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Annual Evolution of Fatty Acid Profile from Muscle Lipids of the Common Carp (*Cyprinus carpio*) in Madagascar Inland Waters

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Annual evolution of muscle lipids fatty acid (FA) from common carp (*Cyprinus carpio*) has been determined in 2001 through monthly samplings in the reserve pond of Sisaony (SIS series) and Itasy Lake (ITA series) of the Madagascar highlands. Total lipids from muscle were extracted and quantified according to the Bligh and Dyer method. FA identification was performed by GC-MS of FA methyl esters and FA pyrrolidides and led to the identification of 41 FA; routine analyses of FA were made by capillary GC. Principal component analysis (PCA) was performed on the data set to compare FA profiles. Lipid content is low, ranging from 0.91 to 1.73% of wet muscle, with a low stage during the hot season (January−April) and a higher stage during the cold season (July−October). Three FA dominated the FA composition: oleic acid (17.0−21.5%), palmitic acid (13.1−16.1%), and linoleic acid (9.6−13.2%). Polyunsaturated fatty acids (PUFA) were present in appreciable amounts: arachidonic acid (AA; 2.9−5.9%), docosahexaenoic acid (DHA; 2.9−6.7%), eicosapentaenoic acid (EPA; 1.9−3.4%), and docosapentaenoic acid (DPA; 1.9−4.3%). Two opposite evolution schemes appear within two groups of FA; on the one hand PUFA (both $n-3$ and $n-6$ series) show a maximum in August−October and a minimum in January−April, and, on the other hand, oleic, palmitic, and linoleic acids show the opposite maxima and minima. PCA results give confirmation of these evolution schemes, the two groups of FA giving opposite high factor loadings on axis 1. The SIS and ITA series are differentiated by axis 2 by mean of minor FA, mostly odd- and branched-chain. Results indicate that common carp, the second most abundant freshwater fish in Madagascar highlands waters, may be an interesting source of dietary PUFA.

KEYWORDS: Carp; fatty acid; evolution; lipids; Madagascar

INTRODUCTION

Inland water fishing is one of the main sources of animal proteins in the continental highland regions of Madagascar, especially in rural areas. In the main areas of fishing, in Itasy and Alaotra Lakes a commercial structure has been developed. Fresh fish are collected for Antananarivo, the main market (1.5 million inhabitants). Official statistics (2001) state ∼240 tons of fresh fish are taken from Itasy (*1*).

The species distribution, originally characterized by a predominance of endemic species (*2*), is nowadays dominated by introduced species (*1*, *2*): tilapia, mainly *Oreochromis niloticus* Lin., fam. Cichlidae, known as Nile tilapia, local name "barahoa", introduced in 1956, representing 53% of captures in Itasy lake; common carp (*Cyprinus carpio* Lin., fam. Cyprinidae, introduced in 1914), representing 32% of captures in Itasy lake; and goldfish (*Carassius auratus* Lin., fam. Cyprinidae, introduced in 1861 for ornamental purposes), representing 14% of captures in Itasy lake.

The occurrence of $n-3$ polyunsaturated fatty acids ($n-3$ PUFA) in the diet has been recognized as having important beneficial properties for the prevention of coronary heart disease (*3*). On the other hand, some researchers have shown that freshwater fish generally contain lower proportions $n-3$ PUFA than marine fish (*4*, *5*). Furthermore, because fish need PUFAs to provide tolerance to low water temperature (*6*), low amounts

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Table 1. Muscle Lipid Content of Carp (*C. carpio*) from Sisaony Station (SIS) and Itasy Lake (ITA)[a]

		Jan	Feb	March	April	May	June	July	Aug	Sept	Oct	Nov	Dec
SIS	mean[b]	1.05	0.98[c]	–[d]	0.93	1.24	1.38[e]	1.59	1.69	1.73	1.57	1.23	1.13
	min	1.01	0.91	–	0.88	1.11	–	1.44	1.64	1.62	1.47	1.18	1.12
	max	1.07	1.05	–	0.98	1.35	–	1.74	1.74	1.84	1.68	1.27	1.23
ITA	mean	0.97	0.95	0.98	0.91	1.19	1.15	1.42	1.56	1.65	1.60	–	–
	min	0.89	0.92	0.78	0.88	1.13	1.03	1.35	1.43	1.50	1.49	–	–
	max	1.05	0.98	1.16	0.94	1.25	1.27	1.50	1.69	1.80	1.71	–	–

[a] Data expressed as percent by weight of fresh muscle. [b] Mean of three fish. [c] Mean of two fish. [d] No collection. [e] Only one fish.

Table 2. Fatty Acid Composition[a] of Total Lipids of Muscle of Carp (*C. carpio*) from Sisaony Station (SIS)

