

Health & Ecological Risk Assessment

Comparison of Temperate and Tropical Freshwater Species' Acute Sensitivities to Chemicals: An Update

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ABSTRACT

Toxicity data for tropical species are often lacking for deriving water quality guidelines (WQGs) and for conducting ecological risk assessment (ERA). To protect and safeguard valuable natural resources and important biodiversity in tropical freshwater ecosystems, a sound framework should be established to assess and manage the ecological risk of an ever-increasing number of chemicals that occur in the tropics. The present study aims to provide a more up-to-date comparison of the species sensitivity distributions (SSDs) between temperate and tropical freshwater species, by incorporating more acute toxicity data that have been documented. Results showed that temperate freshwater species are generally more sensitive to As, Cr, Pb, Hg, carbaryl, chlorpyrifos, DDT, lindane, and malathion than are their tropical counterparts, whereas tropical species tend to be more sensitive to un-ionized ammonia, Mn, chlordane, and phenol. No sensitivity differences were found between temperate and tropical freshwater species to Cu and pentachlorophenol. A general decline in sensitivity trend to chemicals was revealed by comparing taxon-specific SSDs, from crustaceans to mollusks, worms, fishes, and insects. On the basis of calculated 10% hazardous concentration (HC10) ratios from pairwise temperate and tropical SSDs, the temperate-to-tropic safe extrapolation factor was verified and refined as 5 for information. *Integr Environ Assess Manag* 2019;00:000–000. © 2019 SETAC

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INTRODUCTION

The increased discharge of chemical substances into the environment poses a threat to freshwater species and their ecosystems. Given that there are many developing countries with a large human population in the tropics, the water pollution problem in this region is intensified because of the lack of infrastructures to collect and treat wastewater. Given that the biodiversity is substantially higher in the tropics than in the temperate and cold-water regions, the number of species potentially influenced by chemical pollutants is also expected to be greater (Lacher and Goldstein 1997). To safeguard the crucial biodiversity in tropical freshwater ecosystems, there is an urgent need to establish a reliable framework for deriving water quality guidelines (WQGs) and for conducting ecological risk assessment (ERA) of common chemical contaminants in the tropical region (Wepener and Chapman 2012). However, tropical toxicity data are limited compared to their

temperate counterparts (Kwok et al. 2007; Wang and Leung 2015). Due to this data paucity, temperate toxicity data or WQGs are commonly used as surrogates by governments in tropical countries or regions for their WQGs or directly adopted temperate WQGs for protecting their aquatic ecosystems. However, it is still debatable whether temperate toxicity information can provide sufficient protections for the tropics due to geographical differences in species sensitivities to a chemical.

To date, there have been no clear trends in freshwater species sensitivities to chemicals between temperate and tropical regions. For instance, Dyer et al. (1997) found that temperate fish appeared to be more sensitive to DDT than tropical fish, while no significant difference in sensitivity was found for other compounds (carbaryl, lindane, malathion, pentachlorophenol, and phenol). Maltby et al. (2005) did not find any influences of species' geographical distributions on hazard assessment of insecticides. Kwok et al. (2007) extensively compared acute species sensitivity distributions (SSDs) between temperate and tropical freshwater animals, and found that tropical species were relatively more sensitive to 6 out of the 18 chemicals (un-ionized ammonia (NH₃), As, Zn, chlordane, chlorpyrifos, and phenol), while for several

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other chemicals especially metals and metalloids (elements), the opposite trend was found. Nonetheless, the comparison made by Kwok et al. (2007) had a high uncertainty because their study was based on a limited amount of tropical data that could not cover all major taxonomic groups (e.g., absence of mollusks, worms [e.g., annelids], and insects for DDT). In addition, experimental (water) parameters such as hardness, which may have a significant influence on element toxicity to freshwater species (Bianchini and Wood 2008; Brinkman and Johnston 2012), were not considered confounding factors in SSD comparisons by Kwok et al. (2007). The failure to consider these factors may also lead to high uncertainties for the observed temperate and tropical SSD differences for elements. By adding more tropical toxicity data to cover the missing taxonomic groups, and by normalizing toxicity data as a function of water parameters (e.g., temperature and pH to ammonia, and hardness to elements), the certainty of the results can be greatly improved. With more data available, the temperate-to-tropic safe extrapolation factor of 10 developed by Kwok et al. (2007) can also be verified or refined for the tropics so as to improve the accuracy of their predicted no-effect concentrations (PNECs) or WQGs when temperate information is used.

Therefore, the present study had 3 main objectives. First, available acute toxicity data generated after 2005 were added into the data sets of Kwok et al. (2007) for the comparison of temperate and tropical SSDs for 16 common chemicals, in order to fill the data gap of underrepresented or missing taxonomic groups, and to increase the statistical power of the data analysis. The data gaps for these chemicals were also identified for future toxicity testing. Second, the temperate-to-tropic extrapolation factor was verified and refined after supplementing these newly available data. Third, species sensitivities to a particular chemical were compared among different taxa from each of the 2 climatic regions if there were sufficient toxicity data available. The overall results will be essential for chemical regulation and water quality management in tropical and subtropical regions, especially those in Southeast Asia and South Africa.

METHODOLOGY

Data mining

The present study focused on 16 chemicals: NH_3 , 7 elements (As, Cd, Cr, Cu, Pb, Mn, and Hg), 6 pesticides (carbaryl, chlordane, chlorpyrifos, DDT, lindane, and malathion), and 2 narcotics (pentachlorophenol and phenol). Single-species acute toxicity data were collected from databases (USEPA ECOTOX database, ECETOC database, AED, IUCLID), published literature (e.g., Wang and Leung [2015] and Kleinhenz et al. [2018] for NH_3), and government reports. All original documents were carefully examined before extracting the relevant information for the present study. Selected endpoints were median lethal concentrations (LC50s; up to 96 h) for animals and median effective concentrations (EC50s; up to 24 h) for algal growth inhibition.

The reported experimental temperature, pH, and hardness of each toxicity test were also collected accordingly (if any).

Freshwater species were classified as temperate (between 23.5°N and 65°N, and between 23.5°S and 65°S) or tropical (between 23.5°N and 23.5°S) species based on the location of collection or culture from the original study. All original sources of data were also vetted for quality assurance, and data with limited or no reliability (e.g., unacceptable control mortality, missing or extrinsic species locations) were excluded in the current study (Wheeler, Grist et al. 2002; Chapman et al. 2006). The geometric mean was calculated when more than 1 data point was reported for a species. Any outliers detected by Grubb's test (Grubbs 1969) or Tietjen-Moore test (Tietjen and Moore 1972) were also excluded for each data set.

Considering ammonia toxicity as a function of temperature and pH, all temperate and tropical ammonia data were converted to the NH_3 form at pH 7.0 and 20 °C and at pH 7.0 and 25 °C, respectively (Wang and Leung 2015). Similarly, considering hardness-dependent relationships between hardness and toxicity of elements, all toxicity data of Cd, Cr, Cu, and Pb were normalized to a hardness level of $50 \text{ mg} \cdot \text{L}^{-1} \text{ CaCO}_3$ following a least-squares regression (see Supplemental Data Appendix A for data normalization).

Temperate and tropical SSD comparisons

Toxicity data were ranked in ascending order and assigned corresponding percentiles for SSD constructions. Each SSD was then fitted by 7 parametric regression models (lognormal, log-logistic, Gompertz, Fisher-Tippett, Weibull, log-triangular, and Burr Type III), and hazard concentration 5% and 10% (HC5 and HC10) and their 95% confidence intervals (95% CIs) were subsequently determined by the best fit model approach described by Wang et al. (2014). The best fit model should pass both Shapiro-Francia (SF) and Anderson-Darling (AD) tests and has the minimum corrected Akaike information criterion (Min-AICc). For comparisons, nonparametric bootstrap ($n \geq 10$ for an SSD) and bootstrap regression methods were also applied for computing the HCs and 95% CIs. Temperate-to-tropic HC10 ratio and its 95% CI for pairwise SSDs were computed by corresponding best fit model (and nonparametric models) and Monte Carlo simulation approach (resampled 5000 times; SAS version 9.3; Cary, NC, USA). When there were more than 7 data points for a taxon (amphibians, crustaceans, fish, mollusks, and insects) in a data set, a taxon-specific SSD was also constructed.

