

Heavy metals distribution in cetaceans stranded at west and north coasts of Sabah, Malaysia

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Abstract: Heavy metals distribution in the liver, kidney, heart, lung, muscle, melon, muscle and blubber of 5 cetaceans (2 short-finned pilot whales, 2 finless porpoises and 1 spinner dolphin) stranded at the west and north coasts of Sabah, Malaysia in 2015 were examined in this study as a first attempt to establish baseline information. Cd, Cr, As, Pb, Cu, Mn, Fe, Se, and Zn were generally found to be low in these cetaceans. Calves, adolescents, and adult cetaceans examined exhibit differential metal distribution which is normal since diet and maturity stage play the major role on this. Similar pattern of distribution was also found regardless of species. Concentrations Cu, Zn, Fe and Mn in liver and kidney were found to be not affected by species or maturity. Selective accumulation of Cd was found in kidney of adolescent short-finned pilot whale and spinner dolphin (98.65 and 35.04 $\mu\text{g g}^{-1}$ dwt or 54.28% and 67.13% respectively). As and Cd in the calf finless porpoise were significantly high in the muscle compare to other tissues (3.52 and 3.79 $\mu\text{g g}^{-1}$) which could be due to redistribution of harmful metals to protect main organs from toxicity. However, no specific metal toxicity or pollution of local marine environment was found.

Keywords: stranded cetacean, heavy metals, metal distribution, Malaysia, toxicity

Introduction

Some metals naturally present on earth from natural processes like rock weathering and volcanoes activities. But pollution from anthropogenic source had contributed a large amount of them in the ecosystem (Nriagu and Pacyna, 1988; Tang *et al.*, 2014). Metals like Pb, Cd, Cr, and As are predominantly known to be toxic due to their harmful effects even at low concentration while metals like Cu, Zn, Fe, Se, and Mn are essential but are toxic when their levels exceed tolerance limit by body (Wafo *et al.*, 2014). Once exceed tolerance limit, these metals can cause damage or alter normal physiological function in the body.

Cetaceans which are positioned on top of the trophic levels tend to bioaccumulate these metals in their bodies and have a long biological half-time of pollutant elimination (O'Hara and O'Shea, 2001). They accumulate these metals at different concentration in their tissues according to environment condition they live in (Gerpe *et al.*, 2002). Thus they are commonly used as bioindicators or sentinels of the ocean. Studies of these elements in cetaceans are common and the effort is increasing mainly because of heavy metals pollution in marine ecosystem (Honda *et al.*,

1983; Gerpe *et al.*, 2002). After all, pollution is known to be the second biggest threat toward cetaceans after fisheries interaction (Wafo *et al.*, 2014). Yet there is lack of study on metals distribution in cetaceans in Malaysia causing no baseline information for further understanding the welfare of cetaceans in Malaysia.

These metals affect cetaceans even at small concentrations due to the fact that their biochemical roles in metabolic processes (Jakimska *et al.*, 2011). Generally, when a metal in the body exceeded the threshold concentration, it leads to dysfunction of multiple systems such as endocrine system which affect reproduction and growth; interacting with systemic enzymes which affects cellular function (Hylland, 2006). However, different metals react and target different systems in the body. Despite increasing heavy metals levels in the ocean, metals distribution in cetaceans could not be demonstrated in live cetaceans because they are protected animals in Malaysia. Thus, the objective of this paper was to examine the metal distribution in the bodies of cetaceans stranded in Sabah's west and north coasts in 2015.

Materials and methods

Sample collection

Five cetaceans stranded at the west and north coasts of Sabah were collected for this study. Liver, lung, heart, kidney, melon, muscle and blubber were sampled during post-mortem dissection with veterinarian of Wildlife Rescue Unit from Sabah Wildlife Department. The carcasses examined were in

decomposition stages 2 (freshly dead) and 3 (moderately decomposed) and were adequate for chemical analysis of pollutants (Geraci and Lounsbury, 1993). Prior to dissection, species, total length, maturity and sex of the cetaceans were determined and shown in Table 1. Samples were stored into zip-lock bags separately before keeping at -20°C .

Tab. 1: Species, stranding date and location, and main characteristics of stranded cetaceans.

