ECOLOGY OF
FRESHWATER FISH

# Differences in early developmental rate and yolk conversion efficiency in offspring of trout with alternative life histories

**Travis E. Van Leeuwen[1,2], Shaun S. Killen[2], Neil B. Metcalfe[2], Colin E. Adams[1]**

[1]Scottish Centre for Ecology and the Natural Environment, IBAHCM, Graham Kerr Building, Rowardennan, Loch Lomond, Glasgow G63 0AW, UK
[2]Institute of Biodiversity, Animal Health & Comparative Medicine, College of Medical, Veterinary and Life Sciences, University of Glasgow, Glasgow G12 8QQ, UK

Abstract – Partial migration, in which some individuals of a population migrate while other individuals remain resident, is generally associated with ontogenetic shifts to better feeding areas or as a response to environmental instability, but its underlying mechanisms remain relatively unknown. Brown trout (*Salmo trutta*) exhibit partial migration, with some individuals remaining in freshwater (freshwater resident) while others undertake an anadromous migration, where they spend time at sea before returning to breed in freshwater (migrant). We reared full-sibling groups of offspring from freshwater-resident and anadromous brown trout from the same catchment in the laboratory under common garden conditions to examine potential differences in their early development. Freshwater-resident parents produced eggs that were slower to hatch than those of anadromous parents, but freshwater-resident offspring were quicker to absorb their yolk and reach the stage of exogenous feeding. Their offspring also had a higher conversion efficiency from the egg stage to the start of exogenous feeding (so were larger by the start of the fry stage) than did offspring from anadromous parents despite no difference in standard metabolic rate, maximal metabolic rate or aerobic scope. Given these differences in early development, we discuss how the migration history of the parents might influence the migration probability of the offspring.

Key words: alternative life history; ecotype; resident *vs.* migratory; *Salmo trutta*; early development; salmonid metabolism; anadromous vs. nonanadromous; partial migration

## Introduction

In many taxa, there is variation in the propensity of individuals from a single population to migrate between habitats (the phenomenon of 'partial migration', see reviews in Chapman et al. 2011; Dodson et al. 2013). From an evolutionary perspective, the maintenance of such individual variation in migratory tendency implies that the costs and benefits of migration are dependent upon context, with the net benefits not being equal for all individuals. Migration typically incurs increased energetic costs and mortality risks, but may result in increased foraging or breeding opportunities; in the case of species with indeterminate growth, this generally leads to significant growth and size-at-age benefits to migration (Jonsson & Jonsson 1993). While the effect of migration on the adult form is fairly well established there remains a paucity of studies examining the relationship between the migration history of the parents and the early size and development and potentially the migration probability of the offspring.

The commonest form of this intraspecific variation in movement patterns is nonbreeding partial migration (*sensu* Chapman et al. 2011), where migrants and residents breed in sympatry but segregate during

feeding stages of their life cycle. There have been many hypothesised explanations for this category of migration, including competition for resources or breeding opportunities, predation risk trade-offs and intraspecific niche diversity (see Chapman et al. 2011). In all cases, the migration can be viewed as a response to adversity (Taylor & Taylor 1977) or as an individual response to optimise future fitness by selecting a particular environment. However, the degree of adversity and assessment of future fitness will depend on the particular environmental conditions that are experienced at the time and whether or not partial migration is fixed (i.e. influenced by the migration history of the parents) or flexible (condition-dependent). Thus, individuals that by chance have the advantage of prior residence may have greater access to resources and so be less liable to migrate (Sandell & Smith 1991). Similarly, residency may be more likely in those encountering a low local density of competitors [as has been demonstrated experimentally in red-spotted newts *Notophthalmus viridescens* by Grayson & Wilbur (2009)], or those experiencing a higher food supply (as in salmonid fish: Olsson & Greenberg 2004; Olsson et al. 2006; Wysujack et al. 2009). Body size may also affect the selection pressures for/against migration, as larger individuals may generally have less to gain from migration (Dodson et al. 2013; though see Brodersen et al. 2008 for an exception) as they usually have advantages of a higher competitive ability and/or lower risk of starvation and predation (Chapman et al. 2011).

While it is clear that both abiotic and biotic factors are likely to influence the decision to migrate or not, the relative role of genetic (fixed) versus environmental (flexible) causes remains understudied. Berthold & Pulido (1994) provide support for a genetic predisposition for migratory tendency and migration distance in the Blackcap *Sylvia atricapilla*. However, it has also been suggested that partial migration is driven by a complex interaction between the environment and genetics: in the 'threshold model' the triggering of migration depends on whether or not a continuous character ('liability trait') exceeds a genetically predetermined threshold value (Chapman et al. 2011; Dodson et al. 2013). In this scenario, individuals physiologically self-evaluate their performance against this threshold (e.g. of growth rate, body size or physiological condition), with migration being dependent on whether or not the threshold is exceeded (Fleming 1996; Thorpe & Metcalfe 1998; Pulido 2011; Dodson et al. 2013). This model generates variation in migratory tendency between environments, but also generates partial migratory populations even under uniform conditions (as the threshold can vary between individuals); however,

hard evidence of the factors that predispose particular individuals to one or the other strategy is lacking.

A well-documented example of a species exhibiting partial migration is the Brown trout *Salmo trutta*, a polymorphic species that adopts a continuum of life-history strategies, with the two most common being freshwater-resident and anadromous migrant (which grows relatively quickly at sea before returning to freshwater to spawn). Both ecotypes can occur in sympatry, possibly derived from a single gene pool, with both anadromous and freshwater-resident adults having the ability to interbreed and produce offspring capable of adopting either life history (O'Neal. & Stanford 2011). Freshwater-resident and anadromous trout appear indistinguishable during early life, and it is presumed that they only become separable after one or more years, when the migrants turn silver in colour in preparation for entry to sea water (smolting; Jonsson 1985). The major physiological change that is required for the trout to enter sea water means that the option to migrate is also associated with a developmental switch point some months prior to the actual migration (Paez et al. 2011). Jonsson (1985) proposed that migrant brown trout are made up of the slower growing individuals in a population, which migrate in search of more productive habitats. Given that freshwater fluvial ecosystems are often regarded as being food limited (Imre et al.2005), it has also been suggested that metabolic constraints play an important role in determining physiological state and thus migration probability. In the low food environment of their natal river, individuals with a lower growth efficiency, higher food requirement and/or higher metabolic rate (i.e. energy maximisers) will become energetically constrained earlier in life compared to individuals with higher growth efficiency, lower food requirement and/or lower metabolic rates (efficiency maximisers; Metcalfe et al.1995; Forseth et al. 1999; Morinville & Rasmussen 2003; Rosenfeld et al. 2013). Individuals with low growth efficiency and/or higher metabolic rates may therefore migrate in search of more productive habitats (lakes, oceans) to meet their outstanding metabolic needs.

While it is likely that growth history and current body size and physiological condition influence the decision to migrate, this may in part be driven by genetic or parental effects. Given that trout produce large yolky eggs which represent a significant nutritional source for the developing embryo, typically providing the only energy source for the first months of development, there is potential for strong parental effects on offspring development (Mousseau & Fox 1998; Monaghan 2008; Burton et al. 2013), with migration history of the parents potentially influencing the migration probability of the offspring.

