

Differential effects of origin and salinity rearing conditions on growth of glass eels of the American eel *Anguilla rostrata*: implications for stocking programmes

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In this study, growth patterns were monitored in controlled fresh and brackish water (BW) conditions for 7 months during *Anguilla rostrata* glass eel and elver stages. Null hypotheses tested were that there is no significant difference in growth between glass eels (1) collected from two geographic regions typified by different sex ratios, (2) reared in fresh and BW and (3) due to origin \times salinity interactions. It was found that young *A. rostrata* from Mira River (MR, Cape Breton, Nova Scotia, Canada, an area where both males and females occur) grew faster than those from Grande-Rivière-Blanche (Québec, Canada, an area where population are highly skewed towards females; 99–100%). *Anguilla rostrata* from both origins also grew faster in BW, although there was a trend for origin \times salinity interactions whereby this effect was more pronounced for fish from the MR. The results support the hypothesis that salinity can influence growth patterns, as possibly can quantitative genetic differences between *A. rostrata* glass eels from different origins. Possible explanations for these patterns and potential consequences for sex determination and translocation programmes are discussed.

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Key words: *Anguilla*; genetic variation; migration; panmixia; population; sex ratio.

INTRODUCTION

Recruitment of the American eel *Anguilla rostrata* (Lesueur), European eel *Anguilla anguilla* (L.) and Japanese eel *Anguilla japonica* Temminck & Schlegel has declined in recent decades (Casselmann, 2003; Dekker, 2003; Tatsukawa, 2003). The St Lawrence River used to support the most important fishery for *A. rostrata* in Canada, but these have declined from 400 t in 1990 to <72 t in 2007 (ICES, 2008). The numbers of *A. rostrata* yellow eels migrating up the St Lawrence River to Lake Ontario have also suffered a major decline with recruitment in 1999 estimated to be 1000 times smaller than observed 15 years earlier (Castonguay *et al.*,

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1994a; Verreault & Dumont, 2003). Hypothetical causes of anguillid eel decline are numerous and include barriers to migration and hydro-turbine mortality, habitat loss and alteration, fishing, parasitism, pollution, as well as environmental change in the Sargasso Sea and Atlantic Ocean circulation (Castonguay *et al.*, 1994b; Haro *et al.*, 2000; Richkus & Whalen, 2000; van Ginneken *et al.*, 2005; Friedland *et al.*, 2007; Bonhommeau *et al.*, 2008).

Stock translocation is increasingly being considered as a means to increase abundance where declines are particularly pronounced. In *A. rostrata*, elvers from the Maritime Provinces of Canada (New Brunswick and Nova Scotia) have been translocated on a large scale since 2004 to increase abundance in the upper St Lawrence River basin (Richelieu River, Lake Champlain, upper St Lawrence River and Lake Ontario) (ICES 2008; B. Williams & R.W. Threader, unpubl. data). Many questions remain, however, about the genetic consequences of such translocations, as well as their effects on growth, survival rates and sex ratio (Parsons *et al.*, 1977; Knights & White, 1997; Rosell *et al.*, 2005; Chu *et al.*, 2006), or on the ability to migrate back to spawning grounds of the Sargasso Sea (Limburg *et al.*, 2003; Westin, 2003; Shiao *et al.*, 2006; Lin *et al.*, 2007; Okamura *et al.*, 2008). Of particular concern are the pronounced regional differences in sex ratio between areas requiring stocking compared to those where recruitment is high enough to produce sufficient stocking material. For example *A. rostrata* populations in the St Lawrence River basin are female-dominated (99–100%, Castonguay *et al.*, 1994a; Verreault *et al.*, 2009), whereas males are more common in the Maritime Provinces of Canada where *A. rostrata* glass eels or elvers are collected for stocking (Vladykov & Liew, 1982; Jessop *et al.*, 2004). If there is a genetic basis for recruits in the latter areas to tend to mature as males, possibly associated with tendencies to faster growth, this could upset the natural bias towards females in the receiving waters. This might help explain the results of Verreault *et al.* (2009) who translocated 40 000 glass eels in 1999 from Bay of Fundy tributaries to a 400 ha freshwater (FW) lake on the south shore of the St Lawrence River estuary. Males appeared to represent 30% of sampled individuals 4 years after stocking whereas males are absent in neighbouring naturally recruited populations. For *A. anguilla*, stocking elvers from high-recruiting North Atlantic Ocean coastal areas (where males are common) into Baltic and Lithuanian lakes (where females tend to predominate) has also resulted in an early male predominance, but this was followed later by higher abundances of females (Holmgren *et al.*, 1997; Lin *et al.*, 2007). So there was little evidence that this has caused any significant clear switch from female to male-biased ratios. Similar phenomenon was observed in Japanese rivers close to *A. japonica* rearing facilities (Chu *et al.*, 2006). In relation to the above findings, early production of males could be linked with fast grower individuals (Edeline & Elie, 2004) and may be genetically determined. Finally, stocking density can potentially affect sex ratios as it has been observed in Lough Neagh (Rosell *et al.*, 2005).

