Functional Ecology

British Ecological Society

Functional Ecology 2012 doi: 10.1111/j.1365-2435.2012.01999.x

Growth hormone reduces growth in free-living Atlantic salmon fry

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Summary

1. Although life-history theory predicts that juvenile growth rates should be high, there is substantial evidence that most juveniles grow below their physiological maximum. The endocrine system plays an important role in the determination of fundamental life-history traits, and hormones often serve as a link between an organism's environment and the expression of a trait. Particularly, growth is a life-history trait, which is strongly associated with growth hormone (GH) in fish, as well as most vertebrates.

2. To elucidate trade-offs related to elevated GH in fish in a natural environment, we experimentally administrated GH exogenously to juvenile Atlantic salmon using sustained-release GH implants, at an earlier ontogenetic stage than previously achieved (1·5 months). We assessed the effects on growth, dispersal and survival in contrasting environments.

3. Exogenous GH treatment increased the growth rate when fish were fed *ad libitum* in captivity. However, in a natural stream, GH treatment had a significant negative effect on growth and no apparent effect on survival or dispersal. This contrasts with previous studies conducted at later developmental stages, which show either a positive growth effect or no effect of elevated GH levels.

4. This study shows that environmental conditions strongly affect the response to GH and that under some natural conditions, it may also reduce growth. We suggest that the endogenous plasma GH levels may be maximizing growth during early, but not later, juvenile stages in nature.

Key-words: growth enhancement, juveniles, life history, *Salmo salar*, trade-offs

Introduction

In life-history theory, juvenile growth rates have been assumed to be near the physiological maximum, with variation arising primarily as a result of environmental factors, such as temperature and food availability (Stearns 1992). Considered in isolation, rapid growth rate should be beneficial because it reduces time spent at a vulnerable life stage and thus increases the probability of survival until reproduction (Stearns 1992). There is substantial evidence, however, that many organisms grow well below their physiological maximum (Arendt 1997). For example, both growth hormone (GH) treatment (Björnsson 1997) and GH transgenesis (Palmiter *et al.* 1982; Devlin *et al.* 1999) can induce substantial growth increases, indicative of an animal's innate capability to grow faster. Furthermore, animals commonly show compensatory growth following periods of starvation (Metcalfe & Monaghan 2001). As a result, growth rates in many animal populations may have evolved to be submaximal (Arendt 1997).

Life-history theory has traditionally focused on constraints involved in allocation of energy and other resources that creates trade-offs (Stearns 1992). However, less attention has been given to the endocrine regulatory mechanisms that lie beneath these trade-offs and how they are affected by environmental conditions (McGlothlin & Ketterson 2008). In

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most organisms, hormones often serve as a link between an organism's environment and the expression of an appropriate phenotype (McGlothlin & Ketterson 2008) and individuals may differ in the rate of hormone synthesis, release and degradation, leading to variation in circulating hormone levels (McGlothlin & Ketterson 2008). The endocrine system plays an important role in the determination of fundamental life-history traits, such as growth and reproduction, ageing and life span (Sinervo & Licht 1991; Björnsson 1997; Bartke 2008). Particularly, growth is a trait strongly associated with increased levels of GH in most vertebrates such as mammals, fish, reptiles and birds (Guillette, Cox & Crain 1996; Björnsson 1997; Beccavin *et al.* 2001; Bartke 2008).

The benefits associated with increased GH levels have been suggested to be traded off against such costs as increased susceptibility to predation owing to enhanced foraging (Jönsson, Johnsson & Björnsson 1996, 1998), reduced starvation tolerance owing to higher metabolism (Gotthard, Nylin & Wiklund 1994) and reduced developmental stability owing to decreased allocation of energy to maintenance of mature cells (Arendt, Wilson & Stark 2001). Furthermore, recent studies have shown that increased levels of pituitary GH, and thereby increased levels of IGF-1, which stimulates growth and metabolic activity in vertebrates, come with a cost in terms of decreased longevity (Bartke 2008).

