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Integration of community structure data reveals observable effects below sediment guideline thresholds in a large estuary†

Louis A. Tremblay, *[a,b] Dana Clark,[a] Jim Sinner[a] and Joanne I. Ellis‡[a]

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The sustainable management of estuarine and coastal ecosystems requires robust frameworks due to the presence of multiple physical and chemical stressors. In this study, we assessed whether ecological health decline, based on community structure composition changes along a pollution gradient, occurred at levels below guideline threshold values for copper, zinc and lead. Canonical analysis of principal coordinates (CAP) was used to characterise benthic communities along a metal contamination gradient. The analysis revealed changes in benthic community distribution at levels below the individual guideline values for the three metals. These results suggest that field-based measures of ecological health analysed with multivariate tools can provide additional information to single metal guideline threshold values to monitor large systems exposed to multiple stressors.

Environmental impact

Globally, coastal environments are exposed to a range of stressors that challenge the sustainable management of receiving ecosystems. Environmental managers often rely on single pollutant sediment guidelines to characterise potential risk. This study measured 3 metals and characterised the benthic community distribution across a pollution gradient in a large estuary. The analysis of the data showed that changes in benthic community distribution occurred at levels below guideline threshold values for the metals. The results demonstrate the limitations of using sediment quality guideline thresholds based on single contaminant to protect the health of a large estuarine system subject to multiple stressors. This suggests the need for monitoring frameworks to incorporate site-specific data that integrate biological and chemical endpoints.

Introduction

Anthropogenic habitat modification, pollution and over-exploitation of resources adversely affect global biodiversity and ultimately the provision of ecosystem services.[1] A multi-scale spatial model analysis indicated that all marine ecosystems are impacted by human influence.[2] In particular, human modification of coastal zones and the resulting stressors are causing profound ecological changes on estuaries.[3,4] There is evidence worldwide that estuarine and coastal ecosystems have experienced rapid degradation in the last 150–300 years.[5] The health of aquatic ecosystems depend on the quality of sediments that can act both as a source and sink for contaminants.[6,7]

The increasing levels of anthropogenic contaminants released from urban and rural sources lead to complex challenges for environmental and natural resource managers.[8] To enable timely and effective management responses, degradation needs to be detected at an early stage. Assessing the risks of contamination to healthy estuarine ecosystems has traditionally been conducted in two ways, laboratory-based experiments and field assessments. Laboratory-based approaches offer the advantage of tight control over environmental parameters and exposure to contaminants. However, these studies are often not representative of natural conditions and are generally restricted to measures of individual organism health; therefore, they are limited in their ability to measure effects at the scale of population or community.[9,10] Field-based approaches allow measurements of more relevant ecological responses, but measurement of actual exposure to particular contaminants is more difficult and the inferences taken from these studies can be obscured by covarying natural or environmental stressors.[9,10]

Currently, managers rely primarily on water and sediment chemical guidelines developed from toxicity testing databases to assess whether contaminants pose a risk to species and ecosystems.[11,12] Guideline threshold values have been established for single chemicals based on laboratory studies. These guideline

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[a] *Cawthron Institute, 98 Halifax Street East, Private Bag 2, Nelson, 7010, New Zealand. E-mail: Louis.tremblay@cawthron.org.nz; Tel: +64 3 539 3290*

[b] *School of Biological Sciences, University of Auckland, PO Box 92019, Auckland, 1142, New Zealand*

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‡ Current address: King Abdullah University of Science and Technology (KAUST), Red Sea Research Center, Division of Biological and Environmental Science and Engineering, Thuwal, 23955-6900, Saudi Arabia.

values represent concentrations below which there is a low probability of biological effects, and above which there is a high probability of effects.[13] Previous findings demonstrated changes in benthic community composition and functioning at levels below sediment quality guidelines.[14,15]

