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Bioaccumulation of organic pollutants in Indo-Pacific humpback dolphin: A review on current knowledge and future prospects

Edmond Sanganyado, Imran Rashid Rajput, Wenhua Liu*

Marine Biology Institute, Shantou University, Shantou, Guangdong Province, China

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ABSTRACT

Indo-Pacific humpback dolphin (*Sousa chinensis*) are chronically exposed to organic pollutants since they inhabit shallow coastal waters that are often impacted by anthropogenic activities. The aim of this review was to evaluate existing knowledge on the occurrence of organic pollutants in Indo-Pacific humpback dolphins, identify knowledge gaps, and offer recommendations for future research directions. We discussed the trends in the bioaccumulation of organic pollutants in Indo-Pacific humpback dolphins focusing on sources, physicochemical properties, and usage patterns. Furthermore, we examined factors that influence bioaccumulation such as gender, age, dietary intake and tissue-specific distribution. Studies on bioaccumulation in Indo-Pacific humpback dolphin remain scarce, despite high concentrations above 13,000 ng/g lw we previously detected for PFOS, ∑PBDE and chlorinated paraffins. The maximum concentration of organochlorines detected was 157,000 ng/g wt. Furthermore, variations in bioaccumulation were shown to be caused by factors such as usage patterns and physicochemical properties of the pollutant. However, restrictions in sampling inhibit investigations on exposure pathway and toxicity of organic pollutants in Indo-Pacific humpback dolphin. We proposed the use of biopsy sampling, predictive bioaccumulation and toxicity modeling, and monitoring other emerging contaminants such as microplastics and pharmaceuticals for future health risk assessment on this critically endangered marine mammal species.

1. Introduction

Anthropogenic pollutants pose a risk to marine mammals that reside in coastal waters. Discharge of organic pollutants into marine environments has been shown to decrease water quality resulting in loss of habitats and a significant reduction in the species richness (Johnston and Roberts, 2009). The habitat loss and corresponding fragmentation often lead to isolation of marine mammal populations (Deng et al., 2016; Lai et al., 2016), and the resulting decrease in connectivity can cause population decline (Huang et al., 2012; Johnston and Roberts, 2009). Therefore, there is a need for systematic studies on the occurrence, fate, bioaccumulation and toxicity of organic pollutants in marine ecosystems.

Indo-Pacific humpback dolphins (*Sousa chinensis* Osbeck, 1765), also known as Chinese white dolphin, is a medium-sized and robust delphinid widely distributed in estuarine and inshore waters of Indian and western Pacific Ocean and also occurring along the southeastern coastal waters of China (Lin et al., 2016). Recent studies identified five fragmented colonies of this species between Xiamen and Pearl River Estuary (PRE) (Xu et al., 2015). The largest colony of <2000 dolphins dwells in the Pearl River Estuary, which receives huge amounts of organic and inorganic pollutants discharged from the Pearl River Delta, the hub of urbanization and industrialization in South Asia (Huang et al., 2012). In fact, the population of Indo-Pacific humpback dolphins is rapidly declining at an estimated annual rate of 2.46%; suggesting that by 2080, approximately 75% of the current population will be lost (Gui et al., 2017). For that reason, Indo-Pacific humpback dolphins are classified as near threatened on the IUCN Red List of Threatened Species (Lai et al., 2016).

Several studies detected POPs and pollutants of emerging concern in Indo-Pacific humpback dolphins (Gui et al., 2014a; Minh et al., 1999; Wu et al., 2013). These organic pollutants have been shown to cause adverse effects in marine mammals such as disease susceptibility, reproductive and developmental toxicity (Fig. 1) (Bossart, 2011). However, although understanding the exposure of Indo-Pacific dolphins to organic pollutants is critical in risk

* Corresponding author.
E-mail address: whliu@stu.edu.cn (W. Liu).

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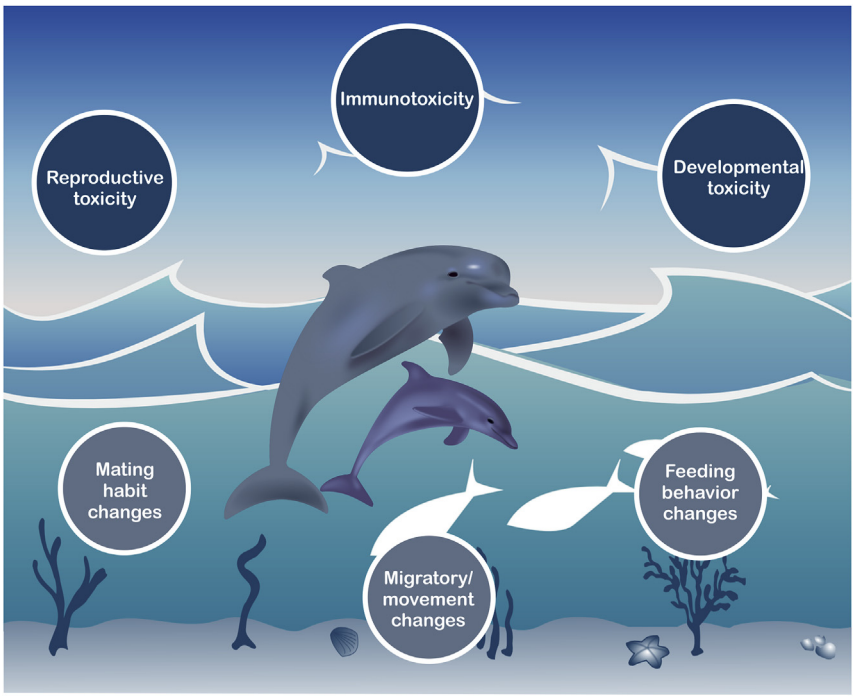

Fig. 1. Potential adverse environmental effects of exposure to organic pollutants in Indo-Pacific humpback dolphins (*Sousa chinensis*).

assessment, there are no reviews summarizing current knowledge. Hence, the aim of this paper is to critically review current data on bioaccumulation of POPs and emerging pollutants in Indo-Pacific humpback dolphin. Furthermore, we identified knowledge gaps and offered recommendation for future research direction. To put the work in context, we also reviewed the state of science regarding the health effects of organic pollutants in Indo-Pacific humpback dolphins. Using the search query "*Sousa chinensis*" on SCOPUS and ISI Web of Science, 279 research articles and reviews were identified (Fig. S1). Following manual screening, we identified 22 publications that focused on bioaccumulation of organic pollutants. However, some studies tended to report the same data twice, hence duplicate data was removed, for example, Minh et al. (1999, 2000).

In this review, we answered the following questions:

1. What are the sources and trends in the amounts of the organic pollutants in Indo-Pacific humpback dolphin?
   a. Legacy pollutants;
   b. Emerging contaminants
2. What the factors that influence bioaccumulation of organic pollutants in Indo-Pacific humpback dolphin?
   a. Intrinsic properties of the contaminant
   b. Anatomy and life history of the Indo-Pacific humpback dolphin
3. What are the knowledge gaps and suggestions for future research?

2. Regulatory frameworks for coastal marine pollution

Since the adoption of the Stockholm Convention, China continues to actively monitor organic pollutants in marine environments. In 1982, the Marine Environmental Protection Law was passed in China to protect marine ecosystems such as the PRE and it was revised in 1999 (Wang et al., 2012). Furthermore, in 2001 administrative regulations and standards were set for near coastal seawaters and technical directives were issued in 2006 (Deng et al., 2016; Tiquio et al., 2017). However, to date, no standards have been set for estuarine zones and the Ministry of Water Resources mandated a Water Management Commission for the Yangtze River and Pearl River estuaries (Deng et al., 2016).

Establishing an emission inventory of organic pollutants is critical in developing regulatory frameworks and source apportionment. In the past ten years, emissions inventories have been established for PFASs, PCBs and PCDDs. Only the usage inventories of pesticides, such as endosulfan, lindane, HCHs and chlordane have been determined at the time of writing (Liu et al., 2016). Furthermore, emissions inventory can be used to predict exposure of marine mammals to organic pollutants. However, there are no emissions inventories developed for most emerging contaminants such as halogenated flame retardants or plasticizers.

The development of effective regulatory frameworks for organic pollutants in marine environments require improvements in chemical and toxicological analysis. The knowledge of exposure to organic pollutants came through analysis of stranded Indo-Pacific humpback dolphins primarily (Gui et al., 2014a; Minh et al., 2000; Wu et al., 2013; Zeng et al., 2015). In a risk assessment study, Guo et al. (2011) observed an increase in the ecological risk of 28.7% for Indo-Pacific humpback dolphins in Xiamen coastal waters between 1996 and 2006. The change in ecological risk corresponded with the urbanization and economic development in the region (Guo et al., 2011). However, between 2006 and 2007, the ecological risk decreased by 15.7% probably due to improvements in wastewater treatment, decline in aquaculture activities and increased environmental consciousness (Guo et al., 2011).