fatty acid[b]	Jan		Feb		April		May		June	July		Aug		Sept		Oct		Nov		Dec	
	mean	SD[c]	mean	SD	mean	SD	mean	SD	mean	mean	SD	mean	SD	mean	SD	mean	SD	mean	SD	mean	SD
12:0	0.43	0.17	0.34	0.11	0.36	0.16	0.45	0.21	0.35	0.37	0.13	0.23	0.05	0.36	0.12	0.45	0.19	0.32	0.21	0.41	0.15
13:0	0.23	0.12	0.18	0.05	0.24	0.09	0.17	0.08	0.20	0.11	0.05	0.16	0.03	0.10	0.04	0.22	0.10	0.13	0.05	0.16	0.10
i-14:0	0.18	0.07	0.20	0.06	0.29	0.11	0.23	0.12	0.19	0.10	0.03	0.22	0.08	0.16	0.07	0.20	0.11	0.19	0.04	0.29	0.07
14:0	1.58	0.32	1.62	0.25	1.72	0.18	1.74	0.23	1.45	1.56	0.19	1.26	0.31	1.62	0.18	1.50	0.24	1.43	0.37	1.68	0.46
14:1*n*–7	0.30	0.11	0.24	0.09	0.30	0.05	0.25	0.08	0.22	0.28	0.11	0.21	0.11	0.18	0.11	0.25	0.09	0.36	0.11	0.12	0.05
i-15:0	0.62	0.19	0.73	0.12	0.56	0.21	0.66	0.29	0.70	0.56	0.34	0.78	0.24	0.48	0.17	0.57	0.13	0.75	0.21	0.81	0.18
ai-15:0	0.51	0.21	0.41	0.14	0.33	0.04	0.51	0.16	0.54	0.35	0.13	0.47	0.11	0.41	0.13	0.38	0.11	0.50	0.16	0.62	0.21
15:0	1.75	0.40	1.53	0.21	1.29	0.16	1.86	0.47	1.54	1.04	0.11	1.41	0.13	1.59	0.12	1.81	0.24	1.21	0.35	1.53	0.41
15:1*n*–8	0.16	0.09	0.20	0.05	0.30	0.08	0.18	0.03	0.25	0.11	0.05	0.28	0.09	0.22	0.05	0.19	0.03	0.19	0.05	0.27	0.07
i-16:0	0.23	0.06	0.31	0.16	0.39	0.15	0.28	0.12	0.34	0.23	0.09	0.37	0.15	0.31	0.17	0.24	0.11	0.31	0.12	0.39	0.14
ai-16:0	0.28	0.10	0.43	0.21	0.63	0.13	0.40	0.16	0.53	0.45	0.13	0.42	0.21	0.55	0.32	0.47	0.12	0.43	0.17	0.63	0.21
16:0	15.2	0.86	15.8	0.94	16.1	0.72	16.0	0.88	15.5	13.9	1.12	13.5	0.45	14.1	0.96	13.2	0.84	14.7	0.85	15.8	1.06
16:1*n*–7	4.54	0.69	4.82	0.38	5.37	0.84	4.36	0.65	6.07	5.42	0.34	6.01	0.60	5.35	0.34	4.02	0.29	4.77	0.48	4.91	0.33
i-17:0	1.10	0.21	0.93	0.31	0.85	0.18	1.20	0.24	0.96	0.80	0.17	1.01	0.12	1.07	0.15	0.91	0.16	0.88	0.24	1.20	0.15
ai-17:0	0.47	0.15	0.52	0.13	0.42	0.10	0.50	0.09	0.42	0.47	0.11	0.37	0.16	0.49	0.21	0.49	0.07	0.33	0.11	0.57	0.13
16:2*n*–4	0.14	0.08	0.15	0.04	0.10	0.06	0.13	0.04	0.10	0.18	0.02	0.15	0.09	0.17	0.06	0.23	0.10	0.19	0.07	0.21	0.09
17:0	2.96	0.47	2.44	0.23	2.56	0.25	1.96	0.12	1.57	2.19	0.21	2.41	0.33	2.15	0.44	2.61	0.33	1.92	0.15	2.22	0.34
17:1*n*–8	2.19	0.29	1.76	0.18	1.81	0.16	1.35	0.24	1.15	1.77	0.18	1.87	0.26	1.60	0.31	2.05	0.16	1.37	0.24	1.89	0.15
i-18:0	1.07	0.18	1.13	0.24	0.84	0.20	0.97	0.16	1.05	1.10	0.21	0.86	0.16	1.01	0.23	0.82	0.11	0.81	0.13	1.01	0.21
ai-18:0	0.57	0.13	0.64	0.10	0.48	0.16	0.56	0.18	0.63	0.52	0.14	0.45	0.11	0.58	0.13	0.58	0.19	0.50	0.21	0.53	0.06
18:0	5.59	0.27	5.71	0.36	4.99	0.56	4.74	0.51	5.83	5.23	0.37	4.86	0.44	4.11	0.36	4.80	0.47	6.30	0.35	5.83	0.41
18:1*n*–9	20.4	1.12	21.2	0.55	19.0	1.26	20.3	0.47	19.4	17.9	0.63	17.0	0.81	18.0	0.69	18.1	1.08	20.8	0.47	20.3	0.86
18:1*n*–7	4.99	0.48	4.62	0.38	4.32	0.54	4.28	0.50	3.76	4.48	0.29	4.96	0.69	4.30	0.24	3.43	0.66	3.96	0.34	4.60	0.30
18:2*n*–6	12.8	1.25	12.9	2.18	12.9	1.81	11.9	2.31	11.3	11.7	2.41	10.7	1.82	9.6	0.98	10.3	1.16	11.7	1.85	12.6	1.27
18:2*n*–4	0.34	0.09	0.34	0.17	0.24	0.05	0.31	0.11	0.28	0.18	0.05	0.33	0.11	0.25	0.06	0.25	0.11	0.33	0.15	0.38	0.18
18:3*n*–6	0.61	0.11	0.67	0.20	0.72	0.16	0.64	0.23	0.59	0.69	0.15	0.67	0.26	0.59	0.23	0.57	0.17	0.67	0.24	0.72	0.20
19:1*n*–10	0.47	0.26	0.38	0.16	0.26	0.11	0.42	0.15	0.35	0.41	0.11	0.22	0.04	0.40	0.16	0.37	0.15	0.29	0.05	0.37	0.12
18:3*n*–3	1.81	0.17	1.73	0.36	2.19	0.41	1.85	0.30	1.65	2.28	0.33	2.02	0.13	1.85	0.34	2.25	0.21	1.72	0.26	1.89	0.37
18:4*n*–3	0.78	0.23	0.66	0.15	0.81	0.23	0.75	0.14	0.61	0.81	0.17	0.62	0.24	0.66	0.16	0.59	0.11	0.78	0.13	0.71	0.16
20:0	0.14	0.06	0.19	0.11	0.21	0.10	0.15	0.05	0.17	0.26	0.10	0.20	0.03	0.15	0.06	0.24	0.03	0.20	0.07	0.17	0.02
20:1	2.49	0.41	1.85	0.26	2.06	0.33	2.47	0.41	2.05	2.22	0.25	1.60	0.42	1.95	0.36	1.81	0.16	1.69	0.24	2.29	0.11
20:2*n*–6	0.60	0.15	0.73	0.20	0.55	0.21	0.61	0.18	0.54	0.45	0.13	0.62	0.17	0.50	0.14	0.57	0.21	0.71	0.17	0.59	0.09
20:3*n*–6	0.68	0.12	0.68	0.16	0.52	0.14	0.66	0.11	0.62	0.47	0.20	0.66	0.10	0.63	0.27	0.71	0.34	0.62	0.25	0.60	0.13
20:4*n*–6	3.30	0.42	3.37	0.84	3.69	0.56	4.26	0.33	4.72	5.21	0.43	5.24	0.23	5.54	0.36	5.58	0.19	4.47	0.27	3.58	0.47
20:3*n*–3	0.42	0.13	0.39	0.11	0.26	0.09	0.40	0.15	0.33	0.35	0.05	0.23	0.05	0.44	0.12	0.43	0.09	0.38	0.10	0.28	0.11
20:4*n*–3	0.28	0.09	0.29	0.03	0.17	0.04	0.25	0.10	0.29	0.16	0.08	0.26	0.04	0.32	0.09	0.39	0.13	0.33	0.11	0.27	0.09
20:5*n*–3	2.21	0.26	2.13	0.32	2.31	0.41	2.20	0.24	2.55	2.71	0.19	2.57	0.21	2.77	0.17	2.84	0.35	2.54	0.23	2.25	0.18
22:4*n*–6	1.17	0.30	1.23	0.19	1.51	0.27	1.78	0.17	1.66	2.14	0.24	2.67	0.44	2.19	0.34	2.35	0.23	1.52	0.18	1.43	0.22
22:5*n*–6	1.27	0.24	1.39	0.30	1.81	0.34	2.02	0.36	2.21	2.16	0.16	2.48	0.38	2.39	0.41	2.64	0.40	2.13	0.28	1.84	0.36
22:5*n*–3	1.76	0.27	1.82	0.21	2.09	0.19	2.18	0.31	2.30	2.80	0.18	3.34	0.17	4.02	0.27	3.82	0.18	2.56	0.38	2.04	0.19
22:6*n*–3	3.23	0.31	3.15	0.47	4.17	0.36	4.15	0.44	5.08	5.79	0.61	6.21	0.31	6.69	0.39	6.66	0.47	5.49	0.41	3.88	0.17

