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# Trace metal content in sediment cores and seagrass biomass from a tropical southwest Pacific Island



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#### ABSTRACT

A unique feature of seagrass among other ecosystem services is to have high phytoremediation potential that is a cost-effective plant-based approach and environmentally friendly solution for metal contamination in coastal areas. The goal of this study was to assess the phytoremediation prospective of seagrass for Cu, Fe, Mn and Zn in Fiji Islands. Heavy metal content was measured in sediments and tissues of the seagrasses *Halophila ovalis*, *Halodule pinifolia* and *Halodule uninervis* to test for local-scale differences. The local study shows that metal concentration in sediment and seagrass tissue was significantly variable, regardless of species and sediment type. Sedimentary concentration of Cu, Fe, Mn and Zn obtained in the present study seemed to be lower than that of previous studies. The results support that *H. ovalis* is a good bioindicator species since it accumulated up to 5-fold more of these metals compared to the *Halodule* species.

## 1. Introduction

Seagrasses play a critical role in the ecology of shallow-marine ecosystems where they stabilize sediment, have an important role in global carbon and nutrient cycling, and may function as a long-term biogeochemical sink for trace metals (Duarte, 1999; Serrano et al., 2016). Their life history characteristics, together with their above ground and below ground biomass support their use as biomonitoring species. As seagrass meadows are exposed to stronger anthropogenic pressures than many other marine communities (Ambo-Rappe et al., 2011), trace element levels in seagrass beds can be a useful indicator for harmful pollutants in such ecosystems. Seagrasses are a primary food for many animals (e.g., green turtles and rabbit fish) and metals sequestered by them may be transferred to higher trophic level consumers (Amado Filho et al., 2004).

Previous research has established that marine angiosperms generally have a high trace metal bioaccumulation capacity because of direct interaction from the water column and sediments in the marine environment via both leaves and root-rhizomes that are sites for ionic uptake

(Ambo-Rappe et al., 2011; Bonanno and Di Martino, 2017). A range of pollutants including heavy metals and pesticides (Bonanno et al., 2020) may significantly contaminate inshore seagrass meadows. This is generally a consequence of environmental pollution in coastal areas, which are most productive marine zones and important sinks of anthropogenic contaminants (Bonanno and Orlando-Bonaca, 2018). Thus, seagrasses are often considered as sentinel species because any change in their distribution (e.g. a loss of covered area) implies an environmental change (Govers et al., 2014; Bonanno and Di Martino, 2017).

Unlike pesticides and acid rain, trace metals exist naturally via weathering and erosion of rocks and soils in marine environments; these include copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn), which are essential micronutrients and critical to support plant metabolism (Boutabia-Trea et al., 2017). The same elements may have toxic effects at high concentrations (Bonanno and Di Martino, 2017). Other, nonessential elements are toxic to organisms even at low levels (e.g., Al, As, Cd, Cr, Hg and Pb) are highly toxic (Bonanno et al., 2020). Trace metals such as Cu and Zn are primarily mobilized through human

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activity such as urban and municipal runoff, industrial effluents, atmospheric deposition, and agricultural activity, whereas other metals such as Fe and Mn have a lithogenic origin (Serrano et al., 2011). Trace metal research with seagrasses has acknowledged their role as effective biomonitors because their tissue metal content often reflects bio-available water or sediment contents (Sanchiz et al., 2001; Ralph et al., 2006). From a management perspective, it is important to know which species are able to accumulate or concentrate specific metals, because different populations of the same species within small geographical distributions, can have widely different tolerances to metal impacts (Ralph et al., 2006). Toxic levels of metals inhibit metabolic activity and interfere with vital pathways, such as photosynthesis (Ralph and Burchett, 1998). Furthermore, plants that survive, do so with decreased growth and slower development (Clijsters and Van-Assche, 1985).

Trace metal contamination in estuarine seagrass meadows has been identified across the world with most studies from the Mediterranean (Bonanno et al., 2020; Morroni et al. 2020), Caribbean (Govers et al., 2014) and Indian subcontinent (Immaculate et al., 2018) and Australia (Prange and Dennison, 2000). However, similar research in the south Pacific region is lacking. In the Fiji Islands, seagrass beds have been identified as an important marine ecosystem (McKenzie and Yoshida, 2020). However, the coastal environments of Fiji are also facing problem of heavy metal pollution (Maata and Singh, 2008). To our knowledge, there is no existing information on the trace metal contents of intertidal seagrasses in Fiji despite a number of studies that have assessed environmental levels of a number of hazardous metals in coastal sediments, seawater and shellfish (e.g., Gangaiya et al., 2001; Morrison et al., 2001; Maata and Singh, 2008; Arikibe and Prasad, 2020). Similarly, Chand et al. (2010) reported levels of metalloid arsenic in surficial sediments to be 334 mg/kg (dry weight) which is almost 30 times greater than the U. S. EPA (2004) cleanup guideline level.

More recently, Pratap et al. (2020) reported that the spatial distribution of metals (Cd, Cr, Cu, Fe, Ni, Pb and Zn) in surficial sediments in Laucala Bay, near Suva, was affected by hydrodynamic regimes. This study confirmed that apart from Cd and Pb, other metals had a medium risk of toxicity, but had a high probability of accumulation and increasing concentration in the near future. Generally, all these studies perceived metal input into Suva coastal area from different sources such as land-based industrial activities like metal fabrication and construcpaint-manufacturing, petroleum storage and garment manufacturing, food processing, sewage treatment plants, as well as direct discharge of domestic and industrial wastes. Overall, it can be noted that factors such as increased economic development, rise in population and urbanization are the major driving forces behind the various anthropogenic sources of heavy metals in the Fijian coastal environment (Pratap et al., 2020). In the Fiji Islands, five species of seagrass from two families (Hydrocharitaceae and Cymodoceaceae) have been reported (Singh et al., 2019).

The few studies on seagrasses in Fiji for example, have focused on morphology, taxonomy and biological/faunal interactions as reported by Singh et al. (2019). We chose to focus on Halodule uninervis, Halodule pinifolia and Halophila ovalis as they are widely distributed throughout the main island coastline (Singh et al., 2019). Brief descriptions of the morphological characteristics of the Halodule plants are identified based on shape of the leaf tip (trident or rounded) and width of the leaves which is either narrow or wide (Den Hartog, 1970). H. pinifolia is often found in the high intertidal to upper subtidal areas, forming homogeneous patches or intermixes with H. uninervis and H. ovalis (McKenzie and Yoshida, 2020). H. uninervis is found from intertidal to 6 m depth, forming dense or patchy beds as well as intermixes with other species including H. pinifolia, Halophila spp. and Syringodium isoetifolium (McKenzie and Yoshida, 2020). Halophila plants are minute and fragile, without stripe-like long leaf blades, but rather oval shaped leaves in pairs and it is the most diverse seagrass group (Kuo, 2020). H. ovalis is found in the intertidal to 10-12 m depth, forming dense or patchy beds (McKenzie and Yoshida, 2020). Moreover, species of both the genus

Halodule and Halophila grow in shallow intertidal to subtidal waters directly adjacent to coastal areas in sand/mud banks.

In Fiji comparative investigation, using sediment and seagrass has yet not been examined as a biomonitoring tool for coastal trace metal contamination and deserve investigation to provide reliable data on marine environmental health. As heavy metal contamination is a great concern at the global, regional and local level because they are not able to be degraded and the toxic effects can be long lasting (Gangaiya et al., 2001). While seagrasses are known as good bioindicators as, they are widespread and sensitive to environmental changes and are able to integrate ecological conditions and processes over various timescales from weeks to years (Govers et al., 2014).

