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Heritability estimates for processing and quality traits in common carp (*Cyprinus carpio* L.) using a molecular pedigree

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Heritability estimates with a microsatellite parentage assignment based pedigree are of special interest in common carp cultured under traditional pond conditions. This method reduces common environment effects as all families can be grown immediately after hatching mixed in the same pond. We applied this method to study genetics of growth and processing traits in common carp at market size (1.5 kg and more). The experimental progeny was established by crossing 147 two-year old males and 8 females (six to eight-year old) of Hungarian synthetic mirror carp population. The fish grew up through three vegetation seasons and at the end a sample of 331 fish was examined for biometrical traits (standard length, body weight, relative head length, relative body height, relative body width), percent fat and processing traits (% processed body, % fillets with skin, % fillets without skin). It was shown that sex had a significant effect on most traits: females were larger and fatter than males, and they had both higher percent processed body and percent fillet with skin. Standard length, body weight, percent fat and relative head length had a high heritability (>0.5), while relative body height, relative body width, percent processed body and fillet yields had a medium heritability (0.2–0.5). We found relatively high positive genetic correlations between body size (standard length and body weight) and percent fat (0.71 and 0.59, respectively), favourable genetic correlations between body size and percent processed body (0.69 for standard length and 0.74 for body weight) and between body size and fillet yields (0.50–0.77). Genetic correlations between body size and body shape (relative head length, relative body height and relative body width) were weak to moderate, thus selection for better growth should have little impact towards more rotund shape. Relative head length had strong negative correlation (–0.7 to –0.9) with percent fat, percent processed body and percent fillet yields. This means that indirect selection for reduced relative head length should be effective in improving of fillet yield.

Keywords: Common carp; *Cyprinus carpio*; Heritability; Muscular fat; Processing; Dressout; Slaughtering; Growth; Genetic correlations; Parentage assignment; Microsatellites

1. Introduction

The common carp, *Cyprinus carpio*, is a major fish species in the world aquaculture production (FAO,

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2005). It is exclusively farmed in traditional extensive or semi-intensive ponds, where fish growth is highly dependent (50% and more) on natural food (plankton and benthos). Hence, environmental conditions have a major impact on the performance of common carp, and high attention has been given to optimization of pond management. The pond management technology is well documented (Horváth et al., 1992) and benefits from centuries of farming knowledge. Genetic improvement could be a complementary way to increase the effectiveness of carp culture.

Studies on the genetic improvement of common carp have been performed by several authors (e.g. Moav and Wohlfarth, 1976; Bakos and Gorda, 1995; Cherfas et al., 1996; Gela et al., 2003; Linhart et al., 2002; Kocour et al., 2005a,b) and reviewed e.g. by Hulata (1995) and Horváth and Orban (1995). Presently, the only widely applied method in common carp culture is intraspecific crossbreeding (Kocour et al., 2005a). Utilization of all-female population, produced from gynogenetic sex reversed offspring and normal females, promises 5–8% higher performance (Cherfas et al., 1996; Kocour et al., 2005b), but is not widely applied due to technical difficulties. In a well documented experiment, mass selection for growth related traits was found ineffective (Moav and Wohlfarth, 1976) and estimated heritabilities for growth traits range from 0.0 to 0.5 (Vandeputte, 2003, for a review). However, some of the past experiments could have been biased due to small number of breeders used, and/or inability to separate common environment and genetic effects (Vandeputte, 2003).

In a previous study with common carp, Vandeputte et al. (2004) used microsatellites for parentage assignment of 240 families produced from 10 dams and 24 sires. Heritability estimates for weight, standard length and Fulton's coefficient of body condition were found to range from 0.33 to 0.37. However, these estimates were obtained from juvenile fish at the age of 8 weeks. Since such results were encouraging and showed a potential for mass selection on growth in common carp, we have undertaken the present study to estimate heritabilities in growth traits at market size. To account for potential future marketing strategy and changing market demands, heritability values were also estimated for quality and processing traits (% fat, % processed body, % fillets with and without skin). The aim of this study was to estimate genetic parameters (heritabilities, correlations) for growth and processing traits in the common carp at three summers of age, using microsatellite parentage assignment to avoid environmental effects common to full-sib families.