[a] For number of fish sampled, see **Table 1**. [b] Percent (w/w) of total fatty acids, determined as FAME by GLC on a Carbowax-20M column. [c] Standard deviation.

should be expected in warmer waters such as in tropical areas such as Madagascar. The consumption of freshwater fish contributes thus significantly to the amount of $n-3$ PUFA in the diet of the highlands population, if we suppose that highland lakes in tropical areas are less warm [the mean water temperature of Itasy lake (*2*) ranges from 16.2 to 24.4 °C].

As stated formerly, common carp represents the second most abundant freshwater fish in captures in Madagascar inland waters and has been strongly encouraged as an aquaculture species by official authorities. Many studies have been conducted on common carp, which is a well-known species throughout the world (*7*), in particular, on its fatty acid (FA) profile. These studies focused mainly on biochemical changes in lipids in response to environmental factors, such as temperature (*8*−*11*) or pollutants (*12*), diseases (*13*), or diverse feed supplementations, peculiarly dietary lipids (*14*, *15*). Some other studies focused on the FA profile on temperate water carp (*16*) or, rarely, tropical water carp (*5*); however, to our knowledge, there has been no study carried out on the influence of season on the FA profile of carp in tropical waters. In this study, we

Table 3. Fatty Acid Composition of Total Muscle Lipids of Carp (*C. carpio*) from Itasy Lake (ITA)

