NIWA
Taihoro Nukurangi

ANZECC methodology used for freshwater copper and zinc guideline value determinations

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Executive summary

The Australian and New Zealand Environment and Conservation Council and Agriculture and Resource Management Council of Australia and New Zealand (ANZECC) guidelines were released in 2000 and are currently in the process of being revised and updated. Revised procedures to derive updated ANZECC guideline values (GVs) for marine and freshwaters provide the basis for the methodology.

This report addresses the following issues relating to the updating of copper and zinc guidelines for marine and freshwaters:

1. whether corrections for water hardness are appropriate for copper in freshwaters
2. maximum metals concentrations which should be acceptable for data acceptability
3. the suitability of the use of the Biotic Ligand Model (BLM) for use in derivation of copper guidelines for marine and freshwaters
4. the use of time-response data for establishing stormwater effects guidelines, and
5. the implication for regulatory policy of a bioavailability-driven guidelines approach.

1 Background

The Australian and New Zealand Environment and Conservation Council and Agriculture and Resource Management Council of Australia and New Zealand (ANZECC) guidelines were released in 2000 [1] and are currently in the process of being revised and updated. Revised procedures to derive updated ANZECC guideline values (GVs) for marine and freshwaters provide the basis for the methodology [2, 3].

This project seeks to update the ANZECC GVs for copper (Cu) and zinc (Zn) in marine and freshwaters following the updated guideline procedures. The ANZECC (2000) freshwater GVs included toxicity modifiers, such as pH for ammonia and water hardness for a number of metals, including copper and zinc. The hardness correction used in the 2000 derivation followed that used by the US EPA in their guideline procedures [4, 5] with all of the toxicity test endpoints values adjusted to a standard hardness value of 30 mg $CaCO_3$ L^{-1}, considered appropriate for the generally soft waters present in Australian and New Zealand. These were largely based on acute toxicity data for fish, but were being applied to modify GVs based on chronic toxicity data. The updated guidance indicates that no hardness adjustment should be undertaken for copper, but that hardness adjustment should be incorporated for other hardness-sensitive metals (cadmium, chromium, lead, nickel and zinc) [2].

In recent years an alternative methodology for guideline derivation internationally has been to use biotic ligand models (BLMs) for metals (including copper, nickel, silver, zinc and manganese) for toxicity prediction after incorporating the effects of the water quality modifying factors [2]. The central notion is that organisms possess a key binding site – the biotic ligand – that interacts with the toxic metal and with other cationic components of the water. These models take solution complexation into account, and also competition by protons and base cations (Na^+, Mg^{2+}, K^+, Ca^{2+}) and binding with dissolved organic carbon (DOC) over a range of pH conditions, by defining and determining equilibrium constants for the reactions of these species, as well as the metal of interest, with the biotic ligand. These derivation procedures are complex and a suite of BLM binding equations must be developed for acute and chronic toxicity data for key species groups (i.e., fish, invertebrates and algae). The multiple BLM equations can then be combined for application to deriving water quality criteria (WQC).

Development of BLMs has so far been based almost entirely on data from laboratory studies. In the US, the Environmental Protection Agency (US EPA) has used a BLM to derive water quality criteria for copper in freshwaters [6]. More recently the BLM has been used to derive predicted no effect concentration (PNEC) values for Europe [7, 8] and the US EPA have released a draft BLM for copper in marine waters [9]. The BLM calculations can be used to adjust toxicity values to a standardised 'normalised' set of water quality conditions prior to deriving water quality GVs, and to make site-specific adjustments to generic WQC to specific local water quality conditions. However, for copper, the US and European BLMs differ in their origin and incorporate different predictive equations. As such, they predict different responses to water quality modifiers, thus requiring specific consideration as to the appropriateness of their use.

The objective to this report is to address issues relating to the selection criteria used for inclusion of marine and freshwater data for the derivation of copper and zinc GV following the updated guidance [2, 3] [1].

[1] These guidance documents were updated in May 2017. Changes are detailed in each of the documents.