To address these issues, we reared brown trout off-spring from eggs of known parentage (i.e. freshwater resident or anadromous) under common garden conditions. Because we knew the migration history of the parents, it provided an opportunity to understand (i) the effects of migration history of the parents on egg size, egg number and egg energy density and (ii) the effects of migration history of the parents on offspring development. Based on the differences in adult size between the two ecotypes (with anadromous fish being considerably larger) and the metabolic constraints hypothesis for migration, we expected that offspring from anadromous parents would be larger from the egg stage through to the start of exogenous feeding, exhibit a higher metabolic rate and/or lower food conversion efficiency and consume their yolk reserves sooner, so causing them to begin feeding on exogenous food sooner than offspring of freshwater residents. Differences in early development between offspring from alternative life histories would provide evidence for parental effects as a potential mechanism underlying the expression of threshold traits and provide potential clarity as to the maintenance of alternative life histories in partially migrating populations.

## Methods

### Broodstock collection

Twenty-four mature freshwater-resident (12 male and 12 female) and 14 anadromous (seven male and seven female) brown trout were captured using electrofishing on 11 October 2013 and 23 October 2013 from two subtributaries of the River Tweed, Scotland. Freshwater-resident trout were collected from above an impassable dam on the Whiteadder River (55° 88′N, 2°57′W), while the anadromous trout were collected from the College Burn (55° 77′N, 2°18′W). Fish were classified as freshwater resident or anadromous based on size and coloration (Eek & Bohlin 1997): freshwater-resident fish were smaller and dark brown in colour with red spots, while anadromous fish were larger and silvery-grey in colour with black spots. There is no history of hatchery releases into either tributary. To further reduce the risk of misclassification, we collected our broodstock from two neighbouring tributaries which contained sufficient numbers of males and females of known life-history type. Average lengths of freshwater-resident and anadromous brown trout males and females were $214.8 \pm 28.8$ mm, $241.9 \pm 39.8$ and $425.0 \pm 27.8$ mm, $397.9 \pm 23.7$ mm (mean $\pm$ SD) respectively. Fish were transported to the Belhaven Trout Company, Scotland, where they were held in two round 1530-l aluminium tanks supplied with

$8.1 \pm 0.4$ °C (mean $\pm$ SD) well water under ambient photoperiod and assessed every 3 days for ripeness.

Ripe fish were anaesthetised, photographed, measured and blotted dry, and their eggs or sperm extruded by abdominal massage. A sample of between 9 and 22 unfertilised eggs from each female was collected and weighed to the nearest 0.0001 g on the day of stripping (freshwater resident $n = 162$; anadromous $n = 139$). These eggs were then frozen and later defrosted, reweighed (9–10 eggs per female) and dried at 40 °C for 12 days so that relative energy density (dry mass to wet mass ratio) of individual eggs could be determined. The remaining eggs from each female were photographed (so that individual eggs could be counted at a later date to determine clutch size) and fertilised with sperm from a haphazardly chosen male of the same life-history origin to create 12 full-sibling freshwater-resident families and 7 full-sibling anadromous families. Freshwater-resident and anadromous fish were artificially spawned from 3 November to 29 November and 17 November to 4 December 2013 respectively.

### Egg rearing, hatching and fish husbandry

Each family of eggs was housed separately in a plastic mesh egg basket, placed in one of two (1 m × 3 m × 0.4 m) rearing troughs supplied with well water and covered with dark plastic sheeting to ensure eggs were in complete darkness. Well water temperature during incubation was $8.1 \pm 0.4$ °C and was similar across the entire rearing period for each family of eggs and did not differ between life-history type and spawning time. Temperatures were recorded once daily along with any dead eggs which were carefully removed.

Eggs were checked daily for hatching; those from freshwater-resident and anadromous fish hatched from 19 December 2013 to 17 January 2014 and 30 December 2013 to 24 January 2014 respectively. Once eggs began to hatch, the newly emerged offspring (alevins) were counted, separated from the remaining eggs and gently placed into a small mesh basket (one per family) located in the same two troughs as the egg baskets. Ten to fifteen alevins from each family were blotted dry and weighed to the nearest 0.0001 g on the day of hatching.

On 31 January 2014, alevins were transported to the Scottish Centre for Ecology and the Natural Environment, Scotland. Families were housed separately in 15 l (50 cm × 30 cm × 15 cm) clear plastic aquaria on a partial recirculation system at a constant temperature of $9.2 \pm 0.2$ °C (mean $\pm$ SD) and ambient photoperiod. The aquaria contained a single air stone and were supplied with water pumped directly from Loch Lomond, which was first treated with an ozone gener-

ator (Sander S1000, Germany) before being discharged into a large sump. Water from the sump was pumped through an in-line 110W UV steriliser (Tropical Marine Center (TMC), Manchester, UK) before entering the aquaria. Return water was gravity fed into a large free standing filter before being discharged back into the main sump. Fish were monitored daily and any mortalities removed. The date of first feeding for each family was determined by visual observations as the point in time when the alevins' yolk sac was fully exhausted and the first individuals began to swim up from the bottom in search of food. Ten individuals from each family were anaesthetised, blotted dry and weighed to the nearest 0.0001 g to determine their mass at first feeding. Additionally, four fish from each family were selected at the point of first feeding for measurement of metabolic rate.

## Measuring maximal metabolic rate

Prior to standard metabolic rate (SMR) being measured overnight, individual fish were sequentially subjected to an exhaustive chase protocol in order to determine maximal metabolic rate (MMR; Killen et al. 2010; Norin 2014). A single fish was introduced into a round circular arena and hand chased for three minutes to exhaustion. The fish was then placed into 1 of 4 respirometry chambers as described below. Once a fish was placed in the chamber, the flush pump was immediately turned 'off' which allowed for the rate of oxygen uptake to be measured and determined as outlined below for SMR measurements. Maximal metabolic rate was determined as the highest oxygen consumption rate over the entire 24-h measurement period. In all cases, this value corresponded to the first measurement immediately following the exhaustive chase protocol.

## Measuring standard metabolic rate

Oxygen uptake was measured over a 24-h period, from approximately 10 AM onwards, using intermittent flow respirometry. Individual fish were placed into 1 of 4 separate (6.0 cm length, 1.4 cm diameter) glass respirometry chambers (Loligo systems, Tjele, Denmark) fitted with OXSP5 sensor spot (PyroScience GmbH, Aachen, Germany). Chambers were secured using a perspex microchamber holder (Loligo systems) and submersed in a water bath housed inside a constant temperature room. An air stone in the water bath of the respirometer apparatus kept the water fully saturated with oxygen. Water temperature averaged 9.2 ± 0.2 °C across all measurements. Opaque plastic partitions prevented visual contact between individual fish during measurements, and all measurements were conducted in the dark to further minimise fish activity

(Cutts et al. 2002). Glass respirometers and tygon tubing were used to prevent possible issues with use of plastics and oxygen permeable materials (Stevens 1992). Oxygen uptake was measured for 25 min every 45 min on a continuous 20 min 'on' and 25 min 'off' cycle. During the 'on' cycle, oxygenated water from the water bath was driven by a single water pump (Eheim 300 universal, Deizisau, Germany) through each of the respirometers. Flow rate was regulated by adjusting the tension of a hose clamp on the outflow side of the pump tubing to prevent swimming and spontaneous behaviour during flushing. After 20 min, the water pump was automatically switched off (Superpro MFRT-1 timer, Somerset, UK) allowing for a decrease in oxygen concentration to be measured during the 25 min 'off' period, during which a peristaltic pump (Masterflex L/S, London, UK) was used to ensure adequate mixing within each respirometer. Water oxygen concentration was measured every 1 s for 25 min during this time period. Oxygen concentration within the respirometer was measured using an oxygen meter (FireStingO$_2$ oxygen meter; PyroScience) fitted with 4 bare fibre oxygen probes; concentrations never dropped below 80% oxygen saturation in this experiment. Probes were calibrated daily, and rates of background oxygen consumption were subtracted from the observed values by measuring the oxygen concentration of water inside each of the respirometers in the absence of fish at the beginning and end of each measurement trial and assuming a linear decrease in oxygen concentration over the measurement period.