The use of a weight of evidence (WOE) approach for the assessment of potential impacts of contaminated sediments is recommended.[16,17] In addition to 'guideline trigger values' based on chemical measures and ecotoxicology, revised frameworks have been proposed that explicitly allow and recommend the consideration of bioaccumulation and ecological health as additional important lines of evidence. Nonetheless, a recent meta-analysis of contaminants study concluded that although the integration of ecology and ecotoxicology has been proposed for over a decade,[18] few study formally consider the ecological effects of contamination.[9] Specifically, it was noted that there is a strong bias towards laboratory studies that investigate the effects of contaminants on individual species (85% of the total number of studies).[9]

The aim of this study was to assess whether ecological health decline, based on community composition changes along a pollution gradient, is occurring at levels below guideline threshold values for selected metals. The approach was focused on the metals only and did not take into consideration other stressors or the potential nutritional implications of the essential metals. Canonical analysis of principal coordinates (CAP) was used to characterise benthic communities along a gradient that integrated spatial changes in the concentrations of three predominant metals in Tauranga Harbour, a large estuary in New Zealand. The structure of benthic communities is considered a sensitive and accurate indicator of environmental health because it integrates the effects of multiple stressors over time.[19–21] The new Australian and New Zealand Environment and Conservation Council (ANZECC) guidelines specifically recognised the use of multivariate tools to characterise the relationships between ecological communities and co-varying contaminants.

Material and methods

Sampling

Tauranga Harbour is a large estuary (approximately 200 km^2) located on the western edge of the Bay of Plenty on New Zealand's North Island (37° 40′S, 176° 10′E). Catchment land use is predominantly pasture and indigenous forest with considerable urbanisation in the south-east, near the city of Tauranga. A field survey was undertaken in intertidal soft-sediment areas of Tauranga Harbour between December 2011 and February 2012. Seventy-four sites of 100 m^2 were selected to represent a variety of habitats including intertidal sand flats, shellfish beds and seagrass meadows (Fig. 1). All sites were intertidal with relatively similar daily fluctuations in depth, temperature and salinity. The coarse grain size fraction of the sediment was used as a proxy for wave exposure, which may have varied across the Harbour. Initial data exploration confirmed that this variable did not confound identification of effects of metals on community composition.[19]

Lead, copper and zinc are important urban estuarine contaminants[22,23] so were selected as the metals of interest in this study. Other physico-chemical parameters (sediment grain-size, organic matter, nutrients and chlorophyll-α) were also measured for use in a preliminary analysis (details in Statistical analyses section) to identify which additional variables were important in explaining benthic community variation within the Harbour.

Cores of 2 cm diameter extending 2 cm deep into the sediment were collected at each of ten randomly selected positions at each site. The replicates were composited into a single sample and the sediment was analysed for metals (lead, copper and zinc), grain size, organic matter (loss on ignition), nutrients (total nitrogen and total phosphorus) and chlorophyll-α. Samples for metal analyses were dried at 30 °C then digested using a combination of nitric and hydrochloric acids, with heating to 95 °C for 30 minutes. Metals were analysed by R. J. Hill Laboratories Ltd (Hamilton, New Zealand) using Inductively Coupled Plasma Mass Spectrometry.[24] Ellis *et al.*[25] provided detailed methodology for the analysis of the other physico-chemical endpoints.

Macrofauna were sampled by collecting cores of 13 cm diameter extending 15 cm into the sediment from each of three randomly selected positions at each site. Core samples were sieved on a 0.5 mm mesh and macrofauna retained on the sieves were preserved in a 70% ethanol solution with seawater. Macrofauna were sorted and identified to the lowest practicable taxonomic resolution and community composition (*i.e.* number and type of taxa and their relative abundances) was used as the response variable in the following statistical analyses.

