

**EFFECTS OF TEMPERATURE, SALINITY AND BODY SIZE  
ON THE ROUTINE METABOLISM  
OF SPOTTED SEATROUT (*Cynoscion nebulosus*) LARVAE**

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**Abstract**

Routine oxygen consumption rates of larval spotted seatrout (*Cynoscion nebulosus*) were measured over a range of temperatures (24, 28, 30 and 32°C) and salinities (5, 10, 20, 35 and 45‰). Larvae (5.0-49.0 mm TL) varying over several orders of magnitude in dry body mass were used to estimate an allometric scaling relationship, resulting in a bi-phasic pattern in the mass scaling of metabolic rate. Oxygen consumption ( $\mu\text{L O}_2 \text{ larva}^{-1} \text{ hr}^{-1}$ ) scaled isometrically with body mass (slope=0.997) for larvae <6.8 mm TL and allometrically (slope=0.797) thereafter. The inflection in the mass-metabolism relationship coincided with the formation of the hypural plate and a change in swimming mode. Temperature and salinity effects on routine oxygen consumption were analyzed using ANCOVA with larval dry weight as a covariate. Temperature and salinity significantly affected routine metabolism during the second phase of growth only. A significant interaction between temperature and salinity was evident at 30 and 32°C during the second phase of

growth. A response surface describing the environmental influences on routine metabolism was developed to provide a bioenergetic basis for modeling environmental constraints on the growth of this species. Ontogenetic changes in the mass scaling of metabolism are discussed in relation to the change in hydrodynamics experienced by larvae.

## **Introduction**

The routine metabolic rate ( $M$ ) can be expressed by the allometric equation  $M=aW^b$ , where  $a$  and  $b$  are constants. If  $b=1$ , then weight-specific metabolic rate remains constant, and total metabolism increases in proportion to weight. When  $b$  is less than 1, total metabolism increases more slowly than weight, and the metabolic rate per unit body mass decreases with increasing body size (Winberg, 1956). This decline in metabolic rate with size has been attributed to an increase in the relative mass of tissue with low metabolic activity (white muscle and bone), combined with a decrease in overall metabolic activity of tissues (Oikawa et al., 1991). If metabolic rate is proportional to body surface area, and different sized bodies are geometrically similar, the value for  $b$  should approximate 0.67 (Schmidt-Nielson, 1984). This theoretical value is rarely observed, but values closer to 0.75 are more common (Peters, 1983). Although the surface area is not important for thermoregulation in fishes, it is important for some fish larvae that utilize cutaneous respiration during the early life stages.

Ontogenetic changes in the relationship between body mass and metabolic rate have been reported by several investigators (Oikawa et al., 1991; Kamler, 1992; Post and Lee, 1996). It appears that in many fish species, the metabolic rates of early life history stages scale isometrically with body mass (Giguere et al., 1988). Post and Lee (1996) described a general bi-phasic pattern for the scaling of metabolic rate with body mass in several species of fish. They proposed metabolic rate increases directly proportional to body mass (isometric) early in life, and later increases less than proportional to body mass (allometric). The location, and importance of the inflection in this relationship, however, is unclear in most species. One possible explanation for the change in scaling of metabolism is the change in swimming efficiency during early development. The physical environment of fish larvae changes from a viscous flow regime to an inertial flow regime as they develop. A change in swimming style from anguilliform to subcarangiform or carangiform accompanies this change in hydrodynamic surroundings (Hunter, 1972). This change is likely to occur very early in development, perhaps about the time of first feeding, and would not explain the prolonged isometric scaling of metabolism for some species, namely,

salmonids. Marine species with small pelagic eggs and larvae will be more influenced by their hydrodynamic surroundings early in life. Spotted seatrout larvae hatch at a length of 1.65 mm TL and begin feeding at 2.6 mm TL (Alshuth and Gilmore, 1995), therefore viscous forces are likely to be important for these small stages.

