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Effect of aquaculture on mercury and polyunsaturated fatty acids in fishes from reservoirs in Southwest China☆

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ABSTRACT

Aquaculture can affect the polyunsaturated fatty acids (PUFA) and mercury (Hg) in fish by altering their diet. Here, planktivorous (silver carp and bighead carp), omnivorous and carnivorous fish with different dietary strategies were selected from two reservoirs, one with on-going aquaculture (WJD) and another without aquaculture (HF) in Southwest China. We compared the total mercury (THg), methylmercury (MeHg) contents and PUFA profiles of fish and their potential diets in these two reservoirs. THg and MeHg contents in omnivorous and carnivorous fish were lower from the WJD Reservoir, which is related to the lower THg and MeHg contents in the artificial fish food. THg and MeHg contents in silver carp from the WJD Reservoir were lower than those from the HF Reservoir, while they were similar in bighead carps from the two reservoirs. The Hg variation in planktivorous fish were inconsistent with that in plankton. THg contents in phyto- and zooplankton from the HF Reservoir were higher than those from the WJD Reservoir, yet their MeHg contents were similar. Artificial fish food which contained higher total PUFA eicosapentaenoic (EPA; 20:5n-3) and docosahexaenoic acid (DHA, 22:6n-3), significantly increased the total PUFA and EPA + DHA contents in carnivorous fish, but had less effect on that in omnivorous fish from the WJD Reservoir. Eutrophication caused by aquaculture reduced total PUFA and EPA + DHA contents of plankton in WJD, yet did not reduce those in planktivorous fish. The impacts of aquaculture on Hg and PUFA accumulated in fish were varied among different fish species, and the mechanism needs further exploration.

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1. Introduction

Mercury (Hg) is a globally distributed toxic pollutant. Methylmercury (MeHg) is one of the toxic Hg forms that can bioaccumulate and biomagnify in food webs (Fitzgerald et al., 2007; Feng et al., 2018; Wang and Wang, 2019; Yan et al., 2019). Inorganic mercury (IHg) can be transformed into MeHg through microbial and chemical methylation in sediments (Yan et al., 2013; Jiang et al., 2018; Liang et al., 2018), especially in eutrophic aquatic system by aquaculture (He et al., 2008; Liang et al., 2016a, 2016b, 2017). Therefore, aquaculture may affect the bioaccumulation of mercury inaquatic organisms via providing artificial feeding and altering the phytoplankton community structure by accelerating eutrophication.

As essential fatty acids (EFAs), polyunsaturated fatty acids (PUFA) are important dietary nutrients for human and animals. PUFA are precursors for important animal hormones and critical biological compounds in regulating cell membrane properties. It is demonstrated that PUFA can enhance the somatic development and reproduction of invertebrates and fish. Unlike plants, animals cannot synthesise PUFA on their own. Most PUFA in aquatic food

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chain was synthesised by phytoplankton and subsequently transferred to organisms at higher trophic levels (Arts et al., 2009; Brett and Müller-Navarra, 2010). Among the PUFA, eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) play important roles in the metabolic functioning, somatic growth and reproduction of fish (Tidwell et al., 2007; Francis et al., 2019; Gergs et al., 2014). While linoleic acid (LIN) is an essential precursor for arachidonic acid (ARA), a-linolenic acid (ALA) is a precursors for EPA and DHA (Arts et al., 2009). Mercury and PUFA can co-occur as dietary sources for fishes, and both were conveyed along aquatic food chain.

Fish is regarded as a high-quality source owing to its high contents of important longer chain (LC) omega-3 (n-3) PUFA. In particular, EPA and DHA are beneficial to neurological development and lowering indices of cardiovascular diseases (Elagizi and Lavie, 2019). Therefore, various investigations have been conducted to estimate the risks of exposure to mercury and other chemical pollutants in fish with omega-3 fatty acids associated health benefits of fish consumption and explore how conveyed along aquatic food webs (Kainz et al., 2006; Ahlgren et al., 1999; Razavi et al., 2014; Taipale et al., 2016; Laird et al., 2018).

EPA and DHA are biomarkers of diatoms and cryptophytes, which synthesise EPA and DHA among phytoplankton species (Ahlgren et al., 1992). Aquaculture and eutrophication are prevailing in China (Wang et al., 2012; He et al., 2008; Feng et al., 2018). The use of artificial fish food and aggravating eutrophication alter the natural structure of food chain, consequently influence Hg bioaccumulation and EFAs profiles in fish. On one hand, feeding with artificial fish food and algal bloom reduce Hg bioaccumulation in fish (Yan et al., 2019; Liu et al., 2012). On the other hand, the transportation and proportion of high-quality fatty acids in the food chain may change with the plankton community structure, which ultimately affects the nutritional value of fish and the ecology health of the basic food chain.

In the present study, we examined two contrasting reservoirs with different degrees of eutrophication and aquaculture intensity in Southwest China. One reservoir has ongoing aquaculture, whereas in the other, although it has a long history of aquaculture, all aquaculture activities ceased 10 years ago. We first compared the Hg content and PUFA profiles in cultured and wild omnivorous and carnivorous fish in these two different aquatic ecosystems. The diets for wild omnivorous and carnivorous fish mainly come from natural food sources, whereas the cultured omnivorous and carnivorous fish mainly feed on artificial diets. Therefore, differences in the Hg contents and PUFA profiles between wild and cultured omnivorous and carnivorous fish can be used to test the effect of artificial feeding on the accumulation of Hg and PUFA in fish. We first hypothesized that farmed fish generally contain lower Hg than wild fish, while it might also contain lower PUFA and EPA + DHA contents since nutritional quality of fatty acids of artificial fish food might be lower than natural diet. Since planktivorous fish (silver carp and bighead carp) remain planktivorous at all life stages (Cremer and Smitherman, 1980), and are therefore directly expected to reflect the Hg and PUFA accumulation from phytoplankton and zooplankton. We finally hypothesised that increased eutrophication in the ongoing aquaculture reservoir would enhance the biomass of Chlorophyta and Cyanophyta, which would decrease the Hg and EPA + DHA contents in plankton and, consequently, reduce the Hg and EPA + DHA contents transferred to planktivorous fish.