fatty acid[a]	Jan		Feb		March		April		May		June		July		Aug		Sept		Oct	
	mean	SD[b]	mean	SD	mean	SD	mean	SD	mean	SD	mean	SD	mean	SD	kmean	SD	mean	SD	mean	SD
12:0	0.38	0.12	0.30	0.13	0.39	0.21	0.41	0.18	0.32	0.05	0.29	0.09	0.32	0.11	0.41	0.06	0.35	0.13	0.30	0.12
13:0	0.27	0.03	0.19	0.09	0.15	0.05	0.19	0.02	0.22	0.06	0.18	0.04	0.21	0.08	0.25	0.09	0.16	0.03	0.20	0.07
i-14:0	0.18	0.02	0.22	0.06	0.16	0.02	0.18	0.04	0.23	0.03	0.27	0.07	0.17	0.01	0.21	0.09	0.18	0.06	0.16	0.02
14:0	1.63	0.23	1.71	0.19	1.40	0.36	1.71	0.38	1.51	0.19	1.67	0.26	1.81	0.32	1.74	0.26	1.63	0.19	1.51	0.25
14:1*n*–7	0.35	0.10	0.20	0.07	0.26	0.03	0.30	0.06	0.24	0.02	0.32	0.10	0.28	0.06	0.26	0.01	0.32	0.05	0.26	0.03
i-15:0	0.59	0.14	0.67	0.17	0.74	0.20	0.62	0.15	0.66	0.28	0.71	0.17	0.78	0.15	0.73	0.23	0.59	0.12	0.62	0.17
ai-15:0	0.30	0.06	0.34	0.11	0.24	0.03	0.34	0.05	0.30	0.02	0.22	0.03	0.35	0.10	0.28	0.06	0.30	0.05	0.23	0.03
15:0	1.73	0.22	1.51	0.17	1.54	0.11	1.65	0.14	1.35	0.09	1.53	0.18	1.56	0.13	1.49	0.15	1.52	0.10	1.38	0.09
15:1*n*–8	0.26	0.03	0.32	0.05	0.19	0.04	0.23	0.01	0.20	0.04	0.29	0.05	0.30	0.02	0.26	0.02	0.18	0.02	0.29	0.03
i-16:0	0.34	0.06	0.42	0.08	0.46	0.05	0.38	0.03	0.40	0.05	0.44	0.10	0.46	0.06	0.43	0.03	0.33	0.06	0.37	0.07
ai-16:0	0.62	0.12	0.69	0.13	0.53	0.10	0.71	0.09	0.68	0.14	0.61	0.07	0.77	0.05	0.61	0.10	0.65	0.03	0.54	0.06
16:0	16.1	0.86	15.9	1.18	16.0	0.80	16.4	0.92	15.9	1.03	15.4	0.75	14.1	0.63	13.9	0.67	13.1	0.82	14.1	0.78
16:1*n*–7	5.14	0.36	4.76	0.58	5.27	0.24	5.40	0.62	6.18	0.56	5.32	0.19	5.42	0.60	5.02	0.30	4.55	0.39	4.64	0.42
i-17:0	0.91	0.10	0.99	0.08	1.12	0.07	0.88	0.11	0.92	0.18	0.87	0.06	0.96	0.12	0.82	0.16	0.75	0.13	0.80	0.14
ai-17:0	0.35	0.07	0.48	0.06	0.30	0.12	0.48	0.09	0.37	0.11	0.28	0.07	0.47	0.05	0.32	0.10	0.42	0.08	0.28	0.05
16:2*n*–4	0.19	0.03	0.23	0.06	0.19	0.05	0.24	0.08	0.20	0.03	0.18	0.01	0.21	0.05	0.26	0.05	0.16	0.03	0.20	0.05
17:0	2.74	0.22	2.81	0.18	2.34	0.23	2.40	0.26	2.87	0.34	2.71	0.10	2.38	0.21	2.21	0.15	2.48	0.24	2.25	0.18
17:1*n*–8	2.00	0.12	1.51	0.22	1.80	0.20	1.65	0.10	2.18	0.29	1.87	0.18	1.60	0.13	1.42	0.10	1.55	0.13	1.61	0.16
i-18:0	1.18	0.20	1.07	0.10	1.14	0.08	0.99	0.11	0.88	0.10	0.97	0.05	1.05	0.06	0.96	0.03	0.85	0.12	0.87	0.17
ai-18:0	0.54	0.10	0.46	0.09	0.57	0.11	0.39	0.03	0.32	0.06	0.36	0.10	0.49	0.10	0.31	0.05	0.31	0.04	0.34	0.07
18:0	5.35	0.38	5.28	0.42	5.38	0.30	5.56	0.26	4.97	0.30	5.32	0.31	5.02	0.21	4.61	0.22	4.83	0.18	5.23	0.56
18:1*n*–9	20.9	0.67	21.2	0.89	21.5	0.75	20.9	1.02	20.6	0.78	19.3	0.91	19.1	0.66	18.2	0.83	16.9	0.76	17.1	0.80
18:1*n*–7	4.29	0.23	4.45	0.47	3.86	0.33	3.93	0.33	4.26	0.19	4.61	0.39	4.01	0.44	3.99	0.18	4.32	0.42	4.14	0.50
18:2*n*–6	12.9	1.22	13.0	1.47	13.2	1.03	13.0	2.06	12.2	1.76	11.8	1.50	11.2	2.11	11.3	1.73	10.3	0.85	10.6	1.16
18:2*n*–4	0.23	0.04	0.20	0.07	0.24	0.02	0.20	0.06	0.16	0.03	0.18	0.05	0.25	0.11	0.19	0.10	0.23	0.10	0.17	0.03
18:3*n*–6	0.84	0.10	0.74	0.12	0.66	0.07	0.82	0.21	0.81	0.16	0.94	0.13	0.94	0.02	0.87	0.14	0.90	0.15	0.86	0.11
19:1*n*–10	0.33	0.03	0.29	0.10	0.33	0.09	0.25	0.11	0.27	0.09	0.37	0.14	0.21	0.07	0.36	0.15	0.25	0.03	0.28	0.06
18:3*n*–3	1.61	0.36	1.86	0.23	2.13	0.16	1.89	0.34	1.62	0.28	1.84	0.42	2.39	0.33	2.05	0.28	1.97	0.41	2.07	0.09
18:4*n*–3	0.70	0.18	0.76	0.22	0.70	0.10	0.70	0.09	0.60	0.16	0.69	0.12	0.81	0.21	0.77	0.17	0.70	0.16	0.82	0.11
20:0	0.27	0.06	0.20	0.05	0.27	0.10	0.20	0.03	0.26	0.07	0.32	0.12	0.22	0.06	0.26	0.07	0.18	0.03	0.16	0.06
20:1	2.27	0.06	1.96	0.32	1.81	0.44	2.10	0.28	1.72	0.19	1.67	0.16	1.96	0.10	1.61	0.12	1.60	0.09	1.69	0.26
20:2*n*–6	0.55	0.09	0.64	0.18	0.57	0.13	0.59	0.16	0.54	0.10	0.56	0.15	0.60	0.09	0.48	0.12	0.55	0.08	0.62	0.10
20:3*n*–6	0.68	0.13	0.68	0.14	0.72	0.10	0.62	0.17	0.71	0.15	0.85	0.09	0.77	0.12	0.67	0.21	0.58	0.14	0.68	0.11
20:4*n*–6	3.21	0.42	2.90	0.33	2.94	0.39	2.75	0.18	3.34	0.31	4.09	0.30	4.91	0.28	5.63	0.50	5.94	0.46	5.62	0.38
20:3*n*–3	0.50	0.10	0.54	0.17	0.42	0.12	0.49	0.06	0.45	0.05	0.59	0.15	0.42	0.11	0.51	0.13	0.40	0.13	0.57	0.10
20:4*n*–3	0.32	0.06	0.34	0.04	0.43	0.07	0.32	0.02	0.38	0.05	0.34	0.10	0.27	0.03	0.30	0.06	0.36	0.08	0.30	0.04
20:5*n*–3	2.14	0.22	2.18	0.17	1.93	0.23	2.31	0.18	2.53	0.15	2.46	0.33	2.73	0.27	3.07	0.18	3.43	0.15	3.06	0.25
22:4*n*–6	1.06	0.20	1.24	0.19	1.42	0.31	1.33	0.42	1.62	0.22	1.94	0.18	2.03	0.06	2.36	0.18	2.52	0.13	2.49	0.15
22:5*n*–6	1.18	0.09	1.45	0.12	1.22	0.06	1.52	0.27	1.65	0.18	2.15	0.15	2.35	0.12	2.41	0.11	2.74	0.10	2.56	0.17
22:5*n*–3	1.62	0.25	2.07	0.14	2.33	0.18	2.14	0.33	2.03	0.17	2.35	0.20	2.79	0.16	3.89	0.14	4.28	0.18	3.95	0.15
22:6*n*–3	3.32	0.28	3.31	0.25	3.14	0.38	2.86	0.42	3.37	0.18	3.17	0.39	3.50	0.30	4.30	0.18	6.53	0.67	6.21	0.42