Other physical properties of water affecting larvae are the temperature and salinity of the water. Spotted seatrout spend their entire life history within estuaries, and larvae are considered both eurythermal and euryhaline. Powell et al. (1989) collected larvae in salinities of 5-40 ‰ in and near Florida Bay, however, little is known on the metabolic cost incurred at different salinities. The presence of cutaneous chloride cells in early stage marine larvae enables them to maintain ion balance. Hiroi et al. (1998) demonstrated a shift in the distribution of chloride cells in Japanese flounder (*Paralichthys olivaceus*) from cutaneous to branchial chloride cells. This shift demonstrates the importance of cutaneous surfaces when larvae are small and in a viscous environment, before gills become an efficient means of ion exchange. We measured routine oxygen consumption to determine the relative metabolic costs for larval spotted seatrout over a range of temperatures, salinities and sizes. Specifically, we tested the following null hypotheses: 1) larval body mass has no effect on metabolism, 2) temperature has no effect on metabolism, and 3) salinity has no effect on metabolism.

## Methods

Spotted seatrout larvae were obtained from the Texas Parks and Wildlife GCCA/CPL Marine Development Center in Corpus Christi, Texas. The captive broodstock were spawned using photoperiod and temperature cycling. After hatching, yolk-sac larvae were shipped overnight to the NOAA Laboratory in Beaufort, NC. Larvae were reared in circular 100 L tanks. When larvae developed pigmented eyes and a functional mouth, rotifers, *Brachionus* sp., were introduced at a concentration of 5 ml<sup>-1</sup> along with algae, *Nannochloropsis* sp. or *Isochrysis* sp.

Routine metabolic rate was determined for 773 individuals at four temperatures, and five salinities. All fish were acclimated for 2-3 days prior to measurement of oxygen consumption. Artificial lights regulated to natural photoperiod were used throughout the experiment. In order to minimize the effects of diurnal variation in metabolic rate, all oxygen consumption measurements were determined from 0800 to 1600 hrs. Experimental fish were removed (by

pipetting) from culture tanks in the morning, before feeding, placed in filtered seawater of the appropriate salinity and temperature, and held for 1-2 hrs. to clear their gut. Trials were conducted in a Gilson differential respirometer on individual larvae (5.0-49.0 mm TL, 15-13190 µg dry weight) in 15 ml respiration flasks, following the procedures of Hoss et al. (1974). Larvae were allowed to acclimate for one hour in the flask before oxygen consumption measurements were made at regular intervals (0.5-1 hr) for a period of 2-6 hr depending on fish size. After each trial, fish were euthanized in MS-222, the notochord or standard length measured under a dissecting microscope, dried at 60°C, and the final dry weight determined.

The hydrodynamic environment experienced by larvae depends on their length and velocity, and the viscosity and density of water. The ratio of these inertial and frictional forces is summarized as the nondimensional Reynolds number ( $Re$ ):

$$Re = U \cdot L / \nu$$

Where  $U$  is the velocity in body lengths (BL)  $\text{sec}^{-1}$ ,  $L$  is the total length, and  $\nu$  is the kinematic viscosity (ratio of viscosity to density). When  $Re$  is less than 30 viscous forces dominate, and at  $Re > 200$  inertial forces dominate. An intermediate zone is recognized at  $30 < Re < 200$ , where the balance between the two forces gradually shifts from a viscous to inertial regime (Fuiman and Webb, 1988). Temperature and salinity both affect the Reynolds number, therefore these were held constant at 30°C and 20‰, and values of kinematic viscosity were taken from Pilson (1998).

The  $\log_e$  normalized metabolism data were subjected to a nonlinear, segmented fitting algorithm (SYSTAT, Wilkinson, 1990) to estimate the inflection point and two linear segments that best fit the data (Post and Lee, 1996). Temperature and salinity effects on the routine metabolic rate were tested in a factorial ANCOVA design with dry weight as a covariate, at a significance level of  $\alpha = 0.05$ .

