

Body shape and fin size in juvenile Atlantic salmon (*Salmo salar*): effects of temperature during embryogenesis

Larry Greenberg, Bror Jonsson, Johnny R. Norrgård, Ann Erlandsson, and Eva Bergman

Abstract: Temperature during egg incubation and early development influences later life stages of fishes, potentially influencing survival. Throughout its distribution, Atlantic salmon (*Salmo salar* Linnaeus, 1758) have experienced population declines, and in view of ongoing global warming, we tested if temperature during the earliest developmental stages modified body shape and fin size when temperatures averaged 2.6 vs. 5.6 °C. This temperature difference simulates increases predicted in climate change scenarios. Based on previous studies, we hypothesized that salmon originating from eggs subjected to cold incubation temperatures would have slimmer bodies and larger pectoral and dorsal fins than salmon from eggs that experienced warmer temperatures. After hatching, the juveniles were raised for 1 year under identical temperatures, after which we measured their body shape and fin areas. We found no support for our hypothesis regarding body shape. Indeed, we found the opposite, with cold-incubated salmon having deeper bodies than warm-incubated salmon. For fin size, the pectoral fins of cold-incubated salmon were larger than for warm-incubated salmon as predicted, but there was no difference in dorsal fin size. These results suggest that global warming may lead to altered body shape and fin size, possibly affecting swimming performance, and thus raise questions about the ecological consequences of the changes.

Key words: ectotherm, phenotypic plasticity, morphology, global warming, incubation temperature, *Salmo salar*, Atlantic salmon.

Résumé : La température durant l'incubation des œufs et le développement précoce influence les étapes subséquentes du cycle biologique des poissons et pourrait influencer leur survie. À la grandeur de son aire de répartition, le saumon atlantique (*Salmo salar* Linnaeus, 1758) a connu des baisses démographiques et, étant donné le réchauffement planétaire en cours, nous avons vérifié si la température durant les toutes premières étapes du développement modifie la forme du corps et la taille des nageoires quand les températures moyennes sont de 2,6 et 5,6 °C. Cette différence de températures simule des augmentations prédites par différents scénarios de changements climatiques. À la lumière d'études antérieures, nous avons postulé que les saumons issus d'œufs assujettis à de basses températures d'incubation auraient des corps plus élancés et de plus grandes nageoires pectorales et dorsales que les saumons issus d'œufs incubés à des températures plus élevées. Après leur éclosion, des poissons juvéniles ont été élevés pendant une année à des températures identiques, après quoi nous avons mesuré la forme de leur corps et l'aire de leurs nageoires. Nous n'avons trouvé aucun appui pour notre hypothèse concernant la forme du corps. C'est en fait le contraire que nous avons constaté, les saumons incubés à basse température présentant des corps plus hauts que les saumons incubés à température plus élevée. En ce qui concerne la taille des nageoires, les nageoires pectorales des saumons incubés à basse température étaient plus grandes que celles des saumons incubés à température élevée, conformément aux prédictions, mais les nageoires dorsales ne présentaient aucune différence. Ces résultats indiqueraient que le réchauffement planétaire pourrait mener à la modification de la forme du corps et de la taille des nageoires, ce qui pourrait avoir une incidence sur la performance de nage, et ils soulèvent des questions concernant les conséquences écologiques de ces changements. [Traduit par la Rédaction]

Mots-clés : ectotherme, plasticité phénotypique, morphologie, réchauffement planétaire, température d'incubation, *Salmo salar*, saumon atlantique.

Introduction

Increased temperature mediated by global warming represents a type of environmental stress with potentially large impacts on the earth's biota (Milligan et al. 2009; Bellard et al. 2012; Paaijmans et al. 2013; Henson et al. 2017). For instance, thermal stress during the earliest life stages can have long-term effects on subsequent

life stages (Burton and Metcalfe 2014; Jonsson and Jonsson 2014). Scenarios for the northern hemisphere, besides predicting a general increase in temperature during the 21st century, also predict relatively larger temperature increases in winter than in summer, and especially at the most northern latitudes (IPCC 2013). For instance, different scenarios for southern Scandinavia produced by the Swedish Meteorological and Hydrological Institute's

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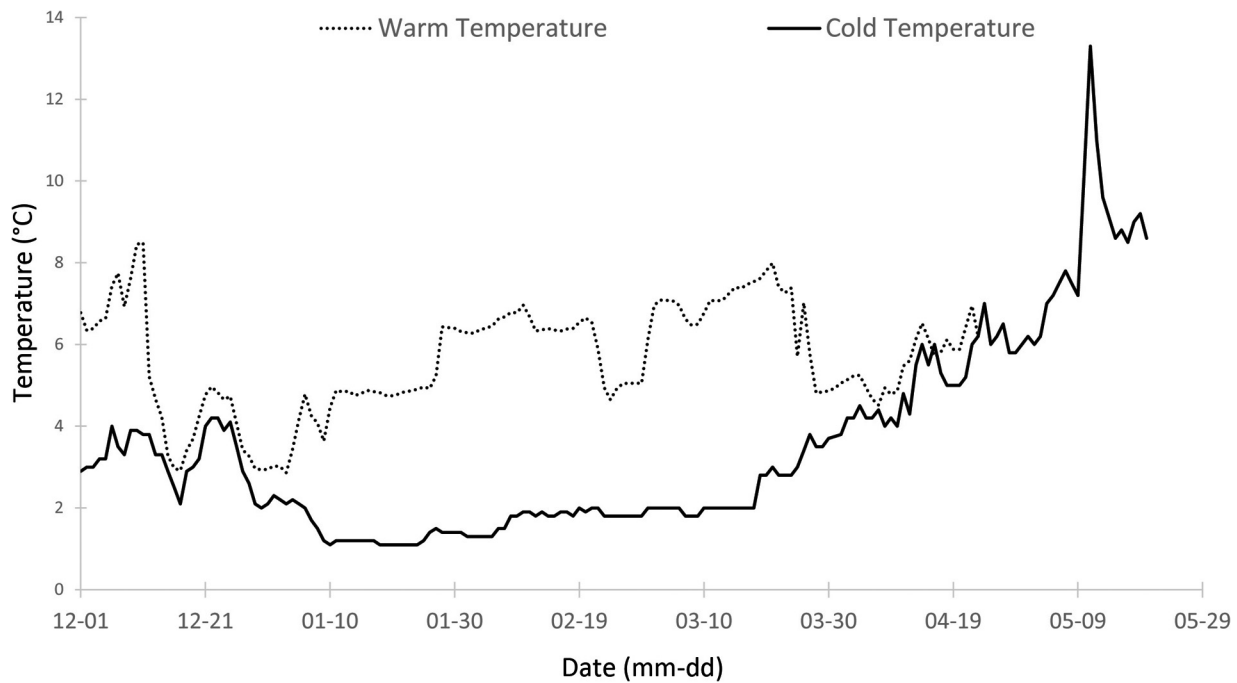
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Fig. 1. Water temperature of the warm-incubated (solid line) and cold-incubated (dotted line) Atlantic salmon (*Salmo salar*) during incubation until feeding was initiated. Dates show month and day in 2015 and 2016.



