Are the existing guideline values adequate to protect soil health from inorganic mercury contamination?

Khandaker Rayhan Mahbub, School of Life Sciences, University of Technology Sydney, NSW 2007, Australia, email: khandaker.mahbub@uts.edu.au, krmjissan@gmail.com

Md Mezbaul Bahar, Global Center for Environmental Remediation, Research and Innovation Division, The University of Newcastle, Callaghan, NSW 2308, Australia, email: mezbaul.bahar@newcastle.edu.au

Mallavarapu Megharaj, Global Center for Environmental Remediation, Research and Innovation Division, The University of Newcastle, Callaghan, NSW 2308, Australia, email: megh.mallavarapu@newcastle.edu.au

Maurizio Labbate, School of Life Sciences, University of Technology Sydney, NSW 2007, Australia, email: maurizio.labbate@uts.edu.au

Author for correspondence:
Khandaker Rayhan Mahbub
School of Life Sciences, University of Technology Sydney
Broadway, NSW 2007, Australia
Email: Khandaker.Mahbub@uts.edu.au; krmjissan@gmail.com

Abstract

Currently, data that guide safe concentration ranges for inorganic mercury in the soil are lacking and subsequently, threaten soil health. In the present study, a species sensitivity distribution (SSD) approach was applied to estimate critical mercury concentration that has little (HC_5) or no effect (PNEC) on soil biota. Recently published terrestrial toxicity data were incorporated in the approach. Considering total mercury content in soils, the estimated HC_5 was 0.6 mg/kg, and the PNEC was 0.12 – 0.6 mg/kg. Whereas, when only water-soluble mercury fractions were considered, these values were 0.04 mg/kg and 0.008 – 0.04 mg/kg, respectively.

Key words: SSD; HC_5; ecotoxicity; bioavailability; safe limits.

Highlights

- Data on terrestrial Hg toxicity are insufficient
- SSD approach was employed to estimate safe concentrations of Hg in soil
- Low levels of Hg could affect terrestrial biota
- Soluble fractions of Hg should be considered to estimate safe Hg limits

1. Introduction

Mercury (Hg) is a heavy metal that is widespread in the biosphere but has no known biological functions, rather it exerts toxicity on living organisms. Soil is one of the most important environments where Hg undergoes numerous chemical and biological reactions, and at certain concentrations disrupts soil health by altering soil biota such as microbes, plants, and animals (Ha et al. 2017; Rice et al. 2014). These bio-geo-chemical changes determine the degree of toxicity that different forms of Hg have toward organisms in different trophic levels (Schaefer 2016). The metallic form of mercury (Hg^0) is the least toxic form because it is not water soluble, and does not bind to animal tissues and are not readily taken up by lower animals or microbes. Hg^0 can be oxidised in the atmosphere to inorganic mercury (Hg^{2+}) which is found in different salt forms such as chloride, nitrate or sulfide. Hg^{2+} is a reactive form that has high affinity to animal/plant tissues and can be taken up by micro- and macro-organisms resulting in many physical and biochemical adversities in the affected biota. Moreover, Hg^{2+} can serve as a substrate for bacterial methylation under anaerobic conditions, such as in sediments and water-logged soils (Mahbub et al. 2017a). The bioaccumulated Hg (after methylation) can enter into the food chain through intoxicated plants or animals, leading to severe acute and chronic disease in humans. Abnormalities in nervous, renal, cardiovascular and reproductive systems were found linked to Hg exposure (Kim et al. 2016; Yassa 2014).

As the divalent and methylated forms of Hg are highly toxic, many industrial countries have developed regulatory limits or guideline values to control the use of Hg in agricultural and industrial practices. The estimation of a critical concentration of Hg in soil above which biological activity may be affected is important, as it constitutes a safe concentration or regulatory limit. Because of the severity of health problems from Hg pollution in waters, most of these regulatory limits are developed for aquatic environments. As such, a large number of studies have been carried out to estimate Hg toxicity in different water environments (Lavoie et al. 2013; Rodrigues et al. 2013). However, soils have not received much attention even though large portions of emitted Hg undergoes various changes in terrestrial environments.

