Carp Genetic Resources for Aquaculture in Asia

Edited by

David J. Penman
Modadugu V. Gupta
Madan M. Dey

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WorldFish Center is one of the 15 international research centers of the Consultative Group on International Agricultural Research (CGIAR) that has initiated the public awareness campaign, Future Harvest.

Contents

Preface

There are over 1 300 species of cyprinids in Asia, and these are utilized by people for food (capture fisheries and aquaculture) and as ornamental species. For people in many parts of Asia, cyprinids form an integral part of their livelihoods, as subsistence or commercial fishers, hatchery operators, nursery operators, seed traders, ongrowers, middlemen, fish sellers, consumers, or producers of ornamental species. The habitats of many of these species are also increasingly threatened by human activities. It is thus vital that we protect this part of our biodiversity, since it sustains human populations as well as having its own intrinsic value.

While there are over 1300 cyprinids species, presently only few of them are used in aquaculture and the potential of many other species in aquaculture is yet to be assessed. Fifteen species are of major aquaculture importance to the countries in Asia and contributed 49 per cent of aquaculture production in the region in 2001.

This publication focuses on cyprinid species that are bred in hatcheries and used in aquaculture and restocking activities. Introduced and threatened species of cyprinids in Asia are also discussed. We are well aware that we have not covered many other cyprinid species that are utilized by people in Asia in various ways: wild species in capture fisheries, self-recruiting species in aquaculture ponds and other water bodies, and aquarist species.

The species nomenclature given in this publication follows that of FishBase (http://www.fishbase.org) and further information on the species dealt with here can be found in this database.

Lastly, the Editors would like to thank the Asian Development Bank for funding the project that led to this publication, and the many people who provided research results and hard-to-find references that are included here.

David J. Penman
Modadugu V. Gupta
Madan M. Dey
Editors

Chapter 1

Importance of Carp Genetic Resources[1]

Modadugu V. Gupta[2], Madan M. Dey[2] and David J. Penman[3]

Fish and fisheries play an important role in the economies of developing countries, contributing to animal protein intake, employment generation, household incomes and foreign exchange earnings. Surveys conducted by the WorldFish Center and FAO show that fish has become an increasingly important source of protein over the last decade in most of the developing countries. Countries with low per capita gross domestic product tend to have a higher share of fish protein in their animal protein consumption (Kent 1997). Studies indicated that demand for fish increases as expenditure/income rises and that higher income groups tend to consume more fish than lower income groups. However, the share of fish (as protein) and the share of fish to the total food expenditure are higher among lower income groups, suggesting that lower income groups are the most dependent on fish (Dey et al. 2004; FAO 1999a; Dey 2000; Dey et al. 2000a; ICLARM 2000). This result is also consistent with the generalization that although less developed countries are not the biggest consumers of fish, they are the most dependent on aquatic resources (FAO 1993; Kent 1997; FAO 1999b), indicating the importance of fish as a primary source of protein among relatively poorer households in these countries. No wonder it is regarded as "poor man's protein" (Williams 1996).

In Asia, on average, almost 30 per cent of the total animal protein intake is derived from fish. Among the Southeast Asian countries, fish protein provides 45 per cent of the total protein consumed (Prein and Ahmed 2000). Although Japan, the European Union and USA have higher per capita consumption of fish and fish products, the share of fish protein in terms of the total animal protein consumption is far less than that in many developing countries.

World fish production in 1999 was estimated at 130 million t, of which only 97 million t was for human consumption (FAO 2003). FAO (1999a) estimated that by the year 2010, demand for food fish will have increased by 13.5-18 per cent or to about 105-110 million t. Against this demand, production from capture fisheries is declining as most of the world's fish stocks have been exploited to their maximum potential or over exploited, indicating it might be difficult to increase yields from capture fisheries in the near future (Williams 1996). During the 1950s and 1960s capture fish production increased by an average of 6 per cent per annum and this declined to 2 per cent during the 1970s and 1980s, falling almost to zero during the 1990s (FAO 2000).

In contrast to declining growth in capture fisheries, aquaculture has been growing at a fast rate, by about 5 per cent per year during the 1950s and 1960s to about 8 per cent during the 1970s and 1980s and over 10 per cent since 1990 (FAO 2000). Aquaculture production has increased from 3.2 per cent of the total fish production in 1950 to 32 per cent in 2000. Aquaculture has become the world's fastest growing food-producing sector, with production more than doubling during 1990-2000, from 13.1 million t in 1990 to 35.6 million t in 2000 (FAO 2003).

Of the total aquaculture production of 45.7 million t in 2000, 45 per cent was contributed by freshwater aquaculture. Production increased from 7.9 million t in 1991 to 20.6 million t in 2000. Asia produces about 91 per cent of the world's freshwater aquaculture production, with China contributing 74 per cent of this (Fig.1). Other major producers of freshwater aquaculture are India (9.89 per cent), Bangladesh (2.77 per cent), Vietnam (1.85 per cent), Indonesia (1.76 per cent), Thailand (1.29 per cent) and the Philippines (0.54 per cent). Growth in freshwater aquaculture in the last decade among these countries differs: Vietnam had the highest growth

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[1] WorldFish Center Contribution No. 1728.
[2] WorldFish Center, Jalan Batu Maung, Batu Maung, 11960 Bayan Lepas, Penang, Malaysia.
[3] University of Stirling, U.K.

of 14.54 per cent per annum during 1990-2000, followed by China (13.6 per cent), Bangladesh (12.94 per cent), Thailand (9.38 per cent) and Indonesia (5.65 per cent).

With this remarkable increase in production, relatively low prices and limited international markets, freshwater fish are expected to become an increasingly important source of animal protein, particularly for those in medium and lower income groups in developing countries, especially in Asia (Dey et al. 2004).

About 242 species of freshwater finfish are cultured globally and of these, carps (Cyprinidae) are the dominant species, especially in Asia. While production of cultured carps is on the increase the amount from capture fisheries from rivers and other natural waters is on the decline due to the shrinking of areas for spawning, over fishing and other human induced habitat degradation. Production of carps from culture has increased from 5.6 million t in 1990 to 16.4 million t in 2001, while the yield from capture fisheries has declined from 0.75 million t in 1990 to 0.54 million t in 2001 (Table 1.1). These human induced changes are leading to threats to the existence of some of the species (Acosta and Gupta, Chapter 7, this vol.). In Asia, aquaculture production increased from 10.8 million t in 1990 to 29.48 million t in 2001, while carp production increased from 5.07 million t in 1990 to 16.42 million t in 2001 (Fig. 1.1).

Table 1.1. Carp production (million t) from capture fisheries and culture during 1990-2001

Year	Production from culture (million t)	Production from capture fisheries(million t)	Total (million t)
1990	5.07	0.47	5.54
1991	5.27	0.54	5.81
1992	6.08	0.39	6.47
1993	7.18	0.45	7.63
1994	8.50	0.51	9,01
1995	9.93	0.55	10.48
1996	11.48	0.43	11,90
1997	12.94	0.38	13.32
1998	13.60	0.38	13.98
1999	14.63	0.38	15.01
2000	15.14	0.33	15.47
2001	16.00	0.31	16.31
Growth rate (%)	11.16%	-2.60	10.44%

Source: FAO 2003. Fisheries Statistics (on line) available at www.fao.org/fi/statist/FISOFT/FISHPLUS.asp [June 2003].

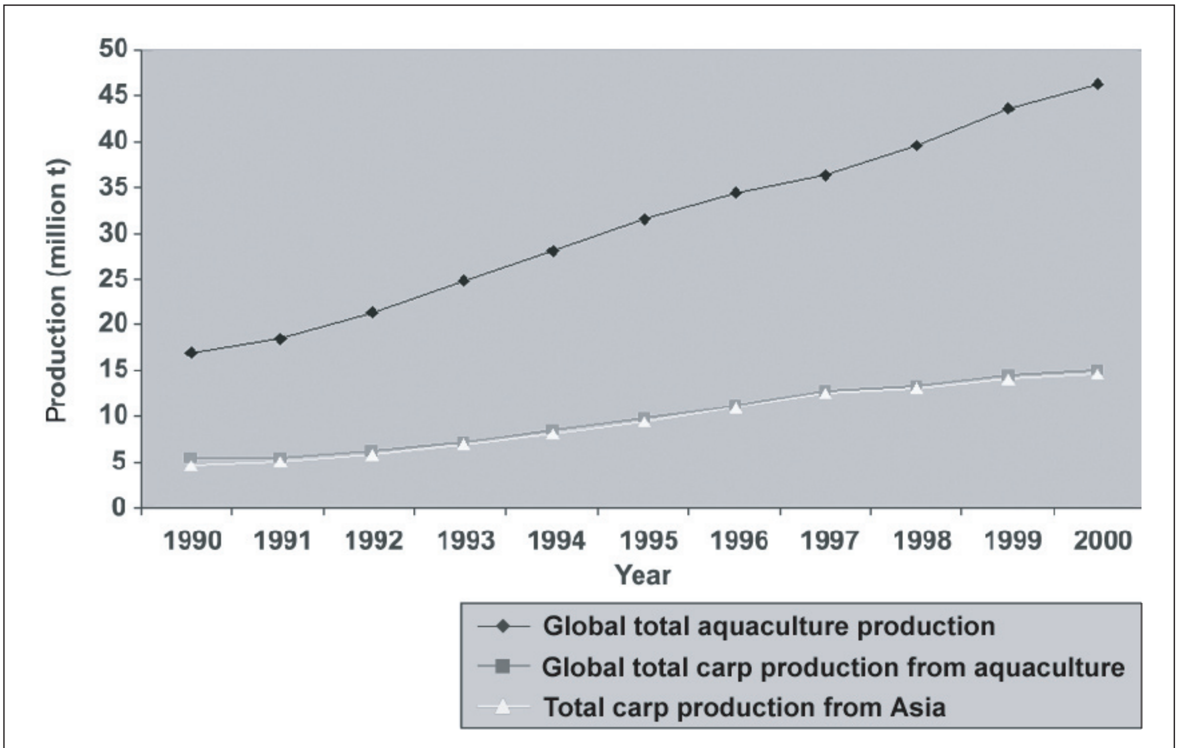

Fig. 1.1. Trends in world aquaculture and carp production (million t) from aquaculture globally and from Asia

Table 1.2. Prominent Cyprinid species used for aquaculture in Asia

Species	Production (t) in 2001[a]
Hypopthalmichthys molitrix	3 546 285
Ctenopharyngodon idella	3 636 367
Cyprinus carpio	2 849 492
Aristichthys nobilis	1 663 499
Carassius carassius	1 527 058
Labeo rohita	833 816
Catla catla	870 085
Cirrhinus cirrhosus	589 841
Parabramis pekinensis	541 115
Mylopharyngodon piceus	190 707
Cirrhinus molitorella	220 118
Barbonymus gonionotus[b]	53 563
Puntius javanicus[b]	26 251
Osteochilus hasseltii	15 319
Leptobarbus hoevenii	1 111
Other cyprinid species	56 440
TOTAL	16 621 067

[a] Represents production of carp from aquaculture in 23 countries of Asia.
[b] *Puntius gonionotus* and *Puntius javanicus* are the same species, now described as *Barbonymus* (*Barbodes*) *gonionotus* by FishBase (2003) [available on line at: www/fishbase.org].
Source: FAO 2003. Fishery statistics: Aquaculture production. Vol. 92/2

Over 99 per cent of the total carp production was contributed by 15 species among which Chinese carps [silver carp (*Hypophthalmichthys molitrix*), grass carp (*Ctenopharyngodon idella*), bighead carp (*Aristichthys nobilis*), crucian carp (*Carassius carassius*) and common carp (*Cyprinus carpio*)] and Indian carps [rohu (*Labeo rohita*), catla (*Catla catla*) and mrigal (*Cirrhinus cirrhosus*)] are the dominant species (Table 1.2 and Fig. 1.2).

Although aquaculture has a long history in Asia, dating back over 2000 years in China, until recently aquaculture has not been subject to the same intensive development of its genetic base as has been the case with terrestrial agriculture. Genetic improvement of fish is estimated to lag behind advances in livestock by nearly 50 years (Eknath et al. 1991). In fact until the 1960s, the aquaculture industry was dependent on seed of carps collected from the wild, but with successful hypophysation of carps in the late 1950s, much of the seed is presently being produced in hatcheries. With this development, there is increasing concern that the genetic bases of many culture species are deteriorating to the extent of lowering the growth (Eknath and Doyle 1990) and the most common stocks used in aquaculture may not be the best performers (e.g. Nile tilapia in the Philippines, Eknath et al. 1993; Dey et al. 2000b; Dey and Gupta 2000). Evidence of inbreeding and unintentional selection among hatchery-based and established farm stocks have been observed (Pullin and Capilli 1988; McAndrew et al. 1993; Eknath 1995).

While remarkable progress has been made in improving the productivity of crops and livestock in the last three to four decades through breeding and selection, it is only in recent years that efforts have been made to harness the benefits of genetic enhancement (e.g. salmon and Nile tilapia). Selective breeding of Nile tilapia (*Oreochromis niloticus*) over five generations has resulted in an

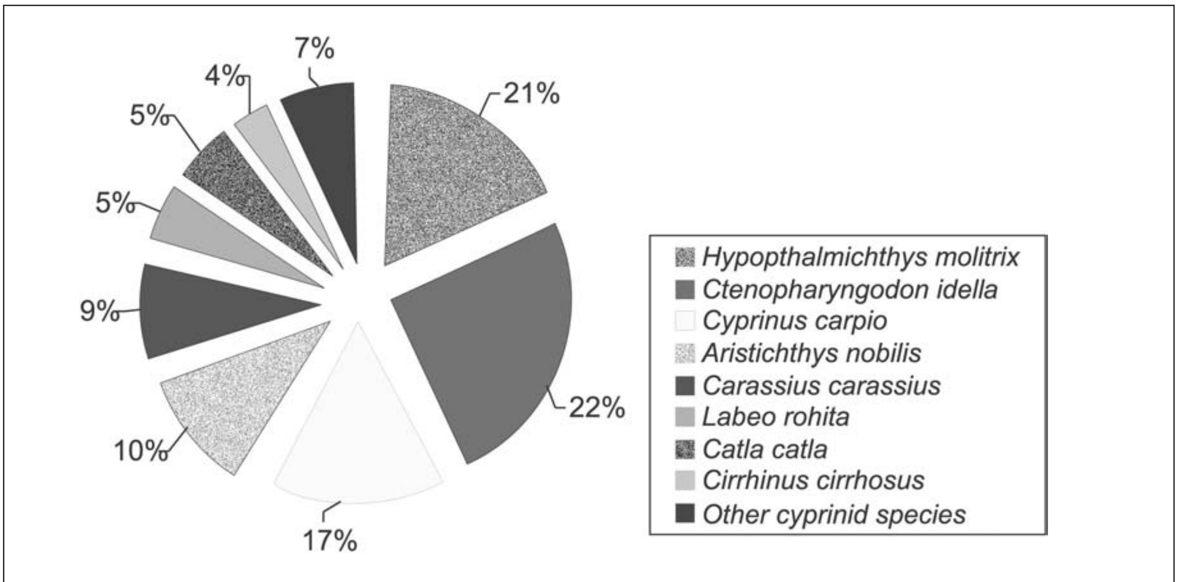

Fig. 1.2. Percentage of contribution of major carp species to the total carp production from aquaculture in Asia during 2001

improved strain, the GIFT strain that has shown 85 per cent higher growth as compared to the existing stocks in the Philippines (Eknath and Acosta 1998). Furthermore, testing of the GIFT strain through on-station and on-farm trials indicated the GIFT strain's faster growth as compared to the existing strains in Bangladesh, China, Thailand and Vietnam (Dey et al. 2000b). This has created interest among a number of major aquaculture producing countries in Asia and research is in progress for genetic improvement of *C. carpio, L. rohita,* silver barb (*B. gonionotus*) and Blunt snout bream (*Megalobrama amblycephala*), besides others, with funding support from the Asian Development Bank (WorldFish Center 2001). While most of these genetic improvement programs have as yet had little impact on aquaculture, there are some examples that demonstrate the kind of impact that can hopefully be expected to become much more widespread in the near future. Over one million tonnes of the Jian strain of common carp is produced each year in China, and several other improved varieties are cultured on a significant scale in China. Genetically improved varieties of several other cyprinid species have been developed and in some cases disseminated in several Asian countries.

In view of the significant growth of aquaculture and the need for increased production, there is a tendency among farmers and government agencies to introduce exotic species. The available records indicate that 259 introductions comprising 42 cyprinid species have taken place in 27 countries of Asia (Acosta and Gupta, Chapter 7, this vol.). In most cases these new species have led to improved aquaculture production, while in some cases they have also resulted in adverse impacts on the aquatic ecosystem.

Conservation and proper utilization of germplasm is a prerequisite for the sustainable management of and increased production from aquatic resources. In view of this, several major carp producing countries in Asia (Bangladesh, China, India, Indonesia, Thailand and Vietnam) have been involved in genetic improvement of carps under a project funded by the Asian Development Bank. As part of this, a review of the carp genetic resources in these countries was undertaken. The results of this are detailed in Chapter 3. The publication also includes a review of progress of genetics research in carps, introductions of carps and their impacts.

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Chapter 2

Carp Production in Asia: Past Trends and Present Status[1]

Madan Mohan Dey[2], Ferdinand J. Paraguas[2], Ramachandra Bhatta[3], Ferdous Alam[4], Miao Weimin[5], Somying Piumsombun[6], Sonny Koeshandrajana[7], Le Thi Chau Dung[8], and N V Sang[9]

2.1 Introduction

Of the total world carp production, 95 per cent was from Asia in 2001. In this region, there are more than 20 main native carp species, contributing about 80 per cent of the total freshwater fish production. China, India, Bangladesh and Indonesia accounted for most of the carp production in Asia: 80 per cent, 12 per cent, 3 per cent, and 1 per cent, respectively. Altogether, they contributed 95 per cent of the world carp production. Low-income people favor carps because of their low price and good taste. In many areas in Asia, carps are the major source of animal protein for the poor.

This chapter focuses on the status and future of carp production in Bangladesh, China, India, Indonesia, Thailand and Vietnam. The analysis is largely based on primary data collected through surveys of 2 025 carp fish farmers conducted during 1998-99. In addition to the primary data, databases compiled by the Food and Agriculture Organization of the United Nations (FAO) and unpublished reports from Asian countries were also used.

The chapter is composed of five sections. Following the introduction, the second section describes recent trends and the current production of carp in the region. The third section discusses the status of the carp industry in the six countries. This includes, among others, recent trends (growth) and the current production of carp species, species-wise production analysis and contribution of carp production to the total aquaculture and freshwater fish production in each country. Section 4 deals with the results of the producer survey conducted by the WorldFish Center and its partner institutions. This includes a description of the profile of carp producers including socioeconomic demography of carp farmers, characteristics of carp farmers and carp farming, stocking characteristics, producers' preference ranking of carp species and traits and their reasons for such preference. The future outlook on carp farming is also discussed in the section. Finally, concluding remarks follow this.

2.2 Recent trends and current production

Carp production is growing rapidly in Asia, increasing from 5.537 million t in 1990 to 16.313 million t in 2001, an annual growth rate of 11 per cent (Table 2.1). Among all the species of carps, Chinese carps i.e. silver carp (*Hypophthalmichthys molitrix*), grass carp (*Ctenopharyngodon idella*), common carp (*Cyprinus carpio*) and bighead carp (*Aristichthys nobilis*) are the major species, accounting for 23 per cent, 22 per cent, 16 per cent and 11 per cent, respectively, of the total Asian carp production in 2001. Indian major carps i.e. rohu (*Labeo rohita*), catla (*Catla catla*), mrigal (*Cirrhinus cirrhosus*) accounted for 5 per cent, 4 per cent and 4 per cent, respectively, in the same period. *H. molitrix, C. idella, C. carpio, A. nobilis* and crucian carp (*Carassius carassius*) are in the top position because of the influence of China, the main world producer of these species.

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1 WorldFish Center Contribution No. 1729.
2 WorldFish Center, Jalan Batu Maung, Batu Maung, 11960 Bayan Lepas, Penang Malaysia.
3 Department of Fisheries Economics, College of Fisheries, University of Agricultural Sciences, Mangalore, Karnataka, 575 002, India.
4 WorldFish Center-Bangladesh Office, House No. 22-B, Road No. 7, Banani, Dhaka 1213, Bangladesh.
5 Freshwater Fisheries Research Center, Chinese Academy of Fisheries Science, Asia-Pacific Regional Research And Training Center for Integrated Fish Farming, Wuxi Fisheries College, Nanjing Agricultural University, No. 1 Qitang, Wuxi 214081, China.
6 Department of Fisheries, Ministry of Agriculture and Cooperatives, Kasetsart University Campus, Phaholyotin Road, Bangkok 10900, Thailand.
7 Research Center for Marine Fisheries, Product Processing and Socioeconomics (RCMFPPSE), Ministry of Marine Affairs and Fisheries, Jl. RM. Harsono No. 8 Ragunan, Jakarta Selatan, Indonesia.
8 Research Center for Aquaculture No.1, Vietnam.
9 Vietnam Agricultural Science Institute, Vietnam.

Table 2.1. Carp Production from aquaculture of the selected countries, Asia and World (in metric tonnes)

Year	Bangladesh		China		India		Indonesia		Philippines		Thailand		Asia		World	
1990			4 093 124	(91.79)	628 157	(63.96)	131 725	(61.89)	4 780	(5.89)	18 945	(19.40)	5 069 717	(77.19)	5 627 775	(73.80)
1991			4 169 740	(90.14)	765 581	(64.59)	113 948	(58.63)	4 897	(5.57)	21 106	(17.17)	5 270 986	(75.61)	5 636 589	(71.62)
1992			4 817 666	(90.25)	894 801	(66.35)	125 018	(58.71)	7 474	(6.42)	28 930	(20.43)	6 076 646	(76.83)	6 433 814	(72.61)
1993			5 731 191	(88.55)	1 037 987	(76.62)	164 548	(67.14)	80	(0.07)	29 958	(18.53)	7 183 165	(78.43)	7 490 870	(74.37)
1994			6 980 027	(88.39)	1 067 280	(74.29)	174 764	(68.45)	4 249	(3.54)	32 954	(18.54)	8 496 347	(78.13)	8 767 159	(74.58)
1995			8 261 854	(87.82)	1 211 033	(76.22)	192 397	(68.75)	2 842	(2.91)	33 601	(16.74)	9 934 215	(78.62)	10 197 660	(75.36)
1996			9 517 916	(86.61)	1 459 506	(86.45)	230 124	(70.00)	2 004	(2.20)	45 174	(19.70)	11 475 070	(79.39)	11 744 451	(76.26)
1997	329 000	(94.76)	10 580 210	(85.56)	1 560 020	(86.86)	181 413	(62.07)	1 865	(1.77)	43 747	(18.22)	12 940 002	(80.98)	13 221 798	(77.82)
1998	381 000	(90.68)	11 228 999	(84.95)	1 551 437	(85.06)	143 822	(52.10)	4 755	(5.47)	50 105	(22.08)	13 604 766	(80.59)	13 916 887	(77.68)
1999	450 000	(87.87)	11 948 233	(84.03)	1 712 193	(83.32)	208 977	(62.55)	10 574	(10.03)	50 570	(20.02)	14 628 684	(79.56)	14 997 943	(76.79)
2000	504 000	(88.25)	12 380 911	(81.62)	1 703 357	(92.36)	224 868	(61.93)	10 682	(9.53)	54 482	(20.10)	15 137 148	(78.48)	15 524 966	(75.85)
2001	530 000	(88.55)	12 892 221	(80.83)	1 964 287	(93.61)	236 363	(58.94)	19 568	(15.82)	60 199	(20.78)	16 003 387	(78.06)	16 427 626	(75.54)

*Figures in parentheses indicate carp production as a percentage of freshwater aquaculture production in that country.
Source: FAO. 2003. Fisheries Statistics [Online] Available: http://www.fao.org/fi/statist/FISOFT/FISHPLUS.asp [2003, June].

A large increase in the total carp production in Asia has taken place mainly due to the very high annual growth rate (12 per cent) in carp aquaculture in the region over the last decade. During 1989-90, carp aquaculture contributed 91 per cent of the total production in Asia, which rose to 96 per cent during 1999-2000. At the same time, carp production from capture fisheries decreased at an average rate of 1 per cent annually.

2.3 Carp aquaculture in Asia

Bangladesh

Carp production accounted for 89 per cent of the total freshwater fish production in the country in 2001 (Table 2.1). Carps are by far the most important species in pond culture operations. *L. rohita, C. catla, C. cirrhosus* and *H. molitrix* together account for more than 78 per cent of (pond) production. About 88 per cent of all fish produced in ponds are carps. Polyculture of carps in ponds is most widely practiced in Bangladesh. Other culture systems include the integration of carp culture with rice farming and with poultry in the ponds. Carp culture in oxbow lakes (i.e. culture-based fisheries) is also practiced. As far as the intensity of polyculture of fish is concerned, pond culture may be rated as traditional to modified extensive[10] (high fingerling stocking rate and low use of feed and fertilizers). However, there are some commercial ponds that are practicing semi-intensive technology.

China

In 2001 China accounted for 79 per cent and 76 per cent of the total carp production of Asia and the world, respectively. Carps are widely cultured, despite the introduction of numerous exotic freshwater species. Eight of the 10 major carp species being cultured are of national economic importance, including black carp (*Mylopharyngodon piceus*), *C. idella, C. carpio, H. molitrix, A. nobilis, C. carassius* and Chinese bream (*Megalobrama amblycephala* and *Parabramis pekinesis*). Production of these eight species reached nearly 12 million tonnes in 2001 and together accounted for about 73 per cent of the total freshwater aquaculture production. *H. molitrix, C. idella, C. carpio, A. nobilis* and

[10] Aquaculture can be broadly classified as extensive, having no feed or fertilizer; semi-intensive, having some fertilizer and/or feed inputs; and intensive, largely reliant on feed inputs (Edwards et al. 1988; Edwards 1993; Molnar et al. 1996; Pullin 1993). As these systems are conceptual stages in a continuum and in actual practice modified to suit farm conditions, they are often categorized as "modified extensive", "modified semi-intensive", "modified intensive", "super intensive", etc. (Pillay 1997).

C. carassius accounted for 93 per cent of the total carp production. However, the share of carp production in the total aquaculture production was decreasing despite the fact that carp production was growing annually by 13 per cent. In 1990, the share was 92 per cent, which declined to 81 per cent in 2001. Growth in production of important species was 9.1 per cent for *C. idella*, 1.5 per cent for *H. molitirix*, 6.4 per cent for *C. carpio*, 19.7 per cent for *C. carassius*, 13.5 per cent for *M. piceus* and 39.5 per cent for *M. amblycephala* and *P. pekinensis*.

Currently, carps are grown in a polyculture system, but carp monoculture is also becoming popular for intensive culture in cages, ponds and running water systems. Culture environments include ponds, lakes, brooks, reservoirs and paddy fields, in a total area of 4 955 000 ha which comprises 1 994 000 ha in ponds, 880 000 ha in lakes, 1 568 000 ha in brooks, 371 000 ha in reservoirs, 1 586 000 ha in rice and paddy fields and 142 000 ha in other freshwater bodies. The highest production comes from ponds (4 474 kg/ha), followed by brooks (1 623 kg/ha), lakes (921 kg/ha) and reservoirs (743 kg/ha). Culture in China is very intensive with relatively high stocking and good use of feed and fertilizers. Carp culture takes place on state owned, collective, family-owned and private commercial farms.

India

Carps dominate aquaculture production in freshwater ponds, cages, pens and recirculating systems and production in inland fisheries. *C. catla*, *L. rohita* and *C. cirrhosus* accounted for 25 to 27 per cent each of the total aquaculture production in the country during 2001. These species together constituted about 85 per cent of the total freshwater aquaculture production during the same period. In freshwater aquaculture, only carp (pond) culture has reached commercial importance. About 93 per cent of the total carp production is coming from aquaculture with an average annual growth rate of 10 per cent during 1990-2001. On the other hand, production from capture fisheries has decreased by 7 per cent during the same period (Table 2.1).

The national productivity of carp culture in ponds more than doubled during the last one and a half decades, from less than 1 tonne/ha/year during the mid 1980s to more that 2 tonnes/ha/year in the early 2000s (Katiha 2000, Ayyappan and Jana 2003). Polyculture takes place in four systems, extensive, improved extensive, semi-intensive or intensive. Polyculture in an improved extensive system forms the major practice all over the country (Wahab et al. 2001). In general, culture may be considered moderately semi-intensive as it is limited by low applications of feed and fertilizers.

Indonesia

Carps are the most important cultured species in Indonesia. Production of carps from aquaculture increased by 6 per cent per annum while capture fisheries decreased by 1 per cent during 1990-2001. The most important species are *C. carpio*, Java barb (*Barbonymus gonionotus*) and Nile carp (*Osteochilus hasseltii*). Indonesia is the world's third largest producer of *C. carpio* after China and the USSR. In 2001, *C. carpio* contributed about 90 per cent of the total carp production.

Cultured carps account for about 86 per cent of all carps produced in the country. Like China, although carp production has been on the increase, its share in the total aquaculture production eventually declined from 62 per cent in 1990 to 59 per cent in 2001.

Presently, carp culture largely takes place in three environments: ponds, floating cages and rice fields (Kontara and Maswardi 1999). The contributions from these systems to total aquaculture production in the country comprised 9.1 per cent from brackish water ponds, 4.1 per cent from freshwater ponds, 1.0 per cent from cages and 2.4 per cent from paddy fields. Production from culture grew by 8.4 per cent annually for brackish water ponds, 7.4 per cent for freshwater ponds and 2.4 per cent for paddy fields during 1987-96; the largest rise in production took place in cage culture, accounting for a 45.8 per cent annual growth during this period (DGF 1999).

Thailand

At present, fish culture in Thailand is one of the fastest growing industries in the region. Carp production from aquaculture had an average annual growth rate of 10 per cent during 1990-2001 where it accounted for about 20 per cent of the total aquaculture production and 56 per cent of the total carp production in the country. Overall carp production increased with an annual average growth rate of 8 per cent during the last decade.

B. gonionotus is the most important carp species in aquaculture (third among the freshwater species after tilapia and catfish) accounting for about 42 per cent of the total carp production in 2001. *C. carpio* accounted for 15 per cent during the same period.

Carp farming is carried out in ponds, paddy fields, ditches and cages. Polyculture in ponds is by far the most popular in terms of the number of farms and culture area. *H. molitrix*, *C. carpio*, *C. cirrhosus* and various species are normally cultured in polyculture together with other omnivorous species like Nile tilapia (*Oreochromis niloticus*). In addition carp culture is integrated with pig and poultry rearing and rice farming.

Vietnam

Rice farmers in Vietnam have practiced traditional carp culture in rice fields and village ponds for a long time. The main cultured species were *C. carpio* and other indigenous species. In the 1960s, with the introduction of *H. molitrix, A. nobilis* and *C. idella* together with induced breeding, freshwater fish culture entered into a new era. In spite of this, carp farming was not important to the rural economy of Vietnam until the introduction of Indian carps *C. catla*, *L. rohita* and *C. cirrhosus*, in the early 1980s. In 1996, carp culture contributed about 29 per cent (0.40 million tonnes) to the total fish production (1.37 million tonnes). The Mekong River Delta plays a most important role in aquaculture, producing about 67 per cent of the total aquaculture production.

Carps are mainly cultured in a polyculture system. The main species are *H. molitrix*, *C. idella*, *A. nobilis*, *L. rohita*, *C. cirrhosus* and local fish species (*Pangasius* sp, *C. carpio*). The culture system is primarily semi-intensive with the use of only minimal amounts of fertilizers, rice bran and other agricultural on-farm and off-farm by-products as feed. In addition, an integrated VAC system (V: garden, A: fish pond, C: livestock) is also common in the Red River Delta.

2.4 Profile of carp producers in Asia

This section discusses the results of the surveys of 2 025 carp producers undertaken by the WorldFish Center and its partner institutes during 1998-99.

Table 2.2. Socio-demographic characteristics of carp producers in Asia. Values shown are averages from each country

Items	Bangladesh	China	India	Indonesia		Thailand	Vietnam	
				RWS[a]	Cages		Northern	Southern
Sampled farm households	540.00	383	409.00	40.00	71.00	284.00	158.00	240.00
Age of farmers (years)	45.00		47.00	46.55	40.87	49.77	43.00	52.00
Gender (%)								
Male	100.00	100	87.00			95.10	43.90	51.40
Female			13.00			4.90	56.10	48.60
Education (years)	8.00	12	7.42	7.43	8.07	5.38	8.80	6.00
Illiterates (%)	11.00		32.70			1.80		4.35
Primary Occupation (%)								
Fish culture	9.00	100	43.70	92.50	94.40	20.10	2.00	7.90
Crop farming	65.00		41.10	2.50	1.40	60.60	87.40	44.60
Animal farming	2.00		2.20		4.20	7.00	10.60	0.80
Others	24.00		12.50	5.00		12.30		46.70
Experience in carp farming (yrs)	13.00	15	6.00	13.00	5.00		10.00	7.00
Gross per capita income (US dollars)	293.00	4 949[b]	1 113.00			2 424.00	575.00	885.00
Income Sources (%)								
Fish culture	14.93	64.00	79.66			20.01	27.60	27.58
Crop farming	28.93	3.00	13.10			13.03	29.40	58.15
Animal farming	3.19	3.00	0.03			48.41	27.30	14.20
Hatchery and seed Production		20.00	6.35				6.20	
Business and salaries	32.55	6.00	0.55				7.40	
Others	20.00	4.00				18.55	0.10	0.08
Average household size (no.)	5.50	3.5	8.00	3.35	3.73	4.65	5.00	5.81

[a] RWS = Running water system: fish are held in pens in small streams and irrigation systems.
[b] The gross per capita income of China refers only to family-based farms. The average gross income of co-operative and state owned farms ranges from US$ 53 179 to 149 135 per farm.

Source: WorldFish Center Field Survey (1998-99).

Socio-demography of carp farmers

The socio-demographic characteristics of the carp producers in Bangladesh, China, India, Indonesia, Thailand and Vietnam are described in Table 2.2 including details on age, gender, sources of income and occupation. The average age of the farmers ranged from 40-52 years. The male head of the family usually carried out carp farming. In Vietnam, the participation of women in aquaculture is as high as 50-56 per cent (for southern and northern Vietnam, respectively) indicating that carp farming is potentially gender neutral. The average educational level of the farmers varied from only five[11] years in Thailand to 12 years in China. The higher educational profile of farmers in China perhaps enhanced production. In India, 32.7 per cent of the farmers were illiterate, a factor which may explain this country's lower level of production. Except for China and Indonesia, fish farming is not the primary occupation of the majority of the fish farmers surveyed.

Experience in carp farming is an important factor influencing the production; carp producers in China were most experienced (15 years) with the least in India (6 years). The average annual gross per capita income of the farmers was as low as US$ 293 in Bangladesh to as high as US$ 4 949 in China. The values for other countries were US$ 2 424 for Thailand, US$ 1 113 for India, US$ 575 for northern Vietnam and US$ 885 for southern Vietnam. In general, the gross per capita income of the carp farmers was above the national average income. Fish culture contributed up to 80 per cent in India and as low as 15 per cent in Bangladesh to household income. The contribution of carp farming to household incomes in India varied among the states, e.g. from 15 per cent in Orissa to 95 per cent in Andhra Pradesh.

General characteristics of carp farmers and carp farming

General characteristics of the carp farms with pond systems in the six countries are shown in Table 2.3. Private owners usually operate the carp farms, except in China and northern Vietnam, where a large proportion of farms are owned by the state or by collectives. In India, the Irrigation Department owns about 30 per cent of the common water bodies, which are used by the Fisheries Department for stocking. Joint ownership is common in India, Thailand and Vietnam.

The size of culture ponds varies among the countries. The average area cultivated by a household is as high as 4.24 ha in India and as low as 1.04 ha in southern Vietnam. In China, the size of a family-based farm is 3.6 ha, on average. State-owned large-scale farms can be as large as 131 ha. The farm area allocated to fishponds was 31 per cent in northern Vietnam, 23.5 per cent in India and 26 per cent in Thailand. The average size of a fishpond in China was 1.70 ha, 1.21 ha in Thailand and 1.16 ha in northern Vietnam. The average size of a fishpond was only 0.20 ha in Bangladesh, where they are used for various purposes in addition to stocking with fish. The polyculture of fish in ponds is the main fish culture system in all these countries, except in Indonesia where monoculture in running water systems and cages are common. In southern Vietnam, 30 per cent of the farmers practiced monoculture. Integrated fish farming and rice fish farming constituted a major part of the aquaculture in Thailand and Vietnam.

Stocking Characteristics

Carp farming practices, including stocking density, species stocked, sources and size of fingerlings, are found in Table 2.4. The average stocking density in fishponds was highest in Thailand (6 7328 pieces/ha), followed by South Vietnam (2 8200/ha) and China (2 7867/ha). *C. catla*, *L. rohita* and *C. cirrhosus* accounted for the largest proportion of stocked fish in India and Bangladesh. *H. molitrix* and *B. gonionotus* were also very important in Bangladesh, accounting for about 20 per cent and 13 per cent of stocked fish, respectively. In China, *H. molitrix* (28 per cent) and *C. carpio* (22 per cent) predominate. In Thailand, *B. gonionotus* accounted for 40 per cent of stocked fish, followed by *O. niloticus* (37 per cent) and *C. carpio* (8 per cent) (this refers only to systems in which carps are produced). In northern Vietnam, *H. molitrix* and *L. rohita* are the dominant carp species stocked, while in southern Vietnam, *B. gonionotus* and *C. carpio* are the dominant carps stocked.

In all the surveyed countries, fingerlings were supplied by private and government hatcheries, except in China where the farmers produced the fingerlings themselves. In Thailand, private hatcheries provided 74 per cent of the total supply of fingerlings, followed by 61 per cent in India and 55 per cent in northern Vietnam.

[11] Although the general level of literacy is quite high in Thailand compared to many other Asian countries, the level of education for carp farmers is low. Commercial fish farmers from the Central Plains of Thailand have a much higher level of education.

Table 2.3. General characteristics of carp farming in Asia

Items	Bangladesh	China	India	Indonesia	Thailand	Vietnam		
				RWS	Cages		Northern	Southern
Farm Area (ha)		3.59[a]	4.24	2.29	2.87	3.98	3.67	1.04
Crop land (%)		8.55[b]	24.76			50.80	43.30	80.69
Water spread area (%)		83.11	44.85			26.04	47.90	18.11
Fish-pond area (%)		17.95	23.51			25.63	31.60	7.94
Homestead area (%)			1.20			5.06	4.80	3.40
Animal farming						0.73		
Unutilized area			5.45				3.90	
Others			0.25			4.40		
Size of the fish pond (ha)	0.20	1.70	0.87			1.21	1.16	0.82
Fish farm area by tenure (%)								
Privately owned	100.00	41.10	62.60	100.00	100.00	90.10	35.00	95.70
State owned		29.60	29.30			0.70	45.00	0.57
Collective		29.30	2.20			8.50	17.80	
Leased /Rented			6.80			0.70		3.73
Others			1.20				2.20	
Type of operation (%)								
Single ownership	86.70	100.00	71.00	100.00	100.00	85.40	88.00	99.12
Joint ownership			26.90			14.60	22.00	0.88
Lease operated	13.30							
Culture period (months)	9-12	8-11	8-12	3-4	3-4	5-12	c	c
Culture type (%)								
Seasonal	26.30		13.00			8.50	8.10	41.42
Perennial	73.70	100.00	87.00	100.00	100.00	91.50	91.90	58.48
Pond system								
Monoculture		4.20				8.50	1.80	30.50
Polyculture	100.00	92.30	100.00			91.50	98.20	69.50
Mono+polyculture		3.50						
Cage culture								
Monoculture					100.00			33.33
Polyculture								66.67
Rice fish farming								
Monoculture						100.00		12.90
Polyculture								87.10
RWS								
Monoculture				100.00				

[a] The average total area refers to small-scale farms. For large-scale state owned farms it is 131.80 ha.
[b] The percentage of pond area refers to the water-spread area.
[c] Not available.
Source: WorldFish Center Field Survey (1998-99).

In Indonesia, where only *C. carpio* were stocked in cages and running water systems, 48-55 per cent of the total supply of fingerlings came from government hatcheries. Middlemen played an important role in supplying fingerlings for cage culture in Indonesia.

Fingerling size at stocking influences the pond yield. Stocking size is expressed in different units among the countries. Stocking size varied between 1-5 cm with an average of 3 cm for Indian major carps in India and Bangladesh. In China, stocking sizes varied between 20-250 g for *H. molitrix* and up to 1 kg for black carps that represent the largest stocking size for Asia. Similarly, in northern Vietnam stocking sizes were large, ranging from 63 g for *H. molitrix* to 382 g for black carp. In southern Vietnam stocking sizes were smaller, ranging from 1.2 g for *L. rohita* to 5.3 g for *A. nobilis.* In Indonesia, the stocking size of *C. carpio* for monoculture in cages and running water system varied between 25-100 g.

Producers' preference ranking for different species

Producers' preferences for stocking of various species are shown in Table 2.5. In Bangladesh, producers ranked *L. rohita* as the most preferred species followed by *H. molitrix*, *C. cirrhosus* and *C. catla*. In China, producers gave highest preference for *C. idella*, followed by *H. molitrix* and *C. carassius*. In India, *L. rohita*, *C. catla* and *C. carpio* were the most preferred. In Thailand, farmers primarily preferred *O. niloticus*, but among carps they preferred *B. gonionotus* and *C. carpio* most. In northern Vietnam, the first preference was for *C. idella* followed by *C. carpio*, *H. molitrix* and *L. rohita* each with equal ranking. In southern Vietnam, farmers preferred *Pangasius hypophthalmus* most, followed by *C. carpio* and *B. gonionotus*.

Table 2.4. Carp farming practices in Asia

Items	Bangladesh	China	India	Indonesia	Thailand	Vietnam		
				RWS	Cages		Northern	Southern
Average stocking density*	10 261.00	27 867.00	18 408.00	56.50	136.56	67 328.00	5 432.00	28 200.00
Species composition	24.10		31.00			4.93	22.90	0.11
(%)	16.13		26.06					0.01
Labeo rohita	16.45		17.77			4.47	7.40	2.68
Catla catla	4.49	21.90	6.44	100.00	100.00	8.37	4.90	17.30
Cirrhinus cirrhosus	2.80	20.90	4.18				8.70	1.54
Cyprinus carpio	19.68	27.96	7.17				28.10	2.83
Ctenopharyngodon	13.04					39.88		20.00
idella	0.55							
Hypophthalmichthys		8.97					2.30	4.33
molitrix		3.11						
Barbonymus								
gonionotus		14.93						
L.calbasu		0.69						
Aristichthys nobilis						36.76		
Mylopharyngodon	2.74	2.23	6.85			4.26	25.70	51.20
amblycephala								
Carassius carassius								
M. piceus								
Tilapia								
Others								
Source of fingerlings								
(%)	5.00	90.00	0.54	2.50	5.72	4.03	23.60	2.00
Own	40.00		61.85	42.50	13.46	74.20	54.50	79.00
Private hatchery	20.00		25.00	55.00	48.08	21.77	7.90	11.00
Government hatchery	35.00	10.00	13.00		32.09		13.80	8.00
Middlemen and others								

* For Indonesia, stocking density is in kg/100 m^2, while the others are in number/ha.
Source: WorldFish Center Field Survey (1998-99).

Table 2.5. Producers' preference for freshwater species in Asia. In Thailand and northern Vietnam, some species had tied rankings

Rank	Bangladesh	China	India	Thailand	Northern Vietnam	Southern Vietnam
1	*L. rohita*	*C. idella*	*L. rohita*	*O. niloticus*	*C. idella*	*P. hypophthalmus*
2	*H. molitrix*	*H. molitirx*	*C. catla*	*B. gonionotus*	*C. carpio /L. rohita/C. cirrhosus/ H. molitrix/ O. niloticus/ M. piceus*	*C. carpio*
3	*C. cirrhosus*	*C. auratus*	*C. carpio*	*C. batrachus*		*B. gonionotus*
4	*C. catla*	*C. carpio*	*C. idella*	*C. carpio*		*L. rohita*
5	*B. gonionotus*	*A. nobilis*	*C. cirrhosus*	*L. rohita/C. cirrhosus*		*C. cirrhosus*
6	*C. carpio*	*M. amblycephala*	*H. molitrix*	Chinese carps		*A. nobilis*
7	*C. idella*	*M. piceus*				*H. molitrix*
8	*L. calbasu*					

Source: WorldFish Center Field Survey (1998-99)

Reasons for preferences of carp species

The reasons given for carp species preference by the producers are listed in Table 2.6. Rapid growth, high market value and better meat quality were important reasons mentioned by farmers. In Bangladesh, *L. rohita* was preferred because of its higher market price, which reflects consumer preference. Farmers in Bangladesh also ranked *H. molitrix* highly, due to its rapid growth. In China, growth was the major factor for selecting *C. idella, H. molitrix* and *C. carpio*. The Chinese farmers also mentioned that better meat quality is a crucial factor for selecting *C. carassius* and *C. carpio*. In India, a majority of the farmers preferred *C. catla* and *L. rohita* because of their higher market prices and *C. carpio* because of more rapid growth. Farmers in Thailand preferred *B. gonionotus* due to its higher growth and ease of culture. Farmers of northern Vietnam preferred *C. idella* due to its rapid growth. The reason for preferring *C. carpio* and *B. gonionotus* in southern Vietnam was their more rapid growth. Earlier studies in Indonesia revealed that preference varied across regions. People from central and east Java do not like red *C. carpio* (Ardiwinata 1981) while it is favored by the people in north Sumatera (Sumantadinata 1995).

Table 2.6. Reasons for preferring particular species

Countries	Species	Reasons
Bangladesh	*L .rohita*	Higher market price
	H. molitrix	Higher growth
	B. gonionotus	Good flavour/taste
China	*C. idella* *H. molitrix* *C. carpio*	Higher growth
	C. auratus *C. carpia*	Better meat quality
India	*C. catla* *L. rohita*	Higher market price
	C. carpia	Higher growth
Thailand	*B. gonionotus* *C. carpio*	Ease of culture Higher growth
	C. carpio	Higher market price
Southern Vietnam	*C. carpio* *B. gonionotus*	Higher growth
Northern Vietnam	*C. .idella* *C. carpio*	Higher market price Higher growth

Source: WorldFish Center Field Survey (1998-99).

Traits preferred by producers

Size, color and shape of the carps preferred by producers are shown in Table 2.7. In some cases, such as in Indonesia, it is known that there is a genetic basis to some of the variations observed in the color and shape of the common carp stocks. However, in most cases (e.g. Indian major carps), it is likely that rearing conditions influenced variations in color and shape.

Farmers in Bangladesh and India preferred fish of one kilogram in size, whereas Thai farmers were willing to grow fish to half that size. Farmers had differing preferences in color and shape. The preferred color for *L. rohita* by Bangladesh farmers was bright and reddish, but Indian farmers preferred brown fish. Thai farmers preferred white *L. rohita*. Farmers in India and Thailand preferred different shapes of *L. rohita*. Indian farmers favored long and thin fish bodies, but Thai farmers preferred shorter and thicker bodies.

Table 2.7. Preferred size, color and shape of the most popular species

Species/	Bangladesh	China	India	Indonesia		Thailand	Northern Vietnam
Trait				RWS	Cages		
L. rohita							
Size (no/kg)	1		1			<2	
Color	Bright & reddish		Brown			White	Bright
Shape	Long & thick		Long & thin			Short & thick	Short & thick
C. catla							
Size (no/kg)	1		1				
Color	Bright & reddish		Brown				
Shape	Short & thick		Short & thick				
C. cirrhosus							
Size (no/kg)	1		1			<2	
Color	Bright		Brown			White	Black green
Shape	Long & thick		Long & thin			Long & thin	Short & thick
C. carpio							
Size (no/kg)	1	1	1	>2	<2		
Color	Bright & yellow	Reddish	Reddish	Greenish	Greenish	Silver/ Green/ Grey	Bright
Shape		Short & thick	Short & thick	Short & highback	Short & high back	Short & thick	Short & deep
B. gonionotus							
Size (no/kg)	3					2-3	
Color	White					White	Bright
Shape	Short & thick					Short & thick	Short & thick
H. molitrix							
Size (no/kg)	<1	2				<2	
Color	Silver white	Silver white				White	Silver
Shape	Long & thick	Short & thick				Short & thick	Short & thick

Source: WorldFish Center Field Survey (1998-99).

For all the species, the most preferred size was one kg/fish in Bangladesh and India, except for *B. gonionotus* in Bangladesh. Farmers using the running-water system in Indonesia preferred smaller sizes for *C. carpio* (> 2 pieces of fish/kg) compared to India and Thailand, where the preferred size was one to two/kg. Preferences of cage farmers in Indonesia were similar to those of farmers in Thailand and India. Farmers in Bangladesh and Thailand showed similar preferences for shape of *B. gonionotus* (short and thick).

2.5 Conclusions

Carps are by far the most important species for freshwater aquaculture of the countries under study, except in Thailand where tilapia and catfish topped the rankings. The major carp species grown in each country varied significantly. This could be due to the difference in production practices of these countries taking into account the bionomic characteristics of the species themselves. The Chinese carps have a long history of culture in China (the P.R. of China and Taiwan) but were less domesticated than *C. carpio* since captive breeding by induced spawning has been widespread only since the 1960s (Pullin 1986).

The ranking of producers' preferences showed that *L. rohita, C. catla, C. idella* and *H. molitrix* were some of the most preferred species in Asia. The reasons for this are rapid growth and better market prices. The top positions of Chinese carps in Asian production are due to the influence of production from China.

Nevertheless, many producers in Thailand, China and India showed a willingness to try new strains if made available. The majority of farmers in Thailand were willing to expand the area of carp farming and to continue the existing mode of operation. Results from the survey also indicated that attitudes toward continuing carp farming were positive in these countries.

Most of the countries cultured carps in a polyculture system. It is of interest to understand the biological and economic reasons for carp polyculture. The feeding niches of some component species are known, but the yield optimization and economic analyses and concepts of balance and competition between species are poorly understood.

The profile of carp producers and production systems varies among the countries. Farmers in China with better educational backgrounds and longer experience have a higher proportion of their income generated through fish farming. Although the average proportion of income from carp farming for Indian farmers is 79 per cent, income from carp farming varies considerably among states. The gross household income of the Chinese farmers is the highest, followed by Thai farmers. The area under fish culture as a proportion of the total farm area varies between 20-30 per cent among the countries. The average size of fishponds ranges from 0.2 ha in Bangladesh to 1.7 ha in China. Farms are mostly privately owned, except in China and northern Vietnam, where state ownership plays an important role.

The average stocking density varied between 5 432 fingerlings per ha in northern Vietnam to 66 927 fingerlings in Thailand. However, the proportion of carps in the total stocking constituted only about 60 per cent in Thailand and 49 per cent in southern Vietnam. Producer preferences for size, color and shape varied among countries. However, there is more uniformity among farmers in size preferences (1-2/kg).

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Chapter 3
Carp Genetic Resources

3.1 Carp Genetic Resources of Bangladesh

M.G. Hussain[1] and M.A. Mazid[1]

3.1.1 Introduction

Bangladesh, a country dominated by the multiple deltas of the Ganges, Brahmaputra and Meghna River systems, has vast water resources and is also rich in biological diversity with approximately 296 fresh and brackish water fish species (including freshwater prawns) and 511 marine species (including marine shrimps). Approximately 93 per cent of inland fishery resources is composed of open water bodies like rivers, floodplains, natural depressions and Kaptai reservoir, which are the natural breeding grounds for all the endemic freshwater fishes including cyprinids. In recent years, inland open water fisheries have been under heavy pressure due to the deterioration of environmental conditions: siltation and soil erosion in river basins; water pollution from industrial, agricultural and municipal wastes; construction of embankments for flood protection; injudicious and destructive fishing practices; outbreak of diseases; and loss of natural breeding grounds as a result of habitat degradation. The reduced carrying capacity and productivity of most of the aquatic environments has affected fish biodiversity. Owing to such natural and man-made changes, a good number of valuable fish species dwelling in freshwater ecosystems have become threatened or endangered and some already face the threat of extinction.

Although the government is making strenuous efforts to rehabilitate the inland fisheries, it has focused its attention on aquaculture, which has a tremendous potential for development. To avoid loss of genetic diversity and inbreeding in hatchery populations, development of improved brood stocks through implementation of effective breeding plans and genetic stock improvement programs for commercially important carp species has recently been identified as an important area of research (Hussain and Mazid 1999a). Genetic studies on carp species using the techniques of chromosome manipulation (viz. gynogenesis and triplody) and sex reversal were initiated first (1986-97) and presently stock improvement of silver barb (*Barbonymus gonionotus*), catla (*Catla catla*) and rohu (*Labeo rohita*) using selective breeding techniques is in progress. In view of conserving biodiversity of threatened carp species, artificial breeding techniques for *Tor putitora*, *Puntius sarana*, *Labeo gonius*, *Cirrhinus ariza*, *Labeo calbasu* and *L. bata* have been developed.

3.1.2 Cyprinid species of importance to aquaculture

3.1.2.1 Endemic cyprinid species

The endemic cyprinid species of Bangladesh can be divided into two sub-groups: (i) "major" carps (e.g. *C. catla, L. rohita, Cirrhinus cirrhosus, L. calbasu*); (ii) "minor" carps (e.g. *L. bata, C. ariza, L. nandina, L. gonius*). Most of the freshwater river systems and floodplains of the country are the natural breeding grounds for all the major and minor carps. There are at least 13 endemic species of carps, from 6 genera, which are of interest to aquaculture in Bangladesh (Table 3.1.1).

Table 3.1.1. List of endemic cyprinid species of Bangladesh of interest to aquaculture

Species	Common name	Local name
Labeo rohita	Rohu	Rui
Catla catla	Catla	Katla
Cirrhinus cirrhosus	Mrigal	Mrigal
Cirrhinus ariza	Reba	Bhangan
Labeo calbasu	Calbasu	Kalibaush
Labeo bata	Bata	Bata
Labeo boga	Boga labeo	Bhangan
Labeo gonius	Gonius	Gonnia
Labeo nandina	Nandina labeo	Nandil
Bengala elanga	Bengala barb	Along
Puntius sarana	Barb	Sarpunti
Tor tor	Tor mahseer	Mahashoal
Tor putitora	Putitor mahaseer	Mahashoal

Source: Rahman (1985), Hasan (1990) and Hussain and Mazid (2001).

[1] Bangladesh Fisheries Research Institute, Mymensingh 2201, Bangladesh.

All the species belonging to the major carp sub-group are the natural inhabitants of the freshwater sections of the rivers of Bangladesh, Burma, northern India and Pakistan (Jhingran and Pullin 1985). In Bangladesh, these species are mostly found in the Padma-Brahmaputra River system (i.e. Padma, Jamuna, Arial Khan, Kumar and Old Brahmaputra rivers) and the Halda River system in Chittagong. Their favorite habitat is the deep pools of these rivers. During the monsoon time they breed in the inundated rivers and flowing waters. Mahseer (*Tor* spp.) are the inhabitants of hilly streams of Mymensingh, Sylhet, Dinajpur districts and Kaptai Lake of the Chittagong Hill tracts. The natural spawning grounds of *Tor* spp. consist of a sandy bottom, pebbles and aquatic weeds, where temperatures are relatively low and dissolved oxygen is sufficient (Pathani 1982).

On the other hand, all the other species belonging to the minor carps are the natural inhabitants of small rivers and floodplains. Shallow freshwater zones of the North-East (Mymensingh, Netrokona and Mohanganj), South-West (Faridpur and Jessore) and North-West (greater Rajshahi area) floodplains in Bangladesh are the natural habitat of minor carps. They naturally breed there and grow to maturity within a short period of time (10-12 months).

3.1.2.2 Exotic carp species

Although Bangladesh is rich in endemic fish genetic resources, introduction of different species of fish (mostly Chinese carps) has occurred since 1960. Such introductions of exotic fish species have not been properly recorded. The only document describing these introductions is by Rahman (1985). Subsequently, BFRI maintained its record of new fish introductions for research purposes (Hussain 1997). A list of different species of introduced species of carps, made on the basis of these records, is in Table 3.1.2. Additionally, stocks of silver carp (*Hypophthalmichthys molitrix*), bighead carp (*Aristichthys nobilis*) and grass carp (*Ctenopharyngodon idella*) were obtained from wild populations in China. Among the introduced carp species, *H. molitrix*, *C. idella*, *A. nobilis*, common carp *(Cyprinus carpio)*, black carp *(Mylopharyngodon piceus)*, *B. gonionotus* and *Tor* spp. are predominant.

