





Original Article

Pollution status and trophic transfer of polycyclic aromatic hydrocarbons in coral reef ecosystems of the South China Sea

Yali Li^{1,2,3,4}, Xinqing Zou^{2*}, Shichun Zou^{1,3,4}, Pu Li^{1,3,4}, Ying Yang^{1,3,4*} , and Junjie Wang⁵ 

¹School of Marine Sciences, Sun Yat-sen University, Zhuhai 519082, China

²Ministry of Education Key Laboratory for Coastal and Island Development, Nanjing University, Nanjing 210093, China

³Southern Marine Science and Engineering Guangdong Laboratory (Zhuhai), Zhuhai 519080, China

⁴Pearl River Estuary Marine Ecosystem Research Station, Ministry of Education, Zhuhai 519082, China

⁵Department of Earth Sciences—Geochemistry, Faculty of Geosciences, Utrecht University, Utrecht 3508 TA, The Netherlands

*Corresponding authors: tel: +86-025-89680605; e-mail: zouxq@nju.edu.cn (X.Z); tel: +86-13427684661; e-mail: yangying6@mail.sysu.edu.cn (Y.Y.)

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Coral reef ecosystems are important fishing grounds in tropical and subtropical areas and have been widely contaminated by chemicals. However, the current understanding of the pollution status and trophic transfer of exogenous chemicals in coral reef ecosystems is still limited. This study aims to characterize the occurrence of 16 priority polycyclic aromatic hydrocarbons (PAHs) in sediments, corals, and other biotas in the trophic-complex coral reef ecosystems in the Xisha and Nansha Islands of the South China Sea. PAH transfer characteristics in the highly diverse coral reef food webs were analyzed through stable isotope ratios and trophic magnification factors (TMFs). PAHs were observed in sediment and biota samples of both Xisha and Nansha coral reef ecosystems. The TMFs ranging from 0.46 to 2.18 for the Xisha food web indicate limited trophic transfer of PAHs, while those ranging from 1.07 to 5.25 for the Nansha food web indicate trophic magnification of PAHs. The octanol-water partition coefficient (K_{ow}) of PAHs may positively influence the PAH biomagnification via accumulation along the trophic levels. This study provides new insights into PAH contamination patterns and their trophodynamic behaviour in highly diverse coral reef ecosystems, which can serve as a scientific basis for ecological risk assessment, biodiversity conservation, and coastal function management in global coral reef systems.

Keywords: biomagnification, coral reef system, PAHs, the South China Sea, trophic transfer

Introduction

Coral reefs, formed in clear tropical waters, constitute the most diverse and productive ecosystem in the ocean (Yu, 2018). They provide valuable ecosystem service functions including high primary productivity, supply of multiple natural resources, and conservation of coastal environments (Cunning et al., 2015; Yu, 2018). Moreover, coral reef regions are important fishing grounds and coral reef fisheries support the high-quality seafood demand for tens of millions of people (Mcmanus et al., 2000). Corals are extremely sensitive to environmental changes and have been regarded as a potential bio-indicator for chemical pollution (Ko et al., 2014; Han et al., 2019). However, due to increasing anthropogenic impacts in recent decades (Gardner et al., 2003;

Fabricius, 2005; Han et al., 2019), e.g. continuous release of toxic chemical substances to the ocean (Meehan and Ostrander, 1997; Ko et al., 2014; Yang et al., 2019), the habitat has been changed and coral reefs all over the world have been experiencing catastrophic declines or even extinction (Thomas and Li, 2000; Ko et al., 2014; Ranjbar Jafarabadi et al., 2017). Many studies have reported coral bleaching (De'Ath et al., 2009; Ko et al., 2014; Xiang et al., 2018), coral reef organisms decline and coral reef ecosystem degradation caused by chemical contamination (Hughes et al., 2017; Xiang et al., 2018; Han et al., 2019). Burke et al. (2011) estimated that various types of pollutants introduced by human activities have affected ~20–25% of coral reefs worldwide.

Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous environmental contaminants and are generated mainly from human activities (Neff, 1979; Li *et al.*, 2019). Pollution of PAHs in the environment is of special concern because of their toxic, mutagenic, carcinogenic, and persistent properties (Kim *et al.*, 2013; Ko *et al.*, 2014; Wang *et al.*, 2017). PAHs are also known as endocrine-disrupting chemicals due to their structural similarity with steroid hormones (Yang *et al.*, 2020). Among hundreds of known PAHs, 16 of them are designated as priority pollutants by the US Environmental Protection Agency (US EPA) according to their prevalence, persistence in the environment, and potential toxicity (IARC, 2007; Ramdine *et al.*, 2012). In aquatic environments, PAHs preferentially partition to sediments and biological membranes because of their hydrophobicity (octanol-water partition coefficient, $\log K_{ow}$ ranging from 3.37 to 7.00) (Neff and Burns, 1996). Once uptake occurs, PAHs are easily accumulated in aquatic organisms due to their high lipophilicity. Accumulation of PAHs via food webs may affect species at higher trophic levels (TLs) (Gray, 2002; Kelly *et al.*, 2007; Bartkowski *et al.*, 2016) and pose threats to the biotas and ecosystem. Furthermore, via trophic transfer, biomagnification of PAHs may eventually affect seafood quality and availability and human health. Therefore, pollution status and trophic transfer of PAHs in aquatic ecosystems should be considered as an important criterion for ecological risk assessment.

So far, studies investigating the trophodynamics of PAHs in aquatic environments are limited and no consensus has been reached on PAH trophic transfer mechanisms. Some results suggest that PAHs are diluted along TLs in the aquatic food webs (Broman *et al.*, 1990; Wan *et al.*, 2007; Khairy *et al.*, 2014; Fan *et al.*, 2017; Qadeer *et al.*, 2019). However, other studies prove the existence of trophic magnification in both freshwater (Wang *et al.*, 2012) and marine food chains (Zhang *et al.*, 2015), which is further supported by both mathematical modelling results from laboratory experiments and measurements from field studies (Richter and Nagel, 2007; Froehner *et al.*, 2011; Arias *et al.*, 2016). The trophic dynamics of PAHs in aquatic ecosystems remain controversial and lack clarity. Therefore, further investigations are warranted to shed light on the PAH trophic transfer behaviour and its influencing factors.

Coral reef systems are a special type of aquatic environments and an ecologically critical zone of tropical and subtropical oceans. The occurrence of PAH pollution in coral reef regions has been reported previously (Ko *et al.*, 2014; Xiang *et al.*, 2018; Han *et al.*, 2019), while the understanding of the mechanism of PAH trophic transfer in coral reef ecosystems remains deficient. The coral reef system in the South China Sea (SCS) is one of the most diverse and well-developed systems in the world (Yu, 2012), but it is increasingly damaged due to aggravating factors such as chemical contamination (Sun *et al.*, 2014, 2016; Li *et al.*, 2019). This study focuses on the pollution status and trophodynamics of PAHs in the coral reef ecosystems in this area, aiming to (1) reveal the pollution status of PAHs in sediments, fish, corals, and other invertebrates in the coral reef ecosystem of the SCS; (2) identify the PAH composition patterns of different compartments in coral reef ecosystems; (3) clarify the trophic transfer characteristics of PAHs through tropical coral reef webs using stable isotope methods and the trophic magnification factors (TMFs). This study provides the first comprehensive assessment of the pollution status, composition pattern, and particular trophodynamics of PAHs in tropical coral reef ecosystems and serves as a first step

towards ecological risk assessment, biodiversity conservation and coastal function management in global coral reef systems.

