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Blood plasma levels of heavy metals and trace elements in white sharks (*Carcharodon carcharias*) and potential health consequences

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ABSTRACT

Heavy metals may adversely affect health in marine organisms. As top predators, sharks may be especially vulnerable to exposure over long lifespans. Here we evaluate plasma levels of 14 heavy metals and 12 trace elements in white sharks, *Carcharodon carcharias*, in South Africa to determine whether they are related to sex, body size, and/or body condition and other health parameters. High levels of mercury and arsenic were found in shark blood at levels considered toxic in other vertebrates. Heavy metal concentrations were not related to body size or sex. Metal concentrations were not related to body condition with exception of copper, which was positively correlated. Protective effects of elements such as selenium, zinc, and iron were not detected. No negative effects on health parameters, such as total leukocytes or granulocyte to lymphocyte ratios were observed. Results suggest that sharks may have protective mechanisms that mitigate harmful effects of heavy metal exposure, providing new opportunities for future studies.

1. Introduction

Due to occupying upper-trophic levels in food webs, predators generally possess higher tissue concentrations of mercury and other toxic metals. In sharks, exposure and heavy metal toxicity is primarily derived via dietary uptake (Pethybridge et al., 2010; Matulik et al., 2017). Heavy metals from the diet are absorbed into the blood and then distributed to various organs, suggesting that levels measured in the blood should correspond to metals being present in tissues (Ollson et al., 1998). The level of heavy metals such as mercury, arsenic, and lead has been evaluated in muscle tissues of several shark species as it relates to toxicity levels for human consumption (Adams and McMichael, 1999; Rumbold et al., 2014; Hammerschlag et al., 2016; Mohammed and Mohammed, 2017; Lavoie et al., 2018). More recently, several studies have investigated the levels of heavy metals present in shark muscle tissues and related them to environmental stressors like pollution (Storelli et al., 2002). Despite this work, it remains poorly known whether there are generally discernible differences in heavy metal exposure among different individuals within a population (i.e. age/size differences, sex differences). Moreover, references intervals for heavy metals in the blood of sharks are not available in the literature. While determining patterns of heavy metal concentrations in sharks is important from a human food perspective, the impacts of heavy metal exposure on the well-being of wild sharks is not well understood.

Given wide-spread population declines of many shark species and their inherent vulnerabilities to anthropogenic threats (Gallagher et al., 2012), it is of interest to determine the effect, if any, of accumulated metals on shark health and fitness (Depew et al., 2012). Sharks exposed to high levels of heavy metals over their lifetime may be at increased risk for numerous pathologies including neurodegenerative effects, deregulated enzymatic and tissue function, compromised immune function, and increased oxidative stress. Heavy metals such as arsenic, mercury, and lead have been found in other species to negatively impact neurological function (Tyler and Allan, 2014; Papp et al., 2006). If

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there were similar neurodegenerative effects on sharks, it might alter various behaviors such as migratory activity or foraging efficiency. Changes in foraging behavior could lead to decreased body condition and have additional ill health effects. Heavy metals can also negatively affect enzymatic and signaling pathways within host tissues due to their interactions with various important mediators (Shen et al., 2013; Hughes, 2002). Mediators of the immune system, for example, are known to be modulated by exposure to various metals and by increased concentrations of certain metals in the blood, with some metals like zinc serving as integral components of immune function (Rink and Kirchner, 2000). Changes in immune function could lead to higher susceptibility to disease (Segner et al., 2012; Zelikoff, 1993; Witeska, 2005).

While exposure to some heavy metals like mercury and lead might be expected to increase oxidative stress, the presence of micronutrient metals such as zinc, selenium, and iron, might provide antioxidant defenses that minimize the impact because they serve as co-factors for key regulatory and antioxidant enzymes (Nam et al., 2011; Barrera-Garcia et al., 2013). For example, selenium has been found to bind with mercury, thereby effectively reducing toxicity (Bjorklund, 2015; Corsolini et al., 2014). Similarly, selenium and zinc have been shown to have protective effects in arsenic toxicity associated with impaired neurological function in mammals and in arsenic-exposed fish (Roy and Bhattacharya, 2006; Zeng et al., 2005; Milton et al., 2004). However, the relationship between levels of various heavy metals and other trace elements found in the blood is not well understood for sharks.

