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Genetic Analysis of Shape in Trout, using image analysis

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ABSTRACT: We used digital images of rainbow trout (lateral view) to fit an ellipse around the circumference of the fish. The values for L and H, obtained from the ellipse, were used to calculate ellipticity as (L-H)/(L+H), and the surface area of the fish as π*1/2 L*1/2H. Heritability of ellipticity and surface area at age 8 months was 0.23 and 0.21. Surface area had near-unity genetic correlation with body weight at same age. Genetic correlations of ellipticity with body weight and surface area were -0.55 and -0.56. Genetic correlation of ellipticity with harvest weight at 14 months was -0.49. Estimates of ellipticity are comparable with those of Nile tilapia and common sole. We conclude that when shape is important, ellipticity should be included in the breeding goal, with a weight that reflects the desired direction of change in shape.
Keywords: fish; shape; ellipse; image analysis; genetics

Introduction

The main trait for genetic improvement in fish breeding programs is increased harvest weight, or ‘growth’. Results from long-term breeding programs have shown that considerable improvement in growth can be achieved. In several species, the increase in growth averages ~14 % per generation (reviewed in Gjedrem et al. (2012)). In the GIFT breeding program (genetically improved farmed tilapia) for Nile tilapia, the increase in harvest weight was ~7% per generation. After 20 generations of selection, commercial harvest weight had increased from 200 grams to over 1.4 kg, after 7-9 months of on growing (reviewed in Komen and Trong (2014)).

Selection for growth in livestock species has led to marked changes in body conformation. In pigs, beef cattle and broilers, selection for growth and/or slaughter weight has led to correlated responses in carcass length and muscle development (Hill (2008)). In fish, shape can be an important trait when fish are sold whole. Selection for growth is suspected to result in correlated responses in shape but evidence is only anecdotal. Simple indicator traits that can accurately predict a change in shape due to selection are currently lacking. Previously, we showed that the shape of common sole (*Solea solea*) can be approximated by using digital image analysis to fit an ellipse which follows the circumference of the image of a fish (Blonk et al. (2010)). Ellipticity is defined as (L-H)/(L+H), with L the long axis and H the short axis of the corresponding ellipse. Values for Ellipticity vary between 0 and 1, with higher values indicating more elongated shapes. Ellipses can also be used to calculate surface area, which can be used as a predictor of body weight. The objective of this study was to estimate genetic parameters for ellipticity and surface area in rainbow trout. Estimates of ellipticity were compared among three commercial species, common sole, Nile tilapia and rainbow trout. Results show that both ellipticity and surface area are traits with moderate heritability. The magnitude of the correlations with harvest weight suggests that ellipticity and surface area can be useful traits to include in a breeding program.

Materials and Methods

Data. We used data from an experiment described in detail in Sae-Lim et al. (2013). In brief, experimental fish were produced by Troutlodge (USA) by mating 58 sires with 100 dams and randomly selecting 25 eggs from each dam. Eggs were pooled and shipped to a commercial farm in Germany where they were communally reared under commercial conditions. At 6 months after hatching, fish were tagged, weighed and stocked together with untagged fish in an outdoor pond. At age 8 months (avg. weight 64.1 g) tagged fish were sorted, photographed (lateral view) and weighed. On-growing continued until fish were harvested at an age of 14 months (avg. weight 376.4 g) and final harvest weight was taken. Pedigree of experimental fish was reconstructed by genotyping each fish and all parents with 9 microsatellite markers. After pedigree reconstruction, data were available for 2,091 fish of age 8 months and 1,992 fish of age 14 months. The average number of fish per sire ranged from 30 to 35; the average number of fish per dam ranged from 17 to 20.

Shape. The digital image of each trout was used to fit an ellipse around the circumference of the fish (lateral view) using ImageJ software (Rasband (2008)). The values for L and H, obtained from fitting the ellipse, were used to calculate ellipticity as (L-H)/(L+H), and the surface area of the fish as π*1/2 L*1/2H.

