



INLAND MYSAP

INLAND Myanmar Sustainable Aquaculture Programme



Management of inbreeding in carp hatcheries in Myanmar

Implemented by:



RESEARCH PROGRAM ON Fish Led by WorldFish



Management of inbreeding in carp hatcheries in Myanmar

Matthew Hamilton

January 2019

Contents

Introduction	3
Current broodstock management in carp hatcheries	3
Key issues	3
Broodstock management and replacement	3
DOF Rohu Genetic Improvement Program	4
Recommendations.....	4
Short term	4
Long term.....	5
Acknowledgements	5
References	5
Appendix 1 Hatcheries visited in 2018	6
Appendix 2. General principles regarding inbreeding control and genetic improvement	7
Introduction	7
Genetic theory	7
Inbreeding control.....	8
Approach 1. Minimise average relatedness in a single strain.....	8
Approach 1.1. Routinely obtain broodstock from wild populations	8
Approach 1.2. Maintain a single strain (i.e. a single closed population)	11
Approach 1.3: Routinely obtain broodstock from a single genetically improved strain in which average relatedness is controlled.....	13
Approach 2: Cross unrelated strains.....	14
Approach 2.1 Cross a hatchery strain with an external unrelated strain	14
Approach 2.2 Maintain multiple unrelated mass selected strains	14
Approach 2.3 Routinely obtain broodstock from two unrelated genetically improved strains	19
Pros and cons of different approaches to inbreeding control	20
Broodstock management for restocking.....	24
Implementation and management of mass-selection programs	24
References.....	28
Appendix 3. Naming strains and documenting their ancestry	31
What is a strain?.....	31





EUROPEAN UNION



Strain details..... 31

Within strain details 32

Naming strains 32

Documenting strain ancestry 32

References..... 33



Introduction

Rohu is the most important aquaculture species in Myanmar and most rohu seed in the country is sourced from hatcheries (FAO 2010). Accordingly, the production of 'quality seed' in hatcheries is integral to the productivity of aquaculture in Myanmar.

The quality of seed produced by a hatchery is influenced by the genetics of the seed and the environment (i.e. husbandry) experienced by the seed. Only the first of these factors is addressed here. With the overarching objective of improving the genetic quality of rohu seed for aquaculture in Myanmar, this report aims to:

- report on current broodstock management in carp hatcheries;
- identify key issues with respect to inbreeding and genetic improvement;
- identify knowledge gaps and constraints with respect to the management of inbreeding and genetic improvement;
- recommend changes to practices; and
- identify research needs.

Current broodstock management in carp hatcheries

To examine current broodstock management in carp hatcheries, Matthew Hamilton visited five Department of Fisheries (DOF) and five private hatcheries across the Sagaing Region, Shan State, Mandalay Region and Yangon region between the 27th of August and the 1st of September (Appendix 1). At these hatcheries, a number of approaches aimed at minimising inbreeding and/or genetically improving rohu in Myanmar were observed. If these, or other, desirable practices can be widely implemented, there is the potential for substantial improvement in the genetic quality of seed produced in Myanmar. Observed desirable practices included:

- replacement of old broodstock with new unrelated broodstock from rivers;
- between-strain crossing of unrelated strains for the production of non-inbred seed for grow out; and
- genetic improvement using a mass selection approach.

Key issues

Broodstock management and replacement

In spite of the aforementioned examples of desirable practices, much of the hatchery-produced seed in Myanmar likely suffers from moderate to extreme levels of inbreeding due to inappropriate broodstock management (see 'Genetic Theory' in Appendix 2). Examples of inappropriate broodstock management practices currently implemented in rohu hatcheries in Myanmar include:

- the establishment of closed hatchery populations from a small number of founders; and
- the replacement of broodstock over multiple generations from spawning events involving a small number of parents.

DOF Rohu Genetic Improvement Program

Under a 'DOF Rohu Genetic Improvement Program' initiated in 2009, two strains were established at each of two hatcheries (Hlawgar and Kume) from a small number of founders collected from different river systems. Under the program, it was intended that all DOF hatcheries be supplied with broodstock from unrelated strains. Supplied hatcheries were then expected to undertake between-strain crosses to produce non-inbred seed for commercial grow out. However, the program has been discontinued and there is no organisation-wide approach to replace aging broodstock supplied under the program. If DOF hatcheries replace their aging broodstock without maintaining the purity of the original DOF strains (see Approach 2.2 of Appendix 2), a substantial increase in the level of inbreeding in seed produced by these hatcheries is likely in coming years.

Recommendations

Short term

1. Hatchery managers and policy makers should be trained to consider and understand the genetic principles underpinning:
 - a. inbreeding depression and genetic improvement, in the context of seed production for aquaculture (see 'Genetic theory' and 'Inbreeding control' in Appendix 2); and
 - b. genetic management of fish stocks in natural water bodies, in the context of restocking programs (see 'Broodstock management for restocking' in Appendix 2).
2. Hatchery managers and policy makers should be encouraged to think in terms of strains and to record strain origins (see Appendix 3).
3. Assuming biosecurity risks can be addressed, hatcheries producing seed for aquaculture should either:
 - a. obtain replacement broodstock from wild populations (see Approach 1.1 in Appendix 2); or
 - b. exchange broodstock with another hatcheries and cross unrelated strains to produce seed for grow out (see Approach 2.1 in Appendix 2).
4. Hatcheries producing seed for restocking should be encouraged to follow the principles outlined in 'Broodstock management for restocking' in Appendix 2.
5. Key hatcheries producing seed for aquaculture should be supported to establish and maintain multiple unrelated mass selected strains (see Approach 2.2 in Appendix 2), as:
 - a. a proof of concept;
 - b. a source of genetically improved stains of broodstock to other hatcheries; and
 - c. training sites
6. Low-cost and easily implemented approaches to externally tagging or marking fish according to strain should be identified (see 'Tagging and marking' in Appendix 2).
7. Appropriately designed 'strain comparison' experiments should be implemented (Ponzoni et al. 2012a), in accordance with animal welfare laws and policies (WorldFish 2004), to:
 - a. Quantify the extent of inbreeding depression in hatchery-produced seed;
 - b. Quantify the extent of genetic improvement in hatchery broodstock; and

- c. Provide a baseline from which to assess future interventions aimed at reducing inbreeding and the genetic improvement of Rohu in Myanmar.

Long term

8. All hatcheries should implement one of the following approaches:
 - a. maintain multiple unrelated mass selected strains (see Approach 2.2 in Appendix 2);
 - b. routinely obtain broodstock from two unrelated genetically improved strains (see Approach 2.3 in Appendix 2); or
 - c. routinely obtain broodstock from a single genetically improved strain in which average relatedness is controlled (see Approach 1.3 in Appendix 2).

Acknowledgements

Production of this report was financially supported by INLAND MYSAP, which is funded by the European Union (EU) and the German Federal Ministry for Economic Cooperation and Development (BMZ) and implemented by WorldFish. I thank Mr Don Griffiths (INLAND MYSAP, WorldFish, Myanmar), Daw (Ms) Aye Aye Lwin (MYCulture, WorldFish, Myanmar) and Mr Lennart Klein (GIZ, Myanmar) for organising hatchery visits. I thank Dr Khin Maung Soe (WorldFish, Myanmar), Daw (Ms) Aye Aye Lwin (MYCulture, WorldFish, Myanmar), Ms Han Nway Htwe (GIZ, Myanmar) and Ms Nann Ohu Khan (INLAND MYSAP, WorldFish, Myanmar) for translating. I thank hatchery managers and staff who generously shared their knowledge when interviewed, including U Win Kyaing, Ms Mie Mie Kaing (DOF), Mr Tun Shwe, Mr Hla Kyaw, Ms Yi Yi Lwin, Mr Zaw Win Aung (DOF), Mr. Khim Maung Myo (DOF), Ms. Thandar Khaing (DOF), Ye Htun Naing (DOF). I also thank Dr Khin Maung Soe (WorldFish, Myanmar) Daw (Ms) Aye Aye Lwin (MYCulture, WorldFish, Myanmar) U Thein Lwin (Hatchery Manager) Dr Myint Swe (MFF) Mr Than Lwin (DOF, Nay Pyi Taw) Mr Lennart Klein (GIZ) Mr Myint Aung (Hatchery Owner, MFF) Mr Thet Tung Aung (Hatchery Manager, MFF) Mr Tin Aung (MFF) Mr Tin Tun (MFF) Mr Aye Naing (DOF) for sharing their knowledge of aquaculture and hatchery management in Myanmar. Furthermore, I thank Mr Don Griffiths, Dr Trong Quoc Trinh (WorldFish) and Professor John Benzie (WorldFish) for their comments on drafts of this report.

References

- Fao (2010). "Fishery and Aquaculture Country Profiles. Myanmar", in: Country Profile Fact Sheets. (Rome: Food and Agriculture Organisation. Fisheries and Aquaculture Department.).
- Ponzoni, R.W., James, J.W., Nguyen, N.H., Mekkawy, W., and Khaw, H.L. (2012). "Strain comparisons in aquaculture species: a manual". (Penang, Malaysia: WorldFish).
- Worldfish (2004). "Animal care, welfare and ethics policy of WorldFish center". (Penang, Malaysia: WorldFish).

Appendix 1 Hatcheries visited in 2018

Name	Ayeyarwady	Nyaung Shwe	U Tun Shwe	U Hla Kyaw	Ms Yi Yi Lwin	Shewbo MFF	Shwebo DOF	Thayet Kone DOF	Nad Yay Kan DOF	Hlawgar DOF
Region	Yangon	Shan	Shan	Shan	Shan	Sagaing	Sagaing	Mandalay	Mandalay	Yangon
Latitude	16.951719	20.685969	20.694167	20.685000	20.743611	22.555833	22.577500	21.990000	21.880560	16.969444
Longitude	96.476077	96.931752	96.938333	96.933611	96.930833	95.699722	95.663333	96.124170	96.091940	96.111111
Features	Formal mass selection program					Mass selection applied	Maintains two strains to avoid inbreeding			Replacing elderly DOF genetic improvement program broodstock with wild caught stock
Date	27-Aug-18	28-Aug-18	28-Aug-18	29-Aug-18	29-Aug-18	30-Aug-18	30-Aug-18	31-Aug-18	31-Aug-18	01-Sep-18
Ownership	Private	Public (DOF)	Private	Private	Private	Private	Public (DOF)	Public (DOF)	Public (DOF)	Public (DOF)
Species other than rohu		Common, Inle, grass, silver barb, catla, silver carp	Common, grass, silver barb	Grass carp, common carp, silver carp, catla, silver barb	Common, grass, plan to collect Inle	GIFT, pacu, pangasius, others		10 others	Silver carp	18 others

Appendix 2. General principles regarding inbreeding control and genetic improvement

Introduction

This Appendix details the pros and cons of a number of approaches that aquaculture hatcheries can adopt to control inbreeding and/or genetically improve the seed they produce for grow out. A brief introduction to the genetic theory underpinning these approaches is provided.

