Heritability estimation for growth-related traits in juvenile wild common carp (Cyprinus carpio L.) in the south of Caspian Sea

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Abstract
Wild common carp of the Caspian Sea is one of the important fish in the north of Iran, which has been introduced to inland aquaculture. Its genetic characteristics are investigated to set up for breeding programs that are still unknown. Using wild common carp in aquaculture is limited for uncontrolled variation in performance of fish derived from undomesticated breeders. A 7×7 factorial mating design was employed to examine genetic effects of dam and sire on growth and heritability of growth-related traits based on dam half-sib and sire half-sib families. Breeders were obtained originally from the Caspian Sea. A total of 1470 offspring were raised in 49 separate fiberglass tanks with common conditions. Body weight and length were recorded at three different ages of rearing. Estimation of heritability of weights ranged from 0.23±0.05 to 0.30±0.07 for dams and ranged from 0.15±0.03 to 0.26±0.04 for sire. Similar results were obtained with length but with less variation (0.24±0.02 to 0.26±0.01 for dams and 0.20±0.02 to 0.22±0.03 for sire). The broad-sense of weight and length are 0.23±0.04 and 0.23±0.01 respectively. The dam and sire effect on growth were significant, indicating the presence of variations among the native fish of the Caspian Sea. Therefore, it is suggested to use a length for selection that is more reliable and less affected by environment than weight at any time of growth and in respect of high phenotypic correlation (0.87-0.95) observed between weight and length. In the present study we even used wild common carp and the heritability was not higher than the domesticated common carp. It means the genetic variation for quantitative traits, for common carp of the Caspian Sea are low or it may be under pressure of inbreeding which should be studied further.

Keyword: Caspian Sea, Common carp, Heritability, Genetic Parameter

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Introduction
At the present, there is a considerable attention in adaptation of genetically wild fish such as wild common carp of the Caspian Sea. This type of fish is preferred for disease resistance and also in the market for its appearance and delicious taste. However, in breeding program management, knowledge of the genetic parameters is required for these species. Therefore, genetic information such as estimation of heritability would be collected in order to select the best breeder. Most of the population of common carp especially in Asia did not undergo the systematic selection program (Hulata, 1995), while nowadays, worldwide production of common carp is 3.2 mil.tons which is more than twice the Salmonids production (FAO, 2005).

Variation in growth rate, hatchability and survival rate of larvae are characteristics of all aquatic animals. Variation is due to various environment factors as well as genetics of fish. Several authors have reviewed the work carried out on genetic improvement in this species, including hybridization, crossbreeding, genome manipulation and selective breeding (Wohlfarth et al., 1987; Hollebecq and Haffray, 1994; Hulata, 1995, Abdolhay et al., 2010). Most papers concluded that the possible use of crossbreeding is the main way to improve this species in contrast to Rainbow trout (Yousefian, 2008), and to a quite low potential for selective breeding, except for disease resistance (Vandeputte, 2003). However, in some other genetic studies moderate levels of genetic variation for growth rate of common carp have been revealed allowing its genetic improvement \((h^2>0.2\), Nenashev, 1966; Nagy et al., 1980). The variables are high among experiments that range from 0.21 (Nenashev, 1969) to 0.58 (Bongers et al., 1997) for weight. This high difference may be due to the design used (sire or dam effects), or could be biased due to the small number of breeders used, and/or the inability to separate common environments and genetic effects (Vandeputte et al., 2008). The differences in heritability also may be due to the differences in domesticated strains of common carp used for investigation. There are not any or very few references on the genetic analysis of wild carp of the Caspian Sea.

The methods were employed in fish breeding to establish genetic parameters, more precise results are obtained by the complete bifactorial dispersion complex resulting from diallele crosses of different degrees of complexity. The external fertilization of eggs in common carp and the high fertility of females facilitate the simultaneous conduction of a large number of crosses. To introduce a wild common carp of the Caspian Sea in aquaculture, genetic parameters of this fish should be identified. Heritability and other expected genetic parameters, response for growth lead us for decision making in the breeding program of this species.