The lack of evidence refuting panmixia in *A. rostrata* (Koehn & Williams, 1978; Avise *et al.*, 1986; Wirth & Bernatchez 2003) has led to accepting the absence of any genetic basis to phenotypic variation observed, including sex ratio. Hence it is commonly accepted that phenotypic variation in *A. rostrata* can be best explained by plastic responses to environmental cues or conditions (Helfman *et al.*, 1987; Krueger & Oliveira, 1999; Oliveira & McCleave, 2002). Consequently, it is implicitly believed that translocated glass eels or elvers will express the

phenotype of anguillid eels that have grown naturally in the area being stocked. Indeed, a number of experimental studies have documented environmentally induced phenotypic responses for several life-history traits in anguillid eels, including growth, and sex ratio (Wiberg, 1983; Colombo *et al.*, 1984; Holmgren *et al.*, 1997). In a widespread and generalist species such as *A. rostrata*, phenotypic plasticity could be adaptive (Via & Lande, 1985, 1987; Schlichting & Smith, 2002; Daverat *et al.* 2006; Crispo, 2008) and influence the probability of persistence of a depleted population by enhancing the ability of individual fish to respond to natural and anthropogenic environmental change (Hutchings *et al.*, 2007). Evidence for plastic responses to environmental conditions, however, does not necessarily preclude some genetic basis for phenotypic variation, or to possible pre-adaptation to a given habitat or selection against FW migratory behaviour (Edeline, 2007).

Regardless of the relative role of environmental *v.* genetic basis of phenotypic variation, there is still little consensus about the proximate factors, as well as developmental timing of sex differentiation in anguillid eels (Beullens *et al.*, 1997; Holmgren *et al.*, 1997; Davey & Jellyman, 2005). It has been proposed that different habitat preferences and environmental characteristics may influence sex distribution and differentiation (Helfman *et al.*, 1987; Oliveira *et al.*, 2001; Tesch, 2003). Density is also generally believed to play an important role in determining sex whereby high density has generally been reported to produce males (Degani & Kushnirov, 1992; Roncarati *et al.*, 1997; Krueger & Oliveira, 1999), although Huertas & Cerdà (2006) showed the opposite. While the exact mechanisms remain to be elucidated, there seems to be growing evidence that sex differentiation (Kuhlmann, 1975; Holmgren *et al.*, 1997; Davey & Jellyman, 2005) as well as habitat preference and survival (Edeline & Elie, 2004, Edeline *et al.*, 2005) are at least partially linked directly or indirectly to growth.

This study tested the null hypothesis that there is no significant difference in growth between *A. rostrata* glass eels (1) collected from two different areas naturally differing in sex ratio, (2) reared in FW and brackish water (BW) and (3) due to origin \times salinity interactions.

