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Effects of salinity on swimming performance and oxygen consumption rate of shiner perch *Cymatogaster aggregata*

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ABSTRACT

Environmental factors add constraints to organismal performance at their extremes, but support optimal performance at energetically beneficial conditions. In aquatic environments, salinity adds costs for ion transportation to the energetic budgets of osmoregulating animals, such as teleost fishes. These additional costs may limit the available energy for important ecological traits of fishes, including maximal and optimal swimming performance, which are required for successful foraging and migration in the wild. Here, we hypothesize that swimming performance, and its related costs, will be optimized at near-isoosmotic salinity, and decline under more saline conditions. Using the euryhaline shiner perch (*Cymatogaster aggregata*) as a model for coastal fishes, we determined critical swimming speeds and oxygen consumption rates during swimming at salinities of 12 g kg^{-1} (near-isoosmotic, brackish, S12) and 31 g kg^{-1} (hyperosmotic, marine, S31). Most tested metrics were unaffected by salinity, including aerobic scope, active metabolic rate and optimal swimming speed. Likewise, critical swimming speed (in body lengths per second, BL s^{-1}) was not significantly different between fishes acclimated to S12 (4.8 ± 0.6 BL s^{-1}) or S31 (5.1 ± 0.5 BL s^{-1}, means ± SD, $n = 5$) suggesting that the fish could swim and hunt for prey equally well regardless of salinity. However, S31 conditions did cause comparatively higher oxygen consumption rates at swimming speeds from 0.5 to 1.5 BL s^{-1}, and a 20% increase in the extrapolated standard metabolic rate (i.e. cost of maintaining bodily functions). Our results confirm that there is an added energetic cost of salinity, but highlight that the cost of osmoregulation appears minimal relative to the energetic demands of swimming, and consequently has no effect on the maximal swimming performance of adult shiner perch. Given the strong salinity gradients naturally encountered in many coastal ecosystems, these data provide an explanation for the capacity of a coastal roaming species to move in an out of coastal habitat zones without significantly compromising their ability to hunt prey, avoid predation and migrate. As climate change locally affects environmental salinity, our results offer valuable insight towards the effects of environmental perturbations on fishes in coastal marine and estuarine habitats.

1. Introduction

Marine organisms can be classified either as osmo-conformers that maintain an internal status similar to the ambient environmental salinity, or as osmo-regulators that change their inner milieu to obtain homeostasis (Meunier et al., 2014). Most marine teleost fishes are osmo-regulators and, while living at oceanic salinities of around 35 g kg^{-1}, typically regulate their extracellular fluids to 9–12 g kg^{-1} (Boeuf and Payan, 2001; Edwards and Marshall, 2012; Zydlewski and Wilkie, 2013) not to compromise physiological processes such as enzyme functionality (Segal and Beem, 2001). Maintaining this osmotic gradient between the body and the ambient environment causes a continuous loss of water over the gill epithelium and other permeable external surfaces in marine teleosts (Evans et al., 2005; Edwards and Marshall, 2012). This is compensated for by gastrointestinal uptake of ambient seawater and subsequent secretion of excess ions through specialized branchial cells and the renal system in processes summarized as osmoregulation (Marshall and Grosell, 2006; Evans, 2008).

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Another mean by which fishes can modulate salinity stress is by decreasing gill permeability, all the while decreasing gas exchange potential, a phenomenon termed the osmo-respiratory compromise (see review by Sardella and Brauner, 2007).