Material and methods

Study area

The coral reef area of China is the eighth largest and accounts for 2.57% in the globe (Xiang *et al.*, 2018). Adjacent to the western border of the “Coral Triangle”, the ecological and economic significance of the coral reefs in the SCS has been highly emphasized since the 1960s (Zou *et al.*, 1966; Burke *et al.*, 2002; Zhao *et al.*, 2016a, b). However, the coral coverage of the SCS, particularly in offshore reef atolls and archipelagos, has dramatically declined from an average of >60 to ~20% over the last 10–15 years (Hughes *et al.*, 2013). Coral reefs in the SCS are mainly distributed in the offshore Dongsha, Xisha, Zhongsha, and Nansha Islands (Figure 1a). Sediment and biota samples were collected in four coral reef atolls, *i.e.* Yongle Atoll (15°46′–17°07′N, 111°11′–112°03′E, Figure 1b), Daoming Atoll (10°40′–10°55′N, 114°19′–114°37′E, Figure 1c), Zhenghe Atoll (10°19′–10°05′N, 114°12′–114°45′E, Figure 1d), and Jiuzhang Atoll (9°42′–10°00′N, 114°15′–114°40′E, Figure 1e), of two offshore archipelagos (Xisha and Nansha Islands) located in a core area of coral reefs in the SCS. Most of these coral reefs have no permanent human habitation. Our study area is approximately 1200 km from Hainan Island and is less impacted by anthropogenic influences compared with reefs in coastal regions. The three atolls of Nansha Islands are close to an important shipping channel connecting the Pacific Ocean and Indian Ocean (*i.e.* Malacca strait-the South China Sea channel). The annual temperature of the whole target area is ~28.1°C (Yu *et al.*, 2006). A detailed description of Yongle Atoll and Jiuzhang Atoll has been presented in a previous study (Li *et al.*, 2019).

Sample collection

Sampling of sediments, fish, corals, and invertebrates excluding corals was conducted on two surveys in (1) Yongle Atoll, Zhenghe Atoll, and Jiuzhang Atoll (Xisha–Nansha voyage, 2016), (2) Doming Atoll (Nansha voyage, 2017). Detailed information on sample location, number, and coral species can be found in Supplementary Table S1 in the Supporting Information. Most of the coral reefs in the sampling region are 0–2 m under the low tide line. A total of 21 surface sediment samples at about 10-cm depth were collected by a diver on the reef platform (Supplementary Table S1); 8 coral samples (Supplementary Table S1) were collected at the reef platform of the Zhenghe Atoll (4 samples collected) and Jiuzhang Atoll (4 samples collected); 6 invertebrates and 22 fish samples were collected at Yongle and Jiuzhang Atolls (Table 1).

All biota samples were obtained through diving and collecting on-site with the aid of fishermen from Tanmen. Representative samples of corals were collected by divers using a hammer and a shovel. Besides corals, fish and other invertebrates were also investigated because they are reef dwellers and important components of the coral reef ecosystem, and are potentially excellent indicators of the pollution status throughout the system. Once on board, the sediment and biota samples were immediately stored in a freezer at –20°C. Reef dwelling organisms were identified at the Nantong Aquatic Products Research Institute. The biota samples were rinsed three times by ultrapure water and their body lengths were measured (data presented in Table 1). The dorsal

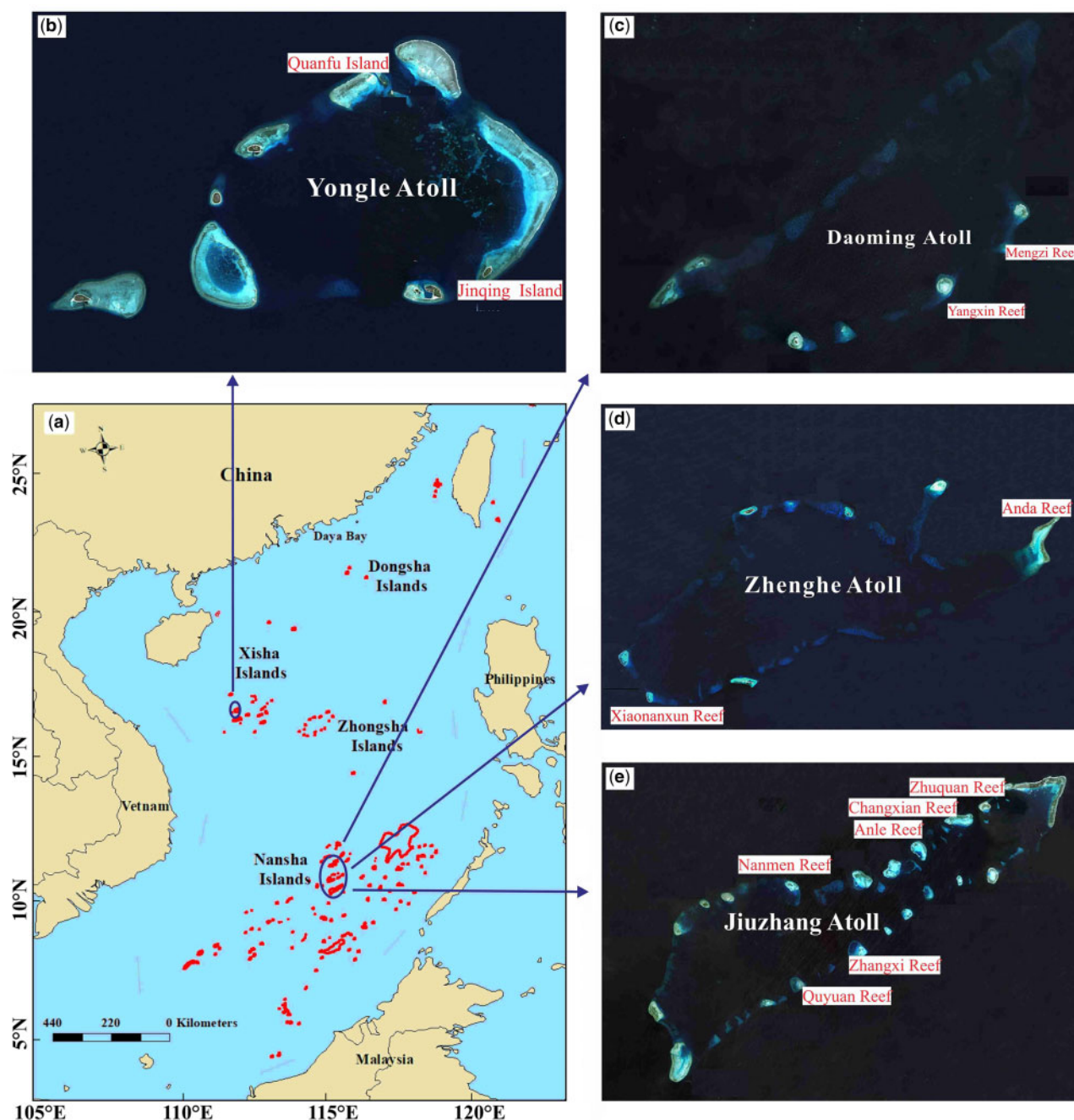


Figure 1. Study area and sampling location in the South China Sea. (a) General location of the Xisha and Nansha Islands, (b) Yongle Atoll in the Xisha Islands, (c) Daoming Atoll, (d) Zhenghe Atoll, and (e) Jiuzhang Atoll in the Nansha Islands.

muscles for fish and soft part of the body (mainly the muscles) for invertebrates were used for PAH-content determination. Sediment and organism tissue samples were dried by vacuum freeze-drying and then grounded into fine powders prior to analysis.