To be consistent, the relative sensitivities between temperate and tropical species (or taxa) to a chemical were determined on the basis of calculated pairwise HC10s and their HC10 ratio from the best fit model. If the 95% CIs of the HC10 ratio did not overlap with the unity, then the 2 SSDs were significantly different (Sokal and Rohlf 2012). One-way analysis of covariance (ANCOVA) was also conducted to compare the slope and/or intercept parameters of the pairwise SSDs (SPSS version 23; Armonk, NY, USA). Relative sensitivities among taxa within a geographical region were

compared by 1-way analysis of variance (ANOVA) and Tukey's post hoc tests (significance level $\alpha = 0.05$; GraphPad Prism version 5.00 for Windows, GraphPad Software, San Diego, CA, USA. www.graphpad.com). Test conditions (temperature, pH, and hardness) of the toxicity tests between temperate and tropical regions were also compared by Mann-Whitney U tests (SPSS: version 23, Armonk, NY, USA). In addition, the exceedance of tropical species being protected based on our derived temperate HC5 for a chemical was also evaluated.

Derivation of temperate-to-tropic safe extrapolation factor

Temperate-to-tropic safe extrapolation factor for all chemicals and classes of elements and pesticides were determined by an inverse prediction approach, respectively (Wheeler, Leung et al. 2002). In brief, temperate-to-tropic HC10 ratios for all chemicals, elements, or pesticides calculated from the best fit parametric model, nonparametric bootstrap approach, and bootstrap regression approach, respectively, were ranked and assigned percentiles for constructing probability distributions. The distributions were then fitted by the lognormal regression model, and overall safe extrapolation factors covering 90%, 95%, and 99% of the test chemicals were calculated. Afterwards, appropriate temperate-to-tropic safety extrapolation factors for all concerned chemicals, elements, and pesticides were estimated, respectively.

RESULTS AND DISCUSSION

Data availability and knowledge gaps

The utility of the SSD approach in establishing WQGs or conducting ERA is highly dependent on availability of the toxicity data and their quality (Wheeler, Grist et al. 2002; Dowse et al. 2013). The data used in SSD constructions with high quality can further increase the certainty of threshold derivations (e.g., HCs) and the accuracy of SSD comparisons. Therefore, all original documents were carefully examined before data extraction, and data of poor quality were not included in the present study. Among all test chemicals, with the exception of the tropical data sets of As (amphibians and fishes), chlordane (fishes only), Cd (5 data points), and Cr (1 data point with data normalization), all data sets contained more than 10 data points from at least 4 taxonomic groups (pie charts in Figures 1 and 2; see Supplemental Data Table S1 for data sets). During the present study, toxicity data gaps were also identified for the 16 selected chemicals (Table S2). For example, taxonomic groups such as crustaceans, mollusks, worms, and insects were all missing in the tropical data sets for As and chlordane, while tropical worms and mollusks are generally missing for phenol and some pesticides (e.g., chlorpyrifos and malathion). The missing of these taxonomic groups could lead to high uncertainties of threshold estimates (e.g., HC10s) and thus influence the accuracy of the temperate and tropical SSDs' comparisons. Therefore, further toxicity tests should be conducted to fill these

knowledge gaps, especially for the tropical data sets of As, chlordane, Cd, and Cr, so that a more accurate comparison between the 2 climate regions can be achieved.

Comparison of temperate and tropical species sensitivities

As demonstrated by previous studies and the present study, tropical aquatic species showed different sensitivities to a number of chemicals when compared with their temperate counterparts. For example, Chapman et al. (2006) found that tropical marine organisms were relatively more sensitive to Cd, least sensitive to Cu, and of intermediate sensitivity to Pb and Zn than those species from temperate and polar regions. In contrast to the results of Chapman et al. (2006), a more comprehensive metaanalysis for 11 chemicals conducted by Wang et al. (2014) showed that there were only small differences in acute toxicities of chemicals between tropical and temperate saltwater organisms, and tropical saltwater species were more sensitive to Cu, Hg, Zn, phenol, and pentachlorophenol than their temperate counterparts. For freshwater ecosystems, Daam and Van den Brink (2010) examined the differences between the sensitivities of temperate and tropical organisms toward pesticides and concluded that the pesticide dissipation rates and sensitivities of freshwater organisms varied among species. Brix et al. (2001) conducted a comparison of the acute sensitivities of temperate (cold- and warm-water) and tropical freshwater species toward Cu, and found that temperate cold-water species appears to be the most sensitive ones, followed by temperate warm-water and tropical species. Kwok et al. (2007) preliminarily found tropical species were relatively more sensitive to 6 of the 18 chemicals tested, while temperate species were generally more sensitive to elements.

In the present study, temperate and tropical freshwater SSDs visually diverged for most chemicals (Figures 1 and 2), indicating different sensitivities. This was characterized by significantly different slope and/or intercept parameters of the pairwise lognormal fitted SSDs (Table S3). The estimated HC5 and HC10 values and their 95% CIs are listed in Table 1 and Table S4 using the best fit model and nonparametric models, respectively. On the basis of calculated temperate-to-tropic HC10 ratios and 95% CIs (Tables 1 and S4), our results demonstrated that tropical freshwater species were more sensitive to NH_3 , Mn, chlordane, and phenol than their temperate counterparts, while temperate freshwater species were more sensitive to As, Cr, Hg, Pb, and 5 out of 6 pesticides (except for chlordane). Temperate and tropical species shared similar sensitivities to Cu and pentachlorophenol.

The observed differences between temperate and tropical SSDs might be driven by quality and quantity of data and by differences in experimental conditions, species and taxonomic compositions and proportions, and taxon-specific sensitivities. For the majority of the tested chemicals, tropical toxicity tests were conducted at a significantly higher temperature and hardness than temperate toxicity tests; Table 2), with overall median differences of 5°C (Mann-Whitney U test: $U = 1.5$, $n_{\text{temperate}} = n_{\text{tropic}} = 16$,