The average contents of mercury in soils range from 0.001 – 1.5 mg/kg, which is related to the soil's property and proximity to an emission site (Kabata-Pendias and Szteke 2015). However, high levels of soil-bound Hg in areas adjacent to the contamination sources have been identified in several studies. In China, 15-119 mg/kg Hg^{2+} was estimated close to a smelting area (Søvik 2008). In different countries in Europe, contaminated soils were reported to contain 5-778 mg/kg inorganic Hg (Moreno-Jiménez et al. 2006). In agricultural soils, Hg concentrations have been reported from background level to approximately 180 mg/kg (Li et al. 2013; Meng et al. 2014; Şenilă et al. 2012). Soil-bound inorganic Hg can linearly accumulate and magnify in important plants such as rice (Li et al. 2013; Meng et al. 2014) which is a staple food in many countries. To protect soils as well as human healt from soil bound Hg, industrial countries like Canada, America, UK, Netherland, Germany and Australia have developed guidlenies for Hg use in residential, agricultural and recreational soils (Mahbub et al. 2017a; Tipping et al. 2010). However, the suggested safe soil Hg limits from different countries lack robustness because of inadequate toxicity data from soil environments; most of the studies being done on observing merely toxic effects, rather than estimating critical doses causing the effects from proper dose-response analyse (Mahbub et al. 2017a).

From our several recent investigations, it has been observed that the degree of toxicity of Hg depends on the biological species inhabited in soils and the soil's physicochemical properties. For instance, soil-bound Hg is highly toxic to soil microorganisms (Mahbub et al. 2016a; Mahbub et al. 2016b) but less toxic to soil invertebrates (Mahbub et al. 2017c) and plants (Mahbub et al. 2017b). Toxic doses also varied depending on varying end points. For instance, a dose required to observe negative effect on earthworm's reproduction rate is different from the toxic dose on their mortality or weight loss (Lock and Janssen 2001). In addition, soil properties such as organic carbon content, pH, and cation exchange capacity play significant roles in bioavailability of Hg in the soil which is directly related to the degree of toxicity (Kim et al. 2016). As significant variation in the toxic doses of Hg in soil has been previously observed, this study was undertaken to consolidate recent toxicity data in the literature with a view to estimating a safe concentration that can be used to protect the majority of biota in the terrestrial habitat.

In the present study, a species sensitivity distribution (SSD) approach was applied to obtain critical Hg concentrations in soil that when exceeded, leads to toxicity. SSD is the recommended approach for ecological risk assessment and is used to predict hazardous concentrations (HC) that may affect a certain percentage of species in a biota, using extrapolation of ecotoxicity data from published literature or databases (Posthuma et al. 2001; US-EPA 2005). This approach has recently been used by others to estimate critical Hg concentrations in water (Rodrigues et al. 2013). Generally, the SSD approach utilized to determine the HC_5 value, which denotes the concentration that affects 5% of the species in an environment. Alternatively, this concentration protects 95% of species. In this study, both total and water-soluble Hg concentrations were considered for the estimation of HC_5. Moreover, the predicted no-effect concentration (PNEC) had been estimated from the same approach. The HC_5 and PNEC values generated in the present study will advance the knowledge of Hg toxicity in terrestrial environments.