Major issues include:

(i) whether corrections for water hardness are appropriate for copper in freshwaters

(ii) maximum metal concentrations that should be acceptable for data acceptability

(iii) the suitability of the use of the Biotic Ligand Model (BLM) for use in derivation of copper guidelines for marine and freshwaters

(iv) the use of time-response data for establishing stormwater effects guidelines

(v) and implication for regulatory policy of a bioavailability-driven guidelines approach.

2 Responses to Major Issues

2.1 Whether corrections for water hardness are appropriate for copper in freshwaters

Water quality ranges widely in Australia and New Zealand for a number of the modifiers affecting toxicity of copper, and other metals, to freshwater species (Table 2-1). Thus, site-specific predictions of WQGVs for a wide range of conditions is ultimately required for water quality management. As stated, the US and EU no longer use a hardness-correction for copper water quality GVs, instead using a BLM which incorporates the effects of hardness along with other water quality modifiers.

We considered that it would be desirable to use this standardised water quality approach prior to undertaking our GV derivations and thus internationally harmonise the applicable BLMs for subsequent site-specific applications. Both the US and the EU Cu-BLMs apply the BLM equations to all laboratory toxicity data to generate a 'normalised' water quality dataset prior to deriving their acute (US only) and chronic WQGs (US and EU). The procedures for the general BLM derivations differ between the two jurisdictions and are summarised in Appendix A.

We evaluated the sensitivity of the US and EU Cu-BLM models to a representative range of water quality parameters for Australian and New Zealand freshwaters (Table 2-1). The US EPA BLM is positively influenced by both water hardness and DOC (Figure 2-1), with the predicted chronic criterion value increasing by 43% as hardness increased from 23 to 100 mg $CaCO_3$ L^{-1} (Table 2-2, CCC values).

In contrast, the European Cu-BLM showed a 27% decrease in the predicted copper guideline value (HC5 (50%)) with a 10x increase in Ca concentration (i.e., a negative response) (Table 2-3). This result was surprising given that the basis of the Cu-BLM is the Hydroqual model as used for the US EPA criteria development (Appendix B). We also evaluated an alternate simplified BLM model, Bio-met, which predicts copper, nickel, and zinc responses to water quality parameters. The Bio-met model [10] also predicted a slight decrease in copper toxicity (19%) with a 10x increase in calcium concentration and otherwise constant pH and DOC conditions (Table 2-4).

All BLM models predict a strong reduction in copper toxicity with increasing DOC concentration and moderate non-linear changes in copper toxicity in response to pH change.

We provided these various predictive outputs to the developers of Bio-met (Adam Peters, wca environment Limited) and Cu-BLM (Stijn Baken, European Copper Institute) for comment on the apparent discrepancy between the BLM models. Their responses highlighted the differing origins of the US and European BLMs, based on acute and chronic toxicity respectively (as outlined in Appendix B) and that the European dataset contains plant toxicity data (micro- and macro-algae) with their representative BLM equations. However, the reasons for the predictive differences were not resolved.

Table 2-1: Ranges for general water quality parameters in Australia and New Zealand. (modified from Batley et al. (2014)[2] to use measured New Zealand dissolved organic carbon (DOC) data)

Parameter	Unit	Range (10th and 90th percentiles)	
		Australia[a]	New Zealand[b]
pH		6.0-7.6	7.2-8.2
Ca	mg Ca L^{-1}	0.4-45	4.2-17.4
Mg	mg L^{-1}	0.5-30	0.7-3.6
Hardness	mg $CaCO_3$ L^{-1}	3-236	16-56
DOC	mg L^{-1}	2-12	0.35-10.5[c]

[a] Data from WCA (2014; pH, n = 6418; Ca, n = 2622; Mg, n = 2653; DOC, n = 5196; hardness, calculated based on Ca and Mg concentrations). It should be noted that the data may not be fully representative of the ranges of these variables in Australian fresh surface waters.

[b] Data from Smith and Maasdam (1984)[11].

[c] DOC for NZ measured for 70 river sites in the National River Water Quality Monitoring Network (NRWQN) (n = 365; median 2.1 mg L^{-1}; Scott et al. (2006) [12]).