2. Sampling sites and methods

2.1. Study sites and sample collection

The study was carried out in two eutrophic reservoirs of the Wujiang River in Guizhou, Southwest China (Fig. 1). The Wujiang

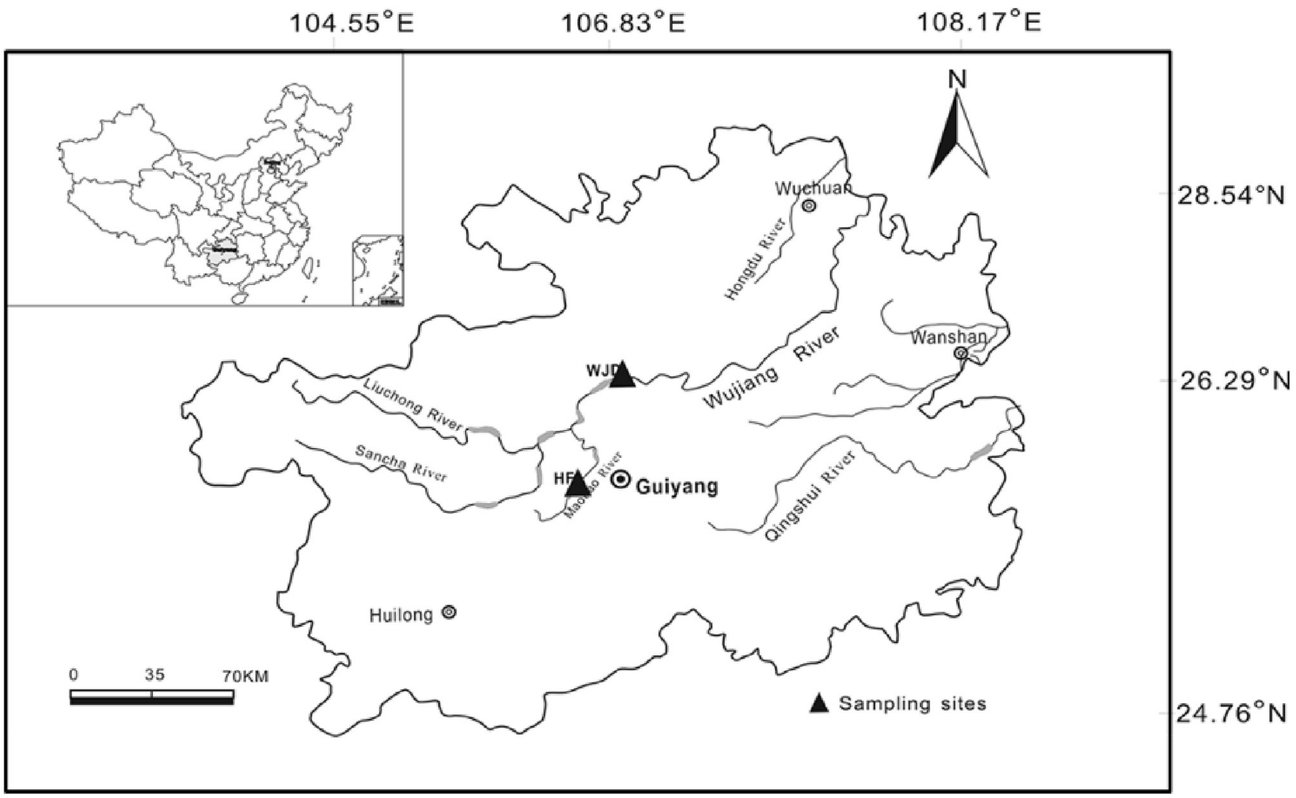

Fig. 1. Location of Wujiangdu (WJD) and Hongfeng (HF) Reservoirs in the Wujiang River Basin.

River is one of the largest tributaries of the Yangtze River. It is a subtropical climate and located in an Hg mineralization zone with high natural Hg release due to the high Hg concentration in the bedrock. Two eutrophic reservoirs with a long history of aquaculture were selected for this study: a) a reservoir with ongoing aquaculture that began in 1999, Wujiangdu Reservoir (WJD; 106°8′26″ E, 26°35′20″ N), located in the main stream of Wujiang River and characterised by a high density of net-caged fish production; and b) the Hongfeng (HF) Reservoir (106°26′16″ E, 26°29′30″ N), the upstream of WJD, located in the tributary of Wujiang River and is the drinking-water source for the city of Guiyang and no aquaculture has taken place since 2007 despite a 12-year history of aquaculture (1995–2007).

Previous studies has showed the similar background environment and aqueous Hg levels in WJD and HF Reservoir. THg and MeHg concentrations in HF ranged from 4.3 to 6.9 and 0.15–0.92 ng/L, respectively (He et al., 2008; Jing et al., 2017), and in WJD ranged from 3.0 to 18.0 and 0.1–0.97 ng/L, respectively (Feng et al., 2018; Zhao et al., 2017; Liang et al., 2017). The significant difference is whether there is on-going aquaculture activity or not. In HF Reservoir without aquaculture, the food sources for wild fish mainly consist of plankton and zoobenthos (Table S1 in supplementary information (SI)), whereas in WJD Rservoir with on-going aquaculture, the food sources for cultured fish consist of plankton and artificial fish foods (Feng et al., 2012; Zhu et al., 2017).

Samplings of the HF and WJD Reservoirs were conducted on different days in July 2016. For analysis of water quality parameters (e.g. pH, total nitrogen (TN) and total phosphorus (TP)), water samples were collected from 0.5 m, 10 m beneath the surface water and 0.5 m above bottom sediment, and dispensed into clean borosilicate glass bottles. Three replicate plankton samples of two sizes (64–112 μm and 112–500 μm) in the two reservoirs were collected vertically from the open water using 64-μm and 112-μm plankton nets. Plankton samples were transferred to polypropylene vials and placed in liquid nitrogen immediately. Zoobenthos and wild fish were only collected in the HF Reservoir. Zoobenthos were collected from littoral sites of the HF Reservoir and hand-picked by screening the sediment through a 500-μm sieve. After being rinsed with pure water, the zoobenthos samples were kept cold in a cooling box and sorted lively in the laboratory by taxa within 24 h of collection. Zoobenthos were not collected from the WJD Reservoir because the cultured fish live in net cages, and thus had no dietary access to the zoobenthos. Artificial fish food and cultured fish were collected from the WJD Reservoir.

Although we targeted the same fish species in the HF and WJD Reservoirs, some cultured fish species were not available in the WJD Reservoir due to the species preference for human consumption. Therefore, we strived to collect fish with the same feeding habits in the HF and WJD Reservoirs. Wild fishes in the HF Reservoir were caught by net fishing or by angling with the help of local fishermen. The collected wild fish consisted of planktivorous fish (silver carp (*Hypophthalmichthys molitrix*) and bighead carp (*Aristichthys nobilis*)), omnivorous fish (crucian carp, rudd (*Scardinius erythrophthalmus*) and sharp belly (*Hemiculter leucisculus*)) and carnivorous fish (*Abbottina rivularis*). The cultured fish in the WJD Reservoir are fed in submerged cages. Cultured fish and artificial fish food were purchased from local fishermen. The sampled cultured fish species consisted of planktivorous fish (silver carp and bighead carp), omnivorous fish (common (*Cyprinus carpio*) and crucian (*Carassius auratus*) carps) and carnivorous fish (catfish (*Silurus asotus*)). The quantity of artificial fish food applied per cultured omnivorous fish was 120–180 g/day while the quantity of artificial fish food applied per cultured carnivorous fish was 80–130 g/day. Three silver carps, four bighead carps, eight omnivorous fish and three carnivorous fish were collected from the HF Reservoir; seven silver carps, eight bighead carps, six omnivorous fish and six carnivorous fish were collected from the WJD Reservoir. Both cultured and wild fish are all 1–2 years old. All the fish samples were transported to the laboratory on the same day. Only dorsal muscle samples of the fish were used for total mercury (THg), MeHg and fatty acids analysis. All samples were kept frozen at −80 °C until lyophilisation and then stored at −80 °C until analysis.