3. Current knowledge on bioaccumulation of organic pollutants

3.1. Mechanism of bioaccumulation in marine mammals

Environmental risk of organic pollutants is often assessed by

determining their persistence, bioaccumulation and toxicity. Indo-Pacific humpback dolphins have a long lifespan that makes them susceptible to chronic exposure to POPs. Furthermore, their high lipid content suggests POPs can easily accumulate in their tissues (Bossart, 2011). However, the amount of organic pollutant that accumulates in the Indo-Pacific humpback dolphins over time could cause significant adverse effects at molecular, tissue, organismal, population, and ultimately ecosystem level (Weisbrod et al., 2009). Hence, understanding bioaccumulation in marine mammals is critical since it influences the internal dose of the potential toxicant (Arnot and Gobas, 2004). However, the concentration in the organisms indicates the balance between uptake, metabolism and elimination of the organic pollutants (Fig. 2). Upon absorption, organic pollutants are distributed to different tissues within the organism where they accumulate and possibly elicit an adverse effect or are metabolized and excreted (Ahrens et al., 2009; Shaw et al., 2012; Waugh et al., 2014). Hence, understanding the toxicokinetics and toxicodynamics of organic pollutants in Indo-Pacific humpback dolphins is important in establishing their toxicological risk.

Bioaccumulation is defined as the uptake of chemicals from the environment through dietary intake, dermal absorption or respiratory transport in air or water (Mackay et al., 2013). In contrast, bioconcentration is the uptake of chemicals from the aqueous environments via dermal absorption and respiratory transport. Hence, bioaccumulation can be considered a combination of dietary intake and bioconcentration (MacKay and Fraser, 2000). Bioaccumulation is often determined using accumulation monitoring in field or laboratory studies, *in vivo* and *in vitro* ADME studies or food web models. Accumulation monitoring is the most widely used technique whereby the amount of organic pollutants is determined in the target organism (C_B) or its prey (C_D). For example, the mean concentrations of PCBs, HCHs and DDTs in *Collichtys lucida*, a prey fish for Indo-Pacific humpback dolphins were found to be 43.2, 4.3 and 4.0 ng/g (Hung et al., 2006a) but 19,979, 1164.7 and 9110.5 ng/g in the predator, Indo-Pacific humpback dolphins (Leung et al., 2005).

Bioaccumulative potential of organic pollutants are determined using the bioconcentration factor (BMF), biomagnification factor (BCF), or bioaccumulation factor (BAF), which are ratios of concentration in organism to concentration in water C_W, diet C_D or both C_T, respectively (Table 1). A study found the BMFs of organochlorines in Indo-Pacific humpback dolphins from PRE were in the order aldrin < HCHs < heptachlor < endrin < CHLs < HCB < dieldrin < PCBs < mirex < pentachlorobenzene < DDTs with values ranging from 3.4 ± 3.4 to 212 ± 160 (Fig. 3) (Gui et al., 2014a). However, there are few studies that determined biouptake factors since most of the dolphins studied were stranded making it challenging to establish concentrations of organic pollutants in their diet or in the exact coastal waters they inhabited.

3.2. Sources and temporal trends in legacy pollutants

Mean concentrations of legacy contaminants and emerging contaminants are shown on Table 2 and Table 3, respectively.

3.2.1. Polycyclic aromatic hydrocarbons

The occurrence of PAHs in marine environments is an issue of major concerns since they are highly persistent, bioaccumulative and toxic. Some PAHs such as benzo(*b*)fluoranthene, benzo[*a*]pyrene and benzo[e]pyrene, are known carcinogens. Furthermore, PAHs enter the marine environment through natural activities such as volcanic eruptions and forests fires or human activities that involve combustion (Bigus et al., 2014). The transport and partitioning of PAHs in different environmental compartments is greatly influenced by their molecular weight. However, the molecular weight is often determined by the combustion temperature; low-molecular weight PAHs are often a result of low-temperature combustion, whereas high-molecular weight PAHs originate from high-temperature combustion such as industrial incineration (Cagnazzi et al., 2013). Thus, a ratio of the low molecular weight PAHs to high molecular weight PAH can be used as a chemical indicator in source apportionment (Bigus et al., 2014). Several studies detected PAHs in different marine samples suggesting Indo-Pacific humpback dolphin could be at risk (Cagnazzi et al., 2013; Leung et al., 2005). A study on effect of geographical distribution on

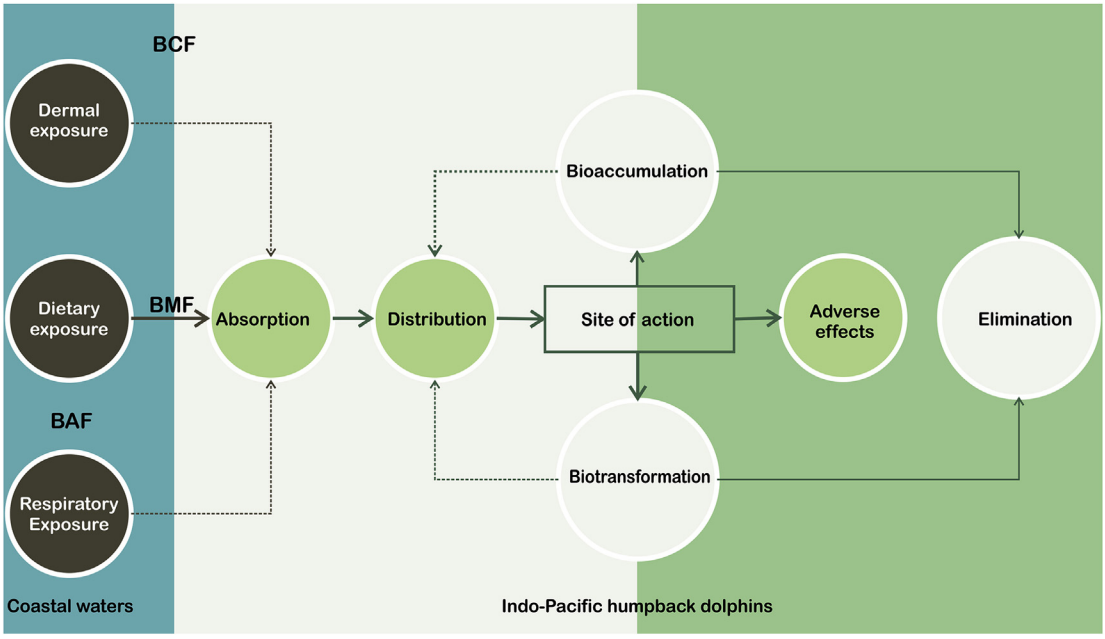

Fig. 2. Mechanism of bioaccumulation of organic pollutants in Indo-Pacific humpback dolphin (*Sousa chinensis*).

Table 1
Definitions and screening criteria for bioconcentration factor (BCF), biomagnification factor (BMF), bioaccumulation factor (BAF), and trophic magnification factor (TMF).

Uptake factor	Definition	Equation	Screening criteria[a]
BCF	Ratio of concentration in organism C_B, to the total concentration in water C_W, or the freely dissolved concentration in water C_f.	$BCF = \frac{C_B}{C_W}$ or $\frac{C_B}{C_f}$	5000 L/kg
BMF	Ratio of concentration in organism C_B to total concentration in diet C_D.	$BMF = \frac{C_B}{C_D}$	–
BAF	Ratio of concentration in organism C_B to total concentration in diet and water C_T.	$BAF = \frac{C_B}{C_T}$	5000 L/kg
TMF	Average change of the BMF (increase means bioaccumulation and decrease means biodilution) per trophic level (TF) in a food web.	$TMF = \frac{\sum_{i=TF}^{n} BMF_i}{n}$	1

[a] Chemicals with uptake factors in marine organisms above these values are considered bioaccumulative (Weisbrod et al., 2009).

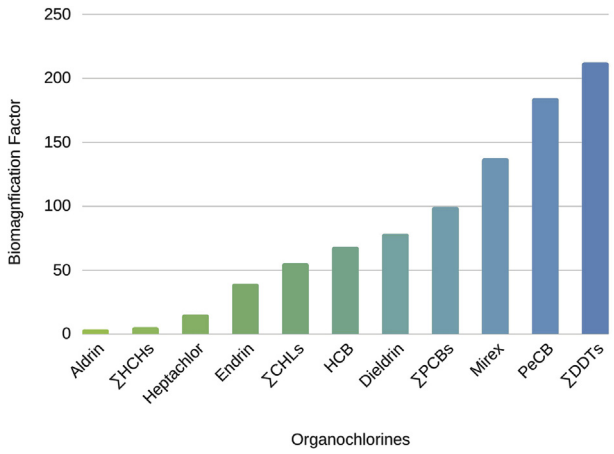

Fig. 3. Biomagnification factors (BMFs) for individual organochlorines in Indo-Pacific humpback dolphins (*Sousa chinensis*) from the PRE, China (Gui et al., 2014a).

bioaccumulation found the median concentration of PAHs in Indo-Pacific humpback dolphin was highest in Xiamen, followed by Hong Kong and Zhuhai, 6,680, 2990 and 2760 ng/g, respectively (Leung et al., 2005). High concentrations of PAHs in Xiamen were attributed to petroleum storage, oil refineries, power stations and wastewater plants within the city and its surrounding areas (Leung et al., 2005).

The source of PAHs that bioaccumulate in marine mammals can be determined using chemical signatures. The ratio of concentration of anthracene to the sum of concentrations of anthracene and phenanthrene is commonly used in source apportionment where a ratio of <0.1 indicates the source of PAHs is petrogenic while above 0.1, it will be pyrogenic (Jiang et al., 2009). A study in Hong Kong found a ratio of 0.98, suggesting the bioaccumulated PAHs originated from a pyrogenic source in Hong Kong. However, the ratio of fluoranthene and pyrene indicate these two compounds in Hong Kong and Zhuhai probably came from petrogenic sources, whereas in Xiamen it was probably from coal, grass and wood combustion (Leung et al., 2005). The ratios were 0.07, 0.39 and 0.70, where a ratio of <0.4 indicates petrogenic source and >0.4 biomass combustion (Jiang et al., 2009). Furthermore, the total concentration of carcinogenic PAHs varied with location. In Xiamen, carcinogenic PAHs comprised of 44.8% of total PAHs, whereas in Hong Kong and Zhuhai it was only 16.8% and 27.1% (Leung et al., 2005). However, additional source apportionment studies using isomer ratios are required to better understand exposure pathways.