Salmonid fishes show a great diversity of life-history patterns and have in recent years become model organisms for studying costs related to rapid growth following different methods for manipulation of growth such as GH transgenesis (Devlin *et al.* 2001) and exogenous GH treatment (Johnsson & Björnsson 1994) in natural and semi-natural environments (Johnsson *et al.* 1999, 2000; Johnsson & Björnsson 2001; Martin-Smith *et al.* 2004; Sundt-Hansen *et al.* 2009). The main endocrine regulator of growth is the GH – insulin-like growth factor I (IGF-I) system (Björnsson 1997; Bjornsson *et al.* 2002). In these salmonids, GH has a strong growth-promoting effect by increasing the hunger state and appetite of the fish (Johnsson & Björnsson 1994; Björnsson 1997) and by stimulating muscle and skeletal growth, as well as lipid mobilization (Björnsson 1997; Bjornsson *et al.* 2002). Growth hormone is also known to have an important osmoregulatory role in preadaptation to seawater during the parr-smolt transformation by stimulating Na^+,K^+-ATPase activity of the gills (Björnsson 1997).

Selection against rapid intrinsic growth has been hypothesized to occur at an early life stage in salmonids (Elliott 1994). Previous studies using exogenous GH treatment have failed to uncover a cost of rapid growth in juveniles that were 5–22 g in weight (Johnsson *et al.* 1999, 2000; Johnsson & Björnsson 2001). However, such a size is typically not reached until at least 2 months of age (i.e. postemergence) in nature, and negative effects of increased GH levels may be expected to be stronger earlier in life when vulnerability to starvation (Kennedy, Nislow & Folt 2008) and predation (Henderson & Letcher 2003) is highest and mortality rates may exceed 50% (Einum & Fleming 2000b). The aim of the present study was therefore to test for GH-mediated effects on growth, dispersal and survival of early juvenile Atlantic salmon (*Salmo salar*) in a natural environment. We predicted that GH, administered exogenously at an early developmental stage, would elevate growth and affect dispersal and survival, and reveal the presence of a trade-off between rapid growth (or high GH levels) and other life-history traits. This was accomplished during a developmental stage, which has previously not been studied with regard to this, but which may be important for evolution of growth rates owing to strong phenotypic selection (Einum & Fleming 2000a,b; Nislow, Einum & Folt 2004).

Materials and methods

STUDY SITE

The field study was conducted in the Stream Osalandsbekken, a small tributary (mean width, 3·8 m) to the River Imsa, which runs adjacent to the NINA Research Station Ims, south-western Norway (58°59′N, 5°58′E). The study stream possesses characteristics typical of a second-order salmon rearing stream of the region (Einum, Sundt-Hansen & Nislow 2006) and contains a natural population of brown trout (*Salmo trutta*), but no salmon owing to migration barriers further downstream. The mean temperature (±SD) during the field study was 19·1 ± 4·2 °C (range, 10·3–25·3 °C).

EXPERIMENTAL FISH

A sample of 10 males and 10 females of Atlantic salmon from the Imsa strain were caught in a fish trap in the river Imsa in November 2005 and used to produce 10 full-sibling family groups. The fertilized eggs were incubated on ambient temperatures at the NINA Research Station. The families were mixed at first feeding, which was initiated on 16 March 2006, and fry were fed *ad libitum* with commercial feed (EWOS AS, Bergen, Norway).