Fry at the point of first feeding was used to standardise for differences in development and hatch timing between families and ecotypes and prevent potential confounding effects of differential yolk sac mass in which little metabolic activity is thought to occur (Kamler 2008; Régnier et al. 2010). The rate of oxygen consumption was determined using the following equation (Ege & Krogh 1914):

$$MO_2 = V_w(\Delta C_w 0_2)/\Delta t$$

where $V_w$ is the volume of water in the respirometer and associated tubing minus the volume of the fish and $\Delta C_w 0_2$ is the change in oxygen tension of the water over time period $\Delta t$ (Steffensen 1989). Oxygen concentration was calculated by correcting PO2 (partial pressure oxygen) for barometric pressure and multiplying by αO2 ($\mu mol \cdot l^{-1} \cdot torr^{-1}$), the solubility coefficient at the observed temperature. Measurements of oxygen uptake were plotted graphically allowing for periods of complete rest to be readily discriminated from spontaneous activity, which appeared as distinct spikes. Standard metabolic rate was estimated using the average of the lowest 10%

of values observed during the respirometry trial (Norin 2014). Following respirometry measures, all fish were anaesthetised, blotted dry and weighed to the nearest 0.0001 g.

## Calculations and statistical analyses

Developmental rate between ecotypes and families was compared by measuring time to reach specific stages of development (i.e. time to hatch, time to first feeding), using accumulated thermal units (ATU, days × average daily temperature) from fertilisation (Taylor et al. 2000) to standardise for slight changes in water temperature and spawning time. Survival rate was quantified as the proportion of eggs per family surviving from fertilisation to hatch. Relative egg energy density was calculated by dividing the dry mass of individual eggs by their wet mass. Conversion efficiency was calculated over three key stages of development by dividing mass at hatching by egg mass, mass at first feeding by hatch mass and mass at first feeding by egg mass. Aerobic scope (AS) was determined by subtracting SMR from MMR. Survival data were arcsine transformed while SMR, MMR, AS, female length and egg number were $\log_{10}$-transformed to linearise the data and meet assumptions of normality and homogeneity of variance.

A general linear model was used to test for the effect of female length and ecotype on clutch size and to compare adult lengths between sex and ecotype (see Table S1). We used linear mixed-effects models (LME) to test for the effect of female length, egg mass and ecotype on hatch time, egg survival, hatch mass, time to first feeding and mass at first feeding. Furthermore, a LME was used to test for the effect of ecotype on egg mass, egg energy density and average conversion efficiency. Lastly to test for differences in SMR, MMR and AS between ecotypes, we again used an LME model but with absolute, rather than mass-specific, values of SMR, AS and MMR using mass as a fixed effect to control for differences in offspring mass. All LME models initially included all possible two way interactions and family as a random factor. Interaction terms and independent variables that were not significant at $P > 0.05$ were removed from the model, which was then rerun to obtain the model of best fit (see Table S1 for breakdown of models). Data for both ecotypes were first analysed together. If a significant interaction with ecotype was found, data for freshwater-resident and anadromous offspring were analysed separately using the above procedure but removing ecotype as a factor. All LME analyses were conducted using the nlme function (Pinheiro et al. 2011) and R version 3.0.1 statistical software (R Core Team, 2013).

## Results

### Broodstock

As expected, anadromous parental fish were larger overall at the time of spawning than freshwater-resident fish ($F_{1,34} = 292.42$, $P < 0.001$). However, there was no significant difference in length between freshwater-resident females and freshwater-resident males at the time of spawning ($F_{1,22} = 3.80$, $P = 0.06$), nor between anadromous females and anadromous males at the time of spawning ($F_{1,12} = 3.87$, $P = 0.07$).

### Egg size

Larger females produced heavier eggs ($F_{15,282} = 73.16$, $P < 0.001$) but not necessarily a greater number of eggs, as clutch size was negatively correlated with egg mass ($F_{15,282} = 11.42$, $P = 0.004$; Fig. 1). There was no significant difference in egg mass between eggs from anadromous females and those from freshwater-resident females (anadromous mean egg size $\pm$ SD: $0.0641 \pm 0.0110$ g; freshwater resident: $0.0611 \pm 0.0150$ g $F_{16,282} = 0.64$, $P = 0.43$). However, after controlling for differences in maternal body size, anadromous females produced a greater number ($F_{16,282} = 5.77$, $P < 0.0001$) of smaller eggs ($F_{15,282} = 10.32$, $P = 0.006$) than did freshwater residents (Fig. 2a,b). We found no significant difference in the energy density of eggs from anadromous and freshwater-resident females ($F_{18,164} = 1.47$, $P = 0.240$).