2. Materials and methods

2.1. Constitution of a pedigreed population of experimental fish

The reproduction and culture of common carp was carried out at the experimental facilities of the University of South Bohemia, Research Institute of Fish Culture and Hydrobiology (Vodňany, Czech Republic). The traits of interest were recorded from generation 2 (G2) fish. G1 offspring were produced in May 2001 using a full factorial cross between 24 G0 males and 10 G0 females of the HSM (Hungarian Synthetic Mirror) line (Vandeputte et al., 2004). Fin samples (1 cm²) were taken on all 34 G0 parents and kept in Eppendorf tubes filled in with 98% ethanol at laboratory temperature. In May 2003, 147 fluent males were collected from the G1 population, and crossed simultaneously with eight suitable G0 HSM females in a full factorial cross to establish the G2. Fin samples were collected on all 155 parents (147 G1 males and 8 G0 females), and stored in 98% ethanol. Only one G0 female was used in both 2001 and 2003 crosses. The full factorial cross was done by pooling an equal quantity of striped eggs from all 8 females. The pool of eggs was then divided into 147 equal aliquots. Each aliquot was fertilized individually by the sperm of the 147 males. This was done in three separate fertilization batches, with 49 egg aliquots fertilized in each batch. The duration between the first and the third fertilization did not exceed an hour. One minute after fertilization all aliquots from one fertilization batch were pooled and egg stickiness was eliminated prior to incubating the eggs in one Zuger jar. Altogether, we incubated eggs in three Zuger jars (three fertilization batches) and nursed larvae in three incubation cradles in the same conditions. At swimming stage, we stocked the larvae in a pond after mixing equal numbers of larvae from each of the three incubation cradles. For more details on the crossing, incubation and larval rearing methodologies used see Vandeputte et al. (2004). The G2 population was reared as a single batch in the same conditions in the same pond under semi-intensive pond management. In September 2003, a random sample of 2000 yearlings of G2 population was individually PIT tagged and fin sampled. G2 population was reared in earthen ponds until October 2005 (through three growing seasons), when they reached market size. From the 2000 collected fin samples of G2, a random subset of 812 fish was genotyped for parentage assignment.

2.2. Parentage assignment

The selected 812 G2 individuals were genotyped for five to eleven microsatellites described by Crooijmans et al. (1997): MFW7, MFW9, MFW11, MFW16, MFW18 and MFW26 for all fish, MFW3, MFW12, MFW20, MFW29 and MFW40 for some fish only. Parentage assignment was performed by exclusion with one mismatch tolerated using VITASSIGN (Vandeputte et al., 2006). Among the 147 G1 sires, 126 (85.7%) could be assigned to a single G0 parental pair, the remaining 21 being assigned to two or more pairs. These 21 sires were considered unrelated in the additive relationship matrix. Among the 812 G2 offspring, 615 (75.7%) could be assigned to a single parental pair. One G2 fish was not assigned, and 197 were assigned to several parental pairs. These 198 fish were excluded from the analysis.

2.3. Selection of slaughtered fish and phenotyping

The 2000 G2 fish were landed and controlled in April 2004, October 2004 and April 2005. In April 2005, 445 of the 615 uniquely assigned fish were still alive. Unfortunately, many fish had mouth deformities. Twenty six fish with strong mouth deformities (scores 2 and 3 in Kocour et al., 2006) were discarded. Out of the remaining 419 fish, we eliminated 25, which were the only offspring of their sire, as they have a very low interest for heritability estimation. The remaining fish were put back in the pond, and harvested in October 2005.