## Results

A bi-phasic relationship in the scaling of routine metabolism with body weight was evident, with the inflection occurring at 6.8 mm TL (Figure 1). The dry weight of the larvae explained most of the variance in oxygen consumption for both phases of growth. During the first phase of growth, the routine metabolic

rate scaled isometrically (slope=0.997). The metabolic rate scaled allometrically (slope=0.797) for larvae greater than 6.8 mm TL.

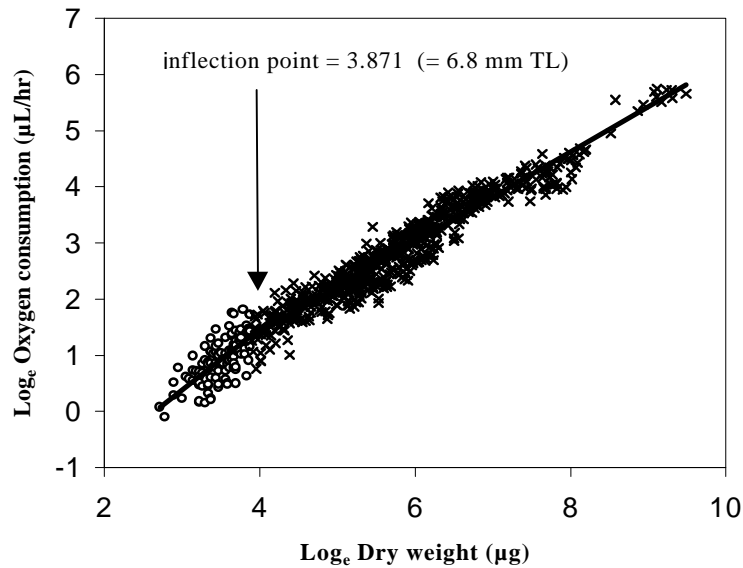


Figure 1. Weight dependence of routine metabolic rate for larval spotted seatrout.

Reynolds numbers were calculated for seatrout through the larval period to relate the influence of swimming speed and body length on the hydrodynamic regime. It is evident that larvae do not escape viscous forces until they reach a length of 6-13 mm TL (Figure 2).

Neither temperature, salinity, nor the interaction term were significant ( $\alpha=0.05$ ) during the first phase of growth, possibly due to increased variability for smaller larvae. During the second phase of growth the effect of temperature was significant ( $P=0.0001$ ). The interaction between temperature and salinity was also significant ( $P=0.0218$ ) for larger larvae. A response surface (Figure 3) was generated to describe the oxygen consumption for a 299 µg spotted seatrout larvae in response to temperature and salinity.

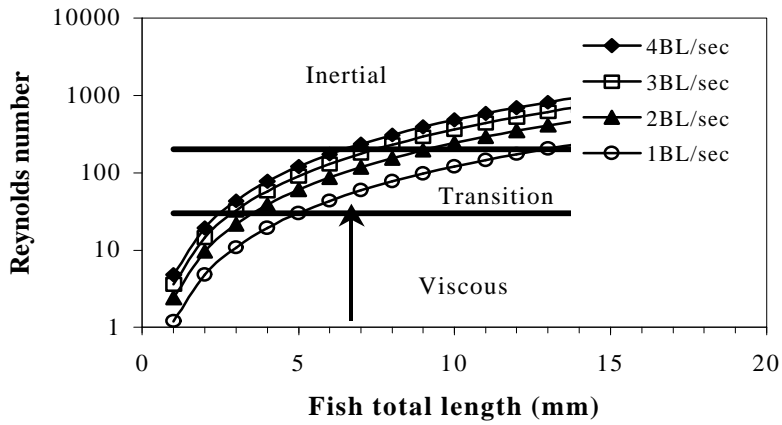


Figure 2. Reynolds numbers for spotted seatrout at different swimming speeds (temperature = 30°C, salinity = 20‰). Arrow indicates size at metabolic inflection point.