2. Materials and methods

2.1. Data collection

Toxicity data were collected from the existing published papers by a literature search using Scopus and Web of Science. Papers from last 20 years (1997 – 2017) were selected, based on data generated from experiments carried out in soil under laboratory conditions. Organisms from three trophic levels – microbes, invertebrates, and plants were chosen which have direct contact with soil. Statistically determined EC_{50} values were considered only when a proper dose-response relation was evident. In contrast, any data failing to demonstrate regression relation (i.e., merely a concentration that has a negative effect on any endpoint) were excluded from this study. As such for soil microbes, data were available for a range of soil enzymatic activities and soil microbial alpha diversity; for soil invertebrates, mortality rate, reproduction inhibition rate, and avoidance rate were available; for plants, only root elongation data were obtained. Based on the literature search, information from the twelve papers that met the above-mentioned criteria were selected for the present study (Table 1). EC_{50} values were either reported in the selected papers or generated from the available data using four parametric logistic model applying IBM SPSS version 17.

2.2.Estimation of critical Hg concentration

The toxicity data were subjected to SSD analysis using the SSD generator downloaded from https://www3.epa.gov/caddis/da_software_ssdmacro.html and HC_5 was determined. The predicted no-effect concentration (PNEC) was estimated by dividing the estimated HC_5 by a factor 1-5 (Rodrigues et al. 2013).

Table 1: Toxicity data of Hg^{2+} for soil micro and macro organisms

Endpoint	EC_{50} (THg mg/kg)	EC_{50} (WHg mg/kg)	Number of soils used and thier properties	Aging period/ site	References
1. Soil microbial activity					
DHA	2.0	NR	One soil, pH 7, Sand 9%, Silt 75%, Clay 15%, TOC 1.12	Laboratory exposure	(Welp 1999)
DHA	13.2	0.05	One soil, pH 7.6, TOC 2%, sand 50%, silt 35% and clay 13%	90 d laboratory exposure	(Mahbub et al. 2016a)
DHA	2.4	0.29	One soil, pH 8.5, TOC 2.2%, sand 42%, silt 44% and clay 13%	90 d laboratory exposure	(Mahbub et al. 2016a)
Nitrification	88	0.27	One soil, pH 7.6, TOC 2%, sand 50%, silt 35% and clay 13%	90 d laboratory exposure	(Mahbub et al. 2016a)
Nitrification	0.7	0.02	One soil, pH 8.5, TOC 2.2%, sand 42%, silt 44% and clay 13%	90 d laboratory exposure	(Mahbub et al. 2016a)
Nitrification	22.6	NR	One soil, pH 7.8, TOC 2.28%	28 d laboratory exposure	(Zhou et al. 2015)
Nitrification	1.59	NR	One soil, pH 7.14, TOC 2.05%	7 d laboratory exposure	(Liu et al. 2010b)
Urease	88	NR	One soil, pH 5.94, OM 26.4 mg/kg, Sand 54%, Silt 30%, Clay 16%	NR	(Yang et al. 2007)
Urease	5.5	NR	One soil, pH 6.19, OM 20.7 mg/kg, Sand 62%, Silt 20%, Clay 18%	NR	(Yang et al. 2007)

	Urease	24	NR	One soil, pH 6.26, OM 31.6 mg/kg, Sand 29%, Silt 38%, Clay 32%	NR	(Yang et al. 2007)
	Urease	20	NR	One soil, pH 6.71, OM 29.4 mg/kg, Sand 22%, Silt 42%, Clay 36%	NR	(Yang et al. 2007)
	Arylsulphatase	0.78*	NR	Three soils, pH 7.2-8.3, OM 1.7-16.8%, Sand 3.9-74%, Silt 16-52.1%, Clay 10-48%	30 d laboratory exposure	(Casucci et al. 2003)
	Arylsulphatase	5.3*	NR			(Casucci et al. 2003)
	Microbial biomass carbon	0.8*	NR			(Casucci et al. 2003)
	Microbial biomass carbon	1.4*	NR			(Casucci et al. 2003)
	Alkaline phosphatase	1.4*	NR			(Casucci et al. 2003)
	Fe (III) reduction	56	NR	Eighteen soils, pH 3.5-7.8, TOC 0.9-11.4%, clay 2-41%	Median EC_{50} of field soils	(Welp and Brümmer 1997)
2. Soil microbial diversity						
	Alpha diversity	25	0.18*	One soil, pH 7.6, TOC 2%, sand 50%, silt 35% and clay 13%	90 d laboratory exposure	(Mahbub et al. 2016b)
	Alpha diversity	57	0.58*	One soil, pH 8.5, TOC 2.2%, sand 42%, silt 44% and clay 13%	90 d laboratory exposure	(Mahbub et al. 2016b)
3. Earth worm's mortality, reproduction and behaviour						
Eisenia fetida	Mortality	152	0.8*	One soil, pH 7.6, TOC 2%, sand 50%, silt 35% and clay 13%	28 d laboratory exposure	(Mahbub et al. 2017c)
	Mortality	294	1.2*	One soil, pH 8.5, TOC 2.2%, sand 42%, silt 44% and clay 13%	28 d laboratory exposure	(Mahbub et al. 2017c)
	Mortality	367	0.8*	One soil, pH 4.2, TOC 2.2%, sand 89%, silt 9% and clay 2%	28 d laboratory exposure	(Mahbub et al. 2017c)
	Reproduction	9.16	NR	One soil, pH 6, TOC 10%	21 d laboratory exposure	(Lock and Janssen 2001)