All these indigenous and exotic carps are used in polyculture in ponds. Some are stocked in floodplains, rivers and reservoirs to enhance fishery production.

3.1.2.3 Induced breeding of fish and recent decline in quality of hatchery stocks

The recent advancement of aquaculture, in particular the carp polyculture and freshwater fish farming in rice fields, seasonal ditches, canals and perennial ponds, is mainly due to the advent of induced breeding or artificial propagation techniques by hypophysation during the late 1950s. In Bangladesh, the production of seed of endemic carps through artificial breeding has become a common practice since 1967 (Ali 1967). Meanwhile, Chinese carps, exotic barbs and catfishes have been introduced and a large number of hatcheries in the private sector (estimated to be about 635) have been established (Ali 1998). These hatcheries presently contribute more than 98 per cent of the total spawn (three-day-old hatchlings) production (see Table 3.1.3) and the rest, a very negligible proportion, is collected from natural sources, mainly rivers and their tributaries (Banik and Humayun 1998).

Table 3.1.2. List of introduced cyprinid species of Bangladesh

Species	Common name	Source	Year of introduction
Ctenopharyngodon idella	Grass carp	Hong Kong Nepal Japan	1966 1979 1970
Mylopharyngodon piceus	Black carp	China	1983
Hypophthalmicthys molitrix	Silver carp	Hong Kong	1969
Aristichthys nobilis	Bighead carp	Nepal	1981
Cyprinus carpio var. communis	Common carp	China Vietnam	1960 1995
Cyprinus carpio var. specularis	Mirror carp	Nepal Hungary	1979 1982, 1996
Barbonymus gonionotus	Silver barb	Thailand	1987, 1994
Tor putitora	Putitor mahseer	Nepal	1991

Source: Rahman (1985), Hasan (1990) Hussain (1997) and Hussain and Mazid (2001).

Table 3.1.3. Number of fish hatcheries and spawn (three-day-old hatchlings) production in Bangladesh

Year	No. of hatcheries		Spawn production (kg)[1]	
	Public sector	Private sector	Natural sources	Hatcheries
1988	77	162	12 533	6 849
1989	84	185	12 236	5 664
1990	89	204	5 128	14 773
1991	89	218	6 855	24 683
1992	89	222	9 342	35 851
1993	102	256	5 069	48 964
1994	102	389	5 872	72 536
1995	141	502	9 144	88 272
1996	141	600	2 399	103 615
1997	141	631	2 824	115 888
1998	141	635	2 885	117 700

[1] 1 kg spawn = approximately 400 000 three-day-old hatchlings
Source: Hussain and Mazid (1999b, 2001).

Table 3.1.4. List of threatened cyprinid species of Bangladesh

Scientific Name	Local name	Critically Endangered	Endangered
Labeo nandina	Nandil	X	
Labeo boga	Bhangan	X	
Labeo gonius	Ghonnia		X
Labeo bata	Bata		X
Labeo pangusia	Ghora maach		X
Labeo calbasu	Kalibaush		X
Cirrhinus ariza	Bhangan		X
Puntius sarana	Sarpunti	X	
Puntius ticto	Tit punti		X
Tor tor	Mahashoal	X	
Tor putitora	Mahashoal	X	

Source: Hussain and Mazid (1999a, 2001).

Recently, in Bangladesh it has been reported that hatchery produced stock, mainly the carps, have shown reduction in growth and reproduction performances, increased morphological deformities, disease and mortalities, etc. This is probably due to genetic deterioration in the hatchery stocks as a result of poor fish brood stock management (i.e. unconscious negative selection and use of undesirable size of breeders for mating), inbreeding depression (due to sib or parent-offspring mating) and ignorance of private hatchery managers regarding overall brood stock management and hatchery operation. Such poor brood stock management and close mating of breeders results in inbred or highly homozygous hatchery populations leaving adverse effects on aquaculture production as well as on open water fisheries in view of stocking of poor quality hatchery bred fish seed in the floodplains and other open water bodies.

3.1.2.4 Degradation of aquatic habitat and threat to natural fish resources

IUCN, Bangladesh (2000) has listed about 54 freshwater fish species of the country as endangered, of which several are critically endangered. Among the endangered species are 11 endemic carp and barb species belonging to the family Cyprinidae (Table 3.1.4).

3.1.3 Documentation and evaluation of genetic resources

3.1.3.1 Morphometric and meristic characteristics of cyprinid stocks

In support of the genetic improvement program of some selected carp species, phenotypic differences, i.e. differences in meristic traits of wild and hatchery stocks of *L. rohita* and morpho-meristic characters, including gonadal differences, of intraspecific hybrid groups of *B. gonionotus* were analyzed.

L. rohita

For the stock improvement program, *L. rohita* and *C. catla* wild stocks were obtained from different river systems of Bangladesh. Initial trials were conducted with *L. rohita* to investigate whether any phenotypic (meristic traits) differences existed among the wild stocks (from the Brahmaputra, Halda and Jamuna Rivers) and a hatchery stock. The traits examined and the range of values observed were as follows: number of dorsal fin rays (13-15), number of pectoral fin rays (14-18), number of pelvic fin rays (8-10), number of anal fin rays (6-8), number of lateral line scale (40-43) and number of vertebrae (30-35). Of these, only the number of pectoral fin rays showed significant ($P<0.05$) differences among the four stocks, where the hatchery stock showed a lower mean value (15.1 ± 1.0) than the three wild stocks.

B. gonionotus

A comparative study was conducted on morphometric and meristic characters and gonadal development of three different intraspecific crosses of *B. gonionotus* (Afroz 1996). The three different intraspecific hybrids of the fish were produced through cross breeding involving two wild stocks collected from Thailand (T) and Indonesia (I) in 1994 and the existing stock in Bangladesh introduced in 1977 (B). The variations of phenotypic characters and in the timing and developmental stages of gonads in such hybrids were investigated.

The means of the different morphometric characters, viz. total length, body weight, body depth, body width, head length, snout length and length of intestine varied significantly (P<0.01) among the three experimental groups and were highest in the hybrid TxB, followed by IxB and BxB. Although some variations were observed in the mean values, range, mode and modal frequencies of the meristic counts, viz. number of lateral line scales, vertebrae, dorsal fin rays, pectoral fin rays, pelvic fin rays, anal fin rays and caudal fin rays, etc. were statistically insignificant (P>0.05). The ratio between the total length and body length and the total length and body width of the hybrids were significantly different (P<0.01). However, regression and correlation analysis revealed that body weight and body depth of all three hybrids and body width of the hybrid TxB were highly correlated with the total length (P<0.01) and this relationship was found to be linear (Begum 1996).

From histological sections prepared in January, well-developed spermatogonia were observed earliest in BxB, followed by IxB, but were absent in TxB. Similar results were also obtained in the three stocks with spermatozoa. The stages of oocyte growth and the seasonal changes in the properties of oocyte stages within the ovaries were observed histologically. It was found that in the peak breeding season when the ovaries contained a huge number of oocytes at vitellogenic and maturation stages, a number of germ cells were still present at the early or late perinucleolus stages. The mean diameter of cell and nucleus of the oocytes were observed to be higher in TxB followed by BxB and IxB, but no significant differences (P>0.05) were noted among the three experimental groups of fish (Begum 1996).

The results of the gonadosomatic index measurements (GSI) in male and female fish of the three fish stocks showed that the GSI of female fish increased from January through May. This pattern was also followed in the case of only TxB males but in IxB and BxB there was a distinct decline in GSI in May. In the females, significant differences (p<0.05) were obtained among GSIs of the three stocks of fish during April and May.

Table 3.1.5 shows the reproductive performance of the three parental stocks of *B. gonionotus* and a pooled crossbred stock derived from diallele crosses used for the evaluation of morpho-meristic characters and reproductive performance in this study (Akhter 1998). Among the observed characters, the crossbred stock showed significantly superior (P<0.05) performance than the purebred stocks, particularly in GSI, egg weight and fecundity. The mean egg diameter was significantly lower (P>0.05) in Thailand stock as compared to other stocks. The highest values were observed in Bangladesh and crossbred stocks. Sperm concentration, fertilization rate and hatching rate of eggs were not significantly different among the experimental groups. Significantly higher values for mean larval size were observed in the Bangladeshi and crossbred groups than in the Thai and Indonesian stocks. Various morphometric and meristic characters showed no significant differences among the purebred and crossbred stocks.

3.1.3.2 Cytogenetics of carps

The only reports available on cytogenetic studies of carp species in Bangladesh are those of Islam et

Table 3.1.5. Reproductive performance of purebred *B. gonionotus* stocks and cross-bred stocks

Characters	Stocks			
	Bangladesh	Thailand	Indonesia	Crossbred
GSI	19.44^{a}	9.20^{a}	14.78^{a}	30.64^{b}
Gonad weight (g)	19.55^{a}	18.60^{a}	37.53^{b}	59.32^{c}
Fecundity	11 7778^{b}	35 597^{a}	13 8553^{b}	269 364^{c}
Egg diameter (µm)	113.35^{b}	95.77^{a}	106.87^{b}	111.40^{b}
Fertilization rate (%)	73.76^{a}	91.77^{a}	91.46^{a}	92.89^{a}
Hatching rate (%)	58.73^{a}	52.31^{a}	52.76^{a}	61.11^{a}
Larval size (mm)	4.25^{b}	3.40^{a}	3.58^{a}	4.15^{b}
Sperm concentration/ml	3.9×10^{10}	5.2×10^{10}	8.9×10^{10}	5.7×10^{10}

Different superscripts mean significant differences between and among the purebred and cross-bred stocks at 5 per cent level. Same superscripts mean no significant differences.
Source: Afroz (1996) and Begum (1996).

al. (1994) and Hussain et al. (1997). Both studies used the chromosome-karyotyping technique to determine the ploidy status of genetically manipulated progeny groups of *L. rohita*. The haploid, diploid and triploid metaphases in *L. rohita* were composed of one (n = 25), two (2n = 50) and three (3n = 75) sets of chromosomes respectively. It has also been reported that both *C. catla* and *C. cirrhosus* bear a similar number of chromosomes in their metaphases to *L. rohita* (Mr. S. Islam, personal communication).

3.1.4 Status of genetic improvement and related research in different species

To counter genetic deterioration through inbreeding, negative selection and hybridization (see 3.1.4.5 and 3.1.5), development of improved brood stocks through implementation of effective breeding plans for commercially important carp and other fish species has been identified as an important area of research in Bangladesh. Genetic improvement research has been initiated for two carp species, *B. gonionotus* and *C. catla*. *L. rohita*, as one of the most important carp species, has also been included for genetic stock enhancement research through selective breeding and chromosome manipulation. In view of conserving threatened carp species, attempts have been made to develop artificial breeding techniques for *Tor* spp., *P. sarana*, *L. gonius*, *L. bata*, *C. ariza* and *L. calbasu*.

3.1.4.1 Stock improvement of *B. gonionotus*

B. gonionotus is a popular species among fish farmers of Bangladesh because it grows well on low protein diets, whether feeding on certain aquatic plants or given supplementary feeds, and can tolerate a wide range of environmental conditions. The species is cultured in perennial ponds, seasonal ponds, road-side dug pits, rice fields, etc., in both polyculture and monoculture systems. Monoculture has been practiced in some seasonal ponds only (about 20 per cent of cultured seasonal ponds). In recent times, *B. gonionotus* has become the main species in rice fish culture because of its high tolerance, rapid growth and higher profit. In Bangladesh, breeding is mainly carried out by hypophysation. In some cases, controlled natural spawning is done by water flushing. Brood stock are normally used for two to three breeding seasons and spawning may be repeated two to three times within an interval of 40-60 days in a single season.

Studies to develop a genetically improved strain and all female *B. gonionotus* using the three available founder stocks (Bangladeshi, Indonesian and Thai), through selective breeding and genetic manipulation techniques are in progress (Hussain 1997). The breeding program was initiated in 1994 involving two wild germplasms obtained from Thailand and Indonesia and the existing local stock. To produce a base population (P_1), the three unrelated founder stocks were bred. In 1997, the first generation (F_1) trials were made through a complete 3x3 diallele crossing experiment to produce nine heterogeneous, outbred genetic groups. For each of the reciprocal crosses, five to eight pairs were mated separately and the best three progeny groups were selected to make 18 full-sib progeny families. These were then communally stocked by mixing equal numbers of larvae (125 individuals) from each family. During the spawning season of 1998, 20 per cent of the best females and males were mass selected from the F_1 communal cross-bred groups and mated to produce the F_2 generation. In 1999, mass selection was carried out again, having 15 per cent F_2 most mature breeders, and used for third generation (F_3) trials. In each generation, evaluation of growth performance was carried out through comparative trials between selected (crossbred) vs. non-selected (BxB) groups (Hussain et al. 2002).

Evaluation of growth performances of nine genetic groups derived from diallele crosses

In this experiment, advanced fingerlings from all nine genetic groups were stocked in replicated communal ponds using PIT tags and it was difficult to maintain homogeneity in size at stocking. Therefore, the initial weights were significantly different among the groups. However, correlation analysis between mean initial weight and final weight of the groups in each pond did not show any significant differences. Thus, it appeared that initial weight was not a significant determinant of the final weight.

***Evaluation of growth performances of F_1 cross-bred and control (BxB) groups of* B. gonionotus**

At harvest, the average mean weight gain by cross-bred and control groups were 63.25 g and 58.88 g, respectively. The data of monthly mean weight showed larger size of the cross-bred group throughout the trial, but no significant differences (P>0.05) were observed between the cross-bred group and the control group at the end.

Evaluation of growth performances of F_2 crossbred and control (BxB) groups

The five-month results obtained in this experiment showed higher growth performance attained by the cross-bred group over the control group, although there was not a significant difference between the two groups (P>0.05). At harvest, sex ratios of both cross-bred and control groups did not show any significant differences from 1:1.

Evaluation of growth performances of F_3 crossbred and control (BxB) groups

Table 3.1.6 shows in detail the fortnightly mean growth data in weight of F_3 cross-bred and control (BxB) groups. During the initial three months (sampling two to sampling six) the growth rate of the cross-bred group did not show any significant difference (P>0.05). During the later 3 months (sampling 7 to sampling 12) the cross-bred group attained significantly (P<0.01) larger mean weight than that of the control group. At harvest, the mean weight gain attained by the cross-bred group and control group were 71.80 g and 58.89 g, respectively.

3.1.4.2 Development of monosex female populations of *B. gonionotus*

Female *B. gonionotus* has a better growth rate than males; therefore, mass production of all female population is desirable using the techniques of gynogenesis and sex reversal. It was previously demonstrated that the meiotic gynogens of this species were all female, suggesting female homogamety (Pongthana et al. 1995). The protocol for the next step in such an approach was to produce neomales (phenotypic male having XX genotype) through feeding androgen hormone treated feed to the gynogenetic fish. These neomales could then be crossed with normal females for mass production of all female seed.

Thus for the production of all female *B. gonionotus* along with large-scale neomale production, research was conducted using imported neomales from the National Aquaculture Genetics Research Institute (NAGRI), Thailand, in view of mating them with females belonging to wild germplasm of Thai and Indonesian stocks. Moreover, production of additional batches of neomales through gynogenesis and sex reversal was initiated (Pongthana et al. 1999).

Studies were also carried out on the sexual dimorphism for weight in *B. gonionotus* through sampling of different populations from different culture systems which indicated sexual dimorphism index for weight (SDI_W) at the age of approximately one year ranging from 1.1-1.7 in different populations and in different culture systems. The proximate composition of the carcass of female and male fish were also compared, where insignificant (P > 0.05) differences were found in the case of moisture, protein and ash content; however, fat showed a significant difference between the sexes (P < 0.05) (Azad 1997).

Table 3.1.6. Evaluation of growth performances of F_3 cross-bred and control (BxB) groups of *B. gonionotus*

Group	Mean Weight (g) at sampling											
	1	2	3	4	5	6	7	8	9	10	11	12
BxB	0.17^a	3.42^a	9.08^a	13.76^a	16.99^a	25.55^a	31.56^b	32.94^b	37.32^b	44.01^b	51.90^b	58.89^b
Cross	0.15^a	4.04^a	9.42^a	14.96^a	19.71^a	29.02^a	36.66^a	39.47^a	46.44^a	51.77^a	66.80^a	71.80^a

Means with the same superscript letter are not significantly different.
Mean values of cross-bred and control groups with the same superscripts are not significantly different at 5 per cent level.

Estimation of generation-wise additive genetic gain

Generation-wise additive genetic gain was estimated approximately by weight between the tested groups and it was observed that F_1, F_2 and F_3 crossbred groups attained respectively 7.5 per cent, 9.78 per cent and 21.92 per cent weight gain over the control (Fig. 3.1.2; Hussain et al. 2002). The preliminary conclusion is that a superior strain of *B. gonionotus* could be developed by several generations of selection.

3.1.4.3 Breeding plans for stock improvement of *C. catla* and *L. rohita*

Breeding and genetic stock improvement program of *C. catla* and *L. rohita* has recently been initiated in Bangladesh. Land races of the species were collected from different river systems of the country, viz. the Halda, Jamuna and Brahmaputra. These wild stocks were screened by investigating differences in extrinsic genetic traits by means of morphological and growth assessment. The two wild stocks (Jamuna and Brahmaputra), along with the existing hatchery stock, were mated to produce three cross-bred (viz. Hatchery female X Jamuna male; Jamuna female X Brahmaputra male; Brahmaputra female X Jamuna male) and three purebred lines (viz. Hatchery female X Hatchery male; Jamuna female X Jamuna male; Brahmaputra female X Brahmaputra male). The selective breeding and line crossing program is in progress using all these lines. Observed phenotypic variations in some of the morpho-meristic characters were investigated and the stocks of hatchery and Brahmaputra were found to be distinctive as compared to the three lines of cross-bred stocks.

3.1.4.4 Genetic stock improvement of carps by other methods

During 1986-97, major studies were conducted on short-term preservation of carp sperm, radiation-induced gynogenesis in *L. rohita* and *C. cirrhosus,* induction of triploidy in *L. rohita* and comparative growth with normal diploid, enhancing ovarian development in *C. catla* through the periodic administration of carp pituitary extract, induction of mitotic gynogenesis and production of clones in *L. rohita.* A brief account of the results of some of these trials is given below.

Radiation and heat shock induced gynogenesis in L. rohita and C. cirrhosus

Meiotic gynogenesis was induced by giving heat shock to eggs fertilized with UV irradiated sperm. The sperm were irradiated with a constant dose of UV-rays for 2 minutes from a distance of 13.5 cm. In *L. rohita*, 4 minutes after fertilization, when heat shock was applied at 40 °C for 2 minutes, gynogenesis was induced in 80-90 per cent of the cases. The optimum heat shock regime was found to be similar for *C. cirrhosus* (Dr. M.S. Shah, personal communication). A temperature of 39 °C was more effective and the induction rate of gynogenesis was 80-100 per cent. Survival of gynogens in both species was low (2-40 per cent) compared to that of the normal control (30-60 per cent).

Induction of triploidy in L. rohita by heat shock treatment and comparative growth with normal diploid

Triploidy was induced in *L. rohita*, by applying heat shock to eggs fertilized with normal sperm at 40 °C for 2 minutes, starting 4 minutes after fertilization (Islam et al. 1994). Their ploidy status was determined karyologically, with triploid induction rates of 60 per cent. The survival rate within the first five to seven days after hatching was recorded at 15 per cent in the heat-shocked group and 25 per cent in the control. Growth in induced triploids after 18 weeks was significantly higher than in diploids (for weight, $P<0.01$, and for standard length, $P<0.05$).

Induction of mitotic gynogenesis and production of clones in L. rohita

In view of improving the stock of *L. rohita*, induction of mitotic gynogenesis and production of clonal lines was initiated during 1993-94 (Hussain et al. 1997). Efforts were made to interfere with the normal functioning of the spindle apparatus during mitotic cell division of fertilized eggs using heat shock treatment, thereby leading to the induction of mitotic gynogenesis in the F_1 generation. Afterwards, putative mitotic gynogenetic alevins were reared as brood stock and a sexually mature female was used to obtain ovulated eggs that were fertilized later with UV irradiated milt. The UV irradiation was carried out for 2 minutes and heat shock of 40 °C for 2 minutes was applied 3-5 minutes after fertilization to produce clonal lines.

Short-term preservation of carp sperm

A study was conducted to keep carp sperm of *L. rohita*, *C. catla* and *H. molitrix* alive and increase its viability for a longer period using Cortlands solution, Hank's salt solution or Tris-buffer with the addition of antibiotics (Islam et al. 1995). Penicillin and streptomycin were added at doses of 200-2 700 IU and 10-13 mg/ml, respectively, to assess optimal doses with different physiological saline solutions. Use of penicillin and streptomycin at the rate of 2 700 IU and 13 mg/ml, respectively, with a 1:4 ratio of sperm to physiological saline (Cortlands solution) was found to increase the viability of sperm up to 96 hours with 28 per cent fertilization, as against 48 hours with 10 per cent fertilization when the sperm was preserved without antibiotics. In addition to the use of antibiotics, Cortlands solution is suitable for *C. catla*, while Hank's salt solution was found better in *H. molitrix*. In *H. molitirix*, the sperm was found to survive for 72 hours with 10 per cent fertilization in Hank's salt solution while the addition of antibiotics raised survival up to 120 hours with 24 per cent fertilization. In the case of *L. rohita*, Tris-buffer was found more suitable than Cortland and Hank's salt solutions, with the addition of antibiotics. When Cortlands solution, Hank's salt solution and Tris-buffer were used with antibiotics for sperm preservation of *L. rohita*, the sperm survived 120 hours in all the cases with fertilization rates of 56, 57, and 61 per cent, respectively.

3.1.4.5 Detection of hybridization between *H. molitrix* and *A. nobilis* in hatchery stocks using DNA microsatellite markers

Hybridization between *H. molitrix* and *A. nobilis* has been reported to occur fairly frequently in commercial hatcheries in Bangladesh. To detect hybridization between these two species, three microsatellite DNA loci were isolated from *H. molitrix* and were used in the analysis of samples collected from hatcheries in different regions of Bangladesh. The analysis indicated that 8.3 per cent of 422 hatchery brood stock that were morphologically identified as *H. molitrix* had *A. nobilis* allele(s) at one or more of the three microsatellite loci, while 23.3 per cent of the 236 fish morphologically identified as *A. nobilis* had *H. molitrix* allele(s) at one or more loci (Fig. 3.1.1). The results suggested that while some of these fish might be F_1 hybrids, others had more complex genotypes, suggesting further generations of hybridization or introgression between the species in hatcheries, with potentially damaging consequences for the integrity of these stocks and their performance in aquaculture. Further analysis of data collected by this survey may reveal associations between hybridization and management practices.

3.1.5 Utilization of genetic resources in aquaculture

Recent regional advances in aquaculture, in particular carp polyculture and other related freshwater fish farming of rice fields, seasonal ditches, canals and perennial ponds, have resulted from the introduction of induced breeding or artificial propagation techniques during the late 1950s. In Bangladesh, seed production through artificial breeding of endemic and exotic carp species has become a common practice.

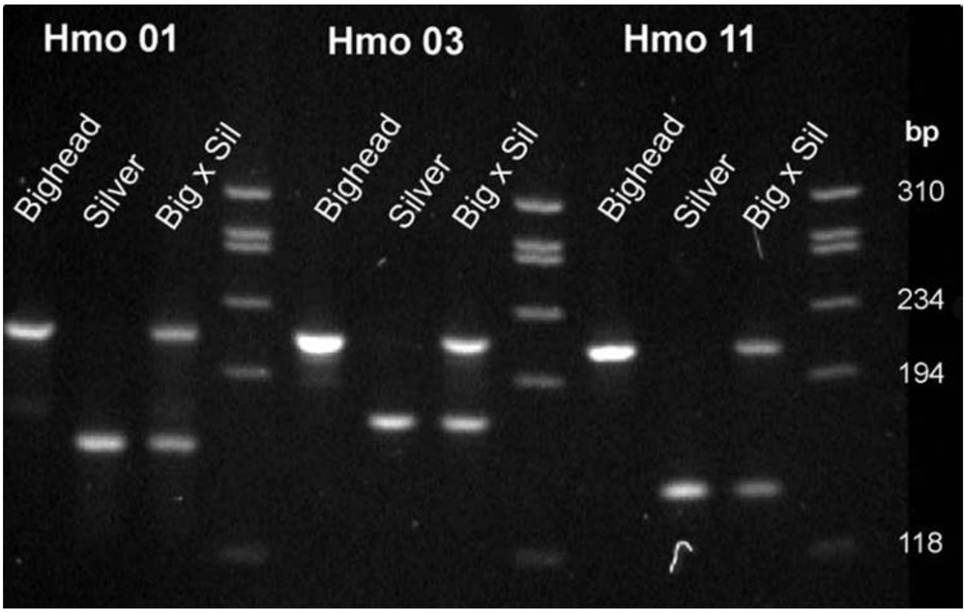

Fig. 3.1.1. Electrophoresis of PCR-amplified microsatellite loci Hmo1, Hmo3 and Hmo11, showing an example of pure *H. molitrix*, pure *A. nobilis* and hybrid (Big x Sil) genotypes for each locus. The right-hand lane of each set contains molecular weight standards, with the sizes in base pairs (bp) given at the extreme right of the figure from Mia et al. 2003

Most hatcheries in Bangladesh rear their own brood stock and usually do not recruit brood stock from natural sources (rivers) although some may obtain brood stock from other aquaculture sources. Many hatcheries, therefore, can be considered as isolated, self-sustaining and genetically closed units (Eknath and Doyle 1990). It is now established that in genetically closed hatchery systems, potential selective pressures exerted on finite and often small culture populations by various farm management practices such as the selection of founder stock, number of breeders maintained, method used for replenishing brood stock, stocking density, feeding regime, etc., can result in "indirect" or negative selection, inbreeding and genetic drift (Doyle 1983).

Data collected from fin fish hatcheries located in Jessore, Bogra, Commilla and Mymensingh in Bangladesh revealed that most of the hatchery operators have no knowledge of simple brood stock management practices and do not follow any principles or guidelines in selecting adequate sized breeders, injecting hypophysation dosage and mating unrelated male and female spawners (Hussain and Mazid 1997). Ignorance of appropriate hatchery procedures leads to negative selection due to the use of fish of undesirable size and leftover brood stock, and mating them generation after generation from closely related or finite populations. Stock deterioration has been observed in hatchery populations due to poor brood stock management and inbreeding. Retarded growth, reduction in reproduction performance, morphological deformities, increased incidence of disease and mortality of hatchery produced seeds of various major carps and *B. gonionotus* have been reported in recent years. Interspecific hybridization in some carp species has recently occurred in Bangladesh (Hussain and Mazid 2001; Mia et al. 2003), either out of scientific interest or shortage of adequate hatchery population (i.e. brood stock). Introgressed hybrids are being produced intentionally or unintentionally by the private hatchery operators and sold to the farmers and nursery operators.

Presumably, a large quantity of such degraded and introgressed hybrid seeds are being used in aquaculture and also stocked in floodplains and open water bodies in Bangladesh. There is widespread concern that mass stocking of such genetically poor quality stocks in the floodplains and related open water bodies might cause serious feral gene introgression problem in wild stocks that would ultimately adversely affect aquaculture and inland open water fish production in the country.

3.1.6 Summary

Bangladesh, a country of deltaic plains dominated by the major river systems like the Ganges, Brahmaputra and Megna, is endowed with unique water resources comprising both inland and marine waters. Along with potential water resources, the country is also rich in the diversity of various fish species.

In recent years the natural fish stocks in Bangladesh have declined due to deterioration of aquatic environments resulting mainly from human interventions.

Relatively little research has been carried out on the cyprinid genetic resources in Bangladesh. However, research is underway on a number of the most important species and it is hoped that improved stocks from selective breeding programs currently being undertaken will be utilized in aquaculture in the near future. Fish genetic manipulation, selection, and biodiversity conservation are of paramount importance in the country and the Government authorities are actively considering establishing a National Center of Fish Genetics Resources and Biodiversity Conservation that will also go a long way in supplementing the global efforts for conserving fish biodiversity.

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3.2 Carp Genetic Resources of China

Zhu Jian1, Wang Jianxin[1], Gong Yongsheng[1] and Chen Jiaxin[1]

3.2.1 Introduction

China is a country vast in territory where the complex climatic and geographical conditions have led to the evolution of a multiplicity of carps. Carps are the major freshwater fish cultured in China and carp culture possesses important economic value in both its culture area and productivity. Carps are not only the favorite target species for culture, but also important species for fishing in natural waters. Their production currently accounts for 83 per cent of the total inland aquaculture production, and covers over 90 per cent of the total landing volume of freshwater fish. Carps grow fast and are suitable for various farming systems. The protein content of carps is high and the production cost is relatively low. Among all carp species cultured in China, common carp (*Cyprinus carpio*) culture has the longest history (more than 2 500 years). *C. carpio* has a lot of morphological variations developed through artificial breeding and natural selection. The Chinese Government has paid much attention to economic development and research on carps. Hence, a variety of techniques including selection, crossing, gynogenesis, polyploidy and introducing new varieties from other regions or countries have been used for genetic improvement of *C. carpio*. Good varieties and strains of *C. carpio*, as well as other carp species, have been achieved. This progress has advanced the culture of carps and increased production.

3.2.2 Cyprinid species of importance to aquaculture

China is very rich in bio-diversity, including rich aquatic germplasm. There are over 3 000 freshwater and marine fish species. Among over 800 freshwater fish, about 140 species are of economic importance, distributed in Changjiang (Yangtze, Yangzi, 44 species), Huanghe (22 species), Zhujiang (Pearl, 30 species) and Heilongjiang (40 species) river systems. It is well known that Chinese carps, such as black carp (*Mylopharyngodon piceus*), grass carp (*Ctenopharyngodon idella*), silver carp (*Hypophthalmichthys molitrix*), bighead carp (*Aristichthys nobilis*), *C. carpio*, crucian carp (*Carassius carassius*) and bream (*Parabramis pekinensis*) are the most important in the Chinese fish industry.

The Cyprinidae is the largest family of fish, with over 2 000 species belonging to more than 200 genera. Among them there are about 410 species, from 110 genera and 10 sub-families, inhabiting Chinese waters (Wu 1964) of which 20-30 carp species are cultured in ponds, lakes, rivers, reservoirs and paddy fields throughout the country. The sub-families, economically important species and cultured carp species in China are listed below.

Sub-family 1: Hypophthalmichthyinae (3 species from 2 genera; *A. nobilis* and *H. molitrix* are endemic to China)

- *Aristichthys nobilis* (Richardson) Bighead carp
- *Hypophthalmichthys molitrix* (Cuvier et Valenciennes) Silver carp
- *H. harmandi* (Sauv.)

Sub-family 2: Cyprininae (26 species from 5 genera in China)

Genus *Carassius*:

- *Carassius carassius* (Linnaeus) Crucian carp
- *C. cuvieri* White crucian carp [introduced from Japan]
- *C. gibelio* (Bloch) Silver crucian carp
- *C. carassius pengzenensis* Pengze crucian carp

Genus *Cyprinus*:

- *Cyprinus carpio*, Common carp
- *C. carpio chilia*
- *C. carpio pellegrini*
- *C. carpio rubrofuscus*
- *C. carpio* var. *jian* Jian carp
- *C. carpio yuankiang* Yuanjiang carp
- *C. carpio* var. *crystallos* Wan'an transparent red carp

[1] Freshwater Fisheries Research Center, Chinese Academy of Fisheries Sciences, Wuxi, China.

- *C. carpio* var. *wan'anensis* Wan'an glass red carp
- *C. carpio wuyuanensis*, CWW Hebao red carp
- *C. carpio singuonensis* Xinguo red carp
- Huanghe carp
- German mirror carp [introduced from Germany]
- Scattered mirror carp [introduced from Russia]
- Songpu mirror carp

New varieties and hybrids:

- Allogynogenetic crucian carp
- Feng carp --- hybrid of *C. carpio singuonensis* x Scattered mirror carp
- Jian carp --- new variety, (*C. carpio wuyuanensis* x *C. carpio yuankiang*, gynogenesis, integration breeding)
- Ying carp --- F_1 scattered mirror carp x CyCa F_2 (CyCa F_2 is F_2 generation of nuclear-cytoplasmic hybrid fish of *C. carpio* nuclei transplanted into *C. carassius* cytoplasm
- Heyuan carp --- *C. carpio* var. *wuyuanensis* x *C. carpio* var. *yuankiang*
- Yue carp --- *C. carpio* var. *wuyuanensis* x *C. carpio*
- Baiyuan carp --- *C. carpio* var. *yuankiang* x *C. pellegrini*
- Tri-crossed carp --- (*C. carpio* var. *wuyuanensis* x *C. carpio* var. *yuankiang*) x Scattered mirror carp
- Backcross carp --- *C. carpio* var. *yuankiang* x (*C. carpio* var. *wuyuanensis* x *C. carpio* var. *yuankiang*)

Sub-family 3: Xenocyprininae (11 species from 4 genera)

- *Plagiognathops microlepis* (Bleeker)
- *Xenocypris davidi* (Bleeker)

Sub-family 4: Leuciscinae (only found in China, with 45 species in 22 genera)

- *Mylopharyngodon piceus* (Richardson) Black carp
- *Ctenopharyngodon idella* (Cuvier et Valenciennes) Grass carp
- *Leuciscus waleckii* (Dybowski)

Sub-family 5: Abramininae (54 species from 17 genera)

- *Megalobrama amblycephala* (Yih) Blunt snout bream, Wuchang fish
- *M. terminalis* (Richardson)
- *Parabramis pekinensis* (Basilewsky) Chinese bream
- *Erythroculter ilishaeformis* (Bleeker)

Sub-family 6: Barbinae (105 species from 27 genera)

- *Cirrhinus molitorella* (Cuvier et Valenciennes) Mud carp
- *Spinibarbus caldwelli* (Nichols)

Sub-family 7: Acheilognathinae (23 species from 6 genera)
(No species of significant importance to aquaculture)

Sub-family 8: Gobioninae (79 species from 19 genera)

- *Hemibarbus maculatus* Bleeker
- *Saurogobio dabryi* Bleeker

Sub-family 9: Schizothoracinae (54 species from 10 genera)

- *Gymnocypris prezewalskii* (Kessler)
- *Schizothorax (Schizopyge) yunnanensis* (Norman)

Sub-family 10: Gobiobotinae (13 species from 1 genera)
(No species of significant importance to aquaculture.)

In addition to these indigenous species and the introduced strains and sub-species of *C. carpio* and *C. carassius*, respectively, which are listed above, rohu (*Labeo rohita*), catla (*Catla catla*) and mrigal (*Cirrhinus cirrhosus*) have been introduced from South Asia.

3.2.3 Documentation and evaluation of genetic resources

Studies on conservation and utilization of germplasm resources started in China in the 1950s. Firstly, resource investigation on the varieties, quantities and distribution of aquatic organisms were conducted in the country. Fish genetic resources and breeding programs were brought into state plans in 1972. Since then, research on genetic resources has received support.

In 1973 and 1981, investigations were conducted to assess changes in fish resources in the

Changjiang River. In 1983, studies on wild stock collections and genetic evaluations of *H. molitrix*, *A. nobilis* and *C. idella* from the Yangtze River, Pearl River and Heilongjiang River were undertaken. From the results of morphological characteristics, biochemical genetics, growth and reproduction traits and population structure, the growth and genetic differences between the three fish species in the three river systems were observed. The fish germplasm in the Changjiang River is thought to be the best among the three river systems (Li et al. 1990).

Genetic characterization of ten major species of freshwater cultured fish in China, of which eight are carps (*M. piceus*, *C. idella*, *H. molitrix*, *A. nobilis*, *C. carpio singuonensis*, *C. gibelio* and *M. amblycephala*, mirror carp) was studied in detail during 1991-95. The parameters studied included morphological characteristics, growth traits, sexual maturation and productivity, oxygen consumption, protein, amino acid, fat, moisture and ash content in muscle, karyotype analysis, DNA content of diploid karyon and isozymes (Li 1998).

In China from the 1970s basic research on fish genetics progressed quickly. Karyotypic analysis of 301 fish species was conducted. Chromosomal localization of HCG gene homology in *C. idella* genome with a heterogenic probe was accomplished in 1989. Electrophoretic analysis and isozymes of major freshwater fish, *M. piceus*, *C. idella*, *H. molitrix*, *A. nobilis*, *M. amblycephala*, *C. carpio* and *C. carassius* involving allele frequency studies on polymorphic loci were undertaken. During 1989-95, biochemical type and its relationship with growth of *C. gibelio* and *H. molitrix* (Changjiang River strain) were studied with an EST genetic marker and the biochemical type of rapid growth was found. The results gave a good base for selection with the biochemical genetic marker.

With the development of international science and technology, the application of molecular biology techniques, such as PCR, mtDNA, RAPD and DNA fingerprinting to study diversity in DNA molecular and gene levels started in the country in the 1990s. Now studies on fish germplasm resources and molecular biology using these techniques are popular in China. DNA sequence and structure of chondriosome gene URFA6L and tRNA gene LYS in *C. carpio*, restriction endonuclease map of mtDNA in *C. carpio*, *C. cuvieri*, *C. gibelio* and *C. carassius* were analyzed in 1991.

3.2.3.1 Molecular genetic markers

Several studies have been carried out on carps using randomly amplified polymorphic DNA (RAPD) markers. Qiu (1996) considered that the application of the RAPD technique in fisheries biotechnology can check and measure the DNA genetic polymorphism of fish genomes, identify the varieties and breeds for aquaculture, and select the brooders for selective breeding and crossing. The RAPD technique is also useful in gene localization and cloning, species classification and molecular evolution studies. Xue et al. (1998) studied *C. idella* from three rivers (Yangtze, Zhujiang and Heilongjiang) and Taihu Lake in China, using 120 random decamer primers. The results showed that each population of *C. idella* has its own unique genome pattern that might be used to identify these different populations of *C. idella*.

Dong et al. (1999) generated 199 RAPD fragments from *C. carpio singuonensis*, German mirror carp and Russian mirror carp, indicating the variation and genetic distance between the three populations. It was presumed that the heterosis between *C. carpio singuonensis* and Russian mirror carp would be the highest.

The similarities and differences of amplified DNA fragments among the multiple species of *C. gibelio*, allogynogenetic *C. gibelio* and red common carp (*C. carpio singuonensis*) were analyzed by Zhou et al. (1998). Forty random decamer primers were used in the RAPD analysis. The results not only provided new molecular evidence for incorporation of heterologous genetic materials in the multiple species, but also demonstrated that heterologous genetic materials in the multiple species were more than that in allogynogenetic *C. gibelio*.

Intra- and inter-population genetic variation of *C. idella*, *C. carpio singuonensis* and *C. carpio* was analyzed by Zhang et al. (1998a) using RAPD markers. It was shown that the genetic distance between *C. idella* and *C. carpio* is larger than that between the intra-specific populations of *C. carpio singuonensis* and *C. carpio* by analyzing the RAPD patterns. Liang et al. (1993a) analyzed the genomic changes of *C. carpio wuyuanensis* (K) and its parents with RAPD.

Microsatellite DNA fingerprinting techniques have been applied to fisheries in recent years

(Zhang et al. 1998b). Zhang et al. (1997) studied the DNA fingerprints of ten types of *C. carpio*. It indicated that the ten types of *C. carpio* are actually six varieties and strains, and that the diversity and differences between them are conspicuous. Jian and Xia (1999) cloned and characterized minisatellite DNA in *A. nobilis*.

3.2.3.2 Immunological and haematological markers

There have been relatively few studies on immunological and haematological markers in carps. Yang et al. (1998) purified immunoglobulin from bile and serum of untreated *C. carpio* and its antigenic, chemical and physical characteristics were studied. The immunoglobulin from bile and serum of *C. carpio* showed identical antigenicity, and similar chemical and physical characteristics. The authors suggested that immunoglobulin in different tissues of the *C. carpio* has the same origin.

Tong et al. (1987) reported the results of a preliminary study on the specificity of red blood cell antigens from varieties (or strains) of *C. carpio*. By means of hemagglutination and serological absorption tests, cross-reactions between special antiserum and RBC of gynogenetic red carp F_2 and gynogenetic red mirror carp F_1 ($RMGF_1$) were investigated. It was found that the reaction values are different between these strains, but the values of random samples in a strain are of the same level. These results show that the genes controlling the red blood cell allotypic antigens of carp are homogenous to a great degree in the same strain. It also appeared that the allotypic antigens of RBC are peculiar to a given carp strain or variety, and it can be considered as one of the markers for fish strain identification.

Lin et al. (1998) carried out a haematological study of two-year-old *H. molitrix* and *A. nobilis*. The blood indices and their annual changes, such as the number of red and white blood cells (RBC and WBC), hemoglobin content (Hb), blood specific gravity (BSG), erythrocyte osmotic fragility (Eof), haematocrit (Ht), erythrocyte sedimentation rate (ESR), differential leukocyte count (DIC) and the normal size (long diameter × short diameter) of the blood cells were investigated. The results indicated that the indices could be used as reference of the normal physiological indices for the two species of fish.

3.2.3.3 Biochemical analysis

The biochemical analysis mainly focused on the nutritive composition in the muscle, protein content, the amounts of amino acid to compare the biochemical composition, meat quality and nutrition value of different fish species. Lu and Bao (1994) analyzed the biochemical composition and nutrition value of *C. carpio* var. *jian*, *C. cuvieri* and F_1 (*C. carpio* var. *jian* × *C. cuvieri*). They showed that the Jian carp contained a higher amount of protein, total amino acids, essential amino acids and taste amino acids. It was concluded that *C. carpio* var. *jian* had a better taste. Hong et al. (1997) analyzed the major nutrition composition in the muscle of Wan'an glassy red carp (*C. carpio* var. *wan'anensis*) that included crude protein, amino acids, moisture, ash and nutritional elements of K, Na, Ca, Mg and microelements Cu, Zn, Fe. Zhang et al. (1988) studied the hydrolysis and free amino acid in the muscles of *C. carpio* and *C. carassius*.

3.2.3.4 Electrophoretic Analysis and Isozymes

Electrophoretic analysis and isozymes technique can be used to analyze the differences between fish species and populations, identify the brooders and their hybrids, check the variety purity and use as a biochemical marker. By using the method of polyacrylamide gel electrophoresis, Ye et al. (1992) investigated sexual differentiation in the electropherogram of serum proteins of *C. carpio*. Wang et al. (1994) analyzed the isozymes of German mirror carp, scattered mirror carp and their hybrid Songpu carp.

Electrophoretic analysis of the isozyme patterns of lactate dehydrogenase (*LDH**), esterase (*EST**) and serum protein of *C. carpio* var. *jian*, *C. carpio wuyuanensis*, and *C. carpio yuankiang* showed that the *C. carpio* var. *jian*'s major isozyme patterns and protein bands are similar to those of *C. carpio wuyuanensis* and *C. carpio yuankiang*, but the expression of the alleles of the former is stronger than that of the latter two (Yuan et al. 1994). The authors also found that the advantageous genetic properties of *C. carpio* var. *jian* were in accord with its distinctive biochemical traits.

Shi et al. (1994) studied the expression patterns of the isozymes *LDH** and *EST** in the hybrids (cis-cross and trans-cross) of (*C. carpio* var. *jian* × *C. cuvieri*) using polyacrylamide gel electrophoretics. Chang et al. (1994) studied the *LDH** and *EST** in Huanghe carp.

3.2.3.5 Cytogenetics - karyotypes of carps in China

Table 3.2.1 shows a summary of information on the karyotypes of Chinese cyprinids that are of importance to aquaculture and fisheries (Yu et al. 1989; Zan and Song 1980; Chen et al. 1996). Some other studies on cytogenetics and karyotypes of carps were found in the literature. Zhang et al. (1995) studied the karyotype of hybrid *C. carassius* and its parents. The parents, *C. cuvieri* (female) and scattered mirror carp (male), both have chromosome numbers of 100. The maternal karyotype is 2n=20m+28sm+38st+14t and the paternal 2n=20m+26sm+30st+24t. There were two types of the karyotypes of the hybrid *C. carassius* (F_1): diploid and triploid. The karyotype of the diploid is 2n=20m+27sm+34st+19t, combining a maternal set and a paternal set of chromosomes. The triploid hybrid had a chromosome number of 150±3.

Table 3.2.1. Karyotypes of Chinese cyprinids of importance to aquaculture and fisheries. 2n: chromosome number of diploid; NF: fundamental arm number

Species	2n	Chromosome types	NF
Aristichthys nobilis	48	6m+36sm+6st	(96)
	48	18m+22sm+8st	88
	48	14m+24sm+10st	
	48	26m+20sm+2st	(96)
Hypophthalmichthys molitrix	48	10m+26sm,st+12t	(84)
	48	14m+24sm+10st	
	48	24m+16sm+8st	(96)
	48	20m+24sm+4st	(96)
	48	18m+22sm+8st	88
Carassius carassius	100	12m+40sm+48st,t	
	100	12m+44sm+44st,t	
	100	22m+30sm+48st,t	
	100	30m+34sm+36st,t	164
	100	24m+30sm+46st,t	154
	100	22m+34sm+22st+22t	156
C. cuvieri	100	12m+36sm+32st+20t	148
	100	20m+28sm+38st+14t	148
C. gibelio	156	42m+74sm+40st,t	272
	162	34m+58sm+42st+28t	254
	162	33m+53sm+76st,t	
C. carassius pengzenensis	166	32m+40sm+18st+76t	238
Cyprinus carpio	100	22m+34sm+22st+22t	156
	100	12m+40sm+48st,t	
	100	28m+22sm+50st,t	
C. carpio chilia	100	22m+30sm+48st,t	
C. carpio rubrofuscus	100	22m+30sm+48st,t	
Plagiognathops microlepis	48	18m+26sm+4st	92
Xenocypris davidi	48	20m+26sm+2st	94
Mylopharyngodon piceus	48	14m+34sm,st	(96)
	48	18m+24sm+6st	90
Ctenopharyngodon idella	48	18m+24sm+6st	90
	48	18m+22sm+8st	
	48	16m+32sm	96
	48	20m+24sm+4st	(96)
Megalobrama amblycephala	48	18m+26sm+4st	92
	48	20m+24sm+4st	
	48	26m+22sm	
	48	16m+24sm+6st	
Parabramis pekinensis	48	14m+26sm+8st	88
	48	26m+22sm	88
Culter erythropterus	48	16m+26sm+6st	90
Erythroculter ilishaeformis	48	16m+26sm+6st	90
Cirrhinus molitorella	50	16m+24sm+10st	90
	50	20m+26sm+2st+2t	96
Spinibarbus caldwelli	100	18m+32sm+26st+24t	150
Hemibarbus maculatus	50	16m+14sm+16st+4t	80
Saurogobio dabryi	50	18m+26sm+6st	94

The karyotypes of the first generation allotetraploid (4N: the hybrid of *C. cuvieri* × *C. carassius* induced allotetraploid), allotriploid (4N male × 2N *C. cuvieri* female), new tetraploid (4N male×2N female, induced allotetraploid) and their parents - *C. cuvieri* and *C. carassius* - were studied by Chen et al. (1998). The results confirmed the relationship and ploidy relation between the first generation allotetraploid, allotriploid, new tetraploid and their three parental species.

Wu et al. (1986) presented karyotypic evidence of induced haploid and diploid gynogenesis and triploidy in carp by means of genome manipulation. Karyotype analysis of the haploid showed that the complement of chromosomes was 6m+20sm+24st,t. It demonstrated that the chromosome components of gynogenetic diploids were derived from two complete genomes. Hence, with normal viability and functional ovaries, the second gynogenetic generation could be employed as a stock for establishing a pure line. The chromosome complement of some triploid individuals having normal functional ovaries consisted of three complete genomes. This evidence suggests that the major reason of the sterility of most induced triploid fish might result from aneuploidy or breakage of chromosomes during temperature shock.

3.2.4 Status of genetic improvement and related research in different species

In recent years the study and practice on genetic evaluation, genetic improvement and utilization of different genotypes of carps are progressing rapidly in China. The methods and techniques commonly used worldwide have been tried and improved. In some fields of fish breeding, China plays a leading role in the world (Lou 1986). Information on carp genetic evaluation, improvement, and utilization has been collected from all resources throughout China, such as research institutions, colleges and libraries.

3.2.4.1 Selective breeding

In China, over 20 fish species have been selected for selective breeding. Some of the major varieties are:

Xinguo red carp (*C. carpio singuonensis*)

More than 1 300 years ago, the variety was cultured in Singuo county. Since 1972, research was undertaken on this native variety by Red Carp Reproduction Farm of Singuo County and Biological Department of Jiangxi University. After six generations of selection, the growth rate increased by 10 per cent per generation. Red individuals in the population accounted for 86.6 per cent. Total production of this variety is about 5 000 tonnes per year. This is the parent of several hybrids: Feng carp, Lotus carp, and allogynogenetic crucian carp.

Hebao red carp (*C. carpio wuyuanensis*, CCW)

The variety has been cultured in Wuyuan County for over 300 years. Since 1961, its biology has been studied by the Institute of Hydrobiology, Academia Sinica and CCW, Research Institute of Wuyuan County. After six generations of selection, its characters tend to be stable, with red individuals accounting for 89.54 per cent. Annual production is about 5 000 tonnes. This variety is the parent of other varieties and hybrids, like Heyuan carp, Yue carp, Tri-back hybrid, and Jian carp.

Pengze crucian carp (*C. carassius pengzenensis*)

The variety has been cultured in lakes of Pengze County. Since 1980, systematic selection has been undertaken by the Fisheries Research Institute of Jiangxi Province. After six generations of selection, its growth rate improved by 56 per cent.

Selected strain of German mirror carp

Mass and family selective breeding of the introduced variety has been undertaken for more than ten years. After selection, the growth rate of F_4 improved by 10.8 per cent. Disease resistance, survival rate and cold resistance of its progeny have been improved.

3.2.4.2 Cross-breeding

Most interspecific hybrids have failed to show characteristics of value to culture. In China, over 100 hybrid combinations have been tried in the family Cyprinidae. The *C. carpio* intraspecific crossbreds with good characters are: Feng carp, Heyuan carp, Yue carp, Baiyuan carp, Tri-crossed carp and Back-cross carp. All of these cross-breds possess economically good characters (such as higher growth rate, lower feed conversion rate, higher fishing rate, etc.). Hence, they have become the main varieties of cultured *C. carpio* throughout China (Zhang and Sun 1994a).

The cross-breds and varieties developed from cross-breeding of *C. carpio* in China are:

Jian carp (*C. carpio* var. *jian*)

Parents of Jian carp are *C. carpio wuyuanensis* and *C. carpio yuankiang*. It is a result of integration of breeding including family selection, inter-line cross combining with gynogenesis. The genetic stability is over 95 per cent. The strain has been extended to 29 provinces and autonomous regions. It is cultured in more than 660 000 ha with an annual production of over 1 million t. The growth rate is 49.75 per cent, 46.8 per cent, and 28.9 per cent higher than that of *C. carpio wuyuanensis* and *C. carpio yuankiang*, and Heyuan carp respectively (Zhang and Sun 1994b; Sun et al. 1994b).

Anti-cold strain of *C. carpio wuyuanensis*

The parents are a native carp strain in Heilongjiang Province and *C. carpio wuyuanensis*. Through F_1 sib mating an individual with anti-cold factor, red color and scale had been obtained in F_2. The individual was used as a parent to keep the anti-cold factor combined with good characteristics from *C. carpio wuyuanensis*. Under the same culture conditions in frigid zone, its growth rate is 10 per cent higher than other varieties.

Feng carp

Feng carp is a hybrid of female *C. carpio singuonensis* and male scattered mirror carp. Its appearance is similar to both female and male parents. In the fingerling stage, the growth rate is higher than its parents: 50 per cent to 62 per cent higher than its mother and 140 per cent more than its father. Because it has obvious hybrid vigor, farmers welcomed it. Comparison between body length and weight of Feng carp and its parents revealed the hybrid vigor both in the fingerling and adult stages.

Ying carp

Ying carp is a hybrid of female scattered mirror carp and male F_2 of a CyCa nuclear-cytoplasmic hybrid, hence it possesses a tri-crossing vigor, its growth rate is 47 per cent and 60.1 per cent higher than its parents at the age of one year old and two years old, respectively.

Heyuan carp

Heyuan carp is a hybrid of female *C. carpio wuyuanensis* and male *C. carpio yuankiang*. It possesses a fast growth rate, strong adaptability and disease-resistance, and high harvesting rate. The hybrid vigor is obvious (Dept. of Fish Breeding, CFRI, 1981)

Yue carp

Yue carp is a hybrid of female *C. carpio wuyuanensis* and male local *C. carpio* from Xiangjiang. It grows 25-50 per cent and 50-100 per cent faster than its female and male parents, respectively. The hybrid vigor is obvious.

Lotus carp

Lotus carp is a hybrid of female scattered mirror carp and male *C. carpio singuonensis*. It grows 40 per cent and 60 per cent faster than its female and male parents, respectively. It adapts to lower temperatures and has obvious hybrid vigor.

Tri-crossed carp

Tri-crossed carp is a hybrid of female Heyuan carp (see above) and male scattered mirror carp. It possesses a tri-crossing vigor and has a fast growth rate, long body and good meat.

Besides these varieties and hybrids mentioned above, German mirror carp and scattered mirror carp are cultured in different zones throughout the country. The scattered mirror carp is the parent of some hybrids.

Further information on cross-breeding of *C. carpio* can be found in Liu and Shen (1993), Liu et al. (1997), Sun et al. (1994a,c) and Wu et al. (1979).

3.2.4.3. Gynogenesis and androgenesis

Research on gynogenesis and androgenesis in fish began in the world as early as the 1950s, and in China projects started from the 1970s.

The progeny of gynogenesis have some particular features and characteristics (Yang et al. 1981; Wu et al. 1986): 1) The progeny are almost the same as the mother in appearance and physiological function; 2) it is easier to set up pure lines; 3) their sex can be controlled; and 4) this can be combined with the technique of sex reversal for producing all-female fish.

Since Romashov achieved diploid artificial gynogenesis in the 1960s (Ye and Wu 1987), the technique has been successfully tried in more than 20 species. In the last decades, Chinese scientists improved the technique and utilized it in goldfish (*C. auratus*), red variety of *C. auratus*, *C. carpio* var. *singuonensis*, *C. idella*, *H. molitrix*, and others. A good example is allogynogenetic *C. carassius*, which is the result of female progeny from gynogenesis of Fangzheng crucian carp (a variety of *C. gibelio*, a natural triploid) crossed with male *C. carpio singuonensis*, and which is welcomed by farmers because of its higher growth rate. The Institute of Hydrobiology, Academic Sinica is the institute involved in developing allogynogenetic *C. carassius*.

Occasionally, androgenesis is induced by distant hybridization. Stanley (1976) reported that androgenetic individuals were produced from the crossing of mirror carp with *C. idella*. Ye et al. (1990) reported that the haploid embryos of *C. carpio* and *C. carassius* were produced from androgenesis induced by γ-Ray (^{60}Co). However, the haploid embryos could not survive when the embryos developed to later stages.

Sun and Zhang (1994) reported on the survival rate, growth, morphological structure and cytological characteristics of gynogenetic Hebao red carp (*C. carpio wuyuanensis*), with comparisons between gynogenetic Hebao red carp and its parents. Wu et al. (1991) described the induction of gynogenesis of the *C. carpio singuonensis* by means of γ-rays (dosage of 1.3×10^{-2} J) or ultraviolet (2350A, 30W×2, 30 min), using the sperm of related species (*C. carassius* or *M. amblycephala*). Then a second generation of gynogenesis was produced, and some of the individuals of the second gynogenetic generation were artificially sex-reversed into "physiological males". By mating physiological males with their sisters a pure line - Red Carp 8305 - was established. Isozymes and transferring loci showed no heterozygosity in this line, although several of these loci were found to be polymorphic in *C. carpio* populations. The serological assay showed that genes controlling the red blood cell allotypic antigens were homogenous to a great extent in Red Carp 8305. The survival rate of scale transplantation between different individuals of Red Carp 8305 was over 87 per cent. It appears that genes controlling MHC are homozygous. Gui et al. (1997) studied the genetic diversity of gynogenetic *C. gibelio* and showed its breeding implication.

3.2.4.4 Polyploid Breeding

Polyploid fish are found on rare occasions in nature. Generally, it is believed that polyploid fish have a higher growth rate and stronger adaptability than that of diploid fish. For this reason, the techniques for inducing polyploidy have become a focus of breeders. The principles and methods of inducing polyploid fish are similar to those of gynogenesis. They mainly include two aspects: 1) to break the spindle fiber, and 2) to restrain the extrusion of the polar body with physical and/or chemical treatments, or the proceeding of fertilized egg's cleavage (Li 1982).