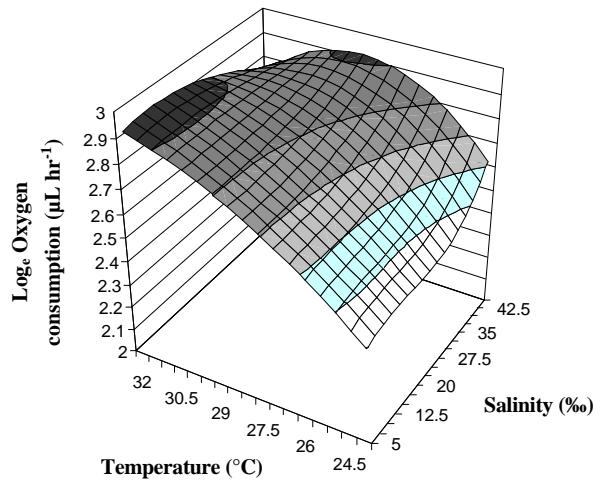


Figure 3. Response surface generated for spotted seatrout larvae during the second phase of growth (dry weight = 299 µg).

## Discussion

A bi-phasic pattern in the mass scaling of metabolic rate was observed. The oxygen consumption rate ( $\mu\text{L O}_2 \text{ larva}^{-1} \text{ hr}^{-1}$ ) scaled isometrically with body mass (slope=0.997) for larvae up until a size of 6.8 mm TL, and then scaled allometrically (slope=0.797) thereafter. The isometric scaling during the first phase of growth is consistent with the conclusions of Giguere et al. (1988) for the larvae of other fish species. The oxygen consumption rates over a wide range of larval size in this study enabled the evaluation of changes in the metabolic scaling. Based on data from this study, spotted seatrout follow the general model of metabolic ontogeny proposed by Post and Lee (1996). The inflection point for spotted seatrout is close to the value they report for sea bream, *Pagrus major*. For both of these marine fishes, the inflection is much lower than their estimates for two freshwater species, rainbow trout, *Oncorhynchus mykiss*, and common carp, *Cyprinus carpio*.

Notochord flexion occurs at 5.7 mm TL in spotted seatrout, and at 6.4 mm TL the urostyle bends upward and formation of the hypural plate begins (Alshuth and Gilmore 1995). The change in metabolic scaling at 6.8 mm TL coincides with the formation of the hypural plate. If we consider an average swimming speed of 2-3 BL  $\text{sec}^{-1}$  for larval fish (Blaxter, 1969), Figure 2 shows the correlation between the inflection point for routine metabolism and the change in hydrodynamic regime experienced by the larvae. These changes in the structure and surroundings of larvae enable them to change swimming modes from anguilliform, to the more efficient subcarangiform or carangiform modes, where increased weight becomes advantageous. This offers them an increased efficiency with size, and may account for the decreasing weight-specific metabolic rate (slope <1) with size after the inflection.

The effect of temperature on the routine metabolism was found to be significant during the second phase of growth. The effects of salinity was temperature dependent, evident by the significant interaction between temperature and salinity at 30 and 32°C during the second phase of growth. The theoretical cost of osmoregulation over a range of salinities calculated by Eddy (1982) was less than 1% for salmonids. Therefore, the energetic cost of ion regulation is likely to be very low, provided functional chloride cells are present. The overall response to salinity is probably affected by other metabolic processes. The response surface describing the influence of environmental factors on the routine metabolism provides a bioenergetic basis for modeling the environmental constraints on the spatial and temporal growth of this species. The decrease in

metabolic rate at high temperatures and salinities does not necessarily indicate increased growth under these conditions. Stressors which reduce metabolic rate may be accompanied by a decrease in consumption, therefore, growth would not be enhanced (Rice, 1990). The effect of high temperatures and salinities on the growth of spotted seatrout larvae cannot be determined until the consumption under these conditions is better understood.

