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Trace element accumulation in *Cassiopea* sp. (Scyphozoa) from urban marine environments in Australia

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Abstract

Jellyfish are robust, short-lived animals, tolerant to a wide range of environmental conditions and pollutants. The benthic jellyfish, *Cassiopea* sp. was collected from five locations along the north eastern coast of Australia and analysed for trace elements to determine if this species has potential as a marine biomonitor. Both the oral arm and bell tissues readily accumulated aluminium, arsenic, barium, cadmium, chromium, copper, iron, manganese and zinc above ambient seawater levels. In contrast, lithium appeared to be actively regulated within the tissues while calcium, magnesium and strontium reflected the ambient environment. The multi-element signatures showed spatial variation, reflecting the geographical separations between locations, with locations closer together showing more similar elemental patterns. The combination of bioaccumulative capacity, life history traits and

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biophysical aspects indicate that this species has high potential as a biomonitor in coastal marine systems.

Keywords: jellyfish, Cassiopea, trace metals, bio-uptake, Australia, biomonitor

1 Introduction

Many trace metals have important cellular and tissue functions within organisms. However, excessive concentrations of some elements can have a detrimental effect on the environment and its inhabitants (Sunda & Huntsman 1998). Heavy metals and organic compounds that have no known
biological function can also be taken up and concentrated in biological tissue (Wright 1977). Although a large body of research exists on the levels, fluxes and cycling of chemicals in marine environments (eg Furness & Rainbow 1990; Sadiq 1992), concentrations of contaminants in water do not necessarily describe their ecological significance to biota.

The use of organisms as biomonitors for trace metals and environmental contaminants is a wellestablished practice in both terrestrial and aquatic environments. The extent to which any particular organism can serve as a biomonitor depends on a number of factors including the system under investigation, life history traits of the organism, the specific contaminants being monitored and the extent of uptake and retention of those contaminants (Rainbow & Phillips 1993).

Gelatinous zooplankton (including jellyfish) can comprise a significant proportion of the total
biomass of pelagic marine assemblages (Shenker 1985; Romeo et al 1992). Jellyfishes can tolerate a wide range of environmental and water quality conditions (Lucas 2001; Purcell et al 2001). They are short-lived generally, capable of rapid population explosions, high growth rates and plasticity of resource allocation. As a consequence, they can have detrimental economic, health and aesthetic effects in coastal systems (Graham et al 2003; Uye & Shimauchi 2005), although they are valuable in some regions as a fisheries resource (Kingsford et al 2000; Pitt & Kingsford 2003).

Although data is limited on the capacity of jellyfish to take up metals, the available information suggests they are capable of accumulating metals and nutrients above ambient seawater concentrations and potentially transferring these further up the foodchain. (Heymans & Baird 2000; Kingsford et al 2000; Fukuda & Naganuma 2001; Fowler et al 2004; Hay 2006). Caurant et al (1999) linked elevated cadmium levels in tissues of certain turtle species with their jellyfish prey. Romeo et al (1992) also

identified that gelatinous zooplankton may have a role in bioconcentration of metals up the food chain.

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Although previously not considered as biomonitors, scyphozoan jellyfish possess many features considered important in biomonitors. Rainbow & Phillips (1983) identified a number of features that were essential for an organism to be considered a suitable biomonitor. These include the capacity to accumulate metals, sedentary behaviour, ease of identification, presence at locations of interest, and tolerance to physico-chemical changes within their environment (Rainbow & Phillips 1993).

The benthic scyphozoan jellyfish, *Cassiopea* spp. is euryhaline, tolerant of physio-chemical changes to its environment, is readily identifiable, can be present at high densities, has a wide distribution in tropical coastal environments and due to its benthic nature is relatively sedentary, and

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as such meets the biomonitoring criteria set out above. *Cassiopea* spp. are typically found in sheltered locations (eg man-made lagoons and lakes, inshore seagrass meadows, mangroves, protected reefs,

etc) and exhibit atypical behaviour of resting upside down on the substrate giving rise to their common name – 'Upside Down Jellyfish'. They possess an endosymbiotic relationship with *Symbodium* spp. and thus are capable of photosynthesis, which is associated with demonstrated efficiencies in uptake of inorganic nutrients and elements from the surrounding water column (Estes et al 2003; Fowler et al 2004).

The objective of this study was to compare trace element concentrations in bell and oral arm tissues of *Cassiopea* sp. collected from urban waterbodies across eastern and northern Australia. Both essential trace elements and priority pollutants were measured and compared to ambient seawater

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concentrations from the collection sites to determine if *Cassiopea* sp. were capable of accumulating trace elements. Trace element signatures in jellyfish and seawater were also compared among locations to determine the potential for jellyfish to provide time-integrated measures of local conditions.

2 Materials & Methods

Cassiopea sp. and water samples were collected from five locations adjacent to urban areas: Myora Drain A & B on the Gold Coast; Lake Magellan A & B on the Sunshine Coast in Queensland; and Lake Alexander in Darwin, Northern Territory, Australia. Locations A & B for Myora Drain & Lake Magellan were separated from each other by approximately 1 - 1.5 km. Five jellyfish and two water samples were collected at each location (Table 1).

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INSERT TABLE 1

2.1 Specimen Handling

Following collection, jellyfish were rinsed with local water to remove any visible sediment or other material adhering to the bell or oral arms. The oral arms were separated from the bell by hand and placed in either clean, acid washed plastic resealable bags or plastic vials. Tissue samples were frozen as soon as practicable and kept at -18° C until they were digested.

