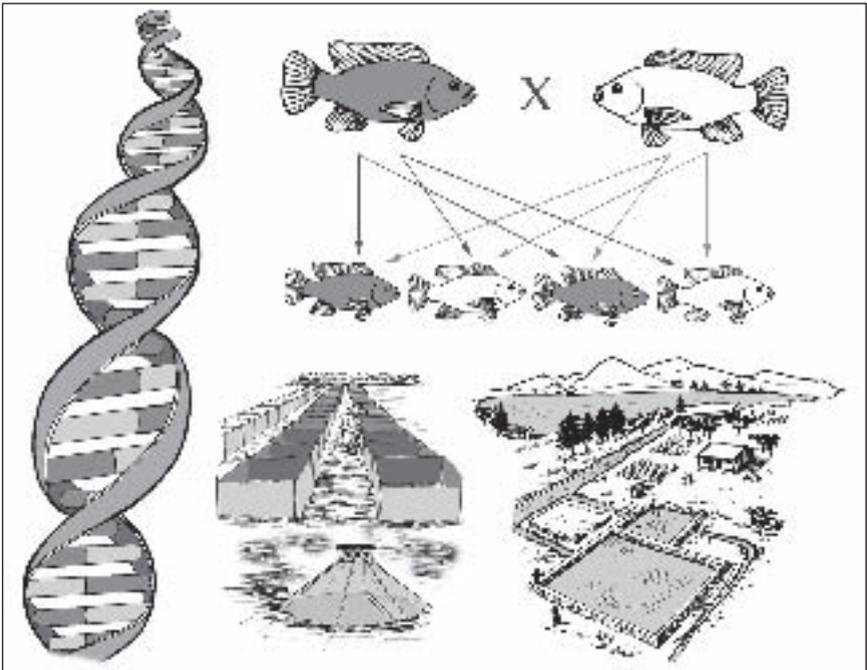


# AQUACULTURE DEVELOPMENT

## 3. Genetic resource management



Cover design, artwork and layout by Emanuela D'Antoni,  
Devin M. Bartley and José Luis Castilla Civit.

# **AQUACULTURE DEVELOPMENT**

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## **PREPARATION OF THIS DOCUMENT**

These Technical Guidelines have been prepared by the Fisheries and Aquaculture Department of the Food and Agriculture Organization of the United Nations (FAO) under the coordination of Devin M. Bartley (Senior Fishery Resources Officer) with the support of the FAO Regular Programme, FAO Commission on Genetic Resources for Food and Agriculture, FishCode (FAO's Programme of Global Partnerships for Responsible Fisheries) and the World Fisheries Trust. The following experts in the field of genetic resource management contributed to individual chapters in the Guidelines: Devin M. Bartley, Malcolm C. M. Beveridge, Randall E. Brummett, Joachim Carolsfeld, R. J. Lawton, Brian J. Harvey, Anne Kapuscinski, Graham Mair, Raul W. Ponzoni, Roger S. V. Pullin, Douglas Tave and Álvaro Toledo. The overall editor for the Guidelines was Devin M Bartley with assistance from the above experts. Layout formatting was by José Luis Castilla; cover design was by Emanuela D'Antoni.

The majority of the coordinating and editing work to produce these guidelines was accomplished while the overall editor was on FAO Staff Development Training in Victoria British Columbia, hosted by the World Fisheries Trust. The support of the FAO Advisory Committee on External Training and the World Fisheries Trust is gratefully acknowledged.

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### **ABSTRACT**

These Technical Guidelines have been developed to support sections of the FAO's Code of Conduct for Responsible Fisheries on aspects of genetic resource management in aquaculture. Guidance is provided on broodstock management and domestication, genetic improvement programmes, dissemination programmes for genetically improved fish, economic considerations in genetic improvement programmes, risk assessment and monitoring, culture based fisheries, conservation of fish genetic resources, gene banks, a precautionary approach and public relations. The effective management of genetic resources, risk assessment and monitoring can help promote responsible aquaculture by increasing production output and efficiency and help minimize adverse impacts on the environment. These benefits of the responsible application of genetic principles to aquaculture should be communicated to consumers, policy-makers, scientists and others interested in responsible fisheries and aquaculture.

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## **BACKGROUND**

1. From ancient times, fishing has been a major source of food for humanity and a provider of employment and economic benefits to those engaged in this activity. However, with increased knowledge and the dynamic development of fisheries, it was realized that living aquatic resources, although renewable, are not infinite and need to be properly managed, if their contribution to the nutritional, economic and social well-being of the growing world's population was to be sustained.

2. The adoption in 1982 of the United Nations Convention on the Law of the Sea provided a new framework for the better management of marine resources. The new legal regime of the oceans gave coastal States rights and responsibilities for the management and use of fishery resources within the areas of their national jurisdiction, which embrace some 90 percent of the world's marine fisheries.

3. In recent years, world fisheries have become a dynamically developing sector of the food industry, and many States have striven to take advantage of their new opportunities by investing in modern fishing fleets and processing factories in response to growing international demand for fish and fishery products. It became clear, however, that many fisheries resources could not sustain an often uncontrolled increase of exploitation.

4. Clear signs of over exploitation of important fish stocks, modifications of ecosystems, significant economic losses, and international conflicts on management and fish trade threatened the long-term sustainability of fisheries and the contribution of fisheries to food supply. Therefore, the Nineteenth Session of the FAO Committee on Fisheries (COFI), held in March 1991, recommended that new approaches to fisheries management embracing conservation and environmental, as well as social and economic, considerations were urgently needed. FAO was asked to develop the concept of responsible fisheries and elaborate a Code of Conduct to foster its application.

5. Subsequently, the Government of Mexico, in collaboration with FAO, organized an International Conference on Responsible Fishing in Cancún in May 1992. The Declaration of Cancún endorsed at that Conference was brought to the attention of the UNCED Summit in Rio de Janeiro, Brazil, in June 1992, which supported the preparation of a Code of Conduct for Responsible Fisheries. The FAO Technical Consultation on High Seas

Fishing, held in September 1992, further recommended the elaboration of a Code to address the issues regarding high seas fisheries.

6. The One Hundred and Second Session of the FAO Council, held in November 1992, discussed the elaboration of the Code, recommending that priority be given to high seas issues and requested that proposals for the Code be presented to the 1993 session of the Committee on Fisheries.

7. The Twentieth Session of COFI, held in March 1993, examined in general the proposed framework and content for such a Code, including the elaboration of guidelines, and endorsed a time frame for the further elaboration of the Code. It also requested FAO to prepare, on a “fast track” basis, as part of the Code, proposals to prevent reflagging of fishing vessels which affect conservation and management measures on the high seas. This resulted in the FAO Conference, at its Twenty-seventh Session in November 1993, adopting the Agreement to Promote Compliance with International Conservation and Management Measures by Fishing Vessels on the High Seas, which, according to FAO Conference Resolution 15/93, forms an integral part of the Code.

8. The Code was formulated so as to be interpreted and applied in conformity with the relevant rules of international law, as reflected in the United Nations Convention on the Law of the Sea, 1982, as well as with the Agreement for the Implementation of the Provisions of the United Nations Convention on the Law of the Sea of 10 December 1982 Relating to the Conservation and Management of Straddling Fish Stocks and Highly Migratory Fish Stocks, 1995, and in the light of, *inter alia*, the 1992 Declaration of Cancún and the 1992 Rio Declaration on Environment and Development, in particular Chapter 17 of Agenda 21.

9. The development of the Code was carried out by FAO in consultation and collaboration with relevant United Nations Agencies and other international organizations, including non-governmental organizations.

10. The Code of Conduct consists of five introductory articles: Nature and Scope; Objectives; Relationship with Other International Instruments; Implementation, Monitoring and Updating and Special Requirements of Developing Countries. These introductory articles are followed by an article on General Principles, which precedes the six thematic articles on Fisheries Management, Fishing Operations, Aquaculture Development, Integration of Fisheries into Coastal Area Management, Post-Harvest Practices and Trade, and Fisheries Research. As already mentioned, the Agreement to Promote

Compliance with International Conservation and Management Measures by Fishing Vessels on the High Seas forms an integral part of the Code.

11. The Code is voluntary. However, certain parts of it are based on relevant rules of international law, as reflected in the United Nations Convention on the Law of the Sea of 10 December 1982. The Code also contains provisions that may be or have already been given binding effect by means of other obligatory legal instruments amongst the Parties, such as the Agreement to Promote Compliance with Conservation and Management Measures by Fishing Vessels on the High Seas, 1993.

12. The twenty-eighth session of the Conference in Resolution 4/95 adopted the Code of Conduct for Responsible Fisheries on 31 October 1995. The same Resolution requested FAO *inter alia* to elaborate appropriate technical guidelines in support of the implementation of the Code in collaboration with members and interested relevant organizations.



## 1 INTRODUCTION

The role of aquaculture in food production, economic development and food security is now well recognized. As the fastest growing food production sector, aquaculture holds promise to help provide a growing human population with food as many of the world's capture fisheries have reached their biological limits of production or have been depleted through over-fishing and habitat degradation. Less well recognized is aquaculture's role in conservation and the recovery of threatened and endangered species. In fact, aquaculture has often been implicated in contributing to the endangerment of aquatic biodiversity.

The aquaculture sector has made significant advances in increased production and environmental protection. However, the sector is now being criticized for degrading the aquatic habitat through release of effluents that include uneaten food, waste products, and pharmaceuticals, and through the escape of farmed fish. There is potential to improve the production, efficiency and environmental sustainability of the sector and the effective management of aquatic genetic resources can assist in addressing all of the above issues. Genetically improved fish (Chapters 4, 5 and 6) grow faster and use food more efficiently, which will produce less waste. Disease resistant fish require less pharmaceutical treatments. Some farmed fish can be made sterile to reduce the chance of them breeding with native species or establishing feral populations. Broodstock management (Chapters 3 and 8), genetic improvement programmes (Chapters 4, 5 and 6), and gene banking (Chapter 10) will help improve production and profitability, as well as assist in protection and conservation of wild resources (Chapter 9). Risk assessment (Chapter 7), adhering to international guidelines (Chapter 2) and a precautionary approach (Chapter 11) will help ensure wise decisions that will protect society and the environment, while at the same time allowing the sector to develop.

Fish genetic resources (FiGR) comprise all finfish and aquatic invertebrate genetic material that has actual or potential value for capture fisheries and aquaculture. This includes DNA, genes, gametes, individual organisms, wild, farmed and research populations, species and organisms that have been genetically altered (for example by selective breeding, hybridization, chromosome set manipulation and gene transfer). How these resources can be used to help aquaculture realize its full potential and conserve valuable wild genetic diversity is the subject of these guidelines.

The purpose of these guidelines is to provide a succinct set of instructions as a framework that can direct policy-makers and senior resource managers

towards improved management of fish genetic resources (FiGR). Throughout these guidelines, management is understood to include use and conservation. Management of genetic resources is approached from a holistic viewpoint that incorporates economics, conservation, risk analysis and uncertainty, as well as increased production and profitability.

### **1.1 Value of genetic diversity and the need for genetic resource management**

Of the over 230 species of farmed aquatic animals and plants for which FAO has statistics, only a few have been the subject of deliberate genetic resource management programmes. Channel catfish, Nile tilapia, Atlantic salmon and many farmed carps are cases that demonstrate the significant gains in production possible from genetic improvement programmes. Only a few culture-based fisheries, usually salmonids, purposefully choose the stocks to release so that they either match or differ completely from the native fishes. One estimate made by a prominent geneticist indicated that the supply gap caused by decreasing output from capture fisheries and the increasing human population could be filled simply by incorporating genetic improvement programmes into already existing aquaculture systems (i.e. no additional farming systems, land or water usage would be required).

Management of FiGR is necessary for more than just increased production. Besides being essential for genetic improvement programmes in aquaculture, genetic resources are the necessary raw ingredients that allow species to adapt to short-term and long-term changes in their environment; they provide species, populations and individuals with the flexibility of dealing with and adapting to changes to their environment, changes both from humans and from natural causes. That is, genetic diversity is necessary for the continued evolution of species. Genetic diversity interacts with environmental variation to produce the variety of shapes, sizes, life-history characters, behaviour, and colours that make aquatic species so valuable and interesting. Some of these differences show up as different colours of fish or as different scale patterns, whereas other differences show up as different migration patterns or reproductive behaviour. Without genetic diversity, there would be no species diversity, no adaptation, no breeds, and no evolution; there eventually would be extinction as climate and habitats change as a result of natural or human actions.

The common carp has by far the longest history of domestication and genetic improvement for aquaculture. Farmed Atlantic salmon, channel catfish and

Nile tilapia have been genetically improved more recently. However, with the success of these breeding programmes (i.e. changing the genetic structure of a wild fish) and the inevitable use of these improved breeds in many farming systems, comes the problem of interaction between genetically improved aquaculture stocks and their wild relatives. These wild relatives often support viable fisheries and will provide new genetic material that can be useful to aquaculture. The aquaculture sector is in an advantageous position to minimize extinction of the wild relatives of farmed species, as was allowed to happen to many in the livestock and crop sectors.

Management of aquatic genetic resources must have defined objectives in order to plan programmes and to judge success and impact. These objectives will depend on the purpose of the aquaculture facility: whether it is maximizing production, maximizing efficiency, reducing inputs, releasing fish for culture-based fisheries, or helping restock threatened or endangered species. Each of these objectives will require different management programmes for aquatic genetic resources.

## **1.2 Relevant articles of the Code**

These guidelines are organized by general subject areas that are important for genetic resource management, rather than by specific articles of the Code. This will allow decision makers and resource planners to find guidance on a specific area of genetics in aquaculture quickly. Given the importance of genetic resource management for a variety of aquaculture objectives, there are several articles of the Code that a particular chapter may help implement. These guidelines provide information on the following articles for the Code (relevant chapters are included).

## **ARTICLE 2 – OBJECTIVES OF THE CODE**

**2e** *facilitate and promote technical, financial and other cooperation in conservation of fisheries (including aquaculture) resources and fisheries management and development (Chapters 2, 5, 6, 7, 9, 10 and 11).*

**2g** *promote protection of living aquatic resources and their environments and coastal areas (Chapters 2, 5, 7, 9, 10 and 11).*

## **ARTICLE 6 – GENERAL PRINCIPLES**

**6.2** *Fisheries management should promote the maintenance of the quality, diversity and availability of fishery resources in sufficient quantities for present and future generations in the context of food security, poverty alleviation and sustainable development. Management measures should not only ensure the conservation target species but also of species belonging to the same ecosystem or associated with or dependent upon the target species (Chapters 7, 9, 10 and 11).*

**6.8** *All critical fisheries habitats in marine and fresh water ecosystems, such as wetlands, mangroves, reefs, lagoons, nursery and spawning areas, should be protected and rehabilitated as far as possible and where necessary. Particular effort should be made to protect such habitats from destruction, degradation, pollution and other significant impacts resulting from human activities that threaten the health and viability of the fishery resources (Chapters 9 and 10).*

**6.12** *States should, within their respective competences and in accordance with international law, cooperate at subregional, regional and global levels through fisheries management organizations, other international agreements or other arrangements to promote conservation and management, ensure responsible fishing and ensure effective conservation and protection of living aquatic resources throughout their range of distribution, taking into account the need for compatible measures in areas within and beyond national jurisdiction (Chapter 2, 5 and 9).*

## **ARTICLE 7 – FISHERIES MANAGEMENT**

**7.2.2.d** *biodiversity of aquatic habitats and ecosystems is conserved and endangered species are protected (Chapter 9 and 10);*

## **7.4 Data gathering and management advice (Chapters 9 and 10)**

**7.5.1** *States should apply the precautionary approach widely to conservation, management and exploitation of living aquatic resources in order to protect them and preserve the aquatic environment. The absence of adequate scientific information should not be used as a reason for postponing or failing to take conservation and management measures (Chapter 11).*

**7.6.8** *The efficacy of conservation and management measures and their possible interactions should be kept under continuous review. Such measures should, as appropriate, be revised or abolished in the light of new information (Chapter 8, 9 and 11).*

## **ARTICLE 9 – AQUACULTURE DEVELOPMENT**

**9.1.2** *States should promote responsible development and management of aquaculture, including an advance evaluation of the effects of aquaculture development on genetic diversity and ecosystem integrity, based on best available scientific information (All chapters).*

**9.1.3** *States should produce and regularly update aquaculture development strategies and plans, as required, to ensure that aquaculture development is ecologically sustainable and to allow the rational use of resources by aquaculture and other activities (Chapters 7, 8, 9 and 11).*

**9.3.1** *States should conserve genetic diversity and maintain integrity of aquatic communities and ecosystems by appropriate management. In particular, efforts should be undertaken to minimize the harmful effects of introducing non-native species or genetically altered stocks used for aquaculture including culture-based fisheries into waters, especially where there is a significant potential for the spread of such non-native species or genetically altered stocks into waters under the jurisdiction of other States, as well as waters under the jurisdiction of the State of origin. States should, whenever possible, promote steps to minimize adverse genetic, disease and other effects of escaped farmed fish on wild stocks. (Chapters 2, 5, 8, 9 and 10).*

**9.3.3** *States should, in order to minimize risks of disease transfer and other adverse effects on wild and cultured stocks, encourage adoption of appropriate practices in the genetic improvement of broodstocks, the introduction of non-native species, and in the production, sale and transport*

*of eggs, larvae or fry, broodstock or other live materials. States should facilitate the preparation and implementation of appropriate national codes of practice and procedures to this effect. (Chapters 3, 4, 5, 8 and 9).*

**9.3.5** *States should, where appropriate, promote research and, when feasible, the development of culture techniques for endangered species to protect, rehabilitate and enhance their stocks, taking into account the critical need to conserve genetic diversity of endangered species (Chapters 3 and 9).*

## 2 INTERNATIONAL SETTING

The Code of Conduct for Responsible Fisheries (CCRF) and the international community have recognized the vital role that genetic resources, including FiGR, play in sustainable development and conservation. As a result, international mechanisms, guidelines and codes of practice have been developed. The Convention on Biological Diversity (CBD)<sup>1</sup> arose from the Earth Summit in 1992 and has more signatories than any other piece of international legislation. It is a legally binding instrument that requires the conservation and sustainable use of biological diversity (including genetic diversity), and the fair and equitable sharing of benefits derived from that use. In recognizing the need for scientific and technological advice in order to implement the articles of the Convention, the CBD established a Subsidiary Body on Scientific, Technical and Technological Advice (SBSTTA). The CBD further established the Cartagena Protocol on Biosafety,<sup>2</sup> a set of binding international protocols on the international movement of living modified organisms, which would include genetically modified organisms (GMOs) (i.e. transgenic organisms). Similar to the CCRF, the CBD recognizes both the need to use and to conserve biodiversity.

The precautionary approach to development is an essential attribute of both the CBD and the CCRF. Aside from agreement to be cautious and use the best information available, there are a variety of opinions on what this approach means in practice; it forms the basis of Chapter 11.

The Convention on International Trade in Endangered Species of Fauna and Flora (CITES) is another significant instrument that impacts on the management of FiGR. CITES restricts the international trade in species that are threatened in the wild – the degree of threat or endangerment indicates how restrictive trade will be. Some aquatic species that are threatened in the wild are also farmed, e.g. sturgeons (*Acipenseriformes*), and arowana or dragon fish (*Scleropages formosus*). International trade in these species must ensure that the species being traded actually come from licensed farms and not from the wild, and that the trade of farmed species does not create a market for the endangered species in the wild. Genetic markers and genetic stock identification have been used to help differentiate species and stocks of wild and farmed species.

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<sup>1</sup> [www.biodiv.org](http://www.biodiv.org)

<sup>2</sup> <http://www.cbd.int/biosafety/default.shtml>. As of August 2008, there were no aquatic GMOs being produced for human consumption.

The Ramsar Convention on Wetlands mandates countries to identify and protect wetlands, including coastal and inter-tidal areas that are of national importance. Primary criteria for establishing importance were the roles wetlands play in maintaining wild biodiversity, primarily waterfowl. However, Ramsar expanded the criteria to include historical use of wetlands as a fishery resource<sup>3</sup> and now allows aquaculture of native species as an acceptable activity in Ramsar sites. However, farming of native species could eventually lead to their domestication and genetic alteration through natural selection to farm environments and breed improvement programmes.

More specific guidelines have been developed by FAO and others that apply indirectly to the management of FiGR. Technical Guidelines on Aquaculture have been developed for general issues relating to FiGR.<sup>4</sup> FAO, the WorldFish Centre (WFC) and other partners established the Nairobi Declaration (Annex 1) on recommendations for importing genetically improved tilapia into Africa. These non-binding resolutions layout a framework that is elaborated here in these guidelines in regards to the responsible use of genetically improved fish in aquaculture.

Fish health concerns play a major role in the trade and movement of aquatic species. Dissemination of genetically improved stocks (Chapter 5) requires adherence to the World Organization for Animal Health (OIE) with regards to transboundary pathogens. Technical guidelines have been established<sup>5</sup> that are consistent with OIE and World Trade Organization (WTO) requirements.

Recently, FAO has made progress with regard to aquatic genetic resources. The lack of coherent fish genetic resources management and of policies is in fact becoming a problem in the recent rapid expansion of aquaculture. A transition to more responsible, sustainable and productive aquaculture has been called for by Members of FAO and the international community. Its success will depend in large measure upon effective management of fish genetic resources.

At its Eleventh Session, the FAO's intergovernmental Commission on Genetic Resources for Food and Agriculture recognized the importance

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<sup>3</sup> [http://www.ramsar.org/res/key\\_res\\_vi.2.htm](http://www.ramsar.org/res/key_res_vi.2.htm)

<sup>4</sup> FAO. 1997. Aquaculture development. FAO Technical Guidelines for Responsible Fisheries. No. 5. Rome, FAO.

<sup>5</sup> FAO. 2007. Aquaculture development. 2. Health management for responsible movement of live aquatic animals. FAO Technical Guidelines for Responsible Fisheries. No. 5. Suppl. 2. Rome, FAO. <ftp://ftp.fao.org/docrep/fao/010/a1108e/a1108e00.pdf>

and vulnerability of aquatic genetic resources, their roles in an ecosystem approach for food and agriculture, and for their contributions to meeting the challenges presented by climate change. It agreed that its 10-year Multi-year Programme of Work should include coverage of aquatic genetic resources for the development of sustainable and responsible fisheries and aquaculture in cooperation with other forums and organizations, such as COFI or UNCLOS.<sup>6</sup>

<sup>6</sup> FAO/CGRFA. 2007. Report of the Eleventh Regular Session of the Commission on Genetic Resources for Food and Agriculture. CGRFA-11/07/REPORT. <ftp://ftp.fao.org/ag/cgrfa/cgrfa11/r11repe.pdf>

### **Box 1. Terminology**

The terminology used to describe organisms that are genetically different from wild types is extremely important because it has legal and policy implications and will influence how well the general public accepts the product or process. Therefore, farmed fish that have been genetically altered in some way by humans must be described clearly and accurately. Unfortunately, a variety of terms have been used to describe genetically altered fish. The usage is not standardized and can lead to consumer confusion and regulatory problems, such as when applying for farming licenses or trade permits. Aquaculturists and government regulators must be aware of these implications.

Fish produced through aquaculture have the potential to become genetically different from their wild ancestors through selection to hatchery and farm environments (Chapters 3 and 9), and/or through purposeful genetic improvement programmes (Chapter 4). In aquaculture, fish farmers seek to farm the best and most profitable fish available and to project an image to consumers that the product is both healthy and natural; consumers are increasingly seeking these qualities from their food. This interface is usually managed through labelling and marketing. The guidelines in this book do not address consumer labelling issues other than in a very general manner (Chapter 12). However, for government oversight of farmed fish and their marketing, it will be crucial to understand the genetic technologies being used and the changes those technologies impart on the farmed organism.

A general term for all human-induced changes to an organism is *genetically altered*. This term should be used as a neutral statement of fact without judging whether the alteration is good or bad, whether it is a result of modern biotechnology or traditional methods, or whether the alteration is deliberate or accidental. This is meant to be a very general term, reflecting the possibility that a genetically altered organism could have environmental or population risks independent of how it became altered (Chapter 7).

The following terms are important to use correctly as they are associated with consumer perception and government oversight. Additional terms can be found in the FAO glossaries.<sup>1</sup>

**Genetically modified organism (GMO):** An organism in which the genetic material has been altered by humans through gene or cell technologies. A genetically modified fish is usually a transgenic fish (i.e. a fish with a gene inserted from another organism in a manner that is not possible through natural processes). At present, there are no genetically modified fish available to the consumer. There are currently several restrictions on the international movement of GMOs. This class of organisms is regulated by the Cartagena Protocol of the CBD<sup>2</sup>. Additionally, many consumer groups are currently against the use of GMOs, including *genetically modified* fish. Thus, a fish farmer wishing to import a fish genetically *improved* through selective breeding, should not use the term *genetically modified*, but instead use *genetically improved through selective breeding (or through traditional breeding)*.

**Hybrid:** Offspring of the mating between parents of different species or varieties. Offspring of matings between parents of the same species are **intra-specific hybrids**, whereas offspring of matings between parents of different species are **inter-specific hybrids**. The distinction is important because some areas have laws against mating different species, or importing inter-specific hybrids, whereas matings or importation of the same species may not be regulated.

**Living modified organism (LMO):** “Living organism that possesses a novel combination of genetic material obtained through the use of modern

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<sup>1</sup> <http://www.fao.org/fi/glossary/default.asp> and [http://www.fao.org/biotech/index\\_glossary.asp](http://www.fao.org/biotech/index_glossary.asp)  
<sup>2</sup> <http://www.cbd.int/biosafety/default.shtml>

biotechnology”. Synonym of GMO used primarily by the Convention on Biological Diversity.

**Polyploids:** Plants or animals having more than 2 sets of chromosomes (called diploids and designated as 2N). Organisms having 3 sets are called triploids (3N), those with 4 sets are tetraploids (4N). The distinction is important because diploids and tetraploids are usually fertile whereas triploids are usually sterile. It is possible to mate tetraploids with diploids to get triploids.

**Traditional breeding:** refers to selective breeding programmes that do not use modern gene manipulation technologies (Chapter 4). Traditional breeding has been practiced and refined for millennia in terrestrial agriculture.

An international group of experts stated that it is more important to understand what actual changes the genetic alteration has caused to the farmed fish, rather than the techniques used to produce that change.<sup>3</sup> That is, addressing questions such as, does the fish consume more food or have better conversion efficiency, does it have wider environmental tolerances, is it fertile, is it more nutritious, can it become invasive, or does it produce new substances that the un-altered fish does not produce are more important in risk assessment (Chapter 7) than what technology was used to create the organism. Current policy, farm practices and public perception do not necessarily recognize this fact; it is recommended that more informative descriptions should be used to describe the actual changes to an organism as a result of genetic technologies.

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<sup>3</sup> Page 253, in Pullin, R.S.V., Bartley, D.M., Kooiman, J. (eds). 1999. Towards Policies for Conservation and Sustainable Use of Aquatic Genetic Resources. ICLARM Conference Proceedings No. 59. Manila, Philippines. “ ... in the formulation of biosafety policy and regulation for living modified organisms, the characteristics of the organisms and of potentially accessible environments are more important considerations than the processes used to produce those organisms.

### **3 BROODSTOCK MANAGEMENT: INBREEDING, GENETIC DRIFT AND DOMESTICATION** <sup>7</sup>

#### **3.1 Introduction**

Aquaculture is not only a critical sector of food production, it is also a necessary component of recreational and commercial fisheries and a required management tool for conservation programmes. As is the case for all types of animal husbandry, aquaculture means that humans must intervene in and manage a species' life cycle. The moment this occurs, we usually produce irreversible changes in the population's gene pool. These changes can be desired, which occurs when selective breeding programmes are conducted to improve growth (Chapter 4) or when domestication creates fish that are better adapted to the hatchery environment. Unfortunately, we also produce undesired, damaging changes in the genome through inbreeding and genetic drift (section 3.3), which lower viability and growth and increase developmental instability. While domestication is beneficial in food fish culture, it is harmful for fish that are stocked in the wild, because fish that are well adapted for a hatchery may be maladapted in the wild (Chapter 8). In this chapter, broodstock management involves the control of inbreeding and genetic drift and the process of domestication.

#### **3.2 Inbreeding**

Inbreeding is the mating of relatives. Inbreeding is one of the three traditional breeding programmes, and it has been used to develop new breeds and can be used in combination with cross-breeding to produce uniform, outstanding individuals (Chapter 4). Even though planned and directed inbreeding can be beneficial, unintentional and unplanned inbreeding will cause problems.

Genetically, inbreeding increases homozygosity in the offspring (i.e. genetic similarity) which means it also decreases heterozygosity (i.e. genetic diversity). Homozygosity is also produced when non-relatives mate and, genetically, the two forms of homozygosity are identical. Even though the two forms of homozygosity are identical, a distinction is made because of the way homozygosity is produced and its consequences.

Relatives are more alike genetically than non-relatives. Consequently, when relatives mate, they produce offspring that are more homozygous than is the

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<sup>7</sup> Contributed by Douglas Tave.

case when non-relatives mate; the closer the relationship between mates, the more homozygous the offspring.

This is of concern, because all animals contain a small number of harmful or deleterious recessive alleles.<sup>8</sup> In most cases an individual is not affected and survives because it has only one copy of the harmful allele, inherited from one of its parents (it is heterozygous); two copies of the allele are needed (one from both parents) to produce the harmful or lethal effect (it is homozygous). Because relatives are more alike than non-relatives, they tend to share the same deleterious recessive alleles. Two non-relatives might only share one or two in common, while relatives usually share more in common; the closer the relationship, the more that are shared. When relatives mate, the pairing and subsequent expression of these deleterious recessive alleles in their offspring produces inbreeding depression—lower growth rate, viability, and fecundity and an increase in the number of abnormalities. Studies in fish have shown that inbred fish have these classical clinical signs of inbreeding depression,<sup>9</sup> as well as a decreased return rate when stocked in the wild.<sup>10</sup>

The negative effects of inbreeding usually do not occur immediately. Inbreeding depression is often delayed (i.e. they might not occur until several generations after inbreeding has begun). How quickly inbreeding depression occurs depends on the amount of inbreeding that has been produced and the trait.

The ideas that were described above can give farmers the erroneous idea that inbreeding is a major reason behind many of their production problems, so they come to the erroneous conclusion that inbreeding has occurred and their stock is no longer of good quality when they observe a deformed individual or yield has declined. Deformities and a decrease in yield are often due to non-genetic factors such as developmental errors, toxins, nutritional deficiencies, or weather and may not be due to inbreeding.

Because inbreeding is the mating of relatives, if individuals can be given unique tags, it is rather easy to prevent inbreeding or to minimize it by

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<sup>8</sup> An allele is an alternate form of a gene.

<sup>9</sup> e.g. Kincaid, H.L. 1976. Effects of inbreeding on rainbow trout populations. Transactions of the American Fisheries Society, 105:273-280; Kincaid, H.L. 1976. Inbreeding in rainbow trout (*Salmo gairdneri*). Journal of the Fisheries Research Board of Canada, 33:2420-2426; Su, G.-S.; Liljedahl, L.-E, Gall, G.A.E., 1996. Effects of inbreeding on growth and reproduction traits in rainbow trout (*Oncorhynchus mykiss*). Aquaculture, 142:139-148.

<sup>10</sup> Ryman, N. 1970. A genetic analysis of recapture frequencies of released young salmon (*Salmo salar*) L. Hereditas, 5:159-160.

preventing parent-offspring, brother-sister, and half-sib matings. If the closest mating allowed is between second cousins, inbreeding will never become a problem.