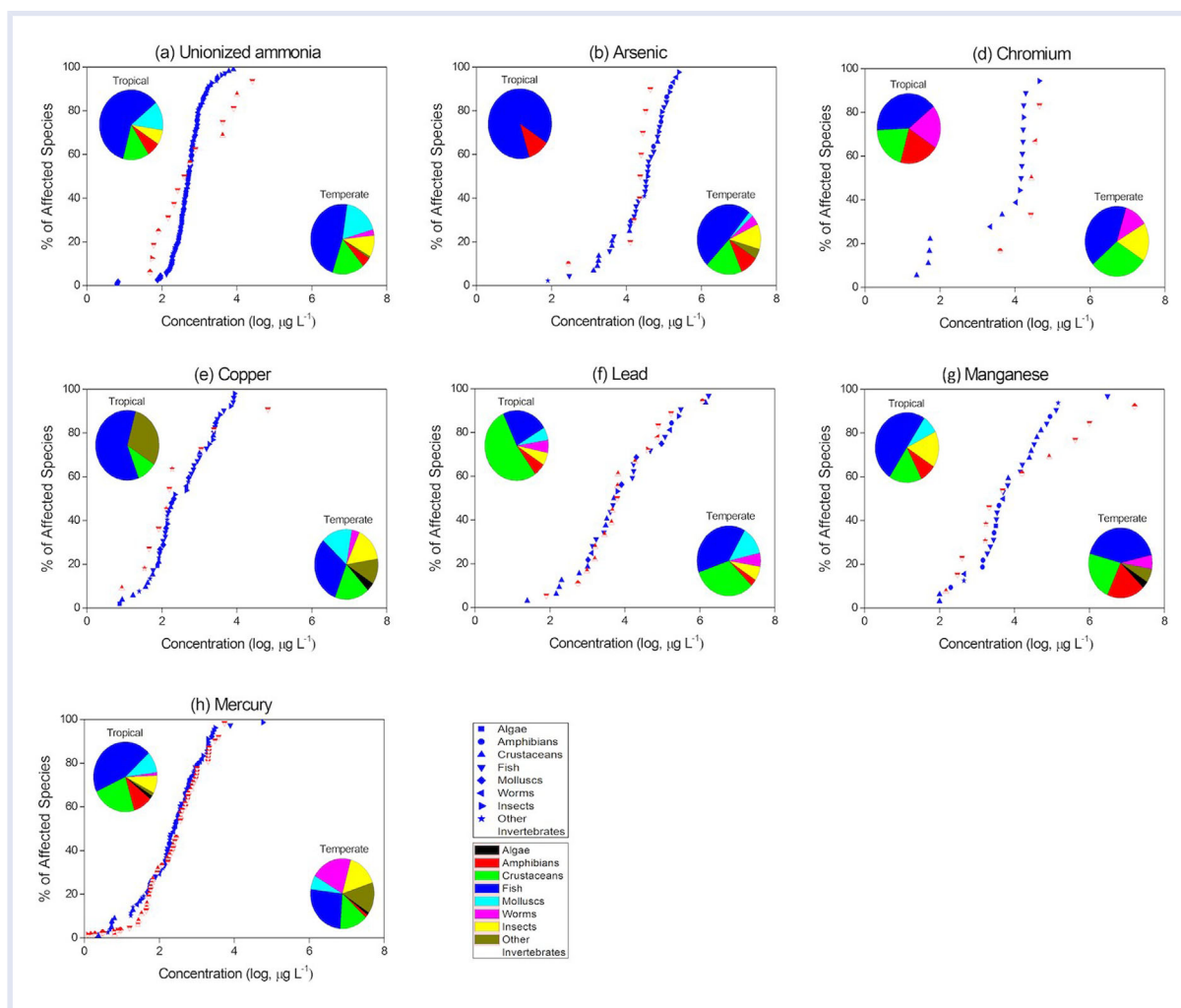


Figure 1. Temperate and tropical species sensitivity distributions (SSDs) for un-ionized ammonia (a) and 7 metals and metalloids, including As (b), Cd (temperate only) (c), Cr (d), Cu (e), Pb (f), Mn (g), and Hg (h). Pie charts represent species compositions for the tropical (upper chart) and temperate (lower chart) distributions. Half-filled symbols (shown in red) indicate tropical data points, whereas fully filled symbols (i.e., shown in blue) indicate temperate data points. Symbols for taxonomic compositions are given in the upper key, while color codes for different taxa in pie charts are presented in the lower key.

$p < 0.05$) and $18 \text{ mg} \cdot \text{L}^{-1} \text{ CaCO}_3$ ($U = 53$, $n_{\text{temperate}} = n_{\text{tropic}} = 12$, $p < 0.05$), respectively. While tropical toxicity tests were conducted at significantly lower pH conditions (median difference 0.2; $U = 56$, $n_{\text{temperate}} = n_{\text{tropic}} = 13$, $p < 0.05$).

The observed test temperature differences may be one of the important factors influencing both the physicochemical properties of chemicals present in the test medium and the physiology of the freshwater organisms; such thermal-mediated changes in these factors can influence the bioavailability, bioaccumulation, excretion and toxicity of the chemical in the organism (Pörtner 2002; Bourgeault et al. 2013). Given that most previous laboratory studies suggested that chemical toxicity increases with temperature (e.g., Cairns et al. 1978; Leung et al. 2000; Heugens et al. 2003; Kwok and Leung 2005), it is commonly believed that tropical species would be more sensitive to chemicals than temperate species (Castillo et al. 1997). However, the present results indicated that most chemicals (10 out of the 15 chemicals) were more toxic to temperate species. This could be because biochemical detoxification and elimination of

toxicants may increase with temperature, which eventually reduces chemical toxicity (Howe et al. 1994), and at the same time, low water temperatures may slow down excretion and detoxification and hence increase toxicity (Sprague 1985). The evaporation rate of chemicals with low vapor pressures (e.g., pesticides) may increase with temperature, and hence reduce the chemical concentration, uptake, and bioaccumulation, and lower toxicity (Magallona 1994).

The toxicity of divalent elements in particular is often inversely related to water hardness (decreased toxicity with increasing hardness [Erickson et al. 1998]). We also found that tropical toxicity tests were generally conducted at significantly higher hardness against As, Cr, Pb, and Hg than their temperate counterparts, while tropical species were sensitive to the 4 elements (lower toxicity at higher hardness). Hardness can affect element toxicity indirectly through nonspecific ionic interferences that decrease the ionic toxicity of the element ion by interfering with element uptake and action through competition at the binding sites (Chapman et al. 1980; Park et al. 2009).