Replicate water samples (50 – 100 ml) were collected and filtered through a $0.45\mu m$ syringe filter and stored in acid washed vials. Water samples were acidified with Suprapur grade nitric acid (HNO₃) to 2% and stored at 4^o C until analysed.

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2.2 Tissue Processing & Analysis

Approximately 3 - 5 g tissue was accurately weighed and digested in five millilitres of concentrated (69%) Suprapur grade HNO₃. Samples were digested for approximately 2 hours, allowed to evaporate to approximately five millilitres. Three to five millilitres of AR grade hydrogen peroxide was added to remove any residual organic carbon. Following digestion, the tissue was made up to 25 millilitres with Milli-Q water. All equipment and containers were pre-cleaned and soaked in 10% HNO₃, triple

Samples were analysed using Varian 820-MS ICP-MS and Varian Liberty Series II AES. ICP-MS

rinsed in deionised water and air dried in a Class 100 laminar flow unit before use.

was used to determine aluminium $(0.5)^{**}$, arsenic (1), barium (0.1), copper (0.1), cadmium (0.05), 90 chromium (1), lithium (0.1), manganese (2), lead (0.05), strontium (0.5), silver (0.1g) and zinc (2), while ICP-AES was used to measure calcium (10), magnesium (10) and iron (10). Elements were selected based on either their importance as essential biological elements or their consideration as anthropogenic or priority pollutants. Due to issues with signal suppression, it was necessary to dilute seawater samples 1:10 (seawater:diluent) prior to analysis.

Subsets of samples were spiked with known concentrations of all elements for quality control purposes and to determine recoveries (90-117%). Indium, gallium and yttrium were used as internal standards to correct for potential instrument drift and matrix effects. Analytical data was checked to ensure signal strength exceeded three standard deviations for all analyses. Digestion blanks were included to ensure integrity of the digestion process. Digestion blanks had low levels of

contamination and tissue data was corrected for blank results before statistical analysis.

2.3 Statistical Procedures

Prior to statistical analysis of tissue data, seawater results that were below detection were recalculated to half the reported detection limit (Section 2.2). Elements below detection in seawater samples were included at half the reported detection limit in statistical analyses.

Analytical data was tested for homogeneity of variance (Bartlett's Test). All tissue data was log_{10} transformed to meet assumptions of normality. Univariate data was analysed using Statistica Version 8.1. Univariate data among locations were analysed using a one-way ANOVA. Comparisons between tissue type (bell, oral arm) were undertaken using a pair-wise two-tailed t-test. Correlations between tissue elements and tissue - water relationships were also performed using Statistica Version 8.1. Bioconcentration plots were prepared by calculating the bioconcentration factor for each metal using ratio of tissue metal concentration in tissue to the seawater metal concentration at each location (Sadig 1992). Principal Component Analyses (PCA) were performed using SYSTAT Version 10 (Crane Software) after log_{10} transformation to describe spatial variation in multi-element signatures.

3 Results

3.1

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Element Concentrations Between & Within Tissues

Trace element concentrations varied among locations and between tissues for most elements 120 (Table 2). Barium and zinc concentrations were significantly higher in oral arm tissues than bell tissue at all locations. Mean concentrations of manganese, arsenic, aluminium and cadmium were also

detection limits for elements (ug/L)

higher in oral arm tissue than bell tissue at all locations but the differences were not always significant (Table 2). Concentrations of copper, iron and strontium were higher in oral arm tissue at some locations and higher in bell tissue at others, although higher bell concentrations of elements were not significant. No significant differences were found between tissues for chromium, calcium, magnesium and lithium. There was no significant relationship between animal size and element concentration, thus size did not confound the spatial comparisons.

There were significant inter-element relationships in both bell and oral arm tissue, although these patterns were not always consistent between the tissues (Table 3). There was a very significant

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positive correlation between Ca-Mg (r = 0.96 bell; r = 0.95 oral arm) in both tissues. Ca-Cd (r = 0.73 bell; r = 0.52 arm) and As-Mn (r = 0.67 bell; r = 0.70 arm) were also positively correlated in both tissues. Other significant positive correlations in both tissues included As-Ba, Ba-Mn, Cr-Zn, Cd-Mg and Mn-Zn (Table 3). In contrast, Cr-Li was negatively correlated in both tissues (r = -0.54 bell; r = -0.52 arm).

For many other elements, the relationships were different for element pairs in the two tissues, with many being significantly correlated in one tissue but not the other (Table 3). Aluminium had significant correlations with a number of elements in the bell tissue but was only significantly correlated with zinc in oral arm tissue. Strontium also correlated positively with a number of other elements in the oral arm tissue but was only negatively correlated with iron in the bell tissue (Table 3). Zinc was also correlated with a number of elements in both tissues but the relationships were not

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Zinc was also correlated with a number of elements in both tissues but the relationships were no consistent between tissues (Table 3). Barium had a number of negative correlations with other elements although again, these patterns varied between the tissues.

INSERT TABLE 2

INSERT TABLE 3

3.2 Comparisons of Tissue and Ambient Seawater Concentration

Most elements were detected in higher concentrations in the jellyfish tissues by at least one order of magnitude compared to ambient seawater (Figure 1). Lithium was the only element found in *Cassiopea* sp. at concentrations below that of seawater. This was consistent across all locations and indicated that the animals were potentially regulating uptake and/or excretion of this element. The relative patterns of element accumulation were similar among locations although the degree of accumulation for some elements (particularly Al, Cd, Mn and Fe) was higher at Lake Alexander than the other locations (Figure 1). Tissue concentrations of calcium, magnesium and strontium were similar to ambient seawater concentrations at all locations. With the exception of magnesium in oral arm tissue and water (p < 0.05, n=5, r=0.94) there were no significant correlations between tissue concentrations of any given metal with the corresponding water concentrations, indicating a lack of regulation in elemental uptake.