MATERIALS AND METHODS

GLASS EELS CAPTURE AND REARING

Anguilla rostrata glass eels were obtained from two sampling locations in north-east Canada in 2007 at river mouths just prior to entering FW, therefore avoiding differential density effect or time spent in FW before experiments. The first sample was collected in Mira River (MR), Cape Breton, on the Atlantic side of Nova Scotia (45°56' N; 60°07' W). Fish from an early and late arrival waves were sampled and shipped at two times (14 June and 29 July; $n = 1000$ in each shipment) in sealed plastic bags inflated with oxygen and containing a small amount of FW collected from the point of capture. Although the sex ratio for MR has not been documented, males are generally observed and sometimes outnumber females in other tributaries along the Atlantic coast of Nova Scotia and New Brunswick (Vladykov & Liew, 1982; Jessop *et al.*, 2004; www.cosewic.gc.ca). The second batch of fish was collected *c.* 900 km north-west of the Nova Scotia sampling site at the mouth of the Grande-Rivière-Blanche [48°78' N; 67°69' W, lower St Lawrence Estuary, Québec, Canada (RB)] where essentially all *A. rostrata* are females (99–100%, Castonguay *et al.*, 1994a; G. Verreault, unpubl. data). *Anguilla rostrata* glass eels from three early and late arrival waves were sampled (15 June, $n = 250$; 1–3 July, $n = 325$; 14–16 July, $n = 280$) using dip nets (mosquito net,

ED Cummings inc. Cat number: L-36-4-MOSQ/14; www.cummingsnets.com). Sampling was carried out at new or full moon periods and occurred at night during flood tides. Experiments were conducted in two independent series of tanks provided with continuous recirculated filtered water without supplemental aeration. Physico-chemical variables (oxygen, nitrites, nitrates, ammonia and pH) were monitored daily and filter back-washed when required. Preventive malachite green treatments were conducted weekly on all tanks simultaneously. Polyvinyl chloride (PVC) tubes were placed on the bottom of tanks to provide shelters to reduce agonistic behaviours. During an acclimation period of 12 days, water temperature was gradually increased from 10 to 22°C and photoperiod set at 12L:12D. Growth of fish from the two geographical origins was compared in two contrasting salinity treatments, FW (salinity 3 range ± 1 , FW) and brackish water (22 range ± 1 , BW). For each treatment there were four replicate tanks for RB and seven for MR (more fish being available for MR), giving a total of 22 tanks. Fish were successively randomly distributed in groups of five into individual tanks, up to a total of 100 fish per tank. The mean initial starting density per tank in biomass terms was $c. 45 \text{ g m}^{-2}$, a density < 10 times the 'low density' criterion in Huertas & Cerdà (2006) and even lower than density used by Knights (1987). *Anguilla rostrata* were fed *ad libitum* twice a day for the first month with frozen ground cod *Gadus morhua* L. roe cubes. From the second month of experiment, commercial trout pellets (0.5 mm starter fish feed, Corey Feed Mills Ltd; www.coreyaqua.com) were mixed with *G. morhua* roe (1:3 proportion) to maximize consumption and nutritional value (Yahyaoui, 1988; Penaz & Vostradovski, 1990; De Silva *et al.*, 2008). Uneaten food and faeces were removed daily.

MEASUREMENTS AND STATISTICAL ANALYSIS

A random sample of fish from each tank was collected and measured at three time periods during the 7 months experiment (Table I). The random procedure was as follows: shelter tubes were first removed from a tank, then all fish were removed by net scoops of $c. 10$ fish, and one per scoop was retained at random until a total of 10 fish were caught. Time 1 (T1) corresponds to the start of the experiment and T2 and T3 to sampling times after 3 months and 7 months respectively. The average number of individuals collected from each tank at each time point was limited to 10 individuals to ensure sufficient fish was left in tanks for long-term rearing and a future sex differentiation study. Individuals were sedated with eugenol and total body length (L_T ; ± 1 mm) and wet mass (M_W ; ± 0.1 mg) measured before freezing in liquid nitrogen for future genetic analyses. Because L_T and M_W measurements were highly correlated from T1 to T3 ($y = 9.395x^{0.3016}$, $r^2 > 0.95$), further statistical analyses were performed and presented for M_W only. All statistics were performed using the MIXED procedure in SAS (SAS Institute Inc.; www.sas.com). Although the Shapiro–Wilk test rejected normality despite various transformation methods attempted, \log_{10} transformation was retained as it best approached normality and homogeneity assumptions. Skewness and kurtosis coefficients were close to normal distribution and because the ANOVA is robust to this situation (Maxwell & Delaney, 2004), it was assumed that departures from normality did not cause significant bias to the results and interpretations. As the number of individuals varied among treatments, the protected Fisher least-square difference (LSD) method was calculated to achieve multiple comparisons. A factorial (nested) ANOVA was used to test the effects of origin ($n = 2$), treatment ($n = 2$) and time ($n = 3$) on elver M_W . These factors were considered fixed, whereas the tank effect ($n = 7$ for MR and $n = 4$ for RB in each treatment) was considered random in the model. In order to take into account the lack of independence of the data, the 'repeated statement' approach was used in the mixed procedure. The analysis was refined with multiple comparisons using the slice-effect test (also known as the simple main effects test), a post-ANOVA procedure in SAS. In all tests, statistical significance was accepted for $P < 0.05$.