Osmoregulation is an energy demanding process and its lowest expenditures are intuitively expected when the osmotic gradient across the fish gills is minimal, i.e. near isoosmotic conditions (9–12 g kg^{-1}, Boeuf and Payan, 2001; Ern et al., 2014). While the mechanistic understanding of osmoregulatory processes at cellular and tissue levels of fishes has considerably increased in the past (see references in Hwang and Lin, 2013), there is still no consensus on the whole-organism magnitude of the energetic expenditure allocated to osmoregulation (Ern et al., 2014). Estimates of the added costs range from around 10 up to 50% (see references in Boeuf and Payan, 2001) of the standard metabolic rate (SMR), a metric that describes the costs for fundamental, life-sustaining processes in resting animals, such as osmoregulation, anabolic processes and basic cardiorespiratory functions (Ern et al., 2014). Salinity can also alter the active metabolic rate (AMR; the peak metabolic rate during activity), possibly due to the osmo-respiratory compromise (Sardella and Brauner, 2007). The ecological importance of AMR and SMR is apparent through the difference between the traits, that is, the metabolic scope, as it defines the swimming ability, and correlates with growth (McKenzie and Claireaux, 2010; Claireaux and Lefrançois, 2007). Whole-animal oxygen consumption rate ($\dot{M}O_2$) has been used as a proxy for metabolic rate since the early 20th century, and helped estimate SMR, AMR, and in turn, aerobic scope (AS), in fishes (Nelson, 2016; Svendsen et al., 2016). Interestingly, previous studies testing effects different salinities on $\dot{M}O_2$ in fishes did not necessarily observe minimal rates near isoosmotic or normal habitat conditions, and there seems to be a lack of a universal trend in the described changes in $\dot{M}O_2$ (Ern et al., 2014). However, only a few studies have so far coupled the effects of osmoregulatory costs with important traits of fishes, such as swimming performance, at the whole-organism level (Rao, 1968; Farmer and Beamish, 1969; Febry and Lutz, 1987).

Swimming performance is required at optimal functioning for successful foraging, predator avoidance and migration (McKenzie and Claireaux, 2010). During swimming, ventilatory flow increases to meet elevated oxygen demands, which in turn create a larger potential for diffusion of water over the gills per Fick's law. This could explain why some studies report the cost of osmoregulation to be activity-related and increase with swimming speed (Rao, 1968; Farmer and Beamish, 1969; Febry and Lutz, 1987). To measure maximum swimming performance in fishes, critical swimming speed (U_{crit}) is often determined by a methodology in which a fish swims against a steady water flow that is increased in step-wise time intervals until fatigue of the tested individual occurs (Brett, 1964; McKenzie and Claireaux, 2010). With regard to salinity, the U_{crit} of several fish species has been shown to decrease at hyperosmotic conditions (Kolok and Sharkey, 1997; Randall and Brauner, 1991), likely caused by the higher cost of swimming and/or the osmo-respiratory compromise in non-isoosmotic conditions.

In the last decades, physiology has been used by conservation biologists to assess and forecast ecological implications of environmental perturbations, such as climate change (Cucco et al., 2012; Teal et al., 2015) fisheries (Hollins et al., 2018), oil spills (Stieglitz et al., 2016), and alien species invasion potential (Marras et al., 2015; Behrens et al., 2017). As environmental salinity is locally expected to change in relation to altering rainfall and river-runoff patterns (Harley et al., 2006; Vuorinen et al., 2015), studies on the effects of salinity on physiological performance of fish are valuable to evaluate dispersal of fish in coastal marine and estuarine environment, now and in the future. Along the Pacific coast of North America, from California to Alaska, the shiner perch (*Cymatogaster aggregata*, Gibbons), is found in shallow bays and calm areas along the coast where it enters estuaries and brackish waters (see references in Triplett and Barrymore, 1960; Kells et al., 2016). This euryhaline life-style makes this teleost species an ideal candidate to test if reduced cost of osmoregulation in brackish water translates to better swimming performance, and could provide insight to the effect of salinity on the species ecology. In this study we aimed to quantify the effects of near-isoosmotic (12 g kg^{-1}, S12) and hyperosmotic salinity (31 g kg^{-1}, S31) on U_{crit}, SMR, and the activity related $\dot{M}O_2$ at various swimming speeds in adult shiner perch. We hypothesize that SMR and the activity related $\dot{M}O_2$ are lower at S12 due to lower costs of osmoregulation, while AMR remaining unchanged, potentially increasing AS and U_{crit}.

