

## Muscle fatty acids as indicators of niche and habitat in Malawian cichlids

**Abstract**—Fatty acid composition in the muscle of eight endemic haplochromine cichlids and one tilapiine cichlid from Lake Malawi was studied by gas-liquid chromatography. The species studied represent a wide variety of niches: Algae combing, shell crushing, feeding on littoral and semipelagic zooplankton, detecting invertebrates hidden in the sediment using expanded lateral line canals, and fish predation in littoral and bathypelagic zones. The sampling was done during the early rainy season. The analysis of fatty acid composition suggests that, despite the reported plasticity in feeding, the niches of the species studied are established and stable. The proportion of eicosapentaenoic acid (20:5n-3) in relation to arachidonic acid (20:4n-6) illustrated the position of these species in the foodweb: equal proportions in algae feeders and clear 20:4n-6 dominance in species feeding on benthic prey. Although the number of replicates was small, discriminant analysis with 31 fatty acids separated the specimens into dense centers according to species. These results encourage the use of chemometric methods for studying ecological relationships among freshwater fauna and ongoing evolutionary processes.

This paper reports for the first time the use of fatty acid signatures in African Great Lakes. Lake Malawi in the East African rift valley is one of the oldest lakes in the world harboring over 500 endemic cichlid fish species. The lake is very deep (770 m) and is warm throughout the year, with the cooler windy season during May–September and hot rainy season in October–April (Patterson and Kachinjika 1995). Most Malawian cichlids have bright breeding colors, special behavioral patterns with nest building, lekking, egg incubation in the mother's mouth, and parental care of fingerlings (Eccles and Trewavas 1989). These endemic cichlids utilize various food sources from blue-green algae, diatoms, benthic and pelagic invertebrates, to hard mollusks, and other fish. They feed by combing algae from the rock surface, sifting sand through their gill slits, scraping the scales of other fish, ambush hunting with death feigning, hitting the throat of an incubating female fish, detecting invertebrates from bottom sediments, and other means.

Due to the reported plasticity of cichlid feeding (Allison et al. 1996) we used the fatty acid signatures to estimate the stability of the niches and the importance of the trophic specializations of this group of fish. Since food sources leave clear traces in the fatty acid composition of the feeder, fatty acids are very useful in determining feeding behavior, ecological niches, foodweb relationships, and the division of food sources among various species (Ackman 1994).

**Samples**—Fatty acid composition was studied in the red muscle tissue surrounding the upper lateral line in nine species of Malawian cichlids (Pisces, Perciformes, Cichlidae). The species represented various niches from algae combing to predation (Table 1). The sampled fish were collected from trawl catches in Mangochi District, in the southeast arm of Lake Malawi in November 1991 and identified by G. F. Turner from University of Southampton, U.K. Littoral spe-

cies *Pseudotropheus zebra*, and *Cynotilapia axelrodi* were obtained from a tropical fish exporter based at Senga Bay.

**Methods**—The tissue samples were frozen immediately and stored at  $-20^{\circ}\text{C}$  in airtight vials. The total lipids in the muscle were extracted according to the method of Folch et al. (1957). Fatty acids were transesterified by heating the oils in 7%  $\text{BF}_3$  in methanol (hexane as cosolvent) in capped test tubes under nitrogen for one hour. The fatty acid methyl esters (FAMES) were then extracted into hexane, and the extract was dried and concentrated. The FAMES were analyzed by a modified analytical Carlo Erba fractovap 2450 gas-liquid chromatograph (Milan) with flame ionization detection (FID) and an Omegawax 320 fused-silica capillary column (30 m, 0.32 mm ID, 0.25  $\mu\text{m}$  film thickness, Supelco). The carrier gas was helium at 12 psig. The oven temperature was kept at  $180^{\circ}\text{C}$  for 8 min and then programmed to rise  $3^{\circ}\text{C min}^{-1}$  to  $230^{\circ}\text{C}$ . The injection port and detector were maintained at  $250^{\circ}\text{C}$ . The integrated peak areas were recorded on a Shimadzu C-R6A Chromatopac computing integrator (Kyoto) and converted to percentage fatty acid by weight by using the theoretical correction factors for FID.

The FAMES were identified by comparing them to authentic standards (Sigma) and marine oils of known composition. The nomenclature of fatty acids was used as the chain length: number of double bonds, and location of the first double bond calculated from the terminal methyl group (n-x). The percentages of fatty acids among species were compared by Kruskal–Wallis nonparametric variance test. The double bond index (DBI) was calculated from the data including all the analyzed fatty acids. Despite small sample sizes discriminant analysis was used as a descriptive tool to illustrate the separation power of fatty acid signatures.