Differences in surface density as biomass m^{-2} or number of individuals due to variable mortality among tanks could interact with calculated mean M_W independent of origin or treatment. Therefore Pearson's correlation was estimated (R Development Core Team; www.R-project.org) between these two variables and mean specific growth rates (G) [$G = 100 (\ln M_2 - \ln M_1) (t_2 - t_1)^{-1}$, where M_1 is mass at time t_1 and M_2 is mass at

TABLE I. Mean \pm S.D. wet mass (M_W) and total length (L_T) at the start of the growth experiment (T1), after 3 months rearing and full pigmentation (T2), and after 7 months of rearing (T3) for *Anguilla rostrata* elvers from different origins (Mira River and Grande-Rivière-Blanche) reared in fresh water (FW, salinity 3) and brackish water (BW, salinity 22)

Time	Mira River			Grande-Rivière-Blanche			
	<i>n</i>	M_W (mg)	L_T (mm)	<i>N</i>	M_W (mg)	L_T (mm)	
FW	T1	90	140 \pm 42	60.3 \pm 2.8	40	150 \pm 41	61.5 \pm 2.9
	T2	50	230 \pm 146	62.9 \pm 6.2	35	290 \pm 86	66.3 \pm 4.7
	T3	75	1680 \pm 1510	102.0 \pm 24.6	45	1270 \pm 691	97.4 \pm 14.0
BW	T1	90	140 \pm 45	59.8 \pm 3.1	40	160 \pm 46	61.9 \pm 2.8
	T2	50	430 \pm 326	69.7 \pm 10.1	35	400 \pm 196	70.6 \pm 7.4
	T3	75	2440 \pm 2210	114.0 \pm 26.2	45	1410 \pm 867	99.1 \pm 16.5

n, the total number of individuals measured at a given time.

time t_2 , Appelbaum *et al.*, 1998] observed in all tanks between T1 and T2 and between T2 and T3.

RESULTS

ORIGIN EFFECTS ON GROWTH

At time T1, there was no significant difference in M_W between fish from both origins but RB fish were significantly longer (Table I, M_W : *t*-test, $P > 0.05$; L_T : *t*-test, $P = 0.001$). Similarly, once randomly distributed among tanks, there was no significant overall M_W difference observed either between origins (Table II, slice effects test: origin \times time, T1: *F*, $P > 0.05$) or salinity treatments (Table II, treatment \times time, T1: *F*, $P > 0.05$).

There was no overall significant main effect of origin on M_W observed between MR and RB (Table III). A significant origin \times time interaction, however, was observed, meaning that there were significant differences in M_W between fish of both origins depending on time period (Table III). Namely, simple main effects test revealed that after 7 months (T3) *A. rostrata* from MR reached a significantly larger M_W than those from RB, either in the FW or BW treatment (Table II and Fig. 1).

TREATMENT EFFECTS ON GROWTH

An overall significant main effect of salinity treatment was observed (Table III) whereby *A. rostrata* reared in brackish water were generally larger for a given origin at a given time (Table II and Fig. 1). There was also a significant salinity \times time interaction, indicating that the extent of the salinity effect varied with the period fish were measured. Indeed, although mean M_W did not differ between treatments at T1, elvers from both origins were significantly larger in BW than FW at both T2 and T3 (Table II and Fig. 1). Although the origin \times salinity interaction was not significant, the mean M_W gain over the whole experiment between *A. rostrata* reared in FW and

TABLE II. Results of the slice effect tests for the factorial (nested) ANOVA of factors explaining wet mass (M_W) variation, including main effects and interactions (see Table I)