Gui et al. (1990) optimized hydrostatic pressure shock conditions for producing triploid transparent colored *C. carassius* by inducing second polar body retention. Gui et al. (1991) studied the induction of tetraploidy in the same strain of *C. carassius*, using hydrostatic pressure and/or cold shocks. Gui et al. (1992) investigated meiotic chromosome behavior, by surface-spreading, air-drying and thin sectioning tests for light and electron microscope examination, in artificial triploid transparent colored *C. carassius* produced by hydrostatic pressure shock. Yu et al. (1991) reported a preliminary study on tetraploid hybrids of natural gynogenetic Fangzhen *C. gibelio*. Fertilized eggs of *C. gibelio* × *C. carpio* were cold shocked and tetraploid hybrids obtained. Sun et al. (1994d) carried out some experiments on the induction of homologous and heterologous triploidy, tetraploidy and gynogenesis in *C. carpio* var. *jian* and *C. carpio* *wuyuanensis*. Another method for producing triploids can be conducted by crossing of tetraploids and diploids. For example, Xiangyun carp (Engineering carp), the new variety developed by Hunan Normal University, is a hybrid of tetraploid (F_3-F_8 of *C. carassius* red var.× native *C. carpio* from Xiangjiang River) and normal diploid (*C. carpio singuonensis*). A new variety of *C. carassius* - Xiangyun crucian carp (Engineering crucian carp) - was also developed by crossing of tetraploid and diploid fish in Hunan Normal University.

Yang et al. (1994) found that triploid *C. cuvieri*, produced by mating heterologous tetraploid (4n) males with diploid (2n) females, were sterile and grew faster than the diploids, suggesting potential commercial value.

Recently, an artificial multiple triploid carp (AM triploid carp) was induced by means of thermal shock to red *C. carpio* x red *C. carassius* hybrid eggs fertilized with mirror carp sperms. The artificial multiple triploid carp contains two sets of *C. carpio* genome and one set of red *C. carassius* genome. The AM triploid carp was developed in the Institute of Hydrobiology, Academic Sinica. Wu et al. (1993) studied an artificial multiple triploid carp, which had a karyotype consisting of three complete genomes from *C. carpio singuonensis*, mirror carp and red *C. carassius*. Ye and Wu (1998) verified the karyotype of the articifial multiple triploid with DNA content measurement using fluorescence microphotometry.

Polyploids can also be induced by distant crossing. Wu et al. (1981, 1988) reported the hybrid of red *C. carpio* and *C. idella*, a tetraploid through automultiplication of chromosomes, possessed features and habits like the maternal parent and could develop into either sex. The authors also reported on the female and male hybrid F_1 backcrossed with both sexes of *C. idella*, and the backcrossing produced triploid offspring, but their features and habits were different. Chen et al. (1987) reported on artificially induced heterogeneous tetraploids in the hybrid of white *C. carassius* (female) and *C. carpio* (male).

In China, it was reported that artificially induced triploid and tetraploid fish have been produced in over 10 species: *C. idella*, *H. molitrix*, *M. amblycephala*, rainbow trout, *C. carpio* var. *wuyuanensis*, the hybrid between *C. carpio* var. *singuonensis* x *C. idella*, the hybrid between

C. carpio var. *wuyuanensis* x *C. cuvieri*, the hybrid between *C. idella* x black bream (*Megalobrama terminalis*), the hybrid between *C. cuvieri* x red goldfish (*C. auratus*, red strain) transparent colored crucian carp, Xiangyun common carp, Hebao red carp, Xiangyun crucian carp and catfish (*Silurus asotus* L). Among them the triploids of Xiangyun carp, Xiangyun crucian carp and *S. asotus* possess commercial value.

3.2.4.5 Sex control

In some species the sexual differentiation of fish influences its growth rate and other economic characteristics. For example, female *C. carpio* and *C. idella* grow faster than males. In order to control the sexes of fish, several methods are available, such as interspecific crossing, inducing of sex-reversal, production of super-male fish, all-female fish, etc. In China, these techniques have been successfully used in *C. carpio* and tilapia.

A combination of sex reversal and gynogenesis to produce all-female *C. carpio* was used in the Institute of Hydrobiology, Academic Sinica. The all-female *C. carpio* is a hybrid of the regular XX female *C. carpio singuonensis* and sex reversed XX male scattered mirror carp. Before sex-reversal, the mirror carp was induced gynogenetic diploid.

3.2.4.6 Mutation breeding

It is a very common technique to obtain mutants using mutagens, both chemical and physical. Since the 1970s, γ-rays and quick-neutrons have been tried to irradiate the gonad, embryo, fry and fingerling of *C. carpio*, *C. idella*, tilapia, etc., in order to obtain mutants. Unfortunately, no effective results have been obtained. Tan et al. (1993) reported results about fish breeding using laser irradiation. A new breed, two tailed brocaded carp - China color carp - was cultivated successfully. It was bred for three generations, but its form did not change.

3.2.4.7 Integration breeding

Experience with fish breeding practices revealed that it is useful to combine two or more breeding techniques together to get new varieties or strains. *C. carpio* var. *jian* is a good example of integration breeding because of its stable genetic characteristics with commercial value and attractive appearance. Zhang et al. (1994) described the development of the *C. carpio* var. *jian* over six generations. Its integration breeding techniques combined family selection, and inter-family crossing with gynogenesis. To date, the new variety has been extended to 31 provinces, municipalities and autonomous regions.

3.2.4.8 Transplant of karyon and cell culture

Since the 1970s, Tong Dizhou and Yan Saoyi (Xue 1996) have developed the basic research dealing with transplanting karyon and cell culture, and utilized the technique and method in commercial fish culture. In this way, they and their colleagues successfully achieved nuclear-cytoplasmic hybrid fish. Five nuclear-cytoplasmic hybrid fish have been produced in China; they are:

- CyCa, *C. carpio* nuclei transplanted into *C. carassius* cytoplasm.
- CaCy, *C. carassius* nuclei transplanted into *C. carpio* cytoplasm.
- CtMe, *C. idella* nuclei transplanted into *M. amblycephala* cytoplasm.
- MeCt, *M. amblycephala* transplanted into *C. idella* cytoplasm.
- Tilapia nuclei transplanted into *C. carpio* cytoplasm.

In order to discover if any changes were induced at the genome level after *C. carpio* nuclei have been transplanted into *C. carassius* cytoplasm, their DNA reassociation kinetics were studied. It was found that the nuclear-cytoplasmic hybrid fish (CyCa) F_3 had very similar properties to *C. carpio* (Wang et al. 1990), suggesting that the nuclear genome was not affected by the heterologous cytoplasm in the nuclear transplantation process. This study proceeded to the next step including research on economics (Pan 1990).

As early as 1914, cell culture of fish started. In order to meet the demand for chromosome studies, the techniques of cell culture have been advanced since the 1970s. The cells from different tissues and organs, like blood cell, fin cell, scale cell, cardiac muscle cell, nephric cell and others, have been cultured in laboratories. These techniques possess practical value, and are able to be used in nuclear transplants.

3.2.4.9 Gene engineering

This technique was first developed in the 1980s in China. Zhu et al. (1985) described the transfer of a mammalian growth hormone gene construct into the fertilized eggs of goldfish and went on to produce transgenic cyprinids with enhanced growth rates (reviewed by Zhu 1992). Cheng (1989) characterized an antifreeze protein (AFP) from the fish *Pseudopleuronectes yokohamae,* and Jiang (1990) cloned the AFP cDNA and expressed it in *E. coli*. The protein could depress the serum freezing point and may have applications in gene transfer. Wei et al. (1992) reviewed the status and prospects of fish gene-engineering research in the early 1990s.

Progress has been made in fish transgenesis complementing traditional breeding programs for improvement of culture traits. Xia (1997) reported that human growth hormone (hGH) had been injected into fertilized eggs of *C. carpio*, *C. gibelio* and *M. amblycephala* and *C. carpio* growth hormone (cGH) transferred into *C. auratus*. Studies on integrating ox and sheep growth hormone with fish have also been carried out in China. Liang et al. (1993b) studied factors affecting egg survival rates and integration frequency of the foreign gene following microinjection of DNA into *C. carpio* eggs. Shen et al. (1993) microinjected a mMT-bGH construct into fertilized eggs of *C. carpio*, *O. mykiss* and *C. auratus* and found evidence for the presence of the exogenous DNA using dot blots and Southern transfer. Wu et al. (1994) described integration, expression, growth enhancement and inheritance of a mMT-hGH construct in *M. amblycephala* and *C. carpio*. Zhao et al. (1996) described integration, expression and growth enhancement of a CMV-cGH construct in *C. carassius*. The foreign DNA can be inserted into the host genome and transferred to the filial generation. The transgenic fish demonstrated growth improvement. In the near future, the practical applications in carp species should be pursued.

3.2.5 Utilization of genetic resources in aquaculture

Management and utilization of wild aquatic resources and introduction of good breeds of fish from other countries are important to make the best use of germplasm resources. Domesticated wild carps and genetically improved carps are widely used in aquaculture in China.

Some of the varieties of *C. carpio* and *C. carassius* developed through crossbreeding and selection have become important on a national scale, e.g. *C. carpio* var. *jian* now has been extended all over China. Four billion Jian carp seeds were produced and supplied to farmers during 1988-2000. The culture area exceeded more than 660 000 ha. The production is over 1 million t annually, which accounts for 50 per cent of the total annual production of *C. carpio* culture in China. In some areas, Jian carp production accounts for 70 per cent of the total fish production. Extension of improved Jian carp also has great potential for poverty alleviation and low-income farmers can benefit more from it. Besides Jian carp, other varieties, such as Xinguo red carp, Hebao red carp, Feng carp, Heyuan carp, Ying carp, allogynogenetic crucian carp, and Pengze crucian carp, although produced in smaller numbers, have significant yields. Triploid Xiangyun carp and Xiangyun crucian carp also have good potential for aquaculture. Section 3.2.4 gives further information on them. Ongoing selection programs such as those on *M. amblycephala* are now producing faster growing stocks that are being produced by hatcheries.

Unlike some of the other Asian countries covered in this book, China has very few introduced cyprinid species of any significance. Stocks and subspecies of the *C. carpio* and *C. carassius* have been imported from Europe and other parts of Asia, respectively. Among them, scattered mirror carp is a good parent fish for carp breeding and German mirror carp, white crucian carp and *L. rohita* appear to be truly introduced cyprinid species that feature in Chinese aquaculture.

3.2.6 Summary

China has many species of cyprinids, some found only in this country. While a few of the species of cyprinids used in aquaculture and restocking in China have been subjected to little, if any, domestication, others have been the subject of much research and the development of improved stocks. China has been a leader in several aspects of aquaculture research, e.g. on gene transfer and nucleus transfer. Selected strains of the *C. carpio* and *C. carassius* contribute a large proportion of the total aquaculture production of these species. Indigenous Chinese cyprinid species (e.g. *C. idella*, *H. molitrix* and *A. nobilis*) have also been introduced into more than 20 countries in Asia, Europe, North and South America and Oceania, where they contribute to aquaculture and fishery production.

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3.3 Carp Genetic Resources of India

P.V.G.K. Reddy[1]

3.3.1 Introduction

Indian fish genetic resources, consisting of over 11 per cent of the world's 20 000 known species of fish, are among of the richest in the world (Das 1994). They include a variety of marine, estuarine/brackish water and freshwater species, most of which are economically important. The development of Indian fisheries started with the enactment of the Indian Fisheries Act of 1897 (Gopakumar 2000). Although in British (pre-independent) India, the importance or need for establishing fisheries research institutes had been felt, the first research department established was the Central Marine Fisheries Research Station in 1947, which has later assumed the status of an institute. Another important development in the area of fisheries was the establishment of the Central Inland Fisheries Research Station, in the same year, at Barrackpore (West Bengal), which also later assumed the status of an institute. This is the parental institute from which many other institutes of today have branched off, such as the Central Institute of Freshwater Aquaculture (CIFA), Central Institute of Brackishwater Aquaculture (CIBA), National Research Centre on Coldwater Fisheries (NRCCF), etc. All these institutes have been established, considering the rich germplasm resources in the country and catering to the needs of all the frontiers of fisheries.

In the prioritization of natural resources, particularly on the food front in India, agriculture occupies the top position, followed by animal husbandry and then fisheries. Over 70 per cent of the people in India live in rural areas, where the main occupation is agriculture. Until the 1960s India had to depend on other developed nations to meet her requirements, particularly of food grains. However, with the establishment of the Agricultural Research Institutes, much progress has been made and new technologies and culture systems have been developed during the post-independence period, especially in the last three to four decades, in almost all the sectors mentioned above. Thus, the inadequacy of agricultural products has been replaced by self-sufficiency.

However, to attain this goal, certain modifications had to be made, mainly by diverting the natural river courses by constructing multipurpose dams, of which developing irrigation networks was one of the major aspects. Although irrigation has contributed to the development of agriculture, enabling farmers to produce two or three crops in a year in the place of a single crop, it has also disturbed or even destroyed the natural habitat, particularly of breeding grounds of the major commercially important fish species, including Indian major carps (i.e. catla *Catla catla*, the rohu *Labeo rohita*, the mrigal *Cirrhinus cirrhosus* and the kalbasu, *L. calbasu*) and some anadromous species like the Ilsa, *Hilsa ilisa*, which migrate from sea to rivers mainly for breeding purposes. These changes have considerably restricted the breeding activities of many species besides the above-mentioned species. It has been reported that there is considerable depletion in fish populations of most of the commercially important fish species, with the exception of major carps (Das and Barat 1990).

Among other human activities that have been causing destruction to the fishery are overfishing (more for commercial purposes than for living) and pollution of the river systems, mainly due to discharge of industrial effluents into the rivers/seas, etc. (Jhingran 1984; Das and Barat 1990). Owing to all these activities, most of the populations of economically important species as well as others have become depleted, endangered or rare. However, regarding Indian major carps, it has been reported that these species may not be under serious threat yet, but they are prone to loss of genetic diversity (Ponniah 1997).

Another great problem caused by human interference is the growing trend of replenishing the rivers and other larger water bodies like lakes

[1] Central Institute of Freshwater Aquaculture, P.O., Kausalyaganga, Bhubaneswar, India.

and reservoirs with hatchery produced fish seed with the intention of making the loss good (Mishra and Raman 1993; Ponniah 1997). As most of the hatcheries produce seed without following any genetic principles or resource management, it may prove disastrous and lead to genetic introgression (Eknath and Doyle 1985). However, when the greater interests of the nation are put ahead of the interests of fish, nothing much can be done with regard to alterations of river systems and increased water abstraction (Das and Barat 1990).

3.3.2 Cyprinid species of importance to aquaculture

3.3.2.1 Indigenous species

Freshwater fish germplasm resources are mainly found in the Ganga and Brahmaputra river networks in the north and the east, and the west coast river network in the south, stretching nearly 18 000 km. Of these, the Ganga River system forms the largest freshwater fish germplasm resource in the country. It is also the natural habitat for many commercially important species, including the Indian major carps. Additionally, some freshwater lakes also contribute to these resources. About 544 species inhabit warm waters of the plains. Besides Indian major carps *C. catla*, *L. rohita*, *C. cirrhosus* and *Labeo calbasu*, freshwater fish include some of the most important species such as minor (peninsular) carps (e.g. *L. bata*, *L. fimbriatus*, *L. gonius*) and *Cirrhinus ariza;* non-cyprinids including large catfish (i.e. *Sperata aor*, *Sperata seenghala*, *Silonia silondia*, *Wallago attu*, *Pangasius pangasius*, *Bagarius bagarius*, *Rita rita*, etc.); and other catfish (such as *Clarias batrachus*, *Heteropneustes fossilis*, *Ompok bimaculatus* and *O. pabda*).

Cold waters in India from a fisheries point of view refer to water bodies with temperatures within the tolerance limits of trouts belonging to the *Salmonidae* family, ranging from 0–20 °C with an optimum range of 10–12 °C (Jhingran 1991). The coldwater fisheries in India constitute about 21 indigenous and seven exotic species of commercial as well as sport interest. Some of the common indigenous cold water cyprinid fish species are *Bangana dero*, *Barbodes hexagonolepis*, *Barilius bendelisis*, *B. gatensis*, *Raiamas bola*, *Schizothorax kumaonensis*, *S. richardsonii*, *Tor putitora*, *T. tor*, etc. This sector is relatively underdeveloped, probably due to the specific environment.

3.3.2.2 Introduced species

A number of exotic fish species have been introduced into India, including Chinese carps, from time to time, for various purposes such as game, culture (food), controlling larvae of harmful insects, etc. The first introduction of exotic species was in the year 1870, with *Carassius carassius* from England for experimental culture (Jhingran 1991). Later as many as 16 other species, including Chinese grass carp (*Ctenopharyngodon idella*), silver carp (*Hypophthalmichthys molitrix*), bighead carp (*Aristichthys nobilis*), common carp (*Cyprinus carpio*), both Bangkok and German strains, etc., have been introduced. Some of them, particularly the Chinese carps and common carp, are thriving well. In coldwater areas, indigenous cyprinids may have to compete with introduced salmonids, such as *Salmo trutta* and *Oncorhynchus mykiss*.

3.3.3 Documentation and evaluation of genetic resources

3.3.3.1 Cytogenetics

Cytogenetic studies on fish species in India were initiated in the 1960s. Most of these studies pertain to karyomorphological aspects and some are on comparative karyological studies of Indian major carps and their hybrids (Khuda-Bukhsh and Manna 1976; Manna and Khuda-Bukhsh 1977a,b; Manna and Prasad 1971, 1977; Khuda-Bukhsh 1979a,b; Khuda-Bukhsh and Barat 1987; Zhang and Reddy 1991; Jana 1993). Karyology was also studied in relation to hybrid fertility and viability (Reddy 1991; Zhang and Reddy 1991), (Reddy et al. 1990a). Modified methodologies have been developed for obtaining well spread and superior metaphase chromosomes in fish for karyomorphology and banding studies (Reddy and John 1986; Banerji 1987; Reddy and Tantia 1992; and Nagpure and Barat 1997).

Chromosomal banding work in fish as a whole is very scanty. Several causes appear to be responsible for this, among which the relatively smaller size and greater number of chromosomes in fish seem to be the main hurdles. Later advances in the area of biochemical and molecular genetics, assumed to be more accurate, have also come along to limit or abandon these studies and divert the interest of the workers. Chromosome banding studies may help in identifying species and the homologous pairs of chromosomes in a karyotype. Nucleolar organizer regions (NORs) may help in tracing back the parentage of a particular hybrid occurring in nature. It is well known that due to the highly compatible nature of the Indian carps to hybridize, the natural occurrence of their hybrids is common. So depending on the NOR bearing chromosomes in parents and the hybrids, the parentage of the latter may be traced.

Karyotypes of Indian major carps

Many researchers have worked out the karyotypes of Indian major carps. These studies have shown the diploid (2n) number in all the four species to be 50 chromosomes, with their types falling broadly under three categories, namely metacentric, sub-metacentric and sub-telo/ telocentric types. Zhang and Reddy (1991) made a comparative study on the karyotypes of Indian major carps (see Table 3.3.1).

Karyotypes of Indian carp hybrids

Table 3.3.1. Comparative karyotypes of Indian major carps (Zhang and Reddy 1991)

Species	2n	Types of chromosomes		
		Metacentric	Sub-metacentric	Sub-telo-/ telocentric
C. catla	50	12	16	22
L. rohita	50	10	18	22
C. cirrhosus	50	12	18	20

Even though over 20 interspecific and intergeneric hybrids were produced among Indian major carps, karyotype studies were made only in the case of Kalbasu-Rohu (Krishnaja and Rege 1975), Kalbasu-Catla and Rohu-Catla (Khuda-Bukhsh and Manna 1976) and Catla-Rohu hybrids (Jana 1993). These studies revealed that though the diploid number of chromosomes is the same in the parent species, in hybrids the number in each type (metacentric, sub-metacentric, telocentric, etc.) is somewhat different. These differences are probably due to differences in the methodology used for preparing the chromosomes.

Karyotypes of hybrids between Indian major carps and common carp

John and Reddy (1987) and Reddy et al. (1990a) have studied the karyotypes of these intergeneric hybrids. These studies were mainly in relation to hybrid fertility. The hybrids between female *C. carpio* and males of Indian major carps (*C. catla*, *L. rohita* and *C. cirrhosus*) possess 74-76 chromosomes in all cases, which is the total of the chromosomes of the parental gametes. However, this genomic status can also be regarded as aneuploid or triploid (as the hybrid is intermediate in chromosome number between the diploid major carp and the ancestrally tetraploid common carp), which usually leads to sterility.

Karyotypes of suspected spontaneous gynogenetic progeny from the cross between *C. carpio* female and *L. calbasu* male (Khuda-Bukhsh et al. 1988) have shown that the morphology of the chromosomes in the F_1 offspring (putative gynogenetics) was slightly different from that of the mother, *C. carpio*, although their external anatomical features were similar. Further, the haemoglobin and transferin bands of F_1 individuals suggest that the F_1 individuals originated from the gynogenetic mode of reproduction, in which the genetic input of the sperm is eliminated and diploidization of the developing egg nucleus takes place by the union of the nucleus from the polar body.

Chromosomal banding studies

Information on chromosomal banding studies on fish species is very scanty, except for some recent studies, particularly on Indian major carps (Barat et. al. (1990); Rishi and Mandhan 1990; John et al. 1992, 1993; Khuda-Bukhsh and Chakraborty 1994; Nagpure 1997). These studies pertain to G & C bands and nucleolar organizer regions (NORs). Rishi and Mandhan (1990) reported the presence of centromeric C-bands in all chromosomes of *L. rohita*, with no intercalary C-bands. The second report on C-banding is on *C. cirrhosus* and *L. rohita* (Khuda-Bukhsh and Chakraborty 1994). These workers reported that in the majority of chromosomes, C-bands were localized in and around the centromere, while in some others it was lacking or showed telomeric (terminal) or interstitial C-bands localization in both species. The observations of Nagpure (1997) on C-band pattern of *L. rohita* have shown multi-site distribution of the band heterochromatin that is contrary to the observations of Rishi and Mandhan (1990). Interestingly, Nagpure (1997) further reports that C-bands are localized only on eight pairs of chromosomes, of which one was sub-metacentric while the others were telocentric. Also in the majority of telocentrics centromeric C-bands were present, with prominent intercalary bands on three pairs, which have also exhibited the centromeric C-bands. John et al. (1992, 1993) made NOR studies on *C. catla, L. rohita, L. calbasu* and *L. bata*. In Catla, the NORs were minute in size and were terminal to the short arms of one sub-metacentic and sub-telocentric chromosome pairs. The NORs in *L. rohita* and *L. calbasu* were similar in size and located on the eleventh pair of chromosomes, while in *L. bata* they were bigger in size and located on the ninth pair of chromosomes. Attempts on G-bands (Khuda-Bukhsh and Tiwari 1994) did not result in any interpretable success.

3.3.3.2 Biochemical and molecular genetic studies

Information on these areas, particularly molecular genetics, is meager.

Biochemical genetic studies

These studies are limited to some preliminary investigations and are mainly based on some isozymes such as Lactate dehydrogenase (*LDH**) and Malate dehydrozenase (*MDH**) patterns. These biochemical studies have been used to discover similarities among Indian carps. The *LDH** patterns in the developing embryos and in eye-lens and red cell hemolysates (Padhi and Khuda-Bukhsh 1989a,b), Ldh C gene expression in liver tissues (Rao et al. 1989) and the electrophoretic pattern in muscle protein (Padhi and Khuda-Bukhsh 1989b) have shown that Indian major carps, *C. catla, L. rohita* and *C. cirrhosus* were similar in many of their biochemical characteristics.

Biochemical genetic studies were used to characterize plasma, haemoglobin and transferin patterns of Indian major carp hybrids produced by crossing *C. catla* with *L. rohita* and *C. cirrhosus* with *L. rohita*. Isozyme studies were used to distinguish between diploid and polyploids (tetraploid) in the case of Indian major carp, *L. rohita*. Intensification of isozyme bands has been observed in tetraploids (Sarangi and Mandal 1996).

Recently, characterization of rohu stocks from different rivers, the founder population for the selection study (see 3.3.4.2 and 3.3.4.3), has been carried out using isozymes. These preliminary studies have indicated genetic variability similar to wild stocks and no genetic contamination was detected.

Molecular genetic studies

Protocols have been developed for genetic stock identification. A study of *L. rohita* stocks from the Ganga River and hatchery (farm) through restriction endonuclease analysis of mitochondrial DNA revealed polymorphism at Hind III restriction site (Padhi and Mandal 1995a, b). Studies have been made on DNA fingerprinting in *L. rohita* and *C. catla* by using BKM2(8) and M13 multilocus probes (Majumdar et al. 1997; Padhi et al. 1998). In these species, M13 gave fewer bands, in spite of higher levels of polymorphism (Majumdar et al. 1997).

Other studies in this area are the use of four tetranucleotide microsatillite probes for differentiating Catla brood from some major hatcheries in Karnataka State, such as the Tunga Bhadra (TBD), the Bhadra Reservoir Project (BRP) and the Kabini Reservoir Project (KRP) and from the wild and hatchery stocks from outside the State (Basavaraju et al. 1997). These studies have shown that despite common sources, the hatchery populations in Karnataka were different from each other, probably due to genetic drift or inadvertent selection.

Mbo l satellite characterization in *C. cirrhosus* has shown that *Mbo*1 fragments are species-specific and not observed in *C. catla* and *L. rohita*. It was also reported that in the hybrids where *C. cirrhosus* is one of the parents (*C. cirrhosus* X *L. rohita* and *C. cirrhosus* X *C. catla*), the mrigal-specific *Mbo*1*l* fragment was inherited by the hybrid.

3.3.3.3 Genetic conservation and preservation

Under the prevailing situation described earlier, obviously it is essential to protect and preserve the valuable fish germplasm of the country. A survey has been initiated by the National Bureau of Fish Genetic Resources (NBFGR), stationed at Lucknow, Uttar Pradesh State, to identify and categorize the endangered and threatened species, particularly those which are commercially important and conserve them either in *in situ* or *ex situ* conditions.

Cryopreservation of fish gametes and gene banking

Cryopreservation of fish gametes is of relatively recent initiation. Protocols have been developed for the successful preservation of milt from fish in

India (Gupta and Rath 1993; Gupta et al. 1995; Ponniah 1997). Cryopreservation of milt of Indian major carps has been carried out at NBFGR and CIFA in India. Besides these carp species, milt of some other fish species including common carp have been successfully cryopreserved at NBFGR. These studies could establish the optimum requirements with regard to extender, activation media, dilution rate, activation period, sperm and egg ratio differences between species, etc. Cryopreserved milt of *L. rohita* was observed to be effective even after eight to nine years of preservation (Ponniah 1997). It has been also reported that safe levels of cryoprotectants have been determined for the embryos of common carp and rohu (Ponniah 1997). These studies are being pursued. It has been reported that the cryopreservation methodology developed by NBFGR and CIFA have been tested under actual field conditions for cross-breeding as well as pure breeding.

It is obvious that development of cryopreservation techniques is essential in view of the fast deteriorating environment, to conserve and preserve the most precious germplasm of the fish species, particularly of the rare, endangered ones and those on the brink of extinction. Cryopreservation is also an effective tool to overcome the problems involved in the transportation of live seed. It makes the maintenance of brood fish, particularly the males (at present), more economical. For example, if female brood fish are maintained in an *in situ* condition, cryopreserved milt can be used for seed production purposes, thus avoiding *in situ* maintenance of male brood stock. Different genotypes can be maintained at one place under a gene banking system. When success on embryo cryopreservation is achieved, several other advantages/benefits can be derived for aquaculture. However, whether it is *in situ* or *ex situ*, both methods, besides being effective, have their own disadvantages/problems. The *in situ* method, although it provides the genome the chance to acclimatize to changes in the environment, requires large infrastructure including manpower, to maintain the stocks. Similarly, although the *ex situ* method is economical, in the long run genomes preserved for several years may find the then prevailing environment hostile when they are revived. Moreover, in the case of animals, unless both male and female gametes or embryos could be cryopreserved, the full advantage cannot be derived (Mc Andrew et al. 1993).

3.3.4 Status of genetic improvement and related research in different species

During the mid-1950s genetic improvement efforts of fish in India were initiated with Indian major carps by simple hybridization, immediately after the success of induced breeding of these carps through the administration of pituitary hormones. As many as six interspecific and 13 intergeneric hybrids have been produced and evaluated for their cultural traits. In the early 1980s, genome manipulation (chromosomal engineering) work was taken up and standard protocols have been developed after the successful induction of artificial gynogenesis and polyploidy (triploidy/tetraploidy) in Indian major carps. Later, during the early 1990s a long-awaited selective breeding program was taken up with *L. rohita* in collaboration with the Institute of Aquaculture Research (AKVAFORSK), Norway and is ongoing.

3.3.4.1 Hybridization

Genetic improvement in the Indian aquaculture scenario, as mentioned above, was initiated with the major carps. A number of hybrids have been produced among the species of Indian major carps and between these carps and Chinese carps, *C. idella*, *H. molitrix*, *A. nobilis* and also *C. carpio.*

Interspecific (intrageneric) hybrids between Indian major carps

As many as six interspecific hybrids were produced between the species of the genus *Labeo* (Table 3.3.2) and their traits were evaluated by a number of researchers (Chaudhuri 1959; Basavaraju et al. 1989 and 1994). Of these, only the hybrids between *L. rohita* X *L. calbasu* and *L. rohita* X *L. fimbriatus* were found to possess some useful traits from an aquaculture point of view. These hybrid crosses have shown high compatibility with fertilization ranging around 80-98 per cent. These hybrids mostly exhibit intermediate traits of their parent species.

Table 3.3.2. Inter-specific crosses among Indian carps

Parent species			Hybrid
Female		Male	
L. rohita	X	*L. calbasu*	rohu-kalbasu
L. calbasu	X	*L. rohita*	kalbasu-rohu
L. rohita	X	*L. gonius*	rohu-gonius
L. rohita	X	*L. bata*	rohu-bata
L. gonius	X	*L. calbasu*	gonius-kalbasu
L. rohita	X	*L. fimbriatus*	rohu-fimbriatus

i) *L. rohita* X *L. calbasu* (rohu-kalbasu) and the reciprocal hybrid

As already mentioned the intermediate traits in these hybrid offspring appeared with regard to the size of the head, body profile and color of the hybrid. The head of the hybrid is smaller than *L. rohita* and slightly bigger than *L. calbasu*. With regard to other traits, the body of the hybrid is broader than *L. calbasu* but slightly narrower than of *L. rohita*. *L. calbasu* possesses a muddy body color, whereas the hybrid has a brighter body color than *L. calbasu*, although not as bright as in *L. rohita*. The growth performance of these hybrids was reported to be faster than the slower-growing parent *L. calbasu*, but not as fast as *L. rohita*.

ii) *L. fimbriatus* X *L. rohita* (fimbriatus-rohu) hybrid

The hybrid has been evaluated in detail with regard to growth, food conversion ratio (FCR) and protein efficiency ratio. With regard to growth, the hybrid has shown faster growth (247.8 g) in a 182-day rearing experiment, when compared to fimbriatus (167.7 g), but not faster than *L. rohita* (324.5 g). Thus the growth in the hybrid is 34.7 per cent higher than in *L. fimbriatus*. The survival in the hybrid has been reported to be greater than in both the parent species, being 90.0 per cent, while it was 86.7 per cent in *L. rohita* and 76.6 per cent in *L. fimbriatus*. In terms of the percentage of weight gained and also the specific growth rate and feed utilization, the hybrid has been reported to perform better than the parent species.

Other interspecies hybrids have not been found to possess any positive traits of interest to aquaculture.

Intergeneric hybrids

Much work has been done on intergeneric hybrids of Indian major carps. As many as 13 intergeneric hybrids have been produced from three genera, *viz. Catla*, *Labeo* and *Cirrhinus* (Table 3.3.3) and evaluated for their traits. Some of the important hybrids are discussed below.

i) *L. rohita* X *C. catla* (rohu-catla) hybrid

This intergeneric hybrid is a cross between female *L. rohita* and male *C. catla*. A good percentage of fertilization has been reported (60-70 per cent). The morphological features of these hybrids, regarding the size of the head, width and color of the body and even the growth were reported to be intermediate. However, this hybrid was reported to possess higher meat quantity over both the parents (Bhowmick et al. 1981). The rohu–catla hybrid possesses a broader feeding spectrum than the parents and thus is able to consume a variety of aquatic microphytes such as *Ceratium* species, particularly available in some of the reservoirs in India. Because of this trait the hybrid has been recommended to be a good substitute to both the parents for stocking in reservoirs.

Table 3.3.3. Inter-generic crosses among Indian carp species

Parent species			Hybrid
Female		Male	
C. catla	X	*L. rohita*	catla-rohu
L. rohita	X	*C. catla*	rohu-catla
C. catla	X	*C. cirrhosus*	catla-mrigal
C. cirrhosus	X	*C. catla*	mrigal-catla
L. rohita	X	*C. cirrhosus*	rohu-mrigal
C. cirrhosus	X	*L. rohita*	mrigal-rohu
C. catla	X	*L. calbasu*	catla-kalbasu
L. calbasu	X	*C. catla*	kalbasu-catla
L. calbasu	X	*C. cirrhosus*	kalbasu-mrigal
L. rohita	X	*C. reba*	rohu-reba
C. catla	X	*L. fimbriatus*	catla-fimbriatus
L. fimbriatus	X	*C. catla*	fimbriatus-catla
L. calbasu	X	*C. reba*	calbasu-reba

ii) *C. catla* X *L. rohita* (catla–rohu) hybrid

This is the reciprocal cross of the above-discussed hybrid. Detailed investigations have been carried out on this hybrid (Alikunhi et al. 1971; Konda Reddy 1977; Konda Reddy and Varghese 1980 and 1983). The compatibility of this reciprocal cross was relatively higher, with fertilization rates ranging around 65 to 80 per cent. The embryonic developmental events of the hybrid as reported by Konda Reddy and Varghese (1983) are quite normal and comparable to that of the parental species. The hybrids did not exhibit any abnormalities. Regarding the morphological features, this cross has shown intermediate traits like its reciprocal hybrid, rohu-catla. Konda Reddy and Varghese (1980) have reported that the mouthparts and the gill rakers of both hybrids are very similar, suggesting the same food habits. The growth of catla-rohu hybrid has been reported to be faster than even catla under monoculture and when cultured with catla (Alikunhi et al. 1971). Similarly, Varghese and Sukumaran (1971) also observed that the hybrid grew better than both parents. This is, however, contrary to the observations of other researchers who reported that the catla-rohu hybrid grows faster than rohu but not as fast as catla (Chaudhuri 1971; Konda Reddy and Varghese 1980; Jana 1993).

iii) *C. catla* X *C. cirrhosus* (catla-mrigal) and their reciprocal hybrids

These hybrid crosses exhibited higher compatibility with fertilization rates up to 97 per cent. As in the previous intergeneric hybrid crosses, these reciprocal hybrids also possessed intermediate phenotypic characteristics. The catla-mrigal hybrid had an elongated body, tending more towards the mrigal (female parent), with a terminal and slightly ventrally placed mouth. In some other traits like the upper lip with a slight thickness and the size of the head, the hybrid resembles catla. It also slightly resembles catla in possessing a little dorsal hump. The reciprocal hybrid of *C. cirrhosus* female and *C. catla* male had an upturned mouth with the upper lip slightly overhanging, unlike its counterpart. The dorsal hump of *C. catla* is clearer in this mrigal-catla hybrid. The filtering mechanism in catla with over 240 gill rakers is well developed, compared to mrigal's 60. The hybrid with over 90 gill rakers was not exactly intermediate, but nearer to the mrigal (Ibrahim 1977). The growth pattern of the mrigal-catla hybrid was reported to be better than its parents when studied under purely experimental conditions. However, the growth of the catla-mrigal hybrid was not as good as its parents. The mrigal-catla hybrid also possessed a wider feeding spectrum over its parents, covering more of the column plankton (catla) and bottom feed (as the mrigal). The superior growth performance of the hybrid over the parent species was attributed to this characteristic.

iv) *L. rohita* X *C. cirrhosus* and the reciprocal hybrid

With fertilization rates over 97 per cent, these hybrid crosses also exhibited high compatibility. The hybrids mostly possessed intermediate traits. However, some of the distinguishing features in the two reciprocal hybrids occur in the *L. rohita-C. cirrhosus* hybrid where there is a visible gap between the anal and caudal fins that is wider than in the *C. cirrhosus-L. rohita* hybrid. Some researchers reported that both hybrid types grew slower than the parent species. However, as reported by others, the growth of the rohu-mrigal hybrid was better than in its parents (Ibrahim 1977).

v) *L. fimbriatus* X *C. catla* and the reciprocal hybrid

The fertilization rate in this cross was moderate. These hybrids were reported to be morphologically so close that it is difficult to distinguish them. The fimbriatus-catla hybrid possessed intermediate traits to its parent species in respect of body profile, coloration of fins and structure of lips. The body of the hybrid was deeper than fimbriatus with a smaller head than catla, the paternal parent. Growth of both hybrid types was reported to be much faster than fimbriatus, the slow growing parent. However, it was slower than in catla (Basavaraju et al. 1994).

vi) *C. catla* X *L. calbasu* hybrid

In this hybrid too, good compatibility was reported. The general appearance, including body color, of the hybrid resembled kalbasu, indicating paternal dominance. Analysis of important morphometric parameters of the hybrid and parent species has also indicated paternal dominance in the traits of the hybrid. However, in possessing a larger body profile and smaller head, the hybrid distinguishes itself from the parents. The food habits of the hybrid indicate that it has a broader feeding spectrum than its parents, thus indicating greater adaptability to different food items, available in different ecological niches of the pond. Regarding the growth rate, the hybrid under various culture/management practices has shown faster growth than kalbasu and compared favorably with catla.

Fertility status of Indian carp hybrids

All these interspecific and intergeneric hybrids have proved to be fertile and attain maturity between two to three years of age. In some cases even F_3 generations were produced (Chaudhuri 1973).

Intergeneric hybrids between Indian major carps and common carp

These hybrids were produced by crossing females of common carp (*C. carpio* var. *communis*) Bangkok strain with males of Indian major carps, viz. *C. catla*, *L. rohita* and *L. calbasu*, and the reciprocal crosses (Alikunhi and Chaudhuri 1959; Khan et al. 1989 and 1990; Reddy et al. 1990b).

Compatibility between these crosses to hybridize is not very high. Although initially a high percentage of fertilization was obtained, the rate of hatching and subsequent viability of the hybrids were relatively poor. Differences were also observed with the reciprocal crosses. Viability was relatively better when the female was a common carp.

Morphologically, although the hybrids appeared intermediate to their parents, statistical analysis of various parameters revealed them to be closer to common carp. Probably due to their sterile nature, the characteristic belly of common carp was lacking in the hybrids. The hybrid exhibited poorly developed or rudimentary gonads. The meat quantity in the hybrid was more than in the parent species. Details of these hybrid crosses are given in Table 3.3.4.

Table 3.3.4. Crosses between Indian carps and *C. carpio*

Parent species			Hybrid
Female		Male	
C. carpio	X	*L. rohita*	Common carp-rohu
C. carpio	X	*C. catla*	Common carp-catla
C. carpio	X	*C. cirrhosus*	Common carp-mrigal
L. rohita	X	*C. carpio*	Rohu-common carp
C. cirrhosus	X	*C. carpio*	Mrigal-common carp

Intergeneric hybrids between Indian and Chinese major carps

The crosses between Indian major carps (*L. rohita, C. catla and C. cirrhosus*) and Chinese carps (*C. idella, H. molitrix* and *A. nobilis*) did not produce any viable hybrids beyond the fingerling stage.

Back-crossed and triple–crossed hybrids

As already mentioned, almost all of the interspecific and intergeneric hybrids of Indian major carps are fertile. A number of back-crossed and some triple–crossed hybrids have been produced (Table 3.3.5). Back-crosses were made with interspecific as well as intergeneric hybrids. Among the interspecific hybrids, the matured kalbasu-rohu female hybrid was back-crossed with male *L. calbasu*. This cross has shown a high rate of fertilization (98 per cent). These kalbasu-rohu-kalbasu back-crossed hybrids resembled the *L. calbasu*. Others are back-crossed intergeneric hybrids, such as the female of kalbasu-mrigal back-crossed with male *L. calbasu* (kalbasu-mrigal-kalbasu) and with male *C. cirrhosus* (kalbasu-mrigal-mrigal), and the female of rohu-catla hybrid back-crossed with male *L. rohita* (rohu-catla-rohu).

Table 3.3.5. List of back-crossed and triple-crossed hybrids

Parent species			Hybrid
Female		Male	
kalbasu-mrigal	X	*L. calbasu*	*kalbasu-mrigal-kalbasu*
kalbasu-mrigal	X	*C. cirrhosus*	*kalbasu-mrigal-mrigal*
kalbasu-rohu	X	*L. calbasu*	*kalbasu-rohu-kalbasu*
rohu-catla	X	*L. rohita*	*rohu-catla-rohu*
kalbasu-mrigal	X	*C. catla*	*kalbasu-mrigal-catla*
kalbasu-rohu	X	*C. carpio*	*kalbasu-rohu-common carp*

The triple-crossed hybrids are those between a kalbasu-mrigal hybrid female crossed with a *C. catla* (kalbasu-mrigal-catla) male and a kalbasu-rohu female crossed with a *C. carpio* (kalbasu-rohu-common carp) male. In all the crosses the fertilization was very high, ranging around 70-98 per cent. Wherever *L. calbasu* was involved the hybrids were dominated by *L. calbasu* traits. No detailed information is available regarding the triple-crossed hybrids.

3.3.4.2 Selective breeding of *L. rohita*

Selective breeding is a classical approach for the genetic improvement of stocks. The method has shown its potential in the case of agriculture and veterinary animals. In aquaculture too, selection has demonstrated its potential. Atlantic salmon and rainbow trout in Norway, the channel catfish in USA and the Nile tilapia in the Philippines are examples. Recent selection work on the *L. rohita* in India is also heading toward the same goal.

Initiation of selection work on *L. rohita* has been carried out since the early 1990s, with base populations collected from five different rivers in the north, viz. the Ganga, the Gomati, the Yamuna, the Sutlej and the Brahmaputra. To these the CIFA farm stock of *L. rohita* was also added. The family selection method is being followed. The full-sib families (groups) are produced following a nested-mating design (Reddy et al. 1998a). Females of *L. rohita* mature when they are two years old. Two generations of selection have been completed and evaluated. The third generation was produced during the year 2000. An average selection response of over 20 per cent was indicated at the second-generation level of selection (Das Mahapatra et al. 2000).

As the project is the first of its kind with regard to Indian major carps, a good deal of experimentation was carried out to develop standard protocols for marking the base populations, deciding mating designs, raising taggable-size fingerlings, identifying suitable tags and other marking techniques like fin-clipping, etc. (Gupta 1994; Saha et al. 1998; and Das Mahapatra et al. 2001).

The base populations were marked with M-procian blue dye at the base of the pectoral fins and by clipping the pelvic fins or a combination of both. Among Indian major carps, *L. rohita* is a very active fish and as such no external tagging device worked for marking the fish. The PIT (passive integrated transponder) tag proved to be the only suitable tag for marking. The tag is implanted in the abdominal cavity of the fish, with the help of a tag implanter. The individuals of full-sib groups are tagged when they are at a minimum size of 8-10 g. The fish have to be anesthetized by keeping them in a 0.3 per cent MS222 solution for about two to three minutes. However, the dose may vary according to the size of the fish (Das Mahapatra et al. 2001).

The spawn of full-sib groups were reared to taggable fingerling size in earthen nursery ponds of 100 m^2, by stocking them @ 50 spawn/m^2. In order to reduce or minimize the environmental/pond effect, the spawn should be raised to taggable fingerling size in as short a time as possible. Once individuals of different families are tagged, they can be stocked and reared together in common/communal ponds to evaluate their breeding values (BV). Those with higher breeding values, say above 50 or maybe 100, are taken as selected ones. Those with breeding values around the mean values are taken as controls. Future generations are produced from these selected and control individuals. The differences in the breeding values of the selected and control progeny will indicate the response to selection.

Selective breeding, although a lengthy process to realize the anticipated genetic gain in a genetic improvement program, allows for the improvement of many traits through family selection, particularly economically or commercially important traits. The speed of the selection process depends on the biology of the species in question. In species like common carp or tilapia that breed more than twice a year, the process may be faster, whereas in major carps of India or China, which take a minimum of two to four years to attain maturity, it is a lengthy process. However, aquaculture selection appears to be the only practically effective process to enhance production. Modern genetic improvement techniques like genome manipulation (chromosomal engineering) or gene transfer (genetic engineering) are still in an experimental stage as far as aquaculture is concerned. However, once these techniques are standardized and applicable, aquaculture may become more cost-effective and sustainable.

3.3.4.3 Cross-breeding/diallele crosses among *L. rohita* stocks

The first cross-breeding/diallele-crossing between different stocks of *L. rohita*, belonging to the Ganga, Yamuna, Sutlej and Brahmaputra, and the CIFA farm stock were carried out under the Indo-Norwegian collaboration project, at the Central Institute of Freshwater Aquaculture (CIFA), Bhubaneswar, during 1995 (Table 3.3.6). The results of these crosses showed low average heterosis effects for both the harvest body weight and survival. The results suggest that systematic crossing of *L. rohita* stocks may not yield any progeny with positive heterosis (Bjarne et al. 2002).

Table 3.3.6. Cross-breeding of different stocks of *L. rohita*

Diallele Cross I	Diallele Cross II
(9 groups)	(9 groups)
Ganga X Ganga (Control I)	Brahmaputra X Brahmaputra (Control I)
Ganga X Local (Farm rohu)	Brahmaputra X Local
Ganga X Yamuna	Brahmaputra X Sutlej
Local X Local (Control 2)	Local X Local (Control 2)
Local X Ganga	Local X Brahmaputra
Local X Yamuna	Local X Sutlej
Yamuna X Yamuna (Control 3)	Sutlej X Sutlej (Control 3)
Yamuna X Local	Sutlej X Local
Yamuna X Ganga	Sutlej X Brahmaputra

3.3.4.4 Genetic improvement of *C. catla*

The University of Agricultural Sciences (UAS), in collaboration with the University of Wales, Swansea and the University of Stirling has taken up this program. The main objectives are to determine the extent of the problem of inbreeding in domesticated (farm) stocks of *C. catla* in Karnataka State and to design and initiate a breeding program for the improvement of cultured *C. catla* stock. The contention that the domesticated (hatchery) stocks are inbred due to poor brood stock management and improper breeding practices is being verified by comparing the levels of genetic variation between the hatchery and wild stocks of *C. catla*. The wild stocks in this study belong to the Ganga, Brahmaputra and Mahanadi. Allozyme, mitochondrial DNA (RAPD) and tetranucleotide techniques are being applied for these investigations.

Some preliminary survey work has been conducted on three main *C. catla* seed producing hatcheries of the state, viz. Tunga Bhadra Dam (TBD), Bhadra Reservoir Project (BRP) and Kabini Reservoir Project (KRP). These studies have indicated that faulty methods of brood stock management, particularly by rearing a few individuals from fingerlings raised from subsequent progeny of two or three families could be the main causes that are likely to lead to inbreeding and negative selection. The rate of inbreeding in smaller hatcheries has been reported to be unacceptably high and attributed to be the probable cause of reduction in the culture performance of the hatchery-produced seed (Basavaraju et al. 1997).

Performance studies of intra and inter-strain crosses between individuals of the same hatchery and between two different hatcheries mentioned above did not show any clear differences in growth performance between the TBD and BRP hatchery stocks, suggesting that neither strain would necessarily be recommended over the other. However, when tested by using four tetranucleotide loci, the stocks of different hatcheries were found to be different from each other (Basavaraju et al. 1997). Inter-hatchery crosses between the strains of TBD and BRP and the reciprocal cross-bred fish have shown apparent enhancement of culture performance over the pure intra-hatchery crosses with a significant increase in the yield. These studies indicate that the hatchery practices are quite likely heading for inbreeding and negative selection of domesticated stocks of Indian major carps. It was suggested that inter-hatchery crosses in the short term and a breeding program based on wild stocks in the longer term should improve yields (Basavaraju et al. 1997).

3.3.4.5 Genome manipulation (chromosome engineering)

Pioneering work has been done on genome manipulation of Indian major carps by developing standard protocols to induce meiotic and mitotic gynogenesis and also polyploidy (triploidy/tetraploidy) for the first time (John et al. 1984, 1988; Reddy et al. 1990b). Induction of triploidy was successfully carried out in grass carp (*C. idella*) and common carp (*C. carpio* var. *communis*) (Reddy et al. 1993, 1998b). The details of inducing gynogenesis and polyploidy are shown in Tables 3.3.7 and 3.3.8, respectively.

Table 3.3.7. Details of induced diploid gynogenesis of Indian carps at CIFA. C.S.- Cold shock, H.S. - Heat shock. Shock treatments were administered to 4-minute-old inseminated eggs in the case of induced diploid meiotic gynogenesis and before the formation of the first cleavage, (22-25 minutes after activation, depending on the ambient temperature: Reddy et al. 1993) in the case of mitotic gynogenesis.

Species	Gynogenesis		Thermal-shock treatment		
	Meiotic	Mitotic	Nature	Intensity	Duration
C. catla	✓	✓	C.S.	12 ^{0}C	10 min
C. catla	✓	✓	H.S.	39 ^{0}C	01 min
L. rohita	✓	✓	C.S.	12 ^{0}C	10 min
L. rohita	✓	✓	H.S.	39 ^{0}C	01 min
C. cirrhosus	✓	✓	C.S.	12 ^{0}C	10 min
C. cirrhosus	✓	✓	H.S.	39 ^{0}C	01 min
L. calbasu	✓	✓	H.S.	40 ^{0}C	01 min
L. calbasu	✓	✓	C.S.	12 ^{0}C	10 min

Induction of meiotic gynogenesis in Indian major carps

Induction of meiotic gynogenesis was successfully done in *C. catla* and *L. rohita* (John et al. 1984) and in *C. cirrhosus* (John et al. 1988). The procedures include the usual denaturing or inactivation of genetic material or DNA content of the penetrating spermatozoa by exposing them to ultraviolet or gamma rays. To restore diploidy in the activated eggs, thermal (cold/heat) shocks were administered. For Indian carps the effective cold shock regime was 12 °C and heat shocks at 39 °C for ten and one minutes, respectively. Cold shocks below 10 °C and heat shocks above 42 °C

Table 3.3.8. Details of induced triploidy/tetraploidy of Indian carps at CIFA. H.S. – Heat shock. Shock treatments administered to 4-7-minute and 20-22-minute-old inseminated eggs for inducing triploidy and tetraploidy, respectively (Reddy et al. (1990b)

Species	Ploidy		Thermal -shock treatment			Success (%)
	Triploidy	Tetraploidy	Nature	Intensity	Duration	
C. catla	X	✓	H.S.	40^{0}C	10 min	30-55
L. rohita	✓	X	H.S.	42^{0}C	01 min	12
L. rohita	X	✓	H.S.	39^{0}C	10 min	70
C. cirrhosus	X	✓	H.S.	39^{0}C-40^{0}C	01 min	10-40

proved to be lethal (John et al. 1984, 1988; Reddy et al. 1993). Genetic inactivation of sperm DNA in Indian major carps was effectively carried out by exposing the sperm to a 15W ultraviolet germicidal tube at a distance of 20 cm and irradiated for 17-20 minutes (John et al. 1984). Before exposing them to UV rays, the milt was diluted in cold Hank's solution (one-part milt and four-parts Hank's solution). The appropriate time for shock treatments in Indian carps is four minutes after activation to induce meiotic gynogenesis. The percentage yield of meiotic gynogens ranged from 12.6-18.1 per cent (John et al. 1984). Again in *C. catla*, heat shocks were more effective than cold shocks, while the reverse was true in the case of *L. rohita* (John et al. 1984).

Induction of mitotic gynogenesis

Reddy et al. (1993) first reported the induction of mitotic gynogenesis in *L. rohita*. They used irradiated milt of *C. catla*, *C. cirrhosus* and *C. carpio*. Reddy et al. (unpublished) also used irradiated milt of *C. idella* to induce mitotic gynogenesis in *L. rohita*, *C. catla*, *C. cirrhosus* and *L. calbasu*. The use of milt from heterologous species, preferably having no or low compatibility to hybridize, may be more desirable for the easy elimination of paternal genomic input and, in the case where milt is improperly irradiated, the offspring would turn out to be haploids as they cannot form into viable hybrids due to incompatibility, and they perish. The same shock treatments administered for inducing meiotic gynogenesis are effective to induce mitotic gynogenesis in Indian carps, but the time of administration after activation differs. Here, the mechanism of diploidization is not by the retention of the polar body, but by blocking the first cleavage in the activated egg (endomitosis technique). The time varies depending on the ambient temperature and perhaps the biology of the species concerned. In the case of Indian major carps the first cleavage was observed to commence 35-40 minutes after activation at ambient temperatures ranging from 29-30 °C.

The yield of mitotic gynogens was low when compared to meiotic gynogens in these carps. Reddy et al. (1993) reported average survival rates ranging from 1.13 per cent with heat shocks to 4.3 per cent with cold shocks in *L. rohita*. The experiences of other workers also show that late shock treatments (mitotic gynogenesis) usually result in lower yields than early shocks (meiotic gynogenesis).

The difference between meiotic and mitotic gynogenesis in a qualitative sense is that the latter produces complete homozygous individuals, as blocking of first mitotic division results in the retention of two identical replicated mitotic products (Mair et al. 1987), unlike the former that may take four to five generations (Nagy et al. 1984).

Induction of polyploidy in Indian major carps

Some preliminary attempts were made to induce triploidy in the Indian major carp *L. rohita* and tetraploidy in *C. catla* and *L. rohita* for the first time by administering thermal shocks to zygotes (Reddy et al. 1990b). In the case of Rohu, zygotes exposed to heat shocks prior to the first cleavage at 39±0.5 °C for two minutes, yielded 70 per cent tetraploids and with the cold shock regime of 10-15 °C yielded only 30-50 per cent tetraploids. In the case of *C. catla*, heat shocks to zygotes at 40 °C for two minutes, prior to the first cleavage yielded 30-65 per cent tetraploids. However, cold shock was not effective.

Induction of triploidy in *L. rohita* was not very successful when heat shocks were administered seven minutes after fertilization at 42±0.5 °C for one to two minutes, resulting in only 12 per cent triploids. This may be an indication that in *L. rohita*, when shock treatments are administered four minutes after fertilization (at the ambient temperatures ranging from 29-30 °C, which usually prevails at the time of the breeding season at the experimental site), the polar body might already be getting separated, as confirmed earlier while inducing meiotic gynogenesis in this species. Moreover, the temperature regime of 42±0.5 °C may be also towards the higher side.

Presently, chromosomal engineering in Indian carp is concentrating on producing allotriploids of their hybrids and evaluating them for hardiness and resistance to parasite infections. It may be appropriate to mention here that among the Indian major carps, *L. calbasu* followed by *C. catla* are resistant to parasite infections, especially the most commonly noticed ectoparasite *Argulus* sp. The other two, *L. rohita* and *C. cirrhosus*, fall victim to the infection and also other diseases like the Epizootic ulcerative syndrome (EUS). Again, comparing *L. calbasu* and *C. catla*, the former is very hardy and can withstand relatively rough handling without developing any bruises unlike the latter. Because of these qualities of *L. calbasu* and *C. catla*, work is being done to produce allotriploids with these carp species, develop hybrids with males of the weaker species like *L. rohita* and *C. cirrhosus*, increase the genomic input from the maternal species with better resistance and study the nature of the allotriploids.

3.3.4.6 Gene transfer or genetic engineering research

On the applied side, some attempts have been made at the National Institute of Immunology (NII), New Delhi, to introduce the growth hormone gene in the embryos of *L. rohita*. About 150 000-200 000 copies of a fusion gene containing murine metallothionine promoter (Mu MT) and the coding sequence of the human growth hormone gene (hGH) were microinjected into 434 fertilized one-celled eggs of *L. rohita*. However, only 50 of these microinjected embryos survived. DNA analysis from gill tissue of the surviving embryos showed that only two of them contained the transgene in their genome, carrying one to two copies of the human growth hormone gene. Thus, these preliminary studies indicated the possibilities of introducing foreign DNA into major carp embryos (Alok et al. 1995).