Therefore, we studied potential trace metal pollution in seagrasses on a local scale to test whether it was possible to detect local-scale differences in trace metal concentrations in both sediments and seagrass plants. Copper, iron, manganese and zinc were selected for this investigation because these metals (Cu, Fe, Mn and Zn) have been recognized as historical contaminants of Laucala Bay (Morrison et al., 2001). For some of the other heavy metals (As, Cd, Hg, and Pb) it was not possible with the resources available to detect concentrations found in samples.

This study was therefore carried out to provide the first report of metal contents in intertidal vegetated sediments and seagrass biomass along the Fijian coast. The aims of the study were to: (i) determine for the first time, contents of four trace metals (Cu, Fe, Mn and Zn) in three tropical seagrass species (*H. ovalis, H. pinifolia* and *H. uninervis*) at two sites; (ii) examine the relationship between metal contents and their environmental compartments (sediments and seagrass tissue); and (iii) reveal any species-specific metal accumulation patterns and identify seagrass species capable of accumulating specific metals.

## 2. Materials and methods

#### 2.1. Study area

The study was conducted at Nasese, Suva  $(18^\circ 9'28''S, 178^\circ 26'47''E)$  and Korotogo, Sigatoka  $(18^\circ 10'12''S, 177^\circ 31'53''E)$ , along the southern coast of Viti Levu, Fiji Islands (Fig. 1). The climate of Fiji may be categorised as an oceanic tropical marine climate with annual rainfall ranging from 1800 to 2600 mm (Singh et al., 2019). The average air temperature is 25 °C but ranges from 18 °C to 30 °C in summer months. The islands have distinct wet and dry zones, with the former located in the southeast region of Viti Levu, while the north and west are usually drier. The dry season is from May to October, the wet season from November to April, and tides are generally diurnal with an annual mean tidal range of 1.1 m. The mean range of neap tides is 0.9 m and spring tides is 1.3 m, and inshore sea surface temperatures have an annual average of between 24 °C to 31 °C.

Surveys were conducted near the two largest river systems in the Fiji Islands, the Rewa River and Sigatoka River located on the southern coast of Viti Levu (Fig. 1). The Rewa River empties into Laucala Bay through the Vunidawa River. The shoreline along the eastern side of Laucala Bay is largely covered by mangroves, and has a wide intertidal zone composed of very fine-grained mud, silt and sand from five relatively smaller rivers (Pohler and Collen, 2006). Situated along the coastline on the eastern side of the Sigatoka River, Korotogo has a wide fringing reef flat interrupted by a narrow channel.

At each location, sampling was performed within intertidal areas in seagrass meadows dominated by either *H. uninervis*, *H. pinifolia* or *H. ovalis*, as well as unvegetated areas adjacent to the seagrass study site. Sampling locations were chosen at random and only the top 10 cm of sediment was analyzed in this analysis. Because previous work (Pohler and Collen, 2006) had indicated that sediments in the area were predominantly mud (<63  $\mu g$  factor is  $\geq 90\%$ ), so no specific particle analysis was completed.

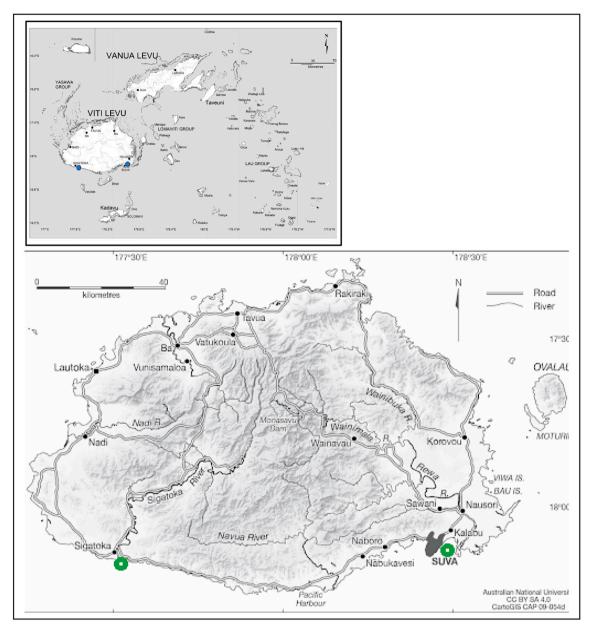


Fig. 1. Map of the Fiji Islands showing the two study sites located in the southeast (Rewa River) and southwest (Sigatoka River) of Viti Levu. Source: The map base layer has been adapted from Lal et al. (2016) and CartoGIS Services Maps Online Australian National University.

#### 2.2. Sediments and tissue sample collection

The fieldwork was conducted in the peak growth period in summer season (November 2017 to February 2018), during which were collected seagrass plants and sediments at Nasese in Laucala Bay, Suva, and Korotogo in Sigatoka. Sites were geolocated in the field using a handheld GPS unit ( $Garmin\ eTrex10$ ). The geographical coordinates for each station at the two sites are reported in Supplementary material, Table S1. These sites were distributed along the coastlines and at depths ranging from 0.1 to 0.5 m. We examined Cu, Fe, Mn and Zn content along all sediment cores and within seagrass tissue. Sampling was conducted using a linear transect and quadrant ( $50\ cm \times 50\ cm$ ) method to obtain sediments and plant tissue within each quadrant ( $50\ cm \times 50\ cm$ ) method to obtain sediments and plant tissue within each quadrant ( $50\ cm \times 50\ cm$ ) method to obtain sediments and plant tissue within each quadrant ( $50\ cm \times 50\ cm$ ) method to obtain sediments and plant tissue within each quadrant ( $50\ cm \times 50\ cm$ ) method to obtain sediments and plant tissue within each quadrant ( $50\ cm \times 50\ cm$ ) method to obtain sediments and plant tissue within each quadrant ( $50\ cm \times 50\ cm$ ) method to obtain sediments and plant tissue within each quadrant ( $50\ cm \times 50\ cm$ ) method ( $50\ cm$ 

To ensure that specimen records are traceable, sample collection data including GPS coordinates, taxonomic information and photographic images recorded during field collections were uploaded to the Barcoding of Life Data (BOLD) system repository (http://www.boldsystems.org) (Singh et al., 2019). This enabled the provision of support for identifications, and permitted easier comparisons between species (Singh et al., 2019).

Sampling at each location was carried out by selecting 15 stations every 5 m along a shore-perpendicular transect line method described by standard Seagrass-Watch methods (http://www.seagrasswatch.org). Within each station, three replicate sediment cores ( $\sim$ 2–5 m apart) were collected at different positions within the quadrant targeting top 10 cm of sediment, from both inside the meadow and an adjacent unvegetated area. The top 10 cm of sediment was within the depth range influenced by root growth (Greiner et al., 2013), and hence analyses were restricted to this layer as the focus of this study was trace metal content within the upper zone. Cores of sediment were sampled at low tide by manually inserting PVC pipe (30 cm length, 5 cm internal diameter) into the substrate. A total of 360 sediment cores and 45 quadrats were sampled to cover all areas of seagrass distribution within 160 ha of the coastal locations.

Sediment samples of known volumes were picked free of living plant tissue and stored in labelled Ziploc® bags and transported to the laboratory for further processing on the same day. In the laboratory, the sediment was cleaned of larger shells, in-fauna, and plant material before drying (Gullström et al., 2018). Sediments were dried at 60 °C for 48 h until a constant weight was achieved and homogenized (Govers et al., 2014).