Species	Stranding date	Stranding location	Sex	Life stage	Total length (m)
Finless porpoise (<i>Neophocaena phocaenoides</i>)	12-Feb	Beringgis, Papar	♀	Adolescent	0.8
Spinner dolphin (<i>Stenella longirostris</i>)	3-Mar	Kota Kinabalu city bay	♂	Adult	1.50
Short-finned pilot whale (<i>Globicephala macrorhynchus</i>)	19-Mar	Likas Bay, Kota Kinabalu	♂	Adolescent	3.36
Finless porpoise (<i>N. phocaenoides</i>)	24-Jun	Survivor Island	♀	Calf	0.70
Short-finned pilot whale (<i>G. macrorhynchus</i>)	10-Dec	Tajau Laut Village, Kudat	♂	Calf	1.60

Each tissue was sub-sampled into five replicates and stored in 5 mL blue cap tubes. The samples were freeze-dried (FreeZone 6L Benchtop, Labconco, Kansas, USA) following Aubail *et al.* (2013). The freeze-dried samples were homogenized with a pulverizer (M6 Yuan Tai Chi Marvelous Grinding Machine, Taiwan).

Metals extraction

Heavy metals extraction was carried out using a microwave digestion system (Milestone Start D, USA). The freeze-dried and homogenized samples of 0.3 g were weighed with an analytical balance and put into 25 mL Teflon digestion tubes. Each Teflon tube was added with 8 mL of 65 % nitric acid (HNO_3) and 2 mL of 30 % hydrogen peroxide (H_2O_2) by following modified method of Ding *et al.* (2014). The samples were digested for 15 min with constant increase of temperature to 120°C , followed by 15 min at this maximal temperature. Each resulted solution was made up to 25 mL with milli-Q water after Teflon tubes were cooled down. Solutions were kept in borosilicate vials and stored at 4°C before determination of heavy metals with an inductive coupled plasma optical emission spectrometry (ICP-OES) (Perkin Elmer, Optima 5300DV, USA).

Quality assurance and control

All glassware used throughout laboratory works were rinsed with 1 N nitric acid followed by ultra-pure water

before use. Blank sample and certified reference material (CRM) DOLT-5 from National Research Council Canada was analyzed using the same method mentioned above. Values of extracted concentration were close to certified values, thus method used is acceptable (Tab. 2).

Tab. 2: Extracted and certified values of CRM (mean \pm standard deviation).

Elements	Certified values (mg kg^{-1})	Detected values (mg kg^{-1})	Recovery (%)
As	34.6 (± 2.4)	27.30 (± 0.40)	79.94
Cd	14.5 (± 0.60)	12.72 (± 0.10)	87.73
Cr	2.35 (± 0.58)	2.76 (± 0.33)	117.49
Cu	35.00 (± 2.40)	40.06 (± 0.50)	114.46
Fe	1070.0 (± 80.0)	991.87 (± 90)	92.7
Mn	8.91 (± 0.70)	9.08 (± 0.10)	101.96
Pb	0.16 (± 0.03)	0.156 (± 0.17)	97.5
Se	8.30 (± 1.80)	10.8 (± 2.40)	130.12
Zn	105.30 (± 5.40)	79.50 (± 2.00)	75.5

Data analysis

Median concentrations of 5 replicates for each metals were used as representative values to calculate the percentage of metals in each tissue for comparison of composition of each metal in different tissues. The data were expressed as median \pm standard error (SE) based on dry weight (dwt), and 95% confidence interval of each median was calculated for the statistical comparison. Wet weight data taken from other studies for comparison purpose was converted

to dry weight using conversion factor 0.28 (Yang and Miyazaki, 2003).

Results

In all the cetacean studied, Fe level was found to be

the highest among all metals (216.5–1710.21 $\mu\text{g g}^{-1}$), followed by Zn (6.81–297.68 $\mu\text{g g}^{-1}$) (Tab. 3). Accumulation of Zn showed no species difference in the 5 cetaceans. Mn concentrations was also found to be high in the livers of all cetaceans examined.

Tab. 3: Concentrations (median \pm se) in $\mu\text{g g}^{-1}$ dwt in each tissue of cetaceans (NA = not analysed; ND = not detected).

