L})$ | $(0.004-0.099)$ | $(0.005-0.053)$ | $(0.007-0.053)$ | $(0.002-0.057)$ |
| Soluble reactive | $0.030 \pm 0.016$ | $0.025 \pm 0.014$ | $0.029 \pm 0.017$ | $0.020 \pm 0.010$ |
| phosphorus $(\mathrm{mg} / \mathrm{L})$ | $(0.001-0.063)$ | $(0.002-0.063)$ | $(0.004-0.112)$ | $(0.001-0.047)$ |

(Figures in the parenthesis are range value)
$\mathrm{T}_{1}$ (carps), $\mathrm{T}_{2}$ (carps + prawn), $\mathrm{T}_{3}$ (carps + prawn + dedhuwa) and $\mathrm{T}_{4}$ (carps +prawn+ pothi)

## Growth and production of carps, SIS and prawn

Species-wise and combined production did not differ significantly ( $\mathrm{P}>0.05$ ) among treatments (Table 3). Final mean weight and daily weight gain of rohu was significantly higher ( $P<0.05$ ) in $T_{1}$ than $T_{2}, T_{3}$ and $T_{4}$ while it was not significantly different $(P>0.05)$ among $T_{2}, T_{3}$ and $T_{4}$. The survival was significantly higher ( $P<0.05$ ) in $T_{3}$ than $T_{2}, T_{1}$ and $T_{4}$ while it was not significantly different ( $P>0.05$ ) among $T_{1}, T_{2}$ and $T_{4}$. Survival of mrigal and bighead carp was significantly higher ( $P<0.05$ ) in $T_{4}$ than $T_{1}, T_{2}$ and $T_{3}$. Survival of silver carp was significantly higher ( $P<0.05$ ) in $T_{2}$ than $T_{1}, T_{3}$ and $T_{4}$ but $T_{1}, T_{3}$ and $T_{4}$ were statically similar.

Table 3. Growth performance of carps, SIS and prawn in different treatments (Mean $\pm$ S.D.)

| Parameters | Treatments |  |  |  |
| :---: | :---: | :---: | :---: | :---: |
|  | $\mathrm{T}_{1}$ | $\mathrm{T}_{2}$ | $\mathrm{T}_{3}$ | $\mathrm{T}_{4}$ |
| Rohu |  |  |  |  |
| Initial mean weight ( $\mathrm{g} / \mathrm{fish}$ ) | $3.7 \pm 0.2$ | $3.3 \pm 0.1$ | $3.6 \pm 0.2$ | $3.8 \pm 0.2$ |
| Initial total weight (kg/100 m${ }^{2}$ ) | $0.11 \pm 0.0$ | $0.10 \pm 0.03$ | $0.11 \pm 0.01$ | $0.12 \pm 0.04$ |
| Final mean weight ( $\mathrm{g} /$ fish) | $221.3 \pm 43.2^{\text {a }}$ | $180.1 \pm 17.7^{\text {b }}$ | $188.1 \pm 4.6^{\text {b }}$ | $183.7 \pm 5.4^{\text {b }}$ |
| Final total weight (kg/100 m${ }^{2}$ ) | $4.67 \pm 1.25$ | $3.96 \pm 0.36$ | $5.39 \pm 1.35$ | $4.24 \pm 0.95$ |
| Survival (\%) | $70.8 \pm 12.9{ }^{\text {b }}$ | $73.8 \pm 1.9^{\text {b }}$ | $95.6 \pm 1.9^{\text {a }}$ | $76.9 \pm 6.7^{\text {b }}$ |
| Daily weight gain ( $\mathrm{g} / \mathrm{f} / \mathrm{d}$ ) | $0.87 \pm 0.17^{\text {a }}$ | $0.71 \pm 0.07^{\text {b }}$ | $0.74 \pm 0.06^{\text {b }}$ | $0.72 \pm 0.02^{\text {b }}$ |
| Total weight gain (kg/100 m${ }^{2}$ ) | $4.57 \pm 1.25$ | $3.85 \pm 0.36$ | $5.28 \pm 1.35$ | $4.12 \pm 0.95$ |
| Mrigal |  |  |  |  |
| Initial mean weight ( $\mathrm{g} /$ fish) | $3.7 \pm 0.2$ | $3.3 \pm 0.1$ | $3.6 \pm 0.2$ | $3.9 \pm 0.2$ |
| Initial total weight (kg/100 m${ }^{2}$ ) | $0.04 \pm 0.01$ | $0.03 \pm 0.01$ | $0.04 \pm 0.01$ | $0.04 \pm 0.02$ |
| Final mean weight ( $\mathrm{g} /$ fish) | $197.3 \pm 7.8$ | $183.7 \pm 3.7$ | $182.7 \pm 2.9$ | $168.8 \pm 4.4$ |
| Final total weight (kg/100 m${ }^{2}$ ) | $1.65 \pm 0.04$ | $1.62 \pm 0.05$ | $1.59 \pm 0.05$ | $1.53 \pm 0.08$ |
| Survival (\%) | $83.8 \pm 1.6^{\text {c }}$ | $88.3 \pm 1.1^{\text {ab }}$ | $86.7 \pm 1.1^{\text {b }}$ | $90.9 \pm 1.6^{\text {a }}$ |
| Daily weight gain ( $\mathrm{g} / \mathrm{f} / \mathrm{d}$ ) | $0.77 \pm 0.05$ | $0.77 \pm 0.05$ | $0.77 \pm 0.05$ | $0.77 \pm 0.05$ |
| Total weight gain (kg/100 m${ }^{2}$ ) | $1.57 \pm 0.34$ | $1.58 \pm 0.12$ | $1.54 \pm 0.18$ | $1.49 \pm 0.44$ |

## Silver carp

| Initial mean weight ( $\mathrm{g} /$ /fish) | $3.8 \pm 0.1$ | $3.8 \pm 0.1$ | $3.8 \pm 0.2$ | $3.7 \pm 0.1$ |
| :---: | :---: | :---: | :---: | :---: |
| Initial total weight (kg/100 m${ }^{2}$ ) | $0.09 \pm 0.01$ | 0.09 $\pm 0.02$ | $0.09 \pm 0.01$ | $0.09 \pm 0.05$ |
| Final mean weight ( $\mathrm{g} /$ fish) | $391.4 \pm 16.2$ | $382.8 \pm 14.4$ | $411.5 \pm 163.8$ | $367.1 \pm 6.9$ |
| Final total weight (kg/100 m${ }^{2}$ ) | $7.40 \pm 0.25$ | $8.11 \pm 0.05$ | $7.74 \pm 1.05$ | $7.23 \pm 0.94$ |
| Survival (\%) | $75.6 \pm 1.6^{\text {b }}$ | $84.8 \pm 0.8^{\text {a }}$ | $76.2 \pm 10.8^{6}$ | $78.8 \pm 3.4^{\text {b }}$ |
| Daily weight gain ( $\mathrm{g} / \mathrm{f} / \mathrm{d}$ ) | $1.55 \pm 0.54$ | $1.51 \pm 0.34$ | $1.65 \pm 0.65$ | $1.46 \pm 0.03$ |
| Total weight gain (kg/100 m²) | $7.31 \pm 3.04$ | $8.02 \pm 0.70$ | $7.75 \pm 3.21$ | $7.14 \pm 0.83$ |

Bighead carp

| Initial mean <br> weight (g/fish) | $3.7 \pm 0.1$ | $3.6 \pm 0.1$ | $3.6 \pm 0.2$ | $3.7 \pm 0.1$ |
| :--- | :--- | :--- | :--- | :---: |
| Initial total weight <br> (kg/ $100 \mathrm{~m}^{2}$ ) | $0.04 \pm 0.01$ | $0.04 \pm 0.01$ | $0.04 \pm 0.00$ | $0.04 \pm 0.01$ |
| Final mean <br> weight (g/fish) | $302.6 \pm 0.3$ | $307.6 \pm 1.2$ | $297.7 \pm 2.8$ | $298.1 \pm 12.0$ |
| Final total weight <br> (kg/ $100 \mathrm{~m}^{2}$ ) | $2.16 \pm 0.08$ | $2.27 \pm 0.05$ | $2.37 \pm 0.07$ | $2.73 \pm 0.02$ |
| Survival (\%) | $71.4 \pm 1.4^{\mathrm{c}}$ | $73.9 \pm 1.2^{\mathrm{c}}$ | $80.0 \pm 1.0^{\mathrm{b}}$ | $91.7 \pm 2.9^{\mathrm{a}}$ |
| Daily weight gain <br> (g/f/d) | $1.19 \pm 0.30$ | $1.22 \pm 0.12$ | $1.17 \pm 0.31$ | $1.18 \pm 0.16$ |
| Total weight gain <br> $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right.$ ) | $2.12 \pm 0.31$ | $2.23 \pm 0.44$ | $2.34 \pm 0.92$ | $2.69 \pm 0.36$ |

## Prawn

| Initial mean <br> weight(g/ Prawn) <br> Initial total weight | - | $0.61 \pm 0.07$ | $0.59 \pm 0.16$ | $0.67 \pm 0.02$ |
| :--- | :--- | :--- | :--- | :--- |
| $\left(\mathrm{~kg} / 100 \mathrm{~m}^{2}\right)$ |  |  |  |  |



Mean values with same superscript in the same row are not significantly different at $\mathrm{P}_{0.05}$.
$\mathrm{T}_{1}$ (carps), $\mathrm{T}_{2}$ (carps + prawn), $\mathrm{T}_{3}$ (carps + prawn + dedhuwa) and $\mathrm{T}_{4}$ (carps +prawn+ pothi)

## Economic analysis

Result showed that the variable cost involved in fish production were not significantly different ( $P>0.05$, Table 4) among treatments. However, gross return was significantly higher ( $P<0.05$ ) in $T_{3}$ than $T_{4}$ and $T_{1}$ but was not significantly different from that of $\mathrm{T}_{2}$. Gross margin was significantly highest $(P<0.05)$ in $T_{3}$ and lowest $(P>0.05)$ in $T_{1}$ where as $T_{2}$ and $T_{4}$ were statistically similar ( $\mathrm{P}>0.05$ ).

Table 4. Economic analysis of different treatments based on about $100 \mathrm{~m}^{2}$ pond in Nepalese currency (NRs.) during experimental period

|  | Treatments |  |  |  |
| :--- | :---: | :---: | :---: | :---: |
| Variables | $\mathrm{T}_{1}$ | $\mathrm{~T}_{2}$ | $\mathrm{~T}_{3}$ | $\mathrm{~T}_{4}$ |
| Gross Return |  |  |  |  |
| Carps | $3087 \pm 116^{\mathrm{ab}}$ | $3138 \pm 165^{\mathrm{ab}}$ | $3378 \pm 224^{\mathrm{a}}$ | $2856 \pm 306^{\mathrm{b}}$ |
| Prawn | - | $815 \pm 815$ | $826 \pm 628$ | $722 \pm 287$ |
| Dedhuwa | - | - | $289 \pm 269$ | - |
| Pothi | - | - | - | $310 \pm 125$ |
| Total Gross Return | $3087 \pm 116^{\mathrm{c}}$ | $3953 \pm 365^{\mathrm{ab}}$ | $4493 \pm 415^{\mathrm{a}}$ | $3888 \pm 182^{\mathrm{b}}$ |
| Variable Cost |  |  |  |  |
| Lime | 75 | 75 | 75 | 75 |
| Urea | 145 | 126 | 145 | 145 |
| DAP | 208 | 182 | 208 | 208 |
| Carps fingerlings | 75 | 75 | 75 | 75 |
| SIS | - | - | 20 | 25 |
| Prawn | - | 175 | 175 | 175 |
| Feed | $1147 \pm 151$ | $1334 \pm 478$ | $1127 \pm 278$ | $1047 \pm 142$ |
| Total Variable Cost | $1650 \pm 151$ | $1967 \pm 161$ | $1825 \pm 276$ | $1750 \pm 142$ |
| Gross Margin | $1437 \pm 241^{c}$ | $1986 \pm 205^{b}$ | $2668 \pm 369^{a}$ | $2138 \pm 111^{b}$ |

Mean values with same superscript in the same row are not significantly different at $P_{0.05}$.
$T_{1}$ (carps), $T_{2}$ (carps + prawn), $T_{3}$ (carps + prawn + dedhuwa) and $T_{4}$ (carps +prawn+ pothi)

## Discussion

All the water quality parameters remained within a suitable range for fish culture. Daily weight gain and mean harvest weight of rohu was found significantly higher $(\mathrm{P}<0.05)$ in $\mathrm{T}_{1}$ than other treatments. This might be due to niche overlapping in terms of space and food with prawn and SIS (dedhuwa and pothi). Dedhuwa is a column feeder with food items like plankton, protozoa, detritus whereas pothi is a bottom feeder and omnivorous feeding on phytoplankton primarily (Wahab and Kadir 2009). Similarly, both juvenile and adult prawns are omnivorous and feed frequently and voraciously on a wide variety of food items (Sandifer and Smith 1985). Rohu is a bottom and column feeder, prefers to feed on plant matter including decaying vegetation (Woynarovich 1975; Pillay 1999) and less adapted to take zooplankton than mrigal (Jhingran 1991). Kadir et al. (2007) also reported that SIS (pothi) affected rohu and did not affect silver carp. According to Kohinoor et al. (1998) SIS (mara and pothi) exerts a negative impact on growth and production of carps (rohu). The survival of rohu, mrigal and silver carp differed significantly ( $\mathrm{P}<0.05$ ) among treatments, which was perhaps due to impact of poisoning in source water. People use Thiodan, the pesticide, to kill fish in the nearby river. The poisoned water entered into the fish ponds through canal and considerable fish mortality took place (Rai et al. 2012).

Net fish yield of carps was not significantly different ( $\mathrm{P}>0.05$ ) among treatments indicating SIS did not have negative effect on carps' production. This can be attributed to partial harvesting system of SIS which thinned down the population of SIS and maintained appropriate density in ponds. Moreover, prawn and SIS might be compatible to carps as suggested by Islam et al. (1999), Wahab et al. (2003), Alim et al. (2005) and Kadir et al. (2006). The net carp yield in the present experiment was lower than reported by Azim et al. (2001), Pandey (2002), Bhakta et al. (2004), Sahu et al. (2007) and average production of Nepal (DOFD 2008). Lower net yield of carps might be due to low survival, insufficient species of carp to utilize all available niches and less natural food availability due to clay turbidity. In the present study, prawn and SIS
contributed $7 \%$ and $12 \%$ on total production. Roos (2001) also reported similar contribution ( $10 \%$ ) of SIS on total fish production.

Gross margin analysis showed that all treatments were profitable. However, gross profit margin was found highest in treatment with carp, prawn and SIS which was due to extra revenue earned by farmers from selling prawn. Prawn has high market value and fetch three times higher price than that of carps. Prawn were sold on NRs. $600 / \mathrm{kg}$ while carps on NRs. $200 / \mathrm{kg}$. Thus, the results of the present study clearly showed that the fish production and profit could be increased further by adding prawn and SIS in the polyculture of carps. Prawn increased profit because it has high market price, whereas SIS (dedhuwa and pothi) increased production through its self recruiting behaviour. Since SIS are nutrient rich fish, increased consumption by farmers improves family health and nutrition.

Carp-SIS-prawn polyculture technology was able to increase household fish production, consumption and income. Therefore, this technology can be an appropriate option for small scale farmers to improve livelihood and nutrition but attention is needed on a sustainable seed supply of prawn.

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Effect of dedhuwa (Esomus danricus), mara (Amblypharyngodon mola) and pothi (Puntius sophore) on carp-prawn production in Chitwan, Nepal

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#### Abstract

In order to assess the effect of adding small indigenous fish species (SIS) dedhuwa (Esomus danricus Hamilton-Buchanan), mara (Amblyparyngodon mola Hamilton-Buchanan) and pothi (Puntius sophore Hamilton-Buchanan) on carp and freshwater prawn (Macrobrachium rosenbergii) production, an experiment was carried out in farmers' pond in Chitwan. The experiment included four treatments with 3 replications of each treatments: $\mathrm{T}_{1}$, (carp: silver carp Hypophthalmichthys molitrix, bighead carp Aristichthys nobilis, rohu Labeo rohita and mrigal Cirrhinus mrigala + prawn), $\mathrm{T}_{2}$ (carp + prawn + dedhuwa), $\mathrm{T}_{3}$ (carp + prawn + mara), and $\mathrm{T}_{4}$ (carp + prawn + pothi). The stocking densities of carp, prawn and SIS were 100 (silver carp 40, bighead carp 15 , rohu 25 , and mrigal 20), 50 , and 300 per $100 \mathrm{~m}^{2}$, respectively. Fish were fed on dough of rice bran and mustard oil cake (1:1) at the rate of $3 \%$ of body weight. Results showed that dedhuwa, mara and pothi did not differ significantly ( $\mathrm{P}>0.05$ ) in terms of production. Production of silver carp and bighead carp was found significantly higher ( $\mathrm{P}<0.05$ ) in $\mathrm{T}_{3}$ and $\mathrm{T}_{4}$ than control indicating no niche overlapping of silver carp and bighead carp with mara and pothi. Based on total production and profit carp + prawn + pothi appeared to be the best.


Keywords: dedhuwa, pothi, mara, prawn, carp polyculture

## Introduction

Small indigenous fish species (SIS) are highly valuable source of macro and micronutrients that play an important role to provide essential nutrients for the people. Vitamins and minerals are found to be much more in small fish than in large fish such as carp. Dedhuwa, mara and pothi are rich in iron, vitamin-A and calcium, respectively. Dedhuwa contains 12 mg iron per 100 gram raw, cleaned parts of the fish which is three times higher than in silver carp. Similarly, vitamin A content in mara is 2,680 Retinol Activity Equivalents (RAE) in 100 g raw, cleaned parts of the fish which is 90 times higher than in silver carp whereas pothi contains 784 mg calcium in 100 g raw cleaned parts while silver carp contains 36 mg calcium (Roos at el. 2006). The bones of SIS are very rich in calcium. Likewise, the eyes, head and viscera of mola are rich in vitamin A. Since SIS are eaten whole, without loss of nutrients from cleaning or as plate waste, their contribution on micro-nutrients intake is higher than large carp. These nutrients are found to be highly bioavailable in SIS. Studies showed that a small production of mola in household ponds in Bangladesh and a daily meal with trey changwa plieng (Esomus longimanus Hamilton-Buchanan) in Cambodia can meet the annual vitamin-A recommendation of 2 million Bangladeshi children and $45 \%$ of the daily median iron requirement of Cambodian women (Roos et al. 2007), respectively. Moreover, SIS are self recruiting fish and are therefore, can be harvested biweekly, monthly, favoring household consumption.

Preliminary research on polyculture of carp with dedhuwa, pothi and prawn in Chitwan showed that there is a potential of such polyculture system in terai (Yadav 2011). Dedhuwa and pothi in carp ponds increased nutrients intake and income generation among farmers. Therefore, there is a need of studies with other high nutrient containing fish such as mara in polyculture. The present experiment was therefore, carried out to assess the effect of dedhuwa, mara and pothi on carp and prawn production in carp polyculture ponds in Chitwan, Nepal.

## Materials and Methods

The experiment was conducted for 270 days at Majhui, Khaireni village development committee-3 of Chitwan. The experiment was conducted in 12 newly constructed ponds of approximately $100 \mathrm{~m}^{2}\left(75 \mathrm{~m}^{2}-133 \mathrm{~m}^{2}\right)$. The experiment was conducted in completely randomized design (CRD). There were four treatments with three replicates of each. Treatments included $\mathrm{T}_{1}$ (carp + prawn), $\mathrm{T}_{2}$ (carp + prawn + dedhuwa), $\mathrm{T}_{3}$ (carp + prawn + mara), and $\mathrm{T}_{4}$ (carp + prawn + pothi). The stocking densities of carp, prawn, and dedhuwa, mara and pothi were 100 (silver carp 40 , bighead carp 15 , rohu 25 , and mrigal $20), 50$, and 300 per $100 \mathrm{~m}^{2}$, respectively. The mean stocking size of silver carp, bighead carp, rohu, mrigal, prawn, dedhuwa, mara and pothi were $2.72 \pm 0.25 \mathrm{~g}, 7.15 \pm 2.04 \mathrm{~g}, 34.84 \pm 2.11 \mathrm{~g}, 6.10 \pm 0.87 \mathrm{~g}, 0.21 \pm 0.01 \mathrm{~g}$, $0.99 \pm 0.01 \mathrm{~g}, 1.39 \pm 0.01 \mathrm{~g}$, and $1.94 \pm 0.04 \mathrm{~g}$, respectively. Water fertility was maintained by applying cow dung, urea and DAP regularly at the rate of 0.4 g $\mathrm{N} / \mathrm{m}^{2} /$ day and $0.1 \mathrm{~g} \mathrm{P} / \mathrm{m}^{2} /$ day (Knud-Hansen et. al.1993). Fish were fed every morning on dough of rice bran and mustard oil cake (1:1) at the rate of $3 \%$ of body weight.

At least $20 \%$ fishes were nefted monthly for growth check sampling. Composite water samples representing the entire pond water column were taken for analysis. Water quality parameters for $\mathrm{DO}, \mathrm{pH}$, temperature, transparency and water depth were measured weekly at $7.00-9.00$ am in situ and water quality parameters for total alkalinity, soluble reactive phosphorous, total ammonium nitrogen and chlorophyll-a were analyzed monthly at Aquaculture Department Laboratory, Rampur. Experimental data were analyzed by one-way ANOVA using SPSS 16.0. Mean differences were compared using DMRT after ANOVA. Differences were considered significant at an alpha level of 0.05 ( $p<0.05$ ). All means were given with $\pm 1$ standard error (S.E.). Dedhuwa, mara and pothi were regularly harvested after three months of stocking because these fish are self recruiting species. Harvesting of these species improves the household consumption and maintains the fish population at desirable level. Final harvesting of all carp, prawn, and dedhuwa, mara and pothi was done on $15^{\text {th }}$ December 2010 after complete draining of each pond. During harvest, all
fishes were counted and weighed separately to assess survival rate and production.