2. Materials and methods

2.1. Animal maintenance

All animal care and experimental protocols followed the guidelines of the Institutional Animal Care and Use Committee at the University of Washington (permit number 4238-03).

Shiner perch (*C. aggregata*) were caught by several beach seining events in July and August 2015 at Jackson Beach, San Juan Islands, Washington, USA (N 48°31′11″, W 123°0′45″), and brought to the experimental facility at the Friday Harbor Laboratories, (University of Washington, Washington, USA), were they were kept at a salinity of 31 g kg^{-1} at temperatures between 12 and 13 °C, in 160 L flow-through tanks (sea water was continuously supplied from a depth of 10 m off the coast from San Juan Islands). After maximally 7 days, the experimental animals were acutely transferred to one of two similar 160 L acclimation tanks, with either a salinity of 12 ± 0.1 g kg^{-1} (S12) or 31.1 ± 0.7 g kg^{-1} (S31, mean ± SD), and a constant temperature of 12.5 ± 0.2 and 12.6 ± 0.2 °C (mean ± SD), respectively. This acute transfer resulted in no visible signs of distress or mortality. Temperature, salinity and dissolved oxygen was measured daily with an YSI 85D probe (YSI; www.ysi.com), and the water was changed twice a day (total exchange 75% d^{-1}). The salinity was adjusted with either ambient sea water or dechlorinated tap water and the water temperature in each tank was regulated by a temperature controller connected to a pump that circulated the water through a cooling unit. The tanks were, furthermore, aerated (dissolved oxygen > 80% air saturation), and the fish experienced a light cycle of 15:9 (light:dark). Total ammonia nitrogen was tested regularly with an API NH_3/NH_4^+ test kit (API; www.apifishcare.com) and never exceeded 0.25 mg L^{-1} at pH 7.9 ± 0.4. Both tanks were divided into sub-compartments, separated with netting (mesh size = 0.15 mm^2), allowing a continuous supply of fish acclimated to the respective salinity for identical amounts of time. Each fish was only used once to avoid pseudo-replication, after which they were released into the wild. Fish were acclimated for 6–7 days to either S12 or S31 (two treatments) before experimentation commenced, and were fasted during the acclimation period to avoid effects of digestion (specific dynamic action) on oxygen consumption rate or swimming performance (Chabot et al., 2016). This period was deemed sufficient acclimation time for plasma osmolality and water content to adjust to salinity change in shiner perch (Triplett and Barrymore, 1960; Divino et al., 2016), and has been reported to be enough time for other euryhaline fishes to stabilize their total organic osmolyte content after acute transfer to different salinities, e.g. milkfish (*Chanos chanos*, Lin et al., 2006) or three-spined stickleback (*Gasterosteus aculeatus*, Divino et al., 2016).

2.2. Swimming respirometry

A swimming respirometer (5.3 L, Steffensen Mk. III type; www.swimtunnels.com) was used to evaluate the critical swimming speed (U_{crit}, measured in body lengths per second, BL s^{-1}) while simultaneously measuring $\dot{M}O_2$ (mmol O_2 kg^{-1} h^{-1}) in relation to swimming speed (U). Five fish were individually examined at each salinity (total n = 10, body mass (BM) = 27.4 ± 3.3 g (mean ± SD), total length (TL) = 12.3 ± 0.5 cm (mean ± SD)). The respirometer was immersed