**Fatty acids**—The conventional view of fatty acids in fish oil is that these oils are composed of one third each of saturated, monounsaturated, and polyunsaturated fatty acids (PUFA). Fish have high nutritional value as a source of the essential fatty acids, n-3, and n-6 PUFA, which cannot be synthesized in the human body. Fish oils rich in long-chain n-3 PUFA help prevent heart and vascular diseases. In feeding ecology, the most useful approach has proved to be tracing of certain fatty acids or arrays of fatty acids (fatty acid signatures). The physical storage of fat differs between fish families from the skin and bellyflap to the muscle. Among the cichlid family such variation is not known to exist, and the red muscle samples used were biochemically comparable.

In the cichlid species studied, fatty acid composition of the muscle total lipids varied greatly. Altogether 73 different fatty acids were analyzed, and the 31 most abundant were included in Table 2. According to the Kruskal–Wallis test, in all individual fatty acids the differences between species were statistically significant ( $P < 0.05$ ); the most striking differences ( $P < 0.001$ ) were found in the fatty acids 15:0

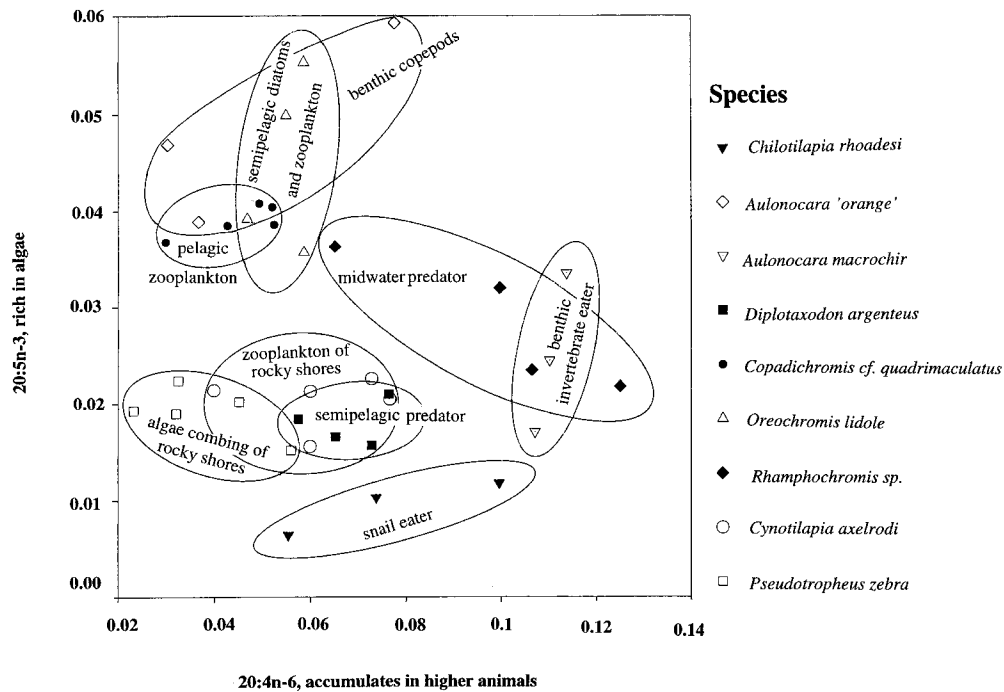


Fig. 1. The wt% of eicosapentaenoic acid (20:5n-3) versus arachidonic acid (20:4n-6) in nine Lake Malawi cichlid species.

(pentadecanoic acid), 18:3n-3 ( $\alpha$ -linolenic acid) and 20:5n-3 (eicosapentaenoic acid).

Fish in different freshwater and marine ecosystems have different general characteristics of fatty acid composition. In the muscle of the Malawian cichlids the main monounsaturated fatty acids (MUFA) were 16:1n-7 and 18:1n-9 (oleic acid), but the longer-chain MUFA 20:1n-9 and 22:1n-11 (docosenoic acid) were almost absent. The planktivorous menhaden *Brevoortia tyrannus* from the North Atlantic shares this feature (Ackman 1994).

When PUFA consumed in the diet are metabolized and

stored in animal tissues, the location of the double bond nearest to the terminal methyl group (n-x) remains unaffected. Among the cichlids the highest levels (5 wt%) of C<sub>18</sub> PUFAs (18:2n-6, 18:3n-3, and 18:4n-3) were found in species belonging to the foodweb that starts with blue-green algae on rocky shores—the detritus and zooplankton-eater *Pseudotropheus* and zooplankton feeder *Cynotilapia*. Temperate freshwater fish commonly possess twice as much 18:2n-6 and 18:3n-3 as marine fish do, but cichlids from Lake Malawi are similar to marine fish in their 18:2n-6 and 18:3n-3 contents.