Rosby Center predict a temperature increase of 3 to 5 °C by 2080, with increases of 1.5 to 9.5 °C (mean of 6.2 °C) in winter and 2.3 to 5.3 °C (mean of 3.7 °C) in summer. Direct effects of temperature on growth of many animals, including poikilotherms, are well studied, but little is known about “carry-over effects” to later life-history stages and almost nothing is known about mechanisms of how developmental effects at the embryonic stage influence variation in later life-history traits (Jonsson and Jonsson 2019; Spinks et al. 2019).

The change in winter temperature may have far-ranging consequences for organisms adapted to current winter conditions. For ectotherms that incubate eggs during winter, such as Atlantic salmon (*Salmo salar* Linnaeus, 1758), warmer temperatures may have subsequent effects on later life stages (Jonsson and Jonsson 2019). Research has revealed that growth and other life-history traits are influenced by egg incubation temperature during winter (Finstad and Jonsson 2012; Jonsson et al. 2014). As this is an economically important species, with many populations decreasing in abundance during recent decades, possibly because of global warming (Chaput 2012; Friedland et al. 2014), it is important to document possible effects of changing temperatures that may influence the ecology and future evolutionary trajectories of this species.

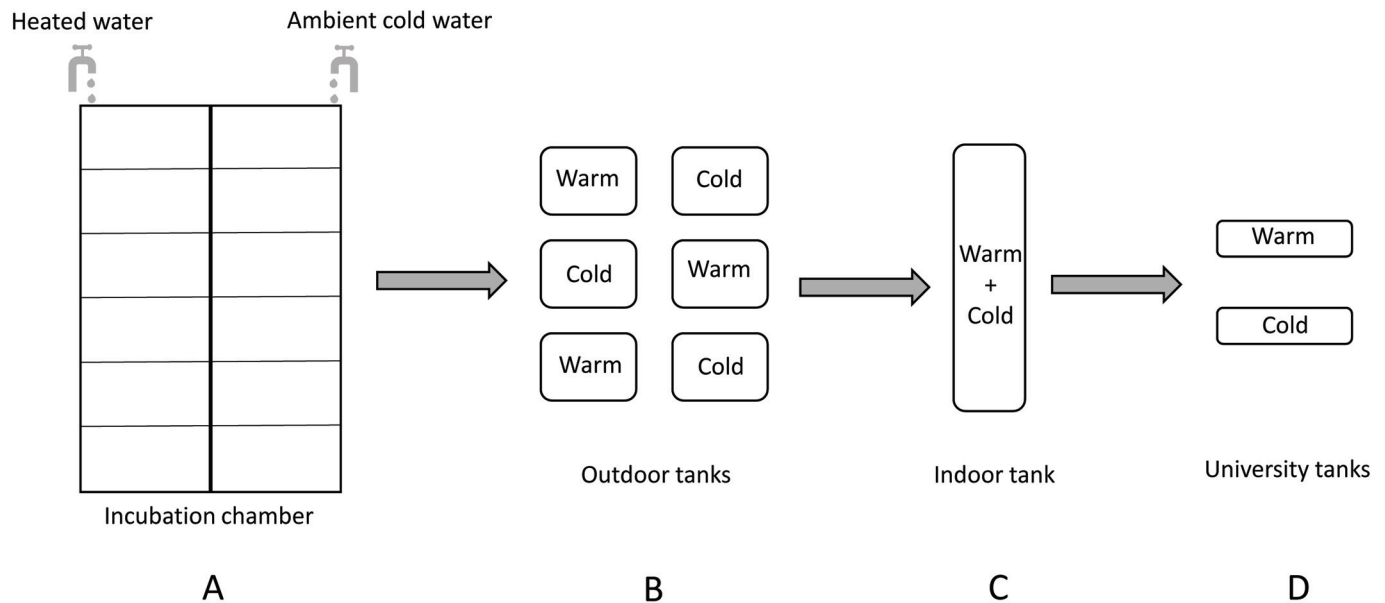
During the latter half of the 19th century, a positive relationship between latitude and the number of vertebrae in fishes was described and became what is referred to as Jordan’s rule (McDowall 2008; Jonsson and Jonsson 2011). In its original formulation, Jordan’s rule was not very specific, referring to fishes in general, but by the early to mid-20th century, the rule was shown to be applicable within a species. This latitudinal effect has since been attributed to early rearing temperature, where temperature is inversely related to the number of vertebrae (McDowall 2008; Sfakianakis et al. 2011a). Since the mid-1900s, it has been well documented for fishes and other ectotherms that temperature conditions during embryogenesis not only influence the number of vertebrae but also other skeletal features, such as the number of fin rays of teleost fishes (Tåning 1952; Billerbeck et al. 1997; McBride and Horodysky