When conducting selective breeding programmes, inbreeding is inevitable, because when you only allow the best to reproduce, you often mate relatives. Minimizing inbreeding during a selective breeding programme is important, because you do not want to use the genetic gain produced via selection simply to counteract inbreeding depression. To prevent this, a number of breeding programmes have been designed to minimize inbreeding during a selective breeding programme.<sup>11</sup> While it is important to prevent the systematic mating of close relatives in selective breeding programmes, incidental (random) matings of close relatives (e.g., brother-sister matings) in large-scale breeding programmes is not as big a problem as it is in small populations, because it is likely that the offspring produced by these matings will be mated to non-relatives in the following generation, which will produce fish with no inbreeding.<sup>12</sup>

While it is easy to give livestock individual marks and thus prevent relatives from mating, it is rather difficult for fish. Therefore, aquaculturists must manage the population as a whole to minimize the accumulation of inbreeding. To do this, aquaculturists must manage the effective breeding number ( $N_e$ ).<sup>13</sup>

One way  $N_e$  is determined is by the number of males and number of females that reproduce and leave viable offspring:

$$N_e = \frac{4 \text{ (number of females) (number of males)}}{\text{(number of females)} + \text{(number of males)}}$$

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- <sup>11</sup> Dupont-Nivet, M.; Vandeputte, M.; Haffray, P.; Chevassus, B. 2006. Effect of different mating designs on inbreeding, genetic variance and response to selection when applying individual selection in fish breeding programs. *Aquaculture*, 252:161-170; Gallardo, J.A.; Lhorente, J.P.; García, X., Neira, R. 2004. Effects of nonrandom mating schemes to delay the inbreeding accumulation in cultured populations of coho salmon (*Oncorhynchus kisutch*). *Canadian Journal of Fisheries and Aquatic Sciences*, 61:547-553; Gjerde, B., GjØen, H.M., Villanueva, B. 1996. Optimum designs for fish breeding programmes with constrained inbreeding. Mass selection for a normally distributed trait. *Livestock Production Science*, 47:59-72; Inbreeding and Brood Stock Management. Fisheries Technical Paper. No 392. Rome, FAO.
- <sup>12</sup> Dupont-Nivet, M.; Vandeputte, M. 2005. Does avoiding full sibs matings preserves genetic variability in a selection scheme? Case of single pair matings. *Aquaculture*, 247:12.
- <sup>13</sup> Hallerman, E. 2003a. Inbreeding. Pages 215-237 in E.M. Hallerman, ed. *Population genetics: Principles and Applications for Fisheries Scientists*. Bethesda, MD, American Fisheries Society; Tave, D. 1993. *Genetics for Fish Hatchery Managers*, 2<sup>nd</sup> ed. New York, Van Nostrand Reinhold; See footnote 14.

Thus,  $N_e$  is determined by the number of males that leave viable offspring, the number of females that leave viable offspring, and by the sex ratio of the brood fish. This means that  $N_e$  is maximized by increasing both the number of males and females that are spawned and by bringing the sex ratio as close to 1:1 as possible. Skewed sex ratios, which are often used in aquaculture, make  $N_e$  far smaller than the number of brood fish that are spawned.  $N_e$  is most strongly influenced by the least represented sex (e.g. when few males are used,  $N_e$  approximates the number of males rather than the total).

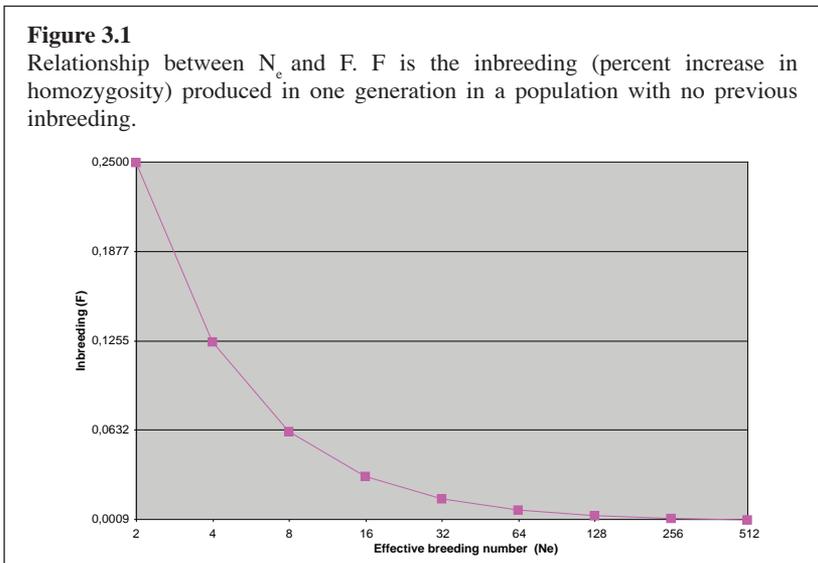
The reason aquaculturists and fisheries biologists need to manage  $N_e$  is that it is inversely related to inbreeding:

$$F = 1/2N_e$$

where  $F$  is the amount of inbreeding produced (0-100%) in a single generation;  $F$  is the percent increase in homozygosity. This formula shows that as  $N_e$  decreases,  $F$  increases (Figure 3.1);  $N_e$  s <50 produce large amounts of inbreeding per generation.

In a closed population, once inbreeding has occurred, it lowers the  $N_e$  of the next generation:

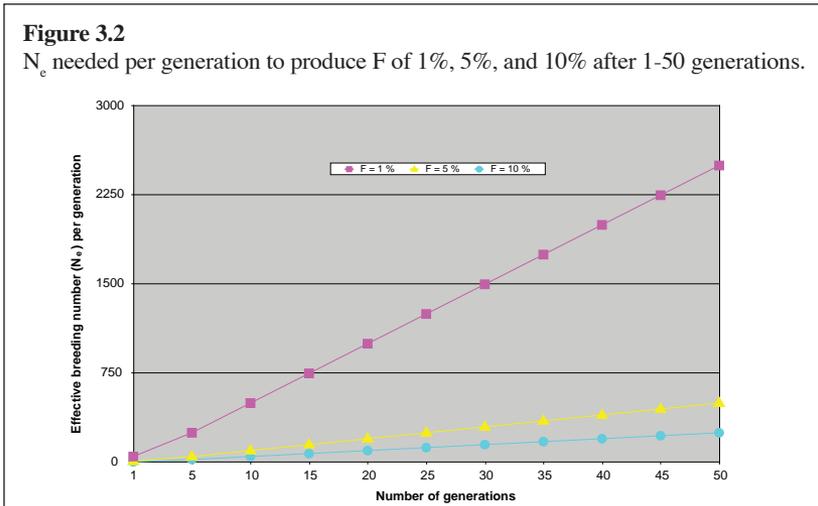
$$N_{eF} = N_e / 1 + F$$



where  $N_{ef}$  is the effective breeding number in a closed population with  $F > 0\%$ . For practical purposes, the total  $F$  that is produced over a series of generations can be calculated by summing the  $F$  that is produced in each generation, without considering previous inbreeding.

How big should  $N_e$  be to minimize inbreeding? Unfortunately, there is no universal value of  $F$  that aquaculturists or fisheries biologists want to avoid, which means there is no universal  $N_e$ .  $N_e$ s ranging from 30-500 have been recommended, 50 being the most common.<sup>14</sup> To calculate a desired  $N_e$ , one must determine what level of genetic risk is acceptable; in this case, it is the maximum amount of inbreeding that is desired after a given number of generations.<sup>15</sup> In addition, the concern about inbreeding, whether  $N_e$  needs to be managed, and how large  $N_e$  must be depends on the purpose/goal of the hatchery or fish farm, if fish are spawned, and how broodstock will be managed.

Figure 3.2 shows the constant  $N_e$ s that are needed to produce  $F$  of 1%, 5%, and 10% for 1-50 generations. The common recommendation of  $N_e = 50$  is effective in minimizing inbreeding for a single generation ( $F = 1\%$ ), but it does a marginal job after 10 generations ( $F = 10\%$ ).



<sup>14</sup> See footnote 11 and 13.

<sup>15</sup> See footnote 11 and 13.

Farmers who acquire brood fish from a breeding center (Chapter 5), spawn them once and then acquire new brood fish, or farmers who simply acquire genetically improved fingerlings for grow-out from “multiplier” hatcheries every growing season (Chapter 6) do not have to worry about inbreeding or managing  $N_e$ . The breeding centers or multiplier hatcheries must manage their stocks to minimize inbreeding, but these farmers do not need to worry about inbreeding.

It may be difficult for subsistence and small-scale farmers who maintain and spawn their own broodstock to manage inbreeding, but they should be encouraged to try, because improving their animal husbandry skills will lead to increased productivity. If they maintain a constant  $N_e = 50$  for 5 generations, they will keep  $F \leq 5\%$ . This recommendation produces good short-term (5 generations) management and this recommendation is not excessive, so many small-scale farmers could incorporate it into yearly work plans.

Large commercial farmers and those who produce fingerlings or conduct selective breeding programmes should try and keep  $F = 5-10\%$ , with  $F = 5\%$  being the desired goal for 10-20 generations, so that selection and domestication aren't being used simply to counteract inbreeding depression. Those who raise fish for fisheries or conservation programs should try and keep  $F = 1-5\%$ , with  $F = 1\%$  being the desired goal for a minimum of 20 generations, since the major management effort in these enterprises is to prevent changes in the gene pool over a long period.

A critical concept in Figure 3.2 is that the  $N_e$ 's are constant  $N_e$ 's.  $N_e$  can be larger than the desired number, but if it is smaller for just one generation, the inbreeding goal will not be achieved. This is because the mean  $N_e$  over a series of  $t$  generations is not the arithmetic mean, but the harmonic mean:

$$N_{e\text{ mean}} = 1/t(1/N_{e1} + 1/N_{e2} + \dots 1/N_{et})$$

This formula shows that the generation with the smallest  $N_e$  has a disproportional impact on mean  $N_e$ .

There are a number of management techniques that can be used to increase  $N_e$ . The most obvious way to increase  $N_e$  is to increase the number of fish that are spawned and produce viable offspring and to spawn a 1:1 sex ratio. One way to increase the number of brood fish that are spawned in order to satisfy production quotas is to keep only a small portion of each family. These simple ideas are contrary to standard fish culture management; aquaculturists tend to spawn as few fish as possible due to the fecundity of fish, and often use a highly skewed sex ratio because this enables them to maintain fewer brood fish.

A second technique is to switch from random mating (the normal practice at most hatcheries) to pedigreed mating.<sup>16</sup> In pedigreed mating, each female leaves one daughter and each male leaves one son as broodstock for the following generation (it can be more than one as long as all leave the same number). Pedigreed mating dramatically increases  $N_e$ , and when the sex ratio is 1:1,  $N_e$  is twice the number that spawn. However, to do this, each family has to be raised in a separate culture unit until fish can be marked to ensure that each parent leaves an offspring of the correct sex.

A third technique is to equalize the number of offspring from each mating, because unequal reproductive success lowers  $N_e$ .<sup>17</sup> However, to do this, each family must be raised in a separate culture unit until family size can be equalized.

A fourth technique is to modify stripping practices.<sup>18</sup> If fish are stripped, milt should not be pooled or added in a sequential manner. These practices cause gametic competition and one male can fertilize most of the eggs, producing an  $N_e$  much smaller than expected.

A fifth technique is to stretch generations. The desired  $N_e$  s in Figure 3.2 are given for generations, not years. A generation is the time interval for the replacement of parents with their offspring. If the goal is to keep inbreeding below a given value for 20 years and the normal procedure is to use a 2-year generation interval, 10 generations will be produced during the 20-year plan. But if the generation interval could be stretched to 3 years, only 7 generations would be produced during the 20 years, which means a smaller  $N_e$  could be used to achieve the desired goal.

Sixth, change the population from a closed to an open population. The above discussion assumed that the population is closed. If 10-25% new brood fish are imported each generation, the amount of inbreeding that is produced

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<sup>16</sup> Tave, D. 1984. Effective breeding efficiency: An index to quantify the effects that different breeding programs and sex ratios have on inbreeding and genetic drift. *Progressive Fish-Culturist*, 46:262-268.

<sup>17</sup> Fiumera, A.C.; Porter, B.A.; Looney, G.; Asmussen, M.A.; Avise, J.C. 2004. Maximizing offspring production while maintaining genetic diversity in supplemental breeding programs of highly fecund managed species. *Conservation Biology*, 18:94-101.

<sup>18</sup> Withler, R.E. 1988. Genetic consequences of fertilizing Chinook salmon (*Oncorhynchus tshawytscha*) eggs with pooled milt. *Aquaculture*, 68:15-25; Withler, R.E. 1990. Genetic consequences of salmonid egg fertilization techniques. *Aquaculture*, 85:326.

can be drastically reduced.<sup>19</sup> In fisheries and conservation management, one approach is to capture and spawn wild brood fish or collect wild-spawned eggs and culture them. Care must be taken if brood fish are collected to avoid broodstock mining (i.e. reducing the number of fish that will spawn naturally in the wild to a dangerous level).

Seventh, a fish farmer can maintain two unrelated populations and produce hybrids. Hybrids have inbreeding of zero; hybridization is often used in plant and animal breeding programmes to eliminate or to counteract inbreeding. If multiple unrelated lines are maintained, a rotational mating programme can be used to prevent inbreeding for a number of generations.<sup>20</sup>

Finally, a factorial mating pattern can be used to increase  $N_e$ , which will minimize inbreeding.<sup>21</sup>

### 3.3 Genetic drift

Genetic drift is random changes in gene frequency—changes that are not due to selection, migration, or mutation. The causes of the random changes can be natural, such as a landslide that divides a population or a storm that kills a large percentage of a population or destroys portions of its habitat, or it can be man-made, which occurs when fish culturists acquire or spawn their fish.

Under normal conditions, the number of fish that reproduce and leave viable offspring is far less than the number of adults; this is especially true in aquaculture. When this subsample spawns, there is a chance that the frequencies of one or more genes will be different in the offspring than they were in the parental generation, and the fewer that are spawned, the more likely that changes will occur. The ultimate effect of genetic drift is the loss of alleles, and the lower the gene frequency the more likely the allele will be lost via genetic drift. Aquaculturists also cause genetic drift when they choose which fish they will buy for their foundation population. The acquisition of fish is critical, and small samples often produce what is called the founder effect—a condition where genetic drift creates a population in which the gene

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<sup>19</sup> Bartley, D.M.; Kent, D.B.; Drawbridge, M.A. 1995. Conservation of genetic diversity in a white seabass hatchery enhancement program in southern California. *American Fisheries Society Symposium*, 15:249-258.

<sup>20</sup> Kincaid, H.L. 1977. Rotational line crossing: An approach to the reduction of inbreeding accumulation in trout brood stocks. *Progressive Fish-Culturist*, 39:179-181; See footnote 11.

<sup>21</sup> Busack, C.; Knudsen, C.M. 2007. Using factorial mating designs to increase the effective number of breeders in fish hatcheries. *Aquaculture*, 273:24-32.

frequencies are markedly different from those of the population from which they originated. The founder stock determines the maximum genetic variance that will exist in a closed population.

The loss of genetic variance makes a wild population more vulnerable to extinction, because it has lost the genetic variability that might have enabled it to adapt to changes in the environment. A number of hatchery stocks have been evaluated, and no matter how much effort was expended in preventing genetic drift, it occurred and reduced genetic variance.<sup>22</sup> The loss of genetic variance via genetic drift was shown to prevent selection for increased growth rate,<sup>23</sup> and has been shown to increase the number of fish with developmental disorders.<sup>24</sup>

The relationship between  $N_e$  and genetic drift is:

$$\sigma^2_{\Delta q} = pq/2N_e$$

where  $\sigma^2_{\Delta q}$  is the variance of the change in gene frequency (the way genetic drift is measured), and  $p$  and  $q$  are the frequencies of alleles  $p$  and  $q$  for a given gene.

As was the case with inbreeding, genetic drift is inversely related to  $N_e$ ; the smaller  $N_e$ , the more likely that genetic drift will change gene frequencies. The effect that a reduction in  $N_e$  can have on gene frequencies via genetic drift is immediate.

Because it is difficult to prevent genetic drift in managed populations, genetic drift must be partitioned into acceptable and unacceptable changes for management purposes. A change in the frequency of an allele from, say, 0.4 to 0.38 might not be critical so that can be classified as acceptable, but

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<sup>22</sup> Allendorf, F.W.; Phelps, S.R. 1980. Loss of genetic variation in a hatchery stock of cutthroat trout. Transactions of the American Fisheries Society, 109:537-543; Hallerman, E.M.; Dunham, R.A.; Smitherman, R.O. 1986. Selection or drift—isozyme allele frequency changes among channel catfish selected for rapid growth. Transactions of the American Fisheries Society, 115:60-68; Vuorinen, J. 1984. Reduction of genetic variability in a hatchery stock of brown trout, *Salmo trutta*. Journal of Fish Biology, 24:339-348.

<sup>23</sup> Tave, D.; Smitherman, R.O. 1980. Predicted response to selection for early growth in *Tilapia nilotica*. Transactions of the American Fisheries Society, 109-439-445; Teichert-Coddington, D.R.; Smitherman, R.O. 1988. Lack of response by *Tilapia nilotica* to mass selection for rapid early growth. Transactions of the American Fisheries Society, 117:297-300.

<sup>24</sup> Leary, R.F.; Allendorf, F.W.; Knudsen; K.L. 1985. Developmental instability as an indicator of reduced genetic variation in hatchery trout. Transactions of the American Fisheries Society 114:230-235.

the change in the frequency of an allele to 0.0 is critical and needs to be prevented, which means that is classified as unacceptable. Consequently,  $N_e$  must be managed to minimize the loss of alleles; since rare alleles are more likely to be lost than common ones, preventing the loss of rare alleles via genetic drift should be the management goal.

The probability of losing an allele of frequency  $q$  via genetic drift in a single generation is:

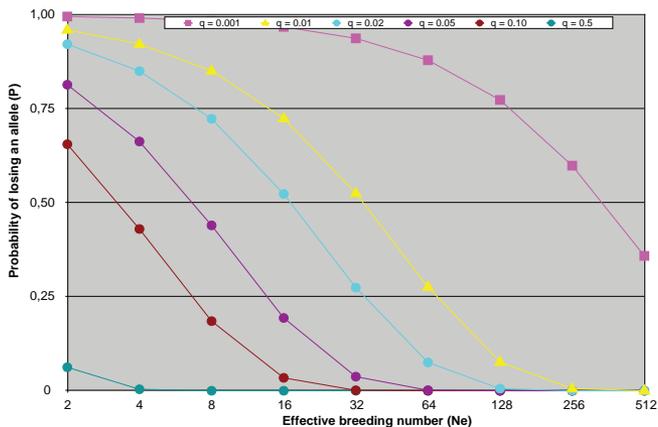
$$P = (1 - q)^{2N_e}$$

The probabilities of losing an allele ( $f = 0.001-0.5$ ) for a single generation are shown in Figure 3.3. Figure 3.3 shows that small  $N_e$  s are needed to prevent the loss of common alleles ( $f \geq 0.2$ ), while large  $N_e$  s are needed for rare alleles ( $f \leq 0.01$ ).

When managing a population's  $N_e$  to minimize genetic drift, one must determine what genetic risk is acceptable; in this case, it is the desired guarantee of keeping an allele ( $1.0 - P$ ) of a specific frequency after a given number of generations.<sup>25</sup> Geneticists and population biologists consider that an allele whose  $f = 0.01$  contributes to polymorphism, so the goal of fisheries

**Figure 3.3**

Probabilities of losing an allele ( $f = 0.001-0.5$ ) for various  $N_e$  s. These probabilities are for a single event (spawning season or acquisition of broodstock).



<sup>25</sup> See footnote 11.

management and conservation programs should be to save alleles whose  $f = 0.01$  (if this is done, more common alleles will also be saved). Saving rare alleles is not as important for food fish farming. If rare alleles improve viability, growth, and other culture traits, domestication will increase their frequency. Because of this, the genetic risk for food fish farmers can be to save alleles whose  $f = 0.05$ .

Constant  $N_e$  s needed to produce a 95% guarantee of saving alleles ( $f = 0.005$ - $0.1$ ) for 1-50 generations are shown in Figure 4. The methodology used to calculate these  $N_e$  s is described in an FAO book on managing inbreeding and genetic drift in hatchery populations.<sup>26</sup> It is easy to prevent the loss of an allele whose  $f \geq 0.05$ , but it can be difficult when  $f \leq 0.005$ .

As was the case for management of inbreeding, farmers who do not spawn fish or who only spawn fish once and then acquire new stock do not need to manage their population to minimize genetic drift. Even though most subsistence or small-scale farmers will not understand genetic drift or its consequences, many can easily incorporate management that will minimize its effects. If they maintain a constant  $N_e = 45$  for 5 generations, they will produce a 95% guarantee of saving an allele whose  $f = 0.05$ . This recommendation is not excessive and produces good short-term (5 generations) genetic management.

For large commercial farmers and those who produce fingerlings or conduct selective breeding programmes, the goal of saving alleles whose  $f = 0.05$  is easily achievable;  $N_e = 59$  will produce a 95% guarantee for 20 generations. The goal of saving alleles whose  $f = 0.01$  should be achievable for fisheries/conservation programs;  $N_e = 297$  will produce a 95% guarantee for 20 generations. The values in Figure 4 are for a single allele. If there are 100 such alleles, a 95% guarantee means that 95 will be saved, while 5 will be lost. The management techniques that were described in the inbreeding section can also be used to increase  $N_e$  in order to minimize genetic drift.

Recommended  $N_e$  s to minimize genetic drift have ranged from 500-5 000, with 500 being the most common.<sup>27</sup> Figure 4 shows that the common recommendation of  $N_e = 500$  will do a good job of minimizing genetic drift;

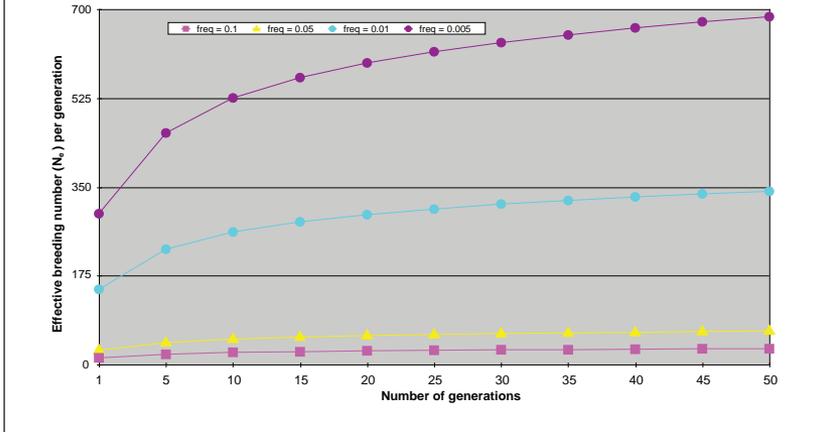
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<sup>26</sup> See footnote 11.

<sup>27</sup> Lande, R. 1995. Mutation and conservation. *Conservation Biology* 9:782-791; Hallerman, E. 2003b. Random genetic drift. Pages 197-214 *in* E.M. Hallerman, ed. *Population genetics: Principles and Applications for Fisheries Scientists*. Bethesda, MD, American Fisheries Society; National Research Council. 2002. *Science and the Endangered Species Act*. Washington, DC. National Academy Press; See footnote 11 and 13.

**Figure 3.4**

$N_e$  needed per generation for 1-50 generations to produce 95% guarantees of saving alleles whose  $f = 0.1-0.005$ .



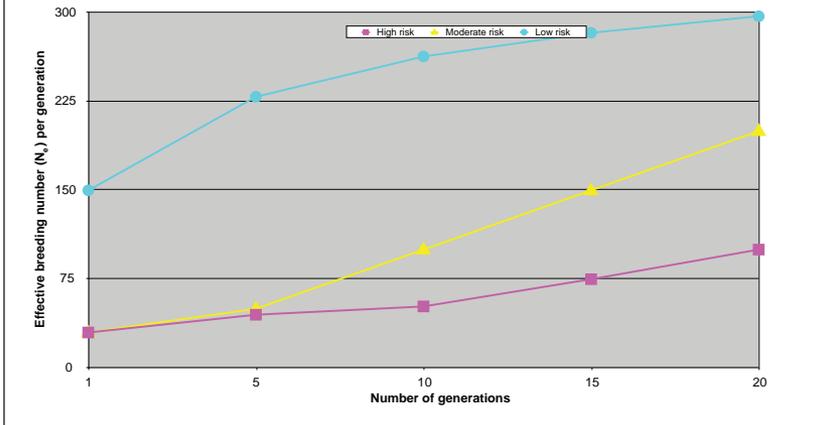
it will produce a 95% guarantee of saving an allele whose  $f = 0.01$  for >50 generations. However, depending on the genetic goal (risk), the desired  $N_e$  can be less than 500, which is often the case for food fish farming.

Since both inbreeding and genetic drift are inversely related to  $N_e$ , it should be managed to minimize both. The information in Figures 3.2 and 3.4 can be combined to create a constant  $N_e$  to achieve both goals; to achieve both goals, the larger  $N_e$  must be used. Figures 3.5 and 3.6 list constant  $N_e$  s needed for food fish and for fisheries/conservation aquaculture, based on different levels of genetic risk.

Figure 3.5 shows that the  $N_e$  needed to minimize the damaging effects of inbreeding and genetic drift are not excessive and can be incorporated into most food fish operations. Even though genetic management is often considered either to be of little value or as inappropriate technology for subsistence or small-scale farmers, those who maintain and spawn their own broodstock can easily incorporate “moderate risk” (Figure 3.5) short-term (5 generations) management within the framework of routine work plans.  $N_e = 50$  will keep  $F \leq 5\%$  and will also produce a 95% guarantee of keeping an allele whose  $f = 0.05$  for 5 generations. In this case, the  $N_e$  needed to manage inbreeding is larger than the  $N_e$  needed to manage genetic drift, so

**Figure 3.5**

$N_e$  needed per generation to minimize inbreeding and genetic drift in hatchery populations on food fish farms.  $N_e$  s are for three options (level of genetic risk): high risk-- $F \leq 10\%$  and a 95% guarantee of keeping an allele whose  $f = 0.05$ ; moderate (acceptable) risk-- $F \leq 5\%$  and a 95% guarantee of keeping an allele whose  $f = 0.05$ ; low risk-- $F \leq 5\%$  and a 95% guarantee of keeping an allele whose  $f = 0.05$ .



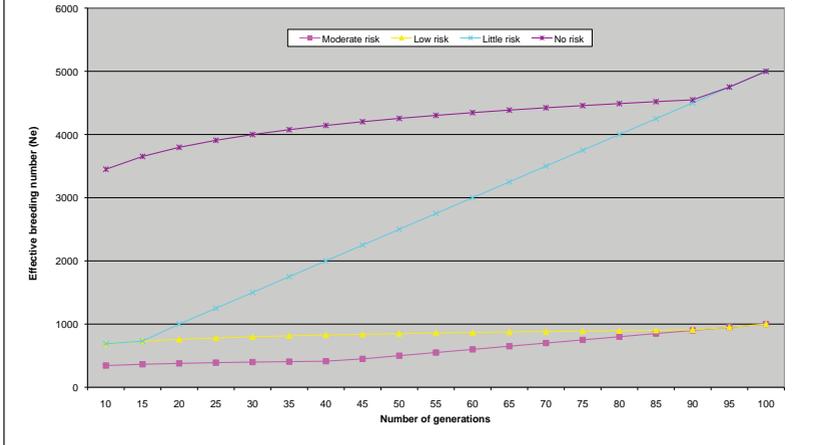
the inbreeding  $N_e$  is the one that is used. Because of this, extension agents only need to explain genetic management in terms of minimizing inbreeding, a concept that's easily understood, since most societies have taboos against consanguineous (blood-related) marriages.

If large commercial farmers, those who produce fingerlings, or those who conduct selective breeding programmes maintain a constant  $N_e = 100$  (this includes the foundation stock), they can minimize genetic problems for 10 generations ( $F \leq 5\%$  and a 95% guarantee of keeping an allele whose  $f = 0.05$  ("moderate risk" in Figure 3.5). However, if they acquire their stock from a farm where  $N_e$  was lower than 100, they will import fish that might already have accumulated a high level of inbreeding or suffered decreased genetic diversity and poor performance due to genetic drift.

The  $N_e$  s in Figure 3.6 are considerable larger, because managing a population's gene pool over a long period of time ( $\geq 10$  generations, with 20 generations being the desired minimum) should be the primary goal for fisheries/conservation-based aquaculture programs, and little genetic

**Figure 3.6**

$N_e$  needed per generation to minimize inbreeding and genetic drift in hatchery populations that are used for fisheries or conservation management projects.  $N_e$  s are for four options (level of genetic risk): moderate risk-- $F \leq 5\%$  and a 99% guarantee of keeping an allele whose  $f = 0.01$ ; low risk-- $F \leq 5\%$  and a 99% guarantee of keeping an allele whose  $f = 0.005$ ; little risk-- $F \leq 1\%$  and a 99% guarantee of keeping an allele whose  $f = 0.005$ ; no risk-- $F \leq 1\%$  and a 99% guarantee of keeping an allele whose  $f = 0.001$ .



risk should be accepted. The constant  $N_e$  s needed for 20 generations are 378-1 000, depending on the genetic risk. Although the “no risk” option in Figure 3.6 is the most desired from a purely genetic view, it is unlikely that it can be incorporated from a management perspective. Consequently, the “low risk” or “little risk” options in Figure 3.6 are those that should be incorporated into this type of work. The “little risk” option in Figure 3.6 combines  $F \leq 1\%$  with a 99% guarantee of keeping an allele whose  $f = 0.005$  rather than  $f = 0.01$ , because by generation 20 the  $N_e$  s are identical. If the combination of  $F \leq 1\%$  and  $f = 0.01$  for less than 20 generations is desired, the  $N_e$  s in Figures 3.2 and 3.4 can be used to produce the desired  $N_e$ .

It has been suggested that an  $N_e \geq 1,000$  will make a population “genetically secure.”<sup>28</sup> An  $N_e = 1,000$  will achieve “moderate” and “low risk” goals for 100 generations and will achieve the “little risk” goal for 20 generations (Figure 3.6).

<sup>28</sup> National Research Council. 2002.

The discussion about calculating  $N_e$  assumes that the population is fairly stable, which is often the case at a hatchery. When this is the case, the  $N_e$  for inbreeding and genetic drift is calculated as described earlier. However, when the population is small, fluctuates wildly over generations, or is declining, the  $N_e$  for inbreeding and genetic drift are different, because the effect of  $N_e$  on the population can be immediate (genetic drift) or delayed (inbreeding). When this is the case,  $N_e$  for genetic drift is called variance effective number ( $N_{ev}$ )<sup>29</sup> and is:

$$N_{ev} = \frac{4N - 2}{V_k + 2}$$

where  $N$  is the number of fish in the parental generation and  $V_k$  is the variance of offspring production.

$N_{ev}$  is far more important for fisheries and conservation management than it is for food fish culture, because genetic drift can have a more damaging effect on the ability of the species to survive in the wild. If the only genetic goal of fisheries/conservation programs is to minimize genetic drift,  $N_{ev}$  should be managed;  $N_{ev}$  is estimated for each generation, independently from those of the previous generations. In this case, the common recommendation of  $N_e$  ( $N_{ev}$ ) = 500 will do a good job of minimizing genetic drift from parents to their offspring for >50 generations (Figure 3.4). However, if  $N_{ev}$  of the population was small in previous generations, an  $N_{ev}$  = 500 will only keep genetic drift-produced problems from getting worse; it will not reverse genetic damage that has already occurred.

One reason to know the  $N_{ev}$  of hatchery-produced fish that are stocked for fisheries or reclamation projects and the  $N_{ev}$  of the wild population into which they are stocked is that the stocking program can actually decrease the overall  $N_{ev}$ .<sup>30</sup> For example, if the  $N_{ev}$  of the stocked fish is small but if they contribute a disproportionate number of offspring to the next generation, the  $N_{ev}$  of the population in the wild can decline. Another way  $N_{ev}$  can decline is if hatchery fish produce large families, while wild parents produce small families; this

<sup>29</sup> Waples, R. S. 2002. Definition and estimation of effective population size in the conservation of endangered species. Pages 147-168 in S.R. Beissinger; D.R. McCullough, eds. Population Viability Analysis. Chicago. The University of Chicago Press.

