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Quantitative genetic parameters for wild stream-living brown trout: heritability and parental effects

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Abstract

Adaptability depends on the presence of additive genetic variance for important traits. Yet few estimates of additive genetic variance and heritability are available for wild populations, particularly so for fishes. Here, we estimate heritability of length-at-age for wild-living brown trout (*Salmo trutta*), based on long-term mark-recapture data and pedigree reconstruction based on large-scale genotyping at 15 microsatellite loci. We also tested for the presence of maternal and paternal effects using a Bayesian version of the Animal model. Heritability varied between 0.16 and 0.31, with reasonable narrow confidence bands, and the total phenotypic variance increased with age. When introducing dam as an additional random effect (accounting for c. 7% of total phenotypic variance), the level of additive genetic variance and heritability decreased (0.12–0.21). Parental size (both for sires and for dams) positively influenced length-at-age for juvenile trout – either through direct parental effects or through genotype-environment correlations. Length-at-age is a complex trait reflecting the effects of a number of physiological, behavioural and ecological processes. Our data show that fitness-related traits such as length-at-age can retain high levels of additive genetic variance even when total phenotypic variance is high.

Introduction

An important question in evolutionary biology is if and how fast a population may adapt to a changing environment. Populations can respond to such challenges by either moving away, adjust the phenotype nongenetically (phenotypic plasticity), or evolve (Gienapp *et al.*, 2008). Populations that live in geographically constrained areas and do not have the option to disperse must consequently cope with the situation at hand. This is often the case for aquatic organisms such as freshwater fishes as they often are physically confined to a given lake or river.

Fish are in general phenotypically plastic, and traits may thus change rapidly. Phenotypic plasticity reflects the individual's ability to respond to different environmental conditions by changing phenotype (Schlichting & Pigliucci, 1998). Phenotypic plasticity is most often regarded as a nongenetic and thus not an evolutionary response (Schluter, 2000), yet plasticity itself can have a genetic basis and be adaptive (Via & Lande, 1985). Plasticity is often visualized as reaction norms (norm of reaction), defined as the phenotypes produced by a given genotype across an environmental gradient (Stearns, 1992). Different reaction norms among populations do indicate that such traits can evolve, even over short time scales (Haugen & Vøllestad, 2000; Jensen *et al.*, 2008; Darwish & Hutchings, 2009). Recent reviews point out that such microevolutionary adaptations commonly occur within a contemporary time frame (Hendry & Kinnison, 1999; Kinnison & Hendry, 2001; Hendry *et al.*, 2007). However, it is assumed that strong directional selection will erode genetic variance (Roff, 1997), and consequently that a lack of genetic variance may be an underappreciated cause of lack of adaptation (Blows &

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Hoffmann, 2005). Further, recent analyses tend to show that phenotypic changes in populations exposed to disturbances sometimes have a genetic basis but the contribution from phenotypic plasticity is often particularly important (Hendry *et al.*, 2008). Thus, before it is possible to give predictions about how populations may respond to changes in the selective regime, it is necessary to have reasonable estimates of genetic variance.

Given its prominent position in the breeder's equation, there are quite a number of estimates of trait heritability available for a large number of species (Roff, 1997). For fish, there are numerous heritability estimates for salmonid species used intensively for fish farming, sea ranching or stock enhancement programs, such as Atlantic and Pacific salmon (*Salmo salar* and *Onchorhynchus* spp) (Carlson & Seamons, 2008). Fewer estimates are available for species not used in aquaculture, even if more studies now are appearing (see Perry *et al.*, 2005; Thériault *et al.*, 2007). Brown trout (*Salmo trutta*) is a species that has not been studied to any large degree in this context. In a recent review, it was noted that only one estimate of heritability was available for brown trout growth rate (Carlson & Seamons, 2008), and this estimate was based on a study on the early growth (to the alevin stage) and performed in a hatchery (Vandeputte *et al.*, 2002). This is unfortunate, as the brown trout is a very common freshwater fish species in temperate Europe and parts of North America (Elliott, 1994) where it is a highly valued species for recreational anglers. It has also been distributed far and wide outside it original distributional area (Elliott, 1989).

Because the total phenotypic variance of a trait is composed of both genetic and environmental elements, quantitative genetic parameters such as heritability are context specific. It is therefore questionable whether estimates from controlled environments (common garden) experiments can be translated to natural situations (Weigensberg & Roff, 1996; Roff, 1997). Thus, it is unfortunate that very few estimates of heritability are derived from wild-reared populations – only 2% of available estimates for salmonid fishes are from wild populations (Carlson & Seamons, 2008). Overall, there are very few studies that successfully have estimated quantitative genetic parameters for wild populations of any taxon (review in Kruuk *et al.*, 2008). Moreover, there are also a number of nongenetic sources of resemblance between individuals (phenotypic plasticity, genotype-environment correlations) that make the distinction between adaptation and nongenetic responses difficult. These effects are very difficult to disentangle in wild populations. The estimation of quantitative genetic parameters such as heritability, and their relation to fitness in wild populations, should therefore be an important challenge for evolutionary biologists in general.

For traits such as length-at-age, a complicating factor is the presence of various parental effects. Especially, maternal effects, defined as any effect of a mother's phenotype on her offspring's phenotype, in addition to the additive genetic effect, are known to be important (Mousseau & Fox, 1998; Green, 2008). Such plastic effects, where environmental stimuli experienced by the mother lead to phenotypic responses in the offspring, can constitute an alternative inheritance system leading to adaptive evolution (Pfennig & Martin, 2008). In fish, it is well known that larger females may produce larger eggs with larger energy reserves and that juveniles hatching from such larger eggs are both larger and have larger energy stores leading to higher survival probability (Einum & Fleming, 1999; Vøllestad & Lillehammer, 2000; Olsen & Vøllestad, 2001c, 2003; Einum *et al.*, 2004). Further, larger and older females may also be able to select better spawning and rearing habitat for the progeny (Crisp & Carling, 1989), potentially giving rise to genotype-environment correlations. Thus, a maternal effect on length-at-age is to be expected. If, and to what degree, paternal phenotypes have any effect on progeny fitness is unknown, and a strong effect is not to be expected. A paternal effect may be mediated through choice of spawning habitat as males compete for access to females (Elliott, 1994; Klemetsen *et al.*, 2003) or through mate choice directly, but not through any direct effect to the progeny (males only transfer DNA to their progeny, no resources otherwise). These effects have not previously been investigated in a wild setting like the one we study here.