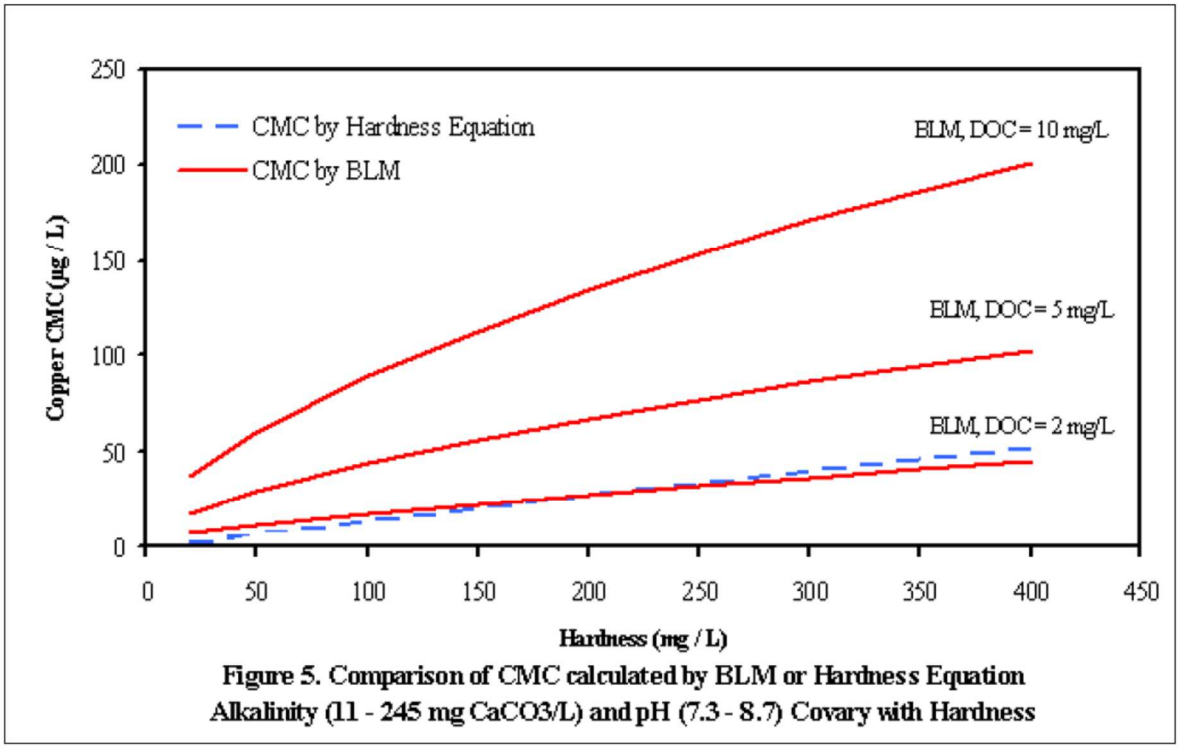


Figure 2-1: Predicted acute toxicity (Criterion Maximum Concentration, CMC) response relationships with DOC and hardness for US BLM [6]. US EPA model (version 2.3.3).

Table 2-2: Predicted acute toxicity (Criterion Maximum Concentration, CMC) and chronic toxicity (Criterion Continuous Concentration, CCC) responses relationships for copper with DOC and hardness for US BLM [6] covering a range representative of Australian and New Zealand waters. US EPA model (version 2.3.3). Shading indicates simulations for different hardness values.

		Final Acute Value (FAV), µg/L	CMC (CMC=FAV/2), µg/L	CCC (CCC=FAV/ACR), µg/L
Hardness 23	pH7, DOC1	134	67	42
Hardness 46	pH7, DOC1	152	76	47
Hardness 100	pH7, DOC1	193	96	60
Hardness 200	pH7, DOC1	268	134	83
Hardness 400	pH7, DOC1	388	194	120
Hardness 100, pH 8	DOC1	309	154	96
Hardness 100, pH 7	DOC1	193	96	60
Hardness 100, pH 6	DOC1	162	81	50
Hardness 100, pH 5	DOC1	185	92	57
Hardness 100, pH 4	DOC1	737	369	229
DOC 2	H100, pH7	272	136	84
DOC 3	H100, pH7	351	175	109
DOC 4	H100, pH7	430	215	134
DOC 5	H100, pH7	509	255	158
DOC 10	H100, pH7	907	454	282