Tissue levels of lead and silver were below detection limit in most animals and all waters.

160 Measurable levels of silver were only found in eight animals at tissue concentrations <2 ug/kg (wet weight) while lead was found in only seven animals with concentrations less than 10 ug/kg (wet weight).

INSERT FIGURE 1

3.3 Spatial Variation in Tissue Elements

There were significant differences in tissue concentrations among locations and between tissues for most elements (Tables 4 & 5). Lithium and chromium were the only elements that were not significantly different among locations for either tissue type. Copper exhibited significant differences among locations for bell tissue but no significant difference among locations for oral arm tissue, while strontium showed the opposite effect (Tables 4 & 5). Variation in barium concentrations was observed at large spatial scales in oral arm tissue but not at local scales (ie between locations A & B for Myora Drain and Lake Magellan). This relationship was not as robust in the bell tissue (Table 5). Bell tissue iron concentrations were significantly different among locations; however, post-hoc tests could not discriminate between them (Table 5).

Samples from Myora Drain A & B had similar elemental concentrations with no significant differences in bell tissue for any element (Table 5). Iron and arsenic were the only elements significantly different in oral arm tissue between the two locations. The similarity in concentration patterns between locations in close proximity to each other reflects a system with a short water

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residence time and regular tidal flushing.

In contrast, the patterns of element concentration for Lake Magellan A & B varied by element and tissue type. Tissues were significantly different in calcium and magnesium. Arsenic, cadmium, iron and strontium concentrations were significantly different in oral arm tissue, while bell tissue was significantly different for copper between the two locations (Tables 2 & 5). The variation in calcium and magnesium concentrations may reflect changes in salinity within the lake between collection dates. Differences in Ca and Mg were also measured in the water samples at these locations. Differences in the concentrations of other elements may be due to the long water residence time in the system (Table 1).

INSERT TABLE 4

Tissue results from Lake Alexander demonstrated no consistent relationship with any other location (Table 5). Significant differences could be identified between Lake Alexander and individual locations in both Myora Drain and Lake Magellan but there were no consistent patterns among the elements. Post-hoc tests separated Lake Alexander from at least one other location for all elements except barium and copper (in bell tissue), and manganese and strontium (in oral arm tissue). Overall, variability was least between locations in closest proximity (eg Myora Drain A & B), with greater variation among locations with the greatest geographical separation. The rate of flushing and length of water residence time is also likely to have influenced variation in tissue concentrations among

locations.

INSERT TABLE 5

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3.4 Patterns in Elemental Fingerprints

Differences in elemental fingerprints were found among locations in both bell and oral arm tissue. Multivariate analysis (PCA) of elements in bell tissue indicated variation among locations was greater for those locations with a wider geographical separation (Figure 2A). Some variation among replicate samples within locations was found in all locations. Total variance in the matrix was 70.4% explained by Factor 1 (41.7%) and Factor 2 (28.7%). Factor 1 was characterised by positive loadings for Mn, Al, As, Zn and Ba (0.872, 0.829, 0.797, 0.726 and 0.707, respectively) and negative loadings for Cu (-0.617). Factor 2 was characterised by positive loadings for Ca & Mg (0.987 & 0.986, respectively) and negative loadings for Ba and Zn (-0.417 & -0.373, respectively). The Myora Drain A & B

210 locations had the greatest similarity in elemental fingerprint. Patterns between Lake Magellan A & B were also very similar although there was greater spread within the Lake Magellan A samples. Lake Alexander was separate to all other locations (Figure 2A).

Multi-element patterns in oral arm tissue were not as discrete as bell tissue (Figure 2B). Overall the total variation in the matrix for oral arms was 65.7% with 35.1% explained by Factor 1 and 30.6% explained by Factor 2. Factor 1 was described by positive loadings for Zn, Cu, Cr and Mn (0.854, 0.833, 0.728 & 0.709, respectively) and a negative loading for Li (-0.562). Factor 2 loadings were positive for Mg & Ca (0.928 & 0.916 respectively) and negative for Fe and Ba (-0.789 & -0.681, respectively). Individual samples within locations displayed greater variability in signatures and there was greater overlap among locations. Although exhibiting greater individual variations, the Myora

220 Drain and Lake Magellan locations remained reasonably distinct from each other. Unlike the bell tissue, the oral arm elemental signature from Lake Alexander reflected an intermediate pattern between Myora Drain and Lake Magellan (Figure 2B). This suggests that different processes were influencing uptake and concentration of elements in oral arm compared with bell tissue.

INSERT FIGURE 2

I Discussion

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Trace Element Uptake and Accumulation

Studies have demonstrated that jellyfishes and other gelatinous plankton are capable of absorbing trace elements from the environment in measurable concentrations (Romeo et al 1992; Hanaoka et al 2001; Fowler et al 2004). However, data on bioaccumulation in scyphozoan and other jellyfishes is limited (Table 6).