3.2.2. Polychlorinated biphenyls

Although use of PCBs has been banned or restricted globally, several studies have detected them abundantly in various environmental compartments in the marine ecosystem (Minh et al., 1999; Wu et al., 2013). Before the ban, PCBs were used in heat exchangers, transformers and hydraulic cylinders because of their high chemical and thermal stability (Bigus et al., 2014). The chemical structure of PCBs comprising two phenyl groups with a varying degree of chlorine substitution. Varying the degree of chlorination and position of chlorines on the phenyl rings yield about 209 unique compounds called congeners. Several studies in China detected PCBs in marine sediment, coastal waters and biota. Hence, Indo-Pacific humpback dolphins are exposed to PCBs.

Data on bioaccumulation of PCBs in Indo-Pacific humpback dolphin is available from 1993 to 2014. A comparison of the published data indicated that the concentration of ∑PCBs in Indo-Pacific humpback dolphin sampled from Pearl River Delta were significantly decreasing with time. For example, samples from 2004 to 2009 had a higher mean concentration of ∑PCBs than samples from 2001 to 2014, with mean concentration of 3250 ± 2880 (Wu et al., 2013) and 1790 ± 2880 ng/g, respectively (Gui et al., 2014b). A significant decrease in average concentration of ∑PCBs was observed between 2004 − 2009 and 1995–2001 ($p < 0.05$) (Minh et al., 1999; Wu et al., 2013). Furthermore, spatial variation in bioaccumulation of PCBs in Indo-Pacific humpback dolphin has been previously reported. Blubber samples from Hong Kong had the highest concentration of ∑PCBs, followed by Xiamen and Zhuhai at 19,978.6 ng/g, 251.4 ng/g and 69 ng/g, respectively.

3.2.3. Organochlorine pesticides

Organochlorine pesticides are a group of highly persistent and bioaccumulative compounds that were globally banned because of their high toxicity. They were predominantly used in agriculture and households for pest control, and examples include DDT, aldrin, endrin and chlordane. A sampling survey conducted between 2004 and 2014 found the mean concentration of organochlorines in blubber samples of Indo-Pacific humpback dolphins from Pearl River Estuary followed the order: DDTs > HCHs > mirex > endrin > CHLs > HCB > dieldrin > aldrin > heptachlor > pentachlorobenzene (Gui et al., 2014a). Between 1993 and 2014, the concentration of ∑DDTs in Indo-Pacific humpback dolphins did not significantly change suggesting the pollutants had not been used recently (Gui et al., 2014a; Minh et al., 1999; Wu et al., 2013). Differences in spatial distribution were observed in an earlier study with concentration in samples from Hong Kong, Zhuhai and Xiamen varying from 9,000, 44,000 to 60,000 ng/g, respectively (Leung et al., 2005). The concentration of HCHs was higher in Hong Kong than in Xiamen or Zhuhai whereas HCB was higher in Zhuhai than in the other two sites (Leung et al., 2005). However, additional studies are required to ascertain spatial trends in bioaccumulation of organochlorine pesticides.

3.3. Sources and temporal trends of emerging contaminants

3.3.1. Brominated flame retardants

Brominated flame retardants are structurally diverse class of compounds commonly added to polymer materials such as electronic equipment and plastics to decrease flammability. In the past

Table 2
Bioaccumulation of legacy contaminants in Indo-Pacific humpback dolphin.

Pollutant	Location	Animal					Analysis		Reference
		Period	Status	Organ	Length, cm	Lipid %	Technique	Mean conc. ng/g, wt	
∑PCBs	Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	33 ± 19	GC-μECD	31,000 ± 28,000	(Ramu et al., 2005)
	Hong Kong	1993–1997	n.r.	Blubber	164 ± 63	49 ± 19	GC–ECD	46 ± 23	(Minh et al., 1999)
	Parangipettai, India	1997	n.r.	Blubber	150	55	GC-ECD	1220	(Karuppiah et al., 2005)
	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	19,979	(Leung et al., 2005)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	251.4	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	69.4	(Leung et al., 2005)
	Great Barrier Reef, Australia	2009–2010	1	Skin	n.r.	n.r.	HRC-GC-ECD	10,382 ± 5308	(Cagnazzi et al., 2013)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	1790 ± 2880	(Gui et al., 2014a)
HCB	Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	33 ± 19	GC-μECD	282 ± 189	(Ramu et al., 2005)
	Hong Kong	1993–1997	n.r.	Blubber	164 ± 63	49 ± 19	GC–ECD	71 ± 68	(Minh et al., 1999)
	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	42.7	(Leung et al., 2005)
	Xiamen	2002–2004		Blubber	198 ± 65	n.r.	GC-μECD	62.7	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	90.1	(Leung et al., 2005)
	Great Barrier Reef, Australia	2009–2010	1	Skin	n.r.	n.r.	HRC-GC-ECD	6.3 ± 3.1	(Cagnazzi et al., 2013)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	63.7 ± 60.3	(Gui et al., 2014a)
∑DDTs	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	9110.5	(Leung et al., 2005)
	Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	33 ± 19	GC-μECD	157,000 ± 139,000	(Ramu et al., 2005)
	Hong Kong	1993–1997	n.r.	Blubber	164 ± 63	49 ± 19	GC–ECD	24 ± 17	(Minh et al., 1999)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	59,542	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	44,367	(Leung et al., 2005)
	Parangipettai, India	1997	n.r.	Blubber	150	55	GC-ECD	19,970	(Karuppiah et al., 2005)
	Great Barrier Reef, Australia	2009–2010	1	Skin	n.r.	n.r.	HRC-GC-ECD	2132 ± 450	(Cagnazzi et al., 2013)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	62700 ± 57700	(Gui et al., 2014a)
∑HCHs	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	1164.7	(Leung et al., 2005)
	Hong Kong	1993–1997	n.r.	Blubber	164 ± 63	49 ± 19	GC–ECD	379 ± 239	(Minh et al., 1999)
	Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	33 ± 19	GC-μECD	657 ± 627	(Ramu et al., 2005)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	227.6	(Leung et al., 2005)
	Zhuhai	2003		Blubber	n.r.	n.r.	GC-μECD	32.8	(Leung et al., 2005)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	960 ± 1140	(Gui et al., 2014a)
	Parangipettai, India	1997	n.r.	Blubber	150	55	GC-ECD	230	(Karuppiah et al., 2005)
∑PAHs	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	3274.6	(Leung et al., 2005)
	Xiamen	2002–2004		Blubber	198 ± 65	n.r.	GC-μECD	6751	(Leung et al., 2005)
	Zhuhai	2003		Blubber	n.r.	n.r.	GC-μECD	2761.6	(Leung et al., 2005)
	Great Barrier Reef, Australia	2009–2010	1	Skin	n.r.	n.r.	LC-FD	51,035 ± 5227	(Cagnazzi et al., 2013)
car∑PAHs	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	550.5	(Leung et al., 2005)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	3026.2	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	750	(Leung et al., 2005)
	Great Barrier Reef, Australia	2009–2010	1	Skin	n.r.	n.r.	LC-FD	2983 ± 1012	(Cagnazzi et al., 2013)
CHLs	Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	33 ± 19	GC-μECD	1860 ± 1980	(Ramu et al., 2005)
	Hong Kong	1993–1997	n.r.	Blubber	164 ± 63	49 ± 19	GC–ECD	756 ± 705	(Minh et al., 1999)
	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	258.6	(Leung et al., 2005)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	249.5	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	221.7	(Leung et al., 2005)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	85.9 ± 81.5	(Gui et al., 2014a)
Mirex	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63.4	47 ± 20	GC-MS	232 ± 233	(Gui et al., 2014a)
Heptachlor	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	93.7	(Leung et al., 2005)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	142.6	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	32.3	(Leung et al., 2005)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	16.7 ± 11.7	(Gui et al., 2014a)
Aldrin	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	39.7	(Leung et al., 2005)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	12	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	n.d.	(Leung et al., 2005)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	14.8 ± 20.9	(Gui et al., 2014a)
Pentachlorobenxene	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	13.9 ± 13.3	(Gui et al., 2014a)
Dieldrin	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	2703.1	(Leung et al., 2005)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	5149.3	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	n.d.	(Leung et al., 2005)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	58.8 ± 55.2	(Gui et al., 2014a)
Endrin	Hong Kong	n.r.	1	Skin	n.r.	n.r.	GC-μECD	624.9	(Leung et al., 2005)
	Xiamen	2002–2004	n.r.	Blubber	198 ± 65	n.r.	GC-μECD	991.4	(Leung et al., 2005)
	Zhuhai	2003	n.r.	Blubber	n.r.	n.r.	GC-μECD	758.7	(Leung et al., 2005)
	Pearl River Estuary	2004–2014	2, 3	Blubber	181 ± 63	47 ± 20	GC-MS	109 ± 93.2	(Gui et al., 2014a)
TCPMe	Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	33 ± 19	GC-μECD	138 ± 158	(Ramu et al., 2005)
	Hong Kong	1993–1997	n.r.	Blubber	164 ± 63	49 ± 19	GC–ECD	99 ± 90	(Minh et al., 1999)
TCPMOH	Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	33 ± 19	GC-μECD	87 ± 78	(Ramu et al., 2005)
	Hong Kong	1993–1997	n.r.	Blubber	164 ± 63	49 ± 19	GC–ECD	134 ± 84	(Minh et al., 1999)

Condition was defined by veterinarians and officials of the Agriculture, Fisheries and Conservation, Hong Kong, as follows: 1 = live; 2 = freshly dead; 3 = moderately decomposing; 4 = advanced decomposing; 5 = mummified remains.