A total of 331 fish, 218 without mouth deformities and 123 with mild mouth deformity (score 1 in Kocour et al., 2006), were processed in November 2005 in the following way: fish were killed by hitting on the head, and mean muscle percent fat was evaluated by Torry Fish Fatmeter®. The percent fat value was the mean of four measurements on the left side of fish body performed evenly on the whole surface (two on the back part and two on the body cavity part). Although we did not estimate the repeatability of the fatmeter values, the correlation between each of the four measurements on the same fish ranged from 0.77 to 0.87. Additionally, the estimated correlation between the mean percent fat and a chemical measurement of fillet fat content on 92 randomly sampled fish was 0.62. After the fat percent evaluation the fish were measured for standard length, head length, (maximal) body height, (maximal) body width and body weight. Then the scales were removed, and the fish were gutted and filleted. Gonads were isolated and skin from both fillets was separated. The

head was cut from the skeleton by a circular cut in front of the pectoral fin girdle so that the fin girdle remained on the body (Gela and Linhart, 2000). Finally, all different body parts were weighed to nearest 0.1 g. The following traits, important in common carp culture, were calculated from the data:

- 1) Relative head length = head length / standard length
- 2) Relative body height = maximal height / standard length
- 3) Relative body width = maximal width / standard length
- 4) Percent processed body = (fillet weight + skin weight + weight of skeleton with remnants) / body weight × 100
- 5) Percent fillets with skin = (fillet weight + skin weight) / body weight × 100
- 6) Percent fillets without skin = fillet weight / body weight × 100

Relative head length, relative body height and relative body width are traits describing body shape, percent processed body and both percent fillets represent main edible parts. Percent fillets without skin is a fundamental trait, as it represents pure flesh with intramuscular bones only. However, fillets without skin are not yet the most demanded commodity on the market of common carp.

2.4. Statistical analyses

For each of the 331 G2 fish slaughtered, the G1 sire and the dam were unambiguously identified. For 280 G2 fish, the G1 sire identified was itself unambiguously assigned to a G0 sire and dam. For 51 G2 fish, the G0 sire and dam were not identified, and therefore the pedigree was only one generation deep. No adequate phenotypes were available for the G0 and G1 fish, and they were only used to complete the pedigree. This practice resulted in a design with 264 full-sibs families produced from 92 sires and 8 dams, with an average full-sibs family size of 1.25 (range 1–5), an average paternal half-sibs family size of 3.6 (range 2–11) and an average maternal half-sibs family size of 41.5 (range 29–52).

The pedigree was used together with the performance records and the fixed effects matrix to estimate heritabilities and genetic correlations with VCE 5.1.2 (Kovac and Groeneveld, 2003). A multivariate animal model included sex (male, female) and mouth deformity status (normal, deformed) as fixed effects, and animal genetic factor as a random effect. Significance of fixed effects was assessed with a single generation

sire and dam model without interactions, using SAS[®]-Mixed procedure. Phenotypic correlations were estimated using SAS[®] as Pearson correlation coefficients between the variables tested, and their standard errors were estimated according to Sokal and Rohlf (1981).

3. Results

3.1. Phenotypic data and fixed effects

The fish were on average 1549 g (Table 1) with a coefficient of variation of 22% for body weight. Sex had a significant effect on all traits except relative body height and percent fillet without skin. Females were larger and fatter than males, and they had higher percent processed body and percent fillet with skin. Percent fillet without skin displayed a pattern similar to fillet with skin, but the difference did not reach statistical significance. This is likely because unequal skinning among individuals increases error variance. Mouth deformities had a significant impact on all traits except relative head length and percent processed body. Fish with deformed mouth were lighter, thinner and had lower fillet yields than normal fish.

3.2. Heritability estimates

Heritabilities of the traits studied are listed in Table 2. Body weight, standard length, percent fat and relative head length had high heritability (>0.5), relative body height, percent processed body and fillet yields had medium heritability (0.2–0.5), and relative body width had low heritability (<0.2).