The focus of this report is on the management of broodstock in hatcheries for the production of seed for aquaculture. However, the management of broodstock in the context of seed production for the restocking of water bodies is briefly addressed.

It is evident that the genetic quality of seed is only one factor affecting the quality of seed produced by hatcheries – the quality of seed can also be impacted by environmental and husbandry (i.e. management) factors, such as suboptimal seed rearing and handling practices. It is also evident that hatcheries have a major role in avoiding and controlling the spread of disease through the adoption of appropriate biosecurity practices (Mohamed Din and Subasinghe 2017). Although not the focus of this report, biosecurity and the disease risks posed by the movement of fish among rivers, hatcheries and farms must be managed and kept front-of-mind when considering approaches to inbreeding control and genetic improvement.

Note that the term ‘fish’ has been used throughout this report to encompass all aquaculture species, including non-vertebrates. Indeed, many of the concepts outlined are also applicable to non-aquaculture species, particularly those that are highly-fecund and in the early stages of domestication.

Genetic theory

Hatchery managers should aim to maximise the **genetic quality** of seed produced for grow out. Although the genetic concepts driving the genetic quality of seed are complex, from the perspective of a hatchery manager primary consideration should be given to i) maximising the average additive genetic value (i.e. average **breeding value**) of the seed produced, and ii) minimising the deleterious impacts of inbreeding, a component of the non-additive genetic value of the seed. The **additive genetic value** of an individual is the component of its **total genetic value** (i.e. genetic quality) that is transmitted from one generation to the next, and the **non-additive genetic value** is that which is not transmitted across generations (assuming the species is diploid and ignoring epistasis).

Inbreeding results from the mating of related parents. The level of inbreeding in an individual, measured by **Wright’s coefficient of inbreeding** (F ; ranging from 0 to 1), is directly proportional to the extent of **additive genetic relatedness** between its parents. In mathematical terms, F of an individual equals half the **coefficient of relationship** between its parents. As an example, if two full siblings (i.e. individuals with the same two parents) are mated, their progeny are highly inbred ($F \geq 1/4$); and if two first cousins are mated, their progeny are less inbred ($F \geq 1/16$). However, if two unrelated parents are mated, their progeny are not inbred ($F = 0$), no matter how inbred the parents themselves are.

The control of inbreeding is important because inbreeding can result in **inbreeding depression**. Inbreeding depression can be manifested as poor growth, poor survival, poor reproductive performance, disease susceptibility and/or morphological deformities (Gjerde et al. 1983; Komen et al. 1992; Evans et al. 2004).

A **closed population** – here considered synonymous with a **strain** – is a population descended from a finite number of founder individuals into which no subsequent introduction of individuals or genes has occurred. Refer to Appendix 3 for guidelines on how to name strains and document their ancestry.

Genetic improvement is here described as the process of making cumulative desirable changes to the average breeding value of a strain, for one or more characteristics. This is achieved by selecting individuals to be mated (i.e. parents) from each generation that are believed to have high breeding values, based on their measured characteristics and/or the characteristics of their relatives.

Genetic improvement requires the presence of **additive genetic variation** (i.e. **additive genetic diversity**) in the population for the characteristic/s under selection. Accordingly, there is an inherent conflict between maximising short-term genetic improvement versus maximising long-term genetic improvement and controlling inbreeding. For example, to maximise short-term genetic improvement, one would select the very best male and very best female as parents to produce one full-sibling family as the next generation. Under this scenario, i) the level of additive genetic diversity in the next generation will be less than if multiple parents had been used to produce multiple families and ii) the progeny of subsequent crosses between full-siblings will be highly inbred.

One means of maintaining additive genetic diversity and minimising future inbreeding in a strain is to control the increase in **average relatedness** among individuals (Meuwissen 1997; Meuwissen and Sonesson 1998). In a strain, average relatedness among individuals increases with each generation. This is unavoidable. However, a number of management strategies can be implemented to minimise the rate of increase in average relatedness. A number of these approaches are detailed below.

Inbreeding control

Methods to control inbreeding in hatcheries producing seed for aquaculture can be classified into two general approaches:

- Approach 1: Minimise average relatedness in a single strain
- Approach 2: Cross unrelated strains

Within these two broad approaches, a number of different strategies can be adopted. Some of these result in the genetic improvement of strains, other do not.

Approach 1. Minimise average relatedness in a single strain

Approach 1.1. Routinely obtain broodstock from wild populations

This approach requires old broodstock to be replaced with individuals sourced from wild (indigenous or introduced) populations. Two means of implementing this approach are evident; i) maintain a single-aged broodstock population, all of which are replaced once



EUROPEAN UNION



animals have reached the end of their useful life as broodstock (Figure A2.1), or ii) maintain a mixed-age population, a proportion of which is replaced each year. However, if the second of these options is adopted and age classes are not distinguished in some way (e.g. with tags or mark, or maintained in separate ponds), there is a risk that old, but small and slow growing individuals, will be retained when they should be replaced, resulting in 'negative selection' (Hussain and Mazid 2005).



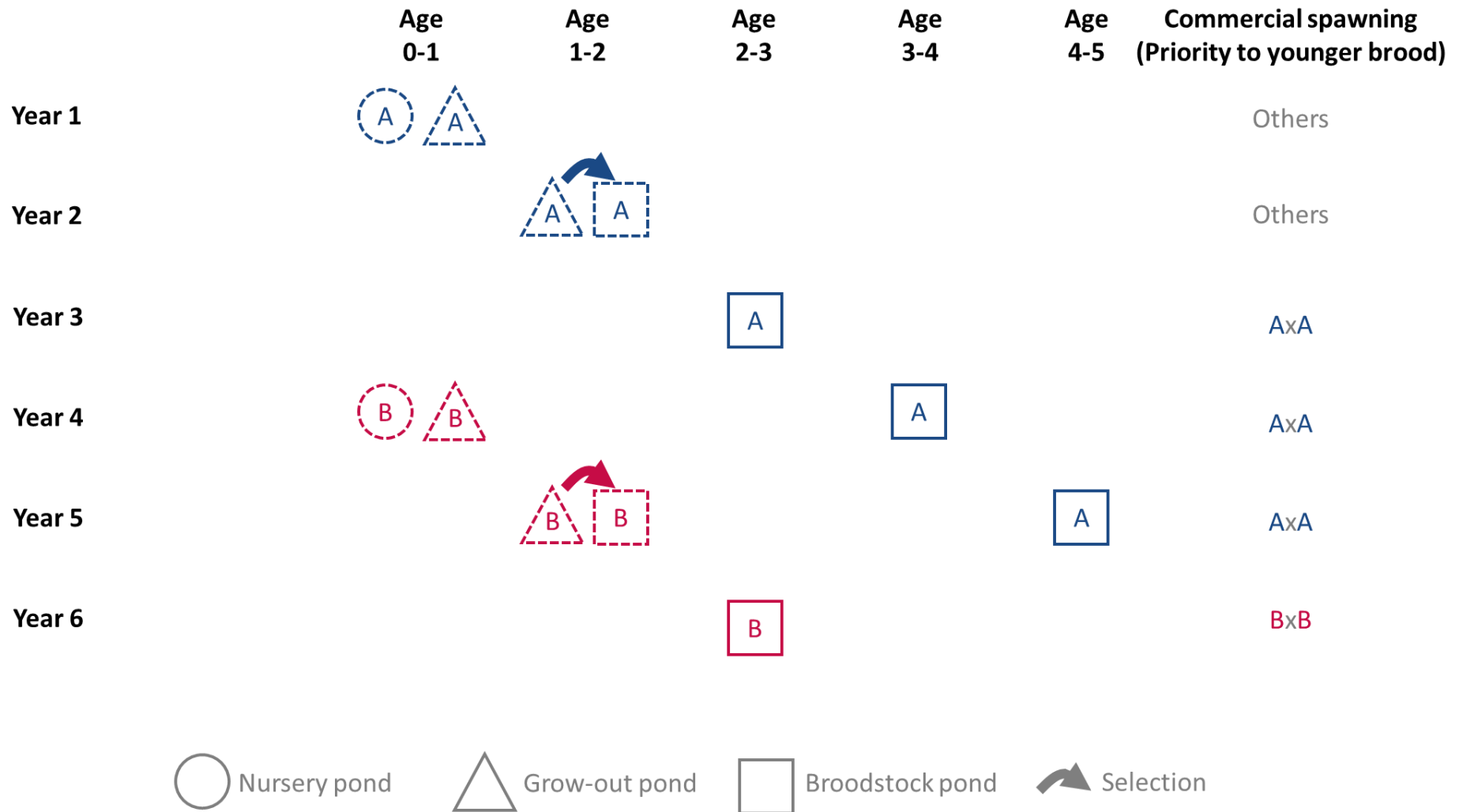


Figure A2.1. Management of broodstock to avoid inbreeding by regularly sourcing fish from wild populations or from a single genetically improved strain in which average relatedness is controlled. Letters refer to strains. Shapes with dashed outlines are not necessary in all circumstances. Note that in species that do not adhere to an annual breeding cycle, the age of animals is best expressed in terms of intervals between spawn runs, rather than years.

To maximise the probability that broodstock obtained from wild populations are unrelated, replacement broodstock should be i) obtained from large water bodies with large populations; and ii) if collected as spawn or fry, obtained at the peak of the spawning season from areas in which the species is prevalent (Hamilton et al. submitted-a; Hamilton et al. submitted-b). Refer to Table 1 for a list of pros and cons for this and other approaches.