<sup>30</sup> Ryman, N; Laikre, L. 1991. Effects of supportive breeding on the genetically effective population size. Conservation Biology, 5:325-329; Waples, R.S.; Do, C. 1994. Genetic risk associated with supplementation of Pacific salmonids: captive broodstock programs. Canadian Journal of Fisheries and Aquatic Sciences, 51 (Supplement 1):310-329.

increases the variance of offspring production, which can actually produce an overall  $N_{ev}$  that is smaller than the combined  $N_{ev}$  s.

### 3.4 Domestication

Domestication is a form of selection that makes an organism more adapted to the culture environment and to all aspects of the management that is used to raise it (i.e. it changes a population's gene pool by selecting for alleles that are able to exploit culture conditions and by eliminating alleles that are less fit in the hatchery). Domestication is a combination of intentional and unintentional selection. Domestication changes the gene pool, so these changes are transmitted to subsequent generations and, over time, the population becomes markedly different. In agriculture, all of the important plants and animals that are raised for food have become domesticated; aquaculturists, on other hand, raise animals and plants that are not domesticated. Domestication is a term that is difficult to quantify, because there is no absolute line that divides wild from domesticated; instead it is continuous process (Domestication has been defined as defined as continuous controlled reproduction for more than 3 generations).<sup>31</sup> We can see an end product (domesticated) and a beginning (wild), but every aquacultured organism is somewhere along the continuum; most are nearer the beginning than the end. Domestication begins when fish culturists take control of the fish's life cycle and determines the conditions under which the fish will be raised (the kind of feed that is used; the stocking rate; water quality management, etc.) and, most especially, which fish will be spawned.

Domestication can change a population in subtle ways due to unintentional selection as the fish adapts to the way a farmer or hatchery manger operates the facility. For example, the way a farmer spawns his fish can produce a stock that responds more readily to hormone injections, or the spawning season can shift if a hatchery manager only spawns fish at the beginning of the spawning season. Choosing brood fish from the first seine haul could select for fish that are easy to harvest, while choosing the fish that remain in the pond after several seine hauls could select for fish that are escape artists. Raising fish under intensive culture condition selects for fish that tolerate degraded, stressful living conditions. Discarding brood fish that thrash about and that are difficult to handle can produce more docile fish.

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<sup>31</sup> Bilio, M. Controlled reproduction and domestication in aquaculture. The current state of the art. Part II. Aquaculture Europe, 32 (3): 5-23.

Simple behavioral modification may or may not be a component of domestication. For example, fish learn when they will be fed and respond to the sound of a feed truck or the noise the feeder makes. This behavior is desirable, because it ensures that the feed will be eaten quickly and less will be wasted. If such behavior has a genetic basis, then it is a part of domestication, but if this learned behavior does not have a genetic basis then it will not be transmitted to the next generation, and it is not domestication.

Domestication is beneficial in food fish culture, because it produces more docile fish, those that thrive on artificial feed which increases growth rate, and those that tolerate crowding, handling, and degraded water quality conditions that might induce stress and subsequent disease. Domestication has been shown to increase growth rate of farmed fish by 2-6% per generation.<sup>32</sup>

However, aquaculture also cultures fish that will be stocked in the wild to support culture-based fisheries (Chapter 8) and in endangered species recovery programmes (Chapter 9), and for this type of management, domestication is detrimental because it can produce unwanted changes in the genome.<sup>33</sup> Unfortunately, the simple act of spawning fish, the way they are spawned, or raising them creates domestication selection, which could produce fish that are less fit in the wild. For example, survival in the wild is tenuous and most fish die when they are fry. Aquaculturists create environments that maximize survival, so genotypes that would have been maladaptive in the wild are not lethal at a hatchery. Improving early survival in a hatchery, a routine practice in aquaculture, is a form of domestication selection that has been shown to have an indirect effect of selecting for smaller egg size which, in turn, decreases viability in the wild. Even if the only form of fish culture is the collection of wild eggs, culturing the fry/fingerlings for a brief period, and then stocking them, domestication selection will occur if survival and reproductive success of the stocked fish is different in the wild than that of their wild counterparts.

Management practices that minimize domestication for fish that will be stocked in the wild are listed in Chapter 8.

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<sup>32</sup> Dunham, R.A.; Smitherman, R.O. 1983. Response to selection and realized heritability for body weight in three strains of channel catfish, *Ictalurus punctatus*, grown in earthen ponds. *Aquaculture*, 33:89-96.

<sup>33</sup> Araki, H.; Cooper, B.; Blouin, M.S. 2007. Genetic effects of captive breeding cause a rapid, cumulative fitness decline in the wild. *Science*, 318:100-103; Heath, D.D.; Heath, J.W.; Bryden, C.A.; Johnson, R.M.; Fox, C.W. 2003. Rapid evolution of egg size in captive salmon. *Science*, 299:1738-1740.

### 3.5 Constraints and opportunities

A major constraint to improve management in this area is the simple fact that most aquaculturists are poorly trained in genetics and feel that this type of management is not necessary—often because they do not understand it. A second constraint is that many aquaculturists are unaware of the long-term benefits to be derived from incorporating genetic management programmes (Chapters 4 and 6), and routinely raise fish that are inbred or have reduced genetic diversity and do not perform as well as genetically undamaged stock would. They also do not realize that improvements in other aspects of fish husbandry (e.g. better feed) may only offset decreases in genetic potential. There are increased costs associated with minimizing inbreeding and genetic drift, but these will be offset by improved production (Chapter 6). A second constraint occurs when hatchery managers and resource managers are erroneously rewarded for producing lots of fish, but fish that do not necessarily perform well because proper genetic management and evaluation aren't done. Fish culturists and resource managers must understand that producing fewer fish that perform better actually improves productivity and resource management. A third constraint is financial in that many hatcheries cannot be expanded, rebuilt, and extra labour cannot be hired, so it is not possible to spawn the required number of fish or minimize domestication for fisheries/conservation programmes. Finally, incorporating genetic management into fish culture must go hand in hand with good aquaculture husbandry and developments in nutrition or fish health.

Fortunately, many leaders and policy-makers are beginning to understand that management of genetic resources is not an abstract concept, but one that can improve food security and ecological stability.

## **4 GENETIC IMPROVEMENT METHODOLOGIES IN AQUACULTURE<sup>34</sup>**

### **4.1 Introduction**

The FAO Code of Conduct for Responsible Fisheries contains several articles that address issues specific to the application of genetic improvement methodologies in aquaculture (Articles 9.1.2, 9.1.3, 9.3.1 and 9.3.3). These articles refer to the risks associated with the development of aquaculture, and specifically of the development and spread of genetically altered stocks and to the genetic and environmental integrity of natural ecosystems. This chapter focuses on reviewing the methodologies used in the genetic improvement of aquaculture species and the extent of their current and likely future application in aquaculture. These methodologies are considered with a view to their short and long term benefit to the development of aquaculture but with references made to the risks posed by their adoption and application. These risks are described in more detail in Chapters 7 and 9. Guidelines are presented for consideration in the application of the various genetic improvement technologies in aquaculture.

As has been indicated in other chapters most forms of human intervention in more than one generation of the life cycle of cultured species will genetically alter the stock through changes in gene frequencies. Chapter 3 has outlined the processes whereby this genetic alteration can occur, namely through inbreeding and genetic drift and the change of gene and allele frequencies (and associated phenotypic traits) through unconscious selection. These genetic changes are generally the result of ignorance of the genetic consequences of management of captive stocks through successive generations. This chapter focuses on the additional consequences associated with the deliberate genetic change of cultured stocks through the application of a range of genetic improvement technologies.

### **4.2 Genetic improvement in aquaculture**

The modern era of genetic improvement in aquaculture has been underway since the mid 1970s with the initiation of selective breeding programmes in Norwegian salmon. It is only over the past two decades that there has been widespread acceptance that genetic improvement has an important role to play in aquaculture development and that very significant genetic gains

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<sup>34</sup> Contributed by Graham C. Mair

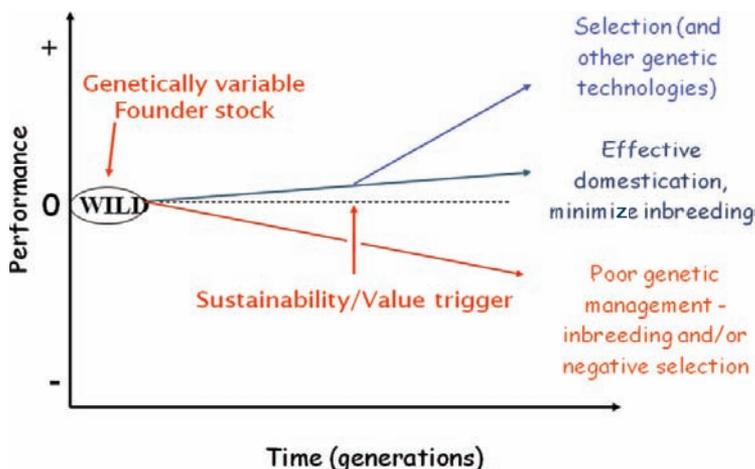
can be achieved through the appropriate application of well planned genetic breeding programmes in aquatic species. As a result there are now many such breeding programmes being implemented worldwide.

There is also a changing perception of the role of genetics in aquaculture and the appropriate timing of genetic interventions. It is now more widely understood that it is important to manage genetic variation from the time that stocks are first domesticated (or introduced) in order to avoid deterioration of stocks through inbreeding, genetic drift and unconscious selection (see Chapter 3), to take advantage of domestication selection and to maximize potential for subsequent genetic improvement (Figure 4.1).

With many aquatic organisms used in aquaculture there are actually more options for genetic improvement than can be readily applicable in higher

**Figure 4.1**

Hypothetical illustration of the different scenarios that can arise with poor vs. good genetic management. Ignorance of genetic management issues has on numerous occasions resulted in a decline in performance of cultured stocks. The objective of a developing aquaculture sector should be to effectively manage genetic diversity in domesticated stocks enabling the benefits from domestication selection to be realised. Management and retention of genetic diversity provides the raw material for the success of selective breeding at such time as an enterprise or production sector reaches a level of maturity, value and/or economic sustainability as to trigger investment in planned genetic improvement.



organisms as a result of their particular biological features and properties. Thus techniques such as chromosome set manipulation, sex control and transgenesis can be applied enabling many interesting improvement options to be explored.

While significant progress is being made and genetically improved stocks are becoming more readily available in aquaculture, the majority of cultured fish are still very similar to wild genotypes. There is a long way to go before aquaculture will attain the status seen today for livestock and crops where production is based almost exclusively on genetically improved varieties and the culture of wild germplasm is not practised.

### **4.3 Approaches to genetic improvement**

This section briefly describes the various genetic improvement methodologies applicable to cultured aquatic species, summarises the progress being made in modern day aquaculture and identifies the key issues for optimising benefits to aquaculture and minimizing risks to the genetic and environmental integrity of wild stocks.

#### ***4.3.1 Selective breeding***

The basis of selective breeding is to select individuals that possess a high additive genetic value for a desired phenotype (trait) as parents such that they can pass on their superior genes to progeny in following generations. In this way it should be possible to shift the mean value of the target trait for the cultured population in the desired direction in each successive generation. In selective breeding programmes it is necessary to minimize loss of genetic variation (e.g. that might arise through inbreeding) during this process to ensure that genetic gains are achieved and sustained for many generations.

Given the long term commitment implicit in initiating selective breeding, it is imperative that the stock to be improved is part of a commercially significant and sustainable aquaculture sector. It is therefore important to review the development and future potential for the sector in question prior to investing in any long term genetic improvement strategy.

A number of other conditions must be met prior to initiating a breeding programme. Firstly, the lifecycle of the species in captivity must be closed. It is then necessary to identify the **commercially valuable** trait or traits to target through a process of estimation of economic weights of traits that can

realistically be modified through genetic improvement. These traits should be variable within the population and preferably quantifiable in reproductively viable animals (traits such as fillet yield which are best measured in animals which have been sacrificed, present greater challenges for inclusion in selective breeding programmes). Ideally traits for selection should have moderate to high levels of heritability, and thus be likely to readily respond to even basic selective breeding. Heritability is a measure of how much of the variability in a particular trait is determined by genetics and varies from 0 (no genetic influence) to 1 (where genetics controls the entire trait). In more formal terms heritability is the proportion of phenotypic variance of a trait contributed by additive genetic variance and reflects the potential for generating a response to selection for that trait. Heritabilities of 0.15-0.5 indicate a trait will respond well to selection although there are procedures and statistical analyses that can effectively select for traits of low heritability.<sup>35</sup> It is also important to determine phenotypic and genetic correlations between traits particularly when developing selection indices in which two or more traits may be combined into a single index value.

A vital first step in initiating a selective breeding programme is the identification or development of an appropriate founder stock. To maximize the potential for long term genetic gain and enhancement of culture performance this founder stock should be highly genetically variable and should be based primarily on the best performing stocks available (where performance data is known or can be obtained). In reality this will likely involve the creation of a composite founder stock using source germplasm of varying origin, very often derived from different genetically discrete populations (domesticated and/or wild) as was the case in at the commencement of a major tilapia breeding programme known as GIFT.<sup>36</sup>

It is thus important to understand that well constructed founder stocks for selective breeding programmes will be genetically distinct from any individual wild population and thus represent a potential threat to the genetic integrity of those populations as, over time, gene frequencies in the wild could be changed by significant introgression of wild with genetically altered cultured

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<sup>35</sup> Gjedrem, T. 2005. Selection and Breeding Programmes in Aquaculture. Springer, Netherlands. Van Vleck, L.D. (1993) « Selection Index and Introduction to Mixed Model Methods », CRC Press Inc., Florida, USA

<sup>36</sup> Eknath, A.E., Bentsen, H.B., Ponzoni, R.W., Rye, M., Nguyen, N.H., Thodesen, J. & Gjerde, B. 2007. Genetic improvement of farmed tilapias: Composition and genetic parameters of a synthetic base population of *Oreochromis niloticus* for selective breeding. Aquaculture 273: 1-14.

stocks. As selective breeding progresses the genetic identity of selected stock is likely to diverge progressively further from any wild stock (See Chapters 3, 4, 8 and 9). One means to address this risk is to develop selected lines only from indigenous stocks (i.e. founders are taken from within discrete local populations) and to then limit their culture to sites within the natural distribution of that population. This option is however likely to be cost prohibitive. Selective breeding focuses on the improvement in quantitative traits (i.e. those phenotypes that are quantitative in nature and continuous in distribution) and growth rate is commonly the first trait considered for improvement. Qualitative traits (such as colour, body or fin shape or sex) are usually controlled by one or two gene loci and can be influenced through gaining an understanding of their inheritance. This is commonplace in ornamental aquaculture sectors but has little relevance to aquaculture for food production.

Growth rate is often the key trait of highest economic value in the majority of aquaculture sectors and this is especially true in extensive and semi-intensive systems where fish are sold whole or gutted and chilled rather than processed. Successful selection for growth can enable fish to be harvested at a larger and more valuable size, to shorten the culture period or possibly to stock animals at higher densities whilst maintaining harvest size. More intensive production systems often give relatively greater priority to traits such as food conversion and fillet yield due to their relatively greater economic importance, although these particular traits are more difficult to select.

It is beyond the scope of this chapter to review the different selection methods and breeding programme designs in any detail.<sup>37</sup> The decision on the method of selection to be used depends on a range of factors including the heritability of the target trait (if known), the type of trait, its ease of measurement in reproductively viable organisms and the biology and reproduction of the species (including fecundity). Table 4.1 summarises the key features of different approaches to selective breeding. The aim of any selection programme is to select the best individuals to be used to create the next generation without losing genetic variation. Breeding programme design invariably involves a series of compromises on what traits or combination of traits are selected for. Ideal designs will include fully pedigreed matings but are rarely feasible and are limited by physical and human resources, numbers of animals, marking/tagging systems (or the capacity to physically separate

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<sup>37</sup> Detailed manuals exist for example Tave, D. 1995. Selective breeding programmes for medium-sized fish farms. FAO Fisheries Technical Paper T352. FAO, Rome.

**Table 4.1** Summary of main properties of the different options for selective breeding design. <sup>a</sup>

<b>Type of Selection</b>	<b>Description</b>	<b>Advantages</b>	<b>Disadvantages</b>
Individual or mass selection	All individuals are pooled and measured. Selection of those to use as future broodstock is based only on the phenotypic value for the target trait in the individual.	Easy and requires few records Individual identification and pedigree records are not required Relatively low cost Can make gains for traits with high heritability	Limited to traits with high heritability Can only be applied to traits that are measured on the breeding candidate Difficult to control inbreeding Important to control key environmental effects such as age, size at stocking, water quality, feeding etc. Potential for highly variable progeny contribution to future generations promoting genetic drift
Within cohort selection	A variation in which broodstock are arbitrarily divided into cohorts with individual selection carried out within cohorts. Inbreeding is avoided by rotational mating of selected individuals with those from other cohorts	Controls inbreeding Pair mating not required Individual identification and pedigree records not required Eliminates environmental effects on individual performance within cohorts Requires limited technical expertise	Works best with traits of high heritability Can only be applied to traits that are measured on the breeding candidate Still requires control of age within cohorts
Within family selection	Multiple families are maintained with individuals selected independently within each family based on the deviation of their trait value from the family mean.	Can control inbreeding by rotational mating between families Relatively easy to manage Useful where environmental variance is common to members of a family Easy to control environmental variance for each family Allows for high selection intensities	Does not fully exploit heritability and thus response to selection is reduced Does not take account of variance in performance between families Can only be applied for traits that can be measured on the breeding candidate Families must be maintained separately or all individuals marked by family

<sup>a</sup> A useful summary of the key features of these approaches to selective breeding are presented in Ponzoni, R.W., Nguyen, N.H & Khaw, H.L. 2006. Importance and implementation of simple and advanced selective breeding programs for aquaculture species in developing countries. Proceedings of the 8<sup>th</sup> World Congress on Genetics Applied to Livestock Production, 13-18 August 2006, Belo Horizonte, MG, Brazil. (Web reference: [http://www.wcgalp8.org.br/wcgalp8/articles/paper/9\\_683-1814.pdf](http://www.wcgalp8.org.br/wcgalp8/articles/paper/9_683-1814.pdf))

**Table 4.1** (continued) Summary of main properties of the different options for selective breeding design

<b>Type of Selection</b>	<b>Description</b>	<b>Advantages</b>	<b>Disadvantages</b>
Combined selection	Selection of individual is based on individual information and information from relatives. Mean values for each family is determined and whole families are retained or culled depending on this mean.	Fully exploits additive genetic variance Inbreeding can be controlled Good for traits with low heritability Good for traits that can't be measured in breeding individuals (e.g. fillet yield) or that introduce risk such as disease challenge tests, as siblings can be selected Models developed to take account of all systematic effects on the target trait	Requires pair mating and family identification Generally expensive requiring large facilities and labour intensive. Costs are reduced if families are marked and stocked in common environment. Requires high levels of genetics expertise Common environment effects should be minimized and family size maintained Requires detailed record keeping including pedigrees Generally need to rear individual families up to tagging leading to tank effects during separate rearing
<b>Adding value to selection</b>	<b>Description</b>	<b>Advantages</b>	<b>Disadvantages</b>
Pooled families and genotyping	Where genetic markers exist that can enable pedigree assignment, individuals to be selected can be pooled at a very early stage and their family of origin determined at harvest.	Permits families to be reared in a common environment throughout the production cycle Defers some costs until harvest reducing risk Less resources required	Differential survival in pooled batches, especially at young ages can lead to uneven family contribution. Family data only obtained at harvest Need to identify selected fish whilst genetic marker analysis is being conducted.
Selection indices	A selection index can be generated based on several commercially important traits weighted by their relative importance. Used as standard in combined selection.	Optimizes gains for multiple trait selection	Requires economic weights for the traits in the breeding objective

families and individuals) and the properties of the species (such as fecundity, generation time and ease of controlled breeding).

With the recent initiation of numerous selective breeding programmes some impressive results are starting to emerge in terms of response to selection with gains in key traits of 13 to 15% per generation being possible in well managed and well resourced breeding programmes. These gains are greater than commonly achieved in livestock breeding programmes and may be a function of the high levels of genetic variability found in cultured aquatic species and the high fecundity of many species enabling higher selection intensities than is possible in less fecund livestock. Clearly the economic value of such significant gains, over several generations, are very significant and Gjedrem's<sup>38</sup> estimate of a 15:1 return on investment in the Norwegian salmon breeding programme is not unrealistic (see also Chapter 6).

Given the relative infancy of selective breeding in aquaculture the selected stocks that are being developed are not yet highly phenotypically divergent from wild types in most cases and are still able to survive in the wild and to interbreed with wild relatives if they escape or are deliberately introduced into the environment from which they were derived. Over time with continued selection for commercially important traits this level of phenotypic divergence will increase to the extent that selected lines will be so specifically adapted as to be dependent on the culture environment and unable to thrive or reproduce away from it as is the case nowadays for many of our livestock and crop species. It is difficult to estimate how long this transition will take but it is likely to be many decades before the majority of aquaculture stocks become this divergent from wild types. During this transition phase selectively bred stocks represent a risk to the genetic integrity of wild stocks and steps should be taken to assess, manage and minimize this risk (Chapters 7&9).

#### ***4.3.2 Hybridization and cross-breeding***

Hybridization is breeding individuals from two separate species while cross-breeding is the mating of two different varieties/stocks within a species. Both these crosses are commonly made with the objective of exploiting non-additive genetic variance through identification of significant positive heterosis, also known as “hybrid vigour”, for commercially important traits. Positive heterosis occurs when the hybrid or crossbred performs better than

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<sup>38</sup> Gjedrem, T. 2000. Genetic improvement of cold-water fish species. *Aquaculture Research* 31: 25-33.

the average of the two parental species or stocks. In practical terms heterosis only becomes really significant when the hybrid or crossbred performs better than either parental species or stocks. When evaluating heterosis it is important to evaluate reciprocal crosses as heterosis can vary depending on the maternal or paternal parent species/stock.

Both cross-breeding and hybridization are relatively simple techniques to master and can have an immediate impact on performance within one generation. However, this benefit is finite and only optimized in the specifically targeted hybrid cross between the original parental lines, unless the parental lines are then selected over generations for their general or specific combining ability, resulting in complex and relatively slow breeding programmes. Some heterosis can be retained in later generations if the numbers of populations crossed is high. Cross-breeding is thus usually looked upon as a potential supplement to a programme for additive genetic improvement, as mentioned above. For example it is possible to negate the effects of in-breeding that might occur in individual mass selected lines by generating production stock by crossing between two such lines. Evidence of significant heterosis for commercially important traits is relatively rare although substantial evidence for heterosis for growth suggests a role for cross-breeding in commercial improvement of oysters.<sup>39</sup>

Due to its relative simplicity there has been considerable research effort to evaluate hybrid crosses between multiple species with hundreds of hybrid crosses being attempted in the past three decades. Considering this large research effort, particularly with cyprinids in Asian aquaculture, there are relatively few hybrids in commercial production. While the use of hybrids in aquaculture may be under reported to some extent this relatively poor rate of return on hybridization research would indicate that commercial benefits of most hybrids are limited or nonexistent. There are a few examples where there has been commercial uptake of hybrids such as hybrid tilapia (*Oreochromis niloticus* x *O. aureus*) in China and Israel, hybrid catfish (*Clarias macrocephalus* x *C. gariepinus*) in Thailand and Southeast Asia, and hybrid striped bass (*Morone chrysops* x *M. saxatilis*). The success of these hybrids is due to “complementary effect” (i.e. specific marketable properties of the combination of the parental species such as high % males in the tilapia hybrids and good product quality attributes in the hybrid catfish) rather than positive heterosis for quantitative traits.

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<sup>39</sup> Hedgecock, D., McGoldrick, D.J. & Bayne, B.L. 1995. Hybrid vigor in Pacific oysters: An experimental approach using crosses among inbred lines. *Aquaculture* 137: 285-298.

Where hybrids are produced commercially a major challenge is the risk of introgression of pure species parental stocks through contamination by hybrids (i.e. backcrossing of hybrids with parents). This should be avoided as once introgression occurs the system breaks down and performance of the supposed  $F_1$  hybrids (which may actually be a mix of  $F_2$  hybrids and backcrosses) will become inconsistent and unpredictable.

Given the relative ease of hybridization between closely related fish species, hybridization can be haphazard, as has been seen for example in the production of major carps in some countries.  $F_1$  hybrids may be used either accidentally or deliberately as broodstock in backcrosses or in the production of  $F_2$  crosses. Over generations this will lead to a general mixing and segregation of genes from the original parental species, known as introgression. With this independent segregation of the genes the resulting phenotypes are highly variable and some of the fish carrying the introgressed genes cannot be easily distinguished from the original pure species. Introgression is now relatively commonplace in tilapia and other interbreeding species groups, where hybrids can be easily produced either artificially or naturally. Where this is occurring in Chinese and Indian major carp (e.g. in Bangladesh<sup>40</sup> where hybrids were originally produced, either out of scientific interest or through reasons of shortage of broodstock of some species) hybrid introgression is very likely to have negative consequences for the widespread carp polyculture systems as a result of loss of the distinct feeding strategies of the pure species, with a resulting decrease in feeding efficiency and production. Where unplanned hybridization events are unavoidable the long term consequences of such ad hoc hybridization events can be minimized if systems are in place that exclude hybrids from use as future broodstock.

It is thus recommended that hybridization in aquaculture should be avoided unless it is part of a systematic strategy to exploit heterosis or complementary effects for commercially important traits. The major challenge in planned cross-breeding and hybridization is to ensure that it is used appropriately, that there are real economic benefits to a hybridization programme and that it is

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<sup>40</sup> Mia, M.Y., Taggart, J.B., Gilmour, A.E., Gheyas, A.A., Das, T.K., Kohinoor, A.H.M., Rahman, M.A., Sattar, M.A., Hussain, M.G., Mazid, M.A., Penman, D.J. & McAndrew, B.J. 2005. Detection of hybridization between Chinese carp species (*Hypophthalmichthys molitrix* and *Aristichthys nobilis*) in hatchery broodstock in Bangladesh, using DNA microsatellite loci. *Aquaculture* 247: 267-273.  
 Simonsen, V., Hansen, M.M., Mensberg, K-L.D., Sarder, R.I. & Alam, S. 2005. Widespread hybridization among species of Indian major carps in hatcheries, but not in the wild. *J. Fish Biol.* 67: 794-808.

managed such that unwanted or uncontrolled introgression does not occur in hatchery or wild stocks.

### ***4.3.3 Chromosome set manipulation***

In fish and shellfish it is possible to manipulate whole sets of chromosomes through disruption of the process of cell division in the newly fertilised egg. These techniques are generally not possible in higher organisms. There are four types of manipulation which have been commonly applied, gynogenesis, androgenesis, triploidy and tetraploidy.

Androgenesis and gynogenesis are forms of induced uni-parental inheritance or parthenogenesis in which respectively the female or male genetic contribution is de-activated by some form of irradiation of gametes and the chromosome complement from the male or female is then doubled. The doubling of the haploid complement is achieved by application of physical (in finfish) or sometimes chemical (mainly in molluscs) shocks to restore diploidy. The resulting gynogenetic or androgenetic diploid progeny are highly or fully inbred depending on how diploidy is restored. Homozygous (i.e. 100% inbred) individuals can be used as the basis for producing isogenic clonal lines in which all fish are genetically identical. Gynogenesis and to a lesser extent androgenesis have been applied to a wide range of finfish and shellfish species and have a number of research and practical applications such as for the elucidation of the genetic basis of sex determination, the rapid induction of inbreeding, genetic mapping and QTL (quantitative trait loci) analysis. Androgenesis can also be used in principle for recovering genotypes from cryopreserved sperm where eggs from the same species are not available. There have however been very few commercial applications of these technologies.

Polyploids are produced in a similar way with application of physical or chemical shocks to normal fertilized eggs with disruption of the second meiosis resulting in triploids with two maternal and one paternal chromosome set. Disruption of mitosis produces tetraploids with a duplicate diploid chromosome complement. Triploids have been produced in a wide range of fish and mollusc species and can be achieved on a commercial scale for some species provided that large scale artificial fertilization of eggs is possible. Viable tetraploids have been produced in only a few commercially important species, predominantly salmonids and oysters.

The main application of chromosome set manipulation in aquaculture has been associated with the sterility of induced triploids which have been

produced in many fish species and several bivalve molluscs. Sterile fish have attractions for aquaculture. Firstly they may put relatively more energy into somatic growth and secondly they provide potential biological containment benefits facilitating the culture of exotic genotypes and possibly in the future, the culture of growth enhanced transgenic fish. However, triploid finfish generally do not grow faster than their diploid counterparts<sup>41</sup> although this may occur post-maturation when the triploids may also have higher dress-out proportions. All induced triploid female finfish produced to date have been shown to be fully sterile; triploid males show more gonad development than females, are generally sterile but rare incidences of fertility of triploid male finfish cannot be completely discounted. Conversely, many studies have shown that triploid bivalves, although not fully sterile in several species, show better performance than control diploids.

The most significant commercial application of chromosome set manipulations are in bivalve shellfish where triploids are cultured widely, for example approximately 50 percent of cultured oysters produced in the United States of America and France are triploids. Crustacea have not proved amenable to chromosome manipulation research due to the challenge of obtaining ovulated eggs for artificial fertilization. However, manipulations have been possible in some species including reports of triploidy having been successfully induced in Penaeid shrimp. Triploids can also be produced from diploid x tetraploid matings in the few species in which tetraploids have been produced and shown to be viable, most notably in oysters<sup>42</sup> and this is likely to represent a more commercially viable and reliable means of mass producing triploids.

Sterile triploids are likely to grow in importance in aquaculture for protection of breeders rights and for biological containment. For this latter application it will be necessary to quickly and cost effectively verify rates of triploidy induction and assess the potential for fertility of triploid fish. These factors are vital in determining the risk that escapes or releases from culture will be reproductively viable. The main challenges for these technologies are to reliably and verifiably produce 100 percent sterile fish in a range of commercially important species, particularly those where the resulting options for biological containment and/or intellectual property protection have significant value.

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<sup>41</sup> Tiwary, B.K., Kirubakaran, R. & Ray, A.K. 2004. The biology of triploid fish. *Reviews in Fish Biology and Fisheries* 14, 391-402.

<sup>42</sup> Guo, X. & Allen, S.K. 1994. Viable tetraploids in the Pacific oyster (*Crassostrea gigas* Thunberg) produced by inhibiting polar body 1 in eggs from triploids. *Molecular Marine Biology and Biotechnology* 3: 42-50.

#### 4.3.4 Sex control

There is a strong commercial incentive to culture single sex (monosex) populations in species in which there is significant sexual dimorphism for commercially important traits and where species become sexually mature within the culture environments before attaining harvest size. There may also be applications of monosex stocks for biological containment although this is less effective than using sterile stocks. These factors combined can profoundly affect the profitability of culture in some species, most notably in the tilapias.

Monosex or near monosex populations can be generated through manual sexing, hybridization, selection and direct and indirect use of hormonal sex reversal. Manual sexing is labour intensive and inefficient and hybrid crosses only apply to specific combinations of species, again notably in the tilapias. It has also recently been shown that there is a genetic basis to temperature dependent sex differentiation in tilapia such that the percentage of males resulting from high temperature treatment of fry can be increased by selective breeding<sup>43</sup>. The most applicable methods for producing monosex stocks are through direct sex reversal using hormones or indirectly through genetic breeding programmes. Direct sex reversal can generally be applied regardless of the sex determining system and has been successfully achieved in a range of species by immersion of eggs and fry in hormone solutions or by feeding with hormone treated diets.<sup>44</sup> The indirect approach through the application of breeding programs, however, requires an understanding of the genetic mechanisms of sex determination in a species, the main factor for success being that it is a monogenic system such as male heterogamety (XX female; XY male) as in salmonids and some tilapia or female heterogamety (WZ female; ZZ male) as in some tilapias and crustacea. Figure 4.2 illustrates alternative breeding programmes to produce all male progeny in species with female heterogamety and all female progeny in species with male heterogamety.