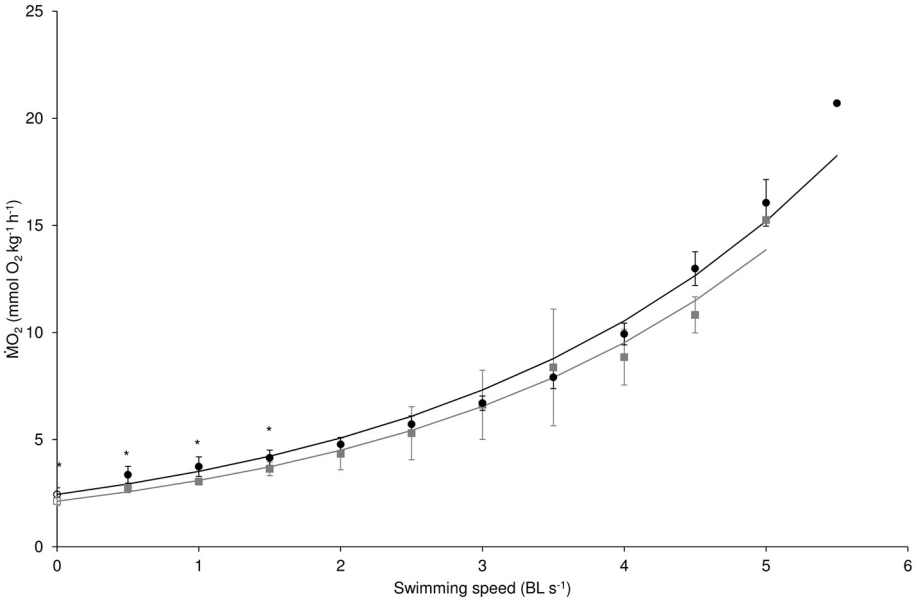

Fig. 1. Oxygen consumption rate ($\dot{M}O_2$) of shiner perch (*C. aggregata*) in relation to swimming speed (U) at salinities of 12 (grey) and 31 g kg^{-1} (black). Closed circles represent mean $\dot{M}O_2$ (± SD), and open circles represent the estimated SMR (± SD). Solid lines show fitted two-parameter exponential functions: $\dot{M}O_2(U) = 2.4441 \cdot e^{0.3658 \cdot U}$ ($r^2 = 0.9809$) at a salinity of 31 g kg^{-1} and $\dot{M}O_2(U) = 2.1279 \cdot e^{0.3748 \cdot U}$ ($r^2 = 0.9898$) at a salinity of 31 g kg^{-1}. Asterisks indicate significant differences ($p < 0.05$, one-way ANOVAs with Welch correction).

Table 1
The difference in oxygen consumption rate ($\dot{M}O_2$; mmol O_2 kg^{-1} h^{-1}) of *C. aggregata* in relation to salinity (31 and 12 g kg^{-1}, S31 and S12, respectively) at different swimming speed (U; BL s^{-1}). Given is the absolute difference in $\dot{M}O_2$ between S31 and S12 ($\Delta\dot{M}O_2$), as well as the fraction that the $\Delta\dot{M}O_2$ constitutes of the $\Delta\dot{M}O_2$ at S31 (Δ%). The sample sizes for the calculations were 5 for both salinities at all swimming speeds but the last, where sample size for S12 was 4. The P values are results of one-way ANOVAs with Welch correction.

U	0.0	0.5	1.0	1.5	2.0	2.5	3.0	3.5	4.0
$\Delta\dot{M}O_2$	0.31	0.59	0.69	0.5	0.44	0.41	0.09	−0.47	1.09
Δ%	20	18	19	12	9	7	1	−6	17
p	0.018	0.024	0.023	0.044	0.275	0.504	0.917	0.724	0.194

in a holding tank, connected to an additional water supply, in which the water was filtered through filter cotton and aerated thoroughly. A recirculation unit, controlled by a temperature regulator relay, pumped the water through a water cooler, which kept the temperature at 12.5 within ± 0.1 °C. The swimming section of the respirometer was 28 × 7.4 × 7.4 cm (length (L) × width (W) × depth (D)), the water recirculation was obtained with an impeller driven by an external electric motor (AC-motor, DRS71, SEW Eurodrive; www.sew-eurodrive.dk), and the speed controlled with a motor controller (Movitrac MCLTE, B0004-101-1-20, SEW Eurodrive; www.sew-eurodrive.dk). Deflectors collimated the flow and rectilinear flow was ensured with a honeycomb (10 cm wide, 0.5 cm cell diameter) positioned upstream the swimming section. A 10 mm horizontal grid marked the rear end of the swimming section. Different water speeds in the swimming section were measured with a 25 mm anemometer (Höntzsch, Waiblingen; www.hoentzsch.com) and were correlated to the related voltage output of the motor controller in a software (Autoresp 1, Loligo Systems; www.loligosystems.com). By entering the fish dimensions (*BM*, *TL*, *D*, *W*), which were measured prior to experiments, the software displayed the real time swimming speed corrected for the solid blocking effect (Bell and Terhune, 1970). Measurements of the fish were taken on top of a wet cloth and the air exposure time was maximally 1 min.