Sample preparation and PAH measurement

Detailed PAH extract method and analytical methods can be found in Li *et al.* (2019). Briefly, dried sediments or biota tissues (5 g) were spiked with a mixture of surrogate standards consisting

of 5 deuterated PAHs including naphthalene-d8 (Nap-D8), acenaphthene-d10 (Ace-D10), phenanthrene-d10 (Phe-D10), chrysene-d12 (Chr-D12), and perylene-d12 (Pre-D12). The samples were extracted with dichloromethane for 48 h using a Soxhlet apparatus. For invertebrate and fish samples, the extract was divided into two sections. One aliquot of the extract was used to measure the lipid content. The remaining aliquot was firstly purified by gel permeation chromatography with SX-3 Bio-Beads (40 g, Bio-Rad Laboratories, USA) and rinsed with dichloromethane/n-hexane (v/v = 1:1). The eluate (90–280 ml) was concentrated by a rotary

Table 1. Overview of information on the biota samples, species identification and contents of lipid, PAHs, and stable isotopes ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$).

Sample	Species	Trophic level	Feeding mode	Sampling area	Location	Sample numbers	body length (cm)	Lipid %	$\Sigma 16\text{PAHs}$ (ng/g dw)	$\delta^{15}\text{N}$ (‰)	$\delta^{13}\text{C}$ (‰)
Invertebrate	Sea cucumber	1.26	Omnivorous	Xisha Islands	Yongle Atoll	1	14	1.25	28.70	3.50	-9.90
	Sleeve-fish	3.62	Omnivorous	Xisha Islands	Yongle Atoll	1	26	5.76	3.60	10.59	-16.86
	<i>Panulirus stimpsoni</i>	4.07	Omnivorous	Nansha Islands	Jiuzhang Atoll	1	16	3.50	16.43	9.72	-16.52
	<i>Tridacnidae spp.</i>	2.12	Omnivorous	Nansha Islands	Jiuzhang Atoll	5	7	5.33	18.68	3.86	-17.31
	<i>Cymatium muricinum</i>	2.20	Omnivorous	Nansha Islands	Jiuzhang Atoll	7	-	2.87	11.32	2.83	-14.36
	<i>Trochus maculatus</i> Linnaeus	1.77	Omnivorous	Nansha Islands	Jiuzhang Atoll	5	-	1.75	16.51	4.12	-11.20
Fish	<i>Lethrinus lentjan</i>	3.77	Micro-carnivorous	Xisha Islands	Yongle Atoll	1	28	3.03	138.50	11.03	-15.36
	<i>Lutjanus gibbus</i>	3.48	Micro-carnivorous	Xisha Islands	Yongle Atoll	1	23	2.30	59.42	10.15	-14.73
	<i>Plectorhinchus chaetodonoides</i>	3.29	Micro-carnivorous	Xisha Islands	Yongle Atoll	1	33	2.43	24.75	9.57	-14.58
	<i>Scarus oviceps</i>	2.58	Omnivorous	Xisha Islands	Yongle Atoll	1	33	3.98	22.56	7.45	-11.94
	<i>Scarus niger</i>	3.26	Micro-carnivorous	Xisha Islands	Yongle Atoll	1	33	5.29	29.84	9.49	-18.10
	<i>Cephalopholis argus</i>	3.60	Omnivorous	Xisha Islands	Yongle Atoll	1	23	3.96	409.28	10.51	-17.44
	<i>Scarus rivulatus</i>	2.13	Omnivorous	Xisha Islands	Yongle Atoll	1	29	1.77	22.38	6.09	-15.90
	<i>Scarus niger</i>	1.84	Omnivorous	Xisha Islands	Yongle Atoll	1	30	6.33	237.13	5.22	-11.12
	<i>Scarus forsteni</i>	2.25	Omnivorous	Xisha Islands	Yongle Atoll	1	27	2.86	22.34	6.46	-12.29
	<i>Scarus ghobban</i>	1.73	Omnivorous	Xisha Islands	Yongle Atoll	1	29	4.25	23.40	4.91	-12.77
	<i>Scarus forsteni</i>	2.09	Omnivorous	Xisha Islands	Yongle Atoll	1	27	2.21	31.99	5.99	-16.10
	<i>Epinephelus quoyanus</i>	3.23	Micro-carnivorous	Xisha Islands	Yongle Atoll	2	16.5-19	10.59	57.37	9.41	-11.96
	<i>Lutjanidae</i>	3.47	Omnivorous	Xisha Islands	Yongle Atoll	1	34	4.90	60.14	10.13	-19.31
	<i>Lethrinus erythropterus</i>	3.60	Micro-carnivorous	Xisha Islands	Yongle Atoll	1	30	2.64	12.79	10.51	-13.96
	<i>Cheilinus fasciatus</i>	3.07	Micro-carnivorous	Xisha Islands	Yongle Atoll	1	37	2.01	15.89	8.92	-14.44
	<i>Plectorhinchus diagrammus</i>	3.23	Micro-carnivorous	Xisha Islands	Yongle Atoll	1	34	2.46	19.27	9.39	-15.77
	<i>Navodon tessellates</i> Günther	3.23	Omnivorous	Nansha Islands	Jiuzhang Atoll	1	25	2.96	63.52	7.20	-21.39
	<i>Scarus niger</i>	4.25	Omnivorous	Nansha Islands	Jiuzhang Atoll	1	24	6.33	139.09	10.27	-14.01
	<i>Pterocaesio tile</i>	3.75	Micro-carnivorous	Nansha Islands	Jiuzhang Atoll	6	16-22	12.97	32.71	8.78	-18.27
	<i>Ballistapus undulatus</i>	4.02	Micro-carnivorous	Nansha Islands	Jiuzhang Atoll	1	22	17.34	92.78	9.59	-16.52
	<i>Scomberomorus niphonius</i>	3.98	Macro-carnivorous	Nansha Islands	Jiuzhang Atoll	1	26	3.96	36.60	9.47	-18.28
	<i>Scomberomorus niphonius</i>	4.08	Macro-carnivorous	Nansha Islands	Jiuzhang Atoll	1	70	0.90	59.55	9.77	-17.82

evaporator to 1 ml and fractionated using a glass column packed with 3 cm of neutral alumina, 3 cm of silica gel, and 1 cm of anhydrous sodium sulfate layered from bottom to top. The concentrated solution was eluted with 50 ml dichloromethane/n-hexane (v/v = 1:1), then concentrated to near dryness by N₂, exchanged with n-hexane and finally concentrated to 100 μL using a rotary evaporator.