[a] Percent (w/w) of total fatty acids, determined as FAME by GLC on a Carbowax-20M column. [b] Standard deviation.

Table 4. Range of Sums of *n*–3 and *n*–6 PUFA and *n*–3/*n*–6 Ratios in Total Muscle Lipids of Carp (*C. carpio*) from Sisaony Station (SIS) and Itasy Lake (ITA)

		Jan	Feb	March	April	May	June	July	Aug	Sept	Oct	Nov	Dec
SIS	Σ *n*–6[a]	20.44	20.98	–	21.73	21.82	21.56	22.80	23.03	21.40	22.71	21.81	21.34
	Σ *n*–3[a]	10.49	10.17	–	11.19	11.78	12.81	14.90	15.25	16.75	16.98	13.80	11.32
	n–3/*n*–6	0.51	0.48	–	0.51	0.54	0.59	0.65	0.66	0.78	0.75	0.63	0.53
ITA	Σ *n*–6[a]	20.38	20.60	20.72	20.60	20.82	22.30	22.82	23.69	23.57	23.39	–	–
	Σ *n*–3[a]	10.21	11.06	11.08	10.71	11.25	11.44	12.91	14.89	17.67	16.98	–	–
	n–3/*n*–6	0.50	0.50	0.53	0.52	0.54	0.51	0.57	0.63	0.75	0.73	–	–

[a] Percent (w /w) of total fatty acids, determined as FAME by GLC.

intend to examine the variations of fatty acid profile through monthly sampling on carp, one of the main freshwater fish species of the Madagascar highlands.

MATERIALS AND METHODS

Sampling. Fish samples were collected monthly from January to December 2001, in two series: the first lot (code ITA) was purchased directly from fish collectors and/or fishermen the day of capture at Itasy Lake, 150 km west of Antananarivo, altitude 1100 m; each monthly sample was composed of three individual fish. No fish was collected during November and December, due to closing of the fishing season. The second lot (code SIS) was caught in the reserve pond (enclosed earthen pond, without feeding) of the experimental pisciculture station of Sisaony, 20 km south of Antananarivo, altitude 1100 m; each monthly sample was also composed of three individual fish when available, due to random captures. In all cases, sampling focused on fish of the most commonly marketed size, that is, 20−30 cm in length, 150−300 g in weight (roughly 1 year old). Individual fish were dissected, and portions of muscle tissue below the dorsal fin, devoid of skin and bone, were kept in ice for <4 h before lipid extraction.

Lipid Extraction. For each analysis, ∼10 g of muscle was homogenized separately using a Warring blender. Lipids were extracted according to the Bligh and Dyer method (*17*). Phase was stored in anhydrous sodium sulfate and evaporated under nitrogen for total lipid content determination.