Here, we ask the question whether or not there is significant heritability for length-at-age in a wild population of brown trout studied in its natural environment. Given that heritability is the ratio between the additive genetic variance for a trait and its total phenotypic variance, we expect that as environmental variability increases heritability decreases. Thus, given that the effect of environmental noise increases with age (adds up over the life span), we expect that the heritability for traits such as length-at-age decreases with time. In addition, we ask if parental phenotype has any effect on traits such as length-at-age – in particular we predict that maternal size is positively correlated with progeny size, but only early in life given previous studies that showed that maternal effects are most pronounced at early life-history stages and tend to vanish later (Perry *et al.*, 2004, 2005). To answer these questions, we use data from a long-term mark-recapture program where potential parental fish were sampled over three spawning seasons. Using microsatellite genotyping, we built pedigrees and use this information to estimate the additive genetic variance using the animal model (Kruuk, 2004; Wilson *et al.*, 2009) in a Bayesian framework, using the MCMCglmm package implemented in R (Hadfield, 2010). We did this for a population where we recently have studied in detail both the breeding system (Serbezov *et al.*, 2010) and the strength of viability selection on length-at-age (Carlson *et al.*, 2008). The

general approach used here should have applicability to a large number of study systems and organisms.

Material and methods

The brown trout

Brown trout breed in fresh water during autumn and winter, usually depositing their eggs in clean gravel in running water. The female digs a depression in the gravel where the eggs are deposited, fertilized and then covered with gravel. After hatching in spring, the alevins remain in the nest for several weeks until the yolk-sac reserves are depleted, and then emerge from the gravel as fry to start feeding exogenously. Mortality is high during the first few weeks after emergence, as the trout now establish feeding territories and the possession of a territory can be crucial for surviving this critical period (Elliott, 1994). Trout may remain in their natal stream throughout their lives, whereas in some populations the fish will migrate to larger rivers, lakes or the ocean to feed during the growth season (summer). Generally, high intra- and interpopulation variability exists for many life-history traits (L'Abée-Lund *et al.*, 1989).

Study site and study population

Brown trout were sampled from a small forest stream in southeast Norway (N: 61°15′, E: 11°51′) (see figure in Olsen & Vøllestad, 2001b) during the period 2002–2007. Twenty-five contiguous stream sections were used as permanent study sites. Site length varied from 32 to 96 m (mean = 60.2 m), spanning a total of 1504 m. There is a small waterfall between station 1 and station 2 preventing upstream migration under most conditions and leading to weak but significant genetic differentiation between trout upstream and downstream the waterfall (Taugbøl, 2008). Below the waterfall, the stream enters the larger river Julussa. Long-term tagging studies indicate that some individuals from Bellbekken migrate downstream, but only one tagged individual has ever been registered to move upstream past the waterfall (own unpublished results).

The brown trout in this population is small-sized, rarely reaching ages older than 6 years and lengths above 20 cm (Vøllestad *et al.*, 2002; Olsen & Vøllestad, 2003, 2005). Fish density is relatively low, but so are growth rates. Long-term mark-recapture studies have shown that survival rate is density dependent, but also that it is strongly influenced by stochastic factors (Olsen & Vøllestad, 2001b; Carlson *et al.*, 2008). Also, growth rate seems to be density dependent, even if the evidence for this is less strong than for survival (Vøllestad *et al.*, 2002). Overall, the fish seem to be stationary showing very little dispersal over scales larger than 100 m (Næsje, 2008).

Fish sampling

The trout population was sampled with a backpack electrofishing apparatus during early summer (June) and just prior to the spawning season in autumn (late September to early October) starting in autumn 2002 and ending autumn 2007. All sites within the stream were usually sampled within a 4–5 day period. Sampling was always performed when conditions for sampling were good (i.e. low water flow, stable weather conditions). At a given sampling occasion, each site was electrofished systematically and thoroughly from the downstream to the upstream limit at least three times (White *et al.*, 1982; Bohlin *et al.*, 1989). Brown trout abundance (excluding age-0 fish) at each site and sampling occasion was estimated using the Zippin multiple-pass removal method (Zippin, 1958; Bohlin *et al.*, 1989). Estimated total abundance for all sites pooled for the different sampling periods varied between 895 and 1413 individuals (age-0 fish excluded) (see Carlson *et al.* (2008) for details). Based on long-term mark-recapture data, it has been estimated that the probability of capturing a given individual during a given sampling session is around 0.6 (Carlson *et al.*, 2008).

Passive integrated transponder tags (Prentice *et al.*, 1990) were used to individually mark all brown trout that were larger than *c.* 50 mm. Some of the smaller fish were individually tagged by injection of a coloured elastomer material just under the skin (Olsen & Vøllestad, 2001a). Irrespective of tagging method, trout were anaesthetized with benzocaine prior to tagging. The adipose fin (or a small clip from the caudal fin) was removed and stored in 1.5-mL tubes with 96% ethanol for later genotyping. The fin clip was also used as an external marker indicating that the fish had been captured and tagged previously. At first capture, a few scales were removed for age determination. The fork length of all fish was measured (to the nearest mm), sex was noted for mature fish during the autumn sessions, and tag number was read for all previously tagged fish. Fish belonging to the 0+ age class and the 1+ age class during the spring sampling could be classified into its age class based on length alone. After handling, the fish were allowed to recover and were then released at the site of capture.

Genotyping

DNA was extracted from the collected tissue samples using a salt-based method similar to that outlined in Aljanabi & Martinez (1997). Tissue samples from a total of 4440 individuals were genotyped for 15 loci that amplified well and were polymorphic (Table 1, and see Results). In brief, PCR amplification was performed in one triplex (SSaD71, SSaD85 and SSaD170), one duplex (CA060177 and TAP2B), and the rest of the loci in simplex as these loci amplified best at slightly different

Table 1 Number of alleles, the expected (H_e) and observed heterozygosity (H_o) and exclusion probability of the first and second parent (P_{first}, and P_{second}; the probability of excluding a randomly chosen non-parent and the probability of excluding a randomly chosen non-parent after the first parent has been assigned, respectively) for 15 microsatellite loci among sampled brown trout in the stream Bellbekken.