Sixteen priority PAHs (naphthalene-Nap, acenaphthylene-Acy, acenaphthene-Ace, fluorine-Flu, phenanthrene-Phe, anthracene-Ant, fluoranthene-Flo, pyrene-Pyr, benzo[a]anthracene-BaA, chrysene-Chr, benzo[b]fluoranthene-BbF, benzo[k]fluoranthene-BkF, benzo[a]pyrene-BaP, indeno[1,2,3-c, d]pyrene-IndP, dibenzo[a, h]anthracene-DahA, and benzo[g, h, i]perylene-BghiP, IARC, 2007) were analyzed by gas chromatography coupled with triple quadrupole mass spectrometry (GC-MS/MS, Thermo Fisher Scientific, TSQ 8000 Evo, USA) with a quartz capillary TG-5MS column (30 m × 0.25 mm × 0.25 μm film thickness). The injector was kept at 270°C. The temperature program firstly increased from 70°C to 140°C at a rate of 25°C min⁻¹, then increased to 300°C at a rate of 5°C min⁻¹ and hold for 4 min. The temperature of the ion source was 280°C. The selected reaction monitoring (SRM) mode was applied in all sample analysis.

In addition, PAHs are commonly classified into two groups of low molecular weight (LMW) and high molecular weight (HMW), referring to PAHs with 2–3 and 4–6 benzene rings, respectively. Both terms were used to discuss the PAH composition in this study.

Quality assurance/quality control

A procedural blank, spiked sample, and duplicate sample were prepared and analyzed with every 12 samples. The relative percentages of analysis differences between duplicate samples were lower than 15%. An external standard method (Wang *et al.*, 2015) was used for quantification with a calibration curves based on 16 PAHs at 9 standard concentrations (1, 10, 50, 100, 200, 400, 500, 800, and 1000 ng L⁻¹). The correlation coefficients ($R^2 > 0.999$) for all calibration curves were checked daily for all targeted PAHs. The recoveries of the surrogates ranged from 45% for Nap—D8 to 126% for Chr-D12 of the spiked concentrations. No detectable amounts of PAHs were found in the blanks.

Trophodynamic analysis

Stable isotopes and TMF models are widely used to investigate the trophodynamics of PAHs in aquatic ecosystems (Fisk *et al.*, 2001; Verhaert *et al.*, 2013). Details of the methods are described as follows:

Stable isotope analysis

Stable nitrogen isotope analysis is widely used to characterize the relative TL in the food web. The nitrogen isotope abundance, expressed as ¹⁵N (‰), increases with dietary assimilation during the trophic transfer process. Therefore, it is used as a proxy of an organism's TL (Verhaert *et al.*, 2013). Stable isotope analyses were performed on 21 sediments, 6 invertebrates, and 22 fish samples in the SCS coral reef ecosystems. The samples were firstly dried at -40°C, then homogenized into fine powders, weighed to about 0.001 mg and encapsulated in 6 × 5 mm Sn capsules to determine their carbon (C) and nitrogen (N) contents as well as carbon isotope abundance ($\delta^{13}\text{C}$) and $\delta^{15}\text{N}$. Stable isotope

measurements were conducted with a Vario ISOPOTE Cube-Isoprime by Elementar. A standard notation is used to express stable isotope results as follows:

$$\delta^{13}\text{C}, \delta^{15}\text{N} = [R_{\text{sample}}/R_{\text{reference}} - 1] \times 1000, \quad (1)$$

where $R = {}^{13}\text{C}/{}^{12}\text{C}$ is used for C and $R = {}^{15}\text{N}/{}^{14}\text{N}$ is used for N.

Data were calibrated using Acetanilide #1 ($\delta^{13}\text{C} = -29.53$ ‰ and $\delta^{15}\text{N} = 1.18$ ‰). Estimated precision for $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ were 0.2 and 0.25‰, respectively.

TLs are calculated from biota $\delta^{15}\text{N}$ values based on the following equation (Post, 2002):

$$\text{TL}_{\text{consumer}} = (\delta^{15}\text{N}_{\text{consumer}} - \delta^{15}\text{N}_{\text{primary consumer}}) / \Delta^{15}\text{N} + 2 \quad (2)$$

where $\text{TL}_{\text{consumer}}$ is the TL of the organism, $\delta^{15}\text{N}_{\text{consumer}}$ is the $\delta^{15}\text{N}$ of the organism, $\delta^{15}\text{N}_{\text{primary consumer}}$ is the mean $\delta^{15}\text{N}$ of a local long-lived primary consumer, 2 is the TL of the primary consumer and $\Delta^{15}\text{N}$ is the trophic enrichment factor or shift in $\delta^{15}\text{N}$ between consecutive TLs (Post, 2002). In this study, the average value of $\delta^{15}\text{N}$ of the reef sediments was used as a baseline. $\Delta^{15}\text{N}$ trophic fractionation value of 3‰ was used as the most appropriate estimate value for non-acid treated muscle tissues (McCutchan *et al.*, 2003; Verhaert *et al.*, 2013). In fact, $\Delta^{15}\text{N}$ can be variable, depending on the surrounding environment of the organism (McCutchan *et al.*, 2003). The exact value of $\Delta^{15}\text{N}$ only affects the absolute TL estimates but not the relationships between chemicals and estimated TLs (Verhaert *et al.*, 2013).

TMF analysis

As the TMF represents the average increase rate per TL, TMFs have been used as a reliable and conclusive tool for bioaccumulation assessments of chemicals (Wan *et al.*, 2007; Verhaert *et al.*, 2013). In this study, TMFs were employed to evaluate the biomagnification potential of PAHs along the TLs.

TMFs are based on the relationships between the TLs and PAH concentrations using single linear regression:

$$\log_{10}(\text{PAH}_{\text{concentration}}) = a + b \times \text{TL} \quad (3)$$

where a and b represent intercept and slope of the linear regression, respectively. The TMF is calculated as follows:

$$\text{TMF} = 10^b, \quad (4)$$

If $\text{TMF} > 1$, the target chemical is considered to be biomagnified in the food web (Verhaert *et al.*, 2013). Furthermore, Pearson's correlation coefficients were calculated between TLs and \log_{10} -normalized concentrations in the biota tissues for all compounds and presented in Table 1.