The potential benefits of monosex female stocks in salmonid aquaculture have been long established and relate to the enhanced availability of female broodstock and to avoiding precocious maturation of males which results in

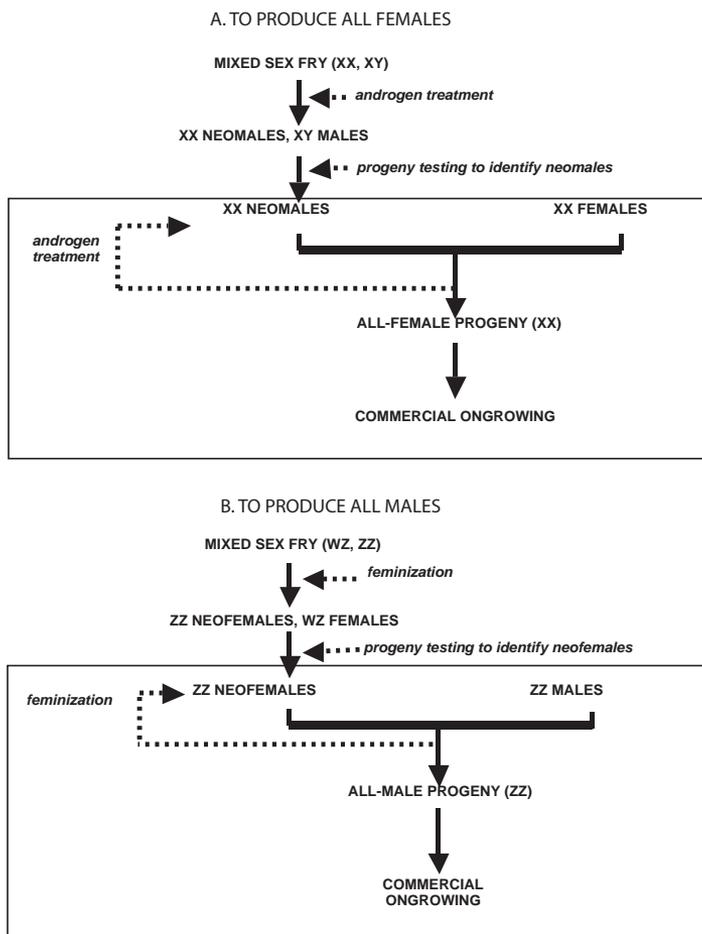
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<sup>43</sup> Wessels, S. and G. Hörstgen-Schwark. 2007. Selection experiments to increase the proportion of males in Nile tilapia (*Oreochromis niloticus*) by means of temperature treatment *Aquaculture* 272, Supplement 1: S80-S87.

<sup>44</sup> Piferrer, F. 2001. Endocrine sex control strategies for the feminization of teleost fish. *Aquaculture* 197: 229-281.

**Figure 4.2**

Illustrations of breeding programmes for production of monosex females (A) for species with male heterogamety and monosex males (B) in species with female heterogamety. Components within the boxes are those steps that can be repeated in cycles as part of commercial production in order to maintain supply of sex manipulated broodstock.



reduced growth, poorer survival and loss of flesh quality post-maturation. Use of all female progeny produced using sex reversed (neomale) broodstock is not universal but there are sectors in some states in which significant proportions of production are monosex.

Monosex male stocks have considerable commercial benefit in a number of species, most notably in tilapia due to problems of both precocious maturation and unwanted reproduction within the production system exhibited by this species. These can also be produced by either direct or indirect masculinization. Sex reversal to male has been achieved in a range of finfish through application of exogenous androgens such as through the administration of diets treated with methyltestosterone during the early life of the animal and is commonplace in tilapia hatcheries worldwide. Breeding programmes for all-male production are relatively easily achieved in female heterogametic species such as blue tilapia (*Oreochromis aureus*) and the giant freshwater prawn (*Macrobrachium rosenbergii*). In male heterogametic species it is also possible to produce genetically all-male progeny through the generation of novel YY “supermales” and such a breeding programme has been developed commercially with Nile tilapia *O. niloticus*.<sup>45</sup>

Sex control programmes are only of relevance to some species for which significant economic benefits will accrue from culture of monosex stocks. Direct induction of sex change by use of hormones is likely to meet social resistance from potential consumers of the treated fish even though studies have shown that excess exogenous hormone disappears from the tissues of the fish shortly after cessation of treatment. More acceptable would be the use of ecologically and ethically sound approaches (such as manipulation of environmental sex determination rather than by hormone treatment). Indirect approaches such as breeding programmes for monosex production are likely to meet with broader acceptance but face the major challenge that they are based on comprehensive understanding of the genetic mechanisms of sex determination which can require considerable research effort.

Sex control can be used as a form of biological containment on the basis that any introductions or escapes from aquaculture would not be able to breed with each other and thus be unable to form sustainable feral populations. Cultured stock would need to be guaranteed 100% monosex for this technology to be

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<sup>45</sup> Mair, G.C., Abucay, J.S., Skibinski, D.O.F., Abella, T.A. & Beardmore, J.A. 1997. Genetic manipulation of sex ratio for the large scale production of all-male tilapia *Oreochromis niloticus* L. *Canadian Journal of Fisheries and Aquatic Sciences* 54, 396-404.

an effective form of containment and it could only be applicable where there are no reproductively compatible species in the receiving environment.

#### **4.3.5 *Transgenesis***

Transgenesis is a genetic engineering technology wherein an isolated gene sequence from one organism is inserted into another organism to confer a new or modified trait. This isolated gene sequence is called a construct and is composed of a functional gene and a promoter gene that acts as a switch to activate the functional gene. Organisms resulting from successful transgenesis are classed as genetically modified organisms (GMO) and thus subject to societal and regulatory concerns. Early research used foreign gene constructs from other species, including terrestrial species. When planning transgenic research it is very important to be fully aware of the risks and concerns over ethics, human health and environmental impacts of transgenic fish and to understand the policy environment in which the research would be conducted and under which any products of the research would be regulated. A recommended response to the risks and concerns over transgenic fish is to focus research and development where appropriate on the production of autotransgenics in which the gene sequence introduced is derived from the same species.

Transgenesis has been a major area of research in fish genetics since the early 1990s. Research in this area is more advanced than in other livestock due to the relative ease of manipulation of reproductive biology in aquatic species. The induction of transgenesis should involve a number of steps: identification of the appropriate target gene and construct development; introduction of the gene into newly fertilized eggs, usually by micro-injection or electroporation; determination of incorporation of the transgene into the host genome; determination of transgene expression; determination of inheritance of the transgene and; quantification of the effect of the transgene on target and non-target traits. The final step in this sequence is of critical importance as it will be necessary to fully characterise the properties of the transgenic fish in order to be able to assess the potential risks associated with its culture. The main target trait of transgenic research in fish to date has been the enhancement of growth rate in aquaculture through the introduction of growth hormone gene constructs. Research has also targeted other traits such as disease and control of reproduction and the focus of transgenic research should be on traits which are difficult to improve through quantitative approaches. Transgenic fish can also be considered as useful models for studies of gene regulation and gene expression and have potential as biofactories to produce valuable pharmaceuticals.

The aforementioned development steps have been successfully completed in a number of finfish species and transgenic lines produced with sometimes dramatic increases in growth performance.<sup>46</sup> Clearly transgenesis has the potential to bring about fairly rapid changes in commercially important traits but awareness of associated risk is critical to planning and implementing such research (see Chapter 7).

Although enhanced performance under culture conditions has been clearly demonstrated for a number of species there are no transgenic food fish currently under commercial production. The only example on the market at present is the GloFish®, a fluorescent transgenic zebrafish which has been approved for sale and is only sold in the United States of America. At the time of writing there is an important test case in which the US Food and Drug Administration (FDA) is evaluating an application for a licence to commercialise a transgenic salmon species for use in aquaculture. While this is a lengthy process any conditional approval by the FDA (likely to be limited to closed, land based production systems) will represent a model for consideration of approvals for commercialisation of other lines of transgenic fish in other states.

Whilst there are some technical reasons behind the lack of commercialisation of transgenic fish the main reason is the concern over the ethical, animal welfare, human food safety and environmental risks associated with the culture of transgenic fish. Policy-makers, resource managers and those contemplating the use of GMOs should become very familiar with issues of environmental risk assessment and management with transgenic fish, both in research and potentially in commercial production.<sup>47</sup>

With solutions now developed for many of the technical constraints to successful application of transgenesis in fish, the main challenges lie in full assessment of environmental, ethical and consumer health risks which currently limit the commercialisation of this technology.

#### ***4.3.6 Genetics markers and marker assisted selection***

A genetic marker is a variation in a gene or sequence of DNA that can be identified by molecular techniques and used to allow identification of

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<sup>46</sup> FAO. 2000. *The State of World Fisheries and Aquaculture 2000*. Rome. 142pp

<sup>47</sup> Kapuscinski, A.R., Hayes, K.R., Li, S. & Dana, G. (eds). (E. M. Hallerman and P.J. Schei, series editors). 2007. *Environmental Risk Assessment of Genetically Modified Organisms*, Vol 3: Methodologies for Transgenic Fish. CABI Publishers. 310 pp.

genotypes and thus identify individuals or groups of interest. Prior to the advances in molecular genetics, isozymes and other proteins were the markers of choice. Nowadays there are a variety of DNA markers, such as mitochondrial DNA (mtDNA) polymorphisms, restriction fragment length polymorphisms (RFLP); random amplified polymorphic DNA (RAPD), repeat sequent markers (mainly microsatellites), amplified fragment length polymorphisms (AFLPs) and single nucleotide polymorphisms (SNPs). The most used markers in aquaculture genetics are microsatellites although AFLPs and SNPs are finding increasing application. Table 4.2 summarises the potential applications for genetic markers in and around aquaculture and lists the preferred markers for different priorities.

Polymorphic DNA markers have now been developed from DNA libraries for the majority of the important aquaculture species including the carps, tilapias, shrimps, salmonids and catfish. These markers have a number of important applications which are being increasingly utilized in aquaculture (mostly in research but there is increasing commercial application) as more enterprises invest in genetic programmes and the cost of genetic marker analysis falls.

The most common application of markers in genetic programmes currently is in parentage assignment in which the efficiency of selective breeding programmes can be enhanced by using genetic markers to identify selected fish to families and thus to individual parents. The removal of the need to keep families separate (either throughout the performance evaluation or at least until they can be physically marked) should reduce the problem of environmental effects on family performance and enable more families to be evaluated which can produce higher selection intensities and increase the response to selection.

In an ideal breeding programme, genetic markers can be used to: (1) characterise potential founder stock(s) to inform the development of a genetically variable base population; (2) to understand natural population structure to inform both the formation of founders stocks and the assessment of the risks posed by culture of genetically altered stocks; (3) to enhance the efficiency of selective breeding through pedigree assignment and (4) to characterize the longer term impact of domestication and genetic management (or mis management) of captive stocks (e.g. to determine loss of genetic variation where effective population size is sub optimal).

Genetic markers can also be used to construct genetic maps in which linked markers are assigned to linkage groups and ultimately to individual

**Table 4.2** Description of practical applications of genetic marker technologies in aquaculture.<sup>a</sup>

<b>Technique</b>	<b>Description</b>	<b>Applications</b>	<b>Remarks</b>
Strain/stock identification	A suite of genetic markers can be used to identify markers that are diagnostic for species or stocks. Preferred markers: Microsatellites, SNPs, AFLP, Isozymes and RAPD.	<ul style="list-style-type: none"> <li>• Discriminating different cultured stocks including protection of breeders rights on improved stocks</li> <li>• Identifying intentional or unintentional hybrid introgression</li> <li>• Identification or confirmation of escapes from aquaculture</li> <li>• Identification of recapture rates in enhanced fisheries</li> </ul>	Markers (of different types) diagnostic for species have been developed for many commercially important species and can be readily accessed. Markers for different stocks or strains generally need to be developed. Such markers may become increasingly important for traceability of aquaculture stocks.
Quantification or characterization of genetic variation	Genetic markers can be used to quantify and characterise levels of genetic variation such as number of alleles per locus, proportion of polymorphic alleles and average heterozygosity. Preferred markers: AFLP and microsatellites.	<ul style="list-style-type: none"> <li>• Determining likelihood and severity of inbreeding</li> <li>• Estimation of current and historic effective population sizes (N<sub>e</sub>)</li> <li>• Comparing the merits of candidate founder stocks for base populations for selective breeding</li> <li>• Confirmation of homozygosity in double haploids</li> </ul>	In genetic programs it is useful to have a standard set of genetic markers to determine baseline levels of variability (along with other applications) in founder stocks so that longer term impacts of domestication and genetic management can be quantified.
Determination of genetic relationships among stocks	Markers can be used to construct genetic relationships between multiple stocks such as phylogenetic trees. Preferred markers: mtDNA, microsatellites, AFLPs	<ul style="list-style-type: none"> <li>• Identifying the origin (e.g. source population) of cultured stocks</li> <li>• Determining the genetic structure of wild populations</li> </ul>	It is very useful to understand the genetic structure of wild fish in order to assess the risks of genetic contamination from aquaculture based on genetically altered stocks and to inform develop policies on translocation. Also useful information when forming genetically variable founder stocks

<sup>a</sup> Adapted from Liu, Z.J. & Cordes, J.F. 2004. DNA marker technologies and their applications in aquaculture genetics. *Aquaculture* 238: 1-37.

**Table 4.2** (continued) Description of practical applications of genetic marker technologies in aquaculture

<b>Technique</b>	<b>Description</b>	<b>Applications</b>	<b>Remarks</b>
Parentage assignment (pedigree determination)	The use of a suite of genetic markers to determine the probability that an individual progeny is derived from a cross of specific two parents Preferred markers: Microsatellites and SNPs	<ul style="list-style-type: none"> <li>• Determine the numbers of broodstock contributing to progeny in pooled spawning and thus estimate <math>N_e</math></li> <li>• Identify family of origin in selecting future broodstock to minimize inbreeding</li> </ul>	Cost of application of marker systems for parentage assignment is reducing (especially for SNPs) and they are increasingly used for identification of families in breeding programmes. Can be a problem in how to identify fish whilst they are genotyped for assignment
Marker Assisted Selection	The selection of a specific marker known to be associated with a commercially important trait (known as a quantitative trait locus – QTL) rather than the traits itself. Requires genetic maps to be constructed Preferred markers: SNPs, microsatellites and others	<ul style="list-style-type: none"> <li>• Currently no commercial application in aquaculture</li> <li>• Few species have been adequately mapped</li> <li>• Potential to be developed for traits that are difficult to improve using traditional approaches</li> </ul>	MAS is being used for genetic improvement for a small number of traits in livestock breeding but is not yet adequately developed or proven for aquaculture.

chromosomes. Genetic markers closely linked to genes that contribute to quantitative traits are known as quantitative trait loci (QTLs). Gene mapping programmes are now on going for several important aquaculture species including the Pacific oyster, salmonids, channel catfish, Nile tilapia, and European sea bass.<sup>48</sup> Linkage maps once developed, can be screened to identify QTLs of interest.

The QTL effect can then be quantified by correlating the inheritance of marker alleles with individual performance for the targeted trait. A number of QTLs for important traits have been identified in fish such as temperature tolerance, growth and disease resistance (e.g. cold tolerance in tilapia<sup>49</sup>).

Marker assisted selection (MAS) is the incorporation of genetic markers linked to QTLs into genetic improvement programmes and has the potential to enhance selection, particularly for traits which might have low heritability or which cannot be measured directly on the breeding individuals. Whilst there are a number of research efforts developing and evaluating QTL there are as yet, no commercial stocks utilising MAS.

The potential benefits of genetic markers in the majority of applications is not contested although the real potential for incorporation of marker assisted selection into breeding programmes and the production and economic gains that will result remain to be verified and this currently represents a major research challenge.

#### **4.4 The current status of genetic improvement and future scenarios**

It is difficult to estimate the proportion of global aquaculture production which is currently of domesticated stock but best estimates would indicate approximately 35% of aquaculture production is of undomesticated, essentially wild stock which are thus not adapted to captive environments. This compares with other forms of agricultural production which is almost exclusively of domesticated and genetically improved germplasm. The benefits of domestication in terms of adaption to captive environments is

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<sup>48</sup> Garber, A.F. and Sullivan, C.V. 2006. Selective breeding for the hybrid striped bass (*Morone chrysops*, Rafinesque x *M. saxatilis*, Walbaum) industry: status and perspectives. *Aquaculture Research* 37: 319-338.

<sup>49</sup> Cnaani, A., Hallerman, E.M., Ron, M., Weller, J.I., Indelman, M., Kashi, Y., Gall, G.A. and Hulata, G., 2003. Detection of a chromosomal region with two quantitative trait loci, affecting cold tolerance and fish size, in an F2 tilapia hybrid. *Aquaculture*, 223(1-4): 117-128.

such that it is recommended that any significant and potentially long term sustainable aquaculture industry should initiate domestication programmes in which the genetic diversity of stocks is effectively managed.

The proportion of global aquaculture production which is based on genetically improved stock (mostly selectively bred but also including monosex and triploid stock) is estimated to be between 10 and 20% so clearly there is scope for very significant increases in production and production efficiency through the widespread implementation of effective genetic improvement programmes with a focus on selective breeding.

Among the challenges facing the future development of aquaculture genetics is the relative lack of resources to support the development of breeding programmes. The resources in deficit include physical, economic and human resources. Well run breeding programmes generating strong genetic gains utilise facilities enabling the raising of multiple families of fish. Given their long term nature, they require sustained funding as it can often be many years before returns on investment in genetic programmes are fully realised through increased income from seed production or through improved production efficiencies (Chapter 6). A further limitation lies in human resources particularly in the area of quantitative genetics which generally requires specialised training to a relatively high level.

There seems little doubt that the consistently rising demand for aquaculture produce will continue to drive the search for improved production efficiencies and genetic improvement is becoming a major component of these efforts. Genetic improvement programmes are set to transform aquaculture stocks over the coming decades with selective breeding at the core of these programmes but with other technologies adding value to these efforts where clear benefits are apparent. Where states are promoting and/or investing in the expansion of aquaculture it is important to be aware of the basic principles of genetic management, the most cost effective approaches to genetic improvement and the environmental and ecological risks associated with the widespread uptake of improved stock by producers. Alongside these technological factors the provision of adequate resources to support the implementation of long term genetic improvement strategies is also critical.

## 5 DISSEMINATION OF GENETICALLY IMPROVED STRAINS AND MATERIAL TRANSFER AGREEMENTS <sup>50</sup>

### 5.1 Introduction

This section covers both (i) the transfer of genetically improved strains from one country to another and (ii) the multiplication and dissemination of germplasm within countries. Although related, there are also issues specific to each and they are therefore dealt with separately.

Article 9.1.2 of the Code of Conduct for Responsible Fisheries requires that “*States should promote the responsible development and management of aquaculture, including an advance evaluation of the effects of aquaculture development on genetic diversity and ecosystem integrity, based on the best available scientific information*”. In Article 9.3, it continues: “*States should conserve genetic diversity and maintain integrity of aquatic communities... should cooperate in the elaboration, adoption and implementation of international codes of practice and procedures for introductions and transfers...*” and “*minimize the risks of disease transfer and other adverse effects on wild and cultured stocks...*”. Technical guidelines for implementation of Article 9 (Aquaculture Development) of the Code were developed in 1997<sup>51</sup> and a wide range of instruments and tools has since been elaborated in the pursuit of responsible and sustainable aquaculture development. These guidelines also strive for consistency with the Convention on Biological Diversity<sup>52</sup> (see also Chapter 2) and other policy advisories designed to ensure wise use of wild and improved genetic resources.<sup>53</sup>

The following sections give general guidance on the dissemination of genetically improved strains between and within countries, with particular

<sup>50</sup> Contributed by R. E. Brummett, M. C. M. Beveridge, R. W. Ponzoni, R. J. Lawton and D. M. Bartley  
<sup>51</sup> FAO (1997). Aquaculture Development. *FAO Technical Guidelines for Responsible Fisheries*. No. 5, <ftp://ftp.fao.org/docrep/fao/003/W4493e/W4493e00.pdf>.

<sup>52</sup> <http://www.cbd.int/default.shtml>

<sup>53</sup> ICES (2004) *Code of Practice on the Introductions and Transfers of Marine Organisms* <http://www.ices.dk/reports/general/2004/ICESCOP2004.pdf>; Hewitt, C.L., Campbell, M.L. & Gollasch, S. (2006). *Alien Species in Aquaculture. Considerations for Responsible Use*. <http://www.iucn.org/dbtw-wpd/edocs/2006-036.pdf>. IUCN, Gland, Switzerland; WorldFish Center (2002) *Nairobi Declaration on Aquatic Biodiversity and Use of Genetically Improved and Alien Species for Aquaculture in Africa*. [http://www.worldfishcenter.org/cms/list\\_article.aspx?catID=39&ddlID=109](http://www.worldfishcenter.org/cms/list_article.aspx?catID=39&ddlID=109). WorldFish Center (2003) *Dhaka Declaration on Ecological Risk Assessment of Genetically Improved dFish*, [http://www.worldfishcenter.org/Pubs/Dhaka%20booklet/Dhaka\\_booklet.pdf](http://www.worldfishcenter.org/Pubs/Dhaka%20booklet/Dhaka_booklet.pdf)

reference to lines of fish improved through traditional selective breeding, as opposed to living modified organisms (LMO's)<sup>54</sup> or transgenic hybrids, which might be better considered as alien species introductions. These guidelines should serve as a starting point for the development of more situation-specific guidelines. The information provided is of technical nature, which is the focus of these guidelines. It does not cover some policy and legal aspects, such as access and benefit-sharing or intellectual property, that also regulate access to, and conditions to use, fish genetic resources.

As mentioned before, this chapter does not focus on the exchange of wild genetic resources, which may be provided to other countries for the purposes of research, breeding and training for aquaculture. Exchange mechanisms for genetic resources for food and agriculture in other sectors, such as crops, have so far received much more international attention than fish genetic resources for aquaculture. These mechanisms normally detail the rights and obligations of the provider and recipient with respect to the materials being transferred. Similar trends could be expected in aquaculture as exchange of fish genetic resources will increase in the next years with breeding programmes developing all over the world.

## **5.2 Transfer of an improved strain to another country**

### ***5.2.1 Introduction***

The Code promotes the use of the *Code of Practice on the Introductions and Transfers of Marine Organisms 2004 developed by the International Council for the Exploration of the Sea (ICES)* and Technical Guidelines on the Responsible Use and Control of Alien Species on the purposeful movement of fish from one country to another and encourage states to make these transfers in such a way that risks to indigenous biological and genetic diversity are minimized. There have been numerous documented cases of competition, predation, disease transfer and habitat damage resulting from the introduction of alien species and these should be treated with utmost caution.<sup>55</sup> In the case of selected lines, there is evidence from salmonids that modified gene

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<sup>54</sup> The Cartagena Protocol on Biosafety defines an LMO as an organism resulting from direct DNA manipulations or the fusion of cells from outside of a taxonomic family.

<sup>55</sup> Sindermann, C.J. 1993. Disease risks associated with importation of non-indigenous marine animals. *Marine Fisheries Review*, **54**, 1-10; McVicar, A. H. (1997) Disease and parasite implications of the coexistence of wild and cultured salmon populations. *ICES Journal of Marine Science*, **54**, 998-1008.

frequencies in fishes planted for stock enhancement or used in aquaculture can, when released into the wild and crossed with the wild genome, reduce the whole lifetime fitness of indigenous populations of the same or closely related species through genetic introgression (i.e. introduction of alleles into the wild population from the improved strain).<sup>56</sup>

### 5.2.2 *Guidance on transfer*

Rather than local, regional or international political boundaries, the biologically more important geographical unit to consider when contemplating a transfer of improved aquatic germplasm is the watershed.<sup>57</sup> Although government agencies should take into consideration both transfers within and from outside the country, a proposed transfer of fish within a watershed across political boundaries might be considered less critically than a transfer from one watershed to another within the same political jurisdiction or country.

In the absence of a dedicated national authority for germplasm transfers, requests for the introduction of improved lines should be made to the highest responsible fisheries official in the importing country (e.g., Director of Fisheries, Environment or Agriculture) on the basis of a sound environmental impact assessment (EIA) and cost: benefit analysis.

EIA guidelines and the analysis of potential negative costs associated with any importation should take into consideration:

- The presence of potentially valuable conspecific genetic diversity in the specific watershed to which the new material is to be imported.
- The presence of other rare or endangered aquatic biodiversity that might be negatively impacted by the introduction.
- The presence of suitable indigenous local species or genetic improvement strategies of existing farmed fish to use as an alternative introduction.

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<sup>56</sup> McGinnity *et al.* 2003. Fitness reduction and potential extinction of wild populations of Atlantic salmon, *Salmo salar*, as a result of interactions with escaped farmed salmon. *Proceedings of the Royal Society of London, Series B*, **270**: 2443-2450; Jonsson, B. and Jonsson, B. (2006) Cultured salmon in nature: a review of their ecology and interactions with wild fish. *ICES Journal of Marine Science* **63**: 1162-1181; Verspoor, E., Stradmeyer, L., Neilsen, J. L. (eds.). 2007. *The Atlantic Salmon. Genetics, Conservation and Management*. Blackwells, Oxford.

<sup>57</sup> The term watershed is used here to refer to interconnected waterbodies, which may be defined at a catchment or sub catchment level.

The ICES Code recommends a framework for the introduction of aquatic organisms covering both alien species and improved lines. The Code is conceptually simple and contains the requirements that any person, agency or business planning to use non-indigenous germplasm should follow. The requirements start with the preparation of a proposal that will be reviewed by an independent body. The results of the review will be communicated back to the proponents for approval, revision or rejection. If the proposal to introduce a new species is approved, then the Code calls for fish health management, monitoring and reporting.

### ***5.2.3 Material Transfer Agreements (MTAs)***

If the request for introduction is approved, the transfer should be consistent with relevant international and national laws such as those related to access and benefit-sharing, property rights or biosecurity. The conditions to access and use such genetic material are normally set through a Material Transfer Agreement. The MTA should be certified by the national empowering body of the importing country and communicated to the FAO Database on Introductions of Aquatic Species (DIAS).<sup>58</sup>

Material transfer agreements can be legally binding agreements that are generally drawn up to document and describe conditions for the transfer of tangible biological materials, including material used in research and genetically improved fish, from one entity to another. An example of an MTA is shown in Annex 5.1.

### ***5.2.4 Protocols for transfer***

The following protocols are based on international codes of practice, which may include one or more of the existing protocols of the different countries, and are included to serve as general guidelines. They may be seen as an addition to the individual national requirements, or they may form the basis for elements of national regulations.

#### ***5.2.4.1 Exporting (transferring) country or organization***

Appended to the Material Transfer Agreement should be specific technical information concerning the requested germplasm, particularly:

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<sup>58</sup> FAO Database on Introductions of Aquatic Species (DIAS) <http://www.fao.org/fi/website/FISearch.do?dom=introsp>, FishBase (<http://www.fishbase.org>)

- scientific and local names of the transferred stock;
- salient features of the transferred stock that make it desirable for importation;
- intended use of the transferred stock and the exact location of that use;
- number of individuals transferred,
- number and type (e.g. full sib, half sibs) of families represented in the transfer;
- age or ontological state (e.g. egg, larvae, post-larvae, swim-up fry, fingerling) of transferred individuals;
- disease and/or pathogen exposure history of the stock;
- genotypic and phenotypic sex of the transferred stock (e.g. normal females, normal males, normal mixed sex, genetically mixed sex but phenotypically all male – hormone treated).

The material to be transferred must be accompanied with a veterinary certification of freedom from prescribed parasites, pathogens and any other biota issued by a competent authority. Shipment water, if any, should be clean and free from suspended particulate matter. If possible, the transferred stock should be disinfected prior to shipment.

Most of this information should have been provided in the original proposal requesting importation of the species into the country. It can be duplicated with the MTA to help ensure compliance with the conditions of the agreement.

#### *5.2.4.2 Importing (receiving) country or organization*

A major concern for importing countries is fish health and the prevention of trans-boundary pathogens. Relevant sections of Technical Guidelines<sup>59</sup> on the subject call for a national aquatic animal health strategy and are summarized here. A formalized national aquatic animal health strategy provides countries with a “road map”, based on national needs and priorities, for achieving the desired aquatic animal health status. The components of a national strategy include: pathogens to be considered, disease diagnosis, health certification and quarantine measures, disease zoning, disease surveillance and reporting, contingency planning, import risk analysis, policy frameworks and regional capacity building.

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<sup>59</sup> FAO. 2007. Aquaculture Development. 2. Health management for responsible movement of live aquatic animals. FAO Technical Guidelines for Responsible Fisheries. No. 5. Suppl. 2. Rome, FAO. <ftp://ftp.fao.org/docrep/fao/010/a1108e/a1108e00.pdf>

Consistent with the World Trade Organization (WTO) and the *Agreement on the application of sanitary and phytosanitary measures* (SPS Agreement), all countries reserve the right to take sanitary and phytosanitary measures necessary for the protection of human, animal, or plant life. In determining the appropriate level of protection (ALOP), relevant economic, social and ecological factors have to be taken into account.

Whenever possible, rather than adult brood fish, stocks should be imported as eggs or as other early life history stages. The longer a fish lives, the more likely it is to come into contact with a pathogen. Also, early life history stages carry less sub-clinical infections than adults, they are easier to maintain in quarantine and eggs cannot transmit certain pathogens, e.g. gill parasites.

Prior to importation, qualified personnel in the importing country should consult the World Organization for Animal Health (OIE) which is the World Trade Organization's standard setting body for fish pathogens, existing literature and disease networking services<sup>60</sup> to identify possible areas for concern in regards to fish health. Every effort should be made to obtain fish from accredited hatcheries that practice good fish health management and ensure the quality of the exporting country's veterinary certification. Upon arrival, shipments should be examined for freedom from prescribed pathogens, e.g. those officially listed by OIE, parasites and other unapproved biological material, such as hitchhiking species for which import was not requested. If diseases are identified, the shipment should be destroyed and disposed of in an appropriate manner, unless effective treatment can be guaranteed.

Quarantine should maintain a group of aquatic animals in isolation with no direct or indirect contact with other aquatic animals, in order to undergo observation for a specified length of time and, if appropriate, testing and treatment, including proper treatment of the effluent waters<sup>61</sup>. The level of quarantine should relate to the risk of disease spread. First time importation of alien species, or species collected from the wild or sources of unknown health status, may require more stringent quarantine levels.

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<sup>60</sup> [http://www.oie.int/eng/ea\\_index.htm](http://www.oie.int/eng/ea_index.htm); Permanent Advisory Network for Diseases in Aquaculture (PANDA; <http://www.europanda.net/>), Aquatic Animal Pathogen and Quarantine Information System (AAPQIS, <http://www.aapqis.org/main/main.asp>).

<sup>61</sup> OIE. 2005. *Aquatic animal health code*. 8th Edn. Paris.  
[http://www.oie.int/eng/normes/fcode/A\\_summry.htm](http://www.oie.int/eng/normes/fcode/A_summry.htm)

It should be understood that physical inspection and quarantine are of only limited effectiveness in preventing transfer of pathogens. Any bacteria or virus to which the imported stock is already immune or only displays sub-clinical symptoms, i.e. appears healthy, can be detected through experimentation and immunoassay but will not be eradicated by holding in isolation. Testing and observation in quarantine may involve co-habitation experiments with local species or placing quarantine animals under increased stress to see if disease problems arise.

