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Breeding plan for common carp in Europe

Hanne Marie Nielsen, Ingrid Olesen and Bjarne Gjerde





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Main office in Tromsø Muninbakken 9–13 P.O. box 6122 NO-9291 Tromsø Norway Tel.: +47 77 62 90 00 Fax: +47 77 62 91 00 E-mail: nofima@nofima.no

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Nofima Marin AS Nofima Marin Muninbakken 9–13 P.O. box 6122 NO-9291 Tromsø Norway Tel.: +47 77 62 90 00 Fax: +47 77 62 91 00 E-mail: marin@nofima.no

Internet: www.nofima.no





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Nofima Marin AS

Nofima Marin P.O. box 6122 NO-9291 Tromsø, Norway Visiting address: Muninbakken 9–13, Tel.: +47 77 62 90 00 Fax: +47 77 62 91 00 marin@nofima.no www.nofima.no

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Summarv:

In this study we present an outline for a selective breeding plan aiming at producing a population of common carp with a high growth rate, a high survival rate, and which is highly resistant to Koi herpes virus (KHV) and the bacteria Aeromonas hydrophila. Different breeding strategies differing in information sources in the selection index and weightings of the traits in the breeding goal were compared. Estimates of heritabilities and genetic correlations were those reported in the EU project EUROCARP (EUROCARP, Project no. 022665). The four traits growth and survival until market size, and resistance to KHV and A. hydrophila were included in the breeding goal. Almost the same response in A. hydrophila was obtained when selecting for resistance for KHV only compared to selection for resistance to both KHV and A. hydrophila. This was due to a low heritability for A. hydrophila and a high genetic correlation between resistance to KHV and resistance to A. hydrophila. The structure of a selective breeding program for common carp should follow the general structure for large-scale breeding programs for aquaculture species, which is divided into breeding nucleus, multipliers and grow out units. A family based selective breeding program is suggested where each of 100 males are mated with two different females to produce 100 half-sib and 100 full-sib families. For each trait, 20 individual should be tested for each full sib family. After tagging, the fingerlings should be communally stocked and reared under commercial grow-out conditions for performance testing. The selective breeding program for common carp should be based on index selection with combined individual and sib selection. Individual body weight and survival should be recorded at second autumn. In order to select for resistance to KHV, challenge tests with separate, tagged individuals from the full-sib families in the breeding nucleus should be performed. Altogether, selection for improved pond survival, resistance to KHV and A. hydrophila will contribute to develop a more robust common carp for farming.

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1 Introduction

In this task we will provide an outline for a selective breeding plan aiming at producing a population of common carp, which has a high growth rate and is highly resistant to Koi herpes virus (KHV) and the bacteria Aeromonas hydrophila. The suggested breeding plan will be based on the results on genetic variation and performance of the offspring of a complete diallel cross between four common carp strains Duna, Amur (landraces), and Tata, Szarvas 15 (farmed races) in the EU project EUROCARP (EUROCARP, Project no. 022665). These offspring were tested for growth (weight and length) and survival in a commercial pond environment and resistance to Koi Herpes virus and the bacteria Aeromonas hydrophila from two independent challenge tests. Results from the diallel cross showed small strain differences for growth, but considerable differences between strains for pond survival. Heterosis was significant with respect to pond survival but varied from negative and up to 29% for weight and length traits and was non-significant for KHV and A. hydrophila. In addition, the results demonstrated substantial additive genetic variation within strains for growth, survival and resistance to KHV, while the additive genetic variation for resistance to A. hydrophila was low. Therefore, the relatively high genetic variation found for growth and resistance to KHV showed good potential for genetic improvement through selective breeding.

In order to suggest a selective breeding plan for common carp, different breeding strategies are compared with respect to different information sources in the selection index and different weightings of the traits in the breeding goal. The different strategies are compared with respect to predicted genetic gain in growth, survival and resistance to KHV and *A. hydrophila* using the magnitude of heritabilities for the traits and the genetic correlations between the traits estimated from the diallel cross experiment in the EUROCARP project. Based on the results from the comparison of the different breeding strategies a breeding plan for common carp is suggested. Aspects such as the number of families (tanks), and numbers of tested sibs are considered in addition to different sources of information and different breeding goals with different sets of relative economic weights.