TREATMENT

During 2–4 May, 1200 fry were anaesthetized with benzocaine, group marked (site- and treatment-specific, visible implant elastomers; Northwest Marine Technology, Inc. Shaw Island, Washington, USA.) and implanted with GH or a sham for release at six different sites in Osalandsbekken (200 fish per site, half GH-treated and half sham-treated). To obtain a similar initial size range, fish were selected to be between 40 and 45 mm in body length (sub-sample measurements (n = 31): mean ± SD body length, 42 ± 1 mm; body mass, 0·69 ± 0·09 g). Following marking, each juvenile was implanted intraperitoneally with sustained-release recombinant bovine GH (bGH; Posilac®; Monsanto Company, St Louis, MO, USA) or a control treatment (sham-implanted) with a corresponding volume of vehicle (sesame seed oil). A 100-μL Hamilton syringe with a fixed needle, 0·45 mm in diameter and *c.* 1 cm in length, was used for this purpose. The Posilac formulation is viscous, requiring considerable pressure to be applied to the piston. To ensure that the correct amount of Posilac was implanted in each juvenile, small plastic discs were attached around the plunger shaft, with the thickness of each disc being equivalent to 2 μL of injection volume. Thus, as each new individual was to be implanted, a disc was removed. The GH treatment represented a dose of 1 mg bGH g^{-1} biomass, previously shown to elicit a growth response (McLean *et al.* 1997). An optical stereoscopic dissecting microscope was used to facilitate accuracy of the

injections, with the needle inserted a little to the left of the anterior portion of the belly. The fish were treated in groups of 50 (half GH and half sham) per release site to avoid the potentially confounding effects of timing of treatment relative to release timing. Following recovery from the anaesthetic, the fry were held in indoor tanks until release on 5 May.

RELEASE

After being transported to the stream, the fry were kept in enclosures for 3 h for acclimation prior to release. They were stocked at six different release sites, separated by 150 m. At each of these sites, 100 GH- and 100 sham-treated fry were released. To induce variation in the intensity of competition among release sites, 400 additional untreated fry of the same origin and size were released at three of the six sites, creating alternating high and low initial densities.

RECAPTURE

Recapture by electrofishing was conducted during 15–18 August 2006, 102–105 days after release. The stream was divided into 50-m sections, starting 150 m below the lowermost release site and ending 350 m upstream of the uppermost release site.

Electrofishing was initiated 150 m below the lowermost release site, and one pass of electrofishing was conducted until the first salmon was encountered. If a section yielded salmon, it was electrofished two to four times depending on catchability of fry (Bohlin *et al.* 1989). All juvenile salmon were anaesthetized and killed, and subsequently stored on ice for 1–8 h before being identified to treatment group and release site and measured for length and weight. Brown trout were also captured to estimate their densities for descriptive purposes. The trout were allowed to recover after sedation, before being released close to their capture site. For dry mass measurements of salmon, carcasses were dried at 60 °C for 3 days. The dry/wet mass (d/wm) ratio is positively correlated with lipid levels (McLean *et al.* 1997). Specific growth rates (SGR) were calculated for body mass M: $SGR_M = 100\,[\ln M_2 M_1^{-1}](d_2 - d_1))$ where $(d_2 - d_1)$is the time interval between measurements (M_1 and M_2) (Ricker 1979).

HATCHERY CONTROLS

Parallel to the field study, GH- and sham-treated fry were kept in indoor tanks ($n = 20$ individuals per tank) to monitor their growth pattern. Four tanks (60 L) were used per treatment (each tank containing a single treatment), and the fry were fed commercial feed (EWOS) *ad libitum* with automatic feeders. The initial body mass and length of these fish were similar to those in the field study. Measurements of body mass and length were conducted approximately once every month until the end of the experiment (6 June, 30 June and 18 August). The mean temperature (±SD) during the field study was 17·5 ± 4·1 °C (range, 8·6–23·2 °C).

STATISTICAL ANALYSIS

Successive removals by electrofishing allowed for the estimation of salmon and trout densities, and hence their numbers in the different sections, while accounting for potential variation in catchability according to the Zippin method (Zippin 1958; Bohlin *et al.* 1989). The total number of survivors from each release group (treatment within release sites) was estimated from the proportion of fry from the different release groups in each section and the total number of estimated fry in each section. Survival rates were arcsine square root transformed prior to statistical analyses. The d/wm ratio was ln-transformed prior to statistical analyses.