Table 1. Sampled Malawian cichlids and their niches, *N* = number of samples.

Species	<i>N</i>	Feeding niche
<i>Pseudotropheus zebra</i> , Boulenger	5	Combing detritus and zooplankton from algae on rock (Genner et al. 1999)
<i>Cynotilapia axelrodi</i> , Burgess	5	Phytoplankton and zooplankton feeder from rocky areas (Genner et al. 1999)
<i>Copadichromis cf. quadrimaculatus</i> Iles	5	Semipelagic zooplankton feeder (Allison et al. 1996)
<i>Aulonocara macrochir</i> , Trewavas	3	Benthic invertebrates hidden in the mud (Eccles and Trewavas 1989)
<i>Aulonocara</i> 'orange'	3	Detritus, benthic copepods (Turner 1996)
<i>Chilotilapia rhoadesi</i> , Boulenger	3	Gastropod mollusk crusher on sandy substrate (Eccles and Trewavas 1989)
<i>Rhamphochromis</i> sp.	4	Midwater predator of fish and invertebrates (Turner 1996)
<i>Diplotaxodon argenteus</i> , Trewavas	4	Predator of the semipelagic shelf area (Turner 1996)
Endemic Tilapiine cichlid <i>Oreochromis lidole</i> , Trewavas	4	Semipelagic diatoms and zooplankton (Turner et al. 1991)

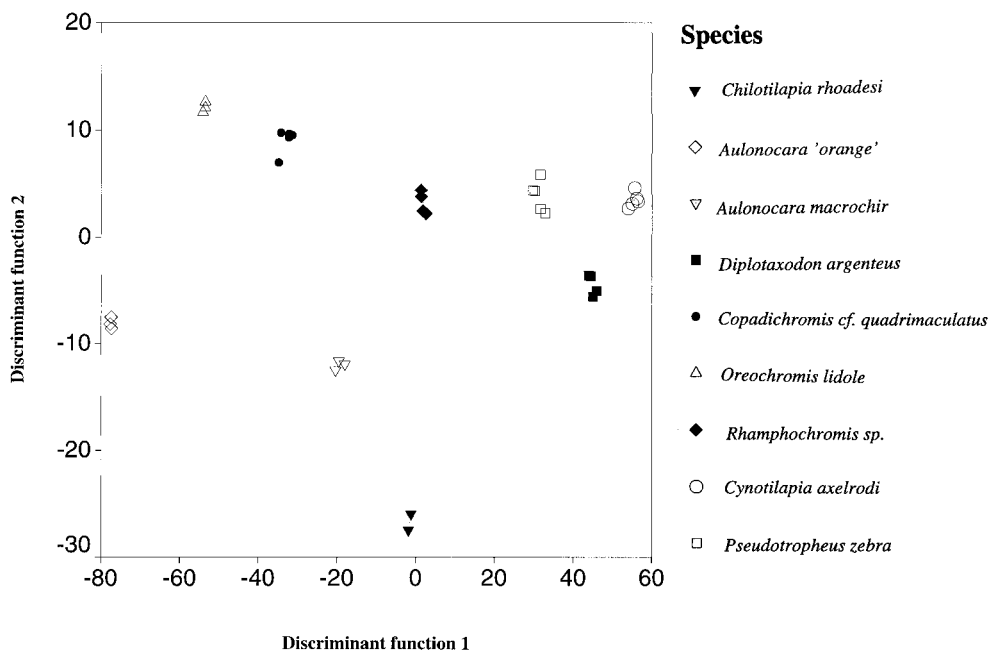


Fig. 2. Discriminant analysis of Lake Malawi cichlids based on the 31 most abundant fatty acids of the red muscle, used for descriptive purposes.

In cichlids the levels of n-3 PUFA (12–22 wt%) were low compared with many freshwater fish from northern latitudes, such as the Nordic vendace *Coregonus albula* (40–50 wt% of n-3 PUFA) (Muje et al. 1989). Freshwater fish and fish from tropical oceans commonly contain large percentages of n-6 PUFA, especially arachidonic acid (Ackman 1994). The levels of n-6 PUFA in the cichlids (8–18 wt%) are among to the highest reported. The Malawian cichlids were especially rich sources of 20:4n-6, as the contents were over three times the average found in northern freshwater fish and were comparable to those of tropical marine fish.