Effect	Origin	Treatment	Time	d.f.	<i>F</i>	<i>P</i>
Origin × time	Mira River			2	638.80	<0.001
Origin × time	Grande-Rivière-Blanche			2	284.78	<0.001
Origin × time			T1	1	0.70	>0.05
Origin × time			T2	1	2.68	>0.05
Origin × time			T3	1	6.13	<0.05
Treatment × time		FW		2	376.58	<0.001
Treatment × time		BW		2	462.50	<0.001
Treatment × time			T1	1	0.03	>0.05
Treatment × time			T2	1	11.49	<0.01
Treatment × time			T3	1	4.30	<0.05

For each slice effect result, *F* and *P* values refer to the significance of a specific factor. For instance, the first two results reported indicate that both the Mira River and Grande-Rivière-Blanche significantly varied with time. Conversely, the next three lines of results indicate that there is a significant difference between origins at T3 only.

TABLE III. Summary of the factorial (nested) ANOVA of factors explaining wet mass variation, including main effects and interactions

Source of variation	d.f.	<i>F</i>	<i>P</i>
Origin	1	0.01	>0.05
Treatment	1	7.43	<0.05
Origin × treatment	1	0.98	>0.05
Time	2	832.68	<0.001
Origin × time	2	6.92	<0.01
Treatment × time	2	5.00	<0.05
Origin × treatment × time	2	2.10	>0.05

BW was greater in MR fish (1540–2300 mg respectively, *i.e.* an increase of 49%) compared to RB fish (1120–1260 mg, 13%). This suggests there might possibly be a gene × salinity interaction difference between glass eels from the two origins (Fig. 2).

CORRELATION BETWEEN SURVIVAL, *ANGUILLA ROSTRATA* NUMBERS, BIOMASS DENSITY AND GROWTH

Variable and inconsistent effects of either surface density (g m^{-2}) or remaining numbers of *A. rostrata* due to mortality loss on growth (*G*) were observed in the two time periods. During T1–T2 and T2–T3, the relationship between the survival and *G* was positive but not significant (T1–T2, $r^2 = 0.090$, $P > 0.05$; T2–T3, $r^2 = 0.171$, $P > 0.05$). During T1–T2, the relationship between the number of surviving fish and *G* was positive but not significant ($r^2 = 0.031$, $P > 0.05$) whereas a significantly negative relationship was observed during T2–T3 ($y = -0.343x + 59.606$; $r^2 = 0.236$, $P < 0.05$). For density, the relationship with *G* during T1–T2 was positive and highly significant ($y = 0.395x - 6.343$; $r^2 = 0.671$, $P < 0.001$), a result similar to that found in the growth experiments of Huertas & Cerdà (2006). No significant

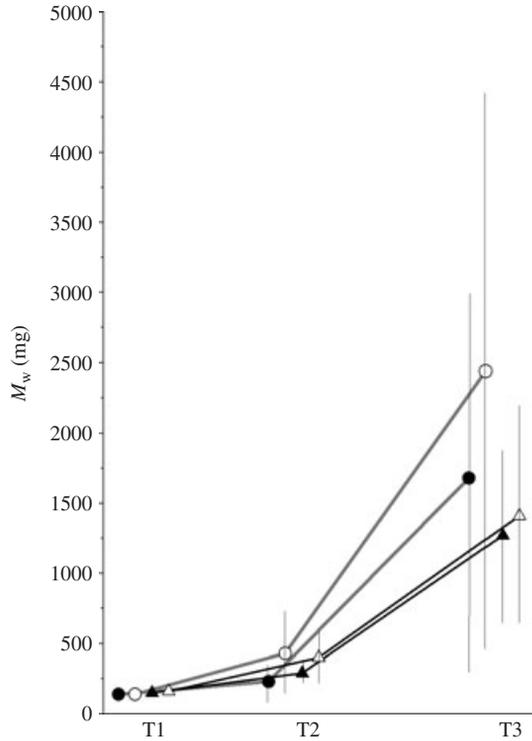


FIG. 1. Growth patterns in wet mass (M_w) observed between *Anguilla rostrata* glass eels and elvers from Mira River, Cape Breton ($n = 4$; ●, ○) and Grande-Rivière-Blanche ($n = 7$; ▲, △) in fresh water (●, ▲) and brackish water (○, △) between the start of the experiment (T1) and after 3 months (T2) and after 7 months (T3). Data points correspond to means \pm s.d. for all treatments combined for a given origin at a given time. Points are slightly offset to make s.d. bars easier to visualize.