Table 3
Bioaccumulation of emerging contaminants in Indo-Pacific humpback dolphin.

Pollutants		Location	Samples					Chemical analysis		Reference
Class	Pollutant(s)		Period	Status	Organ	Length, cm	Lipid %	Technique	Conc. range, ng/g lw.	
HFR	∑PBDE	South China Sea	2003–2011	1–4	Blubber	242 ± 18	35.5 ± 17.9	LC-MS/MS QTrap	691–41,900	(Zhu et al., 2014)
		Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	32 ± 19	GC-MS	n.d. – 6000	(Ramu et al., 2005)
		Hong Kong	2002–2008	2–4	Blubber	174 ± 64	31 ± 15	GC-MS	280–51,100	(Lam et al., 2009)
	BTBPE	South China Sea	2003–2011	1–4	Blubber	242 ± 18	35.5 ± 17.9	LC-MS/MS QTrap	0.0223–0.44	(Zhu et al., 2014)
	DBDE	South China Sea	2003–2011	1–4	Blubber	242 ± 18	35.5 ± 17.9	LC-MS/MS QTrap	0.214–10	(Zhu et al., 2014)
	∑HBCD	Hong Kong	2002–2008	2–4	Blubber	174 ± 64	31 ± 15	GC-MS	32–519	(Lam et al., 2009)
		Hong Kong	1997–2001	n.r.	Blubber	191 ± 66	28 ± 14	LC triple quadruple MS	31–180	(Isobe et al., 2007)
	TBPH	South China Sea	2003–2011	1–4	Blubber	242 ± 18	35.5 ± 17.9	LC-MS/MS QTrap	<0.02–7.55	(Zhu et al., 2014)
		Hong Kong	2002–2008	2–4	Blubber	174 ± 64	31 ± 15	GC-MS	<0.04–5.3	(Lam et al., 2009)
	TBB	South China Sea	2003–2011	1–4	Blubber	242 ± 18	35.5 ± 17.9	LC-MS/MS QTrap	0.061–0.64	(Zhu et al., 2014)
		Hong Kong	2002–2008	2–4	Blubber	174 ± 64	31 ± 15	GC-MS	<0.04	(Lam et al., 2009)
	∑DP	South China Sea	2003–2011	1–4	Blubber	242 ± 18	35.5 ± 17.9	LC-MS/MS QTrap	1.74–63.7	(Zhu et al., 2014)
OCs	TCPMe	Hong Kong	1993–1997	n.r.	Blubber	163 ± 64	46 ± 19	GC-MS	43–240	(Minh et al., 2000, 1999)
		Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	32 ± 19	GC-MS	69–200	(Ramu et al., 2005)
	TCPMOH	Hong Kong	1993–1997	n.r.	Blubber	163 ± 64	46 ± 19	GC-MS	66–270	(Minh et al., 2000, 1999)
		Hong Kong	1995–2001	n.r.	Blubber	187 ± 63	32 ± 19	GC-MS	24–160	(Ramu et al., 2005)
Chlorinated Paraffins	SCCP	Hong Kong	2004–2014	n.r.	Blubber	242 ± 42	49 ± 19	LRMS/GC-ECNI	920–24,000	(Zeng et al., 2015)
	MCCP	Hong Kong	2004–2014	n.r.	Blubber	242 ± 42	49 ± 19	LRMS/GC-ECNI	1400–56,000	(Zeng et al., 2015)
PFC	PFOS	Hong Kong	2003–2007	1–4	Liver	210 ± 62	n.r.	LC-MS	26–693	(Yeung et al., 2009)
		Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	100–13,200	(Lam et al., 2016)
	PFHxS	Hong Kong	2003–2007	1–4	Liver	210 ± 62	n.r.	LC-MS	<0.25–1.31	(Yeung et al., 2009)
		Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	<0.10–22.2	(Lam et al., 2016)
	PFBS	Hong Kong	2003–2007	1–4	Liver	210 ± 62	n.r.	LC-MS	<0.25–0.40	(Yeung et al., 2009)
		Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	<0.10–12.3	(Lam et al., 2016)
	PFHxA	Hong Kong	2003–2007	1–4	Liver	210 ± 62	n.r.	LC-MS	<0.25	(Yeung et al., 2009)
		Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	<0.10–15.5	(Lam et al., 2016)
	∑PFCA	Hong Kong	2003–2007	1–4	Liver	210 ± 62	n.r.	LC-MS	62.9	(Yeung et al., 2009)
		Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	35–2040	(Lam et al., 2016)
	PFOSA	Hong Kong	2003–2007	1–4	Liver	210 ± 62	n.r.	LC-MS	9.51–37.6	(Yeung et al., 2009)
	N-EtFOSA	Hong Kong	2003–2007	1–4	Liver	210 ± 62	n.r.	LC-MS	<0.25–3.37	(Yeung et al., 2009)
	∑PFAS	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	136–15,300	(Lam et al., 2016)
	∑PFSA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	101–13,300	(Lam et al., 2016)
	PFBA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	<0.10–3.36	(Lam et al., 2016)
	PFPeA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	<0.10–7.66	(Lam et al., 2016)
	PFHpA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	0.36–60.8	(Lam et al., 2016)
	PFOA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	1.1169	(Lam et al., 2016)
	PFNA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	2.87–832	(Lam et al., 2016)
	PFDA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	6.74–683	(Lam et al., 2016)
	PFUnDA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	4.52–189	(Lam et al., 2016)
	PFDoDA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	2.76–102	(Lam et al., 2016)
	PFTeDA	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	<0.20–19.9	(Lam et al., 2016)
	PFDS	Hong Kong	2002	n.r.	Liver	251 ± 15	n.r.	UPLC-MS QTRAP	0.36–33.6	(Lam et al., 2016)
Organotin	MBT	Phumrieng Bay, India	2003	n.r.	Liver	211	n.r.	GC-MS	23,000	(Harino et al., 2008)
	DBT	Phumrieng Bay, India	2003	n.r.	Liver	211	n.r.	GC-MS	127,000	(Harino et al., 2008)

Table 3 (*continued*)

Pollutants		Location	Samples					Chemical analysis		Reference
Class	Pollutant(s)		Period	Status	Organ	Length, cm	Lipid %	Technique	Conc. range, ng/g lw.	
	TBT	Phumrieng Bay, India	2003	n.r.	Liver	211	n.r.	GC-MS	15,000	(Harino et al., 2008)
	BTs	Phumrieng Bay, India	2003	n.r.	Liver	211	n.r.	GC-MS	165,000	(Harino et al., 2008)

Condition was defined by veterinarians and officials of the Agriculture, Fisheries and Conservation, Hong Kong, as follows: 1 = live; 2 = freshly dead; 3 = moderately decomposing; 4 = advanced decomposing; 5 = mummified remains.

three decades, enaction of stringent fire prevention regulations globally resulted in an increase in the demand for brominated flame retardants. However, brominated flame retardants are not chemically bound to the polymer materials, thus leach into the environment and they enter the marine coastal waters through estuary discharges. Most brominated flame retardants are semi-volatile and have been shown to undergo long range atmospheric transport in the gas phase - if non-volatile like BDE-209, they attached to airborne particles (Möller et al., 2012). Polybrominated diphenyl ethers (PBDEs) and hexabromocyclododecanes (HBCDs) are the most widely used brominated flame retardants. The Stockholm Convention partially banned some brominated flame retardants because they pose risk to the environment and human due to their high persistence, bioaccumulation potential, and toxicity (Wang et al., 2017). However, several spatial and temporal trends studies detected brominated flame retardants in marine coastal waters (Wang et al., 2017), sediments (Liu et al., 2005; Zhen et al., 2016), atmosphere (Li et al., 2016) and biota (Li et al., 2010; Zhou et al., 2016) suggesting chronic exposure to Indo-Pacific humpback dolphin.