		Short-finned pilot whale								
	Maturity	As	Cd	Cr	Cu	Fe	Mn	Pb	Se	Zn
Kidney	Adolescent	2.55 (± 0.05)	98.65 (± 4.32)	4.90 (± 0.30)	13.55 (± 0.50)	544.93 (± 22.42)	4.84 (± 0.10)	1.63 (± 0.21)	28.99 (± 0.94)	57.73 (± 1.72)
	Calf	0.43 (± 0.09)	1.60 (± 0.01)	8.20 (± 1.08)	23.67 (± 1.60)	433.91 (± 35.02)	9.20 (± 0.30)	8.40 (± 1.75)	13.00 (± 0.58)	88.31 (± 1.27)
Liver	Adolescent	0.94 (± 0.14)	69.96 (± 1.97)	2.75 (± 0.26)	18.19 (± 0.63)	1498.12 (± 53.67)	12.40 (± 0.38)	2.01 (± 0.53)	77.02 (± 4.45)	108.55 (± 15.43)
	Calf	0.53 (± 0.29)	1.59 (± 0.02)	4.79 (± 0.65)	44.93 (± 2.81)	638.78 (± 60.80)	16.97 (± 0.47)	ND	10.07 (± 0.66)	56.47 (± 0.95)
Heart	Adolescent	1.96 (± 0.08)	3.13 (± 0.04)	3.59 (± 0.36)	9.34 (± 0.80)	350.03 (± 55.76)	3.63 (± 0.12)	2.13 (± 0.20)	16.99 (± 2.28)	63.04 (± 10.49)
	Calf	0.26 (± 0.20)	1.43 (± 0.01)	3.75 (± 0.33)	12.00 (± 1.00)	955.66 (± 66.59)	5.67 (± 0.45)	1.52 (± 1.53)	12.14 (± 0.85)	64.34 (± 5.66)
Lung	Adolescent	2.11 (± 0.16)	4.87 (± 0.05)	4.49 (± 0.27)	7.53 (± 1.15)	870.39 (± 44.74)	3.42 (± 0.08)	2.39 (± 0.23)	13.69 (± 0.77)	30.82 (± 5.87)
	Calf	0.26 (± 0.13)	1.40 (± 0.01)	3.83 (± 0.34)	7.39 (± 0.27)	1710.21 (± 24.49)	4.52 (± 0.17)	0.26 (± 0.26)	13.30 (± 0.21)	43.29 (± 2.28)
Melon	Adolescent	0.49 (± 0.11)	1.59 (± 0.13)	2.50 (± 0.15)	9.32 (± 2.06)	166.22 (± 55.37)	2.20 (± 0.27)	ND	12.10 (± 1.42)	8.19 (± 0.89)
	Calf	0.74 (± 0.21)	1.59 (± 0.01)	5.42 (± 1.10)	13.63 (± 2.32)	315.50 (± 14.58)	3.13 (± 0.10)	ND	8.93 (± 0.88)	10.33 (± 1.68)
Muscle	Adolescent	1.06 (± 0.16)	1.87 (± 0.10)	4.05 (± 0.25)	7.97 (± 1.38)	898.82 (± 35.86)	3.05 (± 0.11)	2.15 (± 0.73)	8.57 (± 0.32)	85.71 (± 5.02)
	Calf	0.30 (± 0.04)	1.42 (± 0.00)	3.01 (± 0.10)	6.55 (± 0.24)	373.43 (± 12.34)	2.44 (± 0.04)	1.17 (± 0.08)	9.51 (± 0.23)	54.69 (± 1.68)
Blubber	Adolescent	0.57 (± 0.09)	1.68 (± 0.01)	2.67 (± 0.22)	10.76 (± 1.33)	225.44 (± 20.98)	2.68 (± 0.13)	1.64 (± 0.65)	12.28 (± 0.44)	11.24 (± 0.68)
	Calf	0.94 (± 0.12)	1.21 (± 0.02)	3.90 (± 3.38)	9.32 (± 13.94)	132.31 (± 21.13)	1.96 (± 0.12)	2.12 (± 1.03)	12.18 (± 0.55)	35.12 (± 6.92)
		Finless porpoise								
	Maturity	As	Cd	Cr	Cu	Fe	Mn	Pb	Se	Zn
Kidney	Adolescent	1.50 (± 0.24)	1.34 (± 0.02)	3.36 (± 0.29)	24.75 (± 3.59)	601.82 (± 18.15)	4.49 (± 0.28)	0.81 (± 0.55)	11.14 (± 0.82)	83.12 (± 1.28)
	Calf	0.91 (± 0.14)	1.26 (± 0.01)	5.42 (± 0.37)	25.72 (± 3.87)	541.40 (± 20.29)	3.90 (± 0.18)	1.53 (± 0.17)	6.11 (± 0.94)	84.15 (± 1.29)
Liver	Adolescent	2.03 (± 0.22)	1.30 (± 0.02)	4.37 (± 1.16)	28.99 (± 2.84)	1073.79 (± 38.56)	18.91 (± 1.20)	0.50 (± 0.22)	10.13 (± 0.94)	297.68 (± 25.62)
	Calf	0.85 (± 0.16)	1.41 (± 0.01)	3.62 (± 0.69)	70.43 (± 4.79)	859.01 (± 15.72)	5.37 (± 0.08)	0.35 (± 0.26)	3.06 (± 0.66)	143.42 (± 5.18)
Heart	Adolescent	1.23 (± 0.26)	1.28 (± 0.02)	5.47 (± 0.91)	17.32 (± 3.07)	609.61 (± 105.74)	3.68 (± 0.48)	1.52 (± 0.31)	5.06 (± 0.18)	85.96 (± 16.69)
	Calf	1.67 (± 0.18)	1.90 (± 0.02)	14.29 (± 1.75)	24.34 (± 0.40)	475.86 (± 33.77)	4.41 (± 0.12)	3.63 (± 3.74)	6.07 (± 0.78)	82.99 (± 1.20)
Lung	Adolescent	1.60 (± 0.12)	1.24 (± 0.01)	2.72 (± 0.19)	8.24 (± 0.13)	802.47 (± 15.10)	2.90 (± 0.12)	0.11 (± 0.03)	6.22 (± 0.36)	61.31 (± 1.16)
	Calf	NA	NA	NA	NA	NA	NA	NA	NA	NA
Melon	Adolescent	1.85 (± 0.38)	1.45 (± 0.02)	2.69 (± 0.19)	8.30 (± 0.78)	298.56 (± 33.46)	2.86 (± 0.16)	ND	3.62 (± 0.25)	6.81 (± 1.25)
	Calf	0.64 (± 0.20)	1.48 (± 0.04)	5.81 (± 2.02)	10.17 (± 18.03)	184.82 (± 30.83)	2.36 (± 0.44)	2.57 (± 2.21)	5.53 (± 0.56)	44.37 (± 6.11)
Muscle	Adolescent	1.05 (± 0.07)	1.23 (± 0.00)	3.78 (± 0.17)	7.23 (± 0.28)	454.71 (± 15.91)	2.02 (± 0.05)	2.15 (± 0.59)	5.90 (± 0.29)	51.62 (± 3.31)
	Calf	3.52 (± 0.21)	3.79 (± 0.03)	8.64 (± 0.48)	18.70 (± 0.64)	350.99 (± 21.26)	5.36 (± 0.10)	5.35 (± 0.56)	ND	102.31 (± 3.70)
Blubber	Adolescent	1.50 (± 0.12)	1.43 (± 0.02)	3.53 (± 0.14)	5.53 (± 0.58)	257.16 (± 12.34)	2.34 (± 0.06)	1.64 (± 0.56)	2.96 (± 0.27)	15.72 (± 2.68)
	Calf	2.05 (± 0.37)	3.72 (± 0.19)	10.71 (± 1.12)	8.24 (± 0.58)	216.58 (± 28.48)	4.54 (± 0.30)	3.61 (± 0.91)	3.99 (± 0.67)	43.43 (± 1.85)

