METABOLIC FUEL PREFERENCES IN GILL AND LIVER TISSUES FROM FRESHWATER- AND SEAWATER-ACCLIMATED EEL (ANGUILLA ROSTRATA)

Elizabeth L. Crockett[1], Sarah M. Vekasi[2], and Erin E. Wilkes[3]
[1] Department of Biological Sciences, Ohio University, Athens, OH 45701
[2] Mount Desert Island High School, Mount Desert, ME 04660
[3] Ellsworth High School, Ellsworth, ME 04605

Patterns of metabolic fuel utilization may be altered during specific behavioral activities (e.g., sustained locomotion, migration) or by environmental change (e.g., food availability, temperature). Changes in diet (quantity and quality) are likely to occur for migratory animals, and many migrators must rely on stored energy reserves during this period. For the catadromous teleost Anguilla rostrata, adaptation to seawater (and the preparation to move to seawater) may signal a change in metabolic status that could elicit a switch in preferred metabolic fuels. Egginton (J. Exp. Zool. 237: 173-184, 1986) has shown that in aerobic locomotory muscle, capacities for glucose utilization (indicated by hexokinase activities) as well as fatty acid oxidation (indicated by carnitine palmitoyltransferase activities) are higher in silver (freshwater - migratory) eels compared with yellow (freshwater - nonmigratory) eels. In addition, metabolic fuel selectivity in aerobic locomotory muscle from silver eels appears to favor the oxidation of lipid fuels compared to carbohydrates (Egginton, 1986).

While much attention has been devoted to examining metabolic fuel utilization in muscular tissues (skeletal, cardiac), little is known about metabolic fuel preferences of gill epithelia (Mommsen, In Fish Physiology, v 10. eds. WS Hoar and DJ Randall. Academic Press, Orlando. pp. 203-238, 1984). It seems quite plausible that the metabolic fuel preference of gill may change as the teleost prepares for migration and its transition to the marine environment. Like muscle, gill from A. rostrata may have to rely to a greater extent on storage lipids rather than carbohydrate fuels during its migratory phase. Also, mitochondrial densities of gill chloride cells are elevated in teleosts living in seawater compared with animals raised in lower strength seawater (King et al., Cell Tissue Res. 257: 367-377, 1989). Proliferation of mitochondria in gills from seawater animals may also increase the likelihood that lipids become more important fuels during both the transition to seawater and migration. We have determined capacities for glucose and fatty acid oxidation for gill tissues from freshwater- and seawater-acclimated eels. We have also performed similar experiments with liver tissue to compare with our results for gill. We report maximal ATP yields for the two respective metabolic pathways (glycolysis and mitochondrial ß–oxidation).

Yellow eels were captured in freshwater (Penobscot River, Maine) and acclimated to freshwater (recirculating well water with daily turnover) or seawater (flow-through) for a minimum of three weeks prior to use. Temperatures in freshwater aquaria were matched (within 1°C) to seawater temperatures using chilling units. Animals were anesthetized (0.1% neutralized MS-222 dissolved in either freshwater or seawater depending on acclimation group) and gills were perfused with heparinized saline. Livers were removed and gill tissue was scraped from gill arches on an ice-cold glass stage. Liver (20% w/v) and gill (33% w/v) tissues were homogenized in 1 ml Ten Broeck ground glass homogenizers. Enzyme activities were determined using a Beckman DU 640 spectrophotometer. Assays were conducted at ambient temperatures (28°C ± 1°C). Hexokinase (HK) and carnitine palmitoyltransferase (CPT) activities were measured as the reduction of $NADP^+$ (coupled to glucose-6-phosphate dehydrogenase), and carnitine-dependent reduction of DTNB (by free CoA), respectively. CPT activities were measured with a monounsaturated substrate, palmitoleoyl CoA. While a range of fatty acids may be employed for the CPT assays, it is likely that the monounsaturated substrate will provide activities of CPT that are maximal (Egginton, 1986; Crockett and Sidell, Biochem. J. 289: 427-433, 1993). Since CPT occurs as two forms (CPT I and CPT II), and only CPT I is considered rate-limiting, we estimated CPT I as

half of the total CPT activity. ATP yields were calculated as 30 µmoles ATP/µmole glucose and 105 µmoles ATP/µmole palmitoleoyl CoA.