2.2. Sample analysis

The plankton classification was conducted by the Institute of Hydrobiology, Jinan University (Guangzhou, China). Plankton identification was carried out using a stereomicroscope (Olympus SZX16, Olympus, Japan) and a regular compound microscope (Olympus BX51, Olympus, Japan).

Lipid and fatty acid analyses were conducted at the Inter-University Center for Ecosystem Research, WasserCluster Lunz (Lunz am See, Austria). Lipids of homogenised and freeze-dried samples of plankton, zoobenthos and dorsal fish muscle tissues were extracted in a chloroform–methanol mixture (2:1 v/v) as described elsewhere (Heissenberger et al., 2010). Fatty acids (FA) were trans-esterified to fatty acid methyl esters (FAME) by transferring the lipid extract into H_2SO_4-methanol and incubating at 50 °C for 16 h followed by gas chromatography analysis (Thermo Scientific TRACE) using a 2560 capillary column (100 m, 0.25-mm i.d., 0.2-μm film thickness; Supelco, Sigma-Aldrich, Bellefonte, PA, USA). Helium was used as the carrier gas (flow rate: 1 mL/min). The following temperature ramp was employed: 65 °C for 0.5 min, hold at 195 °C for 15 min after ramping at 40 °C/min, and hold at 240 °C for 10 min after ramping at 2 °C/min. Detection was by flame ionisation (FID). Helium (make-up gas) and air (combustion) had flow rates of 30 and 300 mL/min, respectively. The FID was isothermal at 260 °C, whereas the injector was programmed to increase to 250 °C at a rate of 200 °C/min after holding at 150 °C for 0.5 min. FAME were identified by comparison of retention times with known standards (37-component FAME mix, Supelco 47885-U; bacterial fatty acids 47080-U and the following individual FAME standards: stearidonic acid, O5130 SUFMA, n-3 decosapentaenoic acid, Supelco 47563-U). Quantification of individual FAME components was calculated based on known amounts of injected standard dilutions (2000, 1000, 500, 250, 100, 50 and 2.5 ng/μL). The FA mass fractions are reported as mg FA per g dry weight.

For THg analysis of biota samples, 0.1–0.2 g of freeze-dried samples were digested in HNO_3:H_2SO_4 (7:3 v/v) at 95 °C for 3 h before being measured using gas chromatography (GC)/cold vapour atomic fluorescence spectrometer (CVAFS) (Brooks Rand Model III, Seattle, USA). For MeHg analysis of biota samples, 0.1–0.2 g freeze-dried samples were digested in 5 mL of 25% KOH solution and heated for 3 h at 75–80 °C. The digestion was diluted with Milli-Q water (18.2 MΩ/cm resistivity; Millipore) to a certain volume prior to analysis (Yan et al., 2006). MeHg was separated by GC and then quantified by CVAFS. The quality control consisted of using duplicates, method blanks and certified reference materials (CRMs). Blank spikes and duplicates were taken regularly (>10% of samples). Tort-2 from the National Research Council of Canada was used as the CRM. Recovery of THg and MeHg in the CRM was 106 ± 1.9% and 101 ± 2.0%, respectively. The concentrations of THg and MeHg are reported as dry weight.

For water chemistry analyses, three replicates were collected from each water sampling site from the two reservoirs. Water temperature (T), dissolved oxygen (DO) and pH were measured onsite with a water quality meter (SX751, Sanxi, China). Water transparency was estimated with a Secchi disk (XTY6103481, Xuantaiyi, China). Dissolved organic carbon (DOC) was measured as total organic carbon (TOC) with a TOC analyser (Elementar, High TOC 2, Germany). TN, TP and chlorophyll-a (Chl-a) were determined according to methods described previously (Li and Han, 2007). All the analyses of these parameters were performed following the procedures outlined in the Chinese Standard Methods for Water Quality Analysis (GB 3838-2002).

2.3. Statistical analysis

Eutrophication was assessed by the concentrations of TP, TN, Chl-a and Secchi disk depth in the water column. The modified Carlson trophic level index (Carlson, 1977) (TLI) was used to categorise water bodies as eutrophic ($TLI > 50$), mesotrophic (TLI: 30–50) or oligotrophic ($TLI < 30$). The TLI calculation followed the methods in Wang et al. (2002).

To compare Hg and PUFA contents of the food-web in the HF and WJD reservoirs, the non-parametric Kruskal–Wallis test was used. Differences in the length and weight of fish were tested using analysis of variance. Hg content in wet weight was calculated to dry weight based on moisture content, when necessary to compare with other reported results. All data analyses were performed using IBM SPSS statistics 20 and Origin 9.0.

3. Results and discussion

3.1. Physical and chemical characteristics of water in the reservoirs

The average values of pH, temperature, DOC and DO of water were similar in the two reservoirs (Table 1). The pH values of the two reservoirs (pH 8.0 ± 0.5 for HF and 8.3 ± 0.1 for WJD) indicated neutral and slightly alkaline environments and the water temperatures was ~25 °C. They were both normoxic (DO > 2 mg/L). The nutrient levels were higher in the WJD than HF Reservoir (Table 1).

Table 1
Physical and chemical characteristics of water in Hongfeng (HF) and Wujiangdu (WJD) Reservoirs.

Variables	HF	WJD
Altitude (m)	1240	760
Volume (10^9 m^3)	0.60	2.30
Water retention time (days)	119	90
Secchi disk depth (SD, m)	2.7 ± 0.3	1.5 ± 0.2
pH value	8.0 ± 0.5	8.3 ± 0.1
Temperature (T °C)	24 ± 0.7	26 ± 0.8
Dissolved organic carbon (DOC mg/L)	2.5 ± 0.1	2.6 ± 0.1
Dissolved oxygen (DO mg/L)	5.5 ± 0.2	4.2 ± 0.2
Total nitrogen (TN mg/l)	1.3 ± 0.0	4.4 ± 0.1
Total phosphorous (TP mg/L)	0.05 ± 0.01	0.06 ± 0.01
Chlorophyll-a (Chl-a μg/L)	3.6 ± 0.1	10 ± 9.3*
Trophic level index (TLI)	Mesoeutrophic (46)	Eutrophic (55)
THg_{water} (ng/L)	4.3 ± 0.5	1.4 ± 0.1*
$MeHg_{water}$ (ng/L)	0.2 ± 0.1^a	0.3 ± 0.4^a

Note: "a" reported in Feng et al. (2018).
Values are means ± standard deviation, "*" means level of significance< 0.05.