Locus	No. alleles	H_e	H_o	P_{first}	P_{second}	References
SSa85	5	0.734	0.755	0.312	0.661	O'Reilly *et al.* (1996)
SSsp2213	8	0.775	0.774	0.387	0.753	Paterson *et al.* (2004)
SSaD71	7	0.757	0.750	0.349	0.707	King *et al.* (2005)
SSaD85	11	0.872	0.876	0.589	0.900	Eackles & King (unpublished)*
SSaD170	14	0.858	0.861	0.557	0.883	King *et al.* (2005)
CA060177	7	0.702	0.685	0.278	0.618	Vasemägi *et al.* (2005)
TAP2B	3	0.498	0.490	0.124	0.329	Grimholt *et al.* (2002)
CA040261	11	0.837	0.837	0.510	0.854	Vasemägi *et al.* (2005)
SSaD58	9	0.620	0.619	0.208	0.513	King *et al.* (2005)
SSaD109	6	0.683	0.678	0.263	0.613	King *et al.* (2005)
SSaD157	20	0.837	0.837	0.530	0.877	King *et al.* (2005)
SSaD237	20	0.896	0.914	0.658	0.935	King *et al.* (2005)
MST-85	7	0.733	0.731	0.323	0.682	Presa & Guyomard (1996)
STR-2	12	0.797	0.783	0.442	0.809	Estoup *et al.* (1998)
Strutta-12	15	0.756	0.766	0.377	0.750	Poteaux *et al.* (1999)
Average	10.33	0.757	0.757			
Cumulative exclusion probability		0.999	0.999			

*Available at http://www.ncbi.nlm.nih.gov/nuccore/22094638.

conditions. The protocols used are thoroughly described in Serbezov *et al.* (2010). Samples were subsequently electrophoresed on an ABI Prism® 3100 Genetic Analyzer and analysed with GeneScan® Analysis and Genotyper® software (Applied Biosystems, Foster City, CA, USA) and on an ABI 3730 DNA Analyzer and then analysed with GeneMapper® 3.7 software (Applied Biosystems). As the length of the alleles slightly differed between the two genetic analyzer machines, a plate of 96 individuals was genotyped on both to calibrate the results. Between 12% and 24% of the individuals were genotyped more than once at a particular locus, allowing us to estimate and systemize genotypic error (Hoffman & Amos, 2005). The systematic error rates (Sheehan, 2000) and the stochastic error rates (Wang, 2001) were ~0.5–1% for all loci.

Assignment procedure and pedigree reconstruction

In this pedigree analysis, we combine parentage assignment with sibship reconstruction. We used a full probability Bayesian model for the parentage assignment (Hadfield *et al.*, 2006) that also incorporates phenotypic data (the size of potential spawners and within-stream position of spawners and offspring into the model). Large body size and spatial proximity may increase the parentage probabilities for both males and females. The method is described in detail elsewhere (Serbezov *et al.*, 2010) and only the most important details are reported here.

It was not possible to capture and genotype all potential parents in the stream, and the number is not known with a high degree of certainty. During a particular spawning season, we captured a proportion of the potential parents but some of the actual parents were potentially sampled and genotyped the year before or the year after. We therefore included all fish having a size and age making them candidate parents in the parent input file. These fish do not have a measured size for the actual spawning event, so information on them cannot be used for analyses of parental effects but it can be used for inferring sire and dam effects. Most importantly, the inclusion of these potential parental fish in the parentage input files increased the number of progeny that was assigned a sire by 10% or a dam by 26% in the three seasons, without significantly changing the parentage of offspring that had already been assigned parents observed in the actual spawning season.

Still, some potential parents were never sampled. Genetic information about unsampled successful spawners might however be found in the genotypes of the offspring generation. We used COLONY v2.0 (Wang, 2004; Jones & Wang, 2009) to partition the offspring cohort into full and half-sib families and to infer their parental genotypes. For the 2002 spawning season, the 834 progeny in the offspring cohort could be partitioned into 91 paternal families. Thirteen of these were large half-sib families (> 15 individuals), and the corresponding generated sire genotypes had very high posterior probabilities (mean over loci > 0.999). Two of these genotypes matched sampled parents, but the rest were genotypes that did not match any of the sampled parents, even when allowing for two allelic mismatches. These 11 generated genotypes were included in the input file for parentage analysis as potential fathers, as the offspring in these half-sib families altogether constitute more than

half of the analysed individuals of this particular offspring cohort. The same kind of analysis was performed for the 2003 and 2004 spawning seasons, but it did not reveal any missing highly successful genotypes. Thus, generated parent genotypes were only used for the 2002 spawning season.

Parental effects and heritability

Pedigrees are available for progeny produced during three spawning seasons (2002, 2003, 2004), based on sampling of progeny of various ages from the 2003, 2004 and 2005 cohorts. The pedigrees are complex, with some parents having progeny being sampled during several seasons, some having large numbers of progeny, whereas most parents have only been assigned one or a few progeny. Thus, the data set is unbalanced – a situation that is very common for this kind of data, sampled from natural populations. Here, we estimate variance components from the pedigree data fitting generalized linear mixed models GLMM using MCMC techniques with the R package MCMCglmm (Hadfield, 2010). This package allows the incorporation of special types of variance structures such as those associated with pedigrees (animal model). The animal model (see Kruuk, 2004) is a form of mixed model, where the explanatory terms are a mixture of both fixed and random effects. These analyses readily incorporate unbalanced design datasets containing missing phenotypic measurements; there is also no requirement for balanced design in the pedigree structure (see O'Hara *et al.*, 2008). A range of fixed effects can easily be incorporated in the model and other components of phenotypic variance such as maternal effects or common environmental effects. Also, estimates of variance in the base population are unbiased by any effects of nonrandom mating, inbreeding, selection or evolution during the study period.

The random effects of interest are the additive genetic value of individual animals. Variance components for length-at-age for different age classes were estimated according to the following model:

$$y = \eta + z_1 Year + z_2 Animal + \varepsilon \qquad (1)$$

where η is the overall mean, *Year* is a random effect to describe the variance associated with the sampling years, *Animal* is the additive genetic merit of an individual, and ϵ is a random residual error. The presence of maternal effects implies that residual errors from a simple animal model are no longer uncorrelated, and omitting maternal effects from a model can inflate estimates of heritability. To account for these, we also ran these same models with an added dam random effect:

$$y = \eta + z_1 Year + z_2 Animal + z_3 Dam + \varepsilon \qquad (2)$$

The choice of priors in a MCMC analysis is rarely justified by an objective quantification of the state of knowledge, and the choice of priors can result in different posterior outcomes. Different partitioning of the variance in the priors in our case, however, led to similar results and we ran our animal model with priors that uniformly partition the observed phenotypic variation in body length equally between all random effects. For example, for the model in eqn (1), the specified prior variance for the two random effect parameters, *Year* and *Animal,* and the residuals, was the total phenotypic variance (σ^2_{TOT}) divided by three. We also specify the priors with a low degree of freedom parameter so that little weight is put on the specific variance values. The heritability (h^2) can then be estimated as the ratio between the additive genetic variance (σ^2_A) and the total phenotypic variance (σ^2_{TOT}). The MCMC chains were run for 1 300 000 iterations with a burn-in interval of 300 000 to ensure satisfactory convergence. Parameters and estimated standard errors and confidence intervals were based on sampling 1000 times the posterior parameter distribution.