The purpose of the present study was to evaluate blood concentrations of 14 heavy metals in white sharks (*Carcharodon carcharias*) off South Africa and to investigate how these metals relate to one another, and to 12 other blood chemistry parameters, as well as to differences in sex, size, and body condition. Using these data, we addressed nine specific questions: (1) what are the baseline, reference intervals for blood chemistry and plasma levels of heavy metals and trace elements in the sampled population of South African white sharks? (2) How do heavy metal plasma levels compare to those levels considered toxic in other taxonomic groups? (3) Do blood levels of heavy metals differ among males and females? (4) Do larger sharks have higher concentrations of heavy metals than smaller sharks? (5) Do sharks with higher heavy metal levels exhibit lower body condition? (6) Do sharks with higher heavy metal concentrations exhibit signs of increased oxidative stress, decreased enzyme function, or immunotoxicity? (7) Are there any synergistic or antagonistic relationships between blood parameters that might explain effects of heavy metal exposure on shark well-being? Finally (8) are micronutrient metals such as selenium, manganese, iron, and zinc positively correlated with heavy metal concentrations and/or shark body condition as would be expected if they are providing antioxidant defenses against metal toxicity?

2. Methods

2.1. Sampling and blood analyses

Between March and May of 2012, a total of 43 white sharks were captured and sampled on the R/V Ocearch at five different localities across South Africa: Algoa Bay, False Bay, Gansbaai, Mossel Bay, and Struisbaai. Details on capture and handling methods can be found in Wcisel et al. (2015) and Hammerschlag et al. (2017). Briefly, sharks were captured with baited barbless hooks and carefully lead onto a hydraulic platform. One or two hose(s) were then inserted into the shark's mouth to pump fresh, oxygenated saltwater over the gills. Sharks received antibiotics and electrolyte injections to enhance recovery time.

Sharks were sexed and then measured (in cm) for pre-caudal length (PCL), fork length (FL), total length (TL) and girth. Body condition is typically viewed as an index of overall health, and is usually defined as body mass or body girth relative to body length (Jakob et al., 1996; Green, 2001; Irschick and Hammerschlag, 2014). Here, we calculated body condition as the ratio of shark girth to length (PCL).

Blood was collected via caudal venipuncture using a sterile 20 mL syringe and an 18 gauge needle. Whole blood was used to fill tubes containing sodium heparin as an anticoagulant. Tubes were rocked gently for several minutes before being centrifuged immediately for 5 min at 2000 rpm. Plasma was extracted using a Pasteur pipette into a sterile cryotube; the packed red blood cells were transferred into a separate cryotube. All cryotubes were then stored in liquid nitrogen until returned to the laboratory where they were transferred to a − 80 °C freezer until analyses were performed.

On-board blood smears were prepared from the blood of 15 sharks. Slides were labeled, air-dried, and stored in a slide box. Smears were fixed in ethanol and stained with Diff-Quick (Labor & Technik, Eberhard Lehman GmbH). Estimated total leukocyte counts were done on the thinnest part of smears. Total counts are reported as the mean number of leukocytes counted in 10 high-powered fields (hpf). Differential counts were performed manually using an Olympus binocular light microscope with 400× magnification. A total count of 100 cells were identified to obtain percentages. Leukocytes were identified as granulocytes, lymphocytes, and monocytes. The total granulocyte to lymphocyte ratio (GLR) was calculated for each shark.

Frozen plasma samples were received for chemical and metal analysis. A total of 30 samples were found to be suitable for chemical analysis. All samples were analyzed without any dilution or other modifications. Analyses of trace elements were conducted in South Africa at PathCare Laboratory in Cape Town. The samples were thawed at room temperature before analysing by colorimetry according to manufacturer specifications for human samples on a Beckman Coulter LX 20 (Mikolaenko et al., 2000). The degree of hemolysis in plasma samples was assessed by a semi-quantitative method (Beckman Coulter, LIH test) and any samples measured as severely hemolysed (> 200 mg/dL) were not analyzed. The plasma was tested for Na, K, Cl, urea, Ca, Mg, PO4, protein, albumin, ALP, AST and CK values.

After chemical analyses, the remainder of samples were submitted to V&M Analytical Toxicology Laboratory Services for trace element and metal analysis. The analysis was done by Inductively Coupled Plasma-Mass-Spectrometry or ICP-MS (van der Ven, 2018). Concentrations of 14 metals were done: Cr, Mn, Fe, Co, Ni, Cu, Zn, As, Mo, Cd, Sb, Pb, Hg, and Se. Four of the samples could not be used due to insufficient or dried out samples.

2.2. Data analyses

Descriptive statistics (mean, standard deviation, standard error, minimum and maximum values) were calculated for blood chemistry and plasma levels of heavy metals and trace elements in the sampled population of white sharks. For arsenic and mercury, plasma levels were compared to those found to be toxic in teleosts in other studies and those considered toxic for human consumption, respectively. Differences among males and females for each heavy metal were also evaluated using a *t*-test.