For the collection of plant biomass in each quadrant, all above ground (leaves and stems) and belowground vegetation (rhizomes and roots) were harvested to a vertical depth of 10 cm. Seagrasses were dug from the substrate, cleaned in seawater to remove accompanying debris, and transported in labelled Ziploc® plastic bags to the laboratory. Epiphytes were removed by gently scarping seagrass leaves with a microscope slide. We saw no sign of herbivory (bite or grazing marks) in the collected leave samples. Cleaned seagrass tissue was dried at 60 °C to a constant weight for 48 h (Govers et al., 2014). Dried seagrass material was weighed to determine the dry weight of individual species. Dry samples of each species were pooled within each replicate and homogenized in preparation for trace metal analysis. We measured Cu, Fe, Mn and Zn contents of the sediments and whole plant biomass of the three seagrass species. In addition, bioaccumulation factors (BAF) for each species and trace metal element was calculated using the method described by Lewis and Devereux (2009), which is BAF = concentration in seagrass/concentration in water or sediment (Table 2).

Trace metal concentrations, obtained from Fiji's Great Astrolabe lagoon (Maata and Singh, 2008) were used to compare the trace metal concentrations in Nasese and Korotogo sediment and whole plant samples. The Great Astrolabe lagoon is far from anthropogenic influence and is considered here as a pristine and uncontaminated site, hence, can be used for comparison of heavy metals for studies in Fiji (Maata and Singh, 2008). Iron and manganese were not determined in the Astrolabe lagoon work. Moreover, the value for  $I_{\rm geo}$  was calculated using Eq. (1), where  $C_n$  is the concentration of the heavy metal (n) measured while  $B_n$  is the geochemical background concentration of that particular heavy metal (n) (Muller, 1969). By using the Great Astrolabe value, the geoaccumulation index was calculated.  $I_{\rm geo}$  comprises seven different classes (Table S3).

$$I_{geo} = log_2 [C_n/1.5 B_n]$$
 (1)

## 2.3. Copper, Iron, manganese and zinc extraction

The extraction method of Lindsay and Norvell (1978), which uses diethylenetriaminepentaacetic acid (DTPA) was used to assess Cu, Fe, Mn and Zn contents of sediment samples. In brief, sediment samples weighing 10 g were digested in 20 ml of extracting reagent (double strength reagent - 0.005 M DTPA, 0.01 M calcium chloride and 0.1 M TEA) and centrifuged at 2000 rpm for 15 min. The solution was filtered using a Whatman nucleopore polycarbonate track-etch membrane filter paper, which has a diameter of 42 mm and porosity of 0.4  $\mu$ m (Merck KGaA). The metal content (Cu, Fe, Mn, and Zn) in sediments and whole plant tissues were determined using an atomic absorption spectrometer (AAS – Pekin Elmer®, Pinaacle 900F).

The method used for analysis of plant tissues was described by Prasad and Spiers (1978), and involved dry ashing the plant samples (1 g) for 4 h at 475 °C, then grinding to a powder. Dried plant materials were digested with hydrochloric acid (5 ml) and gently heated for approximately 20 min. The residue was then made up to a 50 ml solution volume using distilled water after filtering through No. 42 Whatman filter paper, and then metal concentrations (Cu, Fe, Mn and Zn) were determined using an atomic absorption spectrometer (AAS – Pekin Elmer®). All analyses were carried out at the chemistry laboratory of the Koronivia Research Station, Ministry of Agriculture in Fiji.

The quality checking was verified through analytical blanks and stability of instrumental recalibration (Bonanno et al., 2020). Analyses were conducted in triplicates and method detection limits were

employed in order to justify the reliability and validity of the method used. In addition, percent recovery for all the metals analyzed was within the acceptable range of 90 and 100%. Based on the standard error given by supplier, our results fell within the acceptable range (95% confidence interval). Wavelengths used for analysis of each metal using the atomic absorption spectrometer are provided in Supplementary material, Table S2.

#### 2.4. Data analysis

#### 2.4.1. Descriptive statistics

Exploratory data analysis was carried out to check the variability and distribution of each variable: Cu, Fe, Mn, Zn and sites. All data were tested for homogeneity of variances and normality with Levene's and Shapiro-Wilk tests, respectively, and where necessary Box-Cox transformed to conform to the assumptions of ANOVA. We assume that high between-site variability is a signal of prevalent local sources of contamination. Testing for differences in the variables studied among communities, meadows and sites was determined using a two-way fixed factor ANOVA. Tukey's HSD (honest significant difference) test was performed to distinguish differences among and between seagrass plant tissues and sediment. Confidence intervals (95%) and p-values obtained for the ANOVA and Tukev's tests were used to consider differences in sediments and seagrass species. Because sites were distributed across three meadows, models included both community and meadow as direct effects, along with their interaction. All statistics were performed using R statistical software version 3.6.3 (R Core Team, 2020) using the following packages: basic, car, and mass.

#### 3. Results

We assessed trace metal contents of seagrasses as an indicator of their level of pollution in coastal areas and their bioavailability in seagrass tissues and sediments. Measurement of trace metals in sediment provided a standard for metal loads in the area, while seagrass tissue content reflects longer-term trace metal accumulation (Govers et al., 2014). Significant variation (p < 0.05) was recorded for the levels of trace metals in sediment and seagrass tissues with the latter consistently displaying high contents of all trace metals determined in this study (Table 5; Figs. 2-5). Iron (Fe) was identified as having the highest element concentration affecting sediments in seagrass meadows at both sites, with it being accumulated most in seagrass tissue. While, Mn had the second highest element level observed in both sediments and tissues of the three seagrass species. In contrast, Cu and Zn were considered the least accumulated anthropogenic pollutant in both sediment and seagrass tissue. Mean values ( $\pm$ SD) and trends in the levels of each metal in both sediments and whole plant tissue are summarized in Table 1.

Seagrass bioaccumulation factor values (BAF) displayed wide variation between seagrass species and are impacted by physical, chemical and biological factors (Lewis and Devereux, 2009; Table 2). This indicates that *H. ovalis* concentrated more trace metals than *H. pinifolia* and *H. uninervis* based on bioaccumulation factor values when comparing whole plant-sediment values obtained for Cu, Fe, Mn and Zn. *H. ovalis* had a higher than anticipated trace metal content between 1 and 5-fold (Table 2) for all metals. In contrast, differences in Cu, Fe, Mn and Zn content in *H. pinifolia* and *H. uninervis* were less than 1.3-fold.

 $I_{geo}$  values were calculated for Cu and Zn elements only and reported in Table 3. On the other hand, Fe and Mn were not determined in Astrolabe lagoon work, thus are not calculated and included in the table. The  $I_{geo}$  values for Cu and Zn from the two sites were mostly within 0–1, which were assigned to be no to moderately contaminated. Results indicated that Korotogo was mostly contaminated with higher levels of Cu and Zn than compared to Nasese, which are mostly introduced into the coastal environments by anthropogenic means. This might be mainly because the area have been quite urbanized and industrialized with construction and hotel developments that could be contributing source

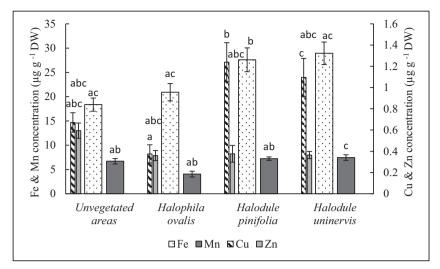


Fig. 2. Mean ( $\pm$ SE) trace metal content ( $\mu$ g g $^{-1}$  dry weight) in sediments in seagrass meadows (with seagrasses separated at the species/genus level) and unvegetated areas pooled. Copper and zinc content in sediment are presented on the secondary vertical (right) axis for reference. The trace metal values are based on a sediment depth of 10 cm. The trace element levels of each sample type vary significantly across vegetated and unvegetated sites; to show this, for each sample type, different letters per different seagrass beds were used; Tukey's test,  $\alpha=0.001$ .