Brominated flame retardants may cause adverse effects in Indo-Pacific humpback dolphin because they have been shown to bioaccumulate in blubber and liver samples. In Hong Kong, the concentration of ∑PBDEs in Indo-Pacific humpback dolphin sampled between 2002 and 2008 ranged from 280 to 51,100 ng/g lw (Lam et al., 2009). The results suggested there was a sharp increase in amount of PBDEs used in the Pearl River Delta region because the maximum ∑PBDEs concentration between 1995 and 2001 was 6000 ng/g lw (Ramu et al., 2005). The sharp increase in bioaccumulation could be attributed to the increase in production of PBDEs between 1995 and 2008. However, no similar trends were observed in ∑HBCDs in the periods 2002–2008 (Lam et al., 2009) and 1997–2001 (Isobe et al., 2007). A traditional industrial mixture of PBDE comprised of Penta-BDE, Octa-BDE and Deca-BDE, of which the first two were banned in China in 2006. Between 1995 and 2001, BDE209, the main constituent of Deca-BDE was not detected in Indo-Pacific humpback dolphin blubber (Ramu et al., 2005). In contrast, samples of blubber from the same region collected between 2003 and 2012 detected a BDE209 concentration of 5.07 ng/g lw (Zhu et al., 2014). A temporal shifting trend was observed between 2003 and 2011 because Penta-BDE, Octa-BDE and Deca-BDE are being replaced by DBDE, PTBPE and DP (Zhu et al., 2014). Plotting the amount of banned brominated flame retardant in Indo-Pacific humpback dolphins against its alternative over time reveals the temporal shifting in their usage. A significant temporal shifting trend was observed between DP and BDE209, DBDPE and BDE209, and PTBBPE and Octa-BDE with $r^2 = 0.67$ and $p = 0.0005$, $r^2 = 0.61$ and $p = 0.002$, and $r^2 = 0.47$ and $p = 0.025$, respectively (Zhu et al., 2014). Therefore, international regulations influence the concentration and type of brominated flame retardant that bioaccumulates in Indo-Pacific humpback dolphins.

3.3.2. Perfluorinated compounds

Perfluorinated compounds are a class of highly fluorinated organic compounds widely used in industrial and domestic products, for example, shampoo, floor polish and lubricants. Since the electronegativity of fluorine is the highest among halogens, the C–F bond is very stable thermally and chemically. Hence, perfluorinated compounds have high thermal and chemical stability, acid and alkaline resistant, amphipathic, low surface tension, and high density (Yan et al., 2015). The physicochemical properties of perfluorinated compounds render them contaminants of major concern in the marine environment because of their high persistence, bioaccumulation potential and toxicity. Hence, perfluorinated compounds have been frequently detected in several environmental compartments including estuaries and marine biota (Wang et al., 2017; Yan et al., 2015; Zhang et al., 2016). In addition, some perfluorinated compounds demonstrated subchronic to acute toxicity by proliferating peroxisomes in rats and mice and inhibiting gap junctional intercellular communication (Zhang et al., 2016). Thus, perfluorinated compounds pose an environmental risk to Indo-Pacific humpback dolphins.

Although some perfluorinated compounds such as PFOS were included in the Stockholm Convention on persistent organic pollutants in 2009, their use continues to increase in China (Table 3) (Lam et al., 2016; Liu et al., 2015b). The concentration range of PFOS, PFHxS, PFHxA and PFBS in Indo-Pacific humpback dolphin between 2002 and 2014 were 100–13,200, <0.10–22.2, <0.10–15.5 and < 0.10–12.30 ng/g, respectively (Lam et al., 2016). In recent years, PFBS has been used as an alternative flame retardant replacing PFOS. A positive temporal shifting trend was observed in a log-linear regression plot of the ratio of the bioaccumulated amount of PFBS to PFOS ($r^2 = 0.50$, $p = 0.049$) (Lam et al., 2016). The usage of PFCAs continues in China as was shown by a positive temporal trend in plots of PFCAs to PFOA ($r = 0.72$, $p = 0.44$) (Lam et al., 2016). Studies in Hong Kong found that PFOS had the highest bioaccumulation compared to other perfluorinated compounds (Lam et al., 2016; Yeung et al., 2009). However, the high concentration of PFOS might be due metabolism of PFOSA, since studies found PFOSA may transform to PFOS (Yeung et al., 2009). Correlations between the amount of pollutants in an organism can be used in establishing the source of the pollutants. A positive correlation was observed between the bioaccumulation of PFOS and PFCAs (such as PFUnDA, PFDA and PFNA) suggesting they had a similar source (Yeung et al., 2009).

3.3.3. Chlorinated paraffins

More than one million tons of chlorinated paraffins are produced globally per year for such use as plasticizers, sealants and flame retardants (Zeng et al., 2015). However, knowledge on their environmental toxicity, fate and bioaccumulation remains scarce. Environmental monitoring and characterization of chlorinated paraffins is challenging because a single technical mixture may

contain more than 4000 congeners. Chlorinated paraffins comprise of polychlorinated n-alkane congeners of varying length and degrees of chlorination commonly used as flame retardants, plasticizers and lubricants. They are classified according to their chain length as short-chain, medium chain and long-chain chlorinated paraffins with C_{10}–C_{13}, C_{14}–C_{17} and C_{18}–C_{30}, respectively (Wei et al., 2016). From the predicted physicochemical properties, chlorinated paraffins probably have high bioaccumulation potential since they have a low water solubility, high octanol-water (log K_{OW}), high octanol–air partition coefficients (log K_{OA}) and semi-volatile (van Mourik et al., 2016). The log K_{OW} of the congeners increase with chain length when their number of chlorine atoms were similar (van Mourik et al., 2016). Hence, chlorinated paraffins detected in blubber samples from Indo-Pacific humpback dolphins collected between 2004 and 2014 in China had concentrations of SCCP lower than MCCP ranging from 920 to 24,000 ng/g and 1400–56,000 ng/g, respectively (Zeng et al., 2015). Although use of short-chain chlorinated paraffins is under review in the Stockholm Convention, a positive temporal trend was observed in a linear regression analysis of their total concentration in Indo-Pacific humpback dolphin each year ($r^2 = 0.82$, $p < 0.05$) (Zeng et al., 2015). However, Zeng et al. (2015) found that each year, the total concentration of short-chain chlorinated paraffins was lower than that of medium chain chlorinated paraffins. A temporal shift from short to longer chain was observed, for example C_{10} abundance decreased each year by 4% (Zeng et al., 2015). Thus, the ratio of medium chain and short chain chlorinated paraffins could be used as an indicator of shifting trends in their production and usage.

Evaluation of the ecological and health risks of chlorinated paraffins is hindered by the scarcity of data on their bioaccumulation and biomagnification in Indo-Pacific humpback dolphins. The BMFs for MCCPs are often higher than those for SCCPs in the freshwater food webs (Houde et al., 2008). A study in Guangdong, China found SCCPs underwent trophic biodilution in an aquatic food web (Sun et al., 2017), whereas trophic magnification was observed in another study in Shanghai, China (Zhou et al., 2018). Therefore, TMFs of chlorinated paraffins may vary between different ecosystems.

3.3.4. Organotin compounds

Although studies on bioaccumulation in Indo-Pacific humpback dolphin remain scarce, organotin compounds have been shown to be highly persistent, bioaccumulative and toxic in the marine environment. Organotin compounds, such as monobutylin, dibutylin, tributylin, monophenyltin and triphenyltin, are frequently detected in the marine environments due to their widespread use as antifouling agents, biocides and polyvinyl chloride stabilizers (Ho et al., 2016). Several studies demonstrated that tributylin cause imposex, that is a superimposition of male sexual characteristics on females, in gastropods (Ho et al., 2016). Butylins were detected in blubber samples from a Indo-Pacific humpback dolphin stranded at Phumrieng Bay with a total concentration of 17,000 ng/g (Harino et al., 2008). In 2008, the International Maritime Organization prohibited the use of organotin compounds as antifouling agents on ships. However, a recent study found organotins remain widespread in Hong Kong's marine ecosystems despite the global ban hence the need to investigate their bioaccumulation and toxicity in the region (Ho et al., 2016).

4. Understudied areas on factors influencing bioaccumulation

4.1. Gender

Bioaccumulation of organic pollutants in Indo-Pacific humpback dolphin may be influenced by gender; however, the gender differences may become more significant with age. Bioaccumulation in females may be low because fat-soluble organic pollutants may be transferred to an offspring during lactation and parturition and lactation (Tuerk et al., 2005). For example, a study on bioaccumulation of POPs found the average concentration of organochlorines in Indo-Pacific humpback dolphins was significantly lower in females than in males (Gui et al., 2014a). Notably, the concentration of HCHs in blubber from male and female Indo-Pacific humpback dolphins ranged from 183 to 3085 ng/g and 344–757 ng/g, respectively (Gui et al., 2014a). However, the concentration of HCHs did not show any significant correlation with age in melon tissue of the Indo-Pacific humpback dolphin (Gui et al., 2014b). Thus, it is possible that HCHs in blubber readily redistribute during lactation and parturition than those in the melon. However, differences were observed in the bioaccumulation of PCBs and DDTs in both melon and blubber samples (Gui et al., 2014a, 2014b). They are only two studies that offer an opportunity to compare concentration difference in males and females between different tissues. Hence, there is limited understanding of the role of reproductive processes in the elimination of organic pollutants in female Indo-Pacific humpback dolphins.

Studies on the effect of gender are often hampered by the size of the samples which are limited by sampling restrictions. A previous study that sampled from the same region as Gui et al. (2014a) did not observe any significant difference between the mean concentrations of DDTs in male and female Indo-Pacific humpback dolphins. Differences in sample size and possibly level of decomposition of the stranded dolphins could account for the difference (Wu et al., 2013). Wu et al. (2013) had a smaller sample size of 15 stranded dolphins, whereas Gui et al. (2014a) sampled 45 freshly dead and moderately decomposing dolphins. Furthermore, to the best of our knowledge, the effect of gender has not been observed in halogenate flame retardants (Zhu et al., 2014), perfluorinated compounds (Lam et al., 2016; Yeung et al., 2009), and short and medium-chain chlorinated paraffins (Zeng et al., 2015). The small samples sizes of the stranded dolphins in these studies ranged from 10 to 25, thus probably inadequate for comparing the bioaccumulative behavior of the emerging contaminants to legacy contaminants. The lack of adequate data on the effect of age in bioaccumulation could be alleviated by developing better sampling strategies.

