

Predictable Effects of Dietary Lipid Sources on the Fatty Acids Compositions of Four 1-Year-Old Wild Freshwater Fish from Poyang Lake

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ABSTRACT: The descriptors linking dietary and fish fatty acids (FAs) compositions in four 1-year-old wild freshwater fish, *Mylopharyngodon piceus*, *Ctenopharyngodon idella*, *Hypophthalmichthys molitrix*, and *Hypophthalmichthys nobilis*, from Poyang Lake were studied. *M. piceus* mainly feeding on crustaceans had the highest relation of polyunsaturated fatty acids (PUFAs; $r = 0.812$) and odd-branched chain fatty acids (OBCFAs; $r = 0.742$) with spiral shells. Correlations between *C. idella* (herbivore) and aquatic plants (PUFAs, $r = 0.995$; OBCFAs, $r = 0.783$) were higher than other diet sources. The strongest correlation for PUFAs ($r = 0.972$) between *H. molitrix* (filter feeder with phytoplankton-feeding preference) and phytoplanktons was observed, followed by zooplanktons, whereas *H. nobilis* (filter feeder with zooplankton-feeding preference) showed the highest association with zooplanktons for PUFAs ($r = 0.895$). The high retainment of dietary FAs in fish body highlighted the potential for tailoring cultured fish FAs. The preferential distributions of n-3 long-chain PUFAs in *sn*-2-triacylglycerols and *sn*-2-phospholipids made fish an alternative for inland people supplementing n-3 PUFAs.

KEYWORDS: Poyang Lake, wild freshwater fish, plankton, lipids, stereospecific analysis

INTRODUCTION

The fatty acids (FAs) compositions of fish are mainly affected by location, temperature, season, gender, species, and especially diet. In fact, many researchers believe FAs compositions of fish could be directly influenced by their dietary FAs. Different FAs from the diet metabolize in fish by different ways: saturated fatty acids (SFAs) and monounsaturated fatty acids (MUFAs) prefer to be oxidized as energy for fish, while polyunsaturated fatty acids (PUFAs) tend to be deposited in the fish body;¹ besides, freshwater fish are capable of elongating 18:2n-6 and 18:3n-3 into long-chain PUFAs (LCPUFAs, $\geq C_{20}$).² Freshwater fish contain high levels of n-6 PUFAs, 18:2n-6 and 20:4n-6, due to their diet rich in plankton.³ Benedito-Palos reported that monoenes, C18 PUFAs, and LCPUFAs were highly associated between dietary and muscle FAs compositions in 1-year-old gilthead sea bream ($r^2 > 0.90$, $p < 0.001$). The amounts of total n-3 LCPUFAs in fish (15.7–28.1%) fed with a high ratio of fish oil (>15) were higher than those (1.90–13.2%) fed with a high ratio of vegetable oil.⁴ It is generally recognized that replacing fish oil with vegetable oil blends in diets will increase the contents of total n-6 PUFAs at the expense of total n-3 LCPUFAs in fish.^{5,6}

Deep-sea fish lipids are the major source of n-3 and n-6 LCPUFAs for human diets, which play an important role in the neurological and visual development for infants, the central nervous system, the inflammation and immune process, and the prevention of cardiac and circulatory disorders.^{7–9} However, because of the great depth of the deep sea, making it hard to exploit, nutraceuticals of 20:5n-3 and 22:6n-3 from marine fish are relatively expensive. Recently, the interest in freshwater fish lipids has increased dramatically because researchers have found that freshwater fish also have good levels of n-3 LCPUFAs (20:5n-3, 1.15–13.8%; and 22:6n-3, 0.94–24.8%), which could

compensate for the lower daily consumption (<100 mg/d) of n-3 PUFAs for inland residents.

Poyang Lake, as a main water source of the Yangtze River and the largest freshwater lake in China, harbors about 136 species of fish. The abundant food organism resources of Poyang Lake, such as phytoplankton (107 species), zooplankton (86 species), zoobenthos (35 species), and submerged plants (7 species) provide fish sufficient ingredients to grow.¹⁰ From 2000 to 2006, the annual fishery production of Poyang Lake was about 34000 tons, which occupied about 70% of the total fisheries in Jiangxi province.¹⁰ In previous studies, we have found that wild freshwater fish from Poyang Lake have higher total n-3 PUFAs (these values are similar to marine fish) than freshwater species from other regions and a considerable amount of odd-branched chain FAs (OBCFAs),^{11–13} which we suppose may relate to the biodiversity of Poyang Lake. Given the significant effects of dietary FAs and negligible impacts of season and fish size on juvenile fish muscle FAs profile,^{4,14} we studied FAs compositions of four 1-year-old cyprinid fish (*Mylopharyngodon piceus*, *Ctenopharyngodon idella*, *Hypophthalmichthys molitrix*, and *Hypophthalmichthys nobilis*) and their staple dietary lipid sources (spiral shells, *Zizania latifolia*, *Phragmites communis*, phytoplankton, and zooplankton) from Poyang Lake accordingly. The selected wild fish, constituting the culturally and economically important “four famous domestic fishes”, have been used in polyculture in China since 1000 years ago, and their feeding preferences are presented in Table 1.

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Table 1. Correlation Coefficients (r) for the 1-Year-Old Wild Freshwater Fish Polyunsaturated Fatty Acids (PUFAs) and Odd-Branched Chain Fatty Acids (OBCFAs) vs Diet PUFAs) and (OBCFAs) from Poyang Lake, Hukou^a

wild fish	body length (cm)	body weight (kg)	environment	water depth (m)	food sources	r							
						spiral shells		aquatic plants		phytoplankton		zooplankton	
						PUFAs	OBCFAs	PUFAs	OBCFAs	PUFAs	OBCFAs	PUFAs	OBCFAs
<i>M. piceus</i>	31.00 ± 1.67 b	0.52 ± 0.14 c	demersal	5–30	carnivorous; mainly feeding on mollusks and crustaceans	0.812 ^c	0.742 ^c	-0.031	-0.244	0.084	0.282	0.063	0.084
<i>C. idella</i>	22.90 ± 3.32 a	0.15 ± 0.06 a	demersal	<30	herbivorous; mainly feeding on higher aquatic plants and submerged terrestrial vegetation; sometimes preying on detritus, insects, and other invertebrates	0.425	0.561 ^b	0.995	0.783 ^c	0.710	0.663 ^c	-0.796	0.001
<i>H. molitrix</i>	31.50 ± 3.54 b	0.34 ± 0.09 b	benthopelagic	<20	filter feeder with phytoplankton-feeding preference; sometimes preying on zooplankton	0.281	0.441	0.318	-0.167	0.972 ^c	0.592 ^b	0.784	0.114
<i>H. nobilis</i>	35.70 ± 0.99 b	0.41 ± 0.03 bc	benthopelagic	>5	filter feeder with zooplankton-feeding preference; sometimes preying on phytoplankton	0.143	0.385	-0.403	0.070	0.807	0.680 ^c	0.895 ^b	0.716 ^c

^aMean values ± SDs ($n = 3$). Values in the same row with different letters for each species show significant differences ($p < 0.05$). Comparison of PUFAs including 18:2n-6, 18:3n-3, 20:4n-6, 20:5n-3, 22:5n-3, 22:6n-3 and OBCFAs including iso15:0, anti15:0, 15:0, iso16:0, anti17:0, 17:0, 19:0, and 23:0, and 7-methyl-6-hexadecenoic acid including 17:1n-8 and 17:1n-7. ^bShows significant differences ($p < 0.05$). ^cShows highly significant differences ($p < 0.01$).