Materials and methods
Experimental fish
The common carp male and females were obtained from wild stock from the Caspian Sea which had been maintained at Fish restocking center of S.R. Sari. For experiments 7 healthy and good shape females ranging from 1.5-2 kg and male of 1-1.5 kg were selected. Both males and
females were induced to spawn by injection of carp pituitary gland collected in the previous winter with a dosage of 2.5 mg/kg for female and 1.25 mg/kg for male. The female were injected two times with 12 hour intervals (10 percent of dosage as initial dose and 90 percent as final dosage). Eggs of fish were collected by stripping after 11 hours of final hormonal injection. A part of the eggs was divided into 7 equal aliquots (30 gr), and placed in 7 separate bowls, milt of each fish was stripped in the laboratory tub. 1 ml aliquots of milt were then added to each of the 7 bowls. After fertilization, the 49 batches of fertilized eggs were incubated into 8 lit glass-jars, separately. Hatching took place 48 hours post fertilization. All newly hatched larvae were then placed in 220 lit plastic-jars and kept and reared up to 4 days. 

**Growth of fish**

The newly larvae were placed in a 2×2×0.5 m$^3$ fiberglass tank, and fed with natural food of rotatoria and naplius of daphnia. After the fry had been fed, they were again fed a high-protein meal-type diet (45% protein). Two hundred larvae were maintained under the same condition up to 40 gram.

**The maintenance of fish**

During the rearing of fish, water quality was controlled every day. Salinity was fixed 1-2 ppt, using well water and running water after mechanical filtration. Dissolved oxygen levels were maintained at 6-8 mg/L, by providing aeration with central electrical blowers and changing the water (0.3 L/s). Water temperature was controlled and maintained at 23±2 °C. The fish were weighted and total lengths were measured at different periods of time. (1st of summer, 1st of winter and 1st of spring).

**Experimental design**

A classical factorial design known as the North Carolina Design II (Roff, 1997), in which 7 brood female were crossed with the same number of males was employed to produce full-sib, half-sib and unrelated progeny. Offspring produced by one female from different males are half-sibs with respect to each other while the individual within each given offspring group treatment is a full-sib. All trails assessed were evaluated according to the following model:

$$Y_{ijm} = \mu + \text{dam}_i + \text{sire}_j + (\text{dam} \times \text{sire})_{ij} + e_{ijm}$$

Where $ijm$ is an individual observation, $\mu$ is the overall mean, $\text{dam}_i$ is the random effect of the $i$th dam, $\text{sire}_j$ is the random effect of the $j$th sire, $(\text{dam} \times \text{sire})_{ij}$ is the random interaction effect of the cross between the $i$th dam and $j$th sire and $e_{ijm}$ is the random residual.

Significance of random genetic and fixed effects of the environment was determined by analysis of variance (ANOVA), variance components and their standard deviation was established using SPSS, version 16.

In the preliminary analysis of this work, the sire × dam interaction effect was not significant; therefore this effect was not included in the model used to generate the present estimation (Previously Backer, 1984 had the same results). The additive genetic Variance $\delta_A^2$ was estimated for the sire component of variance ($\delta d^2$).

The relation: $$\delta_A^2 = 4 \times \delta_s^2$$ (Backer, 1984), and heritability was calculated as the ratio:

$$h^2_s = \frac{\delta_A^2}{\delta_s^2}$$

Where $\delta s2$ is the between sires variance, $\delta d2$ is between dam variance and $\delta e2$ the residual variance. The same is for dam
and the average is used to estimate heritability in general in this experiment.

**Results**

The mean weight and length of offspring at different ages of growing based on diallele crosses are presented in Table 1. The average over all weight of fish at three sampling times was 5.9±1, 35.6±8 and 84.7±22g, respectively. Also, the average lengths of fish during these three times were 9.6±1, 14.7±2 and 19.2±3cm, respectively. A total of 1470 offspring were reared under the same conditions, the overall survival was approximately 94% during 12 months growth.