### Early development

There was no difference in the survival rate of offspring of the two parental ecotypes from the point at which eggs were mixed with sperm to hatching ($F_{17,282} = 0.90$, $P = 0.36$). There was a positive effect of egg mass on time to hatch ($F_{16,16} = 6.39$, $P = 0.02$ Fig. 3a), and a significant effect of parental ecotype, with eggs from freshwater-resident parents taking considerably longer to hatch than those from anadromous parents ($F_{16,16} = 83.38$, $P < 0.001$; Fig. 3a). However, while the mass of the alevin at hatching was positively related to egg mass (Fig. 4a), it did not differ between parental ecotypes, regardless of whether egg size was taken into account (controlling for egg size: $F_{16,16} = 2.48$, $P = 0.13$; ignoring egg size: $F_{17,17} = 0.06$, $P = 0.82$). The mean mass ($\pm$SD) of freshwater-resident and anadromous offspring at hatch was $0.0648 \pm 0.0517$ g and $0.0614 \pm 0.0537$ g respectively. There was no difference in the conversion efficiency of egg mass into hatching mass of the offspring from freshwater-resident and anadromous ecotypes ($F_{17,17} = 0.19$, $P = 0.67$; Fig. 5).
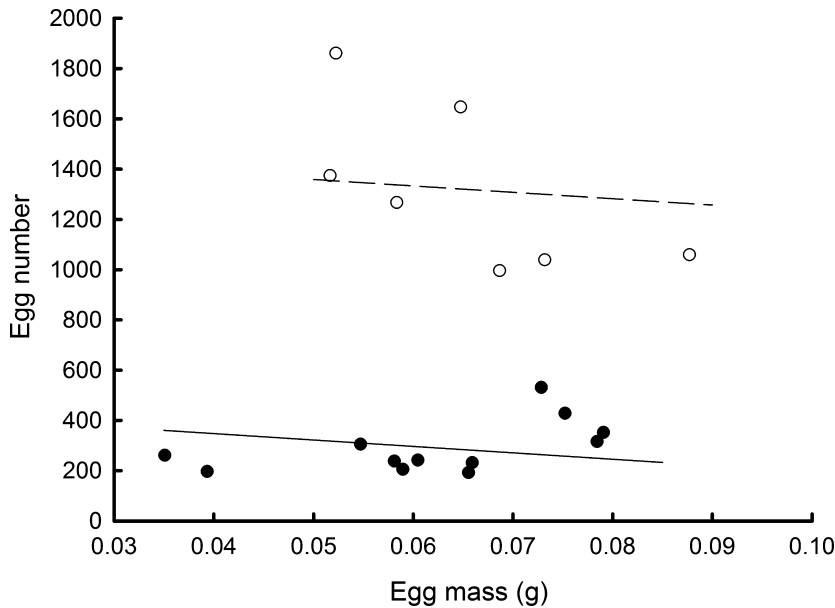
*Fig. 1.* The relationship between number of eggs (clutch size) and egg mass for freshwater-resident (closed circles, solid line) and anadromous (open circles, dashed line) females. See text for statistical analysis.

## First feeding

There was a significant difference between offspring of the two parental ecotypes in the later rate of development, with the time to first feeding of offspring of freshwater residents being considerably shorter than that for offspring of anadromous fish, whether this is taken from the time of egg laying ($F_{17,17} = 5.48$, $P = 0.03$; Fig. 3c) or from the time of hatching ($F_{16,16} = 26.10$, $P < 0.001$; Fig. 3b). Offspring of freshwater-resident parents were significantly heavier at the time of first feeding than those of anadromous parents (mean mass $\pm$ SD $0.1080 \pm 0.0245$ g and $0.0903 \pm 0.0152$ g respectively, $F_{17,171} = 4.42$, $P = 0.05$). We found a positive correlation between egg mass and first feeding mass ($F_{16,16} = 15.03$, $P = 0.001$; Fig. 4b); after controlling for this, offspring from freshwater resident crosses were considerably larger than those from anadromous crosses, whether the analysis ignored ($F_{17,171} = 4.42$, $P = 0.05$; Fig 4b) or controlled for maternal body length ($F_{16,171} = 17.74$, $P < 0.001$). There was no difference in conversion efficiency between the hatch stage and first feeding stage ($F_{17,17} = 1.05$, $P = 0.312$; Fig. 5), but we did find a difference in conversion efficiency between the egg stage and first feeding stage, with offspring from freshwater resident crosses converting egg mass to fry mass more efficiently ($F_{17,17} = 7.04$, $P = 0.02$; Fig. 5).

## Metabolism

Mass was highly significant in all models for SMR, MMR and AS. There was no difference in SMR ($F_{17,53} = 0.032$, $P = 0.58$; Fig 6a), MMR ($F_{17,53} = 1.27$, $P = 0.28$; Fig. 5b) or AS ($F_{17,53} = 1.29$, $P = 0.27$; Fig. 6c) between freshwater-resident and anadromous offspring at the time of first feeding.

## Discussion

Our study demonstrated that offspring from freshwater-resident and anadromous trout ecotypes differ significantly in their size and timing at various stages of early development. Not surprisingly, given the lower productivity of temperate freshwaters relative to marine environments at the same latitude (Gross et al. 1988), adult freshwater-resident trout were smaller than adult anadromous trout (Jonsson 1985; Jonsson & Jonsson 1997). Given their difference in body size, it was not surprising that anadromous trout produced more eggs than did freshwater residents, but these eggs did not differ in absolute size and were actually smaller relative to maternal body size than those of freshwater residents, a pattern consistent with sympatric brown trout populations found in parts of Sweden (Olofsson & Mosegaard 1999).

A reproductive strategy based on a greater number of smaller eggs (as was found for the anadromous females in this study) may be adaptive when habitat quality is high, as this may result in a greater return per reproductive effort (Brockelman 1975; Allen et al. 2008). As food availability in the environment increases, fitness among even the smallest individuals also increases, while the benefits of a larger egg (i.e. larger body size at hatch and/or greater energetic reserves) decreases, making production of more individuals, as opposed to fewer larger individuals adaptive (Rollinson & Hutchings 2013). The eggs of the
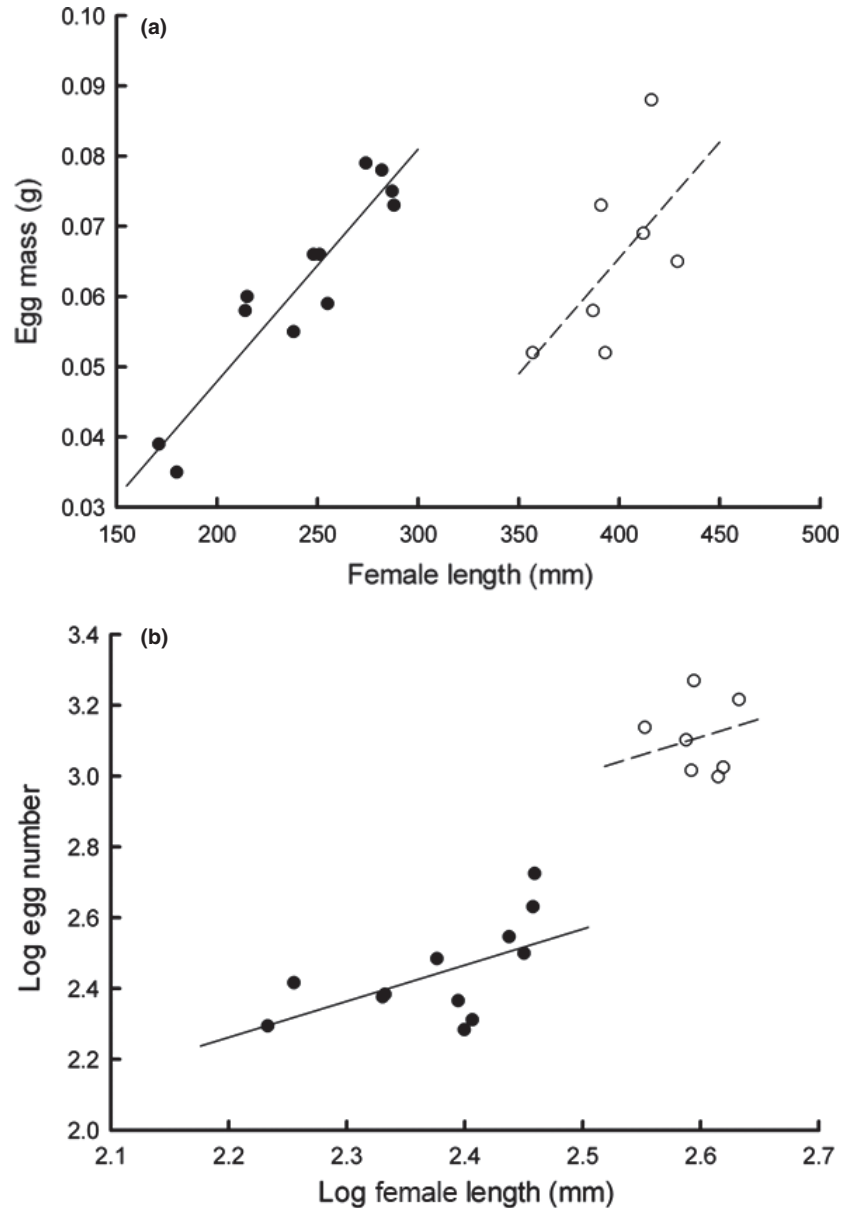
*Fig. 2.* The relationship between female length and egg mass (a) and egg number (b) for freshwater-resident (closed circles, solid line) and anadromous (open circles, dashed line) females. See text for statistical analysis.