Other transgenic work on zebra fish is mostly academic in nature. Pandian et al. (1991) microinjected fertilized eggs of zebra fish with a plasmid containing the rat growth-hormone gene, prior to the first cleavage. Genomic DNA of the embryos developed from the microinjected eggs was extracted and analyzed by slot-blot and Southern-blot hybridization, using labeled plasmid as a probe. The hybridization patterns indicate genomic, extrachromosomal and also mosaic integrations. It was also reported that the concentration of persisting extrachromosomal DNA progressively decreased in F_1 and F_2 generations. The growth rate in these transgenic fish was observed to be high in F_0 and F_1 generations but low in F_2. The extra or higher growth in F_0 and F_1 may be due to transient expression of the extrachromosomal DNA, carrying the growth hormone gene in these generations.

3.3.5 Utilization of genetic resources in aquaculture

Surveys have been and are being conducted in some states like Karnataka and Orissa to assess the status of stocks in major hatcheries against wild stocks, in terms of the inbreeding level in hatchery-bred seed. Biochemical and molecular genetic techniques are in use to identify stocks/ populations and develop genetic markers. Many of the progressive fish farmers are also becoming aware of the importance and effective role that genetics can play in improving the quality of the farm products and boosting the production levels from their farms with assured returns.

In Karnataka, a crossbred *C. catla* from two different hatchery parental stocks was recommended for commercial production in 2001 following trials that demonstrated higher yields in the crossbred than in the parent stocks. A selected common carp stock from Vietnam is also undergoing state level evaluation following promising trials against the local stock.

The genetically improved *L. rohita* has indicated a 35-40 per cent faster growth after two generations of selection in the first set of field trials undertaken in different parts of India. The improved stocks have been disseminated to selected hatcheries for commercial production.

3.3.6 Summary

As agriculture plays an important role in the economy of India, the Government set the priorities in the order of agriculture, animal husbandry and then fisheries. Attention was paid more to developing genetics-based technologies in agriculture and farm animals. After attaining self-sufficiency in these areas, over the past 20-25 years, aquaculture is receiving due recognition through the development of various farming systems. Several aquaculture technologies, such as extensive, semi-intensive and intensive, have already been developed that are designed for marginal, middle level farmers and also industrialists. By these technologies the farmers in India are able to produce 4-10 tonnes of fish/ ha/yr, which otherwise was only 0.6 tonnes/ha/yr. However, as quality increases quantity, for sustainable yields, these technologies need to be supported by using genetically-improved varieties of candidate species. Therefore, the immediate task before us is the exploitation of the genetic potential of the candidate species that are commonly used in aquaculture. To this end, efforts are already being made and several genetic improvement methods are in progress. Selective breeding, which is at present the most practical approach for stock improvement, is underway to improve the growth of *L. rohita*, one of the most important species among Indian major carps. Other modern genetic improvement technologies like chromosomal and genetic engineering technologies under experimentation are also expected to achieve sustainability of aquaculture production.

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3.4 Carp Genetic Resources of Indonesia

Lies Emmawati[1], Achmad Azizi[1], M. Sulhi[1], Subagyo[1] and Atmadja Hardjamulia[1]

3.4.1 Introduction

Indonesia is a country with a high level of biological diversity (Anon. 1994). One element of the biological richness of Indonesia is the freshwater fish. There are approximately 1 050 species, of which about 950 species are found in western Indonesia and Sulawesi (Kottelat et al. 1993) and at least 103 species are found in Sahul Shelf (Schuster 1950). The fish species are characterized by their high levels of endemism. (Kottelat et al. 1993) reported that in Sumatra there are 30 endemic species, in Kalimantan 149 species, in Java 12 species and in Sulawesi 52 species.

Biological richness in freshwater fish also indicates a wealth of genetic resources of freshwater fish. The freshwater fish genetic resources which are presently exploited are the carps, i.e. common carp (*Cyprinus carpio*), Java barb (*Barbonymus gonionotus*), nilem carp (*Osteochilus hasseltii*), jelawat carp (*Leptobarbus hoevenii*), mata merah (*Puntius orphoides*), kancra or soro carp (*Tor tambra*), batak carp (*Neolissochilus thienemanni*) and others. *C. carpio* in particular is one of the economically important species in Indonesia, consisting of several strains that vary in color and body form. In 1998 freshwater fish culture production was about 276 000 tonnes, of which *C. carpio* was a leading species with a production of about 109 900 metric tonnes (39.8 per cent), while the production of *B. gonionotus* and *O. hasseltii* were 23 100 tonnes (8.4 per cent) and 10 800 tonnes (3.9 per cent), respectively (DGF 2000). *C. carpio* is cultivated in ponds, rice fields, bamboo or wooden cages in rivers and in floating net cages in lakes or reservoirs, while *B. gonionotus* and *O. hasseltii* are cultivated in ponds and rice fields.

Nowadays, the fish hatchery and growout industries provide many job opportunities that are generally based in rural areas. In addition, *C. carpio* and freshwater fish in general constitute one of the cheap sources of animal protein for people with low incomes.

3.4.2 Cyprinid species of importance to aquaculture

3.4.2.1 Indigenous species

At least 10 indigenous fish species are used in aquaculture (Table 3.4.1). *B. gonionotus* is the dominant species, particularly in Java. Both government hatcheries and fish farmers produce fry of this Java barb. In East Java they are also

Table 3.4.1. Indigenous cultivated and prospective carp species in Indonesia

Species	Common name	Culture sites
Cultivated species		
1. *Barbonymus gonionotus*	Java carp	Java and Sumatra island
2. *Puntius orphoides*	Merah mata carp	West Java
3. *Barbonymus schwanenfeldii*	Kepiat carp	Riau and West Sumatra
4. *Osteochilus hasseltii*	Nilem carp	West Java and West Sumatra
5. *Leptobarbus hoevenii*	Jelawat carp	Sumatra and Kalimantan
6. *Tor tambra*	Kancra or soro carp	West Java
7. *Tor douronensis*	Semah carp	South Sumatra and Jambi
9. *Tor tambroides*	Tambra/garing carp	Java and Sumatra
10. *Thynnichthys thynnoides*	Ringo carp	Jambi
Prospective species		
1. *Poropuntius huguenini*	Kuleri/kalari/lahoi carp	Kerinci, Jambi (Sumatra)
2. *Osteochilus vittatus*	Medik carp	Kerinci, Jambi (Sumatra)
3. *Neolissochilus thienemannii*	Batak carp	Toba lake (North Sumatra)
4. *Puntioplites bulu*	Bulu-bulu carp	Sumatra and Kalimantan islands
5. *P. waandersi*	Kepiat carp	Sumatra and Kalimantan islands
6. *Barbodes belinka*	Belinka/kapiah carp	Sumatra

[1] Research Institute of Freshwater Fisheries (RIFF), Jalan Raya 2 Sukamandi, Subang 41256, Indonesia.

caught from the Solo River. *P. orphoides* is cultivated in West Java, but at present cultivation of the species is scarce. *L. hoevenii* is cultivated on the islands of Sumatra and Kalimantan and has considerable economic value, but genetic erosion has occurred dramatically on the islands due to overfishing and environmental degradation caused by pollution and silting. Only a few hatcheries exist. In some areas fry are caught from the wild. *Tor tambra* is reared in old ponds in restricted areas in North Sumatra, West Sumatra, West Java (regency of Kuningan) and East Java (regency of Kediri). *Tor douronensis* is reared in Kerinci, a province of Jambi on the island of Sumatra. This species could spawn naturally in ponds after hormone injection. Both of these species are endangered, as is *N. thienemanni* in North Sumatra, which is also scarce. Prospective species for culture are *Puntioplites bulu*, *P. waandersi*, *Barbodes belinka* (Ardiwinata 1981), *Poropuntius huguenini* and *Osteochilus vittatus.*

3.4.2.2 Introduced species

Five species of carps have been identified as introduced species (Table 3.4.2). Of these, *C. carpio* is the dominant freshwater cultured species. Culture of Chinese carps has not developed due to a lack of local market demand. Experience in West Java showed that silver carp (*Hypophthal-michthys molitrix*) grow fast utilizing plankton in sewage ponds, but fish farmers were not interested in developing cultivation of the species due to marketing problems. Grass carp (*Ctenopharyngodon idella*) is utilized to eradicate aquatic weeds such as water hyacinth in Singkarak Lake in West Sumatra.

3.4.3 Documentation and evaluation of genetic resources

Both literature searches and questionnaires were used to compile the information on cyprinid genetic resources in Indonesia.

a. Data search in universities in West Java, Central Java and West Sumatera (University of Indonesia, UI; Gajah Mada University, UGM; Bogor Agriculture University, IPB; Diponegoro University, UNDIP; Jendral Sudirman University, UNSUD; National University, UNAS; Pakuan University, UNPAK; Bung Hatta University, UBH; Siliwangi Univesity, UNSIL; Fisheries College, STP; Academy of Fishery Extention Experts, APP; Djuanda University, UNIDA; Satyawacana University; and Brawijaya University, UNIBRA). Related fishery institutions comprised the Institute for Evaluation and Development of Technology, BPPT; Provincial Fishery Extension Services; Central Fish Hatchery, BBI; and the Research Institute for Freshwater Fisheries, RIFF (Balitkanwar).
b. Documents used as references consisted of students' theses and dissertations, journals, bulletins, reseach papers, abstracts and research newsletters.
c. Compilation of the basic genetic resources was carried out by distributing questionnaires to various Provincial/Subprovincial Fishery Extension Services and Central Fish Hatcheries in West and Central Java. Full observations were conducted in Central Fish Hatcheries competent in *C. carpio* culture, especially in aspects of brood stock management.

3.4.3.1 *C. carpio*: biogeography, current distribution of different strains in Indonesia and evaluation of these strains

Based on biogeography, *C. carpio* is thought to have originated in the Caspian Sea area and later spread to Eastern Europe and China (Balon 1995). Then the fish developed to become a very important culture species. Later in the course of development, *C. carpio* strains developed both naturally and artificially in culture over a relatively long period. The light colored strain *C. carpio* is thought to descend from the Asian *C. carpio* while that with dark color to descend from the European *C. carpio* (Hardjamulia et al. 1982; Suseno et al. 1983).

Table 3.4.2. Cyprinid species introduced into Indonesia.

Species	Common name	Sources and year of introduction
1. *Cyprinus carpio*	Common carp	China and Europe in the Middle Ages (Schuster 1950)
- The Galician variety		The Netherlands in 1927
- The Frankish variety		The Netherlands in 1930
- unknown		
2. *Carassius auratus*	Goldfish	China in the Middle Ages (Schuster 1950)
3. *Ctenopharyngodon idella*	Grass carp	First introduction from Thailand in 1949 (Schuster 1950); also from Taiwan in 1964, 1969 (Djajadiredja et al. 1977)
4. *Hypophthalmichthys molitrix*	Silver carp	Taiwan in 1964, 1994 (Djajadiredja et al. 1977)
5. *Aristichthys nobilis*	Bighead carp	Taiwan in 1964 (Djajdiredja et al. 1977)

The Galicia strain (the "gajah" carp, meaning "elephant" carp) and the Frankish strain (the "kaca" carp, meaning "mirror" carp) came from Holland. Both carps are popular among fish farmers as their flesh is more compact with a small number of spines, and they grow faster than the earlier existing (introduced) strains (which people called the local strains), but during further development, crosses with the local strain took place and it is difficult to distinguish them from the original strain. Similar cases also occurred with the *C. carpio* introduced from Taiwan, Germany, and Japan. These strains were then mixed with the local strains. But it is thought to be possible to find the Koi strain among those farmers who have been succesful in maintaining its purity (Suseno et al.1983).

Based on the results of cooperation between RIFF and the IDRC (International Development Research Centre), Canada, data on the distribution of 9 *C. carpio* strains have been obtained, namely Majalaya, Sinyonya, Punten, Merah (red), Kaca, Kumpay, Domas, Rajadanu, and local. These strains are distributed in nine provinces in Indonesia, i.e. Aceh, West Sumatera, Jambi, Bengkulu, West Java, Central Java, East Java, North Sulawesi, and South Sulawesi. The distribution of these *C. carpio* strains is illustrated in Table 3.4.3.

Table 3.4.3. Distribution of nine *C.carpio* strains in nine provinces in Indonesia (1 = Majalaya; 2 = Sinyonya; 3 = Punten; 4 = Merah; 5 = Kaca; 6 = Kumpay; 7 = Domas; 8 =Rajadanu; 9 =local; and - = no information on strain)

Province	Strain
Aceh	1, 2, 4, 9, -
West Sumatra	1, 2, 3, 4, 5, 9,-
Jambi	2, 3, 4, 6, 9,-
Bengkulu	1,2,3,4,5,6,7,9
West Java	All
Central Java	1,2,3,4,5,6,7,9
East Java	1, 2, 3, 4, 5, 6, 9, -
North Sulawesi	1, 2, 3, 4, 5, 9, -
South Sulawesi	1, 2, 3, 6, 9, -

Hardjamulia and Suseno (1976) describe the phenotypic identification of several varieties of *C. carpio* found in Indonesia as follows:

- **Majalaya** strain: The color of the scales is grayish green and the margin of each scale is darker in color and taller toward the back. The cross-section of the body is more pointed toward the back than that of other strains. The snout is more flattened and the stomach wall is thicker than that of other strains.
- **Sinyonya** strain: The color of the scales is light yellow, the body is slim and the back is shorter than that of the punten strain. The eyes are narrow in adult fish but round in young fish.
- **Punten** strain: The color of the scales is blackish green, the body is relatively shorter than that of other strains, the back is high, and the eyes are rather bulging.
- **Merah** strain: The color of the scales is yellowish red, the body is relatively long and the eyes are somewhat bulging.
- **Kaca** strain: The body form is not particularly distinctive and the scales are irregular with several parts of the body near the back scaleless or covered with only a few large scales. According to Ardiwinata (1981), this strain is also called "gajah" (elephant) carp.
- **Kumpay** strain: The first characteristic is its very long fins. The color is similar to the usual *C. carpio* and the body form is always rounded (Buschkiel *in* Ardiwinata 1981).
- **Taiwan** strain: The color of the scales is yellowish green; the body is relatively longer than the punten strain. The cross-section of the back tends to be round and the eyes are somewhat bulging.
- **Kancra Domas** strain: The body is long; the color of the scales is golden brown or reddish; in the middle of the body is a horizontal line bordering the darker color of the upper part of the body and the golden shiny color of the lower part; and the scales are relativelly small with a less regular pattern (Ardiwinata 1981).
- **German** strain: According to Vaas (1957), the German strain has a body height higher than *C. carpio* in general.

Various trials have been carried out to evaluate the growth and other traits in these strains of *C. carpio*:

- Growth: Among the Majalaya, Punten, Sinyonya and Taiwan strains, with the initial average body weight of 20 g cultured for 60 days in concrete tanks, the Majalaya showed the best growth with the protein retention of 20.45 per cent, the fat retention of 61.23 per cent, and the growth rate of 1.04 per cent/day (Irawan 1986).
- An evaluation of the growth of the Ganefo, local, Punten and Majalaya strains reared in earthen ponds for 60 days showed that the Punten had the best growth with a feed conversion rate of 0.81 (Setyaningrum 1990).

- Spawning and rearing of the local strain in floating net cages showed that spawning of *C. carpio* breeders could be carried out naturally with a hatching rate of 80 and very good larval condition up to the fifth day, but the larval condition worsened afterward (Subagyo et al. 1991).
- The production performance of the *C. carpio* reared in floating net cages of 1 m^3, showed the highest production when fed with floating feed, reaching a production of 138.35 kg/m^3 at a density of 400 individuals, and feed conversion rate of 2.30.
- An evaluation of the growth of the local *C. carpio* (Sebulu, East Kalimantan) reared in cages ("hapa") gave a production of 46.4 kg/ cage, feed conversion rate of 2.3, survival rate of 97.0 per cent (Suryanti et al. 1994a).
- An evaluation of the growth of the local *C. carpio* (Way Rarem, Lampung) reared in cages, showed a production of 41.28 kg/cage, of 1 m^3 on size, daily growth rate of 1.37 per cent with a feed conversion rate of 2.1 (Suryanti et al. 1994b).
- An evaluation of the growth of the Majalaya strain reared in floating net cages in a polyculture system with the Nile tilapia ("nila") showed a decrease in production compared with its monoculture (67.4 per cent) (Jangkaru et al. 1980).
- The hybrid of the Taiwan strain crossed with the local strain, with 40 per cent protein feed for good growth, showed a feed conversion rate of 1.71 with a weight gain of 16.8 g (Suhenda and Djajadiredja 1980).
- The local *C. carpio* (Kerasaan, North Sumatra) evaluated after rearing in floating net cages at a stocking density of 6 kg/ m^2 showed a feed conversion rate of 2.7 (Dharma 1988).
- An evaluation of the local strains (Kuningan, Cianjur, Pandeglang, and Jogyakarta) by measuring the scale circumference showed that the local strain cianjur had a better growth rate (Sudarto 1987).
- Attempts to induce spawning by using cow's hormone gonadotropin with the Wildan Cianjur strain showed that this only stimulated the occurrence of GVM (movement of the nucleus from the center to the periphery), but by using extract of the fish pituitary gland the effectiveness was higher (Sugiayanto 1997).
- The local strain of *C. carpio* was evaluated for its fertility by using several treatments. The best fertility occurred in the treatment without using the "Ringer" solution, in which the fertility reached 91.7 per cent with the hatching rate of 88.14 per cent. The use of a fertilization solution could decrease the hatching rate and percentage of egg fertility (Kuswanto 1996).
- The technique of freezing the milt of *C. carpio* by combining glycerol and chicken egg yolk into a physiological solution as a diluting solvent had a good effect on the fertility and hatching rate of the *C. carpio* eggs. A combination of 10 per cent glycerol and 5 per cent egg yolk in the diluent gave better sperm moility, giving a fertility rate of 40.2-78.3 per cent.
- The pigment cells of *C. carpio* (from Sukabumi), which were red, yellow, white, green and blue, were analyzed. The first chemical analysis was made with a sulfur test to observe carotenoid and pteridin in bromatophores. The second test was done with ammonia to see the presence or absence of melanin. Results of the research showed that erythrophores and xanthophores had similar pigment content, namely carotenoid. Melanophores could be identified by the melanin content (Gustiano 1994).
- Review of characteristics and evaluation of 4 strains of *C. carpio* (Majalaya, Cangkringan, Sinyonya, and Kaca) as based on Hardjamulia (1998) are shown in Table 3.4.4.

3.4.3.2 *B. gonionotus*

The herbivorous Java carp or Ikan Tawes lives in rivers and swamps. In the wild they normally spawn during the beginning of the rainy season, even in silty and turbid water. *B. gonionotus* is an important cultured species, particularly in East Java, Central Java, some parts of West Java and in West Sumatra. In captivity *B. gonionotus* spawns naturally (after drying of the pond bottom and adding new water). The other method used is hormone injection for spawning in hapas. Eggs are collected from the hapas and hatched in hatching funnels.

There are four kinds of *B. gonionotus* that are commonly found in Indonesia, namely the "common" tawes, "bule" tawes, "silap" tawes and "kecupay" tawes (Ardiwinata 1971). Information has been obtained on growth at different salinities, response to supplementary feed, effects of fin cutting on hatching rate, polyculture, monoculture, effect of fertilization, effect of *Azolla* in feed (Indarmawan 1981; Abulias 1984; Komariah 1987; Wirawati 1988; Yandes 1989; Wiyono 1990; Rahmayani 1991; Mulyo 1991;

Table 3.4.4. A Comparison of traits of four strains of *C. carpio* (Hardjamulia 1998) -= no information

Characteristics	Strains			
	Majalaya	Mas Cangkringan	Sinyonya	Kaca
1. Morphometry - Color - Eye form - Standard length vs. height vs. width ratio - Head length vs. standard length ratio - Formula of fin rays - Number of lateral line scales - Number of vertebrae - Fin type - Scale pattern - Form of backbone	 Green, grayish Round, bulging 4.85:1.5:1 1:3.57 D. 4,17-20 P. 26-32 V. 16-18 A. 3-5 C. 18-20 28-37 33-34 Normal Cycloid, larger Convex (curved)	 Merah (red) Round, bulging 5:1.9:1 1:3.35 D. 4.16-19 P. 26-36 V. 14-18 A. 3.5 C. 18-20 - - Normal Cycloid -	 Yellow Narrow 4.78:1.64:1 1:3.87 D. 4.17-21 P. 24 – 32 V. 16 – 18 A. 3.5 C. 18-20 - - Normal Cycloid -	 Green Round 4.89:1.72:1 1:3.8 D. 4.14-20 P. 24-32 V. 16-18 A. 3.5 C. 18-20 37-39 32-35 Normal Cycloid., Indented
2. Genetics and Biochemistry - Number of chromosomes - Fatty Acids (%) · C (14 = 20) · C (16 = 0) · C (16 = 1) · C (18 = 0) · C (18 = 1) · C (18 = 2) · C (18 = 3) - Proximate composition: · Protein · Fats · Carbohydrates · Ash - Protein electrophoresis: · Proportion of polymorphic loci · Number of alleles per locus · Heterozygosity · Polymorphic loci	 100 1.18 9.42 3.29 3.03 21.63 15.72 3.79 84.8 9.05 0.25 5.86 0.174 1.217 0.046 Est-2*, Cpi-2*, Pgm*, Sp-2*	 - - - - - - - - - - - - - - - -	 100 1.37 10.89 3.21 2.68 20.13 16.56 2.50 85.47 8.41 0.24 5.86 0.087 1.087 0.057 Est-2*, Pgm*	 100 1.14 11.25 4.50 2.76 24.52 15.90 2.90 84.37 9.29 0.25 6.07 - - - -
3. Growth · Weight (%) · Daily growth rate · Specific growth rate	 - 3.3-9.9 g/day 200-326.67%	 13.88 - -	 - 6.2-9.9 g/day -	 11.84 3.1-9.9 g/day 293.41-308.9%
4. Disease resistance - to bacteria - to virus - to fungi - to protozoa - Incidence at 4 weeks of age (%): · Trichodina · Metazoae · Cestoda · Crustacea - Incidence at 4 weeks of age (%): · Argulus · Dactylogynus	 Relatively resistant Fair-high Fair—resistant Resistant - - Fair-high Low -high Fair-high Resistant - - -	 Resistant - - Resistant - 100.0 - Not resistant - Resistant - 0 0	 Fair Low Resistant Almost resistant - 73.3 Low Low-resistant Low-resistant Resistant - 26.6 45.0	 Fair Low - Resistant - - Low Low-resistant Low-resistant Resistant - - -
5. Fecundity - Absolute fecundity (per kg of body weight)	 85 000-125 000	 112 000-130 000	 100 000-135 000	 98 000-100 000
6. Eggs diameter (mm)	0.9-1.6	0.7-1.5	0.13-1.5	0.8-1.4
7. Larval length (mm)	4.9-10.5	5.3-5.5	5.2-10.6	5.5

8. Production in: - Running water ponds (kg/m²/year) - Floating net cages (kg/unit/year) - Rice fields (kg/ha/periods) a. Paddy-cum-fish b. Between rice harvest and next planting c. Palawija - Stagnant pond (kg/ha/year)	 5 000-6 500 2 400-3 000 150-200 200-250 150-200 251-350			 4 500-6 000 2 100-2 600 - - - 2 500-5 000
9. Percentage of fillet (%)	40.4-46.3	-	37.8-50.52	41.6-41.7
10. Dressing percentage (%)	48.9-80.0	-	48.2-81.23	50.5-74.35
11. Effective Breeding Number (Ne)	11.7-56.8	20.0-41.0	8.5-34.6	13.5
12. First maturity (month) - Male - Female	8-9 18-24	 8 12	 8-9 18-24	 8 18
13. Feed Conversion Ratio (FCR)	2.01	2.20	2.22	2.15
14. Environmental tolerance - Temperature - pH - Salinity	 18-32 5.5 - 8.5 0-4.0	 - - -	 18-32 18-32 0-4.0	 20-24 - 0-4.0

Suripna 1993; Supriyanto 1995; Pardede 1996). In the Jatiluhur reservoir (in West Java), *B. gonionotus* are able to adapt to the aquatic environment, with a production of 212 t in 1987 (Kartasasmita 1988). Breeding technique using hypophysis extract was found to be more successful resulting in shorter time interval of ovulation (Nuryani 1996). The use of Ovaprim at an optimal dosage of 1.0 ml/kg of body weight could give the best ovulation response (23.68 per cent) (Madyawati 1997) and good results in fertility and hatching rates of eggs (Hasbuna 1997).

3.4.3.3 *L. hoevenii*

L. hoevenii is known locally, among other names, as ikan jelawat, lemak, jelajar and bundung. It is an indigenous species of the islands of Sumatra and Kalimantan, and has a high economic value. This herbivorous fish lives in rivers and oxbow lakes. The population of the species is decreasing dramatically due to overfishing, silting and pollution, and it is threatened to become an endangered species. As an example, the oxbow lake of Takapan in Central Kalimantan was a rich habitat for the species and a fishing area during 1960-1970, but now this species is quite rare.

Culture in ponds and cages gave good growth (Kristanto et al. 1991). In the swamp and pond ecosystem, *L. hoevenii* could grow well when using organic (green) manure (Aida and Said 1993). Polyculture of *L. hoevenii* and "ikan ringo" (*T. thynnoides*) in cages gave better results than monoculture (Sunarno and Reksolegora 1982).

The species can be propagated by applying the pituitary gland extract (Hardjamulia and Atmawinata 1980) and also hormone injection (Kristanto et al. 1994). The number of ovulated eggs varies from 29 000-44 000 per kg of brood fish (Hardjamulia 1992). The eggs are transparent and semi-buoyant.

L. hoevenii is commonly cultivated in ponds extensively using organic (green) manure (Aida and Said 1993). Intensive culture using pellet feed is conducted in cages in rivers. Stocking densities of 200-300 fingerlings of an average individual weight of 2.5 g in cages of 1.0 x 1.2 x 1.2 m^3 produced fish averaging 64 g in a four-month rearing period, with a survival rate of 86 per cent and a feed conversion ratio of 2.14.

3.4.3.4 *O. hasseltii*

O. hasseltii is a cyprinid living in rivers and swamps, and it can be cultured in freshwater ponds, particularly in Garut areas (West Java) and West Sumatra. The fish feed on periphyton consisting of Bacillariophyceae, Desmidiaceae, Chlorophyceae and soft or decayed leaves of higher plants (Hora and Pillay, 1962). In nature this species breeds at the end of the rainy season, but in ponds it can breed throughout the year and requires running water. Based on the scale color, two kinds of *O. hasseltii* may be distinguished, namely the "blackish brown" nilem and "red nilem" (Anon. 1979). Use of the hypophysis extract of the *C. carpio* could induce ovulation in *O. hasseltii* and produce 67.5 per cent larvae (Rachman 1996). Use of the hormone progesterone at a dosage of 8 mg/kg body weight gave the best ovulation and optimal hatching rate (Yunalti 1996).

3.4.4 Status of genetic improvement and related research in different species

The preliminary information on fish genetic improvement was on the Punten strain of *C. carpio* during 1922-30 (Ardiwinata 1981). The aims of the mass selection were to get a green color and high body form. The result of the selection indicated that the ratio of body length to body depth was 2.47 and that the fish belonged to the "big belly" carp type.

Many strains of *C. carpio* were the result of mass selection done for decades by fish farmers for color, body form and growth. The Majalaya strain (named after the site where selection was done in Bandung area, West Java) is a typical green *C. carpio* and has a high body form resembling Punten carp, but the dorsal profile is narrow and the color is greener than the Punten. This strain was used in intensive culture in running water ponds and floating net cages in reservoirs. The strain has been distibuted to many provinces in Indonesia. The Sinyonya strain was originally selected by fish farmers in Kadugedong (Pandeglang, in the Province of Banten). This has a yellow color and a slender body form. At least ten strains of *C. carpio* have been identified (Sumantadinata 1995).

In view of the importance of achieving a sustainable utilization of the carps' genetic resources and improvement of the genetic quality of *C. carpio* in particular, attempts to enhance the genetic quality have been made by the Research Institute for Freshwater Fisheries. Evaluation of different strains collected from across Indonesia (see 3.4.3.1) indicated that the four strains, namely Rajadanu, Wildan Wianjur, Sutisna Kuningan and Cangkringan Yogyakarta, have high potentials for development (Nugroho and Wahyudi 1991). In addition, another strain with a high potential for development is the Majalaya as this is the main culture strain widely used by fish farmers. The first priority of the program for improving genetic quality was an attempt in the selection of the strains having high potential as mentioned above, particulary Rajadanu, Wildan Cianjur, Sutisna Kuningan and Majalaya.

Selection for the improvement of the genetic quality of *C. carpio* was based on the results of earlier research. *C.carpio* generally has heritability values ranging from medium to high, 0.25-0.47, for the trait of weight gain (Smisek 1979; Brody et al. 1981). Heritability values for *C. carpio* between four months and four years of age for the trait of weight gain range from 0.15-0.5 (Nenashev 1966; Smisek 1979; Nagy et al. 1980). These heritability values indicate the potential for exploitation. Thus, an improvement of the genetic quality can be accomplished through a breeding program using a combination of cross-breeding and selection methods. The selection method applied should be based on the interaction between the environment and genotypes, and the performance of the basic population of two-way cross (heterosis, H) in the synthetic population system of the four strains mentioned above (INGA 1997; Dunham 1995).

Hybridization between female *C. carpio* and male *B. gonionotus* produced a hybrid of normal body form (Despharni et al. 1989).

So far there is no report on genetic improvement of other carp species such as *B. gonionotus*, *O. hasseltii* and *L. hoevenii*. Although the culture of the first two species has been carried out for a long time, so far study on their genetic improvement is still neglected. This is probably because both species are regarded as cheap fish. In the case of *L. hoevenii*, the application of induced breeding is quite recent.

3.4.5 Utilization of genetic resources in aquaculture

The surveys carried out by RIFF under a collaborative research project funded by the IDRC identified different strains utilized by farmers in Java, Sumatra and Bali (see 3.4.3.1). Of these, Majalaya is the strain most widely used by the farmers. Beyond this study, exploitation of genetic resources of the carps cultured in Indonesia has not been well documented and consequently has not given significant benefit in optimizing production of fish culture. On the contrary, for *C. carpio*, such exploitation has had a negative effect, namely lowering the genetic quality of the fry (Dharma et al. 1986; Maskur et al. 1996). Such a condition resulted from a decrease in the genetic diversity of the fish due to inbreeding in farmers' hatcheries, that generally have brood stock of inadequate Ne (effective population size) (Maskur et al. 1996; Gustiano 1994 and Gustiano et al. 1997). In fact, there are many farmers who have fewer than 10 brood stock individuals.

3.4.6 Summary

C. carpio is seen as the priority species for freshwater aquaculture and genetic improvement in Indonesia. The genetic resources of *C. carpio* are already fairly diverse, as a result of several introductions in the past, followed by mixing, development of different strains and maintenance through a degree of isolation on farms. As well as the largely unmonitored development of these *C. carpio* strains, it seems likely that some degree of inbreeding has taken place in many hatcheries due to low effective population sizes. An evaluation program of *C. carpio* strains has been undertaken by RIFF, through literature reviews at key institutions, and by collecting and comparing *C. carpio* from different areas of Indonesia. This has resulted in the identification of promising strains for further assessment and the development of a genetic improvement program. Less attention has been given to other native cyprinids that also contribute to freshwater aquaculture production.

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3.5 Carp Genetic Resources of Thailand

Nuanmanee Pongthana[1]

3.5.1 Introduction

The total freshwater fish production in Thailand in 2000 was estimated at 472 500 tonnes, of which 201 489 and 271 011 tonnes were from inland capture fisheries and freshwater aquaculture, respectively. The total carp production was 99 800 tonnes, of which 45 per cent was from capture fisheries and 55 per cent was from culture. The major cyprinid species from capture fisheries are silver barb (*Barbonymus gonionotus*) and common carp (*Cyprinus carpio*) that accounted for 45 318 tonnes. The production from carp culture accounted for 54 481.87 tonnes, of which 46 275.84, 5 538.51, 1 171.87, 438.05 and 1 057.60 tonnes were from *B. gonionotus, C. carpio*, rohu (*Labeo rohita*), Chinese carps and mrigal (*Cirrhinus cirrhosus*), respectively (Table 3.5.1). Most of the research in the past has been carried out on biology, reproduction and breeding techniques. Owing to the poor performance of hatchery produced seed, genetic research has focused on these hatchery produced species.

3.5.2 Cyprinid species of importance to aquaculture

3.5.2.1 Importance of cyprinid species

Freshwater fish in Thailand are an important source of animal protein and are highly affordable for consumers. Tilapia, carps and catfish are some of the freshwater species of economic importance. Among these species, cyprinids are popular farmed species contributing about 21 per cent and 18.6 per cent to the total quantity and value of freshwater fishery production in Thailand.

In Thailand, cyprinid production from aquaculture is growing steadily, while the production from capture fisheries is on the decline. With a robust growth rate and the government intensifying its support for the development of freshwater aquaculture, carp aquaculture production is expected to continue to expand. At the species level, *B. gonionotus, C. carpio* and *L. rohita* are the three major species cultured and caught in the inland freshwaters. *B. gonionotus* dominates both the aquaculture and inland capture fishery production. This is expected as it has a longer culture history, being a native species, than *C. carpio* and *L. rohita*, which are exotic species.

All cultured carp species in Thailand can be spawned through hormonal injections. The hormones normally used for induced ovulation in these species are: 1) pituitary homogenate (PG) from Chinese carps; 2) commercially available human chorionic gonadotropin (HCG); 3) luteinizing releasing hormone (LHRH) mixed with Dopamine antagonist; and 4) "Suprefac" mixed with "Domperidone". Artificial or natural spawning is practiced after ovulation.

Although carps are cultured in six regions in Thailand, the production is concentrated in Northeastern Thailand, which accounts for about 36 per cent of carp culture production. Carps are mainly cultured in ponds and this amounts to about 89 per cent of carp culture production. Among the existing culture systems, i.e. pond, paddy field, ditch and cage, the last has only recently been introduced in Thailand. Carp cage culture is mainly found in the western, northern and eastern regions of the country. Western Thailand accounts for 51 per cent of the total carp production from cage culture.

Carp farmers are incurring a mean total production cost of Baht 21 022 per farm or Baht 8 984 (1 US$ = 38 Baht) per ha. About 73 per cent of this total cost is considered as cash costs and 27 per cent as non-cash costs. As feed cost accounts for about 35 per cent of the total cost, this implies that carp farming is a commercialized aquaculture activity in Thailand. With an average production of 2 398 kg/farm or 1 025 kg/ha, carp farmers are earning a gross income of Baht 36 354/farm or Baht 6 552/ha and a net income of Baht 15 332/

[1] Aquatic Animal Genetics Research and Development Institute (AAGRDI), Pathum Thani, Thailand.

Table 3.5.1 Production from carp culture by region and type of culture in Thailand during 2000

Species	Type	North	Northeast	Central	East	West	South	Total
B. gonionotus	Pond	7 535.76	14 044.45	8 544.66	5 862.89	3 462.29	1 637.84	41 087.89
	Paddy field	120.08	1 663.14	60.83	42.11	30.03	70.62	1 986.81
	Ditch	51.06	7.52	2 570.36	77.71	222.45	27.03	2 956.13
	Cage	72.25	0.50	2.68	44.21	125.37	0.00	245.01
	Total	**7 779.15**	**15 715.61**	**11 178.53**	**6 026.92**	**3 840.14**	**1 735.49**	**46 275.84**
C. carpio	Pond	1 399.27	2 696.58	551.29	47.64	28.68	91.24	4 814.70
	Paddy field	29.47	636.91	16.50	0.04	1.02	0.00	683.94
	Ditch	6.97	2.97	28.86	0.37	0.00	0.10	39.27
	Cage	0.00	0.00	0.60	0.00	0.00	0.00	0.60
	Total	**1 435.71**	**3 336.46**	**597.25**	**48.05**	**29.70**	**91.34**	**5 538.51**
L. rohita	Pond	491.60	243.97	130.96	94.65	77.70	83.99	1 122.87
	Paddy field	2.18	13.35	0.00	0.10	8.92	0.00	24.55
	Ditch	8.32	0.00	1.36	6.05	8.40	0.29	24.42
	Cage	0.00	0.00	0.00	0.03	0.00	0.00	0.03
	Total	**502.10**	**257.32**	**132.32**	**100.83**	**95.02**	**84.28**	**1 171.87**
C. cirrhosus	Pond	221.55	231.48	216.48	336.37	23.04	21.44	1 050.36
	Paddy field	0.00	1.41	0.00	0.09	0.00	0.00	1.50
	Ditch	0.00	0.00	4.24	1.17	0.00	0.33	5.74
	Cage	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Total	**221.55**	**232.89**	**220.72**	**337.63**	**23.04**	**21.77**	**1 057.60**
Chinese carps	Pond	11.73	57.38	224.08	77.78	31.49	31.36	433.82
	Paddy field	0.00	3.01	0.00	0.00	0.00	0.00	3.01
	Ditch	0.07	0.00	0.10	0.00	1.00	0.05	1.22
	Cage	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	Total	**11.80**	**60.39**	**224.18**	**77.78**	**32.49**	**31.41**	**438.05**
All carp species	Pond	9 659.91	17 273.86	9 667.47	6 419.33	3 623.20	1 865.87	48 509.64
	Paddy field	151.73	2 317.82	77.33	42.34	39.97	70.62	2 699.81
	Ditch	66.42	10.49	2 604.92	85.30	231.85	27.80	3 026.78
	Cage	72.25	0.50	3.28	44.24	125.37	0.00	245.64
	Total	**9 950.31**	**19 602.67**	**12 353.00**	**6 591.21**	**4 020.39**	**1 964.29**	**54 481.87**

Source: Department of Fisheries 2003

farm or Baht 6 552/ha. With a farmgate price (Baht 15.16/kg) almost twice the cost of production (Baht 8.77), carp farming can be considered a highly profitable aquaculture activity in the country (ICLARM-WorldFish Center 2001).

Currently, farm prices of carps range from Baht 19-27/kg. Owing to consumer preference for *C. carpio*, it is usually priced higher than *B. gonionotus*. There is considerable fluctuation in the monthly farm prices of carps in Thailand. The farm price of *B. gonionotus* is usually at the lowest level in October and at the highest in December, while the price of *C. carpio* is at the lowest level during November-December and at the highest in April. The lowest and highest levels of farm prices of *B. gonionotus* and *C. carpio* are directly associated with their peak and lean production months.

The Office of Agriculture Economics has conducted several research studies on the production and marketing of freshwater species in Thailand. Specially, these studies focused on: 1) problems of production and marketing of freshwater fish species; 2) socioeconomics of freshwater aquaculture; 3) costs and returns of freshwater aquaculture; and 4) production and resource-use efficiency of freshwater aquaculture.

3.5.2.2 Indigenous species of cyprinids in Thailand

The family Cyprinidae, which includes most of the scaled fish of Thailand, has 4 sub-families, 55 genera and more than 200 species. Most of them are economically important for inland capture fisheries, stocking reservoirs and aquaculture.

The indigenous species used in aquaculture are: *B. gonionotus*, Jullien carp (*Probarbus jullieni*), Jullien's mud carp (*Cirrhinus jullieni*), soldier-river barb (*Cyclocheilichthys enoplos*), *Leptobarbus hoevenii, Catlocarpio siamensis* and *Osteochilus hasseltii*. Of these, *B. gonionotus* is the most important and widely cultured species in ponds and rice fields.

B. gonionotus

B. gonionotus is a native carp species occurring naturally throughout the country in rivers, canals, reservoirs and swamps. It is also cultured in ponds and paddy fields. The production from capture fisheries and culture in 2000 was estimated at 87 300 tonnes. The male:female ratio is 1:1.09. The length (L)-weight (W) relationship in fish of 5-20 cm in total length is Log W=-1.884+3.002 Log L, while in fish of 21-43 cm in total length, it is Log W=1.726+2.909 Log L. The sizes at first maturation are 20.4 cm in females and 18 cm in males. The fish is herbivorous, feeds on blue-green algae, green algae, Euglenophytes, diatoms, characeae, aquatic and terrestrial plants. The spawning season is from May to July. Schooling behavior is found during the spawning season. The fecundity (F)-weight (W) relationship in females is Log F=3.61407+0.89321 Log W. Females of 250 g in weight have about 371 200 eggs per fish. Eggs are non-adhesive, semi-buoyant, yellowish gray in color and 0.6 mm in diameter. Fertilized eggs develop to 2.5 mm hatched fry within 12 hours at 25 °C. Females can grow up to 500-700 g within 8-12 months in pond culture, while males can grow up to only 300 g (Tavarutmaneegul et al. 1992).

P. jullieni

The species is found in rivers and reservoirs throughout the country, except in the south. Over-fishing and habitat destruction by pollution have caused a decrease in wild populations. The male: female ratio is 1:1.59. The length (L)-weight (W) relationship in fish of 3-24 cm in total length is Log W=-21992+3.213129 Log L. In fish of 80-126 cm in total length, the length-weight relationship in females is Log W=-213566+3.202361 Log L, and in males is Log W=-1.16151+2.650352 Log L. The sizes at first maturation are 2 kg in males and 5 kg in females. The species is omnivorous, feeding mostly on molluscs. The spawning season is from December to February. Schooling behavior is found during the spawning season. Eggs are adhesive, semi-buoyant, yellowish brown in color and 2 mm in diameter. Fertilized eggs develop to 0.9 mm hatched fry within 72 hours at 23 °C water temperature (Tavarutmaneegul et al. 1992).

C. jullieni

The species is found naturally in rivers and reservoirs throughout the country, except in the south. The male:female ratio is 1:1. The length-weight relationship is Log W=-5.85364+3.455826 Log L. The fish feeds on plankton, organic detritus, aquatic plants and insects. The spawning season is from June to September. Fish migrate upstream to spawn. The size at first maturation is 11 cm in length. The fecundity (F)-weight (W) and fecundity-length (L) relationships in females are F= $3.1021W^{0.0761}$ and F=$-1.7918L^{0.896}$.Females of 12.9-20 cm in length have 23 500-90 500 eggs per fish. Eggs are semi-buoyant. Fertilized eggs develop to hatched fry within 13 hours at 26.8 °C water temperature (Tavarutmaneegul et al. 1992).

C. enoplos

This fish is a native carp species found naturally in rivers and reservoirs throughout the country, except in the south. The male:female ratio is 1: 1.48. The length (L)-weight (W) relationship is Log W=-1.90585+2.9464 Log L. The size at first maturation is 34 cm in length or 435 g in weight. The species feeds mainly on molluscs. The spawning season is from May to September. The fecundity (F)-weight relationship is Log F= 2.957+0.699 Log W. Females of 45-50 cm in length have 137 200 eggs per fish. Eggs are semi-buoyant, yellowish-gray in color and 1 mm in diameter. Fertilized eggs develop to 3.3 mm hatched fry within 17-19 hours at 28 °C water temperature (Tavarutmaneegul et al. 1992).

L. hoevenii

This fish is a native carp species occurring in the Chao Phaya and Mekong River systems and in

Boraphet Reservoir. Although it is suitable for pond and cage culture, there is little local demand for this species as a food fish. It is being produced as a small fish for the ornamental trade.

C. siamensis

The largest cyprinid fish in Thailand and one of the largest in the world, this species occurs in the Chao Phaya River and some of its tributaries. Its normal size is 1-2 m in length or 70-130 kg in weight. It feeds on plankton and aquatic plants. The spawning season is from July to October. The eggs are semi-buoyant, dark brown in color and 1-2 mm in diameter. Fertilized eggs develop into hatched fry within 11-13 hours at 28-29 °C water temperature (Tavarutmaneegul et al. 1992).

O. hasseltii

This fish is a native species very widely distributed in the rivers, canals, swamps and reservoirs throughout the country. It feeds on aquatic plants, plankton and crustaceans. The spawning season is from May to August. Brood fish of 80-200 g have 30 000-3 000 000 eggs per fish. Eggs are semi-buoyant, and grayish green in color. Fertilized eggs develop into hatched fry within 14-18 hours at 23-27 °C water temperature (Tavarutmaneegul et al. 1992).

3.5.2.3 Introduced cyprinid species

In addition to the indigenous species mentioned above, seven species of carps have been introduced into Thailand and used in aquaculture. They are *C. carpio*, silver carp (*Hypophthalmichthys molitrix*), bighead carp (*Aristichthys nobilis*) and grass carp (*Ctenopharyngodon idella*), rohu (*L. rohita*), mrigal (*C. cirrhosus*) and catla (*C. catla*). Table 3.5.2 summarizes these introductions.

C. carpio

Common carp was first introduced in Thailand more than 80-100 years ago. Later, more strains were introduced as described in Table 3.5.2. The species is found naturally throughout the country in rivers, reservoirs and swamps. It is also cultured in ponds, paddy fields and ditches. The spawning season is from January to December. The eggs are adhesive and 1-2 mm in diameter. Fertilized eggs develop into hatched fry within 48 hours at 25 °C water temperature (Tavarutmaneegul et al. 1992).

Chinese carps

Three species of Chinese carps are found in Thailand: *H. molitrix*, *A. nobilis*, and *C. idella*. Chinese carps were introduced in Thailand from

Table 3.5.2. Introductions of cyprinids into Thailand

Species	Year	From	Origin	Introduction source
Cyprinus carpio				
• Big bellied carp	1912	China	China	DOF*
• Japanese common carp	1964, 1971	Japan	Japan	DOF
• Mirror carp	1970	Germany	Germany	DOF
• P-31	1985	Hungary	FRI, Szavas	DOF
• Selected strain	1997	Vietnam	RIA No. 1	NAGRI*
• Selected strain	1998	Vietnam	RIA No. 1	NAGRI
• Rajadanu strain	1998	Indonesia	RIFF	NAGRI
Carassius auratus (Goldfish)	1980	Japan	China	DOF
Ctenopharyngodon idella	1922	China	China	DOF
Hypophthalmichthys molitrix	1932	China	China	DOF
Aristichthys nobilis	1922	China	China	DOF
Labeo rohita				
• Wild population	1968	India	Ganges basin	DOF
• Wild population	1969	India	Ganges basin	DOF
• Selected strain	1998	India	CIFA	NAGRI
Cirrhinus cirrhosus	1980 1982	Bangladesh Lao PDR	Ganges basin	DOF DOF

* DOF = Department of Fisheries, Thailand; NAGRI = National Aquaculture Genetics Research Institute, presently called the Aquatic Animal Genetic Research and Development Institute (AAGRDI).

China more than 100 years ago. They have been stocked in a number of public water bodies where they have become part of extensive culture fisheries. They are mostly cultured in ponds, stocked in combination with one another in the ratio of 2:1:1 for *C. idella, H. molitrix* and *A. nobilis,* respectively. The spawning season is from May to September. The eggs are semi-buoyant and 3-4.5 mm in diameter. Fertilized eggs develop into hatched fry within 18 hours at 28 °C water temperature (Tavarutmaneegul et al. 1992).

L. rohita

L. rohita was introduced into Thailand from India in 1968 and 1969. Nineteen pairs were first brought to Thailand on November 23, 1968 and another 250 fish were introduced later in 1969. The selected strain from the Central Institute of Freshwater Aquaculture, India was introduced into Thailand in 1998. The hatchery-produced fish have been released in reservoirs and swamps in the north, northeast and central plains since 1972. The species is now found naturally in reservoirs and swamps throughout the country, except in the south. This fish is also cultured in ponds. Females of 1 kg in weight have 122 787 eggs per fish. The eggs are semi-buoyant, yellowish gray in color and 1-1.4 mm in diameter. Fertilized eggs develop into hatched fry 16-17 hours at 26-30 °C water temperature (Tavarutmaneegul et al. 1992).

C. cirrhosus

C. cirrhosus was introduced into Thailand from Bangladesh and the Lao PDR in 1980 and 1982. The first 100 fish were introduced from Bangladesh to Thailand on November 28, 1980 and kept at Chiengmai Inland Fisheries Station for growth trials and breeding studies. Later in August 1982, eight pairs of brood fish of 700-800 g in weight and 500 fingerlings were brought by Dr. M.V. Gupta to Kalasin Inland Fisheries Station from the Lao PDR. Seed produced from these two stocks were put in ponds and reservoirs. The spawning season is from April to September. Females of 1 kg in weight have 100 000 eggs per fish. The eggs are semi-buoyant and 1.5 mm in diameter. Fertilized eggs develop into 6.5 mm hatched fry in 15-20 hours at 26-28 °C water temperature (Tavarutmaneegul et al. 1992).

3.5.3 Documentation and evaluation of genetic resources

3.5.3.1 Cyprinid karyotypes

Cytogenetic studies (chromosome number, karyotype and arm number) have been carried out in some Cyprinids, as summarized in Table 3.5.3.

3.5.3.2 Microsatellite DNA loci in *B. gonionotus*

The spatial genetic structure of *B. gonionotus* populations in Thailand was reported by Kamonrat (1996a). Microsatellite DNA markers were developed from a Thai *B. gonionotus* genomic library and used to study various aspects of the genetics of their populations in Thailand in order to provide the means for evaluating management policies for the species in terms of conservation

Table 3.5.3. Karyotypes of cyprinids found in Thailand (m=metacentric, sm=submetacentric, t=telocentric, st=subtelocentric, a=acrocentric)

Species	2n	Karyotype	Arm number	References
Barbonymus gonionotus	50	-	-	Na-Nakorn and Legrand (1992)
B. schwanenfeldii	50	-	74	Magtoon et al. (1989)
Catlocarpio siamensis	96	9m+27sm+3st+9a	168	Magtoon and Donsakul (1989)
Hypsibarbus wetmorei	48	7m+3sm+15a	70	Magtoon et al. (1988)
Epalzeorhynchos munense	48	7m+5sm+4st+8a	72	Donsakul and Magtoon (1993)
Epalzeorhynchos bicolor	50	10m+2sm+1st+12a	74	Donsakul and Magtoon (1993)
L. rohita	50	7m+3sm+2st+13a	70	Donsakul and Magtoon (1993)
Morulius chrysophekadion	50	-	70	Magtoon et al. (1989)
Osteochilus hasseltii	46	7m+12sm	76	Magtoon et al. (1988)
O. melanopleura	50	10m+5sm+1st+1a	96	Donsakul and Magtoon (1995)
Paralaubuca riveroi	48	8m+9sm+2st+5a	82	Donsakul and Magtoon (1995)
Puntioplites proctozysron	50	10m+3sm+3st+9a	76	Donsakul and Magtoon (1995)
Puntius orphoides	50	7m+8sm+2st+8a	74	Magtoon and Donsakul (1990)
P. stoliczkanus	50	11m+11sm+2st+1a	94	Magtoon and Donsakul (1990)
Rasbora sumatrana	50	13m+8sm+1st+3a	92	Donsakul and Magtoon (1995)
Probarbus jullieni	96	11m+7a	132	Magtoon and Donsakul (1989)
Tor soro	100	9m+7sm+3st+31a	132	Magtoon and Donsakul (1990)

and genetic improvement. Twelve natural populations from three rivers and 29 hatchery stocks from the central and northeast regions of Thailand were studied. Genetic variability was high in both groups of populations. Multidimensional scaling analysis of genetic distances revealed the discreteness apparent between watersheds among natural populations, and between geographic regions among hatchery stocks. High genetic variability within populations and significant genetic differentiation between populations both in native and hatchery stocks indicate rich genetic resources of this species in Thailand. However, there was evidence that stock management may pose a threat of losing or altering the genetic integrity of both natural and hatchery populations. Mixed stock analysis of the fish sampled from the rivers indicated that from 75 per cent to 96 per cent were from hatchery populations. This high genetic contamination of the natural populations was undoubtedly the consequence of restocking programs in which millions of individuals of this species are released in rivers annually. Evidence of a reduction of genetic integrity between regions was also observed due to stock transfer. The results suggested an urgent need for genetically based stock management policies for both natural and hatchery populations.

The potential use of microsatellites for brood stock improvement in aquaculture was also studied. Pedigrees of individuals were successfully established in a large communally reared group by using one to five microsatellites. The ability to identify individuals allowed a complicated genetic experiment and selective breeding to be conducted in places where facilities were limited. Results are considered to be more reliable because environmental variances are accounted for as fish are grown together from birth. In this study, heritability of growth traits in three stocks was estimated where all families were reared together. The estimates ranged from 0.193-0.523 suggesting that selective breeding in this species should result in good progress. However, heterozygosity in the largest individuals was greatly reduced, indicating that rapid inbreeding is very likely in simple mass selection strategies.

3.5.3.3 Heritability estimates in cyprinid species

L. rohita

Studies were undertaken by Supsuksamran (1987) to investigate the growth rate of this species raised in cages and to estimate the heritability as well as the genetic, phenotypic and environmental correlation for body weight, total length, body depth, head length and girth of fish at ages of 118, 202 and 285 days. The half-sib analysis was tested from the parents, 5 dams and 15 sires, with each dam mated to 3 sires. A nested, randomized block design that contained 40 fish/cage, 2 cages/sire was used. The average daily gain and the specific growth rate at ages of 168-195 days were 0.33 g/day and 0.7 per cent day, respectively. Crosses involving the biggest females had the highest average daily gain and specific growth rate. Estimates of heritability for body weight, total length, body depth, head length and girth at ages of 118 202 and 285 days were small and ranged from 0.002-0.138 (Table 3.5.4). Genetic correlation between total length and body depth, total length and head length, total length and girth at 285 days old was also large and ranged from 0.483-1.022. Phenotypic correlation between body weight and total length, total length and body depth, total length and head length, total length and girth at ages of 118 202 and 285 days was large and ranged from 0.903-0.970. The environmental correlation between traits studied at ages of 118 202 and 285 days was large and ranged from 0.901-0.976.

A. nobilis

A study on the genetic parameters of *A. nobilis* was conducted by Nukwan (1987). With a nested design, females of three different sizes were mated with three males each. Fingerlings from each mating were reared separately in hapas for three months, and then transferred to separate cages for nine months. The mean heritability estimates of their offspring at the ages of 124, 208, 292 and 362 days were 0.172, 0.095, 0.152 and 0.035 for body weight, and 0.179, 0.080, 0.095 and 0.036 for the total length, respectively (Table 3.5.4). Genetic, environmental and phenotypic correlation estimates for body weight and total length ranged from 0.007-1.239, 0.927-0.957 and 0.925-0.960, respectively.

B. gonionotus

Estimates of heritability from the female variance of growth traits in *B. gonionotus* were reported (Jitpiromsri 1990). Heritabilities for total length at 111, 170, 231 and 276 days old were 0.01±0.06, 0.04±0.25, 0.17±0.54, and 0.2±0.33, respectively (Table 3.5.4). Heritabilities for body weight at 111, 170, 231 and 276 days old were 0.22±0.09, -0.07±0.32, 0.06±0.63, and 0.29±0.52, respectively

(Table 3.5.4). The heritabilities increased with the number of days indicating the effects of the common environment.

3.5.4 Status of genetic improvement and related research in different species

3.5.4.1 *B. gonionotus*

Gynogenetic diploid *B. gonionotus* were successfully induced by UV irradiation of sperm at 196 uW/cm^2, cold shocking of 2 °C for 10 minutes starting 1.5 minutes after fertilization (Pongthana et al. 1995). Neomale or XX-male (genetically female, but functionally male) in this species produced by hormonally masculinized gynogenetic diploid fry using 17α-methyltestosterone (MT) at 25 mg/kg in the diet, for a duration of four or five weeks starting two weeks post-hatch to produce all-female offspring when crossed with ordinary females (Pongthana et al. 1999).

Comparative growth performances of diploid and triploid *B. gonionotus* were reported by Koedprang and Na-Nakorn (1994). Triploidy was induced by subjecting fertilized eggs to a cold shock of 2 °C for 10 minutes commencing 0.5 minutes after fertilization; this resulted in 64-100 per cent triploidy. The sex ratio in triploids was skewed to males, while small numbers of spermatozoa were observed in triploid males. Nuclear volume in triploid fish was significantly higher than that in diploids. No significant differences in growth between diploid and triploid fish were observed at 10 months of age.

Genetic diversity and socioeconomic aspects of *B. gonionotus* culture in Khon Kaen Province were investigated in order to study the feasibility of developing and differentiating new, locally adapted breeds in situations where there is a focus on market-driven decision making by small-holder commercial and subsistence farmers and hatcheries. Nineteen grow-out farms, six traders and six consumers were investigated. Results from the socioeconomic studies indicated that there are three different models of fingerling/fish distribution from hatcheries to consumers: (1) mobile trading system (hatchery-grow-out farms-consumers); (2) membership system (Hatchery-contract persons/village head men-grow-out farms-fish traders-consumers); and (3) farm gate system (hatchery-grow-out farms-consumers).