2004). Recent studies have also linked the number of vertebrae and body shape, where more vertebrae tend to be associated with an elongated body shape (Aguirre et al. 2019; Corral and Aguirre 2019). Moreover, there is an association between early rearing temperature and body shape. For instance, Georgakopoulou et al. (2007) reported that body shape differentiation of sea bass (*Dicentrarchus labrax* (Linnaeus, 1758)) was influenced by temperature during larval rearing, from the embryo stage to the point of larval metamorphosis. They used two different temperatures (15 and 20 °C) and found that the number of dorsal spines was highest at 20 °C, numbers of pectoral and dorsal lepidotrichia were highest at 15 °C, and the fish had a slenderer shape at 15 °C than at 20 °C. Thermal rearing conditions from eggs to after larval metamorphosis had similar effects on zebrafish (*Danio rerio* (Hamilton, 1822)) (Sfakianakis et al. 2011a). These authors also revealed that there were close associations between body shape and number of vertebrae (see even Corral and Aguirre 2019), which also vary as predicted by Jordan’s rule (McDowall 2008). There is very little known, however, if thermal conditions before egg hatching influence body shape and fin size of fishes (but see Corral and Aguirre 2019).

Both body form and fins size are expected to influence swimming performance of fishes, and previous studies have revealed that larval temperature conditions affect the maximum critical water velocity for juvenile fish (Sfakianakis et al. 2011b). Early temperature also affects muscle development, including the number, size, and distribution of muscle fibers (Johnston 2006). Thus, temperature during embryogenesis may have consequences for body morphology, as fish with slow growth rates during embryogenesis typically develop smaller heads, eyes, and fins and a larger number of vertebrae and fin rays than fish with high growth rates (Barlow 1961).

The aim of this project was to study the effects of water temperature during the earliest development stages (embryo to early fry) on the body shape and fin size of juvenile Atlantic salmon. Based on findings by Georgakopoulou et al. (2007) and Sfakianakis et al. (2011a), we hypothesize that Atlantic salmon juveniles, whose

Fig. 2. History of the Atlantic salmon (*Salmo salar*) from fertilization until body shape and fin size were measured. (A) At the hatchery, the salmon were fertilized on 25 November 2015, placed in the incubation chamber the following day, and heating of the water commenced on 1 December 2015. (B) In spring 2016, the salmon were moved to 2 m² outdoor tanks. (C) In early November 2016, ca. 300 salmon were tagged with PIT tags and moved into a single 3 m² (1450 L) indoor hatchery trough for the winter. (D) In March 2017, the fish were transferred to Karlstad University and placed in two 600 L holding tanks.



eggs are incubated in ambient cold winter water, will have slimmer body shapes and larger pectoral and dorsal fins than conspecifics, whose eggs are exposed to elevated temperatures from fertilization to hatching. Hence, we tested whether or not the effect of temperature on body shape can occur early in life.

Materials and methods

Rearing conditions

Eggs of freshwater resident Atlantic salmon of Lake Vänern, Sweden, were raised in an incubation chamber under two different temperature regimes: a control group that was incubated using water piped in from the nearby Lake Yngen (cold conditions) and an experimental group using the same lake water but heated (warm conditions), approximately 3 °C above ambient temperatures (mean (\pm 1 SD) temperature of 2.6 ± 1.3 vs. 5.6 ± 1.3 °C during heating phase; Fig. 1). The fish came from a stock that historically spawned in the Gullspångsälven River. This stock is currently maintained by rearing eggs in a hatchery, releasing the fish into the River Klarälven, from where they migrate to Lake Vänern and return to Klarälven after 1–4 years in the lake. The gametes used in this experiment came from 12 female and 12 male spawners and were fertilized and reared at the Gammelkroppa fish hatchery, within the Gullspångsälven watershed near Filipstad, Sweden. On 25 November 2015, the eggs were fertilized using four randomly selected spawners (two males and two females) at a time and placed in different trays in the incubation chamber the following day. Heating of the water (Gorenje water heater, model TGR100) commenced on 1 December 2015 and continued until 22 April 2016. Ambient cold and heated water flowed (a flow-through system) through two separate columns of the egg incubation chamber, respectively (Fig. 2). The fertilized eggs were distributed among six trays in the column subjected to ambient (cold-incubated) water temperatures and six trays in the column subjected to elevated (warm-incubated) water temperatures. The eggs were incubated in darkness until hatching, followed by a natural light regime (using Philips Master 36 W, 840 lamps), and water quality is good and periodically tested (e.g., values for 20 March 2018: pH 7.3, conductivity

4.2 mS/m, alkalinity 0.21 meq/L ($1 \text{ mg/L HCO}_3^- = 61 \text{ meq/L}$), total phosphorus $<5 \mu\text{g/L}$, total nitrogen $310 \mu\text{g/L}$).