For triacylglycerols (TAGs) of fish, SFAs and MUFAs were preferred in position 1, whereas PUFAs and short-chain FAs were preferred in position 2, and LCPUFAs was preferred in position 3.¹⁵ Moreover, the distributions of 20:5n-3, 22:5n-3, and 22:6n-3 were governed by the amounts of 20:1 and 22:1 in TAGs. It is believed that FAs in different positions of TAGs and phospholipids (PLs) influence the absorption, metabolism, and function of diet lipids.^{16,17}

This research explored the association between selected fish and dietary FAs profiles of Poyang Lake. Stereospecific analysis of TAGs, phosphatidylcholine (PC), and phosphatidylethanolamine (PE) of the fish was subsequently analyzed to gain a better knowledge of the nutritional and biological effects of fish lipids.¹³

■ MATERIALS AND METHODS

Chemicals. PLs standards from egg yolk including PC (purity >99%), PE (purity >98%), lysophosphatidylcholine (LPC, purity >99%), and lysophosphatidylethanolamine (LPE, purity >99%), lipase from porcine pancreas (type II), phospholipase A₂ from porcine pancreas, and (trimethylsilyl) diazomethane solution were from Sigma-Aldrich Chemical Co. (St. Louis, MO). Standards of TAGs, diacylglycerols, monoacylglycerols, and free FAs were collected from fish lipids using the Hita method with modifications.¹⁸ Standard FAs methyl esters (FAMES, #463) spiked with a mixture of four positional conjugated linoleic acids (CLAs) isomers (#UC-59 M) were obtained from Nu-Chek Prep Inc. (Elysian, MN). Silica gel (ZCX-II, 54–74 μm) and silica gel GF254 TLC plates (20 cm × 20 cm) were purchased from Haiyang Chemical Group (Qingdao, China). *n*-Hexane used in GC was purchased from Merck (Darmstadt, Germany), and other solvents were analytical reagent grade.

Subjects. Four 1-year-old wild freshwater fish (five individuals for each species), *M. piceus* (31.00 ± 1.67 cm; 0.52 ± 0.14 kg), *C. idella* (22.90 ± 3.32 cm; 0.15 ± 0.06 kg), *H. molitrix* (31.50 ± 3.54 cm; 0.34 ± 0.09 kg), and *H. nobilis* (35.70 ± 0.99 cm; 0.41 ± 0.03 kg), and their main food sources (spiral shells, *Z. latifolia*, *P. communis*, phytoplankton, and zooplankton) were captured in July within Poyang Lake of Hukou, Jiangxi province, China. The study was approved by the Hukou fishery company in Jiujiang, Jiangxi province, and conducted by personnel with experience. Samples were collected on a vessel at three sites (the midpoint, the intake, and the outtake of the waterbody, respectively) from 9 a.m. to 4 p.m. on the collection day, identified by experts from the Department of Biology of Nanchang University, and transported in ice to the laboratory. Spiral shells with similar length were captured by a dredge with a 0.5 mm nylon mesh (BD-T30; Beijing Pu lite Instrument Company, China). Phytoplankton were captured by a conical net (15 cm diameter opening) rigged with a 0.064 mm nylon mesh (PTN-25; Beijing Pu lite Instrument Company), and zooplankton were captured by that with a 0.112 mm nylon mesh (PTN-13; Beijing Pu lite Instrument Company).

Fish were weighed, gutted, and filleted. Spiral shells ($n = 20$) were rehydrated with clean water for 3 days before they were decarapaced. Aquatic plants (*Z. latifolia* and *P. communis*) were rinsed with deionized water and then vacuum-dried to a constant weight. Aforementioned samples were homogenized by a blender and stored at -20 °C until analyzed. Phytoplankton and zooplankton were concentrated on 0.22 μm filters, respectively, and then stored at -80 °C before lipid analysis.

FAs Analysis. Sample lipids were extracted by Folch method,¹⁹ and the total lipids were methylated as described by Cruz-Hernandez et al.²⁰ Stereospecific analysis of TAGs and PLs of fish was performed according to published procedures.¹³ The FAMES were analyzed by a GC equipped with a flame ionization detector and a fused silica capillary column (100 m × 0.25 mm × 0.2 μm) coated with 100% cyanopropyl polysiloxane (CP-Sil 88, Chrompack; Middelburg, The Netherlands). The temperature program was 86 min: the initial temperature of the oven was 45 °C for 4 min, increased to 175 °C at a rate of 13 °C/min, and maintained for 27 min, further raised to 215 °C at a rate of 4 °C/min, and finally kept at this temperature for 35 min. Analysis of all peaks was accomplished by comparison of their retention time with FAMES standards. Fish samples were carried out in triplicate.

Table 2. FAs Compositions of the Plankton, Aquatic Plants, and Spiral Shells in Poyang Lake, Hukou, Expressed as % of Total FAs^a