4.2. Age and body length

Differences in bioaccumulation of different age groups may be useful in understanding chronic exposure of organic pollutants in Indo-Pacific humpback dolphin. The age of Indo-Pacific humpback dolphin can be estimated using dental layers (Gui et al., 2014a, 2014b) or growth rings that collate in bones, reproductive activity (Gui et al., 2014c) or body length (Gui et al., 2017). Current age determination techniques are only applicable to stranded and captive Indo-Pacific humpback dolphins. Due to poor metabolism, marine mammals accumulate organic pollutants as they age. However, Gui et al. (2014a, 2014b) found a significant correlation between body length or age with concentration of $\sum$PCBs and $\sum$DDTs ($r^2 \geq 0.536$, $p \leq 0.025$). A significant decrease in concentration of $\sum$DDTs was observed in female Indo-Pacific humpback dolphin with body length above 190 cm ($r^2 = 0.426$) (Gui et al., 2014a). In contrast, Gui et al. (2014a) the concentration of $\sum$DDTs in males showed a significant correlation with age ($r^2 = 0.894$). Pollutant load in small mammals is often higher because they have a higher metabolic rate than bigger mammals. That is, Indo-Pacific humpback dolphin with shorter body length are expected to have higher bioaccumulation as observed in an

interspecies study of the shorter long-finned pilot whales and the longer sperm whales (Pinzone et al., 2015). However, in intra-species studies, the correlation between body length and length of exposure seems to be a more significant factor. Determination of effect of size in either juvenile or adult Indo-Pacific humpback dolphin remains challenging since most studies use size as a determinant of age.

4.3. Physicochemical properties of the organic pollutants

The intrinsic properties of an organic pollutant often determine its fate, transport and bioaccumulation potential in marine environments. Increasing carbon chain length and degree of halogenation often result in a decrease in vapor pressure. Therefore, compounds with lower molecular weight tend to have higher transport and bioaccumulation potential. For example, the concentration of low-molecular PAHs like naphthalene and acenaphthylene in blubber samples than that of high-molecular weight PAHs (Leung et al., 2005). A similar study in Australia found naphthalene and pyrene accounted for 25% of the total PAHs in a skin biopsy study (Cagnazzi et al., 2013). However, in high-chlorinated PCB congeners such as PCBs 128, 188 and 180, were detected in higher concentrations than those with lower chlorination (Minh et al., 1999; Wu et al., 2013). Organic pollutants with a high degree of halogenation are chemically stable, thus increasing their bioaccumulation potential due to chronic exposure. However, since low-chlorinated PCBs are more bioavailable, they can desorb from the sediment and enter the water front resulting in an increase in their bioaccumulation. Thus, the concentration of low-chlorinated PCB congeners increased in Indo-Pacific humpback dolphin between 1993 and 1997 (Minh et al., 1999) and 2004 to 2013 (Gui et al., 2014a). Furthermore, increasing the carbon chain from C_8 to C_{11} among perfluorinated compounds, resulted in an increase in bioaccumulation (Yeung et al., 2009). Similarly, Zeng et al. (2015) found the amount of volatile chlorinated paraffins was lower than that of the less volatile chlorinated paraffins in Indo-Pacific humpback dolphin. The concentration of short-chain and medium-chain chlorinated paraffins ranged from 430 to 9100 ng/g dw and 530 to 23,000 ng/g dw, respectively (Zeng et al., 2015). Therefore, usage profiles of emerging contaminants might be a more significant indicator of bioaccumulation potential than molecular weight. However, there is a lack of conclusive information on the effect of molecular weight on bioaccumulation.

Isomers of organic pollutants often exhibit differences in physicochemical properties, degradation, bioavailability and bioaccumulation in the environment. Thus, a ratio comparing the different isomers of a contaminant can be used as a chemical indicator in source apportionment and bioaccumulation studies. For example, a technical mixture of HCBD comprises of 10–13% α-HBCD, 1–12% β-HBCD and 75–89% γ-HBCD (Covaci et al., 2006). However, bioaccumulation studies in Indo-Pacific humpback dolphin often detect α-HBCD with an α-HBCD to ∑HBCD >0.99 suggesting an isomer-specific bioaccumulation (Isobe et al., 2007; Lam et al., 2009). It is important to note that determination of γ-HBCD in environmental matrices is challenging using GC-MS as it undergoes isomerization at temperatures above 160 °C (Isobe et al., 2007). Thus, to avoid potential artefacts, a liquid chromatograph coupled with a triple quadrupole tandem mass spectrometer technique can be used (Tomy et al., 2004). Previous studies found isomeric ratio increased from phytoplankton to fish as it ranged from 0.49 to 0.78 and 0.76–0.88, respectively (Covaci et al., 2006). Thus, isomer-specific bioaccumulation can be used to determine the feeding habits of the Indo-Pacific humpback dolphin. Although, α-HCH is the dominant isomer in technical mixture constituting about 70%, it rapidly degrades in the environment compared to β-HCH (Leung et al., 2005; Minh et al., 1999; Wu et al., 2013). Furthermore, a study in bioaccumulation of CHLs found *cis*-chlordane, *trans*-nonachlor and *cis*-nonachlor had the highest ratios ranging from 25 to 38% (Wu et al., 2013). Hence, differences in bioaccumulation can be caused by variations in environmental behavior and metabolism in Indo-Pacific humpback dolphin.

4.4. Tissue-specific bioaccumulation

Indo-Pacific humpback dolphins are exposed to organic pollutants primarily through dietary intake since they are air-respiring mammals. Environmental monitoring studies often rely on biopsies of blubber to determine bioaccumulation. Although, a remote biopsy assay study in Atlantic spotted dolphins (Méndez-Fernandez et al., 2016) found no stratification in lipids and contaminants, it has been shown that occurrence of lipophilic pollutants within blubber tissue was stratified in bottlenose dolphins (Ellisor et al., 2013) and southern hemisphere humpback whales (Waugh et al., 2014). The level of stratification was shown to depend on the physicochemical properties of the pollutant, lipid stratification, location of the blubber, and blubber depth (Ellisor et al., 2013; Waugh et al., 2014). Contaminant stratification and its implications is largely unknown in Indo-Pacific humpback dolphin, hence the importance of tissue-specific bioaccumulation studies.

In a study on the tissue distribution of organic pollutants in Indo-Pacific humpback dolphin, highest concentration of 10 organic pollutants investigated were found in melon ($p < 0.05$) (Gui et al., 2014b). Compared to other tissues, melon has a lipid content, thus may act as a reservoir of the lipophilic organic pollutants. Furthermore, previous studies in harbor seals demonstrated that tissue distribution of organic pollutants may vary by age or gender. For example, male pups had more than three and four times mean ∑PBDE than female pups and adult males, respectively (Kruskal–Wallis test: $X^2 = 9.3$, $p = 0.01$) (Shaw et al., 2012). A similar observation was made in Indo-Pacific humpback dolphin where the mean concentrations of ∑PCBs ($p = 0.008$) and aldrin ($p = 0.017$) were significantly higher in the melon tissue of adults than the juveniles (Gui et al., 2014b). However, there was no statistically significant in the total concentration of organohalogens differences between male and female, and adult and juvenile Indo-Pacific humpback dolphins. Since the sample group for each treatment in the study was very small ($n = 2$), it remains difficult to draw out a conclusion on the impact of lipid content and organ function on the bioaccumulation of organic pollutants in the Indo-Pacific humpback dolphins.

Currently, studies on the tissue-specific bioaccumulation of emerging contaminants in Indo-Pacific humpback dolphins remain scarce. However, a study of Indo-Pacific humpback dolphin in India found monobutylin, dibutylin and monophenyltin preferentially bioaccumulated in liver tissues followed by kidneys with concentrations 165,000 ng/g and 37,000 ng/g, respectively (Harino et al., 2008). The bioaccumulation of butylins in Indo-Pacific humpback dolphin followed the following order; liver > kidneys > heart > lung ≈ blubber ≈ muscle (Harino et al., 2008). Interestingly, they detected the lowest concentration in the blubber suggesting using blubber samples in bioaccumulation studies might be underestimating the risk of PFCs to Indo-Pacific humpback dolphins (Ahrens et al., 2009). Unlike, other lipophilic organic pollutants, PFCs preferentially bind to blood plasma rather than the lipid. Furthermore, a whole body burden evaluation found that blood and liver contributed 75%, hence sampling these tissues in environmental monitoring studies may offer a more accurate estimate of bioaccumulation (Ahrens et al., 2009).

4.5. Dietary intake

According to stable isotope analyses, Indo-Pacific humpback dolphins are piscivorous apex predators. Stable isotope analysis is a useful tool for identifying the dietary source ($\delta^{13}C$), that is whether the food source is benthic, pelagic or demersal and nearshore or offshore (Table 4) (Browning et al., 2014). Furthermore, stable isotope analysis is often used in ecology to determine the trophic level of an organism (Figueiredo et al., 2014). Predators often have higher $\delta^{15}N$ than their prey, thus stable isotope enrichment is often observed with increase in trophic level. The $\delta^{15}N$ value of Indo-Pacific humpback dolphin was 14.8%, which was higher than that of Omura's whales (*Balaenoptera omurai*), Fraser's dolphins (*Lagenodelphis hosei*), dwarf sperm whales (*Kogia sima*), pantropical spotted dolphins (*Stenella attenuata*), Risso's dolphins (*Grampus griseus*), and finless porpoises (*Neophocaena phocaenoides*) which ranged from 10.9 to 14% (Liu et al., 2015a). This suggested Indo-Pacific humpback dolphins where at the top of the trophic level in marine ecosystems.