Result and discussion

PAH concentration

Sediments

Figure 2 shows the measured PAH concentrations in the surface sediments of the Xisha and Nansha Islands. The concentration of $\Sigma_{16}\text{PAHs}$ in the surface sediments of the Xisha Islands ranged from 9.00 ng g⁻¹ in dry weight (dw) in the Quanfu Island (QFD) to 16.94 ng g⁻¹ dw in the Jinqing Island (JQD), averaging 13.65 ng g⁻¹ dw. Those of the Nansha Islands ranged from

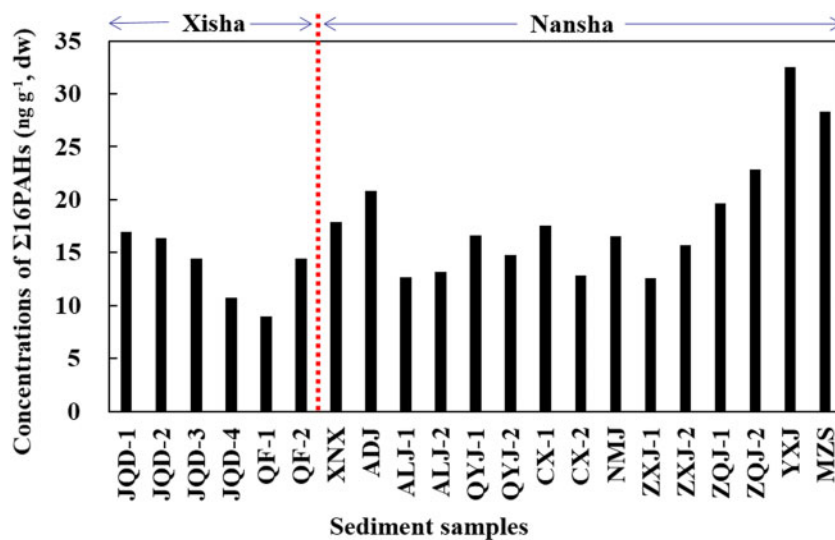


Figure 2. The concentrations of Σ_{16} PAHs in sediment samples collected from the Xisha and Nansha Islands in the South China Sea. Details of the sampling stations along the x-axis can be found in [Supplementary Table S1](#).

12.56 ng g⁻¹ dw in the Zhangxi Reef (ZXJ) to 32.48 ng g⁻¹ dw in the Yangxin Reef (YXJ), averaging 18.29 ng g⁻¹ dw, which was slightly higher than the average in the Xisha Islands.

The concentrations of Σ_{16} PAHs in the surface sediments of the offshore SCS in this study are significantly lower than 67.29–196.99 ng g⁻¹ dw measured in the nearshore coral reef region of the Hainan Island (Xiang *et al.*, 2018) and 120–457 ng g⁻¹ dw measured in the nearshore coral reef region in the Persian Gulf (Ranjbar Jafarabadi *et al.*, 2017). Nevertheless, our measured concentrations are comparable to 2–59 ng g⁻¹ dw measured in the Kenting National Park, Taiwan (Ko *et al.*, 2014). This may be because the nearshore regions receive more terrestrial PAH pollution generated by human activities than the offshore regions.

Corals

All 16 priority PAHs were determined in the coral tissues in the Nansha Islands. The PAH concentrations ranged from 6.77 ng g⁻¹ dw (for *Stylophora danae*) in the Changxian Reef to 69.40 ng g⁻¹ dw (for *Acanthogorgia flabellum*) in the Anle Reef, averaging 19.69 ng g⁻¹ dw for all PAHs. Han *et al.* (2019) also investigated an identical set of PAH compounds excluding naphthalene in the offshore coral tissues in the Xisha and Nansha Islands of the SCS in 2015 and 2016. Combining our results with those from Han *et al.* (2019), we found: (1) the highest concentration in the SCS occurred in *Sandalolitha robusta* in the Nansha Islands (at Xian'e Reef) (Figure 3); and (2) the concentrations in the coral tissues in the Nansha Islands (averaging 74.96 ng g⁻¹ dw) were generally higher than those in the Xisha Islands (averaging 8.85 ng g⁻¹). On the other hand, the PAH concentrations in the coral tissues in the Nansha Islands were lower in this study than those measured by Han *et al.* (2019), which was probably due to the differences in the investigated coral species and sample pretreatment procedures. Nonetheless, both studies show that the PAH concentrations in the coral tissues in the offshore SCS are greatly lower than those in the nearshore region of Kenting National Park (143–1715 ng g⁻¹ dw, Ko *et al.*, 2014) and the nearshore region of the Hainan Island (333.88–727.03 ng g⁻¹ dw, Xiang *et al.*, 2018), possibly because the offshore corals are exposed to less

PAHs from intense human activities on land. The PAH concentrations in sediments from this study are comparable to those from the Kenting National Park, while the PAH concentrations in coral tissues are significantly lower than those of Kenting National Park. These results indicate that the PAH concentrations in corals are not only influenced by terrestrial PAH sources and sediments. Different coral species, various uptake behaviour, and complex metabolic processes may also influence the PAH contents in coral tissues (Ko *et al.*, 2014).

Invertebrates

The lipid contents and concentrations of Σ_{16} PAHs measured in 6 invertebrate species in the Xisha and Nansha Islands are shown in Table 1. The lipid contents in the investigated invertebrates varied from 1.25% for sea cucumbers in the Xisha Islands to 5.33% for *Tridacnidae spp* in the Nansha Islands. The PAH concentrations varied from 3.60 ng g⁻¹ dw for sleeve-fish to 28.70 ng g⁻¹ dw for sea cucumbers. Our measured PAH concentrations in the invertebrates from the offshore Xisha and Nansha Islands are significantly lower than those in cephalopod species (>100 ng g⁻¹ dw, Ke *et al.*, 2017) from the nearshore region of the northern SCS. This indicates that the offshore biota species may be less impacted by PAHs released from anthropogenic activities than nearshore species.

Fish

The PAH concentrations of the coral reef fish in this study were reported in a previous study (Li *et al.*, 2019). In general, the PAH concentrations in the Xisha reef fish ranged from 12.79 ng g⁻¹ dw in *Lethrinus erythropterus* to 409.28 ng g⁻¹ dw in *Cephalopholis argus*, averaging 74.19 ng g⁻¹ dw, while those in the Nansha reef fish ranged from 32.71 ng g⁻¹ dw in *Pterocaesio tile* to 139.09 ng g⁻¹ dw in *Scarus niger*, averaging 67.69 ng g⁻¹ dw (Table 1).

PAH composition

The composition patterns and profiles of PAHs in different compartments of the coral reef ecosystems in the SCS are shown in