Since heavy metals are acquired over the life-span of the animals and concentrations have previously been found to be influenced by shark age/size (Nicolaus et al., 2016, 2017), we used Pearson Correlation to test if either shark length or girth was positively correlated with concentrations of any of the 14 heavy metals. Similarly, given the potential for negative effects of heavy metal toxicity on shark health (Depew et al., 2012), we tested if shark body condition was negatively correlated with heavy metal concentrations. Since the presence of some heavy metals might also influence other blood chemistry parameters, we similarly evaluated if any heavy metals were negatively correlated to plasma chemistry levels such as liver enzymes. Heavy metal exposure can also affect immunocompetent cells of peripheral blood. Total leukocyte counts and the ratio of leukocytes present can reveal immunomodulatory impacts in vertebrates. Granulocyte to lymphocyte

ratio is used as a proxy measure for physiological stress and systemic inflammation in other vertebrates (Davis et al., 2008). The relationship between heavy metals and GLR was investigated using a Pearson Correlation to test whether the presence of heavy metals decreased total leukocyte counts or increased GLR values.

The relationship between heavy metals and trace elements, such as zinc, selenium, and iron, was also investigated using a Pearson Correlation to test whether trace elements were negatively correlated to heavy metals. Of particular interest was the relationship between selenium and mercury, given previously demonstrated protective effects of selenium on mercury toxicity. Accordingly, the molar ratio of selenium to mercury was calculated and correlated against body condition. Finally, a Pearson Correlation was also used to test if trace elements were positively correlated to each other. Data were all log (value + 1) transformed prior to statistical analyses. An alpha level of 0.05 was considered statistically significant.

3. Results

Up to 43 white sharks were sampled for biometric data as well as blood collection. Biometric information for the sharks used in this study are summarized in Table 1. The mean size, TL, was 333.79 cm (N = 43) and mean body condition level was 0.68 (N = 31). Of the sharks sampled, blood chemistry analyses were performed for 29 individuals as shown in Table 2. Values presented here may represent a set of reference ranges for these parameters in this population.

Levels of various heavy metals in plasma were measured for 26 sharks and results are summarized in Table 3. High levels of arsenic and mercury were observed across most sharks in the study. In the case of mercury and arsenic, levels in the blood were greater than those considered toxic in human and fish blood (Fig. 1). There were no significant differences between males and females in levels of these metals. Levels of arsenic and mercury were not correlated with body condition (Fig. 2).

Lead and copper were measured across all sharks. Levels of lead were well below that which is considered toxic. Among all metals tested, copper was the only metal found to be correlated with body condition, and the relationship was positive (Fig. 2, $p = 0.0475$). The level of copper in the blood was also correlated to enzymes, alkaline phosphatase ($p = 0.0487$) and creatine kinase ($p = 0.0104$). Nickel, cadmium, cobalt, and chromium were also measured in the study (Fig. 3). No heavy metals were correlated to any measures of body size (Fig. 4).

Trace elements including zinc, iron, manganese, and selenium were high, but none were correlated to either body size or body condition (Table 3). Zinc levels were positively correlated to iron levels (Fig. 5, $p = 0.0002$). Arsenic and lead were positively correlated with zinc blood levels ($p < 0.010$). The mean molar ratio between selenium and mercury was > 1, but was not found to be correlated to either body size or condition. The level of heavy metal present in the blood was not negatively correlated with total leukocyte count or positively correlated with GLR values for all metals tested, indicating no observed immunotoxic effects. Reference ranges for leukocyte counts and GLR values are presented in Table 4.

Table 1
Biometric information for sharks sampled in this study.

Body measurements	Mean	Std Dev	Std Error	Minimum	Maximum	N
TL	333.79	74.09	11.30	210	505	43
FL	301.72	73.98	11.28	100	462	43
PCL	273.88	60.33	9.31	173	422	42
Girth	190.55	55.88	10.04	99	305	31
Condition	0.68	0.06	0.01	0.55	0.85	31

Table 2
Blood chemistry values for plasma samples for 29 sharks.

Blood chemistry	Mean	Std Dev	Std Error	Minimum	Maximum	N
Na	242.90	47.05	8.74	4.0	264.0	29
K	12.48	44.15	8.20	3.1	242.0	29
Cl	229.17	45.62	8.47	0.0	259.0	29
Urea	439.66	24.87	4.79	345.8	464.6	29
Ca	3.55	0.50	0.09	1.2	3.9	29
Mg	1.32	0.27	0.05	0.9	2.3	29
PO4	2.89	3.69	0.69	1.0	22.0	29
Prot	26.52	12.94	2.40	0.0	83.0	29
Glob	12.97	11.55	2.14	0.0	68.0	29
ALP	12.24	6.14	1.14	3.0	31.0	29
AST	15.24	12.72	2.36	3.0	63.0	29
CK	32.75	41.77	7.89	10.0	195.0	28

Table 3
Total heavy metals and trace elements measured in plasma samples for 26 sharks.