Approach 1.2. Maintain a single strain (i.e. a single closed population)

Approach 1.2.1. Family-based breeding and genomic selection

Family-based breeding programs track the ancestry (i.e. pedigree) of individuals in a strain. This is achieved by maintaining families in separate vessels (e.g. tanks or hapas) after spawning until they reach a size that allows individuals to be tagged and families to be pooled, or with parentage assignment using molecular tools. Cost-reducing techniques, such as walkback selection – whereby only phenotypically superior individuals have their parentage assigned – can also be employed (Sonesson 2005). Inbreeding is controlled in family-based breeding programs i) in the short term, by undertaking crosses between unrelated, or distantly related, individuals; and ii) in the long term, by selecting parents (Figure A2.2) in a manner that minimises the average relatedness among individuals in subsequent generations (Meuwissen 1997; Meuwissen and Sonesson 1998). Family-based breeding programs also enable accurate estimation of each individual's breeding value, by utilising measurement data from not only the individual itself but also its relatives. The more accurate estimated breeding values (EBVs) are, the more accurately the best individuals for mating (i.e. parents) can be selected in each generation and the more rapidly genetic improvement can be achieved in a strain. Furthermore, measurement data from relatives can be used to estimate breeding values for traits that cannot be directly measured on candidate parents (e.g. disease resistance). Genetic parameters, such as heritabilities and genetic correlations (Falconer and Mackay 1996) can also be estimated using data from family based breeding programs. Among other things, genetic parameters are required to predict the extent to which genetic improvement can be achieved in a characteristic. For example, if the heritability of a characteristic is zero, it is not possible to increase the average breeding value of a strain for that characteristic.

Genomic selection combines molecular marker data (generally for tens of thousands of individual single nucleotide polymorphisms; SNPs) with phenotypic (i.e. measurement) data to more accurately identify superior individuals for selection as parents. It is generally implemented within family-based breeding programs and represents an additional level of expense and complexity, albeit one that is increasingly adopted for traits that are expensive or difficult to measure, or cannot be measured on candidate parents (e.g. disease resistance and tolerance) (Banger et al. 2017).

Approach 1.2.2. Maintain a single mass selected strain

Approaches to mass selection with one or multiple strains are described in the 'Implementation and management of mass-selection programs' section below. Also refer to Figure A2.2.

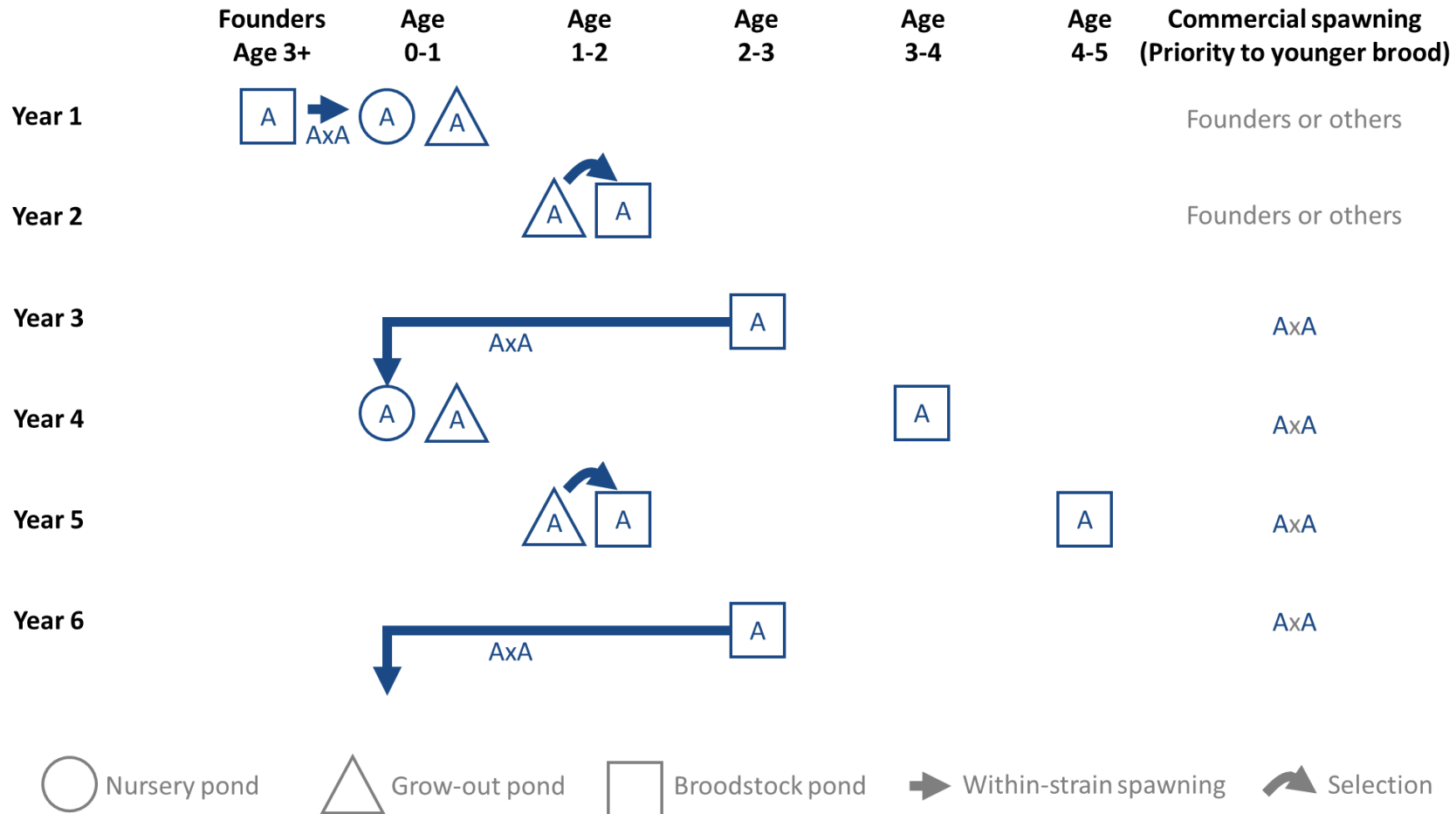


Figure A2.2. A single-strain (A) approach to genetic improvement using family-based or mass selection, suitable for species that reach sexual maturity at age three years. Note that in species that do not adhere to an annual breeding cycle, the age of animals is best expressed in terms of intervals between spawn runs, rather than years.

Approach 1.2.3: Rotational mating among cohorts

Rotational mating among cohorts involves i) the establishment of multiple, generally four or eight, founder cohorts of fish and ii) the systematic transfer of males between cohorts at the time of spawning (Figure A2.3). After each spawning, the progeny cohorts are given the same identifier as the cohort's mothers (e.g. 1 to 8, Figure A2.3).

Rotational mating among cohorts results in considerably less inbreeding than single-strain mass selection (i.e. Approach 1.2.2) (Nomura and Yonezawa 1996; Ponzoni et al. 2012b). If implemented appropriately, it can also achieve ongoing genetic improvement of a population, as it essentially represents a modified form of mass selection (see the 'Implementation and management of mass-selection programs' section below).

Ponzoni et al (2012b) outlined a protocol for rotation mating among cohorts for tilapia, which has been implemented in a number of countries. This approach could be modified for application in other species. Refer to Ponzoni et al (2012b) and the modified eight-cohort mating approach in Figure A2.3 for details.

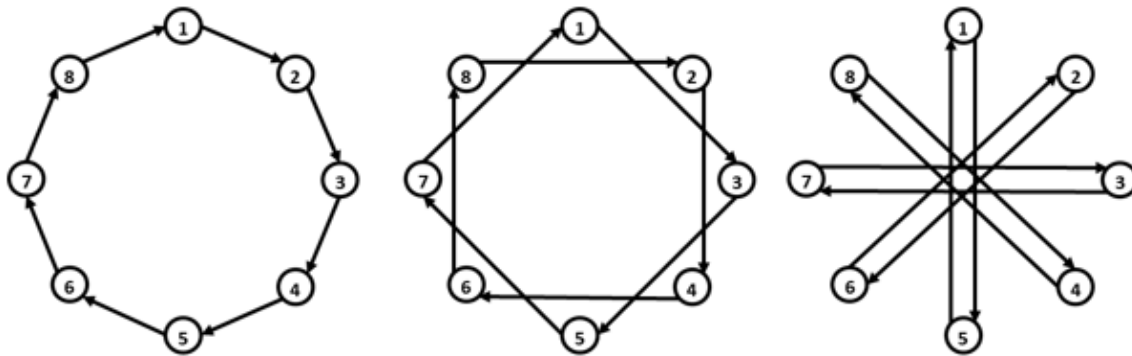


Figure A2.3. Movement of males between cohorts (indicated by arrows) under the eight-cohort rotational mating system. The three cycles depicted should be repeated in sequence indefinitely. This three-cycle approach controls inbreeding more effectively than the two-cycle approach detailed in Ponzoni et al (2012b).

Approach 1.3: Routinely obtain broodstock from a single genetically improved strain in which average relatedness is controlled

This approach is implemented in the same fashion as Approach 1.1 (Figure A2.1) except that replacement broodstock are sourced from a genetically improved strain, in which average relatedness is controlled, instead of genetically unimproved wild populations.

Approach 2: Cross unrelated strains

Approach 2.1 Cross a hatchery strain with an external unrelated strain

Many hatcheries maintain their own strains. Assuming that the largest and healthiest animals have been selected as the parents of broodstock over multiple generations (i.e. negative selection is not an issue, Hussain and Mazid 2005), it is possible that some degree of genetic improvement has occurred in such strains. However, if active measures to control average relatedness have not been implemented, such populations maintained by hatcheries over multiple generations are also likely to produce inbred progeny. In these circumstances, the issue of inbreeding in progeny produced for grow out can be overcome by crossing one inbred strain with another, possibly inbred, but unrelated strain (Evans et al. 2004). In the long term, the adoption of this approach would require a hatchery to intermittently undertake within-strain matings to maintain the 'purity' of its own strain, while routinely obtaining broodstock from an external unrelated strain to cross with and produce seed for grow out. This could involve the exchange of males between hatcheries, to the benefit of both hatcheries, or obtaining broodstock from wild populations (Figure A2.4).

Approach 2.2 Maintain multiple unrelated mass selected strains

This approach involves the genetic improvement of two or more unrelated strains with mass selection and the production of non-inbred seed for grow out by crossing between these unrelated strains. Such an approach ensures that seed supplied for grow out is not inbred, something that cannot be assured under single-strain mass selection (Approach 1.2.2). Within strains, the mating of a large number of dams and sires in each generation is desirable (see the 'Implementation and management of mass-selection programs' section below) as this practice is most likely to retain additive genetic diversity over many generations (Bentsen and Olesen 2002).

Specific approaches to maintaining multiple unrelated mass selected strains are detailed in Figures A2.5 to A2.7. These represent **discrete-generation** (Figure A2.5) and **rolling-front** (Figures A2.6 and A2.7) approaches. A rolling-front approach, once established, requires the same operations to be completed in each year with benefits including:

- smoothing of peaks and troughs in activity across years;
- better utilisation of infrastructure (i.e. nursery, grow out and broodstock ponds are used every year); and
- skill retention – all skills are practiced each year.