All statistical analyses were conducted using the statistical software R, v. 2.9.0. (R Development Core Team 2011). Linear mixed models performed with the function *lmer* in the *lme4* package (Bates & Maechler 2009) were used in analyses of survival, dispersal, individual final body length, final body mass (wet mass), d/wm ratio and dispersal. A model selection procedure based on Akaike Information Criterion (AIC, Burnham & Anderson 2002) was performed to identify the best models. As a first step, we compared models with and without random factors using restricted maximum likelihoods (REML). For all comparisons, the models including the random factors were better than those without (all $\Delta AIC > 2$). We then compared models with different random structures (Schielzeth & Forstmeier 2009): random intercept or random intercept and slope. In analysis of body mass and body length, the best models included recapture site as a random intercept ($\Delta AIC \geq 3{\cdot}94$). For analysis of d/wm ratio, the difference between the models including a random intercept or random intercept and slope was minor (ΔAIC of 1·4).Thus, according to the principle of parsimony, the model including recapture site as a random intercept was kept. For subsequent comparisons of models with different fixed terms, we used maximum likelihoods (ML) and a backwards selection procedure where different fixed effects and their interactions were removed sequentially from the full model until no further model improvement could be attained (Zuur *et al.* 2009). $\Delta AIC < 2$ was used as the selection criterion for enabling removal of both interactions and main effects. We present these final models in the results. The languageR package (Baayen 2008) was used to compute *P*-values for factors in the best models, where a *P*-value is returned based on a MCMC sample.

In the analysis of survival, the full model contained treatment (GH/sham) and released density (high/low) as fixed factors and release site as a random intercept. For analyses of individual final body length, final body mass and the d/wm ratio, fixed factors were treatment, released density and dispersal (i.e. recaptured fish that moved more or < 50 m from their release site), whereas recapture site was included as a random intercept. The inclusion of release site or recapture site as random factors controls for variation in habitat quality (including effects of experienced fish density). Differences in dispersal were tested using a mixed effects model with a binomial error family, and with fixed factors being treatment and release density, and with release site included as a random factor.

Differences in final body mass and length between GH- and sham-treated fry in the hatchery tanks were tested using treatment as a fixed factor and tank as a random intercept.

Results

FIELD EXPERIMENT

Recapture and densities

A total of 335 GH-treated and 345 sham-treated salmon fry were recaptured and identified to release site. The recapture rate did not differ between the two treatments ($\chi^2 = 0{\cdot}03$, $P = 0{\cdot}873$). In addition, 719 of the unmarked salmon fry were recaptured. Thus, the injection and marking treatments

did not appear to influence recapture rates, being 56% and 57% for treated (GH plus sham) and unmarked fry, respectively ($\chi^2 = 0{\cdot}06$, $P = 0{\cdot}796$). Losses from the population owing to emigration appeared to be low, as the first salmon juvenile was found in the section downstream and adjacent to the lowermost release site. Thus, the lowermost two sections (100 m) that were sampled did not yield recaptures, whereas all 18 sections between the lowermost release site and the uppermost release site yielded salmon. Estimated salmon densities ranged from 3·4 to 145·3 100 m^{-2} (mean ± SD, 35·7 ± 36·7). Estimated trout young-of-the-year densities ranged from 10·3 to 84·4 100 m^{-2} (39·1 ± 19·6), and the density of older trout ranged from 7·1 to 75·5 100 m^{-2} (37·8 ± 15·7).

Growth

The best models for growth-related traits (final body mass, length and d/wm ratio) included treatment and dispersal as fixed factors; thus, released density did not have an effect and was excluded from further analysis. The GH-treated fry showed a significantly lower body length, body mass and d/wm ratio than the sham-treated ones (Fig. 1, Table 1). Dispersal was significantly associated with these traits; fry that had moved more than 50 m from release sites were significantly larger than those that had stayed (Table 1, Fig. 2).

Both GH-treated and sham-treated salmon showed positive growth in the stream (Table 2), with a SGR of (mean ± SD) 2·1 ± 0·4% day^{-1} and a mean increase in body mass of 804% over the experimental period.