## Results

Water quality parameters did not vary among treatments ( $\mathrm{P}>0.05$, Table 1 ).

Table 1. Summary of water quality parameters in different treatments during experimental period (Mean $\pm$ S.E.)

| Parameters | Treatment |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
|  | $\mathrm{T}_{1}$ | $\mathrm{~T}_{2}$ | $\mathrm{~T}_{3}$ | $\mathrm{~T}_{4}$ |
| Temperature $\left({ }^{\circ} \mathrm{C}\right)$ | $28.8 \pm 0.2$ | $28.8 \pm 0.5$ | $28.9 \pm 0.4$ | $29.2 \pm 0.1$ |
|  | $(23.1-32.0)$ | $(22.8-31.7)$ | $(23.5-32.4)$ | $(24.0-32.2)$ |
| Dissolved Oxygen | $6.9 \pm 0.2$ | $6.1 \pm 0.7$ | $6.1 \pm 0.7$ | $7.0 \pm 0.1$ |
| $(\mathrm{mg} / \mathrm{L})$ | $(4.1-8.8)$ | $(3.9-8.0)$ | $(3.0-8.4)$ | $(4.4-9.3)$ |
| pH | $8.2 \pm 0.0$ | $8.2 \pm 0.2$ | $8.2 \pm 0.2$ | $8.2 \pm 0.2$ |
|  | $(7.9-8.7)$ | $(7.9-8.8)$ | $(7.8-8.9)$ | $(7.7-8.7)$ |
| Secchi disk depth | $26.2 \pm 0.5$ | $24.1 \pm 1.4$ | $28.6 \pm 0.7$ | $26.6 \pm 0.4$ |
| (cm) | $(17.7-33.3)$ | $(20.7-26.7$ | $(22.3-29)$ | $(19.3-26.3)$ |
| Total alkalinity | $108.6 \pm 5.6$ | $106.4 \pm 2.7$ | $107.7 \pm 5.6$ | $112.7 \pm 4.8$ |
| (mg $/ \mathrm{L}$ Ca $\left.\mathrm{CO}_{3}\right)$ | $(89.0-122.3)$ | $(86.4-125.6)$ | $(90.3-121.4)$ | $(88.9-121.9)$ |
| Chlorophyll-a | $18.2 \pm 1.1$ | $15.1 \pm 3.2$ | $18.5 \pm 1.8$ | $18.8 \pm 1.9$ |
| (mg $\left./ \mathrm{m}^{3}\right)$ | $(7.6-27.3)$ | $(7.1-28.9)$ | $(7.6-31.9)$ | $(6.7-29.3)$ |
| Total ammonium | $0.041 \pm 0.016$ | $0.041 \pm 0.002$ | $0.048 \pm 0.003$ | $0.042 \pm 0.025$ |
| nitrogen $(\mathrm{mg} / \mathrm{L})$ | $(0.019-0.070)$ | $(0.019-0.075)$ | $(0.024-0.079)$ | $(0.029-0.067)$ |
| Soluble reactive | $0.027 \pm 0.006$ | $0.025 \pm 0.007$ | $0.027 \pm 0.006$ | $0.028 \pm 0.004$ |
| phosphorus $(\mathrm{mg} / \mathrm{L})$ | $(0.017-0.036)$ | $(0.001-0.053)$ | $(0.015-0.084)$ | $(0.009-0.093)$ |

Mean values with different superscript letters in the same row are significantly different ( $\mathrm{P}<0.05$ ).

Final total weight and total weight gain of silver carp was significantly higher (Table 2) in $T_{3}$ and $T_{4}$ than $T_{1}$ but was not significantly different than $T_{2}(P>0.05)$. Final mean weight of bighead carp was significantly higher ( $\mathrm{P}<0.05$ ) in $\mathrm{T}_{4}$ than $T_{2}$ but was not significantly different $(P>0.05)$ than $T_{1}$ and $T_{3}$. Total weight gain
of bighead carp was significantly higher ( $\mathrm{P}<0.05$ ) in $\mathrm{T}_{3}$ and $\mathrm{T}_{4}$ than $\mathrm{T}_{2}$. Final mean weight, survival, daily weight gain and total weight gain of rohu and mrigal did not differ significantly ( $\mathrm{P}>0.05$ ) among treatments. Total weight gain of dedhuwa, mara and pothi were $2.56,0.98,4.04 \mathrm{~kg} / 100 \mathrm{~m}^{2}$ in $\mathrm{T}_{2}, \mathrm{~T}_{3}$ and $\mathrm{T}_{4}$, respectively (Table 3). Total weight gain by prawn was less than 0.56 $\mathrm{kg} / 100 \mathrm{~m}^{2}$ in all treatments. Net yield of carp and combined total net fish yield were significantly higher ( $\mathrm{P}<0.05$ ) in $\mathrm{T}_{4}$ than $\mathrm{T}_{1}$ (Table 4).

Table 2. Growth performance of carp in different treatments (Mean $\pm$ S.E.)

| Parameters | Treatments |  |  |  |
| :---: | :---: | :---: | :---: | :---: |
|  | $\mathrm{T}_{1}$ | $\mathrm{T}_{2}$ | $\mathrm{T}_{3}$ | $\mathrm{T}_{4}$ |
| Silver carp |  |  |  |  |
| Initial mean weight (g/fish) | $2.3 \pm 0.2$ | $2.7 \pm 0.1$ | $2.5 \pm 0.0$ | $2.6 \pm 0.0$ |
| Initial total weight ( $\mathrm{kg} / 100 \mathrm{~m}$ ) | 0.09 $\pm 0.00$ | $0.10 \pm 0.00$ | $0.10 \pm 0.00$ | $0.10 \pm 0.00$ |
| Final mean weight (g/fish) | $279.0 \pm 92.4$ | $266.9 \pm 14.1$ | $311.1 \pm 4.5$ | $309.1 \pm 16.6$ |
| Final total weight ( $\mathrm{kg} / 100 \mathrm{~m}^{2}$ ) | $7.54 \pm 0.72^{\text {b }}$ | $9.15 \pm 0.03^{\text {ab }}$ | $11.15 \pm 0.28{ }^{\text {a }}$ | $11.28 \pm 0.82^{\text {a }}$ |
| Survival (\%) | $67.5 \pm 14.7$ | $91.2 \pm 1.9$ | $85.7 \pm 3.9$ | $89.6 \pm 2.5$ |
| Daily fish weight gain (g/fish/d) | $1.02 \pm 0.34$ | $0.97 \pm 0.04$ | $1.14 \pm 0.01$ | $1.13 \pm 0.06$ |
| Total weight gain (kg/100 m²) | $7.54 \pm 0.73^{6}$ | $9.05 \pm 0.03{ }^{\text {ab }}$ | $11.05 \pm 0.28{ }^{\text {a }}$ | $11.18 \pm .826^{\circ}$ |
| Bighead carp |  |  |  |  |
| Initial mean weight (g/fish) | $4.64 \pm 0.9$ | $3.4 \pm 0.4$ | $3.1 \pm 0.3$ | $4.0 \pm 0.3$ |
| Initial total weight ( $\mathrm{kg} / 100 \mathrm{~m}$ ) | $0.11 \pm 0.25$ | $0.08 \pm 0.01$ | $0.07 \pm 0.01$ | $0.09 \pm 0.01$ |
| Final mean weight (g/fish) | $238.7 \pm 25.3^{\text {ab }}$ | $189.2 \pm 22.3^{\text {b }}$ | $289.0 \pm 14.4{ }^{\text {ab }}$ | $326.6 \pm 67.6^{\circ}$ |
| Final total weight ( $\mathrm{kg} / 100 \mathrm{~m}^{2}$ ) | $3.26 \pm 0.49^{\text {ab }}$ | $2.41 \pm 0.27^{\text {b }}$ | $3.93 \pm 0.26^{\circ}$ | $4.33 \pm 0.53^{\text {a }}$ |
| Survival (\%) | $91.0 \pm 5.7$ | $88.4 \pm 8.6$ | $84.9 \pm 8.6$ | 90.6 $\pm 2.3$ |
| Daily fish weight gain (g/fish/d) | $0.86 \pm 0.09$ | $0.68 \pm 0.08$ | $1.06 \pm 0.05$ | $1.19 \pm 0.24$ |
| Total weight gain (kg/100 m²) | $3.09 \pm 0.488^{\text {ab }}$ | $2.32 \pm 0.27^{\text {b }}$ | $3.86 \pm 0.26^{\circ}$ | $4.24 \pm 0.53^{\text {a }}$ |
| Rohu |  |  |  |  |
| Initial mean weight (g/fish) | $40.4 \pm 3.7$ | $36.5 \pm 2.4$ | $28.5 \pm 3.8$ | $41.3 \pm 5.5$ |
| Initial total weight ( $\mathrm{kg} / 100 \mathrm{~m}$ ) | $0.60 \pm 0.05$ | $0.56 \pm 0.02$ | $0.42 \pm 0.05$ | $0.62 \pm 0.08$ |
| Final mean weight (g/fish) | $346.3 \pm 45.2$ | $347.6 \pm 28.7$ | $361.4 \pm 48.8$ | $371.4 \pm 8.0$ |


| Final total weight (kg/100 m²) | $7.73 \pm 0.98$ | $7.78 \pm 0.79$ | $7.11 \pm 0.78$ | $8.16 \pm 0.52$ |
| :---: | :---: | :---: | :---: | :---: |
| Survival (\%) | $89.3 \pm 3.3$ | $92.4 \pm 0.1$ | $87.9 \pm 3.4$ | $78.7 \pm 8.9$ |
| Daily fish weight gain (g/fish/d) | $1.13 \pm 0.17$ | $1.15 \pm 0.09$ | $1.23 \pm 0.16$ | $1.22 \pm 0.02$ |
| Total weight gain (kg/100 m²) | $7.12 \pm 1.02$ | $7.32 \pm 0.76$ | $6.69 \pm 0.75$ | $7.54 \pm 0.44$ |
| Mrigal |  |  |  |  |
| Initial mean weight (g/fish) | $4.1 \pm 0.4$ | $7.5 \pm 3.8$ | $4.3 \pm 0.9$ | $6.6 \pm 2.3$ |
| Initial total weight ( $\mathrm{kg} / 100 \mathrm{~m}$ ) | $0.08 \pm 0.00$ | $0.15 \pm 0.07$ | $0.08 \pm 0.00$ | $0.13 \pm 0.05$ |
| Final mean weight (g/fish) | $350.3 \pm 104.3$ | $340.7 \pm 36.5$ | $352.1 \pm 75.0$ | $437.4 \pm 49.2$ |
| Final total weight ( $\mathrm{kg} / 100 \mathrm{~m}^{2}$ ) | $4.70 \pm 0.54$ | $6.47 \pm 0.52$ | $6.39 \pm 1.44$ | $7.78 \pm 1.30$ |
| Survival (\%) | $67.1 \pm 16.5$ | $88.9 \pm 4.1$ | $95.0 \pm 3.3$ | $90.7 \pm 4.8$ |
| Daily fish weight gain (g/fish/d) | $1.28 \pm 0.38$ | $1.23 \pm 0.14$ | $1.29 \pm 0.27$ | $1.59 \pm 0.17$ |
| Total weight gain (kg/100 m²) | $4.62 \pm 0.54$ | $6.33 \pm 0.60$ | $6.30 \pm 1.54$ | $7.66 \pm 1.27$ |

Table 3. Growth performance of prawn, dedhuwa, mara and pothi in different treatments (Mean $\pm$ S.E.)

| Parameters | Treatments |  |  |  |  |  |  |  |
| :--- | :--- | :--- | :--- | :--- | :---: | :---: | :---: | :---: |
|  | $\mathrm{T}_{1}$ |  |  |  |  | $\mathrm{~T}_{2}$ | $\mathrm{~T}_{3}$ | $\mathrm{~T}_{4}$ |
| Prawn |  |  |  |  |  |  |  |  |
| Initial mean weight $(\mathrm{g} / \mathrm{fish})$ | $0.18 \pm 0.01$ | $0.21 \pm 0.03$ | $0.23 \pm 0.01$ | $0.21 \pm 0.01$ |  |  |  |  |
| (g/prawn) |  |  |  |  |  |  |  |  |
| Initial total weight $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | $0.01 \pm 0.00$ | $0.01 \pm 0.00$ | $0.01 \pm 0.00$ | $0.01 \pm 0.00$ |  |  |  |  |
| Final mean weight $(\mathrm{g} / \mathrm{prawn})$ | $4.29 \pm 4.19$ | $16.67 \pm 8.56$ | $5.73 \pm 2.92$ | $26.53 \pm 8.91$ |  |  |  |  |
| Final total weight $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | $0.03 \pm 0.03$ | $0.31 \pm 0.26$ | $0.04 \pm 0.02$ | $0.56 \pm 0.26$ |  |  |  |  |
| Survival (\%) | $4.44 \pm 4.44$ | $22.08 \pm 18.12$ | $9.18 \pm 4.88$ | $33.86 \pm 13.09$ |  |  |  |  |
| Daily weight gain <br> (g/prawn $/$ day $)$ | $0.02 \pm 0.03$ | $0.09 \pm 0.04$ | $0.03 \pm 0.01$ | $0.14 \pm 0.04$ |  |  |  |  |
| Total weight gain $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | $0.02 \pm 0.02$ | $0.31 \pm 0.26$ | $0.03 \pm 0.02$ | $0.55 \pm 0.26$ |  |  |  |  |


| Dedhuwa |  |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
| Initial mean weight $(\mathrm{g} /$ fish $)$ | - | $0.98 \pm 0.00$ | - | - |
| Initial total weight $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | $0.43 \pm 0.00$ | - | - |
| Final total weight $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | $2.98 \pm 1.78$ | - | - |
| Total weight gain $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | $2.56 \pm 1.80$ | - | - |


| Mara |  |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
| Initial mean weight $(\mathrm{g} / \mathrm{fish})$ | - | - | $1.39 \pm 0.00$ | - |
| Initial total weight $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | - | $0.29 \pm 0.00$ | - |
| Final total weight $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | - | $1.27 \pm 0.13$ | - |
| Total weight gain $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | - | $0.98 \pm 011$ | - |
| Pothi |  |  |  |  |
| Initial mean weight $(\mathrm{g} / \mathrm{fish})$ | - | - | - | $1.94 \pm 0.36$ |
| Initial total weight $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | - | - | $0.58 \pm 0.01$ |
| Final total weight $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | - | - | $4.62 \pm 0.91$ |
| Total weight gain $\left(\mathrm{kg} / 100 \mathrm{~m}^{2}\right)$ | - | - | - | $4.04 \pm 0.92$ |

Mean values with different superscript letters in the same row are significantly different ( $\mathrm{P}<0.05$ )

Table 4. Extrapolated net yield of carp, prawn, dedhuwa, mara and pothi, and combined total net fish yield ( $\mathrm{t} / \mathrm{ha} / \mathrm{yr}$ ) and Apparent Food Conversion Ratio (AFCR) in different treatments (Mean $\pm$ S.E.)

| Parameters | Treatments |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
|  | $\mathrm{T}_{1}$ | $\mathrm{~T}_{2}$ | $\mathrm{~T}_{3}$ | $\mathrm{~T}_{4}$ |
| Carp $(\mathrm{t} / \mathrm{ha} / \mathrm{y})$ | $3.01 \pm 0.19^{\mathrm{b}}$ | $3.38 \pm 0.21^{\mathrm{ab}}$ | $3.77 \pm 0.29^{\mathrm{ab}}$ | $4.14 \pm 0.29^{\mathrm{a}}$ |
| Prawn $(\mathrm{t} / \mathrm{ha} / \mathrm{y})$ | $0.01 \pm 0.00$ | $0.04 \pm 0.03$ | $0.01 \pm 0.00$ | $0.07 \pm 0.00$ |
| Dedhuwa $(\mathrm{t} / \mathrm{ha} / \mathrm{y})$ | - | $0.39 \pm 0.27$ | - | - |
| Mara $(\mathrm{t} / \mathrm{ha} / \mathrm{y})$ | - | - | $0.15 \pm 0.11$ | - |
| Pothi $(\mathrm{t} / \mathrm{ha} / \mathrm{y})$ | - | - | - | $0.61 \pm 0.14$ |
| Total $(\mathrm{t} / \mathrm{ha} / \mathrm{y})$ | $3.02 \pm 0.19^{\mathrm{b}}$ | $3.81 \pm 0.51^{\mathrm{ab}}$ | $3.92 \pm 0.31^{\mathrm{ab}}$ | $4.82 \pm 0.38^{\mathrm{a}}$ |
| AFCR | $3.3 \pm 0.51$ | $2.6 \pm 0.38$ | $2.6 \pm 0.02$ | $2.5 \pm 0.04$ |

Mean values with different superscript letters in the same row are significantly different $(\mathrm{P}<0.05)$.

Both total gross return and return from carp were significantly higher ( $\mathrm{P}<0.05$ ) in $\mathrm{T}_{4}$ than $\mathrm{T}_{1}$ (Table 5). Gross margin was also found significantly higher $(P<0.05)$ in $T_{4}$ than $T_{1}$.

Table 5. Gross margin in different treatments based on $100 \mathrm{~m}^{2}$ pond in Nepalese currency (NRs.) during experimental period

| Parameters | Treatments |  |  |  |
| :---: | :---: | :---: | :---: | :---: |
| Variables | $\mathrm{T}_{1}$ | $\mathrm{T}_{2}$ | $\mathrm{T}_{3}$ | $\mathrm{T}_{4}$ |
| Gross Return |  |  |  |  |
| Carp | $4,639 \pm 285^{\text {b }}$ | $5,186 \pm 308^{\text {ab }}$ | $5,720 \pm 444^{\text {ab }}$ | 6,314 $\pm 454^{\text {a }}$ |
| Prawn | $17 \pm 17$ | $191 \pm 158$ | $25 \pm 14$ | $340 \pm 160$ |
| Dedhuwa | - | $387 \pm 232$ | - | - |
| Mara | - | - | $165.79 \pm 31.37$ | - |
| Pothi | - | - | - | $601 \pm 118$ |
| Total Gross Return | $4,656 \pm 270^{\text {b }}$ | $5,765 \pm 679^{\text {ab }}$ | $5,911 \pm 463^{\text {ab }}$ | $7,256 \pm 501^{\text {a }}$ |
| Variable Cost |  |  |  |  |
| Lime | 50 | 50 | 50 | 50 |
| Feed | 1,284 $\pm 261$ | 1,269 $\pm 309$ | 1,472 $\pm 269$ | 1,657 $\pm 252$ |
| Urea | 531 | 531 | 531 | 531 |
| DAP | 180 | 180 | 180 | 180 |
| Carp fingerlings | 100 | 100 | 100 | 100 |
| SIS | - | 150 | 150 | 150 |
| Prawn | 100 | 100 | 100 | 100 |
| Total Variable Cost | $2,246 \pm 261$ | 2,380 $\pm 309$ | $2,583 \pm 269$ | 2,769 $\pm 252$ |
| Gross Margin $\left(100 \mathrm{~m}^{2}\right)$ | $2,409 \pm 396^{\text {b }}$ | $3,384 \pm 387^{\text {b }}$ | $3,327 \pm 196^{\text {b }}$ | 4,486 $\pm 283^{\text {a }}$ |
| Gross Margin (in'000) (NRs./ha/y) | $323.9 \pm 151.1^{\text {b }}$ | $456.9 \pm 214.9^{\text {b }}$ | $457.7 \pm 113.1^{\text {b }}$ | $605.9 \pm 106.5^{\text {a }}$ |

Mean values with different superscript letters in the same row are significantly different $(P<0.05)$

## Discussion

All the water quality parameters were within the suitable range for carp. Water was muddy brown instead of green during rainy season due to dike run off as ponds were newly constructed and dikes were uncovered. In addition, canal water used to top the pond was also muddy brown. According to Boyd (1990) the non-algal turbidity is caused by suspended clay particles due to erosion of newly constructed pond dikes, run off from the dike and use of muddy canal water during rainy season.

The production of dedhuwa, mara, and pothi did not differ significantly ( $\mathrm{P}>0.05$ ) among treatments. However, their effect on growth and production of carp was apparent. Total weight gain of silver carp was significantly higher $(P<0.05)$ in $T_{3}$ and $T_{4}$ than other treatments. This might be due to no niche overlapping with mara and pothi, which are omnivore while silver carp is a phytoplankton feeder (Wahab and Kadir 2009). Silver carp is a surface feeder while pothi is a bottom feeder. Different feeding habits and habitats reduced interspecific competition between silver carp and mara and pothi. Total weight gain of bighead carp was found significantly higher ( $\mathrm{P}<0.05$ ) in $\mathrm{T}_{3}$ and $\mathrm{T}_{4}$ than $\mathrm{T}_{2}$ indicating that mara and pothi had positive effect on its growth and production. Perhaps no food competition occurred between bighead carp and mara and pothi because both mara and pothi are omnivore (Wahab and Kadir 2009).

Net carp yield and total net fish yield was significantly higher ( $\mathrm{P}<0.05$ ) in $\mathrm{T}_{4}$ than $T_{1}$, which can be attributed to positive effect of pothi on carp leading to better growth and production of carp. The net carp yield in the present experiment was higher than as reported by Miah et al. (1992), Shahabuddin et al. (1994), Mazid et al. (1997), Pandey (2002), Abbas et al. (2010), Rahman et al. (2006), Gharti (2009) and Yadav (2011) from carp polyculture system. The net carp yield in the present experiment was lower than those reported by Wahab et al. (1995) of $2,225 \mathrm{~kg} / \mathrm{ha}$ in 120 days (equivalent to 6,767 $\mathrm{kg} / \mathrm{ha} / \mathrm{yr}$ ), Jena et al. (2002) of $5,843 \mathrm{~kg} / \mathrm{ha}$ and Lakshmanan et al. (1971) of 2,229 to $4,209 \mathrm{~kg} / \mathrm{ha} / \mathrm{yr}$.