Table 3.5.4. Heritability estimates for growth traits in *L. rohita, B. gonionotus*, and *A. nobilis*. TL = total length, BW = body weight, h^2_D = heritability estimated from female variance, h^2_S = heritability estimated from male variance

Species	Traits	h²	References
L. rohita	TL at 118 days old	h^2_D = 0.075±0.155	Supsuksamran (1987)
	TL at 202 days old	h^2_D = 0.046±0.074	
	TL at 285 days old	h^2_D = 0.893±0.094	
	BW at 118 days old	h^2_D = 0.102±0.107	
	BW at 202 days old	h^2_D = 0.024±0.044	
	BW at 285 days old	h^2_D = 0.093±0.088	
	BW at 53 days old	h^2_S = 2.01±0.95 h^2_D = 0.31±0.51	
B. gonionotus	TL at 111 days old	h^2_D = 0.012±0.055	Jitpiromsri (1990)
	TL at 170 days old	h^2_D = 0.044±0.246	
	TL at 231 days old	h^2_D = 0.168±0.541	
	TL at 276 days old	h^2_D = 0.202±0.332	
	BW at 111 days old	h^2_D = -0.217±0.090	
	BW at 170 days old	h^2_D = -0.067±0.322	
	BW at 231 days old	h^2_D = 0.055±0.626	
	BW at 276 days old	h^2_D = 0.291±0.517	
A. nobilis	TL at 124 days old	h^2_D = 0.019±0.159	Nukwan (1987)
	TL at 208 days old	h^2_D = 0.078±0.122	
	TL at 292 days old	h^2_D = 0.038±0.109	
	TL at 362 days old	h^2_D = -0.014±0.039	
	BW at 124 days old	h^2_D = 0.077±0.186	
	BW at 208 days old	h^2_D = 0.069±0.128	
	BW at 292 days old	h^2_D = 0.048±0.156	
	BW at 362 days old	h^2_D = 0.004±0.043	

3.5.4.2 *C. carpio*

The growth performance of Maenam Mun and local strains of *C. carpio* was investigated at the Ubonrachathani Inland Fisheries Development Center (Sihapitukgiat et al. 1992). Fish were communally stocked in six 50-m^2 ponds at the same stocking rate of 2 fish/m^2. Data on the growth rate between these two strains were studied. The Maenam Mun strain was 13 per cent larger than the local strain at the final harvest.

An introductory cross was tested to improve the body configuration of the local strain (Pitsanulok stock) of *C. carpio* aiming to increase the body depth and decrease the head length and depth (Kamonrat 1996b). The selection experiment was initiated in 1985. A series of back-crosses with the local strain of the hybrid between the Hungarian (P-31) and the local strains was carried out. Two local paternal and maternal back-cross lines (P31LOC and LOCP31) were created. After three generations of selection, these two lines showed significant difference in overall body configuration. Both lines had a significantly smaller head, but higher body depth and caudal peduncle than the local strain. The body configuration of this fish appeared to be a maternally predominated inheritance. The LOCP31 had a similar "big belly" as the local strain, while the P31LOC did not show this feature. These findings suggested the possibility of improving the body configuration of the local strain through out-crossing with the Hungarian strain (P-31).

3.5.4.3 Research in progress

1. Selective breeding of *B. gonionotus*.
2. Development of monosex culture in *B. gonionotus.*
3. Using circuli spacing technique to compare the growth rate and brood stock management of different strains of *B. gonionotus*.
4. Structure and genetic variation of *B. gonionotus* stocks from four Inland Fisheries Development Centers of Thailand.
5. Cryopreservation of the XX-male *B. gonionotus* spermatozoa.
6. Comparison of the body shape and growth rate between improved and local strains of *C. carpio*.
7. Genetic improvement of the Hungarian and local strains of *C. carpio*.
8. Allozyme genetic data in three *C. carpio* strains.

3.5.5 Utilization of genetic resources in aquaculture

Most public and private hatcheries use wild captured fish as brood stock with regular replacements. In small hatcheries, cultured fish left in the ponds after final harvest are usually used as brood stock. Poor quality seed fish come mostly from these small hatcheries. No broodstock management has been practiced. Very few hatcheries have used genetically improved breeds as brood fish. According to the restocking programs of the Thai Department of Fisheries, more than 50 million hatchery-produced seed are released in rivers, reservoirs and swamps throughout the country each year. In addition, seed or stock transfer has occurred throughout the country due to most hatcheries being located in the central region. These activities have caused genetic contamination of the hatchery and natural populations.

More than 2 000 XX-neomale *B. gonionotus* brood stocks have been distributed to 20 public and 5 private hatcheries throughout Thailand since 1999. About 100 million all-female *B. gonionotus* fingerlings have been produced from these neomales.

3.5.6 Summary

Many of the cyprinid species found in Thailand are of importance to capture fisheries and a fairly large number can be bred in captivity. A relatively small number of indigenous species (principally *B. gonionotus*) and some introduced species are of significance to aquaculture. Large numbers of fry are produced in government hatcheries for restocking of reservoirs and natural waters, leading to concerns about genetic contamination of natural populations. *B. gonionotus* is the main cyprinid species used for restocking.

Aquaculture of cyprinid species can in general be considered to be a profitable activity in Thailand, with strong recent growth and expected increases in both demand and production over the next few years.

Various genetic studies have been undertaken on cyprinids in Thailand, including karyotypes, population genetics, heritability estimation, selective breeding, the development of monosex production and induced triploidy. Of these, monosex *B. gonionotus* has probably had the largest impact on aquaculture.

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3.6. Carp Genetic Resources of Vietnam

N.C. Dan[1], T.M. Thien[1] and P.A. Tuan[1]

3.6.1 Introduction

The total fish production in Vietnam in 1998 was estimated at 1.67 million tonnes, of which around 538 000 tonnes came from aquaculture (MOFI 1998), providing nearly 35 per cent of the total animal protein intake of the nation.

Aquaculture in Vietnam is now moving from extensive to semi-intensive culture systems and commercial scale. Of the main components of the production system such as seed, feed and fish health management, seed quality is the most important factor to enhance the productivity. Unfortunately, the deterioration of the genetic quality in a number of cultured fish species in the country has been very striking since the late 1970s (Thien et al. 1996). In order to improve aquaculture productivity and the market value of the products, genetic research should receive support in order to develop high quality strains, for improving productivity of aquaculture. Understanding the important role of genetics in aquaculture, in recent years the government has increased its funding for selective breeding research and genetic technology. However, due to a lack of qualified manpower and institutions dealing with this area, such research programs are limited, scattered and poorly coordinated.

A review of the research on fish genetics and breeding conducted over recent years, with the results achieved, is briefly presented here.

3.6.2 Cyprinid species of importance to aquaculture

3.6.2.1 Indigenous cultured fish species

Twenty-eight indigenous fish species under 11 families and 23 genera are currently used in aquaculture in Vietnam, of which nine species originated in the north, 14 species in the south and five species are found in both regions.

The popular indigenous cultured cyprinid species in Vietnam are the Vietnamese silver carp (*Hypophthalmichthys harmandi*), common carp (*Cyprinus carpio*), mud carp (*Cirrhinus molitorella*), black carp (*Mylopharyngodon piceus*), silver barb (*Barbonymus gonionotus*) and some other species (Thien and Dan 1997). (Table 3.6.1).

Table 3.6.1. List of 13 indigenous cultured cyprinid species in Vietnam

Local name	Scientific name
Ca chep	*Cyprinus carpio*
Ca me trang	*Hypophthalmichthys harmandi*
Ca troi	*Cirrhinus molitorella*
Ca tram den	*Mylopharyngodon piceus*
Ca chay mat do	*Squaliobarbus curriculus*
Ca Ven	*Megalobrama terminalis*
Ca bong	*Spinibarbus denticulatus*
Ca diec	*Carassius carassius*
Ca me vinh	*Barbonymus gonionotus*
Ca chai	*Leptobarbus hoevenii*
Ca he	*Barbodes altus*
Ca danh	*Puntioplites proctozysron*
Ca me lui	*Osteochilus hasseltii*

3.6.2.2 Introduced carp species in fresh water aquaculture

Since the 1950s, nine exotic cyprinid carp species have been introduced into Vietnam. The details of introduction of cyprinid species and stocks are in Table 3.6.2.

After the introduction of the Chinese silver carp (*Hypophthalmichthys molitrix*) for aquaculture, widespread introgression occurred between this and *H. harmandi*. Despite some efforts to isolate separate stocks of *H. harmandi* (3.6.2), it is currently thought that most of the stocks of silver carp in Vietnam consist mostly or entirely of *H. molitrix*.

[1] Research Institute for Aquaculture No. 1, Bac Ninh, Vietnam.

Table 3.6.2. Name, year and source of introduction of 9 exotic cyprinid species and stocks into Vietnam

English name	Scientific name	Year introduced	Source
Silver carp	*Hypophthalmichthys molitrix*	1964	China
Bighead carp	*Aristichthys nobilis*	1958	China
Grass carp	*Ctenopharyngodon idella*	1958	China
Rohu	*Labeo rohita*	1982	Laos, Thailand
Mrigal	*Cirrhinus cirrhosus*	1984	Laos
Catla	*Catla catla*	1984	Laos
Mirror common carp	*Cyprinus carpio*	1971	Hungary
Scaled common carp	*C. carpio*	1975	Hungary
Indonesian yellow common carp	*C. carpio*	Before 1975	Indonesia

3.6.3 Documentation of genetic resources

3.6.3.1 Morphological characterization of *C. carpio* varieties and their hybrids

There are eight local and three introduced varieties of *C. carpio* in Vietnam. The local varieties, identified on the basis of morphology and coloration, are white scaled, Bac Can, Ho Tay, South Hai Van, Red, Violet, High Body Depth and Scattered Scale varieties (Trong 1983). The introduced *C. carpio* are Hungarian scaled, Hungarian mirror and Indonesian yellow varieties.

Crossing between Hungarian carp and Vietnamese carp, Hungarian carp and Indonesian carp, Vietnamese carp and Indonesian carp was carried out in the 1980s to investigate their hybrid growth performance. Morphological study on Vietnamese, Hungarian, Indonesian and hybrid *C. carpio* (Table 3.6.3) was also conducted by the Research Institute for Aquaculture No.1 and the University of Hanoi in the same period (Thien and Tuong 1983; Thien 1990).

3.6.3.2 Genetic characterization of C. carpio populations

Transferrin of pure *C. carpio* varieties and their hybrids was analyzed using 12 per cent starch gel electrophoresis. Four banding patterns were observed, designated as A, B, C and D, with eight different phenotypes (Thien and Tuong 1983: Thien 1990) (Table 3.6.4).

Table 3.6.3. Morphology and morphometrics of *C. carpio* varieties and their hybrids (H = Hungarian; Y = Indonesian yellow; V = Vietnamese white)

Descriptions	Varieties			Hybrids		
	H	Y	V	H x V	H x Y	V x Y
Body weight (g)	24.3 ± 1.41	22.2 ± 1.27	16.3 ± 0.61	21.2 ± 0.91	22.8± 1.21	12.9 ± 0.89
Standard length (Cm)	8.2 ± 0.16	3.3 ± 0.18	7.9 ± 0.09	8.1± 0.11	8.2 ± 0.15	6.9 ± 0.15
As % of standard length:						
Maximal body height	1.1 ± 0.19	35.1 ± 0.32	30.9 ± 0.25	5.5 ± 0.25	37.3± 0.28	32.2 ± 0.30
Minimal body height	14.3 ± 0.13	13.0 ± 0.13	11.9 ± 0.10	5.5 ± 0.16	13.9± 0.17	14.4 ± 0.17
Length of head	34.1 ± 0.24	31.9 ± 0.13	31.6 ± 0.16	34.4 ± 0.38	33.1± 0.30	32.1 ± 0.40
Dorsal spine to tip most dorsal ray (DSR)	36.3 ± 0.24	36.6 ± 0.25	33.5 ± 0.16	36.6 ± 0.38	34.4± 0.14	37.1 ± 0.30
Intestine length	174.0 ± 1.6	185.7± 1.8	145.0 ± 1.3	175 ± 1.4	186 ±2.20	165.8 ± 2.10
As % of head length:						
Diameter of eye	26.6 ± 0.32	24.0 ± 0.37	29.6 ± 0.49	25.9 ± 0.45	25.4± 0.30	27.9 ± 0.50
Length of barbell	17.8 ± 0.29	18.6 ± 0.25	18.0 ± 0.21	19.2 ± 0.21	18.2± 0.26	17.4 ± 0.30
No. of lateral line scales	37.7 ± 0.20	32.9 ± 0.25	32.0 ± 0.14	32.6 ± 0.12	33.8± 0.16	33.4 ± 0.15
No. of Dorsal rays	18.9 ± 0.12	18.3 ± 0.16	20.4 ± 0.16	18.0 ± 0.08	18.2± 0.26	19.0 ± 0.15
No. of Anal rays	5	5	5	5	5	5
No. of branched stamens in first bow	24.9 ± 0.20	19.7 ± 0.17	7.7 ± 0.18	20.2 ± 0.20	22.7± 0.23	20.2 ± 0.18
No. of vertebrae	35.8 ± 0.12	35.1 ± 0.08	34.1 ± 0.08	34.4 ± 0.13	35.0± 0.16	34.7 ± 0.20

Table 3.6.4. Distribution (%) of Transferrin phenotypes in different *C. carpio* varieties (HS = Hungarian scaled; HM = Hungarian mirror; Y = Indonesian yellow; V = Vietnamese white; HS x V = hybrid Hungarian x Vietnamese)

Variety or cross	n	AA	AB	AC	BB	BC	BD	CC	DD
HS	58	1.72	-	74.14	1.72	13.80	5.17	1.72	1.72
HM	9	11.10	88.90	-	-	-	-	-	-
Y	33	3.58	9.10	39.39	12.12	27.27	-	12.12	-
V	28	3.02	-	92.84	-	3.58	-	-	-
HS x V	33	-	-	93.96	-	-	3.02	-	-

Table 3.6.5. Distribution (%) of esterase phenotypes in different *C. carpio* varieties (HS = Hungarian scaled; HM = Hungarian mirror; Y = Indonesian yellow; V = Vietnamese white; HxV = hybrid Hungarian x Vietnamese)

Variety or cross	N	FF	FS	SS
HS	58	81.03	18.97	-
HM	9	100.00	-	-
Y	33	-	100.00	-
V	28	28.58	71.42	-
H x V	33	42.42	42.42	3.03

Two banding patterns of serum esterase were obtained, designated as F (fast) and S (slow). There were three esterase phenotypes in pure varieties and their hybrids (Table 3.6.5). Two banding patterns with two phenotypes of pre-albumin were observed in pure and hybrid *C. carpio* varieties (Table 3.6.6). Serum proteins were also analyzed in these strains and the H x V cross-bred (Table 3.6.7).

Table 3.6.6. Distribution (%) of pre-albumin phenotypes in different *C. carpio* varieties (HS = Hungarian scaled; HM = Hungarian mirror; Y = Indonesian yellow; V = Vietnamese white; HxV = hybrid Hungarian x Vietnamese)

Variety or cross	N	FS	SS
HS	58	8.64	91.36
HM	9	-	100.0
Y	33	12.12	87.88
V	28	3.58	96.42
H x V	33	9.09	90.91

3.6.3.3 Morphological characterization of Chinese and Vietnamese silver carps

A restoration program of Vietnamese silver carp (*H. harmandi*) and Chinese silver carp (*H. molitrix*) was carried out at RIA-1 during 1981-85. The research program was focused mainly on morphological and meristic characteristics of two silver carp stocks (Tien 1984: Tien and Thien 1985). Table 3.6.8 summarizes these characteristics and shows that there were some differences between the two stocks, particularly for body height and lateral line scale count.

3.6.3.4 Genetic characterization of *H. harmandi*

Tables 3.6.9 to 3.6.15 describe analyses of various parameters from the blood of *H. harmandi* (Tien and Thien 1985).

H. harmandi and *H. molitrix* could be distinguished through the electrophoretic patterns of prealbumin and esterase in liver (Dat and Thien 1996). Octate dehydrogenase (ODH), malate dehydrogenase (MDH) and lactate dehydrogenase (LDH) in heart, blood and muscle, and esterase (EST) in heart, failed to show any difference between the species. Additionally, both have 48 chromosomes.

Table 3.6.7. Serum protein of various *C. carpio* varieties (HS = Hungarian scaled; HM = Hungarian mirror; Y = Indonesian yellow; V = Vietnamese white; H x V = hybrid Hungarian x Vietnamese)

Varieties	Total protein (g%)	Albumin (g%)	α-globulin (g%)	β-globulin (g%)	δ-globulin (%)
V	60.60 ± 0.041	85.85 ± 0.034	63.63 ± 0.036	56.56 ± 0.036	56.56 ± 0.041
HS	3.03 ± 0.044	1.05 ± 0.066	0.69 ± 0.029	0.63 ± 0.029	66.66 ± 0.04
HM	3.06 ± 0.033	1.06 ± 0.072	0.70 ± 0.025	0.68 ± 0.036	62.62 ± 0.033
Y	3.51 ± 0.033	1.18 ± 0.048	0.84 ± 0.039	0.75 ± 0.033	74.74 ± 0.033
H x V	2.84 ± 0.022	0.90 ± 0.034	0.71 ± 0.026	0.64 ± 0.022	0.59 ± 0.022

Table 3.6.8. Morphological characteristics of Vietnamese *(H. harmandi)* and Chinese *(H. molitrix)* silver carp populations. L: standard length; W: body weight; C: head length; Hc: head height; H: body height; AD: ante dorsal; LP pectoral fin length; PV: length between pectoral and pelvic fins; LA: length anal fin base; PL: caudal peduncle length; LL: number or scale in lateral line. The data given as percentages are percentages of standard length

Characters	*H. harmandi*			*H. molitrix*		
	Original records n=14	Generation I 1984 n=25	Generation II 1988 n=25	Generation I 1984 n=25	Generation II 1988 n=25	Generation III 1990 n=25
L (cm)	21.20 ± 0.50	31.40 ± 0.30	29.00 ± 2.90	29.80 ± 0.20	28.10 ± 0.1	14.00 ± 0.50
W (g)	385.00 ± 20.7	619.70 ± 15.50	507.40 ± 0.20	476.70 ± 7.70	380.40 ± 0.2	41.10 ± 4.70
C (%)	28.40 ± 0.80	29.70 ± 0.20	29.30 ± 0.10	27.80 ± 0.10	28.60 ± 0.1	29.00 ± 0.10
Hc (%)	28.40 ± 0.10	23.50 ± 0.10	24.80 ± 0.10	23.90 ± 0.10	24.70 ± 0.1	
H (%)	33.40 ± 0.80	32.00 ± 0.30	30.10 ± 0.30	29.00 ± 0.20	28.60 ± 0.2	27.50 ± 0.10
AD (%)	55.20 ± 0.80	50.60 ± 0.40	52.10 ± 0.30	49.50 ± 0.20	51.30 ± 0.2	50.70 ± 0.10
LP (%)	22.10 ± 0.60	21.90 ± 0.30	20.40 ± 0.30	21.00 ± 0.10	21.50 ± 0.4	21.90 ± 0.10
PV	21.90 ± 0.80	20.80 ± 0.20	18.70 ± 0.80	21.20 ± 0.40	19.90 ± 0.2	20.50 ± 0.10
LA	17.50 ± 0.90	16.30 ± 0.20	15.30 ± 0.20	15.30 ± 0.20	15.30 ± 0.3	15.00 ± 0.50
PL	13.40 ± 0.60	17.30 ± 0.20	17.40 ± 0.20	17.10 ± 0.30	18.10 ± 0.3	18.90 ± 0.10
LL (No.)	89.50 ± 3.20	87.50 ± 0.50	90.00 ± 0.80	103.60 ± 0.50	103.60 ± 0.7	

Table 3.6.9. Blood cells and haemoglobin count of *H. harmandi*

Blood cells	Range	Mean ± S.D.
Erythrocytes (x10^{6}.mm^{-3})	1.55-2.90	52.52 ± 0.05
Haemoglobin (g%)	7.50-10.7	10.0 ± 0.10
Leucocytes (x10^{3}.mm^{-3})	36.0-87.0	51.25 ± 1.25
- Lymphocytes (%)	75.0-95.0	6.60 ± 0.85
- Monocytes (%)	1.0-10.0	66.66 ± 0.44
- Thrombocytes (%)	0.0-7.0	77.77 ± 0.41
- Polymorphonuclear leucocytes (%)	0.0-4.0	1.00 ± 0.03

Table 3.6.10. Serum protein concentration of *H. harmandi*

Serum protein	mean ± STD (as %)	Mean ± STD (as g %)
Albumin	48.48 ± 0.48	8.08 ± 0.01
α-globulin	16.16 ± 0.24	1.0 ± 0.01
β-globulin	28.28 ± 0.20	58.58 ± 0.01
δ-globulin	25.20 ± 0.28	0.89 ± 0.01

3.6.3.5 Karyotyping of carp species

Research on karyotyping of 17 carp species in Vietnam was carried out by Dung (1990; 1992) and Trong and Dung (1990). About 20–25 fingerlings from each species were karyotyped by colchinice treatment and the chromosomes counted. According to the solid-tissue technique slides were prepared and 353 metaphase chromosome spreads were photographed. Separate chromosomes were measured. Classification of chromosome groups was made based on the ratio between the length of long or short arms according to Levan (1964). The modal chromosome numbers of the studied species are shown in Table 3.6.16.

Table 3.6.11. Erythrocytes, haemoglobin and leucocytes count of *H. harmandi* at various ages

Age of fish	No. of fish sampled	Erythrocytes (10^{6}.mm^{-3})	Hgb (g %)	Leucocytes (10^{3}.mm^{-3})
0+	45	1.73 ± 0.04	8.26 ± 0.06	36.83 ± 0.58
1+	36	2.26 ± 0.02	9.11 ± 0.04	49.62 ± 0.67
2+	45	2.82 ± 0.02	10.33± 0.05	49.90 ± 0.79
4 – 5	33	2.60 ± 0.03	10.21 ± 0.05	57.15 ± 0.63

Table 3.6.12. Concentrations (as %) of different leucocytes of *H. harmandi* at various ages

Age of fish	Lymphocytes	Monocytes	Thrombocytes	Granulocytes
0+	92.87 ± 0.30	5.04 ± 0.02	1.99 ± 0.11	0.53 ± 0.07
1+	89.57 ± 0.63	5.90 ± 0.49	4.90 ± 0.22	0.63 ± 0.08
2+	88.55 ± 0.50	7.49 ± 0.23	4.84 ± 0.20	2.12 ± 0.10
4-5	84.98 ± 0.59	6.64 ± 0.27	6.54 ± 0.27	1.84 ± 0.15

Table 3.6.13. Concentration of serum protein of *H. harmandi* at various ages

Age of fish	Total Protein (g%)	Albumin (%)	α-globulin (%)	β-globulin (%)	δ-globulin (%)
0+	1.73 ± 0.04	19.31± 0.17	13.0 ± 0.15	23.44 ± 0.29	44.23 ± 0.49
1+	2.73 ± 0.08	26.07± 0.65	26.6± 0.25	14.47± 0.69	32.83 ± 0.28
2+	3.36 ± 0.12	34.37± 0.46	25.4 ± 0.50	15.13 ± 0.53	25.1 ± 0.41
4-5	4.01 ± 0.05	32.12 ± 0.31	28.4 ± 0.27	16.95 ± 0.21	22.56 ± 0.29

Table 3.6.14. Erythrocytes, leucocytes and haemoglobin count of *H. harmandi* males and females

Blood cells	Males (mean ± S.D.)	Females (mean ± S.D.)
Erythrocytes (10^{6}.mm^{-3})	2.78±0.03	2.47±0.02
Haemoglobin (g%)	11.14±0.05	9.72±0.04
Leucocytes (10^{3}.mm^{-3})	55.19±0.73	46.96±0.04
Lymphocytes (%)	85.46±0.65	85.06±0.14
Monocytes (%)	6.98±0.29	6.97±0.20
Thrombocytes (%)	5.65±0.25	5.44±0.23
Granulocytes (%)	1.90±0.15	2.50±0.11

Table 3.6.15. Concentrations of serum protein in *H. harmandi* by sex

Sex	Total protein (g%)	Albumin (%)	α-globulin (%)	β-globulin (%)	δ-globulin (%)
Male	3.92	34.24 ± 0.29	25.23 ± 0.38	16.91 ± 0.34	23.63 ± 0.34
Female	3.81	32.16 ± 0.43	28.4 ± 0.73	15.19 ± 0.32	23.75 ± 0.28

Table 3.6.16. Morphological classification of chromosomes in 17 cyprinid species (Source: Dung 1990, 1992; Trong and Dung 1990)

Species	No. of chromosomes	No. of arms	Types of chromosomes			
			Metacentric	Sub-metacentric	Sub-telocentric	Acrocentric
Squaliobarbus curriculus	48	78	18	12	18	
Hemiculter leucisculus	48					
Xenocypris argentea	48	94	20	26	2	
Erythroculter recurvirostrix	48	86	26	12	10	
Toxabramis swinhonis	48	82	22	20		6
			22	12	14	
Rasborinus lineatus	48		20	20		8
	48	84	28	8	12	
Acheilognathus tonkinensis	44					
Carassius auratus	100					
Hypophthalmichthys molitrix	48	88	10	30		8
	48	86	20	18	10	
H. harmandi	48	86	18	20	10	
Mylopharyngodon piceus	48	84	20	16	12	
Cirrhinus molitorella	50	86	20	16	14	
Labeo rohita	50	76	16	10	14	10
Cirrhinus cirrhosus	50	78	16	12	10	12
Barbonymus gonionotus	50	72	8	14	0	28
B. altus	50	82	10	22	4	14
Catla catla	50	72	8	14	14	14

3.6.4 Status of genetic improvement and related research in different species

- **Evaluation of *H. harmandi* and *H. molitrix* and reciprocal hybrids**

This study was carried out using the stocks from the restoration study described in 3.6.3, which were presumed to be representative of the two pure species. Comparisons of the growth rate between these two species indicated that the growth performance of *H. harmandi* and *H. molitrix* was similar, but both reciprocal hybrids grew faster than their parental groups (Table 3.6.17).

Investigations were continued on three-year-old tagged fish of the same populations with communal stocking. The experiments were carried out in three 700 m^2 ponds, with a stocking density of 5 fish/m^2. Table 3.6.18 summarizes the growth performance. *H. harmandi* performed distinctly better than *H. molitrix*. The hybrids from female *H. harmandi* x male *H. molitrix* grew faster than the hybrids from male *H. harmandi* x female *H. molitrix*.

- **Heterosis in intraspecific cross-breeds of *C. carpio***

Eight varieties of local *C. carpio* were investigated, of which white carp, a variety with high viability, is the most popular (Trong 1983). However, this carp and other varieties of Vietnamese *C. carpio* presented slow growth and early maturity. Attempts aimed at obtaining heterosis by crossing among these varieties were not successful.

Two Hungarian *C. carpio* strains (mirror and scale carps) were introduced in Vietnam in 1970 and 1975, respectively. Under Vietnamese conditions, the Hungarian *C. carpio* showed fast growth and late maturation but were easily infected with diseases and possessed low viability. The first hybrid generation (F_1) crossing between

Table 3.6.17. Growth of group 1: *H. harmandi* and *H. molitrix* and their reciprocal hybrids in separate ponds (in 6.5 months); F = female; M = male (An and Thien 1987)

Traits	*H. harmandi*	*H. molitrix*	Female *H. harmandi* x male *H. molitrix*	Female *H. molitrix* x male *H. harmandi*
Initial weight (g)	50.4 ± 1.3 n = 19	56.3 ± 2.9 n = 20	43.4 ± 1.8 n = 15	80.6 ± 4.4 n = 19
Final weight (g)	232.4 ± 8.0 n=34	241.5 ± 7.9 n = 29	317.0 ± 6.2 n = 30	373.1 ± 11.6 n = 30
Increase (g)	182.0	185.2	273.6	292.5
Heterosis (% of midparent)			49.0	59.3

Table 3.6.18. Growth of group II: *H. harmandi* and *H. molitrix* and their reciprocal hybrids (An and Thien 1987)

Traits	*H. harmandi*	*H. molitrix*	Female *H. harmandi* x male *H. molitrix*	Female *H. molitrix* x male *H. harmandi*
First replicate set				
Initial weight (g)	230 ± 9	226 ± 11	330 ± 15	378 ± 14
Final weight (g)	627 ± 16	510 ± 17	716 ± 9	744 ± 18
Increase (g)	397	284	386	366
Heterosis (% of mid parent)			13.3	7.5
Heterosis (% of best parent)			- 2.8	- 7.8
Second replicate set				
Initial weight (g)	296 ± 10	304 ± 16	380 ± 11	432 ± 19
Final weight (g)	549 ± 10	547 ± 17	683 ± 14	695 ± 15
Increase (g)	253	243	303	263
Heterosis (% of mid parent)			22.2	6.4
Heterosis (% of best parent)			19.8	4.0
Third replicate set				
Initial weight (g)	244 ± 7	256 ± 9	330 ± 16	402 ± 14
Final weight (g)	458 ± 9	416 ± 10	578 ± 9	630 ± 14
Increase (g)	214	160	248	228
Heterosis (% of mid parent)			32.8	21.9
Heterosis (% of best parent)			15.8	6.5

Vietnamese white *C. carpio* and Hungarian *C. carpio* showed the best characteristics from their parents, i.e. high survival rate, fast growth and attractive appearance. The survival rate of hybrid fry and fingerlings was much higher than that of Hungarian *C. carpio* (Table 3.6.19). At the same time the survival rate of the hybrids and Vietnamese *C. carpio* was similar.

Hybrid carps grew much faster than Vietnamese *C. carpio*, either in the case of mixed culture in the same ponds or monoculture in separate ponds. In general, the growth rate of the Hungarian *C. carpio* was high, but in most of these experiments the growth rate of hybrids was the highest. The best productivity was obtained in all of the experiments by raising hybrid carp. As a result, tens of millions of larvae, fry and fingerlings of hybrid *C. carpio* from RIA-1 have been provided to farmers annually, thereby contributing to large increases in *C. carpio* culture production in Vietnam. However, due to improper breeding management, the base stocks of *C. carpio* have gradually lost their purity, thus leading to the loss of crossing effectiveness.

- **Mass selection in *C. carpio***

To overcome the difficulty in keeping/maintaining pure *C. carpio* strains for hybridization, mass selection of *C. carpio* was conducted to develop a line having fast growth. This program was started in 1985 from three base stocks, Vietnamese white carp, Hungarian scale carp and Indonesian yellow carp. These *C. carpio* strains were crossed with each other to produce different offspring combinations. Four generations of selection were carried out over six years (1985-91). The selection indices applied in these generations (three *C. carpio* lines per each generation) are shown in Table 3.6.20.

To assess the effectiveness of this mass selection program, the realized heritability of body weight was calculated, based on the body weights of the offspring of the control stock (non-selected fish

Table 3.6.19. Survival rate (%) of fry and fingerling of Vietnamese (V) and Hungarian (H) *C. carpio* and their reciprocal hybrids (VH, HV)

Stage	Crossing	Survival rate (%)		
		1974	1975	1976
Fry	V	51.6	-	71.2
	VH	61.6	70.0	80.0
	HV	60.4	44.3	78.0
	H	22.3	40.0	37.6
Fingerling	V	85.9 ± 9.4	-	78.3 ± 0.2
	VH	94.9 ± 1.9	76.2 ± 2.9	90.0 ± 3.3
	HV	81.4 ± 7.5	76.7 ± 2.3	73.0 ± 11.3
	H	45.7 ± 5.2	38.6 ± 2.4	46.3 ± 5.1

Table 3.6.20. Data describing the mass selection of the hybrid stocks of *C. carpio* (V – Vietnamese, H – Hungarian and Y – Indonesian yellow common carp; F – female, M - male) (Thien and Dan 1994)

Year and generation	Stocks	No. of fish	Body weight (g)	Selection Indices		
				Severity V (%)	Differential S (g)	Intensity i (= S/δ)
1986, F_1	FH x M(YxV)	400	162±6	12.5	99	2.77
	FV x M(YxH)	400	178±4	12.5	84	1.66
	FY x M(HxV)	1720	187±8	7.5	82	1.94
1988, F_2	FH x M(YxV)	248	152±7	10.1	117	1.76
	FV x M(YxH)	258	104±5	9.7	177	2.03
	FY x M(HxV)	253	148±9	9.9	164	1.60
1989, F_3	FH x M(YxV)	75	149±8	33.3	52	1.25
	FV x M(YxH)	243	155±12	32.9	62	0.80
	FY x M(HxV)	74	310±16	33.8	41	0.77
1991, F_4	FH x M(YxV)	200	260±6	20.0	74	1.26
	FV x M(YxH)	209	197±5	19.1	124	1.75
	FY x M(HxV)	189	299±6	25.9	47	1.24

from the same generation) and the selected stock. The realized heritability estimates are presented in Table 3.6.21. A decline in the response to selection was noticed by the F4 generation and a decision was made to change from mass selection to family selection (see below), which is generally more effective for traits with low heritability values.

- **Family selection of *C. carpio***

Family selection of *C. carpio* has been conducted at RIA-1 since 1998, with the main objective to select a *C. carpio* line with fast growth and high survival to improve its production in aquaculture. Using the fifth generation of *C. carpio* from mass selection as the initial material, the program has succeeded in producing two new generations through induced breeding and rearing of juveniles in hapas. The first generation, with about 375 brood fish, were selected from the 5 best families (out of 24). Following the breeding work plan for the second generation, induced breeding was undertaken successfully with 40 families of the selected generation and 23 pairs of the base population. Rearing of juveniles has been carried out entirely in hapas. After harvest, 2 000 fingerlings of the second generation were reared to marketable size for further selection. In the middle of 2001, fish in the grow-out pond were harvested for estimation of genetic parameters. Based on the breeding value (A), 400 fish (200 females and 200 males) were selected as brood stock for the third generation of selection (Dan 1998).

- **Selective breeding of *B. gonionotus***

The program was initiated at the Cai Be Fishery Research Station of RIA-2 in early 1998. The main activities for this year focused on evaluating and selecting a base population as the initial material for the breeding program (Hao 1998). The results achieved for the first year of the program are summarized below.

Collection of material

During the period between early January and the end of February 1998, the brood stock of *B. gonionotus* were collected from six different locations (six stocks) in the southern region and moved to the Cai Be Fishery Research Station for evaluation. Of these, four wild stocks were collected from the river systems of Dong Nai and Ben Tre and branches of the Mekong River, and two stocks came from farms at Can Tho and Tien Giang. The size of these fish ranged from 175.6-371.3 g.

Table 3.6.21. Realized heritability (h^2) of body weight in the hybrid *C. carpio* (F. Hungarian x M. [Vietnamese x Yellow]) (Thien et al. 1996). (1) and (2) refer to the first and second induced spawnings, respectively, from the F4 generation

Year/ Generation	Parent's body weight (g)		Offspring's body weight		Heritability h^2 = R/S
	Control stock	Selected stock	Control stock	Selected stock	
1987 F1	162 ± 6	261 ± 4	180 ± 4	209 ± 6	0.29
1988 F2	218 ± 10	312 ± 21	316	335	0.20
1991 F4 (1)	246 ± 5	334 ± 9	264	268	0.05
(2)	246 ± 5	334 ± 9	288 ± 7	282 ± 5	- 0.07

Induced breeding

The six stocks of *B. gonionotus* were conditioned in earthen ponds until maturity. Induced breeding through hormone injection was carried out both within each stock and in crosses between different stocks. Two breeding sets were attempted and success was achieved the second time, with over 70 per cent of the larvae surviving.

Rearing of fry

The fry obtained from the six stocks and three crosses were reared in earthen ponds and cement tanks for evaluation and comparison of their performance. After four months rearing in an earthen pond, the size of the fish reached 6.5-7.8 g and there was no significant difference in the growth rate among the progenies.

Tagging

Fin clipping and fluorescent tagging were tried. However, the results from both methods need to be evaluated in terms of mortality and color of marks after tagging.

- **Production of all-female *B. gonionotus***

Research on all-female production using techniques for inducting gynogenesis and hormone sex-reversal was done at RIA-1 during 1998. Induction of meiotic gynogenesis in *B. goniontous* was carried out. The sperm were diluted with a 0.85 per cent salt solution to a concentration of 8 x 10^8 sperm.ml^{-1}, and 10 ml of diluted sperm was irradiated in a small petri dish for 60 seconds using an ultraviolet lamp. A cold shock at 2 °C was applied 90 seconds after fertilization at 30 °C to eggs of *B. gonionotus* fertilized with UV-irradiated sperm for 10 minutes. Non-irradiated sperm were used in the diploid control group, and one group of eggs was fertilized with irradiated sperm but was not cold shocked, serving as a haploid control group.

In the haploid control group, all hatchlings died after two days. In contrast, the average survival rates in the diploid control and mitotic gynogenesis groups were 66.7 per cent and 11.5 per cent, respectively. The sex ratios in the gynogenetic fish were from 91.3-97.4 per cent female with a mean of 95.8 per cent, while sex ratios in the control groups was about 1:1. Androgen treatment (17α-methyltestosterone) was orally applied to gynogenetic fish to produce sex-reversed neomale *B. gonionotus*. Gynogenetic fish were fed with 25 mg MT.kg^{-1} diets for 5 weeks. The percentage of males in androgen treated-fish varied from 8.1-36 per cent with a mean of 16.7 per cent male (Thang 1998).

- **Induction of triploidy in *C. carpio***

Nga (1995) carried out studies on the induction of triploidy in *C. carpio*, using cold or heat shocks, in the Southern Branch of Vietnam-Russian Tropical Center and Research Institute for Aquaculture No. 2. The effects of various heat and cold shock parameters on survival rates of embryos were studied, and one trial comparing the growth rate of controls and a cold shocked group was carried out (Table 3.6.22). "Treated" fish grew faster than control fish. However, the experiment was conducted without replication and no information was given on the survival rates of the two groups. In order to identify the proportion of triploid fish induced by cold shocking, 20 fingerlings were used for the karyotyping study, of which 9 individuals possessed chromosome set 2n = 150-156, while the diploid number of chromosomes of the other 11 individuals was 100. Thus the proportion of triploids obtained in this case was 45 per cent. It was concluded that further study was required to improve the technique of producing triploids and to determine the survival, growth rate and gonad development of triploid fish.

Table 3.6.22. Growth performance of normal *C. carpio* and *C. carpio* from cold shock treatments

Date	Pond 1 – normal *C. carpio*		Pond 2 "treated" *C. carpio*	
	Body weight (g)	Total length (cm)	Body weight (g)	Total length (cm)
13 Aug. 1995	3.5	5.7	3.0	6.0
13 Sept. 1995	21.9	11.6	14.3	9.9
13 Oct. 1995	44.6	14.4	44.9	14.2
13 Nov. 1995	71.1	15.7	85.4	16.7
13 Dec. 1995	93.0	18.2	115.5	18.7
13 Jan. 1996	130.0	19.9	134.7	19.5
13 Feb. 1996	167.0	21.9	206.0	23.0

- **Preliminary experiments on generating transgenic *C. auratus***

The first study on gene transfer in fish in Vietnam was carried out at the Institute of Biological Technology, Hanoi (Do et al. 1997). The methods and preliminary results of the study are summarized here.

After hormonal injection for female and male breeders, eggs and sperm were collected by stripping. Fertilization was then carried out by the dry method. 25-30 minutes after fertilization, one-cell embryos formed. The chorion membrane of one-cell stage embryos was removed using trypsin enzyme solution (0.075 per cent concentration). After dechorionation, the embryos were microinjected with a human Growth Hormone (hGH) gene construct. The DNA solution (40 ng/µg in Tris-EDTA) was used for injection. The eggs were then incubated in Holtfreter solution at temperatures of 22-30 °C. The hatching rate of injected eggs without chorion membrane was 20 per cent. The survival rate was 35 per cent for the three-day-old larvae and 3 per cent for the 30-day-old larvae.

3.6.5 Utilization of genetic resources in aquaculture

Aquaculture genetics research in Vietnam began with studies on hybridization of carp species and strains in the 1970s. Of this early research, cross-breeding of *C. carpio* was the most successful. Crossing between Vietnamese white *C. carpio* and Hungarian *C. carpio* produced hybrids showing better performance compared to their parents, i.e. higher survival rate, faster growth and attractive appearance. Also, the hybrids can resist disease better compared to Hungarian *C. carpio*. This hybrid *C. carpio* was disseminated widely and contributed to increased aquaculture production mostly in northern Vietnam. However, the production of this cross-bred declined due to a loss of stock purity. Following this, a mass selection breeding program was conducted for 10 years (1986-95). The fifth selected generation of *C. carpio*, with 33 per cent faster growth compared to the base population was disseminated to farmers, again mostly in northern Vietnam.

Introduced species and stocks have played a significant role in the development of increased aquaculture in Vietnam since the 1970s: the principal contributors to this have been *L. rohita*, *C. cirrhosus*, *H. molitrix*, *C. idella*, and Hungarian *C. carpio* (the last only as a contributor to cross-bred and selected strains).

Awareness of genetics issues has increased among hatchery operators in the last few years, largely due to training programs and extension conducted by the research institutions. This has led to many hatchery operators changing their brood stock management practices and brood stock, for example by exchanging brood stock between local hatcheries or obtaining new brood stock from research institutions. The impacts of this are hard to measure but likely to be fairly widespread.

3.6.6 Summary

About 13 indigenous carp species and five exotic carp species (Chinese and Indian carps) are currently widely cultured in Vietnam. Of these, *L. rohita* and *C. cirrhosus* together account for about 40 per cent of cultured carp, *H. molitrix* about 20 per cent, *C. idella* about 20 per cent and *C. carpio* about 10 per cent. It is estimated that the carp species contribute 75 per cent of freshwater aquaculture production. However, research on the carp genetics resources in the country has been very limited. Some research focused on morphological and genetic characterization of *C. carpio* and *H. molitrix*. The results of this research are useful for classification and selection programs. Cross-breeds and hybridization were tested with several carp species in the 1970s and 1980s. Heterosis was obtained only from hybrids between Hungarian and Vietnamese *C. carpio* strains. Genetic manipulations such as producing all-female *B. gonionotus* using gynogenesis and sex reversal and induction of triploidy in *C. carpio* have been investigated more recently. However, applied results of these programs have been limited to date. Mass selection of *C. carpio* was the main selective breeding program conducted successfully so far. The selection of *C. carpio* is being continued using family selection to replace mass selection. Mass selection of *B. gonionotus* was started recently. Initially, different stocks are being evaluated for this program.

The Vietnamese government has committed itself to a target of increasing aquaculture production from approximately 0.7 million tonne in 2001 to 2.0 million tonnes in 2010, and the value of exports from $1.8 billion (in 2001) to $2.5 billion. Seed quality and genetic improvement are likely to be important factors if these targets are to be achieved. As part of this effort, three national brood stock centers (NBCs) are being constructed. They will be responsible for disseminating and maintaining the genetic quality of existing and improved aquaculture breeds, and will also play a role in genetic improvement programs conducted by the three Research Institutes for Aquaculture. Where different stocks exist for the major cultured cyprinid species, these are currently being evaluated to identify performance differences.

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Chapter 4
Progress in Carp Genetics Research

David J. Penman[1]

4.1 Introduction

Ten years ago, very little was known about the genetics of most of the cyprinid species of interest to aquaculture in Asia, for example the Chinese carps, silver carp (*Hypophthalmichthys molitrix*), bighead carp (*Aristichthys nobilis*) and grass carp (*Ctenopharyngodon idella*); Indian major carps, catla (*Catla catla*), rohu (*Labeo rohita*) and mrigal (*Cirrhinus cirrhosus*); and the silver barb (*Barbonymus gonionotus*). The exceptions to this would have been the common carp (*Cyprinus carpio*) and the goldfish (*Carassius auratus*). Still relatively little is known about the genetics of most of these species, but significant progress has been made in the last decade. Valuable information about the genetic structure of wild and captive populations and traits of interest to aquaculture are now becoming available and our knowledge of these areas should increase greatly over the next decade, as should the applications of genetics to the aquaculture of cyprinids.

This chapter reviews progress in genetics research that is relevant to aquaculture of cyprinid species in Asia. It begins with cytogenetic (4.2) and molecular (4.3) techniques that can be used to provide basic genetic descriptions of cyprinid species and populations, as well as having applications in aquaculture genetics. Hybridization studies, of which many have been carried out, are dealt with next (4.4). Quantitative genetics, the main focus of this publication (4.5), is divided into four subsections (inbreeding and negative selection; current status; stock/strain comparisons; and selective breeding). Chromosome set manipulations (4.6), sex determination and its manipulation (4.7), gene transfer (4.8) and finally applications of cryopreservation (4.9) and tissue culture (4.10) to aquaculture genetics research complete the review.

4.2 Cytogenetics

Basic cytogenetics studies, describing chromosome numbers and morphology, have been carried out on many of the cyprinid species of interest to aquaculture in Asia. Much of this information, and equivalent information on other cyprinids (as well as other groups of fish), is summarized by Klinkhardt et al. (1995). It is evident from a frequency distribution of diploid chromosome numbers for cyprinid species taken from this database that 2n = 50 is by far the most common diploid chromosome number in cyprinids, and also that polyploidisation (tetraploidy and hexaploidy) has occurred in several cyprinids (Fig. 4.1). Ancestrally tetraploid species (approximately 100 chromosomes) are found in the genera *Carassius*, *Cyprinus*, *Spinibarbus* and *Tor*, while *Schizothorax taliensis* and *S. yunnanensis* are ancestrally hexaploid (with 148 chromosomes). In addition to being ancestrally tetraploid, *C. auratus* and *C. gibelio* also have forms that are triploid (relative to the species' ancestrally tetraploid chromosome complement, i.e. they have approximately 150 chromosomes). Reddy (1999) described karyological studies on Indian cyprinid species in some detail. Griffith et al. (2003) demonstrated an increase in the lifespan of fish with larger genomes, from an analysis of a wide range of species. Within individual orders such as the Cypriniformes the relationship was positive but not significant, possibly because of the limited number of samples analyzed.

Perhaps curiously, the number of fundamental chromosome arms (NF) described by various authors in the database of Klinkhardt et al. (1995) showed much more intraspecific variation than the diploid chromosome counts. It is not clear if this reflects genuine polymorphism or differences in interpretation by different authors.

Nucleolus organizer regions (NORs) have been studied in several species of Asian cyprinids (e.g. Carman et al. 1993; John et al. 1993; George et al. 1994; Anjum and Jankun 1998). George et al. (1994) showed that the NORs varied between Indian major carp species. Lakra and Krishna (1996) reviewed chromosome banding

[1] University of Stirling, U.K.

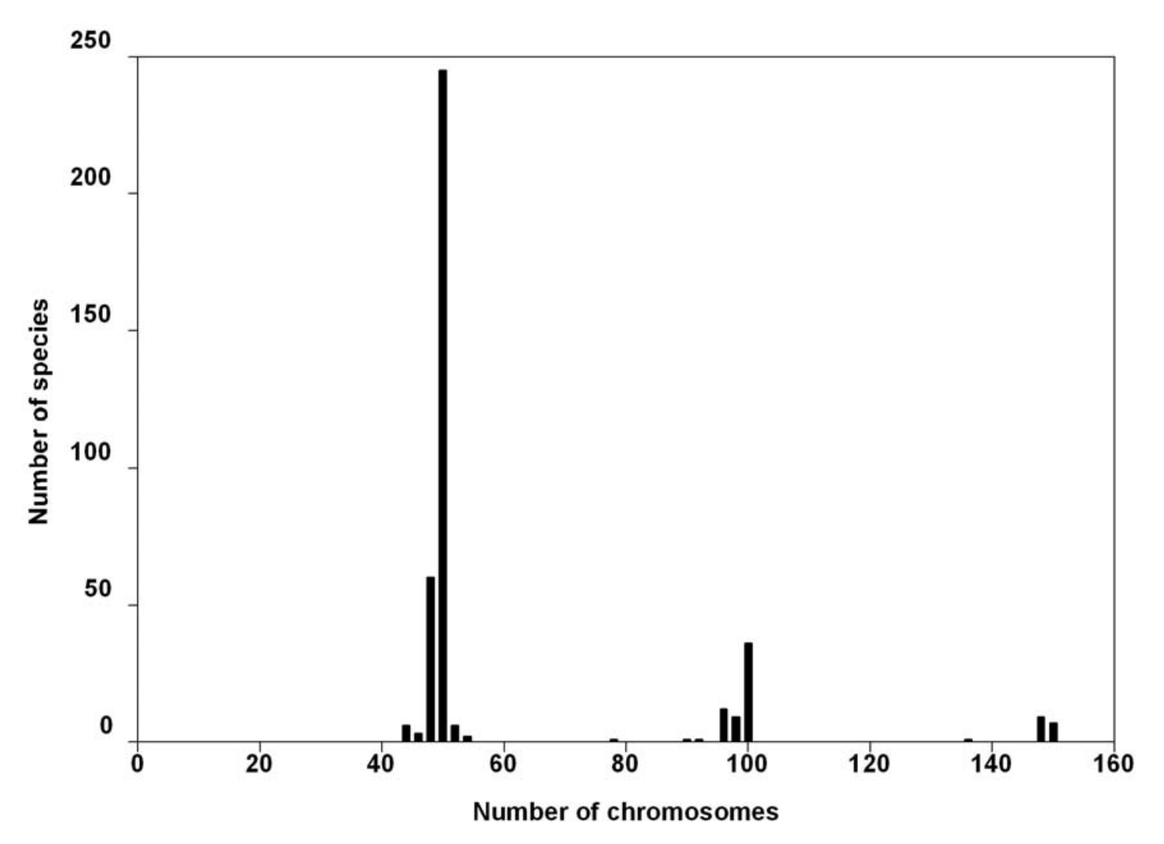

Fig. 4.1. Frequency distribution of the diploid number of chromosomes in cyprinid species (worldwide). Source: Klinkhardt et al. (1995). Where more than one value is given per species in the database, the most common value is shown (in the case of a tie, the lower value is shown). The only cyprinid species in the database that is not shown is *Ptychobarbus dipogon* (2n = 446)

techniques for fish and described G-banding for the first time in Indian major carps. Luo (1998) used trypsin digestion and Giemsa staining to produce chromosome-specific banding in the grass carp. This technique may go some way to overcoming some of the general difficulties in obtaining high resolution banding patterns in fish chromosomes (Goodier and Davidson 1993).

Chromosomes can be used as genetic markers in studies on hybridization, induced polyploidy, gynogenesis and androgenesis. While the techniques involved to prepare metaphase spreads from large numbers of fish or embryos are fairly laborious, they are also well standardized.

Distinguishing triploid and diploid fish using metaphase spreads prepared from hatched embryos is fairly routine and allows analysis at an early stage (Kligerman and Bloom 1977). However, assessment of polyploidy may be more easily carried out using other methods such as semi-automated erythrocyte cell volume quantification (Harrell et al. 1998). Such methods are generally used in *C. carpio* due to the large number of chromosomes in this species (Cherfas et al.1993; Klinkhardt et al. 1995).

Chromosome counts are a good way of verifying haploid controls in gynogenesis or androgenesis experiments, and are generally used in combination with morphological examination (haploid embryos show a typical "haploid syndrome") and scoring survival rates (nearly all haploid embryos die before diploid embryos reach the first feeding stage). Restoration of diploidy via temperature or pressure shocks given to the fertilized eggs following UV treatment of sperm or eggs can be verified by comparing the karyotypes of shocked batches (should be diploid) compared to the UV controls (should be haploid). However, to verify uniparental inheritance in diploid gynogenetic or androgenetic fish and biparental inheritance in diploid controls, molecular genetic techniques such as DNA fingerprinting are more informative.

Interspecific hybridization may not produce the expected diploid animals with one haploid set of chromosomes from each parental species (Chevassus 1983; see 4.4). Altered ploidy levels in such hybridizations should be easily detected, but for parental species with similar karyotypes, analyzing offspring metaphase spreads may not detect some of the more unusual outcomes of interspecific fertilizations, such as gynogenetic or androgenetic progeny. In many situations, allozyme or DNA markers, possibly in combination with karyotype analysis, may be more powerful.

Chromosome polymorphisms have been found in population genetics studies in some species of fish (e.g. variation in B chromosome frequency in the characid *Astyanax scabripinnis*: Néo et al. 2000) but are of limited general application.

Cyprinids are gonochoristic and several aquaculture species have been shown genetically to have apparently fairly simple sex determination systems (generally but not always XX/XY – see 4.7). However, in common with most other species of fish, heteromorphic sex chromosomes appear to be absent or very rare in cyprinids (e.g. Manna and Khuda-Bukhsh 1977; Buth et al. 1991).

Although studies of cyprinid karyotypes yielded some interesting information and applications – for example insights into the role of polyploidization in the evolution of this group, and chromosome counts can be used as markers in chromosome set manipulation experiments – otherwise limited progress has generally been made in such studies. To a large extent this is also true in other groups of fish, but rapid progress is now being made in cytogenetics in certain species of fish (e.g. physical mapping of genes and other DNA sequences). The species where such advances are being made tend to be either high-value aquaculture species or important model species for basic biological research. The latter group includes one cyprinid, the zebra danio (*Danio rerio*), a small cyprinid used as a model species in basic research.

4.3 Genetic markers: species, population and genome analysis

To characterize individuals, populations or species at the genetic level, polymorphic (variable) genetic markers are required. Historically, markers such as allozymes were used, where the product of a gene rather than the DNA sequence itself is examined (polymorphism is detected in the form of bands in different positions in an electrophoresis gel after staining for enzyme activity in the case of allozymes). With the development of a variety of techniques for working directly with DNA sequences, it is now possible to detect DNA polymorphisms directly, both in the nuclear genome and the mitochondrial genome.

Standard techniques can be used for isozymes, multilocus DNA fingerprinting, RAPDs and mitochondrial DNA in different species with relatively little adaptation or prior knowledge of the species, while other markers such as microsatellite DNA loci may require considerable preliminary research to isolate relevant DNA sequences and develop appropriate PCR primers (some microsatellite loci will, however, cross-amplify in related species).

A variety of polymorphic genetic markers, primarily allozymes and different types of DNA markers, can be used for such purposes as phylogenetics, species and hybrid discrimination, population genetic analysis, comparison of levels of genetic variation in hatchery and wild stocks, pedigree analysis in hatchery and experimental breeding, and gene mapping.

Knowledge of the genetic structure of wild populations can be important in conserving genetic resources and managing capture fisheries and identifying potential source populations for the development of aquaculture brood stock. Allozyme analysis has been used to study populations of Chinese carps (*H. molitrix*, *A. nobilis* and *C. idella*) both between and within major river systems. Li et al. (1986) demonstrated differences between natural populations of *H. molitrix*, *A. nobilis* and *C. idella* from the Zhu Jiang (southern China), Chang Jiang (Yangtze: central China) and Heilong (northern China) rivers, using 16 enzyme loci. Allozyme analysis of these three species and black carp (*Mylopharyngodon piceus*) from different areas within the Yangtze River (Xia et al. 1996; Zhao and Li 1996) was carried out to help formulate conservation strategies. The results of Zhao and Li (1996) suggested that there was no significant genetic divergence and that one conservation area in the Yangtze River would be adequate (Swan Oxbow area in the middle reaches: described by Li et al. 1995). However, Lu et al. (1997) used PCR-RFLP analysis of mtDNA to demonstrate that, for three of the four species studied (*H. molitrix, A. nobilis, M. piceus* but not *C. idella*, where mtDNA diversity was lower), juvenile fish from different nursery areas belonged to genetically distinct populations. This implied that these carps could not be managed as a single unit within the Yangtze River – genetically distinct stocks require individual management. The difference between the results from allozyme and mtDNA analyses emphasizes the value of using more than one technique in such studies.

Wang et al. (2003) studied the ND5/6 mtDNA region of *H. molitrix* from Poyang Lake, a major nursery area for this species, and two connected rivers, the Yangtze and the Ganjiang. From analysis of restriction fragment length polymorphisms, they were able to discern differences between the populations in the two rivers and to conclude that the fish in Poyang Lake consisted mainly of the Ganjiang River population.

Genetic markers that show fixed differences between species can be used to monitor hybridization and introgression where these are known or suspected to occur. A low frequency of hybridization occurs in the wild between many pairs of closely related species, but presumably natural selection prevents this from reaching significiant proportions in most cases. In the hatchery environment, hybrids may be deliberately produced for aquaculture, or inadvertent hybridization may take place between similar species. In either case, if F_1 hybrids are used as brood stock, further hybrid or back-cross generations may be produced, which may lead to introgression between the species concerned. This may have deleterious effects on survival and may lead to altered feeding niches in farming systems.