In the case of species that mature at 3 years of age, the rolling front approach necessitates the management of three strains if implemented in full (Figure A2.7). However, in this case, a partial rolling front approach could be implemented by removing Strain C from Figure A2.7.

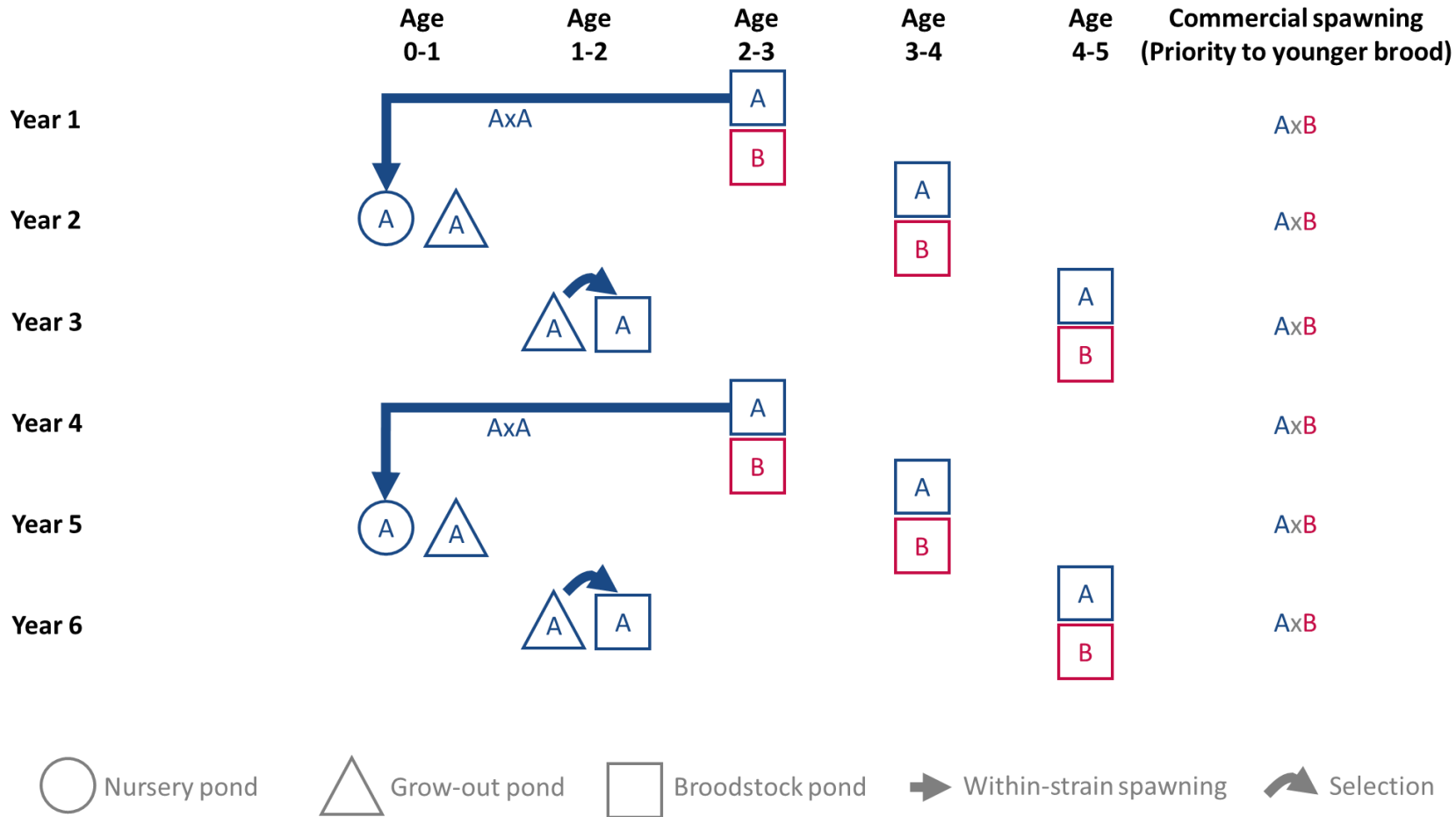


Figure A2.4. An approach to inbreeding control and genetic improvement involving mass selection of a hatcheries own strain and obtaining broodstock of an unrelated strain for commercial production, suitable for species that reach sexual maturity at age three years. Letters indicate strains. Strain A represents the hatchery’s own strain, maintained by intermittently undertaking within-strain matings. Strain B represents broodstock from an external unrelated strain. Note that in species that do not adhere to an annual breeding cycle, the age of animals is best expressed in terms of intervals between spawn runs, rather than years.

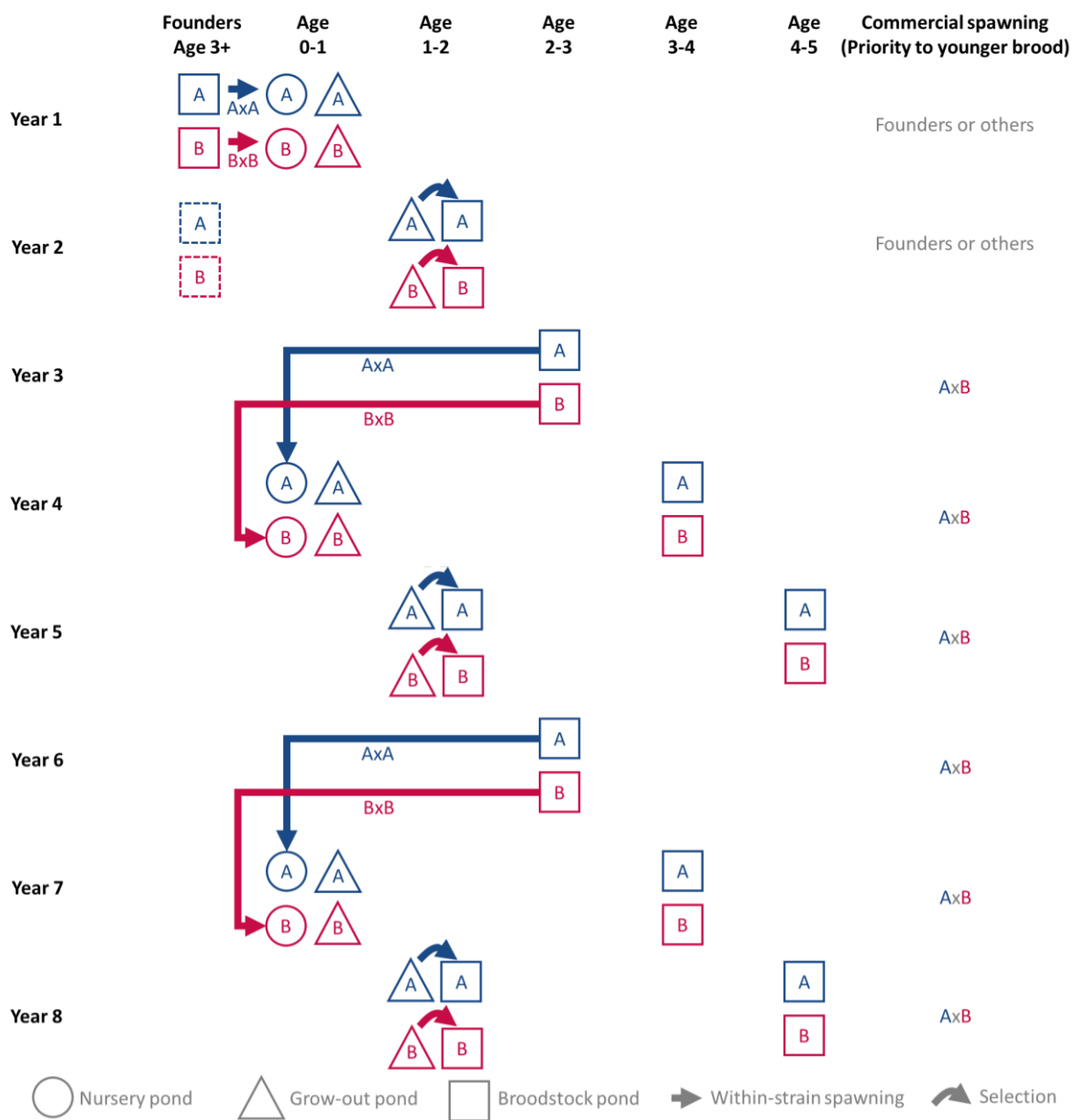


Figure A2.5. A discrete-generation two-strain mass-selection approach to inbreeding control and genetic improvement, suitable for species that reach sexual maturity at age three years. Letters indicate strains. Shapes with dashed outlines are not necessary in all circumstances. Note that in species that do not adhere to an annual breeding cycle, the age of animals is best expressed in terms of intervals between spawn runs, rather than years.