The overall result showed that dedhuwa, mara and pothi did not show adverse effect on growth and production of carp. Production of carp was better in SIS added ponds. Adding SIS in appropriate density enhanced carp production. Carp production was higher in SIS treatments. This can be attributed to partial harvesting system of SIS which thinned the population of SIS and maintained appropriate density in ponds. Moreover, SIS might be compatible to carp as reported by Waheb et al. (2003), Alim et al. (2005), Kadir et al. (2006) and Yadar (2011). Though all treatments were profitable but profit was highest in pothi included treatment due to higher total production. Based on production and profit, carp-prawn-pothi polyculture treatment appears to be the best among the treatments.

In conclusion, polyculture of carp, prawn and SIS: dedhuwa, mara and pothi gave higher total fish production, which ultimately increased household income and fish consumption particularly that of dedhuwa, mara and pothi. Since SIS are rich in micronutrients, increased intake of such fish can improve nutrition of rural poor people. So, polyculture of carp, prawn and SIS could be the better option for rural poor. The approach also helps conservation of local fish species through pond polyculture in Nepal.

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# Polyculture performance of asala (Schizothorax plagiostomus) with rainbow trout (Oncorhynchus mykiss) 

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#### Abstract

An experiment was conducted to assess the growth performance of asala (Schizothorax plagiostomus) and trout (Oncorhynchus mykiss) in mono - and polyculture systems in outdoor raceway ponds ( $8 \mathrm{~m}^{2}$ ) at the Fisheries Research Center, Trishuli for 3 months. The experiment was set up in a Completely Randomized Design (CRD) with 4 treatments and 3 replications. The treatments were: (i) asala monoculture; (ii) trout monoculture; (iii) asala + trout polyculture; (iv) asala + trout polyculture with substrate. Asala were stocked at $25 / \mathrm{m}^{2}$ and trout were stocked at $50 / \mathrm{m}^{2}$. Three ponds were provided with substrate (small boulder about $10 \%$ of surface area in the pond bottom). Fish were fed ad libitum with standard trout feed ( $39.5 \pm 0.5 \% \mathrm{CP}$ ) once a day. Both asala and trout grew steadily and slowly during the entire culture period with significantly higher growth of asala in monoculture ( $0.17 \mathrm{~g} / \mathrm{fish} / \mathrm{d}$ ) than polyculture with trout ( $0.10 \mathrm{~g} /$ fish $/ \mathrm{d}$ ). However, there was no significant difference in the growth of trout between mono - and polyculture. There was no significant difference for all growth and production parameters between with and without


substrate treatments. On overall, 15-16\% net fish yield (NFY) was increased by the addition of asala in trout monoculture systems.

Keywords: asala, rainbow trout, polyculture

## Introduction

Asala (Schizothorax plagiostomus) is a high-value, cold water, native fish species which is well acknowledged for its taste and possesses both food and recreational value. It is declining from its habitat due to over fishing pressure and now undersize fish mostly found in markets (Shrestha 1993; Rai et al. 2002). Thus, there is a need to develop a culture technique to reduce fishing pressure in the river. Previous experiment has demonstrated the possibility of asala culture in raceway ponds.

Rainbow trout (Oncorhynchus mykiss) culture has already been established in cold water of Nepal (Nepal et al. 2002). It is cultured in raceway ponds in monoculture. Water quality deterioration due to waste feed and attached periphytons sometimes cause fish mortality. To utilize such waste food and attached periphytons, asala might be a complementary species with trout. The general objective of this study was to explore the possibility of polyculture between trout and asala. The specific objectives were to compare the growth and survival of trout and asala in mono - and polyculture; and to access the effect of substrate on growth and survival of trout and asala.

## Materials and methods

An experiment was conducted to assess the growth performance of asala in mono and polyculture in outdoor ponds ( $4.0 \mathrm{~m} \times 2.0 \mathrm{~m} \times 1.0 \mathrm{~m}$ ) at the Fisheries Research Center, Trishuli for 3 months. The experiment was set in a Completely Randomized Design (CRD) with 4 treatments and each treatment was replicated thrice. The treatments were: (i) asala monoculture ( $25 \mathrm{fish} / \mathrm{m}^{2}$ ); (ii) trout monoculture ( $50 \mathrm{fish} / \mathrm{m}^{2}$ ); (iii) asala + trout polyculture $(25+50$ fish $/ \mathrm{m}^{2}$ ) and (iv) asala + trout polyculture with substrate ( $25+50$ fish $/ \mathrm{m}^{2}$ ).

Ponds were repaired and disinfected with lime $\left(\mathrm{Ca}(\mathrm{OH})_{2}\right)$ at a rate of 200 $\mathrm{g} /$ pond and filled with river water. Three ponds were provided with substrate (small boulder about $0.7 \mathrm{~m}^{2}$ surface area in the pond bottom - $10 \%$ of surface area) and rest nine ponds were without substrates.

Asala fry of 4.5 g body weight and rainbow trout fry of 0.8 g body weight were stocked at $25 \mathrm{fry} / \mathrm{m}^{2}$ and $50 \mathrm{fry} / \mathrm{m}^{2}$, respectively. During stocking, total weight and individual weight of fry were recorded using electronic balance. About 75 cm water depth was maintained in all the ponds. During the experimental period monthly growth of fry were monitored. Fish were fed with standard trout feed $(39.5 \pm 0.5 \% \mathrm{CP})$ in a day during the entire experimental period. Water temperature, dissolved oxygen, pH and turbidity were monitored daily using thermometer, DO meter, pH meter and nephelometer, respectively. The complete harvesting was done after 3 month by complete draining of pond water. Individual and total batch weight was recorded using electronic balance. Data were analyzed using SPSS Program (version 16) for the analysis of variance (ANOVA). Paired t-test was done for comparison of two means wherever necessary. Means were given with $\pm 1$ standard error and statistical significance was considered at ( $p<0.05$ ) for the effect of treatments.

## Results

The initial weight, harvest weight, growth rate, survival, net fish yield, and apparent feed conversion ratio (AFCR) of asala and trout are presented in Table 1 and 2. The survival of asala ranged from $88-90 \%$ without significant differences between treatments. Similarly, the survival of trout ranged from 91$96 \%$ without significant differences between treatments (Table 1).

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Table 1. Stocking weight, harvest weight, growth and survival of asala and trout in different treatments during the experimental period (Mean $\pm$ SE).

| Parameters | Treatments |  |  |  |
| :---: | :---: | :---: | :---: | :---: |
|  | Asala mono | Trout mono | Asala+Trout | Asala+Trout with substrate |
| Mean stocking weight (g) <br> Asala <br> Trout | $4.5 \pm 0.0^{a}$ | $0.8 \pm 0.0^{a}$ | $\begin{aligned} & 4.5 \pm 0.0^{a} \\ & 0.8 \pm 0.0^{a} \end{aligned}$ | $\begin{aligned} & 4.5 \pm 0.0^{a} \\ & 0.8 \pm 0.0^{a} \end{aligned}$ |
| Mean harvest weight (g) <br> Asala <br> Trout | $20.2 \pm 1.2^{\text {a }}$ | $21.3 \pm 1.6^{\circ}$ | $\begin{aligned} & 14.2 \pm 0.5^{b} \\ & 26.8 \pm 2.5^{a} \end{aligned}$ | $\begin{aligned} & 12.4 \pm 0.5^{b} \\ & 26.0 \pm 2.6^{a} \\ & \hline \end{aligned}$ |
| Growth rate <br> (g/fish/day)  <br> Asala  <br> Trout  <br>   <br>   | 0.17 $\pm 0.00^{\circ}$ | $0.23 \pm 0.00^{a}$ | $\begin{aligned} & 0.11 \pm 0.00^{b} \\ & 0.29 \pm 0.00^{a} \end{aligned}$ | $\begin{aligned} & 0.09 \pm 0.00^{b} \\ & 0.28 \pm 0.00^{a} \end{aligned}$ |
| Survival (\%) <br> Asala <br> Trout | $90.0 \pm 1.4^{\text {a }}$ | $95.4 \pm 0.4^{a}$ | $\begin{aligned} & 89.8 \pm 0.9^{a} \\ & 95.8 \pm 0.6^{a} \end{aligned}$ | $\begin{aligned} & 88.0 \pm 1.5^{a} \\ & 90.9 \pm 1.0^{a} \end{aligned}$ |

Both asala and trout grew steadily and slowly during the entire culture period with significantly higher growth of asala in monoculture ( $0.2 \mathrm{~g} /$ fish $/ \mathrm{d}$ ) than polyculture with trout ( $0.1 \mathrm{~g} /$ fish $/ \mathrm{d}$, Figures 1 and 2). However, there was no significant difference in the growth of trout between mono - and polyculture.


Figure 1. Growth trend of rainbow trout in different treatments during the experimental period.

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Figure 2. Growth trend of asala in different treatments during the experimental period.

The NFY of asala in monoculture ( $3.9 \mathrm{~g} / \mathrm{m}^{2} / \mathrm{d}$ ) was higher than polyculture with trout ( 1.9 and $2.2 \mathrm{~g} / \mathrm{m}^{2} / \mathrm{d}$ ). However, there was no significant difference in the growth of trout between mono - and polyculture. There was no significant difference for all growth and production parameters between with and without substrate treatments. On overall, 15-16\% NFY was increased by the addition of asala in trout monoculture systems (Table 2).

Table 2. Net fish yield (NFY), combined net fish yield, net fish yield increase and apparent feed conversion ratio (AFCR) in different treatments during the experimental period.

| Parameters | Treatments |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
|  | Asala <br> mono | Trout <br> mono | Asala+Trout | Asala+Trout <br> with substrate |
| Net fish yield $\left(\mathrm{g} / \mathrm{m}^{2} / \mathrm{d}\right)$ <br> Asala <br> Trout | $3.9 \pm 0.6^{\mathrm{a}}$ | - | $2.2 \pm 0.3^{b}$ | $1.9 \pm 0.2^{\mathrm{b}}$ |
| Combined net fish yield <br> $\left(\mathrm{g} / \mathrm{m}^{2} / \mathrm{d}\right)$ | $3.9 \pm 0.6^{\mathrm{c}}$ | $10.8 \pm 0.8^{\mathrm{a}}$ | $13.8 \pm 0.3^{\mathrm{a}}$ | $12.7 \pm 1.3^{\mathrm{a}}$ |
| Net fish yield increased <br> by Asala (\%) | - | $16.0 \pm 1.3^{a}$ | $14.7 \pm 1.2^{\mathrm{a}}$ |  |
| AFCR | - | $15.8 \pm 2.5^{\mathrm{a}}$ | $15.2 \pm 0.4^{\mathrm{a}}$ |  |

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The mean water temperature, dissolved oxygen and pH were not significantly different among treatments ( $P>0.05$,Table 3 ). The temperature ranged from 12$22^{\circ} \mathrm{C}$ with an average of around $16^{\circ} \mathrm{C}$ in all treatments (Figure 3). The DO ranged from $8.2-8.5 \mathrm{mg} / \mathrm{L}$ and pH ranged from 7.2-7.4 during experimental period (Figures 4 and 5).

Table 3. Mean and range of water quality parameters in different treatments during the experimental period.

| P Parameters | Treatments |  |  |  |
| :--- | :---: | :---: | :---: | :---: |
|  | Asala <br> mono | Trout- mono | Asala+Trout | Asala+Trout <br> with substrate |
| Temperature $\left({ }^{\circ} \mathrm{C}\right)$ | $16.2^{a}$ | $16.2^{a}$ | $16.3^{a}$ | $16.2^{a}$ |
|  | $(12.6-21.1)$ | $(12.4-21.3)$ | $(12.5-21.5)$ | $(12.6-21.2)$ |
| Dissolved oxygen | $8.2^{a}$ | $8.3^{a}$ | $8.5^{a}$ | $8.5^{a}$ |
| $(\mathrm{mg} / \mathrm{L})$ | $(7.2-9.4)$ | $(7.6-9.3)$ | $(7.6-10.3)$ | $(8.0-9.8)$ |
| pH | $7.2^{a}$ | $7.3^{a}$ | $7.4^{a}$ | $7.4^{a}$ |
|  | $(7.0-7.5)$ | $(7.0-7.6)$ | $(6.9-7.6)$ | $(7.0-7.6)$ |



Figure 3. Weakly means of water temperature in different treatments during the experimental period.

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Figure 4. Weakly means of dissolved oxygen in different treatments during the experimental period.


Figure 5. Weakly means pH of water in different treatments during the experimental period.

## Discussion

The growth of asala in the present experiment was slow ( $0.09-0.17 \mathrm{~g} /$ fish $/ \mathrm{d}$ ) however, it was higher than $0.01 \mathrm{~g} /$ fish $/ \mathrm{d}$ reported by Lamsal (2011). This indicates that the growth of asala was faster in the second year as reported by Sharma (1989) and Tuchiya (1993). Similarly, the growth of trout in the present experiment was $0.23-0.29 \mathrm{~g} /$ fish $/ \mathrm{d}$ which was similar with the report of Rai et al. (2002). The present experiment showed a promising result that the growth of trout was higher in polyculture than monoculture ( $p>0.05$ ). This might be due to improved water quality by utilizing waste feed. This result indicated a positive effect of asala to the trout. In the first experiment, it was observed that the growth of asala was better in the ponds with substrate. However, there was no significant effect of substrate in growth and production of asala and trout in the present experiment. All the water quality parameters were within the preferred range for asala and trout culture and there was no significant difference among treatments. This indicated that addition of asala to the trout had no adverse effect on water quality. The present experiment demonstrated that asala and trout polyculture might be a good alternative to increase feed utilization, maintain good water quality and increase production. Asala was a good complementary species with trout and addition of certain percentage of asala in trout raceways might not harm to the trout. On overall, 15-16\% NFY was increased by the addition of asala to trout monoculture systems. As this experiment was conducted for short duration (only 3 months), an experiment with full culture cycle is recommended to explore the polyculture performance of asala and trout in raceways.

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# Fish diversity of Narayani River System in Nepal 

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#### Abstract

A total of 108 fish species were recorded from different sampling points of the Narayani River system in Chitwan and Nawalparasi district. These species were classified into 9 orders, 27 families and 70 genera. Among the orders, Cypriniformes had the highest number of species (49\%) followed by Siluriformes (29\%), Perciformes (12\%), Synbranchiformes (4\%), Osteoglossiformes (2\%) while Anguiliformes, Beloniformes, Clupeiformes and Tetraodontiformes represented each by about $1 \%$. Cyprinidae has the highest number of species ( $40.7 \%$ ) among the families followed by Sisoridae (10.1\%), Bagridae (7.4\%), Cobitidae (4.6\%), Schilbeidae (4.6\%), Channidae (3.7\%), Balitoridae (2.7\%), Mastacembelidae (2.7\%), Siluridae (2.7\%), Notopteridae (1.8\%), Ambassidae (1.8\%), Nandidae (1.8\%) and Mugilidae (1.8\%). Other families accounted for about $1 \%$ were Anguillidae, Belonidae, Clupeidae, Psilorhynchidae, Anabantidae, Gobiidae, Belontidae, Synbranchidae, Amblycipitidae, Pangasidae, Clariidae, Heteropneustidae, Chacidae and


Tetraodontidae. Various stocks of carps, carfishes, gars, gobies, snakeheads, feather-backs, perch, loaches, eels and puffers were maintained and support livelihoods of people. Several economically important native species are now at risk of extinction. However, cultivation of some species such as Sahar, Tor putitora; Katle, Neolissocheilus hexagonolepis; Asala, Schizothorax richardsonii and Gardi, Labeo dero through induced spawning opened the way to maintain and conserve the indigenous stock. More efforts and appropriate measures are urgently needed.

Keywords: Ichthyofaunal diversity, Rapti-Narayani River system, Fish conservation

## Introduction

Nepal is a Himalayan country possessing extreme topography ranging in altitude from 60-8848 m from mean sea level and has huge water resources covering an area of 818,500 ha (about $2 \%$ ) of the world's natural water resources (Rai et al. 2007). This contains more than 6000 rivers and streams originating from Himalayas, Mahabharat and Siwalik mountain ranges (Gubhaju 2011), including three major rivers namely Koshi, Gandaki and Karnali totaling over 25000 km in length and covering an estimated area of $3,95,000$ ha ( $48.2 \%$, Rai et al. 2007). These major rivers along with other rivers/streams and scores of lakes and reservoirs are the habitat of 232 species of fish from different climatic zones (Shrestha 2008). They belong to 114 genera under 37 families and 11 orders having high commercial potential in terms edibility, sport fishing and as ornamentals. Out of the aforementioned figure (232), there are 217 native species and 15 exotic species (Shrestha 2008). It has been reported that the size of the riverine fishes varies from 280 kg measuring 225 cm Gounch or Thend (Sisorid catfish, Bagarius yarrell/) to the smallest ( 2.5 cm ) yellowish-brown fish, Koshi Hara or Datkitari, Hara hara (Shrestha 2008).

The Narayani River system is one of the three major river systems of Nepal with seven tributaries. It is, therefore, called Sapta Gandaki and drains central Nepal (Figure 1) The Kali Gandaki which originates from Tibetan plateau is regarded as one of the major tributaries and is joined by the Trishuli, another major tributary at Devghat where it attains the name of Narayani. Besides, Marsyangdi, Seti, Budhi Gandaki, Madi and Myagdi Rivers are also considered as major tributaries. These tributaries traverse through varied geographic zones, hence, represent diversified fish species (Dhital et al. 2002). The river Narayani in Chitwan district extends widely and collects Rapti and Rew rivers along with a large number of streams. The Narayani River has an average flow of $283 \mathrm{~m}^{3} / \mathrm{sec}$ and the velocity of water changes from very fast in the higher altitudes to mild in the lower plains. The river receives sewage and wastes from the Bhrikuti Paper and Pulp Factory in Gaindakot Village Development Committee (VDC) in Nawalparasi district. It has a barrage to control water for irrigation and power generation without any fish way in Nepal-India border at Tribenighat of Triveni-susta VDC of Nawalparasi. There is a great impact of barrages and irrigation systems on the aquatic life of upstream and downstream of the river. Faunal composition is affected by interference on the natural system of the river (Rajbanshi 2002; Gubhaju 2012). The fishery resources of the Narayani River system is of great importance for the people of the area as it supports many whose livelihoods depend upon that tremendous fish diversity.


Figure 1. Map of Nepal showing catchment area of the Narayani River system. Circle indicates the Narayani River and its tributaries, which drain central Nepal.

The fragmentary records of fish fauna of the Gandaki River system (Narayani River system) are available (Shrestha 1981, 1990, 1994, Jha et al. 1989, Dhital et al. 2002, Rajbanshi 2002). Shrestha (1981) reported 23 species fish from the Gandaki River system. Edds (1986) recorded 111 species of fish through longitudinal survey of the Kaligandaki-Narayani River system from 100 m to 3000 m asl. Jha et al. (1989) and Dhital et al. (2002) collected 68 and 69 species from the Narayani-Rapti river system in Chitwan, respectively. Shreshta (1990) and Rajbanshi (2002) reported 88 and 85 species from the main cradle of the Narayani River, respectively.

Major rivers along with floodplains support wide range of biodiversity and services to society. The Narayani River systems have vital fish genetic stocks and species complexes. These stocks are greatly affected by alteration of habitat, pollution and overexploitation of aquatic resources. For their proper conservation planning, there is a need to assess species distribution in different geographic locations. Therefore, an effort has been made to investigate the fish diversity of the Narayani River system.

## Materials and methods

A survey was conducted to assess fish bio-diversity of the Narayani River along with its tributaries. Eight fishing spots of upstream and downstream in Chitwan and Nawalparasi districts were monitored from August 2006 to December 2009. The study areas (Figure 2) were divided into following locality (I) Trishuli River at Kuringhat, Trishuli and Marsyangadi confluence at Muglin, Darechowk VDC, Chitwan (II) Narayani River (Trishuli and Kali Gandaki confluence) at Devghat and Narayanghat in Bharatpur Municipality, Chitwan, (III) north eastern bank of Narayani River at Sivaghat, Kabreghat and Kharkhareghat of Mangalpur VDC, Chitwan, (IV) eastern and western bank of Narayani River at Koleghat of Chitwan and Nawalparasi, (V) Rapti River at Meghauli and Rapti -Narayani confluence at Golaghat of Meghauli VDC, Chitwan, (VI) Khageri River, Burhi Rapti, and Lothar River in eastern Chitwan, (VII) Rew River and its feeder streams at Madi in Chitwan National Park, Chitwan, (VIII) north-
western bank of Narayani River at Tribenighat of Triveni-susta VDC of Nawalparasi.


Figure 2. Map of Chitwan district showing study area and location. Black dots indicate sampling points.

Different types of fishing gears such as cast net, gill net and fish traps were used for the collection of fish specimens from these localities. These specimens were preserved in $10 \%$ formalin. Ecological features of the fish habitat and color of fish species were recorded throughout collection. The large specimens were incised lengthwise along the abdomen while the smaller ones were directly put into the formalin. The fish were kept upside down to avoid any damage to caudal fin in the container. For species identification, counts of lateral line scales and fin rays as well as measurement of body were made according to the system developed by Talwar and Jhingran (1991), Jayaram (1999) and Shrestha (2001, 2008). The identified specimens were preserved and kept with proper labeling in the laboratory of Central Department of Zoology, Kirtipur, Kathmandu, Nepal. The genera under their respective families and the species under their respective genera were arranged alphabetically.