The variance components were estimated based on all observations of offspring of a certain age. A subsample of the data set consisting of all individuals that were assigned a mother was used for estimating the maternal length effects, and all the individuals that were assigned a father were used for testing for paternal length effects. The following model structure was used for offspring length-at-season:

$$y = \beta_1 L_{sire} + z_1 Year + z_2 Animal_i + \varepsilon \qquad (3)$$

for the paternal size effects, and

$$y = \beta_1 L_{dam} + z_1 Year + z_2 Animal_i + \varepsilon \qquad (4)$$

for the maternal size effects, where L_{sire} and L_{dam} are the fork length of the assigned sires and dams, respectively, and *Year* and *Animal* are specified as random effects.

Finally, we run three global models for all age classes and seasons pooled, with season and age as fixed effects; one model testing for sire effects including all juveniles with a measured sire length, a corresponding dam model, and a model testing both sire and dam effects simultaneously, based on offspring data with both parents assigned.

Results

The microsatellites used for parentage assignment provided high estimated exclusion probabilities (Table 1). Of the 4440 individuals genotyped, many were either not sexually mature during the three spawning seasons or, if immature, did not belong to the three cohorts studied here. A total of 451 (195 males and 256 females) individual mature fish were observed, measured and genotyped during the 2002–2004 spawning seasons. The mature males tended to be larger than the mature females (mean length (mm) ± SD (range): males, 163.8 ± 23.5 (113–306); females, 150.8 ± 16.3 (112–220)).

In total, 1856 individual progeny belonging to the three cohorts (2003–2005) were genotyped. A proportion of offspring (40.2%) were not assigned any parent, but here, we include those progeny with at least one identified parent. We were able to sample the age classes with reasonable sample sizes up to age 2. We therefore only use fish sampled from the first autumn through the third autumn of life. Some of these juveniles were tagged and subsequently recaptured at later dates, giving a total data set for analysis consisting of 1423 observations. These observations include information about site and time of sampling in addition to age and fork length (Table 2). It was thus possible to infer the family size and breeding structure of this particular set of Bellbekken brown trout (for a more detailed treatment using a larger data set see Serbezov *et al.*, 2010). These analyses revealed that each successful male mated with estimated 1–13 females, whereas each successful female mated with estimated 1–4 males.

When estimating the additive genetic variance using MCMCglmm, we first investigated the animal effect with year as an additional random effect. Overall, total phenotypic variability increased with increasing age (Table 3). The heritability varied between 0.16 and 0.31 and decreased somewhat with age (Fig. 1). When introducing dam as an additional random effect in the model, the estimated heritability somewhat decreased and varied between 0.12 and 0.21. The dam variance components in general were low, constituting overall 7.2 ± 1.8% of the total phenotypic variance. The random year effects were in general rather large, often larger than the residual variance component.

To further investigate the presence of potential parental effects on progeny length-at-age, we introduced dam length or sire length as fixed effects in the animal models with year as random effect (excluding dam as random effect). Overall, both sire and dam lengths were positively correlated with progeny length-at-age (Table 4). All the dam length effects were significantly larger than zero, whereas the sire length effects in general were lower, and some were also negative. For the oldest age classes, total phenotypic variance was very high in these models, driven by very large year variance components. In general, the variance component estimation converged nicely, but some unreasonably large values for the random year component were estimated in these runs.

As a final analysis, and to increase statistical power, we included all age classes in two global models, with age class and season as fixed effects together with either sire length or dam lengths as additional fixed effects. Animal

Table 2 Summary table for mean ± SD (*N*) length-at-age (mm) for three cohorts of brown trout progeny in Bellbekken during 2003–2006. The ANOVA tests for among-year variation in length-at-age; within age estimates sharing the same letter are not significantly different.

	2003	2004	2005	2006	ANOVA
Age – season					
0 – autumn	44.9 ± 4.6[a] (35)	46.0 ± 4.7[a] (101)	40.5 ± 4.9[b] (89)		$F_{2,\ 222} = 32.17$, $P < 0.001$
1 – spring		56.9 ± 4.9[a] (131)	57.9 ± 5.6[a] (219)	54.9 ± 6.7[b] (85)	$F_{2,\ 432} = 8.93$, $P < 0.001$
1 – autumn		73.9 ± 7.7 (115	69.8 ± 6.7 (202)		$F_{1,\ 315} = 24.32$, $P < 0.001$
2 – spring			83.7 ± 8.0 (142)	81.9 ± 8.1 (90)	$F_{1,\ 230} = 2.91$, $P = 0.090$
2 – autumn			93.5 ± 8.9 (128)	97.0 ± 7.0 (85)	$F_{1,\ 211} = 9.05$, $P = 0.003$

Table 3 Results from the animal model for brown trout in Bellbekken. Shown are the estimated variance components (±SE) for length-at-age for various age classes.

Age – season	*N*	V_a	V_{year}	V_{dam}	V_p
Animal model with year as random effect					
0 – autumn	225	8.0 ± 0.1	20.2 ± 1.4		43.9 ± 1.4
1 – spring	435	12.7 ± 0.2	13.2 ± 1.0		45.9 ± 1.0
1 – autumn	318	22.8 ± 0.4	41.7 ± 5.6		94.4 ± 5.6
2 – spring	232	16.2 ± 0.3	36.2 ± 4.0		102.6 ± 4.0
2 – autumn	213	16.2 ± 0.4	90.1 ± 18.8		159.3 ± 18.8
Animal model with year and dam as random effect					
0 – autumn	225	6.1 ± 0.1	16.9 ± 1.1	2.5 ± 0.1	41.8 ± 1.1
1 – spring	435	8.8 ± 0.2	10.6 ± 0.8	3.4 ± 0.1	44.5 ± 0.8
1 – autumn	318	11.1 ± 0.3	35.5 ± 6.2	8.7 ± 0.1	90.8 ± 6.2
2 – spring	232	12.2 ± 0.3	27.5 ± 2.0	7.7 ± 0.2	98.2 ± 2.1
2 – autumn	213	11.8 ± 0.3	102.5 ± 38.5	8.6 ± 0.2	176.0 ± 38.5

Variance components are given for two models – with or without a fixed year effect. Variance components: V_a, additive genetic variance; V_{year}, year variance; V_{dam}, dam variance; V_p, total phenotypic variance.