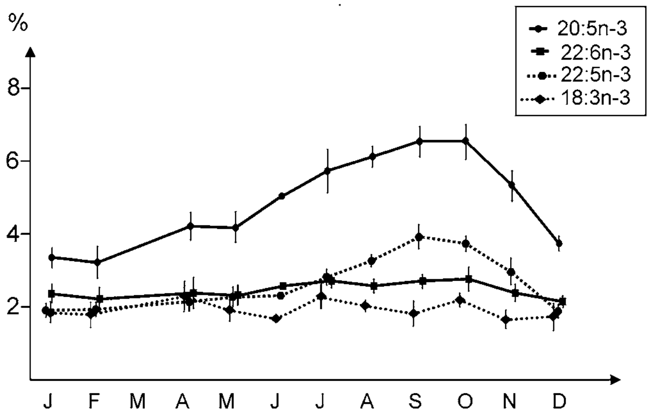

Figure 1. Range of main *n*–3 PUFA of *C. carpio* collected in Sisaony station during the year 2001.

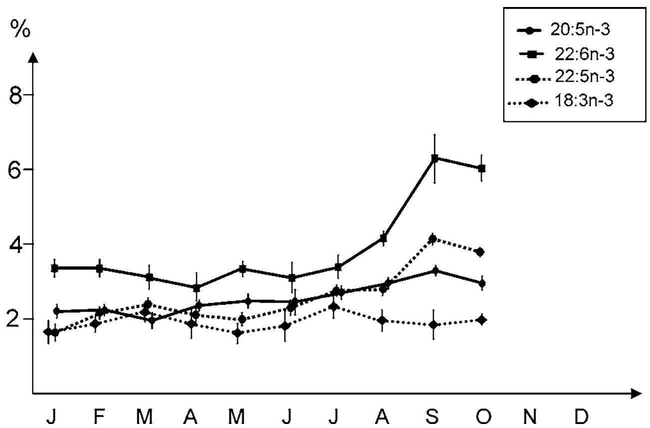

Figure 2. Range of main *n*–3 PUFA of *C. carpio* collected in Itasy Lake during the year 2001.

Gas Chromatography Analyses and Identifications of Fatty Acid Methyl Esters (FAME). FAME were prepared as described by Christie (*18*). A Delsi gas chromatograph equipped with a flame ionization detector (FID) and a fused silica capillary column (25 m long, 0.28 mm i.d.) coated with Carbowax 20M (0.2 μm phase thickness) was used for analyses. Temperatures used were from 180 °C (10 min) to 220 °C, for the column, and 250 °C, for the inlet and detector ovens, raised at 2 °C min^{-1}. Identifications of fatty acids were carried out using mass spectrometry of their FAME and their pyrrolidides and compared to previously published results (*19*, *20*). Combined GC-MS was performed on a Hewlett-Packard model 5890 gas chromatograph instrument equipped with a mass spectrometer detector Hewlett-Packard model 5989A and Hewlett-Packard 9000/345 integrator. A DB1 fused silica capillary column, 30 m × 0.32 mm (i.d.) with a 0.25 μm stationary phase film was used from 170 °C (4 min hold) to 300 °C (3 °C min^{-1}) for FAME and from 200 °C (4 min hold) to 310 °C (3 °C min^{-1}) for *N*-acyl pyrrolidide derivatives. Helium was used as carrier gas, the ion source temperature was 220 °C, and the ionizing voltage was 70 eV.

Statistical Analysis. Principal component analysis (PCA) has been performed by using the data set transformed into centered and reduced variables (standardized PCA). The data set was composed by the mean value for each month for all variables: relative percentages of 41 FAME and the lipid values (%LIP). Data were processed with STAT-ITCF program version 4 (ITCF). PCA has been chosen due to its ability to compare complex sets of data such as FA profiles; moreover, PCA has been previously used successfully to discern FA patterns (*21*−*23*).

RESULTS AND DISCUSSION

Lipid Content. For the two series, lipid content (given in **Table 1**) is low, ranging from 0.93 and 0.91% for SIS and ITA in April to 1.73 and 1.65% in September, respectively. Two distinct stages in lipid content evolution appear: a first stage

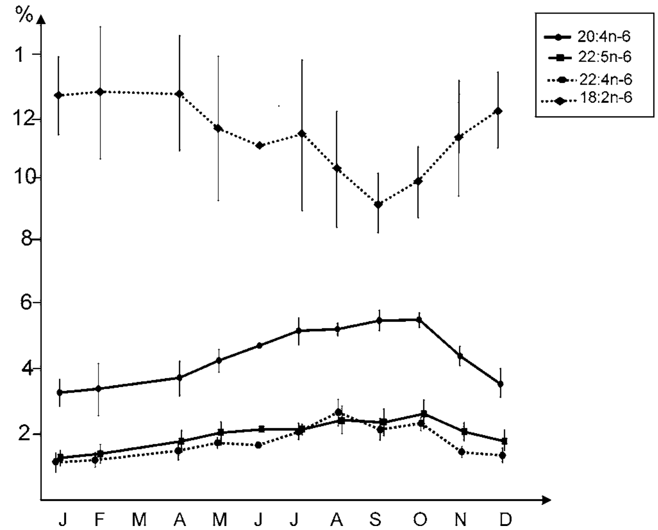

Figure 3. Range of main *n*–6 PUFA of *C. carpio* collected in Sisaony station during the year 2001.