## Results and discussion

The present study revealed that the river had diversified fishes belonging to 9 orders, 27 families, 70 genera and 108 species. Among the orders, Cypriniformes had the highest number of species (49\%) followed by Siluriformes (29\%), Perciformes (12\%), Synbranchiformes (4\%) and Osteoglossiformes (2\%, Figure 3). Interestingly, the orders Anguilliformes, Beloniformes, Clupeiformes, and Tetraodontiformes each were represented by single species; Anguilla bengalensis, Xenentodon cancila, Gudusia chapra, and Tetraodon cutcutia respectively. The percentage composition of the fishes of different orders is given in Figure 3.


Figure 3. Percentage composition of fishes among different orders
Cyprinidae had the highest number of species ( $40.7 \%$ ) among the families followed by Sisoridae ( $10.1 \%$ ), Bagridae (7.4\%), Cobitidae (4.6\%), Schilbeidae ( $4.6 \%$ ), Channidae (3.7\%), Balitoridae (2.7\%), Mastacembelidae (2.7\%), Siluridae (2.7\%), Notopteridae (1.8\%), Ambassidae (1.8\%), Nandidae (1.8\%) and Mugilidae ( $1.8 \%$ ). Other families accounted for about $1 \%$ were Anguillidae, Belonidae, Clupeidae, Psilorhynchidae, Anabantidae, Gobiidae, Belontidae, Synbranchidae, Amblycipitidae, Pangasidae, Clariidae, Heteropneustidae, Chacidae and Tetraodontidae.

Taxonomic position, local name, localities and status of fish species are listed in the Table 1.

Table 1. Systematic position and status of fish species in the Narayani River.

| Order, family, sub-family, genus, species, local name, locality and status | Order, family, sub-family, genus, species, local name, locality and status |
| :---: | :---: |
| I. Order: Anguilliformes Suborder: Anguilloidei Family: Anguillidae <br> 1. Anguilla bengalensis (Gray), Raj bam, I to VIII, R, M | II. Order: Beloniformes Suborder: Belonoidei Family: Belonidae <br> 1. Xenentodon cancila(Ham), Kauwa machha, Dhonga, Thunge, I, to VIII, C, LM |
| III. Order: Clupeiformes <br> Suborder: Clupeioidei <br> Family: Clupeidae <br> 1. Gudusia chapra (Ham.), Suiya, VIII, R |  |
| IV.Order: Cypriniformes <br> Family: Cyprinidae <br> Subfamily: Cyprininae <br> 1. Catla catla (Ham.), Vakura, V \& VIII, R <br> 2. Chagunius chagunio (Ham.), <br> Chaurahi, I \& III, C <br> 3. Cirrhinus mrigala mrigala (Ham.), <br> Naini, III, V \&VIII, R <br> 4. Cirrhinus reba (Ham.), Rewa, III to VIII, C <br> 5. Cyprinion semiplotus (Mc-Clelland), Chepti, <br> Mahanga, III to VIII, R, M <br> 6. Labeo angra (Ham.), Basarhi, Rohu, II to VIII, C, LM <br> 7. Labeo bata (Ham.), Rohu, Bata,V to VIII, C, LM <br> 8. Labeo calbasu (Ham.) Karaunchh, | Subfamily: Cultrinae <br> 23. Salmostoma acinaces (Val.) Chelha, V toVIII, C <br> 24. Salmostoma bacaila (Ham.) Chelha, VIII, UC <br> 25. Securicula gora (Ham.) Chelha, VIII, UC <br> Subfamily: Rasborinae <br> 26. Amblypharyngodon mola (Ham.), Mara, Mola, Dhawai, VIII, R <br> 27. Aspidoparia morar . (Ham.), Chepua, I \& VIII, C <br> 28. Barilius barna (Ham.), Faketa, I, II \& V, C <br> 29. Barilius barila (Ham.), Faketa, Motia, I ,II to V, R <br> 30. Barilius bendelisis (Ham.), |

9. Labeo dero (Ham.), Gardi, I to VIII, C
10. Labeo gonius (Ham.), Kursa, V to VIII, U C
11. L. rohita (Ham.), Rohu, III to VIII, UC
12. Labeo pangusia (Ham.), Hande, Kalaunchh, I to VIII, R
13. Neolissocheilus hexagonolepis (McCl), Katle, I to III, R
14. Osteobrama cotio cotio (Ham.), Gurda, VIII, C
15. Puntius chola (Ham.), Sidhare, III to VIII, C
16. Puntius conchonius (Ham.), Sidhare, III to VIII, UC
17. Puntius phutunio (Ham.), Sidhare, VIII, R
18. Puntius sarana sarana (Ham.), Darhi, III to VIII, C
19. Puntius sophore (Ham.), Gujarahta, III to VIII, C
20. Puntius ticto (Ham.), Tikulia sidhre, III to VIII, C
21. Tor Putitora (Ham.), Mahaseer, Sahar, I \& IV, R, M
22. Tor for (Ham), Mahaseer, Sahar, I \& IV, R, M

Order: Cypriniformes (contd.)
Family: Cyprinidae
Subfamily:Garrinae
42. Crossocheilus lativs latius (Ham.), Budhuna, III to V, C
43. Garra annandalei (Hora), Budhuna,Lohari II to VI, UC
44. Garra gotyla (Gray), Nakuro Budhuna, II to VI, C
31. Barilius tileo (Ham.), Faketa, III and $V, C$
32. Barilius vagra (Ham.), Faketa, IV and VIII, RS
33.Brachydanio rerio (Ham.), Dedhwa, III \& VIII, RS
34. Danio aequipinnatus (Ham.), Chithari Pothi, Pataki, III to VI, RS
35. Danio devario (Ham.), Pataki, III to $\mathrm{VI}, \mathrm{RS}$
36. Esomus danricus (Ham.), Dedhwa, III to VIII, C
37. Parluciosoma daniconius (Ham.), Dedhwa, III to VIII UC
38. Raiamas bola (Ham.) Galara,Hasta III to VIII, C
39. Raiamas guttatus (Ham.) Galara,Hasta, III to VIII, C
40. Schizothorichthys progastus (McCl.), Asla, I to III, R
41.Schizothorax richardsonii (Gray),Asla, I to III,R

Order Cypriniformes (contd.)
Family: Cobitidae
Subfamily: Cobitinae
49. Lepidocephalus guntea (Ham.), Nakati, III to VIII, C
50. Somileptus gongota (Ham.), Goira, III to VIII,R
Subfamily: Botiinae
51. Botia almorhae (Gray), Baghe, II to VIII, R

| Family: Psilorhynchidae <br> 45. Psilorhynchus pseudecheneis (Menon \& Datta), Tite, I to III, R <br> Family: Balitoridae <br> Subfamily: Nemacheilinae <br> 46. Acanthocobitis botia (Ham.), Goira, III to IV, C <br> 47. Nemacheilus corica (Ham.), Gadela, I to III,UC <br> 48. Schistura beavani (Gunther), Goira, III to IV, C | 52. Botia dario (Ham.), Baghe, Il to VIII, R <br> 53. B. lohachata (Chauduri), Baghe, II to VIII, UC <br> V. Order: Osteoglossiformes <br> Suborder: Notopteroidei <br> Family: Notopteridae <br> 1. Notopterus notopterus (Pallas), Golahi, Patra, III to VIII, C <br> 2. Chitala chitala (Ham.), Moi, Chitala, III to VIII, R |
| :---: | :---: |
| VI. Order: Perciformes <br> Suborder: Percoidei <br> Family: Ambassidae <br> 1. Chanda nama (Ham.), Chuna, III to VIII, C <br> 2. Parambassis ranga (Ham.), Chuna, Chanarbijua, III to VI, C <br> Family: Nandidae <br> Subfamily: Nandinae <br> 3. Nandus nandus (Ham.), Dhedhari, III to VIII C <br> Subfamily: Badinae <br> 4. Badis badis (Ham.), Kotari, Khesaki, Chepti,VIII, R <br> Suborder: Anabantoidei <br> Family: Anabantidae <br> 5. Anabas testudineus (Bloch), Kabai, VIII, R | VII. Order: Siluriformes <br> Family: Amblycipitidae <br> 1. Amblyceps mangois (Ham.), Paharisinghi, Paidani, Chilni, Aagomachha, III to VIII, C <br> Family: Bagridae <br> 2. Aorichthys aor (Ham.), Kubratengar, III to VIII, C <br> 3. Aorichthys seenghala (Sykes), Sarangatengar, Kanti, III to VIII, C <br> 4. Mystus bleekeri (Day.), Tengasi, III to VIII, RS <br> 5. M.cavasius (Ham.), Tengara, III to VIII, RS <br> 6. M.menoda (Ham.), Tengara, VIII, R <br> 7. M.tengara (Ham.), Tengara, Ill to VIII, C <br> 8. M.vittatus(Bloch), Tengna, III to VIII, C <br> 9. Rita rita (Ham.), Rita, Tengar, III |


| Suborder: Gobioidei | to VIII, UC |
| :---: | :---: |
| Family: Gobiidae |  |
| Subfamily: Gobiinae | Family: Siluridae |
| 6. Glossogobius giuris (Ham.), Bulbule, Bulla, III to VIII, C | 10. Ompok bimaculatus (Bloch), Lodara, Nanaria III to VIII, C |
| Suborder: Mugiloidei Family: Mugilidae | 11. Ompok pabda (Ham.), Pabda III to VIII, R |
| 7. Rhinomugil corsula (Ham.), Hurra, Thadiya, VIII, R | 12. Wallago attu (Bl. \& Schn.), Barari, III to VIII, C |
| 8. Sicamugil cascasia (Ham.), Hurra, Piyarpeti,VIII, R | Family: Schilbeidae Subfamily: Ailiinae |
| Family: Belontidae Subfamily: Trichogasterinae | 13. Ailia coila (Ham.), Jalkapoor, IV to VIII, R |
| 9. Colisa fasciatus (Bloch \&Schneider) | Subfamily: Schilbeinae |
| Kotari, III to VIII, C | 14. Clupisoma garua (Ham.), Jalkapoor, IV to VIII, C |
| Sub-order: Channoidae Family: Channidae | 15. Eutropiichthys vacha (Ham.), Suha, III to VIII, R |
| 10. Channa marulius (Ham.), Saura, III to VIII, C | 16. Eutropiichthys murius(Ham.), Jalkapoor, IIII to VIII, R |
| 11. Channa orientalis (Bl.\& Schn.), Hile, Bhoti, III to VIII, C | 17. Pseudeutropius atherinoides(Bloch), Jalkapoor, III |
| Kasurhati, III to VIII, C | Family : Pangasidae |
| 13. Channa striatus (Bloch), Saura, Gajahari, III to VIII, C | 18. Pangasius pangasius (Ham.) Jalkapoor, VIII, R <br> Family : Sisoridae |
| VIII. Order: Synbranchiformes Suborder: Synbranchoidei | 19. Bagarius bagarius (Ham.), Gonch, Thenda, I to VIII, UC |
| Family: Synbranchidae <br> 1. Monopterus cuchia (Ham.), Andho bam, III to VIII, C | 20. Bagarius yarrelli (Sykes), Gonch, Thenda, I to VIII, UC (Largest fish of Nepal) |
|  | 21. Gagata cenia (Ham.), Gogata, Datkitari IV to VIII, UC |

Suborder: Mastacembeloidei
Family: Mastacembelidae
Subfamily: Mastacembelinae
2. Macrognathus ara/(Bl. \& Schn.), Chuche bam, III to VIII, C
3. Macrognathus pancalus (Ham.), Dhare bam, III to VIII, RS
4. Mastacembelus armatus (Lacepede), Bam, III to VIII, RS

## IX. Order: Tetraodontiformes

Suborder: Tetraodontoidei
Family:Tetraodontidae
Subfamily: Tetraodontinae

1. Tetraodon cutcutia (Ham.), Phuwa machha, Kitkitia, Kichkichauwa V and VIII, R
2. Glyptothorax cavia (Ham.), Kapre, II to IV, R
3. G.indicus (Talwar), Kursimlo, III to $\mathrm{V}, \mathrm{R}$
4. G. pectinopterus (McCl.), Kapre, III to VIII, R
5. G.telchitta (Ham.), Chepti, III to VIII, C
6. Hara hara (Ham.), Datkitari, V to VII, R(Smallest fish of Nepal)
7. Nangra viridescens (Ham.), Katenga VII \& VIII, R
8. Pseudecheneis sulcatus (McCl), Vedra, I to IV, UC
9. Sisor rhabdophorus (Ham), Chheparo machha, Girgitiya VIII, R

Family: Clariidae
30. Clarias batrachus (Linnaeus), Mangur, III to VIII, C

Family : Heteropneustidae
31. Heteropneustes fossilis (Bloch), Singhi, III to VIII, C

Family : Chacidae
32. Chaca chaca (Ham.), Kirkire VIII, R

The status of each species is given as: common (C), uncommon (UC), rare (R), resident (RS), migratory ( $M$ ) and local migrant (LM). Localities I to VIII are given as mentioned in the 'Materials and methods'. Common: Frequently noted during sampling period; Uncommon: Very few represent in the sample; Resident: Never migrate according to local fishermen; Rare: very few, sometimes absent; only repeated sampling reveal the presence of the species.

Results showed that, in general, the Rapti/ Narayani River ecosystem supports diverse stock of carps, cat fishes, perches, feather backs, eels, gobies, puffers, yellowtails, loaches, mullets and so on. Upper stretches of the river is dominated by important coldwater fishes such as Schizothorax sp, Schizothoraichthys sp, Neolissocheilus hexagonolepis, and Tor sp. In the middle and lower stretches of the river, the species found were Barilius sp, Labeo sp, Aorichthys sp, Cirrhinus sp, Macrognathus sp, Mastacembelus armatus, Mystus cavasius, Heteropneustes fossilis, Clarias batrachus, Channa spp, Clupisoma garua, Eutropichthys spp, Pseudeutropius atherinoides, Ailia coila, Wallago attu, Monopterus cuchia, and Bagarius yarrelli. Out of 108 species, common (49), uncommon (14) rare (37) and resident (8) were observed (Figure 4). Among these, Barilius bendelisis, Puntius spp., Esomus danricus, Mystus spp., Glossogobius giuris, Monopterus cuchia, Macrognathus sp., Notopterus notopterus, Aorichthys spp. and Channa spp. were frequently observed. Labeo gonius, Pseudecheneis sulcatus, Gagata cenia, Bagarius yarrelli, Rita rita, Securicula gora, and Nemacheilus corica were uncommon. Similarly, Tor sp., Schizothorax sp., Neolissochilus hexagonolepis, Gudusia chapra, Sicamugil cascasia, Sisor rhabdophorus and Chitala chitala were very rare. Further, Amblypharyngodon mola, Chaca chaca, Rhinomugil corsula and Anabas testudineus were also very rare and collected from canals in Nawalparasi only. Most of these species have high market value and preferred by people. However, they are caught only from the natural water bodies and have not yet been cultured in Nepal.


Figure 4. Status of fish species in the Narayani River system

The genus Glyptothorax spp, Schizothorax sp. Tor spp. and Labeo dero were collected from Kali-Gandaki-Trishuli confluence near Devghat. These species were also collected from Trisuli River at Kuringhat and Muglin while they were very rare in the downstream except Labeo dero. Similarly the mullet Sicamugil cascasia and feather backs Chitala chitala were collected downstream section of the river only. Most of the fishes from this river were collected from the middle and lower stretches. Garra spp and Crossocheilus latius found mostly in the upper and middle stretches were also collected from the lower reaches of river during monsoon. Tetraodon cutcutia was collected from Rapti-Narayani confluence at Golaghat (Khoriamohan) of Chitwan and Trivenighat of Nawalparasi. The Sisorid cat fish Bagarius yarrellii is the largest fish while hara, Hara hara is smallest fish of Nepal. Some species such as Colisa sp, Botia sp Brachydanio sp and Danio sp were also found which could be utilized as ornamental fishes.

Various researchers reported different numbers of fish species from the Narayani River system. Amongst them, Shrestha (1981) reported 23 species fish from the Gandaki River system. Edds (1986) recorded 111 species of fish through longitudinal survey of the Kali Gandaki-Narayani River system from 100 m to 3000 m asl. Jha et al. (1989) and Dhital and Jha (2002) collected 68 and 69 species fish from the Narayani-Rapti River system in Chitwan, respectively. Shreshtha (1990) and Rajbanshi (2002) reported 88 and 85 species from the main cradle of the Narayani River. Further, Shrestha (2008) reported 170 species from the Sapta Gandaki River system and their tributaries from different altitudinal gradient. Present survey recorded 108 species of fishes from the range of Kurintar, Mugling, Devghat, Golaghat (Khoriamohan) along with several tributaries in Chitwan district to Tribenighat in Nawalparasi only. Decrease in species diversity might be due to watershed developments that are taking place at a rapid rate, particularly through dam building and associated habitat destruction (Shrestha 2008) and the barrage to control water for irrigation and power generation without any fish way. Faunal composition is affected by interference on the natural system of the river (Rajbanshi 2002; Gubhaju 2012). Specially, during summer, downstream near barrage was completely dry, which affected the migration of important migratory fishes.

Fishing community of Tribenighat reported declining catches due to degradation of fish habitats and barrage without fish-way which obstructed spawning migration also. Regulation of water flow by damming or irrigation has pronounced effect on fishery resources of major rivers (Gubhaju 2012).

Research on some species such as Tor putitora, Labeo dero, Neolissocheilus hexagonolepis and Schizothorax richardsonii is underway for the purposes of conservation and commercial use. However, more research is needed on whether there are any significant effects of human activities on the fish population through habitat alterations, poisoning and other illegal fishing, over exploitation and new species introduction in the river system.

## Conclusion

Fish diversity of the Narayani River system was found affected by over exploitation of fishery resources and loss of habitat. Abundance of fish species of the river had changed from highly preferred to least preferred species indicating that many of them were at risk. Fish catch data should be recorded and collected to confirm the fishermen's claim of fish decline and appropriate measures should be applied for species conservation.

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Fish diversity and their contribution in livelihoods of fishers' in Koshi River basin

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#### Abstract

This study was carried out to assess fish diversity and contribution of fishery on livelihood of fisher community in Koshi River at Madhuban and West Kusaha Village Development Committee (VDC) of Sunsari district. Fish species were collected from $1^{\text {st }}$ week of July 2007 to February 2008. Altogether 60 fish species belonging 7 orders, 20 families and 40 genera were collected from Koshi River. Among them, 23 were common, 9 were uncommon, 17 were rare, and 11 were resident. According to the fishers, poisoning and haphazard catches were the main reasons of the stock depletion. Out of 65 fishers in the VDCs, $46 \%$ ( 30 respondents) were selected randomly for the household survey. Landless ( $26.7 \%$ ) and less land holding (23.3\%) were the main reasons for depending on riverine fishery to fulfill the basic needs of life. Fishers were found to catch from 0.5 to over 3 kg fish/day from the river. Majority of fishers earned NRs 2,500 to 3,000 per month and the average contribution to the total income was $60 \%$. Besides fishing, fishers were also involved in seed supply and marketing of farmed fish. Due to large number of fishers, competition was higher among fisher compared to fish traders.


Key words: Koshi River, fishers, fish diversity, fishing implements, fish catch

## Introduction

The Koshi River is the largest river of Nepal and is the largest tributary of the River Ganges. The Koshi Tappu Wildlife Reserve (KTWR) is an important wetland area in the Sapta Koshi River basin. The site consists of the natural river floodplains, including numerous low islands, extensive mudflats, oxbow lakes, freshwater marshes and grasslands. It is the only wetland area in Nepal of international importance (Thakur 2006). Large number of local communities residing in 16 different Village Development Committees (VDCs) spreading over three districts (Karki et al. 2006) depend on these wetlands for their livelihoods (Shrestha et al. 2006).

In Nepal, fish have wide distribution corresponding to variation in the climate and altitude. Directorate of Fisheries Development (2007) reported 127 species from Koshi River while Menon (1949) collected 11 families of fish comprising 26 genera and 52 species, Shrestha (1990) recorded 108 fish species and Yadav (2002) reported 96 fish species.

## Materials and methods

The study was carried out from July 2007 to February 2008 at Madhuvan and West Kusaha VDC of Sunsari district along with the Koshi River. The study covered deprived fisher community and fish traders of Koshi buffer zone area. Out of 65 fishers, $46 \%$ (i.e. 30 fishers) were selected from both VDCs for the household survey. Simple random sampling was used to select the fishers. To assess fish diversity, the species were collected during whole study period from Koshi River. Abundance and status of fish species were assessed through catch observation, interviews and discussions with fishers. The species were then categorized as common, uncommon, rare, and resident. Books related with fish identification were used as guidance for the identification of fish species. Also an experience of local fishers was given consideration during sampling of fish species for their local names. The collected fish species from Koshi River were
identified through morphometric measurements and meristic counts. The specimens were preserved in $10 \%$ formalin ( $40 \%$ formaldehyde) solutions. Both the primary and secondary data were analysed using Statistical Packages for Social Sciences (SPSS) and Microsoff Excel. Conventional analysis was done using frequencies and percentages to assess the socio-economic characteristics and income. Similarly, species distribution, species depletion, and status were analyzed using descriptive statistics.

## Results and discussion

## Distribution and size of land holding

The land holding size of the fishers varied from less than 0.05 ha to 1 ha. Average land holding size of fisher was 0.16 ha (Table 1).

Table 1. Distribution of land and land holding size at Madhuvan and West Kusaha VDC, Sunsari

| Description | Households (no.) | Percentage (\%) |
| :--- | :---: | :---: |
| Households with land | 22 | 73.3 |
| Households without land | 8 | 26.7 |
| Average land holding (ha/HH) | 0.16 |  |
| Landless | 8 | 26.7 |
| $<0.05$ ha | 7 | 23.3 |
| $0.05-0.1$ ha | 2 | 6.7 |
| $0.1-0.2$ ha | 4 | 13.3 |
| $0.2-0.5$ ha | 7 | 23.3 |
| $0.5-1$ ha | 2 | 6.7 |
| Above 1 ha | - | - |
| Total | 30 | 100 |

Involvement on fishing
There are various fisher communities in the buffer zone area. Among fisher community, Mallah, Godhi, Mukhiya, Bahardhar, Bin, Sardar, Jhagar are the
poorest among the poor and scattered. The Bahardar and Mukhiya communities represent the fisher communities in the area. Around $30 \%$ fishers adopted this business from their ancestor and they were involved since long time. Similarly, $63.3 \%$ had more than 5 years fishing experience, while very few fishers $(6.7 \%$ ) had less than 2 years experience. Majority of fishers in the study area are involved in fishing as part time job (50\%) followed by full time (20\%) and occasional (30\%).