$\dot{M}O_2$ measurements were carried out with an intermittent-flow approach (Steffensen et al., 1984; Steffensen, 1989; Svendsen et al., 2016) in loops of 15 min, consisting of a 240 s flush period, a 60 s wait period and a 600 s measuring period. After entering the respirometer, a fish was kept at 0.5 BL s^{-1} until their $\dot{M}O_2$ reached a steady state (around 11 h) (Roche et al., 2014). After this, a U_{crit} protocol was initiated (Brett, 1964), starting at 0.5 BL s^{-1} and a swimming speed increase by 0.5 BL s^{-1} every 45 min (three $\dot{M}O_2$ determinations for each speed before U_{crit}). The 45 min per speed was chosen to avoid increasing U_{crit} as a result of short time per speed (Farlinger and Beamish, 1977), making it more comparable to other studies and less affected by burst swimming and anaerobic swimming performance. The experiment was terminated when the fish rested its tail at the rear end grid for more than three seconds. U_{crit} was determined as $U_{crit} = U_i + U_{ii}\frac{t_i}{t_{ii}}$, where U_i is the last swimming speed that fish completed (BL s^{-1}), U_{ii} is the increment (BL s^{-1}), t_i is the time that the fish endured at the last speed (s), and t_{ii} is the time prescribed for one swimming speed (s) (Brett, 1964). The swimming fish were monitored through a USB camera mounted above the swimming respirometer and the experimental setup shielded with tarps to avoid visual disturbances.

Oxygen partial pressure ($kPaO_2$) was measured every second with a fiber optic oxygen meter (Fibox 3, Precision Sensing GmbH; www.presens.de), calibrated to 100% air saturated water at the respective salinity, as well as deoxygenated water in a 10 g L^{-1} $NaSO_2$ solution. No temperature compensation was used, as the system temperature was controlled within 0.1 °C. Data acquisition was done with Autoresp 1. The program calculated the time specific oxygen decrease (α; $kPaO_2$ h^{-1}), and $\dot{M}O_2$ by $\dot{M}O_2 = \frac{\alpha V_{RE}\beta}{BM}$, where V_{RE} is the total volume of the respirometer minus the volume of the fish (L; the animal density was assumed to be 1.0 kg L^{-1}), β is oxygen solubility constant (14.863 and 13.211 μmolO_2 L^{-1} kPa^{-1} for S12 and S31, respectively (Green and Carritt, 1967)) and *BM* is the body mass of the fish (kg). The oxygen content of the water never decreased below 19.11 kPa. Background respiration (BR) was assessed before and after a trial by measuring the decrease in oxygen content in the respirometer without a fish, with a water speed of 6 cm s^{-1}, a wait period of 400 s and a measuring period of 2700 ($R^2 > 0.95$). The BR were around 30% of SMR, and subtracted from the $\dot{M}O_2$ according to Svendsen et al. (2016).

For each fish, a two parameter exponential function ($\dot{M}O_2 = a \cdot e^{b \cdot U}$) was fitted to the $\dot{M}O_2$ as a function of U (excluding the last speed, as the fish did not swim throughout the prescribed period of 45 min). The exponential function was used to determine the standard metabolic rate (SMR) and the active metabolic rate (AMR) by extrapolation, using the $\dot{M}O_2$ at 0 BL s^{-1} and the $\dot{M}O_2$ at U_{crit}, respectively. The optimal swimming speed (U_{opt}), that is, the speed where the fish use the least energy per distance (mmol O_2 kg^{-1} BL^{-1}), was calculated for each fish according to Stieglitz et al. (2016).