Enchytraeus albidus	Reproduction	22	NR	One soil, pH 6, TOC 10%	42 d laboratory exposure	(Lock and Janssen 2001)
Folsomia candida	Reproduction	3.26	NR	One soil, pH 6, TOC 10%	28 d laboratory exposure	(Lock and Janssen 2001)
Eisenia andrei	Avoidance	128.3	NR	One soil, pH 6, TOC 10%, Sand 70%, Clay 20%	2 d laboratory exposure	(Buch et al. 2017a)
Eisenia andrei	Avoidance	206.2	NR	One soil, pH 4, OM 24 g/kg, Sand 50%, Silt 15% Clay 35%	2 d laboratory exposure	(Buch et al. 2017a)
Eisenia andrei	Avoidance	168.2	NR	One soil, pH 4, OM 26 g/kg, Sand 53%, Silt 15% Clay 32%	2 d laboratory exposure	(Buch et al. 2017a)
Pontoscolex corethurus	Avoidance	266	NR	One soil, p H 6, organic carbon 10%, Sand 70%, Clay 20%	2 d laboratory exposure	(Buch et al. 2017a)
P. corethurus	Avoidance	300	NR	One soil, p H 6, organic carbon 10%, Sand 70%, Clay 20%	2 d laboratory exposure	(Buch et al. 2017a)
P. corethurus	Avoidance	295	NR	One soil, pH 4, OM 24 g/kg, Sand 50%, Silt 15% Clay 35%	2 d laboratory exposure	(Buch et al. 2017a)
Eisenia andrei	Mortality	153	NR	One soil, pH 6, TOC 10%, Sand 70%, Clay 20%	14 d laboratory exposure	(Buch et al. 2017a)
Eisenia andrei	Mortality	113	NR	One soil, pH 4, OM 24 g/kg, Sand 50%, Silt 15% Clay 35%	14 d laboratory exposure	(Buch et al. 2017a)
Eisenia andrei	Mortality	110	NR	One soil, pH 4, OM 26 g/kg, Sand 53%, Silt 15% Clay 32%	14 d laboratory exposure	(Buch et al. 2017a)
P. corethurus	Mortality	203	NR	One soil, p H 6, organic carbon 10%, Sand 70%, Clay 20%	14 d laboratory exposure	(Buch et al. 2017a)
P. corethurus	Mortality	194	NR	One soil, p H 6, organic carbon 10%, Sand 70%, Clay 20%	14 d laboratory exposure	(Buch et al. 2017a)
P. corethurus	Mortality	220	NR	One soil, pH 4, OM 24 g/kg, Sand 50%, Silt 15% Clay 35%	14 d laboratory exposure	(Buch et al. 2017a)
Eisenia andrei	Reproduction	10	NR	One soil, pH 6, TOC 10%, Sand 70%, Clay 20%	91 d laboratory exposure	(Buch et al. 2017a)
Eisenia andrei	Reproduction	7	NR	One soil, pH 4, OM 24 g/kg, Sand 50%, Silt 15% Clay 35%	91 d laboratory exposure	(Buch et al. 2017a)