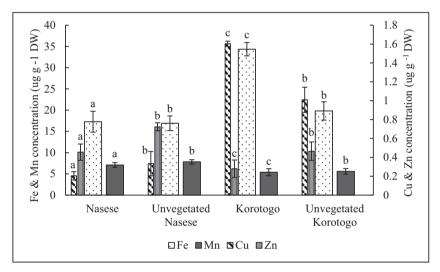


Fig. 3. Mean ( $\pm$ SE) trace metal content ( $\mu$ g g  $^{-1}$  dry weight) in sediments for seagrass meadows (with the three seagrasses, that is, Ho, Hp, and Hu sp., merged; for seagrass name abbreviations, see Fig. 2) and unvegetated areas from two Fijian sites. The trace metal values are derived from pooled seagrass meadows and unvegetated areas to a sediment depth of 10 cm. Copper and zinc contents of sediment are presented on the secondary vertical (right) axis for reference. The trace element levels of each sample type vary significantly across vegetated and unvegetated sites; to show this, for each sample type, different letters per different sites were used; Tukey's test,  $\alpha=0.001$ .

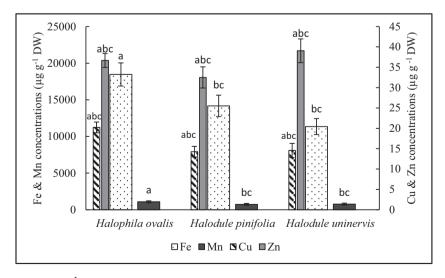


Fig. 4. Mean ( $\pm$ SE) trace metal content ( $\mu$ g g<sup>-1</sup> dry weight) in seagrasses species meadows from two Fijian sites. Copper and zinc contents of sediment are presented on the secondary vertical (right) axis for reference. The trace element levels of each sample type vary significantly across species; to show this, for each sample type, different letters per different seagrass species were used; Tukey's test,  $\alpha = 0.001$ .

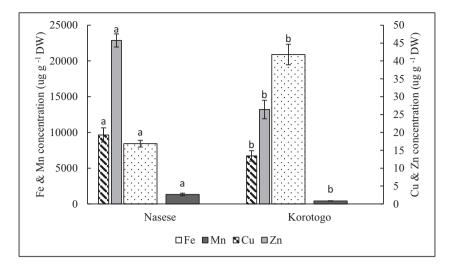


Fig. 5. Mean ( $\pm$ SE) trace metal content ( $\mu$ g g<sup>-1</sup> dry weight) in seagrasses species meadows from two Fijian sites. Copper and zinc contents of sediment are presented on the secondary vertical (right) axis for reference. The trace element levels of each sample type vary significantly across the two sites; to show this, for each sample type, different letters per different site were used; Tukey's test,  $\alpha = 0.001$ .

Table 1
Mean ( $\pm$ SD) trace metal contents ( $\mu$ g g<sup>-1</sup> dry wt.) of sediments and seagrass species. Sediment values are at depth of 10 cm for the combined 15 plots for both study sites.

Study site Habitat	Sediment					Plant					
	Cu	Fe	Mn	Zn	Trend	Cu	Fe	Mn	Zn	Trend	
Nasese											
Halophila ovalis	0.11 $\pm$	15.02 $\pm$	6.84 $\pm$	0.36 $\pm$	Cu < Zn < Mn	20.33 $\pm$	10,782.93 $\pm$	1329.27 $\pm$	42.27 $\pm$	Cu < Zn < Mn	
	0.09	10.30	2.29	0.28	< Fe	6.65	2191.02	920.96	8.00	< Fe	
Halodule	0.32 $\pm$	17.93 $\pm$	7.55 $\pm$	$0.60 \pm$	Cu < Zn < Mn	17.07 $\pm$	7493.07 $\pm$	1251.13 $\pm$	42.67 $\pm$	Cu < Zn < Mn	
pinifolia	0.33	9.95	2.38	0.52	< Fe	6.06	2146.90	685.60	6.50	< Fe	
Halodule	0.17 $\pm$	18.89 $\pm$	6.87 $\pm$	0.40 $\pm$	Cu < Zn < Mn	20.47 $\pm$	7005.60 $\pm$	1380.87 $\pm$	52.27 $\pm$	Cu < Zn < Mn	
uninervis	0.09	8.32	1.92	0.20	< Fe	10.42	1098.98	497.21	6.82	< Fe	
Unvegetated	$0.33~\pm$	16.92 $\pm$	7.82 $\pm$	0.72 $\pm$	Cu < Zn < Mn						
area	0.11	6.09	3.18	0.36	< Fe						
Korotogo											
Halophila ovalis	$0.64 \pm$	26.81 $\pm$	$1.25~\pm$	0.36 $\pm$	Zn < Cu < Mn	20.13 $\pm$	26,137.00 $\pm$	852.87 $\pm$	$31.07~\pm$	Cu < Zn < Mn	
	0.54	3.91	0.56	0.24	< Fe	7.66	5123.15	204.99	6.99	< Fe	
Halodule	2.15 $\pm$	37.26 $\pm$	6.91 $\pm$	0.15 $\pm$	Zn < Cu < Mn	11.47 $\pm$	20,861.33 $\pm$	266.33 $\pm$	22.33 $\pm$	Cu < Zn < Mn	
pinifolia	0.48	8.73	1.38	0.10	< Fe	7.14	5891.36	73.20	12.46	< Fe	
Halodule	2.02 $\pm$	38.97 $\pm$	8.02 $\pm$	0.32 $\pm$	Zn < Cu < Mn	8.67 $\pm$	15,700.00 $\pm$	175.20 $\pm$	25.83 $\pm$	Cu < Zn < Mn	
uninervis	0.47	7.28	3.95	0.15	< Fe	2.09	5672.34	39.79	10.58	< Fe	
Unvegetated	1.01 $\pm$	19.81 $\pm$	5.58 $\pm$	0.46 $\pm$	Zn < Cu < Mn						
area	0.52	8.32	2.58	0.38	< Fe						

**Table 2** Bioaccumulation factor (BAF) values and ratios for trace metal content ( $\mu g g^{-1}$  dry weight) in the seagrasses. BAF values were calculated based on Lewis and Devereux (2009).

Species	Ratio (conc. seagrass/conc. sediment)							
	Cu	Fe	Zn	Mn				
Halophila ovalis	4.7	2.3	1.2	2.6				
Halodule pinifolia	1.0	1.3	1.0	1.0				
Halodule uninervis	1.2	1.0	1.2	1.0				

of Cu and Zn in the coastal environment. While it is not possible at this stage to fully explain these patterns, they do provide a means of distinguishing between the samples.

The results obtained for the trace metals in seagrass bed sediments were also compared to the results of other studies in Indo-Pacific region as shown in Table 4. The concentration of Cu, Fe, Mn and Zn obtained in the present study seemed to be lower than that of previous studies. When compared to the pristine environment i.e., the Great Astrolabe Reef in

Geo-accumulation index ( $I_{geo}$ ) for the studied metals at the two sites. Background concentration of metals used in this study is from the Great Astrolabe reef in Kadavu, Fiji; Fe and Mn were not determined.