Dispersal

Of the recaptured fish, 82% of the GH-treated and 81% of the sham-treated fry were recaptured within 50 m of the release site (Fig. 3). The estimated proportion of fry that moved more than 50 m upstream from the release site was 17·9 ± 9·2% for the GH-treated fry and 19·7 ± 3·8% for the sham-treated fry (only one individual moved more than 50 m downstream). None of the fixed factors remained in the model for dispersal after the selection procedure.

Survival

The estimated survival was 58 ± 14% and 60 ± 6% for GH- and sham-treated fry, respectively. None of the fixed factors remained in the model for survival after the selection procedure.

HATCHERY TANKS

The GH-treated fry had a significantly higher growth in terms of final body mass ($t_{138,\ 8} = -3{\cdot}39$, $P < 0{\cdot}001$) and final length ($t_{138,\ 8} = -2{\cdot}32$, $P = 0{\cdot}028$) than the sham-treated ones (Fig. 4, Table 2). The best model explaining final body mass and final body length included treatment as a fixed factor and tank as a random factor. Fry in the hatchery growth control tanks had a SGR of (mean ± SD) 2·56 ± 0·38% day^{-1} and a mean increase in body mass of 1400% over the experimental period. The specific growth rate was 2·61 ± 0·3% day^{-1} and 2·43 ± 0·4% day^{-1} for the GH- and sham-treated fish, respectively.

Discussion

This study shows that exogenous GH treatment enhances growth of Atlantic salmon fry kept in indoor tanks and fed *ad libitum*, as expected from a range of previous studies, including Atlantic salmon parr of the same strain and kept at the same indoor facilities (Neregård *et al*. 2008). However, the present study also shows that GH treatment reduces growth of conspecifics released into a natural stream, in contradiction to our predictions. There are several nonexclusive factors that may explain this diametrically different effect of GH treatment on the growth of fish in different environments. Firstly, the exogenous GH treatment in parr and presmolt of Atlantic

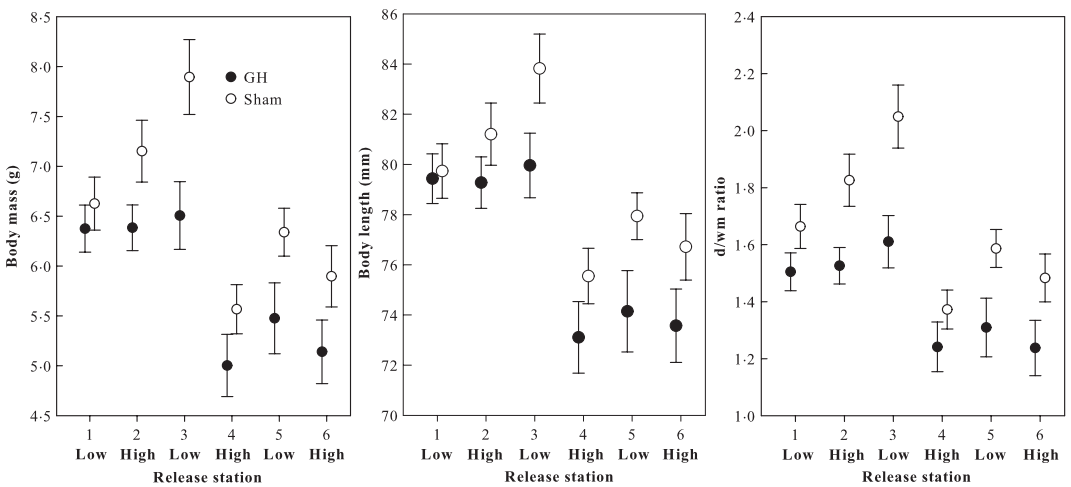

Fig. 1. The body mass, body length and dry/wet mass (d/wm) ratio (±1 SE) of growth hormone- and sham-treated Atlantic salmon fry by release site (high or low density) in stream Osalandsbekken. The release sites are numbered from lowermost to uppermost position in the stream.