Statistical analyses

Data were transformed in some cases to meet the test requirements, limit the effect of outliers, and reduce the weight of dominant taxa. Optimal transformations were performed on the data, if necessary preliminary analyses were conducted by multivariate linear regression to select variables that explain the maximum variation in the community data cloud using Distance-based Linear Modelling (DistLM[26]). DistLM was analysed using square-root transformed Bray–Curtis similarities with a backward selection procedure based on the AIC selection criteria. Variables included in the analysis were percentage mud (<63 μm), organic matter, total nitrogen, total phosphorus, lead, copper, zinc and chlorophyll-α. This analysis indicated that copper and lead were important in explaining the variation in benthic community distribution in the Harbour.

Copper and lead were correlated with zinc so a principal component analysis (PCA) was used to generate a single variable that would characterise an overall gradient corresponding to increases in the concentrations of all metals in the field. The PCA was performed on the basis of log-transformed metal concentrations (copper, lead, and zinc) using the PRIMER (version 6.1.13) computer program,[27] where the PC1 axis explained 85.5% of the variance (PC1 contamination). This axis was subsequently used as the contaminant gradient in the multivariate analyses.

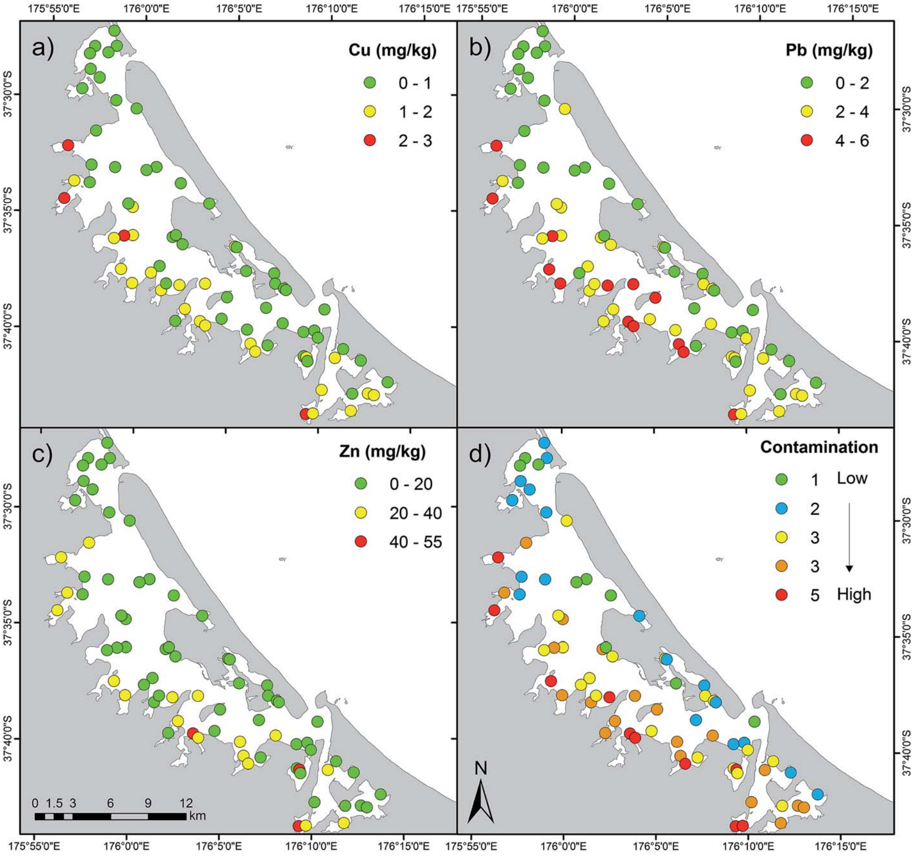

Fig. 1 Sediment metal concentrations (mg kg^{-1}) for 74 sites in Tauranga Harbour, (a) copper, (b) lead, (c) zinc and (d) corresponding contamination groupings as determined using canonical analysis of principal coordinates (CAP) relating benthic community data to a metal contamination gradient based on copper, lead and zinc.