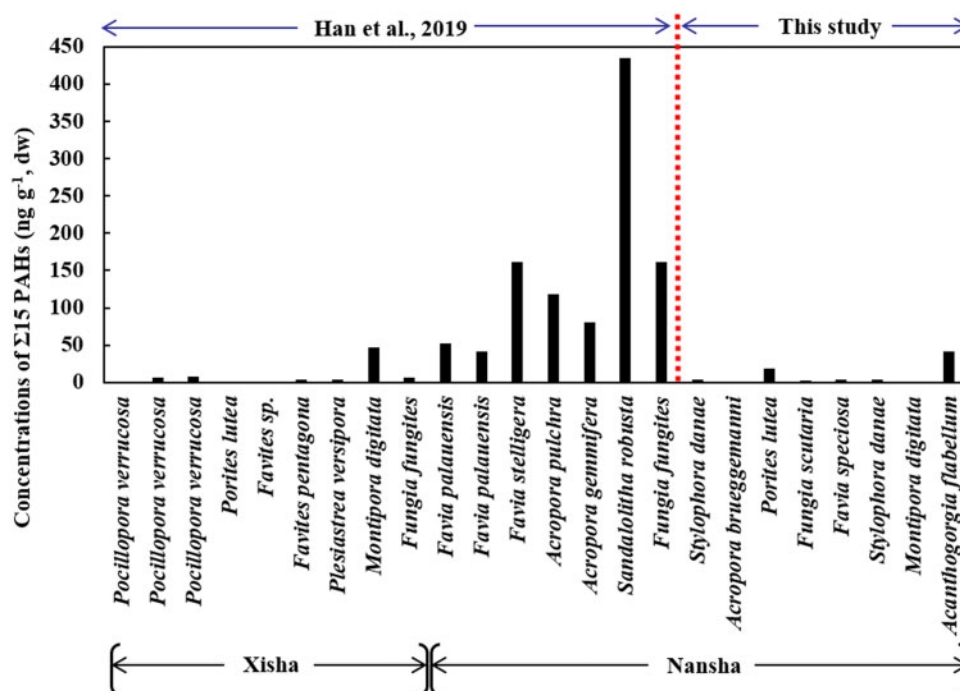


Figure 3. The PAH concentrations in the coral tissues in the Xisha and Nansha Islands measured by Han et al. (2019) and this study.

Figure 4 and Supplementary Figure S1. Four compartments comprising sediments, fish, coral tissues, and other invertebrates (excluding corals) presented different composition patterns. The LMW PAHs (*i.e.* with 2–3 benzene rings) with a total proportion of 83.57–91.62% dominated the entire coral reef ecosystem.

The predominant PAH species in the sediments was 3-ring PAHs (accounting for 76.06% of the total PAHs) (Figure 4), which is consistent with the PAH concentration in seawater reported by Han et al. (2019). 5 + 6-ring and 2-ring PAHs with the fractions of 12.34 and 11.50% ranked second and third, respectively.

Different from the PAH composition in sediments, 2-ring PAHs were dominant in coral tissues with a proportion of 63.43% (Figure 4). The proportions of 3-ring, 4-ring, and 5 + 6-ring PAHs in the total PAHs in corals were 26.37, 7.25, and 2.95%, respectively. On average, the proportion of 5 + 6-ring PAHs in coral tissues (2.95%) was lower than that found in sediments (12.34%).

The dominant PAH species in invertebrates was 3-ring PAHs (49.66%), followed by 2-ring (41.96%), 4-ring (8.32%), and 5 + 6-ring PAHs (0.07%). LMW PAHs (with 2–3 rings) dominated the PAH composition in the fish muscles: 2-ring and 3-ring PAHs accounted for 44.83 and 44.54% of the average total PAH content, respectively.

In coral reef ecosystems, almost all the sediments consist of the debris of coral tissues (Kench et al., 2005; Li et al., 2020) while corals and other invertebrates are preyed on by reef fish. The differential PAH composition patterns in the four compartments in a coral reef ecosystem indicates a necessity for further research into the trophic transfer mechanisms of PAHs.

Trophodynamics of PAHs

Food web structure

Nitrogen stable isotope ratios, feeding modes, and TLs of the biota species in the Xisha and Nansha Islands are presented in

Table 1. The TLs ranged from 1.26 for sea cucumbers to 3.77 for *Lethrinus lentjan* in the Xisha food web and from 1.77 for *Trochus maculatus* Linnaeus to 4.25 for *Scarus niger* in the Nansha food web. This result indicates that the food chain length in the Xisha Islands could be longer than that in the Nansha Islands.

Trophodynamics of PAHs

Tables 2 and 3 summarize the slope, r^2 , significance level (p) and calculated TMFs for the Xisha and Nansha food webs. The TMFs of individual PAHs are evaluated based on the differences in the regression slopes between the \log_{10} -transformed concentrations (lipid weight, lw) and the TLs of the biotas (Figure 5). Among the target PAHs, positive correlations ($p=0.05$ – 0.40) were observed between the PAH concentrations for Flu, Pyr, BaA, Chr and Bghip and TLs in the Xisha food web. Positive correlations ($p=0.08$ – 0.40) existed between the concentrations of all target PAH species and TLs in the Nansha food web (Figure 5).

According to the criterion of biomagnification in the food web (*i.e.* TMF > 1), the TMF values ranged from 0.46 (for Nap) to 2.18 (for BghiP), with an average of 1.12 in the Xisha food web, indicating a limited trophic transfer of PAHs (Table 2). The TMF values ranged from 1.07 (Ace) to 5.25 (for BbF+BkF) with an average of 1.71 in the Nansha food web (Table 3), suggesting potential biomagnification of PAHs. Despite p -values > 0.05, the TMFs in both food webs indicate low possibility of trophic biodilution in the coral reef ecosystems in the SCS. Although the average TMF is only slightly higher than 1 in the Xisha food web, biomagnification may also exist because TMFs only represent the average trophic accumulation condition based on lipid-normalized contaminant concentrations and associated TLs and may fail to capture the possible biomagnification (Verhaert et al., 2013). The TL of an organism determined by the stable nitrogen isotope ratio represents a long-term trophic status and the PAH concentration