2 Comparison of different alternative breeding plans

2.1 Genetic parameters of traits in the breeding goal

The following four traits, which show additive genetic variation and are economically important, could possibly be included in the breeding goal;

- a) Growth until harvest size (weight at second autumn),
- b) Pond survival (survival from second spring to second autumn),
- c) Resistance to Koi herpes virus (KHV) from a challenge test,
- d) Resistance to the bacteria Aeromonas hydrophila from a challenge test.

The magnitude of genetic variation (heritability) of the four different traits assumed are based on estimates presented by Nielsen et al. (2009) and Ødegård et al. (2009) and are shown in Table 1.

Table 1	Heritabilities (± S.E.) assumed for growth (weight at second autumn), pond
	survival (survival from second spring to second autumn), resistance to Koi herpes
	virus (KHV), and resistance to Aeromonas hydrophila (AH) (from Nielsen et al.,
	2009 and Ødegård et al., 2009)

Trait	Heritability ± S.E.	Mean	Phenotypic sd.
Growth	0.50 ± 0.12	1180 g	325 g
Pond survival	0.34 ± 0.09	78 %	41 %
Resistance to KHV	0.79 ± 0.15	5 %	22 %
Resistance to AH	0.04 ± 0.03	39 %	47 %

In general, weight at tagging, weight at first autumn, and weight at second spring are weakly genetically correlated with weight at second autumn (-0.53 to 0.47). These relatively low correlations indicate that weight recorded earlier than at second autumn is a poor predictor of weight at second autumn, which may be explained by the very different rearing environments (tank, pond, temperature) in the different growth periods. Therefore body weight at second autumn is chosen as the trait in the breeding goal.

With respect to survival, survival in the pond from second spring to second autumn shows significant genetic variation, whereas survival in earlier growth periods shows no or very little genetic variation. In addition, breeding for survival in earlier periods may be uneconomically because there is no potential income associated with the fish surviving these periods. However, mortality losses from second spring to second harvest represent large economic losses of fish having consumed large amounts of feed and that are almost ready for marketing. There may also be a welfare problem related to breeding for fish survival in earlier growth periods, because some of the fish, which survives in earlier periods but not until final harvesting, may have poor body condition (small and thin fish). Therefore survival of the fish should be recorded as survival from second spring to second autumn. Resistance to KHV and *A. hydrophila* can presently not be measured on potential selection candidates. Instead these disease traits can be recorded on sibs of the selection candidates using challenge tests (see Dixon et al., 2009; Jeney et al., 2009).

The genetic correlations between weight at second autumn, pond survival from second spring to second autumn, and resistance to KHV and *A. hydrophila* are presented in Table 2. The genetic correlation between weight and pond survival and between resistance to KHV and *A. hydrophila* is high. This means that selection for growth will lead to correlated response in pond survival. Likewise, selection for reistance to KHV will increase resistance to *A. hydrophila*. However, the genetic correlations between pond survival and resistance to both KHV and *A. hydrophila* are low and not significantly different from zero. The genetic correlations of growth with resistance to KHV and *A. hydrophila* were not estimated but are assumed to be zero.

Table 2Genetic correlations and their standard errors between growth, pond survival,
and resistance to Koi herpes virus and <u>Aeromonas hydrophila</u> (Nielsen et al.,
2009; Ødegård et al., 2009)

Trait ¹	Pond survival	Resistance to Koi virus	Resistance to Aeromonas hydrophila
Growth	0.65 ± 0.15		
Pond survival		-0.236 ± 0.213	-0.002 ± 0.313
Resistance to Koi virus			0.608 ± 0.283

At present there are no estimates of the economic values for the four traits. Therefore, different strategies will be compared with respect to different weightings of the traits in the breeding goal.

2.2 Selection and mating of broodstock

When comparing the different testing and selection strategies it is assumed that index selection will be used to select broodstock. Individual and sib selection is combined such that the breeding values for the different traits are based on information obtained from both the individual and it's full and half sibs. However, the source of information used will differ between the traits and in the different studied alternatives.

A breeding program, where each male is mated to two females and where 20 full sibs per full sib family and per trait are tested, is assumed. Thus, the information available for breeding value estimation is obtained from the breeding candidate (for body weight and pond survival) and its 19 full sibs and 20 paternal half sibs (for each trait).