Heavy metals	Mean (μg/L)	Std Dev	Std Error	Minimum	Maximum	N
Cr	2.88	1.24	0.24	1.3	5.4	26
Mn	24.83	37.04	7.26	0.5	189.5	26
Fe	1485.80	862.93	169.23	427.5	4685.7	26
Co	3.64	1.67	0.33	0.9	7.1	26
Ni	7.05	29.74	5.83	0.0	152.8	26
Cu	398.62	127.58	25.02	218.9	883.9	26
Zn	457.09	103.04	20.21	274.8	868.5	26
As	833.43	781.09	153.18	252.4	4520.3	26
Mo	0.50	1.68	0.33	0.0	8.7	26
Cd	0.09	0.08	0.02	0.0	0.2	26
Sb	0.28	0.35	0.07	0.0	1.5	26
Pb	1.58	3.22	0.63	0.2	16.3	26
Hg	146.98	67.25	13.19	36.0	265.5	26
Se	159.50	36.09	6.95	112.8	247	27

4. Discussion

To our knowledge, here we provide the first published accounts of blood plasma concentrations of heavy metals and trace elements in wild sharks. The level of heavy metals present in the blood of white sharks was high for mercury, arsenic, and copper, when compared to those levels found thus far in elasmobranch studies measuring heavy metals in tissue samples such as muscle and liver (Mull et al., 2012). The mean level of mercury found in white shark plasma exceeded that which is considered toxic for humans (EFSA, 2015; Bernhoft, 2012; Taylor et al., 2014). The level of arsenic present in the blood of these sharks was substantially higher than that recorded in studies where arsenic was measured in tissues, where it is expected to be concentrated. Levels were also higher than those expected to cause adverse effects in teleosts such as catfish, salmon, and carp (Kavitha et al., 2010; Kumar and Banerjee, 2016; Budiati, 2010). Similarly, the level of arsenic was higher than that considered toxic to humans (FDA, 2016). For copper, the levels found in all sharks tested were higher than that considered acutely lethal in juvenile rainbow trout and salmon (Price, 2013). Levels of other metals were lower than those expected to cause toxicity. For example, the level of lead present was significantly lower than that expected to be toxic to fish or mammals (Gidlow, 2004).

Blood levels were not significantly different between males and females for arsenic and mercury. No sex differences were observed across any metals or trace elements tested. Previous studies have indicated differences in observed concentrations of heavy metals in the muscle and liver of dogfish sharks among male and female specimens. However, differences were primarily due to differences in the growth rate for males and females and growth following sexual maturity (Endo et al., 2013).

Several studies have shown a positive relationship between shark size and heavy metal concentrations (Endo et al., 2008; Nicolaus et al.,

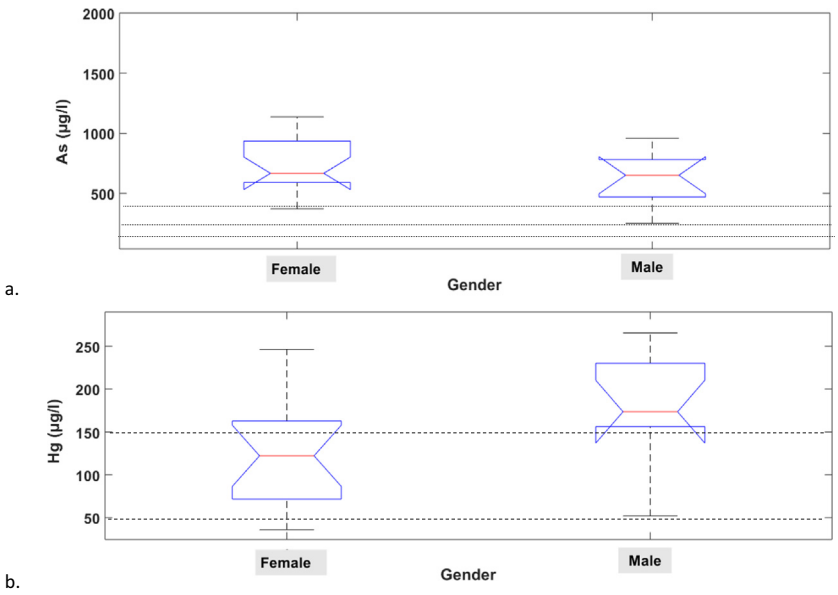

Fig. 1. Levels of total arsenic and mercury in plasma samples across sampled sharks. No significant sex differences were observed in heavy metal content. A. Mean level of arsenic in the blood was significantly higher than that considered toxic in human (1 µg/L) and fish blood (100–300 µg/L in salmon and carp) denoted by dotted lines. Highest arsenic level observed in the study for a female shark was excluded as an outlier (4520.3 µg/L). B. The level of mercury found in the blood was significantly higher than that considered toxic in humans (– –).