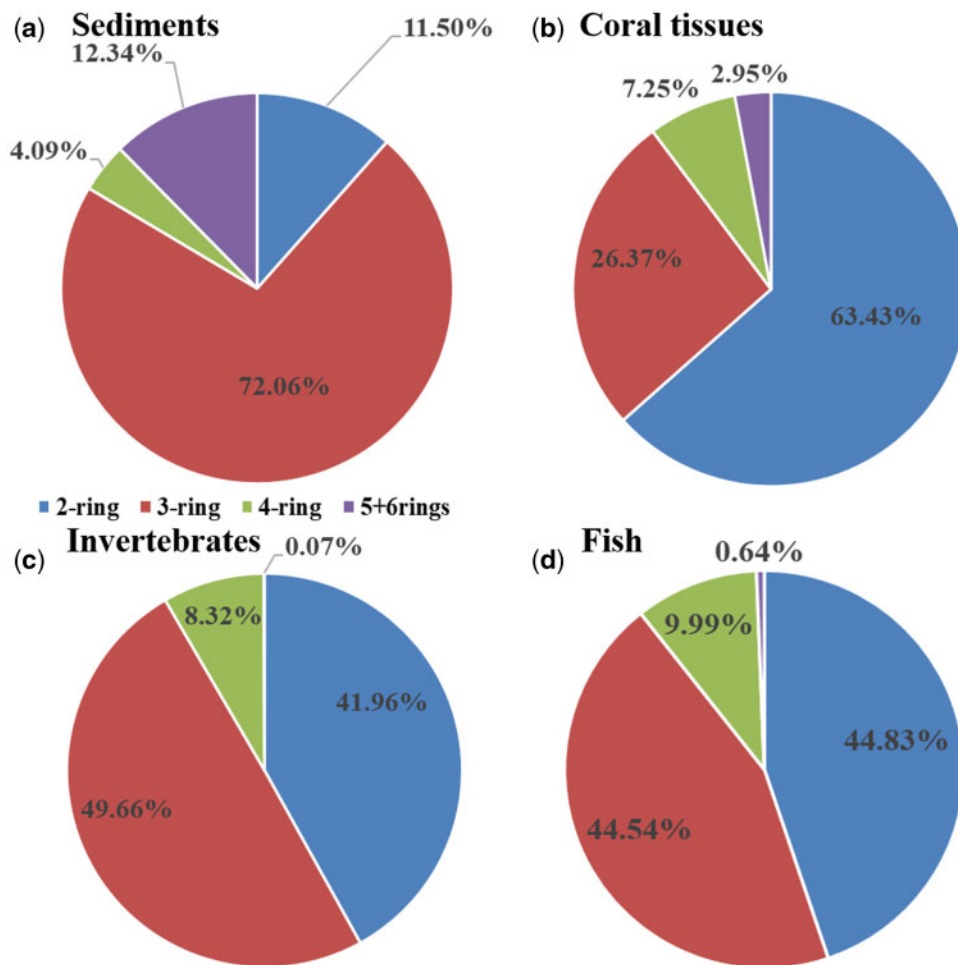


Figure 4. The composition of PAHs regarding ring size in sediments, fish, coral tissues, and invertebrates (coral excluded) in the Xisha and Nansha Islands.

Table 2. Trophic magnification factors (TMFs) of trophic levels (TLs) versus PAH concentrations in \log_{10} (ng g^{-1} , lipid weight) in the Xisha food web.

Compound	Slope	r^2	TMF	p	Log K_{ow}
Nap(X1)	-0.333	0.11	0.46	0.094	3.34
Acy (X2)	-0.1162	0.07	0.77	0.138	3.85
Ace (X3)	-0.0411	0.01	0.91	0.374	3.89
Flu (X4)	0.1647	0.04	1.46	0.218	4.14
Phe (X5)	-0.063	0.01	0.86	0.356	4.39
Ant (X6)	-0.1394	0.02	0.73	0.33	4.39
Flo (X7)	-0.0087	0.00	0.98	0.47	4.9
Pyr (X8)	0.2001	0.09	1.59	0.128	4.9
BaA (X9)	0.351	0.06	2.24	0.252	5.32
Chr (X10)	0.038	0.01	1.09	0.395	5.32
BbF+BkF (X11)	-0.0936	0.01	0.81	0.276	5.95
BaP (X12)	-0.306	0.19	0.49	0.282	5.95
BghiP (X13)	0.3379	0.09	2.18	0.054	6.45

Table 3. Trophic magnification factors (TMFs) of trophic levels (TLs) versus PAH concentrations in \log_{10} (ng g^{-1} , lipid weight) in the Nansha food web.

Compound	Slope	r^2	TMF	p	Log K_{ow}
Nap (N1)	0.11	0.0733	1.28	0.210	3.34
Acy (N2)	0.08	0.0421	1.20	0.272	3.85
Ace (N3)	0.03	0.0074	1.07	0.401	3.89
Flu (N4)	0.05	0.0149	1.12	0.360	4.14
Phe (N5)	0.09	0.0474	1.24	0.260	4.39
Ant (N6)	0.23	0.0762	1.71	0.220	4.39
Flo (N7)	0.15	0.1069	1.43	0.163	4.9
Pyr (N8)	0.06	0.0185	1.15	0.345	4.9
BaA (N9)	0.23	0.1798	1.70	0.111	5.32
Chr (N10)	0.20	0.0952	1.57	0.078	5.32
BbF+BkF (N11)	0.72	0.2873	5.25	0.080	5.95
BaP (N12)	0.38	0.1498	2.39	0.120	5.95
DahA (N13)	0.07	0.0161	1.19	0.372	6.5

measured from an organism tissue is the counteractive result of PAH uptake and elimination. Moreover, the PAH concentration may be higher in other fatty tissues (e.g. the gut) (Bandowe *et al.*, 2014), while only the PAHs in muscle tissues were analyzed in

this study. Thus, the TMFs calculated in this study may underestimate the actual biomagnification level in the SCS.

Trophic transfer characteristics are important in the health and environmental risk assessment, and thus attracting intensive

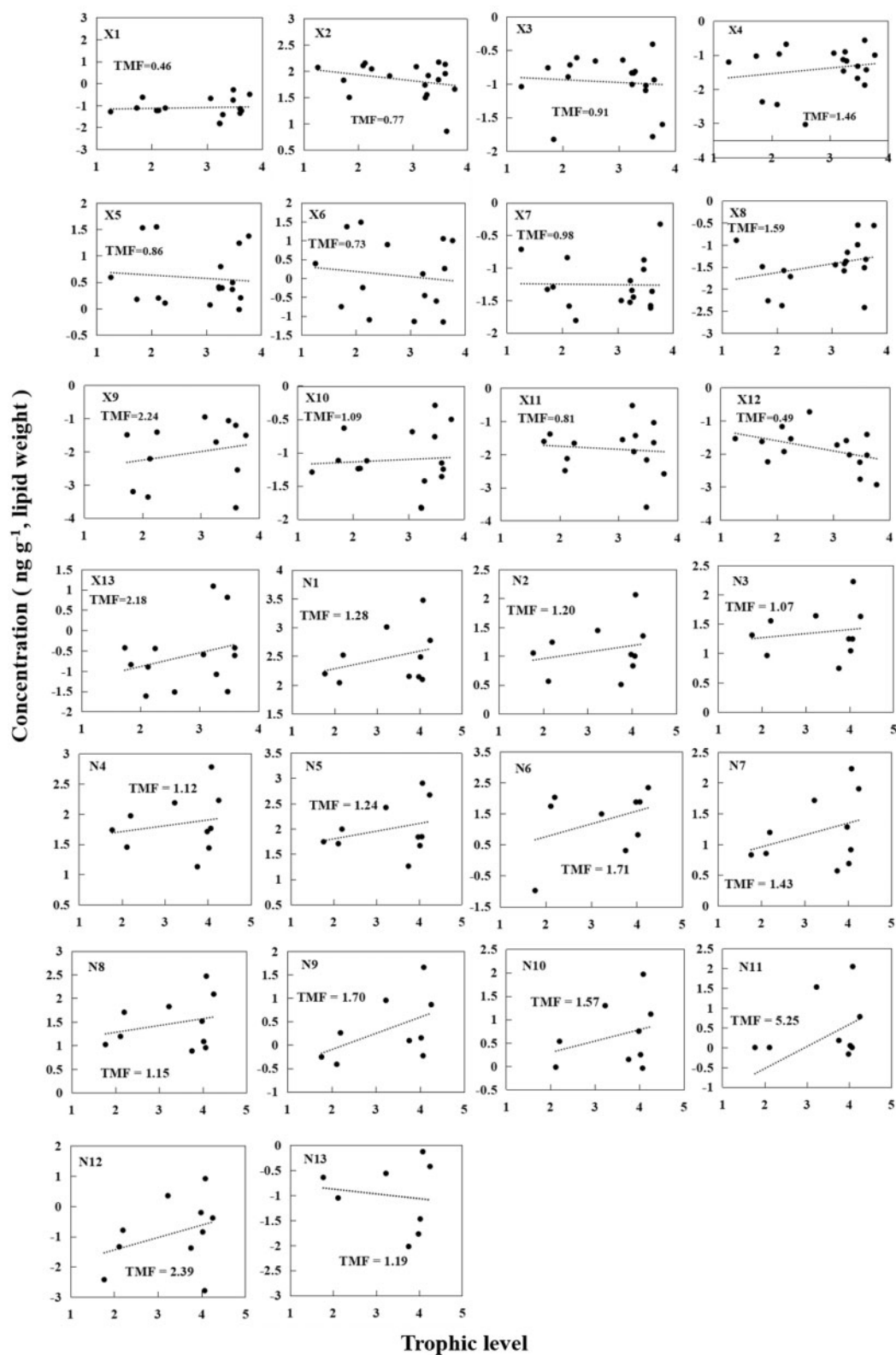


Figure 5. Relationships between the PAH concentrations (lw, ng g⁻¹) and trophic levels for the Xisha and Nansha food webs. The figure numbers of X1–X13 and N1–N13 correspond to the PAH species in Table 3.