Tab. 3: Continued.

		Spinner dolphin							
Maturity	As	Cd	Cr	Cu	Fe	Mn	Pb	Se	Zn
Kidney	3.26 (±0.19)	35.04 (± 0.41)	3.45 (±0.17)	26.96 (±0.54)	803.53 (±13.14)	5.45 (±0.06)	1.53 (±0.61)	28.23 (±2.22)	118.28 (±1.29)
Liver	3.28(±0.41)	8.70 (±0.23)	2.89 (±0.13)	41.53 (±1.95)	720.39 (±41.72)	9.61 (±0.67)	2.20 (±0.24)	53.74 (±1.90)	115.40 (±2.37)
Heart	2.82(±0.35)	1.88 (±0.17)	3.62 (±0.22)	25.16 (±4.12)	461.24 (±25.72)	5.20 (±0.48)	1.83 (±0.87)	20.81 (±1.26)	149.11 (±18.61)
Lung	2.27(±0.29)	2.88 (± 0.10)	3.55 (± 0.27)	11.70 (±3.96)	851.37 (± 84.23)	4.13 (± 0.27)	4.34 (± 1.00)	24.71 (± 1.48)	98.15 (±4.40)
Melon	1.69(±0.31)	1.21 (±0.03)	5.11 (±0.58)	7.48 (±1.68)	332.85 (±39.11)	2.50 (±0.19)	0.87 (±0.30)	10.28 (±0.87)	22.42 (±3.81)
Muscle	1.38(±0.10)	1.26 (±0.02)	4.01 (±0.70)	7.54 (±0.22)	477.13 (±12.84)	1.87 (±0.05)	2.28 (±0.23)	11.10 (±0.53)	36.17 (±1.99)
Blubber	2.08(±0.10)	1.22 (±0.07)	5.17 (±1.40)	2.98 (±18.12)	299.66 (±17.99)	1.74 (±0.09)	2.07 (±0.79)	9.74 (±0.44)	40.43 (±8.45)

Short-finned pilot whales

The heavy metal distribution in adolescent and calf was different. In the adolescent short-finned pilot whale, Cd in the kidney was significantly higher compare to other tissues (98.65 µg g⁻¹) (Tab. 3 and Fig. 1). Significantly high levels of essential metals were distributed in the liver of the adolescent. On the other hand, high levels of non-essential metals (As, Cd, and Cr) were preferentially accumulated in the kidney. In the calf specimen, Pb was significantly high in the kidney (62.34% or 8.4 µg g⁻¹) compared to other tissues (Tab. 3 and Fig. 1). Fe and Zn were the most abundant metals in the body of both short-finned pilot whales.