2016; Nicolaus et al., 2017). Unexpectedly, such a pattern was not found for white sharks in this study as no metals were significantly correlated to body size. For example, mercury was not related to any measure of body size tested. However, heavy metal levels in these previous studies were measured in the muscle and liver tissues rather than in circulation as in the present study. Tissue samples such as muscle and liver may bioaccumulate metals over time whereas, here, levels of mercury and other metals measured in the blood may be more transient and, therefore, less related to the growth and/or age of the animal. It may also indicate that the source of high levels of heavy metals measured are not due solely to diet and, instead, are more closely linked to environmental conditions. The question remains whether the high levels observed in blood indicate that levels in tissues would be significantly higher still and that exposure is concurrent with time of collection.

Another unexpected study result was that sharks with higher heavy metal concentrations did not exhibit lower levels of body condition. In fact, shark body condition was not correlated with any heavy metal, except copper and the relationship was positive. Copper is an essential nutrient and is involved as a co-factor for various critical enzymes in the body that protect against oxidative stress and, as such, levels present in these sharks may be providing some protective effects and positively impacting condition. These results suggest that shark body condition is not adversely affected by any heavy metal or trace element tested even when blood values exceed those considered toxic in related taxonomic groups like teleosts.

In addition to no apparent negative effects of heavy metal concentrations on white shark body condition, sharks with high levels of metals in the blood did not exhibit alterations in enzyme concentrations that might be indicative of oxidative stress as would have been expected. However, specific markers of oxidative stress such as superoxide dismutase (SOD) were not evaluated in this study so it is difficult to draw any major conclusions regarding the effect of heavy metals on oxidative stress. Similarly, high levels of heavy metals were not associated with changes in total leukocyte counts or the granulocyte to lymphocyte ratio or GLR, in these animals. Since leukocyte counts are only one possible measure of immune toxicity and counts were only available for a subset of sharks in this study, it is difficult to discern

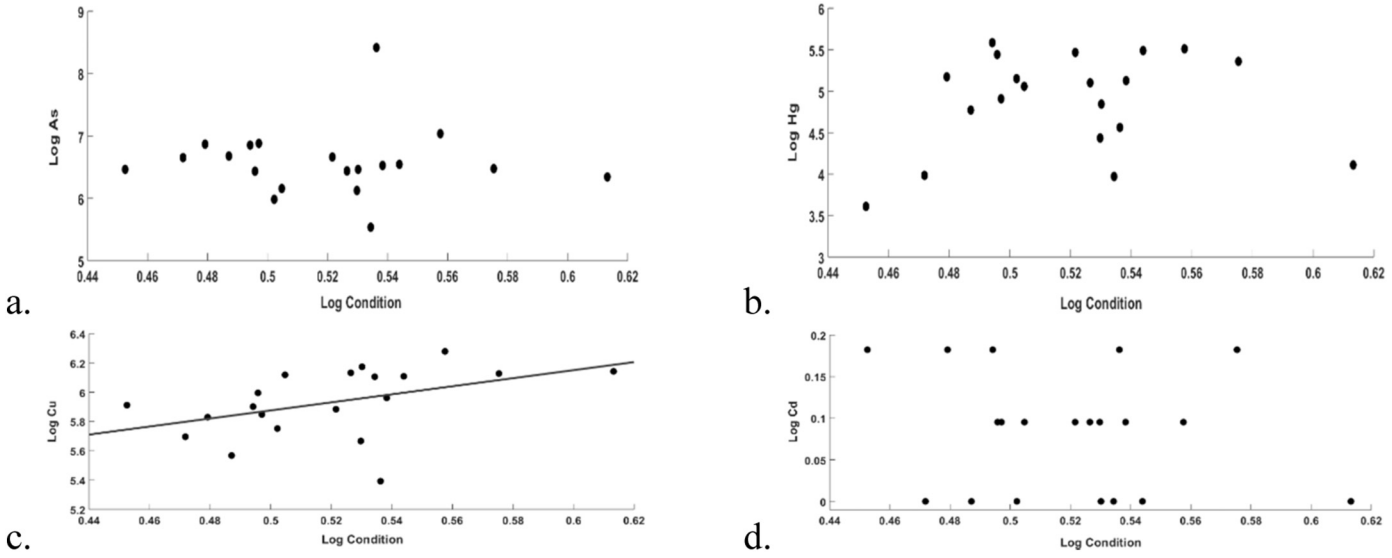

Fig. 2. Correlations between heavy metals (a) arsenic, (b) mercury, (c) copper, and (d) cadmium and body condition in sampled sharks. Copper was the only metal tested that was correlated to body condition.