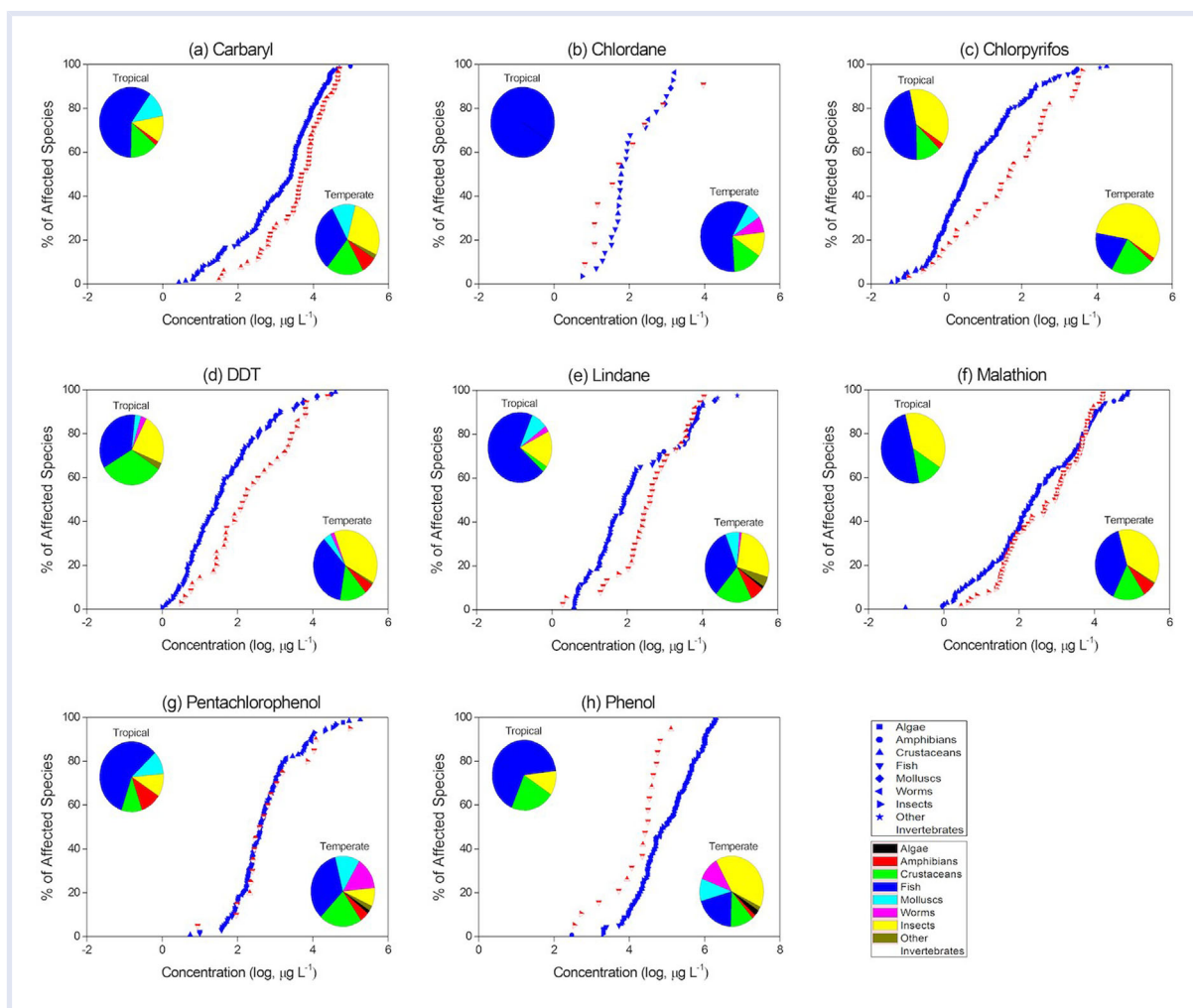


Figure 2. Temperate and tropical species sensitivity distributions (SSDs) for pesticides and narcotics, including carbaryl (a), chlordane (b), chlorpyrifos (c), DDT (d), lindane (e), malathion (f), pentachlorophenol (g), and phenol (h). Conventions as for Figure 1.

Because pH can affect the equilibrium of aqueous ionic chemicals, especially elements, the differences in the test pH for elements might play a part in driving the observed differences between temperate and tropical species sensitivities. It is commonly reported that element toxicity increases with decreasing pH (McDonald et al. 1989; Campbell 1995; Pynnönen 1995). However, we found that tropical tests for As and Mn were generally conducted at lower pH than tropical ones, while tropical species were found to be less sensitive to both elements. Such responses might be explained by competition between H^+ and ionic elements, decreasing the binding of elements on cell-surface ligands at the cell surface and altering the cell's permeability to certain elements and reducing toxicity at low pH (Wang et al. 2016).

To minimize the potential influences of water parameters on chemical toxicity to freshwater species, and thus introduce bias in the SSD comparisons, all toxicity data of ammonia (temperature- and pH-dependent) and Cd, Cr, Cu, and Pb (hardness-dependent) were normalized and used in our metaanalyses. Nonetheless, the insufficient data availability of other water parameters such as alkalinity, dissolved

organic carbon, dissolved O, or ionic strength inherently limited our ability to investigate their potential influences on the observed temperate and tropical SSD differences for the elements. Further study should be conducted when there is more information available to investigate the potential effects of these confounding factors on the observed species sensitivity differences between the 2 geographical regions alone or in combination with temperature, pH, or hardness for the elements of concern.

Representativeness and compositions of species in a data set might be other drivers introducing uncertainty for SSDs comparisons. Thus, the general data sets were separated into major taxonomic groups and relative sensitivities between or among different taxa were also compared in the present study. Using all available toxicity data, more temperate subdistributions were available than the ones from the tropical regions for the chemicals being examined (Supplemental Data Figures S1 and S2). Within a geographical region, relative sensitivities of taxa to different elements expressed a similar trend, while those to NH_3 , pesticides, and narcotics varied among taxa (see HC10s in the Table S5). For elements, crustaceans were generally the most sensitive

Table 1. Temperate and tropical hazardous concentrations 5% and 10% with their 95% confidence intervals for each chemical and the corresponding temperate-to-tropic HC10 ratio from the best fit model

Chemical (temperate/tropical)	N (nr of common species for both data sets)	HC5 (95% CI; $\mu\text{g} \cdot \text{L}^{-1}$)	HC10 (95% CI; $\mu\text{g} \cdot \text{L}^{-1}$)	Best fit model	Min-AICc	W'	AD	Temperate-to-tropic HC10 ratio (95% CI; $\mu\text{g} \cdot \text{L}^{-1}$)	Tropical species being protected by temperate HC5 (%)
Un-ionized ammonia									
Temperate	110 (4)	129 (124, 134)	181 (177, 185)	Burr Type III	-978	0.56	0.20	4.2 (2.8, 8.0)	62%
Tropical	15 (4)	28 (9.0, 38)	43 (20, 55)	Burr Type III	-93	0.56	0.41		
Arsenic									
Temperate	43 (0)	754 (606, 950)	2000 (1660, 2420)	Burr Type III	-317	-0.68	1.51	0.2 (0.01, 0.5)	99%
Tropical	9 (0)	6250 (4764, 16600)	9010 (7580, 17700)	Burr Type III	-45	0.69	0.86		
Cadmium									
Temperate	25 (1)	8.9 (6.8, 13)	22 (17, 32)	Burr Type III	-155	-1.01	0.71	NA	NA
Tropical	1 (1)	NA	NA	NA	NA	NA	NA		
Chromium									
Temperate	17 (0)	763 (8.3, 4940)	1550 (25, 6650)	Burr Type III	-69	0.27	1.15	0.6 (0.3, 0.9)	96%
Tropical	5 (0)	1040 (30, 3080)	2480 (199, 5480)	Weibull	-15	-1.19	1.04		
Copper									
Temperate	51 (0)	19 (12, 27)	32 (23, 39)	Burr Type III	-331	0.50	0.58	1.9 (0.9, 3.3)	88%
Tropical	10 (0)	11 (5.7, 19)	17 (10, 25)	Burr Type III	-56	1.58	0.51		
Lead									
Temperate	31 (5)	118 (77, 199)	270 (198, 382)	Burr Type III	-222	-0.33	0.24	0.5 (0.3, 0.9)	99%
Tropical	17 (5)	318 (144, 560)	560 (336, 811)	Burr Type III	-107	0.21	0.26		
Manganese									
Temperate	31 (2)	175 (105, 618)	384 (264, 874)	Burr Type III	-203	-0.14	0.29	2.3 (1.2, 7.1)	90%
Tropical	12 (2)	94 (33, 123)	170 (88, 218)	Burr Type III	-75	-0.28	0.38		
Mercury									
Temperate	77 (4)	8.4 (7.6, 9.3)	17 (16, 19)	Burr Type III	-598	-0.45	0.61	0.7 (0.6, 0.8)	96%
Tropical	62 (4)	12 (10, 14)	24 (21, 27)	Burr Type III	-461	0.40	0.34		
Carbaryl									

(Continued)

Table 1. (Continued)