Spinner dolphin

Cd was found to be significantly high in the kidney (35.04 µg g⁻¹) (Fig. 1 and Tab. 3) compare to other non-essential metals examined which showed no significant difference in other tissues (Fig. 1). As for essential metals, Fe was abundant in all tissues, followed by Zn. Cu, Mn, and Se were significantly high in the liver compared to the kidney (Fig. 1).

Finless porpoise

Concentrations of non-essential metals in both finless porpoises were low. Fe was the most abundant metal in the body of both finless porpoises as well and its concentration was significantly high in the liver (Fig. 1 and Tab. 3). This was followed by Zn and its concentration was also significantly high in the liver (Fig. 1 and Tab. 3).

However, there were difference in metal distribution between adolescent and calf. In the adolescent finless porpoise, harmful metals (As, Cr and Pb) were about equally distributed in the kidney,

liver, and heart (Fig. 1). For the calf finless porpoise, As and Cd were preferentially accumulated in the muscle (Fig. 1). Se was not detected in the muscle of this calf.

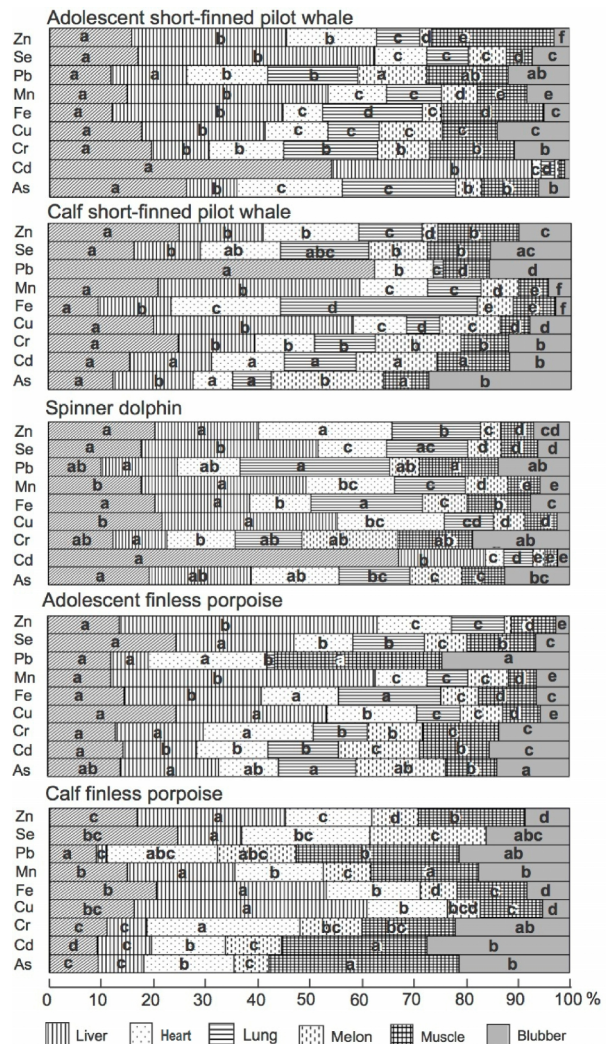


Fig. 1: Bar graphs showing ratios (% of each element total) by species. For each element, ratios marked by different letter are significantly different (95% confidence intervals).

Discussion

Generally, the concentrations of heavy metals present in the body of stranded cetaceans were low. None of the heavy metals examined was found to exceed a level which induces organ damage (Shlosberg et al., 1997; Das et al., 2003; Jakimska et al., 2011). This can be seen in Tables 4, 5 and 6 where comparison of results from current study with other studies were summarized for the liver.