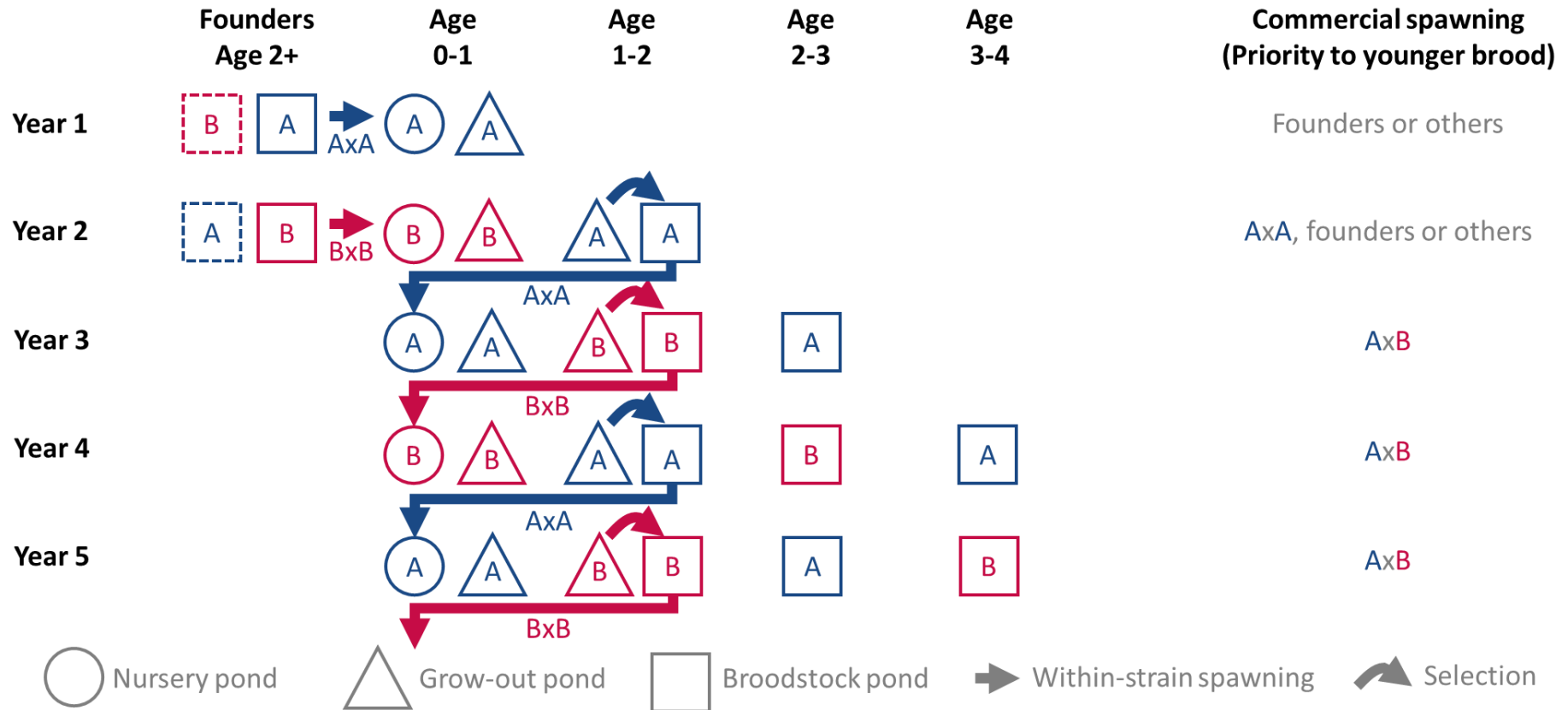


Figure A2.6. A rolling-front two-strain mass-selection approach to inbreeding control and genetic improvement, suitable for species that reach sexual maturity at age two years and in which commercial mating between males and females of different ages is practical. Letters indicate strains. There should be no genetic relationships between founders of different strains and minimal genetic relationships among founders of the same strain. Shapes with dashed outlines are not necessary in all circumstances. Note that in species that do not adhere to an annual breeding cycle, the age of animals is best expressed in terms of intervals between spawn runs, rather than years.

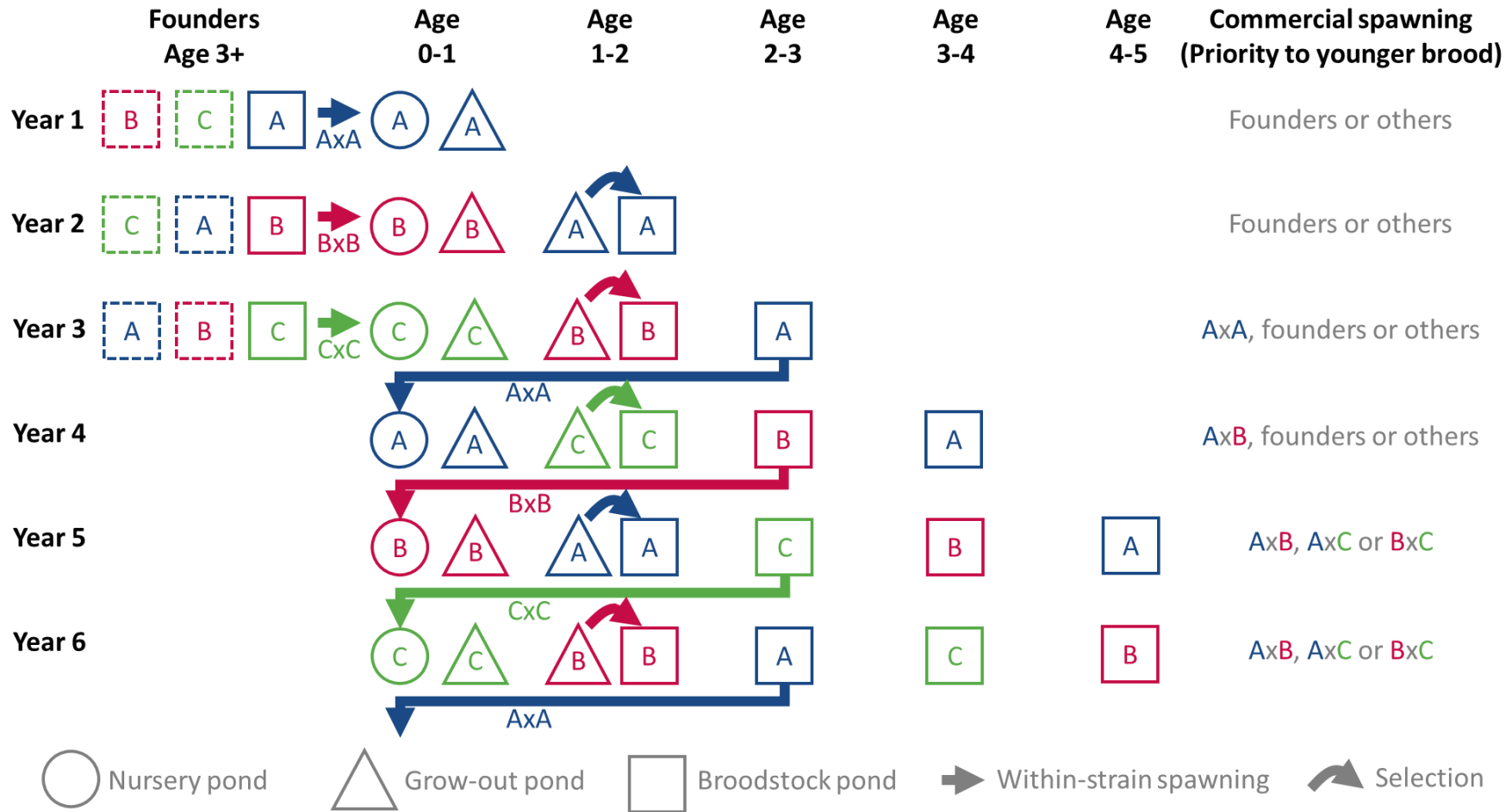


Figure A2.7. A rolling-front three-strain mass-selection approach to inbreeding control and genetic improvement, suitable for species that reach sexual maturity at age three years and in which commercial mating between males and females of different ages is practical. Letters indicate strains. There should be no genetic relationships between founders of different strains and minimal genetic relationships among founders of the same strain. Shapes with dashed outlines are not necessary in all circumstances. Note that in species that do not adhere to an annual breeding cycle, the age of animals is best expressed in terms of intervals between spawn runs, rather than years.

Approach 2.3 Routinely obtain broodstock from two unrelated genetically improved strains

This approach is similar to Approach 1.3. It depends on Approach 2.2, or equivalent, being implemented elsewhere and represents a means of multiplying improved strains for distribution to grow out farms (Figure A2.8). It is most suited to large organisations, or collaborations among organisations, with multiple hatcheries. In these circumstances, a small number of **nucleus hatcheries** implementing Approach 2.2, can distribute genetically improved broodstock to **multiplier hatcheries**. In other circumstances, nucleus hatcheries may be reluctant to supply external hatcheries with broodstock, in the absence of long-term funding (e.g. from governments or non-government organisations), a revenue stream from multiplier hatcheries, appropriate quality control practices in multiplier hatcheries to avoid reputational damage and/or intellectual property protections.

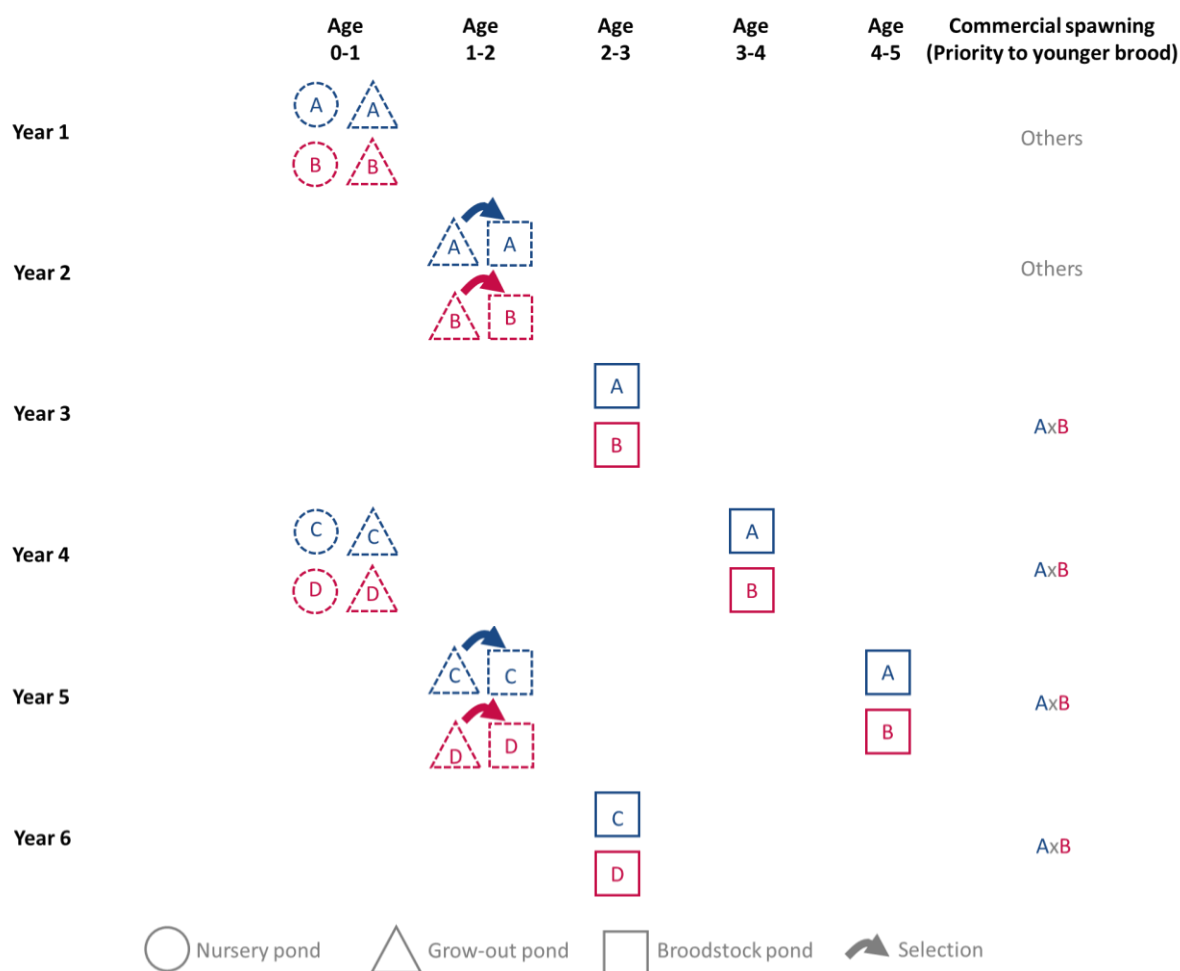


Figure A2.8. Management of two unrelated genetically improved strains routinely obtained from external hatcheries, suitable for species that become sexually mature at age 3 years. Shapes with dashed outlines are not necessary in all circumstances. Note that in species that do not adhere to an annual breeding cycle, the age of animals is best expressed in terms of intervals between spawn runs, rather than years.