DistLM was run seven times to obtain the percentage explained (R^2) by this contamination gradient. The relative percentages explained by different components were then determined by variance partitioning.[28,29] A canonical analysis of principal coordinates, or CAP,[30,31] based on Bray–Curtis similarities,[32] was applied to characterise how the soft sediment macrofaunal communities of the estuary change along the contamination gradient. All CAP analyses were performed using specialised software, written in FORTRAN and available as an executable file (CAP.exe) or in PRIMER (version 6.1.13) and PERMANOVA (version 1.0.3) with permission from M. Anderson, University of Auckland.

The model output was used to classify sites along the metal contamination gradient from 'low contaminants' to 'high contaminants' to indicate strong changes in community composition. *K*-Means partitioning[33] of the PC1 contamination axis was used to identify possible groupings along the contamination gradient (contamination groups). The optimal number of sites occurring in each group was determined using the Calinski–Harabasz criterion.[34] It is important to note that references to 'low contaminants' and 'high contaminants' are relative to the sampling sites. For example, a site ranked as having 'high contaminants' would have high metal concentrations relative to other sites within Tauranga Harbour, but compared to other locations, it might not be considered highly impacted. The taxa that contributed to differences in community composition along the contamination gradient were identified using SIMPER in PRIMER (version 6.1.12). Values were calculated on raw data rather than transformed data, as changes in the relative abundance of dominant taxa were considered to be important in capturing changes in communities associated with contaminants.

Comparison with existing sediment quality guidelines

Existing sediment quality guideline values were converted to determine their position along the contamination gradient and facilitate comparison with multivariate benthic community data.[14] The contamination gradient (PC1 contamination) is a linear combination of copper, zinc and lead concentrations, so new samples can be positioned along the axes, provided the concentrations of these metals have been measured. For principal component analysis (PCA), the eigenvector weights provide coefficients for a linear combination of the original variables that will yield the principal component scores. The following equation was used to determine the position of existing sediment quality guidelines along the PC1 contamination axis:

$$\text{PC1 contamination} = 0.323(X_{\text{Cu}}) + 0.546(X_{\text{Pb}}) + 0.773(X_{\text{Zn}})$$

where X equals the log concentration of that metal (copper, lead or zinc) in the sample minus the mean log concentration of that metal across the full set of 74 samples (mg kg^{-1} dry weight). The mean log concentrations used were -0.27899 for copper, 0.72093 for lead and 2.681024 for zinc.

Results

Metal concentrations in Tauranga Harbour

Metal concentrations in the Harbour tended to be higher in inner areas compared with outer sites (Fig. 1). The Uretara Estuary site had the highest copper and lead concentrations (3.0 and 5.6 mg kg^{-1}, respectively) while zinc concentrations were highest at the outer Te Puna Estuary site (55 mg kg^{-1}).

Community changes

Total abundance ranged from 28 to 333 organisms per core and averaged 117. One hundred and twenty-nine taxa were found in the Harbour with the number of taxa per site ranging from 10 to 39. Sixty-two percent of taxa were identified to species level, another 10% were identified to genus and the remaining 28% were identified to family or higher. The most numerically dominant organisms were, in order of decreasing abundance, *Heteromastus filiformis*, Corophiidae, *Prionospio aucklandica*, Phoxocephalidae, *Austrovenus stutchburyi*, *Linucula hartvigiana*, Oligochaetes, *Aonides trifida* and Nereidae.

Variance partitioning methods showed that metal contamination alone explained 7.5% of the observed variation in benthic communities, explaining more variation than the other two key variables identified, percentage mud (4.8% variation explained) and nutrients (PCA of total nitrogen, total phosphorous and chlorophyll-α, 2.7% variation explained). Data for all variables are summarised in Table S2.† The CAP analysis related benthic community taxa to the contamination gradient (PC1 contamination axis) generated from the concentrations of lead, copper and zinc at each site. A strong gradient of community change was observed in response to metal concentrations in the sediment ($R^2 = 0.71$) suggesting that CAP can be used to characterise aspects of how benthic communities change with increasing level of those metals.