Chemical (temperate/tropical)	N (nr of common species for both data sets)	HC5 (95% CI; $\mu\text{g} \cdot \text{L}^{-1}$)	HC10 (95% CI; $\mu\text{g} \cdot \text{L}^{-1}$)	Best fit model	Min-AICc	W'	AD	Temperate-to-tropic HC10 ratio (95% CI; $\mu\text{g} \cdot \text{L}^{-1}$)	Tropical species being protected by temperate HC5 (%)
Temperate	143 (8)	14 (13, 15)	38 (34, 41)	Burr Type III	-1062	0.18	0.27	0.1 (0.08, 0.2)	99%
Tropical	51 (8)	106 (86, 138)	270 (226, 338)	Burr Type III	-357	0.27	1.15		
Chlordane									
Temperate	27 (1)	18 (14, 21)	22 (18, 25)	Burr Type III	-155	-1.19	1.04	3.5 (1.6, 8.6)	67%
Tropical	10 (1)	4.6 (1.7, 6.1)	6.3 (2.6, 7.9)	Burr Type III	-60	0.50	0.58		
Chlorpyrifos									
Temperate	135 (8)	0.15 (0.14, 0.16)	0.27 (0.25, 0.29)	Fisher-Tippett	135	1.58	0.51	0.5 (0.3, 0.8)	88%
Tropical	32 (8)	0.15 (0.10, 0.20)	0.50 (0.36, 0.64)	Log-normal	32	-0.33	0.24		
DDT									
Temperate	103 (8)	1.6 (1.5, 1.7)	2.7 (2.6, 2.9)	Fisher-Tippett	-844	0.21	0.26	0.4 (0.3, 0.6)	99%
Tropical	34 (8)	3.5 (2.3, 4.8)	6.7 (4.3, 8.8)	Burr Type III	-230	-0.14	0.29		
Lindane									
Temperate	85 (5)	4.1 (3.7, 4.7)	6.7 (6.1, 7.5)	Burr Type III	-581	-0.28	0.38	0.2 (0.1, 0.3)	>99%
Tropical	36 (5)	20 (13, 36)	38 (27, 58)	Burr Type III	-241	-0.45	0.61		
Malathion									
Temperate	135 (20)	1.6 (1.4, 1.8)	5.1 (4.5, 5.6)	Log-normal	-980	0.40	0.34	0.5 (0.4, 0.6)	>99%
Tropical	63 (20)	4.6 (4.1, 5.3)	11 (9.3, 12)	Burr Type III	-429	0.18	0.27		
Pentachlorophenol									
Temperate	129 (6)	52 (49, 57)	79 (75, 83)	Burr Type III	-1042	1.31	0.98	0.8 (0.6, 1.1)	99%
Tropical	19 (6)	76 (58, 96)	101 (81, 123)	Burr Type III	-151	0.42	0.95		
Phenol									
Temperate	154 (7)	2840 (2040, 3680)	6520 (5310, 7540)	Burr Type III	-1077	-3.29	0.43	2.5 (1.4, 6.7)	90%
Tropical	18 (7)	1160 (589, 2290)	2610 (1510, 4490)	Burr Type III	-105	-1.33	0.25		

AD = statistic of the Anderson-Darling test; AICc = Akaike information criterion; HC5 = hazardous concentration 5%; HC10 = hazardous concentration 10%; W' = statistic of the Shapiro-Francia test.

Table 2. Comparison of median values of temperature, pH, and hardness used between temperate and tropical toxicity tests for 19 chemicals^a

Chemical	Median temperature (°C)			Median pH			Hardness (mg · L ⁻¹ CaCO ₃)		
	Temperate	Tropical	Difference	Temperate	Tropical	Difference	Temperate	Tropical	Difference
Unionized ammonia	20	25	5 ^b	7.9	7.5	-0.4	86	NA	NA
Arsenic	20	22	2	7.6	7.1	-0.5	49	218	169 ^b
Cadmium	20	27	7 ^b	7.9	7.5 (n=2)	NA	50	NA	NA
Chromium	21	25	4 ^b	7.6	7.3	-0.3	50	268	218 ^b
Copper	20	26	6 ^b	7.6	7.6	0.0	89	90	1.0
Lead	20	25	5 ^b	7.3	7.3	0.0	48	114	66 ^b
Manganese	20	25	5 ^b	7.6	6.9	-0.7 ^b	117	78 (n=2)	NA
Mercury	19	26	7 ^b	7.6	7.6 (n=1)	NA	55	238	183 ^b
Carbaryl	20	22	2	7.5	7.5	0.0	92	110	18 ^b
Chlordane	20	26	6 ^b	7.3	7.3	0.0	47	85	38 ^b
Chlorpyrifos	21	26	4 ^b	7.7	7.0 (n=2)	NA	110	199 (n=2)	NA
DDT	19	27	8 ^b	7.2	7.5	0.3	44	83	39 ^b
Lindane	20	26	6 ^b	7.4	7.4	0.2	107	72	-35 ^b
Malathion	22	26	4 ^b	7.2	7.4	0.2	77	81	4.0
Pentachlorophenol	20	22	2	7.7	7.6	-0.1	167	195	28 ^b
Phenol	20	24	4 ^b	7.6	7.3	-0.3	100	72	28 ^b

^aThe data with data points fewer than 3 were not used in Mann-Whitney *U* test (NA).

^bIndicates significantly different medians based on Mann-Whitney *U* test, under level of significance $\alpha = 0.05$.

species, followed by mollusks, worms, and fishes, while insects were the least sensitive (for As [temperate], Cu [temperate], Pb [temperate], Mn [temperate], Hg [temperate and tropical]). For NH₃, mollusks were the most sensitive species, followed by fishes, crustaceans, and insects. For pesticides and narcotics, crustaceans were the most sensitive taxon to carbaryl (temperate and tropical), chlorpyrifos (temperate), and malathion (temperate and tropical), while insects were surprisingly the most sensitive taxon to chlorpyrifos (tropical), DDT (temperate and tropical), lindane (temperate), and phenol (temperate). On the contrary, fishes (temperate) were the most sensitive taxa to pentachlorophenol, while insects (temperate) were the least sensitive. The results of taxon-specific SSDs' comparisons suggested that freshwater crustaceans were generally the most sensitive groups to the tested chemicals. Therefore, crustaceans could be considered as sensitive taxon for adoption in future toxicity testing and risk assessment.

Taxon-specific temperate-to-tropic HC10 ratios were also calculated for crustaceans, fishes, and insects (Table S6). There was no clear trend in sensitivities of insects to the examined chemicals between both regions. Temperate crustaceans were found to be generally more sensitive to their tropical counterparts for Pb, Hg, carbaryl, DDT, and malathion, while temperate fishes were either more sensitive or shared similar sensitivity to the fishes from the tropics. On the contrary, tropical fishes tended to be more sensitive to

NH₃. Such taxon-specific differences and their taxonomic compositions and proportions in the both data sets can partially contribute to the observed temperate and tropical SSDs differences. For example, the temperate and tropical data sets for malathion were all dominated by crustaceans, fishes, and insects (pie charts in Figure 2f), whereas corresponding temperate taxa were all more sensitive than their tropical counterparts. Although one cannot conclude that this is the only cause of the difference in species sensitivities to a chemical (e.g., malathion), further studies are required to test the hypothesis that individual temperate taxonomic groups (e.g., algae, amphibians, mollusks, and worms) are more sensitive than their corresponding tropical counterparts.

Temperate-to-tropic safe extrapolation factor

Extrapolation factors (also referred to as "assessment factors" or "uncertainty factors") are often applied in WQGs and ERA as an alternative (conservative) approach when there was limited or no ideal toxicity information for a species at a desired experimental scenario (see Table 3 for current commonly used extrapolation factors for WQGs and ERA). In the present study, temperate-to-tropic safe extrapolation factor proposed by Kwok et al. (2007) was also evaluated and refined by incorporating more freshwater toxicity data inclusion, especially for tropical species. Using HC10 ratios derived from corresponding best fit models, extrapolation

Table 3. Types and magnitudes of extrapolation factors applied in derivation of water quality guidelines and ecological risk assessment