Table 1. Analyses on individual data testing for the effect of treatment [growth hormone (GH) or sham], dispersal (moved more than 50 m or not) and recapture site on body length, body mass, dry/wet mass (d/wm) of Atlantic salmon fry in stream Osalandsbekken. *P*-values are computed from MCMC analyses

	Effect	Value	SE	d.f.	*t*	*P*	SD
Body length	Treatment	2·22	0·69	649	3·23	0·001	–
	Dispersal	5·45	1·30	649	4·19	< 0·001	–
	Recapture site	–	–	–	–	–	2·39
Body mass	Treatment	0·66	0·16	649	4·16	< 0·001	–
	Dispersal	1·52	0·32	649	4·78	< 0·001	–
	Recapture site	–	–	–	–	–	0·59
D/wm ratio	Treatment	0·03	0·01	649	6·23	< 0·001	–
	Dispersal	0·04	0·01	649	4·55	< 0·001	–
	Recapture site	–	–	–	–	–	0·02

Table 2. Summary of final body length (mm), body mass (g) and dry/wet mass (d/wm) ratio of recaptured GH- and sham-treated Atlantic salmon fry in stream Osalandsbekken and in the hatchery controls

	Field study				Hatchery controls			
	GH (n = 333)		Sham (n = 345)		GH (n = 62)		Sham (n = 76)	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Body length	77	10	79	9	97	8	90	3
Body mass	5·89	2·25	6·59	2·35	11·3	3·11	9·72	4·14
D/wm ratio	0·239	0·015	0·245	0·013	–	–	–	–

salmon can increase metabolic rate (Seddiki *et al.*1995; Herbert, Armstrong & Björnsson 2001), and GH-transgenic Atlantic salmon show a higher metabolic rate than non-transgenic salmon (Cook, McNiven & Sutterlin 2000). Furthermore, the temperatures in the last 2 months of the study were relatively high, with a mean temperature of 21·5 °C, whereas the optimal temperature for fast growing Imsa salmon when fed to satiation is 18·1 °C (Elliott & Hurley 1997). An elevated metabolic rate in combination with high water temperatures may have induced a greater energy loss in GH-treated than in sham-treated fry, explaining the growth differences. The lower d/wm ratio of the GH-treated fry, indicating a lower lipid reserve (McLean *et al.* 1992), supports the notion that GH-treated fish suffered a higher energy loss. The different growth response to exogenous GH treatment in contrasting environments is in accordance with earlier studies on salmonid species (Johnsson *et al.* 2000; Johnsson & Björnsson 2001). However, neither of these reported negative effects of GH relative to sham treatment on growth; rather, the response was either positive or absent. The fish in our study were too small for blood sampling, making it difficult to know natural levels of endogenous GH. Thus, for any of our results, we cannot exclude the possibility that our manipulation with exogenous GH resulted in unnatural levels that produced an effect on growth (elevating it in the captive fish or reducing it in the free-living fish) that would not have occurred under natural levels of the hormone. Furthermore, previous experiments show that levels of the bovine GH are elevated as long as 3 months after the implant (Gahr *et al.* 2008; Neregård *et al.* 2008).

Hypotheses explaining evolution of submaximal growth rates are usually framed within the growth-survival trade-off (Gotthard 2000; Brodin & Johansson 2004; Biro *et al.* 2006). However, none of the studies using exogenous GH treatment of salmonid fish, including the present one, have been able to demonstrate survival effects (Johnsson *et al.* 2000; Johnsson & Björnsson 2001). However, we cannot exclude the possibility for negative survival effects of elevated plasma GH levels at even earlier developmental stages. Some studies have shown that genotypes that have slow growth early in life tend to grow faster late in life and vice versa, a phenomenon termed convergent growth (Dmitriew, Blows & Rowe 2010). One explanation for this could be that the selection on size at maturity is strong relative to earlier juvenile stages, resulting in a canalization of the size at maturation (Metcalfe & Monaghan 2001; Flatt 2005). However, for fish that have indeterminate growth, this selection might be lower. Selection may be