3.2. Hg and PUFA in fish diets

3.2.1. Hg and PUFA contents in fish diets

In the HF Reservoir, the diet of the omnivorous and carnivorous fish consisted of phytoplankton, zooplankton and zoobenthos. In the WJD Reservoir, the omnivorous and carnivorous fish mainly fed on artificial fish food composed of corn, soybean and marine fish. Silver carp and bighead carp are planktivorous, and silver carp mainly fed on phytoplankton and bighead carp mainly fed on zooplankton instead of artificial fish food in both the HF and WJD Reservoirs (Table S1 in SI).

The THg and MeHg contents in the fish dietary sources from the two reservoirs are shown in Table 2. The THg and MeHg contents of the artificial fish food in the WJD Reservoir were significantly lower than those of the phyto- and zooplankton in both reservoirs and the zoobenthos in the HF Reservoir ($p < 0.05$), which may reduce the dietary Hg transferred to the cultured omnivorous and carnivorous fish in the WJD Reservoir. The THg contents of phytoplankton and zooplankton in the HF Reservoir were higher than those in the WJD Reservoir ($p < 0.05$), whereas their MeHg contents were not significantly different ($p > 0.05$).

The total PUFA and important EPA + DHA contents of the fish diets from the two reservoirs are shown in Table 2. The PUFA profiles of these diets, including the contents of LIN, ALA, ARA, EPA and DHA are shown in Fig. 2.

The total PUFA contents of the benthos and artificial fish food were higher than those of the phyto- and zooplankton (Table 2) due to their higher LIN contents. LIN was the predominant PUFA in the zoobenthos and artificial fish food. The average LIN content of the zoobenthos in the HF Reservoir was 7.0 ± 6.5 mg/g while the average LIN content of the artificial fish food for omnivorous and carnivorous fish in the WJD Reservoir were 13.3 ± 1.2 and 15.0 ± 1.2 mg/g, respectively. The LIN contents of the zoobenthos and artificial fish food were higher than those in the phyto- and zooplankton in both reservoirs. The LIN contents of the phyto- and zooplankton in the HF Reservoir were 1.0 ± 0.4 and 0.7 ± 0.2 mg/g, respectively, and 0.6 ± 0.2 and 0.6 ± 0.2 mg/g in the WJD Reservoir, respectively (Fig. 2). The phyto- and zooplankton contained higher EPA + DHA contents than that of the zoobenthos in the HF Reservoir, while the EPA + DHA content of the zoobenthos in the HF Reservoir was lower than that of the artificial fish feeds in the WJD Reservoir (Table 2). In particular, the EPA content of the artificial fish food for omnivorous fish (0.6 ± 0.02 mg/g) was the lowest of all the fish foods. However, the DHA content of the artificial fish food for carnivorous fish (3.5 ± 2.1 mg/g) was the highest of all the fish foods from the two reservoirs (Fig. 2), which may be due to marine fish ingredients being added to the artificial fish foods. Fish is rich in DHA and marine fish often contain higher EPA + DHA than

Table 2
Total mercury (THg), methlymercury (MeHg) contents (ng/g dw), polyunsaturated fatty acids (PUFA) (mg/g dw) and eicosapentaenoic acid (EPA) + docosahexaenoic acid (DHA) contents (mg/g dw) of diet sources for fish in Hongfeng (HF) and Wujiangdu (WJD) Reservoirs.

	Phytoplankton		Zooplankton		Zoobenthos	Artificial fish feeds
	HF	WJD	HF	WJD	HF	WJD
n	3	3	3	3	24	6
THg	150 ± 0.6	32 ± 0.6	134 ± 0.6	60 ± 1.2	211 ± 213	14 ± 12
MeHg	7.2 ± 0.4	8.9 ± 1.4	4.2 ± 0.3	4.3 ± 0.7	95 ± 80	2.5 ± 3.2
Total PUFA	9.0 ± 0.9	5.3 ± 0.8	9.8 ± 1.3	4.9 ± 0.5	12 ± 10	16 ± 5.2
EPA + DHA	5.4 ± 1.2	0.5 ± 0.3	5.3 ± 1.6	0.4 ± 0.1	1.7 ± 1.6	3.9 ± 2.6

Note: Values are means ± standard deviation, "n" is the number of samples.