## Household income source

Fishers in the study area were found to be involved in different occupational activities. Around $36.1 \%$ fishers had the main source of income from fishing and $3.4 \%$ fishers had both fishing and agricultural farming occupation. Fishing was the main source of income followed by wage earning (17.6\%). In the study area, the school going children were $13.7 \%$ (Table 2).

Table 2. Population distribution of the sampled households by occupation at Madhuvan and West Kusaha VDC, Sunsari

| Occupation | Fishers |  |
| :--- | :---: | :---: |
|  | Households (no.) | Percentage (\%) |
| Farming | 2 | 1 |
| Fishing and farming | 7 | 3.4 |
| Fishing/Fish farming | 74 | 36.1 |
| Fishing and fish business | 8 | 3.9 |
| Housewife | 5 | 2.4 |
| Study | 28 | 13.7 |
| Job | 0 | 0 |
| Business | 1 | 0.5 |
| Wage labor | 36 | 17.6 |
| Mat weaving | 7 | 3.4 |
| Abroad job | 0 | 0 |
| Unemployment | 37 | 18 |
| Total | 205 | 100 |

## Fishing implements and methods

The common fishing implements used by the sampled fishers were Cast net, Gill net, Duwalo, Tapi, and Fishing rod (Figure 5). The result showed that Cast net was the common fishing gear used by the fishers. All the sampled fishers used Cast net with addition to Gill net (86.7\%), Duwalo (6.7\%), Tapi (3.3\%), and Fishing rod $(43.3 \%)$. Fishers used Tapi to catch the fishes in ditches and it was not taken as important fishing gear by majority of fishers, most of the fishers had owned Cast net and Gill net.

## Fish diversity

For the study of fish diversity in the Koshi River, the site from Rajabas to Koshi barrage was selected for the sampling of fish. Total of 60 fish species were collected from Koshi River from July 2007 to end of February 2008. Fish species collected from various sites of this river belong to 7 order, 20 families and 40 genera. Classification of the fish species (order, family, genus, species) including their local name is presented in Table 3. Maximum fish species collected from Koshi River belonged to order Cypriniformes (49\%) and minimum from order Beloniformes (2\%), Osteoglossiformes (2\%) and Tetraodontiformes (2\%). Cypriniformes order dominated with 30 fish species followed by Siluriformes (13), Perciformes (11), Synbranchiformes (4), Beloniformes (1), Osteoglossiformes (1), and Tetraodontiformes (1) (Figure 1). Among the family, Cyprinidae dominated with 25 species followed by Bagridae (5), Channidae (4), Mastacembelidae (3), Cobitidae (3), Balitoridae (2), Nandidae (2), Ambassidae (2), Siluridae (2), Sisoridae (2), and Schilbeidae (1). The single families collected in Koshi River were Belonidae, Notopteridae, Belontidae, Heteropneustidae, Synbranchidae, Clariidae, Tetradontidae, Chacidae, and Gobiidae.

Table 3. Fish checklist of Koshi River

| $\begin{aligned} & \hline \mathrm{S} . \\ & \mathrm{N} . \end{aligned}$ | Order | Family | Genus | Species | Local name |
| :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | Beloniformes | Belonidae | Xenentodon (Reg.) | X. cancila (Ham.) | Kauwa machha |
| 2 | Cypriniformes | Balitoridae | Acanthocobatis (Peters) | A. botia (Ham.) | Gadelo |
| 3 | Cypriniformes | Balitoridae | Nemacheilus (Bleeker) | N. corica (Ham.) | Gadelo |
| 4 | Cypriniformes | Cobitidae | Botia (Gray) | B. Iohachata (Chaudhari) | Baghi |
| 5 | Cypriniformes | Cobitidae | Somileptes (Swai.) | S. gongota (Ham.) | Lata |
| 6 | Cypriniformes | Cobitidae | Lepidocephalus (Ble.) | L. guntea (Ham.) | Lata |
| 7 | Cypriniformes | Cyprinidae | Labeo | L. pangusia (Ham.) | Latani |
| 8 | Cypriniformes | Cyprinidae | Barilius (Ham.) | B. barna (Ham.) | Faketa |
| 9 | Cypriniformes | Cyprinidae | Garra (Ham.) | G. annandalei (Hora) | Buduna |
| 10 | Cypriniformes | Cyprinidae | Aspidoparia (Heckel) | A. morar (Ham.) | Bhegna |
| 11 | Cypriniformes | Cyprinidae | Chagunius | C. chagunio (Ham.) | Rewa |
| 12 | Cypriniformes | Cyprinidae | Crossocheilus (Vank.) | C. latius | Dhurla |
| 13 | Cypriniformes | Cyprinidae | Rasbora (Ham.) | R. daniconius (Ham.) | Khasara |
| 14 | Cypriniformes | Cyprinidae | Branchydanio (Weber) | B. rerio (Ham.) | Zebra |
| 15 | Cypriniformes | Cyprinidae | Barilius (Ham.) | B. guttatus (Day) | Jalkapoor |
| 16 | Cypriniformes | Cyprinidae | Tor | T. putitora (Ham.) | Sahar |
| 17 | Cypriniformes | Cyprinidae | Labeo | L. boga (Ham.) | Tilke |
| 18 | Cypriniformes | Cyprinidae | Danio (Ham.) | D. devario (Ham.) | Gairtirrhi |
| 19 | Cypriniformes | Cyprinidae | Puntius | P. sarana (Ham.) | Darhi |
| 20 | Cypriniformes | Cyprinidae | Puntius | P. chola (Ham.) | Pothi |
| 21 | Cypriniformes | Cyprinidae | Labeo | L. angra (Ham.) | Thed |
| 22 | Cypriniformes | Cyprinidae | Barilius (Ham.) | B. vagra (Ham.) | Hakrahi |
| 23 | Cypriniformes | Cyprinidae | Puntius | P. sophore (Ham.) | Pothi |
| 24 | Cypriniformes | Cyprinidae | Barilius (Ham.) | B. barila (Ham.) | Tile |


| 25 | Cypriniformes | Cyprinidae | Aspidoparia (Ham.) | A. jaya (Ham.) | Solhi |
| :---: | :---: | :---: | :---: | :---: | :---: |
| 26 | Cypriniformes | Cyprinidae | Labeo | L. gonius (Ham.) | Kursa |
| 27 | Cypriniformes | Cyprinidae | Labeo | L. rohita (Ham.) | Rohu |
| 28 | Cypriniformes | Cyprinidae | Tor | T. tor (Ham.) | Sahar |
| 29 | Cypriniformes | Cyprinidae | Cirrhinus | C. mrigala (Ham.) | Naini |
| 30 | Cypriniformes | Cyprinidae | Barilius (Ham.) | B. bendelisis (Ham.) | Fageta |
| 31 | Cypriniformes | Cyprinidae | Cirrhinus | C. reba (Ham.) | Rewa |
| 32 | Osteoglossiformes | Notopteridae | Notopterus <br> (Lacepede) | N. notopterus | Golhai |
| 33 | Perciformes | Ambassidae | Chanda (Ham.) | C. nama (Ham) | Nata |
| 34 | Perciformes | Ambassidae | Pseudambassis (Bleeker) | P.ranga (Ham.) | Chanda |
| 35 | Perciformes | Belontidae | Colisa (Cuv.) | C. fasciatus (Ham.) | Katara |
| 36 | Perciformes | Channidae | Channa | C. punctatus | Garai |
| 37 | Perciformes | Channidae | Channa (Scopoli) | C. orientalis (Bloch) | Chenga/Hile |
| 38 | Perciformes | Channidae | Channa (Scopoli) | C. striatus (Bloch) | Saura |
| 39 | Perciformes | Channidae | Channa (Scopoli) | C. marulius (Ham.) | Bhorha machha |
| 40 | Perciformes | Gobiidae | Glossogobius (Gill.) | G. giuris (Ham.) | Bulla |
| 41 | Perciformes | Nandidae | Badis (Bleek) | B. badis (Ham) | Jharki kotari |
| 42 | Perciformes | Nandidae | Nandus (Valen) | N. nandus | Dhalle |
| 43 | Siluriformes | Bagridae | Mystus (Scopoli) | M. cavasius (Ham.) | Tengra |
| 44 | Siluriformes | Bagridae | Aorichthys (Wu.) | A. aor (Sykes.) | Kanti |
| 45 | Siluriformes | Bagridae | Mystus (Scopoli) | M. bleekeri (Day.) | Tengra |
| 46 | Siluriformes | Bagridae | Mystus (Scopoli) | M. tengara (Ham.) | Tengri |
| 47 | Siluriformes | Bagridae | Aorichthys (Wu.) | A. seenghala (Sykes) | Gochara/Kan ti |
| 48 | Siluriformes | Chacidae | Chaca (Grey) | C. chaca (Ham.) | Khirkiri |
| 49 | Siluriformes | Claridae | Clarias (Scopoli) | C. batrachus | Magur |
| 50 | Siluriformes | Heteropneustidae | Heteropneustes (Muller) | H. fossilis (Bl.) | Singhi |
| 51 | Siluriformes | Schilbeidae | Eutropiichthys | E. vacha (Ham.) | Bachawa |

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|  |  | (Bleeker) |  |  |  |
| :--- | :--- | :--- | :--- | :--- | :--- |
| 52 | Siluriformes | Siluridae | Ompok (Lacepede) | O. bimaculatus <br> (Bloch.) | Pabata |
| 53 | Siluriformes | Siluridae | Wallago (Bleeker) | W. attu | Buhari |
| 54 | Siluriformes | Sisoridae | Gagata (Bleeker) <br> 55 | Siluriformes | Sisoridae |



Figure 1. Distribution of fish species of Koshi River collected during July to mid February 2008.

## Status of fish species

Fish fauna of Koshi River varied according to the seasonal calendar. Out of 60 species, 23 were common, 9 were uncommon, 17 were rare, and 11 were resident. Through the catch observation, interviews, and discussions with fishers the abundance and status of fish species were categorized (Table 4).

Table 4. Abundance and status of fish species of Koshi River

| Fish species | Abundance | Status |
| :--- | :--- | :--- |
| Acanthocobatis botia | Common | Small hill stream fish, found in June to Aug. |
| Aorichthys aor | Resident | Medium sized fish, found in July to Sept. |
| Aorichthys seenghala | Resident | Found during August to Nov. |
| Aspidoparia jaya | Common | Local migrants, found all year round. |
| Aspidoparia morar | Resident | Mostly found in December to June |
| Badis badis | Rare | Mostly found in June to August. |
| Barilius barna | Resident | Mostly found in July to November. |
| Barilius barila | Resident | Small sized fish, found in May to Sept. |
| Barilius bendelisis | Common | Small sized fish, found in May to Sept. |
| Barilius guttatus | Resident | Found in June to October. |
| Barilius vagara | Resident | Small sized fish, found in May to Sept. |
| Botia lohachata | Common | Hill stream fish, found in July to Sept. |
| Branchydanio rerio | Common | Found mostly during Oct. to January. |
| Chaca chaca | Rare | Ugly fish, found rarely. |
| Chagunius chagunio | Uncommon | Common food fish, found in June to Aug. |
| Chanda nama | Uncommon | Small sized fish, found in Oct. to Jan. |
| Channa marulius | Uncommon | Large sized fish, found all year round. |
| Channa orientalis | Rare | Small sized fish, found all year round. |
| Channa punctatus | Rare | Medium sized fish, found all year round. |
| Channa striatus | Uncommon | Medium sized fish, found all year round. |
| Cirrhinus mrigala | Rare | Important food fish. |
| Cirrhinus rewa | Common | Important food fish, found in June to Sept. |
| Clarias batrachus | Rare | Common food fish, found all year round. |
| Colisa fasciatus | Rare | Migratory fish, found in July to Sept. |
| Crossocheilus latius | Common | Mostly found in July to Nov. |
| Danio devario | Common | Found mostly during October to Jan. |

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| Eutropichthys vacha | Uncommon | Mostly found during August to Nov. |
| :--- | :--- | :--- |
| Gagata cenia | Uncommon | Medium sized fish. |
| Garra annandalei | Resident | Hill stream fish, mostly found in June to Sept. |
| Glossogobius giuris | Common | Migratory fish, mostly found in July to Sept. |
| Glyptothorax pectinopterus | Uncommon | Hill stream fish, mostly found in June to Sept. |
| Heteropneustes fossilis | Rare | Mostly found all year round. |
| Labeo angra | Common | Mostly found in June to Aug. |
| Labeo boga | Common | A common food fish. |
| Labeo gonius | Common | Mostly found in August to Dec. |
| Labeo pangusia | Rare | Mostly found in July to November. |
| Labeo rohita | Rare | Mostly found in June to Aug. |
| Lepidocephalus guntea | Common | Mostly found in May to June. |
| Macrognathus aral | Uncommon | Medium sized fish found in stagnant pool. |
| Macrognathus pancalus | Common | Mostly found during September to Dec. |
| Mastacembelus armatus | Common | Medium sized fish found in stagnant pool. |
| Monopterus cuchia | Common | Muddy fish, Found during July to Nov. |
| Mystus bleekeri | Resident | Small sized fish found during June to Nov. |
| Mystus cavasius | Resident | Small sized fish found during June to Nov. |
| Mystus tengara | Resident | Mostly found in May to August. |
| Nandus nandus | Rare | Found in stagnant water pools in June to Nov. |
| Nemacheilus corica | Rare | Mostly found in May to June. |
| Notopterus notopterus | Rare | Mostly found from July to November. |
| Ompok bimaculatus | Uncommon | Medium sized fish. |
| Parambassis ranga | Rare | Found in stagnant water pools in June to Nov. |
| Puntius chola | Common | Small sized fish common everywhere. |
| Puntius sarana | Common | Small sized fish common everywhere. |
| Puntius sophore | Common | Small sized fish common everywhere. |
| Rasbora daniconius | Common | Mostly found in July to September. |
| Somileptes gongota | Common | Mostly found in May to June. |
| Tetraodon cutcutia | Rare | Small sized fish. |
| Tor putitora | Rare | Migratory, mostly found in July to Sept. |
| Tor tor | Rare | Migratory, mostly found in July to Sept. |
| Wallago attu | Common | Large sized fish, found in July to Dec. |
| Xenentodon cancila | Common | Migratory fish, found all year round. |
|  |  |  |

## Fish catch

Maximum catch of fish was found in the month of September to November and minimum in February to July. In these months, the fishers catch the fish 1 kg to more than $3 \mathrm{~kg} /$ day, whereas minimum catch in Koshi River was found 0.5 to 1 kg .day during February to July. Average fish catch was decreasing or almost no catch during December to January; it may be due to the cold weather when the fishermen feel difficulty to get inside the water.

## Reasons of fish species depletion

The fish in the Koshi River is decreasing day by day due to several factors (Figure 2). According to the fishers in the study area the main reason of fish species depletion in Koshi River is poisoning and haphazard catch (46.7\%), followed by poisoning and over fishing (20\%), over fishing (16.7\%), haphazard catch (10\%), and poisoning (6.7\%). Among the several traditional methods of poisoning, use of extracts from local plants is common. Illegal methods of fishing i.e. use of explosives, electro fishing, and poisoning were mostly used by the non-fisher groups in the study area.


Figure 2. Reasons of fish species depletion in Koshi River.

## Income status

## Total income

The result showed that majority of fishers earned NRs. 2,500 to 3,000 (53.3\%) and 2,000 to $2,500(26.7 \%)$ per month. While, other $20 \%$ fishers' earning varied from NRs. 1,000 to 5,000 per month.

## Contribution of fish in total income

The average contribution of fishing to the total income of fishers was $60 \%$. Other sources of income which contributed to their total income were; agriculture (15\%), business (13\%), and wage labour (6\%). None of the fishers had any contribution by abroad job and services.

## Income distribution

Gross annual income per fisher varied from NRs.10,000 to 65,000 (Table 5).

Table 5. Distribution of Yearly Income of fishermen in Madhuvan and West Kusaha VDC, Sunsari (in 000 'NRs.)

| Income level <br> per year (NRs.) | Mid point | Number of | Percent of | Percent of | Cumulative percent |  |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| respondents | respondents | income | Respondent | Income |  |  |
| 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| $10-40$ | 25 | 4 | 13.3 | 8.7 | 13.3 | 8.7 |
| $40-45$ | 42.5 | 11 | 36.7 | 14.8 | 50 | 23.5 |
| $45-50$ | 47.5 | 7 | 23.3 | 16.5 | 73.3 | 40 |
| $50-55$ | 52.5 | 2 | 6.7 | 18.3 | 80 | 58.3 |
| $55-60$ | 57.5 | 4 | 13.3 | 20 | 93.3 | 78.3 |
| $60-65$ | 62.5 | 2 | 6.7 | 21.7 | 100.00 | 100. |
|  |  |  |  |  |  | 00 |
|  | 287.5 | 30 | 100 | 100 |  |  |

## Marketing system

The fishers and fish traders were main performer of the marketing system in study area. The fishers were also found to be involved in selling activity at the fishing sites. Most of the fishers brought their collections at the collection center, after that fish traders were involved in collecting, buying, grading, transporting, and financing. There were numerous fishers in the river and hence had to face a high competition among them whereas the traders were few in numbers and had major influence in marketing. The fishers were found to be selling their collection directly to the local consumers and fish traders. The fishers sold small sized fish in local market and large sized in distance market or to the fish traders.

## Marketing channel and market margin

The length of marketing channel was found to depend on the quantity of fish to be moved, consumer demand and collection of fish. The fish supplied through various channels to the main market from the collection center. Collector, supplier, wholesalers, retailers, and consumers were main actors in the channel. The consumer demand of Koshi fish is very high in Biratnagar and Dharan fish market. Average fish supplied in Biratnagar and Dharan fish market was $20-100 \mathrm{~kg}$ /day and $20-50 \mathrm{~kg}$ /day, respectively. In Biratnagar fish market, the wholesalers fixed the price of fish through auction and sell the fish to the retailer.

## Market efficiency

A comparison of efficiency of different marketing channel is presented in Table 6. Among the three marketing channels, the index of marketing value was observed highest (i.e. 1.18) in Biratnagar market (channel c). Therefore this marketing channel was most efficient one because of low marketing cost and margins. However, it was observed that among the three marketing channels, the fish supplied to both Dharan and Biratnagar market (channel b) was the dominant. The existing marketing scenario showed that the fish collectors were found to receive low share on consumer's rupees compared to middle men.

Table 6. Measurement of marketing efficiency of Koshi fish in buffer zone area during 2007

| Location of fish <br> market | Per unit <br> cost <br> (NRs./kg) | Per unit <br> margin <br> (NRs./kg) | Total selling <br> price <br> (NRs./kg) | Producer's <br> price <br> (NRs./kg) | Modified <br> marketing <br> efficiency |
| :--- | :---: | :---: | :---: | :---: | :---: |
| Dharan (a) 16 104 250 130 1.08 <br> Both Dharan and <br> Biratnagar (b) 20 100 250 130 1.08 <br> Biratnagar (c) 15 95 240 130 1.18${ }^{2}$ |  |  |  |  |  |

## Conclusions

Fisher households are the most dependent on wetland resources and Koshi River, and have minimal access to alternative sources of income. It is necessary to provide alternative sustainable livelihood options for fishers' community with adequate support to conserve the ecological resources and fish diversity in Koshi River. Though, this study period was less than a year and is not sufficient to explain the species diversity, composition and frequency of occurrence with seasonal changes, it provides ample data and information in fisheries and aquaculture of the buffer zone area. However, it is recommended to carry out such study at least for complete one-year cycle to observe monthly and seasonal variations.

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Estimation of fish catch and maximum sustained yield in a part of Kali Gandaki River

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#### Abstract

In order to compare fish catch and maximum sustained yield (MSY) between upstream and downstream of Kali Gandaki hydro-dam, daily fish catch data was collected from July 2011 to June 2012. The fish yield in upstream based on Henderson and Welcomme's, and Ryder's morphoedaphic model was $42.75 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ and $6.38 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$, respectively. The actual fish yield estimated from daily fish catch data was $6.26 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$, which was very close to later model. Similarly, the fish yield in downstream based on Henderson and Welcomme's, and Ryder's morphoedaphic model was 78.99 and 11.79 $\mathrm{kg} / \mathrm{ha} / \mathrm{y}$, respectively, but the actual fish yield estimated from daily fish catch data was $9.68 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$. In upstream, gardi was the dominant fish (54.03\%) followed by katle ( $35.87 \%$ ), sahar ( $6.07 \%$ ) and others ( $4.03 \%$ ), respectively. In downstream too gardi was the dominant fish (74.23\%) followed by jalkapur $(12.61 \%)$, lahare ( $5.04 \%$ ), and others ( $8.12 \%$ ), respectively. MSY value was $123 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ and $10 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ in upstream and downstream, respectively. However, the resulted MSY value was doubfful and must be reconfirmed by the further study.


Keywords: fish catch, maximum sustained yield, morphoedaphic model, fishing effort

## Introduction

Kali Gandaki River originating from Tibet plateau is one of the big rivers of Narayani River basin. The river has formed the world's deepest gorge between Dhaulagiri and Annapurna. It joins Andhi stream at Mirmi of Syangia district. The dam was built across Kali Gandaki River at Mirmi forming a reservoir of 65 ha with depth ranging from 14 to 20 meter. The river joins the Trisuli River at Devighat and is then called Narayani River. It is 47 km long from the dam site to powerhouse. The river is about 120 km long from the point of its appearance to Mungling. It is one of the seven rivers of Gandaki River system and finally flows down to Narayani River.