**Ecological implications**—Studying the n-6 PUFA/n-3 PUFA ratios may reveal ecological and foodweb relationships. We chose to plot the percentages of 20:5n-3 (eicosapentaenoic acid), which is plentiful in algae, against its counterpart in n-6 PUFA, 20:4n-6 (arachidonic acid), which accumulates in higher animals (Fig. 1). On the graph the algae comber *Pseudotropheus* was on the left side with the rock-dwelling zooplanktivore *Cynotilapia*. The species from a pelagic habitat, the diatom and zooplankton-feeder *Oreochromis*, the zooplankton-feeder *Copadichromis* and also *Aulonocara* “orange”, an undescribed species probably feeding on benthic copepods, were grouped into the corner of high 20:5n-3 and low 20:4n-6. It is surprising, that the fatty acid signature of *Aulonocara* “orange” differed greatly from that of *Aulonocara macrochir*. *A. macrochir*, which feeds on benthic invertebrates is situated in the corner of low 20:5n-3 and high 20:4n-6 with the midwater predator *Rhamphochromis*. The third species of this corner, the snail crusher *Chilotilapia* has the lowest n-3/n-6 PUFA ratio and the highest percentages of polyunsaturated 22:4n-6 (adrenic acid) and 22:5n-6.

The fatty acid composition of species from similar envi-

ronments are more similar to each other than are species from similar feeding niches—the algae comber *Pseudotropheus* is close to *Cynotilapia*, a zooplankton feeder in a rocky habitat; the bottom invertebrate eater *A. macrochir* is close to the snail crusher *Chilotilapia*, and the pelagic zooplankton feeder *Copadichromis* is close to the pelagic diatom, and zooplankton feeder *Oreochromis*. *Oreochromis* does not belong to Haplochromines as *Copadichromis* and the other studied species do, but is a member of distantly related cichlid tribe Tilapiine. Apparently, species in the same habitat are similar to each other because they belong to the same foodweb. The fatty acids of the predators *Rhamphochromis* and *Diplotaxodon* included more n-6 PUFA in relation to n-3 PUFA than did their potential prey, which may indicate trophic accumulation of n-6 PUFA in tissues and also consumption of some benthic prey.

Certain fatty acid details can be used to ascertain the trophic location of a species. For example, *A. macrochir* and benthic *Chilotilapia* included the same levels of iso- and anteiso-branched 17:0, which are usually good indicators of detritivory and bacterial lipids in the foodweb. These benthic species also possess more 20:1n-9 than the other species studied. *Aulonocara* “orange” lacks these traces, and its fatty acid signature is reminiscent of the pelagic species *Oreochromis* and *Copadichromis*. *Aulonocara* “orange” probably feeds more on algae and zooplankton, and less on bottom invertebrates. Another good example was the high  $C_{18}$  PUFAs of the algae-feeding cichlids.

The Lake Malawi endemic cichlids have clear specializations in their jaw structure, but they have been reported to have dietary plasticity in response to fluctuations in availability of prey (Allison et al. 1996). Temporary changes in the diet of fish that feed on zooplankton have been shown to affect the fatty acid content of muscle only slightly (Linko

Table 2. Principal fatty acids in total lipids of muscle from nine species of Lake Malawi cichlids (wt%, mean  $\pm$ SD). Characterization of the main food source under the scientific name. SAT, MUFA, PUFA, n-3 PUFA, and n-6 PUFA = the sums of the percentages of all identified saturated, monounsaturated, polyunsaturated, n-3 polyunsaturated, and n-6 polyunsaturated fatty acids, respectively. n-3/n-6 = the ratio of the sum of n-3 PUFA to the sum of n-6 PUFA. DBI = double bond index: average number of double bonds in a fatty acid molecule. *i* and *ai* = *iso*- and *anteiso*-branched fatty acids, respectively.