relationship, however, was seen during T2–T3 ($r^2 = 0.048$, $P > 0.05$). So there was no significant effect of density between T2 and T3 when most of the growth difference between origins took place. Altogether, the effect of variation in survival, density or fish numbers among aquaria on differences in mean M_w observed between origins and treatments cannot be ruled out since the number of fish, although not surface density, apparently suppressed G during T2–T3. These results, however, suggest that neither of these variables were consistent in explaining the overall growth patterns.

DISCUSSION

A major objective of this study was to compare growth under experimental conditions in *A. rostrata* collected from two geographic areas where fish differ in sex ratio. More specifically, the null hypothesis was that there is no significant difference in growth between *A. rostrata* glass eels (1) collected from two locations typified

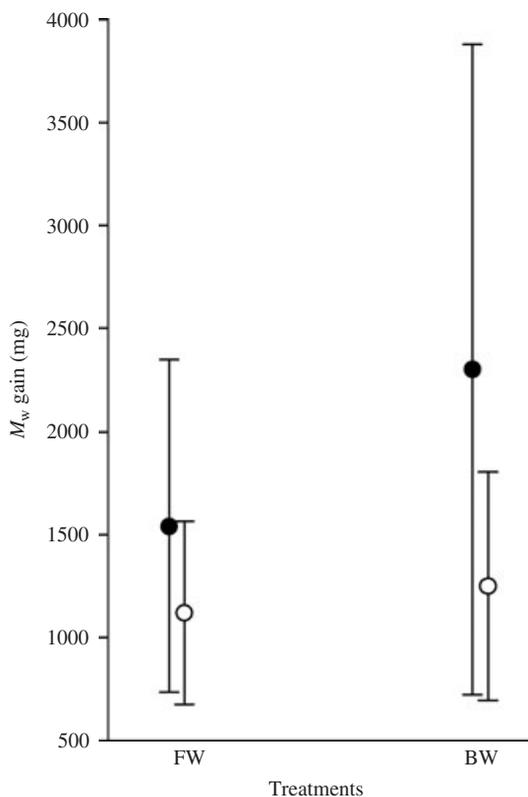


FIG. 2. Mean \pm s.d. fresh mass (M_w) gain over the whole 7 month experiment between *Anguilla rostrata* elvers from Mira River, Cape Breton (●) and Grande-Rivière-Blanche (○) reared in fresh water (FW) and in brackish water (BW). Data correspond to the (M_w) gain of all elvers for a given origin and treatment measured at the end of the experiment. Points are slightly offset to make s.d. bars easier to visualize.

by different sex ratios, (2) reared in FW and BW and (3) due to origin \times salinity interactions.

The first two null hypotheses were rejected as fish originating from MR (Nova Scotia) grew significantly faster than those from Grande-Rivière-Blanche (Québec) over the 7 month experimental period (Table II). Also a significant plastic response was observed as *A. rostrata* from both origins grew significantly faster in brackish than FW. Although the third null hypothesis was not rejected, there was a trend for origin \times salinity interactions whereby mean differences in growth between FW and BW was qualitatively more pronounced for *A. rostrata* from MR (49% higher in BW) than those from Grande-Rivière-Blanche (13% higher in BW). Hence these results support the hypothesis that both origin of *A. rostrata* glass eels and salinity influenced growth in this study. Although these results support the hypothesis of a genetic basis for growth differences between fish from the two geographic areas, environmental and experimental factors also need to be considered. Firstly, different growth propensities might be affected by migratory histories and sampling. For example, RB *A. rostrata* glass eels have to migrate *c.* 850 km further than MR fish