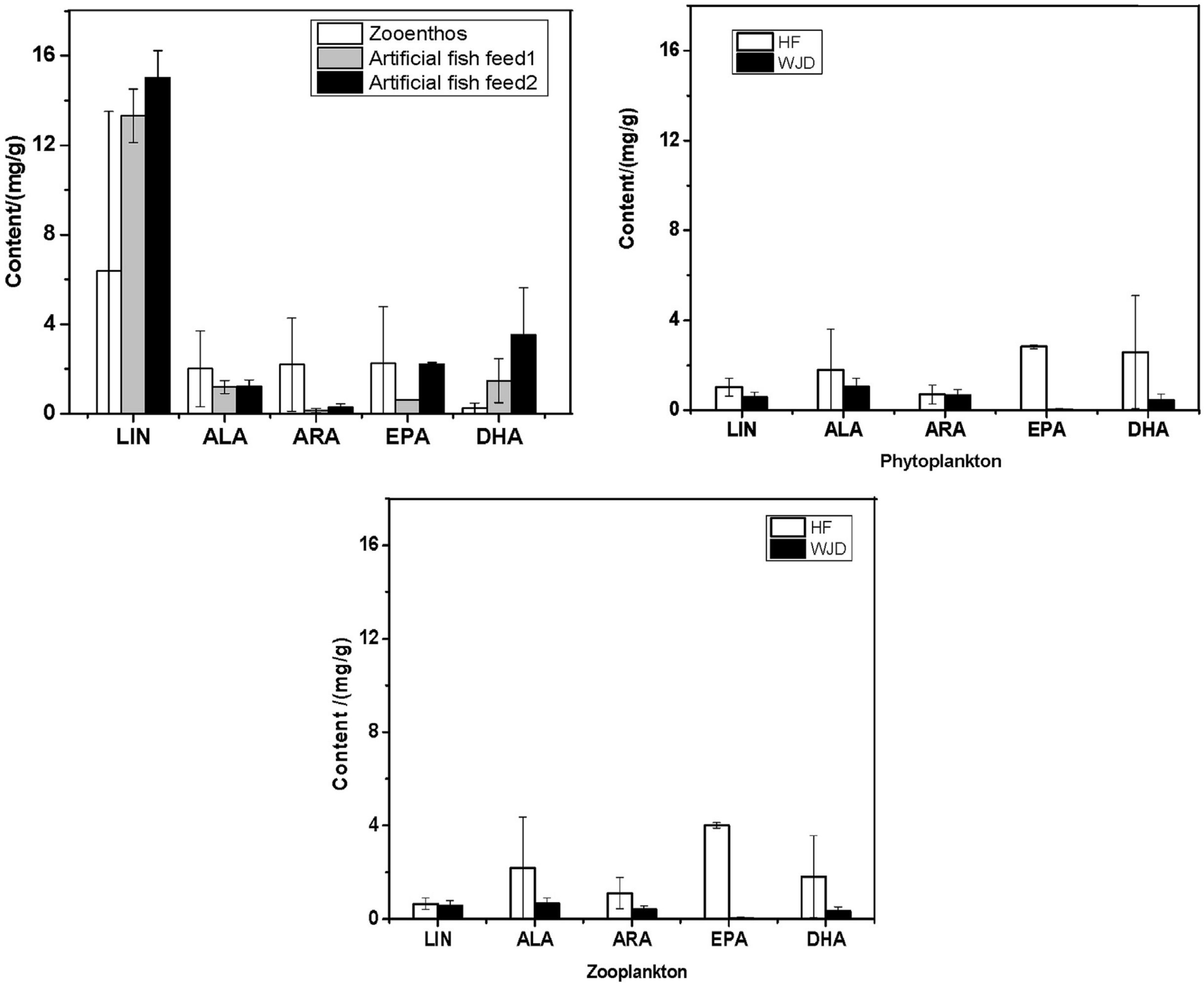

Fig. 2. Polyunsaturated fatty acids (PUFA) profile in zoobenthos in Hongfeng (HF) Reservoir, fish foods in Wujiangdu (WJD) Reservoir and phytoplankton and zooplankton in HF and WJD. Artificial fish feed 1: artificial fish feed for omnivorous fish; artificial fish feed 2: artificial fish feed for carnivorous fish. LIN: linoleic acid (C18:2n-6), ALA: α-linolenic acid (C18:3n-3), ARA: arachidonic acid (C20:4n-6), EPA: eicosapentaenoic (C20:5n−3), DHA: docosahexaenoic acid (C22:6n−3).

freshwater fish (Copeman et al., 2002; Gapasin and Duray, 2001; Li et al., 2011; Kris-Etherton et al., 2000).

The total PUFA contents of the phytoplankton and zooplankton in the HF Reservoir were almost twice ofthose in the WJD Reservoir. The ALA contents of the phyto- and zooplankton in the HF Reservoir were almost two times higher than those in the WJD Reservoir. The EPA + DHA contents of the phyto- and zooplankton in the HF Reservoir were almost 10 times higher than those in the WJD Reservoir (Table 2). The DHA contents of the phyto- and zooplankton in the HF Reservoir were almost 6 and 80 times higher than those in the WJD Reservoir, while the EPA contents were 94 and 6 times higher than those in the WJD Reservoir (Fig. 2). The markedly higher EPA and DHA contents of the plankton of the HF Reservoir indicates a higher dietary quality for planktivorous fish (Guo et al., 2016).

3.2.2. *Effect of eutrophication on Hg and PUFA in fish food sources*

Eutrophication can affect the accumulation of Hg and the PUFA profile in plankton by altering the biomass and composition of phytoplankton. A long history of aquaculture has increased the inputs of nutrient into both the WJD and HF Reservoirs, leading to eutrophication, which may decrease the Hg and PUFA concentrations in plankton. Compared to oligotrophic systems, the MeHg and PUFA contents of plankton in both reservoirs were lower than those with plankton of similar sizes from oligotrophic lakes and reservoirs reported in Canada, which the MeHg content of plankton was in the range 24–68 ng/g d.w. and total PUFA 13.9–21.6 mg/g d.w., and, in particular, higher EPA (7–13 mg/g) and DHA (10–14 mg/g) (Kainz et al., 2006) were observed comparing with the present study.

The trophic status of the WJD Reservoir was slightly higher than that of the HF Reservoir due to its ongoing aquaculture, while phytoplankton density in the WJD Reservoir (2.38×10^6 cells/L) (Huang et al., 2013) was significantly lower than that in the HF Reservoir (37.34×10^6 cells/L) (Huang et al., 2015). Fig. 3 shows the species composition of phytoplankton and zooplankton in the HF and WJD Reservoirs. Chlorophyta was the most abundant phytoplankton group in the two reservoirs, contributing 51% and 45% of the total phytoplankton in the HF and WJD reservoirs, respectively. The proportion of diatoms was similar in the HF and WJD reservoirs, accounting for 25% and 16% of the total phytoplankton, respectively. However, the proportion of Cyanophyta was 35% of phytoplankton in the WJD Reservoir, which was twice that of the HF Reservoir (14%). The results suggested that eutrophication shifted the phytoplankton abundance towards cyanobacteria.

The zooplankton density was similar in the WJD (21 ± 2 cells/L) and HF (23 ± 5 cells/L) reservoirs. However, the zooplankton taxa composition was different in the two reservoirs: 71% of

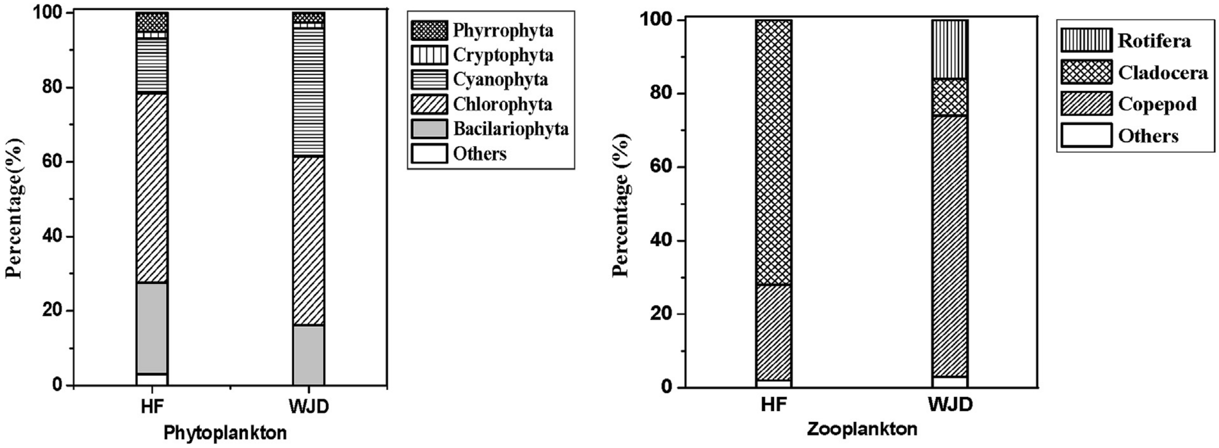

Fig. 3. Percentages of phytoplankton (64–112 μm) and zooplankton (112–500 μm) in Hongfeng (HF) and Wujiangdu (WJD) Reservoirs.

zooplankton in the WJD Reservoir were carnivorous copepods (cyclopoid) and 16% of the WJD Reservoir zooplankton were omnivorous rotifers, while 72% of zooplankton from the HF Reservoir were herbivorous cladocerans and no rotifers were identified (Fig. 3). The effects of ongoing aquaculture on the zooplankton community in the WJD Reservoir might be related with the increasing planktivorous fish stock biomass. It has been suggested that increasing planktivorous fish can decrease cladocerans but increase rotifers and copepods (Yang et al., 2010). Thus, ongoing aquaculture changed the phytoplankton and zooplankton taxa composition but had less effect on their density.