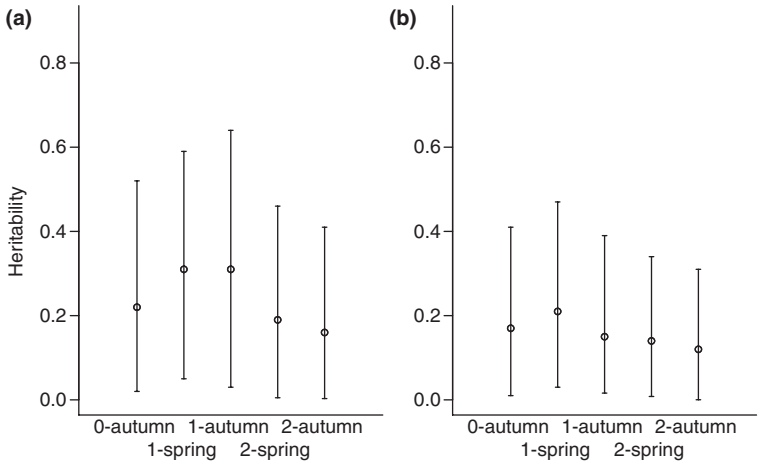

Fig. 1 Heritability estimates (means ± confidence intervals) for brown trout length-at-age of various age classes, extracted from for Animal models with (a) no dam effects and (b) with dam effects. Age class (sampling) is given as age in years and sampling season.

Table 4 Animal model results with sire length or dam length as fixed effects, and animal and year as random effects.

Age – season	*N*	Sire/dam effect	β
0 – autumn	172	Sire	−0.002 ± 0.001
	169	Dam	0.031 ± 0.001
1 – spring	297	Sire	0.009 ± 0.001
	326	Dam	0.042 ± 0.001
1 – autumn	212	Sire	0.022 ± 0.001
	235	Dam	0.084 ± 0.001
2 – spring	126	Sire	0.040 ± 0.001
	184	Dam	0.049 ± 0.002
2 – autumn	117	Sire	−0.001 ± 0.001
	165	Dam	0.105 ± 0.002

The table shows parameter estimates (β) ±SE for the dam and sire length effects.

Table 5 Summary results from animal models testing for either sire or dam length effects on total length-at-age for brown trout progeny in Bellbekken.

	Sire model	Dam model	Sire + dam model
N	1060	1275	657
V_a	33.8 ± 0.1	42.3 ± 0.1	37.3 ± 0.2
V_{year}	226.1 ± 19.7	199.3 ± 10.9	21.6 ± 1.4
V_r	30.7 ± 0.1	23.1 ± 0.1	25.2 ± 0.1
h^2	0.20 (0.03–0.37)	0.25 (0.04–0.47)	0.48 (0.27–0.63)
Sire length	0.022 ± 0.001		0.008 ± 0.0001
Dam length		0.078 ± 0.001	00.071 ± 0.001

V_a, additive genetic variance; V_{year}, year variance component; V_r, residual variance; h^2, heritability (with confidence limits). Animal and year are random effects, with age class and season as fixed effects. Parameter estimates are given for the dam and sire length effects (β; ±SE).

and year were random effects. The heritability was lower than for the single-age models but was estimated with higher precision (Table 5). The sire and dam length effects were both significantly larger than zero, with the dam length effect being larger than the sire length effect. As a final analysis, we tested for sire and dam length effects at the same time. This leads to reduced sample size, because MCMCglmm does not allow missing values for the fixed factors. The dam length effect was still large, whereas the sire length effect was much smaller – but still significant (Table 5).

Discussion

The main aim of this study was to investigate how much additive genetic variance is underlying important life-history traits for selection to work on in a wild population of brown trout. We also ask how strong maternal and paternal effects might be on these traits. We have thus estimated, for the first time, the additive genetic variance for length-at-age for wild-living brown trout. Overall, we document high levels of additive genetic variance translating into heritability varying from 0.16 to 0.31. Further, we demonstrate significant maternal and paternal effects with larger parents in general producing larger progeny – this parental effect lasted for more than 2 years.

Our results show that the level of additive genetic variance and the associated heritability for a complex life-history trait such as length-at-age are relatively large. Estimates of quantitative genetic parameters often have high error rates, and thus low power (Roff, 1997, 2002). In the present study, using the newly developed Bayesian version of the Animal model, we have been able to assess h^2 with rather high precision. Overall, the h^2 estimates for length-at-age for various age classes of trout, and thus based on partly independent data sets, were in the same range. These h^2 estimates are also in the range of values commonly reported for traits such as length-at-age and growth rate for salmonid fishes (see review in Carlson & Seamons, 2008). Total phenotypic

variance increased with increasing age, overall leading to a reduced importance of the additive genetic variance with increasing age.

There are a few studies available reporting estimates of heritability for life-history traits from wild salmonid fishes (e.g. Garant *et al.*, 2003; Thériault *et al.*, 2007). These studies show that various life-history traits have, indeed, significant levels of additive genetic variance. However, most studies estimating quantitative genetic parameters are from the laboratory. A recent review clearly showed that estimates of heritability from natural systems in general were lacking (Carlson & Seamons, 2008) – the main reason for this is probably that obtaining this type of data is labour intensive and complicated. The paucity of estimates from the wild is unfortunate since quantitative genetic parameters are context specific (Roff, 1992, 1997; Carlson & Seamons, 2008), and they are also strongly dependent upon the statistical model used for estimating these parameters (Wilson, 2008). Thus, it is problematic to make inferences with parameters estimated in the laboratory to natural situations, as it is problematic to use hatchery-reared and domesticated animals in the experiments. To predict the effect of changes in selection strength and direction, it will be necessary to estimate these parameters in the wild under natural conditions (Crozier *et al.*, 2008; Kuparinen *et al.*, 2009).