Site/habitat	Cu	Zn
Nasese		
Halophila ovalis	0.0008	0.0024
Halodule pinifolia	0.0022	0.0041
Halodule uninervis	0.0011	0.0027
Unvegetated area	0.0022	0.0049
Korotogo		
Halophila ovalis	0.0043	0.0025
Halodule pinifolia	0.0150	0.0010
Halodule uninervis	0.0140	0.0022
Unvegetated area	0.0071	0.0031
Background value	29.6	29.5

Kadavu, the concentrations of all metals analyzed at both sites are quite low. This is mainly because the plants absorb the seagrass bed metal elements. Since, seagrasses show different element accumulation

Table 4 Comparison of the sedimentary trace metal contents ( $\mu g g^{-1}$ ) in Nasese and Korotogo with other studies done in the Indo-Pacific region.

Study site	Cu	Fe	Mn	Zn	Reference
Nasese	0.11-0.32	15.02-18.89	6.84–7.55	0.36-0.60	Present study
Korotogo	0.64-2.15	26.81-38.97	1.25-8.02	0.15-0.36	
Laucala Bay	44.2–170	13,733-68,492	_	15.5-15.7	Pratap et al., 2020
Great Astrolabe reef	22-88	_	_	10–164	Morrison and Nagasima, 1999
Rewa River	4.5-6.0	2200-7800	_	5.5-584	Deo, 2000
Lami	5-1980	_	_	52-514	Gangaiya et al., 2001
Suva Harbour	21.4-143	14,000-48,700	_	40.2–269	Maata and Singh, 2008
Tonga	2-180	1550-114,000	_	6–208	Morrison and Brown, 2003
Kiribati	0.3-13.9	_	_	1.2–77	Redfern, 2006
New Zealand	14–48	_	_	97–896	Redfern, 2006
India	214.4	3600-34,600	70–818	41–184	Mishra et al., 2020
China	18.23-69.34	-	519.25–1212.79	71.32–143.64	Ahamad et al., 2020

Table modified from Pratap et al. (2020).

patterns among the two study sites (Tables 1–3); they may act as biological filters, favoring the decrease of metals.

#### 3.1. Variability of sedimentary and plant Cu

Seagrass habitats had higher mean Cu content ( $0.90\pm0.10~\mu g~g^{-1}$ ) than nearby unvegetated habitats ( $0.67\pm0.09~\mu g~g^{-1}$ ) at both sites (Fig. 2). The mean Cu content in seagrass sediment ranged from 0.01 to 3.35  $\mu g~g^{-1}$  and was up to one and a half times the levels detected in nearby unvegetated sediments (0.20 to 2.11  $\mu g~g^{-1}$ ) (Fig. 2). When comparing pooled vegetated habitats versus unvegetated areas, Cu contents were significantly different (Table 5; Fig. 2). The high between sites variability were recorded in H. ovalis, H. pinifolia and H. uninervis beds, while unvegetated sediments did not show significant difference (Table 5). Similarly, when comparing vegetated habitats with unvegetated areas at both sites, there was a significant effect on mean Cu contents (Table 5; Fig. 2), which was also apparent between Nasese and Korotogo. The results demonstrated that Korotogo vegetated and unvegetated areas stored more Cu compared to the Nasese vegetated and unvegetated areas (Fig. 3).

The mean Cu content in seagrass plant tissue was  $16.36\pm0.90~\mu g~g^{-1}$  and ranged from 3 to 53  $\mu g~g^{-1}$ . *H. uninervis* had the highest mean plant tissue Cu content (above and below ground combined) of  $11–53~\mu g~g^{-1}$ , with a mean of  $20.47\pm2.69~\mu g~g^{-1}$ . *H. ovalis* had the second highest mean plant tissue Cu content of  $10–34~\mu g~g^{-1}$ , with a mean of  $20.33\pm1.72~\mu g~g^{-1}$ , while *H. pinifolia* had the lowest mean plant tissue Cu content of 4–33, with a mean of  $11.47\pm1.84~\mu g~g^{-1}$  (Fig. 4). There was a significant interaction demonstrated between seagrass species and Cu

content (Table 5; Fig. 4). However, significant differences between the three seagrass species were not apparent. The mean Cu content in seagrass tissue differed significantly among sites (Table 5; Fig. 5). Moreover, the sedimentary and seagrass tissue Cu content was highly significant in the two sites in Fiji (Table 5; Fig. 5).

## 3.2. Variability of sedimentary and plant Fe

Seagrass habitats had a higher mean Fe content ( $25.81 \pm 1.33 \ \mu g^{-1}$ ) than nearby unvegetated habitats ( $18.36 \pm 1.36 \ \mu g \ g^{-1}$ ) at both sites (Fig. 2). The mean Fe content in seagrass sediment ranged from 6.34 to 55.37  $\mu g \ g^{-1}$  and was up to one and a half times the content in nearby unvegetated sediments ( $7 \text{ to } 40 \ \mu g \ g^{-1}$ ) (Fig. 2). When comparing pooled vegetated habitats versus unvegetated areas, Fe content were significantly different (Table 5; Fig. 2). However, the only significant variability in Fe content was demonstrated in *H. pinifolia* beds between *H. ovalis*, *H. uninervis* and unvegetated sediments compared to the other interactions. When comparing vegetated habitats and unvegetated areas at both sites, there was a significant effect on mean Fe content (Table 5; Fig. 2), which was also apparent between Nasese and Korotogo. Korotogo vegetated and unvegetated areas were found to store more sedimentary. Fe compared to Nasese vegetated and unvegetated areas (Fig. 3).

The mean Fe content in seagrass plant tissue was  $1466.32\pm860.66$  µg g $^{-1}$  and ranged from 4467 to 34,668 µg g $^{-1}$ . *H. ovalis* had the highest mean plant tissue Fe content (above and below ground combined) of 15,563–34,668 µg g $^{-1}$ , with a mean of 26,137.00  $\pm$  1322.79 µg g $^{-1}$ . *H. pinifolia* had the second highest mean plant tissue Fe content of

Table 5 Summary of two-way ANOVA and post hoc Tukey's test for the effects of site, unvegetated and vegetated areas, Cu, Fe, Mn and Zn on sediments and seagrass tissue of *Halophila ovalis*, *Halodule pinifolia* and *Halodule uninervis*. Bold indicates statistically significant effects at  $p \le 0.05$ .

	Cu			Fe			Mn			Zn		
	df	F value	p value	df	F value	p value	df	F value	p value	df	F value	p value
Sediments												
Site	1	105.69	< 0.001	1	37.09	< 0.001	1	270.12	< 0.001	1	11.81	< 0.001
Unvegetated * vegetated	6	31.51	< 0.05	4	17.24	< 0.05	4	18.79	< 0.05	4	2.87	< 0.05
Ho * unvegetated	0.19		0.79	-2.56		0.79	0.57		0.74	0.23		0.05
Hp * Ho	0.81		< 0.01	6.67		< 0.01	0.46		0.87	0.017		0.99
Hu * Ho	1.38		< 0.05	-2.03		0.95	2.95		< 0.01	0.04		0.99
Hp * unvegetated	-0.43		0.01	-9.23		< 0.05	0.12		0.73	0.22		0.08
Hu * unvegetated	0.10		0.98	-0.52		0.99	3.28		< 0.0001	0.19		0.36
Hu * Hp	0.57		< 0.05	11.38		< 0.05	2.49		0.0003	0.03		0.99
Sample * site	0.36		< 0.001				-4.70		0.0000	-0.18		0.003
Whole plant samples												
Site	1	20.56	< 0.001	1	284.6	< 0.001	1	93.54	< 0.001	1	97.23	< 0.001
Seagrass tissue	3	4.09	< 0.05	3	104.7	< 0.05	3	15.98	< 0.05	3	6.32	< 0.05
Ho * Hp	-6.06		0.06	10,429.36		0.0000	624.46		0.002	1.43		0.96
Ho * Hu	-5.77		0.08	7848.69		0.0000	-637.99		0.001	-3.22		0.69
Hp * Hu	0.30		0.99	-2580.67		0.09	64.85		0.96	6.55		0.04
Sample * site	-5.8		0.0004	12,185.49		0.0000	-876.21		0.0000	-15.59		0.0000