Since dietary intake is the primary route of exposure in Indo-Pacific humpback dolphins, the concentrations of organic pollutants in the prey species could be used to predict their health risk (Hung et al., 2006a). Using the food web approach, the BMFs of DDTs, pentachlorobenzene, mirex, and PCBs in Indo-Pacific humpback dolphins where estimated as 212 ± 160, 184 ± 170, 137 ± 150, and 99 ± 83, respectively (Gui et al., 2014a). However, since the diet of Indo-Pacific humpback dolphins is not fully known, and only 10 prey species were used to determine the BMFs, the obtained values could be an underestimate or an overestimate. Furthermore, the accumulation of organic pollutants in the prey fish might be influenced by their migratory behavior and the sea layer they reside. The concentration of different organochlorine pesticides in the prey fish was less than 10 ng/g lw in all 10 species. However, the concentration of aldrin, endrin and mirex was higher in amphidromous fish namely *Johnius belengerii*, *Trichiurus lepturus* and *Odontamblyopus rubicundus* (Gui et al., 2014a). Dieldrin and pentachlorobenzene were detected mainly in pelagic prey fish (*Clupanodon thrissa* and *Coilia mystus*) at concentrations ranging from 0.08 to 3.06 ng/g lw (Gui et al., 2014a). Therefore, when determining the bioaccumulation factor of organic pollutant in Indo-Pacific humpback dolphin from the prey fish, it is important to weight the contribution of each prey fish family.

Neonatals are exposed to organic pollutants through lactation. The concentration of $\sum$PCBs and $\sum$OCs in Indo-Pacific humpback dolphin milk was 2490 ng/g lw and 15,880 ng/g lw, respectively (Parsons and Chan, 1998). Previous studies detected a high concentration of $\sum$PCBs and $\sum$OCs, 2390 ± 99 ng/g lw and 25 ± 4 ng/g lw, respectively (Parsons and Chan, 1998). However, the overall contribution of lactation in transferring contaminants remains understudied due to sampling restrictions and in adequate methods of determining age.

4.6. Health effects and environmental risk assessment

Data on biological effects of organic pollutants on Indo-Pacific humpback dolphins remain scarce due to sampling restrictions and an incomplete understanding of the population dynamics of Indo-Pacific humpback dolphins. However, toxicological studies for establishing the adverse effects of organic pollutants and pathogens on marine mammals can be conducted using *in vitro* cell cultures (Jin et al., 2013). For example, previous studies found Carvan dolphin kidney cells from Atlantic bottlenose dolphin may be used as a toxicological model for investigating health effects of hydrocarbon contaminants (Carvan et al., 1994). Jin et al. (2013) derived fibroblast cells from the skin of stranded Indo-Pacific humpback dolphins. Recently, an *in vitro* study showed that CYP1A1 was up-regulated when cultured skin fibroblast cells from Indo-Pacific humpback dolphins were exposed to varying concentrations of HCB, DDTs, CHLs, and HCHs (Jia et al., 2015). However, a lot of uncertainty remains on extrapolation of biological effects observed at molecular level to population level. Hence, the absence of whole animal toxicity studies hampers the environmental risk assessment of Indo-Pacific humpback dolphins using the persistence, bioaccumulation potential and toxicity models.

Understanding the type and severity of potential adverse effects of exposing Indo-Pacific humpback dolphins to organic pollutants is critical for research and regulatory purposes. Environmental risk assessment encompasses hazard identification, exposure assessment, dose-response assessment, and risk characterization (Fig. 4). Although much information is available on toxicity of organic pollutants and their exposure levels in marine mammals, their dose-response data remains scarce, particularly for endangered species such as Indo-Pacific humpback dolphins (Hung et al., 2006b). Determination of threshold levels such as, no observed adverse effect level (NOAEL), lowest observed adverse effect level (LOAEL) or effective maximal concentration (EC_{50}) of an organic pollutant is critical for ascertaining their potential hazard. Assessing threshold levels in Indo-Pacific humpback dolphins is

Table 4
Prey fishes of *Sousa chinensis* adapted from Gui et al. (2014a).

Species	Family	Species niche[a]	Migratory behavior[a]	Family dietary %	Common name
Johnius belengerii	*Sciaenidae*	Demersal	Amphidromous	17.2	Belanger's croaker
Collichthys lucidus	*Sciaenidae*	Demersal	Oceanodromous	17.2	Croaker
Clupanodon thrissa	*Clupeidae*	Pelagic	Anadromous	37.0	–
Coilia mystus	*Engraulidae*	Pelagic-neritic	Amphidromous	–	–
Harpadon nehereus	*Synodontidae*	Benthopelagic	Oceanodromous	–	Bombay duck
Cynoglossus bilineatus	*Cynoglossidae*	Demersal	–	–	Fourlined tonguesole
Sillago sihama	*Sillaginidae*	Reef-associated	Amphidromous	–	Silver sillago
Arius sinensis	*Ariidae*	Demersal	–	–	–
Mugil cephalus	*Mugilidae*	Benthopelagic	Catadromous	20.1	Mullet
Odontamblyopus rubicundus	*Taenioididae*	Benthopelagic	Amphidromous	–	–
Odontamblyopus lacepedii	*Taenioididae*	Benthopelagic	–	–	–
Trichiurus lepturus	*Trichiuridae*	Benthopelagic	Amphidromous	–	Largehead hairtail
Pseudosciaena crocea	*Sciaenidae*	Demersal	Oceanodromous	17.2	Croceine croaker
Argyrosoma macrocephalus	*Sciaenidae*	Demersal	–	17.2	–
Psenopsis anomola	*Stromateidae*	Benthopelagic	–	2.4	Japanese butterfish
Leiognathus brevirostris	*Leiognathidae*	Demersal	Amphidromous	3.6	Shortnose ponyfish
Pomadasys sp.	*Haemuliade*	Demersal	–	14.3	–

[a] Data obtained from FishBase (Froese and Pauly, 2017).

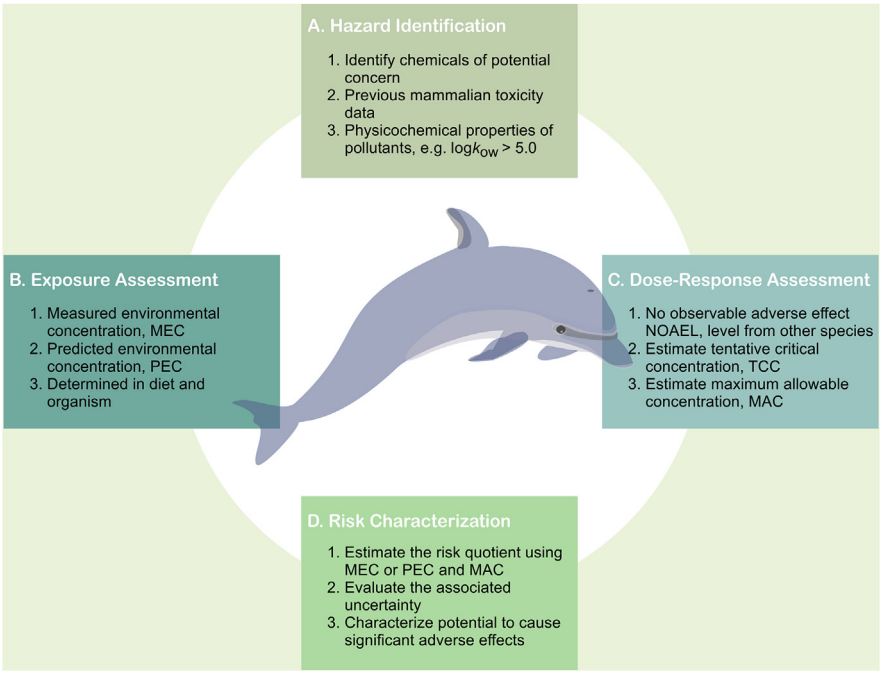

Fig. 4. An overview of critical steps in environmental risk assessment of Indo-Pacific humpback dolphin (*Sousa chinensis*).

challenging because there are ethical restrictions on conducting dosimetry studies on them since they are endangered. Assuming there is evolutionary conservation of primary pollutant target, tentative critical concentration (TCC) also known as toxicity reference value (TRV) can be used as an estimate of the NOAEL by adjusting for the differences in body weight between the surrogate ($BW_{surrrogate}$) and Indo-Pacific humpback dolphin ($BW_{dolphin}$) when dose-response data is not available (Equation (1)) (Lam et al., 2016). To establish the risk associated to dietary intake, a food web approach is often employed involving estimating the maximum allowable concentration (MAC) (Equation (2)) followed by determination of the risk quotient (RQ) (Equation (3)) (Hung et al., 2006a).

$$TCC_{dolphin} = NOAEL_{surrogate}\left(BW_{surrogate}/BW_{dolphin}\right)^{1/4} \quad (1)$$

$$MAC_{diet} = \frac{TCC_{dolphin} + averaging\,time + BW_{dolphin}}{ingestion\,rate + exposure\,frequency + exposure\,duration} \quad (2)$$

$$RQ = \frac{contaminant\ concentration\ in\ diet}{MAC_{diet}} \quad (3)$$

In previous studies, environmental risk of organochlorines and perfluorinated compounds in Indo-Pacific humpback dolphins was assessed using food web approach and accumulation monitoring, respectively. Using food web approach, the MAC values of organochlorines in prey fish ranged from 80.97 to 11,856 μg/kg, for PCB-218 and chlordane, respectively (Hung et al., 2006a). However, the risk due to organochlorines was low with all RQ values > 0.01. However, the food web approach uses single values for the ingestion rate, exposure frequency, exposure duration or body, thus it fails to account for seasons, habitat location, and age and sex of the Indo-Pacific humpback dolphins. Alternatively, environmental risk can be determined using a probabilistic assessment of organic pollutant exposure (Lam et al., 2008). This involves establishing the probability of concentration of organic pollutants in tissue samples to exceed the TCC values. A study in South China Sea found 35% of PFOS concentrations detected in liver samples of Indo-Pacific dolphins were greater than the corresponding TCC values that caused hepatic toxicity in monkeys and rats (Lam et al., 2016). The results suggested that current concentrations of PFOS in liver samples could cause significant adverse effects on Indo-Pacific humpback dolphins. However, using TCC may underestimate or overestimate the risk of an organic pollutant because modes of actions vary between species and the toxicity could be mediated through multiple receptors (Krafft and Riess, 2015). Therefore, there is additional need to develop tools for accurately extrapolating effects between species and individual to population level, which is most relevant for environmental management.