The warm- and cold-incubated Atlantic salmon initiated hatching on 16–20 February 2016 (after 429 degree-days) and on 16 April (after 367 degree-days), respectively. When external feeding was initiated (i.e., after yolk absorption was completed), the salmon were moved from the 12 incubation trays into six 2 m² (500 L) outdoor tanks. This was done by combining fish from two trays from the same temperature treatment into each outdoor tank, resulting in three tanks with warm-incubated fish and three with cold-incubated fish (Fig. 2). The date for first feeding of commercial pellets (Aller Aqua, Denmark) was 22 April for the warm-incubated salmon and 20 May for cold-incubated salmon. Hence, from the point when all fish were feeding externally, the salmon from the two egg incubation treatments were subjected to similar conditions. Between 22 April and 20 May, conditions differed slightly. Fish from both incubation treatments experienced the same temperature (same source water: 7.6 ± 1.8 °C) and light regime, but differed in that cold-incubated fish were subjected to artificial lighting and warm-incubated fish were subjected to natural light. The fish were fed daily rations of 4%–6% of their body mass, dependent on temperature. As during egg incubation, a flow-through system was used, with water piped in from the same nearby lake. Hence, temperature and light varied with outdoor conditions. On 2–4 November 2016, approximately 300 fish, 50 from each tank, were hand-netted, anesthetized with MS-222, and tagged with passive integrated transponder (PIT) tags. These fish were then placed in a single indoor hatchery trough (3 m², 1450 L) for the winter (Fig. 2). As before, a flow-through system using lake water was used, and temperature and light varied with outdoor conditions. In March 2017, the fish were then transferred to Karlstad University and placed in two 600 L holding tanks kept at the same temperature as at the hatchery (7 °C; Fig. 2). The fish were slowly acclimated at a maximum of 1 °C per day until temperature reached 13 °C and then maintained at this temperature for the duration of this study and subsequent foraging studies. The aquaria were filled with filtered tap water, and an Eheim filter and pump

recirculating system was used to maintain water quality. Approximately 25%–40% of the water was changed every week. The fish were fed pellets (Aller Aqua, Denmark) and thawed red chironomids once per day.

Body shape and fin measurements and analysis

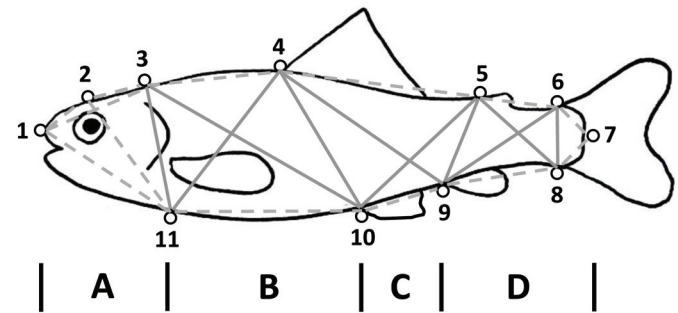
In June 2017, the fish were collected from the tanks and identified with a handheld PIT-tag reader. The fish were anesthetized with MS 222 and then weighed, measured for length (total length), and placed on a round tray for photographing. The dorsal fins were held up by hand using a pair of tweezers. A Canon XA10, mounted on a tripod directly above the fish, was used to take the photographs. The fish were cared for and handled in accordance with Swedish legal requirements, based on a permit from the Swedish Animal Welfare Board, Department of Agriculture (permit Dnr 88-2013).

A total of 62 individuals, 30 from the cold group (99.0 ± 14.3 mm (mean \pm 1 SD); 9.6 ± 4.0 g (mean \pm 1 SD)) and 32 from the warm-incubated group (95.6 ± 22.6 mm; 9.1 ± 4.3 g), was used. The fish did not differ in size (length: $t_{[60]} = 0.70$, $P = 0.48$; mass: $t_{[60]} = 0.45$, $P = 0.65$). The photographs of the dorsal fin area could be properly measured for 49 of the 62 salmon (19 cold-incubated and 30 warm-incubated fish); the others were discarded mostly because of incomplete fins due to fin erosion (common in hatchery raised fish), but also poor-quality photographs. For the pectoral fin, all but one photo from the cold-incubated group ($N = 29$) and all of the warm-incubated fish ($N = 32$) could be analyzed. The fin areas were measured using the computer program tpsDig2w32 version 2.30 (Rohlf 2017). The arithmetic scale was set in the program using the diameter of the tray (16.6 cm diameter) on which the fish were lying. To investigate whether there were statistically significant size differences in the area of dorsal fins and pectoral fins between the groups, an ANCOVA was performed, and controlled for body size. Before running the ANCOVA, a principal components analysis (PCA) of the size variables (mass, length, and body area) was performed as all three size variables correlated positively with each other (>0.965). In the ANCOVA, the PCA component of the size variables was then used as a covariate. The assumption of homogeneity of variance (Levene's test) was fulfilled.

The body shape of the 62 fishes was examined by selecting 11 landmarks that were easily defined on the photographs (Fig. 3). The following landmarks, indicated by a number from 1 to 11, were used to describe body shape: 1, tip of the snout at the upper jaw; 2, on the dorsum, directly above the eye; 3, the posterior portion of the neurocranium; 4, anterior margin of the dorsal fin; 5, anterior margin of the adipose fin; 6, dorsal portion of the caudal peduncle; 7, the midpoint at the base of the caudal fin; 8, the ventral portion of the caudal peduncle; 9, anterior margin of the anal fin; 10, anterior margin of the pelvic fin; 11, directly under of the anterior edge of the pectoral fin. These landmarks encompassed the different regions of the body, namely the head region, the anterior mid-region, the posterior mid-region, and the caudal region. In addition, the maximum body depth and standard length were measured from the photographs to be able to compute the fineness ratio (standard length/body depth), a common measure of elongation relative to a fish's transverse sectional diameter, with larger values indicative of a more streamlined body shape (Aguirre et al. 2019).