from the spawning area in the Sargasso Sea. Anguillid glass eels enter river mouths in temporal waves which may be genetically differentiated (Maes *et al.*, 2006). Thus a difference in sampling time associated with different glass eel waves could explain the distinct growth patterns observed between origins. *Anguilla rostrata* from MR and RB, however, were sampled over approximately the same period and were at the same developmental stages (Strubberg's pigmentation stages V to VI A2; Tesch, 2003) and experiments were conducted on pooled individuals representing different waves. In experiments, differences in density (either in terms of biomass, g m^{-2} or number of individuals m^{-2}) may have occurred between T1 and T2 and between T2 and T3 because of variable mortality among tanks, *e.g.* due to poor feeding, stress and cannibalism. To minimize such influences, *A. rostrata* were fed *ad libitum* and shelters were placed in all tanks in order to reduce agonistic behaviour (Knights, 1987). Starting densities were relatively low compared to those used in previous studies (Knights, 1987; Huertas & Cerdà, 2006). Another factor that could have affected growth among tanks is the number of fish of different sizes or the particularly slow or fast growth rates in one or few individuals (Knights, 1987). The high s.d. in M_W (Table I and Fig. 1) indicate individual growth rates were indeed very variable but only 10 *A. rostrata* were individually measured in each tank at each time period, which is not sufficient to rigorously compare the occurrence of extremes in L_T frequency distribution.

COMPARISONS WITH PREVIOUS STUDIES

No experimental studies have compared anguillid elvers from different origins before this study. Vladykov & Liew (1982) reared *A. rostrata* elvers from two different streams in eastern Canada in the same FW pond but they did not control environmental conditions, and the significance of their results was not investigated statistically. The present results, however, corroborate their general observation that *A. rostrata* elvers collected from two different rivers revealed differential growth pattern in identical rearing conditions. In contrast, more literature is available regarding the effect of salinity on growth in *Anguilla*: laboratory and field studies on different Anguillid species have generally shown better growth in sea water or BW than FW. In *A. rostrata*, Jessop *et al.* (2004, 2008) reported higher growth rates in marine and estuarine fish relative to FW fish in their natural environment, which could be induced by higher habitat productivity (Jessop *et al.*, 2004). In *A. anguilla*, Melia *et al.* (2006) also reported that body growth was faster in brackish than in FW natural environments. Moreover, Edeline *et al.* (2005) clearly showed that *A. anguilla* elvers reared in salt water grew faster than in FW under controlled conditions, irrespective of food availability. As proposed by Edeline & Elie (2004), several physiological factors could explain the differential effect of salinity on growth, including difference in standard metabolic rate which could be higher in FW (Knights, 1985), food intake and conversion, and hormonal stimulation such as the salt water stimulation of the growth hormone (GH)/insulin growth factor-1 (IGF-1) axis (Beckman *et al.*, 1998; Kalujnaia *et al.*, 2007a, b). These authors observed also that preference for FW was linked to higher locomotor activity, which probably results in poorer growth. Edeline *et al.* (2005) also hypothesized that 'genetic factors' could be involved in the link between salinity preference and anguillid glass eel growth. The results of the present study offer some support for this hypothesis, given that the origin \times salinity

interaction tended to differ between *A. rostrata* glass eels from the two study sites reared in the same laboratory conditions in aquarium replicates.

LINK BETWEEN EARLY GROWTH AND SEX DIFFERENTIATION

Although proximate factors as well as developmental timing of sex differentiation remain poorly understood, there is growing evidence that sex is at least partially determined by growth (Beullens *et al.*, 1997; Holmgren *et al.*, 1997; Davey & Jellyman, 2005). Based on life history theory, Helfman *et al.* (1987) predicted that slow-growing anguillid eels should develop as females since they attain a larger size than males during their lifetime which may compensate for fitness reduction associated with slower growth in early age. The few studies that have evaluated the influence of early growth rate on sex differentiation tend to support Helfman *et al.*'s (1987) prediction (Holmgren, 1996; Melia *et al.*, 2006).

POSSIBLE EXPLANATIONS FOR A GENETIC BASIS FOR GROWTH DIFFERENCES

The results of the present study which suggests a possible genetic basis for growth differences corroborate other indirect evidence suggesting quantitative genetic differences among *A. rostrata* from different locations, albeit other factors can also be involved. First, regional differences in recruitment patterns indicate the possibility of at least partial demographic independence among various eel stocks (Castonguay *et al.*, 1994a; Cairns *et al.*, 2007). Also, female *A. rostrata* silver eels from the St Lawrence River are much larger than those of the Gulf of St Lawrence, and form a sharp disjunction in size distribution c. 150 km upstream from the mouth of the St Lawrence River estuary (Cairns *et al.*, 2007). Finally, the difference in growth reported by Vladykov & Liew (1982) and Verreault *et al.*, (2009) also suggested that there might be genetically based regional differences for these traits. What could be possible explanations for genetic differences in growth between sites?