Investigations into the extent of uptake and / or accumulation of elements from the surrounding water by jellyfish are even more limited. Cimino et al (1983) found tissue concentrations between 100

times (Al, As, Cd, Co, Hg, Mn, Pb, Sn & Zn) and 10 000 times (Fe) greater than typical seawater concentrations in tentacles of *Pelagia noctiluca*. The current study showed that *Cassiopea* sp. accumulated iron and aluminium at concentrations greater than 1000 times ambient seawater concentrations. Other metals including copper, zinc, cadmium, manganese and arsenic were accumulated greater than 10 fold above ambient seawater concentrations (Figure 1), while lithium seemed to be actively regulated in the tissues.

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Tissue copper concentrations in *Cassiopea* sp. were comparable to the literature for other jellyfish (Table 6), although accumulation in this study was lower than levels reported for other marine organisms (eg Rainbow & Phillips 1993). This may reflect the high water content in jellyfish and the different metabolic requirements of other phyla (eg molluscs, crustaceans).

Zinc is a key component of the metalloenzyme carbonic anhydrase. Carbonic anhydrase is particularly abundant in organisms with a symbiotic association with photosynthetic algae (Furla et al 2000) and the presence of zooxanthellae may therefore be an important contributor to tissue concentrations of zinc within symbiotic jellyfish. Concentrations of zinc measured in tissues from this study were higher than reported literature values for non-symbiotic jellyfish species for both oral arm

and bell tissues (Table 6). This is consistent with the observation that zooxanthellae potentially influence zinc concentrations in the jellyfish. Zinc concentrations were below detection in waters at all locations indicating that *Cassiopea* sp. must either be efficient at obtaining zinc from food and water, or recycling zinc.

INSERT TABLE 6

The uptake and accumulation of non-essential elements in this study was variable. Chromium was present in low concentrations in the water but present at higher levels in the tissues, while arsenic and cadmium, although below detection in the seawater, were found in measurable concentrations in all animals (Table 2). This suggests that tissue concentrations of these metals may reflect a time-integrated measure of metal concentrations in the water (either from pulse or press events).

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Cadmium generally correlates with phosphate in seawater (Lares et al 2002) and uptake of this element may be linked with nutrient levels at the sampling locations. Phosphate was not measured in this study but there were no obvious signs of eutrophication (eg colour, algal blooms etc) at any location during sampling that would suggest pulses or presses of nutrients including phosphates.

Tissue concentrations of arsenic vary among scyphozoan jellyfish and differences were found between tissues in this study and others (Table 6). Investigations of arsenic in *Mastigias papua* (Hanaoka et al 2001) found that levels in oral arm tissue containing zooxanthellae were higher than bell tissue but lower than levels in gastric filaments and gonads. Hanaoka et al (2001) also found similar differences between umbrella (bell) tissues and other tissues in other asymbiotic jellyfish species. The arsenic results for *Cassiopea* sp. (this study) are within the range of literature data, with bell concentrations generally higher than other studies. In contrast, arsenic concentrations in oral arm

270 bell concentrations generally higher than other studies. In contrast, arsenic concentrations in oral ar tissue from this study tended to be lower than for other studies that focussed on zooxanthellae

containing tissues (Table 6).

Lithium was the only element present at lower concentrations in tissue samples than seawater samples. Kszos et al (2003) showed lithium was toxic to freshwater organisms in the absence of sodium. However, in marine environments, sodium levels are sufficiently elevated that lithium is not generally a problem (Sadiq 1992). It is interesting that lithium appears to be actively regulated within the body in this study, although the mechanism/s and requirements for regulation are not known. It is possible that this result reflects a lag between lithium uptake and change in water concentration but it is unlikely that pulses of lithium occurred at all five locations immediately prior to sampling.

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The correlations in metal accumulation in the tissues provided some significant relationships. Positive correlations such as calcium and magnesium in tissue were not unexpected given the osmoconforming nature of the animal (Arai 1997). Other relationships however, were more difficult to explain. It is recognised that metal transport sites, whilst having a high affinity for metals, may not have a high selectivity and can bind with non-essential metals of similar geometry (Worms et al 2006).

Given the variability in element correlation between the tissues, it is feasible that elemental absorption and sequestration may occur in at different locations within the jellyfish. The ability of the zooxanthellae to migrate between the tissues within amoebocytes (Estes et al 2003) may allow ready migration of elements between the tissues as well. In addition, it is possible that there may be deliberate absorption of some elements as a detoxification mechanism (Wang & Rainbow 2005). Food as a source of metal uptake may be important in explaining some of these patterns, and has been identified by many authors as a potentially significant source (eg Depledge & Rainbow 1990).

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identified by many authors as a potentially significant source (eg Depledge & Rainbow 1990). Importantly, the presence of these correlations indicates that uptake and sequestration of elements may not be independent from each other.

Relationships between tissue and water for the different elements for the most part were not significant. However, a grab water sample may not reflect the time integrated exposure of the jellyfish to elements. It is likely that for many of the elements, tissue concentrations were the result of long term uptake and/or sequestration of elements while the water measurements reflected the current water quality. Again, the contribution of diet in tissue element concentrations could obscure any relationships between tissue and water concentrations. There was a positive correlation between tissue magnesium and water magnesium in the oral arm (p < 0.05, n = 5, r = 0.94) and is likely to be due to the osmoconforming nature of jellyfish (Arai 1997).