2.3 Comparison of different breeding strategies

Seven different breeding strategies are investigated (Table 3). Strategies 1-4 differ with respect to sources of information included in the selection index. In alternative 1, breeding values for growth and pond survival are predicted based on own phenotype and phenotypes of sibs, whereas there is no information about the two disease resistance trait. In alternative 2, selection is for only growth, survival and resistance to KHV. However, due to the high genetic correlations between KHV and *A. hydrophila*, a correlated genetic improvement in resistance to *A. hydrophila* is also expected. In alternative 3, breeding values for growth and

pond survival are predicted based on own phenotype and phenotypes of sibs, whereas breeding values for both resistance to KHV and resistance to *A. hydrophila* are based on sibs of the selection candidates. In alternative 4 it is assumed that individual can be selected for resistance to KHV, which assumes that the fish can be used as selection candidates after being challenge tested. By strategies 5, 6 and 7, the effect of different relative weighting of the traits in the breeding goals on genetic improvement of the four traits are investigated assuming index sources as in strategy 2. The applied economic values were standardized per phenotypic standard deviation (Table 1) and expressed as relative economic values.

	Traits included in breeding goal	Index source	Relative Economic value
	Growth	Individual + sibs	1
Otroto m / 4	Pond survival	Individual + sibs	1
Strategy 1	Resistance to KHV	-	1
	Resistance to A. hydrophila	-	1
	Growth	Individual + sibs	1
Otroto m / O	Pond survival	Individual + sibs	1
Strategy 2	Resistance to KHV	Sibs	1
	Resistance to A. hydrophila	-	1
	Growth	Individual + sibs	1
Strotomy 2	Pond survival	Individual + sibs	1
Strategy 3	Resistance to KHV	Sibs	1
	Resistance to A. hydrophila	Sibs	1
	Growth	Individual + sibs	1
Strotomy 4	Pond survival	Individual + sibs	1
Strategy 4	Resistance to KHV	Individual + sibs	1
	Resistance to A. hydrophila	Sibs	1
	Growth	Individual + sibs	1
Strotomy E	Pond survival	Individual + sibs	2
Strategy 5	Resistance to KHV	Sibs	1
	Resistance to A. hydrophila	-	1
	Growth	Individual + sibs	1
Otroto m / C	Pond survival	Individual + sibs	1
Strategy 6	Resistance to KHV	Sibs	2
	Resistance to A. hydrophila	-	1
	Growth	Individual + sibs	1
Strotom, 7	Pond survival	Individual + sibs	1
Suralegy /	Resistance to KHV	Sibs	1
	Resistance to A. hydrophila	-	2

Table 3 Description of the seven different breeding strategies studied

Based on the heritabilities (Table 1) of the traits and their genetic correlations (Table 2), the expected genetic superiority of the selected fish for each of the four studied traits for each of the seven breeding strategies (Table 3) were calculated (Table 4) assuming a selection intensity of one for each trait).

Table 4Expected genetic superiority of selected males and females after one round of
selection (assuming a selection intensity of one) for harvest body weight, pond
survival, resistance to Koi herpes virus (KHV) and resistance to <u>A. hydrophila</u>
expressed as percentage fish survived, and accuracy of selection (correlation
between true and estimated breeding value) for the seven studied breeding
strategies

	<u>Trait</u>				
	Body weight (g)	Pond survival (%)	Resistance to KHV (%)	Resistance to <i>A. hydrophila</i> (%)	Accuracy of selection
Strategy 1	178	11.1	0	0	0.71
Strategy 2	169	10.6	2.4	0.6	0.75
Strategy 3	169	10.6	2.4	0.5	0.75
Strategy 4	167	10.4	3.1	0.7	0.76
Strategy 5	163	11.6	1.7	0.4	0.73
Strategy 6	149	9.3	4.2	1.0	0.71
Strategy 7	152	10.8	3.9	0.8	0.71

The expected genetic superiority for harvest varied from 12.6 (strategy 6) to 15.1 (strategy 1) percentage of the mean and thus substantial for all strategies. About the same genetic superiority is obtained for *A. hydrophila* when selecting for resistance to KHV only (strategy 2) compared to selection for resistance to both KHV and *A. hydrophila* (strategy 3). This is because of the low heritability for *A. hydrophila* and the relatively high genetic correlation (0.6) between resistance to KHV and resistance to *A. hydrophila*. In addition, if it will be possible in the future to select for own performance for resistance to KHV (strategy 4) even higher resistance to both KHV and *A. hydrophila* could be obtained because the intensity of selection will be higher for these traits.