Statistical analysis. Heritability, phenotypic (r_p) and genetic (r_g) correlations were estimated using restricted maximum likelihood in a multivariate animal model (ASReml; Gilmour et al. (2009)). Each trait was modeled as:

$$Y_{ij} = \mu + A_i + FS_j + e_{ij},$$

where y_{ij} is the observation of the *i*th individual from the *j*th full-sib family, μ is the overall mean and A is the random additive genetic effect of the *i*th animal. *FS* is the random full-sib common environmental effect, modelled without a pedigree, to correct for environmental effects common to full-sibs, e.g. incubator effects and environmental maternal effects. *e* is the random residual term. To estimate genetic correlations, the full-sib effect was excluded from the model. Potential selection bias due to selective mortality was accounted for by including body weight at tagging as reference trait (Sae-Lim et al. (2013)).

Results and Discussion

Figure 1 shows the distribution of Ellipticity at 8 months. Ellipticity values varied between 0.5 and 0.7 and followed a normal distribution. Mean ellipticity was 0.63. Estimates of heritability are given in Table 1. Heritability of ellipticity was moderate, 0.23 ± 0.09. Heritability estimate of surface area (0.21) was higher than of body weight at 8 months (BW_8: 0.18), but lower than heritability of BW at 14 months (0.23). Genetic correlations of ellipticity with BW_8 and surface area were similar and highly negative (Table 2: -0.55). Negative values indicate that at 8 months, fish with higher weights will have lower ellipticity values, i.e. they are 'rounder' or less elongated. Ellipticity values at 8 months also showed a negative correlation (-0.49) with harvest weight at 14 months. This suggests that fish with lower ellipticity values at 8 months will have higher final harvest weight. Larger trout tend to be more round and breeders will favor more elongated salmon-like phenotypes for selection (Haffray et. al. (2013)). Kause et al. (2003) estimated the genetic correlation between body weight at 2 and 3 years and shape, defined as a categorical trait: 'slender', 'medium' or 'rotund'. Genetic correlations were positive (0.37 - 0.57) showing that larger fish will be more rotund. This correlation is undesirable when trout is marketed as whole carcass. Our results confirm the observations from Kause et al., (2003) and show that ellipticity can be used already at an early age to correct for shape during selection.

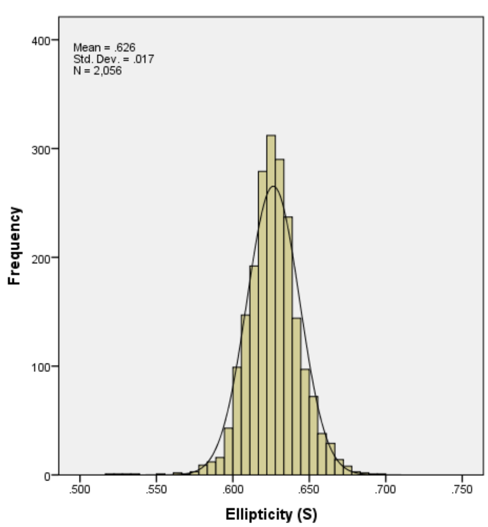

Figure 1. Distribution of shape measured with digital image analysis of rainbow trout during sorting.

Table 1. Genetic parameter estimates for traits measured at age 8 months and 14 months.

Trait	*V*a	*V*e	h^2	c^2
BW_8	51.0	216.4	$0.18^{\pm 0.11}$	$0.07^{\pm 0.04}$
$ELLIP_8$	0.0001	0.0002	$0.23^{\pm 0.09}$	$0.02^{\pm 0.03}$
$SURFS_8$	238824	820126	$0.21^{\pm 0.11}$	$0.05^{\pm 0.04}$
BW_{14}	1736.3	5087.1	$0.25^{\pm 0.10}$	$0.00^{\pm 0.03}$

*V*a = additive genetic variance, *V*e = residual variance, h^2 = heritability, c^2 = common environmental effect, BW_8 = body weight at 8 months, $ELLIP_8$ = ellipticty at 8 months, $SURFS_8$ = surface area at 8 months, BW_{14} = body weight at harvest (14 months). SE in superscript.

Table 2. Genetic (lower diagonal) and phenotypic correlations (upper diagonal).