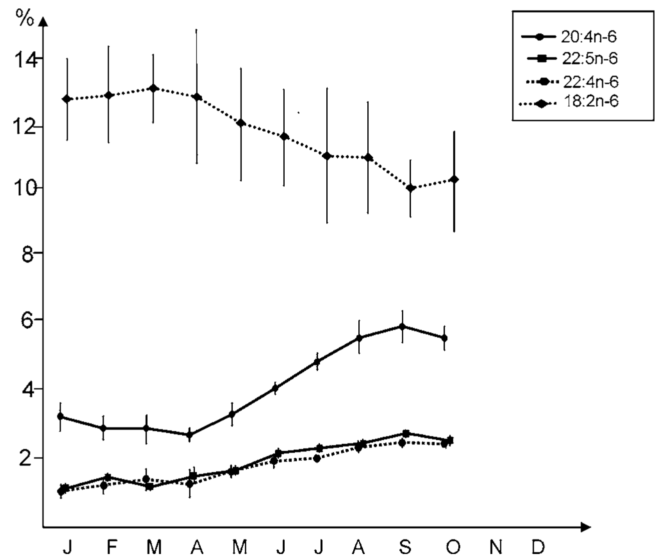

Figure 4. Range of main *n*–6 PUFA of *C. carpio* collected in Itasy Lake during the year 2001.

when lipid content is relatively low, close to 1% during the period January−April (warm season), and a higher stage when lipid content exceeds 1.4−1.5%, in July−October (cold season), for both series.

The lipid content, ranging between 0.9 and 1.7%, is relatively low, in comparison to data generally found in other freshwater fish: Rahman et al. (*5*) found 1.17−34.0% in Malaysian fish, with a value of 1.92% for common carp, whereas Aggelousis and Lazos (*16*) pointed out values of 0.6−3.5% for freshwater fish from Greece. The data indicate that common carp is a lean fish, according to Bennion (*24*). Kiener (*2*) noticed that Madagascar highlands waters, situated in geological crystalline formations, are low in minerals; thus, the food chain is relatively poor. This could explain the generally low levels of fat in carp in the waters of Madagascar. We also note that the period of lowest fat content corresponds to the spawning period for the carp, that is, the hot season (*2*); during this period, dietary and novel-synthesized lipids are mostly used for maturation of eggs, which need considerable amounts of FA (*25*).

Fatty Acid Profile. As seen in **Tables 2** and **3** and **Figures 1**−**6**, three FA dominate the profile, with generally >10% throughout the year: oleic acid (18:1n−9), with concentrations

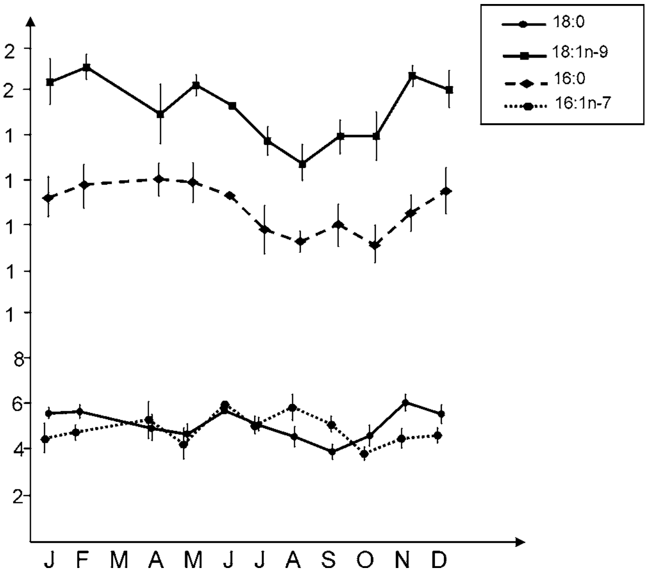

Figure 5. Range of main saturated and monounsaturated fatty acids of *C. carpio* collected in Sisaony station during the year 2001.

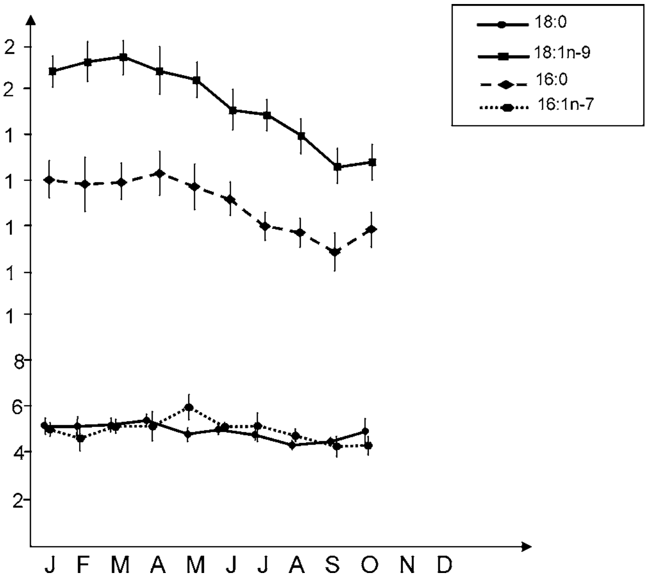

Figure 6. Range of main saturated and monounsaturated fatty acids of *C. carpio* collected in Itasy Lake during the year 2001.

ranging from 17.0 (SIS, August) to 21.5% (ITA, March); palmitic acid (16:0), at 13.1 (ITA, September) to 16.1% (SIS, April), and linoleic acid (18:2*n*−6, LA), at 9.6 (SIS, September) to 13.2% (ITA, March).