In the results above we assumed that inbreeding was considered (e.g. mating of related fish was avoided). In addition, we assumed the same selection intensity for all strategies. In practise, selection intensity would differ dependent on the number of families and differ between traits dependent on whether the trait are based on own performance (within family selection) or on performance of relative (between family selection).

When increasing the relative economic weight on resistance to KHV (strategy 6) or *A. hydrophila* (strategy 7), resistance to KHV and *A. hydrophila* increases as expected. However, increased economic weights on resistance to KHV and *A. hydrophila* change selection emphasis from growth towards disease resistance such that the genetic improvement for growth decreases.

Based on these results, alternative 2 seems to be the best strategy because relatively high genetic improvement is obtained for growth, survival and resistance to both KHV and *A. hydrophila*, without challenge testing for *A. hydrophila*. However, it should be noted that strategy 2 may not be the best alternative when expressed in economic terms. However, because the economic values for the four studied traits currently are unknown, the strategies can not be compared in monetary units.

3 Suggestion of a breeding plan for common carp in Europe

3.1 Genetic material in the base population

A selective breeding program for common carp in Europe could be initiated using the families produced in the EUROCARP project (Jeney et al., 2009). The relative performance of the four different strains (Tata, Amur, Duna, and Szarvas 15) is known from the diallel cross experiment. Some animals from these families, which all have pedigree information, were produced in 2006 and are kept in a pond managed by the Fish Biology Department at the Research Institute of Fisheries, Aquaculture and Irrigation (HAKI), based at Szarvas in Hungary and could be ready for mating in year 2009.

When performing the selection and mating in 2009 the breeding candidates should not only be selected based on their estimated breeding values which may result in candidates from only a limited number of the 16 purebred and crossbred groups of the diallel cross. Instead candidates should be selected from a large proportion of the different groups to secure a broad representation of the four strains in future generations.

3.2 General structure of a selective breeding program for carp

The structure of a selective breeding program for common carp is suggested to follow the general structure for large-scale breeding programs for aquaculture species (Figure 1), which is divided into different layers; breeding nucleus, multipliers and grow out units (Gjerde, 2005). In the breeding nucleus, a continuous, accumulating selection response for the traits under selection is produced. In the multiplier units, fingerlings from the nucleus are reared until they reach sexual maturity, where after they reproduce and thus supply seed to individual farmers/grow out units.



Figure 1 Figure showing the structure and the main elements of a fish breeding program (Gjerde, 2005)

The main advantage of breeding programs that utilize sib information in the selection decisions is their flexibility and superior efficiency for improving traits of low to medium heritability and traits that cannot be recorded on live animals such as resistance to diseases. Another advantage of a family based selection program is that it allows for parallel testing in diverse rearing environments for investigation of possible genotype by environment interactions.

3.3 Traits in the breeding goal

The breeding goal should include the four traits growth; (harvest weight), pond survival (survival during last summer growth season, usually from second spring to second autumn), resistance to KHV and resistance to *A. hydrophila*. Outbreaks of mortality caused by koi herpesvirus (KHV) has been found in many countries all over the world, and the disease is currently considered as one of the most risky factors affecting populations of farmed common carp (Pokorova et al., 2005). The bacteria *A. hydrophila* is associated with various disease problems in carp aquaculture production worldwide, and infection often follows situations with elevated stressing of the fish (Nielsen et al., 2001). Altogether, selection for improved pond survival, KHV and *A. hydrophila* will contribute to develop a more robust common carp for farming.

3.4 Selection of broodstock

The selective breeding program for common carp should be based on index selection with combined individual and sib selection, as described under the different breeding strategies. Selection for growth should be based on own performance as well as harvest weight of full and half sibs at second autumn. Likewise, pond survival should be based on own

performance and performance of full and half sibs, whereas resistance for KHV should be based on survival of challenge tested full and half sibs. Due to the high genetic correlations between resistance to KHV and resistance to *A. hydrophila*, genetic improvement in resistance to *A. hydrophila* is expected without having to test sibs for resistance to *A. hydrophila*. This will reduce the costs of the breeding program and suffering of tested fish considerably.