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Although data on the long term accumulation of trace metals in *Cassiopea* sp. are limited, Fowler et al (2004) used radiotracers on asymbiotic *Aurelia aurita* and symbiotic *Cassiopea andromeda* to assess efficiencies of uptake and half-life retention of Zn, Co, Ag and Cd. *C. andromeda* was found to be very efficient at both accumulating and retaining radiotracers from seawater (Fowler et al 2004). This capability occurred under both light and dark conditions. Results for *A. aurita* were lower than for *C. andromeda* but still measurable with shorter retention times for *A. aurita*, indicating that zooxanthellae may be important in trace element retention (Fowler et al 2004). The reported

biological half life of zinc in Cassiopea sp. (Fowler et al 2004) was comparable to the 66 days

310 reported for the sea snail *Rapana venosa* (Onat & Topcuoğlu 1999) and longer than the biological half lives of 16.7-30.1 and 25.1 days reported for *Crassostrea gigas* and *Mytilus smarangdium* respectively (Han et al 1993). Fowler et al (2004) is one of the few studies to date that quantifies the efficiencies of uptake, and demonstrates that scyphozoan jellyfish have the potential to accumulate and retain metals over time with similar efficiencies to other invertebrate species.

Differences found in elemental signatures between oral arms and bell tissues in this study could be due to several factors. Different tissue functions and densities of zooxanthellae will certainly contribute to the variation (Estes et al 2003). The variation in uptake and half-life efficiencies in *A. aurita* and *C. andromeda* (Fowler et al 2004) suggest that zooxanthellae may explain differences in accumulation and sequestration of trace elements. Jellyfish may also actively transport elements

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within their bodies leading to differences between uptake and storage tissues. Elemental concentrations in prey and undigested food could also contribute to variation in tissue elemental loads, as could other differences in uptake mechanisms (eg direct absorption from the environment).

4.2 Spatial Variation in Elemental Signatures

Multivariate analyses have been used widely in ecological studies and more recently it has also been used to try and characterise processes and trace anthropogenic pollutants to their sources (eg Del Valls et al 1998; Zitko 1989). The locations in this study reflected a variety of urban marine environments with different catchment characteristics, adjacent population densities, and water turnover rates. Spatial variation was found among locations, separated by kilometres to hundreds of kilometres and was consistent for both tissue types. Accumulation patterns in bell tissue were more discrete than oral arm tissue (Figure 2). Notwithstanding this, it was still possible to separate locations based on the oral arm signature. Variation was, as predicted, smallest between locations separated by hundreds of metres (eg Myora Drain A & B; Lake Magellan A & B). In addition, the patterns of accumulation tended to reflect differences in the extent of the adjacent urban areas. The sedentary nature of *Cassiopea* sp. is likely to be fundamental to this capability, as individual animals do not readily move large distances within their environment.

Biomonitoring Potential of <u>Cassiopea</u> sp.

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4.3

Rainbow & Phillips (1993) identified a number of key criteria that organisms need to have in order to be considered an effective biomonitor. *Cassiopea* sp. has a wide distribution in tropical environments, in particular coastal marine environments. It is largely sedentary due to its atypical behaviour of resting upside down on the substrate and which also makes its identification relatively simple. Due to its presence in coastal marine systems, *Cassiopea* sp. is considered euryhaline and thus tolerant physico-chemical changes in its environment. These attributes meet many of the criteria for biomonitors (Rainbow & Phillips 1993). A primary attribute of biomonitors is the ability to

accumulate elements of concern. This study and the few other studies to date (Table 6) indicate that *Cassiopea* sp. (among others) is capable of accumulating elements above ambient seawater concentration (Figure 1). The extent of this capability requires further exploration to determine limitations to uptake and also retention times within tissues.

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5 Conclusions

This study has demonstrated that *Cassiopea* sp. is capable of accumulating elements above ambient seawater concentrations. It is also possible to discriminate different geographical populations based on chemical signatures in their tissues. Whilst their persistence in polluted systems has been documented, historically jellyfish have not been considered useful biomonitors. We suggest it would be useful to consider them as a future tool in the biomonitoring toolbox, as they meet many of the identified criteria.

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FIGURE CAPTIONS

Figure 1: Elemental bioconcentration in *Cassiopea* sp. tissues as a function of ambient seawater concentration measured at each location. Equation to calculate bioconcentration = (tissue concentration/water concentration). A= Bell tissue; B= Oral arm tissue. Bioconcentration >1 is considered accumulation; bioconcentration = 1 reflects no accumulation; bioconcentration <1 reflects regulation / excretion

Figure 2: Results of multivariate PCA for multi-element signatures among locations in *Cassiopea* sp. tissues. (A) Bell tissue percent of variation explained by Factor 1 = 41.7%; Factor 2 = 28.7%. (B) Oral Arm Tissue percent of variation explained by Factor 1 = 35.1%; Factor 2 = 30.6%. Data $\log_{10}+1$ transformed prior to analysis to reduce contribution from elements with highest concentrations

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Tables

Table 1: Sampling locations and site descriptions. For Lake Alexander collection (A) = Jellyfish, (W) = Water

Sampling Legation	Site	Latitude /	Site Description	Collection Date	Estimated
Location	Name &	Longitude			Water
Myora Drain, Coombabah Gold Coast	Reference Myora	S 27 ⁰ 54'05.2" E 153 ⁰ 22'38.0"	Urban drain adjacent Coombabah Wetland, 1.0 km east of outlet to Coombabah Creek	1 June 2007	Turnover <1 day
Myora Drain, Coombabah Gold Coast	Drain A Myora	S 27 ⁰ 53'52.9" E 153 ⁰ 22'03.6"	Urban drain adjacent Coombabah Wetland, 0.2km west of outlet to Coombabah Creek	20 July 2007	< 1 day
Lake Magellan, Pelican Waters, Sunshine Coast	Drain B Lake Magellan A	S 26 ⁰ 49' 27.6" E 153 ⁰ 06' 37.4"	Urban lake adjacent to piped outlet to canal. Located below small urban park	3 June 2007	21 days
Lake Magellan, Pelican Waters, Sunshine Coast Lake	Lake Magellan B	S 26 ⁰ 49' 43.5" E 153 ⁰ 06' 44.6"	Urban lake adjacent to piped inlet from canal. Located below small urban park	(A) 18-20	21 days
Alexander, Darwin	Lake Alexander	S 12 ⁰ 24' 47.8" E 130 ⁰ 49'55.0"	Urban lake surrounded by Parkland mangroves, coastal beach and urban residential	October 2006 (W) 17 Aug 2007	< 14 days