K-Means partitioning identified five groups along the contamination gradient (Fig. 1). Metal concentrations increased with increasing group number (*i.e.* increasing metal contamination; Table 1). These groups differed from one another in community composition by more than 60%, on average. In particular, the communities found in the least contaminated group (group 1) were, on average, 71% dissimilar from those in the most contaminated group (group 5; see Table 2 for taxa contributing most to the differences between groups). Most of the sites were assessed to be moderately impacted by metals; *i.e.* the CAP placed them in group 2 (24% of sites), group 3 (24% of sites) or group 4 (26% of sites). Group 5 sites (highest metal contamination) tended to be situated in inner Harbour areas while sites in groups 1 and 2 were located further out (Fig. 1). Two univariate community measures were assessed; abundance and species richness. No significant differences in average abundance were found between the five groups. Significant differences ($p < 0.01$) in average species richness were only found between group 5 and groups 1, 2 and 3 and between group 2 and 4.

Table 1 Tauranga Harbour contamination groups and associated mean metal concentrations (mg kg^{-1} ± standard error)

Group	n	Copper	Lead	Zinc
1	9	<1 ± 0	0.8 ± 0.1	6.0 ± 0.5
2	18	<1 ± 0	1.4 ± 0.1	9.9 ± 0.3
3	18	0.7 ± 0.1	2.3 ± 0.1	14.6 ± 0.5
4	19	1.1 ± 0.1	3.3 ± 0.2	21.2 ± 0.6
5	10	1.8 ± 0.2	4.3 ± 0.3	36.4 ± 3.0

Comparison with other sediment quality guideline values

Metal concentrations in Tauranga Harbour were well below existing sediment quality guideline values. The most conservative guideline values (MacDonald's threshold effects level[11]) were more than double the highest recorded value for zinc, and five to six times the highest values for lead and copper (Table 1). For each set of guidelines, the combined position of the three metals values was located on the PC1 contamination axis to compare with the five contamination groups, which represent

Table 2 Taxa contributing most to the differences between group 1 (least contaminated) and group 5 (most contaminated) together with their average abundances per core and key information

Taxa	Group 1	Group 5	Key information
Increased			
Corophiidae	0.1	37.3	Medium-sized burrowing amphipod, suspension or deposit feeder, freely motile
Heteromastus filiformis	2.0	24.0	Small capitellid polychaete, deposit feeder, limited motility
Oligochaetes	4.6	9.6	Small predator/scavengers, freely mobile
Decreased			
Prionospio aucklandica	11.5	9.9	Small spionidae polychaete, deposit feeder, freely motile
Phoxocephalidae	8.9	3.6	Amphipod, deposit feeders/scavengers/predators, freely motile
Anthopleura aureoradiata	10.3	2.2	Anemone, suspension feeder, commonly living on *Austrovenus* shells
Austrovenus stutchburyi	8.9	2.0	Bivalve, suspension feeder, freely motile

Table 3 Existing sediment quality guidelines from various sources, along with their positions on the PC1 contamination axis. ISQGs are the interim sediment quality guidelines. Values for metals are in mg kg^{-1} dry weight

Source	Guideline	Zn	Cu	Pb	PC1 contamination
ANZECC[12][a]	ISQG-low	200	65	50	5.204
ANZECC[12]	ISQG-high	410	270	220	7.028
Long *et al.*[35]	Effects-range low (ER-L)	150	34	46.7	4.735
Long *et al.*[35]	Effect-range median (ER-M)	410	270	218	7.023
MacDonald[11]	Threshold effects level (TEL)	124	18.7	30.2	4.157
MacDonald[11]	Probable effects level (PEL)	271	108	112	6.043

[a] The Australian and New Zealand Environment and Conservation Council.