10,568–34,303 µg g<sup>-1</sup>, with a mean of 20,861.33  $\pm$  1521.14 µg g<sup>-1</sup>, while *H. uninervis* had the lowest of 6628–27,823 µg g<sup>-1</sup>, with a mean of 15,700.00  $\pm$  1464.59 µg g<sup>-1</sup> (Fig. 4). There was a significant interaction between seagrass species and Fe content (Table 5; Fig. 4). Significant differences were apparent for *H. ovalis* between *H. pinifolia* and *H. uninervis*, while both *H. pinifolia* and *H. uninervis* did not differ significantly from each other. The mean Fe in seagrass tissue differed significantly among sites (Table 5; Fig. 5). The sedimentary and seagrass tissue Fe content comparison was highly significant in two sites in Fiji (Fig. 5).

## 3.3. Variability of sedimentary and plant Mn

Seagrass habitats had lower mean Mn content  $(4.05\pm0.59~\mu g~g^{-1})$  than nearby unvegetated habitats  $(6.70\pm0.57~\mu g~g^{-1})$  at both sites (Fig. 2). The mean Mn content in seagrass sediment ranged from 0.61 to 15.93, and in nearby unvegetated sediment varied from 1.98 to 17.10 (Fig. 2). When comparing pooled vegetated habitats versus unvegetated areas, Mn content were significantly different (Table 5; Fig. 2). Moreover, significant variability was observed in *H. uninervis* beds between *H. ovalis, H. pinifolia*, and bare sediment; whereas no significant difference were demonstrated for *H. ovalis, H. pinifolia*, and bare sediments. When comparing vegetated habitats and unvegetated areas across both sites, there was a significant effect on mean Mn storage (Table 5; Fig. 3), which was also apparent between Nasese and Korotogo. Nasese vegetated and unvegetated areas stored more sedimentary Mn compared to Korotogo vegetated and unvegetated areas (Fig. 3).

The mean Mn content in seagrass plant tissue was  $869.28\pm76.12~\mu g^{-1}$  and ranged from 99 to 3580  $\mu g~g^{-1}$ . H. uninervis had the highest mean plant tissue Mn content (above and below ground combined) of  $845-2760~\mu g~g^{-1}$ , with a mean of  $1380.87\pm128.38~\mu g~g^{-1}$ . H. ovalis had the second highest mean plant tissue Mn content of  $388-3580~\mu g~g^{-1}$ , with a mean of  $1329.27\pm237.79~\mu g~g^{-1}$ , while H. pinifolia had the lowest mean plant tissue Mn content of  $579-3193~\mu g~g^{-1}$ , with a mean of  $1251.13\pm177.02~\mu g~g^{-1}$  (Fig. 2). There was a significant interaction observed between seagrass species and Mn content (Table 5; Fig. 4). Significant differences were apparent for H. ovalis and between H. pinifolia and H. uninervis, while both H. pinifolia and H. uninervis did not differ significantly from each other. The mean Mn in seagrass tissue differed significantly among sites (Table 5; Fig. 5). Likewise, the sedimentary and seagrass tissue Mn content was highly significant between two sites in Fiji (Fig. 5).

## 3.4. Variability of sedimentary and plant Zn

Seagrass habitats had lower mean Zn content  $(0.37\pm0.03~\mu g~g^{-1})$  than nearby unvegetated habitats  $(0.59\pm0.07~\mu g~g^{-1})$  at both sites (Fig. 2). The mean Zn content in seagrass sediment ranged from 0.002 to  $2.17~(\mu g~g^{-1})$ , and in nearby unvegetated sediments it varied from 0.17 to  $1.88~\mu g~g^{-1}$  (Fig. 2). Furthermore, when comparing pooled vegetated habitats versus unvegetated areas, Zn contents were significantly different (Table 5; Fig. 2). However, significant differences between all the sediment samples were not observed during pairwise comparisons. Meanwhile, when comparing vegetated habitats and unvegetated areas at both sites, there was a significant effect on mean Zn storage (Table 5; Fig. 2), which was also apparent between Nasese and Korotogo. Nasese vegetated and unvegetated areas stored more sedimentary Zn, compared to Korotogo vegetated and unvegetated areas (Fig. 3).

The mean Zn content of seagrass plant tissue was  $36.07\pm1.45~\mu g~g^{-1}$  and ranged from 0.5 to 68  $\mu g~g^{-1}$ . *H. uninervis* had the highest mean plant tissue Zn content (above and below ground combined) of 42–68  $\mu g~g^{-1}$ , with a mean of 52.27  $\pm$  1.76  $\mu g~g^{-1}$ . *H. pinifolia* had the second highest mean plant tissue Zn content of 33–54, with a mean of 42.67  $\pm$  1.68  $\mu g~g^{-1}$ , while *H. ovalis* had the lowest mean plant tissue Zn content of 19–47, with a mean of 31.07  $\pm$  1.80  $\mu g~g^{-1}$  (Fig. 4). There was a significant interaction demonstrated between seagrass species and Zn

content (Table 5; Fig. 4). However, the three seagrass samples failed to differ significantly from each other. The mean Zn content in seagrass tissue differed significantly among sites (Table 5; Fig. 5). Both sedimentary and seagrass tissue exhibited a highly significant Zn content between the two sites in Fiji (Fig. 5).

#### 4. Discussion and conclusion

The trace metals analyzed in this study (Cu, Fe, Mn and Zn) were selected as they represent the most common trace and toxic metals affecting coastal communities (Roberts et al., 2008). These metals are also essential micronutrients for plants and are not necessarily toxic but many anthropogenic activities increase their natural concentrations causing pollution (Sanz-Lazáro et al., 2012). Additionally, Ambo-Rappe et al. (2011) pointed out that once threshold levels of trace elements are reached, it affects both root cellular structure and photosynthesis. In some areas, seagrasses represent the greatest heavy metal reservoir, and the ecosystem can remobilize metals through litter production and in food chains (Amado Filho et al., 2004). This contrasts with the relative immobilization which occurs in unvegetated sediments (Amado Filho et al., 2004). To understand trace element cycling in seagrasses, it is important to study the accumulation trends in the whole plants followed by further investigation into accumulation patterns among their compartments such as leaves, roots, rhizomes and epiphytes (Sanz-Lazáro

This study is the first to focus on seagrass trace metal contents in whole plant tissues, seagrass meadows and compare these with unvegetated areas, based on earlier recognition of these metals (Cu, Fe, Mn and Zn) as historical contaminants in Laucala Bay, Fiji (Morrison et al., 2001). *H. ovalis, H. pinifolia* and *H. uninervis* were selected for this study because they are common along the Fijian coastlines. Moreover, Short et al. (2014) highlighted that in the tropical Western Pacific region, *H. ovalis, H. pinifolia* and *H. uninervis* are considered pioneer species growing rapidly and surviving well in unstable or depositional environments. Similar to other studies, we found significant variation in metal contents with respect to species and site (Table 5). This study demonstrated that trace metal content varied among the three seagrass species, prominently including high Fe content in sediments and whole plant tissue.