Eisenia andrei	Reproduction	7	NR	One soil, pH 4, OM 26 g/kg, Sand 53%, Silt 15% Clay 32%	91 d laboratory exposure	(Buch et al. 2017a)
P. corethurus	Reproduction	11	NR	One soil, p H 6, organic carbon 10%, Sand 70%, Clay 20%	91 d laboratory exposure	(Buch et al. 2017a)
P. corethurus	Reproduction	12	NR	One soil, p H 6, organic carbon 10%, Sand 70%, Clay 20%	91 d laboratory exposure	(Buch et al. 2017a)
P. corethurus	Reproduction	13	NR	One soil, pH 4, OM 24 g/kg, Sand 50%, Silt 15% Clay 35%	91 d laboratory exposure	(Buch et al. 2017a)
			4. Plant's growth			
Iseilema membranaceum (Barcoo)	Root growth	200	1.4	One soil, pH 7.6, TOC 2%, sand 50%, silt 35% and clay 13%	28 d laboratory exposure	(Mahbub et al. 2017b)
	Root growth	10	0.41	One soil, pH 8.5, TOC 2.2%, sand 42%, silt 44% and clay 13%		
	Root growth	224	1.9	One soil, pH 4.2, TOC 2.2%, sand 89%, silt 9% and clay 2%		
Dichanthium sericeum (Qld blue)	Root growth	126	1.32	One soil, pH 7.6, TOC 2%, sand 50%, silt 35% and clay 13%	28 d laboratory exposure	(Mahbub et al. 2017b)
	Root growth	123	0.82	One soil, pH 8.5, TOC 2.2%, sand 42%, silt 44% and clay 13%		
	Root growth	ND	1.9	One soil, pH 4.2, TOC 2.2%, sand 89%, silt 9% and clay 2%		
Sporobolus africanus (Tussock)	Root growth	209	1	One soil, pH 7.6, TOC 2%, sand 50%, silt 35% and clay 13%	28 d laboratory exposure	(Mahbub et al. 2017b)
	Root growth	132	0.82	One soil, pH 8.5, TOC 2.2%, sand 42%, silt 44% and clay 13%		
	Root growth	ND	0.15	One soil, pH 4.2, TOC 2.2%, sand 89%, silt 9% and clay 2%		

*estimated from available data of 30 days study; THg – total mercury; WHg – water soluble Hg; TOC – total organic carbon; OM – organic matter, NR- neither $WHgEC_{50}$ nor WHg was reported; ND- not detected/statistically not significant. A total of 34 different soils with varying physicochemical properties were observed.

3. Results and discussion

Different species of plants, animals, and microbes have been used as indicator organisms in long term and short term exposure experiments to estimate the toxicity of Hg in soil environments, but not as extensively as the toxicological assessments in water environments. Plants are higher organisms, and their uptake rate of Hg through their root system is very low because of the presence of barriers in the root tips (Patra and Sharma 2000). Plants also accumulate elemental Hg from the atmosphere through the leaves which is then translocated to other organs. At certain concentrations, Hg^{2+} is reported to exert oxidative stress (Israr et al. 2006; Tamás and Zelinová 2017), disrupt membrane structure (Ma 1998), damage DNA (Dogan-Topal et al. 2018), reduce the uptake of minerals and nutrients (Tangahu et al. 2011), interfere cell division (Azevedo et al. 2018) and disrupt chlorophyl synthesis (Liu et al. 2010a), photosynthesis and transpiration rates (Rai et al. 2016). Although a lot is known about toxic effects of Hg on plants, there is a scarcity of data where a proper dose-response relationship was reported for terrestrial plants to predict a safe Hg limit. Only one study is available where three Australian native plants namely *Iseilema membranaceum* (Barcoo), *Dichanthium sericeum* (Qld blue) and *Sporobolus africanus* (Tussock) were used in a 28 d laboratory experiment in three soils of different physicochemical properties (Mahbub et al. 2017b). The other studies report only Hg uptake and toxicity related syndromes in different plant parts harvested in contaminated fields (Azevedo and Rodriguez 2012; Mahbub et al. 2017b; Nagajyoti et al. 2010).