*Genetic parameters of weight and length*

The effect of sire and dam as well as the interaction effects are presented in Table 2. Effect of dam and sire on weight and length at 1st spring were highly significant. The data of sire, dam and total heritability estimated for weight and length gain at different ages are presented in Table 3.

| Table 1: Mean and standard deviation of weight and length at 3, 6 and 12 months |
|---------------------------------------------|-----------------|-----------------|-----------------|
| Sampling                                   | 1th Summer Weight | 1th Winter Weight | 1th Spring Weight |
| mean± SD                                   | 5.88±1.36       | 35.65±8.1       | 84.66±21.6       |
| Range                                      | 3.4-9.5         | 7.3-11.8        | 11.1-18.8        |
| No. of offspring                           | 1470            | 1470            | 1470            |

| Table 2: Probability of significance of genetic factors by analysis of variance |
|---------------------------------------------|-----------------|-----------------|
| Parameters                                  | Weight          | Length          |
| Effect of Dam                               | 0.003           | 0.005           |
| Effect of Sire                              | 0.001           | 0.004           |
| Effect of Dam×Sire                          | 0.277           | 0.324           |

| Table 3: Heritability estimated for growth rate matured at three intervals during the rearing period. |
|---------------------------------------------|-----------------|-----------------|-----------------|
| Variable                                    | H²♀♀           | H²♂♂           | H²♀♂♂♂♂         |
| Weight at 1st Summer                        | 0.30±0.07       | 0.26±0.04       | 0.28±0.05       |
| Weight at 1st Winter                        | 0.23±0.05       | 0.17±0.03       | 0.20±0.04       |
| Weight at 1st Spring                        | 0.26±0.04       | 0.15±0.03       | 0.21±0.03       |
| Length at 1st Summer                        | 0.26±0.01       | 0.22±0.02       | 0.24±0.01       |
| Length at 1st Winter                        | 0.24±0.02       | 0.22±0.03       | 0.23±0.02       |
| Length at 1st Spring                        | 0.25±0.02       | 0.20±0.02       | 0.23±0.02       |
Discussion

Literature resources focusing on heritability of quantitative traits were quite poor due to unsuitable experimental models used (low number of breeders, no replication) or missing methodology of testing in details (Vandeputte et al., 2003).

A few problems in the setup of experiments were reported in estimating genetic parameters in carp arising from the inability of fish tagging at hatching and subsequent need to rear families separated in many different ponds. Thus, the increment of environmental variance confounds with genetic effects. (Vandeputte, 2003).

To obtain the genetic and environmental effects microsatellites are used for progeny assay as Vandeputte et al., 2004 suggested eliminating the need for separate rearing full-sib or half-sib families. That can confound genetic and environmental effects but this technique is very expensive and cannot be applied for a number of research centers. Therefore for obtaining the genetic parameter of wild common carp of the Caspian Sea, the classical old method was used (Nenashev, 1966; Kirpichnikov et al 1993; Tanck et al., 2001). The maximum number of the offspring with triplicates were reared in separate rearing equal facilities in order to reduce the bias of measuring genetic parameters.

The dam and sire had significant effects on both weight and length after the fish start off their growth trials (35 g) but it was greater when fish had reached an average weight and length of 100 g and 100 cm respectively. The significant genetic variations of growth trials in this study indicate that selective breeding could be implemented at juvenile stage.

The heritability for weight estimated in this study was in the range of (0.15-0.30) which is in the usual range of common carp (Nenashev, 1966, 1969; Smisek, 1981; Moave and Wohlfarth, 1976, see review by Vandeputte, 2003), but lower than those of Vandeputte et al, 2008 that is 0.21 to 0.44 in total for domesticated common carp. Actually, there are two subspecies of common carp from Europe and Asia (Balon, 1995). Within each subspecies partially in the European one, the genetic distance between population is low (Desvignes et al., 2001). The population genetic studies showed lower variables of domestic carp when compared with wild strains (Kohlmann et al., 2003). Vandeputte, 2003 stated, the lower variability of domesticated strain indicates that in many cases, they were propagated with a small effective number of breeders, resulting in some inbreeding. Although it has to be confirmed with estimates of genetic variation for quantitative traits, this initial inbreeding in the domesticated
strains could somehow hamper the possibility for genetic gain from selective breeding.

The heritability of wild common carp was not higher than the domesticated common carp. It means the genetic variation for quantitative traits, for common carp of the Caspian Sea are low or it may be under pressure of inbreeding which should be studied further. On the other hand, the individual score concluded for estimating genetic parameters was not enough.