Mercury in plankton, especially phytoplankton, is mainly absorbed from water (Mason et al., 1995, 1996). Both the THg concentration in water and the THg contents of the phyto- and zooplankton in the HF Reservoir were higher than those in the WJD Reservoir ($p < 0.05$), while their MeHg contents were not significantly different ($p > 0.05$; Tables 1 and 2). The trends in THg and MeHg contents of the phyto- and zooplankton in the HF and WJD Reservoirs were consistent with the patterns of THg and MeHg concentrations in the water (Table 1). The higher trophic status in the WJD Reservoir altered the plankton composition, yet it didn't significantly increase the plankton biomass, so the eutrophication of WJD had no dilution effect on plankton Hg.

Certain fatty acids indicate the presence of certain phytoplankton groups (Taipale et al., 2009; Gladyshev et al., 2010). Therefore, the composition and density of phytoplankton can affect the PUFA profile of plankton. EPA and DHA are exclusively synthesised by diatoms and cryptophytes among phytoplankton species (Ahlgren et al., 1992), while Chlorophyta and Cyanophyta are the main producers of LIN and ALA (Arts et al., 2009). The similar PUFA profiles of phyto- and zooplankton in both two reservoirs suggest that the PUFA profile of zooplankton is mainly determined by the PUFA profile of its diet (Table 2 and Fig. 2). The higher total PUFA contents of the phyto- and zooplankton in the HF Reservoir than those in the WJD Reservoir ($p < 0.05$) are mostly due to higher algal density in HF (Table 2). Further, the lower EPA and DHA contents of the phyto- and zooplankton in the WJD Reservoir than those in the HF Reservoir ($p < 0.05$) may be associated with the higher percentages of Cyanophyta in the WJD Reservoir (Fig. 3).

3.3. Hg and PUFA in fish

3.3.1. Hg and PUFA contents in fish

Diet types can affect Hg contents and fatty acids composition in fish. For the same fish species, Hg contents generally increased with fish age, size and trophic level (Dang and Wang, 2012; Liu et al., 2014; Wang et al., 2012). While it is also suggested that larger fish with higher growth rate should have lower Hg concentrations that smaller ones with lower growth due to somatic growth dilution effect (Ward et al., 2010). In this study, we compared fish with same feeding habits in HF and WJD, which had similar age and trophic levels, but different diet structure and sizes. The fish sizes associated with their growth rate. Thus, we suggested that diet and growth rate can be important factors for the difference of the fish Hg in these reservoirs.

The contents of Hg, total PUFA and EPA + DHA in fish are shown in Table 3. The PUFA profiles of omnivorous and carnivorous fish are shown in Fig. 4 and the PUFA profiles of planktivorous fish are shown in Fig. 5.

The THg and MeHg contents of the cultured omnivorous and carnivorous fish were the lowest in all fish species ($p < 0.05$). The THg and MeHg contents of the wild omnivorous fish in the HF Reservoir were 17 and 12 times higher than those of the cultured omnivorous fish in the WJD Reservoir, respectively, while the THg

Table 3
Total mercury (THg), methlymercury (MeHg) (ng/g dw), polyunsaturated fatty acids (PUFA) (mg/g dw) and eicosapentaenoic acid (EPA) + docosahexaenoic acid (DHA) contents (mg/g dw) of fishes in Hongfeng (HF) and Wujiangdu (WJD) Reservoirs.

	Silver carp		Bighead carp		Omnivorous fish		Carnivorous fish	
	HF	WJD	HF	WJD	HF	WJD	HF	WJD
n	3	7	4	8	8	6	3	6
THg	258 ± 4.4	127 ± 52	247 ± 51	240 ± 73	514 ± 334	29 ± 17	302 ± 127	70 ± 70
MeHg	90 ± 6.4	24 ± 9.1	93 ± 46	101 ± 5.7	179 ± 99	14 ± 3.5	75 ± 7.6	24 ± 16
Total PUFA	1.3 ± 0.4	17 ± 8.0	6.9 ± 0.3	8.9 ± 2.5	18 ± 13	11 ± 5.7	5.5 ± 5.6	36 ± 30
EPA + DHA	0.3 ± 0.3	12 ± 2.8	5.2 ± 0.3	6.5 ± 2.0	4.7 ± 1.7	5.0 ± 1.7	1.3 ± 1.0	4.3 ± 2.0

Note: Values are means ± standard deviation, "n" is the number of samples.

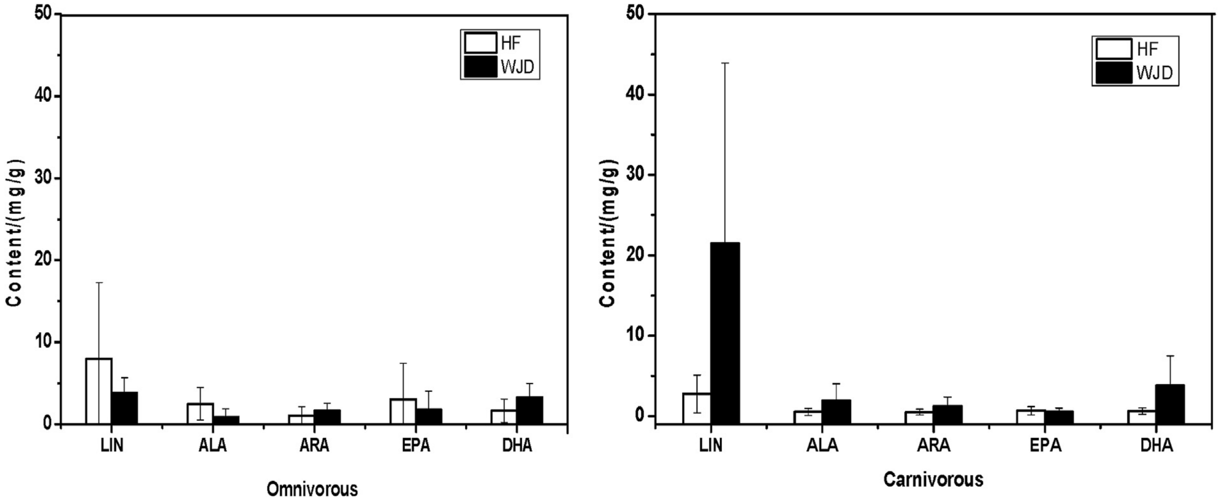

Fig. 4. Polyunsaturated fatty acids (PUFA) content (mg/g dw) in omnivorous and carnivorous fish in Hongfeng (HF) and Wujiangdu (WJD) Reservoirs. LIN: linoleic acid (18:2n-6), ALA: α-linolenic acid (18:3n-3), ARA: arachidonic acid (C20:4n-6), EPA: eicosapentaenoic (20:5n−3), DHA: docosahexaenoic acid (22:6n−3).