FAs	phytoplankton	zooplankton	Z. <i>latifolia</i>	P. <i>communis</i>	spiral shells	FAs	phytoplankton	zooplankton	Z. <i>latifolia</i>	P. <i>communis</i>	spiral shells
4:0	—	—	0.51	0.3	—	t18:1n-9	0.38	0.57	—	0.23	0.15
iso6:0	0.28	0.35	—	1.07	—	t18:1n-7	—	—	—	—	0.09
anti7:0	—	—	—	—	—	t18:1n-6	0.13	1.28	0.10	—	—
iso8:0	—	—	—	0.08	0.43	18:1n-12	—	—	0.12	—	0.30
8:0	0.11	0.24	—	—	0.29	18:1n-9	19.83	10.73	3.73	3.90	8.99
iso9:0	—	—	0.18	—	—	18:1n-7	—	2.79	0.47	0.35	1.65
anti9:0	0.61	0.84	0.11	0.42	0.44	18:1n-6	0.05	0.29	0.89	1.82	—
9:0	—	—	—	0.05	0.10	18:1n-5	0.24	0.25	0.46	0.31	0.14
iso10:0	—	2.39	—	0.99	0.58	18:1n-4	—	—	0.34	0.47	0.12
10:0	0.06	0.19	—	0.06	—	18:1n-3	0.18	0.22	—	—	0.49
iso11:0	0.24	0.51	—	—	—	20:1n-12	—	—	—	—	4.05
anti11:0	0.04	0.41	0.11	—	—	22:1n-9	0.27	0.25	0.69	—	0.05
iso12:0	0.16	0.30	0.14	—	0.89	total cis MUFAs	23.45	18.15	9.96	8.90	19.12
12:0	0.09	0.09	7.33	0.14	0.50	total trans MUFAs	0.75	2.54	0.19	0.70	1.69
iso13:0	0.03	0.09	1.7	1.45	0.18	tt18:2n-6	—	—	0.19	0.17	—
anti13:0	0.05	0.04	2.26	—	0.16	total trans DUFAs	—	—	0.19	0.17	—
iso14:0	—	—	0.32	—	1.63	10c12t/9t11c-18:2	—	—	2.08	0.98	—
14:0	1.37	1.89	0.29	0.13	1.93	10t12c-18:2	0.20	1.27	—	—	—
iso15:0	0.19	0.35	0.22	—	—	cc-CLAs	—	—	0.36	0.17	0.21
anti15:0	—	0.19	0.16	0.22	—	11t13t-18:2	0.46	0.60	0.12	—	—
15:0	0.34	0.42	0.21	0.2	4.07	8t10t/9t11t/ 10t12t-18:2	—	—	—	—	0.17
iso16:0	—	0.33	0.09	0.11	0.39	total CLAs	0.66	1.87	2.56	1.15	0.38
16:0	17.75	17.62	17.47	17.83	18.53	18:2n-6	27.88	17.26	16.01	17.36	5.22
iso17:0	0.08	0.51	—	—	—	18:3n-6	0.27	0.57	—	—	—
anti17:0	0.13	0.06	—	—	1.93	20:2n-6	0.08	0.16	—	—	6.15
17:0	0.66	0.91	0.39	0.39	0.09	20:3n-6	0.04	0.08	—	—	0.24
18:0	5.85	5.63	1.08	1.73	6.29	20:4n-6	1.08	1.66	0.09	—	10.77
19:0	0.04	0.07	0.08	0.12	—	22:2n-6	2.15	8.19	—	—	0.57
20:0	0.34	0.25	0.66	0.26	0.15	22:4n-6	—	—	—	—	2.89
22:0	0.35	0.29	—	0.43	0.04	22:5n-6	0.66	0.91	—	—	0.83
23:0	0.15	0.22	—	—	0.79	18:3n-3	5.51	5.51	30.80	37.00	4.13
24:0	0.18	0.18	—	—	0.63	20:3n-3	0.07	0.13	3.20	0.09	0.52
26:0	0.06	—	—	—	—	20:5n-3	2.03	2.95	—	—	2.78
total SFAs	29.27	34.55	33.47	26.19	39.06	22:3n-3	—	—	0.31	—	—
11:1	—	—	—	4.91	—	22:5n-3	0.17	0.19	—	—	1.72
14:1n-5	—	—	—	—	0.05	22:6n-3	2.48	3.64	—	—	1.09
t16:1n-8	0.14	0.44	—	—	0.26	total PUFAs	43.06	43.14	53.16	55.77	37.12
t16:1n-7	0.11	0.26	—	—	0.81	total n-3 LCPUFAs	8.75	17.92	3.60	0.09	6.11
16:1n-9	0.43	0.55	2.35	1.31	0.31	total n-6 PUFAs	32.16	28.85	16.10	17.36	26.67
16:1n-8	—	—	—	0.17	—	total n-3 PUFAs	10.24	12.42	34.31	37.09	10.23
16:1n-7	1.68	2.04	0.44	0.52	2.41	n-6/n-3 PUFAs	3.14	2.32	0.47	0.47	2.61
16:1n-6	0.23	0.26	—	—	0.25	total (n-6 + n-3)/ SFAs	1.45	1.19	1.51	2.08	0.94
16:1n-5	—	—	0.08	—	—	total BCFAs	1.81	6.37	5.29	4.34	6.63
17:1a	—	—	—	—	0.21	total SOCFAs	1.19	1.62	0.68	0.76	5.05
17:1n-8	0.14	0.14	0.38	—	—	total UOCFAs	0.54	0.77	0.38	4.98	0.21
17:1n-7	0.40	0.63	—	0.07	—	total OBCFAs	3.54	8.76	6.35	10.08	11.89
t18:1n-12	—	—	0.09	0.47	0.38						

^aTotal SFAs, total saturated fatty acids; 17:1a, 7-methyl-6-hexadecenoic acid; total cis MUFAs, total cis monounsaturated fatty acids; total trans MUFAs, total trans monounsaturated fatty acids; total trans DUFAs, total trans diunsaturated fatty acids; total CLAs, total conjugated linoleic acids; total PUFAs, total polyunsaturated fatty acids; total n-3 LCPUFAs, total n-3 long-chain polyunsaturated fatty acids; total n-6 PUFAs, total n-6 polyunsaturated fatty acids; total n-3 PUFAs, total n-3 polyunsaturated fatty acids; total BCFAs, total branched chain fatty acids; total SOCFAs, total saturated odd carbon fatty acids; total UOCFAs, total unsaturated odd carbon fatty acids; total OBCFAs, total odd-branched chain fatty acids; and —, not detected.

Statistical Analysis. FAs compositions were analyzed with one-way analysis of variance, and mean values were compared using Duncan's test. Bivariate correlations between fish and dietary FAs compositions

were examined by Pearson correlation coefficient (r). The significance level was set at $p < 0.05$. All statistical analyses were carried out using SPSS 18.0 software for windows (SPSS Inc., Chicago, IL).