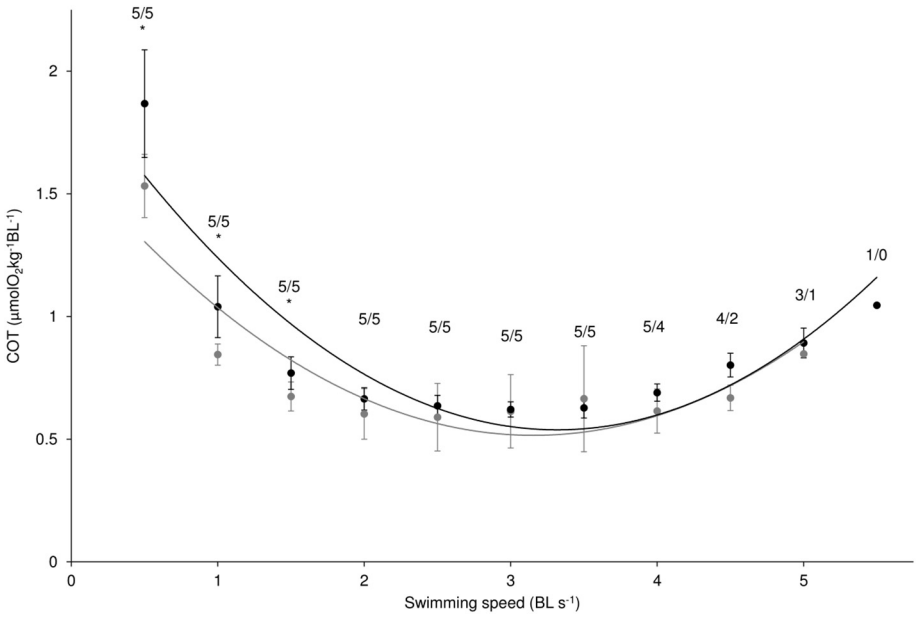

Fig. 2. Cost of transport (COT) of shiner perch (*C. aggregata*) in relation to swimming speed (U) at salinities of 12 (grey) and 31 g kg^{-1} (black). Solid lines show fitted second degree polynomials: $U_{opt} = 0.1306 \cdot x^2 - 0.8663 \cdot x + 1.975$ ($r^2 = 0.8361$) at a salinity of 31 g kg^{-1} and $U_{opt} = 0.1121 \cdot x^2 - 0.7072 \cdot x + 1.631$ ($r^2 = 0.7987$) at a salinity of 12 g kg^{-1}. Asterisks indicate significant differences ($p < 0.05$, one-way ANOVAs with Welch correction).

2.3. Statistical analyses

All statistics were performed in SPSS statistics 24 (IBM; www.IBM.com), using an α of 0.05. *BM*, *TL*, U_{crit}, all $\dot{M}O_2$ and U_{opt} data were normally distributed at all swimming speeds at both salinities (Shapiro-Wilk's tests; all $p > 0.05$). Variance heterogeneity was detected for $\dot{M}O_2$ between the two salinities at 3.5 BL s^{-1} (Levene's Test; $p = 0.041$). *BM*, *TL*, U_{crit} and $\dot{M}O_2$ measurements (at 0.5–4.0 BL s^{-1}) as well as SMR, AMR, aerobic scope (AS; the difference between AMR and SMR) and U_{opt} were compared between salinities with one-way ANOVA with Welch correction due to variance heterogeneity.