freshwater-resident crosses hatched later than did those of anadromous crosses, but began feeding earlier and at a heavier mass. As there was no difference in our study in relative energy density of the egg or mass at hatching between the offspring of the two ecotypes, this meant that freshwater-resident offspring had a higher efficiency of conversion from egg mass to fry mass, although differences in energy density of the eggs between ecotypes have been noted elsewhere (Jonsson & Jonsson 1997).

The negative relationship between egg size (corrected for maternal body length) and the time taken to complete the stage from hatching to the start of exogenous feeding has been supported in other taxa (although see Pakkasmaa & Jones 2002 and Rollinson & Hutchings 2010 for exceptions). For example,

Dziminski et al. (2009) demonstrated that larger eggs of the quacking frog (*Crinia georgiana*) had a shorter larval period and were larger at metamorphosis. Large eggs generally give rise to large offspring at hatching (Reznick 1991; Roff 1992), which can confer benefits of greater competitive ability (Einum & Fleming 1999) and hence preferential access to food (Cutts et al.1999), although the incurred benefits of a larger body size at hatch may be species dependent and may differ based on the degree of parental care and the dependence level of the offspring on exogenous food resources. However, given that salmonid fry must compete for preferred feeding territories when they have emerged from the gravel at the start of exogenous feeding (Cutts et al. 1999; Van Leeuwen et al.2011), a process in which prior residence is
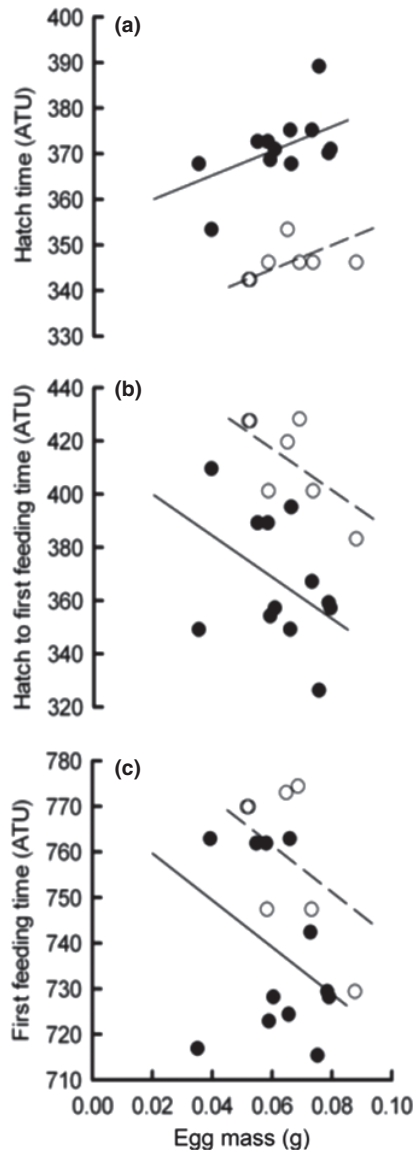
Fig. 3. The relationship between three development milestones [from fertilisation to hatching (a), from hatching to first feeding (b) and from fertilisation to first feeding (c)] and egg mass for freshwater-resident offspring (closed circles, solid line) and anadromous offspring (open circles, dashed line). Accumulated thermal units (ATU = days × average daily temperature). See text for statistical analysis.
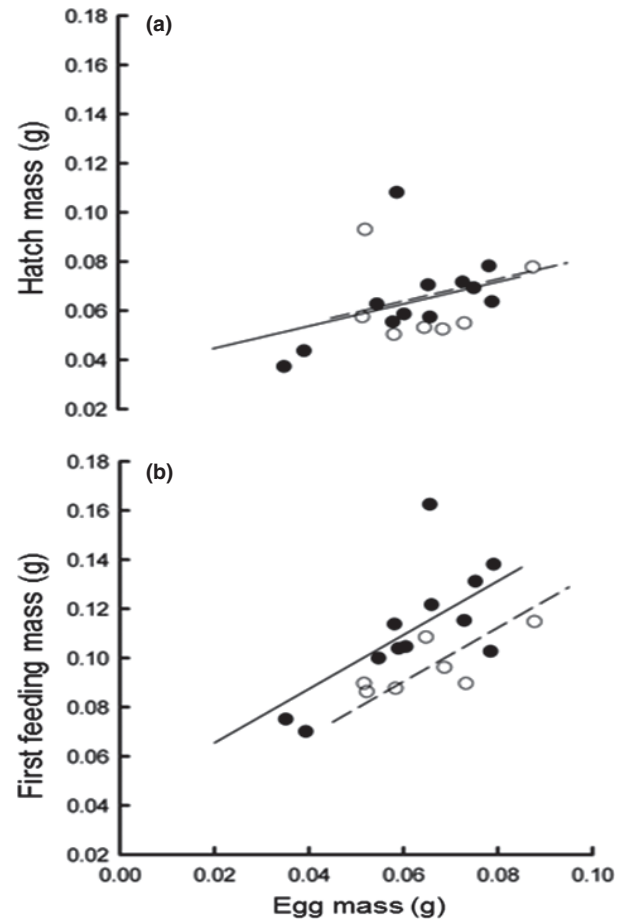


Fig. 4. The relationship between two developmental milestones [hatching (a) and first feeding (b)] and egg mass for freshwater-resident offspring (closed circles, solid line) and anadromous offspring (open circles, dashed line). See text for statistical analysis.

also important (Sloman & Armstrong 2002), a further advantage of producing larger eggs is that they had a reduced time taken to develop to the first feeding stage.