Pros and cons of different approaches to inbreeding control

Table 1. Pros and cons of different approaches available to hatcheries to control inbreeding and genetically improve seed for aquaculture.

Approach	Pros	Cons	Conclusion
1.1. Routinely obtain broodstock from wild populations	<ul style="list-style-type: none"> Low cost Technically simple Controls average relatedness and inbreeding Does not rely on partnerships or collaborations Does not require the use of tags or marks 	<ul style="list-style-type: none"> Does not allow genetic improvement Does not allow estimation of genetic parameters May not be suitable for introduced species Requires ongoing access to wild-caught fish Biosecurity risk from external broodstock 	Recommended if approaches allowing genetic improvement are not feasible and biosecurity risks are managed
1.2.1. Family-based breeding and genomic selection	<ul style="list-style-type: none"> Controls average relatedness and inbreeding Allows genetic improvement Allows use of genetic relationships to optimise genetic improvement Allows estimation of genetic parameters Does not rely on partnerships or collaborations Suitable for introduced and indigenous species Does not require ongoing access to wild-caught fish No biosecurity risk from external broodstock 	<ul style="list-style-type: none"> High cost Technically complex Requires the use of tags 	Recommended only if funds and technical expertise are available over the long-term

Approach	Pros	Cons	Conclusion
1.2.2. Maintain a single mass selected strain	<p>Low cost</p> <p>Technically simple</p> <p>Allows genetic improvement</p> <p>Does not rely on partnerships or collaborations</p> <p>Suitable for introduced and indigenous species</p> <p>Does not require the use of tags</p> <p>Does not require ongoing access to wild-caught fish</p> <p>No biosecurity risk from external broodstock</p>	<p>Does not guarantee control of average relatedness or inbreeding</p> <p>Does not allow use of genetic relationships to optimise genetic improvement</p> <p>Does not allow estimation of genetic parameters</p>	<p>Does not guarantee control of inbreeding – Approach 2.2 is preferred</p>
1.2.3: Rotational mating among cohorts	<p>Low-moderate cost</p> <p>Technically simple</p> <p>Controls average relatedness and inbreeding</p> <p>Allows genetic improvement</p> <p>Does not rely on partnerships or collaborations</p> <p>Suitable for introduced and indigenous species</p> <p>Does not require the use of tags</p> <p>Does not require ongoing access to wild-caught fish</p> <p>No biosecurity risk from external broodstock</p>	<p>Does not allow use of genetic relationships to optimise genetic improvement</p> <p>Does not allow estimation of genetic parameters</p> <p>Requires large number of ponds</p> <p>Prone to failure due to human error or flooding</p>	<p>Only recommended if sufficient ponds are available, and human error and flooding issues are addressed</p>

Approach	Pros	Cons	Conclusion
1.3 Routinely obtain broodstock from a single genetically improved strain in which average relatedness is controlled	<p>Low cost</p> <p>Technically simple</p> <p>Controls average relatedness and inbreeding</p> <p>Allows genetic improvement</p> <p>Suitable for introduced and indigenous species</p> <p>Does not require the use of tags or marks</p> <p>Does not require ongoing access to wild-caught fish</p>	<p>Does not necessarily allow use of genetic relationships to optimise genetic improvement</p> <p>Does not necessarily allow estimation of genetic parameters</p> <p>Relies on partnerships or collaborations</p> <p>Biosecurity risk from external broodstock</p>	<p>Recommended if a suitable genetically improved strain is available, its long-term supply is assured and biosecurity risks are managed</p>
2.1 Exchange broodstock with another hatchery	<p>Low cost</p> <p>Technically simple</p> <p>Controls inbreeding</p> <p>Allow genetic improvement</p> <p>Suitable for introduced and indigenous species</p> <p>Does not require ongoing access to wild-caught fish</p>	<p>Does not guarantee control of average relatedness within strains</p> <p>Does not allow use of genetic relationships to optimise genetic improvement</p> <p>Does not allow estimation of genetic parameters</p> <p>Requires the use of tags or marks</p> <p>Relies on partnerships or collaborations</p> <p>Biosecurity risk from external broodstock</p>	<p>Recommended if a competent and willing partner to exchange broodstock can be found, Approaches 2.2 or 2.3 are not feasible and biosecurity risks are managed</p>

Approach	Pros	Cons	Conclusion
2.2 Maintain multiple unrelated mass selected strains	<p>Low-moderate cost</p> <p>Technically simple</p> <p>Controls inbreeding</p> <p>Allows genetic improvement</p> <p>Does not rely on partnerships or collaborations</p> <p>Suitable for introduced and indigenous species</p> <p>Does not require ongoing access to wild-caught fish</p> <p>No biosecurity risk from external broodstock</p>	<p>Does not guarantee control of average relatedness within strains</p> <p>Does not allow use of genetic relationships to optimise genetic improvement</p> <p>Does not allow estimation of genetic parameters</p> <p>Requires the use of tags or marks</p>	<p>Recommended if Approach 1.2.1 is not feasible</p>
2.3 Routinely obtain broodstock from two unrelated genetically improved strains	<p>Low cost</p> <p>Technically simple</p> <p>Controls inbreeding</p> <p>Allows genetic improvement</p> <p>Suitable for introduced and indigenous species</p> <p>Does not require ongoing access to wild-caught fish</p>	<p>Does not guarantee control of average relatedness within strains</p> <p>Does not allow use of genetic relationships to optimise genetic improvement</p> <p>Does not allow estimation of genetic parameters</p> <p>Requires the use of tags or marks</p> <p>Relies on partnerships or collaborations</p> <p>Biosecurity risk from external broodstock</p>	<p>Recommended if genetically improved strains are available, their long-term supply is assured and biosecurity risks are managed</p>

Broodstock management for restocking

Where restocking programs involve locally indigenous species the maintenance of genetic variation and the genetic integrity of the local population should be given consideration in the management of hatchery broodstock. Accordingly, in the case of indigenous species, broodstock management practices appropriate for aquaculture seed production may not be appropriate for the production of seed for restocking purposes (FAO 2008; Valiquette et al. 2014).

Approach 1.1 is generally the most appropriate for the restocking water bodies with locally indigenous species. Ideally, in adopting Approach 1.1, broodstock should be replaced each spawning season with a large number of sexually mature males and females (e.g. > 50 of each), sourced from the water body to be restocked. It is particularly important to frequently replace broodstock if the number of seed to be restocked each spawning season is substantive, relative to the number of naturally-spawned fish of comparable age, in the waterbody. However, if an indigenous species is threatened with extinction, or is not prevalent, in the water body to be restocked, Approach 1.1 is unlikely to be appropriate and the establishment and maintenance of a genetically 'pure' *ex situ* strain may be necessary (FAO 2008). Such a population would need to be carefully established, to ensure the genetic integrity and diversity of the founders, and managed to maintain genetic diversity and limit inbreeding. Approaches 1.2.1 or 1.2.3, with random 'selection' of broodstock, could be adopted as broodstock management strategies to achieve these goals.

Where restocking programs involve non-indigenous species, the maintenance of genetic integrity and diversity are of lesser importance and broodstock management practices aimed at achieving genetic improvement, along with inbreeding control, may be appropriate. However, the impact of the release of non-indigenous species on other species should be considered from a genetic, cultural, fishery productivity and ecological perspective. This is particularly the case if a species is not yet present in a waterbody. An example of an adverse impact from the introduction and restocking of a non-indigenous species is the case of common carp (*Cyprinus carpio*) in Inle lake, Myanmar. The locally indigenous, commercially important and culturally significant Inle carp (*Cyprinus intha*) is now considered endangered with extinction for reasons including competition from, and hybridisation with, common carp (Hlaing 2014).

Implementation and management of mass-selection programs

Mass selection, also known as **phenotypic selection**, is the process of selecting the 'best' individuals within each generation of a strain as parents of the next generation (Bentsen and Olesen 2002; In et al. 2016). Under mass selection, the measured phenotype (i.e. measured value of a characteristic) of an individual is assumed to be proportional to its breeding value for the characteristic in question. This is an imprecise means of estimating a breeding value. However, mass selection has been practiced by farmers of domesticated crops and terrestrial animals for millennia and has been responsible for marked genetic improvement in these species.

The benefits of mass selection over family-based selective breeding and genomic selection are its simplicity and low cost. Its primary limitation is that genetic relationships among individuals within strains are unknown, meaning that:

- the degree of relationship among selected parents is unknown, with implications for
 - inbreeding control, and
 - the maintenance of additive genetic diversity, required for long term genetic improvement;
- it cannot be used to select individuals on the basis of the performance of their relatives, a key to making genetic improvement in traits that cannot be directly measured on candidate parents (e.g. disease resistance); and
- it does not allow the estimation of genetic parameters (e.g. heritability and genetic correlations), without which the magnitude of genetic improvement through selection cannot be accurately predicted.

However, in the absence of well-resourced family-based breeding programs, substantial genetic gains are possible through mass selection (Bentsen and Olesen 2002). Furthermore, the existence of multiple mass selected strains would represent a good foundation for any future family-based breeding population, as they would not only have undergone a degree of genetic improvement but, together, also retained a substantial proportion of additive genetic diversity (Knibb et al. 2014). Furthermore, issues associated with inbreeding, caused by unknown relationships among individuals in mass selected strains, can be curtailed to some extent by:

- mating a large number of parents, to produce a large number of families, each generation (Approaches 1.2.2, 1.2.3, 2.1 and 2.2, Bentsen and Olesen 2002);
- adopting a rotational mating approach (Approach 1.2.3, Ponzoni et al. 2012b); or
- maintaining two or more unrelated mass selected strains and crossing between them to produce non-inbred seed for grow out (Approaches 2.1 and 2.2).