Capacities for ATP generation are 8-9 times greater for glucose than for the lipid fuel, palmitoleoyl CoA, in gills from either freshwater- or seawater-acclimated eels (Figure 1). In contrast with gill, liver tissues can generate at least 4-fold more ATP from oxidation of the lipid fuel compared with ATP yields calculated for carbohydrate utilization. CPT activity is 2-fold greater in hepatic tissues from seawater-acclimated eels than from freshwater-acclimated animals (data not shown). While our small sample size precludes a statistically significant result, the trend implies that there is an even greater capacity for ATP production from the lipid compared with the carbohydrate fuel in liver from seawater-acclimated animals (Figure 1).

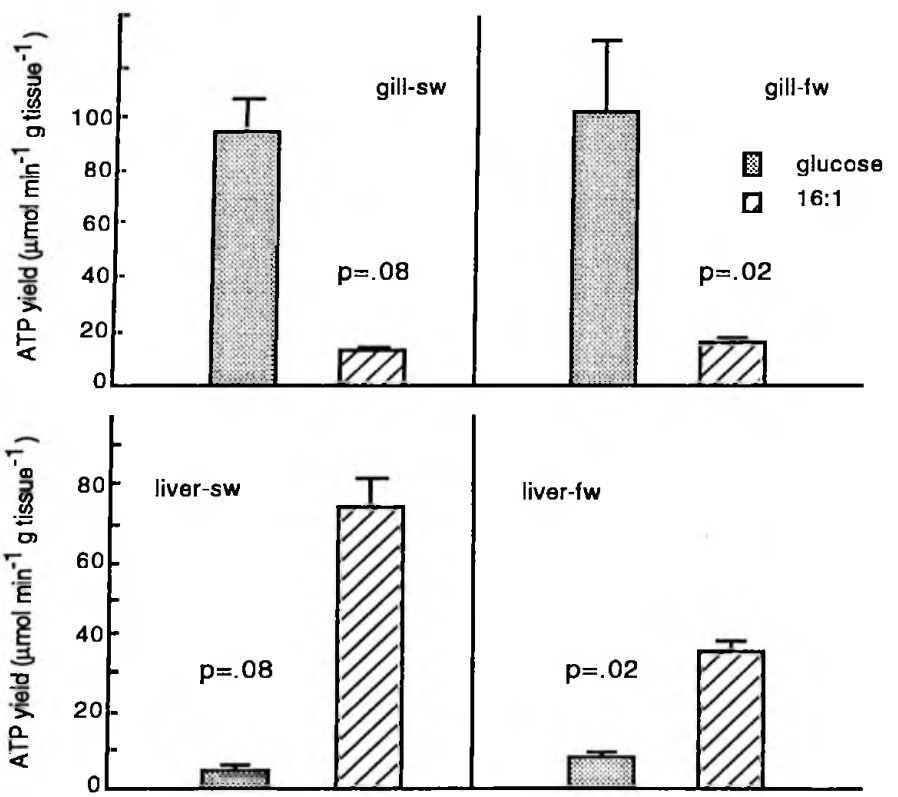

Figure 1. Capacities for ATP generation from the oxidation of glucose or palmitoleoyl CoA (16:1). Top panel: gill from seawater-acclimated (sw) and freshwater-acclimated (fw) animals. Bottom panel: liver from seawater-acclimated (sw) and freshwater-acclimated (fw) fish. Values represent mean ± SEM (sample sizes range from 2 to 4).

Our results suggest that gill tissue is largely reliant on glucose oxidation in freshwater. While a similar result emerges from our study with seawater-acclimated animals, a larger sample size is required to confirm this trend. In marked contrast, hepatic tissues show preference for the oxidation of fatty acid fuels. In addition, the capacity for fatty acid oxidation in liver is increased 2-fold in seawater-acclimated animals compared with freshwater counterparts.

Research supported by a New Investigator Award from MDIBL (Blum/Halsey Scholar Award) and start-up funds from Ohio University to ELC. SMV and EEW were recipients of fellowships from the Hancock County Scholars Program funded by NSF ESI-9452682.