Trait	BW_8	$ELIPS_8$	$SURFS_8$	BW_{14}
BW_8		$-0.33^{\pm 0.03}$	$0.95^{\pm 0.03}$	$0.64^{\pm 0.02}$
$ELLIP_8$	$-0.56^{\pm 0.11}$		$-0.44^{\pm 0.02}$	$-0.22^{\pm 0.03}$
$SURFS_8$	$0.99^{\pm 0.01}$	$-0.55^{\pm 0.11}$		$0.65^{\pm 0.02}$
BW_{14}	$0.73^{\pm 0.07}$	$-0.49^{\pm 0.13}$	$0.77^{\pm 0.07}$	

BW_8 = body weight at 8 months, $ELLIP_8$ = ellipticty at 8 months, $SURFS_8$ = surface area at 8 months, BW_{14} = body weight at harvest (14 months). SE in superscript.

Genetic correlation of body weight at 8 months was near unity with surface area, estimated from digital images. Furthermore, the genetic correlation of surface area with harvest weight was 0.77, higher than that of body weight at 8 months with harvest weight (Table 2). These results show that image analysis can be used instead of body weight to estimate and predict body weights. Image analysis is less prone to systematic errors than physical weighing, and can be automated with high throughput. Image analysis of young fish can be used to discard malformed fish, and for selection to correct for undesired shapes (Blonk et al. (2010)).

Table 3 summarizes estimates of heritability and genetic correlations between Ellipticity and harvest weight for rainbow trout, Nile tilapia and common sole. In sole and tilapia, ellipticity was measured at harvest. Results show that ellipticity is a trait with low to moderate heritability, moderately correlated to harvest weight. In both common sole and rainbow trout, genetic correlations with harvest weight are significant and negative. Negative correlations indicate that selection for higher harvest weight will change the shape of fish and make them more round. In case of sole, this is undesired as sole is marketed whole, and customers will pay lower prices for fish that do not have an ideal shape. Blonk et al., (2010) showed that including shape in the breeding goal resulted in a 13% reduction in response in harvest weight when zero change in shape was desired. The response in shape is different from what has been observed in Nile tilapia. In this study (Trong et al., ((2013)) ellipticity values were calculated from manual measurements on length, height and thickness. Ellipticity was then calculated along the three axes (mid-sagittal: L-H, transverse: L-T, frontal: H-T). Values are given in Table 3. Heritability of ellipticity is lower than for trout and sole, which could be due to the fact that manual measurements were used instead of image analysis. Genetic correlations indicate that Nile tilapia, selected for high harvest weight will develop a more elongated and rounder 'torpedo'-like shape. This response in tilapia is different from that observed in trout or sole. Juvenile Nile tilapia change shape as they reach sexual maturity, growing more in height than in length. However, selection has been for considerable larger weights at harvest at the same harvest age without any apparent correlated response in onset of sexual maturity (Komen and Trong (2014)). Hence, the observed correlation between shape and harvest weight might actually represent selection for a more juvenile phenotype.

Table 3. Summary of genetic parameter estimates for ellipticity in fish.

Species	h^2	r_g	r_p
Trout	$0.23^{\pm 0.09}$	$-0.49^{\pm 0.13}$	$-0.22^{\pm 0.03}$
Sole*	$0.34^{\pm 0.11}$	$-0.44^{\pm 0.25}$	$-0.30^{\pm 0.04}$
Tilapia**			
E_{L-H}	$0.08^{\pm 0.04}$	$0.47^{\pm 0.21}$	$0.12^{\pm 0.03}$
E_{L-T}	$0.14^{\pm 0.04}$	$-0.15^{\pm 0.22}$	$-0.17^{\pm 0.03}$
E_{H-T}	$0.08^{\pm 0.04}$	$-0.42^{\pm 0.21}$	$-0.22^{\pm 0.03}$

E= Ellipticity; r_g and r_p: genetic and phenotypic correlations of elliptitcity with harvest weight. SE in superscript. * = Blonk et al. (2010) ** = Trong et al. (2013)

Conclusion

The shape of a fish can be approximated by a set of ellipses, fitted along the three main axes: length-height, length-thickness, and height-thickness. Heritability for ellipticity and surface area in rainbow trout is moderate, indicating good prospects for selection. Image analysis can be advantageous over physical weighing in situations where it is difficult to get accurate measurements. Ellipses, fitted on digital images, can be used to calculate ellipticity and surface area, which in trout has near-unity genetic correlation with body weight (at same age). Genetic correlations between ellipticity and surface area or body weight are negative, showing that selection for higher weights will produce more round fish. In cases where shape has commercial value, it is recommended to include shape as a trait in the breeding goal, with a weight that reflects the desired direction of change.

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