Seagrass sediments from Korotogo and Nasese both showed greater Fe and Mn content compared to plant tissues, and similarly, unvegetated areas showed higher content of these two metal elements. In contrast, micronutrient (Cu and Zn) contents were the lowest and least accumulated in all seagrass species and sediments at both sites. Cu and Zn are essential for the growth of seagrass and in general, their concentrations are low, i.e.,  $\leq \! 10 \, \mu g \, g^{-1}$  for Cu and  $\leq \! 30 \, \mu g \, g^{-1}$  for Zn (Immaculate et al., 2018). Additionally, sediment contents of Zn and Cu at Nasese were lower than tissue contents of all three seagrass species. In contrast, Morrison et al. (2001) reported that intertidal sites at Laucala Bay showed high content of Cu and Zn possibly due to pollution from a local industrial area, less than 1 km upstream from the Vatuwaqa River which discharges effluents into the Bay.

Pratap et al. (2020) reported Zn content in water samples from Laucala Bay ranging between  $76.9 \pm 0.9$  to  $119.0 \pm 3.0 \, \mu g \, g^{-1}$ , which is well within the ranges reported here for seagrass tissue and sediments. The authors reported a decrease in Zn content for the area over several years, which may result from a decrease in anthropogenic sources and implementation of an Environmental Management Act (EMA) by the Government of Fiji (Pratap et al., 2020). The contributing source of Cu and Zn in the coastal environment could have been from the urbanization and industrialization of these areas (Pratap et al., 2020). The present study assessed Zn concentrations in sediment and identified it as being available at the third highest level. Our data shows that Zn was recorded at the highest content in tissues of *H. uninervis* (52.27  $\pm$  1.76  $\mu g \, g^{-1}$ ) at Nasese, while the lowest level was recorded in *H. pinifolia* (22.33  $\pm$  3.22  $\mu g \, g^{-1}$ ) tissues at Korotogo. The results obtained for Zn in

the present study were compared to the results of Mishra et al. (2020) for the same three seagrass species where H. pinifolia (35–69.17  $\mu g g^{-1}$ ) had accumulated higher Zn content, while H. ovalis Zn content was within this range (Table S4).

Mishra et al. (2020) reported that the Cu content of the seagrass (H. ovalis, H. pinifolia and H. uninervis) from India varied from 3.79 to  $60.83~\mu g~^{-1}$  which is comparable to those reported in the three seagrass species in this study (8.7 to  $20.5~\mu g~^{-1}$ ). Nonetheless, the low Cu and Zn contents of seagrasses reported in this study, from both study sites, suggest moderate trace metal (Table 3) pollution, and such levels may be considered background levels for a tropical coastal locality. The Cu and Zn contents detected reflect that these may be mobilized by human activities, in contrast to Fe and Mn which are mainly of lithogenic origin (Serrano et al., 2011).

The results showed that *H. ovalis* ( $5668 \pm 139.66$  ind. m $^{-2}$ ) had a 1.4-fold lower shoot density (Table S5) than that of *H. pinifolia* ( $6450.00 \pm 200.15$  ind. m $^{-2}$ ) and *H. uninervis* ( $8069.00 \pm 189.99$  ind. m $^{-2}$ ). Whole plant biomass of *H. ovalis* ( $3522.00 \pm 159.22$  g DW m $^{-2}$ ) was 3-fold lower than that of *H. uninervis* ( $9693.20 \pm 244.86$  g DW m $^{-2}$ ); while *H. pinifolia* was 1.5-fold lower than that of *H. uninervis* (Table S5). The Fe contents of all three seagrasses sampled in this study were high with *H. ovalis* accumulating the highest level of  $26,137.00 \pm 1322.79 \,\mu g \,g^{-1}$  at the Korotogo site. Although in general, the plants with higher biomass tend to accumulate higher metals, this seems not to be the case in the present study. This may result from smaller seagrass species such as *H. ovalis* that have thinner roots underlying surface sediments that may allow easier access to elements and a greater surface to volume or biomass ratio compared to larger seagrass species (Duarte et al., 1998).

Furthermore, H. ovalis roots are located within the first few centimeters of sediment and are likely to extract Fe more rapidly compared to H. pinifolia and H. uninervis, which have a more complex root and rhizome network (pers. obs.). Fe content was around 1 to 5-fold higher in seagrass biomass than in sediments at both sites in this study. The results also show that, of those tested, Fe was the metal element with the greatest absorption in sediments of both vegetated and unvegetated areas. Prange and Dennison (2000) reported Fe as an essential element required by enzymes and proteins during photosynthesis and respiration processes. They suggested that higher Fe content in some plants may cause photosensitivity linked to Fe accumulation in chloroplasts, while Duarte et al. (1995) suggested a minimum of 600 mg kg<sup>-1</sup> was required for Fe content to be sufficient for seagrasses. Overall, the seagrasses sampled at the two Fijian sites in this study had an average Fe content of 25,281.60 and 62,698.33  $\mu g\ g^{-1},$  respectively, at Suva and Sigatoka, which are well above the average global values reported for seagrasses (e.g., 412 mg kg<sup>-1</sup>, Anton et al., 2018). The concentration of Fe obtained in the current study seemed to be more than sufficient.

Moreover, our results are in line with the findings of Pratap et al. (2020) and other studies in the Pacific region (Table 4) who reported high Fe contents in water samples from Laucala Bay (ranging from 47,067 to  $68,492 \,\mu g \, g^{-1}$ ) and sediments. Although the present study did not measure elements in water however, concentration in the water, sediment and seagrass might be different but may be correlated. These authors suggested that the Fe content showed low increases over the past 25 years that could be due to minimal anthropogenic or natural weathering processes in the area. On the other hand, our study also showed that seagrasses at the Korotogo site accumulate more Fe in their tissues than those at Nasese. Similar trend were observed in the whole plant tissues of the three seagrass species reported by Mishra et al. (2020) where Fe content was the highest element (795–1886  $\mu g g^{-1}$ ) accumulated in H. pinifolia (Table S4). However, Fe content in the present study (7493-26,137 µg g<sup>-1</sup>) was much higher than reported by Mishra et al. (2020); this could be because the uptake of the elements is influenced by their available concentrations in the surrounding sediment.

The highest Mn content was recorded in H. uninervis beds (8.02  $\pm$  1.02  $\mu g\,g^{-1})$  at Korotogo, however, the overall Mn accumulation seemed

to be more in seagrass tissues from Nasese. Patterns of Mn accumulation was reasonably consistent between the samples analyzed, indicating that they all responded similarly and irrespectively of the total Mn content, and thus, may be considered representative of the total metal availability in the environment. Elevated Mn contents in sediments and seagrass biomass have been cited for other seagrass species (Prange and Dennison, 2000). Analysis of the biological concentration factor showed that *H. ovalis* accumulated up to five-fold higher Cu concentration compared to one-fold for both the *Halodule* species. Similarly, Fe concentration was found to be one-fold for both *Halodule* species compared to two-fold for *H. ovalis*. On the other hand, bioaccumulation factor values showed that *H. ovalis* accumulated approximately two and a half times more Mn than the two *Halodule* species. While, bioaccumulation factor for *H. ovalis*, *H. pinifolia* and *H. uninervis* showed Zn concentration to be approximately one-fold.