The dam and sire effects on early growth are very important in breeding programs. The dam and sire had significant effects on early growth which have been reported for some cultured fish species (Wang et al., 2006), including common carp (Vandeputte et al., 2004). The pronounced effect of dam on early growth was hypothesized that this could be due to a maternal phenotypic effect (Herbinger et al., 1995; Wangila and Dick, 1996). The maternal effect was greater than sire (Table 2) and even greater than some previous works in common carp (Vandeputte et al., 2004). This difference may be due to the design of the experiment. The dam effect is expected to include both additive-genetic effects and maternal phenotypic effects as Falconer and Mackay, indicated previously in 1996. Gjerdrem (1992), in a review of eight reported heritabilities found that those estimated from dam components of variance, suggesting strong common environmental and /or non-additive genetic effects.

The results of wild common carp are in contrast with those of Gjerdrem (1992), the average dam effect was 0.21 higher than sire. Estimation of correlation between trials was analyzed. Correlation between two trials and among one trial at different stages of growing will help the breeders evaluate the stocks (Su et al., 2002). In this study, correlation between weight and length for all ages was quite high and near to unit, indicating highly positive correlation between genetic parameters related to weight and length (0.9 ±0.03). The correlation between different stages of growth was also very high (0.87-0.95). The correlation between growth at different stages before maturation has been reported in culture fish species of rainbow trout (Su et al., 2002) and European sea bass (Saillant et al., 2006). This indicated that growth estimated at early stages could be used as a predictor of growth at later stages. The heritability estimates (up to 0.3) obtained in this study indicates that a substantial fraction of the selection differential would be expected to be gained in offspring of selected parameters.

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تخمین وراثت پذیری برای صفت های وابسته به رشد در ماهی کبور
وخشی جنوب دریای خزر
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خلاصه
ماهی کبور وخشی با پیمودن دریای خزر، یکی از مهم‌ترین ماهیان شمال ایران محسوب می‌گردد که به آبی‌پروری معرفی شده است. در تحقیق حاضر خصوصیات زنبیلی‌ای آن مورد بررسی قرار گرفته که در برنامه‌های اصلاحی مورد استفاده قرار گیرد. استفاده از کبور وخشی در آبزی پرورش به دلیل تتنوع کننده نشده مولدین محدد می‌باشد. طرح تلاقی‌ای فاکتوریل 7×7 برای آزمون اثر مولدین نر و ماده بر روی رشد و وراثت پذیری این صفت بر اساس خصوصیات برادر و خویه‌های ناتین انجام گرفت. مولدین از دریای خزر صید گردیده‌اند و پس از تلاقی 74 مولدین نر و ماده، فرزندان آن‌ها به تعداد 1470 بچه ماهی در 64 حوضه‌ی فرعی گل‌السیاه جاده‌ای بنا شرایط یکسان پرورش یافتند. طول و وزن بدن در سه دوره رشد نهایی ثبت گردید. تخمین وراثت پذیری وزن در 0.43±، 0.70± و 0.70± برای ماهیان ماده و 0.23± و 0.43± برای ماهیان نر به دست آمد. وراثت پذیری کل وزن و طول به ترتیب 0.43± و 0.43± به دست آمد. اثر مولدین نر و ماده بر رشد معنی‌دار بوده و نشان دهنده وجود تتنوع در ماهیان وخشی کبور دریای خزر می‌باشد. در این تحقیق بیشترین گردیده برای انتخاب ماهیان بر اساس طول ماهی انتخاب صورت گرفته و برای صفت طول نسبت به وزن کمتر تحت تأثیر شرایط محیطی قرار داشته و از طرف دیگر هم‌گیانه شدنی‌ی بین طول و وزن مشاهده شده است (0.43±). در مطالعه حاضر علی رغم اینکه از ماهیان وخشی کبور استفاده گردیدن، ولی میزان وراثت پذیری آن در مقایسه با ماهیان پرورشی خیلی زیاد نبوده است. این نتیجه به معنای که تتنوع زنبیلی‌ای برای صفت‌های کمی برای ماهی کبور دریای خزر کم بوده و نیاز باعث هم‌خوانی قرار دارد و با پدیده‌ی مشابهی بررسی قرار گیرد.

واژگان کلیدی: دریای خزر، ماهی کبور، وراثت پذیری، خصوصیات زنبیلی

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