Nevertheless, quarantine does give authorities the opportunity to observe the stock for a period of time, which might give indications of problems. Quarantine in an appropriate facility should be for a period of at least 28 days but must be determined by the specific pathogens under consideration. Upon arrival in quarantine, introductions should be disinfected in a prophylactic bath and, if feasible, put on an oral course of broad-spectrum antibiotics. All water, packing materials, containers or other associated shipping materials should be sterilized or destroyed.

Quarantine sites must be secure against escapes and discharges of water. Water must be safely disposed of. If the quarantine unit suffers a disease outbreak treatment is sometimes possible. However chemical therapy can cause other problems such as antibiotic resistance and should be used under expert advice. When the outbreak cannot be controlled, diseased stocks should be destroyed and disposed of after sterilization in an approved manner. Water quality at the quarantine unit should be monitored at regular intervals and periodic checks for introducible parasites and diseases carried out. A list of known parasites, diseases and pathogens should be maintained and the exporter advised in case of unexpected occurrence of parasites or pathogens.

Original imports should not be transferred to natural environments. The ICES Code recommends distributing only the F1 generation of imported species following quarantine of the original parents.

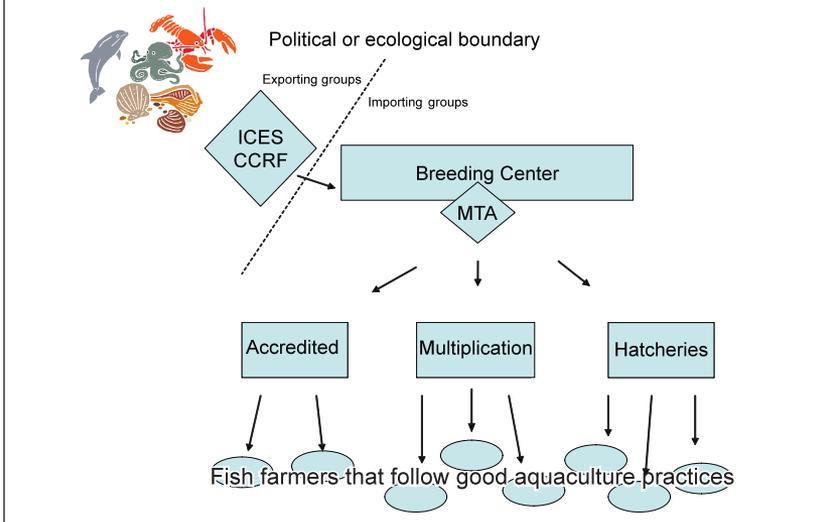
Zoning areas of aquaculture use and conservation (Chapter 9) also apply to fish health management. Countries may establish zones where certain pathogens are known to exist and disease free zones; zones should be based on ecological criteria rather than on political boundaries. Movement of animals between zones where the same pathogens exist or from a disease free zone would not be problematic. Animals may not be moved from a zone having pathogens that are absent in the receiving zone.

### 5.3 Dissemination of an improved strain within a country as part of a rational aquaculture development strategy

Given that original imports should not be transferred to natural environments, a process of multiplication must be carried out prior to the dissemination of seed from genetically improved strains.<sup>62</sup> Improved strains should be disseminated through a system of accredited hatcheries and breeding centers (Figure 5.1). Accreditation of hatcheries that function as multipliers of the improved stocks from the breeding centers should be carried out by an evaluation team of the regional breeding centre. Accredited hatcheries

**Figure 5.1**

Dissemination system for the introduction and use of genetically improved strains in aquaculture. Diamonds represent guidelines and codes of practice that should be followed before dissemination. There should be no movement of germplasm except as noted by the arrows. MTA = Material Transfer Agreement; ICES and CCRF refer to the recommendations and guidance in the International Council for the Exploration of the Sea Code of Practice on Introductions and the FAO Code of Conduct for Responsible Fisheries, respectively.



<sup>62</sup> *Pioneering Fish Genetic Resource Management and Seed Dissemination Programmes for Africa: adapting principles of selective breeding to the improvement of aquaculture in the Volta Basin. Workshop Proceedings, 27-30 March 2007, FAO, Rome.*

must meet technical requirements established by the evaluation team and have an agreement with the breeding center concerning standard operating management and dissemination procedures.

The main objective of developing a hatchery accreditation system is to ensure implementation of guidelines on maintaining genetic quality of fingerlings supplied by the hatcheries to farmers and safeguarding native genetic resources. It is recommended that:

- In order to receive genetically improved seed hatchery operators must apply for accreditation to the breeding center; the application would be reviewed on the basis of a set of criteria that could include the elements listed here as well as other relevant information (e.g. facilities, experience, location, earlier performance).
- Brood stock would be supplied by breeding centers to the accredited hatchery and replaced following a well defined protocol and on a needs basis.
- Hatcheries being considered for accreditation should be well managed and follow best aquaculture practices according to the judgment of qualified technical staff.
- A system of good record keeping of supplied brood stock or fry to the hatchery should be implemented.
- A system to monitor distribution of fingerlings from accredited hatcheries to producers should be implemented in order to monitor the geographical distribution of genetically improved stocks. This would enable assessments of potential economical and environmental impacts of the improved strains being disseminated.
- Hatcheries should implement quality control measures, and their accreditation status should be regularly reviewed.

## 5.4 Discussion

There has been considerable movement of alien species and strains for aquaculture purposes,<sup>63</sup> but very little evaluation of their impacts, either good or bad.<sup>64</sup> Governments are requested to maintain records on the introduction and subsequent distribution of alien species and genetically improved stocks

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<sup>63</sup> FAO Database on Introductions of Aquatic Species (DIAS) <http://www.fao.org/fi/website/FISearch.do?dom=introsp>, FishBase (<http://www.fishbase.org>)

<sup>64</sup> A notable exception is, *An Impact Evaluation of the Development of Genetically Improved Farmed Tilapia and Their Dissemination in Selected Countries* by Asian Development Bank. ADB 2005; available at [www.adb.org/publications](http://www.adb.org/publications).

in their countries and to report the information to FAO. FAO maintains a Database of Introductions of Aquatic Species (DIAS) that also contains information on impacts. The coverage of alien species is increasing and allows for better decisions on introducing alien species; there is no comparable information source on the impacts of genetically improved strains.

Many movements of improved stocks and alien species are poorly controlled, even though there is wide recognition that control is needed due to the risks involved. The Hazard Analysis and Critical Control Point Approach (HACCP)<sup>65</sup> is being promoted by the aquarium trade and fish and wildlife scientists in some areas, primarily to reduce risks of importing countries bringing in hitchhikers and pathogens and to improve public awareness. HACCP is also being promoted by salmon farmers in order to reduce the likelihood of escapes. MTAs present a way of helping improve controls, but to date they have been little used in aquaculture and fisheries transfers.

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<sup>65</sup> See [http://seagrant.umn.edu/downloads/ais-haccp\\_manual.pdf](http://seagrant.umn.edu/downloads/ais-haccp_manual.pdf) for guidance on applying the HACCP principles to aquatic invasive species.

## Annex 5.1

### *Material Transfer Agreement*<sup>1</sup>

The following example of a Material Transfer Agreement is based on one currently used by the WorldFish Center.

To: The request for improved germplasm should be made to a competent authority that has legal and political authority to disseminate the material.

I/we order the following material:

A list of material being requested should be attached here including the detailed description of the material, its intended use and location of use as listed in the text.

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I/we agree

- to abide by the provisions in the Convention on Biological Diversity;
- to preclude further distribution of germplasm to locations at which it could have adverse environmental impact;
- not to claim ownership over the material received, nor to seek intellectual property rights over that germplasm or related information;
- to ensure that any subsequent person or institution to whom I/we make samples of the germplasm available, is bound by the same provision;
- that the responsibility to comply with country's biosafety and import regulations and any of the recipient country's rules governing the release of genetic material, is entirely mine/ours;
- to follow the quarantine protocols suggested by the FAO Technical Guidelines on Health Management for Responsible Movement of Live Aquatic Animals and the WorldFish Center;
- that when germplasm is transferred beyond the boundaries of our country, we will abide by the relevant international codes and guidelines, e.g. the CCRF, ICES, and the OIE.

Date: .....

Name of person or institution requesting the germplasm: .....

Address: .....

Shipping address (if different from the above): .....

Authorized signature: .....

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<sup>1</sup> From the International Network for Genetics in Aquaculture (INGA) [www.worldfishcenter.org](http://www.worldfishcenter.org)

## 6 ECONOMIC CONSIDERATIONS RELEVANT TO GENETIC IMPROVEMENT PROGRAMMES<sup>66</sup>

### 6.1 Evidence about genetic improvement

In terrestrial animal and plant species genetic improvement programmes have made a substantial contribution to productivity increases and to industry viability. By contrast, most aquaculture stocks in current use in developing countries are genetically similar or inferior to wild, undomesticated counterparts.<sup>67 68</sup> There is evidence indicating that genetic improvement programmes implemented in aquatic animal species can have the same positive effect they have had in livestock and crops. The Genetically Improved Farmed Tilapia (GIFT)<sup>69</sup> (*Oreochromis niloticus*) and Jayanti rohu<sup>70</sup> (*Labeo rohita*) are two examples in developing countries; The genetic improvement programmes implemented in these two species were modeled on the successful project with Atlantic salmon (*Salmo salar*) initiated in the 1970s in Norway. These improved strains are very appealing and valuable to farmers due to their greater growth and survival rates.

### 6.2 Limiting factors to the widespread adoption of the technology

Proof about genetic improvement can be easily obtained under controlled, experimental conditions, in which a set of necessary records are systematically kept. However, the ‘visibility’ of genetic gains in aquatic animals under farming conditions is extremely low. Important traits from a production viewpoint, such as growth rate, survival, and freedom from disease, are not only influenced by genetics, but also, and to a large extent, by the environment. This makes it difficult, if not impossible, to precisely ascertain the cause of observed changes in the production system. Furthermore, genetic

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<sup>66</sup> Contributed by Raul W. Ponzoni.

<sup>67</sup> Eknath, A.E. 1991. Simple broodstock management to control indirect selection and inbreeding: Indian carp example. NAGA, The ICLARM Quarterly 738: 13-14.

<sup>68</sup> Brummett, R.E., Angoni, D.E. and Pouomogne V. 2004. On-farm and on-station comparison of wild and domesticated Cameroonian populations of *Oreochromis niloticus*. Aquaculture 242, 157-164.

<sup>69</sup> Gupta, M. and Acosta B. 2004. From drawing board to dining table: The success story of the GIFT project. NAGA, WorldFish Center Quarterly 27, (3&4), 4-14.

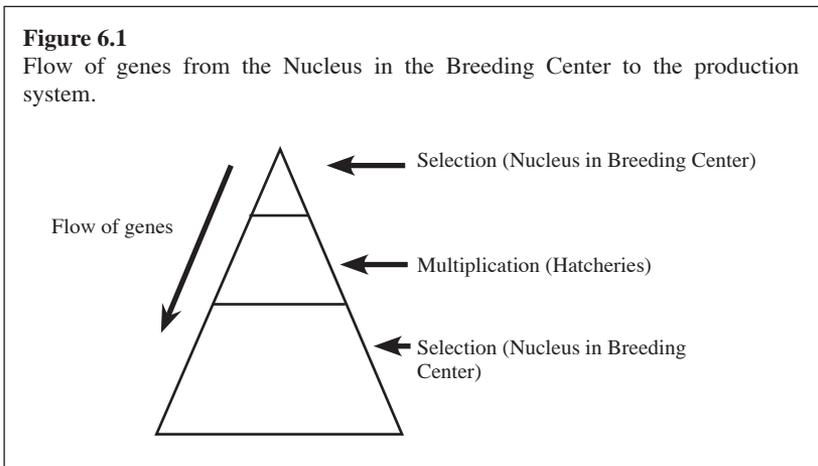
<sup>70</sup> Mahapatra, K., Jana, R.K., Saha, J.N., Gjerde, B. and Sarangi N. 2006. Lessons from the breeding program of Rohu. In: Ponzoni, R.W., Acosta, B., Ponniah, A.G. (eds.), Development of aquatic animal genetic improvement and dissemination programs: Current status and action plans, WorldFish Center Conference Proceedings 73, Penang, Malaysia, pp. 34-40.

improvement programmes require an initial investment, as well as recurrent annual expenditure to run them. In view of these costs, government institutions may remain unconvinced about the wisdom to invest in such programmes unless clear benefits to the nation can be confidently anticipated. In order to generate information that can assist in making logical decisions about genetic improvement, economic considerations at two critical levels have to be made, namely, when defining the programme's breeding objectives, and when assessing the costs and benefits of implementing the programme within a reasonable time horizon. These two levels are, of course, related, but they are best dealt with separately.

### 6.3 Breeding objectives

In animal production, genetic improvement typically takes place in a very small fraction of the population. The genetic improvement achieved in that 'elite' or 'nucleus' of superior animals is multiplied and disseminated to the production systems (Chapter 5). The flow of genes is graphically illustrated in Figure 6.1. The implementation of a genetic improvement programme in a relatively small number of animals can be enough to service a very large population involved in production. The nucleus supplies brood stock to hatcheries (multipliers of genetically improved stock). In turn, the fry produced by hatcheries are grown out in the production sector.

With this industry structure (Figure 6.1; see also Chapter 5) farmers produce virtually all the fish for consumption. Hence, the breeding objective must be



defined according to farmers' interests, considering the nucleus and the dependent hatcheries as sectors servicing farmers. The biological traits included in the breeding objective must be those that influence profit, i.e. income, expense, or both, at the farm level. They are shown for a simple case in Table 6.1.

A profit equation has the form:

$$\text{Profit (P)} = \text{Income} - \text{Expense}$$

This equation can be expressed as a function of the biological traits in Table 1. Scaling it up to a production unit of 1000 fish stocked we may write:

$$P = 1000 [(W) (S/100) (\text{price per unit weight of fish}) - \text{FI} (\text{price per unit weight of feed})] - K$$

where: W is weight at harvest, S is the percent survival to harvest time, FI is the total amount of feed consumed per fish to harvest time, and K represents fixed costs. Fixed costs are those that a producer incurs in no matter what the level of production is, and can be ignored when deriving the economic value for each trait. This equation enables the estimation of the economic value for each trait in the breeding objective. The economic values usually differ between traits because of the unit of measurement, their expression in the production system, and because of their relative economic importance. For instance, survival rate is expressed in all fish stocked, but market weight in only those that survive to market. Also, if the feed price is low (high) relative to fish price, then feed intake will have a lower (greater) economic value than harvest weight.

Assigning economic values to the traits in the breeding objective enables the calculation of genetic gains in economic units. The inclusion of traits associated with expense as well as those associated with income is very

**Table 6.1** Biological traits included in the breeding objective.

Effect on profit	Trait	Logic for inclusion
Income	Harvest weight (W)	Fish are marketed on a weight basis, heavier fish generally fetch a greater price. Fast growing fish will reach a particular weight faster than slow growing fish.
	Survival rate (S)	Greater survival results in a greater number of fish available for consumption or for sale.
Expense	Feed intake (FI)	Feed is a major production cost. Greater growth rate may result in greater feed consumption.

important because if only income traits are included, the economic worth of genetic gain may be overestimated. The economic values for each trait can be evaluated numerically by computing the difference  $P^* - P$ , where  $P$  is the profit at the average value for all traits, and  $P^*$  is the corresponding value after increasing the trait in question by one unit, while leaving the other traits at the average value. Using the equation for  $P$  above, we find that the economic values for  $W$ ,  $S$  and  $FI$  are US\$0.85, US\$3.00 and - US\$0.56, respectively.

#### **6.4 Costs and benefits of a genetic improvement programme**

Whereas there are several ways of manipulating the genetics of aquatic animals (e.g. polyploidy, cross-breeding), selective breeding is the only approach whereby the gains achieved can be multiplied, transmitted to other animals and passed on from generation to generation. This paper focuses exclusively on selective breeding. Annual responses to selection often look negligible when compared with the gains that may be achieved through expansion, improved nutrition and intensification of the production system. However, response to selection measured in one population does not provide a good measure of the potential impact of genetic gains. With an adequate industry structure, the small but cumulative responses to selection achieved in a nucleus undergoing genetic improvement, can be passed over to a multiplier tier of hatcheries and in turn, from hatcheries to farmers (Figure 6.1; Figure 5.1 Chapter 5). This potential for expression of small accumulated changes in thousands or millions of animals is what makes genetic improvement programmes one of the most powerful and cheapest means of increasing the efficiency of aquaculture.

#### **6.5 Factors affecting the economic benefit and the benefit/cost ratio of genetic improvement programmes**

There is an established methodology that is generally used in studies about the economic consequences of implementing a genetic improvement programme.<sup>71</sup> The results of such studies are dependant on the assumptions made about the numerous factors that may affect the outcome. Table 6.2 lists such factors and provides numerical values that cover a range of plausible scenarios. In practice, one can test the robustness of the assumptions made by testing the sensitivity of the results to realistic deviations from such

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<sup>71</sup> Ponzoni R.W., Nguyen, H.N. and Hooi Ling Khaw. 2007. Investment appraisal of genetic improvement programs in Nile tilapia (*Oreochromis niloticus*). *Aquaculture* 269, 187-199.

assumptions. The values shown in Table 6.2 were used in the calculation of the economic benefit (EB) and benefit/cost ratio (BCR) resulting from the genetic improvement programme. When several values are shown for a given parameter, the one in bold was used as a reference to generate “base results” (Table 6.3), other values being used in the sensitivity analysis (see section 6.8).

## 6.6 General usefulness of the results

The conduct of economic appraisals of genetic improvement programmes

**Table 6.2** Parameter values for economic evaluation of selective breeding programme.

Parameter	Value(s) <sup>a</sup>
<b>Economic parameters</b>	
Initial investment in programme	50 000, <b>75 000</b> , 100 000 US\$
Discount rate	<b>0.05</b> , 0.10, 0.15 d (fraction)
Discount factor	Computed from d values $r = 1 / (1+d)$
Annual (recurrent) costs	30 000, <b>60 000</b> , 90 000 US\$
Price of fish (farm gate)	<b>0.001</b> , 0.0015, 0.002 US\$/g
Cost of feed	0.00056 US\$/g
Number of years over which scheme is evaluated	10 years
<b>Biological parameter</b>	
Generation interval in females	1.0 year
Generation interval in males	1.0 year
Heritability estimates	W values = 0.2, <b>0.3</b> , 0.4; S values = 0.05, <b>0.08</b> , 0.12; FI values = 0.16, <b>0.25</b> , 0.3
Cumulative feed intake	400 g
<b>Operational parameters</b>	
Year when first returns are obtained	<b>2</b> , 3, 4 years
Number of fish marketed for slaughter/year <sup>b</sup>	(1) 2.205; (2) <b>6.6248</b> ; (3) 47.32; (4) 338.0 in millions
Harvest weight	300 g
Survival rate	85 %

<sup>a</sup> When several values are presented, the value in bold was used as a reference to generate “base results”, whereas the other values were used in the sensitivity analysis.

<sup>b</sup> The figures correspond to different reproductive technological levels, from a very low one, to higher ones. Level 1 corresponds to poor management and natural spawning in ponds; Level 2 is as Level 1 but with good management; Level 3 uses reproduction in hapas, egg collection from the mouths of females and artificial incubation in the nucleus, and natural spawning with good management in hatcheries; Level 4 assumes that reproduction in hapas (as described for Level 3) is used in both the nucleus and in hatcheries.

**Table 6.3** Discounted cash flow ( $d = 5\%$ ), economic benefit and benefit/cost ratio for the base situation.

Year	Discount factor	Discounted returns	Discounted costs (000's US\$)	Economic benefit (000's US\$)	Benefit/cost ratio
0	1.0	0	0	-75	-
1	0.952	0	57.14	-132.14	0
2	0.907	130.56	111.56	-56.01	0.7
3	0.864	379.23	163.39	140.84	1.6
4	0.823	734.48	212.76	446.73	2.6
5	0.784	1 185.60	259.77	850.83	3.5
6	0.746	1 722.64	304.54	1 343.10	4.5
7	0.711	2 336.40	347.18	1 914.21	5.5
8	0.677	3 018.35	387.80	2 555.56	6.5
9	0.645	3 760.62	426.47	3 259.15	7.5
10	0.614	4 555.90	463.30	4 017.60	8.5

is especially useful from a national perspective, where decision-makers will focus on the calculation of what additional wealth to the nation would emerge from the implementation of such a programme. The findings are also applicable to a vertically integrated firm controlling the nucleus breeding center, the hatcheries and the production sector (Figure 6.1). The results strongly suggest that very favorable returns on investment can be obtained from genetic improvement (Table 6.3). Even for the very conservative values assumed for the base level of factors in Table 2, EB and BCR were extremely favorable, at four million US\$ and 8.5, respectively, after 10 years of programme implementation. The “break-even point”, that is, the moment when profit turns from negative to positive, occurs in year 3.

## 6.7 Positioning the base parameter values in a real life context

The base parameter values were chosen here to represent a very conservative scenario. For instance, when both fish price and reproductive efficiency were set close to the lower limit of the values that can be expected, EB turned from negative to positive by the third year of programme implementation (Table 6.3), and by year 10 the BCR was 8.5. In practice, the fish price is likely to be greater, and using very simple and inexpensive technology the reproductive efficiency of the fish can be greater. Hence, the EB and BCR obtained with the base parameter values should be taken as the minimum that can be expected from a genetic improvement programme such as the one in question.

## 6.8 Sensitivity analysis

The factors that can affect EB and BCR (Table 6.2) may be grouped into three categories: (i) Biological (heritability values, accounting for feed intake), (ii) Economic (initial investment, annual cost, discount rate, price of fish), and (iii) Operational (year when first return occurs, reproductive efficiency).

### 6.8.1 Biological parameters

The effects of two biological factors were studied, namely, the heritability values for the traits in the breeding objective, and the approach taken regarding feed intake. Greater heritabilities resulted in greater genetic gain and consequently in greater EB and BCR. Partly, the heritability value is a property of the trait and the population in question, but it may be improved by reducing the environmental variance by managerial means. Although EB and BCR were only moderately sensitive to rather large variations in the heritabilities, management practices that may lead to reduced environmental variance in the nucleus should be adopted whenever possible. The production of progeny from synchronized spawnings and its grow-out in standard and uniform conditions are examples of such practices.

With regards to feed intake, despite a lack of genetic parameters for this trait in most cultured species, it should be included in the breeding objective because generally feed is a major cost in aquaculture production. The parameter values used for feed intake were based on a number of assumptions, but note that ignoring feed intake involves more radical assumptions, namely, that feed requirements do not increase with greater growth rate, or that the cost of the additional feed is zero; the latter assumption is certainly not correct. With regards to the former, there is experimental evidence indicating that in Atlantic salmon there is a correlated response in feed intake, as well as in feed efficiency, to selection for growth rate.<sup>72</sup> Also, in brown trout (*S. trutta*) there is a correlated response in feed intake, but there is no change in the efficiency of feed utilization.<sup>73</sup> These experimental results, coupled with the importance of feed costs in the production system, provide ample justification for the inclusion of the trait in the breeding objective. Ignoring feed intake in the breeding objective would result in a gross overestimate of the benefit of a

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<sup>72</sup> Thodesen, J. 1999. Selection for improved feed utilization in Atlantic salmon. Doctor Sci. Thesis, Agricultural University of Norway, 108 pp.

<sup>73</sup> Mambrini, M., Labbe, L., Randriamanantsoa, F. and Boujard, T. 2006. Response of growth selected brown trout (*Salmo trutta*) to challenging feeding conditions. *Aquaculture* 252, 429-440.

genetic improvement programme emphasizing growth rate. This result is consistent with what is observed in terrestrial animal species.<sup>74</sup> Although it is unlikely that feed intake will be measured in any breeding programmes in developing countries, the estimation of phenotypic and genetic parameters for this trait by research institutions would be highly desirable to increase our confidence on the parameter values used for genetic evaluations and in predicting responses to selection.<sup>75</sup>

### ***6.8.2 Economic parameters***

EB and BCR were both insensitive to the magnitude of the initial investment, whereas the annual cost of the programme had a greater effect on BCR than on EB. By contrast, discount rate, had a greater effect on EB than on BCR. The discount rate ( $d$ , Table 6.2) is the interest rate used in calculating the present value of expected future benefits and costs. The discount factor ( $1/(1+d)^y$ , Table 6.2) is the factor that transforms expected benefits or costs in any future 'y' year into present value terms. The choice of a discount rate in a study such as this is always open to debate. In the present context the costs and benefits are being assessed from the viewpoint of society as a whole (as distinct from an individual firm or person), and the discounting technique is used to express such costs and benefits in terms of net present value. This net present value can then be compared to that obtained from alternative uses of the limited resources a nation may presently have for investment. In the present case, despite the assumed low reproductive rate, even at a high discount rate of 15 percent EB remained highly positive and BCR was about 75 percent of that for the base situation.

The price of fish had a large effect on both EB and BCR. Although prices are most often beyond planners' and farmers' control, bigger fish often fetch greater prices in the market, so an added (and not accounted for) benefit of the selection programme could be better prices in the future.

### ***6.8.3 Operational efficiency***

The year when first returns occur is likely to be a reflection of how soon the programme gets fully underway, including the distribution of stock to hatcheries. There may be delays in the latter activities despite on-going

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<sup>74</sup> Ponzoni, R.W. 1992. Genetic improvement of hair sheep. FAO Animal Production and Health Paper no. 101, 168 pp. (Rome, Italy).

<sup>75</sup> Doupe, R.G., Lymbery, A.J. 2003. Toward the genetic improvement of feed conversion efficiency in fish. *J. World Aquacult. Soc.* 34, 245-254.

genetic gain in the nucleus. The earlier returns occur, the better, but even with a delay of two years EB and BCR were still highly favorable.

The reproductive efficiency assumed for the base situation (Table 6.2) was considered to be the lowest level at which a genetic improvement programme should be entertained, and one that can be easily improved with readily available and affordable technology. Despite this it resulted in a very favorable EB and a BCR of 8.5 after 10 years (Table 6.3). Reproductive efficiency Level 3 can be achieved with simple and inexpensive technology, and it can be easily targeted in a national genetic improvement programme. In Level 4 with even more improved reproductive efficiency both EB and BCR increased in an extraordinary manner. It may be argued that to achieve a greater reproductive efficiency in hatcheries an additional government investment would be required to transfer technology to hatchery managers. Modeling showed that despite substantial additional investment to train hatchery personnel, EB and BCR were still very favorable and worth the investment.

#### ***6.8.4 Summary of sensitivity analysis***

- Management practices in the nucleus that may reduce environmental variance and thus increase heritabilities are likely to have a moderate effect on profitability.
- The cost of increased feed intake as a correlated response to selection for greater growth rate should be taken into consideration to avoid gross over-estimations of the EB and BCR of the programme.
- Initial investment, annual costs and choice of discount rate are likely to have a relatively small effect on EB and BCR, whereas the effect of the price of fish can be substantial.
- The earlier the first returns are achieved the greater EB and BCR will be. However, the greatest contribution to EB and BCR came from improvements in the reproductive efficiency at the level of both the nucleus and the hatcheries. This last factor, reproductive efficiency, is the one likely to have the greatest impact on EB and BCR.

### **6.9 Chance of success**

The results presented in the earlier sections are of a deterministic nature (use of mathematical equations to predict results) implicitly assuming a total certainty of outcomes. However, genetic improvement by selection is a stochastic process, involving sampling of genes when the parents of each generation are chosen and when those parents produce progeny. A way of assessing the

**Table 6.4** Upper and lower limits (95 % probability) for EB and BCR for the different levels of reproductive efficiency.

<b>Reproductive efficiency<sup>A</sup></b>	<b>Limit for EB and BCR</b>	<b>EB (millions US\$)</b>	<b>BCR</b>
Level 1	Upper	1.17	3.17
	Lower	0.79	2.46
Level 2	Upper	4.60	9.53
	Lower	3.44	7.40
Level 3	Upper	36.11	68.08
	Lower	27.90	52.82
Level 4	Upper	261.25	486.32
	Lower	202.56	377.30

<sup>A</sup> See Table 6.2 for definition of Levels 1 to 4.

probability of success of a genetic improvement programme is by looking at the anticipated variability in response to selection.<sup>76</sup> The coefficient of variation calculated using equations provided by Nicholas (1989) was low enough to inspire confidence in the programme's outcome, and if confidence limits were set for EB and BCR these fell within favorable values even for the lowest level of reproduction studied (Table 6.4). Therefore, the risk of failure due to technical reasons is extremely low. Of course, failure due to natural disasters or to lack of continuity of purpose can occur but it is very difficult to deal with this kind of causes in a systematic manner.

## 6.10 Concluding remarks

Economic considerations in genetic improvement programmes are necessary in order to logically assign relative emphasis to different traits in the breeding objective. In turn, these enable the assessment of the economic impact of the programme on industry as a whole. The methodology used illustrates the multiplicity of factors that can influence the impact of a genetic improvement programme. The factors to which the economic benefit and the benefit/cost ratio are most sensitive can be identified and given greatest attention Both EB and BCR were most sensitive to reproductive efficiency in the nucleus

<sup>76</sup> Nicholas, F.W. 1989. Incorporation of new reproductive technology in genetic improvement programmes. In Hill, W.G. Mackay, T.F.C. (eds), Evolution and animal breeding, CAB International, Wallingford, U.K., pp. 203-209.

and in hatcheries, a factor that determines the number of fish upon which the genetic improvement is expressed. This quantitative finding is consistent with the generalized perception that multiplication and dissemination of improved strains or breeds is of paramount importance in a comprehensive approach to genetic improvement. The model (see footnote 71) can be used to investigate other factors that one may suspect will influence the outcome of a genetic improvement programme (e.g. less frequent transfer of brood stock to hatcheries, expression of only a fraction of the selection response in the nucleus in the production environment due to genotype by environment interaction). It can be used 'in reverse', to examine the wisdom of setting up a genetic improvement programme for hatchery and production sectors of specific sizes.

With conservative reproductive efficiencies (Level 2 in Table 6.1), attractive EB and BCR values of over four million US\$ and 8.5, respectively, can be obtained. Implementing available, proven, and inexpensive reproductive technology (Level 3 in Table 6.1) resulted in EB and BCR increases to over 32 million US\$ and 60, respectively. With easily cultured species (e.g. tilapia), because of its feasibility and impact reproductive efficiency Level 3 should be the initial target in national genetic improvement programmes, with a view to upgrading to Level 4 as skills in hatcheries are enhanced.

From a national viewpoint, investing in genetic improvement programmes in cultured aquatic animals is a wise decision. Additionally, the availability to producers of a "high performing" strain can act as a stimulus to the adoption of better practices in other areas (management, nutrition, animal health, marketing).

## 7 RISK ASSESSMENT AND MONITORING IN GENETIC IMPROVEMENT PROGRAMMES<sup>77</sup>

### 7.1 Introduction

Genetic improvement programmes (Chapters 4, 5 and 6) raise the need to assess and manage ecological risks imposed by intentional introductions and unintended escapes of improved organisms into aquatic ecosystems. Ecological risk assessment and management is fundamentally a social process guided by scientific information and analysis. The importance of human values is clear in the definitions of risk assessment terms:

- **Risk** refers to the likelihood of harm occurring from a specified hazard or set of hazards.
- **Harm** refers to undesirable consequences to humans and the things that they value.
- **Hazard** refers to an event that has the potential to produce harm.

Risk assessment processes in natural resource arenas, therefore, often incorporate stakeholder<sup>78</sup> deliberations with scientific analysis. International expert consultations co-led by the FAO have identified some important elements of ecological risk assessments for genetically improved fish<sup>79</sup> and an international team has produced the first global synthesis of current approaches and methodologies.<sup>80</sup>

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<sup>77</sup> Contributed by Anne R. Kapuscinski.