The computer program tpsDig2w32 program was used to measure 23 Euclidean distances between the 11 landmarks (Fig. 2), and the distances formed a box-truss network (Strauss and Bookstein 1982) of distance variables, representing the body shape of the fish. Since the values of the distance variables are related to the size of the fish, each variable was divided by the geometric mean of all variables for that individual (Jungers et al. 1995). After the distance variables were standardized for size, a Kolmogorov–Smirnov test and a Shapiro–Wilk test were performed, showing that the variables followed a normal distribution ($P > 0.05$). A logistic regression was then used to test if the body shape metrics could

predict if and how well the cold-incubated and warm-incubated groups could be separated. As there were 23 body shape metrics and 62 fish, the number of variables needed to be reduced. Hence, a 23×23 Pearson correlation matrix was computed to identify issues of collinearity. We found that the variables associated with the body contour (along the anterior–posterior axis) were often correlated with the different metrics that traversed the body along the dorsal–ventral axis. Hence, we removed the body contour metrics, retaining only a network of six adjacent triangles, or 10 lengths, that traversed the body, excluding the most anterior metrics (Fig. 3). Correlations between these 10 lengths were weak, with correlation coefficients less than about 0.4 (16% explained variation), averaging 0.18 ± 0.12 (mean \pm 1 SD) in absolute value. The resultant 10 variables were 3–11, 3–10, 4–11, 4–10, 4–9, 5–10, 5–9, 5–8, 6–9, and 6–8 (Fig. 3). Statistical analyzes were performed in IBM SPSS Statistics 24.



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Results

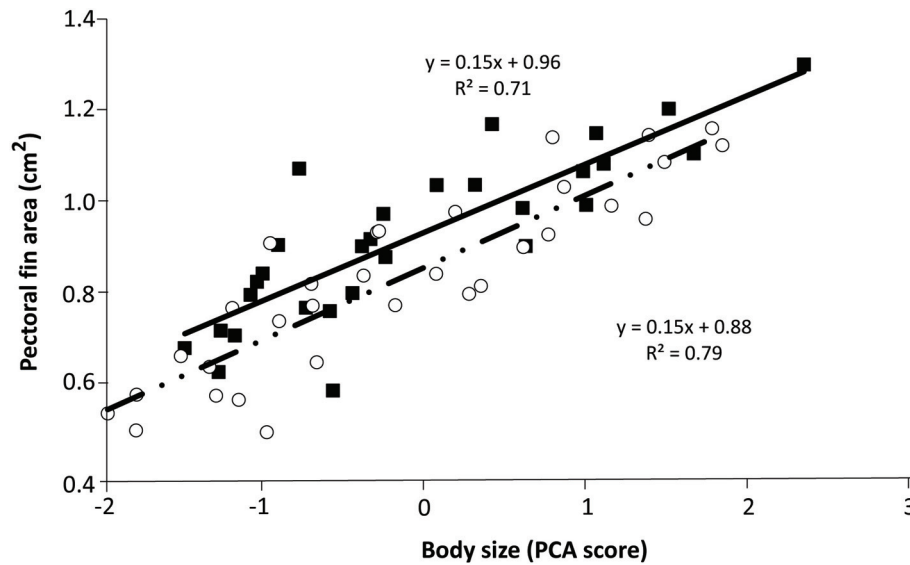
Fin area

There was a significant linear relationship between body size and pectoral fin area for fish from both incubation temperatures (cold-incubated fish: $y = 0.15x + 0.96$, $R^2 = 0.71$; warm-incubated fish: $y = 0.16x + 0.88$, $R^2 = 0.79$). The area of the pectoral fins was greater in the cold group (mean \pm 1 SD: 0.96 ± 0.17 cm²) than in the warm-incubated group (mean \pm 1 SD: 0.88 ± 0.18 cm²), and this difference was significant when controlling for body size (ANCOVA, $F_{[1,58]} = 10.164$, $P = 0.002$; Fig. 4). There was not a significant linear relationship between body size and dorsal fin area for fish from both incubation temperatures, nor was there a significant difference between the relationships when controlling for body size (ANCOVA, $F_{[1,46]} = 0.65$, $P = 0.799$).

Body shape

Logistic regression on the 10 body shape metrics produced a significant model (Omnibus, $\chi^2_{[10]} = 61.175$, $P < 0.001$) with a good fit (Hosmer–Lemeshow test, $\chi^2_{[8]} = 1.700$, $P = 0.99$). The logistic regression function correctly predicted 86.7% of the fish belonging to the cold-incubation group and 93.8% to the warm-

Fig. 4. Relationship between pectoral fin area and body size for cold-incubated (open circles, dash-dot-dot line) and warm-incubated (solid squares, solid line) Atlantic salmon (*Salmo salar*).



incubated group, for an overall prediction of 90.3% correct. There were four metrics, located along the entire length of the fish that significantly contributed to the equation (Figs. 3 and 5). These metrics are distance 3–11 from the head region, distance 4–11 from the anterior mid-region, distance 5–10 from the posterior mid-region, and distance 6–8 from the caudal region (Table 1). When comparing body shape, the cold-incubated fish had deeper bodies than the warm-incubated fish (Fig. 5). The fineness ratio also differed between fish from the two incubation temperature treatments, with warm-incubated fish having a larger fineness ratio (and thereby more elongated body) than cold-incubated fish (ANOVA, $F_{[1,60]} = 31.6$, $P < 0.0001$; Fig. 6).