Metal distribution: Interspecies (similar pattern regardless of species)

Relatively higher concentrations of Fe and Zn than other essential metals were detected in all the tissues of the 5 cetaceans, which is in agreement with study of Zhou et al., (2001) on common dolphins (*Delphinus delphis*). Concentrations of Zn were found to be not affected by species or maturity. Results were comparable to those found in other species of cetaceans (Caurant et al., 1994; Shoham-Frider et al., 2002; Kunito et al., 2004; Kamel et al., 2014). In fact, concentration of Zn in the liver was found to be not affected by age or sex (Caurant et al., 1994; Endo et al., 2007). This metal together with Cu does not usually reflect the habitat pollution because they are constantly being regulated by homeostasis in cetacean tissues (Monaci et al., 1998). The large amount of Zn was due to its importance in metabolism, proper functioning of enzymes (Jakimska et al., 2011), rapid development and differentiation of tissues as observed in other young cetaceans (Das et al., 2003) and aids in dermal protection and wound healing (Yang et al., 2002). High level of Fe in all tissues is realistic with the reason Fe is extremely important for cetacean's efficient oxygen transportation all over the body to fit their need in diving (Wafo et al., 2014).

Significantly high concentration of Cd was detected in the kidney followed by the liver of the spinner dolphin and the adolescent short-finned pilot whale (Fig. 1). Cd is stored in the liver temporarily before absorbed and bioaccumulated in the kidney (Jakimska et al., 2011) due to presence of proteins suitable for metal binding in the kidney (Das et al., 2003). The kidney Cd levels of both specimens were far below the level to induce renal dysfunction which is at 200 $\mu\text{g g}^{-1}$ dwt in human (Elinder and Järup, 1996). Marine mammals are known to be able to tolerate much higher levels Cd compare to human (Das et al., 2003).

In current study, liver Pb in all specimens ranged

from not detected (calf short-finned pilot whale) to 2.20 $\mu\text{g g}^{-1}$ dwt (spinner dolphin). This is far below subclinical effects or poisoning level and lethal level which is reported to be 10 $\mu\text{g g}^{-1}$ dwt (Jakimska et al., 2011) and approximately 12.9 $\mu\text{g g}^{-1}$ dwt (3.6 $\mu\text{g g}^{-1}$ wwt) (Shlosberg et al., 1997) respectively. Pb is often found in trace amount as it is poorly absorbed by body. But when present and slowly digested in the stomach for prolong period of time, it causes chronic toxicity (Osweiler et al., 1978) as in the case of accident ingestion of air gun pellet (40% Pb) by a captive bottlenose dolphin (*Tursiops truncatus*) which the dolphin died from progressive liver damage (Shlosberg et al., 1997).

In current study, all metals in melon of the 5 specimens were generally low (Tab. 3). This is because bioaccumulation of metals mainly relies on protein as binding site. Melon contains high lipid and low fiber which make it poor accumulation site (Honda et al., 1982). Significantly high liver Fe in the short-finned pilot whales and finless porpoises is also in agreement with those reported in bottlenose and striped dolphins (*Stenella coeruleoalba*) (Roditi-Elasar et al., 2003; Wafo et al., 2014). High Fe is commonly stored in the liver for the production of essential iron proteins such as myoglobin and haemoglobin (Yang et al., 2003). In contrast, Fe was found to be the significantly low in the melon and blubber due to the low affinity of Fe for lipid (Yang et al., 2003).

Significantly higher liver Mn were observed in the 5 specimens which is regarded as normal since Mn is an essential metals needed as cofactor enzymes to facilitate various metabolic processes (Wafo et al., 2014).

Comparison on metal distribution pattern (by species)

The distributions of every metal in the two short-finned pilot whales in this study were different due to different maturity stages. The adolescent could naturally accumulate more heavy metals from fish consumption compare to the calf which depended on milk. Within the metals considered toxic, the highest concentration measured was Cd in the kidney of the adolescent. This is due to the fact that diet of short-finned pilot whale consists of cephalopods which contain high Cd concentration and are the main source of Cd to its predators (Bustamante et al., 2008). This observation was in agreement with other squid eaters such as sperm whale, and pygmy sperm whale also contained high kidney Cd (133 – 426 and 412.6 $\mu\text{g g}^{-1}$ dwt respectively) (Marcovecchio et al., 1994; Holsbeek et

Tab. 4: Comparison for heavy metals in the liver of larger dolphins from different marine regions ($\mu\text{g g}^{-1}$ dwt).