Table 3. FAs Compositions of the 1-Year-Old Wild Freshwater Fish in Poyang Lake, Hukou, Expressed as % of Total FAs^a

FAs	<i>M. piceus</i>	<i>C. idella</i>	<i>H. molitrix</i>	<i>H. nobilis</i>	FAs	<i>M. piceus</i>	<i>C. idella</i>	<i>H. molitrix</i>	<i>H. nobilis</i>
iso8:0	—	—	—	0.27 ± 0.04	23:0	0.30 ± 0.01 a	0.23 ± 0.05 a	—	0.32 ± 0.14 a
iso10:0	—	—	0.13 ± 0.03	—	total SFAs	28.61 ± 0.56 a	31.97 ± 0.17 b	29.83 ± 0.64 ab	35.05 ± 2.29 c
12:0	—	0.10 ± 0.04 a	0.17 ± 0.06 a	0.51 ± 0.08 b	14:1n-5	0.03 ± 0.00 a	0.04 ± 0.01 a	0.07 ± 0.00 b	—
iso13:0	—	0.05 ± 0.01	—	0.25 ± 0.13	t6:1n-8	0.16 ± 0.02 a	0.15 ± 0.01 a	0.23 ± 0.01 b	0.22 ± 0.01 b
anti13:0	—	0.05 ± 0.01	—	0.28 ± 0.11	t6:1n-7	0.03 ± 0.00 a	1.74 ± 0.04 d	0.86 ± 0.02 c	0.71 ± 0.13 b
13:0	—	0.07 ± 0.03 a	0.12 ± 0.03 ab	0.16 ± 0.01 b	t6:1n-6	—	—	0.07 ± 0.01	—
iso14:0	—	0.07 ± 0.01 a	0.10 ± 0.01 b	0.18 ± 0.00 c	16:1n-9	0.47 ± 0.04 ab	0.73 ± 0.05 c	0.70 ± 0.01 bc	0.42 ± 0.25 a
14:0	1.26 ± 0.30 a	1.76 ± 0.08 b	3.23 ± 0.30 d	2.25 ± 0.17 c	16:1n-8	—	—	0.08 ± 0.02 a	0.60 ± 0.30 b
iso15:0	0.19 ± 0.03 a	0.67 ± 0.02 c	0.62 ± 0.03 b	0.81 ± 0.00 d	16:1n-7	6.18 ± 0.25 b	6.05 ± 0.09 b	8.45 ± 0.13 c	4.22 ± 0.15 a
anti15:0	0.03 ± 0.02 a	0.19 ± 0.02 b	0.18 ± 0.01 b	0.21 ± 0.01 b	16:1n-6	0.06 ± 0.01 a	—	0.21 ± 0.04 b	0.20 ± 0.01 b
15:0	1.01 ± 0.10 c	0.83 ± 0.08 b	0.89 ± 0.02 bc	0.63 ± 0.00 a	16:1n-5	0.06 ± 0.01 b	0.05 ± 0.00 ab	0.04 ± 0.00 a	—
iso16:0	0.43 ± 0.01 b	0.61 ± 0.08 c	0.21 ± 0.00 a	—	17:1a	0.25 ± 0.02 a	0.37 ± 0.11 a	0.23 ± 0.00 a	0.31 ± 0.11 a
16:0	16.06 ± 0.32 a	20.41 ± 0.13 c	19.43 ± 0.15 c	17.64 ± 1.30 b	17:1n-12	0.04 ± 0.00 a	0.19 ± 0.00 b	0.31 ± 0.01 c	—
iso17:0	2.79 ± 0.03	—	0.04 ± 0.01	—	17:1n-10	—	0.03 ± 0.00	—	—
anti17:0	0.26 ± 0.02 b	0.38 ± 0.03 c	0.12 ± 0.08 a	—	17:1n-8	0.76 ± 0.00 d	0.56 ± 0.01 b	0.60 ± 0.02 c	0.42 ± 0.00 a
17:0	1.04 ± 0.00 b	1.14 ± 0.02 c	0.68 ± 0.01 a	1.25 ± 0.00 d	17:1n-7	0.11 ± 0.01 c	0.08 ± 0.00 b	0.06 ± 0.01 a	—
iso18:0	0.28 ± 0.00 b	0.18 ± 0.03 a	0.37 ± 0.00 c	—	t18:1n-12	—	—	0.22 ± 0.00	—
18:0	4.72 ± 0.20 a	4.45 ± 0.04 a	4.05 ± 0.16 a	8.19 ± 0.99 b	t18:1n-9	0.73 ± 0.03 d	0.35 ± 0.01 c	0.07 ± 0.01 a	0.18 ± 0.00 b
19:0	0.14 ± 0.02 b	0.36 ± 0.00 d	0.12 ± 0.00 a	0.25 ± 0.00 c	t18:1n-7	0.05 ± 0.02	—	0.13 ± 0.11	—
20:0	0.10 ± 0.00 a	0.30 ± 0.01 bc	0.25 ± 0.05 b	0.37 ± 0.08 c	t18:1n-6	—	—	0.19 ± 0.10	—
22:0	—	0.14 ± 0.01 bc	0.12 ± 0.01 b	—	18:1n-12	0.31 ± 0.05	—	—	—
18:1n-9	25.81 ± 0.65 d	12.67 ± 0.07 b	18.31 ± 0.14 e	11.21 ± 0.03 a	20:2n-6	1.28 ± 0.03 c	0.79 ± 0.00 b	0.36 ± 0.00 a	0.36 ± 0.11 a
18:1n-7	2.66 ± 0.06 a	2.99 ± 0.03 b	4.09 ± 0.14 d	3.38 ± 0.01 c	20:3n-6	0.37 ± 0.02 a	0.98 ± 0.01 d	0.51 ± 0.02 c	0.43 ± 0.03 b
18:1n-5	0.24 ± 0.01 c	0.17 ± 0.01 b	0.09 ± 0.00 a	0.66 ± 0.00 d	20:4n-6	11.68 ± 0.04 c	3.47 ± 0.11 b	1.65 ± 0.14 a	3.51 ± 0.56 b
18:1n-3	0.48 ± 0.00	0.56 ± 0.00	—	—	22:4n-6	0.91 ± 0.02 c	0.34 ± 0.01 b	0.13 ± 0.01 a	0.38 ± 0.06 b
20:1n-15	0.05 ± 0.04	—	—	0.44 ± 0.00	22:5n-6	1.87 ± 0.02 c	0.77 ± 0.02 a	1.13 ± 0.06 b	3.46 ± 0.02 d
20:1n-12	—	0.07 ± 0.01	—	—	18:3n-3	1.27 ± 0.04 a	11.62 ± 0.21 d	9.51 ± 0.12 c	6.42 ± 0.43 b
20:1n-9	0.79 ± 0.03	—	—	0.46 ± 0.00	20:3n-3	0.33 ± 0.01 a	0.39 ± 0.01 a	0.52 ± 0.05 b	0.63 ± 0.06 c
22:1n-9	—	0.05 ± 0.01	—	1.09 ± 0.03	20:5n-3	1.41 ± 0.02 a	1.69 ± 0.02 b	3.47 ± 0.11 c	5.75 ± 0.15 d
24:1n-9	—	—	0.05 ± 0.00	—	22:3n-3	0.06 ± 0.01	—	—	—
total cis MUFAs	38.18 ± 0.55 d	24.47 ± 0.16 b	33.05 ± 0.21 c	23.74 ± 0.01 a	22:5n-3	1.08 ± 0.02 a	1.25 ± 0.05 ab	1.07 ± 0.01 a	1.53 ± 0.35 b
total trans MUFAs	0.97 ± 0.02 a	2.24 ± 0.06 c	1.77 ± 0.07 b	1.11 ± 0.12 a	22:6n-3	3.41 ± 0.06 db	2.96 ± 0.06 a	3.48 ± 0.18 b	12.24 ± 0.28 c
tl18:2n-6	—	0.11 ± 0.01	1.15 ± 0.07	—	total PUFAs	29.84 ± 0.13 a	38.98 ± 0.35 b	30.78 ± 0.25 a	39.47 ± 0.90 b
ct18:2n-6	0.15 ± 0.04	—	—	—	total n-3 LCPUFAs	6.29 ± 0.02 a	7.27 ± 0.62 ab	8.36 ± 0.25 b	21.11 ± 1.37 c
tc18:2n-6	0.08 ± 0.01	—	—	—	total n-6 PUFAs	21.31 ± 0.06 d	19.36 ± 0.08 c	8.91 ± 0.20 a	11.46 ± 0.20 b
total trans DUFAs	0.23 ± 0.05 b	0.11 ± 0.01 a	1.15 ± 0.07 c	—	total n-3 PUFAs	7.57 ± 0.04 a	18.45 ± 0.25 b	18.05 ± 0.32 b	26.56 ± 0.70 c
9c11t-18:2	0.03 ± 0.00	—	0.06 ± 0.01	—	n-6/n-3 PUFAs	2.82 ± 0.00 d	1.05 ± 0.01 c	0.49 ± 0.02 b	0.43 ± 0.00 a
cc-CLAs	0.13 ± 0.05 b	0.10 ± 0.02 ab	0.05 ± 0.02 a	—	total (n-6 + n-3)/SFAs	1.00 ± 0.01 d	1.18 ± 0.02 c	0.87 ± 0.03 a	1.14 ± 0.10 c
11t13t-18:2	0.21 ± 0.01 a	0.30 ± 0.01 a	2.40 ± 0.09 b	—	total BCFAs	3.98 ± 0.12 d	2.20 ± 0.11 c	1.77 ± 0.04 a	2.00 ± 0.08 b
8t10t/9t11t/10t12t-18:2	0.35 ± 0.01 b	0.67 ± 0.01 c	0.16 ± 0.01 a	1.44 ± 0.06 d	total SOCFAs	2.49 ± 0.05 b	2.63 ± 0.14 b	1.81 ± 0.16 a	2.61 ± 0.21 b
total CLAs	0.74 ± 0.03 a	1.07 ± 0.02 b	2.67 ± 0.09 d	1.44 ± 0.06 c	total UOCFAs	1.16 ± 0.04 b	1.23 ± 0.05 b	1.20 ± 0.06 b	0.31 ± 0.02 a
18:2n-6	5.15 ± 0.05 b	12.80 ± 0.16 c	4.67 ± 0.15 b	3.32 ± 0.64 a	total OBCFAs	7.63 ± 0.15 d	6.06 ± 0.28 c	4.78 ± 0.09 a	5.34 ± 0.17 b
18:3n-6	0.05 ± 0.00 a	0.20 ± 0.00 b	0.46 ± 0.02 c	—					