3.3. Correlations between quantitative traits

As usual there were very high genetic and phenotypic correlations between length and body weight ($r_A=0.97$, $r_P=0.92$). Correlations between percent fillet with skin and without skin were also very high ($r_A=0.96$, $r_P=0.87$), but interestingly the heritability of fillet yield with skin is almost twice as high as that of fillet yield without skin (0.38 vs. 0.21). The rest of phenotypic correlations are mostly moderate (below 0.5 in absolute value) except between yields. We found relatively high positive genetic correlations between body size (standard length and body weight) and percent fat (0.71 and 0.59, respectively), favourable correlations between body size and percent processed body (0.69 with standard length and 0.74 with body weight) and body size and fillet yields (0.50–0.77). Genetic correlations between body size and body

Table 1
Growth and processing traits of common carp at the end of the 3rd growing season ($n=331$), with effects of sex and mouth deformity

	Sex effect			Mouth deformity effect			
	Mean±S.D.	Male LS mean±S.E.	Female LS mean±S.E.	Sig. level	Normal LS mean±S.E.	Deformed LS mean±S.E.	Sig. level
	Standard length	352.8±27.0	345.6±2.8	355.0±2.6	***	355.0±2.5	345.6±2.9
Body weight	1549±341.9	1449±35.1	1571±33.1	***	1602±32.1	1419±36.4	***
Mean % fat	5.20±1.91	4.57±0.20	5.58±0.19	***	5.38±0.19	4.77±0.21	***
Relative head length	0.292±0.015	0.289±0.002	0.294±0.001	***	0.293±0.001	0.291±0.002	NS
Relative body height	0.398±0.022	0.396±0.004	0.397±0.004	NS	0.400±0.003	0.394±0.004	**
Relative body width	0.204±0.011	0.202±0.001	0.200±0.001	*	0.205±0.001	0.201±0.001	***
% Processed body	67.2±2.2	66.2±0.2	67.9±0.2	***	67.1±0.2	67.0±0.2	NS
% Fillet with skin	41.1±2.2	40.6±0.2	41.4±0.2	**	41.2±0.2	40.7±0.2	*
% Fillet without skin	32.1±2.0	31.8±0.2	32.2±0.1	NS	32.3±0.2	31.7±0.2	*

*** $P<0.001$, ** $P<0.01$, * $P<0.05$, NS: not significant.

shape (relative head length, relative body height and relative body width) were weak to moderate. Relative head length seems quite informative, as it had strong negative correlations (-0.7 to -0.9) with percent fat, percent processed body and percent fillet yields (Table 2).

4. Discussion

4.1. Phenotypic data

The fish reached commercial size in three growing seasons, as usual in the Czech Republic. The phenotypic variation in fish body weight was within expected range, with a CV of 22%. Sex had a significant effect on growth, but also on yields. The effect was especially important on percent processed body (66.2% in males, 67.9% in females), mostly caused by the fact that more males than females mature in the 3rd spring, giving a gonadosomatic index of 2.4% in males vs. 1.1% in females. Similar results were found in the three summers old common carp by Kocour et al. (2005b) in a study comparing all-female and mixed sex populations. It shows one possible practical use of all-female populations at that age in the local climate. However, this advantage of females disappeared at four-summer-old carps due to female maturation leading to an increase of their gonadosomatic index (Kocour et al., 2005b).

As expected, there was a negative effect of mouth deformities on growth. This was already demonstrated at earlier growth stages (Kocour et al., 2006) together with the fact that mouth deformities are brought about essentially by (unidentified) environmental factors. Here, we assumed that the mouth deformity had a purely environmental effect on the traits studied, which can be accounted for by its inclusion as a fixed effect in the statistical model. The syndrome provoked by the mouth deformity was quite coherent with a lower ability to feed. Deformed fish had lower body weight and length, lower percent muscle fat, thinner body (lower relative body height) and lower fillet yields.