By ensuring that a large number of parents are spawned each generation (i.e. no less than 50 dams and 50 sires, Bentsen and Olesen 2002) and that a large number of families are generated, the probability that closely related individuals are selected as parents (i.e. the probability of a **genetic bottleneck**) in the following generation is diminished. However, even under this conservative implementation of mass selection, there is the possibility of a genetic bottleneck occurring in strains. For example, if most families die or one family performs substantially better than all others – due to differences in genetics, environment/husbandry prior to family pooling, or the time of spawning – then a high proportion of individuals selected as parents will be siblings. The adverse impacts of a genetic bottleneck on inbreeding and additive genetic diversity cannot be reversed in subsequent generations of an individual strain. Some specific guidelines, aimed at minimising the drawbacks of mass selection, are detailed below.

Establishment of founder populations

General principles for the establishment of founder populations are:

- there should be no known or suspected genetic relationships between founders of different strains;
- there should be no, or minimal, genetic relationships among founders of the same strain; and
- there should be no less than 50 male and 50 female founders (Bentsen and Olesen 2002).

Possible sources of founders are other captive strains, farm stocks and wild stocks. However, for non-indigenous species wild stocks may not be available. The advantage of using other captive strains as founders is that previous management and selection may have resulted in a degree of genetic improvement, assuming no negative selection (Hussain and Mazid 2005). However, captive strains and farm stocks are prone to high levels of average relatedness - limiting future genetic improvement and the ability to control inbreeding.

Strain founders can be sourced from multiple strains (e.g. both wild and hatchery strains). However, if merging strains for mass selection, all matings should be between strains and no more than two strains should be merged in any one generation. This is to avoid possible overrepresentation of selections from as small number of families in the next generation due to i) differences in genetic improvement among families from different parental strains; and/or ii) superior performance of families derived from between-strain crosses, compared with those from within-strain crosses, due to release from inbreeding depression (i.e. positive **heterosis**).

If two or more strains are to be established (see Approach 2.2), each from different captive strains or wild populations, it is possible that some between-strain crosses will express greater levels of heterosis than others in future commercial crosses. The expression of such heterosis becomes increasingly predictable (i.e. fixed) as the average relatedness of individuals within strains increases over generations – a phenomenon exploited in the genetic improvement of some crops (e.g. hybrid corn). However, between-strain heterosis can only be fully addressed and/or exploited through well designed strain comparison trials (Ponzoni et al. 2012a), which require financial and technical resources that are beyond the capacity of most hatcheries. Accordingly, hatcheries implementing Approach 2.2 should select founders on the assumption that the establishment of multiple strains, each from different captive strains or wild populations, is unlikely to result in adverse outcomes (i.e. negative heterosis), unless data from previously conducted strain comparison trials provide evidence to the contrary.

To maximise the probability that founders obtained from wild populations are unrelated, they i) should be obtained from large water bodies with large populations; and ii) if collected as spawn/fry, obtained at the peak of the spawning season from areas in which the species is prevalent (Hamilton et al. submitted-a; Hamilton et al. submitted-b).

Ideally, all founders should be fin-clipped and samples stored and archived appropriately for future pedigree assignment, estimation of genetic relatedness and/or genomic studies. This may require advice and support from external organisations, particularly if tissue samples require long term storage.

Spawning to obtain new broodstock (within-strain mating)

It is desirable in a mass selection program to:

- maximise the number of parents that contribute to the next generation (ideally contributions from each parent should be equal);
- maximise the number of full-sibling families to which each parent contributes; and
- if possible, avoid mass spawning due to the associated uncertainty of parental contributions (Lind et al. 2009).

At least 50 dams and 50 sires should be used to produce new broodstock (i.e. regenerate strains, Bentsen and Olesen 2002). Ideally, each sire would be mated with each female and equal numbers of progeny from each full-sibling family retained. This could be achieved, approximately at least, in many finfish species by:

1. inducing and strip spawning each of the males;
2. obtaining equal quantities of milt from each male – excess milt could be used for commercial seed production by crossing with an unrelated strain;
3. using milt-extenders to allow short-term milt storage;
4. pooling milt from all sires and mixing;
5. inducing and strip spawning each of the females;
6. obtaining equal volumes of eggs from each female – excess eggs could be used for commercial seed production by crossing with an unrelated strain;
7. separately fertilising the eggs of each female with equal volumes of the pooled milt; and
8. pooling fertilised eggs and rearing according to normal procedures.

Mating in mass selection programs should be undertaken over a short time frame, ideally in a single day. If this is not possible, fish spawned at different times should be maintained separately up to the point of selection (Figures A2.2 and A2.4 to A2.7). If different aged fish are grown out together it is not possible to determine if differences in performance at the time of selection are the result of difference in age or genetics.

Selection

Large and healthy fish should be selected as parents but other traits may also be selected for, such as external shape and colour. The very best individuals should be retained to produce new broodstock (within-strain mating). However, additional broodstock may be retained for commercial production (between-strain mating). Points to consider at the time of selection follow.

- The more traits that are selected for, the less the genetic improvement achieved for any one trait. For example, if both colour and size are selected for, the genetic improvement for size will be less than if size alone had been selected for.
- The extent to which genetic improvement can be achieved in any one generation is influenced by i) the heritability of the trait/s under selection, which cannot be quantified in a mass selection program, ii) the proportion of fish selected (i.e. the intensity of selection), and iii) the extent of additive genetic diversity.
- No less than an average of 25 progeny per parent should be grown out. However, increases in the rate of genetic improvement by growing out substantially larger

numbers of progeny per parent (e.g. 50) are likely to be limited (Bentsen and Olesen 2002). Future broodstock requirements also need to be considered when determining the number of individuals to be grown out.

- Inadvertent selection of unfavourable characteristics (e.g. early maturity is potentially selected for if parents are mated before all individuals are sexually mature).

Tagging and marking

If multiple mass selected strains are to be maintained, tags or marks identifying the strain to which each fish belongs should be applied at the time of selection (Figures A2.4 to A2.7). For ease of management, externally-visible tags/marks are best. Tags or marks are required even if broodstock are to be maintained according to age class in separate broodstock ponds, due to the risk of infrastructure failure, flooding and human error – particularly at the time of between-strain mating to produce seed for growout (Figures A2.4 to A2.7).

Research may be required into the development of appropriate methods of externally tagging/marking fish (Basavaraju et al. 1998; CDOF 2004; McKenzie et al. 2012). For example, clipped fins tend to grow back relatively rapidly in many warm water species, including carp (family Cyprinidae), and repeated clipping would be required if this approach was adopted in these species – with associated labour costs and a risk of damaging valuable broodstock.

If tags or marks fail on individuals (i.e. the strain of an individual is uncertain), such fish must not be used to produce new broodstock. If small in number, such fish could be used for the production of seed for grow out.

Broodstock maintenance

If using a rolling front approach, maintaining broodstock in ponds according to age class is likely to simplify management (Figures A2.6 and A2.7) but is not necessary if fish are tagged or marked according to strain. Furthermore, multiple species can be maintained in ponds as long as it is certain that between-species hybridisation does not occur in planned matings.

Population backup

Ideally, all strains should be maintained and distinguishable (e.g. tagged or maintained in separate ponds) on more than one site. In the event of a catastrophic infrastructure failure, disease outbreak or mass mortality at one site, the strains are then less likely to be lost.

References

- Bangera, R., Correa, K., Lhorente, J.P., Figueroa, R., and Yáñez, J.M. (2017). Genomic predictions can accelerate selection for resistance against *Piscirickettsia salmonis* in Atlantic salmon (*Salmo salar*). *BMC Genomics* 18, 121. doi: 10.1186/s12864-017-3487-y
- Basavaraju, Y., Devi, B.S.R., Mukhatyakka, G., Reddy, L.P., Mair, G.C., Roderick, E.E., and Penman, D.J. (1998). Evaluation of marking and tagging methods for genetic studies in carp. *Journal of Biosciences* 23, 585-593. doi: 10.1007/bf02709169
- Bentsen, H.B., and Olesen, I. (2002). Designing aquaculture mass selection programs to avoid high inbreeding rates. *Aquaculture* 204, 349-359. doi: 10.1016/S0044-8486(01)00846-8

- Cdof (2004). "Canada Department of Fisheries and Oceans Animal-User Training Template 6.0 Marking and Tagging of Finfish".
- Evans, F., Matson, S., Brake, J., and Langdon, C. (2004). The effects of inbreeding on performance traits of adult Pacific oysters (*Crassostrea gigas*). *Aquaculture* 230, 89-98. doi: <https://doi.org/10.1016/j.aquaculture.2003.09.023>
- Falconer, D.S., and Mackay, T.F.C. (1996). Introduction to quantitative genetics. Forth edition. Harlow: Longman
- Fao (2008). "Aquaculture development. 3. Genetic resource management. FAO Technical Guidelines for Responsible Fisheries. No. 5, Suppl. 3". (Rome: Food and Agriculture Organization of the United Nations).
- Gjerde, B., Gunnes, K., and Gjedrem, T. (1983). Effect of inbreeding on survival and growth in rainbow trout. *Aquaculture* 34, 327-332. doi: [https://doi.org/10.1016/0044-8486\(83\)90212-0](https://doi.org/10.1016/0044-8486(83)90212-0)
- Hamilton, M.G., Mekkawy, W., and Benzie, J.a.H. (submitted-a). Sibship assignment to founders of a Bangladeshi *Catla catla* breeding population. *Genetics Selection Evolution*
- Hamilton, M.G., Mekkawy, W., Kilian, A., and Benzie, J.a.H. (submitted-b). Single nucleotide polymorphisms (SNP) reveal sibship among founders of a Bangladeshi rohu (*Labeo rohita*) breeding population. *Frontiers in Genetics*
- Hlaing, M.M. (2014). Preserving a critical fishery resource in Inle lake, Myanmar for sustainable fisheries and food security. *Fish for the People* 12, 24-29
- Hussain, M.G., and Mazid, M.A. (2005). "Carp genetic resources of Bangladesh," in *Carp genetic resources for aquaculture in Asia*, eds. D.J. Penman, M.V. Gupta & M.M. Dey. (Penang, Malaysia: WorldFish), 16-25
- In, V.V., O'connor, W., Dove, M., and Knibb, W. (2016). Can genetic diversity be maintained across multiple mass selection lines of Sydney rock oyster, *Saccostrea glomerata* despite loss within each? *Aquaculture* 454, 210-216. doi: 10.1016/j.aquaculture.2015.12.030
- Knibb, W., Whatmore, P., Lamont, R., Quinn, J., Powell, D., Elizur, A., Anderson, T., Remilton, C., and Nguyen, N.H. (2014). Can genetic diversity be maintained in long term mass selected populations without pedigree information? — A case study using banana shrimp *Fenneropenaeus merguensis*. *Aquaculture* 428-429, 71-78. doi: <https://doi.org/10.1016/j.aquaculture.2014.02.026>
- Komen, J., Wiegertjes, G.F., Van Ginneken, V.J.T., Eding, E.H., and Richter, C.J.J. (1992). Gynogenesis in common carp (*Cyprinus carpio* L.). III. The effects of inbreeding on gonadal development of heterozygous and homozygous gynogenetic offspring. *Aquaculture* 104, 51-66. doi: [https://doi.org/10.1016/0044-8486\(92\)90137-A](https://doi.org/10.1016/0044-8486(92)90137-A)
- Lind, C.E., Evans, B.S., Knauer, J., Taylor, J.J.U., and Jerry, D.R. (2009). Decreased genetic diversity and a reduced effective population size in cultured silver-uppé pearl oysters (*Pinctada maxima*). *Aquaculture* 286, 12-19. doi: DOI 10.1016/j.aquaculture.2008.09.009
- Mckenzie, J., Parsons, B., Seitz, A., Kopf, R., Mesa, M., and Phelps, Q. (2012). Advances in fish tagging and marking technology. Bethesda Maryland: American Fisheries Society
- Meuwissen, T.H.E. (1997). Maximizing the response of selection with a predefined rate of inbreeding. *Journal of Animal Science* 75, 934-940
- Meuwissen, T.H.E., and Sonesson, A.K. (1998). Maximizing the response of selection with a predefined rate of inbreeding: Overlapping generations. *Journal of Animal Science* 76, 2575-2583
- Mohamed Din, M., and Subasinghe, R. (2017). "Basic biosecurity manual for tilapia hatchery technicians in Bangladesh." (Penang, Malaysia: CGIAR Research Program on Fish Agri-Food Systems. WorldFish).
- Nomura, T., and Yonezawa, K. (1996). A comparison of four systems of group mating for avoiding inbreeding. *Genetics Selection Evolution* 28, 141