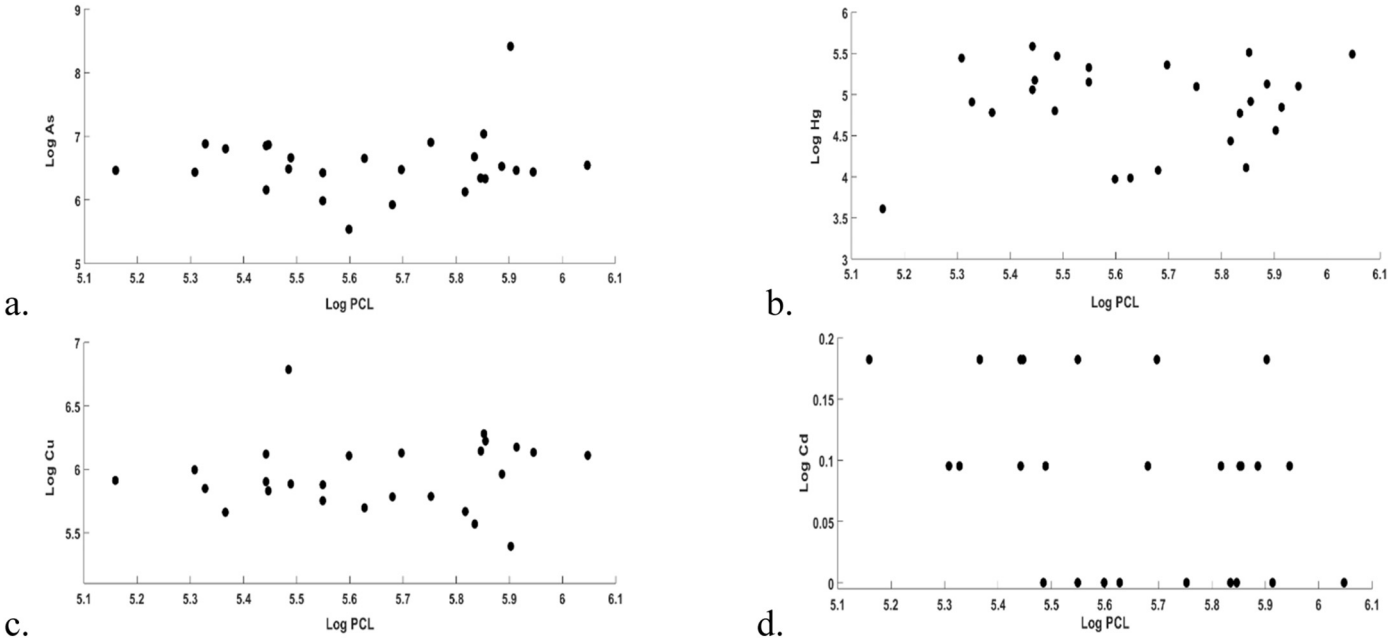

Fig. 3. Correlations between heavy metals (a) arsenic, (b) mercury, (c) copper, and (d) cadmium and body size in sampled sharks. There was no correlation between heavy metals and body size across sharks.