Types of extrapolation factors	Magnitude	Extrapolation factors ^a	References
Intra-order extrapolation	Plant sensitivity to herbicides	15	Fletcher et al. 1990
Intra-class extrapolation		≥ 300 (80%)	Fletcher et al. 1990
Interspecies extrapolation	Species within genus	10 (95%); 16.3 (99%)	Calabrese and Baldwin 1993
	Genera within family	11.7 (95%); 16.9 (99%)	Calabrese and Baldwin 1993
	Families within order	99.5 (95%); 145 (99%)	Calabrese and Baldwin 1993
	Orders with class	64.8 (95%); 87.5 (99%)	Calabrese and Baldwin 1993
	Classes within phylum	1000 (95%)	Sloof et al. 1986
Acute-to-chronic ratio (ACR)		10	USEPA 1991; OECD 1995
		40	ECETOC 1993
	Un-ionized ammonia-specific	20 ^b	Wang and Leung 2015
		10 ^c	Wang and Leung 2015
LOEC-to-NOEC ^c		10	USEPA 1985
Aquatic assessment factor	Canada	0.01; 0.05	Ontario Ministry of Environment 1984; CCME 1991
Field-to-laboratory extrapolation		10	USEPA 1991; OECD 1995
Freshwater-to-saltwater extrapolation		7.2 (95%); 8.8 (99%)	Leung et al. 2001
Temperate-to-tropic extrapolation	Saltwater ecosystem	2	Wang et al. 2014
	Freshwater ecosystem (un-ionized ammonia-specific)	4	Wang and Leung 2015
	Freshwater ecosystem	10 (95%); 40 (99%)	Kwok et al. 2007
		5	The present study

ECETOC = European Centre For Ecotoxicology and toxicology of Chemicals; LOEC: lowest-observed effect concentration; NOEC: no-observed effect concentration; OECD = Organisation for Economic Co-operation and Development; USEPA = US Environmental Protection Agency.

^aThe percentile in parentheses indicates the potential variability being captured.

^bThe ACR was derived based on pairwise freshwater acute and chronic toxicity data of un-ionized ammonia with normalization at pH 7.0 and 20 °C.

^cThe ACR was derived based on pairwise freshwater acute and chronic toxicity data of un-ionized ammonia without normalization at pH 7.0 and 20 °C.

factors covering 95% of all tested chemicals (except for Cr), elements and pesticides were estimated as 4.4 (3.5, 5.9), 3.5 (2.2, 7.7) and 3.7 (1.5, 9.1), respectively from the distribution (Figure 3 and Table S7). Considering the current results and practicality for ease of use, we recommend the refined safe extrapolation factor as 5, which is smaller than the one (i.e., 10) derived by Kwok et al. (2007). Arguably, such a difference is likely because of the enlarged data sets (pesticides) or exclusion of data without data normalization (NH₃ and elements) being used in the present study. Comparatively, when we conducted similar extrapolation factor derivations using nonparametric fitting models, larger values were computed (e.g., 11 (7.4, 18) and 13 (9.3, 20) using all

chemicals; Figures S3 and S4 and Table S7). Therefore, 10 instead of 5 should be considered during temperate-to-tropic extrapolations when one is applying the nonparametric bootstrap or bootstrap regression approach.

Compared to the traditional default extrapolation factor approach (e.g., 10 for LOEC-to-NOEC extrapolations; Table 3), our probabilistic distribution approach appears to be more reliable because these observations are data driven and account for inherent variability of differences in sensitivity between temperate and tropical freshwater species. When there are sufficient data for a chemical or a class of chemicals, chemical-specific (e.g., NH₃-specific temperate-to-tropic extrapolation factor of 4 recommended by Wang and Leung

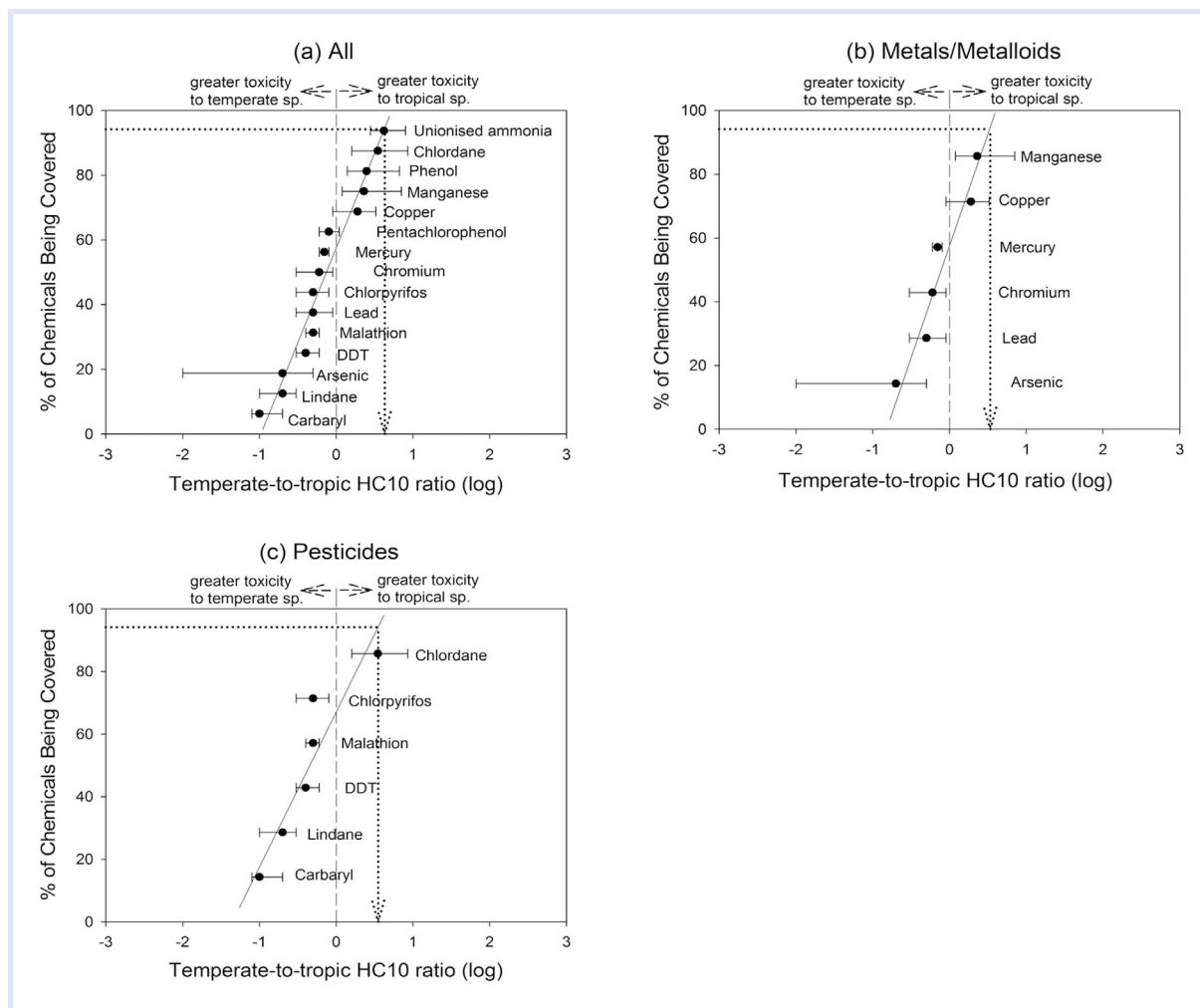


Figure 3. The relationship between temperate and tropical hazardous concentration for 10% of species (HC10) ratios based on the best fit model for all chemicals (a), metals and metalloids (b), and pesticides (c). Horizontal error bars denote the 95% confidence intervals (95% CIs) of the HC10 ratios, and vertical arrows (i.e., dotted lines) indicate the safe extrapolation factors at 95% of chemicals being covered.