EUROPEAN UNION



Ponzoni, R.W., James, J.W., Nguyen, N.H., Mekkiy, W., and Khaw, H.L. (2012a). "Strain comparisons in aquaculture species: a manual". (Penang, Malaysia: WorldFish).

Ponzoni, R.W., Nguyen, N.H., Khaw, H.L., and Rodriguez Jr., B.M. (2012b). "Considerations about effective dissemination of improved fish strains.". (Penang, Malaysia: WorldFish).

Sonesson, A.K. (2005). A combination of walk-back and optimum contribution selection in fish: a simulation study. *Genetics Selection Evolution* 37, 587-599. doi: 10.1051/Gse:2005020

Valiquette, E., Perrier, C., Thibault, I., and Bernatchez, L. (2014). Loss of genetic integrity in wild lake trout populations following stocking: insights from an exhaustive study of 72 lakes from Quebec, Canada. *Evol Appl* 7, 625-644. doi: 10.1111/eva.12160



Appendix 3. Naming strains and documenting their ancestry

What is a strain?

In the context of this report:

- a strain is a population, descended from a finite number of founder individuals that is managed in captivity (i.e. a closed population);
- the founders of a strain may be sourced from the wild or be members of another strain; and
- a new strain is defined i) when wild founders are collected, ii) at the time of mating after members of a strain have been moved from one river/hatchery to another (i.e. river to hatchery or hatchery to hatchery), iii) when a strain is split, and iv) when strains are merged.

When merging strains, all crosses should be made between strains.

A strain may be partitioned into multiple 'age cohorts' (or generations, if generations do not overlap).

Strain details

The approach outlined below allows individual strains to be identified and their ancestry reconstructed, in isolation from a centralised database. It provides a general structure to document strain ancestries in the absence of perfect information about the origins of all founders and strains.

Where known, the following data should be recorded.

- **Species scientific name** (e.g. 'Labeo rohita' for rohu carp).
- **Strain type** – Type 'N' for a strain comprised of founder individuals taken from an unmanaged wild (indigenous or introduced) population (N for 'natural' or 'nature'), and Type 'S' a strain with founders sourced from other Type N or Type S strain/s.
- **Date of capture** for Type N strains, or **date of mating** among founders for Type S strains, recorded to the closest year, month or day (YYYY-MM-DD; 0000 or 00 if unknown). If collections/matings are made over a number of days, the first date of collection/mating should be specified.
- Location of the **place of collection** for Type N, or **hatchery** where founders were initially mated for Type S:
 - Two-letter (Alpha-2) ISO country code (ISO 2013) (e.g. 'MM' for Myanmar);
 - Postal code (0 if postal code unknown); and
 - Place of collection (river and location) for Type N strains, or name of hatchery for Type S strains.
- Unique **individual, organisation and/or project code identifier** for the collector of Type N strains, or a unique **strain identifier within the hatchery** for Type S strains.
- **Strain identifier** (e.g. the short 'common' name under which the strain is marketed).
- Other salient details depending on context and availability of information. For example, the:

- number of founding dams and sires from each ancestral strain;
- family identifiers of founders and or average relatedness if from a family-based breeding program;
- selection history (e.g. number of mating events since inception and number of parents per mating event, traits under selection, intensity of parental selection); and/or
- FishBase strain identifier.

Within strain details

Within strains, ‘age cohorts’ (or generations) can be defined. The date of the ‘mating event’ that generated each ‘age cohort’ should be recorded to the closest year, month or day (YYYY-MM-DD; 0000 or 00 if unknown). If matings are made over a number of days, the first date of mating should be specified.

Naming strains

Strains can be named and uniquely identified according to the formats outlined in Figures A3.1 and A3.2.

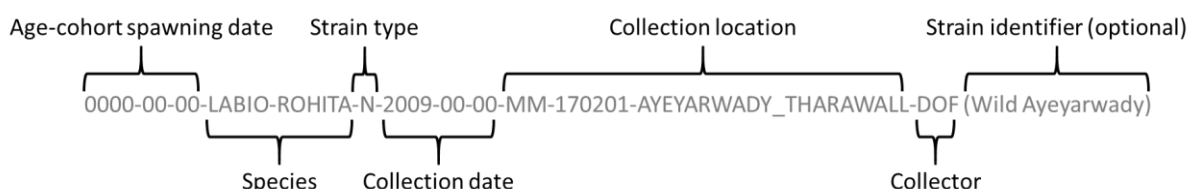


Figure A3.1. Naming convention for strains comprised of founder individuals taken from wild (indigenous or introduced) populations (i.e. Type N strains). Note the use of dashes, to partition data fields, and underscores to avoid blank spaces within data fields.

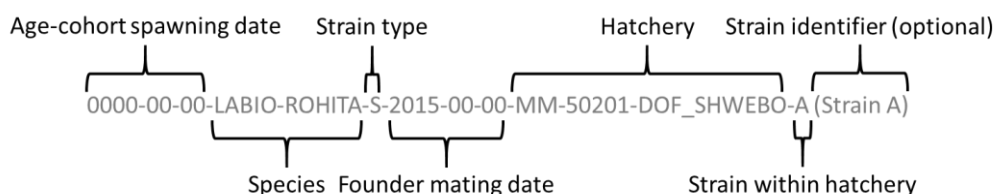


Figure A3.2. Naming convention for strains with founders sourced from other strain/s (i.e. Type S strains). Note the use of dashes, to partition data fields, and underscores to avoid blank spaces within data fields.

Documenting strain ancestry

As general rule, two strains with a common strain in their ancestry should not be mated to produce seed for grow out. This ensures that mated individuals are not related and their progeny are not inbred.

Below is a semi-fictional example showing two strains A and B. Information regarding the ancestry of the strains is imperfect (grey text). In this case ‘Strain A’ is descended from the ‘Kume Ayeyarwady’ and ‘Kume Bago’ strains; which are descended from the ‘Wild Ayeyarwady’ and ‘Wild Bago’ strains, respectively. ‘Strain B’ is descended from ‘Wild

Ayeyarwady' and 'Shwebo Native' and 'Shwebo Native' is descended from 'Wild Native'. It is evident from Figure A3.3 that Strains A and B do not have ancestral strains in common and thus can be mated to produce non-inbred seed for grow out.

Note that the term 'Wild Ayeyarwady' has been used in Figure A3.3 to describe two different collections, to highlight the confusion that can be caused by using short 'common' strain identifiers. This issue often arises in the case of genetically improved strains, where the same name is often used to identify what are in reality different strains with potentially different levels of genetic improvement and inbreeding.

	A	B	C	D	E	F	G	H	I	J	K	L	M	N	O	P	Q	R	S	T	U	V	W	X	Y
1	0000-00-00-LABIO-ROHITA-S-2015-00-00-MM-50201-DOF_SHWEBO-A (Strain A)																								
2	2012-00-00-LABIO-ROHITA-S-2009-00-00-MM-05162-DOF_KUME-THARAWALL (Kume Ayeyarwady)																								
3	0000-00-00-LABIO-ROHITA-N-2009-00-00-MM-170201-AYEYARWADY_THARAWALL-DOF (Wild Ayeyarwady)																								
4	2012-00-00-LABIO-ROHITA-S-2009-00-00-MM-05162-DOF_KUME-BAGO (Kume Bago)																								
5	0000-00-00-LABIO-ROHITA-N-2009-00-00-MM-0-BAGO_MOE_NYE-DOF (Wild Bago)																								
6																									
7	0000-00-00-LABIO-ROHITA-S-0000-00-00-MM-50201-DOF_SHWEBO-B (Strain B)																								
8	0000-00-00-LABIO-ROHITA-N-0000-00-00-MM-0-AYEYARWADY_MOE_GONJ-DOF_SHWEBO (Wild Ayeyarwady)																								
9	0000-00-00-LABIO-ROHITA-S-0000-00-00-MM-50201-DOF_SHWEBO-NATIVE (Shwebo Native)																								
10	0000-00-00-LABIO-ROHITA-N-0000-00-00-MM-0-NATIVE-DOF_SHWEBO (Wild Native)																								
11																									

Figure A3.3. A semi-fictional strain ancestry for two strains, 'Strain A' and 'Strain B'. Ancestral strains are listed below each descendant strain and are indented.

References

Iso (2013). "ISO 3166-1:2013. Codes for the representation of names of countries and their subdivisions -- Part 1: Country codes". International Organization for Standardization "ISO 3166 Maintenance agency").