The relative time at which mothers spawn in the breeding season may also influence the viability of their offspring (Skoglund et al. 2012). Elliott & Hurley (1998) demonstrated that larger females within a population of anadromous brown trout spawned earlier in the season than did smaller members of the population. This did not appear to be the case in our study population, as the migratory females (which were larger than freshwater residents) tended to be ripe later than the freshwater residents, although these fish were held in the laboratory and were artificially stripped. Moreover, for a given egg size, the offspring from freshwater-resident crosses were larger at the onset of exogenous feeding than those from anadromous crosses and reached that stage sooner. It would therefore be expected (assuming the observed patterns in our study are representative of those found in sympatric populations) that the offspring from freshwater-resident crosses would have a competitive advantage over anadromous offspring during early ontogeny. Given this initial size/ prior residence advantage, the temporal stability of size based/dominance hierarchies in territorial juvenile salmonids (Bachman 1984; Abbott & Dill 1985; Nakano 1995), and the likelihood of higher growth supporting a resident life history (Olsson & Greenberg 2004; Olsson et al. 2006), it seems likely that a higher proportion of offspring from freshwater-resident crosses would remain freshwater resident whereas a higher proportion of offspring from anadromous
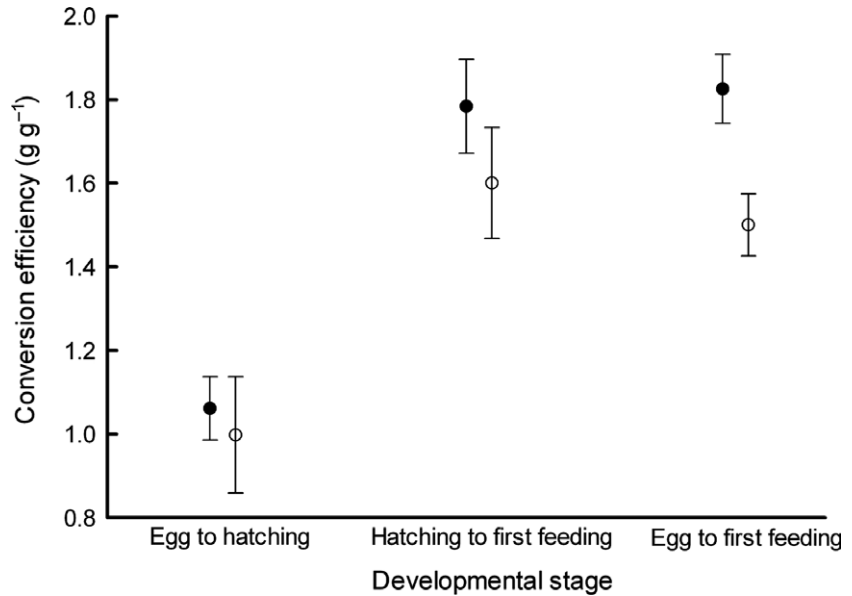
*Fig. 5.* The relationship between conversion efficiency at three developmental milestones (egg to hatching, hatching to first feeding and egg to first feeding for freshwater-resident offspring (closed circles) and anadromous offspring (open circles). Error bars represent standard error of the mean. See text for statistical analysis.

crosses would undergo at least some form of migration to maximise their growth opportunity.

It is unclear what factors contributed to the differences in body size between the two offspring types at the start of exogenous feeding although it is likely that these arose from a difference in conversion efficiency between the egg stage and the start of exogenous feeding, with offspring from freshwater-resident parents converting a given egg mass into a greater fry mass. Morinville & Rasmussen (2003) demonstrated that individual migrant brook trout (*Salvelinus fontinalis*) had a higher food consumption rate and lower growth efficiency in the year prior to migration compared to sympatric resident brook trout. Therefore, the pattern of higher conversion efficiency in offspring from freshwater-resident parents described at the embryo stage here may be maintained through ontogeny, although this remains to be tested in brown trout. Although not tested at the egg or alevin stage, one possible explanation for a difference in conversion efficiency would have been differences in minimal (SMR) or maximal (MMR) metabolism although no subsequent differences in SMR, MMR or AS were observed at the first feeding stage in our study. This result suggests that at least during the early fry stages, differences in overall baseline energy expenditure and aerobic capacity are similar between offspring types.

Given the differences in development between the two types of offspring in our study, we suggest that the migration history of the parents has the potential to influence the migration probability of the offspring. What remains unclear is whether the differences have a genetic basis (i.e. due to genetic differences between anadromous and freshwater residents), or are parental effects, possibly arising as a by-product of differing environments experienced by the two types of parents prior to spawning. There is a strong theoretical basis for variation in maternal provisioning of the egg arising as a by-product of differing environments. Mothers living in low food environments often exhibit reduced growth, smaller adult size and lower lipid reserves and in turn tend to produce fewer but larger eggs (Taborsky 2006), whereas those in high food environments show greater growth, larger size and higher lipid reserves and tend to produce a greater number of smaller sized eggs (Burton et al. 2013). Furthermore Braun et al.(2013) demonstrated that in years when migration was challenged by high water discharges, female Sockeye salmon (*Oncorhynchus nerka*) invest less in gonadal development and produce smaller but not fewer eggs, consistent with other studies demonstrating a negative relationship between egg size and migration cost (Fleming & Gross 1990; Kinnison et al. 2001). Therefore, given both the likelihood of nonbreeding partial migration to generate variation in the prebreeding environment (Burton & Metcalfe 2014) and the further constraints due to migration, it follows that anadromous females should be producing a greater amount of smaller eggs compared to freshwater-resident females.

A limitation of this study is that the age and spawning history (maiden spawners versus repeat spawners) of the females was not known. Reid & Chaput (2012) found that repeat spawning females of the closely related Atlantic salmon had a higher fecundity than maiden spawning females but produced smaller eggs with lower survival. Furthermore, in other species, it has also been shown that younger females for a given body size tend to produce smaller eggs (Quinn et al. 2011; Burton et al. 2013). Given
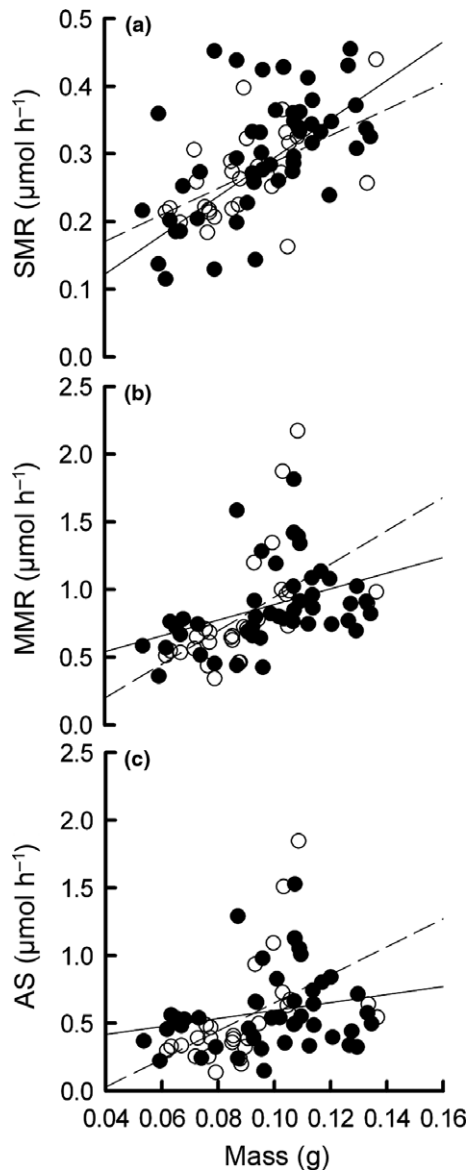
Fig. 6. The relationship between metabolic rate (SMR, MMR and AS) and mass for freshwater-resident offspring (closed circles, solid line) and anadromous offspring (open circles, dashed line). See text for statistical analysis but note that there were no significant differences between freshwater-resident and anadromous offspring in all metabolic measures.

the lower probability of repeat spawning in anadromous fish (due to higher mortality associated with migration) compared to freshwater-resident fish, it is possible that age and spawning history of the females could have influenced the patterns that we observed. A second limitation is that we were unable to determine whether the differences between offspring were primarily due to genetic or maternal effects. This would be difficult to establish given that the resident-anadromous dichotomy by its very nature prevents the use of the standard approach of rearing the parents in a common garden to rule out maternal effects. It is possible that the ontogenetic patterns described

here may be reduced or accentuated in the wild. Freshwater-resident and anadromous females may use different spawning habitats and have different spawning behaviour. For example, larger fish (i.e. anadromous fish in our study) have been shown to dig deeper nests and utilise larger substrates (Van den Berghe & Gross 1984) which could potentially influence developmental characteristics which may not necessarily be representative under benign laboratory conditions (Olsson & Persson 1988).