[2015]) or class-specific (e.g., 3.5 (2.2, 7.7) for elements and 3.7 (1.5, 91) for pesticides; Table S7) extrapolation factors can be derived. Overall, our recommended extrapolation factor of 5 can provide adequate protection for all tested chemicals when their temperate information is borrowed as surrogates by the tropics. Alternatively, for a particular chemical for which temperate species are more sensitive (smaller than 1 HC10s; Table 1), its temperate information can be directly used without extrapolation factor adjustment (for potential overprotection concern). But it is important to note that such a relaxation must be further validated through field studies. For instance, despite the fact that there being no differences in sensitivity between temperate and tropical species to Cu, only 88% of tropical species would be protected when our interim temperate HC5s are adopted as a conservative surrogate (Table 1). If the estimated temperate HC5 (median: $32 \mu\text{g} \cdot \text{L}^{-1}$) is divided by our recommended extrapolation factor of 5, an interim tropical HC5 of $6.4 \mu\text{g} \cdot \text{L}^{-1}$ could be derived, which can provide sufficient protection (more than 99% of protection) for corresponding tropical species to Cu.

Our derived temperate HC5s were then used as surrogates directly and the probabilities that tropical species protected were also estimated (Table 1). For example, if temperate HC5s were used as surrogates for tropical species directly, 38%, 10%, 33%, and 10% of tropical species would not be protected when exposed to NH_3 , Mn, chlordane, and phenol, respectively (e.g., tropical fish *Cirrhinus mrigala*; see more examples in Table S8). If these temperate HC5s are divided by our recommended extrapolation factor of 5, the estimated interim tropical HC5s can be sufficiently protective for the 4 chemicals (more than 99% of protection). Therefore, when there are limited or no tropical toxicity data for a chemical, temperate information (toxicity data or WQGs) can be applied as surrogates and divided by our recommended of extrapolation factor of 5 by tropical countries or regions for their data dossier development, interim WQG establishment, and screening-level risk assessment of the chemical. This can provide important economic welfare benefits (e.g., reduce cost of protection and use of animals in toxicity testing) with an appropriate and protection level.

Because only acute toxicity data were used in our metaanalyses, further studies should be conducted using chronic toxicity to investigate whether the patterns of differences in sensitivity between temperate and tropical freshwater species toward the tested chemicals are similar to the acute ones, and to evaluate or refine our recommended temperate-to-tropical extrapolation factor of 5. No clear trend (larger or smaller) existed when we compared the measured water background concentrations from the tropics (e.g., for Cd, Cr, Cu, and Pb; Table S9) with the interim tropical chronic WQGs estimated from temperate chronic WQGs recommended by the US Environmental Protection Agency (USEPA) and a factor of 5. Therefore, field studies are also necessary to validate whether applying the extrapolation factor of 5 to surrogate WQGs (based on temperate information) can adequately protect freshwater species and their ecosystem integrity.

CONCLUSIONS

The results of the present comprehensive and up-to-date metaanalysis showed that temperate freshwater species were generally more sensitive to most chemicals of concern (i.e., As, Cr, Pb, Hg, carbaryl, chlorpyrifos, DDT, lindane, and malathion) than their tropical counterparts. On the contrary, tropical freshwater species were found to be more sensitive to un-ionized ammonia, Mn, chlordane, and phenol, while there were no sensitivity differences between temperate and tropical freshwater species to Cu and pentachlorophenol. Comparison of a limited number of taxon-specific SSDs showed that crustaceans were the most sensitive taxon that are suitable for toxicity testing and risk assessment of chemicals. Based on the HC10 ratios for the test chemicals, an extrapolation factor of 5 is recommended when using temperate freshwater toxicity information as a conservative surrogate for predicting chemical toxicity to tropical freshwater species or deriving tropical WQGs.

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Data Accessibility—Data are available upon request to Zhen Wang at wangzhen.wz@hotmail.com.

SUPPLEMENTAL DATA

Appendix A. Data normalization for ammonia and metals

Figure S1. Temperate and tropical taxon-specific species sensitivity distributions for un-ionized ammonia and 7 metals.

Figure S2. Temperate and tropical taxon-specific species sensitivity distributions for pesticides and narcotics.

Figure S3. The relationship between temperate and tropical hazardous concentration for 10% of species ratios based on the nonparametric species sensitivity distributions for all chemicals (excluding As and Cr with number of data points < 10 in tropical data sets) (a) and pesticides (b).

Figure S4. The relationship between temperate and tropical hazardous concentration for 10% of species ratios based on bootstrap regression species sensitivity distributions (SSDs) for all chemicals (a), metals and metalloids (b), and pesticides (c).

Table S1. Summary of all available temperate and tropical data for the 16 tested chemicals

Table S2. Data availability of taxonomic groups in constructing freshwater species sensitivity distributions

Table S3. Comparison of temperate and tropical log-normal species sensitivity distributions, using 1-way analysis of covariance

Table S4. Temperate and tropical hazardous concentrations 10% and 95% confidence intervals for each chemical and corresponding temperate-to-tropic HC10 ratio from nonparametric bootstrap and bootstrap regression approaches, respectively

Table S5. Taxon-specific hazardous concentrations 10% and their 95% confidence intervals derived from the best fit model which must pass both Shapiro-Francia and AD tests, and have the minimum corrected Akaike information criterion (Min-AICc)

Table S6. Taxon-specific temperate-to-tropic hazardous concentration 10% ratios and their 95% confidence intervals

Table S7. Estimated temperate-to-tropic safe extrapolation factors derived from probability distributions using pairwise temperate and tropical HC10 ratios of all 15 chemicals (except for Cd), metals, and pesticides

Table S8. Summary of tropical species that cannot be protected when derived temperate HC5 values were used directly by the tropics

Table S9. Summary of interim tropical water quality guidelines derived from temperate acute and chronic WQGs as surrogates and divided by our recommended temperate-to-tropic extrapolation factor of 5.

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REFERENCES

- Bianchini A, Wood CM. 2008. Does sulfide or water hardness protect against chronic silver toxicity in *Daphnia magna*? A critical assessment of the acute-to-chronic toxicity ratio for silver. *Ecotoxicol Environ Saf* 71: 32–40.
- Bourgeault A, Ciffroy P, Garnier C, Cossu-Leguille C, Masfaraud J, Charlatcka R, Garnier J. 2013. Speciation and bioavailability of dissolved copper in different freshwaters: Comparison of modelling, biological and chemical responses in aquatic mosses and gammarids. *Sci Total Environ* 452–453:68–77.
- Brinkman SF, Johnston WD. 2012. Acute toxicity of zinc to several aquatic species native to the Rocky Mountains. *Arch Environ Contam Toxicol* 62:272–281.
- Brix KV, Deforest DK, Adams WL. 2001. Assessing acute and chronic copper risk to freshwater aquatic life using species sensitivity distributions for different taxonomic groups. *Environ Toxicol Chem* 20:1846–1856.
- Cairns J Jr, Buikema AL Jr, Heath AG, Parker BC. 1978. Effects of temperature on aquatic organism sensitivity to selected chemicals. *Bull – Va Water Resour Res Cent* 106:1–88.
- Calabrese EJ, Baldwin LA. 1993. Performing ecological risk assessments. Boca Raton (FL): Lewis.