Discussion

Based on findings by Georgakopoulou et al. (2007) and Sfakianakis et al. (2011a), we hypothesized that the warm-incubated salmon would have a deeper body shape than the cold salmon, but this was not corroborated by our study. Indeed, we found the opposite, with Atlantic salmon incubated at ambient cold temperatures having a deeper body shape than conspecifics incubated at elevated temperatures. The observed difference in body shape may be related to the number of vertebrae, but it may also reflect differences in musculature, as incubation temperature has been shown to affect muscle development, with effects on ultrastructure, number, and phenotype of muscle fibers of juvenile fish (Johnston 2006; Scott and Johnston 2012). Notwithstanding the situation for body shape, we did find support for our hypothesis that the pectoral fins of the cold-incubated salmon had a larger area than the warm-incubated salmon, but no relationship was observed for dorsal fins, possibly due to the smaller sample size. Thus, body shape and fin size responses to environmental change may be difficult to predict from general patterns and rules, as for example Jordan's rule. There are often exceptions to these rules; for example, Täning (1952) reported different types of V-shaped responses for the relationships between both the number of fin rays and vertebrae with temperature for anadromous brown trout (*Salmo trutta* Linnaeus, 1758). The V-shaped relationship for the number of vertebrae had a minimum at 6 °C and higher numbers if incubated at both colder and warmer temperatures, but an inverted V-shape for fin rays, with the highest number of fin rays between 8 and 10 °C. We do not know how well meristic

relationships from brown trout fit for Atlantic salmon. Our warm-incubated group was incubated at about 6 °C, about 3 °C warmer than the cold group, and this should, based on Täning (1952), give fewer vertebrae and more fin rays in the warm-incubated salmon than in the cold-incubated salmon. We did not measure the number of skeletal elements, but assuming body shape and fin area are related to the number of vertebrae and fin rays, respectively, it seems likely that a slenderer body shape and a larger fin area would indicate a larger number of vertebrae and fin rays (cf. Aguirre et al. 2019; Corral and Aguirre 2019), but this remains to be examined in the present case.

We do not know when during early development that the morphology of juvenile Atlantic salmon is sensitive to the thermal environment. We expect that there are developmental windows when salmon are particularly thermo-sensitive as found for a number of organisms (Fawcett and Frankenhuis 2015). As the temperatures of cold- and warm-incubated groups were similar in December, we assume that the morphology of Atlantic salmon was affected by thermal conditions after this first month. Generally, it appears that developing poikilothermic organisms are most sensitive to temperature conditions during the earlier phases of embryonic incubation (Henry and Uljaszek 1996; Lindström 1999), which is certainly the case for environmental effects on sex determination that occur in the middle trimester of embryogenesis (Bull 1983). Perhaps, this is a sensitive period influencing morphological differentiation as well.

Our results confirm that salmonids exhibit great morphological plasticity in response to environmental changes. Plasticity in vertebral counts and number of fin rays has been known for a long time (Täning 1952; Barlow 1961; Currens et al. 1989). More recently, it was shown that counts of vertebrae and number of fin rays influence body shape and fin sizes as revealed for the genus *Rhoadsia* Fowler, 1911, which is endemic to western Ecuador and northwestern Peru (Aguirre et al. 2019). Similarly, Mexican tetra (*Astyanax mexicanus* (De Filippi, 1853)) raised in 20 and 28 °C water had a higher number of vertebrae and a more elongated body shape than fish raised at temperatures in between (Corral and Aguirre 2019). Environmental influence on body shape has also been shown for Chinook salmon (*Oncorhynchus tshawytscha* (Walbaum in Artedi, 1792)), where those from an ephemeral stream had deeper bodies than those that came from continuously

Fig. 5. Photographs of a typical example of a cold-incubated (10.5 cm total length) and warm-incubated (10.8 cm total length) Atlantic salmon (*Salmo salar*), illustrating the effect of incubation temperature on body shape. The four lines on each fish show the length metrics that contributed significantly to the logistic regression (region A at points 3–11; region B at points 4–11; region C at points 5–10; region D at points 6–8). Colour version online.