Species	Location	As	Cd	Cr	Cu	Fe	Mn	Pb	Se	Zn	Ref.
Short-finned pilot whale (<i>G. macrorhynchus</i>)	Kota Kinabalu	0.94 (± 0.14)	69.96 (± 1.97)	2.75 (± 0.26)	18.19 (± 0.63)	1498.12 (± 53.67)	12.40 (± 0.38)	2.01 (± 0.53)	77.02 (± 4.45)	108.55 (± 15.43)	1
	Cumberland Island National Seashore	-	40.36-70.71	-	-	-	-	-	81.43- 220.00	-	2
Long-finned pilot whale (<i>G. melas</i>)	Faroe Islands	0.71-1.96	117-325	-	17.5-25.71	-	-	-	48.57-82.14	192.86-389.29	3
Risso's dolphin	Cape Cod	1.30-2.60	0.05-131.00	-	9.90-20.30	-	-	0.05-0.91	2.30-695.00	-	4
(<i>G. griseus</i>)	Israeli Mediterranean Croatian Adriatic Sea	- 1.60-16.75	51.07* 2.28-42.50	-	21.82* -	45.60* -	9.43* -	- 0.78-4.11	1350* -	129.29* -	5 6
Sperm whale (<i>P. macrocephalus</i>)	Southern North Sea	-	52-175	<0.06-0.3	5.3-11.9	1990-2810	-	<1-3.2	5.8-43	90-125	7
Pygmy sperm whale (<i>K. breviceps</i>)	Argentina coast	-	27.14*	-	36.79*	-	-	-	-	582.86*	8

* n = 1

Ref.: 1 (Current study); 2 (Stoneburner, 1978); 3 (Caurant et al., 1994); 4 (Meador et al., 1993); 5 (Shoham-Frider et al., 2002); 6 (Bilandžić et al., 2012); 7 (Holsbeek et al., 1999); 8 (Marcovecchio et al., 1994)

Tab. 5: Comparison for heavy metals in the liver of smaller dolphins from different marine regions ($\mu\text{g g}^{-1}$ dwt).

Species	Location	As	Cd	Cr	Cu	Fe	Mn	Pb	Se	Zn	Ref.
Spinner dolphin (<i>S. longirostris</i>)	Kota Kinabalu	3.28 (± 0.41)	8.70 (± 0.23)	2.89 (± 0.13)	41.53 (± 1.95)	720.39 (± 41.72)	9.61 (± 0.67)	2.20 (± 0.24)	53.74 (± 1.90)	115.40 (± 2.37)	1
Short-beaked common dolphins (<i>D. delphis</i>)	Portuguese coast	-	8.93	-	11.61-32.18	303.21-1892.86	2.68-32.57	-	-	115.71-375.00	2
	Algerian west coast	-	1.10-1.61	-	1.71-141.54	354.43-690.57	-	0.93-14.71	-	83.86-251.25	3
Long-beaked common dolphin (<i>D. capensis</i>)	Brazilian coast	2.3*	2.55*	0.42*	27.7*	1320*	12.4*	0.062*	30*	158*	4
	Kawana & Taiji	-	0.14-39.64	-	12.75-54.29	199.29-341.07	4.64-23.96	0.11-2.29	-	94.64-436	5
	Croatian waters (Adriatic Sea)	1.00 -18.07	0.43-11.18	-	-	-	-	0.14-0.34	-	-	6
Striped dolphin (<i>S. coeruleoalba</i>)	Israeli Mediterranean	-	0.25-32.14	-	3.93-75.00	460.71-2121.43	14.64*	-	-	82.14-339.29	7
	French Mediterranean	-	0.01-11.4	-	10-193	265-2958	2.0-18.1	-	1.4-2350	43-370	8
	Brazilian coast	1.2*	7.83*	0.23*	33.4*	1810*	12.3*	0.074*	190*	287*	3

Tab. 5: Continued

Species	Location	As	Cd	Cr	Cu	Fe	Mn	Pb	Se	Zn	Ref.
Bottlenose (<i>T. truncatus</i>)	Croatian Adriatic Sea	0.39–18.07	0.27–16.00	–	–	–	–	0.13–1.36	–	–	6
	Israeli Mediterranean	–	0.43–3.93	–	15.36–85.71	582.14–3007.14	4.64–23.21	–	–	53.57–410.71	7
Franciscana (<i>P. blainville</i>)	Brazilian coast	0.72–2.4	0.129–3.87	0.21–3.1	17.5–88.0	546–1330	10.4–16.1	0.008–0.070	6.8–30	111–156	3
	Argentina coast	–	11.79 ^a	–	57.14 ^a	–	–	–	–	297.86 ^a	9
Estuarine dolphin (<i>S. guianensis</i>)	Brazilian coast	0.27–1.6	0.19–2.19	0.26–5.1	14.5–39.3	330–1470	5.91–15.1	0.028–0.197	4.7–170	117–348	4
Atlantic spotted dolphin (<i>S. frontalis</i>)	Brazilian coast	0.68–0.71	5.89–56.0	0.29–0.98	33.3–47.9	468–1270	12.3–17.4	0.037–5.12	27–130	146–575	4

^amean, n = 1

Ref.: 1 (Current study); 2 (Zhou et al., 2001); 3 (Kamel et al., 2014); 4 (Kunito et al., 2004); 5 (Honda et al., 1983); 6 (Bilandžić et al., 2012); 7 (Roditi-Elasar et al., 2003); 8 (Wato et al., 2014); 9 (Marcovecchio et al., 1994)

Tab. 6: Comparison for heavy metals in the liver of porpoises from different marine regions ($\mu\text{g g}^{-1}$ dwwt).