______regroves, coasts residential

Table 2: Trace elements (mean ± 1 SE) in *Cassiopea* sp. tissues (wet weight) with 2-tailed pair-wise t-test between tissues for each location. ns p>0.05; *p<0.05; **p<0.01; ***p<0.005. df=4. All concentration expressed as ug/kg except Ca, Mg and Fe (mg/kg) wet weight

Element	Tissue	GC	р	EC	р	SC	р	JB	р	LA	р
Al	Bell Oral Arm	358 <u>+</u> 81 1323 <u>+</u> 484	ns	251 <u>+</u> 73 628 <u>+</u> 96	***	1727 <u>+</u> 680 5418 <u>+</u> 2874	ns	481 <u>+</u> 117 1142 <u>+</u> 284	ns	6975 <u>+</u> 2577 6681 <u>+</u> 2128	ns
As	Bell Oral Arm	45.9 <u>+</u> 6.6 69.2 <u>+</u> 10.6	*	84.1 <u>+</u> 9.0 153 <u>+</u> 25	*	94.0 <u>+</u> 21.1 135 <u>+</u> 15	ns	170 <u>+</u> 15 378 <u>+</u> 38	**	143 <u>+</u> 25 156 <u>+</u> 25	ns ns
Ba	Bell Oral Arm	14.4 <u>+</u> 0.7 19.4 <u>+</u> 1.7	*	12.9 <u>+</u> 0.6 21.3 <u>+</u> 1.7	**	27.0 <u>+</u> 5.7 53.3 <u>+</u> 7.3	***	22.8 <u>+</u> 1.7 47.6 <u>+</u> 4.8	*	20.2 <u>+</u> 2.1 30.8 <u>+</u> 4.4	*
Ca	Bell Oral Arm	372 <u>+</u> 29 429 <u>+</u> 36	ns	427 <u>+</u> 18 446 <u>+</u> 20	ns	258 <u>+</u> 11 272 <u>+</u> 17	ns	342 <u>+</u> 7 412 <u>+</u> 28	ns	466 <u>+</u> 19 430 <u>+</u> 20	ns
Cd	Bell Oral Arm	7.58 <u>+</u> 0.97 8.29 <u>+</u> 1.48	ns	7.11 <u>+</u> 0.69 12.4 <u>+</u> 2.4	ns	3.31 <u>+</u> 1.41 5.42 <u>+</u> 1.18	ns	4.86 <u>+</u> 0.56 13.1 <u>+</u> 1.1	***	15.1 <u>+</u> 2.1 40.6 <u>+</u> 8.0	*
Cr	Bell Oral Arm	108 <u>+</u> 30 104 <u>+</u> 35	ns	110 <u>+</u> 47 117 <u>+</u> 51	ns	141 <u>+</u> 84 141 <u>+</u> 74	ns	121 <u>+</u> 52 162 <u>+</u> 54	***	88.8 <u>+</u> 39.5 116 <u>+</u> 50	ns
Cu	Bell Oral Arm	124 <u>+</u> 9 119 <u>+</u> 14	ns	79.4 <u>+</u> 13.1 134 <u>+</u> 29	ns	36.8 <u>+</u> 7.6 142 <u>+</u> 26	**	79.8 <u>+</u> 10.9 196 <u>+</u> 16	***	64.2 <u>+</u> 16.8 139 <u>+</u> 27	*
Fe	Bell Oral Arm	1.79 <u>+</u> 0.65 3.46 <u>+</u> 0.46	*	4.38 <u>+</u> 2.21 12.9 <u>+</u> 2.7	*	8.73 <u>+</u> 2.69 38.8 <u>+</u> 3.6	***	1.52 <u>+</u> 0.36 13.4 <u>+</u> 1.8	***	19.2 <u>+</u> 7.8 13.0 <u>+</u> 3.7	ns
Li	Bell Oral Arm	120 <u>+</u> 32 137 <u>+</u> 55	ns	94.1 <u>+</u> 39.3 124 <u>+</u> 41	ns	52.6 <u>+</u> 17.9 52.7 <u>+</u> 19.3	ns	71.1 <u>+</u> 30.2 70.6 <u>+</u> 31.9	ns	154 <u>+</u> 33 127 <u>+</u> 31	ns
Mg	Bell Oral Arm	1050 <u>+</u> 79 1251 <u>+</u> 115	ns	1221 <u>+</u> 69 1257 <u>+</u> 29	ns	744 <u>+</u> 28 734 <u>+</u> 44	ns	979 <u>+</u> 13 1034 <u>+</u> 75	ns	1297 <u>+</u> 55 1251 <u>+</u> 55	ns
Mn	Bell Oral Arm	123 <u>+</u> 15 354 <u>+</u> 42	**	248 <u>+</u> 25 687 <u>+</u> 80	***	402 <u>+</u> 159 644 <u>+</u> 166	**	338 <u>+</u> 48 1077 <u>+</u> 105	***	590 <u>+</u> 200 669 <u>+</u> 246	ns
Sr	Bell Oral Arm	7340 <u>+</u> 699 9184 <u>+</u> 560	ns	8022 <u>+</u> 707 10343 <u>+</u> 799	**	5812 <u>+</u> 862 6970 <u>+</u> 828	***	7228 <u>+</u> 518 10666 <u>+</u> 874	*	8358 <u>+</u> 647 7634 <u>+</u> 599	ns
Zn	Bell Oral Arm	1245 <u>+</u> 178 1897 <u>+</u> 170	***	1040 <u>+</u> 121 2431 <u>+</u> 575	ns	2633 <u>+</u> 441 4835 <u>+</u> 836	*	2959 <u>+</u> 421 7559 <u>+</u> 828	***	2085 <u>+</u> 275 4999 <u>+</u> 833	*
	6										
C)										
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Table 3: Correlations among trace elements in bell and oral arm tissue of Cassiopea sp. n=25 for both tissues