Fatty acid	Pseudotr. z. (rocky algae)	Cynotil. a. (zoo plankton)	Rhamphoc. (littoral predator)	Oreochr. lid. (pelagic algae)	Copadichr. quadr. (pelag. zooplan.)	Diplotax. arg. (pelag. pred.)	Aulon. m. (mud invertebr.)	Auton. 'or.' (benth. copepods?)	Chilot. rho. (shells)
14:0	6.4 $\pm$ 1.27	3.2 $\pm$ 1.13	2.5 $\pm$ 1.50	3.1 $\pm$ 0.60	6.9 $\pm$ 1.55	2.8 $\pm$ 0.87	2.3 $\pm$ 1.06	4.5 $\pm$ 1.54	2.2 $\pm$ 0.16
15:0	0.9 $\pm$ 0.18	0.7 $\pm$ 0.18	0.7 $\pm$ 0.22	1.1 $\pm$ 0.18	1.4 $\pm$ 0.22	0.8 $\pm$ 0.10	1.2 $\pm$ 0.25	2.6 $\pm$ 0.80	1.6 $\pm$ 0.27
<i>i</i> 15:0	0.8 $\pm$ 0.18	0.4 $\pm$ 0.21	0.2 $\pm$ 0.06	0.4 $\pm$ 0.07	0.6 $\pm$ 0.07	0.3 $\pm$ 0.07	0.4 $\pm$ 0.18	0.2 $\pm$ 0.11	0.3 $\pm$ 0.23
<i>ai</i> 15:0	0.3 $\pm$ 0.14	0.2 $\pm$ 0.11	0.1 $\pm$ 0.02	0.3 $\pm$ 2.81	0.3 $\pm$ 0.08	0.1 $\pm$ 0.03	0.2 $\pm$ 0.11	0.1 $\pm$ 0.07	0.1 $\pm$ 0.16
16:0	22.8 $\pm$ 1.28	19.1 $\pm$ 0.63	22.8 $\pm$ 1.34	24.7 $\pm$ 2.81	22.8 $\pm$ 0.13	22.4 $\pm$ 1.29	18.7 $\pm$ 2.40	17.8 $\pm$ 0.68	21.6 $\pm$ 3.30
17:0	1.2 $\pm$ 0.26	1.1 $\pm$ 0.20	1.0 $\pm$ 0.08	1.1 $\pm$ 0.27	0.9 $\pm$ 0.06	0.8 $\pm$ 0.09	1.7 $\pm$ 0.16	1.1 $\pm$ 0.27	1.5 $\pm$ 0.38
<i>i</i> 17:0	0.6 $\pm$ 0.10	0.6 $\pm$ 0.15	0.4 $\pm$ 0.07	0.3 $\pm$ 0.08	0.2 $\pm$ 0.13	0.2 $\pm$ 0.04	1.2 $\pm$ 0.16	0.5 $\pm$ 0.20	1.4 $\pm$ 0.16
<i>ai</i> 17:0	0.5 $\pm$ 0.12	0.3 $\pm$ 0.09	0.3 $\pm$ 0.11	0.3 $\pm$ 0.07	0.2 $\pm$ 0.16	0.2 $\pm$ 0.10	0.6 $\pm$ 0.12	0.2 $\pm$ 0.10	0.7 $\pm$ 0.07
18:0	6.5 $\pm$ 1.20	8.2 $\pm$ 0.82	9.0 $\pm$ 1.45	8.1 $\pm$ 0.32	5.9 $\pm$ 0.66	9.4 $\pm$ 0.68	8.9 $\pm$ 0.67	6.0 $\pm$ 1.29	7.7 $\pm$ 1.00
20:0	0.2 $\pm$ 0.10	0.4 $\pm$ 0.11	0.2 $\pm$ 0.15	0.4 $\pm$ 0.43	0.2 $\pm$ 0.07	0.2 $\pm$ 0.09	0.3 $\pm$ 0.08	0.2 $\pm$ 0.04	0.6 $\pm$ 0.69
16:1n-9	0.5 $\pm$ 0.04	0.2 $\pm$ 0.05	0.2 $\pm$ 0.05	0.3 $\pm$ 0.13	0.3 $\pm$ 0.10	0.2 $\pm$ 0.05	0.2 $\pm$ 0.03	0.3 $\pm$ 0.06	0.4 $\pm$ 0.14
16:1n-7	12.8 $\pm$ 2.41	6.2 $\pm$ 2.06	5.6 $\pm$ 3.44	7.4 $\pm$ 1.30	15.0 $\pm$ 3.63	8.3 $\pm$ 2.43	5.4 $\pm$ 1.30	17.2 $\pm$ 5.69	6.5 $\pm$ 1.73