Species	Location	Life stage	As	Cd	Cr	Cu	Fe	Mn	Pb	Se	Zn	Ref.
Finless porpoise (<i>N. phocaenoides</i>)	Kota Kinabalu	Adolescent	2.03 (± 0.22)	1.30 (± 0.02)	4.37 (± 1.16)	28.99 (± 2.84)	1073.79 (± 38.56)	18.91 (± 1.20)	0.50 (± 0.22)	10.13 (± 0.94)	297.68 (± 25.62)	1
		Calf	0.85 (± 0.16)	1.41 (± 0.01)	3.62 (± 0.69)	70.43 (± 4.79)	859.01 (± 15.72)	5.37 (± 0.08)	0.35 (± 0.26)	3.06 (± 0.66)	143.42 (± 5.18)	
Dall's porpoise (<i>P. dalli</i>)	Sanniku coast, Japan	Adult	–	36.79	0.59	34.54	1517.86	18.21	0.086	27.36	130.00	2

Ref.: 1 (Current study); 2 (Yang et al., 2006)

al., 1999). This phenomenon was not observed in the calf because mammary milk contains low Cd concentration ($0.0014 \mu\text{g g}^{-1}$ dwt) (Frodello *et al.*, 2002). Another reason for low Cd concentration detected in calf is because this element does not cross placenta (O'Hara and O'Shea, 2001) and often range from not detected to extremely low level in cetacean foetus ($0.003 \mu\text{g g}^{-1}$ wwt) (Honda *et al.*, 1982; Fujise *et al.*, 1988).

Cu and Se were significantly high in the liver of adolescent short-finned pilot whale (Fig. 1). This was also reported in other pilot whales (Meador *et al.*, 1993). In marine mammals, the levels of Cu and Se are determined by both physiological needs and environmental exposure which make it difficult to interpret (Meador *et al.*, 1993). The high Cu, Zn and Se levels in this adolescent can only be explained by physiological need. The significantly high Se level in the liver was found in adolescent but not calf due to the need to mitigate mercury bioaccumulation (by forming mercury selenide) in older cetacean (Ostertag *et al.*, 2014). Significantly high liver Cu and Zn could be explained by result of starvation. The adolescent was thought to be in an emaciated condition before death because the stomach was compacted with plastic sheets at sampling which make no room for food. In the study by Debacker *et al.* (2000) on guillemots (*Uria aalge*), concentrations of Cu and Zn in the liver, kidney and muscle were found to increase directly with cachexia severity.

The highest measured concentration metal in the spinner dolphin was Cd in the kidney. This is in agreement with Wafo *et al.*, (2014) who stated that kidney Cd was 4 times higher in the kidney compared to the liver. Its distribution was also same as striped dolphins; kidney > liver > lung (Wafo *et al.*, 2014; Bilandžić *et al.*, 2012). The significantly high accumulation of Cu, Zn, and Se in the liver in this adult specimen is inconsistent with the study on striped dolphin (Wafo *et al.*, 2014).

Cd and As in calf finless porpoise of current study were preferentially accumulated in the muscle which was in contrast to other marine mammal studies which stated that high Cd and As concentrations are commonly found in the kidney (Yang *et al.*, 2006; Das *et al.*, 2003). This was perhaps caused by starvation and toxicity which leads to metal redistribution from contaminated organs (liver or kidney) to muscle as a protection mechanism from toxicity (Das *et al.*, 2003).

Conclusion

According to the results, liver and kidney were found to be the most contaminated organs due to the role of these two organs in detoxifying and re-distributing metals. Blubber was least contaminated. However, heavy metals levels and distribution patterns in local's stranded cetaceans were generally in agreement with the accumulation patterns reported in other studies, regardless of marine regions. Current study represents the first report of heavy metals in cetaceans stranded in Malaysia. Through this study, no specific concern was found regarding contamination of local marine resources. However, it is encouraged such study to be carried out continuously to monitor the changes in long term.

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