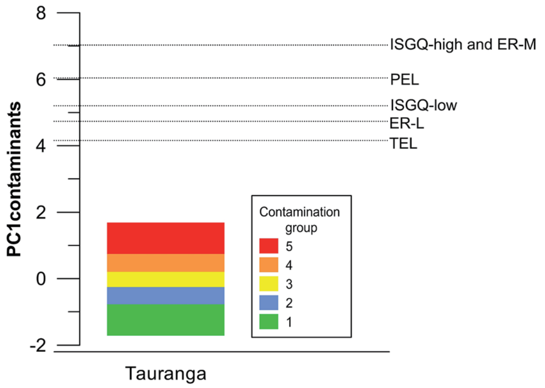

Fig. 2 Values for the five contamination groups derived in the present study along the contamination gradient (PC1 contamination). PC1 contamination from the sediment quality guidelines from Table 3 are shown along the axis.

a range of sediment contaminant loadings and impacts (Table 3; Fig. 2).

Discussion

This study used multivariate methods (CAP) to quantify ecological health and demonstrated changes in benthic community composition were highly correlated with three metals in the field. The five contamination groups defined in this study showed impacts at levels well below current ANZECC and international guidelines for the three metals. Clear differences in community composition were observed between each of the partitioned groups and the adjacent groups along the contamination gradient. Taxa contributing the most to differences between the least and most impacted sites were Corophiidae amphipods, *Heteromastus filiformis* polychaete worms and Oligochaetes worms (increased abundance with increased impact) and the polychaete *Prionospio aucklandica*, Phoxocephalidae amphipods, the cockle *Austrovenus stutchburyi* and the anemone *Anthopleura radiata* (decreased abundance with increased impact).

Evidence from the literature generally supports the differing sensitivities of these taxa. *H. filiformis* and Oligochaetes were assigned to relatively tolerant eco-groups (IV and V) by the AZTI Marine Biotic Index[36] and *H. filiformis* abundance increased in response to metal contamination in a study in Auckland.[14] However, unlike the current study, Hewitt *et al.* found that abundance of Corophiidae amphipods declined with increasing metal contamination.[14] The Corophiidae family covers a wide range of species that may differ in terms of their function and sensitivities to environmental factors,[36] so responses to metal contamination may vary depending on which species are present.

We identified *Austrovenus stutchburyi* and *Anthopleura aureoradiata* as sensitive to metal contamination as previously reported.[14] The abundance of these species was negatively correlated with sediment copper concentrations in another field study.[37] The anemone *Anthopleura radiata* is often found attached to cockle shells so may act as a proxy for *A. stutchburyi* rather than being sensitive to contaminants itself. De Luca-Abbott[38] also showed that sediments contaminated with lead, zinc and polycyclic aromatic hydrocarbons (PAHs) had sublethal effects on *A. stutchburyi* in the field. A manipulative field experiment found declines in the abundance of *P. aucklandica* in treatments spiked with copper, zinc or a mixture of copper, zinc and lead but no change in abundances were found for *A. stutchburyi* or Oligochaetes.[39] As with Corophiidae amphipods, Phoxocephalidae amphipods encompass a broad range of genera. However, 95% of the genera for which eco-groups were available (approximately 25%) were assigned to sensitive eco-groups (I & II; ref. 36). The decline in Phoxocephalidae is consistent with other studies reporting reduced abundances of amphipods in sediments contaminated with metals.[40–42] Laboratory-based bioassays have also shown the sensitivity of amphipods to copper and zinc.[43,44] However, amphipods are a broad group with potentially differing functions and sensitivities, therefore, comparison at this level may not be valid.

These results suggest that field-based measures of ecological health analysed with multivariate tools can provide additional information to single metal guideline threshold values to monitor large systems exposed to multiple stressors. This approach provides an additional line of evidence, as recommended in the revised ANZECC guidelines, which recommended use of additional lines of evidence based on 'ecological health' in addition to 'trigger guideline' values.[45] The objective of an ecological assessment in a weight of evidence (WOE) assessment is to obtain information that can help ascertain whether the ecology of a location has been negatively or extensively impacted.[17] In this study, the multivariate analysis of chemistry and benthic community data provided additional information that can be used in a WOE risk assessment process to characterise the health of a complex environment.[16]