4.2. Genetic parameters

First of all, we would like to point out that due to the single batch rearing of the progenies from the larval stage, genetic parameters are not likely to be strongly biased by common environmental effects.

Still, the heritability estimate obtained for body weight is high (0.70). All previous studies in common

Table 2
Heritabilities (bold, diagonal), genetic correlations (below diagonal) and phenotypic correlations (italics, above diagonal) for growth and processing traits in common carp after 3rd growing season ($n = 331$)

	Standard length	Body weight	Mean % fat	Rel. head length	Rel. body height	Rel. body width	% Processed body	% Fillet with skin	% Fillet without skin
Standard length	0.69 ± 0.10	0.92 ± 0.02	0.34 ± 0.05	-0.18 ± 0.05	-0.17 ± 0.05	-0.03 ± 0.06	0.35 ± 0.05	0.46 ± 0.05	0.42 ± 0.05
Body weight	0.97 ± 0.01	0.70 ± 0.08	0.40 ± 0.05	-0.05 ± 0.06	0.15 ± 0.05	0.28 ± 0.05	0.35 ± 0.05	0.43 ± 0.05	0.41 ± 0.05
Mean % fat	0.71 ± 0.16	0.59 ± 0.14	0.58 ± 0.09	-0.14 ± 0.05	-0.01 ± 0.06	0.17 ± 0.05	0.20 ± 0.05	0.37 ± 0.05	0.20 ± 0.05
Relative head length	-0.36 ± 0.12	-0.26 ± 0.12	-0.82 ± 0.15	0.54 ± 0.12	0.42 ± 0.05	0.39 ± 0.05	-0.09 ± 0.05	-0.25 ± 0.05	-0.23 ± 0.05
Relative body height	-0.14 ± 0.17	0.08 ± 0.11	-0.63 ± 0.08	0.53 ± 0.11	0.32 ± 0.06	0.74 ± 0.04	0.02 ± 0.06	-0.15 ± 0.05	-0.11 ± 0.05
Relative body width	0.18 ± 0.23	0.34 ± 0.11	-0.14 ± 0.18	0.18 ± 0.21	0.62 ± 0.12	0.15 ± 0.05	0.00 ± 0.06	-0.11 ± 0.05	-0.08 ± 0.05
% Processed body	0.69 ± 0.12	0.74 ± 0.12	0.66 ± 0.15	-0.75 ± 0.13	0.00 ± 0.17	0.10 ± 0.22	0.28 ± 0.06	0.63 ± 0.04	0.54 ± 0.05
% Fillet with skin	0.77 ± 0.10	0.73 ± 0.11	0.76 ± 0.10	-0.83 ± 0.08	-0.14 ± 0.16	0.00 ± 0.22	0.79 ± 0.13	0.38 ± 0.09	0.87 ± 0.03
% Fillet without skin	0.50 ± 0.19	0.60 ± 0.16	0.64 ± 0.11	-0.86 ± 0.05	-0.07 ± 0.16	-0.14 ± 0.21	0.75 ± 0.18	0.96 ± 0.04	0.21 ± 0.07

All estimates given ± standard error.

carp have found lower estimates, between 0 and 0.48 (Nenashev, 1966; Nenashev, 1969; Nagy et al., 1980; Smíšek, 1981; Tanck et al., 2001). Our previous heritability estimates for juvenile weight, which was done on a sample of the G1 population, was also lower (0.30–0.33; Vandeputte et al., 2004). The present dataset is small (331 fish) and this may be thought as a reason for lack of precision. Nevertheless, the standard error estimate shows that the heritability of body weight clearly differs from zero.