The heritability of length-at-age for this natural population of brown trout was in the range 0.16–0.31 and decreased with age. Overall, environmental variance was large and increased with progeny age. Length-at-age or associated traits, such as growth rate and age at maturity, are important fitness-related traits (Roff, 1992; Stearns, 1992) because of their influence on survival and reproduction. Given that they are closely associated with fitness, it is often assumed that they are influenced by strong directional or stabilizing selection, leading to erosion of additive genetic variance (Mousseau & Roff, 1987; Roff, 1997; Carlson & Seamons, 2008). However, it may also be that heritability is low because the environmental component of the phenotypic variance is very high leading to low heritability even when additive genetic variance is relatively high (Price & Schluter, 1991; Merilä & Sheldon, 2000). This may be so especially under natural conditions where environmental conditions vary on short temporal and spatial scales. This environmental and developmental noise may add up over time, at least for traits expressed during the same life stage such as for the brown trout here.

Given this large environmental effect, and the assumed strong relationship between size and fitness, we should expect low heritability decreasing with age. A weak tendency for this was observed here. The brown trout in Bellbekken is small-sized, mature at a young age and small size, and in general have a short reproductive life (Olsen & Vøllestad, 2001b, 2003, 2005; Vøllestad *et al.*, 2002). As evidenced from mark-recapture studies, there are strong density dependent and environmental effects on survival (Carlson *et al.*, 2008). One conclusion to be drawn from this is that under current environmental conditions selection on size at age, and thus on growth rate, is very weak. This should lead to the prediction that the fitness surface for traits such as growth rate and length-at-age is flat. This is indeed what was found in an independent analysis of selection differentials and size-dependent survival probabilities for the younger age classes in this population of trout (Carlson *et al.*, 2008). In conclusion, length-at-age does not appear to be strongly related with survival probability in this population. On the other hand, larger females produce larger and more eggs (Olsen & Vøllestad, 2003) and have, as also do larger males, higher reproductive success (Serbezov *et al.*, 2010). Given that selection is not strong and directional, and that stochastic processes are important, relatively high levels of additive genetic variance can be maintained. This indicates that these traits may indeed respond rapidly to selection, given the right environmental context.

On top of these genetic and environmental effects, there are a number of potential parental influences. The maternal (dam) effect was consistently small (c. 7% of total phenotypic variance) but stable. By including the random dam effect in our models, the level of additive genetic variance decreased, indicating that maternal effects may be important. In our analysis, we tried to evaluate to what extent parental characteristics (i.e. maternal and paternal length) influenced progeny length-at-age. The main result was that both maternal and paternal size had a significant influence on progeny length. Larger progeny tended to have a larger than average parent. The maternal effect is probably either because of differences in egg quality or because of differences in the choice of spawning habitat (a genotype – environment correlation). In general, larger brown trout females produce larger eggs that in turn produce larger juveniles (Einum & Fleming, 1999; Vøllestad & Lillehammer, 2000; Olsen & Vøllestad, 2003). This size advantage can be transferred into a later fitness advantage, especially under arduous conditions (Einum & Fleming, 1999, 2000). One possible way this could work is if the larger juveniles that hatch from larger eggs are able to acquire and defend better feeding territories (Keeley & Grant, 1995). The weaker but still significant paternal effect on juvenile length-at-age cannot be explained in the same way. The paternal effect can either be through transfer of 'good genes', through choice of high quality partners, or through some genotype-environment correlation. The final analysis where we tested for maternal and paternal size effects in a global model indicated that the maternal effect was the stronger. It also did indicate that the paternal size effect probably works through some kind of mate choice. In a detailed study of the breeding system of the Bellbekken trout, we have found that large male trout tend to mate

with the larger female trout (Serbezov *et al.*, 2010), and larger parents tend to have higher reproductive success. This size-assortative mating may indeed explain the relationship between maternal and paternal size and progeny size. The paternal effect is thus indirect and through mate choice, and the maternal effect may be direct through the effect of egg size or indirect through the choice of spawning site (as a form of genotype-environment correlation). The benefits of larger size, which are at best marginal during earlier life stages, might thus become more pronounced later in life because of the specific breeding system of the trout.

To conclude, we have shown for a natural population of brown trout that the level of additive genetic variance, and thus heritability, for length-at-age for various age groups can be relatively high. Further, we found significant maternal and paternal effects showing that larger parents tended to produce progeny with larger length-at-age. In total, these results show that brown trout in this system has the potential to respond to environmental challenges, both by evolutionary and by plastic adjustments.

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References

Aljanabi, S.M. & Martinez, I. 1997. Universal and rapid salt-extraction of high quality genomic DNA for PCR-based techniques. *Nucleic Acids Res.* **25**: 4692–4693.

Blows, M.W. & Hoffmann, A.A. 2005. A reassessment of genetic limits to evolutionary change. *Ecology* **86**: 1371–1384.

Bohlin, T., Hamrin, S., Heggberget, T.G., Rasmussen, G. & Saltveit, S.J. 1989. Electrofishing – theory and practice with special emphasis on salmonids. *Hydrobiologia* **173**: 9–43.

Carlson, S.M. & Seamons, T.R. 2008. A review of quantitative genetic components of fitness in salmonids: implications for adaptation to future change. *Evol. Appl.* **1**: 222–238.

Carlson, S.M., Olsen, E.M. & Vøllestad, L.A. 2008. Seasonal mortality and the effect of body size: a review and an empirical test using individual data on brown trout. *Funct. Ecol.* **22**: 663–673.

Crisp, D.T. & Carling, P.A. 1989. Observations on siting, dimensions and structure of salmonid redds. *J. Fish Biol.* **34**: 119–134.

Crozier, L.G., Hendry, A.P., Lawson, P.W., Manuta, N.J., Battin, J., Shaw, R.G. & Huey, R.B. 2008. Potential responses to climate change in organisms with complex life histories: evolution and plasticity in Pacific salmon. *Evol. Appl.* **1**: 252–270.

Darwish, T.L. & Hutchings, J.A. 2009. Genetic variability in reaction norms between farmed and wild backcrosses of Atlantic salmon (*Salmo salar*). *Can. J. Fish. Aquatic Sci.* **66**: 83–90.

Einum, S. & Fleming, I.A. 1999. Maternal effects of egg size in brown trout (*Salmo trutta*): norms of reaction to environmental quality. *Proc. R. Soc. Lond. B Biol. Sci.* **266**: 2095–2100.

Einum, S. & Fleming, I.A. 2000. Selection against late emergence and small offspring in Atlantic salmon (*Salmo salar*). *Evolution* **54**: 628–639.

Einum, S., Kinnison, M.T. & Hendry, A.P. 2004. Evolution of egg size and number. In: *Evolution Illuminated. Salmon and Their Relatives* (A.P. Hendry & S.C. Stearns, eds), p. 510. Oxford University Press, Oxford.