Multi species of indigenous fish exist in Kali Gandaki River and its tributaries (Edds 1986). Number of fish species inhabiting the various river systems increases with the increasing size of the river and river basin (Welcomme 1985). This is because the number of ecological niches remains greater in larger river systems than in small ones. It is necessary to have information on existing fish species, fish catch, catch per unit effort and fish yield per unit area for planning the fisheries management. Morphoedaphic index (MEI), which predicts potential fish harvest as a power function of total dissolved solids divided by mean depth, is regarded as a model of fish production systems. Henderson et al. (1973), and Henderson and Welcomme (1985) have demonstrated MEI as an estimation of fish yield and standing crop in reservoirs. The applicability of MEI to estimate fish yield in river is still unknown. The main objectives of the study was to determine fish yield based on MEI model and compare its value with the actual fish catch data collected from up and downstream of Kali Gandaki hydro-dam and to estimate the maximum sustained yield.

## Materials and methods

Fish catch data were collected from the commercial fishermen all the year round from July 2011 to June 2012. Collected fish were sorted out according to species, and respective fish species were counted and weighed. Fishing effort
was expressed as fishermen because fishing gears varied from cast net, gill net, lift net to long lined loop and angling rod.
In upstream, the fishing area included Mirmi Reservoir and Andhimuhan area and covered nearly 80 ha. The annual average depth was 17 m . In downstream, the fishing area covered 50.16 ha of Kali Gandaki River from Thulobagar to Tipua. The annual average depth was 5.2 m . Estimated fish yield was computed using MEI of Ryder (1965);
MEI=T/Z Yield $=2 \sqrt{ }$ MEI

Where, T designated as the total dissolved solids, more conveniently calculated as the product of 0.65 and conductivity ( $\mu \mathrm{mhs} / \mathrm{cm}$ ) and $Z$ the average depth in meter. Henderson and Welcomme's morphoedaphic model was used to calculate fish catch (C) expressed as $\mathrm{kg} / \mathrm{ha} / \mathrm{y}$;
$\mathrm{C}=14.3136 \times \mathrm{MEI}^{0.4681}$

Conductivity was measured every month at different depths to obtain the mean value. The average depth was determined measuring the mean cross sections of the river.

The maximum sustained yield (MSY) and optimum fishing effort (OFE) were calculated using the formula;
MSY $=a^{2} / 4 b$ and OFE $=a / 2 b$

Where, $a$ and $b$ were intercept and slope of catch effort curve.

## Results and discussion

The mean conductivity of upstream Mirmi Reservoir area was $266 \mu \mathrm{mhos} / \mathrm{cm}$ and the mean depth was 17 m . Based on the MEI model (Henderson and Welcomme 1974) the fish catch in the upstream was $42.75 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ whereas the yield was $6.38 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ based on Ryder's model. The actual fish catch was
$6.26 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ based on daily collected data, which was very close to the Ryder's model.

The mean conductivity of downstream area was $278 \mu \mathrm{mhos} / \mathrm{cm}$ and the mean depth was 5.2 meter. Based on the MEI model (Henderson and Wellcomme 1974) the fish catch was $78.99 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$. On the contrary, the yield was 11.79 $\mathrm{kg} / \mathrm{ha} / \mathrm{y}$ based on Ryder's model which was close to the actual fish catch of $9.68 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ calculated based on daily fish catch data.

Catches obtained from 6 flood plain rivers in Asia ranged from 24.17-78.17 $\mathrm{kg} / \mathrm{ha} / \mathrm{y}$ (Welcomme 1985) which was higher than the fish catch obtained both in upstream and downstream in the present study. Joshi and Nepal (2004) also reported higher fish yield from morphoedaphic model, actual fish yield and maximum sustained yield were $61 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}, 53 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ and 93 $\mathrm{kg} / \mathrm{ha} / \mathrm{y}$, respectively from Riialghat, a part of Trishuli River than calculated in the present study. Though fish yield of $6-9 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ seemed low but the production in the reservoir increases over the pre-impoundment conditions. The fish catch increased with the increased efforts. Both catches and efforts remained highest from October to March (Table 1). The highest catch and effort in those particular months was attributable to lower water velocity and upstream moving of fishes during spawning period. Fishing efforts declined in the monsoon due to heavy flood and the catch dropped accordingly. The average catch per unit effort was $1.2 \mathrm{~kg} / \mathrm{head} / \mathrm{y}$ which was very low compared to that of downstream ( $4.4 \mathrm{~kg} / \mathrm{head} / \mathrm{y}$ ). In downstream, both catches and efforts remained highest from March to April (Table 2) corresponding to lower water velocity and upstream migration.

Table 1. Fish catch data collected from Mirmi Reservoir used to estimate maximum sustained yield and optimum fishing effort, 2011/2012.

| Months | Catch (Number) | Catch (Kg) | Effort (No. of gears) | Catch/effort |
| :--- | :--- | :--- | :--- | :--- |
| July | 5 | 18 | 5 | 3.60 |
| August | 22 | 17 | 6 | 2.83 |
| September | 82 | 28 | 77 | 0.36 |


| October | 468 | 78 | 397 | 0.20 |
| :--- | :--- | :--- | :--- | :--- |
| November | 62 | 16 | 44 | 0.36 |
| December | 186 | 39 | 157 | 0.25 |
| January | 370 | 94 | 382 | 0.25 |
| February | 97 | 27 | 59 | 0.46 |
| March | 214 | 71 | 197 | 0.36 |
| April | 85 | 22 | 77 | 0.29 |
| May | 66 | 27 | 59 | 0.46 |
| June | 105 | 64 | 57 | 1.12 |
| Total | 1762 | 501 | 1517 |  |
| $\mathrm{Y}=1.405-0.004^{*} \mathrm{X}$ |  |  |  |  |
| $\mathrm{Y}=\mathrm{a}-\mathrm{bX}$ |  |  |  |  |
| $\mathrm{a}=1.405$ |  |  |  |  |
| $\mathrm{~b}=-0.004$ |  |  |  |  |
| Optimum fishing effort $\mathrm{f}=\mathrm{a} / 2 \mathrm{~b}=176$ |  |  |  |  |
| $\mathrm{MSY}=\mathrm{a}^{2} / 4 \mathrm{~b}=123 \mathrm{~kg} / \mathrm{ha} / \mathrm{yr}$ |  |  |  |  |

In upstream, actual fishing efforts used was 1517. More use of gill nets with large mesh size than cast net with small mesh size and hooks might have builtup dense population of small fishes. Application of higher fishing efforts might overexploit fish resources but the exploitation rate was quite reasonable to maintain the desired fish population in the natural water. In downstream, actual fishing efforts used were 110 .

The concept of maximum sustained yield (MSY) is the optimum number of fish caught from the population without long-term changes in the fisheries management. MSY value was $123 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ in upstream (Table 1). The actual fish yield was very low, just $6.26 \mathrm{~kg} / \mathrm{ha} / \mathrm{yr}$ which was far below the value of MSY, an indication of under exploitation. In downstream, MSY value was 10 $\mathrm{kg} / \mathrm{ha} / \mathrm{y}$ but the actual fish yield was $9.68 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$, similar to MSY value.

Table 2. Fish catch data collected from Thulobagar to Tipuwa used to estimate maximum sustained yield and optimum fishing effort, 2011/2012

| Months | Catch <br> (Number) | Catch <br> $(\mathrm{Kg})$ | Effort <br> (No. of gears) | Catch/effort |
| :--- | :--- | :--- | :--- | :--- |
| July | 11 | 11 | 9 | 1.22 |
| August | 10 | 10 | 8 | 1.25 |
| September | 10 | 11 | 7 | 3.00 |
| October | 11 | 11 | 3 | 3.67 |
| November | 5 | 10 | 5 | 2.00 |
| December | 0 | 0 | 0 | 0.00 |
| January | 53 | 16 | 7 | 2.29 |
| February | 0 | 0 | 0 | 0.00 |
| March | 487 | 178 | 38 | 4.68 |
| April | 107 | 102 | 10 | 10.20 |
| May | 16 | 112 | 20 | 5.60 |
| June | 4 | 13 | 3 | 4.33 |
| Total | 714 | 484 | 110 |  |
| $\mathrm{Y}=2.185+0.109^{*} X$ |  |  |  |  |
| $\mathrm{Y}=\mathrm{a}-\mathrm{bX}$ |  |  |  |  |
| $\mathrm{a}=2.185$ |  |  |  |  |
| $\mathrm{~b}=0.109$ |  |  |  |  |

Optimum fishing effort $f=a / 2 b=10$
MSY $=a^{2} / 4 b=10 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$
Regarding the species composition, gardi was the dominant fish contributing $54.03 \%$ followed by katle ( $35.87 \%$ ), sahar ( $6.07 \%$ ) and others ( $4.03 \%$ ) to the total catch in upstream. In downstream also, gardi was the dominant fish (74.23\%) followed by jalkapur (12.61\%), lahare (5.04\%), and others (8.12\%), respectively (Table 3). The reason might be regular stocking of gardi fingerlings to the river as a part of fisheries enhancement by Nepal Agricultural Research Council. Out of the 157 fish species reported from the Kali Gandaki River System, 57 species were collected from the Kali Gandaki River during different level of study conducted by the Kali Gandaki "A" Hydroelectric Project (Shrestha and Chaudhary 2004). Wagle et al. (2000) collected 24 fish species
under 7 families from Kali Gandaki and Aandhi Khola near Kali Gandaki hydroelectricity dam, out of them 10 species were commonly available, 12 species were fairly common and 2 species were rarely available according to local fishers.

The estimated fish density based on collective catch data in upstream and downstream was $22 /$ ha and $14 / \mathrm{ha}$, respectively. Fish stocking in the reservoir ranges from 100-500 depending upon the natural food availability (Sreenivasan 1977). Fish density seemed quite low for natural water body. These resulted data must be reconfirmed by the further study.

Table 3. Species composition of fishes catches in upstream and downstream

| Species | Fish catch |  |  |  |  |
| :--- | :--- | :--- | :--- | :--- | :---: |
|  | Upstream |  |  | Downstream |  |
|  | Number | $\%$ | Number | $\%$ |  |
| Gardi (Labeo dero) | 952 | 54.03 | 530 | 74.23 |  |
| Hade (Labeo pangusia) | 16 | 0.91 | 0 | 0 |  |
| Katle (Neolissocheilus hexagonolepis) | 632 | 35.87 | 4 | 0.56 |  |
| Sahar (Tor putitora) | 107 | 6.07 | 10 | 1.4 |  |
| Asala (Schizothorax spp.) | 54 | 3.06 | 0 | 0 |  |
| Raibam (Anguilla bengalensis) | 0 | 0 | 26 | 3.64 |  |
| Gonch (Bagarius yarelli) | 0 | 0 | 17 | 2.38 |  |
| Jalkapur (Pseudeutropius murius) | 1 | 0.06 | 90 | 12.61 |  |
| Lahare (Garra annandalel) | 0 | 0 | 36 | 5.04 |  |
| Bhedra (Glyptothorax cavia) | 0 | 0 | 1 | 0.14 |  |
| Total | 1762 | 100 | 714 | 100 |  |

## Conclusion

This study concluded that actual fish yield was higher ( $9.68 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ ) in downstream than in upstream ( $6.26 \mathrm{~kg} / \mathrm{ha} / \mathrm{y}$ ) based on daily fish catch data. Regarding the species composition, gardi was the dominant fish species in both upstream and downstream of Kali Gandaki hydro-dam.

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# Response of fish species to riverbed extraction in the Tinau River, Nepal 

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#### Abstract

In order to assess the effects of gravel extraction on the species diversity and abundance of fish species, an experiment was carried out in the Tinau River, which is one of the major spring-fed rivers flowing through Siwalik piedmont zone of Nepal. The effects on the species richness and abundance of fish species in this river before and after the extraction were compared based on past studies and sampling carried out in November 2010 (Post-monsoon) and June 2011 (Pre-monsoon) by using electro-fishing gear. The study was focused on the effects of riverbed extraction on fish fauna in the impact zone. The sampling was carried out in three different zones, reference zone, impact zone, and recovery zone. The research showed that the reference and recovery zones were suitable for fish habitat, whereas the impact zone was unsuitable for the


fish fauna to survive owing to the habitat destruction. The research also showed that both species richness and abundance of fishes were significantly reduced in the impact zone as compared to the reference and recovery zones.

Keywords: Tinau River, riverbed materials extraction, fish habitat destruction, impacts

## Introduction

According to origin, water availability and stream order, The Tinau River is class-II, perennial and third order stream (Dahal et al. 2012). The river originates from Lemthem Phant of Palpa district. The highest elevation of its watershed is 1496 m (Okharkot), the highest spring source is originated from 1200 m elevation and the lowest elevation is 175 m in plain Terai. The total length of this river is 95 Km and basin area of Tinau River is $1081 \mathrm{Km}^{2}$. The basin area extends from $27^{\circ} 15^{\prime} \mathrm{N}$ to $27^{\circ} 45^{\prime} \mathrm{N}$, and $83^{\circ} 15^{\prime} \mathrm{E}$ to $83^{\circ} 45^{\prime} \mathrm{E}$. The width of this river varies from place to place. Generally the width is very small in upstream and it is wider in the downstream. The average annual flow of this river is $24 \mathrm{~m}^{3} / \mathrm{s}$. The main tributaries of Tinau are: Lakuri Khola, Pasdi Khola, Pugdi Khola, Budhare Khola, Sukhaura Khola, Rithe Khola, Bhobadi Khola, Sisne Khola, Khadal Khola, Hulandi Khola, Suridi Khola, Bhaiskatta Khola, Jhumsa Khola, and Dhoban Khola (Kharel 2002; Guragain 2012). The majority of these tributaries join the main Tinau River before the confluence of Dobhan Khola. Then the river flows from north to south through a gorge section of Churia hills before entering into the Terai plain at Butwal. In this reach, the slope of the river is significantly steep. As the river debouches into Terai plain, it spreads into a wet piedmont fan and splits into two distributaries viz. Tinau and Dano (Gyawali and Dixit 1999; Guragain 2012).

In the past decades there were many fish species and number along the length of the Tinau River. Thus, many fishers were engaged to capture fishes through local fishing implements (Sharma 1996). However, the fish species and number has decreased in these days and the fishers' livelihood has adversely impacted (Dahal et al. 2013). There may be various causes of reduction in fish species
and abundance but one of the major can be the excessive gravel extraction and destruction of fish habitat which cause the reduction in species and number of fishes (Kondolf 1998; http://www.threeissues.sdsu.edu; http://www.davidsuzuki.org; http://www.academia.edu). Although the history of gravel extraction in the Tinau River dates back to many decades, the devastating effects of gravel extraction started affer the dawn of democracy and particularly after the enforcement of local self-governance Act and Regulation (Local Self Governance Act 1999 and Regulation 2000). The local selfgovernance Act and Regulation has provided the right for the use of local resources such as riverbed materials for their development. Therefore, local administrative bodies like Village Development Committees (VDCs), Municipalities and District Development Committees (DDCs) started to extract the riverbed materials massively throughout the country. These local bodies started to award the contracts for the extraction of riverbed materials for their fund rising. The Rupandehi DDC also started to award the contracts for extraction of riverbed materials in the Tinau River after the promulgation of the Decentralization Act.

Human interventions in the river environment like gravel extraction directly affects the fish habitats. It spoils the spawning ground for fishes, blocks the migratory path by creating the river fragmentation (http://www.academia.edu). This type of disturbances could be observed in the Tinau River too. Riverbed extraction induces lowering of ground water table, creates disturbance in the aquatic habitat of wetlands, change in river morphology and loss of aquatic habitat as a whole through bank erosion, slope instabilities, river incision, head cutting, and damage to river equilibrium (USEPA 1995; Trites and Suzanne 2009).

Considering the aforesaid overall impacts of in-stream gravel mining, researchers must play their role to seek the best alternatives of use of gravel as construction materials. Similarly, they have a duty to conserve the existing nature and natural resources in a sustainable way for the future generations. The main objective of this study is to find the effects of gravel extraction on fish
species diversity (species richness and abundance) at various locations of the Tinau River with varying scale of activities, and to compare the variations on fish species diversity before and after gravel mining in the Tinau River.

## Materials and methods

Tinau River originates from the Mahabharat Mountain in Palpa district and passes through the Terai plain of Nepal before joining to the West Rapti River in India. The total length of the selected reach of the river is about 28 Km (Figure 1). Three sampling stations (sites) named as Reference Zone (Site A), Impact Zone (Site B) and Recovery Zone (Site C) were fixed along the 28 km length of the river (Figure 1).


Figure 1. Sampling sites in Tinau River showing three zones, viz. reference zone, impact zone and recovery zone.

The length of reference zone is 2 Km , impact zone is 25.54 Km and recovery zone is 1 Km (Figure 1). However, the selected reach length for the sampling for this study for reference zone 1 Km ., for the impact zone 1 Km ; and for the recovery zone 500 m were considered. Thus, the distance from site $A$ to $B$ is 1 Km , site B to C is 25.54 Km . Global Positioning Systems (GPS) was used to determine the geographical locations of the selected sites. Geographical
location of site A is $83^{\circ} 27^{\prime} 55^{\prime \prime}-83^{\circ} 27^{\prime} 47^{\prime \prime}$ (longitude), $27^{\circ} 42^{\prime} 34^{\prime \prime}-27^{\circ} 47^{\prime} 13^{\prime \prime}$ (latitude); site B is $83^{\circ} 27^{\prime} 47^{\prime \prime}-83^{\circ} 27^{\prime} 21^{\prime \prime}$ (longitude), $27^{\circ} 41^{\prime} 42^{\prime \prime}-27^{\circ} 41^{\prime} 11^{\prime \prime}$ (latitude) ; and site $C$ is $83^{\circ} 25^{\prime} 37^{\prime \prime}-83^{\circ} 24^{\prime} 06^{\prime \prime}$ (longitude), $27^{\circ} 52^{\prime} 30^{\prime \prime}-$ $27^{\circ} 30^{\prime} 49^{\prime \prime}$ (latitude).

Sampling was carried out in three zones $A, B$ and $C$ by using electro-fishing gear (Honda, G* V50). Reference, impact and recovery zones were separated by using USEPA protocol. Site A was the reference zone (less disturbed site, upstream of Tinau bridge at Butwal); site $B$ was the impact zone downstream of Tinau bridge at Butwal; and site $C$ was the recovery zone, downstream of Tinau bridge at Bethari (where the riverbed extraction ends) (Figure 1). The sampling team was composed of 5 members. In each site, the fish sampling was done in two runs; run 1 and run 2 respectively. The time span for each run was 20 minutes. Catch per Unit Effort (CPUE) was taken as the abundance, which was estimated as the number of fish captured in 10 minutes of sampling. The equipment like grip net for catching shocked fishes, bucket for collection of captured fishes and weighing machine were used during sampling at sites. Consistent sampling design for each site in each season (i. e. pre and post monsoon were chosen for this study) was applied to avoid biased results. The numbers of sampling sites were 3 and numbers of samples collected were 6. All captured fishes were identified in the field with the help of expert. The statistical analysis of the fish samples was done by using SPSS-15. The first sampling (i.e. post -monsoon) was done in November 2010, and the second sampling (i.e. pre-monsoon) in June 2011. The evaluation of existing fish species was done on the basis of fish species richness and abundance (CPUE) of the fish species compared with the previous studies conducted by Sharma (1996) and Jha (2006b). Chi - square $\left(\chi^{2}\right)$ test was carried to find out the yearly relationship with fish catch. Similarly, Two-way ANOVA was conducted to find out the differences of fish composition among different sites and seasons.

## Results

The result of the study is presented in the figures below (Figure 2, 3, 4, 5, 6 and 7) according to seasons and sites. These figures signify the fish species and
number of fishes captured in pre and post monsoon at sites $\mathrm{A}, \mathrm{B}$ and C respectively. Similarly, site wise fish species and abundance at site $A, B$ and $C$ is presented in Figure 8. The result showed that the total numbers of fish species as well as total number of catch were lower in the impact zone compared to the reference and recovery zones.


Figure 2. Fish species and number at site $A$ in pre-monsoon


Figure 3. Fish species and number at site A in post-monsoon


Figure 4. Fish species and number at site $B$ in pre-monsoon


Figure 5. Fish species and number at site $B$ in post-monsoon


Figure 6. Fish species and number at site C in pre-monsoon


Figure 7. Fish species and number at site C in post-monsoon


Figure 8: Fish species and abundance at site $A, B$ and $C$ in pre and post monsoon

Chi-square $\left(\chi^{2}\right)$ test revealed that there was a significant relationship of different fish species with the year in the impact zone ( $\chi^{2}=19.44$, d.f. $=4, p<0.05$ ) while comparing present study (2011) with the previous study in 2005. Two way ANOVA showed significant variation in fish species at different sites ( $\mathrm{F}=$ 119.23, d.f. $=2, p<0.05$ ) with an indication that the impact zone had low number of fish species. However, there was no significant difference of fish species between pre- and post-monsoon seasons.

## Discussion

Jha (2006b) collected fish samples from two locations (upstream and downstream of Butwal city) along the Tinau River between the year 2004-2005 and reported 33 numbers in upstream and 50 numbers in downstream with equal species richness in both the cases during pre-monsoon. The major fish species in the impact zone during that period (2004-2005) in pre-monsoon were Barilius barila, Puntius conchonius, Puntius sophore, Schistura beavani and Lepidocephalus guntea (Jha 2006b). The fish species captured in this study was Barilius vagra only in pre-monsoon. Similarly, fish species captured in the year 2004-2005 at site B in post-monsoon were Barilius barila, Botia lohachata, Channa punctatu, Garra gotyla, Lepidocephalus guntea, Pseudechaeneis conchonius, Puntius sophore, Schistura beavani and Crossocheilus latius (Jha 2006b). In this study, the captured fish species were Barilius vagra, Garra gotyla, Puntius sarana, Puntius sophore, and Schistura beavani in post-monsoon.