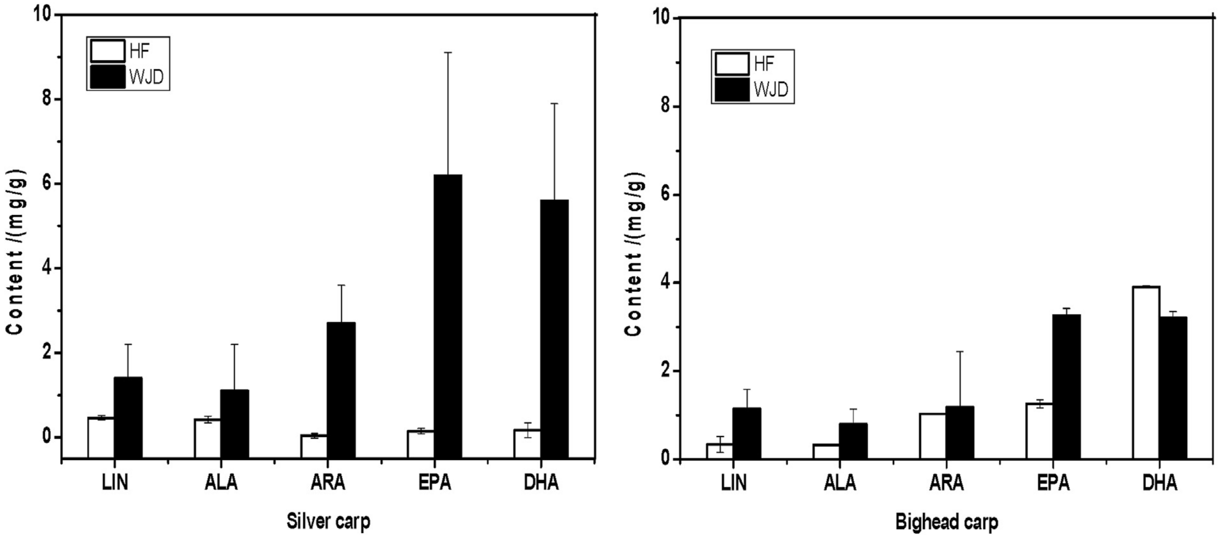

Fig. 5. Polyunsaturated fatty acids (PUFA) content (mg/g dw) in silver carp and bighead carp in Hongfeng (HF) and Wujiangdu (WJD) Reservoirs. LIN: linoleic acid (18:2n-6), ALA: α-linolenic acid (18:3n-3), ARA: arachidonic acid (C20:4n-6), EPA: eicosapentaenoic (20:5n-3), DHA: docosahexaenoic acid (22:6n-3).

and MeHg contents of the wild carnivorous fish in the HF Reservoir were four and three times higher than those of the cultured carnivorous fish in the WJD Reservoir, respectively. The total PUFA contents of the cultured omnivorous fish in the WJD Reservoir were slightly higher than those in the wild omnivorous fish in the HF Reservoir ($p > 0.05$) and their EPA + DHA contents were similar ($p > 0.05$). LIN, ALA and EPA contents of the wild omnivorous fish in the HF Reservoir were significantly higher than those of the cultured omnivorous fish in the WJD Reservoir ($p < 0.05$), while the DHA content of the cultured omnivorous fish in the WJD Reservoir was higher than that of the wild omnivorous fish in the HF Reservoir ($p < 0.05$). Different to omnivorous fish, the total PUFA contents of the cultured carnivorous fish in the WJD Reservoir were almost six-fold higher than those of the wild carnivorous fish in the HF reservoir and were also significantly higher than those of the cultured omnivorous fish in the WJD Reservoir ($p < 0.05$). The higher total PUFA contents of the cultured carnivorous fish in the WJD Reservoir is attributed to its higher LIN content (21.5 ± 22.3 mg/g), which is almost 10 times higher than that of the wild carnivorous fish in the HF Reservoir. In addition, the EPA + DHA content of the carnivorous fish in the WJD Reservoir was three times higher than that in the HF Reservoir and, in particular, the DHA content of the cultured fish was six times higher than that of the wild fish in the HF Reservoir.

The THg and MeHg contents of silver carp were higher in the HF Reservoir than those in the WJD Reservoir ($p < 0.05$), while they were similar in bighead carp in both reservoirs ($p > 0.05$). The total PUFA and EPA + DHA contents of silver carp in the WJD Reservoir were significantly higher than those in the HF Reservoir ($p < 0.05$) while the total PUFA and EPA + DHA contents of bighead carp in the HF and WJD Reservoirs were similar ($p > 0.05$). The five individual PUFA contents of silver carp in the WJD Reservoir were all significantly higher than those in the HF Reservoir ($p < 0.05$), especially for EPA and DHA, which were 40 and 30 times higher than that in the HF Reservoir, respectively. LIN and EPA contents of bighead carp in the WJD Reservoir were three and two times higher than those in the HF Reservoir, respectively.

3.3.2. Effect of aquaculture on Hg and PUFA in omnivorous and carnivorous fish

In the HF Reservoir, omnivorous fish mainly fed on phytoplankton, zooplankton and zoobenthos, while carnivorous fish mainly fed on zooplankton and zoobenthos. In the WJD Reservoir, both omnivorous and carnivorous fish mainly fed on artificial fish food. The Hg contents in artificial fish food in the WJD Reservoir were lower than those in phytoplankton, zooplankton and zoobenthos in the HF Reservoir (Table 2). The THg and MeHg contents of the omnivorous and carnivorous fish in the WJD Reservoir were lower than those in the HF Reservoir (Table 3). The possible reasons include the lower THg and MeHg contents in the artificial fish food and the higher growth rate of omnivorous and carnivorous fish in the WJD Reservoir. Dietary uptake is the dominant pathway of mercury uptake by fish (Hall et al., 1997; Wang et al., 2010). Therefore, it is a more important for fish to accumulate mercury from dietary than waterborne uptake, although the mercury in water from WJD is slightly lower than that in HF. For fish with similar feeding habits, the size of the cultured fish feeding on artificial fish food was higher than that of the wild fish (Table S2). Both the wild and cultured fish were 1–2 years old, however the sizes of cultured omnivorous and carnivorous fish in the WJD Reservoir were significantly higher than those of the wild omnivorous and carnivorous fish in the HF Reservoir ($p < 0.05$), which is mainly attributed to the high food supply of artificial fish food (Table S2 in SI).