The heritability of percent fat obtained using Torry Fish Fatmeter was also high (0.58), and in accordance with what is seen in other species (0.55 in chinook salmon, Jopson et al., 2002; 0.36–0.72 in rainbow trout, Chevassus et al., 2002; 0.25 realized h^2 in rainbow trout, Quillet et al., 2005). This high heritability shows that selection for increased or decreased lipid content is feasible. However, the favourable direction for selection is not necessarily clear, as consumers may ask for less fat products, but fat deposits may be an asset for overwintering survival in carp (Steffens, 1996). Two-trait selection for weight and lower fat content would also decrease the genetic gain to be obtained for growth in carp, as there is a positive genetic correlation (0.59) between percent fat and body weight. The heritability of relative body height (0.32) is of similar magnitude to the realized heritability given by Ankorion et al. (1992) (0.47 for upwards selection, 0.33 for downwards selection) and to an average 0.36 heritability of body shape in rainbow trout (Kause et al., 2004). This confirms the ability to select for body shape in common carp, which is also confirmed by the fact that another body shape parameter, the relative head length, seems to be heritable ($h^2=0.54$).

Interestingly, percent processed body and fillet yields were heritable ($h^2=0.21$ –0.38). This is in accordance with data on other species ($h^2=0.45$ for gutted yield and 0.33 for fillet yield in rainbow trout, Kause et al., 2002; 0.36 for gutted yield in rainbow trout, Gjerde and Schaeffer, 1989; 0.30 for fillet yield in black sea bream, Doupe and Lymbery, 2005). The heritability of percent fillet with skin is higher than that of percent fillet without skin (0.38 vs. 0.21), but the genetic correlation between the two is close to unity (0.96). This is probably due to a lower repeatability of the fillet without skin due to variations in the efficiency of skin removal. Percent fillet without skin is commercially more important trait. To increase this trait, it would be however advisable to select on percent fillet with skin. These results show the theoretical feasibility of selection for improved yields in common carp. However, because the evaluation of fillet yield is lethal for breeding candidates, this implies that

selection should be done on the basis of slaughtering traits of their relatives. This practise would require identification of families, either by separate rearing or by genotyping. In this context, the highly negative genetic correlation between relative head length and fillet yield without skin (–0.86) is interesting, because relative head length can easily be measured on the live breeding candidates. Moreover, relative head length is more heritable than fillet yield, so for the same intensity, the relative response between indirect selection on relative head length and direct selection on fillet yield should be $r = \frac{h_X r_A(X, Y)}{h_Y} = 1.37$ (Falconer, 1960). Therefore, indirect selection for reduced relative head length should be effective in improving fillet yield in common carp. Cibert et al. (1999), with more sophisticated morphometric analysis but with a smaller sample size ($n=42$) proposed that the best morphotype for fillet yield should have a lower anterior dorsal height. It does not seem to be the case here, as both genetic and phenotypic correlations between relative body height and fillet yields, although negative, are small and hardly different from zero. One interesting point is the fact that genotypes with high fillet yield seems to have also high muscular percent fat ($r_A=0.64$), despite a quite low phenotypic correlation ($r_P=0.20$). Therefore, selection for increased fillet yield may also lead to fatter animals, which may or may not be an advantage as already pointed out.

The genetic correlation between body weight and relative body height was weak (0.08), and the one between body weight and relative body width was positive but moderate (0.34). Therefore, selection for increased body weight should slowly change body shape towards more “rotund” fish, similar to what was demonstrated in rainbow trout (Kause et al., 2003). If selection is undertaken on standard length, the effects on shape should be even reduced ($r_A=0.18$ with relative body width and –0.14 with relative body height). Additionally, rotund fish in common carp are not necessarily unwanted and may even sometimes be specifically selected for.

Finally, we would like to stress that although our results are interesting as they are the first data on genetic parameters of processing and quality traits in the common carp, they should be taken with care, because of the small sample size used. Moreover, there are many strains of common carp, and these results are only valid for the HSM strain held at USB RIFCH, Vodňany. We feel that the heritability estimated for body weight is very high, and not fully compatible with previous estimations as well as the failure of mass selection for growth reported by Moav and Wohlfarth (1976).

Therefore, the current results should be validated by a selection experiment.

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