Fish species and numbers were reduced in the impact zone (site $B$ ) as compared to the past decade. The massive extraction has been occurring in this site and fish habitat has been completely destructed in this stretch of the Tinau River. Therefore, the reduction in fish species and abundance in this stretch of the river might be due to the over extraction of riverbed materials. During survey of this study river fragmentation could be observed at site B (Impact zone). The existence of river fragmentation ultimately disturbs the river continuum process thereby reducing fish migration (Jungwirth 1998). Ultimately, there is a reduction in fish species and abundance.

Extraction of construction materials can have various costly effects near the mining sites and even far. Fertile land of stream sides, timber resources and wildlife habitat of riparian areas are lost due to over extraction. Furthermore, such activities degrade the aquatic habitat. Ultimately it causes the loss in fisheries productivity, aesthetic beauty, bio-diversity and recreational potential of the river (USEPA 1995). Mining of riverbed materials improves in flood control and channel stability; however, it results in destruction of aquatic and
riparian habitats notably (Kondolf 1993). It also has impacts on velocity, discharge, roughness of bed, turbidity, temperature, sediment transportation capacity, bed elevation, and sub-stratum composition. Any modification of above mentioned parameters can cause changes in channel configuration and flood paths of the river (Kondolf 1993). This fact as stated by Kondolf, and GSI, could be observed in the Tinau River clearly.

Anthropogenic activities on river banks like rock mining, extraction of sand, stone and boulders affect fish habitat quality and pose major threat to fishes. Similarly deforestation and removal of bank vegetation result in drastic changes of stream habitat eliminating shades for the aquatic biota. In the same way, removal of tree canopy, on the other hand, reduces driffing material such as fruits, vegetables, insects that bring changes in the bio-climatic conditions of the river (Shrestha 2008). Furthermore, it changes ichthyo-fauna. The significance of biological integrity is now viewed as the most important life supporting system on earth as it refers to a balanced, integrated, adaptive system too (Jha 2006a), however, it could not be found in the Tinau River. Thus the extraction of riverbed materials from the Tinau River without considering its effects has degraded the aquatic habitat and lost fish species richness and number and as whole river integrity.

Comparison of past studies with the present fish species richness and abundance Jha (2006b) reported (from the same site) much higher species, number and abundance - The species and number has reduced during this study period (2011). The comparison of past study and present study is presented in Figure 9.


Figure 9. Seasonal comparison on fish diversity and abundance between the present study (2011) and Jha (2006).

Seasonal variations in the species composition of fishes showed high faunal diversity in post-monsoon season in the upstream section of the Tinau River with dominance of Garra annanadalei, whereas midstream section of the river (impact zone) showed drastic fall in both species richness as well as abundance of fish fauna. During the post monsoon there is a moderate rise in species composition. The lowermost stretch of the river also showed high fish diversity during post-monsoon, but with poor abundance in comparison to the fish captured in pre-monsoon.

There may be various causes of decline in fish species, however one of the most important and major causes is the over extraction of riverbed materials leading to habitat destruction. Rupandehi DDC allowed extraction of riverbed materials from the Tinau River exceeding its replenishment rate (Figure 10).


Figure 10. Riverbed extraction during the period of 2004-2011 (Source: DDC, 2011) modified by Dahal et al. (2012).

Tinau being a potential for riverbed materials, mining of these materials started (in a massive way) from last decade using excavators although it existed here since the last quarter of century (Guragain 2012). However, heavy machines entered into the river only in 2004. The riverbed materials were also exported to India in a huge quantity since the period of 2004-2011 (DWIDP 2011). Figure 10 shows extraction is more than the deposition causing the deficit of construction materials. The deficit of sediment in the river was about $53.50 \%$. During past decade Rupandehi DDC including other local administrative bodies have collected large amount of revenue from the riverbed materials. The income generated by the local administrative bodies from the riverbed materials within Rupandehi district from the Tinau River is presented in Table 1.

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Table 1: Income generated by local bodies from riverbed materials.

| $\begin{aligned} & \mathrm{S} . \\ & \mathrm{N} . \end{aligned}$ | Local Body | 2005 | 2006 | 2007 | 2008 | 2009 | 2010 | Total Incon (NRs) | USD, \$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| 1 | P. Amawa | 1444082 | 1013203 | 3397268 | 3226775 | 1526950 | 3676500 | 14284778 | 160521 |
| 2 | Shankarnagar | 4661150 | 4512528 | 6645561 | 3288742 | 4320880 | 7424110 | 30852971 | 346702 |
| 3 | Aanandaban | 4697530 | 5598399 | 5703860 | 7924000 | 7957953 | 10665265 | 42547007 | 478110 |
| 4 | Tikuligadh | 4560000 | 3688000 | 4483700 | 6700000 | 8196000 | 9180090 | 36807790 | 413617 |
| 5 | Chilhiya | 805000 | 1246832 | 1317753 | 1460425 | 1725000 | 2431000 | 8986010 | 100978 |
| 6 | HatiBangai | 1246100 | 1040000 | 2120500 | 1152500 | 1924060 | 1745000 | 9228160 | 103699 |
| 7 | Motipur | 550688 | 768938 | 1371570 | 1767000 | 1458159 | 1775200 | 7691555 | 86432 |
| 8 | Farsatikar | 587666 | 848316 | 1950878 | 648000 | 1682500 | 972000 | 6689360 | 75170 |
| 9 | Mainahiya | 366900 | 123622 | 430915 | 448399 | 568516 | 272000 | 2210352 | 24838 |
| 10 | Gonaha | 449700 | 470617 | 469508 | 389667 | 2320200 | 2345000 | 6444692 | 72420 |
| 11 | Butwal Municipality | 75098308 | 75915110 | 96363923 | 77479638 | 162355008 | 155145847 | 642357834 | 7218315 |
| 12 | DDC Rupandehi | 59451551 | 60720688 | 64918062 | 69969900 | 161794140 | 158705120 | 575559461 | 6467687 |
|  | tal (NRs.) | 53918675 | 155946253 | 189173498 | 174455046 | 355829366 | 354337132 | 1383659970 | 15548489 |
|  | uivalent, USD \$=NRs. 88.39) | 1729618 | 1752402 | 2125784 | 1960389 | 3998532 | 3981763 | 15548488 |  |

Source: DDC (2011) modified by Dahal et al. (2012)

The number of fishes recorded in the reference zone (site A) was 82 in the present study. The abundance of reference zone (Site A) during the present study was similar to the upstream side studied by Jha (2006b). Less anthropogenic disturbances could be the reason for such a high number of fishes in this site. The illegal fishing activities in the Tinau River in upstream side were banned for 4 years due to the case filed by Forum for Environmental Awareness and Legal Public Concern (FEALPEC) in Chief District Office (CDO), Palpa. Thus, the fish number in the upstream side of the Tinau River was comparatively high.

At impact zone (Site B), very less number of fish was captured, viz. 5 in premonsoon and 24 in post- monsoon respectively. Similarly, the fish abundance (CPUE) was 2.5 and 12 during pre- and post-monsoon respectively. Jha (2006b) captured 50 and 83 numbers of fishes in the same site with an
abundance of 39.75 and 40.40 in pre- and post-monsoon respectively. The massive riverbed extraction caused destruction of breeding habitat for laying eggs especially for local fishes as indicated by Boudaghpour (2008) in his study. The destruction of appropriate fish habitats in the Tinau River due to over extraction of riverbed materials could be the main reason behind such a decline in fish species richness and abundance in the middle stretch (impact zone) of the river.

The rate of disappearance of native fishes is high throughout the country. There are various causes of disappearance of fish species like damming, poisoning, water pollution, over fishing and habitat destruction (Gurung 2003). Although the scope of the work was set only on the impact of gravel extraction on fish species richness and abundance, other stressors observed during the investigation included over-fishing, poisoning, and damming including habitat destruction. Therefore, other stressors should be included in the study so as to get the better picture of the decline in the fish species, which needs further investigation.

## Conclusions

The river bed extraction was carried in a high scale in the Tinau River to generate the revenue for the local administrative bodies. This has created several environmental degradations including habitat destruction for aquatic biota including fish. The comparison of fish species richness and abundance showed that the fish species has been badly affected from the gravel extraction in the Tinau River over a decade, especially in the impact zone. The adverse impact of extraction of riverbed materials was significant on impact zone (site B), whereas the impact was not significant at reference and recovery zones (Site A and site C ) owing to the less anthropogenic disturbances. The information of this study could be useful for the researchers, policy makers, environmentalists and conservation activists.

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## ANNEX

PHOTOGRAPHS OF THE SAMPLING


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Present status of freshwater prawn (Macrobrachium rosenbergi) post larvae collection in the coastal region of Bangladesh

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#### Abstract

Government of Bangladesh banned wild freshwater prawn (Macrobrachium rosenbergii) post larvae (PL) collection due to indiscriminate fishing and high levels of by-catch in the coastal water. In order to know the current status of wild PL collection in the coastal areas of Khulna, Bagerhat, Peroipur and Noakhali districts in Bangladesh, Focus group discussion was carried out. It was observed that bag net and pull-net are commonly used to collect wild PL.


The peak season for PL collecting is mid-May to mid-June in all locations. The highest rate of capture was observed at mid-June to mid-July in Khulna and Noakhali district, May to June in Bagerhat and Peroipur district. The value chain from collectors to farmers was longest for Noakhali and shortest for Khulna. The price of PL was found highest during mid-March to mid-April. Fishermen at Noakhali earned less than rest locations. Both men and women and children are found to involve in PL collection whereas trading is done solely by men. The freshwater prawn sector is facing a critical short-supply of good quality PL and the availability of wild PL is decreasing at an alarming rate. Therefore, it is suggested to promote better hatcheries to produce high quality PL.

Keywords: Macrobrachium, PL, wild, value chain, coastal region

## Introduction

In Bangladesh, freshwater prawn (Macrobrachium rosenbergii) farming has become one of the most important sectors of the national economy and attracted considerable attention for export potential. The production mostly relies upon wild post larvae (PL) caught in southwest region of Bangladesh. Only about $15 \%$ of the total demand originated from the 70 commercial prawn hatcheries in the year 2008-2009, accounting for approximately 108 million post larvae (DoF 2010). It was also reported that only $2 \%$ of a total of 2000 million PL originated from hatcheries (Paul 2008). Although number of commercial hatcheries increased with demand of PL but only 16 hatcheries were in operation in the year 2000 (Ahmed et al. 2008) due to the low quality of the PL. The hatcheries could not meet the demand and the quality remained questionable to the farmers. Government of Bangladesh banned collection of PL from the wild but this could not stop the collection due to the short supply from the hatcheries. Fishing for wild PL in the coastal regions continues to intensify, threatening other marine and riverine fish stocks.

Indiscriminate fishing of wild PL with high levels of by-catch have affected biodiversity in coastal ecosystem and has, in turn, provoked the enforcement of restrictions on wild PL collection. This affected thousands of rural poor who are involved in prawn PL collection at the coastal rivers of Khulna, Bagerhat, Satkhira, Bhola, Barisal, Barguna, Perojpur, Patuakhali, Laxmipur, Feni, Noakhali, Cox's Bazar and Chittagong districts (Abedin et al. 2001). Researches showed negative impacts on marine fisheries from the intensive PL collection in coastal areas (Canonizado et al. 1998). Most of the fishermen use set bag net that captures almost everything found in the surface layers. The only species with a commercial application is prawn PL (locally referred to as Golda) and shrimp PL (locally referred to as Bagda) while the rest of the catch is discarded. It was reported that more than $2 \times 10^{9} \mathrm{PL}$ (prawn/shrimp) are collected each year from wild sources while about $2 \times 10^{12}$ other species of fish and zooplankton are destroyed as a consequence (Habib 1999). Moreover, it was also reported that more than 1341 other fish fry are caught during the collection of a single prawn PL (Abedin et al. 2001) and around 50 species of finfish juveniles and 28 species of shellfish juveniles are wasted per net per day (Rahman 2008). For this reason Department of Fisheries (DoF 2002) of Bangladesh imposed a ban on wild PL collection in 2000. The rationale for the ban was to protect biodiversity from the harmful effects of intensive PL fishing in the coastal zone. However, the lack of alternative livelihoods for poor people engaged in PL fishing and short supply of hatchery PL resulted in limited success of the ban. That is why the present research was undertaken to know the present status of PL collection from nature in some selected coastal area of Bangladesh.

## Materials and methods

Field data collection was carried out between April and June of 2012. The study sites included Khulna, Bagerhat, Peroipur and Noakhali districts (Figure 1), situated in the coastal areas of the Bay of Bengal (Table 1). To get a better understanding of the current status of wild prawn PL capture, eight Focus group discussions (2 for each location) were conducted with the PL collectors, and one with the PL traders in each districts. The Focus group discussions with an open-
ended questionnaire and primary data were gathered by field survey using prepared questionnaires specific for the purpose. Data from the interviews were later coded and entered into Microsoff Excel soffware for producing descriptive statistics.


Figure 1. Shaded color shown studied locations (K: Khulna, B: Bagerhat, P: Perojpur, H: Hatiya island in Noakhali)

Table 1: Study sites and GPS reading

| Village | Sub-district | District | River |  | GPS reading |  |  |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :---: |
|  |  |  |  | East | North |  |  |
| Kismotfultola | Batiaghata | Khulna | Kazibacha | 0893040.18 | 224448.13 |  |  |
| Khuriakhali | Sharonkhola | Bagerhat | Bhola | 0895055.18 | 221846.93 |  |  |
| Zianagar | Zianagar | Perojour | Kocha | 0895800.36 | 222921.09 |  |  |
| Charchenga | Hatiya | Noakhali | Meghna | 0910202.28 | 220856.26 |  |  |

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## Results and discussion

## Post larvae collection method

The collection of prawn PL is traditionally done from rivers by netting. Two types of nets were seen in use for PL collection, set bag net locally called behundi nets (Figure 3) and pull-nets (Figure 4). Natural PL collection is highly dependent upon tides as PL moves with the water forces. During low tide, PL move downstream and during high tide, upstream. During high-tide fishermen set behundi nets across the river, and during low tide only on the riverbank. PL collection using rectangular shaped pull-nets is done during low tide on the riverbank.

## Set bag net (Behundi Jal)

Behundi nets are operated by boat anchored against the current. The length of the behundi net varies between 8 and 20 meters (average 11.5 meter), and the opening spans between 6 and 15 meters (average 9 meter). It is usually set at 3 to 8 meters (average 7 meter) depth (Figure 2).


Figure 2. Behundi net in operation during high tide

## Pull net

The PL collectors tie the pull-nets around their waist with a rope and pull the net about 0.2 km in the riparian of river by foot (Figure 3). At each low tide, collectors spend up to three hours during which they are able to net two or
three times. The length of the bamboo frame varies between 1.5 to 2 meter (average 1.75 meter) and the width varies between 0.9 to 1.5 meters (average 1.2 meter). The length of net varies from 3 to 4 meter with a cod end of about 0.5 meter.


Figure 3. Pull net in operation during low tide

## Season of PL collection

Seasons are mentioned in Bengali months. The relationship of Bengali and English months is shown in Table 2. Availability of prawn PL differs from season to season and between locations (Table 3). The highest rate of capture per person/day was observed at Ashar in Khulna (300-400 PL day ${ }^{-1}$ ) and Noakhali (300-450 PL day ${ }^{-1}$ ) district, Joishtho in Bagerhat (300-500 PL day ${ }^{-1}$ ) and Peroipur ( $250-450 \mathrm{PL}^{-1}$ day ${ }^{-1}$ ) district. The duration of capture season varied between 4 to 5 months with peak seasons between Baishak and Ashar (Table 4). Highest numbers of prawn PL are collected (600-1000 per person/day) 2 times in a month; during full moon and new moon. The capture rate (PL per person/day) has been decreased to 150 in the year 2011 from 700 in 1980 at Noakhali, to 135 in 2011 from 3500 in 1985 at Peroipur, to 300 in 2011 from 3000 in 1985 at Bagerhat and to 125 in 2011 from 3000 in 1985 at Khulna (Table 5).

Table 2. Relationship of Bengali and English months

| Baishak | Joishtho | Ashar | Shrabon | Vadro | Asshin | Kartik | Augrahaion | Poush | Magh | Falgun | Chaitra |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- | :--- |
| April- | May- | June- | July-Aug | August- | September | October- | November- | December | January- | February | March- |
| May | June | July |  | September | -October | November | December | -January | February | -March | April |

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Table 3. Seasonwise variation in PL collection in 2011

| Bengali month | Captured rate (PL per person/day) |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
|  | Noakhali | Peroipur | Bagerhat | Khulna |
| Chaitra | Negligible | $20-30$ | $40-60$ | $10-20$ |
| Baishak | $25-40$ | $200-400$ | $250-400$ | $40-60$ |
| Joishtho | $250-400$ | $250-450$ | $300-500$ | $200-300$ |
| Ashar | $300-450$ | $10-20$ | $50-70$ | $300-400$ |
| Shrabon | $20-25$ | negligible | $30-40$ | $5-10$ |
| Vadro - Falgun | Negligible | negligible | negligible | negligible |

Note: Negligible refers less than 5
Table 4. Locationwise peak season of PL collection in 2011

| Location | Capture duration | Peak season |
| :--- | :--- | :--- |
| Noakhali | Baishak- Shrabon (4 months) | Joishtho -Ashar |
| Peroipur | Chaitra- Ashar (4 months) | Baishak- Joishtho |
| Bagerhat | Chaitra - Shrabon (5 months) | Baishak- Joishtho |
| Khulna | Chaitra - Shrabon (5 months) | Joishtho -Ashar |

Table 5. Year wise variation of average PL collection (PL per person/day)

| Year | Noakhali | Peroipur | Bagerhat | Khulna |
| :--- | :--- | :--- | :--- | :--- |
| 1985 | not practiced | 3500 | 3000 | 3000 |
| 1990 | 700 | 1000 | 2000 | 750 |
| 1995 | 500 | 700 | 800 | 350 |
| 2000 | 500 | 300 | 500 | 250 |
| 2005 | 350 | 200 | 500 | 175 |
| 2011 | 150 | 135 | 300 | 125 |

## Sorting and counting of PL

During netting, there are many other species caught as by-catch which are discarded on the banks or in the river after sorting. For sorting PL, either a white colored spoon, an oyster shell, tin made plates or a plastic container is

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used (Figure 4). After sorting, PLs are kept in river water in earthen pot, aluminum or plastic containers.


Figure 4. PL sorting and counting activities and equipment used

## Market chain analysis

Value chain in Noakhali
One of the biggest markets for prawn fry trading in the southwest region is Foyla, located in Rampal upazilla of Bagerhat district. A major portion of the wild PL comes from Hatiya Island of Noakhali district. The value chain of wild PL collection in Hatiya to farming in southwest region is shown in Figure 5. There were about 100 hawkers on Hatiya Island who buy PL at different locations of the island and go to Charchenga Bazar in Hatiya on foot or by non-mechanized van to sell the PL to Mohajon, a type of local commission agent. There were about 15 Mohajons in Hatiya who stayed at Char Chenga Bazar to trade prawn fry. During the peak season a Mohajon buys about 25000 to 30000 PL from the hawkers every day. The price paid by the Mohajons is about Tk. 1100 per thousand PL. The Mohajons then sell the PL to the PL traders who come to Charchenga Bazar from Khulna and Bagerhat region.

After buying of PL from the Mohajons at the rate of roughly Tk. 1300 per thousand PL, the PLs are kept in aluminum bowls ( 30 L capacity to carry 10000 PL ) or in plastic barrels ( 15 L capacity to carry 5000 PL ). The PLs are then shipped to Rampal in Bagerhat district, by boats powered by diesel engines.

Boats differ in engine power, but most common is 33 HP . The journey from Hatiya to Rampal is about 350 km by boat and takes about 20 hours, consuming 150 L of diesel (for a return journey 300 L diesel assuming a 33 HP engine). The carrying capacity is on average 40 aluminum bowls containing about 0.4 million PL in total. From the PL collection at the Meghna River in Hatiya to Rampal, 10\% mortality is expected by the stakeholders. After which the PL travel 15 km by road from Rampal to Foyla market. This transport is carried out by a mechanized van (locally called Nosimon) and takes half an hour, consuming 0.5 L of diesel. The carrying capacity of a mechanized van is 10 aluminum bowls containing 0.1 million PL. Therefore, 2 L diesel ( $0.5 \mathrm{~L}^{*} 4$ ) is needed to carry 0.4 million PL to Foyla market.


Figure 5. Value chain of wild PL (collector to farmers) from Noakhali

The main Aratder (shop vendor of PL trading business) in Foyla buys the PL at a rate of Tk. 2500 per thousand PLs from the traders. They then sell these with a profit of Tk. 100 per thousand PLs. The Aratder then sell the PL to the Foria (middleman) who, in turn, supplies local Arats (market place) at different
farming locations, charging Tk. 3000 per thousand PLs. The local Aratder sells at the rate of Tk. 3300 per thousand PL. There are some groups of people locally called Patilwala (carries PL in container), who buys PL from local Arat and sell these to the prawn farmers at farm-gate at the rate of Tk. 3500 per thousand PL. Some big farm owners buy PL directly from the main Arat of Foyla, while mostly farmers buy from local Arat or from Patilwala.

## Value chain in Peroipur

The collectors of Zianagar village on the bank of Kocha River sell PL to Foria-1 at riverside. Foria-1 then sells the PL either directly to the farmers at farm-gate or to Aratders in Fakirhat and Foyla (Bagerhat district) which are about 35 km away. Based on farming location, farmers buy PL either directly from main Arat at Bagerhat or from local Foria-2, local Arat or Patilwala (Figure 6).