Unlike plants, invertebrate animals in soils have been used more elaborately as indicator organisms to estimate safe Hg limits in the soil. At toxic concentrations, Hg can cause death, weight loss, lead to behavioural abnormalities, and interfere with reproduction rates in different species of terrestrial invertebrates. There are few studies (Table 1) where a proper dose-response relationship was established to estimate a Hg concentration that affects any of the endpoints. Most of these studies used different species of earthworms as they are considered a reliable bioindicator of soil pollution. The issue here is, the estimated toxic concentrations of Hg can vary depending on the species used and the endpoints observed (Buch et al. 2017b). Therefore there is a need to combine data obtained from different species of organisms where several endpoints are observed. To include soil invertebrates in the present study, data were obtained from studies where different species of *Eisenia*, *Pontoscolex*, *Enchytraeus*, and *Folsomia* were used to monitor the effect of Hg on their behaviour, mortality, weight loss and reproduction rate (Table 1).

Many studies have demonstrated that microbes are the most affected organisms in a contaminated area (Harris-Hellal et al. 2009; Liu et al. 2014; Mahbub et al. 2017a). Therefore to predict a safe Hg concentration that protects organisms from all trophic levels, microbes can be used as the most reliable indicators. Changes in microbial community structure, diversity and functions are common in contaminated environments (Müller et al. 2001; Zappelini et al.

2015). Therefore, establishing a proper dose-response curve and subsequent estimation of HC values from microbial endpoints can provide reliable secondary data for establishing guideline values. Hence we obtained a wider range of data that covers various soil microbial functions which included dehydrogenase enzyme activity (DHA), soil nitrification rate, urease activity, arylsulphatase activity, alkaline phosphatase activity (AP), Fe (III) reduction, microbial biomass carbon content (MBC) and total microbial alpha diversity (Table 1). These endpoints were observed to respond in a varying manner with Hg gradients.

After plotting the data on SSD calculator, we observed a sigmoidal pattern of distribution of the species affected by different Hg concentrations (Figure 1 and 2). Considering total Hg concentrations in soil, a HC_5 value of 0.6 mg/kg (confidence interval 0.25-1.45) was estimated. Whereas, this value was much lower when we considered water-soluble Hg fractions, i.e., 0.04 mg/kg (CI 0.01-0.15). Because, water-soluble Hg fractions are potentially bioavailable to soil biota, the estimated HC_5 of 0.04 mg/kg indicates that very low concentration of bioavailable Hg is sufficient to exert toxicity to soil organisms. The estimated PNEC values for total Hg and water-soluble Hg were 0.12 – 0.6 mg/kg and 0.008 – 0.04 mg/kg respectively. The only other similar study was by Tipping et al. (2010) who used chronic toxicity data from the years of 1973 to 1997 and data from their experiments on microbial activities. They expressed the HC_5 as 0.13 µg/g soil and 3.3 µg/g organic material. Our approach includes both chronic and acute toxicity data from more recent years as listed in Table 1. In another study, De Vries et al. (2007) emphasized on Hg content in soil solutions and estimated HC_5 value of 0.02–0.08 mg/m^3, however, the study included only 11 data points from the literature. Above all, the HC_5 values estimated in the present study are lower than many guideline values set by different industrial countries, notably Australia (1 mg/kg), Canada (6.6 mg/kg) and the US (2.3 mg/kg) (Mahbub et al. 2017a).