^aMean values ± SDs ($n = 3$). Values in the same row with different letters for each species show significant differences ($p < 0.05$). Total SFAs, total saturated fatty acids; 17:1a, 7-methyl-6-hexadecenoic acid; total cis MUFAs, total cis monounsaturated fatty acids; total trans MUFAs, total trans monounsaturated fatty acids; total trans DUFAs, total trans disaturated fatty acids; total CLAs, total conjugated linoleic acids; total PUFAs, total polyunsaturated fatty acids; total n-3 LCPUFAs, total n-3 long-chain polyunsaturated fatty acids; total n-6 PUFAs, total n-6 polyunsaturated fatty acids; total n-3 PUFAs, total n-3 polyunsaturated fatty acids; total BCFAs, total branched chain fatty acids; total SOCFAs, total saturated odd carbon fatty acids; total UOCFAs, total unsaturated odd carbon fatty acids; total OBCFAs, total odd-branched chain fatty acids; and —, not detected.

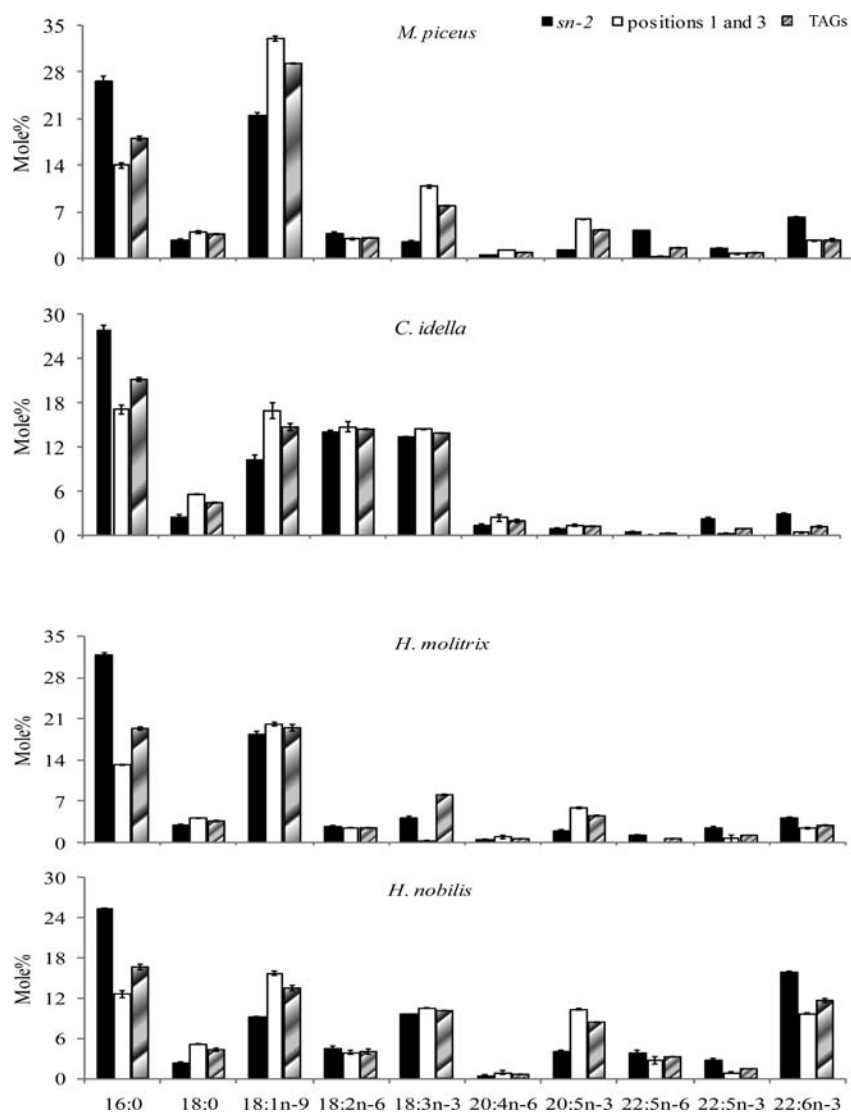


Figure 1. Positional distribution of major FAs in TAGs of four wild freshwater fish from Poyang Lake (mol %, $n = 3$). Mean values with standard deviations are plotted as bars.