Valenta et al. (1990, 1991) studied enzyme polymorphism in *A. nobilis* and *H. molitrix* in hatchery populations in what was then Czechoslovakia, and concluded that several loci could be used to distinguish between these species and to identify F_1 hybrids. Egelnof (1996) demonstrated that *H. molitrix* and *A. nobilis* used in aquaculture and enhanced fisheries in Bangladesh could be distinguished on the basis of allozyme variation. Zhang et al. (1999) concluded that RAPD analysis could also be used for the same purpose, as well as revealing intraspecific polymorphism. Song et al. (1994) constructed restriction maps of the mtDNA of these two species. Differences between the species were observed: however the maternal inheritance of mtDNA does limit its usefulness in monitoring hybridization. Introgression between *H. molitrix* and *A. nobilis* has been suggested to be a major reason for the decline in performance of *H. molitrix* stocks in Bangladesh and a preliminary study using species-diagnostic microsatellite loci detected some hybrid genotypes in hatchery brood stock (Mia et al. 2002). Likewise, Thien and Tien (1988) suggested that one reason for a decline in the quality of Harmandi silver carp (*Hypophthalmichthys harmandi*) in Vietnam could be due to uncontrolled hybridization of this species and *H. molitrix* imported from China. Payusova and Tselikova (1993) described genetic (allozyme) differences between these two species, which could presumably be used to determine the extent of such hybridization.

Microsatellite DNA loci exhibit the highest levels of genetic variation that can currently be routinely detected and attributed to alleles of single loci (O'Connell and Wright 1997; Carvalho and Hauser 1998). They have tremendous potential to increase the power of genetic analysis of a number of situations. Kamonrat (1996), McConnell et al. (2001b) and Kamonrat et al. (2002) described microsatellite loci from *B. gonionotus*. Kamonrat (1996) applied some of these loci to study several population genetics- and aquaculture-related questions in this species in Thailand. A low but significant level of genetic discreteness was observed between natural populations from different watersheds (Chao Phraya and Mekong rivers), but mixed stock analysis indicated that 75 and 90 per cent of the fish captured from these two river systems, respectively, were of hatchery origin. This could be interpreted as a success in terms of the hatchery-based restocking program, but the lack of a specific policy for restocking and transferring brood stock between hatcheries located in the different watersheds carry the threat of eroding the genetic integrity of the natural populations.

The pedigrees of individual *B. gonionotus* in a large communally reared group could be successfully established by using one to five microsatellites (Kamonrat 1996). This allowed estimation of heritability values (which ranged from 0.193-0.523) without the requirement of separate rearing facilities and/or complex tagging systems, and removed several potentially large environmental sources of phenotypic variance from the experiment.

Polymorphic genetic markers have been applied to the analysis of *C. carpio* populations. This species is thought to have originated in the Caspian Sea area and later spread to Eastern Europe and China (Balon 1995). Both allozyme (Kohlmann and Kersten 1999) and mtDNA (Gross et al. 2002; Zhou et al. 2003) polymorphisms indicated a major division between the European and East Asian populations, with a further division between koi carp and the other East Asian populations studied (see also 4.5.2: given the long history of domestication of *C. carpio*, much of the analysis of further subdivision of Asian *C. carpio* populations is dealt with in this publication in the context of strain development). Microsatellites have also been developed for *C. carpio* (Crooijmans et al. 1997; Aliah et al. 1999). A study utilizing allozyme, microsatellite and mtDNA markers to look at *C. carpio* populations throughout its distribution range concluded that these populations clustered into two groups, Europe/Central Asia (*C. carpio*) and East/Southeast Asia (*Cc. haematopterus*) (Kohlmann et al. 2003), with no justification for a separate Central Asian subspecies (*C. aralensis*). David et al. (2003) estimated that about 60 per cent of 59 microsatellite primer pairs studied in *C. carpio* amplified duplicate loci, supporting other evidence for the tetraploid origin of this species. They furthermore suggested that the tetraploidy may have arisen by hybridization (allotetraploidy) and that this may have occurred relatively recently (approximately 12 million years ago).

Among the Indian major carps and other cyprinid species in South Asia, many species-specific markers have been detected, for example using esterases (Gopalakrishnan 1997), mtDNA RFLP (Padhi and Mandal 1995) and *Mbo*I satellites (Padhi et al. 1998). Reddy (1999) summarized much of this information. Padhi and Mandal (1997) detected inadvertent hybridization resulting from multispecies spawning in a major carp hatchery by probing *Eco* RI digested DNA with a ribosomal RNA gene probe. Barman et al. (2003) reported species-specific RAPD DNA markers for four major carp species.

In contrast, there have been few applications of such techniques to the study of intraspecific genetic variation in wild or captive cyprinid populations in South Asia. Polymorphism for several enzyme loci has been characterized in *L. rohita* (NBFGR 1999) and an ongoing study is characterizing eight riverine populations of this species. Naish et al. (1997) reported an ongoing study on wild and hatchery populations of *C. catla*, using tetranucleotide microsatellites to examine both the genetic structure of the wild populations and comparative levels of genetic variation in wild and hatchery populations. Basavaraju et al. (2000) and Mair et al. (2000) described preliminary results from this study and a parallel one using PCR-RFLP analysis of mtDNA on the same groups of fish (see 4.5.1). NBFGR (1999) reported that the primers for one of the microsatellite loci developed for *C. catla* (G1: Naish and Skibinski 1998) also amplified a polymorphic locus in *L. rohita*, with four alleles being found in three wild populations. McConnell et al. (2001a) described five more polymorphic microsatellite loci from *C. catla*. A number of microsatellite sequences from *L. rohita* have been lodged in GenBank (http://www.ncbi.nlm.nih.gov/Genbank index.html).

Given the multiple threats to wild cyprinid populations in many parts of Asia including the observed declines in the populations of several species (e.g. Tsai et al. 1978; Dehadrai et al. 1994; Sreenivasan 1995; Lu et al. 1997), further genetic studies to help understand population structure, genetic diversity, etc., should have priority status.

Relatively few DNA sequences have been entered into the GenBank database for most of the cyprinids being considered in this publication (see list of species in Appendix 1). While *C. carpio* and *C. idella* had 11 140 and 591 nucleotide sequences respectively in the GenBank database in January 2004, none of the other species had more than 50 entries each, with most having very few. This can be contrasted to 584 674 entries for *D. rerio*, one of the major model species for genome analysis. While the zebra danio is the only cyprinid to have had the full capacity of modern genomics techniques applied to the analysis of its genome (e.g. Postlethwait et al. 1999; Kelly et al. 2000; Clark 2003), some of these resources have also been developed for *C. carpio*, including microsatellite markers (Crooijmans et al. 1997), a bacterial artificial chromosome library with two-fold coverage of the haploid genome (Katagiri et al. 2001) and an extensive range of ESTs and other DNA sequences (GenBank). It should be noted that the number of entries for *C. carpio* and *C. idella* has increased remarkably in approximately two years, reflecting the potential of genomics research to produce large numbers of microsatellite markers, expressed sequence tags (ESTs), etc., in a short time.

Microsatellites and other markers are already being applied to population genetics studies and for family identification in aquaculture genetics research, and in the longer term marker-assisted selection (using genetic markers closely linked to quantitative trait loci) is expected to improve the efficiency of selective breeding for at least some traits. Sun and Liang (2000) described a genetic linkage map that has been constructed for *C. carpio* and *Cyprinus pellegrini*, using microsatellites developed from *C. carpio*, crucian carp (*Carassius carassius*) and *D. rerio*, as well as RAPDs and other DNA markers. This map spanned 5 789 cM in 50 linkage groups.

4.4 Hybridization

Interspecific hybridization has been undertaken in a number of cyprinid species in many countries with the objective of manipulating sex ratio, producing sterile animals, improving growth and a few other traits. Although the desired or expected outcome of interspecific hybridization is generally an organism carrying one haploid set of chromosomes from each parental species, Chevassus (1983) described several different possible outcomes. These include diploid, triploid or tetraploid hybrids, haploid or diploid gynogenetics and haploid or diploid androgenetics. Several such "unexpected" progeny types have been observed from cyprinid hybridizations (Krasznai 1987; Wu 1990). For example, Stanley and Jones (1976) observed gynogenetic and androgenetic progeny as well as diploid hybrids in a *C. carpio* x *C. idella* cross. Thus it is accurate to describe the process as "heterospecific insemination" (Chevassus 1983), although there might be some uncertainty about the outcome of this process in terms of the genome of the progeny. Such possibilities emphasize the value of verifying the genetic nature of progeny from heterospecific inseminations, using for example karyotyping, allozymes or DNA markers in addition to morphological or morphometric analysis.

Wu (1990) reported that during 1960-90, over 100 combinations of "distant hybridizations" were carried out in China, including crosses between families, sub-families, genera and species (it was not explicitly stated that all of these hybridizations were between cyprinid species, but only one non-cyprinid species was referred to in the text and reference list of this review). In general, more distant combinations of parental species gave less viable hybrid offspring – no hybrids between different families survived to the hatching stage; hybrids between sub-families gave some viable, sterile hybrids with very high rates of abnormalities, while five hybrids between different genera gave viable, fertile hybrid offspring. One of these intergeneric hybrids, *C. carpio* (Jian variety) x *C. carassius*, was evaluated for aquaculture in China (Sun et al. 1994) and considered to be promising. Liu et al. (2001) found that F_2 hybrids of red *C. carassius* x *C. carpio* produced diploid eggs and diploid sperm, resulting in tetraploidy in the F_3 and subsequent generations. These tetraploid hybrids were fertile, and gave rise to sterile triploid hybrids in back-crosses to either parent species. These have been produced on a large scale in China (Liu et al. 2001). This appears to be the first case of artificial synthesis of a bisexual hybrid tetraploid form in fish (Gomelsky 2003).

Research has also been carried out on hybridization between Chinese carp species elsewhere in the world, principally in India, Eastern Europe and the USA. Marian et al. (1986) described characteristic karyological, biochemical and morphological markers of *H. molitrix*, *A. nobilis* and their hybrid. Issa et al. (1986) described better growth and survival of the reciprocal hybrids over the parental species during the fingerling period, and Kammerad (1991) stated that the female *H. molitrix* x male *A. nobilis* showed faster growth and better survival than *H. molitrix* during three years of culture.

C. idella x *A. nobilis* hybrids were produced in the USA as a potential way to obtain sterile fish for aquatic weed control, in an attempt to mitigate the controversy over the use of *C. idella* in the country (Sutton et al. 1981). Induced triploidy and monosex female production were also investigated in *C. idella* for the same reason (Shelton 1987; Thomas 1994). The *C. idella* x *A. nobilis* hybrid progeny were a mixture of diploids and triploids (Allen and Stanley 1983; Beck and Biggers 1983a and b; Beck et al. 1984). The hybrids do not appear to be used at present, although triploid *C. idella* are (e.g. Thomas 1994).

Kowtal (1987) and Reddy (1999) reviewed the hybridizations of cyprinid species carried out in India. Research on cyprinid hybridization began in India shortly after the earliest successful induced breeding through hypophysation in 1957 (Kowtal 1987). More than 40 hybrids have been produced. These include hybrids between species within the genus *Labeo*, hybrids between species in different genera of Indian carps (*Catla*, *Cirrhinus* and *Labeo*), hybrids between *C. carpio* and Indian major carps, hybrids between the Indian and Chinese carps, hybrids between *C. carpio* and Chinese carps, and hybrids between the Chinese carp species.

Almost all of the hybrids produced among Indian major carps exhibited intermediate traits of their parental species (Reddy 1999). The majority of these hybrids were also fertile. Langer et al. (1991) produced an F_2 generation of *C. catla* x *L. rohita* hybrids. Khan et al. (1990) and Reddy et al. (1990) described how three *C. carpio* x Indian major carps (*C. catla*, *L. rohita* or *C. cirrhosus*) hybrids were sterile, apparently due to aneuploidy (74–76 chromosomes) resulting from having one ancestrally tetraploid parent (*C. carpio*, 2n = 100 chromosomes) and one diploid parent (Indian major carp, 2n = 50 chromosomes). However, Khuda-Bukhsh et al. (1988) reported that *C. carpio* x *L. calbasu* progeny were gynogenetic in origin, following analysis of the karyotype and electrophoresis of haemoglobin and transferrin.

Some of these hybrids have been reported to have some potential for aquaculture and reservoir stocking, for example the *C. catla* x *L. rohita* hybrid for reservoir stocking (Natarajan et al. 1976; Somalingam et al. 1990); *C. carpio* x Indian major carp hybrids for aquaculture due to their high growth rate and flesh content, low seine net escapability and sterility (Khan et al. 1990); and *C. catla* x *L. fimbriatus* hybrids for higher dress out percentage due to smaller head and growth nearly equivalent to that of *C. catla* (Basavaraju et al. 1995). However, to date there has been little actual use of these hybrids.

A novel form of hybridization, "nucleocytoplasmic hybridization" was developed in fish species in China by microinjecting the nucleus of one species into the egg of another (Tung et al. 1973). Chen et al. (1990) took this a stage further by using electrofusion instead of microinjection to combine the nucleus and eggs of different cyprinid species. While the technique also has the potential to be used to produce clonal lines of animals (in a cytoplasmic environment which could be conspecific or heterospecific), to date it has mostly been used to study interactions between the nucleus and cytoplasm of different species, mostly cyprinids.

Despite the large body of research on hybridization among cyprinid species, there is little evidence of significant production of any of these hybrids in Asia except for *C. carpio* hybrids in China (Bartley et al. 1997, 2000; FAO 1999), although Bartley et al. (1997) do point out that hybrid production in general may be underreported in FAO aquaculture statistics.

4.5 Quantitative genetics

Kirpichnikov (1981) stated that "selection should begin simultaneously with the domestication of new species of freshwater fishes; the delay with selective breeding may result in the decreased diversity of the genetic structure of the cultivated species or variety and may even lead to rapid degeneration." In a newly domesticated species or population (i.e. when a captive breeding population is initiated from a wild one, and maintained over generations), some genetic changes will take place without any active intervention by the hatchery managers, as a result of the new environment and the changed selection pressures it places on the population. A lack of application of genetic principles, or misguided attempts to select for improved performance, may result in deterioration of the performance of the stock. Closely linked to this, and also frequently caused by poor management, is inbreeding.

Conversely, the application of genetic principles (in the context of the biology of the species and hatchery practices) should be able to prevent or minimize inbreeding and improve the performance of the population.

4.5.1 Hatchery management: Inbreeding and inadvertent selection

Cyprinids can be particularly prone to the negative effects of poor genetic management. High fecundity means that a new generation of brood stock can be produced from very few parents in most species. Long generation times in most cultured cyprinids can have both positive and negative effects. Long generation times should slow down the rate of inbreeding per year, but mixing fish from different classes in the same pond for future replacement brood stock and selecting later within such a group, may narrow the genetic base. It is also common practice in some hatcheries to retain fish for future brood stock from fry or fingerlings remaining after the bulk have been sold off. These are quite likely to have been subjected to some form of selection (e.g. size, net escapability) that may affect performance traits. Similar practices may happen in combined hatchery/grow-out farms, where larger fish are sold for consumption and smaller fish retained for brood stock replacement (Hussain and Mazid 1999).

Negative effects of inbreeding on traits of importance to aquaculture have been demonstrated in several species of fish, including cyprinids. Kirpichnikov (1981), summarizing some of his earlier work, concluded that even moderate levels of inbreeding lead to inbreeding depression in *C. carpio*, with a reduced growth rate and survival and the appearance of "phenodeviants". The same author also quoted work by Moav and Wohlfarth (1968) and Wohlfarth and Moav (1971), who concluded that one inbreeding generation after the crossing of sibs retarded the growth of *C. carpio* by 10-20 per cent, and was accompanied by decreasing viability and a marked increase in the number of malformations. Kincaid (1983) studied the effects of inbreeding on a variety of traits in rainbow trout (*Oncorhynchus mykiss*) by setting up a series of planned crosses that would give known levels of inbreeding (25 or 50 per cent) relative to an outbred control. For nearly every trait a significant reduction in performance was observed under both hatchery and field conditions.

While these negative effects can be demonstrated in planned inbreeding experiments, and practices that are likely to have negative genetic effects on performance can be identified, it is generally difficult to quantify the degree of inbreeding or negative selection and the effects of such practices in existing stocks of fish in hatcheries and farms. There are a number of possible reasons for this:

- Poor or non-existent records in the hatchery of the relevant data, e.g. number of spawners actually contributing to brood stock replacement (this may be lower than the total number of spawners used in breeding), age at first spawning, age at removal from brood stock, etc.

Eknath and Doyle (1990) managed to estimate N_e (effective population size) and ΔF (rate of inbreeding) for major carp populations in hatcheries in Karnataka, India but few others have managed to obtain adequate data to estimate inbreeding levels in this way.

- Molecular genetics techniques (e.g. microsatellite DNA loci, mitochondrial DNA, allozymes) have been used to estimate levels of genetic variation in hatchery populations (e.g. Allendorf and Ryman 1987; Billington and Hebert 1991; Ferguson 1995), but without access to the source population in its original state (or to samples from earlier generations in the same hatchery) these data may be of limited usefulness.

If samples were taken from the hatchery population at the time of its founding or if the hatchery population was taken from a single wild population that is still extant and presumably unaltered, then assessment of any genetic changes should be relatively straightforward. For example, Fiumera et al. (2000) used microsatellite loci to study a captive breeding program for *ex situ* conservation of a Lake Victoria cichlid, *Prognathochromis perrieri*, over several generations, and concluded that the effective population sizes in the subpopulations held in different institutes were 2.5-7.7, far below the actual number of adults maintained in each generation (mean 32-243). However, such closely-monitored situations are likely to be rare in the context of most Asian cyprinid hatcheries. More frequently, the source population will be unknown or unavailable. The hatchery may not operate as an entirely closed unit or the hatchery population may have been founded from more than one source.

Reed and Frankham (2001) surveyed studies on populations from a wide variety of species, and concluded that there was no consistent relationship between molecular genetic variation and quantitative genetic variation. This suggests that, in general, the results of molecular genetic studies have to be used with caution when making inferences about variation for quantitative genetics parameters, and that where possible quantitative genetic variation should be estimated directly, e.g. by estimating heritability for traits of interest.

- Perceived deteriorations in the performance of aquaculture stocks over time have led to suggestions of genetic deterioration (e.g. Hussain and Mazid 1999). However, accurate documentation of such changes is seldom available.

Even if reductions in performance can be verified, it is very difficult to separate genetic changes from environmental factors that may have contributed to declining performance (e.g. changes in culture practice or increasing water pollution). There is a parallel between this and the measurement of genetic improvement through deliberate selection (see below), where genetic changes need to be separated from improvements in culture practices and other environmental factors. Genetic controls are used in selective breeding and could be used to estimate the degree of genetic change where inbreeding or negative selection has been suggested, e.g. fish taken from the source population and grown side-by-side with the tested stock, or cryopreserved sperm from the base hatchery population. Again, however, these are not often available.

Eknath and Doyle (1985 a, b) concluded that negative selection was taking place in *L. rohita* and *C. cirrhosus* hatchery populations in Karnataka, India, in that selection of larger brood stock fish for spawning actually selected slower growing, late maturing fish.

- In unmonitored hatchery populations, complex genetic changes may take place. For example, "green-fingered" selection may be having some positive effect but may be counteracted by inbreeding.

It is evident in the case of *C. carpio* that "unscientific" selection over many generations of domestication has led to genetic changes and to a wide variety of different body and fin shapes, growth rates, scale patterns and colors. Unintentional selection during domestication will also result in adaptation of wild populations to the hatchery environment. If these are operating at the same time as management practices that result in inbreeding, the net changes in the population are likely to be complex and hard to evaluate.

4.5.2 Current status of domestication and strain development

Distinct domesticated strains have only really been developed in *C. carpio*. From its natural origins in Central Asia, it spread first naturally into Asia and Europe and then more recently has been introduced to almost every country in the world with suitable habitat and climatic conditions (FAO 1998). During its long history of domestication (Balon 1995; Hulata 1995) in several areas, stocks and strains have been developed with a variety of characteristics. These include very obvious differences in scale pattern, coloration and body shape, as well as variation in growth rate and other quantitative traits. The most striking morphological variation is observed among ornamental strains (koi). In many cases, the origins and development of such stocks and strains cannot be traced because records were not kept and the selection applied to develop such varieties was not scientifically applied or monitored over generations. Most countries in which aquaculture of *C. carpio* is important have stocks with genetic differences for traits of economic importance, good examples in Asia are in China, Indonesia and Vietnam (Li and Wang 2001; Sumantadinata 1995; Thien and Trong 1995). These have come from a mixture of importations and local differentiation.

Because of the long history of domestication and transfers, it has become very difficult to identify and study wild populations of *C. carpio*, or to be sure of the geographical origin of some captive populations. Brody et al. (1979) studied enzyme and transferrin loci in two domestic populations of European origin and two of Chinese origin. Differences in allele frequencies were found between the European and Chinese populations at most polymorphic loci. Kohlmann and Kersten (1999) studied enzyme polymorphism in sixteen populations of *C. carpio* in Germany. Four of these were of Asian origin (the Amur River in Russia; the Red River in Vietnam; Koi from Japan; Ropsha selected strain bred in Russia from wild Amur River carp and local domesticated carp). There was a clear differentiation between the populations of Asian and European origin. The authors concluded that these two clusters corresponded to the two still existing subspecies *C. carpio haematopterus* (East Asia) and *C. carpio carpio* (Europe). However, all of the countries that are the focus of this publication have imported *C. carpio* of European origin and used these in the development of commonly used aquaculture stocks, so even in the Asian countries such as China and Vietnam with indigenous *C. carpio* (Froese and Pauly 2001; Trong 1995), it is likely that some introgression has taken place between European and Asian genotypes. In the Asian countries with no indigenous *C. carpio*, it is likely that current stocks are predominantly or entirely of European origin, on the basis of the known history of introductions (see Chapter 6).

4.5.3 Stock/strain comparisons and cross-breeding

As *C. carpio* is the only cyprinid species in which distinct stocks/strains have been developed, it follows that with few exceptions stock/strain comparisons, including studies on heterosis and genotype-environment interaction, have been conducted on this species. A few studies have been conducted on other cyprinid species to compare wild populations from different sources. Comparison between existing populations, stocks or strains can be an important initial part of developing a breeding program, so this is dealt with here, before selective breeding (see 4.5.4).

Israeli scientists conducted a series of experiments comparing different strains of *C. carpio* and their cross-breds, much of which focussed on comparisons of Chinese and European carp. Moav et al. (1975) compared three European stocks (Našice, with a very high height:length ratio; Gold, an inbred line with gold body coloration; and Dor-70, the product of a selection experiment in Israel), one stock of Chinese origin (Big-Belly) and eight cross-breds (mostly from crosses between these four stocks), in five different environmental conditions. The average growth rate of the Big-Belly carp across all five environments was much poorer than that of the European carp, while all of the cross-breds showed heterosis. When the genotype-environment interaction of the four purebred groups was represented as a linear function of the environment (i.e. the mean weight of the group as a function of the overall mean weight of all of the fish), the European carp showed much higher responsiveness to changes in the environment. The cross-breds showed an intermediate but complex responsiveness with a high degree of heterosis. Našice and Gold showed evidence of high levels of inbreeding, reflected in the superior performance of the Našice x Gold cross-bred over the parental strains, while Dor-70 did not show any evidence of inbreeding depression despite some inbreeding over the previous five generations.

Wohlfarth et al. (1983) conducted a similar study with less groups and a more refined experimental design to minimize the effects of inbreeding on the performance of the tested groups. All three groups of fish in the experiment were cross-breds - Chinese origin (Taiwanese x Hong Kong), European origin (Dor-70 x Našice) and Chinese x European (Taiwanese x Dor-70). They were communally stocked into a variety of environments. In environments with a mean overall growth rate of >5 g.day^{-1}, the European cross-bred were the fastest growing of the three groups, while in environments with overall mean growth rates of 2-5 g.day^{-1}, the Chinese x European cross was the fastest growing. The Chinese cross-bred grew faster than the European cross-bred in the poorest environment (which had an overall mean growth rate of approximately 2 g.day^{-1}) and extrapolation of the results suggested that the Chinese cross-bred would also have grown faster than the Chinese x European group in environments with an overall mean growth rate of <1 g day^{-1}. Wohlfarth et al. (1986) compared similar groups of fish in ponds with different inputs (manure and/or supplementary feeds), and observed that the Chinese strain showed the fastest relative growth in poor conditions, while the European strain showed the fastest growth under improved conditions. These results suggested that the European and Chinese *C. carpio* strains are each adapted to the typical culture environments in which they were developed, that genotype-environment interac-tions can be very important and that heterosis for growth can also be strong.

Wohlfarth et al. (1975) looked at a range of other traits in the same groups of carp studied by Moav et al. (1975). They concluded that sexual dimorphism for weight was greater in the Big-Belly than the European strain (cross-breds were intermediate, suggesting additive inheritance for this trait), viability under poor conditions was higher in the Big-Belly strain (mortality rates were intermediate in the Big-Belly x European crosses but heterosis in the direction of lower mortality was observed in the European crosses, suggesting inbreeding in the latter) and the Big-Belly had much higher seine net escapability (Big-Belly x European crosses showed slight dominance in the direction of higher seine net escapability).

Hulata et al. (1980) found higher percentages of gonad abnormalities in F_1 Chinese x European *C. carpio* and in back-crosses between the F_1 and either parent. It was suggested that this was evidence of a partial reproductive barrier between the two geographic groups.

Evaluation of three strains of *C. carpio* in Thailand (local, Indonesian and fifth generation selected Vietnamese strain) and their cross-breds indicated that the local strain grew faster than the Vietnamese strain, but survival was lower. The Indonesian strain grew faster than the local strain, but with lower survival. The Indonesian strain had significantly faster growth among the nine groups (six cross-breds and three pure strains). No crosses performed better than the parent pure strains for growth. Hence it was planned to undertake combined family and mass selection for growth (Anon. 2001).

Wohlfarth (1993) reviewed heterosis for growth rate in *C. carpio*. He concluded that heterosis for growth in this species is a common but not universal phenomenon, limited by genetic factors, genotype-environment interactions, and age and weight of fish. It appeared that the cross-bred advantage was limited to relatively young and small fish and he suggested that this could be of particular importance in warm climates where *C. carpio* could reach market size during their first year. It is worth noting that Flajšhans and Gall (1995) stated that 80 per cent of Hungarian and almost 100 per cent of Israeli *C. carpio* production was based on cross-breds and that they are also used commercially in the former Soviet Union (Hulata 1995). Linhart et al. (2002) stated that practically all *C. carpio* stocked in commercial ponds in the Bohemia area of the Czech Republic are F_1 cross-breds between two distant strains. Bakos and Gorda (1995) gave more details on the cross-breeding program for *C. carpio* in Hungary, including attempts to increase the heterosis effect by reproducing the parental lines by gynogenesis. One gynogenetic hybrid out of 21 tested resulted in a 10 per cent higher growth rate. No positive heterosis on the survival rate was observed in the gynogenetic hybrids. Wang and Xia (2002) suggested that heterosis should be higher in crosses involving more distantly related parent stocks: however, this study was based on very few crosses.

Sixteen crosses among four strains of *C. carpio* (Rajadanu, Wildan Cianjur, Majalaya and Sutisna Kuningan) were evaluated in Indonesia for developing a synthetic base population for selective breeding. Heterosis for growth ranged from 0.2 - 4.9 per cent and for survival from 1.6 - 4.7 per cent (Anon. 2001).

A number of intraspecific hybrids (cross-breds) of *C. carpio* have been produced and are being widely used in aquaculture in China. These include not only F_1 cross-breds between two parental strains, but also three-way crosses and populations derived from further generations of breeding from initial cross-breds (Anon. 2001, Li and Wang 2001; Zhu et al. Chapter 3.2, this vol.). After 587 days of monoculture, Jianhuang carp showed 12 and 32 per cent higher growth than female and male parents, respectively. Jianhe carp showed 21 per cent higher growth as compared to the male parent and 8 per cent lower than the female parent (Anon. 2001).

Gela et al. (2003) found evidence for significant genotype x environment interaction for several traits when four *C. carpio* crosses were compared under low (350 m above sea level) and high (750 m ASL) altitude conditions in Central Europe. A Hungarian strain (HSM) was used as a common maternal parent, crossed to four paternal strains (HSM, wild Amur carp, Ropsha carp and Tata carp) to produce the four evaluated crosses. The traits showing significant strain x altitude interaction included the total weight, fillet weight, index of head length and gonadosomatic index.

Li et al. (1987) studied the growth rates of wild and hatchery populations of *H. molitrix* and *A. nobilis* from the Changjiang (Yangtze) and Zhujiang rivers in China. Although the *H. molitrix* from the Zhujiang River grew faster than those from the Changjiang River at the fingerling stages, the Changjiang River fish grew faster in trials in the second year of life and onwards. The wild stocks outgrew the hatchery stocks in both cases. *A. nobilis* from the Changjiang River grew faster than those from the Zhujiang River. However, they suggested that poor nutrition and environmental conditions for captive brood stock could reduce the performance of their offspring compared to offspring from wild brood stock. This emphasizes that when comparing stocks of different origins, ideally the brood stock as well as the tested offspring should be held under identical environmental conditions to produce reliable estimates of genetic differences in the analyzed traits. Li et al. (1987) noted that fry from the Zhujiang River were distributed more widely due to the earlier spawning in the more southerly Zhujiang River region, but suggested that the Changjiang River fish were more suitable for culture. Yang and Li (1996) also noted faster growth rates of wild Yangtze River *H. molitrix* and *C. idella* compared to a hatchery population (three generations from the wild), although the differences were not significant. Li et al. (1987) opined that stocks of slow-growing *H. molitrix* introduced into Southeast Asian countries probably originated from the Zhu River system and suggested replacement with stocks from the Yangtze River.

Reddy et al. (2002) compared five riverine stocks and one farmed stock of *L. rohita* (Ganga, Yamuna, Gomati, Brahmaputra, Sutlej and CIFA farm stock) in earthen ponds in Orissa, India. These were grown communally either in monoculture or polyculture with *C. catla* and *C. cirrhosus.* The final body weight and survival rates of the wild stocks were equal to or better than the farmed one, while large differences were found between half-sib and full-sib groups within the stocks. Ranking of groups between monoculture and polyculture was highly consistent, indicating a lack of genotype x environment interaction in this study. Gjerde et al. (2002) studied heterosis in two 3 x 3 diallel crosses involving the same stocks of *L. rohita.* They concluded that heterosis for harvest weight was low or negative and heterosis for survival was negligible, indicating little value in using cross-breeding for genetic improvement in these rohu stocks.

Comparison of three populations of *B. gonionotus* (introduced Thai and Indonesian strains and a "local" population of Thai origin) and six cross-breds between these populations in Bangladesh did not show any significant differences in growth apart from one cross-bred that was significantly smaller at harvest than the other eight groups (Hussain et al. 2002). Evaluation of six indigenous populations of *B. gonionotus* in southern Vietnam indicated genotype x environment interaction (Anon. 2001).

Vandeputte et al. (2002) used a hemi-isogenic scaly *C. carpio* as an internal control in a comparison of the performance of three French mirror carp strains. The tested strains were grown separately from each other, in replicated ponds, and fish from the control strain were added to each pond. Although no significant differences were detected between the mirror carp strains (possibly because of a lack of genetic differentiation between these strains), the method did allow for an assessment of traits at the early stages (the first trait to be studied was at five weeks post-resorption) and had a greater power to detect any differences than a comparable experiment using replicated separate ponds for each stock without an internal control. In the latter design, environmental variation between "replicate" ponds reduces the ability of such a study to detect genetic differences between strains.

In summary, differences in performance between populations and strains of the same species have been clearly demonstrated in some cases, as have heterosis and genotype-environment interactions. However, it is hard to draw general conclusions about the prevalence of the latter two. Additionally, there may be significant amounts of additive genetic variance within populations as well as differences between populations. In developing a breeding program where two or more different populations are initially available, it is probably best to start by comparing the populations (and if possible families within populations) and their F_1 cross-breds in the relevant range of culture systems. F_1 cross-breds have been extensively used for commercial production in *C. carpio* in China, Israel and some European countries, while crosses involving two or more different populations have been used as the basis for selective breeding programs in several species (see 4.5.4).

4.5.4 Selective breeding

Selective breeding has been the basis of nearly all of the genetic improvement that has taken place in terrestrial agricultural animals and plants since the earliest stages of domestication. This genetic improvement has been the result of centuries of on-farm breeding and the recent application of modern breeding and selection theory: it is estimated that selection has increased the productivity of modern breeds of farm animals by at least two to three times in the last 50 years (Bentsen and Olesen 2002). In contrast, Gjedrem (1997) estimated that less than 1 per cent of aquaculture production worldwide in 1993 came from genetically improved breeds.

Although aquaculture organisms differ from agricultural mammals and birds in several important ways (e.g. higher fecundity and smaller post-embryonic size), the principles of selective breeding can also be applied to their genetic improvement.

Some of the most important points in applying quantitative genetics theory to the selective breeding of fish are:

- The nature of the base population in which the selection is started: this should be made up from the best performing wild populations and/or captive stocks available and should contain good levels of genetic variation.

- The traits that are to be selected should be clearly defined, if necessary prioritized (e.g. on the basis of economic importance) and heritability values estimated in the base population. It is very hard to give "typical" heritability values for a trait or a species, since these will depend on the population and the environment in which the estimation is made. Fig. 4.2 shows a frequency distribution of heritability values for traits related to aquaculture production from a range of fish species, including *C. carpio*.

- Conceptually, the simplest form of selection is what is known as "mass selection" (sometimes also called "individual selection"). This can often be very effective, but the lack of knowledge of family structure can lead to problems, e.g. inbreeding if individuals from only a few families are (inadvertently) being selected in each generation. Bentsen and Olesen (2002) describe ways of designing mass selection programs while avoiding high inbreeding rates. Within-family selection and between-family selection can be used, or combined selection, which should be the most efficient genetically. Falconer and Mackay (1996) described these in some detail, including some of the factors that may influence the choice of selection method.

- Cross-breeding (see 4.5.3) is sometimes a valuable alternative to selective breeding (e.g. if heritability values are low but heterosis is high) or an additional technique (if selected lines show heterosis in crosses).

Five generations of mass selection for a faster growth rate (size at seven months in pond culture) in *C. carpio* in Israel did not yield any response, while selection for slower growth yielded a relatively strong response for the first three generations (Moav and Wohlfarth 1976). The authors suggested that this stock of *C. carpio* ("the local strain of the domesticated European carp") had already reached a selection plateau for fast growth rate (and thus had diminished additive genetic variance for growth), due to a strong correlation between growth rate and reproductive fitness. They also suggested that the response to selection for slower growth demonstrated residual, mostly non-additive, genetic variance for growth rate, with a strong dominance component for fast growth. Inbreeding may have been a complicating factor in this study. Despite the lack of improvement through mass selection, subsequent between-group selection from within the "high" line

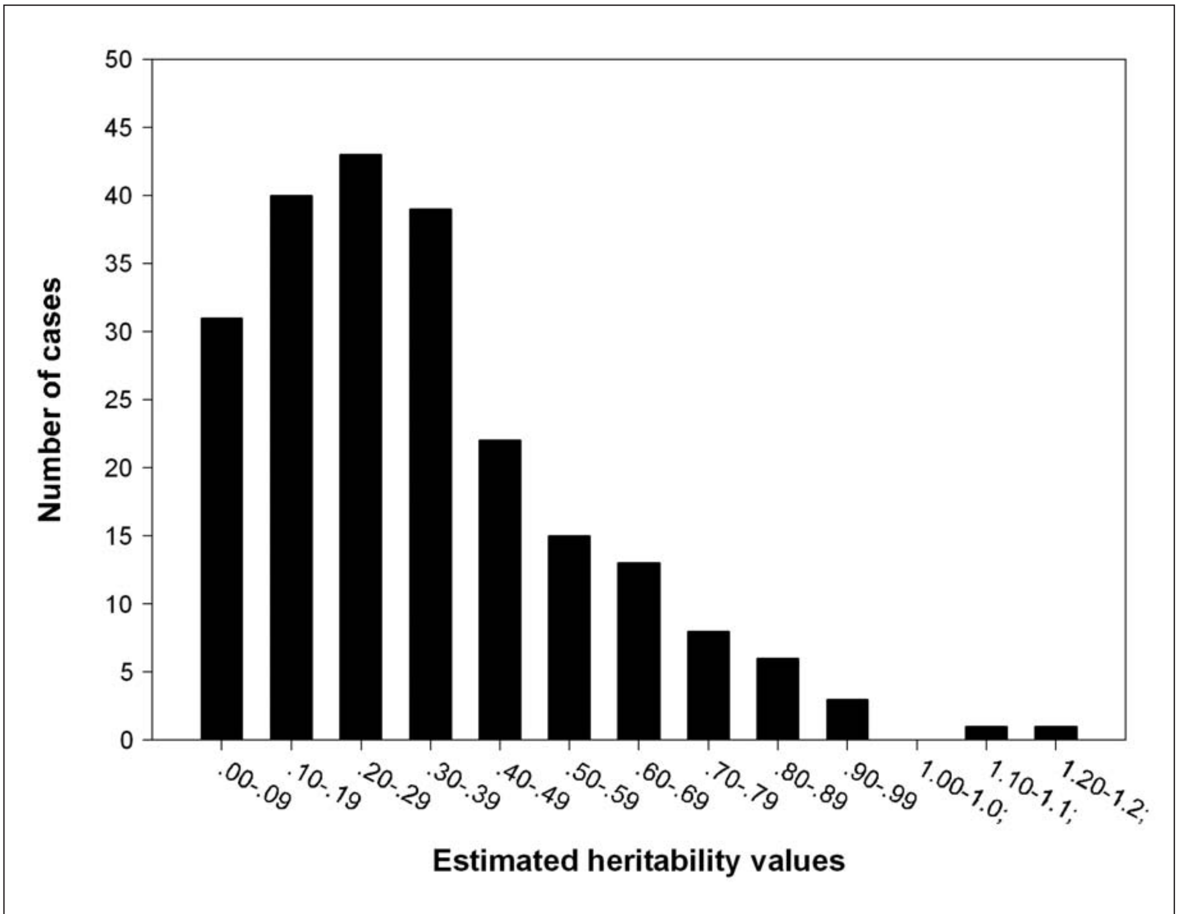

Fig. 4.2. Frequency distribution of heritability values for a range of production-related traits in *C. carpio, Oreochromis niloticus*, *Salmo salar, Oncorhynchus mykiss* and *Ictalurus punctatus* (data from Table 4.1 of Tave 1993)

resulted in the fast-growing Dor-70 line, which also produced high-yielding cross-breds (Moav and Wohlfarth 1976) that became the basis of commercial carp production in Israel (Wohlfarth 1993). Although some inbreeding must have accompanied the selection that resulted in the Dor-70 line, cross-breds involving Dor-70 as a parental line rarely exceeded the growth of Dor-70 by a meaningful amount (Wohlfarth 1993).

Eknath (1990) suggested that the lack of response to mass selection for growth rate in the experiments of Moav and Wohlfarth (1976) impeded the adoption of the selection approach to carp genetic improvement for a long time despite promising results obtained from other species.

Ankorion et al. (1992) estimated realized heritabilities for body shape (height/length ratio) in *C. carpio* through one generation of mass selection. Both up and down selection showed strong responses (heritability estimates were 0.47±0.06 and 0.33±0.10, respectively). The authors considered that the heritability estimates might have been inflated due to the structure of the base population (which was composed of two cross-breds, with a common female parent and two different male parents). There was no significant phenotypic correlation between body shape and growth rate.

Other selection programs on *C. carpio* have emphasized the importance of starting with a genetically diverse base population. In these examples, two or more different stocks have been used, sometimes with the aim of selecting more than one trait for improvement. Kirpichnikov (1972, 1981) and Babouchkine (1987) summarized the development of the Ropsha *C. carpio* by selecting from hybrids of the Galician European carp and the Amur wild carp. After producing groups of cross-breds and back-crosses in the initial generations, mass selection was used within three different lines. The Ropsha carp possesses good resistance to low temperature, high general viability and rapid growth in the first and second years and is well adapted to cultivation in the northern and northwestern regions of the former USSR. Kirpichnikov et al. (1993) described the development of the Krasnodar *C. carpio*, which was selected for resistance to dropsy and also for high growth rate. The base populations for this program were a local mirror carp stock (L), the Ropsha carp (R) and a Ukrainian-Ropsha cross (UR). In initial tests, the Ropsha carp were the most resistant to dropsy, while after five, six and seven generations of mass selection the UR fish were the most resistant. The effectiveness of selection in improving dropsy resistance in the R and L stocks was questionable, but UR x R and UR x L cross-breds also showed higher resistance and UR x L cross-breds showed some heterosis for embryonic survival and growth in two-year-old fish. Preliminary experiments on the L stocks after several generations of mass selection indicated that family selection could be successfully used to improve dropsy resistance.

Many strains/varieties of *C. carpio*, a principal aquaculture species in China with a production of 2.05 million tonnes in 1999 (20 per cent of total freshwater fish production: Li and Wang 2001) have been developed during its long history of culture. The Jian carp was developed in China through a six-generation combined breeding program involving family selection, inter-line crossing and gynogenesis (Zhang and Sun 1994). It has been described as the first "artificial" breed or variety of *C. carpio* in China (Doyle et al. 1994; Sun et al. 1995). Improvement of the growth rate in pond culture over several generations has been demonstrated (Zhang et al. 1995), although a highly significant genotype x environment interaction was observed when growth was compared in ponds and cages. The Jian carp were shown to grow faster than several other varieties of *C. carpio* in China (Sun et al. 1995), and is widely cultured in over 660 000 ha of ponds in China, with an estimated annual production of over 1 million tonnes (Anon. 2001). Further selection of Jian carp for one more generation during 1999-2000 resulted in increased growth by 6 per cent and 9 per cent in mono and polyculture (Anon. 2001).

Selective breeding studies are in progress in China to produce: (i) stable (color) fish with improved growth; and (ii) ornamental fish with different pigmentation patterns in Oujiang common carp. The results to date indicate the existence of strong genetic inheritence of pigmentation patterns (Li and Wang 2001).

From eight stocks of *C. carpio* in Vietnam, double hybrids between the Vietnamese white carp, Hungarian scaled carp and Indonesian yellow carp were used as the starting material for a selective breeding program (Thien et al. 1987; Thien and Trong 1995). Six generations of mass selection were carried out in three hybrid lines (Thien 1997). Thien (1997) reported that the realized heritability in one of these lines in the early generations was 0.20-0.29. After five generations of mass selection, the growth rate had increased by 33 per cent compared to the base population. However, realized heritability decreased to nearly zero by the sixth generation. To overcome this, family selection has been initiated (Thien et al. 2001).

In cyprinid species other than *C. carpio*, selective breeding has generally been started from base populations entirely or partially composed of fish taken directly from the wild, which presumably ensures relatively high levels of genetic variation in most cases. Two generations of selective breeding of *L. rohita* for faster growth, from a base population composed of five wild riverine stocks and one farm stock (see 4.5.3), resulted in 35-40 per cent faster growth in field trials undertaken in different parts of India (Reddy, chapter 3.3, this vol.; Anon. 2002).

Additive gain in terms of growth from two generations of mass selection of *B. gonionotus* in Bangladesh from a synthetic base population formed from three strains (local, Thai and Indonesian) was estimated at a mean of 7.2 per cent per generation, while the synthetic base population grew 7.5 per cent faster than the nonselected local stock, giving a total gain of 21.9 per cent (Hussain, chapter 3.1, this vol.; Hussain et al. 2002). Mass selection of *B. gonionotus* undertaken in Thailand for two lines, the Chao Phraya and Mekong River, resulted in 30 per cent higher growth in the Chao Phraya line with a realized heritability of 0.4 for body weight at seven months (Anon. 2001).

Selective breeding of blunt snout bream (*Megalobrama amblycepahala*) in China started in 1986 through the establishment of two selected lines, a control group and an inbred line (Li and Cai 2003). Five generations of mass selection resulted in an average improvement in growth of 4.0 and 5.8 per cent per generation in the two selected lines. Three generations of full-sib mating resulted in a decline in the growth rate of 17 per cent relative to the control line, i.e. an average of 5.7 per cent per generation. The reduction in growth rate through inbreeding was thus approximately equal to the gain from mass selection, which is a clear reminder of the potential damage that can be done by poor brood stock management.

In conclusion, it is apparent that there is much scope for the appliance of quantitative genetics to improve the performance of cultured carps. This may take the form of obtaining one or more stocks (from the wild or other domesticated stocks) on the basis of their performance and/or level of genetic variance, managing stocks to minimize inbreeding or avoid inadvertent selection, applying selective breeding to improve performance for important traits, or cross-breeding where heterosis is observed. Ideally, a breeding program should take all of these factors into consideration. In practice, limitations in financial, physical or human resources have often restricted the scale and complexity of breeding programs for carps in Asia. For example, selective breeding involving the identification of individual families is generally more efficient than mass selection, but requires additional resources for physical identification (separate rearing for each family followed by physical tagging or marking of each family before communal ongrowing) or identification using highly polymorphic microsatellite markers.

4.6 Chromosome set manipulations and polyploidy

Gynogenesis occurs naturally in the genus *Carassius*, with many different clonal lines existing, some of them triploid (e.g. Nakanishi and Onozato 1987; Zhu 1990; Cherfas et al. 1994; Dong and Taniguchi 1996; Umino et al. 1997; Zhou et al. 2000). Induced gynogenesis involves inactivating the paternal (sperm) genome and then restoring diploidy either through suppression of the second meiotic division ("meiotic" gynogenesis) or suppression of the first mitosis ("mitotic" gynogenesis). Most of the more recent protocols for gynogenesis have used ultraviolet irradiation to inactivate the paternal genome, and temperature (cold or heat) shocks to restore diploidy.

Diploid gynogenesis has been induced experimentally in a wide variety of cyprinid species, including *M. piceus* (Rothbard and Shelton 1993), *C. idella* (Stanley 1976; Xia et al. 1990), Indian major carps (John et al. 1984; reviewed by Hussain 1996 and Reddy 1999), *B. goniontus* (Siraj et al. 1993; Pongthana et al. 1995), *H. molitrix* (Mirza and Shelton 1988; Xia et al. 1990) and most notably in the *C. carpio* (Nagy et al. 1978; Linhart et al. 1986; Hollebecq et al. 1986; Komen et al. 1988; Komen 1990; Sumantadinata et al. 1990).

Androgenesis has been investigated less than gynogenesis. In general it appears to be more difficult to successfully produce diploid androgenetic fish than the equivalent mitotic gynogenetics, due to the difficulties of irradiating unfertilized eggs appropriately. However, one laboratory in the Netherlands has been highly successful in developing and using androgenesis in research on the *C. carpio* (Bongers et al. 1994, 1997a, b, c, 1999) and a few other laboratories have also carried out research on induced androgenesis in *C. carpio* (e.g. Grunina et al. 1995; Ponniah et al. 1995) and *C. carassius* (Fujikawa et al. 1993).

Gynogenesis and androgenesis have been used to investigate sex determination and to produce monosex female cyprinids (see 4.7), to study the effects of inbreeding (e.g. Komen et al. 1992c), to produce fully inbred clonal lines (Komen et al. 1991), to study gene-centromere recombination rates (reviewed by Komen 1990) and to study quantitative genetics parameters. Sumantadinata et al. (1990) studied variance in quantitative characters in gynogenetic *C. carpio* and were able to determine the genetic contribution to this. Bongers et al. (1997a and b) used androgenetic lines to investigate the genetic factors influencing gonad development in *C. carpio*. Bongers et al. (1998) reviewed the use of genetically uniform strains of *C. carpio* in experimental animal research.

While gynogenesis and androgenesis have made little direct impact on aquaculture apart from contributing to monosex female production and in some selective breeding programs, they provide interesting tools for basic research underpinning aquaculture genetics and are likely to make further contributions in the future.

Triploidy has been investigated in certain species of cyprinids, notably *C. carpio* (Gervai et al. 1980; Hollebecq et al. 1986; Cherfas et al. 1993, 1994; Reddy et al. 1998; Basavaraju et al. 2002) and *C. idella* (Cassani and Caton 1985; Thompson et al. 1987; Allen and Wattendorf 1987). In most of the studies on triploidy in *C. carpio* no clear advantages in terms of the growth rate have been demonstrated (Gervai et al. 1980; Cherfas et al. 1994, and papers cited therein). Triploidy could in theory be used to prevent ovary development and spawning, and increase growth rates, in early maturing strains, but the evidence for this is ambivalent. Wu (1990) gave brief information on two trials in which the *C. carpio* triploids were more than twice the size of diploids at five months old. Reddy et al. (1998) described a trial conducted in a single pond in which a batch of *C. carpio* originating from heat-shocked eggs and containing approximately 80 per cent triploids/ 20 per cent diploids were grown (without any separate, unshocked, diploid controls). Nine months after stocking, the triploids were 53 per cent larger than their diploid siblings (mean weights 59.46 g and 38.83 g, respectively). Basavaraju et al. (2002) described several trials comparing the growth rates and other parameters in triploid and diploid *C. carpio* in Karnataka, India. Although these trials continued for several months beyond the typical time of first maturation in diploid common carp, the triploid fish did not show any superiority over diploids apart from higher dressout percentages.

In the grass carp, the induction of triploidy was investigated in connection with control of reproduction of this exotic species that is used for aquatic weed control in the USA (Allen and Wattendorf 1987). This is in fairly widespread use, e.g. in the state of Florida (Thomas 1994).

Triploidy could at least in theory be used anywhere to control reproduction of alien species, domesticated strains, transgenic fish, etc., and is already used in commercial aquaculture in a limited range of species. However, many Asian cyprinid hatcheries allow brood stock to spawn in tanks or ponds without stripping eggs and sperm. As *in vitro* fertilization is necessary for the induction of triploidy (to give access to newly fertilized eggs and to allow the precise timing of the shocks involved relative to fertilization), the incentive to produce triploids would have to be high to bring about such a change in the hatchery breeding technique. Crosses between diploid and tetraploid brood stock can produce triploid offspring without the need to induce triploidy (e.g. in rainbow trout: Myers and Hershberger 1991), but the production of tetraploid brood stock is very difficult in most species where this has been attempted (very low larval viability; poor fertility in females: Thorgaard et al. 1990; Pandian and Koteeswan 1998; Rothbard et al. 2000; Arai 2001) and fertilization rates in diploid female x tetraploid male crosses are also low, probably because of the large size of the sperm (Thorgaard et al. 1990).

4.7 Sex determination and its manipulation

Most of the cyprinid species that are cultured in Asia (e.g. Indian major carps, Chinese carps) are harvested long before sexual maturation and before any significant sexual dimorphism is observed in traits that are of relevance to aquaculture. However, there are exceptions. The two most noticeable are *B. gonionotus* and *C. carpio*. *B. gonionotus* generally matures at one year old, and even during growout significant sexual dimorphism in the growth rate may be observed, with females growing faster than males (Pongthana et al. 1999). Ripe ovaries are often found in harvested females, and are consumed in many countries where this species is grown, adding value to the females for culture. In some of the regions of Asia in which the *C. carpio* is cultivated, sexual maturation is observed in fish as young as six months old (Reddy et al. 1998; Basavaraju et al. 2002.). While this may or may not be a problem in terms of breeding and fry production before harvest, a significant proportion of the body weight at harvest can be gonad (the gonadosomatic index can exceed 20 per cent in females). Females also grow slightly faster than males. Sexual dimorphism for growth appears to be larger in Asian strains of *C. carpio* than in European strains (Wohlfarth et al. 1975).

The sex determination systems of several species of cyprinids of interest to Asian aquaculture have been investigated using gynogenesis and/or hormonal masculinization and progeny testing. All-female gynogenetics have been produced in *C. idella* (Stanley 1976), *C. carpio* (Nagy et al. 1978), *A. nobilis* (Shelton 1986a: cited by Castelli 1994), *H. molitrix* (Mirza and Shelton 1988) and *B. gonionotus* (Pongthana et al. 1995), while all-female progeny have been observed in at least some of the crosses between hormonally masculinized fish and normal females in *C. carpio* (Nagy et al. 1981; Wu et al. 1990), *C. idella* (Boney et al. 1984; Shelton 1986b) and *B. gonionotus* (Pongthana et al. 1999: there were also some batches containing a small proportion of males in this study). Bongers et al. (1999) produced viable YY *C. carpio* by androgenesis. These results suggest that female homogamety (i.e. an XX/XY type of sex determination mechanism) is found in these species. However, there are some species of cyprinids that have, or appear to have, other types of sex determination mechanisms. For example, fully inbred lines of the *D. rerio*) contained both males and females (Streisinger et al. 1981), and the European barbel (*Barbus barbus*) has a female heterogametic (ZW/ZZ) system (Castelli 1994); gynogenetic and control batches of the rosy barb (*Puntius conchonius*) showed very variable sex ratios (Powell 2000). Komen et al. (1992a and b) demonstrated that a recessive mutation (*mas-1*), first found in a homozygous inbred line of *C. carpio*, caused masculinization of genetic females (XX).

In Israel, all female *C. carpio* populations have been established by sex reversing XX-gynogenetic females to males and using these males for breeding. This resulted in a 10-15 per cent increase in yield (Gomelsky et al. 1994).

Mirza and Shelton (1988) developed a breeding plan for the production of monosex female *H. molitrix*, to control reproduction of an exotic species used for water quality improvement in the USA. Rothbard et al. (2000) produced triploid all-female *C. idella* by inducing triploidy (see 4.6) in eggs fertilized by XX neomales. In Asia, there has been some application of monosex female breeding plans for the species where sexual dimorphism is observed in aquaculture, namely *C. carpio* (Wu et al. 1990) and *B. gonionotus* (Pongthana et al. 1999). Fig. 4.3 shows the breeding plan initially developed for monosex female *B. gonionotus* by Pongthana et al. (1995, 1999), although this has subsequently been modified to combine mass selection for the growth rate with all-female production. In the modified plan, neomales are produced in each selected generation by gynogenesis and sex reversal.

Sex-reversal in carps using steroid hormones, required for purposes such as the breeding plans outlined above, appears to be more difficult to achieve routinely than in cichlids or salmonids. This appears to be because the onset of sexual differentiation (the "labile period" for sex-reversal) occurs later in cyprinids and may be dependant on size as well as age (Shelton et al. 1995), thus making the exact timing of treatments difficult. There may also be genetic variation in *C. carpio* for response to treatment with 17α-methyltestosterone (Komen et al. 1993). Table 4.1 presents information from sex-reversal studies on several cyprinid species used in Asian aquaculture.