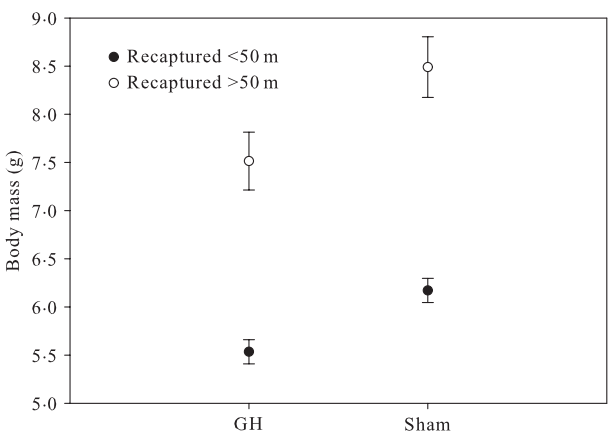

Fig. 2. The mean body mass (± 1 SE) of growth hormone- and sham-treated Atlantic salmon fry recaptured within ± 50 or > 50 m from their release site in stream Osalandsbekken.

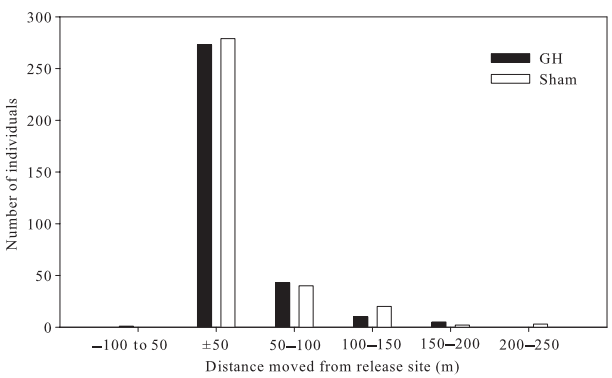

Fig. 3. The distance moved by the recaptured growth hormone- and sham-treated Atlantic salmon fry in stream Osalandsbekken.

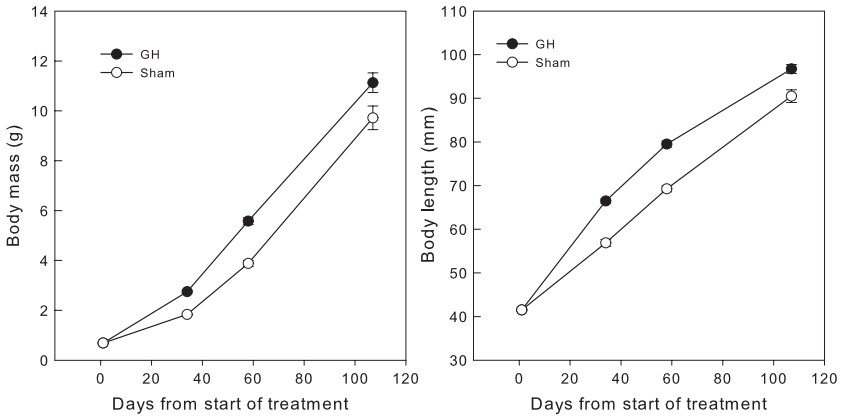

Fig. 4. Mean body mass and body length (±1 SE) of growth hormone- and sham-treated Atlantic salmon fry fed *ad libitum* in hatchery tanks.

acting differently on growth rate and consequently GH levels, across size classes and ontogenetic stages. Because competition is intense and selection for being large is particularly strong immediately after emergence from the nest (Brännäs 1995; Einum & Fleming 2000a,b; Nislow, Einum & Folt 2004; Einum, Sundt-Hansen & Nislow 2006), the fitness consequences of having submaximal growth may be more severe during this stage. Thus, the GH levels at the fry stage may be maximized to the prevailing environmental conditions.