In conclusion, the results of this study demonstrate that offspring from freshwater-resident and anadromous life-history strategies differ considerably in early development but in a way that is consistent with literature-based predictions for triggers of migration (i.e. migratory fish have smaller eggs for a given maternal body size, while fry from larger freshwater-resident eggs begin feeding sooner and at a larger size). Therefore, we suggest that parental effects (either direct or environmentally mediated through differences in rearing environments) may be an important mechanism underlying the expression of threshold traits and play a significant role in the perpetuation of nonbreeding partial migration within populations.

## Acknowledgements

## References

Abbott, J.C. & Dill, L.M. 1985. Patterns of aggressive attack in juvenile steelhead trout (*Salmo gairdneri*). Canadian Journal of Fisheries and Aquatic Sciences 42: 1702–1706.

Allen, R.M., Buckley, Y.M. & Marshall, D.J. 2008. Offspring size plasticity in response to intraspecific competition: an adaptive maternal effect across life-history stages. American Naturlist 171: 225–237.

Bachman, R.A. 1984. Foraging behavior of free-ranging wild and hatchery brown trout in a stream. Transactions of the American Fisheries Society 113: 1–32.

Berthold, P. & Pulido, F. 1994. Heritability of migratory activity in a natural bird population. Proceeding of the Royal Society of London 257: 311–315.

Braun, D.C., Patterson, D.A. & Reynolds, J.D. 2013. Maternal and environmental influences on egg size and juvenile life-history traits in Pacific salmon. Ecology and Evolution 3: 1727–1740.

Brockelman, W.Y. 1975. Competition, the fitness of offspring, and optimal clutch size. American Naturalist 109: 677–699.

Brodersen, J.P., Nilsson, A., Hansson, L.A., Skov, C. & Brönmark, C. 2008. Condition-dependent individual decision-making determines cyprinid partial migration. Ecology 89: 1195–1200.

Burton, T. & Metcalfe, N.B. 2014. Can environmental conditions experiences in early life influence future generations? Proceeding of the Royal Society of London DOI: 10.1098/rspb.2014.0311.

Burton, T., Hoogenboom, M.O., Beevers, N.D., Armstrong, J.D. & Metcalfe, N.B. 2013. Among-sibling differences in the phenotypes of juvenile fish depend on their location within the egg mass and maternal dominance rank. Proceeding of the Royal Society of London DOI: 10.1098/rspb.2012.2441.

Chapman, B.B., Bronmark, C., Nilsson, J.A. & Hansson, L.A. 2011. The ecology and evolution of partial migration. Oikos 120: 1764–1775.

Cutts, C.J., Metcalfe, N.B. & Taylor, A.C. 1999. Competitive asymmetries in territorial juvenile Atlantic salmon, *Salmo salar*. Oikos 86: 479–486.

Cutts, C.J., Metcalfe, N.B. & Taylor, A.C. 2002. Juvenile Atlantic Salmon (*Salmo salar*) with relatively high standard metabolic rates have small metabolic scopes. Functional Ecology 16: 73–78.

Dodson, J.J., Aubin-Horth, N., Theriault, V. & Paez, D.J. 2013. The evolutionary ecology of alternative migratory tactics in salmonid fishes. Biological Reviews 88: 602–625.

Dziminski, M.A., Vercoe, P.E. & Roberts, D.J. 2009. Variable offspring provisioning and fitness: a direct test in the field. Functional Ecology 23: 164–171.

Eek, D. & Bohlin, T. 1997. Strontium in scales verifies that sympatric sea-run and stream-resident brown trout can be distinguished by colouration. Journal of Fish Biology 51: 659–661.

Ege, R. & Krogh, A. 1914. On the relation between the temperature and the respiratory exchange in fishes. Internationale Revue der gesamten Hydrobiologie und Hydrographie 7: 48–55.

Einum, S. & Fleming, I.A. 1999. Maternal effects of egg size in brown trout (*Salmo trutta*): norms of reaction to environmental quality. Proceeding of the Royal Society of London 266: 2095–2100.

Elliott, J.M. & Hurley, M.A. 1998. Predicting fluctuations in the size of newly emerged sea-trout fry in a Lake District stream. Journal of Fish Biology 53: 1120–1133.

Fleming, I.A. 1996. Reproductive strategies of Atlantic salmon: ecology and evolution. Reviews in Fish Biology and Fisheries 6: 379–416.

Fleming, I.A. & Gross, M.R. 1990. Latitudinal clines: a trade-off between egg number and size in Pacific salmon. Ecology 71: 1–11.

Forseth, T., Næsje, T.F., Jonsson, B. & Harsaker, K. 1999. Juvenile migration in brown trout: a consequence of energetic state. Journal of Animal Ecology 68: 783–793.

Grayson, K.L. & Wilbur, H.M. 2009. Sex- and context-dependent migration in a pond-breeding amphibian. Ecology 90: 306–312.

Gross, M.R., Coleman, R.M. & McDowall, R.M. 1988. Aquatic productivity and the evolution of diadromous fish migration. Science (Washington) 239: 1291–1293.

Imre, I., Grant, J.W.A. & Cunjak, R.A. 2005. Density-dependent growth of young-of-the-year Atlantic salmon in Catamaran Brook, New Brunswick. Journal of Animal Ecology 74: 508–516.

Jonsson, B. 1985. Life history patterns of freshwater resident and sea-run migrant brown trout in Norway. Transactions of the American Fisheries Society 114: 182–194.

Jonsson, B. & Jonsson, N. 1993. Partial migration: niche shift versus sexual maturation in fishes. Reviews in Fish Biology and Fisheries 3: 348–365.

Jonsson, N. & Jonsson, B. 1997. Energy allocation in polymorphic Brown Trout. Functional Ecology 11: 310–317.

Kamler, E. 2008. Resource allocation in yolk-feeding fish. Reviews in Fish Biology and Fisheries 18: 143–200.

Killen, S.S., Atkinson, D. & Glazier, D.S. 2010. The intraspecific scaling of metabolic rate with body mass in fishes depends on lifestyle and temperature. Ecology Letters 13: 184–193.