4.8 Gene transfer

The first published study on gene transfer in fish (Zhu et al. 1985) involved the introduction of

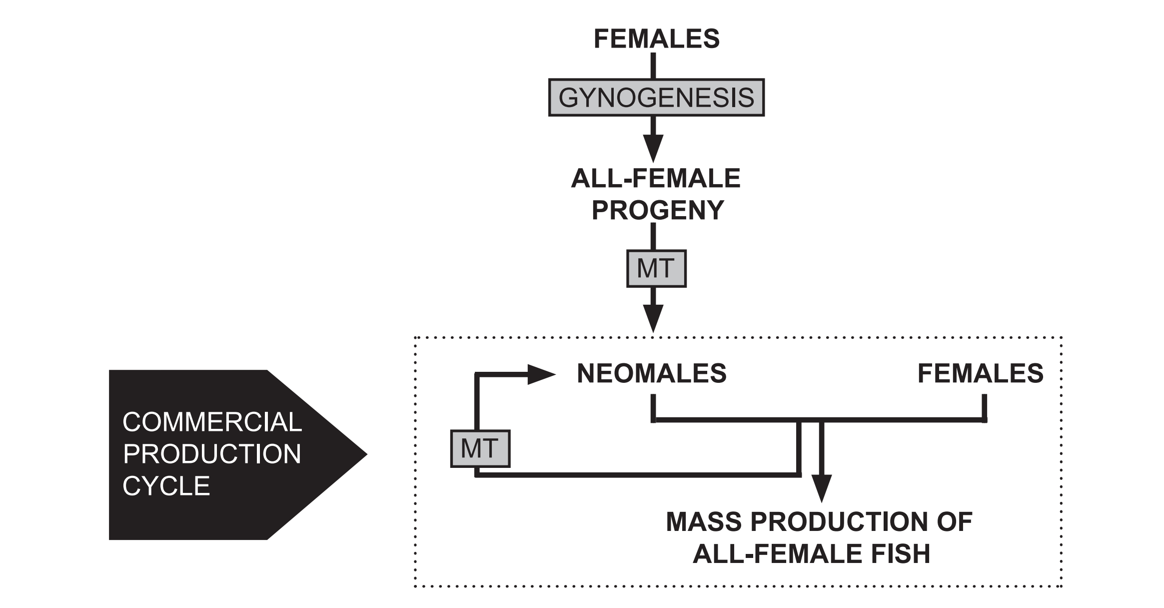

Fig. 4.3. Mass production of all-female *B. gonionotus*. MT = masculinization with 17α-methyltestosterone. After Pongthana et al. (1995, 1999)

Table 4.1. Examples of sex-reversal in cyprinid species. DPH = days post-hatch; MT = 17α-methyltestosterone; E2 = 17β-estradiol; n.d. = not determined

Species	Steroid treatment	Start time (DPH)	Duration of treatment (days)	Results and comments	Reference
C. carpio	MT (100 $mg.kg^{-1}$ in feed); gynogenetic fry	26-62	36	71-86.7% males, rest undifferentiated when treated at 25°C; size effect; hermaphrodites at 20°C	Nagy et al. (1981)
	MT (100 $mg.kg^{-1}$ in feed); normal fry	42	35	92.7% males (earlier or later treatments gave high % of sterile fish)	Komen et al. (1989)
	MT (100 $mg.kg^{-1}$ in feed); gynogenetic fry	26-40	40	Highest % males (up to 96.6%) in control groups in recirculating systems	Gomelsky et al. (1994)
	E2 (25-125 $mg.kg^{-1}$ in feed); normal fry	21-70	35	No effect on sex ratio	Komen et al. (1989)
C. idella	5 mg MT in silastic implant; normal fry	112	n.d.	98% male or intersex	Boney et al. (1984)
	5 mg MT in silastic implant; gynogenetic fry	70	n.d.	86% male or intersex	Boney et al. (1984)
B. gonionotus	MT (25 $mg.kg^{-1}$ in feed); gynogenetic fry	14	28 or 35	33.9, 29.8% males	Pongthana et al. (1999)
H. molitrix	5 mg MT in silastic implant; normal fry	124 or 319	n.d.	78.9, 90.9% male or intersex	Mirza and Shelton (1988)

mammalian growth hormone coding sequences driven by mammalian metallothionein promoters (pMThGH) into *C. auratus* eggs. Zhu (1992) reviewed the early research on this subject and described how linearized cloned foreign genes were microinjected into fertilized, dechorionated cyprinid eggs at the single-cell stage using the expelled second polar body as a guide to the target area for microinjection. This led to the generation of mosaic transgenic fish (with copies of the foreign gene integrated into the chromosomes of some, but not all, cells) and germline inheritance of the foreign genes. Expression of the foreign genes could be detected through Northern blots, radioimmunoassay and enhanced growth in the transgenic fish. Transgenic *C. carpio* and *C. carassius* were respectively 9.4 per cent and 78.4 per cent larger than the controls. Changes in body proportions were also observed in transgenic carp and loach, with these fish being deeper in relation to length than the controls. Wei et al. (1993) and Fu et al. (1998) also described growth trials on *C. carpio* using the pMThGH fusion gene. Transgenic fish were 22.7 per cent to 52.1 per cent larger than the controls. Fu et al. (1998) concluded that the transgenic carp were more efficient in utilizing dietary protein than the controls across a range of 20-40 per cent dietary protein content.

Further developments in China described by Zhu (1992) included the gene transfer by electroporation and an "all-fish" growth hormone gene construct, pCAgGH, in which expression of a *C. idella* growth hormone gene was controlled by a *C. idella* β-actin promoter. This "all-fish" construct was developed with two aims: to try to increase the efficiency of growth enhancement and to produce transgenics that were more likely to be acceptable to consumers. It has been suggested that transgenics receiving only rearranged genes from the same species should be called "autotransgenics", as opposed to "allotransgenics", where all or part of the introduced DNA construct comes from another species (Beardmore 1997).

Other research groups attempted to enhance growth in *C. carpio* using DNA constructs that were at least partially of fish origin (see Table 4.2). A construct in which the *Oncorhynchus mykiss* growth hormone cDNA was expressed using the long terminal repeat promoter of the Rous sarcoma virus (RSVrtGHcDNA) were used by a group in the USA, while constructs using the carp β-actin promoter and *O. tshawytscha* (FV-1/csGH) or carp growth hormone (FV-2/cGH) were used by a group in Israel. These groups achieved similar levels of growth enhancement to that obtained with pMThGH. Transgenic carp were up to 58.5 per cent larger in the studies in the USA (Zhang et al. 1990; Chen et al. 1993). In the Israeli studies two groups of transgenic carp were 42.2 per cent and 70.6 per cent larger than the controls in a winter growth trial (although the controls were slightly larger than the transgenics at the start of the trial, the transgenics showed higher specific growth rates during the trial), while three groups of transgenic *C. carpio* all showed a lower specific growth rate (SGR) than the controls in a summer growth trial (Hinits and Moav 1999).

Most research on growth enhancement via gene transfer in cyprinids has been carried out on *C. carpio*. While there is some data on growth in transgenic *C. carassius* (Zhu 1992; Xu et al. 1991) and *D. rerio* (Sheela et al. 1998), published information on other cyprinids is mostly related to methodology for gene transfer, e.g. on Indian major carps (Alok and Khillan 1989; Reddy et al. 1991; Tantia et al. 1991; Venugopal et al. 1998) and *M. amblycephala* (Wu et al. 1994).

While growth enhancement has been achieved in *C. carpio* using GH constructs, the levels of enhancement demonstrated in experiments to date do not compare with that achieved in salmonids (e.g. transgenic mean weight >10 times the size of control mean: Devlin et al. 1995) and tilapia (e.g. transgenic mean weight >3 times the size of control mean: Rahman and Maclean 1999). Dunham (1999), reviewing the literature on transgenic fish, suggested that the response (growth enhancement) appears to be greatest in unimproved stocks. This seems to be confirmed by experiments carried out by Devlin et al. (2001) in salmonids. Rainbow trout from a slow-growing wild stock showed dramatic growth enhancement following growth hormone gene transfer (OnMTGH1) while a fast-growing domesticated strain indicated relatively modest growth enhancement.

Given the extremely long history of domestication and selection for *C. carpio* (Balon 1995; Hulata 1995) and the relatively short history of domestication of most other cyprinids, it is attractive to speculate that transgenic growth enhancement in cyprinids such as Indian major carps and Chinese carps could be larger than that observed in *C. carpio*. However, the studies reported above on *C. carpio* utilized different DNA constructs from those used in salmonids and tilapia, which may also have been a factor. Promoter efficiency, other DNA sequences in the construct (introns, enhancers, vector, etc.), integration position affects, DNA methylation, etc., can all affect expression of transgenes (Iyengar et al. 1996).

Transgenic fish are now widely used for basic research on developmental biology, gene regulation and function, etc. (see Table 4.2 for examples) *D. rerio* has become one of the most important model species for such studies. While proving valuable for basic research, some of the knowledge generated should also filter through into aquaculture-related developments. These can be divided into two broad areas: improved efficiency in generating transgenic organisms (transfer efficiency, targetted integration, control over location, timing and level of expression, etc.) and greater ability to modify a variety of commercially important traits. While attempts have been or are being made to modify traits of fish such as freeze and cold tolerance (Shears et al. 1991; Fletcher et al. 1992; Wang et al. 1995), disease resistance (e.g. Jiang 1993; Jia et al. 2000; Dunham et al. 2002; Sarmasik et al. 2002; Zhong et al. 2002), carbohydrate metabolism (Pitkänen et al. 1999) and sterility (Alëstrom et al. 1992; Uzbekova et al. 2000) in fish, to date the results from such research have generally been much less encouraging than those from transgenic growth enhancement, or the research is still at an early stage.

Major concerns exist over consumer issues and potential environmental impacts of transgenic fish. Wu et al. (2003) address some of these issues. "All-fish" DNA constructs were used to produce transgenic growth-enhanced *C. carpio*, to lessen consumer worries about eating fish expressing non-piscine genes. Trials were carried out on mice fed with transgenic or non-transgenic fish, and the digestibility of the growth hormone in the human digestive tract was considered. They concluded that the "all-fish" GH-transgenics met the principle of "substantial equivalence" to the non-transgenics. To address the potential environmental impact, Wu et al. (2003) also produced a sterile triploid transgenic, by crossing diploid transgenic *C. carpio* with tetraploid *C. carassius* x *C. carpio* hybrids (Liu et al. 2001). The benefits to fish farmers using the transgenics were estimated to be 52 per cent higher than from using normal common carp. Producing triploids in this way should avoid the possibility of a remnant small proportion of diploids (i.e. temperature or pressure shock "failures") that could breed. Wu et al. (2003) stated that the "all-

Table 4.2. Gene transfer in cyprinids: examples of DNA constructs including sequences from cyprinids (1-3) and DNA constructs that have been introduced into cyprinids (4-5). (*D. rerio* examples quoted represent only a sample of papers/subjects related to this species)

Role of cyprinid	DNA sequences	Species	References
1. Promoters/ regulatory sequences from cyprinids	Carp myosin, β-actin regulatory sequences	*C. carpio, C. idella*	Liu et al. (1989, 1990a and b); Cavari et al. (1993); Moav et al. (1993, 1995); Alam et al. (1996); Alam and Maclean (1997); Müller et al. (1997); Hinits and Moav (1999)
2. Coding sequences from cyprinids	α-globin gene	*C. carpio*	Yoshizaki et al. (1991a and b); Fu and Aoki (1992)
3. "All-cyprinid" DNA constructs	Actin promoter/ GH coding sequence	*C. carpio/C. idella (respectively)*	Zhu et al. (1993)
4. Cyprinid recipients for gene transfer (non-cyprinid DNA constructs)	Non-piscine GH construct	*C. idella, L. rohita, C. cirrhosus, M. ambycephala, C. carassius, C. auratus, D. rerio*	Zhu et al. (1985); Alok and Khillan (1989); Chen et al. (1989); Hayat et al. (1991); Hernandez et al. (1991); Pandian et al. (1991); Powers et al. (1991); Xu et al. (1991); Chen et al. (1993); Cui and Zhu (1993); Wei et al. (1993); Xie et al. (1993); Erdelyi et al. (1994); Wu et al. (1994); Chatakondi et al. (1995); Sun et al. (1995); Fu et al. (1998) ; Sheela et al. (1998)
	Piscine GH construct	*C. carpio, L. rohita, C. cirrhosus, C. catla, C. auratus, D. rerio*	Chen et al. (1990, 1993); Zhang et al. (1990); Hayat et al. (1991); Powers et al. (1992) ; Cavari et al. (1993); Zhang et al. (1993); Zhao et al. (1993); Venugopal et al. (1998)
	Rainbow trout vitellogenin gene	*C. carpio*	Hayat *et al.* (1991)
	Non-piscine reporter gene	*C. carpio, C. carassius, C. auratus, D. rerio, Puntius conchonius*	Hallerman et al. (1990); Yoon et al. (1990); Zelenin et al. (1991); Khoo et al. (1992); Müller et al. (1993); Guillen et al. (1996); Kang et al. (1999)
	Rat Gap-43 gene construct	*C. carpio*	Shimbo et al. (1993)
	Bacterial plasmid	*D. rerio*	Stuart et al. (1988)
	Mammalian *Hox* promoters	*D. rerio*	Westerfield et al. (1992)
5. Cyprinid recipients for gene transfer (DNA construct partially or totally of cyprinid origin)	Reporter gene with carp myosin, β-actin regulatory sequences	*C. auratus, D. rerio*	Moav et al. (1993); Williams et al. (1996); Müller et al. (1997)
	Common carp β-actin promoter + salmon GH or carp GH	*C. carpio*	Moav et al. (1995); Hinits and Moav (1999)

fish" transgenic *C. carpio* were waiting for public acceptance and governmental authorization.

Cloned gene constructs have been shown to be expressed strongly when injected directly into the muscle of fish (Hansen et al. 1991; Rahman and Maclean 1992). From this and studies on mammals, "DNA vaccination", where proteins expressed from the injected DNA stimulate anti-body production, is being developed as an alternative to more traditional vaccination techniques for fish (e.g. Heppell et al. 1998; Kanellos et al. 1999). A U.S. patent has been taken out on this process (Davis 2001). The injected DNA does not appear to integrate into the fish genome (Vaughan et al. 1999), so the vaccinated fish cannot be described as transgenics. Goldfish have been shown to produce anti-bodies to foreign proteins expressed from plasmids injected into muscle (Russell et al. 2000), while the carp β-actin promoter has been shown to be suitable for driving expression of foreign proteins in this situation (Gomez-Chiarri and Chiaverini 1999).

4.9 Cryopreservation

Successful cryopreservation of sperm of many cyprinids, like many other groups of fish, has become fairly routine at least under laboratory conditions (Withler 1982; Kumar, 1988, 1989; Gupta and Rath 1991; Chen et al. 1992a and b; McAndrew et al. 1993; Maisse 1996; Ponniah et al. 1999). In contrast, cryopreservation of eggs and embryos has not been successful, and appears likely to remain elusive (Chen et al. 1988; Calvi et al. 1996; Lubzens et al. 1996; Zhang et al. 1997). While cryopreservation of sperm alone still has applications in relation to genetics research and practice in aquaculture, the inability to store eggs or embryos in the same way places major limitations on most of these. For example, while cryopreserved sperm can form a gene bank of sorts for a wild or domesticated stock of fish, without cryopreserved eggs from the same stock live fish can only be "reconstituted" from the gene bank using androgenesis (Penman et al. 1996), and even then the mtDNA will be of maternal origin. Otherwise eggs from a later generation of the same stock (if available) or from a similar stock will have to be used.

Cryopreserved sperm can be used to monitor the progress of selective breeding without maintaining a separate (live) control line in addition to the selected line(s). The difference between the means of the current generation x current generation and current generation x earlier generation crosses should represent half of the genetic progress over this period, and the progress per generation can be calculated from this.

One factor that has limited the successful application of sperm cryopreservation at the field level has been poor reproducibility of freezing protocols (Rana and Gilmour 1996). While programmable coolers can give highly reproducible cooling rates in different samples, simpler types of apparatus (e.g. freezing straws in liquid nitrogen vapor in a polystyrene box or in the neck of a Dewar vessel) can give highly variable cooling rates and thus lower success. Improved portable sperm freezing systems have been designed (e.g. Magyary et al. 1996) and other attempts have been made to efficiently scale up sperm cryopreservation methods (Lubzens et al. 1997).

4.10 Tissue culture

Cultured cells can be used for a variety of purposes associated with research on genetics of cultured cyprinids. Examples of such applications are:

(i) Short-term leucocyte culture to provide an abundant source of high-quality metaphase spreads for studies on the cytogenetics of the source species. In general, such cultures are a much better source of mitotic chromosome spreads than solid tissues. Chromosome preparations can also be made from longer term tissue cultures, but chromosome rearrangements and losses in such cultures may limit their usefulness for this purpose (e.g. Li et al. 1994).
(ii) Tissue cultures can be used to test expression of gene constructs ultimately intended for gene transfer (Moav et al. 1991; Bearzotti et al. 1992; Fu et al. 1993; also see 4.8). Transformation and expression assays are often more easily carried out in tissue culture than in whole organisms.
(iii) Fish embryonic stem (ES) cell lines could be genetically transformed and then introduced into developing embryos, giving rise to chimeric fish. Transgenic offspring would be generated from the gametes produced from the stem cells that had become part of the developing gonadal tissue in the host fish. ES lines could thus form a "bridge" between *in vitro* and *in vivo* manipulations of animal

genomes. The elements of such a system have been demonstrated in the mouse and the medaka, *Oryzias latipes* (Hyodo and Matsuhashi 1994; Sun et al. 1995; Hong et al. 2000) but not to date in any cyprinids.

(iv) Nucleus transplantation (see also 4.4). While nuclei of cells from dissociated early embryos have given rise to the most successful rates of nucleus transplantation (e.g. Tung et al. 1973), tissue culture could provide large numbers of cells for this process, particularly if scaling up using electrofusion instead of microinjection (Chen et al. 1990). Although nucleus transplantation has been achieved using donor cells from tissue culture, chromosome rearrangements and losses occurring in long-term cultures could limit the usefulness of such an approach.

Primary cultures and cell lines have been set up from several species of cyprinids, including *C. carpio* (e.g. Horiuchi et al. 1979; Fijian et al. 1983; Chen et al. 1987; Moritomo et al. 1996; Weyts et al. 1997; Degani et al. 1999), major carps (e.g. Rao et al. 1997; Sathe et al. 1997; Joseph et al. 1998), *C. idella* (e.g. Deng et al. 1985; Zuo et al. 1986; Li et al. 1988; Yang et al. 1992; Zeng et al. 1993) and ginbuna crucian carp (e.g. Hasegawa et al. 1997). Very few cyprinid cell lines are held in, and thus available from, centralized cell culture collections – the ECACC (European Collection of Cell Cultures: http://www.ecacc.org/) listed four cyprinid cell lines (CLC and EPC from *C. carpio*; CAR from *C. auratus*; and FHM from fathead minnow, *Pimephales promelas*) among 25 fish lines from a total of 985 lines, while the ATCC (American Type Culture Collection: http://www.atcc.org/) did not list any cyprinid cell lines among nine fish cell lines.

4.11 Summary

In the last ten years or so, progress has been made in several areas of basic and applied research on the genetics of the carp genetic resources of relevance to Asia. For example, China has continued its progress on selective breeding of *C. carpio* and all of the other countries where cyprinids are of importance to aquaculture now have breeding programs for one or more important cyprinid species. Most of these selective breeding programs have begun by comparing different stocks of the target species, including studies of genotype x environment interaction and heterosis, and have then used mass selection to improve growth-related traits. Some have used more complex forms of selection or selected for more than one trait. In other directions, there have been some studies on population genetics of important cyprinid species, development of monosex female production in appropriate species, and further advances in research on transgenic fish.

With some notable exceptions, there has been limited impact of genetic improvement on aquaculture production to date in most of the countries in Asia where carps are important in aquaculture. Dissemination of improved breeds is now receiving a greater degree of attention, including the targetting of poorer farmers.

Priority areas for future research and development on the genetic resources of cyprinids in Asia should include:

- A greater understanding of the population genetic structure and biodiversity of indigenous cyprinids, to support both conservation efforts and exploitation for aquaculture.
- The continued application of genetics research to develop improved breeds for aquaculture.
- Greater interaction with fish farmers, hatcheries, etc., and dissemination of improved breeds and the technologies involved.

Molecular genetics is a rapidly advancing field that has already given valuable tools such as polymorphic molecular markers and gene transfer. It is likely that further developments in this area will have applications in research on the genetics of cyprinids in Asia, hopefully to the benefit of both conservation and exploitation of these resources.

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Chapter 5
Constraints to Increased Yields in Carp Farming: Implications for Future Genetic Research[1]

Madan Mohan Dey[2], Ferdous Alam[3], Miao Weimin[4], Somying Piumsombun[5], Ramachandra Bhatta[6], Le Thi Chau Dung[7] and Ferdinand J. Paraguas[2]

5.1. Introduction

The Asian countries, particularly Bangladesh, China, India, Indonesia, Thailand and Vietnam are the largest producers of carps in the world. Various culture technologies and carp strains and varieties have been developed or introduced to these countries. The adoption of carp farming in Asia has greatly improved fish production. However, large difference in production levels exists among farms in each country and between countries, even in China, where carps have been cultured for thousands of years. Several issues related to carp production in Asia are important to the research community:

1. What are the production constraints?
2. What should be the priorities for research?
3. What is the best research strategy according to these priorities?

To meet the urgent need for increased carp production in developing countries of Asia (Delgado et al. 2003; Dey et al. 2004), the problems that affect carp yield must be clearly identified. Planners and policy-makers need information on the relative importance of various problems so that they can design and implement strategies to solve these problems. This chapter has the following specific objectives: (i) to identify the gap between the potential and actual yield (yield gap); (ii) to estimate the contribution of various technical constraints to the yield gap; and (iii) to suggest appropriate research strategies to solve these constraints.

5.2 Conceptual Framework and Survey Methodology

Following previous studies on similar subjects (Widawsky and O'Toole 1990, 1996; Dey et al. 1996; Lin and Shen 1996), the fundamental assumptions of this study are: 1) there exists a yield gap between potential and actual yield or there is a possibility of increasing potential yield; 2) the yield gap is due to a number of constraints; 3) the specific contribution of individual constraints can be estimated and cumulatively account for part, but not all, of the yield gap; and 4) based on the estimated value of yield loss, technical constraints can be ranked in order of importance.

Yield gap analysis has been used in agriculture for many years (IRRI, 1977, 1979; De Datta et al. 1978; Widawsky and O'Toole 1990; Evenson et al. 1996; Dey et al. 1996). Yield gap studies can demonstrate how close farm yields are to the maximal potential yield with the available technology. They can also suggest how to improve production more efficiently through the extension of present available technology or by developing new technologies.

Three definitions of yield gap are found in the literature. Yield gap I is defined as the difference between the yield observed on experimental stations and the best practice yield on farmers' fields. This yield gap probably is largely attributable to inherent differences in the biophysical environments (micro and macro) between the experimental station and typical farmers' fields, which cannot be easily managed

[1] WorldFish Center Contribution No. 1730.
[2] WorldFish Center, Jalan Batu Maung, Batu Maung, 11960 Bayan Lepas, Penang, Malaysia.
[3] WorldFish Center-Bangladesh Office, House No. 22-B, Road No. 7, Banani, Dhaka 1213, Bangladesh.
[4] Freshwater Fisheries Research Center, Chinese Academy of Fisheries Science, Asia-Pacific Regional Research and Training Center for Integrated FishFarming, Wuxi Fisheries College, Nanjing Agricultural University, No. 1 Qitang, Wuxi 214081, China.
[5] Department of Fisheries, Ministry of Agriculture and Cooperatives, Kasetsart University Campus, Phaholyotin Road, Bangkok 10900, Thailand.
[6] Department of Fisheries Economics, College of Fisheries, University of Agricultural Sciences, Mangalore, Karnataka, 575 002, India.
[7] Vietnam Agricultural Science Institute, Vietnam.

or eliminated. Production research or changes in socioeconomic conditions can do little to exploit yield gap 1. Yield gap II is defined as the difference between actual and best practice yields on farmers' fields, given a particular technology. Yield gap II is caused by technological constraints (e.g. disease, soil problem, water quality problem, adverse climate/weather, etc.) and/or socioeconomic constraints (e.g. lack of credit, poor knowledge, input unavailability, tradition and attitudes, poor institutions, etc.). It is a measure of the yield that could be gained through technological and policy interventions to overcome constraints. This gap can be overcome if practical, profitable and sustainable means to control constraints are developed. Yield gap III is the difference between the theoretical potential yield and highest experimental yield. It represents the potential increase in biological efficiency, and is attributed to genetic and management improvement conceived but not yet developed or perfected. By definition this yield gap cannot be measured.

In this study the focus is on the technological constraints component of yield gap II. To get information on the yield gap and factors contributing to yield gap II, around 1900 carp farmers in six countries of Asia (Bangladesh, China, India, Indonesia, Thailand and Vietnam) were interviewed. Sampled farmers were asked to report their maximum, minimum and normal (average) yields obtained from particular pond(s)

Table 5.1. Yield gap II in carp culture by intensity level

Intensity Level[a]	Bangladesh	China	India	Indonesia[b]	Thailand	Northern Vietnam
"Low" intensity						
No. of farms	295	122	96	39	95	27
Potential (max) farm yield (mt/ha)	6.31	21.39	6.79	89.10	9.50	6.97
Average (actual) farm yield (mt/ha)	3.06	6.77	2.80	48.17	3.60	3.47
Yield gap II (mt/ha)	3.25	14.62	3.99	44.30	5.90	3.50
Yield gap II as % of max farm yield	54.88	68.36	58.76	49.72	62.11	50.19
"Medium" intensity						
No. of farms	146	155	232	71	105	56
Potential (max) farm yield (mt/ha)	6.69	28.95	14.62	96.10	10.60	10.11
Average (actual) farm yield (mt/ha)	3.23	11.69	3.20	57.40	3.70	3.53
Yield gap II (mt/ha)	3.46	17.26	11.42	38.70	6.90	6.58
Yield gap II as % of max farm yield	51.77	59.63	78.11	40.27	65.09	65.08
"High" intensity						
No. of farms	99	157	81	31	48	28
Potential (max) farm yield (mt/ha)	8.45	40.77	16.89	280.50	11.30	12.16
Average (actual) farm yield (mt/ha)	3.91	16.61	3.78	200.70	4.40	4.05
Yield gap II (mt/ha)	4.54	24.16	13.11	79.80	6.00	8.11
Yield gap II as % of max farm yield	53.76	59.26	77.62	28.45	53.09	66.69
Overall[b]						
No. of farms	540	434	409		248	111
Potential (max) farm yield (mt/ha)	7.13	27.18	14.94		10.58	10.38
Average (actual) farm yield (mt/ha)	3.26	12.08	3.20		3.78	3.65
Yield gap II (mt/ha)	3.87	15.10	11.74		6.80	6.73
Yield gap II as % to max farm yield	54.30	55.56	78.58		64.27	64.84

[a] Although most of the fish farms surveyed are semi-intensive in nature (except for some intensive pond culture in China and some intensive cage culture in Indonesia), the sample farmers were grouped into three intensity levels ("low", "medium" and "high") to see whether the yield gap varies with the changes in the intensity level. For all countries except Indonesia, the culture system/environment is pond polyculture under all intensity levels. For Indonesia, it is running water, floating cage and double floating cage for "low", "medium" and "high" intensity levels, respectively.

[b] Given the fact that the Indonesian sample from three culture systems /environments (i.e., running water, floating cage and double floating cage) represent three different levels of input intensity (low, medium and high); we have not done any analysis for the overall sample.

Source: Field surveys conducted by the authors..

during the last 10 years. Yield gap II was calculated as the difference between the maximum farm-level yield and the average actual farm-level yield. The adoption of a single maximum yield from the sample is not wise as it may be an outlier and hence may exaggerate the magnitude of the yield gap. To overcome this potential problem, the average of the top 5 per cent of the total sample was used as the maximal farm level yield.

A total of 1 883 fish farmers, who are knowledgeable about various technical details of fish farming, were interviewed in six countries[8] using a pre-designed questionnaire to collect information on yield gaps and yield losses by technical constraints. The interviews were conducted by experts who are also knowledgeable about local fish farming conditions, sometimes in the presence of one or more local fisheries/aquaculture workers. The survey covered pond polyculture in Bangladesh, China, India, Thailand and Vietnam, and monoculture of *C. carpio* in a running water system and cages in Indonesia.

5.3 Technical Constraints to Carp Production in Asia

Yield gap analysis

The overall average yield gap II in carp culture of the six countries examined is more than 50 per cent of the potential yield (Table 5.1). This yield gap is considerably larger than the country specific rice yield gaps estimated by the International Rice Research Institute (IRRI) and its partners in the early 1990s (Evenson et al. 1996). This may be due to the fact that, compared with rice cultivation, carp culture is carried out in a more complex environment. That is, farm fish production is more influenced by the environmental conditions than is rice cultivation.

Comparisons among the different countries indicate that on average Indonesia has the smallest yield gap for carp culture. In contrast, countries with considerable environmental variation and a diversity of culture systems, such as Bangladesh, China, India and Thailand, have relatively higher yield gaps. The average ratio of the yield gap to the potential yield is between 54 and 79 per cent in all countries, except Indonesia. Although most of the fish farms surveyed were semi-intensive in nature (except for some intensive pond culture in China and some intensive cage culture in Indonesia), the sample farmers were divided into three intensity levels ("low", "medium" and "high" to see whether the yield gap varies with the changes in the intensity level. In general, the yield gap is higher in higher intensity farms, as is the average yield.

Yield loss analysis

To explain the yield gap II in present carp culture in the six different countries, yield loss analysis was undertaken. It was expected that the yield loss analysis by technical constraints would provide insights into identifying priority areas for further genetic research to improve production. Two kinds of technical constraints, abiotic (e.g. water, soil, temperature, etc.) and biotic (e.g. disease) were identified by biologists and included in the study. The financial loss caused by different factors (constraints) was estimated, based on the producer survey data. The results of this analysis are in Table 5.2. The outcome of the Indonesian study could not be incorporated into this analysis because data collection differed from that in other countries.

The reported total annual financial loss caused by various biotic and abiotic factors ranged from US$243/year/ha (Bangladesh) to US$1 691/year/ha (China). Financial loss as a percentage of the total yield was highest in northern Vietnam and Thailand (53 and 54 per cent, respectively) and lowest in Bangladesh (14 per cent). Such wide differences mainly result from factors included in the study by different countries or institutions. Given the fact that the average yield gap II is about 65 per cent of the potential yield (i.e. yield gap is about 185 per cent of the average yield) (Table 5.1), the result of the yield loss explained only a small portion of the gap II. As hypothesized earlier, technical constraints account for only a part of the total yield gap II. Yield loss estimated under this study did not include an analysis of the socioeconomic factors. Many of the small-scale fish farmers in Asia are risk averse, i.e. they are not prepared to take the financial risks involved in intensification.

The study revealed that water quantity and quality (specifically, high turbidity and low dissolved oxygen) and diseases (bacterial and viral) are factors contributing most to yield loss. Soil problems and extreme temperatures also

[8] The number of farmers interviewed in each of the countries are given in Table 5.2.

contribute to the total yield loss among farmers in Thailand. Among the important factors contributing to the total yield loss, low dissolved oxygen and disease are related to both the fish and farm management. The results presented in Table 5.2 indicates that the losses per hectare due to low DO and diseases are higher in countries with a more intensive production system (e.g. China). Improved farm management can reduce the likelihood of low dissolved oxygen and outbreaks of disease. However, it is not possible to avoid losses from these factors in today's carp culture practices, especially with the tendency to increase the farming intensity level. Therefore,

Table 5.2. Yield loss (US$/year/ha) caused by various factors in five participating countries

Factor	Bangladesh	China	India	Thailand	Northern Vietnam	Average
Water quality	170.98	776.48	269.86	139.17	470.07	365.31
High turbidity	109.28	-	-	26.68	-	27.19
Plankton bloom	21.17	77.48	5.50	21.11	-	25.05
Filamentous algae/weeds	4.52	-	-	68.53	-	14.61
Low Dissolved Oxygen	36.01	442.48	264.36	22.85	470.07	247.15
Pollution	-	256.52	-	-	-	51.30
Water	55.74	-	211.51	341.81	378.35	197.48
Shortage of water	8.48	-	211.51	89.88	378.35	137.64
Flooding	47.26	-	-	251.93	-	59.84
Soil Problem	8.42	-	-	184.51	-	38.59
Acidity	-	-	-	108.54	-	21.71
Sedimentation	-	-	-	5.48	-	1.10
Seepage	8.42	-	-	70.49	-	15.78
Disease	8.11	625.06	247.71	128.71	354.15	272.75
Virus	2.58	253.09	-	61.91	-	63.52
Bacteria	5.53	258.58	247.71	66.80	-	115.72
Parasite	-	113.39	-	-	-	22.68
Temperature	-	84.90	-	123.85	15.29	44.81
High	-	47.21	-	28.72	-	15.19
Low	-	30.37	-	95.13	15.29	28.16
Abnormal fluctuation	-	7.32	-	-	-	1.46
Others	-	119.79	-	189.78	48.41	71.60
Reduced growth	-	61.33	-	-	-	12.27
Easily affected by disease	-	58.46	-	-	-	11.69
Others	-	-	-	-	-	-
Total loss (US$/ha)	243.24	1 691.13	729.08	1 231.68	1 281.56	1 035.34
Average gross output (US$/ha)	1 715.12	10 797.11	2 124.53	2 343.42	2 374.07	3 870.85
Loss % of the yield	14.18	15.66	34.32	52.56	53.98	34.14

Source: Field surveys conducted by the authors

Table 5. 3. Percentage of each factor of the total yield loss

Factor	Bangladesh	China	India	Thailand	Northern Vietnam	Average
Water quality	70.29	45.91	37.01	11.30	36.68	40.24
High turbidity	44.92	0.00	0.00	2.17	0.00	9.42
Plankton bloom	8.70	4.58	0.75	1.71	0.00	3.15
Fila. Algae/weed	1.86	0.00	0.00	5.56	0.00	1.48
Low Dissolved Oxygen	14.80	26.16	36.26	1.86	36.68	23.15
Pollution	0.00	15.17	0.00	0.00	0.00	3.03
Water quantity	22.91	0.00	29.01	27.75	29.52	21.84
Shortage of water	3.49	0.00	29.01	7.30	29.52	13.86
Flooding	19.43	0.00	0.00	20.45	0.00	7.98
Soil Problem	3.46	0.00	0.00	14.98	0.00	3.69
Acidity	0.00	0.00	0.00	8.81	0.00	1.76
Sedimentation	0.00	0.00	0.00	0.44	0.00	0.09
Seepage	3.46	0.00	0.00	5.72	0.00	1.84
Disease	3.33	36.96	33.98	10.45	27.63	22.47
Virus	1.06	14.97	0.00	5.03	0.00	4.21
Bacteria	2.27	15.29	33.98	5.42	0.00	11.39
Parasite	0.00	6.70	0.00	0.00	0.00	1.34
Temperature	0.00	5.02	0.00	10.06	1.19	3.25
High	0.00	2.79	0.00	2.33	0.00	1.02
Low	0.00	1.80	0.00	7.72	1.19	2.14
Abnormal fluctuation	0.00	0.43	0.00	0.00	0.00	0.09
Others	0.00	7.08	0.00	15.41	3.78	5.25
Reduced growth	0.00	3.63	0.00	0.00	0.00	0.73
Easily affected by disease	0.00	3.46	0.00	0.00	0.00	0.69
Total Loss	100.00	100.00	100.00	100.00	100.00	100.00

Source: Field surveys conducted by the authors

geneticists and other scientists may look into the possibilities of initiating research to develop strains of fish that are resistant to disease and tolerant to low dissolved oxygen (DO). Apart from tolerance to low dissolved oxygen and resistance to disease, other problems such as intolerance to extreme temperatures may also be solved through genetic research. A closer examination of these data, however, shows that genetic gains may be able to reduce the total loss by only 18 per cent. On the other hand, 80 per cent of the total loss can be reduced by better management and other types of research (Table 5.4). Genetic research can also reduce yield gap III by increasing the highest potential yield (i.e. by pushing the yield frontier further). It is, however, important to note that there is a biological and ecological limit to which yield can be increased.

5.4 Conclusion

This chapter represents a first step towards identifying priorities for genetic research in Asia through economic analysis. The present yield gap and yield losses caused by various factors identified provide guidance to develop a strategy to further improve the production of carp culture in Asia and the world. Important traits that could be improved through genetic methods to increase the production quantity and output quality have been identified.

It is, however, important to note that the data presented in this chapter on yield loss are measures of demand for yield increasing research. For identifying genetic research priorities, the supply side of the research (i.e. research discovery

Table 5.4. Percentage of each factor of the total yield loss classified by potential solutions

Factor	Bangladesh	China	India	Thailand	Northern Vietnam
Genetic research	18.14	71.75	70.24	35.15	65.90
Disease (total)	3.33	38.91	33.98	11.62	27.80
Virus	1.06	15.76	-	5.59	-
Bacteria	2.27	16.10	33.98	6.03	-
Parasite	-	7.06	-	-	-
Soil Problem	-	-	-	10.29	-
Acidity	-	-	-	9.80	-
Sedimentation	-	-	-	0.49	-
Water quality	14.80	27.55	36.26	2.06	36.90
Low Dissolved Oxygen	14.80	27.55	36.26	2.06	36.90
Temperature	-	5.29	-	11.18	1.20
High	-	2.94	-	2.59	-
Low	-	1.89	-	8.59	1.20
Abnormal fluctuation	-	0.46	-	-	-
Other form of research	1.86	-	-	23.32	-
Filamentous algae/weeds	1.86	-	-	6.19	-
Others (predators)	-	-	-	17.13	-
Management	80.00	20.79	29.76	41.53	33.50
Water quality	53.63	4.82	0.75	4.31	-
High turbidity	44.92	-	-	2.41	-
Plankton bloom	8.70	4.82	0.75	1.91	-
Water quantity	26.38	15.97	29.01	37.22	29.70
Shortage of water	3.49	-	29.01	8.11	29.70
Flooding	19.43	-	-	22.74	-
Pollution	-	15.97	-	-	-
Seepage	3.46	-	-	6.36	-
Others	-	-	-	-	3.80

Source: Field surveys conducted by the authors.

process) would also need to be considered. Some problems are more difficult than others for research to address - they require more time or money to investigate, or have been less explored previously and there may be greater uncertainty associated with conducting research on them. For example, it is currently more feasible to develop disease resistant fish strains than to develop strains tolerant to low DO. There is a need to collect additional information on the likelihood, possible time length and cost of solving the constraints identified.

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Chapter 6
The Status Of Introduced Carp Species In Asia[1]

Belen O. Acosta[2] and Modadugu V. Gupta[2]

6.1 Introduction

Cyprinids are the most important group in aquaculture in Asia, accounting for 49 per cent of the total aquaculture production Asia in 1999 (FAO 2001a), and they also account for a large number of the introductions of tropical/subtropical finfish in the region (de Silva 1989). Introduction is defined as the act of intentionally or accidentally transporting and releasing an organism into an environment outside its present range (ICES 1988). In fish, the main reasons for deliberate introductions include increased production, employment, generation of foreign exchange, biological control and recreation (FAO 1996). While tremendous benefits can be and have been derived from fish introductions, there are also indications that movement of alien cyprinid species, both across international borders and across watersheds, has led to negative effects in natural ecosystems. The available information on species introductions, however, reports mainly on movements across country boundaries and very seldom on movements across zoogeographical boundaries or watersheds.

The Asian countries, in particular Bangladesh, China, India, Indonesia, Thailand and Vietnam, have vast carp genetic resources that are an important element of their aquaculture production systems. Hence, in recent years, the governments of these countries have been recognizing the need to exercise a precautionary approach in the introduction of any aquatic organism, including carps.

Information on the introduced species is a pre-requisite for implementing programs aimed at the conservation of genetic resources. This chapter discusses the status of introductions of cyprinid species across international borders in Asia. The data used were derived mainly from the FishBase[3] records of fish introductions, the Food and Agriculture Organization (FAO) of the United Nations database on species introductions, the International Network on Genetics in Aquaculture (INGA)[4] records on germplasm transfer and reports from INGA member countries. These include each instance in which a non-native species is moved into a recipient country, even if the species has already been moved into that recipient country before.

6.2 Species introduced and sources

A total of 259 introductions of cyprinid species have been recorded in the 27 countries of Asia. Of these, 72 per cent (186 records) were from countries within Asia, 10 per cent (26 records) were from countries outside Asia and another 18 per cent (47 records) were from "unknown" countries (Annex 6.1). The available records indicate introductions of forty-two cyprinid species. Fig. 1 shows the top five most often introduced cyprinid species based on the number of records. Common carp (*Cyprinus carpio*) was the most often introduced species in the region (52 records), followed by the grass carp (*Ctenopharyngodon idella*) (29 records), silver carp (*Hypopthalmichthys molitrix*) (28 records), bighead carp (*Aristichthys nobilis*) (20 records) and goldfish (*Carassius auratus auratus*) (15 records). The high frequency of introductions of *C. carpio* could be the result of this species having the longest history of culture and domestication among the cyprinids. It is believed that the species has been farmed in China for over 2000 years.

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[1] WorldFish Center Contribution No. 1731.
[2] WorldFish Center, Jalan Batu Maung, Batu Maung, 11960 Bayan Lepas, Penang, Malaysia.
[3] FishBase is an information system with key data on the biology of all fish. It has been developed by the WorldFish Center (formerly ICLARM) in collaboration with the Food and Agriculture Organization (FAO) of the United Nations and many other partners (Froese and Pauly 2002). Information on fish introductions in FishBase is an updated version of the original of Welcomme (1988) and has been expanded to include records of FAO, INGA and literature searches (Casal and Bartley 2000).
[4] INGA is a global forum for applied fish breeding and genetics. It plays an important role in national, regional and international genetics research aimed at improving productions from aquaculture operations and conservation of aquatic genetic resources. It has a membership of 13 countries from Asia, the Pacific and Africa and 12 advanced scientific institutions, international and regional organizations, with the WorldFish Center as the member coordinator (Gupta and Acosta 2001).

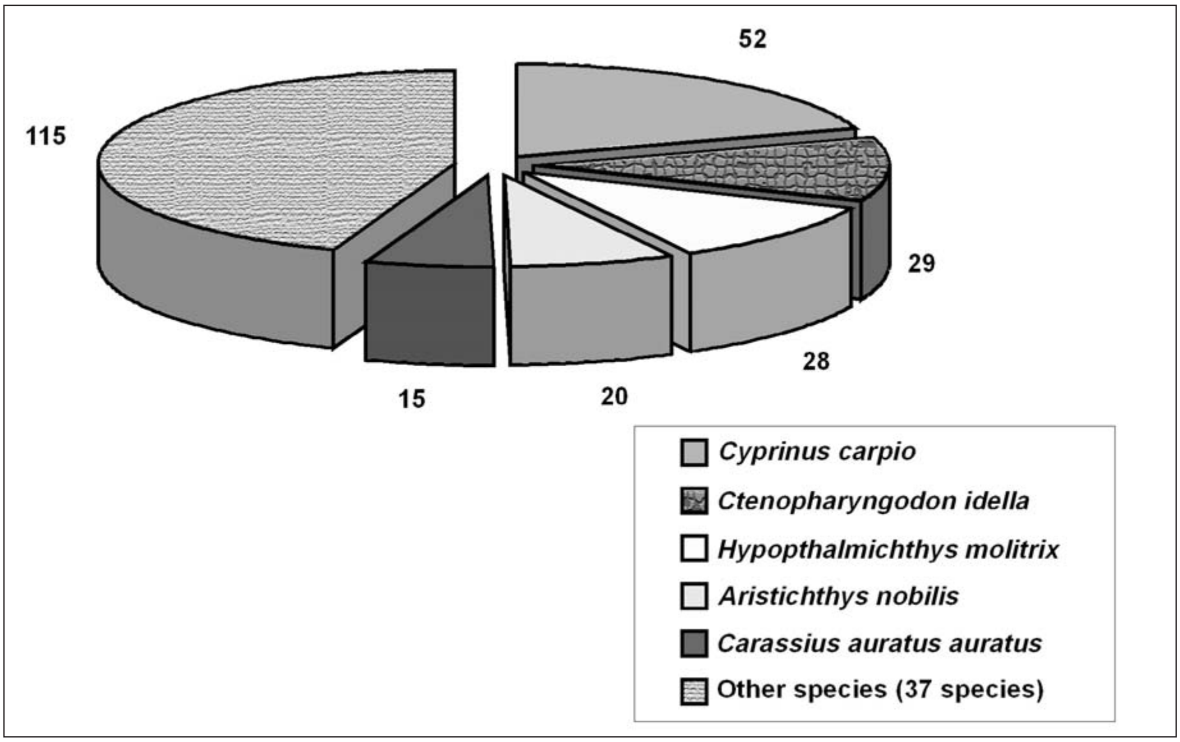

Fig. 6.1. Most commonly introduced cyprinid species based on the number of records (data based on the table in Annex 6.1)

6.3 Dates of introductions

Fig. 6.2 summarizes the number of cyprinid species introduced into the countries in Asia over time. Some of these introductions were made before the 18th century, with 38 records (or 14.7 per cent of the total) before 1950. *C. carpio* was the most frequent species among these early introductions. Apart from common carp, the Chinese carps were also introduced from the Pearl and Yangtze rivers to different provinces of China, Thailand, Singapore and Malaysia. The 4-5 cm fry were transported in wooden tubs on board ships until at least 1948, when fry were successfully transported by air for the first time (Lin 1949).

Subsequent introductions took place in the 1950s but the number of records for this period is only 11, or 4.2 per cent. During the 1960s, the frequency of introductions was at its highest (20 per cent or 52 records). The success of induced breeding through hypophysation, that improved fry supply, probably explains the increase of introductions during this decade. Over the next two decades, introductions became less frequent. Only 29 (11 per cent) and 31 (12 per cent) introductions were recorded in the 1970s and 1980s, respectively. From the 1990s to 2001, there was a further reduction in the recorded frequency of introductions (6.6 per cent or 17 records), probably because: (i) most of the non-native species that are good candidates for aquaculture had already been introduced in many countries; (ii) heightened awareness of the risks involved in introductions in terms of impact on the environment, biodiversity and introductions of pathogens; and (iii) there is now greater focus in the countries to improve the genetic quality of their native carp stocks. About 31.3 per cent (81 records) of cyprinid introductions have no indications of the period or year the fish were brought into the country. The period of introduction was listed in FishBase as either "unknown" or left blank. The relatively high number of unknown time of transfer and unrecorded transfer of carps in the region may also be a reflection of accidental introductions or deliberate introductions without authorization from governments. In India, for instance, *A. nobilis*, *Cirrhinus molitorella* and *Mylopharyngodon piceus* have been introduced without authorization (Ponniah 1997).

6.4 Reasons for introductions

The earliest introductions of cyprinids in Asia were carried out mainly by the Chinese who "imported" both exotic species and culture systems into many Southeast Asian countries where they settled, rather than buying and consuming still relatively abundant local wild fish. Hence, it may

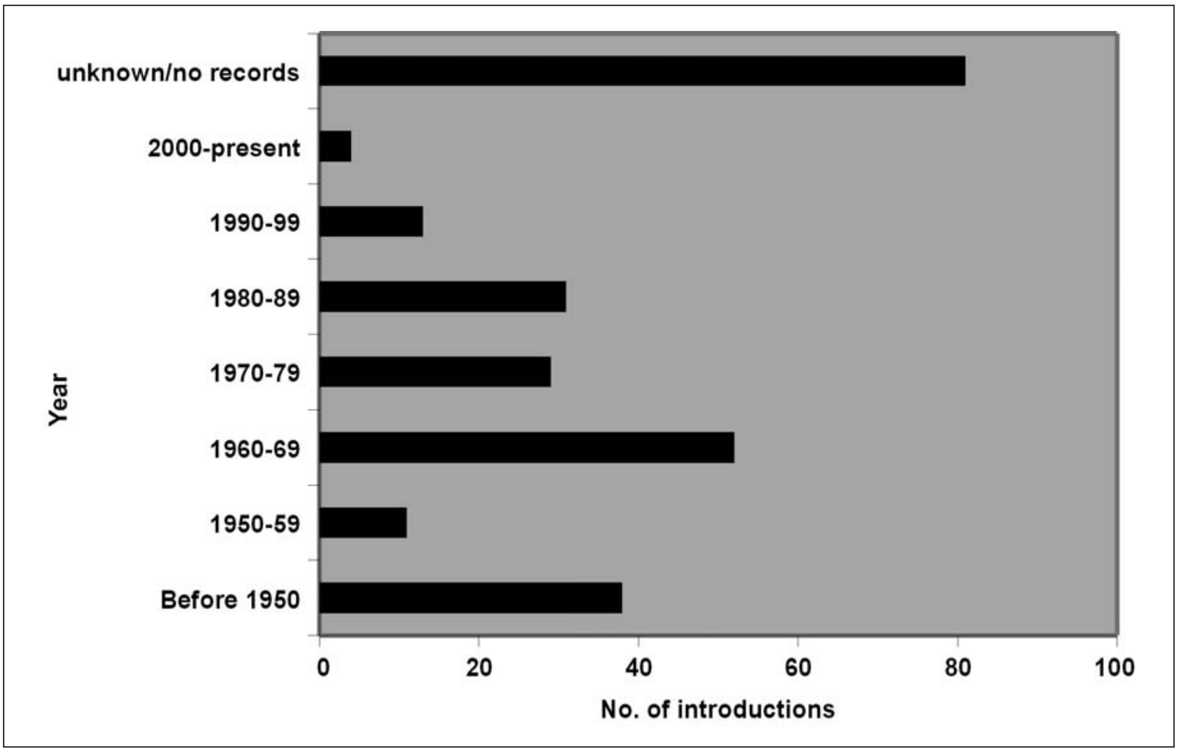

Fig. 6.2. Cyprinid introductions in Asia per decade (data based on the table in Annex 6.1)

be surmised that early introductions were due to cultural preference for Chinese carps based on nostalgia and well-established culinary tradition. Convenience may also have played a role for the predominantly urban-based Chinese in Southeast Asia, because fish culture could provide a predictable, abundant, year-round supply of fish in contrast to often seasonal capture fisheries (Edwards et al. 1997).

Subsequent to early introductions, cyprinids were introduced mainly to enhance fisheries/ aquaculture production and thereby to increase the available animal protein sources. Based on FishBase records, Fig. 6.3 summarizes the reasons for introducing carps in Asia. Use in aquaculture operations was the most frequent (69 per cent or 175 records) reason for introducing carps in all countries in the region. The main reason for introduction was primarily to broaden the species spectrum in fish farming and increase yields through better utilization of trophic niches. This reflects the greater focus given by the countries in the region to aquaculture, in contrast to developed countries where fish are most often introduced to support recreational fishery activity. The other major reasons given were ornamental (10.4 per cent or 27 records) and research (9.3 per cent or 24 records). In a few instances, phytoplankton/ weed control, snail control and angling/sport were also indicated as the purpose of the introductions (1.5 per cent or 4 records). There were also several cases of accidental or unknown reasons for the introductions (9.7 per cent or 25 records).

6.5 Effects of introductions

The known effects of introducing carps in Asia are summarized in Annex 1. These are categorized as to whether they are known to have resulted in significant ecological and socioeconomic impacts.

FAO (2001b) reported that among the cyprinids, *A. nobilis*, *C. auratus*, *Catla catla*, *Labeo rohita* and *M. piceus* are the species generally viewed as "beneficial" where introduced. In terms of socioeconomic effects, the introduction of Indian and Chinese major carps in Asian countries has paved the way for the progress made in carp aquaculture. This is more evident in the six main carp producing countries in the region: Bangladesh, China, Indonesia, India, Thailand and Vietnam. In Bangladesh, where a number of exotic major carps (*H. molitrix*, *A. nobilis*, *C. idella*, *M. piceus*, *C. carpio*, *Tor putitora* and *Barbonymus gonionotus*) were introduced for aquaculture as early as the 1950s, these species (except for *T. putitora*) have become a part of aquaculture and fishery operations (Hossain 1997), for polyculture in ponds or for stocking in floodplains, rivers and

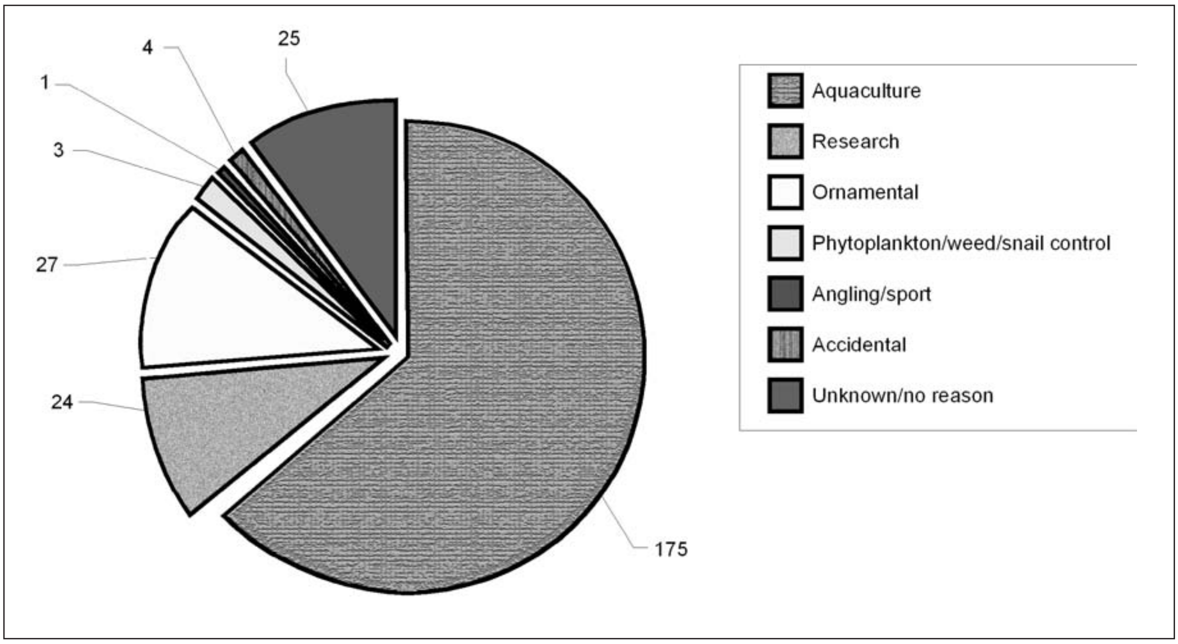

Fig. 6.3. Reasons for introduction of cyprinids based on a number of records

reservoirs to enhance natural fisheries (Gupta et al. 1997). In India, *H. molitrix* and *C. idella*, which occupy two important ecological niches hitherto vacant in the traditional polyculture system, have played a significant role in the country's aquaculture ever since their introduction in 1959 (Tripathi 1989). *H. molitrix* , *C. idella* and different strains of *C. carpio* that have been introduced into India form an important component of polyculture of indigenous and exotic species. In Indonesia, *C. carpio*, which was believed to be the first species introduced in the country, has become the most common fish cultured in freshwater ponds, cages and running water systems (Eidman 1989). In Thailand and Vietnam, the introduced *C. carpio*, *L. rohita*, and *C. cirrhosus* are used for aquaculture. For example, in northern Vietnam, *C. carpio* has been sustained because the species is bred at the household level and rice/*C. carpio* culture has been carried out locally for generations and been adapted to local circumstances (Edwards et al. 2000). Apart from *C. carpio*, the Indian major carps, *L. rohita* and *C. cirrhosus* have also become economically important cultured species in Vietnam as they are easy to breed in hatcheries and grow fast in different culture systems (Thien and Dan 1997).

While the introduction of major carp species has proven to be a boon in aquaculture and optimized the yield in ponds, the accidental and deliberate stocking of these in open waters has, in a few instances, resulted in negative ecological effects. The introduction of *C. auratus*, *C. idella*, *C. carpio* and *H. molitrix* in India is viewed as controversial in that they have created ecological problems of various kinds but are, at the same time, perceived as being extremely useful socioeconomically (FAO 2001a; FAO 2001c). These species have made a substantial contribution to food production but have also been implicated in threatening endemic species in the country. This has been the case in India where *H. molitrix* introduced in Gobindsagar Reservoir reportedly resulted in a sharp decline of the dominant native species *C. catla* due to overlapping feeding habits and habitat. Production of *H. molitrix* in the reservoir increased from 0.60 tonnes to 334 tonnes in 10 years (from 1977-78 to 1987-88), while *C. catla* production, which was 210 tonnes in 1977-78, declined to 36.7 tonnes in 1987-88. Similarly, in Kalgarhi Reservoir the *C. catla* population declined as a result of competition with *H. molitrix* (Das 1997).

In Girna and Krishnarajasagar reservoirs (India), the introduction of *C. carpio* has resulted in the decline of *Cirrhinus* sp. *C. carpio*, because of its habit of stirring up the bottom while feeding, has the reputation of muddying the waters it occupies. This shades out macrophytes, chokes benthic invertebrates, and through the more rapid recycling of phosphate contributes to accelerated eutrophication. As a result, the composition and abundance of the native fish fauna are affected (FAO 2001b).

6.6 Need for and development of a precautionary approach

Global efforts through fisheries regulations, conventions, treaties and agreements are now emerging to protect biodiversity and minimize the risks caused by human actions and development activities.

One of the risks associated with species introductions is the spread of disease. International codes of practice, technical guidelines which describe, at least in part, standardized protocols for minimizing the risks of disease associated with movements of aquatic animals are currently available (Subasinghe et al. 2001). Examples of these are the International Council for Exploration of the Sea (ICES) Code of Practice on the Introductions and Transfer of Marine Organisms (ICES 1995) and the European Inland Fisheries Advisory Commission (EIFAC) Codes of Practice and Manual of Procedures for Consideration of Introductions and Transfer of Marine and Freshwater Organisms (Turner 1988). Within the Asia-Pacific region, the Network of Aquaculture Centres in the Asia-Pacific (NACA), in cooperation with the Office International des Epizooties (OIE) and FAO have initiated measures to minimize disease risks caused by the transboundary movement of aquatic organisms. These include, among others, the establishment of a disease surveillance and reporting system in the region (Subasinghe et al. 2001) and production of a set of technical guidelines on health management for the responsible movement of live aquatic animals in Asia (FAO and NACA 2000, 2001). These efforts complemented the existing FAO Code of Conduct for Responsible Fisheries that provides a global framework for the sustainable use and conservation of biological diversity (FAO 1995).