Bell	Tissue											
	Al	As	Ba	Ca	Cd	Cr	Cu	Fe	Li	Mg	Mn	Sr
As	0.475^{*}											
Ba	0.488^*	0.534^{**}										
Ca	0.101	0.127	-0.396									
Cd	0.294	0.134	-0.077	0.729^{***}								
Cr	-0.050	0.109	0.131	0.233	0.248							
Cu	-0.543**	-0.324	-0.494*	0.440^{*}	0.300	0.362						
Fe	0.671***	0.271	0.161	-0.090	0.051	-0.199	-0.580***					
Li	0.324	0.061	0.023	0.260	0.231	-0.542**	-0.096	0.317				
Mg	0.102	0.143	-0.411*	0.970^{***}	0.723***	0.191	0.430^{*}	-0.094	0.275			
Mn	0.589***	0.674^{***}	0.543**	0.189	0.252	0.433*	-0.452*	0.346	-0.208	0.120		
Sr	-0.336	-0.139	0.179	-0.187	-0.293	0.044	0.033	-0.454*	-0.361	-0.254	0.063	
Zn	0.002	0.131	-0.010	0.717***	0.690***	0.606***	0.318	-0.305	-0.116	0.663***	0.464^*	0.095
								- (*		
Ora	Arm Tis	ssue										
	Al	As	Ba	Ca	Cd	Cr	Cu	Fe	Li	Mg	Mn	Sr
As	-0.100											
Ba	0.322	0.526**										
Ca	-0.132	0.168	-0.406*				V					
Cd	0.324	0.202	-0.134	0.525**								
Cr	0.267	0.058	-0.056	0.278	0.270							
Cu	0.124	0.326	0.216	0.100	0.317	0.811***						
Fe	0.279	0.313	0.592***	-0.596***	-0.220	-0.071	0.084					
Li	-0.175	-0.024	-0.212	0.222	0.081	-0.552***	-0.533**	-0.176				
Mg	-0.090	0.005	-0.539**	0.948***	0.494^{*}	0.108	-0.123	-0.629***	0.370			
Mn	0.139	0.702***	0.477*	0.202	0.079	0.238	0.292	0.273	-0.266	0.071		
Sr	-0.200	0.249	-0.101	0.714***	0.157	0.539**	0.423*	0.333	-0.192	0.566^{**}	0.497^{*}	
Zn	0.454^{*}	0.593***	0.682***	-0.128	0.340	0.455*	0.695***	0.431*	-0.464*	-0.323	0.542**	0.140

* significant at p < 0.05, ** significant at p <0.01, ** * significant at p < 0.005

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Table 4: One-way ANOVA trace element concentrations in water, bell and oral arm tissue. Data log10 transformed. n.d. = not determined (results below reported detection limit). ns p>0.05; *p<0.05; **p<0.01; ***p<0.005. df (Water) = 4,9; df (Bell & Oral Arms) = 4,20.

Element	Water			Bell			Oral Arm					
	MS	Residual	р	MS	Residual	р	MS	Residual	р			
Ag	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d			
Al	0.426	0.013	***	1.580	0.086	***	0.766	0.110	***			
As	n.d	n.d	n.d	0.240	0.024	***	0.352	0.021	***			
Ba	0.040	0.000	***	0.072	0.008	***	0.179	0.010	***			
Ca	0.011	0.000	***	0.049	0.002	***	0.0399	0.004	***			
Cd	n.d	n.d	n.d	0.266	0.022	***	0.436	0.020	***			
Cr	0.003	0.006	ns	0.078	1.10	ns	0.281	0.942	ns			
Cu	0.035	0.007	ns	0.208	0.028	***	0.040	0.030	ns			
Fe	0.300	0.000	***	0.510	0.128	*	0.570	0.029	***			
Li	0.019	0.001	***	0.354	0.436	ns	0.292	0.440	ns			
Mg	0.007	0.000	***	0.044	0.002	***	0.051	0.004	***			
Mn	0.145	0.000	***	0.256	0.047	***	0.153	0.035	*			
Pb	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d	n.d			
Sr	0.010	0.000	***	0.022	0.009	ns	0.034	0.007	**			
Zn	n.d	n.d	n.d	0.197	0.019	***	0.306	0.023	***			