## References

- Alshuth, S. and R. G. Gilmore. 1995. Egg and early larval characteristics of *Pogonias cromis*, *Bairdiella chrysoura* and *Cynoscion nebulosus* (Pisces: Sciaenidae), from the Indian River Lagoon, Florida. ICES C.M. 1995/L:17, Biol. Oceanogr. Ctte., 21pp.
- Blaxter, J. H. S. 1969. Development: eggs and larvae. Pp 177-252 *In* Fish Physiology, vol. 3., W. S. Hoar and D. J. Randall eds. Academic Press, New York.
- Eddy, F. B. 1982. Osmotic and ionic regulation in captive fish with particular reference to salmonids. *Comparative Biochemistry and Physiology* 73B:125-141.
- Fuiman, L. A. and P. W. Webb. 1988. Ontogeny of routine swimming activity and performance in zebra danios (Teleostei: Cyprinidae). *Animal Behaviour* 36:250-261.
- Giguere, L. A., B. Cote, and J.-F. St-Pierre. 1988. Metabolic rates scale isometrically in larval fishes. *Marine Ecology Progress Series* 50:13-19.
- Hiroi, J., T. Kaneko, T. Seikei and M. Tanaka. 1998. Developmental sequence of chloride cells in the body skin and gills of Japanese flounder (*Paralichthys olivaceus*) larvae. *Zoological Science* 15:455-460.
- Hoss, D. E., W. F. Hettler and L. C. Clements. 1974. Effects of thermal shock on larval estuarine fish- ecological implications with respect to entrainment in power plant cooling systems. Pp 357-371 *In* J. H. S. Blaxter ed. *The early life history of fish*. Springer-Verlag, New York

- Hunter, J. R. 1972. Swimming and feeding behavior of larval anchovy, *Engraulis mordax*. U. S. Fishery Bulletin 70:821-838.
- Kamler, E. 1992. Early life history of fish: an energetics approach. Chapman & Hall, London.
- Oikawa, S., Y. Itazawa and M. Gotoh. 1991. Ontogenetic change in the relationship between metabolic rate and body mass in a sea bream *Pagrus major* (Temminck and Schlegel). Journal of Fish Biology 38:483-496.
- Peters, R. H. 1983. The ecological implications of body size. Cambridge University Press. Cambridge.
- Pilson, M. E. Q. 1998. An introduction to the chemistry of the sea. Prentice Hall. Upper Saddle River, New Jersey.
- Post, J. R. and J. A. Lee. 1996. Metabolic ontogeny of teleost fishes. Canadian Journal of Fisheries and Aquatic Science 53: 910-923.
- Powell, A. B., D. E. Hoss, W. F. Hettler, D. S. Peters and S. Wagner. 1989. Abundance and distribution of ichthyoplankton in Florida Bay and adjacent waters. Bulletin of Marine Science 44:35-48.
- Rice, J. A. 1990. Bioenergetics modeling approaches to evaluation of stress in fishes. American Fisheries Society Symposium 8:80-92.
- Schmidt-Nielson, K. 1984. Scaling: why is animal size so important? Cambridge University Press. Cambridge.
- Wilkinson, L. 1990. SYSTAT: the system for statistics. SYSTAT Inc. Evanston, Illinois.
- Winberg, G. G. 1956. Rate of metabolism and food requirements of fishes. Nauchn. Tr. Beloruss. Gos. Univ. Im. V.I. Lenina. (Translated from Russian by Fisheries Research Board of Canada Translation Series Number 194, 1960.)

### **Acknowledgements**

This study was supported through funding from the National Oceanic and Atmospheric Administration Coastal Ocean Program. Yolk sac larvae were provided by D. Abrego, Texas Parks and Wildlife GCCA/CPL Marine Development Center in Corpus Christi, Texas. The authors wish to thank W. F. Hettler for providing oxygen consumption data at 30°C used in this analysis, J. Burke and G. Fisher for assisting in the culture of larvae at the Beaufort Laboratory, and E. Bevilacqua for help with statistical analysis.