RESULTS AND DISCUSSION

Total FAs Profile of Four Wild Freshwater Fish Diet.

The proportions of total PUFAs (43.06–55.77%) predominated over total SFAs (26.19–34.55%) and total MUFAs (9.60–24.20%) in *Z. latifolia*, *P. communis*, phytoplankton, and zooplankton, whereas total SFAs showed the maximum value (39.06%) in spiral shells (Table 2). 16:0 fluctuated modestly from 17.47% in *Z. latifolia* to 18.53% in spiral shells. The total OBCFAs of fish diets was from 3.54% to 11.89%. The total cis MUFAs varied between 8.90% of *P. communis* and 23.45% of phytoplankton, with 18:1n-9 as the richest isomer. The total CLAs showed the highest level in *Z. latifolia* (2.56%) and the lowest level in spiral shells (0.38%). It has been reported that CLAs in diet could decrease the levels of SFAs, MUFAs, and total lipids but increase the levels of n-3 PUFAs of fish.²¹ CLAs could improve antioxidation, immunity, and growth, regulate cholesterol and TAGs levels in blood, and prevent atherosclerosis.²² 18:2n-6 of plankton (>17%), 18:3n-3 of aquatic plants (>30%), and 20:4n-6 (>10%) of spiral shells showed exclusive dominance over other PUFAs, respectively. *P. communis* and spiral shells could be distinguished from other species by 11:1

(4.91%) and 22:4n-6 (2.89%), respectively. Zooplankton and phytoplankton had the highest and lowest contents of 20:5n-3, 22:5n-6, 22:5n-3, and 22:6n-3, respectively, with spiral shells in between (exception for 22:5n-3), while these LCPUFAs were barely detected in aquatic plants.

Fish FAs Profile Tailored by Diets. Significant differences were observed for most of the FAs among different fish species (Table 3). Among the selected fish, total SFAs and total PUFAs decreased with the order of *H. nobilis* > *C. idella* > *H. molitrix* > *M. piceus*, whereas total MUFAs increased as *H. nobilis* < *C. idella* < *H. molitrix* < *M. piceus*. 16:0 predominated in total SFAs from 16.06% to 20.41%, followed by 18:0. Our studied fish showed similar levels of total SFAs (28.61–35.05%) to freshwater fish from other regions (23.10–35.60%) but lower levels than most marine fish (30.10–40.60%).²³ Total MUFAs varied between 24.85% in *H. nobilis* and 39.15% in *M. piceus*, of which 18:1n-9 was the most abundant. Similar levels of total MUFAs were found in freshwater fish (22.60–39.60%) and marine fish (27.60–37.60%) from other regions.²³ Interestingly, although CLAs are mainly from ruminant products, small amounts of CLAs (0.74–2.67%) were found in our fish lipids, which may be attributed to their diets containing a small

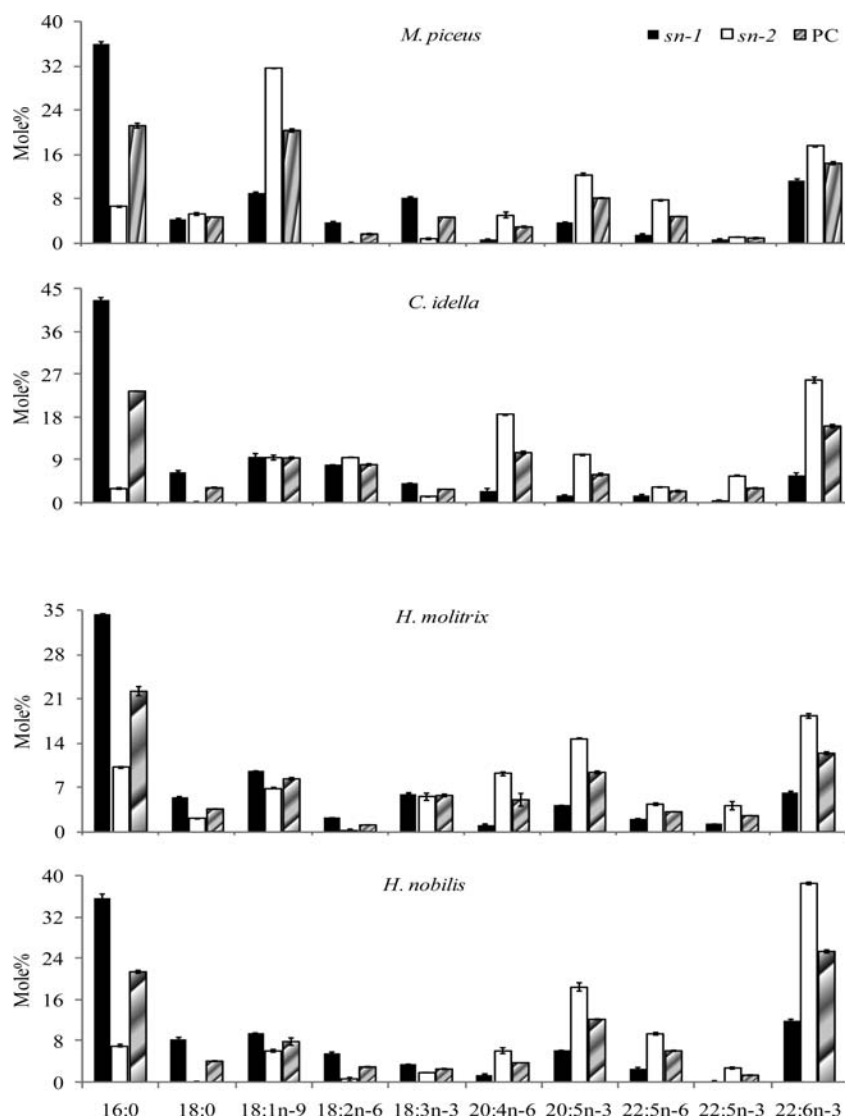


Figure 2. Positional distribution of major FAs in PC of four wild freshwater fish from Poyang Lake (mol %, $n = 3$). Mean values with standard deviations are plotted as bars.