- Campbell PG. 1995. Interactions between trace metals and aquatic organisms: A critique of the free-ion activity model. In: Tessier A, Turner DR, editors. Metal speciation and bioavailability in aquatic systems. Chichester (UK): Wiley. p 45–102.
- Castillo LE, De la Cruz E, Ruepert C. 1997. Ecotoxicology and pesticides in tropical aquatic ecosystems of Central America. *Environ Toxicol Chem* 16:41–51.
- [CCME] Canadian Council of Ministers of the Environment. 1991. A protocol for the derivation of water quality guidelines for the protection of aquatic life, Canadian water quality guidelines. Winnipeg (MB). Appendix 9.
- Chapman GA, Ota S, Reht F. 1980. Effects of water hardness on the toxicity of metals to *Daphnia magna*. Corvallis (OR): USEPA Office of Research and Development.
- Chapman PM, McDonald BG, Kickham PE, McKinnon S. 2006. Global geographic differences in marine metals toxicity. *Mar Pollut Bull* 52:1081–1084.
- Daam MA, Van den Brink PJ. 2010. Implications of differences between temperate and tropical freshwater ecosystems for the ecological risk assessment of pesticides. *Ecotoxicology* 19:24–37.
- Dowse R, Tang D, Palmer CG, Kefford BJ. 2013. Risk assessment using the species sensitivity distribution method: Data quality versus data quantity. *Environ Toxicol Chem* 32:1360–1369.
- Dyer SD, Belanger SE, Carr GJ. 1997. An initial evaluation of the use of Euro/North American fish species for tropical effects assessments. *Chemosphere* 35:2767–2781.
- [ECETOC] European Centre for Ecotoxicology and Toxicology of Chemicals. 1993. Environmental hazard assessment of substances. Brussels (BE). Technical Report 51. p 103.
- Erickson RJ, Brooke LT, Kahl MD, Venter FV, Harting SL, Markee TP, Spehar RL. 1998. Effects of laboratory test conditions on the toxicity of silver to aquatic organisms. *Environ Toxicol Chem* 17:572–578.
- Fletcher JS, Johnson FL, McFarlane JC. 1990. Influence of greenhouse versus field testing and taxonomic differences on plant sensitivity to chemical treatment. *Environ Toxicol Chem* 9:769–776.
- Grubbs FE. 1969. Procedures for detecting outlying observations in samples. *Technometrics* 11:1–21.
- Heugens EHW, Jager T, Creyghton R, Kraak MHS, Hendriks AJ, van Straalen NM, Admiraal W. 2003. Temperature-dependent effects of cadmium on *Daphnia magna*: Accumulation versus sensitivity. *Environ Sci Technol* 37:2145–2151.
- Howe GE, Marking LL, Bills TD, Rach JJ, Mayer F. 1994. Effects of water temperature and pH on toxicity of terbufos, trichlorfon, 4-nitrophenol and 2,4-dinitrophenol to the amphipod *Gammarus pseudolimnaeus* and rainbow trout *Oncorhynchus mykiss*. *Environ Toxicol Chem* 13:51–66.
- Kwok KWH, Leung KMY. 2005. Toxicity of antifouling biocides to the intertidal harpacticoid copepod *Tigriopus japonicus* (Crustacea, Copepoda): Effects of temperature and salinity. *Mar Pollut Bull* 51:830–837.
- Kwok KWH, Leung KMY, Lui GSG, Chu VKH, Lam PKS, Morrirt D, Maltby L, Brock TCM, Van den Brink PJ, Warne MSJ, Crane M. 2007. Comparison of tropical and temperate freshwater animal species' acute sensitivities to chemicals: Implications for deriving safe extrapolation factors. *Integr Environ Assess Manag* 3:49–67.
- Lacher TE, Goldstein MI. 1997. Tropical ecotoxicology: status and needs. *Environ Toxicol Chem* 16:100–111.
- Leung KMY, Taylor AC, Furness RW. 2000. Temperature-dependent physiological responses of the dogwhelk *Nucella lapillus* to cadmium exposure. *J Mar Biol Assoc UK* 80:647–660.
- Leung KMY, Morrirt D, Wheeler JR, Whitehouse P, Sorokin N, Toy R, Holt M, Crane M. 2001. Can saltwater toxicity be predicted from freshwater data? *Mar Pollut Bull* 42:1007–1013.
- Magallona ED. 1994. Impact of pesticides on tropical ecosystems with emphasis on rice production in the Asian countries. In: Widianarko B, Vink K, van Straalen NM, editors. Environmental toxicology in South East Asia. Amsterdam (NL): University Pr. p 19–31.
- Maltby L, Blake N, Brock TC, van den Brink PJ. 2005. Insecticide species sensitivity distributions: Importance of test species selection and relevance to aquatic ecosystems. *Environ Toxicol Chem* 24:379–388.
- McDonald DG, Reader JP, Dalziel TRK. 1989. The combined effects of pH and trace metals on fish ionoregulation. In: Morris R, Taylor EW, Brown DJA, Brown JA, editors. Acid toxicity and aquatic animals. Cambridge (MA): Cambridge Univ Pr. p 221–242.
- [OECD] Organisation for Economic Co-operation and Development. 1995. Guidance document for aquatic effects assessment. Paris (FR). OECD Environment Monographs 92.
- Ontario Ministry of Environment. 1984. Water management: Goals, policies, objectives and implementation procedures of the Ministry of Environment. Toronto (ON).
- Park EJ, Jo HJ, Jung J. 2009. Combined effects of pH, hardness and dissolved organic carbon on acute metal toxicity to *Daphnia magna*. *J Ind Eng Chem* 15:82–85.
- Pörtner HO. 2002. Climate variations and the physiological basis of temperature dependent biogeography: Systemic to molecular hierarchy of thermal tolerance in animals. *Com Biochem Phys A* 132:739–761.
- Pynnönen K. 1995. Effect of pH, hardness and maternal pre-exposure on the toxicity of Cd, Cu and Zn to the glochidial larvae of a freshwater clam *Anodonta cygnea*. *Water Res* 29:247–254.
- SAS Version 9.3. 2011. The data analysis for this paper was generated using SAS software, Version 9.3 of the SAS System for Windows. Cary (NC): SAS Institute Inc.
- Sloof W, Van Oers JAM, de Zwar D. 1986. Margins of uncertainty in ecotoxicological hazard assessment. *Environ Toxicol Chem* 5:841–852.
- Sokal RR, Rohlf FJ. 2012. Biometry: The principles and practice of statistics in biological research. 4th ed. New York: W.H. Freeman and Co. p 937.
- Sprague JB. 1985. Factors that modify toxicity. In: Rand GM, Petrocelli SR, editors. Fundamentals of aquatic toxicology. Bristol (UK): Taylor & Francis. p 124–163.
- SPSS version 23. IBM Corp. Released 2015. IBM SPSS Statistics for Windows, Version 23.0. Armonk (NY): IBM Corp.
- Tietjen GL, Moore RH. 1972. Some Grubbs-type statistics for the detection of several outliers. *Technometrics* 14:583–597.
- [USEPA] US Environmental Protection Agency. 1985. 40 CFR Part 141. National primary drinking water regulations. *Fed Regist* 50:46944–46946.
- [USEPA] US Environmental Protection Agency. 1991. Technical support document for water quality-based toxics control. Washington (DC). EPA/505/2-90-001. p 315.
- Wang Z, Kwok KWH, Lui GCS, Zhou GJ, Lee JS, Lam MHW, Leung KMY. 2014. The difference between temperate and tropical saltwater species' acute sensitivity to chemicals is relatively small. *Chemosphere* 105:31–43.
- Wang Z, Leung KMY. 2015. Effects of unionised ammonia on tropical freshwater organisms: Implications on temperate-to-tropic extrapolation and water quality guidelines. *Environ Pollut* 205:240–249.
- Wang Z, Meador JP, Leung KMY. 2016. Metal toxicity to freshwater organisms as a function of pH: A meta-analysis. *Chemosphere* 144:1544–1552.
- Wepener V, Chapman PM. 2012. South African ecotoxicology – Present status and future prognosis. *Afr J Aquat Sci* 37:229–234.
- Wheeler JR, Grist EPM, Leung KMY, Morrirt D, Crane M. 2002. Species sensitivity distributions: Data and model choice. *Mar Pollut Bull* 45:192–202.
- Wheeler JR, Leung KMY, Morrirt D, Sorokin N, Rogers H, Toy R, Hold M, Whitehouse P, Crane M. 2002. Freshwater to saltwater toxicity extrapolation using species sensitivity distributions. *Environ Toxicol Chem* 21:2459–2467.