5. Future research directions

5.1. Sampling strategy and technique

Sampling is probably the most critical step in environmental monitoring and characterization. However, it is the main drawback in studying bioaccumulation of organic pollutants because Indo-Pacific humpback dolphins are 'near threatened' mammals. Sample sizes and frequency was limited in previous studies because sampling was only conducted on stranded animals. Therefore, there is need for development of a non-evasive sampling technique such as biopsy sampling. In biopsy sampling, samples are collected from a patch of skin using an arrow of varying sizes. The skin biopsy can be used for a wide range of chemical and toxicological analyses, including pollutant residue analysis, monitoring of a photo-identified animal, biomarker survey, and fibroblast cell cultures (Fossi et al., 2003). Recently, Cagnazzi et al. (2013) employed skin biopsy sampling using PAXARMS biopsy system to investigate bioaccumulation of organochlorines and PAHs in Chine white dolphin found in southern Great Barrier Reef, Australia. However,

there is concern on the effect of biopsy sampling on the health of the animals. A previous study found Indo-Pacific humpback dolphins reacted slightly to the dart and the sampling wound healed in 21 d (Jefferson and Hung, 2008). A major challenge with biopsy sampling is the heterogenous distribution of lipids and lipophilic contaminants in the blubber. Hence, there is need for validation of biopsy sampling to establish the levels of stratification. Furthermore, knowledge of the age and gender of the Indo-Pacific humpback dolphin is important in understanding the trends in bioaccumulation. There is need for development of molecular techniques to determine age or gender of the dolphins. For example, concentrations of thyroid and adrenal hormones in blood samples of Atlantic bottlenose dolphins (*Tursiops truncates*) were shown to change with age (St. Aubin et al., 1996). Skin biopsy sampling may prove to be a viable alternative to sampling stranded Indo-Pacific humpback dolphins.

5.2. Exposure pathway

Understanding the exposure pathway of organic pollutants in Indo-Pacific humpback dolphins is a critical step in environmental risk assessment and in designing mitigation efforts. Future research should focus on understanding the source of an organic pollutants helps in formulating environmental management programs that reduces or eliminate marine pollution. Although the routes of exposure of legacy contaminants in marine environments are mostly known, knowledge of the source, fate and transport of emerging contaminants such as TBPH, TBB, PDBPs, SCCP and MCCP remain scarce. Further research is required for these compounds and classes of compounds for establishing their exposure pathway in Indo-Pacific humpback dolphins.

5.3. Predicting bioaccumulation

Since sampling Indo-Pacific humpback dolphins is restricted, development of accurate predictive models are critical in understanding their ecological risk. Quantitative Structure-Activity Relationships can be used to predict bioaccumulation in Indo-Pacific humpback dolphin. The log K_{OW} is a good predictor for bioaccumulation of highly lipophilic compounds in lipid-rich tissues. There was a significant positive correlation between the concentration of organic pollutants with log $K_{OW} < 5.5$, $p < 0.05$ in spotted seals and log K_{OA} (Moses et al., 2015). Hence, fugacity-based predictive models could be used to estimate the bioaccumulation of such organic pollutants in Indo-Pacific humpback dolphin. However, most environmental fate models predict fugacity and bioaccumulation using octanol-water partition coefficient, which might not be adequate for amphipathic compounds such as perfluorinated compounds. For such compounds, to systematically understand their bioaccumulation determination of concentration of pollutants in water, sediments, organism in each trophic level immediate to the Indo-Pacific humpback dolphin and tissue distribution is imperative (Houde et al., 2006).

5.4. Monitoring of additional emerging contaminants

There are several contaminants of major concern in coastal marine waters, besides the ones profiled in this paper, that can potentially cause adverse effects in Indo-Pacific humpback dolphin. Pharmaceuticals are biologically active compounds designed to effect change at low does, hence they pose an environmental risk to Indo-Pacific humpback dolphins. The discharge of pharmaceuticals in coastal waters through wastewater effluent may continue to increase due to rapid urbanization (Wei et al., 2016). High pH values common in marine water often increase lipophilicity of most pharmaceuticals, thus result in an increase in their bioaccumulation potential (Fabbri and Franzellitti, 2015). Hence, high discharge of pharmaceuticals into marine ecosystems may induce adverse biological effects in Indo-Pacific humpback dolphins. However, since they are more than 5000 pharmaceuticals on the market, the target pollutants need to be prioritized using production volume or read-across method (Rand-Weaver et al., 2013). Therefore, there is a need to assess exposure and risk of residues of pharmaceutical and their transformation products in Indo-Pacific humpback dolphins.

Microplastics are frequently and abundantly detected in marine environments, thus could potentially cause adverse effects in Indo-Pacific humpback dolphin. The main sources of microplastics in marine ecosystems are mechanical abrasion and photodegradation of large plastics, plastic pellets, cosmetics, and clothing fibers. Marine organisms that ingest microplastics may suffer chemical and physical damage (Jabeen et al., 2017). Furthermore, persistent, bioaccumulative and toxic compounds such as bisphenol A, phthalates, brominated flame retardants and nonylphenols are commonly used as plastic additives (Hermabessiere et al., 2017). Several studies found a correlation between amount of microplastics and lipophilic organic pollutants (Hermabessiere et al., 2017). A recent study in China detected microplastics in coastal fish, with the amount of plastics in demersal fish higher than in pelagic fishes ($p < 0.05$) (Jabeen et al., 2017). Therefore, there is need to determine the exposure scenario and potential ecological risk of microplastics to Indo-Pacific humpback dolphin.

5.5. Health effects and environmental risk assessment

The direct and indirect health effects of organic pollutants are a major cause of concern for the health of Indo-Pacific humpback dolphins. Although the biological effects of organic pollutants, such as organochlorines and PAHs, are well documented in some mammals, ecotoxicological data on Indo-Pacific humpback dolphin remain scarce. For example, 60% bottlenose dolphins inhabiting waters highly contaminated with PCBs suffered anemia (Schwacke et al., 2012). The study found the blubbers had low levels of thyroid hormone and reduced proliferation of T-lymphocyte suggesting high susceptibility to infectious diseases (Schwacke et al., 2012). High concentrations of PCBs have been reported in the shallow coastal waters of South East Asia, thus risking Indo-Pacific humpback dolphins to PCB-related health effects (Wang et al., 2015). Future research, should investigate the potential effect of organic pollutants on the immune system of Indo-Pacific humpback dolphins. Considering the difficulty in accessing samples, there is need for development of techniques that predict health effects. One technique that could be beneficial is the read-across method where toxicity can be predicted using a model organism. However, the utility of read-across method depends on the evolutionary conservation of the primary target of the toxicant (Rand-Weaver et al., 2013). Thus, future studies should explore evolutionary conservation of organic pollutant receptors between Indo-Pacific humpback dolphins and other aquatic organisms. A previous study identified several genes involved in immune response of Indo-Pacific humpback dolphins using de novo transcriptome analysis (Gui et al., 2013). Transcriptome analysis may be useful in identification of genes that are critical in the environmental health of the Indo-Pacific humpback dolphins.

6. Conclusion

Efforts in environmental monitoring and characterization should probably increase since bioaccumulation of organic pollutants in Indo-Pacific humpback dolphin may continue to increase

due to rapid economic growth and urbanization. Although studies on exposure pathways and bioaccumulation remain scarce, previous studies demonstrated spatial and temporal trends particularly for legacy contaminants such as PCBs, DDTs and PAHs. The temporal trends observed in bioaccumulation of emerging contaminants were probably due to usage patterns that shifted with passing of restrictive legislations. However, systematic characterization of impact of organic pollutants is hindered by restrictions in sampling since Indo-Pacific humpback dolphin are near threatened marine mammals. Development and validation of sampling techniques such as skin biopsy sampling might be helpful in future studies. Furthermore, future studies should probably focus on the factors that influence bioaccumulation to understand the exposure pathways and risk of organic pollutants, particularly emerging contaminants such as halogenated flame retardants, perfluorinated substances, pharmaceuticals and microplastics.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at https://doi.org/10.1016/j.envpol.2018.01.055.

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