Our results demonstrated the improved ability of multivariate methods to detect changes in community composition compared with univariate measures such as taxa abundance, richness and biotic indices, as reported previously.[46,47] Simple univariate measures do not differentiate amongst different types of taxa, which limits their ability to detect changes in community composition across different sites.[19,48] Accordingly, the multivariate constrained ordination method used in this study was able to detect community shifts in response to metals whereas the univariate methods were unable to detect differences in taxa abundance across the five contamination groups and only detected differences between the most and least impacted sites for species richness. The ability of univariate measures to only differentiate between the most and least disturbed sites, but not smaller relative differences, has been reported elsewhere[19,25] and we suggest that multivariate methods are more appropriate for future studies that aim to determine differences in community in response to stressors.

Our study moved beyond single stressor effects to examine the combined effects of three important estuarine contaminants: lead, copper and zinc. Research has shown that synergistic and antagonistic interactions are more common in nature than simple additive interactions,[49] creating uncertainty in the prediction of contaminant effects and ecological resilience.[50,51] A manipulative field study with copper, lead and zinc showed differences between communities subjected to mixed metal treatments and those with individual metal treatments, suggesting cumulative effects.[39] The potential interactions arising from the effects of these metals were accounted for in the analysis by modelling the community response to all three metals concurrently. The effects of other stressors were excluded by conducting a constrained ordination that assessed benthic variation only in response to the metal stressor data.

While the analyses revealed that contamination explained a relatively low amount of total variation, the ecological significance of weak relationships is increasingly recognised.[52,53] For instance, a study in a rocky intertidal zone demonstrated that some 'weak' average interaction strength may nonetheless be important by magnifying spatiotemporal variation in community structure.[54] A similar conclusion was reached in another New Zealand study using a similar approach to assess the health of a major estuarine system in Auckland.[14] Hewitt *et al.*[14] also observed strong fauna composition changes along a contaminant gradient of copper, lead and zinc, with communities near the ends of the gradient exhibiting 90% dissimilarity in taxa composition. Again, these changes to community composition occurred below the ANZECC ISQG-low guideline levels.[14] The ability to detect ecosystem health changes at lower metal concentrations may be a result of: (i) differences between field and laboratory test data, and/or (ii) differences in guidelines based on single contaminants in relation to multiple stressors situations.

There are limitations to the use of benthic community changes to guide environmental management. Field-based approaches can be resource intensive, time consuming and are not designed to provide early warning signals of impact or cause and effect relationships. The results of this study are based on correlative patterns and it is not always possible to identify all factors like organic pollutants that could influence community structure. We disregarded organic pollutants after 97% of 325 compounds (pesticides, PAHs, phenols and phthalates) measured at eight of the sites were below detection limits or at concentrations below their guideline levels.[55] Further manipulative experiments would be required to prove causality and derive site-specific guidelines for key stressors. However, our results show that laboratory-derived metal guidelines may not always protect biological communities from observable effects. This may be of particular concern in the presence of multiple stressors in the environment (ref. 14 and 37; and this study). We concur with O'Brien and Keough's[9] recommendation that to assess the risks of contamination on ecosystems, there is a need for studies to fill the knowledge gaps on ecological effects at all levels of biological organisation. Monitoring frameworks integrating chemical and biological endpoints (*e.g.* biomarkers in receptor species) along temporal and spatial gradients are needed to identify the cause(s) (stressors) responsible for the effects on benthic fauna.[56] The biomonitoring framework must carefully consider the selection of biota indicators and ecosystem-specific stressors.[57] The resulting knowledge should be easy to interpret and meaningful to risk assessors and managers and policy makers.[58]

The ordination model showed that copper, lead and zinc were highly correlated to the observable effects on benthic communities. This suggests ecological health declines may occur at metal concentrations below guideline values. The results from this study demonstrate the benefits of field-based ecological knowledge to complement guideline values to assess the health of large estuarine systems subjected to multiple stressors as previously observed.[59]

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