The best fish (the fish with the highest breeding values) within a given family should be selected as parents of the next generation of fish. Breeding values for growth should be estimated using BLUP (best linear unbiased prediction) using a sire/dam model including the tank effect (the environmental effects common to full sibs reared in the same tank). Breeding values for KHV should be based on a linear repeatability model or test day model (see e.g. Ødegård et al., 2006, 2009), which utilize all information in the data.

We suggest using 200 families instead of 100 families (as in the EUROCARP project) because using 200 families will yield lower rate of inbreeding. For example, with 3200 selection candidates, truncation selection based on BLUP breeding values for a single trait with a heritability of 0.4, rate of inbreeding will be 0.017 and 0.009 with 100 and 200 families, respectively. A way to avoid high rates of inbreeding is by using optimum contribution selection, a method that maximizes genetic gain with a restriction on the rate of inbreeding (Meuwissen, 1997; Grundy et al., 1998). Lately tools based on optimum contribution selection, which can handle a large number of selection candidates, have become available and applied in aquaculture breeding schemes (Hinrichs et al., 2006).

In order to produce minimum 200 families when using a 1 male to 2 female ratio, 100 males and 200 females are needed. However, not all selected fish are expected to produce high quality milt and egg such that more sires and dams need to be selected.

3.5 **Production of full and half sib families**

For each year class, min. 200 families should be produced using a nested mating design where each male is mated to two different females. Fertilized eggs from each family should be placed on a fine meshed nylon cloth attached to a steel frame and placed at a 45° angel into separate 0.5 m² fiber glass tanks with a water volume of 250 L as described by Jeney et al. (2009). When the fertilised eggs hatch, the nylon cloth should be removed from the tanks. When reaching an average size of 12-15 grams, an equal number of fingerlings from each family should be individually tagged (for example with PIT-tags).

The number of selection candidates reaching sexual maturity should be around 20 per family. The number of tagged animals per family will depend on expected survival rates from hatching until recording time. This means that for a breeding scheme, where the fish is tested for growth rate, pond survival and resistance to KHV, 40 fingerlings per family should be tagged. With 200 families this corresponds to 8000 fish in total per year class. Additional fish should be stocked according to the mortality loss expected before the performance can be recorded.

3.6 Performance testing of fish for growth, survival and resistance to KHV

After tagging, the fingerlings should be communally stocked and reared under commercial grow-out conditions for performance testing. Individual body weight and survival should be recorded at second autumn every year at a fixed age of the fish. Effective selection for resistance to KHV requires controlled challenge test experiments with separate, tagged groups from the full-sib families in the breeding nucleus as described in more details by Dixon et al (2009). The test must be carried out in an isolated test facility, and the test fish should be discarded after the test. Challenge testing with KHV can be conducted using cohabitants. A single test should be carried out in which animals at an age of about 9 month from each family are exposed to the Koi herpes virus (see e.g. Dixon et al. 2009). The course of the mortality must be closely monitored. Dead fish should be recorded every day until the end of the test; i.e., at least 50 percent mortality. Preferably, the test should be prolonged and thus terminated at a higher mortality which will make it possible to use a repeatability test day model to the data.

In addition, it is suggested to include a field test with tagged fish from the breeding nucleus (see Figure 1), which should be performed at a commercial farm in order to get estimates about survival and growth of the fish in a realistic farm environment. In order to test survival, use of antibiotics could be avoided.

3.7 Strategies to monitor genetic gain

The realized genetic gain obtained for the traits under selection should routinely be estimated using BLUP methodology, which allow for separation of the genetic and environmental trends in the data accumulated over generations of selection. In order to obtain unbiased and accurate estimates of genetic gain some of the breeders (about 5 to 10%) should be reused in subsequent generations. Individually tagged breeders, which are kept alive at the breeding nucleus facility can be used for this purpose and used to perform repeated matings, or mated to some of the new selection candidates, after one generation of selection in the breeding program. The genetic gain obtained in the selective breeding program can be estimated by comparing average breeding values of the reused breeders with breeding values of the breeders from the latest reproduction of families in the breeding program. The offspring groups of the 'old' and 'new' sires and dams should be reared communally to obtain unbiased estimates of the genetic gain.

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