Kinnison, M.T., Unwin, M.J., Hendry, A.P. & Quinn, T.P. 2001. Migratory costs and the evolution of egg size and number in introduced and indigenous salmon populations. Evolution 55: 1656–1667.

Metcalfe, N.B., Taylor, A.C. & Thorpe, J.E. 1995. Metabolic rate, social status and life history strategies in Atlantic salmon. Animal Behavior 49: 431–436.

Monaghan, P. 2008. Early growth conditions, phenotypic development and environmental change. Philosophical Transactions of the Royal Society of London 363: 1635–1645.

Morinville, G.R. & Rasmussen, J.B. 2003. Early juvenile bioenergetic differences between anadromous and resident brook trout (*Salvelinus fontinalis*). Canadian Journal of Fisheries and Aquatic Sciences 60: 401–410.

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

Nakano, S. 1995. Individual differences in resource use, growth and emigration under the influence of a dominance hierarchy in fluvial red-spotted masu salmon in a natural habitat. Journal of Animal Ecology 64: 75–84.

Norin, T. 2014. Intraspecific variation in metabolic rates of fishes: causes, consequences and consistency, Ph.D. thesis. Aarhus University, Denmark.

Olofsson, H. & Mosegaard, H. 1999. Larger eggs in resident brown trout living in sympatry with anadromous brown trout. Ecology of Freshwater Fish 8: 59–64.

Olsson, I.C. & Greenberg, L.A. 2004. Partial migration in a landlocked brown trout population. Journal of Fish Biology. 65: 106–121.

Olsson, T.I. & Persson, B.G. 1988. Effects of deposited sand on ova survival and alevin emergence in brown trout (Salmo trutta L.). Archiv fur Hydrobiologie 113: 621–627.

Olsson, I.C., Greenberg, L.A., Bergman, E. & Wysujack, K. 2006. Environmentally induced migration: the importance of food. Ecology Letters 9: 645–651.

O'Neal., S.L. & Stanford, J.A. 2011. Partial migration in a robust Brown trout population of a Patagonian river. Transactions of the American Fisheries Society 140: 623–635.

Paez, D.J., Brisson-Bonenfant, C., Rossignol, O., Guderley, H.E., Bernatchez, L. & Dodson, J.J. 2011. Alternative developmental pathways and the propensity to migrate: a case

study in the Atlantic salmon. Journal of Evolutionary Biology 24: 245–255.

Pakkasmaa, S. & Jones, M. 2002. Individual-level analysis of early life history traits in hatchery-reared lake trout. Journal of Fish Biology 60: 218–225.

Pinheiro, J., Bates, D., DebRoy, S. & Sarkar, D. & the R Development Core Team. 2011. NLME: Linear and Nonlinear Mixed Effects Models. R package version 2.13.1.

Pulido, F. 2011. Evolutionary genetics of partial migration – the threshold model of migration revis(it)ed. Oikos 120: 1776–1783.

Quinn, T.P., Seamons, T.R., Vøllestad, L.A. & Duffy, E. 2011. Effects of growth and reproductive history on the egg size fecundity trade-off in steelhead. Transactions of the American Fisheries Society 140: 45–51.

R Core Team 2013. R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing. ISBN 3-900051-07-0, URL http://www.R-project.org/.

Régnier, T., Bolliet, V., Labonne, J. & Gaudin, P. 2010. Assessing maternal effects on metabolic rate dynamics along early development in brown trout (*Salmo trutta*): an individual-based approach. Journal of Comparative Physiology 180: 25–31.

Reid, J.E. & Chaput, G. 2012. Spawning history influence on fecundity, egg size, and egg survival of Atlantic salmon (Salmo salar) from the Miramichi River, New Brunswick, Canada. ICES Journal of Marine Science 69: 1678–1685.

Reznick, D.N. 1991. Maternal effects in fish life histories. In: Dudley, E.C., ed. The unity of Evolutionary Biology. Portland, OR: Dioscorides Press, pp. 780–793.

Roff, D.A. 1992. The evolution of life histories: theory and analysis. London, UK: Chapman and Hall.

Rollinson, N. & Hutchings, J.A. 2010. Why does egg size increase with maternal size? Effects of egg size and egg density on offspring phenotypes in Atlantic salmon (*Salmo salar*). Evolutionary Ecology Research 12: 949–960.

Rollinson, N. & Hutchings, J.A. 2013. Environmental quality predicts optimal egg size in the wild. American Naturalist 182: 76–90.

Rosenfeld, J.S., Bouwes, N., Wall, C.E. & Naman, S.M. 2013. Successes, failures, and opportunities in the practical application of drift-foraging models. Environmental Biology of Fishes 97: 551–574.

Sandell, M. & Smith, H.G. 1991. Dominance, prior occupancy, and winter residency in the great tit *(Parus major)*. Behavioral Ecology and Sociobiology 29: 147–152.

Skoglund, H., Einum, S., Forseth, T. & Barlaup, B.T. 2012. The penalty for arriving late in emerging salmonid juveniles: differences between species correspond to their interspecific competitive ability. Functional Ecology 26: 104–111.

Sloman, K.A. & Armstrong, J.D. 2002. Physiological effects of dominance hierarchies: laboratory artefacts or natural phenomena? Journal of Fish Biology 61: 1–23.

Steffensen, J.F. 1989. Some errors in respirometry of aquatic breathers: how to avoid and correct for them. Fish Physiology and Biochemistry 6: 49–59.

Stevens, D.E. 1992. Use of plastics materials in oxygen-measuring systems. Journal of Applied Physiology 72: 801–804.

Taborsky, B. 2006. Mothers determine offspring size in response to own juvenile growth conditions. Biology Letters 2: 225–228.

Taylor, L.R. & Taylor, R.A.J. 1977. Aggregation, migration and population mechanics. Nature 265: 415–421.

Taylor, E.B., Kuiper, A., Troffe, P.M., Hoysak, D.J. & Pollard, S. 2000. Variation in developmental biology and microsatellite DNA in reproductive ecotypes of kokanee, *Oncorhynchus nerka*: implications for declining populations in a large British Columbia lake. Conservation Genetics 1: 231–249.

Thorpe, J.E. & Metcalfe, N.B. 1998. Is smolting a positive or a negative developmental decision? Aquaculture 168: 95–103.

Van den Berghe, E.P. & Gross, M.R. 1984. Female size and Nest Depth in Coho salmon (*Oncorhynchus kisutch*). Canadian Journal of Fisheries and Aquatic Sciences 41: 204–206.

Van Leeuwen, T.E., Rosenfeld, J.S. & Richards, J.G. 2011. Failure of physiological metrics to predict dominance in juvenile Pacific salmon (*Oncorhynchus* spp): habitat effects on the allometry of growth in dominance hierarchies. Canadian Journal of Fisheries and Aquatic Sciences 68: 1811–1818.

Wysujack, K., Greenberg, L., Bergman, E. & Olsson, I. 2009. The role of the environment in partial migration: food availability affects the adoption of a migratory tactic in brown trout *Salmo trutta*. Ecology of Freshwater Fish 18: 52–59.

## Supporting Information

Additional Supporting Information may be found in the online version of this article:

**Table S1.** A table showing the full and reduced models used in the analyses along with AIC scores for the model selection.