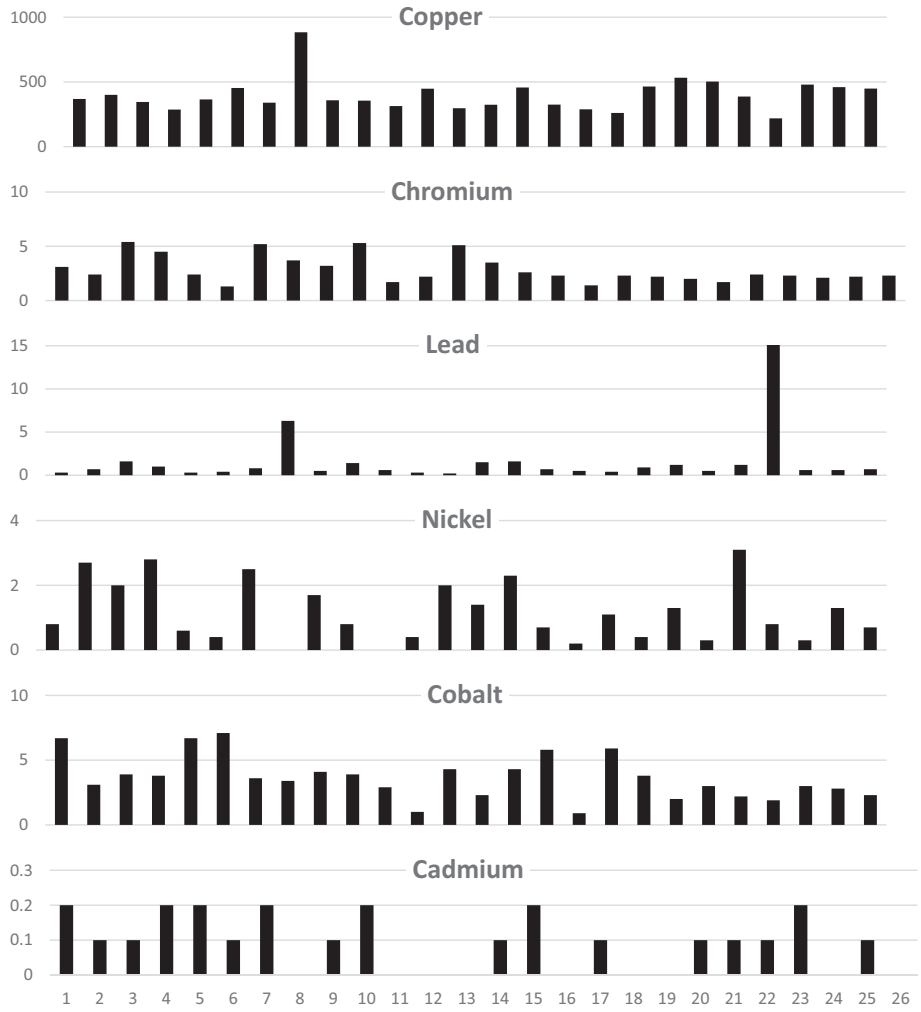

Fig. 4. Plasma levels (ug/L) of copper, chromium, lead, nickel, cobalt, and cadmium in the blood of 26 white sharks ordered from smallest to largest (PCL range 173–422 cm) along the x axis.

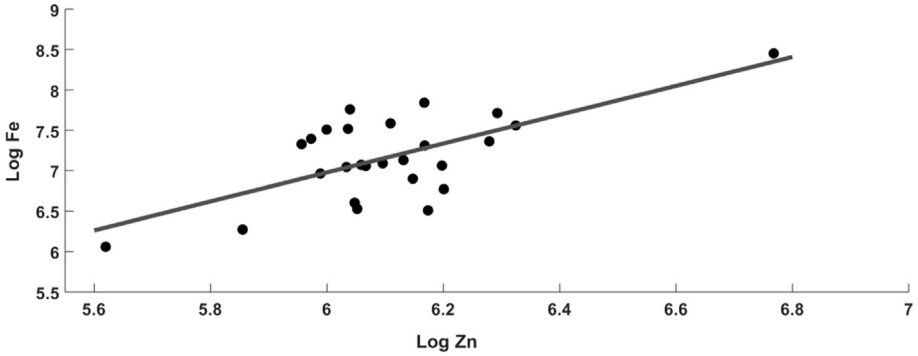

Fig. 5. Positive correlation between plasma iron and zinc levels. Among trace elements measured in this study, a positive relationship was found between iron and zinc levels across all sharks ($p = 0.0002$).

Table 4
Total leukocyte counts (estimated from peripheral blood smears, HPF 400×) and GLR values.

	Mean	Minimum	Maximum	N
Total Leukocyte Count (x 10^3)	9–10	4–5	17–18	15
GLR (Total granulocytes/total lymphocytes)	2.11	0.85	4.44	15

whether sharks with high levels of heavy metals in the blood are affected immunologically.

There was a lack of synergistic or antagonistic relationships between blood parameters and heavy metal concentration. Initial hypotheses in this study predicted that the presence of trace elements such as selenium, manganese, iron, and zinc would be protective against any oxidative stress associated with heavy metal concentrations given their role as co-factors for antioxidant enzymes and, in some cases, their ability to directly bind certain metals (Jomova and Valko, 2011). However, no relationships between these trace elements and heavy metals or with shark body condition were observed in this study.

In teleost studies, mercury toxicity is associated with disruption of enzymatic function as it binds sulfur in the cysteine residues of proteins. Among these are selenium dependent enzymes since mercury has a high affinity for selenium (Spiller, 2018). Selenium, however, also binds and sequesters mercury and is expected to have a protective effect on mercury toxicity (Sormo et al., 2011; Burger et al., 2013). However, there is substantial variation among different species of fish in these ratios and it is yet unclear how reliable ratios are in predicting overall toxicity and health outcomes. In this study, the molar ratio of selenium to mercury exceeded one for most individuals, but these ratios showed no correlation with shark body condition. Therefore, it is unclear whether selenium is providing any protective effects or whether levels of mercury in the blood are having a detrimental impact on shark health.

Exposure in the aquatic environment to high levels of arsenic can be lethal for some organisms (Bhattacharya and Bhattacharya, 2007). We realize that in marine environments, inorganic forms of arsenic are reduced and converted to less toxic organic forms by microorganisms, plants, and other marine organisms (Duker et al., 2005) and that measuring total arsenic in the blood does not allow us to speculate about how much of the arsenic present is potentially toxic to the shark. However, if only some of the arsenic measured in blood is of a toxic form, then it would be expected to have sublethal physiological impacts, such as increasing oxidative stress, immunotoxicity, or altering enzymatic function (Kavitha et al., 2010; Greani et al., 2017; Dringen et al., 2016). However, the high levels of arsenic found here were not correlated with any trace elements or to levels of liver enzymes such as AST and ALP.