16:1n-5	0.2 $\pm$ 0.10	0.2 $\pm$ 0.05	0.2 $\pm$ 0.07	0.2 $\pm$ 0.09	0.3 $\pm$ 0.07	0.3 $\pm$ 0.02	0.7 $\pm$ 0.12	0.4 $\pm$ 0.10	0.2 $\pm$ 0.15
17:1n-8	0.8 $\pm$ 0.13	1.0 $\pm$ 0.35	0.7 $\pm$ 0.42	0.8 $\pm$ 0.18	0.8 $\pm$ 0.15	0.7 $\pm$ 0.05	1.7 $\pm$ 0.12	2.5 $\pm$ 0.57	1.4 $\pm$ 0.17
18:1n-9	9.2 $\pm$ 0.67	9.4 $\pm$ 0.99	8.6 $\pm$ 2.40	8.5 $\pm$ 1.74	6.0 $\pm$ 0.82	10.8 $\pm$ 1.20	10.0 $\pm$ 2.45	9.8 $\pm$ 2.06	12.5 $\pm$ 5.72
18:1n-7	3.1 $\pm$ 0.46	2.6 $\pm$ 0.46	3.0 $\pm$ 0.68	3.5 $\pm$ 0.68	2.1 $\pm$ 0.25	3.3 $\pm$ 0.50	2.7 $\pm$ 0.55	3.0 $\pm$ 0.34	2.3 $\pm$ 0.21
18:1n-5	0.3 $\pm$ 0.08	0.5 $\pm$ 0.14	0.1 $\pm$ 0.04	0.1 $\pm$ 0.06	0.3 $\pm$ 0.03	0.1 $\pm$ 0.03	0.3 $\pm$ 0.05	0.2 $\pm$ 0.12	0.3 $\pm$ 0.15
20:1n-9	0.2 $\pm$ 0.06	0.3 $\pm$ 0.09	0.1 $\pm$ 0.09	0.4 $\pm$ 0.15	0.2 $\pm$ 0.05	0.3 $\pm$ 0.12	0.9 $\pm$ 0.35	0.4 $\pm$ 0.08	0.6 $\pm$ 0.31
24:1n-9	0.3 $\pm$ 0.12	0.6 $\pm$ 0.28	0.4 $\pm$ 0.39	0.3 $\pm$ 0.24	0.3 $\pm$ 0.12	0.4 $\pm$ 0.19	0.3 $\pm$ 1.15	0.2 $\pm$ 0.03	0.2 $\pm$ 0.22
16:2n-4	0.6 $\pm$ 0.26	0.3 $\pm$ 0.11	0.3 $\pm$ 0.16	0.4 $\pm$ 0.23	0.8 $\pm$ 0.19	0.3 $\pm$ 0.14	0.4 $\pm$ 0.02	0.6 $\pm$ 0.32	0.3 $\pm$ 0.04
18:2n-6	2.4 $\pm$ 0.48	2.2 $\pm$ 0.42	1.7 $\pm$ 0.37	1.3 $\pm$ 0.31	1.7 $\pm$ 0.08	1.5 $\pm$ 0.10	1.2 $\pm$ 0.30	1.1 $\pm$ 0.23	0.7 $\pm$ 0.62
20:2n-6	0.2 $\pm$ 0.02	0.3 $\pm$ 0.09	0.1 $\pm$ 0.11	0.2 $\pm$ 0.14	0.3 $\pm$ 0.07	0.2 $\pm$ 0.05	0.4 $\pm$ 0.04	0.2 $\pm$ 0.08	0.7 $\pm$ 0.09
18:3n-3	2.2 $\pm$ 0.61	2.4 $\pm$ 0.37	1.0 $\pm$ 0.21	0.9 $\pm$ 0.30	1.4 $\pm$ 0.13	0.8 $\pm$ 0.11	0.5 $\pm$ 0.07	0.6 $\pm$ 0.08	0.4 $\pm$ 0.26
20:3n-6	0.3 $\pm$ 0.09	0.5 $\pm$ 0.17	0.4 $\pm$ 0.06	0.3 $\pm$ 0.06	0.4 $\pm$ 0.08	0.4 $\pm$ 0.10	0.7 $\pm$ 0.05	0.4 $\pm$ 0.10	0.7 $\pm$ 0.63
20:3n-3	0.4 $\pm$ 0.17	0.5 $\pm$ 0.07	0.2 $\pm$ 0.13	0.3 $\pm$ 0.06	0.1 $\pm$ 0.08	0.2 $\pm$ 0.12	0.1 $\pm$ 0.06	0.1 $\pm$ 0.08	0.1 $\pm$ 0.08
18:4n-3	0.4 $\pm$ 0.10	0.3 $\pm$ 0.09	0.1 $\pm$ 0.11	0.2 $\pm$ 0.06	0.4 $\pm$ 0.08	0.1 $\pm$ 0.02	0.1 $\pm$ 0.02	0.3 $\pm$ 0.08	0.2 $\pm$ 0.23