Elliott, J.M. 1989. Wild brown trout *Salmo trutta*: an important national and international resource. *Freshw. Biol.* **21**: 7–19.

Elliott, J.M. 1994. *Quantitative Ecology and the Brown Trout*. Oxford University Press, Oxford.

Estoup, A., Rousset, F., MIchalakis, Y., Cornuet, J.-M., Adriamanga, M. & Guyomard, R. 1998. Comparative analysis of microsatellite and allozyme markers: a case study investigating microgeographic differentiation in brown trout (*Salmo trutta*). *Mol. Ecol.* **7**: 339–353.

Garant, D., Dodson, J.J. & Bernatchez, L. 2003. Differential reproductive success and heritability of alternative reproductive tactics in wild Atlantic salmon (*Salmo salar* L.). *Evolution* **57**: 1133–1141.

Gienapp, P., Teplitsky, C., Alho, J.S. & Merilä, J. 2008. Climate change and evolution: disentangling environmental and genetic responses. *Mol. Ecol.* **17**: 167–178.

Green, B.S. 2008. Maternal effects in fish populations. *Adv. Mar. Biol.* **54**: 1–105.

Grimholt, U., Drabløs, F., Jørgensen, S.M., Høyheim, B. & Stet, R.J.M. 2002. The major histocompatibility class I locus in Atlantic salmon (*Salmo salar* L.): polymorphism, linkage analysis and protein modelling. *Immunogenetics* **54**: 570–581.

Hadfield, J.D. 2010. MCMC methods for multi-response generalized linear mixed models: the MCMCglmm R package. *J. Stat. Softw.* **33**: 2.

Hadfield, J.D., Richardson, D.S. & Burke, T. 2006. Towards unbiased parentage assignment: combining genetic, behavioural and spatial data in a Bayesian framework. *Mol. Ecol.* **15**: 3715–3730.

Haugen, T.O. & Vøllestad, L.A. 2000. Population differences in early life-history traits in grayling. *J. Evol. Biol.* **13**: 897–905.

Hendry, A.P. & Kinnison, M.T. 1999. The pace of modern life: measuring rates of contemporary microevolution. *Evolution* **53**: 1637–1653.

Hendry, A.P., Nosil, P. & Reisenberg, L.H. 2007. The speed of ecological speciation. *Funct. Ecol.* **21**: 455–464.

Hendry, A.P., Farrugia, T.J. & Kinnison, M.T. 2008. Human influences on rates of phenotypic change in wild animal populations. *Mol. Ecol.* **17**: 20–29.

Hoffman, J.I. & Amos, W. 2005. Microsatellite genotyping errors: detection approaches, common sources and consequences for paternal exclusion. *Mol. Ecol.* **14**: 599–612.

Jensen, L.F., Hansen, M.M., Pertoldi, C., Holdensgaard, G., Mensberg, K.-L.D. & Loeschcke, V. 2008. Local adaptation in brown trout early life-history traits: implications for climate change adaptability. *Proc. R. Soc. Lond. B Biol. Sci.* **275**: 2859–2868.

Jones, O.R. & Wang, J. 2009. COLONY: a program for parentage and sibship inference from multilocus genotype data. *Mol. Ecol. Res.* **10**: 551–555.

Keeley, E.R. & Grant, J.W.A. 1995. Allometric and environmental correlates of territory size in juvenile Atlantic salmon (*Salmo salar*). *Can. J. Fish. Aquatic Sci.* **52**: 186–196.

King, T.L., Eackles, M.S. & Letcher, B.H. 2005. Microsatellite DNA markers for the study of Atlantic salmon (*Salmo salar*) kinship, population structure, and mixed-fishery analyses. *Mol. Ecol. Notes* **5**: 130–132.

Kinnison, M.T. & Hendry, A.P. 2001. The pace of modern life II: from rates of contemporary microevolution to pattern and process. *Genetica* **112–113**: 145–164.

Klemetsen, A., Amundsen, P.-A., Dempson, J.B., Jonsson, B., Jonsson, N., O'Connell, M.F. & Mortensen, E. 2003. Atlantic salmon *Salmo salar* L., brown trout *Salmo trutta* L. and Arctic charr *Salvelinus alpinus* (L.): a review of aspects of their life histories. *Ecol. Freshw. Fish.* **12**: 1–59.

Kruuk, L.E.B. 2004. Estimating genetic parameters in natural populations using the ''animal model''. *Philos. Trans. R. Soc. Lond. B Biol. Sci.* **359**: 873–890.

Kruuk, L.E.B., Slate, J. & Wilson, A.J. 2008. New answers for old questions: the evolutionary quantitative genetics of wild animal populations. *Annu. Rev. Ecol. Syst.* **39**: 525–548.

Kuparinen, A., Garcia de Leaniz, C., Consuegra, S. & Merilä, J. 2009. Growth-history perspective on the decreasing age and size at maturation of exploited Atlantic salmon. *Mar. Ecol. Prog. Ser.* **376**: 245–252.

L'Abée-Lund, J.H., Jonsson, B., Jensen, A.J., Sættem, L.M., Heggberget, T.G., Johnsen, B.O. & Næsje, T.F. 1989. Latitudinal variation in life-history characteristics of sea-run migrant brown trout *Salmo trutta*. *J. Anim. Ecol.* **58**: 525–542.

Merilä, J. & Sheldon, B.C. 2000. Lifetime reproductive success and heritability in nature. *Am. Nat.* **155**: 301–310.

Mousseau, T.A. & Fox, C.W. 1998. *Maternal Effects as Adaptations*. Oxford University Press, Oxford.

Mousseau, T.A. & Roff, D.A. 1987. Natural selection and the heritability of fitness components. *Heredity* **59**: 181–197.

Næsje, T. 2008. Dispersal in stream-living brown trout (*Salmo trutta*). MSc Thesis, *Department of Biology,* University of Oslo, Oslo, Norway.

O'Hara, R.B., Cano, J.M., Ovaskainen, O., Teplitsky, C. & Alho, J.S. 2008. Bayesian approaches in evolutionary quantitative genetics, *J. Evol. Biol.* **21**: 949–957.

Olsen, E.M. & Vøllestad, L.A. 2001a. An evaluation of visible implant elastomer for marking age-0 brown trout. *North Am. J. Fish. Manage.* **21**: 967–970.

Olsen, E.M. & Vøllestad, L.A. 2001b. Estimates of survival of stream-dwelling brown trout using mark-recaptures. *J. Fish Biol.* **59**: 1622–1637.