nd nd 0.197

Element	Tissue	GC	EC	SC	JB	LA
Al	Bell	а	а	b, c	a, b	с
	Oral Arm	a, b	а	b, c	a, b	с
As	Bell	а	a, b	a, b	b	b
	Oral Arm	а	b	b	с	b
Ba	Bell	a, d	a, d	b, c, d	c, d	d
	Oral Arm	a, c	a, c	b	b, c	с
Ca	Bell	a, c	a, d	b	с	d
	Oral Arm	а	а	b	а	a
Cd	Bell	а	а	b, c	a, c	a
	Oral Arm	a, b	а	b	a	С
Cr	Bell	а	а	а	а	a
-	Oral Arm	a	a	a	a	a
Cu	Bell	а	а	b	a	a b
cu	Oral Arm	a	a	a	a	a
Fe	Bell	а	а	а	a	а
	Oral Arm	a	b	c	b	b
Ci	Bell	а	а	a	a	а
	Oral Arm	a	a	a	a	a
Mσ	Bell	acd	acd	h	c	d
	Oral Arm	a, e, u a	a	b	a	a
Mn	Bell	а	a h	ab	h	b
	Oral Arm	a	a, b	a, b	b	a, b
Sr	Bell	а		я	а	a
51	Oral Arm	a, b, c	a, b	a, c	a, b	a, b, c
Zn	Bell	a h	h	C	C	3.0
	Dell	<i>a</i> , 0	• 0	1.	1	a, c

Table 5: Post –hoc test (Bonferroni p<0.05) from one-way ANOVA for trace element concentrations among locations. Locations sharing the same letters are not significantly different from each other.

Table 6: Trace element concentration in tissues of scyphozoan jellyfish from the literature. Results are mean for tissue type. # indicates concentration as ug/g dry weight, all others ug/g wet weight. Results below detection <d.l. Results from current study (both wet and dry) are the means from all locations.

Species	Location Collected	Tissue Type	Ag	Al	As	Ba	Ca	Cd	Cr	Cu	Fe	Li	Mg	Mn	Pb	Sr	Zn	Source
Pelagia noctiluca	Straits of Messina, Sicily, Italy	Tentacles		0.71	1.23		119	0.055		0.19	33		11.7	1.15	0.33		3.01	Cimino et al 1983
Pelagia noctiluca	Bay of Villefranche- sur-Mer, Cote d'Azur, France	Whole animal						0.4#		2.0#				2	0.6#		46#	Romeo et al 1987
Auralia sp.	Coastal waters Karachi, Pakistan	Whole animal					18800#	0.04#		16.1#	178.8#		75700#	0.04#			19.9#	Siddiqui et al 1988
Unidentified	Pertuis charentais, coastal waters, France	Unknown						0.27#			6	0						Caurant et al 1999
Chrysaora melanaster	Coastal waters Yoshimi, Japan	Umbrella Other			0.074 0.428													Hanaoka et al 2001
Pelagia noctiluca	Coastal waters Yoshimi, Japan	Whole animal			0.197													Hanaoka et al 2001
Mastigias papua	Coastal waters Yoshimi, Japan	Umbrella Oral Arm Gastral Filament /Gonad Zooxanthellae Tissue			0.039 0.073 0.578 0.308			5										Hanaoka et al 2001
Stromolophus nomurai	Coastal waters Yoshimi, Japan	Umbrella Oral Arm Tentacle			0.183 0.111 0.664													Hanaoka et al 2001
<i>Cassiopea</i> sp.	Gold & Sunshine Coasts, Queensland;	Bell	<d.l.< td=""><td>1.959 39.5[#]</td><td>0.107 2.16[#]</td><td>0.019 0.383[#]</td><td>373 7520[#]</td><td>0.008 0.161[#]</td><td>0.114 2.30[#]</td><td>$0.077 \\ 1.41^{\#}$</td><td>7.12 144[#]</td><td>0.098 1.98[#]</td><td>1058 21300[#]</td><td>0.340 6.85[#]</td><td><d.l.< td=""><td>7.352 148[#]</td><td>1.993 40.2[#]</td><td>Current Study</td></d.l.<></td></d.l.<>	1.959 39.5 [#]	0.107 2.16 [#]	0.019 0.383 [#]	373 7520 [#]	0.008 0.161 [#]	0.114 2.30 [#]	$0.077 \\ 1.41^{\#}$	7.12 144 [#]	0.098 1.98 [#]	1058 21300 [#]	0.340 6.85 [#]	<d.l.< td=""><td>7.352 148[#]</td><td>1.993 40.2[#]</td><td>Current Study</td></d.l.<>	7.352 148 [#]	1.993 40.2 [#]	Current Study
	Territory, Australia	Oral Arm	<d.1< td=""><td>3.038 54.3[#]</td><td>0.178 3.18[#]</td><td>0.034 0.607[#]</td><td>398 7110[#]</td><td>0.016 0.286[#]</td><td>0.128 2.29[#]</td><td>0.146 2.61[#]</td><td>16.33 292[#]</td><td>0.102 1.82[#]</td><td>1106 19750[#]</td><td>0.686 12.3[#]</td><td><d.l.< td=""><td>8.959 160[#]</td><td>4.344 77.6[#]</td><td>Current Study</td></d.l.<></td></d.1<>	3.038 54.3 [#]	0.178 3.18 [#]	0.034 0.607 [#]	398 7110 [#]	0.016 0.286 [#]	0.128 2.29 [#]	0.146 2.61 [#]	16.33 292 [#]	0.102 1.82 [#]	1106 19750 [#]	0.686 12.3 [#]	<d.l.< td=""><td>8.959 160[#]</td><td>4.344 77.6[#]</td><td>Current Study</td></d.l.<>	8.959 160 [#]	4.344 77.6 [#]	Current Study