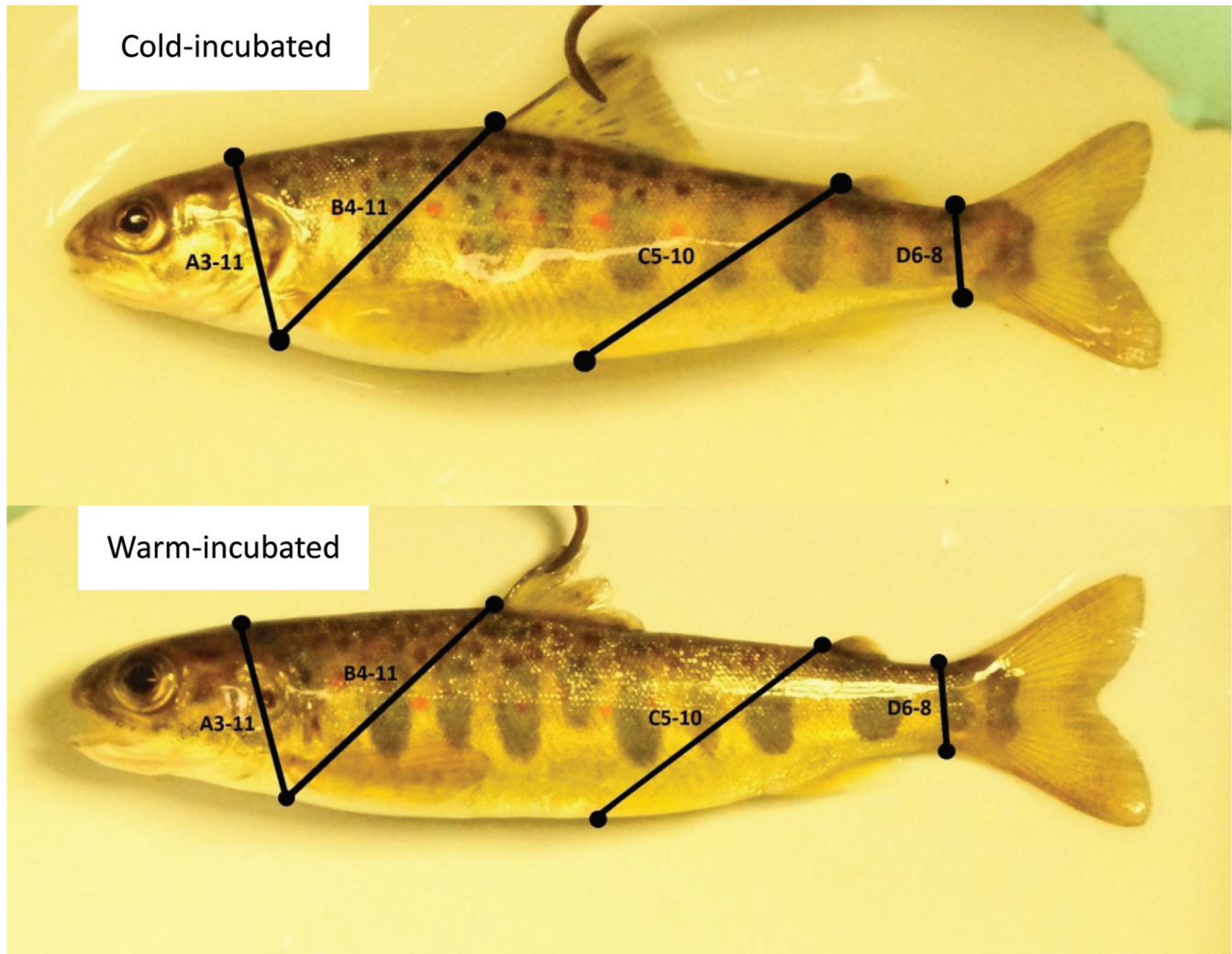
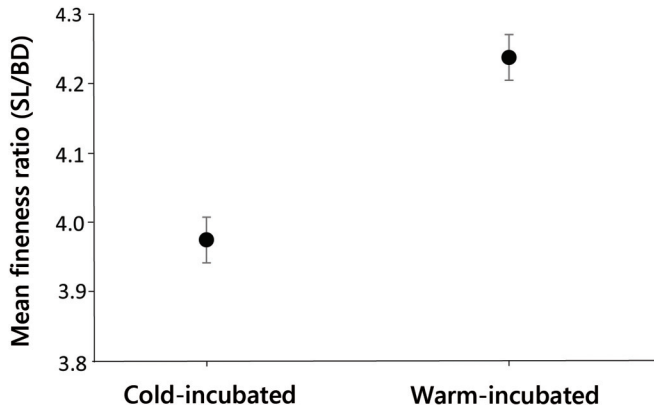


Table 1. Results of logistic regression for the 10 selected morphological variables describing body shape for warm-incubated and cold-incubated Atlantic salmon (*Salmo salar*).

Variable	B	SE	Wald	Probability	Exp(B)	Cold	Warm
3–11	79.86	40.02	3.98	0.046	4.8×10^{34}	0.96 ± 0.02	0.95 ± 0.02
3–10	0.57	14.09	0.002	0.968	1.8	2.11 ± 0.04	2.12 ± 0.05
4–11	66.40	32.95	4.06	0.044	6.9×10^{28}	1.57 ± 0.03	1.54 ± 0.04
4–10	-24.98	34.70	0.52	0.472	1.4×10^{11}	1.27 ± 0.02	1.26 ± 0.03
4–9	-33.71	21.02	2.57	0.109	2.3×10^{15}	1.79 ± 0.04	1.81 ± 0.04
5–10	58.61	23.61	6.16	0.013	2.8×10^{25}	1.63 ± 0.04	1.60 ± 0.04
5–9	-29.64	40.33	0.54	0.462	1.3×10^{13}	0.86 ± 0.02	0.85 ± 0.02
5–8	-20.89	44.86	0.22	0.642	8.5×10^{10}	0.88 ± 0.02	0.89 ± 0.03
6–9	-27.65	40.46	0.47	0.494	9.8×10^{13}	1.21 ± 0.03	1.21 ± 0.03
6–8	232.24	82.08	8.01	0.005	7.2×10^{100}	0.47 ± 0.01	0.46 ± 0.01
Constant	79.86	40.02	3.98	0.046	4.0×10^{-94}		

Note: B is the regression coefficient; SE is the standard error; exp(B) is the odds ratio. There is one degree of freedom for the Wald test for each variable. The mean and standard deviation for each variable (each length divided by the geometric mean for all lengths) for cold- and warm-incubated salmon are also shown. Omnibus test: $\chi^2_{[10]} = 61.175$, $P < 0.001$. Hosmer-Lemeshow test: $\chi^2_{[8]} = 1.700$, $P = 0.99$.

Fig. 6. The mean fineness ratio (standard length (SL)/body depth (BD)) for cold- and warm-incubated Atlantic salmon (*Salmo salar*). Error bars represent one standard error. $N = 30$ for cold-incubated fish and $N = 32$ for warm-incubated fish.



flowing streams (Bowen and Marchetti 2016). In another study, young-of-the-year brown trout and salmon raised at two water velocities differed in body shape, with brown trout becoming more streamlined and salmon less so at the higher stream velocity (Pakkasmaa and Piironen 2000). Pakkasmaa and Piironen (2000) also found that the size of the dorsal and caudal fins differed with flow conditions. Rajput (2013) reported fin and body shape differences related to different habitat variables for the snow trout (*Schizothorax richardsonii* (Gray, 1832)) in tributaries to the upper Ganges River, India. For instance, snow trout in the coldest tributary had the largest relative pectoral and dorsal fin area, whereas those from the warm tributaries had small dorsal and pectoral fins paired with a more elongated body shape. Here, we show that a thermal difference of only 3 °C during the embryo to the alevin stage is enough to yield such a difference in body shape and pectoral fin size in young Atlantic salmon. Both incubation temperatures are encountered by Atlantic salmon embryos and alevins in natural rivers.