Naylor (1994) stressed the importance of institutionalizing quarantine programs in developing countries to minimize the risks of disease and other factors that may contribute to loss of biodiversity and damage to ecosystems as a result of species introductions. Although existing agreements, international codes of practice and guidelines have placed emphasis on precautionary measures such as quarantine, these are still left to the responsibility of individual countries. Hence, in many instances of species introductions, these measures have not been fully implemented. International cooperation is one mechanism to ensure effective implementation of these regulatory measures. The INGA, which the WorldFish Center has been coordinating since 1993, provides assistance to 13 developing member countries (9 are from Asia) in the exchange of fish germplasm, for evaluation, direct use in aquaculture or utilization in breeding programs. Protocols and quarantine procedures formulated by the network based on international codes of practice and the Material Transfer Agreement are being followed in the transfer of germplasm through the network (Gupta and Acosta 2001). To avoid the risk of contaminating wild gene pools through interbreeding, it has been the policy of INGA not to transfer species that are endemic to potential recipient countries. INGA has also not made any new introductions, i.e. brood stock exchange has only been supported where a non-endemic species has previously been introduced into the recipient country.

Globally, governments and the international community have recognized that one of the major threats today to native biodiversity is the impact of alien invasive species.[5] In response to this, the IUCN has drawn up guidelines for the prevention of biodiversity loss caused by such species. Its intention is to assist all governments and management agencies to give effect to Article 8 (h) of the Convention on Biological Diversity, which states that: "Each Contracting Party 'shall, as far as possible and as appropriate: …(h) prevent the introduction of, control or eradicate those alien species which threaten ecosystems, habitats or species'" (IUCN 2001). The Conference of the Parties to the Convention on Biological Diversity elaborated this during discussions at the Sixth Meeting of the Subsidiary Body on Scientific, Technical and Technological Advice in April 2002 and this version states that "no first-time intentional introduction of an alien species should take place without the authorization from a competent authority unless it is known that an alien species poses no threat to biological diversity. A science-based risk assessment, including environmental impact assessment, should be carried out as part of the evaluation process before coming to a decision on whether or not to authorize a proposed introduction. States should make all efforts to knowingly permit only those species that are unlikely to cause unacceptable harm to ecosystems, habitats or species" (UNEP/CBD/COP 2001).

[5] Alien invasive species means an alien species that becomes established in natural or semi-natural ecosystems or habitat, is an agent of change, and threatens native biological diversity (IUCN 2001).

The WorldFish Center addresses research and policy issues that have impacts on aquatic biodiversity through its contributions to the work of the Convention on Biological Diversity, FAO, IUCN and other international bodies/ organizations (ICLARM 1999). In line with this effort and prompted by the need to promote public awareness and strict implementation of conservation measures, the WorldFish Center, in collaboration with its partners, held expert consultation meetings on ecological risk assessment of introductions of genetically improved and modified aquatic organisms and conservation of biodiversity (Gupta et al. 2004; WorldFish Center 2002, 2003). The meeting, organized in Bangladesh in 2003 (WorldFish Center 2003), recognized that existing institutional mechanisms, polices and legal frameworks related to introductions do not adequately cover issues posed by improved strains and suggested that effective institutional frameworks, monitoring and enforcement mechanisms be established at national and local levels as appropriate. The meeting also suggested that transparent, objective and practical methodologies be adopted and promoted for assessment of risks associated with the dissemination of improved strains of fish (WorldFish Center 2003).

6.7 Conclusion

The benefits derived from introductions of cyprinid species in the region are evident in terms of improving overall fish yields through aquaculture. The introductions of this group of fish for aquaculture has resulted in significant contributions to improve human nutrition and alleviate rural poverty. The development of world aquaculture has been mostly in freshwater environments (58.7 per cent) and mainly in Asia (FAO 2000). The freshwater aquaculture production in this region is dominated by finfish, particularly the major carp species.

Although problems have been reported in a few cases, none of the carp species introduced has been regarded as a pest,[6] nor have any carps caused major negative environmental impacts. However, the many cases of "unknown" introductions of carp species in the region that are probably unauthorized by the governments, as evidenced by the incomplete entries in some of the FishBase records, and the few reported negative effects of alien cyprinid species on endemic species still merit attention. This indicates that stringent regulatory measures are still lacking in many of these countries and careful assessment of the risks was probably not done when the exotic carp species were introduced. This affirms the need for implementation of control measures on movements across international boundaries and across watersheds, of any aquatic organism, even if previous introductions of such a species have been generally perceived as beneficial.

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[6] A pest is defined as an introduced species whose behavior makes it useless for anything but forage for other fish and causes a disproportionate amount of environmental nuisance (FAO 2001b).

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Annex 6.1

History of carp introductions in Asia

Country	Species	English name	Source	Year introduced	Reason for introduction	Significant ecological effects	Significant socioeconomic effects	Reference
Afghanistan	*Ctenopharyngodon idella*	Grass carp	China	-	Aquaculture	-	Yes-beneficial	Welcomme 1988
	Cyprinus carpio	Common carp	China	Unknown (1970-79)	Aquaculture	-	-	Welcomme 1988
Bangladesh	*Aristichthys nobilis*	Bighead carp	Nepal	1981	Aquaculture	-	-	Hussain and Mazid 1997
	Barbonymus gonionotus	Silver barb	Thailand	1977	Aquaculture	-	Yes-beneficial	Rahman 1989
			Thailand	1987, 1994	Research			Hussain and Mazid 1997
			Indonesia	1994	Research			Hussain and Mazid 1997
	C. idella	Grass carp	Hong Kong	1966	Aquaculture	-	-	Hussain and Mazid (this volume)
			Hong Kong	1969	Aquaculture	-	-	Welcomme 1988
			Japan	1970	Aquaculture	-	-	Hussain and Mazid 1997
			Nepal	1979	Aquaculture	-	-	Hussain and Mazid 1997
	C. carpio	Common carp	China	1960	Aquaculture	-	-	Hussain and Mazid (this volume)
		Common carp	Hungary	1982, 1996	Aquaculture	-	-	Hussain and Mazid (this volume)
		Common carp	Vietnam	1995	Research	-	-	INGA 2002
	C. carpio	Mirror carp	Nepal	1979	Aquaculture	-	-	Hussain and Mazid 1997
	Hypopthalmichthys molitrix	Silver carp	Hong Kong	1969	-	-		Hussain and Mazid (this volume)
	Mylopharyngodon piceus	Black carp	China	1983	Aquaculture	-	-	Hussain and Mazid (this volume)
	Tor putitora	Putitor Mahseer	Nepal	1991	Unknown	Unknown	Unknown	FAO 1997; Hussain and Mazid (this volume)
Bhutan	*A. nobilis*	Bighead carp	Nepal	1983	Aquaculture			Csavas 1983
	Cirrhinus cirrhosus	Mrigal	Unknown	1985	Aquaculture	-	-	Welcomme 1988
	C. idella	Grass carp	Nepal	1983	Aquaculture	-	-	Csavas 1983

	C. carpio	Common carp	Nepal	1983	Aquaculture	-	-	Csavas 1983
	H. molitrix	Silver carp	Unknown	1984	-	No		
	Labeo rohita	Rohu	India	1985	Aquaculture	-	-	Welcomme 1988
	T. putitora	Putitor mahseer	India	1969	Ornamental	Unknown	Unknown	FAO 1997
Brunei	*C. carpio*	Common carp	Unknown	-	Aquaculture	-	Yes-beneficial	FAO 1996
Cambodia	*C. idella*	Grass carp	Unknown	-	Aquaculture	-	-	Davidson 1975
	C. carpio	Common carp	Unknown	-	Aquaculture	-	-	Alikunhi 1966
China	*A. nobilis*	Bighead carp	Hong Kong	Unknown	Aquaculture			
	B. gonionotus	Silver barb	Thailand	1986	Aquaculture	-	Yes-beneficial	Welcomme 1988
	Carassius auratus	Goldfish	Japan	1976	Aquaculture	-	Yes-beneficial	Welcomme 1988
	C. cuvieri	-	Japan	1976	Aquaculture	-	-	Tan, Y. and H.E. Tong 1989
	Catla catla	Catla	India	1973	Aquaculture	Probably no-undecided	Unknown	FAO 1997
			Bangladesh	1973	Aquaculture	Probably no-undecided	Unknown	Tan, Y. and H.E. Tong 1989
	C. carpio	Common carp	F. Germany	1982	-	-	-	Tan Jo-Jun and Tong He-Yi 1989
	L. rohita	Rohu	Thailand	1978	Aquaculture	Probably no-undecided	Probably no-beneficial	FAO 1997
	Orthodon microlepidotus	Sacramento blackfish	USA	1982	Aquaculture	-	-	Welcomme 1988
	Pseudorasbora parva	Stone moroko	Unknown	Unknown (1970-79)	Accidental	Yes-adverse	-	Kottelat and Whitten 1996
Hong Kong	*C. molitorella*	Mud carp	Unknown	-	Aquaculture	-	-	Roberts 1997
	C. idella	Grass carp	China	Unknown	Aquaculture	-	-	Shireman and Smith 1983
	C. carpio	Common carp	Unknown	-	Aquaculture	-	-	Man. and Hodgkiss 1981
	Opsariichthys bidens	-	China	1994	Aquaculture	Probably no-undecided	Unknown	FAO 1997
India	*A. nobilis*	Bighead carp	Bangladesh	1987	Aquaculture	Probably yes-undecided	Probably no-undecided	FAO 1997
			Japan	1987	Aquaculture	Probably yes-undecided	Probably no-undecided	FAO 1997
	B. gonionotus	Silver barb	Indonesia	1972	Aquaculture	-	-	Shetty, Nandeesha and Jhingran 1989
	C. auratus	Goldfish	Japan	Unknown	Ornamental	Yes-undecided	Probably no-undecided	FAO 1997
	C. carassius	Crucian carp	UK	1870	Ornamental	Probably yes-adverse	Probably yes-undecided	FAO 1997

	C. idella	Grass carp	Hong Kong	1959	Aquaculture	Probably no-beneficial	Yes-beneficial	FAO 1997
	C. carpio (German strain)	Common carp	Sri Lanka	1939	Aquaculture	Yes-adverse	Yes-beneficial	Jhingran 1991; FAO 1997
	(Bangkok strain)		Thailand	1957	Aquaculture	-	-	Jhingran 1991
			Vietnam	1995	Research	-	-	INGA 2002
			Hungary	2000	Research	-	-	INGA 2002
	(wild Amur carp)		Hungary	2000	Research	-	-	INGA 2002
			Indonesia	2001	Research	-	-	INGA 2002
	H. molitrix	Silver carp	Japan	1959	Aquaculture	Probably yes-adverse	Probably yes-beneficial	Shetty et al. 1989
	H. molitrix	Silver carp	Hong Kong	1959	Aquaculture	Probably yes-adverse	Probably yes-adverse	Shetty et al. 1989
	Puntius lateristriga	Spanner barb	Malaysia	-	Unknown; ornamental	Probably no-undecided	Probably no-undecided	FAO 1997
	P. oligolepis	Checkered barb	Indonesia	-	Ornamental	Probably no-undecided	Probably no-undecided	FAO 1997
	P. semifasciolatus	Chinese barb	Japan	-	Ornamental	Probably no-undecided	Probably no-undecided	FAO 1997
	P. tetrazona	Sumatra barb	Unknown	-	Ornamental	Probably no-undecided	Probably no-undecided	FAO 1997
	Tinca tinca	Tench	UK	1870	Phytoplankton control	-	-	Welcomme 1988
Indonesia	*A. nobilis*	Bighead carp	Taiwan	1969	Aquaculture			Eidman 1989
	B. gonionotus	Java barb	Unknown	1963	Aquaculture			
	C. auratus	Goldfish	China	Unknown	Aquaculture	-	Yes-beneficial	Welcomme 1988
		Goldfish	Taiwan	1987	Research	-	-	Eidman 1989
	C. chinensis	Chinese mud carp	Taiwan	1969	Aquaculture	-	Yes-beneficial	Eidman 1989
			Unknown	-	Aquaculture	-	-	Roberts 1997
	C. idella	Grass carp	Singapore	1915	Research	-	-	Welcomme 1988
			Thailand	1915	Research	-	-	Welcomme 1988
			Japan	1915	Research	-	-	Welcomme 1988
			Malaysia	1915	Research	-	-	Welcomme 1988
	C. carpio	Common carp	China	-	Aquaculture	-	Yes-beneficial	Welcomme 1988
			Netherlands	1927	Aquaculture	-	-	Eidman 1989
			Taiwan					Schuster 1950
			Japan					Schuster 1950
			Germany					Schuster 1950
	H. molitrix	Silver carp	China, Japan	1964	Research	-	-	Eidman 1989
					Research	-	-	Eidman 1989

			Taiwan	1967	-	-	-	Eidman 1989
	C. molitorella	Mud carp	Malaysia and Singapore	1915	Aquaculture	-	-	Eidman 1989
	Osteochilus hasseltii	Silver sharkminnow	Indonesia	1937	Unknown	-	-	Welcomme 1988
	T. tinca	Tench	Netherlands	1927	Unknown	-	-	Welcomme 1988
Iran	*A. nobilis*	Bighead carp	unknown	-	Aquaculture			
	C. auratus	Goldfish	unknown	-	Aquaculture	-	-	Coad 1996
	C. carpio	Common carp	unknown	-	Aquaculture	-	Yes-beneficial	Coad 1996
	H. molitrix	Silver carp	USA	-		Probably no	Probably yes	
			Unknown	-	Fisheries	Yes-adverse	-	Coad 1996
Iraq	*C. auratus*	Goldfish	Unknown	-	Aquaculture	Probably yes-beneficial	Unknown	FAO 1997
	C. idella	Grass carp	Japan	1968	Aquaculture	-	-	Shireman and Smith 1983
	H. molitrix	Silver carp	Unknown	1966-69	-	Yes	Probably no	
Israel	*A. nobilis*	Bighead carp	Germany	1973	Aquaculture	-	-	Golani and Mires 2000
	C. auratus	Goldfish	Germany	1985	Ornamental	-	-	Golani and Mires 2000
	C. carassius	Crucian carp	Unknown	-	Angling/sport	-	-	Welcomme 1988
	C. catla	Catla	India	1953	Aquaculture	-	-	Golani and Mires 2000
	C. idella	Grass carp	China	1965	Weed control	-	-	Golani and Mires 2000
	C. carpio	Common carp	Yugoslavia	1927-28	Aquaculture	-	Yes-beneficial	Golani and Mires 2000
	H. molitrix	Silver carp	Japan	1966	Aquaculture	-	Yes-beneficial	Golani and Mires 2000
	M. piceus	Black carp	Unknown	-	Snail control	-	-	Golani and Mires 2000
Japan	*A. nobilis*	Bighead carp	China	Unknown	Aquaculture			
	C. catla	Catla	India	1960	Aquaculture	-	-	Welcomme 1988
			Pakistan	1960	Aquaculture	-	-	Welcomme 1988
	C. chinensis	Chinese mud carp	China	1965	Aquaculture	-	-	Welcomme 1988
	C. cirrhosus	Mrigal	India	1960	Aquaculture	-	-	Chiba et al. 1989
	C. idella	Grass carp	China	1878	Fisheries	-	-	Chiba et al. 1989
	C. carpio	Common carp	Germany	1905	-	-	-	Chiba et al. 1989
	H. molitrix	Silver carp	Nepal	1967		Unknown	Unknown	
			China	1878-1940	Accidental	-	-	Chiba et al. 1989
	L. rohita	Rohu	India	1960	Aquaculture	-	-	Welcomme 1988
	M. piceus	Black carp	China	1970	Accidental	-	-	Welcomme 1988
	T. tinca	Tench	Netherlands	1961	Unknown	-	-	-
Jordan	*A. nobilis*	Bighead carp	Germany	1973	Aquaculture			Krupp and Schneider 1989

	C. auratus	Goldfish	Unknown	-	Unknown	-	-	Krupp and Schneider 1989
	C. catla	Catla	Unknown	-	Unknown	-	-	Krupp and Schneider 1989
	C. idella	Grass carp	Japan	1965	Unknown	-	-	Krupp and Schneider 1989
	C. carpio	Common carp	Yugoslavia	Unknown (1931-34)	Aquaculture	-	Yes-beneficial	Krupp and Schneider 1989
	H. molitrix	Silver carp	Japan	1966	-			Krupp and Schneider 1989
	M. piceus	Black carp	Unknown	-	Unknown	-	-	Krupp and Schneider 1989
Korea	*A. nobilis*	Bighead carp	Taiwan	1963	Aquaculture		-	Welcomme 1988
	C. auratus	Goldfish	Japan	1972	Aquaculture	-	Yes-beneficial	Welcomme 1988
	C. idella	Grass carp	Japan	1963	Aquaculture	-	-	Welcomme 1988
	C. carpio	Common carp	Israel	1973	Aquaculture	-	-	Welcomme 1988
	H. molitrix	Silver carp	Japan	1963	Aquaculture	-	-	Welcomme 1988
Lao PDR	*C. catla*	Catla	Thailand	1977	Aquaculture	-	-	ICCILMB 1981*
	C. cirrhosus	Mrigal	India	1977	Aquaculture	-	-	ICCILMB 1981
			Thailand	1977	Aquaculture			ICCILMB 1981
	C. idella	Grass carp	Unknown	-	Aquaculture	-	-	Davidson 1975
	C. carpio	Common carp	India	1977	Aquaculture	-	-	ICCILMB 1981
			Thailand	1977	Aquaculture	-	-	ICCILMB 1981
	C. carpio (Hungarian)	Common carp	Vietnam	1996, 1998	Aquaculture	-	-	INGA 2002
	L. rohita	Rohu	India	1977	Aquaculture	-	-	
Malaysia	*A. nobilis*	Bighead carp	China	19th century	Aquaculture	-	Yes-beneficial	Welcomme 1988
	B. gonionotus	Silver barb	Indonesia	1958	Aquaculture (polyculture in ponds)			
	C. catla	Catla	India	1960	Aquaculture	-	-	Ang et al. 1989
	C. chinensis	Chinese mud carp	China	-	Aquaculture	-	-	Welcomme 1988
	C. cirrhosus	Mrigal	India	1960	Aquaculture	-	-	Welcomme 1988
	C. idella	Grass carp	China	Unknown (18th century)	Aquaculture	-	-	Welcomme 1988
	C. carpio	Common carp	China	Unknown (18th century)	Aquaculture	-	Yes-beneficial	Welcomme 1988
	H. molitrix	Silver carp	China	18th century	Aquaculture		Yes-beneficial	FAO 1997
	L. rohita	Rohu	India	1960	Aquaculture	-	-	Welcomme 1988

Myanmar	*C. idella*	Grass carp	India	1969	Aquaculture	-	-	Shireman and Smith 1983
	C. carpio	Common carp	Unknown	-	Aquaculture	-	-	Alkunhi 1966
	Devario malabaricus	Malabar danio	Unknown	-	Ornamental	-	-	Welcomme 1988
Nepal	*A. nobilis*	Bighead carp	Hungary	1971	Aquaculture	Unknown	Probably yes-beneficial	FAO 1997
	C. carassius	Crucian carp	Unknown	-	Aquaculture	-	Yes-beneficial	Shrestra 1994
	C. idella	Grass carp	India	1965	Aquaculture	Unknown	Probably no-beneficial	FAO 1997
	C. carpio	Common carp	India	1956	Aquaculture	Unknown	Probably yes-beneficial	FAO 1997
	H. molitrix	Silver carp	India	1965	Aquaculture	Unknown	Probably yes-beneficial	FAO 1997
			Japan	1967	Aquaculture	Unknown	Probably beneficial	Manandhar 1995
Pakistan	*A. obilis*	Bighead carp	China	Unknown	Unknown			
	C. catla	Catla	India	Unknown	Aquaculture	-	-	Welcomme 1988
	C. cirrhosus	Mrigal	India	Unknown	Aquaculture	-	-	Welcomme 1988
	C. idella	Grass carp	China	1964	Aquaculture; weed control	Probably no	Probably yes-beneficial	FAO 1997
	C. carpio	Common carp	UK	1964	Aquaculture	Unknown	Yes-beneficial	FAO 1997
			Thailand	1964	Aquaculture	Unknown	Yes-beneficial	FAO 1997
	H. molitrix	Silver carp	China	1982-83	Aquaculture	Yes	Probably no	FAO 1997
			Nepal	1982-83	Aquaculture	Yes	Probably no	FAO 1997
	L. rohita	Rohu	India	-	Aquaculture	-	-	Welcomme 1988
Philippines	*A. nobilis*	Bighead carp	Taiwan	1968	Aquaculture			Juliano et al. 1989
	Balantiocheilos melanopterus	Tricolor sharkminnow	Unknown	Unknown (1970-79)	Ornamental			
	Danio albolineatus	-	Unknown	Unknown	Ornamental			Aquarium Science Association of the Phil., Inc. (ASAP) 1996
	B. gonionotus	Silver barb	Indonesia	1956	Aquaculture (introduced as pituitary donor)			Juliano et al. 1989
	Barbonymus schwanenfeldii	Tinfoil barb	Unknown	Unknown	Ornamental	-	-	ASAP 1996
	C. auratus	Goldfish	Unknown	Unknown	Ornamental	-	-	ASAP 1996
	C. carassius	Crucian carp	Unknown	1964	Aquaculture	-	Yes-beneficial	Welcomme 1988
	C. catla	Catla	India	1967	Aquaculture	-	-	Juliano et al. 1989
	C. cirrhosus	Mrigal	India	1967	Aquaculture	-	Yes-beneficial	Welcomme 1988

	C. idella	Grass carp	China	1964	Aquaculture	-	Probably yes-beneficial	Welcomme 1988
	C. carpio	Common carp	Hong Kong	1915	Aquaculture	-	Yes-beneficial	Juliano et al. 1989
			Vietnam	1995	Research	-	-	INGA 2002
	Devario aequipinnatus	Giant danio	Unknown	Unknown	Ornamental	-	-	ASAP 1996
	D. devario	Sind danio	Unknown	-	Ornamental	-	-	ASAP 1996
	D. malabaricus	Malabar danio	Unknown	Unknown	Ornamental	-	-	ASAP 1996
	D. rerio	Zebra danio	Unknown	Unknown	Ornamental	-	-	ASAP 1996
	H. molitrix	Silver carp	China	1964	Aquaculture	-	-	Welcomme 1988
			Taiwan					
	L. rohita	Rohu	India	1964	Aquaculture	-	-	Welcomme 1988
	Leptobarbus hoevenii	Mad barb	Unknown	(1980-89)	Ornamental	-	-	ASAP 1996
	O. hasseltii	Silver sharkminnow	Indonesia	1957	Unknown	-	-	Juliano et al. 1989
	P. conchonius	Rosy barb	Unknown	-	Ornamental	-	-	ASAP 1996
	P. lateristriga	Spanner barb	Unknown	-	Ornamental	-	-	ASAP 1996
	P. tetrazona	Sumatra barb	Unknown	-	Ornamental	-	-	ASAP 1996
	P. titteya	Cherry barb	Unknown	(1970-79)	Ornamental	-	-	ASAP 1996
	Rasbora caudimaculata	Greater scissortail	Unknown	-	Ornamental	-	-	ASAP 1996
	R. dorsiocellata	Eyespot rasbora	Unknown	-	Ornamental	-	-	ASAP 1996
	R. elegans	Twospot rasbora	Unknown	-	Ornamental	-	-	ASAP 1996
	Tanichthys albonubes	White cloud mountain minnow	Unknown	-	Ornamental	-	-	ASAP 1996
Singapore	*A. nobilis*	Bighead carp	China	Unknown	Aquaculture			
	C. auratus	Goldfish	Japan	Unknown (1900-97)	Ornamental	Probably yes-beneficial	Probably yes-beneficial	FAO 1997
			China	Unknown	Ornamental	Probably yes-beneficial	Probably yes-beneficial	FAO 1997
	C. chinensis	Chinese mud carp	China	Unknown	Aquaculture	-	-	Welcomme 1988
			Unknown	Unknown	Aquaculture	-	-	Roberts 1997
	C. idella	Grass carp	China	Unknown	Aquaculture	-	-	Welcomme 1988
	C. carpio	Common carp	China	Unknown	Aquaculture	-	-	Welcomme 1988
	P. semifasciolatus	Chinese barb	China	Unknown	Accidental	-	-	Lever 1996
	H. molitrix	Silver carp	China	Unknown	-	No	-	
Sri Lanka	*A. nobilis*	Bighead carp	China	1948	Aquaculture			
	B. gonionotus	Silver barb	Indonesia	1968	Aquaculture	-	-	Welcomme 1988
	C. carassius	Crucian carp	Europe	1915	Unknown	-	-	Welcomme 1988
	C. catla	Catla	India	1942	Aquaculture	Unknown	Probably yes-beneficial	FAO 1997

	C. cirrhosus	Mrigal	India	1981	Aquaculture	Unknown	-	Welcomme 1988
	C. idella	Grass carp	China	1948	Aquaculture; weed control	Unknown	Unknown	FAO 1997
	C. carpio	Common carp	Europe	1915	Aquaculture	Unknown	Unknown	FAO 1997
	H. molitrix	Silver carp	China	1948	Aquaculture	Unknown	Unknown	De Zylva 1994
	L. rohita	Rohu	India	1981	Aquaculture	-	-	Welcomme 1988
Syria	*C. carpio*	Common carp	Unknown	Unknown	Aquaculture	-	-	Welcomme 1988
Taiwan	*A. nobilis*	Bighead carp	China	Unknown	Aquaculture			
	C. auratus	Goldfish	China	Unknown	Aquaculture	-	Yes-beneficial	Welcomme 1988
	C. cuvieri	-	Japan	-	Aquaculture	-	-	Shen (ed.) 1993
	C. chinensis	Chinese mud carp	China	-	Aquaculture	-	Yes-beneficial	Welcomme 1988
	C. molitorella	Mud carp	Unknown	-	Aquaculture	-	-	Roberts 1997
	C. idella	Grass carp	China	Unknown	Aquaculture	-	Yes-beneficial	Welcomme 1988
	C. carpio	Common carp	Japan	Unknown	Aquaculture	-	Yes-beneficial	Welcomme 1988
	L. hoevenii	Mad barb	Indonesia	1979	Research	-	-	Welcomme 1988
	M. piceus	Black carp	China	-	Aquaculture	-	Yes-beneficial	Welcomme 1988
	H. molitrix	Silver carp	China	Pre-18th century	Aquaculture		No	Welcomme 1988
Thailand	*A. nobilis*	Bighead carp	China	1932	Aquaculture		No	Welcomme 1988
	C. auratus	Goldfish	China	Unknown	Ornamental			Welcomme 1988
	C. carassius	Crucian carp	Japan	1980	Aquaculture	-	-	Welcomme 1988
	C. catla	Catla	Bangladesh	1979	Aquaculture	-	-	Welcomme 1988
	C. chinensis	Chinese mud carp	China	-	Aquaculture	Unknown	-	Welcomme 1988; Rainboth 1996
	C. cirrhosus	Mrigal	Japan	1980	Aquaculture	-	-	Piyakarnchana 1989
			Bangladesh	1980	Aquaculture			
			Lao PDR	1982	Aquaculture			
	C. idella	Grass carp	China	1922	Aquaculture			
			China	1932	Aquaculture	No	-	Welcomme 1988
			Hong Kong	1932	Aquaculture	No	-	Welcomme 1988
	C. carpio	Common carp	China	1912, 1913	Aquaculture	-	Yes-beneficial	Welcomme 1988
			Germany	1913	Aquaculture	-	Yes-beneficial	Welcomme,1988
			Japan	1913	Aquaculture	-	Yes-beneficial	Welcomme 1988
			Japan	1964, 1971	Aquaculture			
			Germany	1970	Aquaculture			

			Hungary	1985	Aquaculture			
			Israel	-	Aquaculture	-	Yes-beneficial	Welcomme 1988
	(selected strain)		Vietnam	1997-98	Research	-	-	Pongthana (this vol.)
	(*Rajadanu* strain)		Indonesia	1998	Research	-	-	INGA 2002
	H. molitrix	Silver carp	China	1913	Aquaculture	Yes-beneficial	Yes-beneficial	FAO 1997
	H. molitrix	Silver carp	China	1932	Aquaculture			
	L. rohita (wild population)	Rohu	India	1968-69	Aquaculture	-	-	Welcomme 1988; Pongthana (this vol.)
	L. rohita (genetically improved)	Rohu	India	1998	Research	-	-	
	M. piceus	Black carp	China	1913	Aquaculture	-	-	Welcomme 1988
Vietnam	*A. nobilis*	Bighead carp	China	1958	Aquaculture	Yes-beneficial	Yes-beneficial	FAO 1997
	C. auratus	Goldfish	China	Unknown	Aquaculture	-	Yes-beneficial	Welcomme 1988
	C. catla	Catla	Lao PDR	1984	Aquaculture	Probably yes-undecided	Probably yes-undecided	FAO 1997
	C. cirrhosus	Mrigal	Lao PDR	1984	Aquaculture	Yes-beneficial	Yes-beneficial	FAO 1997
			India	1997	Research	-	-	INGA 2002
	C. idella	Grass carp	China	1958	Aquaculture	Yes-beneficial	Yes-beneficial	FAO 1997
	C. carpio	Common carp	Hungary	1971, 1975	Aquaculture	Yes-beneficial	Yes-beneficial	FAO 1997, Thien (this vol.)
			Indonesia	Before 1975	-	-	-	Thien, this vol.
	H. molitrix	Silver carp	China	1958, 1964, 2000	Aquaculture	Yes-beneficial	Yes-beneficial	FAO 1997; Thien (this vol.)
	L. rohita	Rohu	Thailand	1982	Aquaculture	Yes-beneficial	Yes-beneficial	FAO 1997
			Lao PDR	1982	Aquaculture	Yes-beneficial	Yes-beneficial	FAO 1997

*ICCILMB 1981 = Interim Committee for Coordination of Investigations of the Lower Mekong Basin, 1981.

Chapter 7
Threatened Cyprinid Species In Asia[1]

Belen O. Acosta[2] and Modadugu V. Gupta[2]

7.1 Introduction

Asia is home to about 40 per cent of the world's species of flora and fauna. The region's aquatic environment is among the most diverse in the world (ADB 2000). It has rich native carp species diversity with a total of 1 333 cyprinid species (Froese and Pauly 2001). Of these, 15 are of major aquaculture importance and contributed 49 per cent of total aquaculture production in the region in 2001. The Chinese carps: silver carp (*Hypopthalmichthys molitrix*), grass carp (*Ctenopharyngodon idella*), bighead carp (*Aristichthys nobilis*), common carp (*Cyprinus carpio*), and crucian carp (*Carassius carassius*) and Indian major carps: rohu (*Labeo rohita*), catla (*Catla catla*) and mrigal (*Cirrhinus cirrhosus*) have continuously dominated global carp production and accounted 71.3 per cent of the total freshwater aquaculture worldwide (FAO 2003). This indicates the important role of carp resources as a major provider of fish protein not only to growing populations in Asia but also to the entire world.

Despite the considerable potential for continued growth in cyprinid aquaculture production in Asia, there are increasing concerns about the status of native cyprinids especially those that are important to aquaculture in the region. During the last 25 years, Asia has seen remarkable economic and social transformation that consequently has resulted in increases in developmental activities and enormous pressure on the region's natural resources. Degradation of the aquatic environment and water pollution have taken their toll on its biological resources and threatened its rich natural diversity (ADB 2000).

In view of the importance of cyprinids to the present and future status of Asian aquaculture (small-scale and industrial scale), it is necessary to have information on endangered/threatened species to assist with the region's conservation programs. This chapter reports on the status of such cyprinids in Asia and the factors that are threatening them.

7.2 Threatened species

Globally, 179 cyprinid species have been recorded as threatened (Froese and Torres 1999). The World's Conservation Union's (IUCN) Red Book and country reports list 56 cyprinid species as "threatened" in 14 countries in Asia (Annex 7.1). However, it is likely that the figure is an underestimate. This does not appear to be a very comprehensive assessment of freshwater fish fauna and hence there is the possibility of more species than those reported being either extinct or threatened (Bruton 1995; Gupta 1999; Froese and Torres 1999).

Of the 56 threatened cyprinid species listed in Annex 7.1, 40 are from the IUCN Red list and the other 16 species are from country reports. The genera with the most threatened endemic species in the region are *Puntius* (20 species) and *Labeo* (8 species). This list of threatened cyprinid species also indicates that the natural range of distribution of five of these cyprinids extends to more than one country in the region. These species are *Amblypharyngodon microlepis, Balantiocheilos melanopterus, Chela caeruleostigmata, Labeo porcellus* and *Probarbus jullieni* (Annex 7.1, Table 7.1).

Based on Annex 7.1, 14 species are categorized as vulnerable, 19 as endangered, 22 as critically endangered and one as extinct (Fig. 7.1). The vulnerable, endangered and critically endangered categories[3] in this paper are from the IUCN Red list and follow the criteria for classifying

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[1] WorldFish Center Contribution No. 1732.
[2] WorldFish Center, Jalan Batu Maung, Batu Maung, 11960 Bayan Lepas, Penang, Malaysia.
[3] Definitions by the IUCN (Groombridge 1993): **endangered** – taxa in danger of extinction and whose survival is unlikely if the causal factors continue operating. Included are taxa whose numbers have been reduced to a critical level or whose habitats have been so drastically reduced that they are deemed to be in immediate danger of extinction. Also included are taxa that may be extinct but have been definitely been seen in the wild in the past 50 years; **vulnerable**- taxa that are likely to move into the "endangered" category in the near future if the causal factors continue operating. Included are the taxa of which most or all of the population(s) are decreasing because of over-exploitation, extensive destruction of habitat or other environmental disturbances, whose ultimate security has not been assured; and taxa with population(s) that are still abundant but are under threat from severe adverse factors throughout their range.

Table 7.1. Threatened cyprinid species that are endemic to more than one country in Asia (based on IUCN records of threatened species in FishBase)

Name of species	Countries	Conservation status
Amblypharyngodon microlepis	Bangladesh, India	Vulnerable
Balantiocheilos melanopterus	Brunei Darussalam, Cambodia, Indonesia, Lao PDR, Malaysia, Thailand and Vietnam	Endangered
Chela caeruleostigmata	Cambodia, Lao PDR, Thailand	Critically endangered
Labeo porcellus	India, Sri Lanka	Critically endangered
Probarbus jullieni	Cambodia, Lao PDR, Thailand, Vietnam	Endangered

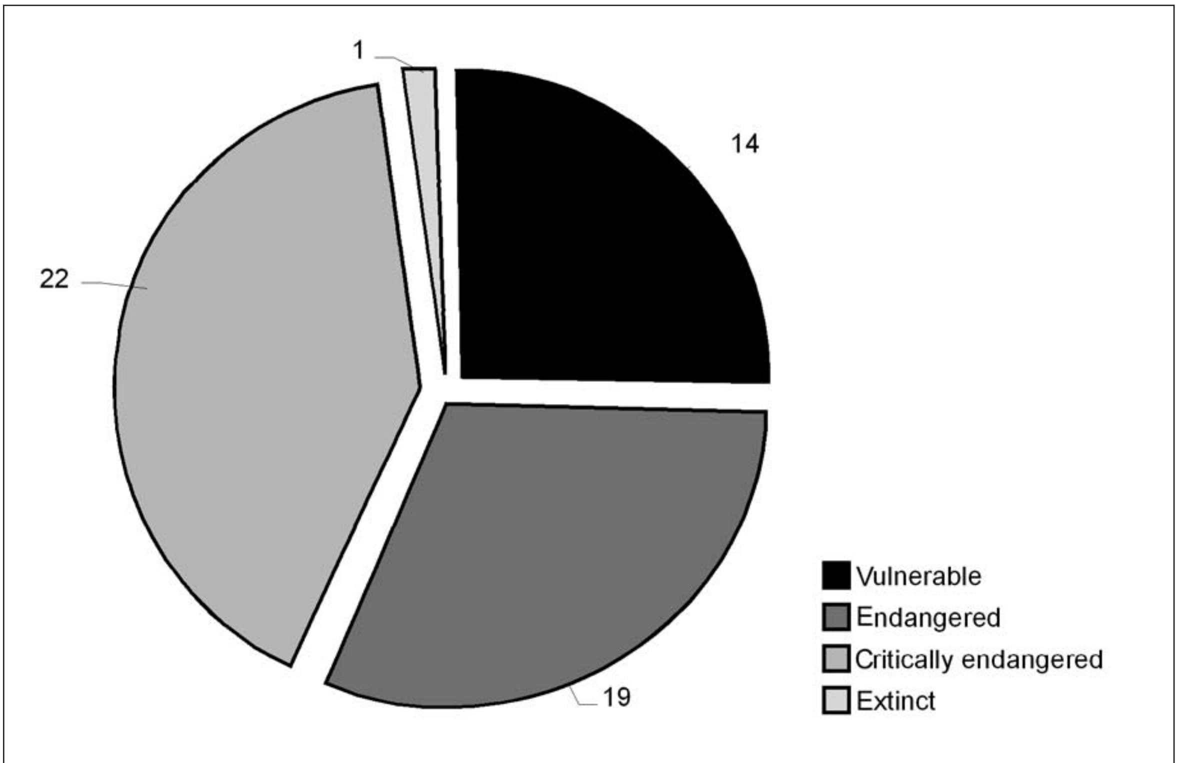

Fig. 7.1. Number of threatened cyprinid species per category in Asia (based on IUCN records of threatened species in FishBase and country reports).

threatened species. These criteria are based on biological factors related to rate of decline, population size, geographic distribution, and degree of population and distribution fragmentation (IUCN 2001). However, some of the endangered and extinct categories reported in this paper are from country reports (Bangladesh and China) and information is lacking as to the basis on which these countries have made these classifications.

In terms of global importance, 12 of the 56 threatened cyprinid species in the region are classified in FishBase as being of commercial importance or of potential interest to aquaculture. These species belong to the genera *Labeo* (5 species), *Puntius* (2 species), *Tor* (2 species), *Balantiocheilos, Chela* and *Probarbus* (1 species each) (Annex 7.1).

Based on the IUCN records of threatened species in FishBase, Bangladesh, China and the Philippines are the countries in Asia with the highest number of threatened cyprinid species. Among the Asian countries, only China (Yunnan Province) has an endemic cyprinid species (*Cyprinus yilongensis*) that has been listed as "extinct" (Xie and Chen 2001). This confirms reports that China's aquatic biodiversity is under stress: it has been estimated that some 98 fish species are either extinct, endangered or threatened in China (Li 1997). In the Philippines, many species of the genus *Puntius* that are endemic in streams and lakes have been driven near to the brink of extinction (WWF 2004a). IUCN (Harrison and Stiassny 1999) considers 12 of the 18 cyprinid species listed in Annex 7.1 from the Philippines to be "possibly extinct".

7.3 Threats to native diversity of cyprinids

One of the factors that can threaten native species is the adverse impact of introduced species, including predation, disruption of ecological processes, competition for food and space, infestation with alien parasites and diseases, habitat degradation and hybridization (Bruton 1995). Indiscriminate introductions of aquatic organisms from one habitat into another, where they have never been before, have serious risks and represent a significant threat to aquatic biodiversity (Welcomme 1988).

In freshwater and estuarine environments, species extinctions, and threatened and endangered aquatic species are most common and mainly result from habitat loss and degradation (Smith 1999; Froese and Torres 1999). This holds true for cyprinids in Asia, where the greatest threat to endemic populations stems from degradation of aquatic ecosystems more than the effects of species introductions.

For instance in India, there is depletion in the populations of almost all economically important fish species including major carps in all the rivers. This appears to be mainly due to man-made stresses through over-fishing and alteration of the environment through the construction of dams, hydro-power plants, etc., which prevents the fish from migrating to their natural breeding and feeding grounds (Reddy 1999). Sarkar and Ponniah (2000) reported that in northeast India alone, 22 endemic cyprinid species are categorised as threatened. Another 22 cyprinid species have also been reported to be threatened in the Western Ghats, India (Shaji et al. 2000).

IUCN (2002) confirmed that the major native cyprinid species in South Asian countries that breed naturally in flowing river waters are at great long-term risk due to destruction of the region's ecosystem of rivers. These include pollution, inadequate water flow during lean season due to draw-off for irrigation, lack of bypass facilities in weirs, barrages and dams, use of explosives and fine mesh nets, bank-to-bank net laying and other destructive practices, lack of controls and/or enforcement, etc. Asian Development Bank (2000) reported that Asian rivers have 4 times more than the world average of suspended solids and 1.4 times the biological oxygen demand.

In Bangladesh, the natural cyprinid stocks have declined due to degradation of aquatic environments and reduction of many wetlands and water areas due to natural causes and man-made changes resulting in the loss of many habitats of riverine and floodplain endemic species (Hussain and Mazid 1997). More recently, concerns have been raised about the threat to wild endemic fish populations from large-scale induced breeding operations, inappropriate artificial reproduction practices and the large-scale stocking of domesticated, genetically degraded fingerlings in floodplains, and large-scale escapes of cultured stocks due to flooding (Rajts et al. 2002).

WWF (2004b) reported that in China, the strong pressure of the rapidly increasing human population and, with most of it concentrated around river courses, the large-scale infrastructures such as dams and aquatic pollution have brought severe damage to the Yangtze River's remaining biodiversity.

In the Philippines, the near extinction of some native cyprinid species (*Puntius lindog*, *P. baoulan* and *P. tumba*) in Lake Lanao has been attributed to soil erosion due to mining and agricultural development that has degraded water quality and to introduction of alien aquatic species such as common carp (*Cyprinus carpio*) and goby (*Glossogobius giurus*) (WWF 2004a).

7.4 Conclusion

The threat to cyprinids and other genetic resources in Asia and elsewhere is likely to worsen if pressures imposed on their populations due to environmental degradation, over-exploitation, disturbance by the introduction of alien species, etc., continue.

If the contribution of carp aquaculture to total fish production is to be sustained, there has to be an emphasis on the conservation of carp genetic resources in the region. IUCN (1996), however, cautioned that the category of threat is not necessarily sufficient to determine priorities for conservation action. It should also include numerous other factors concerning conservation action such as costs, logistics, chances of success and taxonomic distinctiveness of the species.

Reddy (1999) stressed that in view of the deteriorating aquatic environment, the first necessary step should be *in situ* conservation of germplasm resources of at least all the economically important species. IUCN's Global Biodiversity Strategy calls for the establishment of national strategies in all countries and a comprehensive world *in situ* and *ex situ* genetic conservation system to be operational by 2010 (Bruton 1995). While governments in Asia have now realized the importance of living aquatic resources, only in recent years have some governments initiated the process for developing national policies and plans for conservation and utilization of biodiversity. In addition to enactment of national laws/regulations for conservation, there is a need for regional collaboration in the implementation of regulations especially for countries in the regions with common watersheds/borders and endemic species (Gupta 1999). An example is mahseer (*Tor putitora*), a species that is native to Pakistan, Northern India, Nepal and Bangladesh. The present plight of the species, which is critically endangered in Bangladesh, has become a matter of serious concern to South Asian countries. To address this problem, the IUCN South Asia Sustainable Use Specialist Group will develop a Mahseer Conservation and Management Plan and will play a leading role in river conservation projects (IUCN 2002).

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Annex 7.1

Threatened cyprinid species in different countries in Asia

Country	Species	Status	Global importance	Reference
Bangladesh	*Amblypharyngodon microlepis*	Vulnerable*	Low importance in fisheries; commercial importance as aquarium fish	Talwar and Jhingran 1992; Hilton – Taylor 2000
	Cirrhinus reba	Endangered	-	Hussain and Mazid (this volume)
	Labeo bata	Endangered	Interest to aquaculture	Hussain and Mazid (this volume)
	L. boga	Critically endangered	Interest to aquaculture	Hussain and Mazid (this volume)
	L. calbasu	Endangered	Interest to aquaculture	Hussain and Mazid (this volume)
	L. gonius	Endangered	Interest to aquaculture	Hussain and Mazid (this volume)
	L. nandina	Critically endangered	Interest to aquaculture	Hussain and Mazid (this volume)
	L. pangusia	Endangered	-	Hussain and Mazid (this volume)
	Puntius sarana	Critically endangered	Interest to aquaculture	Hussain and Mazid (this volume)
	P. ticto	Endangered	-	Hussain and Mazid (this volume)
	Tor tor	Critically endangered	Interest to aquaculture	Hussain and Mazid (this volume)
	T. putitora	Critically endangered	Interest to aquaculture	Hussain and Mazid (this volume)
Brunei Darussalam	*Balantiocheilos melanopterus*	Endangered*	Commercial importance in aquaculture and as aquarium fish	Kottelat 2001; Hilton – Taylor 2000
Cambodia	*B. melanopterus*	Endangered*	Commercial importance in aquaculture and as aquarium fish	Rainboth 1996
	Chela caeruleostigmata	Critically endangered*	Commercial importance as aquarium fish	Rainboth 1996
	Probarbus jullieni	Endangered*	Commercial importance in fisheries; aquaculture - experimental	Rainboth 1996
China	*Acheilognathus elongatus*	Endangered*	-	IUCN 1994
	Barbodes exiguus	Endangered	-	Li 1997
	B. cogginii	Endangered	-	Li 1997
	Cyprinus crassilabris	Endangered	-	Li 1997
	C. longipectoralis	Endangered	-	Li 1997
	C. megalopthalmus	Endangered	-	Li 1997
	C. micristius	Endangered*	-	IUCN 1994
	C. yilongenesis	Extinct	-	Li 1997

	Sinocyclocheilus anatirostris	Vulnerable*	-	Proudlove 1997; Hilton – Taylor 2000
	S. angularis	Vulnerable*	-	Proudlove 1997; Hilton – Taylor 2000
	S. anopthalmus	Vulnerable*	-	IUCN 1990
	S. cyphotergous	Vulnerable*	-	Proudlove 1997; Hilton – Taylor 2000
	S. microphthalmus	Vulnerable*	-	Proudlove 1997; Hilton – Taylor 2000
	Tor yunnanensis	Endangered*	-	Baillie and Groombridge 1996
India	*A. microlepis*	Vulnerable*	Low importance in fisheries; commercial importance as aquarium fish	Talwar and Jhingran 1992; Hilton – Taylor 2000
	L. porcellus	Critically endangered*	Fisheries: minor commercial importance	Talwar and Jhingran 1992; Hilton – Taylor 2000
Indonesia	*B. melanopterus*	Endangered*	Commercial importance in aquaculture and as aquarium fish	Kottelat 2001; Hilton – Taylor 2000
	Rasbora baliensis	Vulnerable*	-	Kottelat et al. 1993; Hilton – Taylor 2000
	R. tawarensis	Vulnerable*	-	Kottelat, et al. 1993; Hilton – Taylor 2000
Iran	*Iranocypris typhlops*	Vulnerable*	-	Proudlove 1997; Hilton-Taylor 2000
Japan	*Rhodeus ocellatus smithii*	Critically endangered*	-	Kimura and Nagata 1992; Hilton-Taylor 2000
Lao PDR	*B. melanopterus*	Endangered*	Commercial importance in aquaculture and as aquarium fish	Kottelat 2001; Hilton-Taylor 2000
	C. caeruleostigmata	Critically endangered*	Commercial importance in aquaculture and as aquarium fish	Rainboth 1996; Hilton-Taylor 2000
	P. jullieni	Endangered*	Commercial importance in aquaculture	Roberts 1992; Hilton-Taylor 2000
Malaysia	*B. melanopterus*	Endangered*	Commercial importance in aquaculture and as aquarium fish	Kottelat 2001; Hilton – Taylor 2000
Philippines	*Cephalakompsus pachycheilus*	Critically endangered*	-	Herre 1924; Hilton-Taylor 2000
	Mandibularca resinus	Critically endangered*	Minor interest to fisheries	Herre 1924; Hilton-Taylor 2000
	P. amarus	Critically endangered*	-	Herre 1924; Hilton-Taylor 2000
	P. baoulan	Critically endangered*	-	Hilton-Taylor 2000
	P. clemensi	Critically endangered*	-	Hilton-Taylor 2000
	P. disa	Critically endangered*	-	Herre 1953; Hilton-Taylor 2000
	P. flavifuscus	Critically endangered*	-	Herre 1924; Hilton-Taylor 2000
	P. hemictenus	Vulnerable*	-	Herre 1924; Hilton-Taylor 2000
	P. herrei	Critically endangered*	Commercial importance in fisheries	Herre 1953; Hilton-Taylor 2000

	P. katolo	Critically endangered*	-	Herre 1953; Hilton-Taylor 2000
	P. lanaoensis	Critically endangered*	-	Herre 1924; Hilton-Taylor 2000
	P. lindog	Vulnerable*	-	Herre 1924; Hilton-Taylor 2000
	P. manalak	Critically endangered*	Subsistence fisheries	Herre 1924; Hilton-Taylor 2000
	P. manguaoensis	Vulnerable*	-	Herre 1924; Hilton-Taylor 2000
	P. sirang	Vulnerable*	-	Herre 1953; Hilton-Taylor 2000
	P. tras	Critically endangered*	-	Hilton-Taylor 2000
	P. tumba	Vulnerable*	Low importance in fisheries	Herre 1924; Hilton-Taylor 2000
	Spratellicypris palata	Critically endangered*	Commercial importance in fisheries	Herre 1953; Hilton-Taylor 2000
Sri Lanka	*L. fisheri*	Endangered*	-	Talwar and Jhingran 1992; Hilton – Taylor 2000
	L. porcellus	Critically endangered*	Low importance in fisheries	Talwar and Jhingran 1992; Hilton – Taylor 2000
	P. asoka	Endangered*	Commercial importance as aquarium fish	Pethiyagoda 1991; Hilton-Taylor 2000
	P. bandula	Critically endangered*	For likely future use in aquaculture	Pethiyagoda 1991; Hilton-Taylor 2000
	P. martenstyni	Endangered*	Commercial importance in fisheries	Pethiyagoda 1991; Hilton-Taylor 2000
Thailand	*B. melanopterus*	Endangered*	Commercial importance in aquaculture and as aquarium fish	Kottelat 2001; Hilton – Taylor 2000
	C. caeruleostigmata	Critically endangered*	Commercial importance as aquarium fish	Rainboth 1996; Hilton-Taylor 2000
	P. jullieni	Endangered*	Commercial importance in fisheries; aquaculture – experimental; gamefish	Roberts 1992; Hilton-Taylor 2000
Vietnam	*B. melanopterus*	Endangered*	Commercial importance in aquaculture and as aquarium fish	Kottelat 2001; Hilton – Taylor 2000
	P. jullieni	Endangered*	Commercial importance in fisheries and in aquaculture	Roberts 1992; Hilton-Taylor 2000
	C. caeruleostigmata	Critically endangered*	Commercial importance as aquarium fish	Rainboth 1996

* Data obtained from the FishBase record of threatened species (www.fishbase.org); based on IUCN categories of threatened species.

APPENDIX 1

Nomenclature of cyprinid species mentioned in this publication from FishBase[1])

Latest nomenclature of carp species (from FishBase)	Synonyms
Acheilognathus elongatus	*Acheilognathus elongates*
Acheilognathus tonkinensis	*Acanthorhodeus tonkinensis*
Amblypharyngodon microlepis	
Aristichthys nobilis	
Balantiocheilos melanopterus	
Barbodes belinka	
Barbodes cogginii	*Barbodes coggili*
Barbodes exiguus	*Barbodes exigiua*
Barbodes hexagonolepis	
Barbodes sarana	*Puntius sarana*
Barbonymus altus	*Barbodes altus*
Barbonymus gonionotus	*Barbodes gonionotus; Puntius gonionotus; Puntius javanicus*
Barbonymus schwanenfeldii	*Barbodes schwanenfeldii*
Barbus barbus	
Barilius bendelisis	
Barilius gatensis	
Bengala elanga	*Bengala elonga*
Carassius auratus	*Carassius auratus*
Carassius carassius	
Carassius cuvieri	
Carassius gibelio	
Catla catla	
Catlocarpio siamensis	
Cephalakompsus pachycheilus	
Chanodichthys erythropterus	*Erythroculter ilishaeformis*
Chanodichthys flavipinnis	*Erythroculter recurvirostrix*
Chela caeruleostigmata	
Cirrhinus chinensis	
Cirrhinus cirrhosus	*Cirrhinus mrigala*
Cirrhinus jullieni	
Cirrhinus molitorella	
Cirrhinus reba	
Ctenopharyngodon idella	
Culter erythropterus	
Cyclocheilichthys enoplos	
Cyprinus carpio	*Cyprinus carpio*
Cyprinus longipectoralis	
Cyprinus megalophthalmus	
Cyprinus micristius	
Cyprinus pellegrini	
Cyprinus yilongensis	*Cyprinus yilongenesis*
Danio albolineatus	*Brachydanio albolineatus*
Danio rerio	
Devario aequipinnatus	

Latest nomenclature of carp species (from FishBase)	Synonyms
Devario devario	*Danio devario*
Devario malabaricus	*Danio malabaricus*
Epalzeorhynchos bicolor	*Labeo bicolour*
Epalzeorhynchos munense	*Labeo eythrurus*
Gymnocypris przewalskii	*Gymnocypris prezewalskii*
Gymnodiptychus pachycheilus	*Acheilognathus crassilabris*
Haplochromis perrieri	*Prognathochromis perrieri*
Hemibarbus maculatus	
Hemiculter leucisculus	
Hypophthalmichthys harmandi	
Hypophthalmichthys molitrix	
Hypsibarbus wetmorei	*Barbodes daruphani*
Iranocypris typhlops	
Labeo ariza	*Cirrhinus ariza*
Labeo bata	
Labeo boga	
Labeo calbasu	
Labeo fimbriatus	
Labeo fisheri	
Labeo gonius	
Labeo nandina	
Labeo pangusia	
Labeo porcellus	
Labeo rohita	
Leptobarbus hoevenii	
Leuciscus waleckii	
Mandibularca resinus	
Megalobrama amblycephala	
Megalobrama terminalis	
Morulius chrysophekadion	
Mylopharyngodon piceus	
Neolissochilus thienemanni	
Opsariichthys bidens	
Orthodon microlepidotus	
Osteochilus hasseltii	
Osteochilus melanopleurus	*Osteochilus melanopleura*
Osteochilus vittatus	
Parabramis pekinensis	
Paralaubuca riveroi	
Pimephales promelas	
Plagiognathops microlepis	
Poropuntius huguenini	
Probarbus jullieni	
Pseudorasbora parva	
Ptychobarbus dipogon	*Diptychus dipogon*
Puntioplites proctozysron	
Puntioplites waandersi	

[1] Species nomenclature follows that of FishBase 2004 [available on line at: http://www.fishbase.org].

Latest nomenclature of carp species (from FishBase)	Synonyms
Puntius amarus	
Puntius asoka	
Puntius bandula	
Puntius baoulan	
Puntius clemensi	
Puntius conchonius	*Barbus conchonius*
Puntius disa	
Puntius flavifuscus	
Puntius hemictenus	
Puntius herrei	
Puntius katolo	
Puntius lanaoensis	
Puntius lateristriga	
Puntius lindog	
Puntius manalak	
Puntius manguaoensis	
Puntius martenstyni	
Puntius oligolepis	
Puntius orphoides	*Barbodes orphoides*
Puntius semifasciolatus	
Puntius sirang	
Puntius stoliczkanus	
Puntius tetrazona	
Puntius ticto	
Puntius tras	
Puntius tumba	
Raiamas bola	
Rasbora baliensis	
Rasbora caudimaculata	
Rasbora dorsiocellata	
Rasbora elegans	
Rasbora sumatrana	
Rasbora tawarensis	
Rasborinus lineatus	
Rhodeus smithii	*Rhodeus ocellatus (also valid)*
Saurogobio dabryi	
Schizothorax kumaonensis	
Schizothorax richardsonii	
Schizothorax taliensis	
Schizothorax yunnanensis	
Sinilabeo dero	*Bangana dero*
Sinocyclocheilus anatirostris	
Sinocyclocheilus angularis	
Sinocyclocheilus anophthalmus	
Sinocyclocheilus cyphotergous	
Sinocyclocheilus microphthalmus	
Sperata aor	*Osteobagrus aor*
Sperata seenghala	*Osteobagrus seenghala*
Spinibarbus caldwelli	
Spinibarbus denticulatus	
Spratellicypris palata	
Squaliobarbus curriculus	

Latest nomenclature of carp species (from FishBase)	Synonyms
Tanichthys albonubes	
Thynnichthys thynnoides	
Tinca tinca	
Tor douronensis	
Tor putitora	
Tor soro	
Tor tambra	
Tor tambroides	
Tor tor	
Toxabramis swinhonis	
Xenocypris argentea	*Xenocyprinis argentea*
Xenocypris davidi	

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