A first explanation could be that such differences are associated with population genetic structure, which would however contradict the current paradigm of panmixia within *A. rostrata* (Avisé *et al.*, 1986; Wirth & Bernatchez, 2003). Arguably, population genetics studies performed to date in *A. rostrata* have all been based on a relatively modest number of sampling sites and genetic markers, and none have included samples from the St Lawrence River basin. Consequently, the null hypothesis of no genetic difference between *A. rostrata* from this area and other parts of the species' range remains to be rigorously tested. Moreover, the absence of statistically significant genetic divergence in neutral markers such as microsatellites does not necessarily preclude the possibility of adaptive population genetic differentiation. Indeed, genetic divergence can develop much more rapidly in adaptive than in selectively neutral traits or loci, despite levels of gene flow that would prevent or eliminate differentiation at selectively neutral loci (Conover *et al.*, 2006).

A significant genetic difference in growth between geographic areas does not necessarily imply the occurrence of population structure. An alternative hypothesis could be that this reflects either non-random dispersal or habitat selection, or

differential mortality associated with individual genetic variation within a single panmictic population. For example, Edeline *et al.* (2005) observed individual variation for salinity preference among *A. anguilla* glass eels from a single location and hypothesized that the link between individual behavioural variation in salinity preference and growth rate is related to some genetic factors. One such genetic factor could be individual heterozygosity, which has been shown to be positively associated to growth and survival in *A. anguilla* (Pujolar *et al.*, 2005). Edeline *et al.* (2007) further proposed that genetic differences among individuals could explain alternative dispersal tactics, whereby fast growing *A. anguilla* would tend to remain in lower reaches and brackish and salt water, while those adopting a slow-growing strategy would be more likely to migrate further inland, and may have a better survival probability (Edeline & Elie, 2004). In *A. rostrata*, Wang & Tzeng (1998) documented variation in time at metamorphosis and growth among elvers collected along the North American Atlantic Ocean coast. They observed a non-random distribution of individuals with faster-growing and earlier metamorphosing leptocephali migrating to the mid point of the range whereas those migrating further north were characterized by slower growth and delayed metamorphosis. Similarly, *A. rostrata* glass eels from the north grew more slowly than those to the south.

These observations, along with the results of the present study that differential growth between anguillid elvers from different sampling sites is apparently maintained when reared in similar conditions, lead to the hypothesis that regional variation in growth and time at metamorphosis could result from either non-random dispersal (Edeline *et al.*, 2007) or differential survival associated with variation in individual genetic characteristics. Both processes could result in regional genetic variation and growth among individuals from a same cohort within an otherwise panmictic population. Such a phenomenon has previously been documented in marine invertebrates that have a random larval dispersal followed by differential mortality in specific habitats where they settle (Veliz *et al.*, 2004, 2006).

RELEVANCE FOR ANGUILLID EEL CONSERVATION AND MANAGEMENT

Results from this study raise issues regarding current stocking practices in the upper St Lawrence River basin and elsewhere. If there are indeed genetically based differences in sex ratio and growth between *A. rostrata* of distinct origins, this could imply that stocking of areas where only females are found (such as the upper St Lawrence River system) with glass eels from areas with variable proportions of males may reduce the proportion of females produced in waters being stocked. Such consequences would call for a re-examination of stocking practices. Also, if slower growing anguillid eels tend to migrate farther upstream in order to avoid competition with faster-growing individuals as proposed by Edeline *et al.* (2007), such stocking practice could potentially be detrimental for the remnant local anguillid eel stock. Furthermore if there is an association between individual growth characteristics and habitat preference (*e.g.* downstream *v.* upstream) as proposed by Edeline *et al.* (2007) anguillid eels from downstream locations stocked at upper locations may have a reduced fitness and hence may not contribute significantly to spawning escapement. More studies, including genetic analyses, are required to clarify these issues.

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