<sup>78</sup> Anyone who has an interest in an issue, or anyone who shares the burden of the risks resulting from a particular decision. An individual or representative of a group affected by or affecting the issues in question.

<sup>79</sup> Gupta, M.V.; Bartley, D.M.; Acosta, B.O. (eds). 2004. *Use of genetically improved and alien species for aquaculture and conservation of aquatic biodiversity in Africa*. WorldFish Center Contribution No. 1707. Penang, Malaysia. 107 pp. Nairobi Declaration in Gupta *et al.*, 2004.

WorldFish Center. 2003. Dhaka Declaration on Ecological Risk Assessment of Genetically Improved Fish. WorldFish Center Contribution No. 1704, Penang, Malaysia.

Pullin, R.S.V.; Bartley, D.M.; Kooiman, J. (eds). 1999. *Towards policies for conservation and sustainable use of aquatic genetic resources*. ICLARM Conf. Proc. 59, 277 pp.

<sup>80</sup> This chapter represents and draws extensively from the work of 44 natural and social scientists and policy specialists from 19 countries, begun at a workshop at the WorldFish Center in 2005 and published in a refereed book. They concluded that their synthesis of risk assessment and management methodologies applies broadly to different kinds of genetically improved lines in aquaculture. Kapuscinski, A. R.; Hayes, K.R; Li, S; Dana, G. (eds). 2007. *Environmental risk assessment of genetically modified organisms: Volume 3, methodologies for transgenic fish*. CAB International, Wallingford, UK, 304 pp.

These guidelines address predictive risk assessment, in order to predict the likelihood and consequences of potentially harmful events before and during dissemination of genetically improved fish. The focus is on possible ecological harm to wild populations of aquatic species or the ecosystems that support these species; ecological harm can involve undesirable changes at the genetic, population, community or ecosystem level. The guidelines also address risk management, including monitoring, as part of dissemination programmes.

## **7.2 The Code of Conduct**

Genetic improvement programmes should not undermine the goals of conserving genetic diversity in wild aquatic species and protecting the integrity of aquatic communities and ecosystems, as stated in Articles 6.2, 7.2.2., 9.1.2, 9.31 and 9.3.5 of the Code of Conduct. Stakeholder participation in the risk assessment process is supported by Articles 6.13 and 6.16. Article 9.1.2 gives a clear basis for incorporating ecological risk assessment and management into genetic improvement programmes:

States should promote responsible development and management of aquaculture, including an advance evaluation of the effects of aquaculture development on genetic diversity and ecosystem integrity, based on best available scientific information.

## **7.3 Principles**

***7.3.1 Frameworks for ecological risk assessment and management differ across nations but all effective frameworks contain similar systematic steps that build upon each other.<sup>81</sup>***

***7.3.2 The entire ecological risk assessment and management process should integrate interdisciplinary scientific analysis with multi-stakeholder deliberation.***

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<sup>81</sup> Hayes, K.R.; Kapuscinski, A.R.; Dana, G.; Li, S.; Devlin, R.H. 2007. Introduction to environmental risk assessment for transgenic fish. Pages 1-28 in Kapuscinski *et al.* (eds) (see footnote 80).  
Nelson, K.C.; Basiao, Z.; Cooper, A.M.; Dey, M.; Lorenzo Hernandez, M.; Kunawasen, S.; Li, S.; Fonticciella, D.; Ratner, B.D.; Toledo, M.I.; Leelapatra, W. 2007. Problem formulation and options assessment: science-guided deliberation in risk assessment of transgenic fish. Pages 29-60 in Kapuscinski *et al.* (eds) (see footnote 80).

Credible frameworks for risk assessment and management have certain steps in common (Table 7.1). Responsible agencies should identify who will conduct the various steps in their risk assessment and management framework, identify required areas of expertise, identify relevant stakeholders and decide on how to involve experts and stakeholders in the process.<sup>82</sup> In linking scientific analysis with multi-stakeholder deliberation, each political jurisdiction will need to determine the level of stakeholder participation that fits with its society and available resources. Transparent and equitable deliberation among relevant stakeholders can enhance legitimacy and public trust of the risk assessment conclusions and risk management recommendations and improve the quality of the assessment because it:

- allows all concerns to be recognized;
- incorporates stakeholders' important knowledge about the system, such as information about wild fish in the area, which may be unknown to the technically oriented risk analysts;
- incorporates perspectives of stakeholders at key points in the process; and
- assures that risk assessment conclusions and risk management approaches are meaningful to the stakeholders.

***7.3.3 Each ecological risk assessment should be organized around a hazard chain of events that starts with potential entry of the genetically altered organisms into the ecosystem and defines the subsequent events that pose potential harm.***

The need to assess genetic risks and other ecological risks stems from the changes in genetic makeup and traits of the genetically altered organism. Numerous steps in a risk assessment will require empirical data on these changes compared to the population(s) currently farmed in the geographical area and compared to any wild relatives<sup>83</sup> in the aquatic ecosystem, and how those changes might or might not lead to ecological harm. Figure 7.1 presents a generalized example of a chain of events that would have to occur to end up with ecological harm.

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<sup>82</sup> For further information on incorporating multi-stakeholder deliberations: Hayes *et al.* 2007 and Nelson *et al.*, 2007 (see footnote 81); and Nelson, K.C.; Banker, M.J. 2007. *Problem formulation and options assessment handbook: A guide to the PFOA process and how to integrate it into environmental risk assessment (ERA) of genetically modified organisms (GMOs)*. GMO-ERA Project. Available at: [www.gmoera.umn.edu](http://www.gmoera.umn.edu).

<sup>83</sup> Any species in the ecosystem with which the genetically altered fish can interbreed, including the same species as the genetically altered fish or a closely-related species.

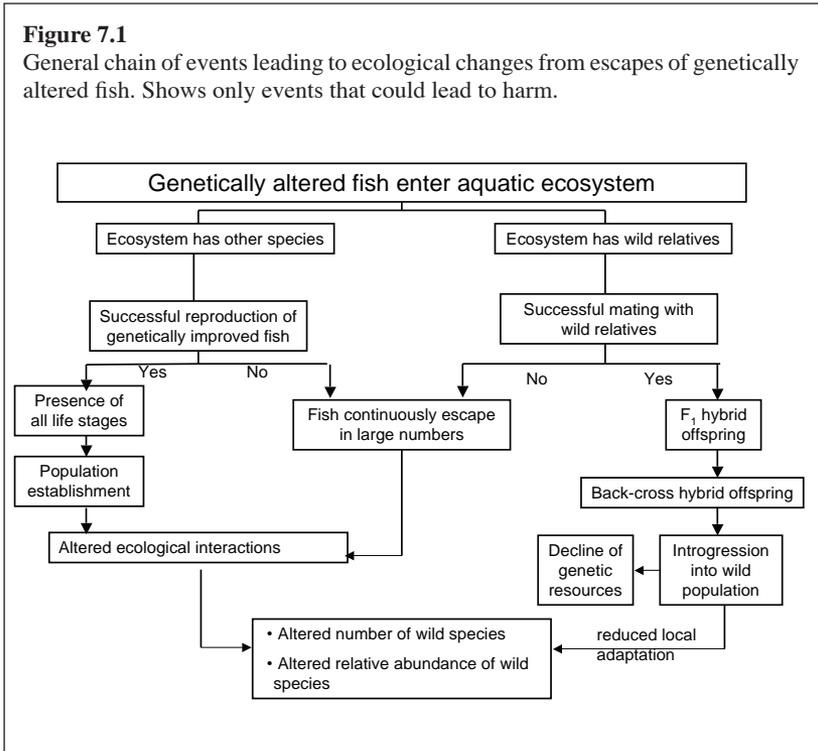
**Table 7.1** Steps found in most risk assessment and management frameworks. Stakeholder participation should be integrated with technical analysis throughout, particularly to address questions in italics.\*

Multi stakeholder deliberation integrated at key points in each step	STEP	KEY QUESTIONS ADDRESSED AT THIS STEP
	<b>Risk Assessment</b>	Identify and prioritize hazards
<b>Risk estimation</b>	Estimate exposure to each prioritized hazard, and likelihood of harm resulting from hazard exposure.	What is the hazard exposure and how likely is the hazard?  What would be the harms of realization of the hazard and how severe are they?
Estimation is quantitative (when possible), semi-quantitative, or qualitative.		What are conclusions of the risk assessment (matrix of estimated likelihood of harm plotted against severity of harm)?  How certain is the knowledge used to identify hazards, estimate likelihood, and predict harms? Which uncertainties can be eliminated? Which uncertainties need to be treated throughout the assessment?
Identify and analyze uncertainties		
<b>Risk Management</b>	Risk reduction planning	What can be done to reduce risk to acceptable levels, either by reducing the likelihood or mitigating the consequences? Are the risk reduction measures acceptable?
Implementation of plan		
Monitoring		Are the monitoring activities acceptable? How effective are the implemented measures for risk reduction? Are they as good, better or worse than planned for?
Remedial action		What remedial (corrective) action will be pursued if findings are unacceptable? Did the action adequately resolve the concern(s)?

\* Hayes *et al.*, 2007 and Nelson *et al.*, 2007 (see footnote 81); Nelson and Banker, 2007 (see footnote 82).

**Figure 7.1**

General chain of events leading to ecological changes from escapes of genetically altered fish. Shows only events that could lead to harm.



**7.3.4 Early in the risk assessment process, relevant experts and stakeholders should deliberate to describe the event chain of concern, identify and prioritize the hazards and harms along the chain, and agree on acceptable levels of risk.**

These outcomes from deliberations among relevant experts and potentially affected interested stakeholders give decision-makers a socially trusted basis for allocating limited resources on assessing the higher priority hazards and harms. The rest of the risk assessment and management process thus focuses on the selected priority hazards (Table 7.1).<sup>84</sup>

<sup>84</sup> Further guidance on prioritizing hazards is in Hayes *et al.*, 2007 (see footnote 81).

### 7.3.5 Focus the risk assessment and management on measurable end points for the prioritized hazards.

It is essential to carefully choose measurable end points for the ecological changes that stakeholders and analysts have agreed are undesirable.<sup>85</sup> Risk analysts can then focus on estimating the likelihood and severity of harm for each endpoint (Figure 7.2). Risk assessment end points (what the risk assessment is trying to protect) should be identified for each prioritized hazard along the event chain (Figure 7.1). When it is difficult to assess an end point of main interest, risk analysts should identify and assess measurement end points (what they can actually measure) that are good scientific indicators of whether a specific ecological harm will or will not occur. For example, if genetically altered fish prey on a wild species which stakeholders agree to protect at a specific abundance level (assessment endpoint), it may be easier to assess effects on survival of wild adults (measurement endpoint) than to predict changes in overall abundance of the wild species.

**Figure 7.2**

Schematic of a qualitative risk assessment matrix for estimates of likelihood (vertical axis) and severity (horizontal axis) of harm. Quantitative risk assessments are preferable to qualitative or semi-quantitative assessments but require more data.

Likelihood of harm	Frequent				Greatest risk
	Almost never	Lowest risk			
		Very low			Very high
		Severity of harm			

<sup>85</sup> End points explicitly express the valued elements of the ecosystem that the interested parties are trying to protect by performing the ecological risk assessment (Hayes *et al.*, 2007, see footnote 81).

The ability to provide honest and accurate predictions of risk diminishes as the length of the hazard event chain increases due to increasingly complex and cascading interactions between the genetically altered organisms and wild species and their habitats. Thus, it is wise to establish a careful balance between reality, complexity and stakeholder concerns by choosing assessment end points that are clearly relevant to these concerns, but occur earlier (rather than later) in event chains. An interdisciplinary team of experts should define endpoints (preferably through deliberations with multiple stakeholders) and identify appropriate assessment methods and existing data. Relevant expertise will vary case-by-case and would ideally include aquatic biologists and ecologists and persons trained in risk assessment methods.

### ***7.3.6 Case-by-case risk assessment and management***

Any culture programme can change the genetic makeup and traits of the cultured organisms (Chapter 3 & 4). No method of genetic improvement inherently poses greater or lesser environmental risks than other methods. Instead, risks need to be assessed case-by-case, based on characteristics of the aquaculture production system (especially its patterns and frequency of escapes into nature), the genetically altered organisms and the potentially affected ecosystems.<sup>86</sup>

## **7.4 Assessing genetic effects<sup>87</sup>**

Gene flow from genetically altered individuals to wild relatives is a major process through which genetically altered fish may affect wild fish populations. The main concerns are whether gene flow results in introgression (incorporation) of genes from the improved organisms into wild gene pools, and whether this leads to harmful genetic and ecological consequences (Figure 7.1). Risk assessors should assess end points in a chain of events that must occur to end up with introgression. They can do this by partitioning the assessment into two major endpoints, entry and introgression; and further partitioning these into sub-component events that should be easier to assess than treating entry or introgression as a single variable.<sup>88</sup>

<sup>86</sup> There is broad agreement on the need for case-by-case ecological risk assessment; e.g. the Cartagena Protocol on Biosafety, Article 15 and Annex III. See also Bellagio Statement in Pullin, R.S.V.; Bartley, D.M.; Kooiman, J. (eds). 1999. *Towards policies for conservation and sustainable use of aquatic genetic resources*. ICLARM Conf. Proc. 59, 277 pp.

<sup>87</sup> Kapuscinski, A.R.; Hard, J.J.; Paulson, K.; Neira, R.; Ponniah, A.; Kamonrat, W; Mwanja, W; Fleming, I.A.; Gallardo, J.; Devlin, R. H.; Trisak, J. 2007. Approaches to assessing gene flow. Pages 112-150 in Kapuscinski *et al.* (eds) (see footnote 80).

<sup>88</sup> Extensive guidance for assessing sub-components is in Kapuscinski *et al.*, 2007 (see footnote 87).

Predicting the likelihood and genetic effects of these events requires data on how the genetic alteration affects the fitness<sup>89</sup> of the farmed fish, and on how that fitness might change if fish escape into the environment and interbreed with wild relatives in the environment. Also required are specific baseline data about the wild relatives, such as population genetic structure and spatial distribution of breeding adults. It can be a daunting task to collect case-specific empirical data for assessing all sub-components of gene flow. To reduce data needs, risk analysts can pursue a step-by-step strategy of assuming that a specific event leading to entry or introgression will occur (instead of obtaining data to estimate its likelihood) and then proceed to estimating the next event in the chain.<sup>90</sup>

Introgression of altered genotypes into wild populations could result in several declines in genetic resources (Figure 7.1): altered frequencies of native alleles, loss of genetic distinctiveness, or loss of genetic variation in the affected wild population. Introgression could lead to outbreeding depression due to disruption of co-adapted gene complexes of the wild population. These genetic changes can reduce the fitness of wild populations and reduce their ability to adapt to environmental change such as climate change or habitat transformation (e.g. from dams or other construction). Such risks are of particular concern for wild populations already in decline or in a species' center of origin.

## 7.5 Assessing ecological effects<sup>91</sup>

Genetically altered organisms may have ecological effects beyond their possible effects on the genetics of wild populations (Figure 7.1). Ecological effects are even possible when there is no interbreeding of farmed fish with wild populations. Adding a new element to an ecosystem can trigger the ecosystem to shift from an initial state to a new state. The purpose of assessing ecological risks is to predict whether a new state might occur that involves socially undesired changes for instance, species extinctions, altered population abundance, and or altered ecosystem functions.

<sup>89</sup> The degree to which an organism succeeds at passing on its genes to future generations. Fitness is determined by the joint effect of key traits spanning the entire life cycle of the organism, such as juvenile and adult viability, fecundity, fertility, mating success, and age at sexual maturity. Muir, W.M.; Howard, R.D. 2001. Fitness components and ecological risk of transgenic release: a model using Japanese medaka (*Oryzias latipes*). *American Naturalist* 158:1-16.

<sup>90</sup> Guidance for this strategy appears in Kapuscinski *et al.*, 2007 (see footnote 87).

<sup>91</sup> Devlin, R.H.; Sundström, L.F.; Johnsson, J.I.; I.A Fleming, I.A.; Hayes, K.R.; Ojwang, W.O.; Bambaradeniya, C.; Zakaraia-Ismail, M. 2007. Assessing ecological effects of transgenic fish prior to entry into nature. Pages 151-187 in Kapuscinski *et al.*, (eds) (see footnote 80).

Predictive assessment of ecological risks should involve four phases that build upon each other.<sup>92</sup>

- (1) characterize specific biotic and abiotic properties of the receiving ecosystem(s) that the genetically altered fish might affect;
- (2) measure intended and unintended changes in traits of the genetically altered fish, focusing on changes that could alter their interactions with the ecosystem;
- (3) determine anticipated interactions between the genetically altered fish and the ecosystem, such as interference competition with another fish species or grazing of aquatic vegetation; and
- (4) estimate the scale and likelihood of ecological effects resulting from each interaction of the genetically altered fish with the ecosystem.

In each phase, assessors should integrate information from several sources including experts and appropriate stakeholders; baseline data about potential receiving ecosystems (e.g. from field surveys); and empirical data from well-designed experiments that incorporate semi-natural conditions. Risk assessors should determine appropriate confinement of genetically altered fish in risk assessment experiments,<sup>93</sup> taking into consideration available resources and current unknowns about these fish. Even when applying this systematic four-phase approach, predictive risk assessment of ecological effects will be a complex task involving significant sources of uncertainty. Genetically altered fish may behave differently in risk assessment experiments than in nature, especially due to genotype-environment interactions, reducing the value of applying the results to natural environments. Studies to obtain case-specific data on ecological consequences should simulate a range of ecological conditions representative of the potentially affected aquatic ecosystem.

## 7.6 Uncertainty analysis<sup>94</sup>

All risk assessments are subject to uncertainty. The reliability of an ecological risk assessment depends on identifying and treating the various sources of uncertainty. To ‘treat’ an uncertainty means to analyse, eliminate (resolve) or carry it through the chains of calculations and judgments of the entire

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<sup>92</sup> Extensive guidance for carrying out each phase appears in Devlin *et al.*, 2007 (see footnote 91).

<sup>93</sup> Guidance on semi-natural experiments and confined experiments appears in Kapuscinski *et al.*, 2007, chapters 5, 6 and 8 (see footnote 80).

<sup>94</sup> Hayes, K.R.; Regan, H.M.; Burgman, M.A. 2007. Introduction to the concepts and methods of uncertainty analysis. Pages 188-208 in Kapuscinski *et al.*, (eds) (see footnote 80).

risk assessment. Systematic identification and treatment of uncertainties can help inform when to apply a precautionary approach (Chapter 11). Different types of uncertainty arise from different mechanisms and risk analysts have developed appropriate mathematical and qualitative methods to identify, treat and communicate each type.<sup>95</sup> It is critically important to build capacity and practical experience in applying these methods to risk assessment of genetically altered fish. Parties responsible for carrying out ecological risk assessments should receive training to:

- identify uncertainties through appropriate stakeholder-expert deliberation methods;
- treat the uncertainties using appropriate methods or recruit properly trained experts to do this;
- understand the results from treating each identified uncertainty; and
- represent and communicate the uncertainty treatments in a reliable and transparent manner.

## 7.7 Ecological risk management

Ecological risk management aims to reduce identified risks to acceptable levels.<sup>96</sup> It can include confinement measures and monitoring programs. When a risk assessment identifies likely but manageable risks, a risk management plan should be developed and implemented as an integral part of dissemination of genetically improved fish. Risk management plans should be based on conclusions from a risk assessment so that they focus on the prioritized risks and are backed by the shared understanding among those who participated in the assessment.

### 7.7.1 Confinement of genetically altered organisms<sup>97</sup>

No confinement method is 100% effective, so risk managers should consider the use of multiple and reinforcing confinement measures and best management practices<sup>98</sup> for confinement. Multiple confinement methods may be needed

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<sup>95</sup> A summary of methods to treat sources of uncertainty is in Hayes *et al.*, 2007 (see footnote 94).

<sup>96</sup> Using multi-stakeholder deliberations to agree upon acceptable levels of reduced risk will increase social acceptance of the decision.

<sup>97</sup> Mair, G.C.; Nam, Y.K.; Solar., I.I. 2007. Risk management: Reducing risk through confinement of transgenic fish. Pages 209-238 in Kapuscinski *et al.*, (eds) (see footnote 80).

<sup>98</sup> Best management practices will vary depending on the aquaculture system and may be very difficult to implement in resource-poor contexts. General guidance on best management practices is in Mair *et al.*, 2007 (see footnote 97).

to reduce the number of escapees from an aquaculture system to acceptable levels. Confinement measures can focus on preventing escapes or reducing effects if escapes occur. Physical barriers e.g., lethal water temperatures or pH; mechanical barriers e.g., screens; and geographical barriers e.g., raising a marine species in an inland closed seawater system (Chapter 9) can be used to prevent escapes. Biological barriers, such as induced triploidy which makes adults of some fish species functionally sterile, can be used to reduce gene flow (thus reduce genetic risks) and population establishment (thus reduce ecological risks). But sterilization does not eliminate all environmental risks. Escaped, sterile fish might still compete with wild fish for limited resources or engage in courtship and spawning behavior, disrupting breeding in wild populations.<sup>99</sup>

### ***7.7.2 Monitoring for presence and ecological effects of genetically altered organisms.***<sup>100</sup>

The best way to detect escapes and early signs of undesired ecological changes is through a well-designed monitoring program that integrates typical fisheries field sampling methods with statistical techniques and uses DNA-based genetic markers to detect genetically altered individuals. Monitoring should be designed to detect one or more end points at various ecological levels:

1. presence of genetically altered individuals in the ecosystem;
2. presence of first-generation hybrid offspring (from successful reproduction between escapees and wild relatives);
3. presence of back-cross hybrid offspring (from successful reproduction between first-generation hybrids and wild relatives);
4. presence of genetically altered individuals at all life stages;
5. population change of both genetically altered and wild individuals; and
6. changes in the number of local aquatic species and their relative abundance.

End points 1-5 may occur over one to several generations after genetically altered fish enter an ecosystem, allowing relatively early detection of ecological effects. It is easier and faster to detect these end points than to

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<sup>99</sup> Agricultural Biotechnology Research Advisory Committee (ABRAC). 1995. *Performance Standards for Safely Conducting Research with Genetically Modified Fish and Shellfish*. Parts I and II, USDA, Office of Agricultural Biotechnology, Washington D.C. Available at: [www.isb.vt.edu/perfstands/psmain.cfm](http://www.isb.vt.edu/perfstands/psmain.cfm)

<sup>100</sup> Senanan, W.; Hard, J.J.; Alcivar-Warren, A.; Trisak, J.; Zakaria-Ismail, M.; Lorenzo Hernandez, M. 2007. Risk management: post-approval monitoring and remediation. Pages 239-271 in Kapuscinski, A.R. *et al.* (eds) (see footnote 80).

monitor for community-level changes in species composition (end point 6). This last end point may take many generations to manifest, is harder to detect, and could also result from other hazards (e.g. habitat damage), making it difficult to distinguish effects due to the genetically altered organisms. For example, relatively early detection of genetically altered fish at all life stages in a monitored area (end point 4) would indicate that these individuals are reproducing well enough to interact extensively with other species. Longer-duration and more complex monitoring would be needed to determine whether interactions between these genetically altered fish and other species lead to undesired changes in fish community composition (end point 6).

Early monitoring can allow remedial responses (or contingency plans in Chapter 11) at the earliest point possible. Remedial responses may include improving confinement measures, removing genetically altered fish from the wild (rarely feasible and likely quite costly) and restricting further use of the genetically altered fish in aquaculture. Decision makers should realize that it is extremely difficult and costly to remedy adverse ecological effects once they have become widespread. Monitoring can also confirm a risk assessment's conclusion of ecological safety. A monitoring programme should have a high probability to detect changes that actually occur by using *inter alia* appropriate sampling designs, scientific tools and data analyses.<sup>101</sup>

## 7.8 Constraints and opportunities

Ecological risk assessment and management of genetically altered fish are complex and demand considerable resources. Methodologies are evolving and practical experience with them is limited. The need to build human and institutional capacity is widespread. Major needs are to.<sup>102</sup>

- fill key gaps in baseline ecological and genetic data and improve access to existing databases;
- further develop broadly useable methods for ecological risk assessment and management of genetically altered fish;
- develop in-depth risk assessment training programs for persons needed to run risk assessment processes (managers, scientists, facilitators), as well as to help policy-makers understand how outcomes can inform their decision-making;

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<sup>101</sup> Extensive guidance on these aspects of monitoring is in Senanan *et al.*, 2007 (see footnote 100).

<sup>102</sup> Nairobi Declaration, Dhaka Declaration (see footnote 2); Kapuscinski *et al.* (eds), 2007 (see footnote 80).

- strengthen international collaborations for conducting risk assessment studies under semi-natural and confined conditions;
- strengthen institutional frameworks needed to govern risk decision-making in this area; and
- promote networks among relevant institutions, as well as international cooperative programs to address the above needs.

Efforts to meet these needs will also assist in the conservation of aquatic biodiversity and responsible development of aquaculture. Better baseline data on key components of natural fish communities (e.g. on genetic diversity of wild populations and on factors affecting species composition) can help prioritize efforts in aquatic biodiversity conservation, inform the design of aquaculture zones and conservation zones (Chapter 9), and inform strategies for climate change adaptation in the fisheries sector. Other concerns about ecological impacts of aquaculture (e.g. raising alien species or effluent discharges) and other development activities (e.g. dam construction) require systematic risk assessment frameworks and some similar methodologies. Thus, broadly useable methods and training programmes will improve ecological risk assessment in the aquatic sector in general.

## **7.9 Conclusion**

Ecological risk assessment of genetically improved fish before their dissemination, and ecological monitoring after dissemination, is necessary to achieve broad benefits without undermining the conservation of aquatic biodiversity. Systematic risk assessment approaches allow policy-makers to focus limited resources for risk assessment on the highest priority issues. Appropriate scientific techniques should be incorporated with multi-stakeholder deliberations. This makes it possible to reach agreement on the prioritized hazards, utilize the most relevant existing knowledge, focus tests and data collection on filling the most important information gaps, apply uncertainty analysis to improve the quality of the conclusions, and improve understanding of the issues and social trust in the risk assessment process and conclusions. Well-designed monitoring is essential for detecting early signs of undesired effects of genetically altered fish in natural ecosystems. Effective monitoring, however, is complex and requires considerable technical expertise and a long-term commitment.

Risk assessment and management is a complicated endeavor and has not been used extensively in aquaculture. As aquaculture expands and uses more genetically improved organisms, there is an urgent need to refine and apply risk

analysis processes involving scientists, multi-stakeholders and government regulatory agencies. Pro-active risk assessment and management can help steer aquaculture of genetically improved organisms towards practices that protect nature while supporting successful fish farming.

## 8 CULTURE-BASED FISHERIES<sup>103</sup>

### 8.1 General principles

For the purpose of these guidelines, culture-based fisheries (CBF) mean capture fisheries that are maintained by stocking with material raised within aquaculture installations. The “material” is usually early life-history stages, but may also include juveniles or adults. There are three broad categories of CBF:

1. Those where the stocked material is meant to breed with each other and with the local species, thus increasing or re-establishing the local stocks;
2. Those where the stocked material is meant to breed with each other, but not with local species, thus creating a new fishery stock;
3. Those where the stocked material is not meant to breed at all.

Terminology suggested by an international group working on coastal fisheries suggests use of

- restocking, the release of cultured juveniles into wild population(s) to restore severely depleted spawning biomass to a level where it can once again provide regular, substantial yields;
- stock enhancement, the release of cultured juveniles into wild population(s) to augment the natural supply of juveniles and optimize harvests by overcoming recruitment limitation; and
- sea ranching, the release of cultured juveniles into unenclosed marine and estuarine environments for harvest at a larger size in “put, grow and take” operations.

In order to manage genetic resources effectively in CBF, it is essential to understand which of the above objectives are being sought. It is recognized that these categories are not discrete. For example fish in category 3 may breed. This is not the measure of CBF success, but must be factored into risk analysis. Success of culture-based fisheries will depend on the social, economic, and ecological contexts in which they are applied. The use of hatcheries to support fisheries is a fishery management tactic that must be integrated into an overall management plan for the fishery or water body. Simply releasing organisms into a water body with no provisions for resource management, no control of fishers and fishing practices, and no protection of habitat will not succeed. The guidelines given here focus on genetic resources management in CBF.

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<sup>103</sup> Contributed by Devin M. Bartley.

## **8.2 Genetic resource management plan for culture-based fisheries**

The use of hatchery-raised organisms in fishery management has often not met the desired objectives of increasing fishery production. This is partially because aquaculturists produce young fish that survive well in the hatchery, but then releases them to survive in completely different wild environments (Chapter 3). A domesticated fish that is adapted to regular feedings on formulated diets will not survive well in most natural habitats. Therefore, a genetic resource management plan for CBF must be very different from the breed improvement plans outlined in chapters 4-6. General management plans are outlined below for the three main categories of CBF.

### ***8.2.1 Culture-based fisheries where the stocked material is meant to breed with the local species***

When the CBF objective is to restock or increase natural reproduction of natural populations of local species, genetic resources management should strive to recreate the natural level of genetic diversity in the stocked material. The hatchery environment should be as natural as possible so that no artificial selection pressures are introduced. This requires choosing the correct strain to stock, as well as modifying hatchery and grow-out techniques to minimize artificial or inadvertent selection.

#### ***8.2.1.1 Choosing the correct stock***

The material to be stocked should match the genetic diversity of the natural populations. This is best achieved by using wild-caught broodstock. If wild broodstock are in short supply, as in the case of many endangered or locally extinct natural populations, genetic stock identification should be used to identify a very similar stock. Where genetic data are not available for stock identification, surrogate information can be used, e.g. choose stocks from same aquatic habitat (water body or specific watercourse, such as a tributary) with similar life history, growth, color, shape and behaviour characters. Transfers of stocks among different watersheds or ecoregions is to be avoided. Broodstock management (Chapter 3) to optimize effective population size and reduce genetic drift should be followed as soon as brood fish are ready to be mated.

For long-term programmes it is desirable to develop a rotational breeding plan where broodstock are used to produce material for stocking, they are then released back into the wild, and then new broodstock are brought into

the hatchery. The timing of this rotation will depend on the success of the programme and the availability of natural broodstock. For species that spawn only once (e.g. Pacific salmon) or where killing broodfish is necessary for achieving fertilization (e.g. some sturgeons) this rotation will not apply.

#### *8.2.1.2 Choosing the correct hatchery procedures*

Fish will adapt in the short term to hatchery management practices and, in the longer-term, hatchery conditions will exert selective pressures on them (Chapter 3). Hatchery procedures should be designed so as to minimize these influences (i.e. to reduce domestication selection) when the stocked material is meant to survive and/or to breed in the wild. Guidelines on specific breeding protocols to minimize inbreeding and loss of genetic diversity are discussed in Chapter 3.

Typical modifications to hatcheries to decrease artificial selection include:

- Provision of live food, from the wild if possible, rather than formulated feed;
- Provision of more natural habitat with gravel, plants, and shelters, rather than sterile tanks and raceways;
- Provision of limited amount of predators to teach predator avoidance;
- Natural light/dark cycles;
- Release of younger fish that have not adapted to hatchery conditions. However, this should be assessed as older fish may survive better;
- Spawn fish over the entire spawning season (i.e. do not simply collect lots of spawn when convenient in order to meet production goals);
- Do not transfer fish among hatcheries that are located in different watersheds or tributaries in order to meet production goals.

#### ***8.2.2 Culture-based fisheries where the stocked material is meant to breed with each other, but not with local species***

This can arise when the fish stocked for CBF have different reproductive strategies from local species, because they are different species or, if the same as the local species, have different migration patterns or other behaviour, such as mate preferences. The most common example is the regular stocking for CBF of alien species or of specific strains of fish, such as salmon. If fertile fish are being released to breed, self-sustaining populations would be expected to develop thus negating the need for continued stocking.