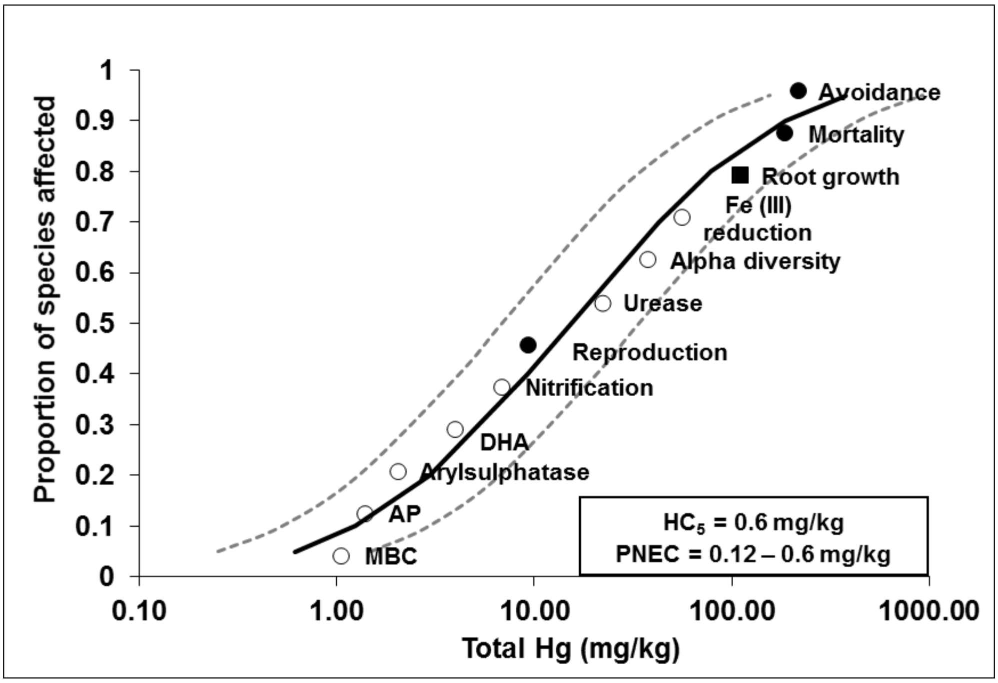

Figure 1: SSD plot of total Hg concentrations in soil and proportion of species affected. The estimated HC_5 is 0.6 mg/kg (CI 0.25-1.45), PNEC is 0.12 – 0.6 mg/kg, R^2=0.95, n=50. Each point on the Y-axis represents the mean of replicate observations of an endpoint as labeled in certain species (total 12 endpoints used) (Table 1). The

unbroken black line is a central tendency, and the grey dashed lines are upper and lower limits at 95% CI. Open circles represent soil microbes, closed circles represent soil invertebrates and closed square represents plants.