Figure 6. Value chain of wild PL (collector to farmers) from Peroipur

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## Value chain in Bagerhat

The collectors of Khuriakhali village on the bank of Bhola River sell PL at Sharonkhola depot to the Aratder at the rate of Tk. 800-3000 per thousand PL depending on season. The batch then goes to farmers at different locations (Bagerhat, Khulna, Satkhira, Narail, Jessore) directly or indirectly through middlemen, as shown in Figure 7. At each stage, chain actors add a profit of about Tk. 100-150 per thousand PL to value.


Figure 7. Value chain of wild PL (collector to farmers) from Bagerhat

## Value chain in Khulna

The collectors of Kismoffultola village, working on the banks of Kazibacha River, sell their PL to the Foria-1 at the riverside. Sometimes, however, they have to walk to nearby market places ( 0.5 km away) if there are no Forias nearby. Fishermen can stock their captured PL for up to one week without supplying any feed, but in most cases they will sell it on daily basis. Most of the time, Foria-1 will sell its PL directly to the farmers around Khulna at a profit of Tk. 1000 per thousand PL. In some cases, Foria-1 sells PL to another larger Foria-2 that accumulates PL from several Forias to supply big farm owners (Figure 8).


Figure 8. Value chain of wild PL (collector to farmers) from Khulna

## Pl transportation system

Depending upon the distance, a wide range of vehicles are used to transport the PL from the collectors to the farmers (Figure 9), including hand (a) or motor (b) powered boats, bicycles (c), motorcycles (d), and pedal (e) or motor powered (f) vans. Aluminum containers (10-30 L) each holding 5000-10000 PL , are commonly used to transport the PL.


Figure 9. a) Hand powered boat, b) Motor boat, c) Bicycle, d) Motor cycle, e) Non-mechanized van, f) Mechanized van (Nosimon)

## Price of PL

The fishermen sell their captured PL at different prices depending on season (Table 6). The price at the collectors varied greatly between locations (in Tk. per thousand PL) from 400 (Shrabon) to 1500 (Baishakh) in Noakhali; from 1200
(Ashar) to 3000 (Chaitra) in Peroipur; from 800 (Shrabon) to 3500 (Chaitra) in Bagerhat; and from 1000 (Shrabon) to 3500 (Chaitra) in Khulna. A major influence on the price retrieved by the collectors is the costs related transportation that needs to be covered by the traders, e.g. Noakhali is far away than Bagerhat.

Table 6: Seasonwise price variation of PL

| Bengali month | Fishermen's average price (Tk. per thousand PL) |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
|  | Noakhali | Peroipur | Bagerhat | Khulna |
| Chaitra | - | 3000 | 3500 | 3500 |
| Baishak | 1500 | 2700 | 3000 | 3000 |
| Joishtho | 1000 | 2000 | 2200 | 2000 |
| Ashar | 550 | 1200 | 1500 | 1500 |
| Shrabon | 400 | - | 800 | 1000 |

A major increase in price was observed over the last decades (Table 7) with an at least two-fold increase in all regions.

Table 7: Annual average prices of PL in different regions (based on fishermen sales prices)

| Year | Region wise price (Tk. per thousand PL) |  |  |  |
| :--- | :--- | :--- | :--- | :--- |
|  | Noakhali | Perojpur | Bagerhat | Khulna |
| 1985 | - | 100 | 150 | 175 |
| 1990 | 95 | 250 | 300 | 300 |
| 1995 | 200 | 450 | 500 | 550 |
| 2000 | 500 | 800 | 1000 | 1000 |
| 2005 | 750 | 2000 | 2000 | 2000 |
| 2011 | 1000 | 2500 | 2600 | 2750 |

## Gender involvement

Behundi nets are mainly operated by male fishermen, whereas pull-nets are mainly used by women and children of age below 14 years (Table 8). All of the other actors in the value chain (traders) are male.

Table 8: Gender involvement among wild PL collectors and traders

| Location | Fishermen for pulling net (\% involvement) |  | Traders |  |  |  |
| :--- | :--- | :--- | :--- | :--- | :--- | :--- |
|  | Child | Women | Men | Child | Women | Men |
| Noakhali | 40 | 57 | 3 | 0 | 0 | 100 |
| Peroipur | 25 | 65 | 10 | 0 | 0 | 100 |
| Bagerhat | 20 | 75 | 5 | 0 | 0 | 100 |
| Khulna | 15 | 80 | 5 | 0 | 0 | 100 |

## Conclusion

The freshwater prawn sector is facing a critical short-supply of high quality hatchery PL. As a result of indiscriminate fishing over last three decades, the availability of wild PL is decreasing at an alarming rate and price of PL is rapidly increasing, making prawn farming economically unsustainable. Eleven years after the ban on wild PL capture was imposed, little or no progress has been made due to the short supply of hatchery PL, high demand for wild PL and a lack of alternative income source of the poor fishermen. Government actions are therefore needed to promote better hatcheries to produce high quality PL. Otherwise the promising prawn sector will be undermined by shortages in a near future.

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# Oxygen uptake in hillstream cyprinid Schizothorax richardsonii (Gray) 

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#### Abstract

Schizothorax richardsonii (Gray) is a freshwater obligate water breathing hillstream fish. A study on Schizothorax richardsonii was carried out to assess the oxygen requirement of the fish in Ilam district, Nepal. Oxygen uptake in relation to body weight was measured using flow through cylindrical glass respirometer at an ambient temperature of $21.0 \pm 0.7^{\circ} \mathrm{C}$. Data were analyzed using logarithmic transformations and relationship between body weight (W) and oxygen uptake $\left(\mathrm{V}_{2}\right)$ and expressed by the allometric equation $\mathrm{VO}_{2}=$ $\mathrm{aW}^{\mathrm{b}}$. The oxygen uptake per unit time increased with increase in body weight from $1.40 \pm 0.15 \mathrm{mlO}_{2} / \mathrm{h}$ to $4.78 \pm 0.00 \mathrm{mlO}_{2} / \mathrm{h}$ within the body weight ranged from $2.60 \pm 0.42 \mathrm{~g}$ to $47.8 \pm 0.00 \mathrm{~g}$. A positive and high degree of correlation $(\mathrm{r}$ $=0.9490 ; \mathrm{P}<0.001$ ) was observed between the above parameters. The weight specific oxygen uptake however, decreased with increase in body weight from $0.55 \pm 0.04 \mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$ to $0.10 \pm 0.00 \mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$ within the same range of body


weight. There was a negative and highly significant correlation ( $r=0.9796$; $\mathrm{P}<0.001$ ) between these two variables. The higher rate of oxygen consumption per unit body weight $\left(\mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}\right)$ in small fishes has been related to their higher metabolic rate. An average body weight of 19.24 g Schizothorax richardsonii consumed $3.45 \mathrm{mlO}_{2} / \mathrm{h}$ and $0.29 \mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$.

Keywords: oxygen uptake, body weight, Schizothorax richardsonii, Buchche Asala, Hillstream fish

## Introduction

Schizothorax richardsonii, the spotted snow trout (Buchche Asala) is a coldwater obligate water breathing fish belonging to the Subfamily Schizothoracini and Family Cyprinidae of the Order Cypriniformes (Nelson 1994). It is a hillstream and potamodromus fish having adhesive structure in the form of irregular folds of skin on the chin as a hill stream modification to suit the habitat (Ojha 2002). The gills of the fish have modified to suit the torrential and hyperoxic water of the hillstream.

Oxygen uptake ( $\mathrm{V}_{2}$ ) has long been considered as the valid measure of metabolic rate in animals (Muire and Hughes 1969; Hughes and Gray 1972). The rate of oxygen uptake in fish depends on the body weight, surface area of gills, permeability of the blood water tissue barrier, state of fish, season, time and water quality parameters. The measurement of metabolic rate in terms of oxygen uptake, gives an estimate of the energy requirement of the fish (Fry 1957; Prosser 1973; Dejours 1975). The rate of oxygen uptake increases with the increase in metabolic activity. Though the oxygen uptake $\left(\mathrm{VO}_{2}\right)$ is not a linear function of the body weight (W) it follows the allometric equation ( $\mathrm{VO}_{2}=$ $\mathrm{aW}^{\mathrm{b}}$ ) as used by Huxley (1924), Zeuthen (1953), Winberg (1956), Ultsch (1973), Hughes et al. (1974) and many more. Rübner (1883) was probably the first investigator to work on the body weight and oxygen uptake relationship in animals. After that Ojha and Singh 1981; Roy 1983; Kunwar et al. 1989; Gallaugher et al. 2001; Kumari 2003; Das et al. 2003; Bang et al. 2004; Jha
et al. 2008; Kumari et al. 2010 worked on water breathing fish and Rooi (1984), Subba (1999) and Subba and Ghosh (2011) on hillstream fishes.

Knowledge of oxygen requirements of a particular species is a prerequisite for their economical and successful transportation, stocking and culture (Kamler 1976). The present study was carried out to investigate the rate of routine oxygen uptake in relation to various weight groups and to establish a relationship between them during summer season at an ambient temperature of $21.0 \pm .07^{\circ} \mathrm{C}$.

## Materials and methods

Eighteen live specimens of Schizothorax richardsonii of 8 different body-weight groups, collected from Puwa Khola in llam district, Nepal in May 2006 were acclimatized in a hand dug excavate for two weeks at Gumaantaar of Shangrumba VDC, Ward No 5 in the same district. The excavate was filled with seepage water and fish were kept under flowing water condition feeding with algae wrapped around pebbles, insects, rotten leaves and chironomid larvae twice a day i.e. at 9:00 AM and 6:00 PM. The fish was kept starved for at least 12 hrs before experimentation.

The routine oxygen uptake was measured in a flow through cylindrical glass respirometer as described by Munshi and Dube (1973). Fish was weighed in a balance and acclimatized inside the respirometer for 2 hours before taking the observations for the determination of dissolved oxygen concentration. The rate of water flow in the respirometer was maintained at 1.5 litres per hour i.e. 25 ml per minute. Ambient and expired water samples were collected thrice at hourly intervals for each specimen. Oxygen concentrations were calculated separately for ambient and expired water using Winkler's volumetric method (Welch 1948) and their difference gave the value of oxygen uptake ( $\dot{\mathrm{V}} \mathrm{O}_{2}$ ) of the fish. Oxygen uptakes per unit time $\left(\mathrm{mlO}_{2} / \mathrm{h}\right)$ and per unit body weight $\left(\mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}\right)$ were calculated. The frequency of opercular movements was observed and average value was calculated. The values of water temperature, dissolved oxygen and pH were recorded during the experiment.

The obtained data were analyzed by linear logarithmic transformations using least square regression method to establish a relation between body weight and oxygen uptake for the experimental fish. For the analysis MS Excel soffware was used. The equation ( $\left.\mathrm{VO}_{2}=a \mathrm{~W}^{\mathrm{b}}\right)$ was used to show the allometric relationship where $\mathrm{VO}_{2}$ is oxygen uptake, ' $a$ ' is the intercept, ' $b$ ' is the slope of the regression line and $W$ is the body weight of the fish.

## Results

Results of regression analysis showing relationship between body weight and oxygen uptake have been summarized in Table 1 and 2 and depicted in Figure 1 A and 1 B .

Table 1. Mean values (Mean $\pm$ S.D.) of oxygen uptake $\left(\mathrm{VO}_{2} ; \mathrm{mlO}_{2} / \mathrm{h}\right)$ and weight specific oxygen uptake ( $\mathrm{vO}_{2} ; \mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$ ) in relation to body weight (W) in Schizothorax richardsonii at $21.0 \pm 0.7^{\circ} \mathrm{C}$

| Body weight | Opercular <br> ( g ) | Oxygen uptake $\left(\mathrm{VO}_{2}\right)$ <br> frequency <br> (Beats/minute) |  |
| :---: | :---: | :---: | :---: |
| $2.60 \pm 0.42$ | $114 \pm 4$ | Per unit time <br> $\left(\mathrm{mlO}_{2} / \mathrm{h}\right)$ | Per unit body weight <br> $\left(\mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}\right)$ |
| $3.70 \pm 0.64$ | $110 \pm 3$ | $2.40 \pm 0.15$ | $0.55 \pm 0.040$ |
| $12.70 \pm 1.12$ | $137 \pm 7$ | $3.94 \pm 0.25$ | $0.55 \pm 0.01$ |
| $17.50 \pm 0.71$ | $106 \pm 2$ | $4.01 \pm 0.30$ | $0.31 \pm 0.05$ |
| $24.20 \pm 1.55$ | $132 \pm 1$ | $4.03 \pm 0.36$ | $0.16 \pm 0.01$ |
| $31.70 \pm 1.34$ | $152 \pm 4$ | $4.27 \pm 0.65$ | $0.14 \pm 0.02$ |
| $42.30 \pm 0.00$ | $156 \pm 0$ | $4.36 \pm 0.00$ | $0.10 \pm 0.00$ |
| $47.80 \pm 0.00$ | $144 \pm 0$ | $4.78 \pm 0.00$ | $0.10 \pm 0.00$ |
| Average 19.24 | 131 | 3.45 | 0.29 |

In Schizothorax richardsonii oxygen uptake per unit time ranged from $1.40 \pm 0.15 \mathrm{mlO}_{2} / \mathrm{h}$ to $4.78 \pm 0.00 \mathrm{mlO}_{2} / \mathrm{h}$ within the body range of 2.60 to 47.8 g when the dissolved oxygen content of ambient water was recorded in the range of 5.0 to 7.4 ppm and pH 8.1 to 9.3 at $21.0 \pm 0.7^{\circ} \mathrm{C}$. The bilogarithmic plots of the oxygen uptake with respect to the body weight showed a straight line with a slope (b) of 0.3839 (Figure 1A). The intercept (a) was found to be $1.1730 \mathrm{mlO}_{2} / \mathrm{h}$. The oxygen uptake per unit time increased with the increase in the body weight by a power of 0.3839 (Table 2). A positive and high degree of correlation ( $r=0.9490 ; p<0.001$ ) was observed between the two parameters whereas the weight specific oxygen uptake decreased with an increase in the body weight. It decreased from $0.55 \pm 0.04$ $\mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$ to $0.10 \pm 0.00 \mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$ of the same body weight (Table 1). The $\mathrm{log} / \log$ plots of both variables gave a straight line with a slope value -0.6227 (Fig. 1B) and the intercept value $1.1928 \mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$. A negative and highly significant correlation ( $\mathrm{r}=0.9796 ; \mathrm{p}<0.001$ ) was found between the two variables (Table 2). It has been calculated that Schizothorax richardsonii of 19.24 g average body weight consumed $3.45 \mathrm{mlO}_{2} / \mathrm{h}$ and $0.29 \mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$. The allometric equations showing the relationship between body weight and oxygen uptake have been summarized in Table 3.

Table 2. Values of intercept (a), regression coefficient (b) and correlation coefficient ( $r$ ) showing the relationship between body weight and oxygen uptake $\left(\mathrm{vO}_{2}\right)$ and the same for opercular frequency in Schizothorax richardsonii at $21.0 \pm 0.7^{\circ} \mathrm{C}$.

| Parameters |  | Intercept <br> (a) | Regression <br> coefficient <br> (b) | Correlation <br> coefficient $(\mathrm{r})$ |
| :---: | :---: | :---: | :---: | :---: |
| Oxygen uptake <br> $\left(\mathrm{VO}_{2}\right)$ | $\dot{\mathrm{VO}}_{2}\left(\mathrm{mlO}_{2} / \mathrm{h}\right)$ | 1.1730 | 0.3839 | 0.9490 <br>  <br>  <br>  <br>  <br>  <br>  <br> $\left(\mathrm{HIO}_{2} / \mathrm{m} / \mathrm{g} / \mathrm{h}\right)$ |
| 1.1928 | -0.6227 | 0.9796 <br> $(\mathrm{P}<0.001)$ |  |  |
| Opercular | (Beats $/ \mathrm{minute})$ | 98.0237 | 0.1033 | 0.7480 <br> frequency |
|  |  |  |  | $(\mathrm{P}<0.05)$ |



Figure 1. A. Bilogarithmic plots showing the relationship between body weight and oxygen uptake $\left(\mathrm{mlO}_{2} / \mathrm{h}\right) \mathrm{B}$. Bilogarithmic plots between body weight and weight specific oxygen uptake ( $\mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}$ ) in Schizothorax richardsonii at $21.0 \pm 0.7^{\circ} \mathrm{C}$.

Table 3. Allometric equations showing the relationship between body weight (W) and oxygen uptake $\left(\mathrm{HO}_{2}\right)$ in Schizothorax richardsonii at $21.0 \pm 0.7^{\circ} \mathrm{C}$

| Allometric equation ( $\left.\mathrm{VO}_{2}=\mathrm{aW} \mathrm{W}^{\mathrm{b}}\right)$ |  |
| :---: | :---: |
| $\mathrm{VO}_{2}\left(\mathrm{mlO}_{2} / \mathrm{h}\right)$ | $\mathrm{VO}_{2}\left(\mathrm{mlO}_{2} / \mathrm{g} / \mathrm{h}\right)$ |
| $\begin{aligned} & \mathrm{VO}_{2}=1.1730 \times \mathrm{W}^{0.3839} \\ & \text { or, } \log \mathrm{vO}_{2}=\log 1.1730+0.3839 \times \log \mathrm{W} \\ & \text { or, } \log \mathrm{VO}_{2}=0.0693+0.3839 \times \log \mathrm{W} \end{aligned}$ | $\begin{aligned} & \mathrm{VO}_{2}=1.1928 \times \mathrm{W}^{-0.6227} \\ & \mathrm{~N} \text { or, } \mathrm{Log} \mathrm{VO}_{2}=\log 1.1928+(- \\ & 0.6227) \times \log \mathrm{W} \\ & \text { or, } 10 \mathrm{Dog} \mathrm{O}_{2}=0.0766- \\ & 0.6227 \times \log \mathrm{W} \end{aligned}$ |

The opercular frequency (opercular beats/minute) has been recorded from 106 to 156 for the same body weight with an average of 131 beats/minute for a fish of 19.24 g average body weight (Table 1). The relationship was found weakly significant ( $r=0.7480$; $p<0.05$ ) between body weight and opercular frequency (Table 2).

## Discussion

The rate of oxygen uptake in any animal is related to the magnitude of the energy requirements of the animal which is dependent on its body weight, organismic (Bertalanffy 1957) and environmental factors (Dejours 1976). Similarly in fishe too, it is considered as the valid measure of metabolism and is related to the body weight (Fry 1971). In Schizothorax richardsonii the oxygen uptake increased with increasing body weight. The slope value (b) was estimated 0.3839 which was less than in fishes like Salmo gairdneri ( 0.86 ; Jager and Dekkers 1975), S. trutta (0.88; Jager and Dekkers 1975), Mystus cavasius (0.66; Ojha and Singh 1981), Cirrhinus mrigala (0.80; Roy and Munshi 1984), Glossogobius giuris (0.63; Singh and Munshi 1985), Labeo rohita (0.85; Kunwar et al. 1989), and Catla catla (0.75; Kunwar et al. 1989) and was close to the values in the fishes like Colisa fasciatus ( 0.37 ; Ojha et al. 1988) and Notopterus notopterus (0.42; Kumari 2003). The observed value in this hill stream cyprinid came to be less than those in other hill stream fishes like Garra lamta (0.87; Rooj 1984), Noemacheilus rupicola (1.20; Rooj 1984) and Glyptothorax telchitta (0.93; Subba 1999). The rate of weight specific oxygen uptake decreased with increasing body weight with the slope value -0.6227 . It indicated that the weight specific oxygen uptake was more in smaller fishes than the larger ones. It was in agreement with the previous findings (Oiha and Singh 1981; Roy and Munshi 1984). Prigogine and Wiame (1946) indicated that for thermodynamic reasons the metabolic rate can never increase but only decrease with the increase of body weight in organisms. This may be the reason that the slope (b) for oxygen uptake against the body weight had never been found to be higher than 1 . The high rate of oxygen consumption per unit body weight in small fish has been related to their high metabolic rate (Fry 1957). In addition, decreased weight specific oxygen consumption may be due
to the fact that larger fishes are more sluggish than smaller ones (Roy 1983). Another possible cause may be the increase in inert tissue like skeleton, metabolites, and blood-water diffusion barrier in larger fishes. Generally, the oxygen uptake is proportional to the respiratory area. Thus, weight specific decrease in respiratory area also might be the cause of weight specific decrease in oxygen uptake.

The observed value of opercular movement (beats/minute) was observed 106156 with the mean of 131 in Schizothorax richardsonii (Table 1.) which was greater than that in Catla catla (24-35; Kunwar 1984), Labeo rohita (35-50; Pandey 1988) and Sparisoma viridae (63-76; Van Rooii and Videler 1996). The same trend has been recorded in the hill stream fishes such as $60-74$ in Garra lamta and 60-88 in Noemacheilus rupicola (Rooj 1984) and 85-91 in Glyptothorax telchitta (Subba 1999). It reveals that Schizothorax richardsonii, having the higher value of opercular movement, needs large amount of water to fulfill the oxygen requirement for its metabolic activity. The regression analysis did not show any relation between body weight and opercular frequency because oxygen requirement might be the function of concentration of dissolved oxygen in water, efficiency of gills and activity i.e. state of fish, etc.

In conclusion, at $21.0 \pm 0.7^{\circ} \mathrm{C}$ Schizothorax richardsonii of 1.0 g required $1.1730 \mathrm{ml} \mathrm{O}_{2} /$ hour and it increased with the increase of body weight by a power of 0.3839 . The opercular frequencies observed under experimental condition were 131 beats / minutes which showed that the fish ventilated the large amount of water to fulfill the oxygen requirement for its metabolic activity.

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[^0]:    *Locus G3PDH*was monomorphic in low line.