The release of viable alien fish that are capable of reproducing is considered the most risk-prone type of stock enhancement (Chapter 7). Management of these stocks requires in-depth knowledge of the genetics and natural history of a species or stock. Even then, the natural history characters and behaviour of a strain may change when it encounters a new environment. Guidelines have been created to advise on responsible procedures on the use of alien species (Chapter 5).<sup>104</sup> Assuming that these guidelines have been followed and that CBF based on alien fish have been determined to be an acceptable management option, a stock should be chosen that has appropriate behavioural characteristics (e.g., timing and location of migration), and genetic resources management measures should be planned and implemented to optimize  $N_e$  and avoid domestication selection (see Chapter 3).

### ***8.2.3 Culture-based fisheries where the stocked material is not meant to breed***

In many CBF there is no intention or possibility of creating self-sustaining populations. In these circumstances, genetic resource management should strive to optimize productivity and reduce negative impacts on the ecosystem. The production of sterile fish is the best means to reduce the chance of stocked fish breeding with local species. Creation of triploids, i.e. adding another set of chromosomes, is the most common method of producing sterile fish. Triploidy can be induced by temperature, pressure or chemical treatments to fish gametes and developing embryos. This is easily accomplished in many species such as oysters, salmon and trout, but difficult on a commercial scale for others, such as tilapia.

The release of individuals of a single sex, i.e. monosex, has also been used to reduce chance of reproduction. Monosex groups can be made either by genetic manipulation or by administering sex hormones at the proper time. Combining induced triploidy with monosex production would further reduce chance of unwanted breeding.

Monitoring of the stocked material is necessary to ensure that the hatchery is producing the desired product (i.e. that the stocked material is all triploids or all of the desired sex).

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<sup>104</sup> For example, International Council for the Exploration of the Sea ICES Code of Practice on the Introductions and Transfers of Marine Organisms 2004. [www.ices.dk/reports/general/2004/ICESCOP2004.pdf](http://www.ices.dk/reports/general/2004/ICESCOP2004.pdf)

It is also possible to control breeding by controlling fishing effort and through choice of habitat that is stocked. Fish are often stocked in temporary waterbodies that dry up before they can reproduce, in enclosed water bodies that lack connection to critical spawning habitats, or in areas with intense fishing pressure where 100% of the stocked fish are taken. However, these conditions are not always 100% effective at preventing all reproduction, because some fish may move to areas where reproduction is possible or fishing less intense. The use of sterile fish in these circumstances would further reduce the chance of unwanted reproduction.

### **8.3 Monitoring, assessments and reporting**

As with all fisheries, monitoring of CBF is essential and for this the stocked material must be identifiable. Hatchery marking programmes are being mandated in many parts of the world to assess hatcheries' contributions to CBF. Physical tags can identify initial contributions, but if stocked fish reproduce in the wild only genetic tags can indicate hatcheries' contributions to subsequent generations. Increase in abundance of stocked species is sometimes, but not always, an indicator of hatchery contribution to a fishery; favourable changes in the environment or better fishery management may also promote natural increases.

Additionally, stocked fish have in some cases been shown to displace local con-specific stocks. This is a situation to be avoided and another reason why the ability to differentiate among hatchery and wild stocks is important in the overall assessment of stocking as a management strategy.

A precautionary approach to developing CBF requires the development of reference points (Chapter 11); target reference points to indicate desirable situations a fish farm will strive to achieve and limit reference points to indicate conditions to be avoided and then regular monitoring to see to what extents the reference points are met. The reference points should relate to stated objectives, risk assessment and measures of success (see Chapter 11).

Where hatcheries release viable organisms that are capable of reproduction in order to support CBF, there is the possibility that self-sustaining populations will develop and thus negate the need for further stocking. This might be especially true in recovery programmes for endangered species that would combine stocking with habitat improvement and improved legislation. Monitoring and honest discussions with stakeholders are required to determine whether further stocking is still needed after self-sustaining populations have been established.

FAO collects information on the numbers and species that are released into open waterbodies, including natural waterbodies, semi-natural waterbodies such as reservoirs, and other managed waters such as rice fields. In order to evaluate the contributions of CBF to national and global fishery production, hatchery managers should convey comprehensive and timely information on all releases for CBF to their national statistics offices, for forwarding to FAO.

## **9 CONSERVATION OF WILD FISH GENETIC RESOURCES AND AQUACULTURE<sup>105</sup>**

### **9.1 Introduction**

In unanimously adopting the Code, States recognized that conservation is a necessary element of responsible use. This chapter addresses the responsible use of wild fish genetic resources (FiGR) for aquaculture, emphasizing their conservation. Wild FiGR are a very valuable subset of all of the FiGR that are available for current and future use in aquaculture and related research. Wild FiGR are free-living FiGR in nature and minimally changed by human activities, though it is becoming difficult to find any completely unchanged wild populations (see section 9.2). Wild is therefore a relative term, meaning as wild as possible in changing circumstances.

Like all FiGR for aquaculture, wild FiGR comprise DNA, genes, gametes, individual organisms and populations (Chapter 1). Although not mentioning wild FiGR explicitly, the Code includes them implicitly in all of its references to biodiversity, cultured stocks, living aquatic resources, genetic diversity, wild stocks, cultured stocks and genetically altered stocks and, thereby, calls for their management (i.e., conservation and sustainable use) and for care for their habitats. The purpose of this chapter is to provide guidance to aquaculture policy- and decision makers so that they can promote responsible aquaculture, protect valuable wild FiGR and, where necessary, contribute to their recovery.

### **9.2 Wild fish genetic resources**

Wildness in fish is a special quality, widely recognized by naturalists and conservationists, as well as by commercial and sport fishers and fish consumers. However, true aquatic wilderness is shrinking and the wildness of free-living fish populations is easily compromised. Capture fisheries, loss of habitat and degradation of the aquatic environment reduce the genetic diversities of aquatic populations and other biodiversity. Aquatic protected areas become less natural the more intensively they are managed and the more they are influenced by non-protected areas around them.

Many self-sustaining fish populations in nature have been derived from purposeful stocking, from fish escaped from aquaculture and from fish discarded from aquaria. Such populations include alien and native species.

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<sup>105</sup> Contributed by Roger S.V. Pullin.

Those descended from fish that were wild types or genetically close to wild types still represent wild FiGR. Those descended from fish that were at various stages of domestication, including distinct strains, hybrids and other genetically altered forms, are feral fish; analogous to the feral livestock that are descended from by animals that escaped from farms and ranches. Feral forms are naturally selected back from domestication to fitness in the wild. Feral fish represent valuable FiGR for capture fisheries and for aquaculture and related research. They are not wild FiGR *per se* but should be included with wild FiGR for management purposes.

The following types of fish all contribute to the diversity of wild FiGR: wild type native species; free-living alien species, descended from introductions and releases of wild fish; and free-living populations of species that extended their former natural ranges when barriers were removed, e.g. introductions into the Mediterranean via the Suez Canal. Many of the world's wild fish populations are distinguishable from their farmed relatives by their location, appearance, behavior and, above all, by biochemical genetic characterization. Although some farmed fish populations are wild types because they were collected as wild seed - for example, mollusc spat - most are genetically different from their free-living relatives in wild populations, with markedly different frequencies for many alleles.<sup>106</sup> Even where no purposeful selection or other genetic alteration is applied, successive generations of captive reproduction yield fish that differ increasingly from wild types (Chapters 3 and 4).

In the broadest sense, wild FiGR for aquaculture comprise not only those of farmed fish species but also those of other species in all of the ecosystems that support aquaculture production; for example, wild fish populations that are harvested for making aquaculture feeds and the plankton and microorganisms on fish farms that provide feeds, oxygen and waste processing. Therefore, the genetic resources of these organisms, upon which aquaculture production depends, must also be documented and conserved through appropriate measures applied to capture fisheries and to the health of the ecosystems in which aquaculture is practiced.

### 9.3 Importance for aquaculture

Domestication and genetic improvement of most farmed fish are far behind those for cultivated plants and livestock. Captive reproduction and breeding

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<sup>106</sup> Elliot, N. and Evans, B. 2007. Genetic change in farm stocks: should there be concern? *World Aquaculture*, 36 (1): 6-8.

programmes have been established for many species of farmed fish but not for all. Therefore, some fish are still farmed as wild types or as undomesticated populations that are close to wild types. Seaweed farming also relies heavily on the propagation of wild types. If domestication of fish is defined as continuous controlled reproduction for more than 3 generations, only 30 species of farmed fish, out of 103 for which 2004 production exceeded 1 000 mt, can be termed domesticated<sup>107</sup> (Chapter 3). Capture-based aquaculture (CBA),<sup>108</sup> aquaculture-based fisheries (CBF – Chapter 8) that involve wild-collected fish seed or hatchery seed from wild-collected broodstock, and capture fisheries that provide feeds and feed ingredients for aquaculture all harvest wild fish. As new technologies for captive reproduction become available, the farming of wild and undomesticated fish will diminish but wild FiGR will remain important for aquaculture, for use in fish breeding programmes and related research. This is analogous to the continuing importance of the wild relatives of cultivated plants as sources of genetic diversity to be tapped by plant breeders, despite huge progress in plant genomics. The same will apply to farmed fish, even as fish genomics advance and modern genetic technologies are increasingly used in aquaculture. Aquaculture will face inevitable challenges from, for example, new and more virulent diseases, climate change and the need to cut production costs and raise productivity by improvement in a wide range of performance traits. Most of the FiGR that can contribute to meeting these challenges are wild FiGR. They are extremely valuable public goods that are vulnerable and, in many cases, vanishing. Therefore, it is important first to recognize that wild FiGR are vital for the future sustainability and profitability of aquaculture and, second, to invest adequately in their characterization and conservation, so as to ensure their continued availability.

## **9.4 Approaches to management**

### ***9.4.1 Categorization and prioritization***

Wild fish populations can become genetically differentiated when there is a reduction in the exchange of genes (gene flow) among them and when there are different selective pressures from the environment (Chapter 3). They are found as small populations with high rates of gene flow; partially

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<sup>107</sup> Bilio, M. Controlled reproduction and domestication in aquaculture. The current state of the art. Part II. Aquaculture Europe, 32 (3): 5-23.

<sup>108</sup> Ottolenghi, F.; Silvestri, C.; Giordano, P.; Lovatelli, A. and New, M. 2004. Capture-based aquaculture. The fattening of eels, groupers, tunas and yellowtails. Food and Agriculture Organization of the United Nations, Rome, Italy. 308p.

isolated subpopulations sometimes having local adaptations; more isolated local populations often having local adaptations; isolated, distinct closed populations; and metapopulations connected through migrations. For any species used in aquaculture, the overall goal should be to maximize the continued availability of as much wild genetic diversity as possible.

The genetic diversity of a species is usually represented by variations across its geographical range, with the more isolated and undisturbed populations often being the most distinct. The key is to gather sufficient genetic data to characterize as much as possible of the genetic diversity of the species, and in so doing to identify the wild populations that represent the most significant contributions to that diversity. In the conservation literature, these may be called conservation units or evolutionarily significant units. They represent important components of the total genetic diversity within a species. Moreover, some local fish populations, though superficially similar to others, are distinct, cryptic species and as such have unique and valuable genes.<sup>109</sup>

Prioritizing among a wide diversity of wild FiGR for conservation and arriving at consensus on management measures are difficult, especially where genetic data are limited. A highly precautionary approach is recommended, assigning high priority to conservation of FiGR that are clearly distinct and which represent significant contributions to the overall wild genetic diversity of the species, as far as that is known, but also assuming that all other FiGR are potentially important. Advice from professional geneticists should be sought to make the most of all information to hand and to remedy information gaps.<sup>110</sup>

High priority wild FiGR for conservation include populations in separate waterbodies and watercourses, on and around different islands, and in different bays and estuaries. Geographical isolation usually indicates distinctiveness and potential value of wild FiGR. For highly migratory species, this criterion of isolation applies particularly to breeding populations and early life history stages. Potentially distinct and valuable wild FiGR are also indicated by different migration patterns, spawning seasons, and other behaviour. Populations close to the natural centres of genetic diversity of species are usually important as wild FiGR and should be given high priority for conservation, but it is also important to conserve representative populations

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<sup>109</sup> Thorpe, J.P.; Solé-Cava, A.M. and Watts, P. 2000. Exploited marine invertebrates: genetics and fisheries. *Hydrobiologia*, 420: 165-184.

<sup>110</sup> Pullin, R.S.V. 2000. Management of aquatic biodiversity and genetic resources. *Reviews in Fisheries Science*, 8 (4): 379-393.

across the entire natural range of a species, particularly those close to its limits and in extreme habitats: for example, the most northerly and southerly populations and those in hot springs or high salinities. Expert advice from conservation geneticists should be sought to prioritize among wild FiGR for conservation. Where such advice is not easily obtainable, it can be sought from international organizations, including FAO, the World Conservation Union (IUCN),<sup>111</sup> the secretariats of international conventions – for example, the Convention on Biological Diversity,<sup>112</sup> the Convention on International Trade in Endangered Species,<sup>113</sup> and the Convention on the Conservation of Migratory Species of Wild Animals.<sup>114</sup>

#### ***9.4.2 Intersectorial perspectives***

Article 9.1.3 provides for the sharing of resources among aquaculture and other sectors: “*States should produce and regularly update aquaculture development strategies and plans, as required, to ensure that aquaculture development is ecologically sustainable and to allow the rational use of resources shared by aquaculture and other activities.*” This requires intersectorial perspectives. Conservation of wild FiGR is part of nature conservation, which is a sector in its own right. The habitats of wild FiGR and their waters are used by humans to varying extents for agriculture, aquaculture, conservation of wildlife, forestry, industry, mining, nature conservation, navigation, power generation, recreation and tourism, water supplies to human settlements and industry, and waste treatment and disposal. Conservation of wild FiGR must contend with the needs of all these other sectors, as they must all contend with each other’s needs.

Reconciling aquaculture with conservation of free-living wild FiGR is particularly difficult. Some waters that present opportunities for aquaculture also contain wild FiGR of high national and sometimes international importance. Fish farmers need and should be permitted to farm the most productive and profitable fish species and strains, as in agriculture, subject to their compliance with biosafety, biosecurity, other environmental safeguards and legal access and ownership. However, fish that have escaped from farms and pathogens from fish farms can have adverse impacts on wild FiGR and on other wild biodiversity and habitats.

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<sup>111</sup> [www.iucn.org](http://www.iucn.org)

<sup>112</sup> [www.biodiv.org](http://www.biodiv.org)

<sup>113</sup> [www.cites.org](http://www.cites.org)

<sup>114</sup> [www.cms.int](http://www.cms.int)

As aquaculture expands in watersheds, coastal areas and the open sea, policy-makers and regulators must increasingly consider which fish they will allow to be farmed in which locations, and the conservation of wild FiGR is a factor here. The four options, in order of increasing restrictiveness and precaution in pursuit of conservation goals, are: 1. permit the farming of any fish; 2. permit only the farming of a native fish species; 3. permit only the farming the strain of a native species that is typical of that locality – note here, however, that the farmed fish strain(s) will soon become genetically different from the local wild strain(s); and 4. prohibit all aquaculture. Choosing among these options is difficult. Aquaculture development gains must be balanced against losses of and changes to wild FiGR, other biodiversity and habitats. The approach recommended here is to follow the general provisions of the Code to allow only development of responsible aquaculture, which implies the setting and pursuit of nature conservation goals, including conservation of wild FiGR, and the safeguarding of interests of other sectors. Taking an intersectoral perspective is the key to achieving a balance between development and conservation. Even when limited to a few sectors – for example, to aquaculture, nature conservation and water resources management – an intersectoral perspective here benefits those and other sectors that depend upon aquatic ecosystem health and services.

Stakeholders in these and other sectors should meet, discuss and arrive at a balanced consensus, based upon mutually agreed compromises, sacrifices and sharing of benefits. This will often be difficult because, historically, many of the institutions for aquaculture and for conservation have been separate, with aquaculture development and oversight proceeding independently of the setting and pursuit of conservation goals. Intersectoral institutions are not yet well developed, though their establishment is implied in the Code for the furtherance of responsibility in aquaculture. Therefore, development of intersectoral institutions, to work for harmony among aquaculture, conservation and other sectors, should be pursued urgently. The intersectoral perspective must be maintained not only prior to and during the development of aquaculture but also through ongoing and indefinite oversight of aquaculture and its intersectoral relationships. This is also recognized in Article 7.6.8 which requires that “*conservation and management measures and their possible interactions should be kept under continuous review.*”

### **9.4.3 Twinning aquaculture and conservation**

Twinning the development and oversight of aquaculture with measures for and monitoring of the conservation of wild FiGR, is recommend as a logical

means to ensure both the sustainable use and long-term conservation of wild FiGR.<sup>115</sup> Twinning requires the zoning of areas that are designated for aquaculture and areas for conservation that are completely off-limits to and isolated from aquaculture and fish farm waters, as well as from the impacts of other potentially disruptive sectors. In well chosen aquaculture areas, a wide choice of fish can be farmed, provided that conservation of wild FiGR is fully assured in twinned conservation areas, such as nature reserves and sacred sites. But twinning is more than just separate zoning of aquaculture and conservation of wild FiGR. It must involve co-policy-making, integrated action, co-monitoring and especially co-financing, with both sectors advancing interdependently. Use and conservation then become *twinned* management objectives and are co-funded continuously thereafter.

Conservation areas that fit the strict criteria defined here for twinning will not always be available. Many nature reserves and aquatic protected areas, though lacking isolation from impacts of aquaculture, fishing and other sectors and sometimes allowing rational use of their living aquatic resources, including fishing, play vital roles in conservation of FiGR.<sup>116</sup> Where, despite best efforts, it proves impossible to identify and to establish one or more conservation areas in a given ecosystem, such as a watershed or coastal zone, because of historical or present ecological and social circumstances, the concept of twinning can be widened, nationally and internationally. The main requirement is aquaculture development anywhere that could compromise the integrity of wild FiGR is linked to *in situ* and complementary *ex situ* conservation of those wild FiGR somewhere.

#### **9.4.4 In situ conservation**

By convention, the distinct varieties, strains and breeds of cultivated plants, farmed fish and livestock are called *in situ* genetic resources when located in the farms that are their natural surroundings. Their free-living wild relatives in nature are also called *in situ* genetic resources. Well-managed aquatic protected areas are *in situ* genebanks for wild FiGR (Chapter 10), though this role is not often recognized and their management often lacks adequate

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<sup>115</sup> Pullin, R.S.V. *in press*. Aquaculture and conservation of fish genetic resources: twinning objectives and opportunities, p. 00-00. *In* Pioneering Fish Genetic Resource Management and Seed Dissemination Programmes for Africa: Adapting Principles of Selective Breeding to the Improvement of Aquaculture in the Volta Basin and Surrounding Areas. CIFA Occasional Paper No. 29. FAO: Accra, Ghana.

<sup>116</sup> Ramsar Convention on Wetlands ([www.ramsar.org](http://www.ramsar.org)), Parties to which consider the presence of important fish populations as a criterion for designation of Ramsar sites.

gathering and use of genetics data. *In situ* wild FiGR are found only in natural or relatively undisturbed habitats. The two main requirements for *in situ* conservation of any population of wildlife in any protected area are: i) to maintain a genetically effective population size; i.e., an number of effective breeders ( $N_e$ ), so as to avoid the inbreeding depression and loss of genetic variation to which small, isolated populations are always at risk (see also Chapter 3);<sup>117</sup> and ii) to pay equal attention to the management of their habitats, so as to prevent their degradation or loss. Unless the latter is successful, the FiGR targeted for conservation will be changed or lost. The continued presence and integrity of the waters and biological communities that host particular wild FiGR must be assured, in the face of challenges by *inter alia* climate change, dam construction, droughts, floods, introductions of alien species and diseases, overfishing pollution, siltation and water abstraction. In this respect, *in situ* conservation of wild FiGR faces the same constraints as all nature conservation, but the threats to wild fish, especially freshwater and highly migratory fish, are greater than those for all other vertebrate groups used as food by humans.

*In situ* conservation of threatened and important wild FiGR should not be abandoned because the populations that remain for conservation purposes have low  $N_e$ s. Small populations of wild FiGR conserved *in situ* contribute to the overall conservation effort for a given species and are particularly important where they represent rare or sole remaining examples of a genetically distinct local population, such as a riverine or lacustrine race. *In situ* conservation of wild FiGR has operational and opportunity costs and these must be recognized and shared by public and private beneficiaries.

One of the key issues with respect to all *in situ* wild genetic resources, including FiGR, is how to ensure their responsible collection from nature, avoiding in particular over-collection and unauthorized collection, and their exchange and fair use thereafter. In 1993, the Member Nations of FAO negotiated an International Code of Conduct for Plant Germplasm Collecting and Transfer,<sup>118</sup> the objectives of which can all be applied to wild FiGR. The Convention on Biological Diversity<sup>119</sup> – particularly its Articles: 8, on *in situ* conservation, and especially 8j, on equitable sharing of benefits; 15, on access to genetic resources; 17, on exchange of information; and 18 on technical and scientific cooperation - and many other international and national instruments

<sup>117</sup> Frankham, R. 1995. Conservation genetics. *Annual Review of Genetics*, 29: 305-327.

<sup>118</sup> FAO. 1994. *International Code of Conduct for Plant Germplasm Collecting and Transfer*. Food and Agriculture Organization of the United Nations: Rome, Italy. 20p.

<sup>119</sup> [www.biodiv.org](http://www.biodiv.org)

provide for the management of all biodiversity, including implicitly *in situ* wild FiGR, but have so far been applied much more extensively to other wild genetic resources, especially to the wild relatives of cultivated plants.

#### 9.4.5 *Ex situ conservation*

Conservation of FiGR as live fish is called *in vivo* conservation. All *in situ* conservation of wild FiGR is *in vivo*, as fish populations of various sizes. The *ex situ* conservation of wild FiGR can be either *in vivo* as individuals or populations held in research establishments and aquaria, or *in vitro* as cryopreserved sperm, and more rarely as embryos and as any tissues containing DNA. *Ex situ/in vitro* conservation of wild FiGR as cryopreserved sperm is by far the most important technology available (Chapter 10). The absence of comparable technology for cryopreservation of the eggs and embryos of all farmed finfish and of most farmed aquatic invertebrates means that cryopreserved sperm can only be used to fertilize eggs from live females. However, cryopreservation of sperm is still a very important means of conserving wild FiGR, especially threatened wild FiGR, and for providing wild FiGR in breeding programmes and related research.

*Ex situ/in vivo* conservation of wild FiGR, in research collections and aquaria, faces the same constraints as all captive breeding for conservation purposes in zoos and other establishments: chiefly, that captive-bred populations become genetically different from their wild relatives, that the facilities available often constrain effective population size ( $N_e$ ) and that security of funding is often limited. Public-private partnerships can help to mobilize more resources for *ex situ* conservation of wild FiGR, sharing the costs and benefits, though public funding will usually have to take the lead. *Ex situ/in vivo* collections of wild FiGR are kept for research purposes by many public funded organizations, especially universities, as well as by the private aquaculture sector. Public and private aquaria are also *in vivo* fish gene banks and some of their fish can be FiGR for aquaculture. *Ex situ/in vivo* collections of wild FiGR should be managed to keep them as genetically close to wild type as possible, minimizing loss of genetic variation (Chapters 3, 4 and 10).

*Ex situ* conservation of wild FiGR should be considered first as complementary to their *in situ* conservation, with high emphasis on the latter. However, where no or few undisturbed and accessible free-living populations of important FiGR remain, *ex situ* conservation becomes the main or only approach to ensure their long-term conservation and availability. As recommended above for *in situ* conservation, all efforts to conserve threatened and important wild

FiGR *ex situ* are valuable and contribute to the overall conservation of genetic diversity for a given species. As with the conservation of rare animals in zoos and rare breed trusts, this applies even where cryopreserved genetic material is representative of only a few individuals or populations and where *in vivo* populations have low  $N_e$ s.

Wherever aquaculture development and conservation of wild FiGR for aquaculture are undertaken, concurrent provisions should be made for all necessary current and foreseen *in situ* and *ex situ* conservation of wild FiGR. The twinning approach is again recommended here, with appropriate institutional development and capacity building for both *in situ* and *ex situ* FiGR conservation methods.

## 9.5 Information

Accurate and up to date information is of paramount importance for the effective management of wild FiGR. For effective zoning of aquaculture and *in situ* wild FiGR conservation areas, wild FiGR must be fully documented, including as far as possible genetic characterization. Only with such information can they be prioritized for conservation. Thereafter, information must still be collected to monitor the status of *in situ* populations and, where applicable, complementary efforts in *ex situ* conservation. This information should be shared and disseminated in a variety of formats such as genetic databases, scientific journals, and on-line open access sources. FishBase<sup>120</sup> is a good example of an information system that can be used for recording and disseminating such information, from its own contents that relate to wild FiGR and from linkages to other relevant databases. The FAO Species Identification Programme<sup>121</sup> and Aquaculture Fact Sheets<sup>122</sup> contain taxonomic descriptions, based on morphology, with only limited genetic data. However, information systems for wild and other FiGR are likely to change as the scope and demand for this information grow. Guidance on new developments in FiGR information sources can be sought from the FAO Commission on Genetic Resources for Food and Agriculture. Moreover, with conservation genetics increasingly applied to a wide range of taxa, information on wild FiGR is increasingly available from national, regional and international nature conservation organizations.

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<sup>120</sup> [www.fishbase.org](http://www.fishbase.org)

<sup>121</sup> <http://www.fao.org/fi/website/FIRetrieveAction.do?dom=org&xml=sidp.xml>

<sup>122</sup> <http://www.fao.org/fi/website/FISearch.do?dom=culturespecies>

National and local inventories, i.e. computerized lists and databases, of wild FiGR should be established from an inclusive perspective, to comprise all free-living fish populations - wild, feral and others - and their accessible individuals, gametes, DNA and genes. This approach recognizes that wildness is a relative attribute. Inventories should include for each population and for other forms of wild FiGR: accurate and authoritative specific (and where applicable intraspecific) identification and scientific nomenclature, references to sources of local and indigenous knowledge and nomenclature, distinguishing characteristics, genetic characterization data, conservation status, history of use in aquaculture and main threats.

Site-specific, management of *in situ* wild FiGR requires broader information sources and planning instruments because it comprises both the management of the FiGR *per se* and the management of their habitats. Information must, therefore, be sourced from all the sectors that could have adverse impacts on the latter, including all likely changes to the surrounding watershed or coastal zone and especially any foreseeable changes in water quality and quantity. Some of the methods to be applied here, such as Geographical Information Systems are long-established, though their application in conservation genetics is relatively new. Managing natural habitats specifically for FiGR conservation is also relatively new and published information about experiences and guidelines are limited. Medical practice faces a similar situation in striving to synthesize and disseminate recent information in order to maximize effective actions, and it has been suggested that conservation could learn from some its approaches to information processing.<sup>123</sup>

Information sources about different types of fish habitats are generally less well developed than those for fish biology, and every individual situation of a fish habitat and its wild FiGR will have some unique features. The need to understand fish habitat ecology is a key requirement for conservation<sup>124</sup> and advice from aquatic ecologists that can be applied to management of *in situ* wild FiGR is increasing. A good example is the list of sites for which Ecopath analyses have been completed as information for ecosystem-based management.<sup>125</sup> Expert advice on information for managing habitats in conservation of wild FiGR should be sought from professional aquatic

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<sup>123</sup> Fazey, J., Salisbury, J.G., LindenMayer, D.B., Maindonald, J. and R. Douglas, 2004. Can methods applied in medicine be used to summarize and disseminate conservation research? *Environmental Conservation*, 31 (3): 190-198.

<sup>124</sup> Rice, J.C. 2005. Understanding fish habitat ecology to achieve conservation. *Journal of Fish Biology*, 67 (Supplement B): 1-22.

<sup>125</sup> [www.ecopath.org](http://www.ecopath.org).

ecologists and geographers. Where such advice is not easily obtainable, it can be sought in the first instance from IUCN.

## 9.6 Conservation aquaculture for endangered fish

The term endangered is used here in a broad sense, comprising species listed by the Convention in International Trade in Endangered Species,<sup>126</sup> all species categorized in the Red List of IUCN as threatened (where three subcategories - vulnerable, endangered, and critically endangered – are defined),<sup>127</sup> and all species and other taxa termed endangered in national legislation. International lists are important; however, lists should also be made at a national or local level of endangered species that are locally important and that may be endangered. Aquaculture decision makers could request such lists from national fisheries or environment officers. The main strategies for conservation of all endangered species are to protect and to rehabilitate their natural habitats from degradation and to protect their populations from adverse impacts.

Captive breeding can be also used to augment remaining wild populations and, where there have been local extinctions, for reintroductions.<sup>128</sup> When applied to endangered fish, this can be termed conservation aquaculture, but its interventions must be integrated into an overall resource management strategy involving *inter alia* conservation areas, fishery management and well managed access to natural resources. Captive breeding and the production of hatchery seed have been used to assist with the conservation and use of a wide range of endangered fish, including: the Mekong giant catfish; mahseers; giant clams; ornamental species such as arowana; paddlefish and sturgeons; and several species, subspecies and runs of salmon and trout.

Many public aquaria have some endangered fish among their collections, but the large captive breeding efforts of zoos to assist conservation of endangered animals, particularly birds and mammals, have not yet been matched by similar efforts for fish. Guidelines have been published for captive breeding as an aid to conservation of endangered fish species.<sup>129</sup> As with all *ex situ* breeding of wild fish, the main principle for captive breeding to assist in the conservation of endangered fish is to keep captive broodstock and their

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<sup>126</sup> [www.cites.org](http://www.cites.org)

<sup>127</sup> IUCN. 1994. IUCN Red List Categories. IUCN, Gland, Switzerland. 21p.

<sup>128</sup> IUCN. 1998. IUCN Guidelines for Re-introductions. IUCN, Gland, Switzerland and Cambridge, U.K. 10p.

<sup>129</sup> Huntley, R.V.; Langton, R.W. 1994. Captive Breeding Guidelines. Aquatic Conservation Network, Inc., Ottawa, Ontario, Canada. 62p.

progeny as genetically close as possible to the wild type populations that are being augmented or re-established (Chapter 3). However, for endangered fish that are close to extinction, the situation can be so serious that any captive breeding, even if compromising these genetic goals and reliant on very low  $N_e$ s, is better than none.

## 9.7 Summary

Wild fish genetic resources (FiGR) represent the majority of the genetic diversity that is available for the further domestication and genetic improvement of farmed fish.

Many wild FiGR are threatened with genetic change or extinction. These wild relatives of farmed and potentially farmable aquatic species must be valued and protected in order to ensure their future availability for use in aquaculture.

With adequate recognition of the value of wild FiGR and sharing of the costs and benefits of their conservation, there is still time and opportunity for aquaculture to avoid losses of wild genetic resources to the extents that have been experienced in the livestock and crop sectors.

*In situ* conservation of wild FiGR should be recognized as part of the nature conservation sector, and should be pursued through intersectorial action and cooperation.

*Ex situ* conservation of wild FiGR to complement *in situ* efforts for aquaculture is an important option and captive breeding can assist conservation of some endangered fish.

For all aspects of the management of wild FiGR, accurate and up to date information is of paramount importance.

Conservation of wild FiGR should be accorded adequate importance in funding allocations and in the sharing of natural resources with other sectors.

## 10 BANKING AQUATIC GENETIC RESOURCES<sup>130</sup>

### 10.1 Introduction

A gene bank is a managed collection of genetic resources. Gene banks are necessary whenever the genetic resources fundamental to farming and harvesting animals and plants are threatened. While modern genetic techniques make it possible to bank any plant or animal tissue that contains DNA, most gene banks are collections either of whole organisms, their reproductive cells or early life stages. A good indication that a collection is actually a bank is if one can make a withdrawal from it. The technologies used for aquatic gene banking are as applicable to industry (broodstock collections, prospecting for new genetic material) as they are for traditional conservation.