Olsen, E.M. & Vøllestad, L.A. 2001c. Within-stream variation in early life-history traits in brown trout. *J. Fish Biol.* **59**: 1579–1588.

Olsen, E.M. & Vøllestad, L.A. 2003. Microgeographical variation in brown trout reproductive traits: possible effects of biotic interactions. *Oikos* **100**: 483–492.

Olsen, E.M. & Vøllestad, L.A. 2005. Small-scale spatial variation in age and size at maturity of stream-dwelling brown trout, *Salmo trutta*. *Ecol. Freshw. Fish.* **14**: 202–208.

O'Reilly, P.T., Hamilton, L.C., McConnell, S.L. & Wright, J.M. 1996. Rapid analysis of genetic variation in Atlantic salmon (*Salmo salar*) by PCR multiplexing of dinucleotide and tetranucleotide microsatellites. *Can. J. Fish. Aquatic Sci.* **53**: 2292–2298.

Paterson, S., Piertney, S.B., Knox, D., Gilbey, J. & Verspoor, E. 2004. Characterization and PCR multiplexing of novel highly variable tetranucleotide Atlantic salmon (*Salmo salar* L.) microsatellites. *Mol. Ecol. Notes* **4**: 160–162.

Perry, G.M.L., Laplante, B., Audet, C. & Bernatchez, L. 2004. Shifting patterns in genetic control at the embryo-alevin boundary in brook charr. *Evolution* **58**: 2002–2012.

Perry, G.M.L., Audet, C. & Bernatchez, L. 2005. Maternal genetic effects on adaptive divergence between anadromous and resident brook charr during early life history. *J. Evol. Biol.* **18**: 1348–1361.

Pfennig, D.W. & Martin, R.A. 2008. A maternal effect mediates rapid population divergence and character displacement in spadefoot toads. *Evolution* **63**: 898–909.

Poteaux, C., Bonhomme, F. & Berrebi, P. 1999. Microsatellite polymorphism and genetic impact of restocking in Mediterranean brown trout (*Salmo trutta* L.). *Heredity* **82**: 645–653.

Prentice, E.F., Flagg, T.A. & McCutcheon, C.S. 1990. Feasibility of using implantable passive integrated transponder (PIT) tags in salmonids. *Am. Fish. Soc. Symp.* **7**: 317–322.

Presa, P. & Guyomard, R. 1996. Conservation of microsatellites in three species of salmonids. *J. Fish Biol.* **49**: 1326–1329.

Price, T. & Schluter, D. 1991. On the low heritability of life-history traits. *Evolution* **45**: 853–861.

Roff, D. A. 1992. *The Evolution of Life Histories. Theory and Analysis*. Chapman & Hall, New York.

Roff, D.A. 1997. *Evolutionary Quantitative Genetics*. Chapman & Hall, New York.

Roff, D.A. 2002. *Life History Evolution*. Sinauer Associates, Inc., Sunderland, Massachusetts, USA.

Schlichting, C.D. & Pigliucci, M. 1998. *Phenotypic Evolution: A Reaction Norm Perspective*. Sinauer Associates, Inc., Sunderland, Massachusetts, USA.

Schluter, D. 2000. *The Ecology of Adaptive Speciation*. Oxford University Press, Oxford.

Serbezov, D., Bernatchez, L., Olsen, E.M. & Vøllestad, L.A. 2010. Mating patterns and determinants of individual reproductive success in brown trout (*Salmo trutta*) revealed by parentage analysis of an entire stream living population. *Mol. Ecol.* in press.

Sheehan, N.A. 2000. On the application of Markov chain Monte Carlo methods to genetic analyses on complex pedigrees. *Int. Stat. Rev.* **68**: 83–110.

Stearns, S.C. 1992. *The Evolution of Life Histories*. Oxford University Press, Oxford.

Taugbøl, A. 2008. Fine-scale genetic structure of brown trout (*Salmo trutta*). MSc thesis, Department of Biology, University of Oslo, Oslo, Norway.

Thériault, V., Garant, D., Bernatchez, L. & Dodson, J.J. 2007. Heritability of life-history tactics and genetic correlations with body size in a natural population of brook charr (*Salvelinus fontinalis*). *J. Evol. Biol.* **20**: 2266–2277.

Vandeputte, M., Quillet, E. & Chevassus, B. 2002. Early development and survival in brown trout (*Salmo trutta fario* L.): indirect effects of selection for growth rate and estimation of genetic parameters. *Aquaculture* **204**: 435–445.

Vasemägi, A., Nilsson, J. & Primmer, C.R. 2005. Seventy-five EST-linked Atlantic salmon (*Salmo salar* L.) microsatellite markers and their cross-amplification in five salmonid species. *Mol. Ecol. Notes* **5**: 282–288.

Via, S. & Lande, R. 1985. Genotype-environment interaction and the evolution of phenotypic plasticity. *Evolution* **39**: 505–522.

Vøllestad, L.A. & Lillehammer, T. 2000. Individual variation in early life-history traits in brown trout. *Ecol. Freshw. Fish.* **9**: 242–247.

Vøllestad, L.A., Olsen, E.M. & Forseth, T. 2002. Growth-rate variation in brown trout in small neighbouring streams: evidence for density-dependence? *J. Fish Biol.* **61**: 1513–1527.

Wang, J.L. 2001. A pseudo-likelihood method for estimating effective population size from temporally spaced samples. *Gen. Res.* **78**: 243–257.

Wang, J. 2004. Sibship reconstruction from genetic data with typing errors. *Genetics* **166**: 1963–1979.

Weigensberg, I. & Roff, D.A. 1996. Natural heritabilities: can they be reliably estimated in the laboratory? *Evolution* **50**: 2149–2157.

White, G.C., Anderson, D.R., Burnham, K.P. & Otis, D.L. 1982 *Capture–Recapture and Removal Methods for Sampling Closed Populations*. pp. 1–235. Los Alamos National Laboratory, Los Alamos, New Mexico, USA.

Wilson, A.J. 2008. Why h^2 does not always equal V_a/V_p? *J. Evol. Biol.* **21**: 647–650.

Wilson, A.J., Réale, D., Clements, M.N., Morrissey, M.M., Postma, E., Walling, C.A., Kruuk, L.E.B. & Nussey, D.H. 2009. An ecologist's guide to the animal model. *J. Anim. Ecol.* **79**: 13–26.

Zippin, C. 1958. The removal method of population estimation. *J. Wildl. Manage.* **22**: 82–90.

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