Zinc and iron levels were positively correlated across individuals in this study. A positive relationship between zinc and iron has been found in other studies where it has been shown that zinc and iron interact extensively. In fact, there is significant interplay between the homeostatic, regulatory mechanisms at the cellular level for each micronutrient and one can influence the availability and function of the other in eukaryotic cells (Knez et al., 2015; Ehrensberger and Bird, 2011). Zinc was up-regulated in animals with high levels of arsenic and lead. Whether high levels of either zinc or iron are capable of off-setting potential ill effects associated with heavy metal concentrations in the blood remains an open question for further inquiry. This is of particular interest in the case of the very high levels of arsenic observed. Heavy metals can mimic and/or replace essential elements. For example, essential zinc can be replaced by cadmium and biologically important phosphates can be replaced by arsenates. If any of these mechanisms are present in white sharks, they were not supported by the results of this study where shark condition was not affected by high levels of heavy metals in the blood.

A positive relationship between arsenic and lead with zinc was observed in shark blood. This may reflect environmental conditions where the sharks live rather than the biological availability or synergism between them. Previous studies have shown that marine environments with sediments high in arsenic and lead tend to also be high in zinc levels (Luo et al., 2010; Marmolejo-Rodriguez et al., 2010). This suggests that blood levels in sharks may reflect environmental variation in these metals present in the local environment. In South Africa, a study conducted in 2011 found that metal levels were not significantly increased from studies done in 1985 and that they were less than those measured in other coastal areas (Sparks et al., 2017). However, neither arsenic nor mercury were measured.

Taken together, the results of this study show that sharks are capable of being exposed to high levels of arsenic and mercury and that their overall condition is not adversely affected. Questions arise about whether sharks may have additional mechanisms in place to bind and/or manage these metals in the blood so that they are less able to negatively impact shark well-being. The presence of metallothionein or other metal binding proteins has not been carefully examined in sharks and may contribute to the metabolism of heavy metals in the blood (Wang et al., 2014). Additionally, shark proteins, including immunological mediators and enzymes, are considered robust, resistant molecules that can persist and be functional in the urea-rich environment of shark tissues (Trischitta et al., 2012). It may be possible that this affords them some additional protection against oxidative stress and other harmful impacts of heavy metal exposure.

One of the limitations in this study was the lack of other hematological or health data that could have confirmed the health status in the individuals sampled. Reference intervals for total leukocyte counts and GLR values in normal, healthy white sharks are not available for this population, making it difficult to draw broad conclusions about the health of these sharks. Furthermore, blood smears were only available for a subset of sharks in the study. Additionally, for some metals like

arsenic, it may be possible to test specifically in the blood for various arsenic types, such as arsenobetaine, that could help us evaluate the level of toxicity the animals are likely experiencing. Another deficit here is that there was no measure of metals in the immediate area at time of collection. Although recent studies in the bays where sharks were sampled were done on heavy metal contamination, they were not concurrent with sampling in this study. Another limitation is that we did not have data on muscle or liver concentrations of heavy metals, which may show patterns that differ from the blood. It is also unclear precisely how long various metals remain in the blood and organotropism has not yet been clearly described for elasmobranchs. Therefore, to understand how these metals and elements are metabolized in sharks, it would be informative to conduct a study where blood levels of metals are compared to levels present in the tissues in the same animals. Potentially, it may be necessary to conduct a study on captive sharks that can be exposed to various metals over a time course and then blood sampled to (1) assess how these metals are metabolized in the blood and tissues and (2) measure the physiological response using various shark health parameters. Studies like this might help inform studies of wild populations and how they are being affected by exposure to heavy metals in either the aquatic environment or as part of their diet. It might also help confirm results of this study that suggest little consequences to white sharks with high levels of heavy metals in the blood in terms of either body size or body condition.

This is the first study to our knowledge where blood levels of heavy metals and trace elements have been measured in wild sharks. Here, we provide a set of reference intervals for these metals in this population and the results suggest that the levels of heavy metals present in blood are not related to body size and are not adversely affecting body condition in these animals. Interestingly, relationships between various heavy metals and protective anti-oxidant micronutrients like selenium and iron were not observed in this study. Taken together, this suggests that other aspects of shark physiology are providing sharks the ability to manage the levels of heavy metals in the blood and possibly mitigate their impacts on shark health and fitness. This provides an exciting area for future investigation.

Declaration of interests

None.

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