3. Results

The critical swimming speed (U_{crit}) was on average 5.1 ± 0.5 BL s^{-1} at S31 and 4.8 ± 0.6 BL s^{-1} at S12 (mean ± SD), and not significantly different ($p = 0.388$). At swimming speeds from 0.5 through 1.5 BL s^{-1} the $\dot{M}O_2$ was significantly lower ($p < 0.05$) at S12 (Fig. 1), and the absolute difference in $\dot{M}O_2$ was between 0.69 and 0.50 mmol O_2 kg^{-1} h^{-1} (Table 1). Between 2.0–4.0 BL s^{-1}, no significant difference in $\dot{M}O_2$ could be detected between the two salinities. The U_{opt} was 3.2 ± 0.2 BL s^{-1} at S31 and 2.9 ± 0.4 BL s^{-1} but not significantly different ($p = 0.233$) (Fig. 2). At S12 the extrapolated SMR (i.e. $\dot{M}O_2$ at swimming speeds of 0 BL s^{-1}) was significantly lower than at S31 by 0.31 mmol O_2 kg^{-1} h^{-1}, equivalent to a 20% change. Neither AMR nor aerobic scope was different between the two salinities ($p > 0.05$) (Fig. 3).

4. Discussion

4.1. Effects of salinity on oxygen consumption rate during swimming

The difference in SMR between the two salinity exposures was 20%, which is in accordance with previously published studies on the effect of salinity on whole-animal $\dot{M}O_2$, in which the value typically ranges from 10–30% of SMR (Boeuf and Payan, 2001; Ern et al., 2014). This is often ascribed to cost of osmoregulation, yet it is still debated to what extent whole-animal $\dot{M}O_2$ is suited to measure this process, as theoretical biochemical estimates of the cost of osmoregulation are only a few percent of SMR (Boeuf and Payan, 2001). Importantly, the $\dot{M}O_2$ in the present study increased approximately fivefold from SMR to AMR and the ventilatory flow must therefore have increased with at least the same magnitude in order to meet the oxygen demand in the tissues. Such an increase in ventilatory flow would enhance the osmotic diffusion potential, leading to a higher energetic need for osmoregulation if osmotic balance is to be maintained. However, the absolute difference in $\dot{M}O_2$ between S12 and S31 did not increase, at least at the swimming speeds where they were significantly different. This indicates either that the increase in ventilation does not cause a severe osmotic challenge in *C. aggregata*, or the fish do not allocate extra energetic expenditure to maintain osmotic homeostasis during exhaustive exercise. Exhaustive exercise protocols do cause osmotic imbalance in marine teleosts (Byrne and Beamish, 1972; Wood, 1991), and while anaerobic metabolism waste products cause ionic imbalance, it is possible that the dehydration due to an increased osmotic diffusion potential over the gills may contribute to the osmotic imbalance. If the fish is not increasing its osmoregulation effort under activity induced osmotic distress, regaining osmotic balance would be an integrated part of paying back the oxygen debt after exhaustive exercise in fish.

4.2. Effects of other factors changing with salinity

Salinity changes other important factors that are not related to osmoregulation *per se*. For instance, cortisol, an important control hormone for osmoregulation in marine teleosts, increased $\dot{M}O_2$ when injected in freshwater acclimated cutthroat trout parr (*Oncorhynchus clarki clarki*) (Morgan and Iwama, 1996). This shows that the cost of osmoregulation is influenced by both ion regulation and hormonal control. Another important factor changing with salinity is the oxygen solubility of the water (Verberk et al., 2011), which is 11% lower at S31 than at S12 at 12.5 °C. This could imply an additional cost of ventilation at S31 in order to the meet oxygen demand as a secondary consequence of changing salinity, in addition to the cost of osmoregulation. Alternation of these factors could, in addition to the cost of osmoregulation, decrease the swimming performance and make it more energetically costly to swim in S31. However, no differences could be found in U_{crit} or U_{opt}, and both parameters were even slightly higher at S31 than at S12. Water viscosity also changes with salinity, at 12.5 °C from $1.252\,10^{-6}$ m^2 s^{-1} to $1.222\,10^{-6}$ m^2 s^{-1} at S31 and S12, respectively. It is known that at a high Reynolds number ($Re > 1000$), typical for juvenile and adult fishes, swimming is dominated by inertial forces, and changes in viscosity have little effect on swimming performance (Fuiman and Batty, 1997; Hunt von Herbing and Keating, 2003). Here, the Re of the fish at U_{crit} were 2389 and 2667 at S31 and S12,