20:4n-6	3.8 $\pm$ 1.29	6.2 $\pm$ 1.44	9.9 $\pm$ 2.51	5.5 $\pm$ 0.56	4.5 $\pm$ 0.95	6.8 $\pm$ 0.84	11.0 $\pm$ 0.33	4.8 $\pm$ 2.57	7.6 $\pm$ 2.22
22:4n-6	0.8 $\pm$ 0.25	1.1 $\pm$ 0.27	1.1 $\pm$ 0.08	0.8 $\pm$ 0.13	0.8 $\pm$ 0.09	1.2 $\pm$ 0.31	2.2 $\pm$ 0.34	0.6 $\pm$ 0.25	2.7 $\pm$ 0.53
20:5n-3	1.9 $\pm$ 0.26	2.0 $\pm$ 0.27	2.8 $\pm$ 0.69	4.5 $\pm$ 0.91	3.9 $\pm$ 0.16	1.8 $\pm$ 0.23	2.5 $\pm$ 0.82	4.8 $\pm$ 1.03	1.0 $\pm$ 0.28
22:5n-6	2.4 $\pm$ 0.75	3.7 $\pm$ 0.58	3.4 $\pm$ 0.49	1.8 $\pm$ 0.40	2.1 $\pm$ 0.41	2.7 $\pm$ 0.56	2.5 $\pm$ 0.24	0.7 $\pm$ 0.30	4.2 $\pm$ 0.97
22:5n-3	3.1 $\pm$ 0.61	3.5 $\pm$ 0.64	3.1 $\pm$ 0.77	4.7 $\pm$ 0.73	4.1 $\pm$ 0.38	3.3 $\pm$ 0.56	3.7 $\pm$ 1.12	3.8 $\pm$ 1.50	3.0 $\pm$ 0.80
22:6n-3	8.6 $\pm$ 3.10	13.3 $\pm$ 1.86	13.6 $\pm$ 2.91	9.4 $\pm$ 2.84	9.0 $\pm$ 2.46	14.6 $\pm$ 2.74	8.8 $\pm$ 2.11	7.6 $\pm$ 3.53	7.1 $\pm$ 2.51
SAT	40.5 $\pm$ 2.63	35.0 $\pm$ 1.57	37.5 $\pm$ 1.21	40.2 $\pm$ 3.00	39.5 $\pm$ 2.62	37.5 $\pm$ 1.95	35.7 $\pm$ 2.54	33.5 $\pm$ 1.00	38.5 $\pm$ 1.00
MUFA	27.6 $\pm$ 2.83	21.2 $\pm$ 2.89	19.2 $\pm$ 3.98	21.8 $\pm$ 2.41	25.5 $\pm$ 3.45	24.8 $\pm$ 3.09	22.5 $\pm$ 3.52	34.2 $\pm$ 7.13	25.1 $\pm$ 7.33
PUFA	26.8 $\pm$ 4.30	36.1 $\pm$ 3.42	37.7 $\pm$ 3.31	30.0 $\pm$ 2.53	29.2 $\pm$ 3.86	33.8 $\pm$ 4.92	33.8 $\pm$ 5.21	25.4 $\pm$ 8.89	28.0 $\pm$ 7.02
n-3PUFA	16.5 $\pm$ 2.89	22.0 $\pm$ 2.46	20.9 $\pm$ 2.90	19.9 $\pm$ 2.71	18.9 $\pm$ 2.81	20.8 $\pm$ 3.22	15.7 $\pm$ 4.00	17.2 $\pm$ 5.86	11.8 $\pm$ 3.78
n-6PUFA	9.7 $\pm$ 1.70	13.8 $\pm$ 1.75	16.7 $\pm$ 3.05	9.7 $\pm$ 1.17	9.5 $\pm$ 1.40	12.6 $\pm$ 1.73	17.7 $\pm$ 1.18	7.6 $\pm$ 3.14	15.9 $\pm$ 3.64
n-3/n-6	1.7 $\pm$ 0.09	1.6 $\pm$ 0.25	1.3 $\pm$ 0.42	2.1 $\pm$ 0.47	2.0 $\pm$ 0.19	1.7 $\pm$ 0.09	0.9 $\pm$ 0.16	2.3 $\pm$ 0.21	0.7 $\pm$ 0.17
DBI	1.52 $\pm$ 0.24	1.94 $\pm$ 0.16	2.03 $\pm$ 0.15	1.70 $\pm$ 0.16	1.65 $\pm$ 0.20	1.93 $\pm$ 0.23	1.83 $\pm$ 0.25	1.58 $\pm$ 0.39	1.61 $\pm$ 0.30