Therefore, the analysis of biological concentration factor led to the conclusion that seagrass H. ovalis can be a good bioindicator for some of these metals as pollutants of coastal sediment. Lewis and Devereux (2009) pointed out that Zostera marina and P. oceanica have been recommended as bioindicators of environmental condition on numerous occasions and as agents of phytoremediation due to their bioaccumulation ability. However, the authors suggested that bioaccumulation is affected by the time of collection (growing, nongrowing seasons) and the tissue (above and belowground) analyses (Lewis and Devereux, 2009). Moreover, there is a need to investigate bioaccumulation in these seagrasses during non-growing season and analyze separately above and belowground tissues that were not carried out in the present study. Once a study of biological (leaves, roots and rhizome) has been completed, it may be possible to determine which individual materials are storing most of these metal elements. Globally, there are some toxicity assessment of trace elements on seagrass physiology of H. ovalis (Ambo-Rappe et al., 2011) and H. uninervis (Prange and Dennison, 2000). However in Fiji, trace metal toxicity assessment on seagrass physiology or growth have not been reported therefore, further investigation are needed to provide details and understanding the impacts of these on the plant and coastal environment.

As a result, Fe and Mn distributions in sediments and seagrass tissue at both the sites displayed a similar pattern of variation that varied significantly in sediment but not in seagrass tissues (Table 5). The results show that the close relationship between Fe and Mn may result from the two metals having the same source. This implies that Fe and Mn, which are of lithogenic origin, may result from relatively high terrigenous inputs to coastal sediments from local sources such as catchments and surface runoff (Serrano et al., 2011). We found that seagrass species were not Fe or Mn deficient, but higher than anticipated content occurred in carbonate sediment because of their lithogenic origin. In contrast, Cu and Zn, which mobilize through human activities such as urban and industrial effluents, agricultural activity, sewage and boating, were low and reflected a positive correlated pattern. The most obvious explanation for these low levels could be the implementation of an Environmental Management Act (EMA) by the Government of Fiji in 2005 as pointed out by Pratap et al. (2020). Additionally, the interdependency between these metals (Fe and Mn; Cu and Zn) means that reduction in the availability of one metal and the simultaneous increase in another may result in uncertainty in predicting responses.

Our results indicate that variation in the trace metal content of within different ecosystems could be due to various land-use activities in the surrounding area, even in different regions of the same ecosystems. While it is not possible at this stage to fully explain this, they do provide a means of guiding between samples metal content. These findings indicate that trace metal contents recorded in vegetated and unvegetated areas may be site-specific, suggesting further investigations should be performed to clarify point source pollution in local areas. Moreover, the surrounding environmental conditions of seagrasses should be controlled to avoid any metal pollution effect (Prange and Dennison, 2000). Therefore, from an environmental management perspective

there is a need to know which seagrass species are able to accumulate or concentrate particular metals for mitigation of coastal metal pollution. Our study shows that H. ovalis has a remarkable ability to take-up and accumulate these metals more efficiently and effectively compared to the other two seagrass species. However, it is worth mentioning that literature reports of growth rate of H. uninervis (1.53 cm daygenerally higher than H. ovalis (0.69 cm day<sup>-1</sup>) as reported by Mishra et al. (2021) and is generally acknowledged of the two genus. It is also noteworthy that H. ovalis accumulated up to five-fold higher contents of the four trace metals tested, compared to H. pinifolia and H. uninervis. This could be because the species has a strong ability to grow clonally, which plays a critical role in its adaptation to various environments (Erftemeijer and Stapel, 1999). Rozaimi et al. (2013) suggested Halophila species invest more energy in rapid clonal propagation, resulting in high turnover rates and Vermaat et al. (1995) indicated that H. ovalis is the fastest growing seagrass species in tropical seagrass beds.

Additionally, Prange and Dennison (2000) reported that *H. ovalis* and *H. spinulosa* concentrated more trace metals than *Zostera capricorni* and *H. uninervis*. Results of the present study concur with this pattern, suggesting that *H. ovalis* is a trace metal accumulator and as a consequence, may be more prone to the toxic effects linked with sequestration of trace metals from the environment that may provide a source of contamination to seagrass consumers (Prange and Dennison, 2000). Ambo-Rappe et al. (2011) observed that the higher concentrations of Cu inhibited the growth and increased leaf asymmetry of *H. ovalis*. The authors noted that the plant actively takes up Cu; however, increased concentrations may exceed metabolic requirements and result in a toxic impact.

This observation suggests that metal element uptake efficiency may be higher for smaller species, which grow faster and therefore have a greater nutrient demand per unit biomass (Duarte et al., 1998). This also gives an indication that different seagrass species are capable of accumulating metals at different rates from the environment as shown by the two Halodule species in this study. The reason for these differences is unclear; however, factors to consider would be the relative metal availability in unvegetated areas for uptake by seagrass plants i.e., from the water column and sediment that can change from site to site, and which also depends on metal supply (Llagostera et al., 2011). Findings of our study are consistent with this statement in showing this to be true for the three seagrass species analyzed. On the other hand, our results also indicate that sediment resources for trace metal elements are likely to be greater in the uppermost substrate layers where belowground organs of H. ovalis have been observed to grow within 2–5 cm depth compared to 5-10 cm depth of the two *Halodule* species (pers. obs.). Moreover, further experimental investigation will help to clarify this phenomenon to increase our understanding of seagrass trace metal accumulation.

Detection of trace metals in the three seagrass species investigated in this study supports previous evidence that seagrasses are able to accumulate trace metals from the marine environment (Prange and Dennison, 2000; Sanz-Lazáro et al., 2012; Govers et al., 2014). Our results show trace metal accumulations within seagrasses appear to be influenced more by the content of biologically available metals within their surroundings, and ensures that seagrasses are reliable sources for metal pollution monitoring in the area. Prange and Dennison (2000) reported that increased rainfall and associated runoff may serve to increase trace metal supply to seagrass habitats, and localized developments can mobilize latent metal sources. Additionally, Waycott et al. (2011) projected that rainfall will increase in the Pacific by 5–20% as a result of climate change, increasing runoff and supply of toxic chemicals such as herbicide diuron from catchments to seagrass meadows.

Management strategies are therefore needed to consider minimizing potential metal pollution. The presence of metals in both water and sediment has been shown to inhibit the growth of seagrass (Ambo-Rappe et al., 2011). The results of this study demonstrate the potential use of seagrass as a benchmark for measuring and monitoring trace metal contamination in the marine environment. The accumulation capacity of the different seagrass species in Fiji clearly indicates trace element

accumulation in seagrass is a species-specific phenomenon and the various seagrass species in Fiji are potential indicators of different trace element concentration in the environment. Likewise, similar trends in content were reported in other seagrasses as discussed above. However, the principal limitation of the present study is that the trends displayed are similar between total biomass and sediment samples collected to measure the elements. Thus, the data does not provide clear indication which tissues (roots, rhizomes and leaf) or species can be used as bio-indicators warranting further investigation at local scale.

Seagrasses are often considered useful metal bioindicators and this assessment is validated in this study by comparing, for the first time, metal concentrations found in *H. ovalis*, *H. pinifolia* and *H. uninervis* with those found in sediments in seagrass vegetated and unvegetated areas. Furthermore, this study showed that *H. ovalis*, *H. pinifolia* and *H. uninervis* might have greater bioaccumulation capacity than previously considered for the four metals studied (Cu, Fe, Mn and Zn). Results suggest that the trace metal contents of seagrass tissues vary with species, endorsing the findings of other studies. Further, our data show the potential before using seagrasses as tools to assess trace metal pollution.

## CRediT authorship contribution statement

Shalini Singh: Conceptualization, Funding acquisition, Data curation, Methodology, Investigation, Formal analysis, Visualization, Writing – original draft, Writing – review & editing. Monal M. Lal: Supervision, Writing – review & editing. Paul C. Southgate: Writing – review & editing. Morgan Wairiu: Supervision, Writing – review & editing. Awnesh Singh: Project administration, Supervision, Writing – review & editing.

#### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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## Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.marpolbul.2021.112745.

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