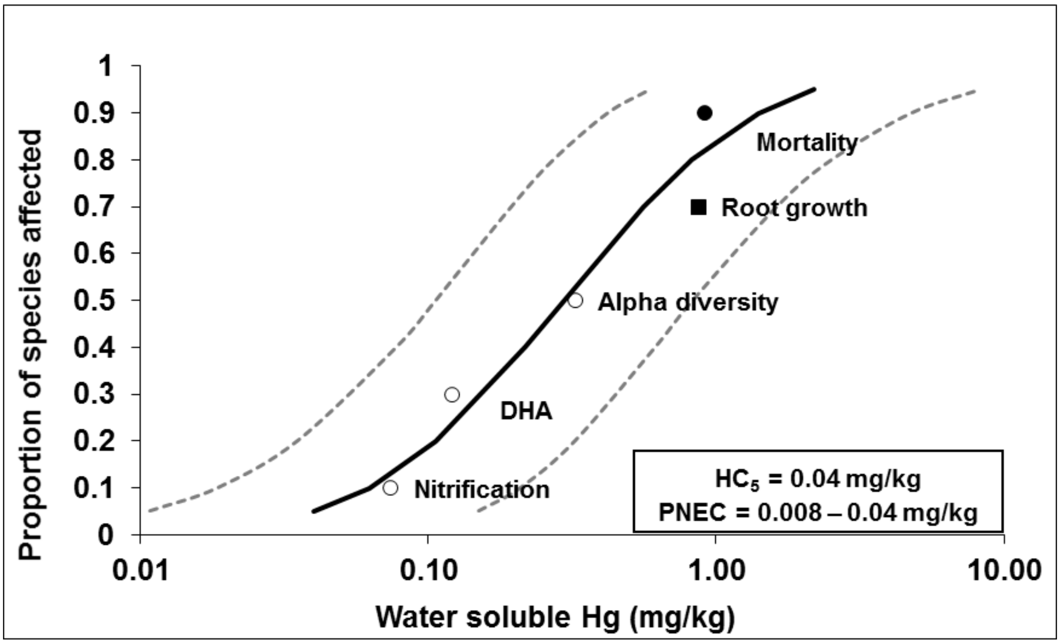

Figure 2: SSD plot of water-soluble Hg concentrations in soil and proportion of species affected. The estimated HC_5 is 0.04 mg/kg (CI 0.01-0.15), PNEC is 0.008 – 0.04 mg/kg, R^2=0.91, n=18. Each point on the Y-axis represents the mean of replicate observations of an endpoint as labeled in certain species (total 5 endpoints used) (Table 1). The unbroken black line is a central tendency, and the grey dashed lines are upper and lower limits at 95% CI. Open circles represent soil microbes, closed circles represent soil invertebrates and closed square represents plants.

The bioavailable fractions of Hg in the soil cannot be predicted by measuring total Hg content. Rather, it largely depends on soil physicochemical properties, such as organic carbon content, pH, mineral contents and clay contents. The selected papers from where toxicity data obtained in the present study used 34 soils with varying physicochemical properties. The important soil properties that influence the Hg bioavailability such as organic carbon content, pH and sand/clay content were in various ranges which indicate that the bioavailable fractions of Hg in the studied soils could have been different (Table 1). Organic matter-rich soils have always been reported to contain a very little amount of soluble fractions of Hg (Biester et al. 2002; Skyllberg 2012). Hence, different soils with similar amounts of total Hg can display varying amounts of soluble Hg (Millán et al. 2006; Skyllberg et al. 2006). This clearly suggests that measuring total Hg content may not predict the real toxicity of Hg in a soil. Alternatively, soluble fractions may be a better predictor to use in toxic dose determination approaches. However, adequate data are not available where toxic doses have been determined based on soluble fractions of Hg in soil; hence, the estimation of a true toxic dose remains a challenge. More eco-toxicological studies are required where soluble fractions of Hg in soils would be considered to determine critical safe limits. Most of the data used in the present study were generated from laboratory-based experiments that might be different in field scenario. However, field data lacks appropriate controls and often contain multiple contaminants making it difficult to validate the toxicity against particular contaminant. Therefore, toxicological studies conducted in the laboratory with appropriate controls are best suited to estimate the potential toxicity of any contaminant.

4. Conclusion

The current soil Hg guideline values developed by the industrialised countries seem to be inadequate for the protection of soil biota given these are based on limited toxicity data. Therefore, we have derived the HC_5 values (0.6 mg/kg and 0.04 mg/kg for total and water-soluble Hg respectively) and safe Hg concentrations based on wider toxicity data available to-date including from our own toxicological studies, and we believe these are more scientifically defensible and appropriate for use as guideline values for Hg in soils. On the other hand, toxicity data based on water-soluble Hg are scarce. Therefore we recommend the future ecotoxicological studies should consider the water-soluble Hg fractions in soil.

5. Declaration of Interests

None

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