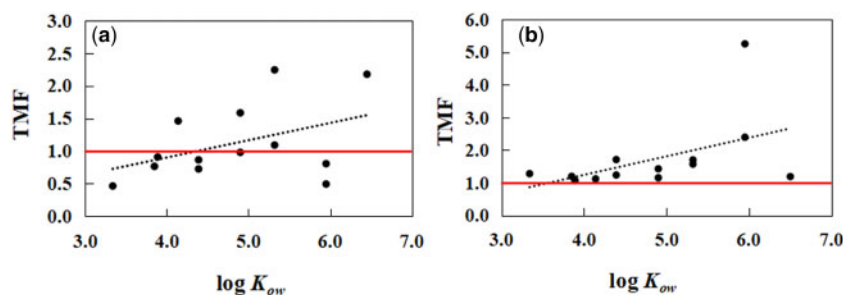


Figure 6. Relationships between the $\log K_{ow}$ values and TMFs of PAHs in the coral reef food webs of (a) the Xisha and (b) Nansha Islands. The $\log K_{ow}$ values were obtained from Han *et al.* (2019). The red line represents the TMF value equal to 1 and the black dotted line represents the linear regression line between the TMF and the $\log K_{ow}$.

research efforts (Arnot and Gobas, 2006). Currently, both biodilution and biomagnification of PAH trophic transfer in aquatic food webs have been reported in the limited existing studies. Wan *et al.* (2007) reported the trophic biodilution of PAHs with TMFs ranging from 0.11 to 0.45 in the coastal seawater food web in the Bohai Bay, North China. Similar biodilution phenomena were also observed by Tao *et al.* (2018) and Qadeer *et al.* (2019) in some freshwater food webs. In contrast, Wang *et al.* (2012) reported biomagnification and trophic transfer of PAHs with TMFs values ranging from 0.91 to 1.90 ($p > 0.05$) in the Taihu Lake, North China. The biomagnification phenomenon was further supported by laboratory exposure experiments for the Atlantic and Pacific marine ecosystems carried out by Arias *et al.* (2016) and mathematical modelling for a freshwater food chain in the Southern Brazil by Froehner *et al.* (2011). Clarification of the ambiguity in the trophic transfer characteristics of PAHs will significantly help assess their fate and potential effects on an aquatic ecosystem (Qadeer *et al.*, 2019).

The average TMF values of PAHs in the food webs of the two targeted coral reef systems in the offshore SCS determined in this study are higher than those reported in other aquatic environments and indicate a potential biomagnification effect. The differences between TMFs observed in the offshore SCS coral reef food webs and those in other aquatic ecosystems are possibly attributed to the differences in ecosystem characteristics, such as ecosystem type and scale, dominant species, food-web structure, habitat environment, and feeding habits, as well as potential of biotransformation or metabolism of PAHs in different organisms (Walters *et al.*, 2016; Sun *et al.*, 2017). Consequently, a combination of these influencing factors may contribute to the trophic transfer of PAHs in an aquatic ecosystem and a certain level of biomagnification (Bekele *et al.*, 2019; Liu *et al.*, 2019). Environmental conditions, such as dissolved organic matter, suspended particle concentrations and water temperature, may also influence the trophodynamics of chemicals within an ecosystem (Sun *et al.*, 2017; Wang *et al.*, 2019). The biotransformation/metabolism of PAHs within the organisms and their potential risks to biotas and human health require further investigations.

$\log K_{ow}$ is frequently used to represent the biomagnification potential of hydrophobic compounds in aquatic food webs (Wan *et al.*, 2007; Bekele *et al.*, 2019; Wang *et al.*, 2019). K_{ow} of PAHs was thus examined in this study regarding the biomagnification of PAHs in marine food webs. In Figure 6, the TMFs of all PAHs are plotted against $\log K_{ow}$. The TMFs of PAHs in both the Xisha and Nansha ecosystems and the $\log K_{ow}$ ($p > 0.05$) are positively correlated. Our results differ from those observed in the Bohai

Bay of China, where the TMFs and $\log K_{ow}$ values are not correlated (Wan *et al.*, 2007), but agree with the observation in the Taihu Lake food web (Wang *et al.* 2012), where the TMFs and $\log K_{ow}$ show a positive linear relationship with a marginal statistical significance ($p = 0.057$). The positive relationships between the TMFs and $\log K_{ow}$ of PAHs indicate that chemical properties, such as lipophilicity, may have important effects on the chemical trophic transfer in a food chain.

Conclusions

This study provides the latest scientific finding and a comprehensive assessment of pollution status, composition patterns, and trophic transfer of PAHs in the coral reef ecosystems of the Xisha and Nansha Islands in the SCS. The PAH status was investigated in sediment, coral, and other biota samples from the coral reef ecosystems. Stable isotope analyses and the TMF method were adopted to further clarify the trophic transfer behaviour of PAHs. The concentration of the total PAHs in sediments (9.00–32.48 ng g^{-1} dw), corals (6.77–69.40 ng g^{-1} dw), invertebrates (3.60–28.70 ng g^{-1} dw), and fish (12.79–409.28 ng g^{-1} dw) were determined. The TMFs of the Xisha food web ranged from 0.46 to 2.18, indicating a limited trophic transfer of PAHs, while the TMFs of the Nansha food web ranged from 1.07 to 5.25, indicating a trophic magnification of PAHs. The reasons for differences in trophodynamics among ecosystems may be attributed to different ecosystem characteristics (*e.g.* the food chain length). Octanol-water partition coefficient (K_{ow}) of PAHs may have positive influences on the PAH biomagnification. This study provides the first insight into the trophic transfer characteristics of PAHs in highly diverse coral reef ecosystems and reveals their biomagnification phenomena along TLs. These results may serve as a scientific basis for ecological risk assessment, biodiversity conservation, and coastal function management in global coral reef systems and lay a foundation for future studies on the partitioning of PAHs within the organisms, transformation *in vivo* and potential risks to biotas and human health.

Supplementary data

Supplementary material is available at the ICESJMS online version of the manuscript.

Data availability

The data underlying this article are available in the article and in its online supplementary material.

Acknowledgements

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