proportion of CLAs mentioned above (Table 2). The proportions of 18:2n-6 and 18:3n-3 in *C. idella* were higher than other fish, with 12.80% and 11.62%, respectively. *M. piceus* predominated in the content of 20:4n-6 (11.68%); *H. nobilis* of 22:5n-6 (3.46%), 20:5n-3 (5.75%), 22:5n-3 (1.53%), and 22:6n-3 (12.24%). The ratios of n-6/n-3 PUFAs (0.43–2.82) and (n-6 + n-3) PUFAs/SFAs (0.87–1.18) in all of the studied fish were in accordance with FAO/WHO recommendations (n-6/n-3 PUFAs, <4; (n-6 + n-3) PUFAs/SFAs, >0.4–0.5), which will be beneficial for improving human nutrition.²⁴ Otherwise, the imbalance intake of n-6 and n-3 PUFAs could lead to inflammation, dysfunctions of the immunological system, cancer, and cardiovascular disease.^{25,26} The n-3 LCPUFAs in our study ranged from 6.29% to 21.11%, which were similar to most of the freshwater fish (2.20–19.60%) but lower than marine fish (14.90–35.20%) reported by Li et al., who explained that this discrepancy may be due to the different dietary FAs compositions of freshwater and marine fish.²³ It has been reported that the levels of n-3 and n-6 PUFAs in fish, such as 20:5n-3, 22:6n-3, and 18:2n-6, can be altered by fish feeds enriched with fish oil or vegetable oils. Fish oil could improve the utilization of diet lipid, while vegetable oil containing a low

level of cholesterol could influence the digestion, absorption, and transportation of lipid in fish.^{27–29} Besides, researchers link the high amounts of PUFAs in freshwater fish to their feed preference of phytoplankton, which are usually rich in 18:2n-6 and 18:3n-3 and high activity of $\Delta 6$ and $\Delta 5$ desaturase and elongase.^{30,31} It is interesting to note that the levels of PUFAs (29.84–39.47%) in the fish selected were higher than those in marine fish from the Mauritanian coast (17.78–22.17%)³² and freshwater fish (11.42–22.21%) from the Indus River,³⁰ which may be ascribed to the contribution of diverse food organism resources provided by Poyang Lake. The correlation of PUFAs compositions of diets against fish is shown in Table 1, where correlation coefficients (r) and p values were considered for 18:2n-6, 18:3n-3, 20:4n-6, 20:5n-3, 22:5n-6, 22:5n-3, and 22:6n-3 at detectable contents in all of the analyzed samples. The highest and strongly evident relation of PUFAs ($r = 0.812$, $p < 0.01$) was established between *M. piceus* and spiral shells. A higher correlation was reported between *C. idella* and aquatic plants for PUFAs ($r = 0.995$, $p > 0.05$) than the other three diet species. *H. molitrix* and phytoplankton were observed with the especially significant and highest correlation for PUFAs ($r = 0.972$, $p < 0.01$), followed by zooplankton ($r = 0.784$, $p > 0.05$).

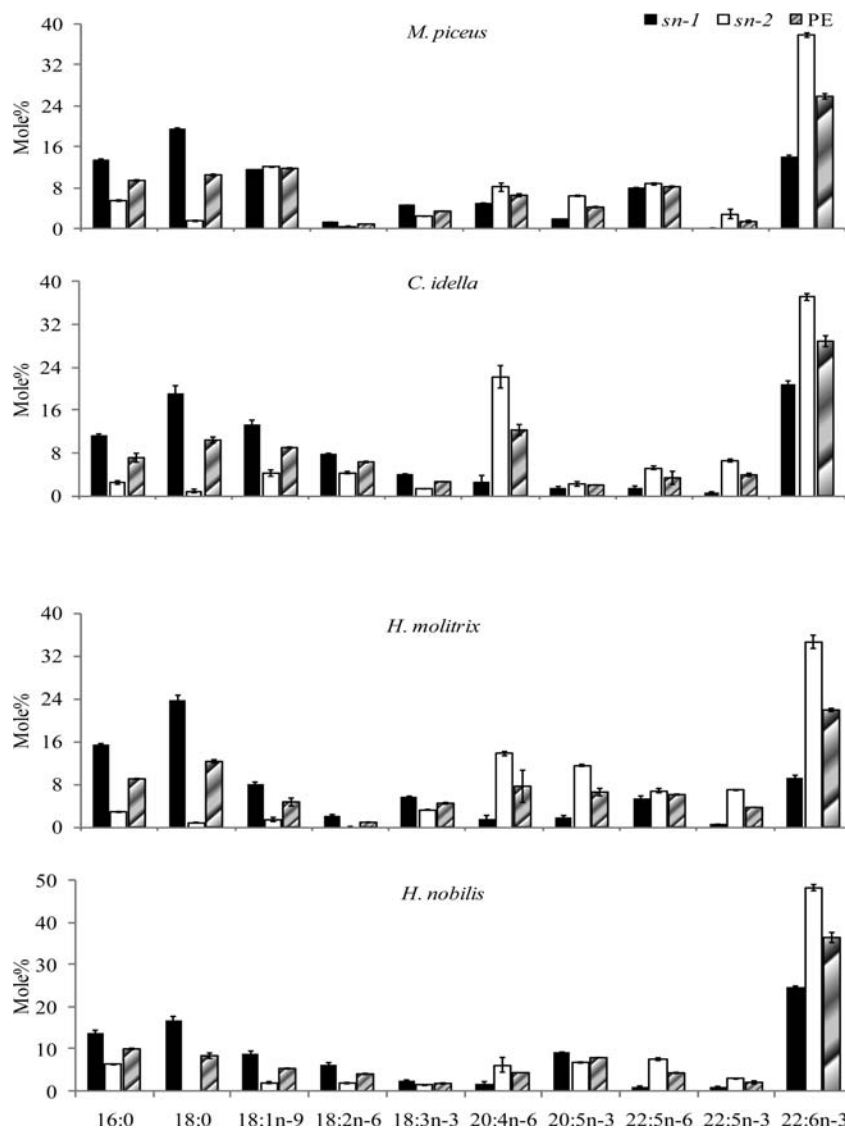


Figure 3. Positional distribution of major FAs in PE of four wild freshwater fish from Poyang Lake (mol %, $n = 3$). Mean values with standard deviations are plotted as bars.