What then is the mechanism behind the negative effect of exogenous GH treatment on growth as observed in the present study? The natural conditions in the stream affect the response to the GH manipulation in form of reduced growth. This may be due to the natural feeding conditions in the stream, which may have diminished the ability of GH-treated fry to reach their growth potential. Commercial feed has higher lipid (and energy) content than the natural feed (Bell, Ghioni & Sargent 1994), which may explain part of the growth difference observed between GH-treated fish in the hatchery and in the river. In GH-transgenic salmon, much of the growth enhancement diminishes when the fish are reared under naturalized conditions (natural food, presence of predators, complex habitat) compared with hatchery conditions (Sundström *et al*. 2007). Suppression of maximal growth rates has also been observed in cotton rats (*Sigmodon hispidus*), where growth rates were manipulated by subjecting them to elevated thyroid hormone levels (Derting 1989). When food abundance was high, the growth rates of the manipulated rats exceeded those of the control groups. However, when food was restricted, the growth rate of the control groups exceeded that of the manipulated group (Derting 1989). The benefits of the 'high-activity' strategy typical of growth-enhanced fish (Sundt-Hansen *et al*. 2009) may not only be related to resource levels *per se*, but also to the spatial/temporal distribution and other characteristics of available food resources, which differ between the hatchery and the wild. For example, a generally high activity level and prey search rate may be less beneficial when resource distribution is clumped and/or unpredictable in time (Adriaenssens & Johnsson 2011), or if it results in cryptic prey being overlooked (Johnsson & Kjallman-Eriksson 2008). This may ultimately constrain the growth of GH-treated Atlantic salmon fry in natural environments where they must search for food resources (as opposed to hatchery conditions).

When a life-history trait such as growth rate is selected through consecutive generations, later-acting selection pressures might oppose early-acting ones, creating a situation termed ontogenetic conflict (Chippindale, Gibson & Rice 2001; Rice & Chippindale 2001). Although simultaneous conflicting selection pressures are well documented (e.g. the effect of behaviour on growth vs. predation; Schluter, Price & Rowe 1991; Brodin & Johansson 2004), relatively less is known regarding conflicting selection pressure across ontogeny. One exception is given by the effect of seed size in oak (*Quercus ilex*), which is negatively correlated with post-dispersal seed predation, but positively correlated with subsequent germination and seedling survival (Gomez 2004). Based on the present study, a similar ontogenetic conflict may exist regarding the optimal levels of GH at different life stages in Atlantic salmon juveniles, and that opposing selection on GH levels at different life stages may lead to the evolution of submaximal growth at later life stages.

This study indicates that under certain, natural conditions, GH do not stimulate but actually reduce growth. Thus, the often assumed notion that it promotes growth may be unwarranted. Despite showing a physiological capability to grow very fast as a result of GH implants (as shown in the indoor tanks in the present study), the environmental conditions in the wild did not support such a growth for Atlantic salmon fry. On the contrary, the fry that had GH implants showed a poorer growth in the wild than those with a sham treatment. Thus, the optimal growth rate in a certain environment may not necessarily be the maximum growth rate possible, and this indicates that selection for rapid growth is constrained by costs that prevent evolution of maximum growth. Further investigation is needed to be able to properly distinguish between the ontogenetic differences in response to exogenous GH treatment from differences owing to environmental

conditions. As the environmental conditions and resource availability may vary a considerable degree both in space and time, the outcome of GH manipulation may not necessarily be the same in other locations, given different environmental conditions. Thus, growth conditions that change over time may cause selection on GH levels to fluctuate, and this may allow for a large number of genetic growth strategies to exist, even on a local scale (Dmitriew 2011).

Acknowledgements

We thank the three anonymous reviewers and editor for valuable comments on the manuscript. We also thank Anders Finstad and the staff at NINA research station Ims for technical assistance. The recombinant bovine growth hormone formulation (Posilac) was kindly provided by Monsanto Company, St Louis, MO, U.S.A. This work was supported by the FUGE programme of the Research Council of Norway and the Swedish Research Council for Environment, Agricultural Sciences and Spatial Planning (Formas). The study was conducted according to national regulations for the treatment and welfare of experimental animals.

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Received 28 March 2011; accepted 26 March 2012
Handling Editor: Keith Sockman