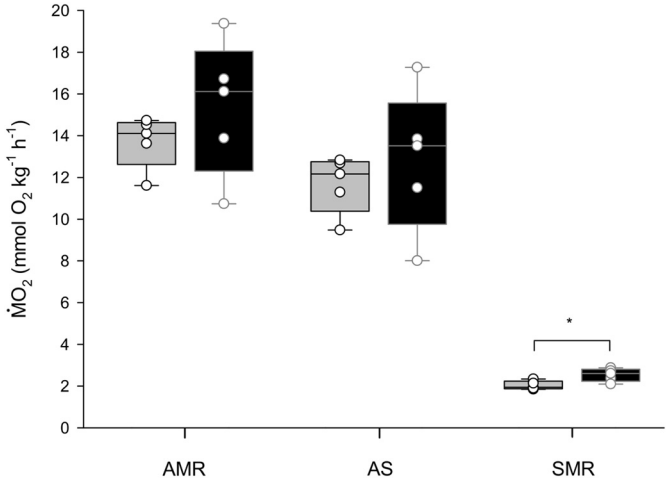

Fig. 3. Estimates of active metabolic rate (AMR), aerobic scope (AS) and standard metabolic rate (SMR) of shiner perch (*C. aggregata*) at salinities of 12 (grey, $n = 5$) and 31 g kg^{-1} (black, $n = 5$). Individual estimates (scatter plot symbols) were obtained using a two-parameter exponential function and $\dot{M}O_2$ measurements made at different swimming speeds. Significant differences in SMR are indicated by an asterisk ($p < 0.05$ ANOVA with Welch correction).

respectively, which is in the range of *Re* expected to be little affected by changes in viscosity. Overall, changing the salinity from S31 to S12 had little effect on swimming performance of *C. aggregata*, regardless of the many changing environmental factors.

4.3. Ecological implications

The ambient environment, in particular temperature, oxygen content, and salinity set the fundamental parameters for a fish species' distribution in the wild (Teal et al., 2015). Furthermore, sub-lethal environmental conditions can have effects on fish fitness (Wikelski and Cooke, 2006). Locally, climate-driven changes in rainfall and river runoff will affect the environmental salinity of coastal marine and estuarine areas (Harley et al., 2006; Vuorinen et al., 2015), consequently altering species composition and the associated prey-predator interactions, interspecific competition for food resources, and availability of suitable spawning grounds. *Cymatogaster aggregata* is a coastal roaming species, also found in estuaries (Triplett and Barrymore, 1960), where their migration might be in the search for food, to avoid predators or to spawn. The results of the present study showed no effect of salinity on swimming performance, despite an apparent energetic cost of osmoregulation in seawater at low speeds, which may provide a good explanation for the capacity of a coastal roaming species to move in an out of coastal habitat zones without significantly compromising their swimming ability. Furthermore, it shows a high adaptability for the species to potential changes in environmental salinities in the future.

Many fish species are potentially affected by changes in environmental salinity, for instance Atlantic cod (*Gadus morhua*) migrating through haloclines (Neuenfeldt et al., 2007), European perch (*Perca fluviatilis*) that conduct winter/spawning migrations between streams and estuaries (Christensen et al., 2016) and California killifish (*Fundulus parvipinnis*) in tidal ponds with extreme hypersaline water (Feldmeth and Waggoner, 1972). Knowledge on the effect of salinity on swimming performance and oxygen consumption rates of these species could be valuable targets for future studies.

4.4. Conclusions

Here we show that marine, hyperosmotic conditions, caused an increase in $\dot{M}O_2$ in relaxed and slowly swimming fishes, while at swimming speeds higher than 1.5 BL s^{-1} swimming $\dot{M}O_2$, AMR, and AS, remained unaffected. Importantly, the effect of salinity on $\dot{M}O_2$ was not high enough to significantly affect critical swimming speeds, and in turn the capacity for essential ecological tasks, such as hunting for prey, evading predators or migration. The results of the present study provide important knowledge on the interplay between individuals and environmental salinity, in an era where climate change is expected to locally affect environmental salinity in coastal marine and estuarine areas.

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