H. nobilis showed an evidently positive and highest association with zooplankton for PUFAs ($r = 0.895$, $p < 0.05$) (Table 1). A strong association between FAs profile and food habits is observed in the four freshwater fish (Table 1). Table 3 shows that *H. molitrix* and *H. nobilis* exhibited higher contents of n-3 PUFAs (18.05–26.56%) than n-6 PUFAs (8.91–11.46%), while *M. piceus* and *C. idella* displayed lower contents of n-3 PUFAs (7.57–18.45%) than n-6 PUFAs (19.36–21.31%). This is in accordance with the results of Li et al., who attributed the discrepancy to the fact that *H. molitrix* and *H. nobilis* (typical filter feeder fish) mainly feed on the plankton rich in 20:5n-3 and 22:6n-3.²³ The amounts of 20:5n-3, 22:6n-3, and total n-3 LCPUFAs declined as zooplanktons > phytoplanktons > spiral shells > aquatic plants (Table 2), so *H. nobilis* (mainly feed on zooplankton) had the highest proportions of 20:5n-3 (5.75%), 22:6n-3 (12.24%), and total n-3 LCPUFAs (21.11%), followed by *H. molitrix* (mainly feed on phytoplankton), *M. piceus* (mainly feed on spiral shells), and *C. idella* (mainly feed on aquatic plants) with the lowest proportion of 22:6n-3. However, as compared with *M. piceus*, the contents of 20:5n-3 and total n-3 LCPUFAs in *C. idella* were a little higher, which is

probably due to the higher amounts of 18:3n-3 in aquatic plants (>30%) (Table 2) and the higher ability of *C. idella* to convert 18:3n-3 into LCPUFAs (Table 3).^{33,34} It has been found that crustacean zooplankton are capable of accumulating more 20:4n-6, 20:5n-3, and 22:6n-3 than its diets (phytoplankton), which is proved again by our results that the contents of LCPUFAs in phytoplankton were lower than those in zooplankton (Table 2).³⁰

Table 3 described that considerable amounts of OBCFAs, usually originating from plankton or bacteria, were found in fish (4.78–7.63%), which was in agreement with our aforementioned studies.^{11,13} Table 1 describes the correlation of OBCFAs compositions (iso15:0, anti15:0, 15:0, iso16:0, iso17:0, anti17:0, 17:0, 19:0, 23:0, 7-methyl-6-hexadecenoic acid, 17:1n-8 and 17:1n-7) of diets against fish at detectable contents in all of the analyzed samples. The strongly highest evident relation of OBCFAs ($r = 0.742$, $p < 0.01$) was established between *M. piceus* and spiral shells. A higher correlation was reported between *C. idella* and aquatic plants for OBCFAs ($r = 0.783$, $p < 0.01$) than the other three diet species. As compared with spiral shells, aquatic plants, and zooplankton, the highest relation ($r = 0.592$, $p < 0.05$) was reported for OBCFAs between *H. molitrix*

and phytoplankton. *H. nobilis* showed an evidently positive and highest association with zooplankton for OBCFAs ($r = 0.716$, $p < 0.01$). The total OBCFAs decreased with the order of spiral shells (11.27%) > *P. communis* (10.09%) > zooplankton (8.52%) > phytoplankton (3.31%), and the dietary OBCFAs responses for total OBCFAs in selected fish were well characterized as *M. piceus* (7.65%) > *C. idella* (5.87%) > *H. nobilis* (5.25%) > *H. molitrix* (4.77%). It has been well documented that OBCFAs function as human antitumor agents, modulators of neutrophil functions, and inhibitors of counteracting the decreases in liver glycogen and serum glucose during starvation.^{35–37} Unsaturated odd-chain FAs (UOCFAs), such as 11:1, 15:1, and 17:1, ranged from 0.31–1.23% for fish to 0.21–4.98% for their diets. UOCFAs could maintain cell membrane fluidity and permeability under low temperature as an alternative to PUFAs, which are more prone to oxidation.³⁸ Thus, fish have potential for serving as one of the nutritional supplements to improve the health status of human.

FAs Distributions in TAGs and PLs of Four Wild Freshwater Fish. Figures 1–3 describe that our results are similar with previous reports,^{13,39} where the principal FAs in the TAGs and PLs fractions of *M. piceus*, *C. idella*, *H. molitrix*, and *H. nobilis* are displayed. In each species, PC had a higher level of 16:0 and lower 18:0 than PE, respectively, with TAGs in between (exception for 18:0 of *M. piceus*). The amounts of C18 unsaturated FAs mainly decreased with the order of TAGs > PC > PE, whereas 20:4n-6, 22:5n-6, 22:5n-3, and 22:6n-3 increased as TAGs < PC < PE. PC predominated in the content of 20:5n-3 (6.00–12.21%), followed in sequence by PE (2.00–8.04%) and TAGs (1.28–8.4%). 22:6n-3 in PLs (>12%) presented exclusive dominance over other PUFAs of all species.

For positional FAs distributions in TAGs of all species, 16:0, 18:2n-6, 22:5n-6, 22:5n-3, and 22:6n-3 showed a preference for the *sn*-2-position, while 18:0, 18:1n-9, 18:3n-3, 20:4n-6, and 20:5n-3 were primarily esterified in positions 1 and 3, with the exception of *C. idella* for 18:2n-6 and *H. molitrix* for 18:3n-3 (Figure 1). It is well-known that the bioavailability of FAs in the *sn*-2-position of TAGs is better than positions 1 and 3, especially for long-chain saturated FAs ($\geq C_{14}$).^{40,41}

Regarding positional FAs distributions in PC (Figure 2) and PE (Figure 3) in all of the freshwater fish, 16:0, 18:0, 18:1n-9, 18:2n-6, and 18:3n-3 were preferentially distributed in the *sn*-1-position. LCPUFAs (20:4n-6, 20:5n-3, 22:5n-6, 22:5n-3, and 22:6n-3) were mainly located in the *sn*-2-position, with the exception of 18:0 and 18:1n-9 for *M. piceus*, 18:2n-6 for *C. idella*, and 20:5n-3 for *H. nobilis*.

It has been proved that the preferential distribution of PUFAs in *sn*-2-position of PLs is linked to being protected against oxidative damage.⁴² Besides, the absorption of PUFAs from PLs could be better absorbed and metabolized in the body. As PLs are hydrolyzed to lysophospholipids and free FAs by phosphate lipase A₂, lysophospholipid could combine with free FAs and monoglycerides and then form as water-soluble mix particles to pass the unstirred water layer of small intestinal villi.²¹ Carnielli et al. reported that the absorption of 22:6n-3 from egg yolk PLs (88.3%) was superior to that from unicellular microorganisms TAGs (80.6%).⁴³

In summary, high linear correlations between dietary and studied fish FAs compositions from Poyang Lake were reported for PUFAs ($r > 0.8$) and OBCFAs ($r > 0.6$), respectively. Because FAs taken up directly from the diets could be highly retained in the fish body, food organism resources of Poyang Lake strengthen the potential for tailoring cultured fish FAs compositions to

improve their nutritional value. The study also shows that the distributions of n-3 PUFAs of fish, like 20:5n-3 and 22:6n-3, in the *sn*-2-position make fish lipids a good choice for Chinese inland residents to satisfy international recommended daily allowances (250 mg/day) of n-3 PUFAs. However, to get a better knowledge of the association between diets and fish structural FAs distributions, the stereospecific analysis of TAGs and PLs in fish diets should be further investigated.

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Notes

The authors declare no competing financial interest.

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