The main PUFA other than LA are arachidonic acid (20:4*n*−6, AA), at a level ranging from 2.9 (ITA, February) to 5.9% (ITA, September), docosahexaenoic acid (22:6*n*−3, DHA), ranging from 2.9 (ITA, April) to 6.7% (SIS, September), eicosapentaenoic acid (20:5*n*−3, EPA), ranging from 1.9 (ITA, March) to 3.4% (ITA, September), and docosapentaenoic acid (22:5*n*−3, DPA), ranging from 1.6 (ITA, January) to 4.3% (ITA, September). Linolenic acid (18:3*n*−3) content is comparatively low, from 1.6 (ITA, January) to 2.4% (ITA, July). The total *n*−3 PUFA (**Table 3**) ranges from 10.2 (SIS, February) to 17.7% (ITA, September) and *n*−6 PUFA (including LA) from 20.4 (ITA, January) to 23.7% (ITA, August). Other major FA are the usual FA of fish lipids such as stearic acid (18:0), with contents ranging from 4.6 to 6.3%, and palmitoleic acid (4.0−6.2%). One can note also the presence of odd-chain and/or

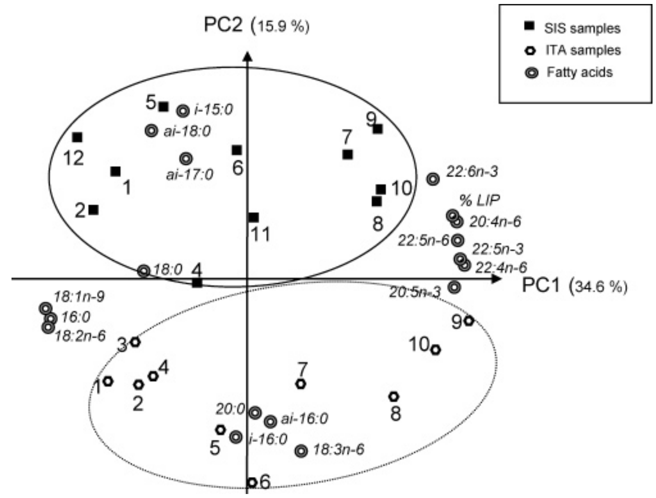

Figure 7. Two-dimensional plot of the FAME profiles of muscle from carp investigated by PCA for the year 2001. Each month is designated by its number. The amount of variance relative to each axis and the FAME providing the major contribution are given.

branched FA, in fair concentrations (near 1%). These FA have been stated as usually found in fish lipids and often attributed to planktonic or bacterial origin (*26*).

The FA profile is thus characterized by a dominance of SA and MUFA, representing ∼40% of the total FA, a high amount of linoleic acid (10−13%), and PUFA, ranging, for the *n*−6 series (LA excluded), from 10 to 13% and for the *n*−3 series from 10 to 17%. Two opposite evolutions appear within two groups of FA: on the one hand, major SA, MUFA, and LA, showing a minimum in August−October and a maximum in January−April; on the other hand, PUFA, both *n*−6 (excluding LA) and *n*−3 (excluding linolenic acid), with opposite minima and maxima. Other major FA do not seem to show a peculiar evolution scheme. As shown in **Table 4**, the *n*−3/*n*−6 ratio is lowest in the period January−April (∼0.5) and rises to 0.8 in September. The low ratio value is due to high levels of LA.

The FA profile generally conforms to those reported in the literature: high levels of SA and MUFA are regularly reported for freshwater fish, both in temperate (*16*) and in tropical waters (*5*). Chatakondi et al. (*27*) found a level of 11.5% of LA in common carp, whereas Ackman et al. (*28*) reported about 40−50% SA and 24−39% MUFA in Indian carps, with ∼5% each of LA and linolenic acid. Higher levels of *n*−6 than *n*−3 PUFA are also reported in the literature: Ackman et al. (*28*) found an AA level of 5−10%, slightly higher than in the present work. In regard to major FA, no difference appears between the two series; both have the same evolution pattern, and amounts of individual FA are always close, considering the value of standard deviation.

Statistical Analysis. Figure 7 shows a plot of observations, represented by the mean of samples, and most representative variables on the 1−2 plane. There appear two groups of variables having major contributions to axis 1 (34.6% of total information). Major PUFA and lipid contents are loaded on the positive part of axis 1 and palmitic, oleic, and linoleic acids on the negative part of this axis. All variables within each group show high correlation factors (near 0.9). Monthly plots of both series seem to develop between these two poles, corresponding to minima and maxima cited formerly: maximum PUFA and lipid contents around August−October and correlative minimum of palmitic, oleic, and linoleic acids, whereas the period January−April shows inversion. PCA confirms the evolution pattern of FA profiles. Accumulation of lipids in muscle during the cold season (July−October) is correlatively followed by an increase of PUFA levels and decreases of SA, MUFA, and LA during the hot (spawning) season. Highly negative correlations between the two groups suggest that during the cold season, the fish converts actively dietary LA to *n*−6 PUFA, probably via the well-known biosynthetic pathways (*29*). The *n*−3 pathway from linolenic acid is not clearly demonstrated, linolenic acid levels showing no neat evolution; PCA shows the tendency of linolenic acid to get close to the PUFA group.

The two series coming from Itasy Lake (ITA) and the experimental station of Sisaony (SIS) are clearly distinguished by means of axis 2 (15.9% of total information), corresponding mainly with minor FA, such as branched ones. Among them, the most representative are *i*-15:0, *ai*-18:0, and *ai*-17:0 for the positive branch, corresponding to the SIS series; and *i*-16:0 and *ai*-18:0, for the negative one (ITA series). A minor PUFA, 18:3*n*−6, seems also to characterize the ITA series. Thus, separation between the two series may reflect differences of composition of plankton in the rearing environment (*26*).

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