et al. 1992). Thus fatty acids are good indicators of long-term feeding patterns. Sampling was done during early rainy season, just after the productive period with high nutrient loading and high turbulence (Patterson and Kachinjika 1995). If these have resulted in plankton blooms and opportunistic feeding behavior in Malawian cichlids, it has not been sufficient to unify the fatty acid signatures. It seems highly probable that the ecological niches of the species studied here are clearly differentiated. This conclusion is further supported by the clear separation of species in discriminant analysis (Fig. 2).

Similar questions of Malawian cichlids have been studied using entirely different technique, stable isotope ratios (Bootsma et al. 1996; Genner et al. 1999). Tests of concordance of these two methods might be a very powerful way of unraveling trophic interactions in these and other systems. The present results encourage the use of chemometric methods, especially fatty acid studies, for studying ecological relationships among freshwater fauna and investigating the ongoing evolutionary processes.

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### References

ACKMAN, R. G. 1994. Animal and marine lipids, p. 292–328. In B. S. Kamel and Y. Kakuda [eds.], *Technological advances in im-*

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## Invertebrate recolonization patterns in the hyporheic zone of a gravel stream

**Abstract**—Invertebrate recolonization at short-term exposures (2–192 h) in the hyporheic zone of a gravel stream revealed significant depth-specific differences. In the shallow hyporheic zone (0–20 cm), mean meiofaunal density increased asymptotically stabilizing within 66 h, a similar but nonsignificant pattern was found for macrofauna. Permanent meiofaunal members (e.g., rotifers, chironomids, cyclopoids) were the fastest colonizers of the traps. At the deeper hyporheos (20–50 cm) meiofauna and macrofauna entered the pipes rapidly (2 h) but neither a power curve nor an asymptotic curve described their colonization pattern. Nonlinear regression analysis indicated a significant relationship between amounts of interstitial sediment entering the pipes and abundances of macrofauna and meiofauna.

- proved and alternate sources of lipids. Blackie Academic and Professional, Chapman and Hall.
- ALLISON, E. H., K. IRVINE, A. B. THOMPSON, AND B. P. NGATUNGA. 1996. Diets and food consumption rates of pelagic fish in Lake Malawi, Africa. *Freshw. Biol.* **35**: 489–515.
- BOOTSMA, H. A., R. E. HECKY, R. H. HESSLEIN, AND G. F. TURNER. 1996. Food partitioning among Lake Malawi nearshore fishes as revealed by stable isotope analysis. *Ecology* **77**: 1286–1290.
- ECCLES, D. H., AND E. TREWAVAS. 1989. Malawian Cichlid fishes. The classification of some haplochromine genera. *Lake Fish Movies*, H. W. Dieckhoff, Arenbergstrasse 27, 4352 Herten.
- FOLCH, J., M. LEES, AND G. H. SLOANE-STANLEY. 1957. A simple method for the isolation and purification of total lipides from animal tissues. *J. Biol. Chem.* **226**: 497–509.
- GENNER, M. J., G. F. TURNER, S. BARKER, AND S. J. HAWKINS. 1999. Niche segregation among Lake Malawi fishes? Evidence from stable isotope signatures. *Ecol. Lett.* In press.
- LINKO, R. R., M. RAJASILTA, AND R. H. HILTUNEN. 1992. Comparison of lipid and fatty acid composition in vendace (*Coregonus albula* L.) and available plankton feed. *Comp. Biochem. Physiol.* **103A**: 205–212.
- MUJE, P., J. J. ÅGREN, O. V. LINDQVIST, AND O. HÄNNINEN. 1989. Fatty acid composition of vendace (*Coregonus albula* L.) muscle and its plankton feed. *Comp. Biochem. and Physiol.* **92**: 75–79.
- PATTERSON, G., AND O. KACHINJIKI. 1995. Limnology and phytoplankton ecology. The fishery potential and productivity of the pelagic zone of Lake Malawi, p. 1–68. Njassa. [ed.] A. Menz. U.K. SADC, Natural Resources Institute, ODA.
- TURNER, G. F. 1996. *Offshore cichlids of Lake Malawi*. Cichlid Press, Germany. 240 pp.
- , A. S. GRIMM, O. K. MHONE, R. L. ROBINSON, AND T. J. PITCHER. 1991. The diet of *Oreochromis lidole* and other chambo species in Lakes Malawi and Malombe. *J. Fish Biol.* **39**: 15–24.

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Mobile and sedentary invertebrates inhabiting streambeds are exposed to forces of flowing water and can be displaced during periods of high discharge when stones are scoured, overturned, or rolled downstream and finer substrates are stirred or shifted (Mackay 1992). These physical disturbances promote a diverse invertebrate community adapted for recolonizing denuded areas of streambeds. The process of recolonization and subsequent changes in species presence and abundance has been intensively targeted to benthic macroinvertebrates inhabiting the streambed surfaces. The rich literature on experimental studies dealing with benthic recolonization indicates that it may be a deterministic process (Cushing and Gaines 1989), where important factors include