### 10.2 *In situ* and *ex situ* gene banks

A gene bank can be *in situ* or *ex situ*, a distinction based largely on its physical location. *Ex situ* banks, which can be collections of DNA, genes, single cells, seeds or whole organisms, are remote from the organism's natural or farmed habitat; they are the commonest kind of gene bank, and the one most familiar to the public. *In situ* banks are populations of organisms protected along with their natural or farmed habitat; they are less common than *ex situ* banks, but may be more palatable to agencies and the public (see Chapter 12). While the Convention on Biological Diversity (CBD) regards *ex situ* banks as "complementary" to *in situ* ones, both explicitly address Articles 7.2.2 and 9.3.1 (*in situ* and *ex situ* banks) and 9.3.5 (*ex situ* banks) of the CCRF. They are equally important for aquatic genetic resources.

Gene banks for aquatic organisms are much more recent than the seed banks and livestock insemination centres familiar to many people. The biggest difference is that, unlike domesticated plants and animals, aquatic organisms are still captured from wild ecosystems or from farmed stocks, so their preservation in gene banks should involve preservation of natural habitats (aquaculture systems are not yet threatened). Loss of habitat means that the option of an *in situ* gene bank of wild species no longer exists for many wild plants and animals, but remains very much available for finfish, shellfish and aquatic plants. Managers of aquatic gene banks must thus be clear about the breadth of options for conserving genetic resources of farmed aquatic species, for which an *in situ* bank can include not only a live, "on farm"

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<sup>130</sup> Contributed by Brian Harvey.

collection of a particular breed, but also a portion of the habitat of its wild relatives (Chapter 9). In this chapter, only the first *in situ* option, namely on-farm conservation, is considered.

### 10.3 History

The first gene banks for aquatic organisms were small collections of cryopreserved sperm gathered by researchers interested in wild populations of finfish. Their most obvious utility, however, was for safeguarding the results of aquaculture breeding programs. Many of the collections that followed were short-lived due to poor investment planning, poor technology and lack of government buy-in. A number of “living gene banks” (again, mostly finfish) also arose in the form of captive broodstock collections in state or private hatcheries.

Today, managed *ex situ* collections of aquatic animal germplasm and whole organisms are maintained by national, state and indigenous governments, private companies, academics and NGOs. Some are part of a concerted national effort at aquatic germplasm conservation. While these *ex situ* banks are widespread, their terminology and technologies need to be standardized and lines of communication set up. Partnerships between groups enormously strengthen any program, and should be sought.

### 10.4 Guidance on banks of cryopreserved gametes and embryos

A gene bank represents an unusually long commitment to maintaining infrastructure. Although relatively easy to set up, gene banks are hard to maintain over decades, which is their natural time frame. They *can* be successfully used on a small scale (for example, on a single farm) but the livestock model, which uses a central (and centrally funded) storage and records centre is probably the best long-term bet. This multi-user model is the one contemplated in the following discussion.

The sperm of many species of freshwater finfish has been successfully cryopreserved (frozen indefinitely in liquid nitrogen). Fish spermatozoa present few serious technical problems, although progress has been hampered by a low quality scientific literature on the topic, reflecting many empirical attempts uninformed by cryobiological theory. Researchers and those establishing gene banks should consult recent reviews<sup>131</sup> for more in-

<sup>131</sup> A recent example is Tiersch, T., and Mazik, P. (eds). 2000. Cryopreservation in aquatic species. World Aquaculture Society, Baton Rouge. 439 pp.

depth technical guidance and are encouraged to disseminate their experiences widely, including in the peer-reviewed literature.

Fish sperm is generally frozen and stored in plastic straws. The actual freezing can now be done in the field using portable, low cost equipment. It is not yet possible to freeze finfish eggs. Sperm and ova of some shellfish have, however, been successfully frozen, and the larvae of bivalves (oyster, clam, scallop, mussel) are well suited to cryopreservation; several national research programs currently target bivalve gene banking. Gene banks should currently target fish spermatozoa or bivalve ova and larvae.

Cryopreserved sperm, ova and larvae are stored in liquid nitrogen. Secure storage should be sought in a livestock breeding centre amenable to contracting out space and manpower. Duplication in another site is an extra safeguard but is practical only for small collections. If the species being conserved has not been cryopreserved before, the main cost for this kind of *ex situ* bank is in developing or acquiring the technology; sources include academic and government researchers, although some private fish farms have also invested in refinement of existing techniques.

### **10.5 Guidance on living gene banks (broodstock collections)**

Isolated collections of “pure” brood lines of live fish have long been part of large-scale hatchery programs that produce fish for sale to other farms, for conservation and for release to the wild. The main requirements of this or any other kind of living gene bank are that the conserved stocks remain secure and that their genetic diversity is maintained. They must, however, be bred, which imposes selective forces and inevitably distances them from their original wild state (see Chapters 3 & 9). Captive breeding of endangered fish populations has become a familiar part of the gene banking scene. Broodstock collections can also be maintained in academic research laboratories and public aquaria.

### **10.6 Data management**

While much effort has been spent to develop software for managing plant and livestock gene bank accessions, and the existing international agreements on gene banking have stimulated a fair degree of standardization, the majority of fish gene banks still rely on crude in-house record-keeping systems based on widely available spreadsheet software. Most of these home-grown systems fail when asked to provide good records of withdrawals, exchanges and replacements;

none of them can account for the broad range of data that plant and livestock gene banks normally maintain. While the requirements for fish gene banking will differ somewhat depending on location and the kind of bank, data that must be accounted for will usually include provenance (what was collected, where and by whom, and under what legal arrangement); identification (species and, where possible, population genetics); and subsequent use (removal and re-deposit of samples, by whom and for what purpose).<sup>132</sup>

### **10.7 Policy implications**

Given the appropriate containers, cryopreserved genetic resources are far easier to transport, over any distance, than living ones. Those doing so must be aware of national and international legislation on introductions, transfers and disease control.

Few governments, even those which are parties to the CBD have policies on aquatic gene banking. Yet the CBD enshrines precisely those principles that demand such policy – namely access to genetic resources and sharing the benefits derived from them. These principles affect every group possibly interested in gene banking: communities, the aquaculture industry, indigenous groups, NGOs and fisheries and environment ministries. Access to genetic resources, especially those removed from their natural habitat and stored for later use, can rapidly become politically or legally difficult. Each group must therefore understand the policies of other involved group before embarking on a gene banking program, and reach prior agreement on access, storage and use of those resources. There is so far no standard format or general principles for such agreements specific to aquatic genetic resources.

Resource management and development agencies, especially international agencies, should work toward standardization of terminology, policies, technologies and record keeping; additional policies may need to be developed as the field of genetic resource characterization advances.

### **10.8 Establishing an aquatic gene bank**

For any group wishing to establish an *ex situ* aquatic gene bank, the following steps should be followed:

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<sup>132</sup> SpermSaver – Gene Bank Management Software. 2005. World Fisheries Trust, Victoria BC, Canada. This is a beta version of fish gene banking software that addresses all these areas available from World Fisheries Trust ([www.worldfish.org](http://www.worldfish.org)).

- find a long term institutional home for the programme (e.g. a fisheries or agriculture agency) and a long term physical home for secure storage (e.g. a state or private livestock insemination station);
- secure short-term funding (e.g. granting agencies) for research and long-term funding (primarily government) for secure storage;
- acquire the technology from academia or in-house research funded as above;
- train field staff regularly on technology, data management, permitting and legislation;
- survey and incorporate into a gene bank management plan all relevant environmental and fisheries legislation and regulations, including those on disease control, transfer of live animals and their gametes, and endangered species;
- develop policies on acquisition and release of material and in particular with regard to access to genetic resources and the sharing of benefits arising from its use;
- make the links to providers of associated data on accessions (for example, modern DNA analysis allows for fine-level characterization of genetic structure; a standardized aquatic gene banking system would incorporate the results of such analyses); and
- develop a mission statement and hire a communications specialist to promote the objectives, terms of use and policies of the gene bank with all partners.

## 11 A PRECAUTIONARY APPROACH<sup>133</sup>

To ensure that aquaculture development proceeds in a responsible manner, the international community through, for example, the Convention on Biological Diversity (CBD) and the FAO Code of Conduct for Responsible Fisheries, many national governments, NGOs and others are calling for the adoption of a precautionary approach.

All development has impact. Society wishes to benefit from the development of new technologies and genetically improved species for culture, while at the same time society expects government to protect it from any harmful effects of that development. Balancing developmental progress and the adverse impacts from progress is the essence of a precautionary approach to the use of genetically altered species (Chapter 2) in aquaculture.

There is still a high level of uncertainty and debate on the probability and magnitude of many of the adverse impacts of genetically altered species on the environment and on aquatic biodiversity. Current understanding of many species, aquatic ecosystems and the forces that structure them is often not adequate to predict accurately how a biological community or ecosystem will respond to the introduction of genetically altered species.

### 11.1 An approach

The precautionary approach advocated by FAO and CBD states that where there are threats of serious or irreversible damage, lack of full scientific certainty shall not be used as a reason for postponing cost-effective measures to prevent environmental degradation. Elements of the precautionary approach developed for capture fisheries and introduced species<sup>134</sup> follow.

- Reference points should be established to help determine desirable situations and undesirable impacts, e.g. target and limit reference points. For example Maximum Sustainable Yield could be considered a target reference point, whereas occurrence of not more than a given number of escaped farmed fish in the wild would be a limit reference point. Some potential reference points are listed in Table 11.1. Resource managers should develop quantitative values for the reference points listed in Table 11.1.

<sup>133</sup> Contributed by Devin M. Bartley.

<sup>134</sup> FAO. 1996. Precautionary Approach to Capture Fisheries and Species Introductions. FAO Technical Guidelines for Responsible Fisheries No. 2. FAO, Rome.

**Table 11.1** Possible reference points for the application of a precautionary approach to genetic resources management in aquaculture. T and L are Target and Limit reference points, respectively.

Purpose of establishing a reference point	What to measure for reference point
<b>Genetic</b>	
To establish acceptable level of inbreeding (L)	- Inbreeding coefficient (F) (Chapter 3)
To establish acceptable level of gene flow/introgression between farmed and wild stocks (L)	- Number of wild and farmed fish exchanging genes - Change in gene frequency in wild stocks
To establish acceptable number of fish to be used as broodstock (T)	- Effective population size ( $N_e$ ) (Chapter 3) of broodstock
To ensure sterile aquaculture product	- Number of triploid fish in hatchery product
To conserve rare genes in culture (T)	- Effective population size ( $N_e$ ) (Chapter 3) - Gene frequency in hatchery stocks
<b>Native stock abundance</b>	
To assess impact of escapes	- Number of escaped fish from aquaculture - Percent decline in native fishes
To establish level of endangerment (L)	- Reduction in population size over a given period of time (e.g. 10 years or 3 generations)
To establish acceptable fishery impacts (T and L)	- Fishing mortality; - Maximum Sustainable Yield.
To establish risk of extinction (L)	- Effective population size - Probability of extinction within a given time (e.g. 5 yrs) - Decrease in population size (e.g. order of magnitude decrease over a period of time)
<b>Pathogens</b>	
To prevent spread of disease (L)	- Levels of specific pathogens in farmed and wild populations (often 0 is set as a target and limit reference point for pathogens)

- Undesirable outcomes, as well as corrective or preventative measures, should be identified, including the prohibition or enforced cessation of activities that carry unacceptable risks or have already had unacceptable adverse impacts. Pre-agreed actions or contingency plans should be implemented in a timely manner when limit reference points are approached, or when adverse impacts are apparent. Thus monitoring of aquaculture facilities, local species and the environment is necessary to know when reference points are reached. Such actions could include switching to sterile fish if breeding with local species is a problem or changing containment

or location of facilities. Conversely, if good culture practices are used and no adverse impacts are monitored, additional development following the same approach could be planned.

- Priority should be given to maintaining the productive capacity of the resource where there is uncertainty as to the impact of development. In capture fisheries, this means that priority is given to conservation of stocks over harvesting the stocks when there is uncertainty. This can be extended to aquaculture where the productivity of local stocks should be maintained when there is uncertainty as to the risk of genetically altered species adversely affecting them. This may require locating fish farms in areas away from valuable local resources (Chapter 9).
- The impacts of development should be reversible within the time frame of 2 - 3 decades. This element renders as non-precautionary the use of reproductively viable, genetically altered species in many situations, none the less a precautionary approach can be followed. Species introduced for aquaculture have naturalized and established self-sustaining populations in many instances; the eradication of such populations (i.e. the reversibility of the impact) is difficult or impossible, especially in marine areas, large inland water bodies and wetlands, and extensive river systems.
- The burden of proof should be placed according to the above requirements and the standard of proof should be commensurate with risks and benefits (i.e. a higher standard of proof would be required when risks relative to benefits are high). The precautionary approach has often been taken to mean that the burden of proof rests with those proposing the use or development of a resource (i.e. the aquaculture facility must prove that a genetically altered species will have no adverse impact). This is the “guilty until proven otherwise” approach. The application of this, in real situations, is very complicated. All cases for allowing or prohibiting aquaculture activities should be based, to the greatest extents possible, on sound scientific information and opinion.

## 11.2 Conclusions

A precautionary approach acknowledges uncertainty and establishes mechanisms to deal with potential problems. Such mechanisms may involve *inter alia* policies, management programmes, risk management, monitoring systems and changes in management or development based on experience. Thus, this approach has much in common with adaptive management. The

requirement to perform environmental impact assessment or to follow codes of practice, such as the those developed by the European Union<sup>135</sup>, the International Council for the Exploration of the Sea and the European Inland Fisheries Advisory Commission (ICES/EIFAC) (Chapter 5) are excellent precautionary devices that help determine whether the use of genetically altered species should be undertaken.

The precautionary approach is action in the face of uncertainty, and in advance of and during development. The approach does not call for a lessening of research or less effort to reduce uncertainty. Action must be taken with the best scientific information available and to improve the scientific information available.

The application of a precautionary approach should weigh benefits and risks (Chapter 7). Thus, in areas with needs for increased protein or economic opportunities, aquaculture and the use of genetically altered species may provide benefits that other types of agriculture or development would not provide. Thus, a higher level of risk may be justified when benefits to a needy area are expected to be substantial. However, the needs of future generations must also be considered, especially if short-term interventions pose risks to maintaining their breadth of options for availability and use of wild genetic resources and aquatic ecosystems.

A precautionary approach to the use of genetically altered species in aquaculture requires the mobilization of significant effort in regards to management, monitoring and research. Reference points will be critical and, for the present, they are not well agreed for acceptable levels of genetic diversity or for numbers of escaped farmed animals necessary to cause adverse impacts. Countries should strive to apply the approach and provide information to national policy-makers and to FAO so that uncertainty is reduced, lessons can be learned and information can be disseminated to a wider audience.

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<sup>135</sup> EU Directive 90/220, on the release of genetically modified organisms in to the environment.

## 12 PUBLIC RELATIONS AND CONSUMER AWARENESS<sup>136</sup>

### 12.1 Introduction

Consumer acceptance of genetically altered organisms from aquaculture is critical to the success of a breeding programme. Not only will people decide whether or not to purchase the farmed product, they can also put pressure on policy-makers that can influence legislation governing the import and use of genetically altered organisms.

Public awareness is not considered in the original CCRF, except in a very general manner. Article 6.16 on General Principles recommends that, *States should ... promote awareness of responsible fisheries (including aquaculture) through education and training...*. Yet public acceptance of genetically altered products is increasingly important in aquaculture, its role in livelihoods and its potential impact on the environment. The Convention on Biological Diversity and Agenda 21 both cite public awareness as crucial to sustainable development and for effective public participation in decision-making.<sup>137</sup>

This chapter alerts decision makers to some of the issues around public relations. Problems result from a *lack of information* or *different points of view*. Both kinds of problem can be averted if the users and managers of genetic technologies establish lines of communication with stakeholders – and with each other. The goals of this chapter are to make decision makers and advocates of the application of genetic technologies aware of some non-technical issues that can influence the success of genetic resource management programmes, and to propose elements of a general communication or public relations strategy to help disseminate accurate information.

### 12.2 Communication strategy

A communication strategy is needed to help promote the responsible use of genetic technologies because consumers and the general public in most of the world do not understand how their food is produced. Confusing terms, inconsistently used terms, exaggerated claims of success or disaster, complicated subject matter, deliberate attempts to hide information or influence public opinion add to consumer confusion, even mistrust, of genetic

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<sup>136</sup> Contributed by Devin M. Bartley

<sup>137</sup> Raymond, R.D. 1999. Agricultural research and the art of public awareness. Pages 217-224 in Pullin, R.S.V., D.M. Bartley and J. Kooiman (eds) Towards Policies for Conservation and Sustainable Use of Aquatic Genetic Resources. ICLARM Conf. Proc. 59. 277p.

technologies. This is extremely unfortunate because the responsible use of appropriate genetic technologies can greatly benefit the consumer and the environment.

The communication strategy should have defined objectives and a defined target audience. A successful approach to communication is “framing”<sup>138</sup> a subject area. Framing deliberately focuses on certain parts of an issue (inside the frame), while omitting other aspects (outside the frame), in order to meet the objective and elicit support from an audience (e.g. consumer or policy-maker). For example, in a strategy to promote acceptance of genetically improved fish, the frame could include the cost savings from growing or buying fish that can be produced more efficiently, and not focus on the technical details of how that fish was produced.

A communication strategy may have to “reframe” an issue by changing the current focus. For example some groups have “framed” aquaculture as using too much land and natural fish in the production of aquaculture feeds. By stressing reduced land and feed requirements associated with producing genetically improved fish it is possible to “reframe” the discussion in a more positive light (Table 12.1).

None of the above is to suggest that promoters of genetic resource management should conceal, withhold or distort information. They should be pro-active by disseminating positive and accurate information on the advantages of genetic resource management.

Other elements that can help create the “frame” are presented below.

### ***12.2.1 Know your audience***

Know your audience is the most basic rule of public awareness. The “public” is composed of numerous diverse groups with different interests. These different interests will dictate their information needs. Current sociological research has demonstrated that people often make decisions not on the basis of science or logic, but on deeply held preconceptions or on very simple principles. Consumers want to feel good about what they are buying, either because it is good for their health, good for the environment or good value for

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<sup>138</sup> Annex 2: Sink or Swim: mobilizing key audiences through strategic communication. Suzanne Hawkes and Liz Scanlon IMPACS, September 2006. ([worldfish.org/images-pdfs/Projects/sinkorswim.pdf](http://worldfish.org/images-pdfs/Projects/sinkorswim.pdf))

**Table 12.1** “Framing” genetic management in aquaculture helps stress positive aspects to promote acceptance of genetic improvement programmes.

<b>Current “Frame” concerning genetic technologies</b>	<b>Suggested focus of a new frame</b>
Genetic technologies are costly	Genetic technologies are cost-effective by producing an organism that grows well and uses less inputs. Genetic technologies can be used to produce a specific color or shape of fish that consumers would pay a premium for.
Genetic technologies are complicated	Genetic technologies are often based on traditional animal breeding practices. The reproductive biology of fishes makes application of genetic technologies easy.
Genetic technologies are bad for native biodiversity and environment	Genetic technologies in aquaculture can reduce adverse environmental impacts. They can be used to produce organisms that have reduced ability to interact with wild ones; by growing more efficiently there will be less waste going into the environment; by having increased disease resistance there will be less chance of disease transmission and less pharmaceuticals used. Genetic resource managers in aquaculture should demonstrate that they place a high value on wild genetic diversity – it is the raw material for all genetic improvement programmes.
Genetic technologies benefit large companies	Benefits of decreased production costs will be passed on to the consumer.
Genetic technologies produce a product that consumers are afraid of, e.g. unhealthy, bad tasting, strange	Genetic technologies can be used to produce a healthy fish that has no ingredients not found in wild relatives.
Genetic technologies are harmful to farmed organisms	Improved domestication and production efficiencies from farming genetically improved fish will mean fish are less stressed in the culture environment, they feed better, have lower levels of aggressive interactions and will be less susceptible to diseases.

money, aquaculturists want access to lucrative markets, policy-makers want to do what is best for the majority of their constituents.

Consumers will be more strongly influenced by reduced prices for high quality genetically improved fish that are grown more efficiently and with less environmental impact. Policy-makers will be influenced by growing consumer and business demand for these traits. The growth of “organic” agriculture products and eco-certified capture fisheries is an indication that consumers want to buy a product that has reduced environmental impacts, as well as a product that is economical.

Because consumers may have strong feelings that are difficult to change, and existing laws are difficult to change, surveys should be conducted to ensure that any genetic technology used in production is accepted by consumers and will not have any associated legal or trade restrictions. For example, hybridization between different species is prohibited or requires special permits in some areas. Although at present there are no aquatic genetically modified organisms (GMO's) (i.e. transgenics) available to consumers, some are likely to be developed and approved in the future. Thus, consumer and trade partner acceptance of this technology should be examined before using it.

### ***12.2.2 Establish partners to help promote genetic management programmes***

Proponents of genetic technologies in aquaculture will need to partner with numerous stakeholders to ensure that the technologies are given a chance, used responsibly, and accepted by consumers and policy-makers (see also Chapter 9 on multi-sectoral approaches). Aquaculture is being criticized for causing adverse environmental impacts because of over-use of certain inputs and high discharge of contaminants. Genetic programmes that reduce these impacts through more efficient production should find wide acceptance in the aquaculture industry and conservation sectors.

Partnerships will promote confidence in the product produced and credibility in the information disseminated by genetic improvement programmes. The “Shrimp Consortium”<sup>139</sup> composed of international development and conservation groups and donor institutions would serve as an excellent example of how such partnerships could work in the promotion of genetic improvement programmes.

While much has recently been made of the role of aquaculture in “filling the supply gap” arising from limited production from capture fisheries, aquaculture is only one solution to this problem and genetic improvement programmes can help. Conflicts between aquaculture and capture fisheries based on competition and access to resources have developed and could pose a threat to both sectors. Efforts should be made to conserve and protect wild fishery resources (Chapter 9), to help in promoting partnerships and to avoid conflicts.

It should be recognized that there are areas where aquaculture is not appropriate, regardless of whether or not genetic technologies are used. It is best not to spend time fighting these battles that may alienate partners and

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<sup>139</sup> <http://www.worldwildlife.org/cci/dialogues/shrimp.cfm>

result in failed aquaculture operations. The twinning strategies of Chapter 9 and the designation of areas where aquaculture is limited or excluded should be loudly embraced by the aquaculture sector so that other areas more appropriate for aquaculture can be developed fully, using the best species and strains available.

### ***12.2.3 Learn from other sectors***

The terrestrial farming sectors are more advanced than aquaculture in the use of genetic technologies and there are good lessons to be learned from them. Some lessons include the following:

First, it should be stressed that the benefits of genetically improved fish will be passed onto the consumer. The plant biotechnology sector is experiencing strong consumer resistance to the use of genetically modified organisms, whereas the pharmaceutical sector routinely uses modern genetic engineering with little public resistance. One reason for this is that the public perceives the benefits of genetic engineering in plants to benefit only the industry, whereas the use of genetic biotechnologies by the pharmaceutical company is perceived to benefit sick people.

Second, ethical issues matter. Consumer concerns have been expressed for the welfare of genetically engineered livestock and for general growing conditions of farmed animals. Similar concerns have arisen to a limited extent for farmed and genetically altered fish. Genetic alterations that may cause deformities should be avoided and it should be emphasized how genetically improved fish will have improved welfare in culture because of increased domestication. Food security issues and intellectual property protection that could deprive farmers of adequate food have arisen in the crop sector. Seeds for crops essential for rural communities were genetically sterilized so that farmers could not replant them. Advocates of genetic improvement programmes should be aware of how genetic improvements may impact food security of rural communities.

Finally, labelling is a controversial issue with which all sectors are dealing. Guidelines on eco-labelling fishery have been produced by FAO and partners and guidelines on aquaculture products are under development; the Marine Stewardship Council and the Forest Stewardship Council have developed private industry guidelines. These existing guidelines do not address genetic criteria yet. Some inter governmental fora have mandated labelling of certain terrestrial products from modern biotechnology (e.g. GMOs) and some

organic labeling schemes do not allow certain genetic technologies. In light of the sensitive and complicated nature of this field, discussions on how to use genetic information in these guidelines is not yet at a sufficiently advanced level where guidance can be given at this time. It is recommended that genetic resource managers and proponents of genetic technologies in aquaculture follow this rapidly advancing field and engage partners as recommended above to help develop an informed way forward.

#### ***12.2.4 Use accurate terminology consistent with national and international legislation***

The field of genetics is complicated and often controversial. Accurate terminology and correct use of terms and principles will help in communicating useful and accurate information and in avoiding problems associated with misunderstanding (see Box “some terminology” in Chapter 2). Glossaries exist to help understand this complicated arena.<sup>140</sup>

### **12.3 Conclusion**

The benefits of genetic management programmes in aquaculture are substantial, but often poorly understood by the general public and policy-makers. Communicators (see footnote 138) state that new ideas are first embraced by a small number of “innovators”; then slowly by others. When 15 percent of a group adopt the idea, it can successfully spread. Promoters of genetic technologies and breeding programmes need to communicate the positive aspects of these programmes to a wide audience and seek partnerships with other users of aquatic resources and civil society to help reach this 15 percent level of acceptance. The responsible use of genetic technologies can help aquaculture produce more food more efficiently and with less environmental impact. Once this is realized by a large audience it will help aquaculture integrate into multisectoral local community development plans. These facts should be part of an overall communication strategy that helps build public relations and consumer confidence in genetically improved fish.

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<sup>140</sup> FAO glossaries exist on biotechnology ([www.fao.org/biotech/index\\_glossary.asp](http://www.fao.org/biotech/index_glossary.asp)); fisheries ([www.fao.org/fi/glossary/default.asp](http://www.fao.org/fi/glossary/default.asp)); and aquaculture ([www.fao.org/fi/glossary/default.asp](http://www.fao.org/fi/glossary/default.asp)).



## ANNEX 1

### NAIROBI DECLARATION<sup>1</sup>

#### CONSERVATION OF AQUATIC BIODIVERSITY AND USE OF GENETICALLY IMPROVED AND ALIEN SPECIES FOR AQUACULTURE IN AFRICA

##### BACKGROUND

Fish are a critical source of animal protein to the people of Africa, and aquatic resources play a central role in sustaining rural and urban livelihoods across much of the region. Yet, for the continent as a whole, per capita supply of fish is declining and current projections of supply and demand indicate that this gap will continue to grow in the coming decades. If this gap is to be bridged, capture fisheries need to be sustained and the potential of aquaculture realised. In doing so attention needs to be given to protecting the rich aquatic biodiversity of Africa, especially the rich diversity of freshwater fish and its role in sustaining capture fisheries and providing species for aquaculture.

At present, fish production from aquaculture in Africa is low. However as population increases, together with demand for fish, the aquaculture sector is projected to grow. For this to happen, a wide range of constraints need to be addressed and a greater range of management practices considered. Pond and broodstock management will need to be improved, a wider range of feeds developed, and market access improved.

In addition, there is considerable potential for improving performance of the fish species and strains used. At present many of the fish used in aquaculture in Africa are derived from undomesticated stocks. This contrasts with crops, livestock and poultry where large increases in production have been achieved through application of breeding programs and other genetic improvement procedures. However, while improved strains and introduced species have potential to increase production there is clear risk of escape into the wild, and possible negative impacts on biodiversity. If the full potential for sustainable aquaculture in Africa is to be realised these concerns need to be addressed.

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<sup>1</sup> Gupta, M.V., Bartley, D.M., Acosta, B.O. (eds) 2004. Use of Genetically Improved and Alien Species for Aquaculture and Conservation of Aquatic Biodiversity in Africa. The WorldFish Conference Proceedings No. 68. Declaration available at [www.cta.int/pubs/nairobi/declaration.pdf](http://www.cta.int/pubs/nairobi/declaration.pdf)

## **RECOMMENDATIONS**

### **1. Quality seed**

Given that aquaculture from small-scale, low-input systems to large-scale intensive systems can achieve potential benefits from genetic enhancement, quality seed should be made available and used in conjunction with proper broodstock and farm management.

### **2. Genetics in broodstock management**

Since genetic resources in cultured populations can be degraded as a result of captive breeding, genetic aspects of broodstock management need to be a basic element within all aquaculture and stock enhancement programmes.

### **3. Responsible introductions**

Introductions of fish, including genetically improved strains and alien species, may have a role in the development of aquaculture. Any movement of fish between natural ecological boundaries (e.g. watersheds) may involve risk to biodiversity and there is need for refinement and wider application of protocols, risk assessment methods, and monitoring programs for introductions of fish, including genetically improved strains and alien species. States have an important responsibility in the development and implementation of such protocols and associated regulations, the establishment of clear roles and responsibilities, and capacity building. Such efforts should be linked to obligations pursuant to the Code of Conduct for Responsible Fisheries, the Convention on Biological Diversity and other relevant international agreements.

### **4. Conserving wild stocks**

Unique wild stocks of important tilapia species still exist in many parts of Africa. Priority areas should be identified and managed as conservation areas in which introductions of alien species and genetically improved strains should be prevented.

### **5. Transboundary problems in fish transfer**

The majority of issues and problems associated with movement of fish and the use of genetically improved strains are common to most African countries. Countries are encouraged to: (a) look beyond borders for examples of workable policies and legislation, adopt them where appropriate to fill national policy gaps and harmonize them where necessary; and (b) use existing regional bodies or form new bodies to assist in coordinating management activities taking into account ecological realities, in particular transboundary watersheds.

## **6. Strengthening access to information**

Baseline information on fish genetic diversity, environmental integrity and aquaculture practices exists, but it is neither comprehensive nor easily accessible. The existing mechanisms for collection and dissemination of information need to be strengthened.

## **7. Controlling pathogen movement**

Internationally accepted codes and protocols for reducing the risk of transboundary movement of pathogens (the term pathogen used here includes parasites) through movement of fish including alien species do exist, but they do not address any specific needs regarding genetically improved species. States and other relevant bodies should evaluate the existing codes and protocols for reducing the risk of transboundary movement of pathogens through movement of fish including alien species and genetically improved strains, and adapt them for African conditions.

## **8. Raising awareness of risks of fish introductions**

Policy-makers, enforcement agencies, stakeholders and the general public need to be made aware of issues related to, and the need for, policy on the movement of alien species and genetically altered species, and this should be high on national agenda.

## **9. Engaging stakeholders**

Some policies relevant to movement of fish seem difficult to implement, are unknown to users, create conflicts of interest, or are viewed as restrictive, in part because they have been developed with limited consultation and participation. Formulation of policy and legislation concerning fish movement should seek to engage all stakeholders in a participatory process. In addition, governments should establish advisory groups with links to independent and scientifically competent expert bodies such as FAO, IUCN and ICLARM (now the WorldFish Center).

## **10. Liability for adverse environmental impacts**

Although economic benefits can be derived through the use of alien and/or genetically improved species in aquaculture, in many cases, those to whom benefits accrue do not bear the costs associated with adverse environmental impacts. In view of this, there should be provision for liability, compliance (e.g. incentives) and restoration within policies and legislation concerning the movement and use of alien and genetically improved fish species in aquaculture.



These technical guidelines have been developed to support sections of FAO's Code of Conduct for Responsible Fisheries on aspects of genetic resource management in aquaculture. Guidance is provided on broodstock management and domestication, genetic improvement programmes, dissemination programmes for genetically improved fish, economic considerations in genetic improvement programmes, risk assessment and monitoring, culture-based fisheries, conservation of fish genetic resources, gene banks, a precautionary approach and public relations. The effective management of genetic resources, risk assessment and monitoring can help promote responsible aquaculture by increasing production output and efficiency, and help minimize adverse impacts on the environment. The benefits of the responsible application of genetic principles to aquaculture should be communicated to consumers, policy-makers, scientists and others interested in responsible fisheries and aquaculture.

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