Differences of PUFA profiles in carnivorous fish from HF and WJD are generally consistent with that in their dietary supply (Table 3). In contrast, the variation in the PUFA profiles of the cultured and wild omnivorous fish showed a large discrepancy with that of their diets. We speculate that was because the food sources for omnivorous fish were more diverse than those of carnivorous fish in the HF Reservoir and, in addition to plankton and zoobenthos, there might be other food sources yet to be identified.

3.3.3. Effect of aquaculture on Hg and PUFA in planktivorous fish

The MeHg content of silver carp in the WJD Reservoir was lower than that in the HF Reservoir, while the MeHg content of the phytoplankton in the HF and WJD Reservoirs were similar. The THg content of bighead carp in both reservoirs were similar, while the THg content of the zooplankton in the HF Reservoir was higher than that in the WJD Reservoir. The PUFA profile of silver carp and bighead carp were inconsistent with those of the phytoplankton and zooplankton, which is in contrast to what Razavi observed in eutrophic reservoirs in eastern China (Razavi et al., 2014), who suggested that eutrophication can reduce the contents of Hg, EPA and DHA in bighead carp via changing the plankton density and species composition. In contrast, the higher trophic status in the WJD Reservoir did not increase the plankton density or influence Hg bioaccumulation and the PUFA profile of planktivorous fish. In our study, the status of eutrophication (indicated by carlson trophic level index) of the HF and WJD Reservoirs was similar, and thus may have had no significant effect on the Hg content and PUFA profile of planktivorous fish. Besides, unlike wild fish in the HF Reservoir, the fish in the WJD Reservoir were kept in net cages and were less active than fish in the HF Reservoir, leading to differences in growth, behaviour, physiology and morphology between the two reservoirs, which might affect the bioaccumulation of Hg and PUFA in fish (Arechavala-Lopez et al., 2012; Einum and Fleming, 1997; Handelsman et al., 2010).

Eutrophication caused by aquaculture didn't result in significant difference in the toxic Hg and beneficial EPA + DHA contents of fish in HF and WJD. But we observed that the Hg and EPA + DHA contents of fish in these two reservoirs were lower than that in oligotrophic aquatic systems. The Hg contents of fish in the HF and WJD reservoirs were within the range of freshwater fish Hg in China (the ranges of THg and MeHg was 8.5–274.10 and 8.0–680.0 ng/g w.w., respectively) (Yan et al., 2019). It has been widely reported that Hg contents of freshwater fish in China are lower than those from oligotrophic aquatic systems in Europe (Lars, 2003; Rask and Metsälä, 1991) and North America (Evans et al., 2005; Kainz et al., 2006; Lockhart et al., 1972), and it was assumed that the fish sampled from China were younger and had a higher growth rate, which together reduced the bioaccumulation of Hg (Liu et al., 2012; Zhang et al., 2007; Razavi et al., 2015). The EPA + DHA contents of bighead carp in the HF and WJD Reservoirs approached that of eutrophic reservoirs in eastern China (4.55–7.40 mg/g) (Razavi et al., 2014), while the EPA + DHA contents were lower than those of planktivorous fish in Canada (13.6 ± 4.8 mg/g) (Kainz et al., 2004, 2006). The EPA + DHA contents of omnivorous and carnivorous fish in the HF and WJD Reservoirs were lower than those of oligotrophic freshwater fish in Sweden (omnivorous: 12.8 ± 2.9 mg/g, carnivorous: 5.7 ± 1.2 mg/g) (Ahlgren et al., 1999). We suggest that most reservoirs in China are eutrophic and more prone to the occurrence of phytoplankton, *e.g.* cyanobacteria and green algae, which do not synthesise LC-PUFA, including EPA and DHA. Lakes in Europe and North America, however, are mostly oligotrophic where the dominant plankton species are diatoms, thus more EPA and DHA can be synthesised and transferred to fish. In the present study, the EPA + DHA contents of artificial food approached those of plankton, but they were still less efficient than those of plankton in oligotrophic lakes (Kainz et al., 2004). Therefore, fish in the HF and WJD reservoirs may retain low LC-PUFA contents because of the generally low dietary LC-PUFA supply in these eutrophic ecosystems.

3.3.4. Assessments of risks and benefits of cultured fish consumption for human health

The average THg and MeHg concentrations in cultured fish (n = 27) from WJD Reservoir were 30.7 ± 29.1 and 7.9 ± 7.1 ng/g w.w, respectively. The average THg and MeHg concentrations in wild fish (n = 18) from HF Reservoir were 74.6 ± 50.5 and 27.1 ± 15.4 ng/g w.w, respectively. No samples exceeded the national limit recommended by the Standardization Administration of China (500 ng/g w.w. for MeHg) (GB 2762-2012, 2013). It indicates that fish consumption does not cause health risks due to mercury exposure. In term of nutrition PUFAs, the total PUFAs contents of cultured fish in WJD (16.13 ± 16.39 mg/g d.w.) was generally higher than the wild fish in HF (13.1 ± 12.2 mg/g d.w.), which were comparable with the wild freshwater fish in some aquatic systems (the average range from 8.09 to 20.10 mg/g) (Williams et al., 2017; Gladyshev et al., 2017; Laird et al., 2018). The EPA + DHA contents of fish in WJD (21.2–52.2 mg/g w.w.) were higher than those in HF (1.4–26.0 mg/g w.w.) as well. Therefore, for general adult with daily consumption of 227 g fish (USEPA, 2019), all the cultured fish species in WJD can provide the recommended intake of EPA + DHA (250 mg/day) (EFSA, 2010), and the carnivorous catfish in WJD Reservoir was optimal choice owing to its highest EPA + DHA.

4. Conclusions

We showed that cultured omnivorous and carnivorous fish contained lower THg and MeHg contents than the wild ones, yet their total PUFA and EPA + DHA contents were higher than the wild ones. As to planktivorous fish, THg and MeHg contents of silver carp in aquaculture reservoir were lower than that without aquaculture, while THg and MeHg contents of bighead carps were similar in these two reservoirs. Total PUFA and EPA + DHA contents of planktivorous fish in aquaculture reservoir were higher than that without aquaculture. Higher trophic status of aquaculture reservoir had no significant effect on Hg contents in plankton, but it likely altered the plankton community structure, which consequently decreased the total PUFA and EPA + DHA contents in plankton. In summary, aquaculture may have reduced the Hg bioaccumulation and overall enhanced the PUFA content in cultured fish. Our study suggested that the concumption of cultured fish present no mercury health risk and can be a benefit source of PUFA for human health. However, eutrophication caused by aquaculture likely poses impact on the ecological health of pelagic food web.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2019.113543.

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