Body shape and fin size influence the swimming performance of fish. A more streamlined body improves swimming performance (Webb 1984; Swain 1992; Ackerly and Ward 2016), and there are indications that link swimming performance to small differences in the number of vertebrae (Swain 1992; Aguirre et al. 2019) and to incubation temperature (Lim et al. 2020). The higher fineness ratio for the warm-incubated salmon (4.2) than for the cold-incubated salmon (4.0) indicates that they had a more streamlined body shape. In fact, a fineness ratio of 4.5 is commonly described as the optimal value for endurance swimming (Walker et al. 2013). In terms of fins, caudal fin size is important for propulsion, whereas the other fins largely influence balance, stability, maneuverability, and water flow pattern around the caudal fin (Helfman et al. 2009; Walker 2010). The ecological consequences of the smaller pectoral fin size in the warm-incubated salmon than in the cold-incubated salmon in the present study are unclear, but the observed difference may have repercussions for their lifestyle (Rouleau et al. 2010).

A more streamlined fish in warmer water means reduced power required for swimming (Priede 1985). This in turn will reduce cruising and foraging costs, even if these may not be high for a sit-and-wait predator feeding on drifting food items (Priede 1985). Recently completed experiments indicate that aerobic scope at 13 °C is lower for juvenile brown trout when produced from eggs incubated at a mean incubation temperature of 7 °C than at 4 °C (Durtsche et al. 2021). Aerobic scope may affect growth rates, which in turn affect body shape. Previous studies have found that juvenile Atlantic salmon produced from warm-incubated eggs

grow faster and become larger than those from cold-incubated eggs (Finstad and Jonsson 2012; Jonsson et al. 2014; Burgerhout et al. 2017). Thus, the effects on body shape observed in this study may be an indirect consequence of a modified growth rate rather than a direct effect of temperature during early development on body shape allometry (Devlin et al. 2012). Specifically, there may be a general tendency for fast growth to be associated with a more streamlined body in salmon as previously found for the Bahamian mosquitofish (genus *Gambusia* Poey, 1854) (Araújo et al. 2017), although this pattern may depend on the developmental temperature that the fish experience (Ramler et al. 2014). The temperature of 13 °C used for the study of aerobic scope for brown trout (Durtsche et al. 2021) is around the optimal temperature for growth for brown trout fed on invertebrates or pellets (Elliott and Hurley 2000; Forseth et al. 2009). Possibly, Atlantic salmon produced from eggs incubated at a high temperature may also have a reduced aerobic scope. If so, then a more streamlined body shape may allow them to forage at a lower energetic cost.

Energy saving may not be the only advantage of the more streamlined body shape of the warm-incubated group as argued by Priede (1985). Equally important may be an improved opportunity for predator avoidance. For example, Arnett and Kinnison (2017) found that western mosquitofish (*Gambusia affinis* (Baird and Girard, 1853)) and eastern mosquitofish (*Gambusia holbrooki* Girard, 1859) responded phenotypically to early exposure to predator cues from largemouth bass (*Micropterus salmoides* (Lacepède, 1802)) and bluegill sunfish (*Lepomis macrochirus* Rafinesque, 1819) by developing more streamlined and hydrodynamic body shapes. Thus, the mosquitofish experienced less drag and could move more efficiently and quickly through water when pursued by a predator (Fu et al. 2013). On the other hand, others have pointed out that there may be a trade-off between different types of swimming (Webb 1984; Langerhans 2009). Cruising or steady swimming describes constant-speed swimming and is involved, for example, in searching for food, whereas unsteady swimming, such as “C-start” bursts, describes more complicated swimming involving changes in velocity and direction and is typically associated with predator-prey and social interactions (Langerhans 2009). Thus, the broad caudal area for the cold-incubated salmon over the warm-incubated ones, may reflect differences in the amount of caudal muscle mass. If so, then warm-incubated fish may be less proficient at burst swimming, and thus their ability to avoid predators may be compromised relative to cold-incubated fish. It is unclear what predation pressure will look like in a future climate, nor is it clear if the altered body shape observed here reflects differences in caudal muscular development, and if this has effects on the ability of salmon to escape predators.

The mechanism allowing plasticity in shape in response to temperature during early development is unknown. As stated above, it may be an indirect consequence of a modified growth rate (Devlin et al. 2012). Whether it be a consequence of altered growth or some other reason, epigenetic regulation may be involved (Scott and Johnston 2012). Simonet et al. (2013) showed that there is an inverse relationship between epigenetic DNA methylation and embryonic temperature, affecting the capacity to silence gene transcription. Furthermore, there is evidence indicating that that phenotypically plastic adaptations can be mediated by epigenetic mechanisms (Schlichting and Wundt 2014; Vogt 2017). For instance, Carballo et al. (2018) showed strong indications of epigenetic influence on growth of juveniles initiated by embryonic temperatures for Senegalese sole (*Solea senegalensis* Kaup, 1858) (Carballo et al. 2018).

In summary, temperature during early development resulted in morphological differences in body shape and fin size, which might affect swimming performance, with consequences for later survival. With continued global increases in atmospheric CO₂, temperature is expected to increase during this century,

and thus warmer winters may have far-ranging consequences for ectotherms that incubate eggs during winter. This may include effects on swimming performance, predator avoidance, and energy budgets for ectotherms during their entire lives.

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