2.1.1 Recommendations for freshwater Cu GV derivations for hardness incorporation:

1. That the toxicity database be restricted to:

    a. Data with measured copper concentrations in exposure solutions (i.e., all nominal data excluded).
    b. Maximum copper concentrations of <0.5 mg L^{-1} for chronic endpoints.
    c. Hardness measurements must be included.
    d. Toxicity data restricted to hardness values <200 mg $CaCO_3$ L^{-1} (i.e., values below the practical upper limit for Australian and New Zealand waters).
    e. Laboratory data only with low ($\leq$ 1 mg L^{-1}) DOC concentrations (i.e., excluding elevated DOC testing data and natural waters testing where DOC may have been elevated).
    f. Test data within the pH range of 6.5 to 8.0.

2. That selected **acute** toxicity data be **hardness adjusted to 30 mg $CaCO_3$ L^{-1}** based on the US EPA hardness slope for low DOC (i.e., 0.5 mg L^{-1})
3. That selected **chronic** toxicity **not be hardness adjusted** prior to guideline derivation

The recommendations above were used in the updating of the Cu freshwater guidelines [13].

2.2 Whether corrections for pH are appropriate for copper in freshwaters

Corrections for pH have been historically ignored in guideline derivations, yet they are a critical control on the free metal ion concentration. For example, free copper concentration decreases by over two orders of magnitude over the pH range of most natural waters (Figure 2-2), being particularly affected in some freshwater systems having high pH and alkalinity, where the proportion of free copper ion is likely to be extremely low [14].

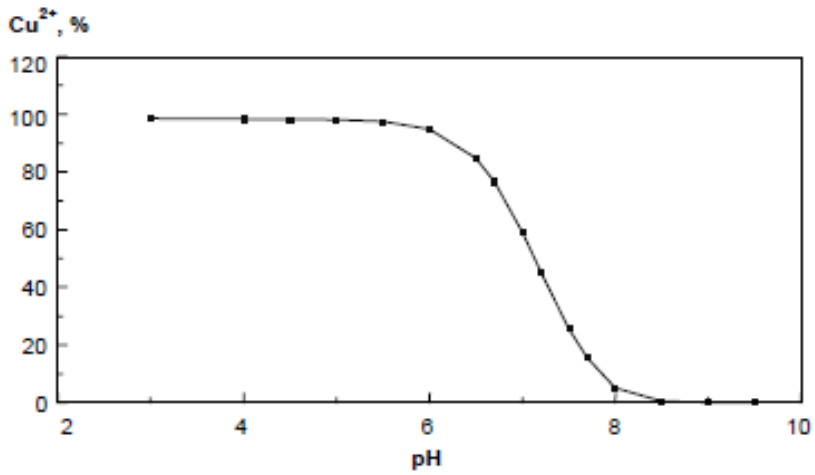
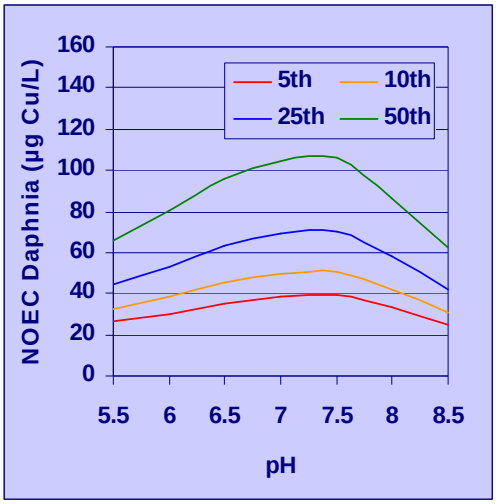


Figure 2-2: Effect of pH on free Cu^{2+} concentration in a freshwater sample. Composition 100 µg L^{-1} total Cu, hardness 30 mg $CaCO_3$ L^{-1} [14].

The ANZECC (2000) [1] GV derivation did not adjust for pH and neither did the US EPA BLM-derived criteria for copper [6]. However, the more recent European Cu-BLM and Bio-met models do incorporate a pH-modifying function based on the results of chronic toxicity testing for fish, invertebrates and algae. An important difference between the chronic fish/invertebrate and algae BLM is related to the differences in sensitivity with changing pH (Figure 2-3). Indeed, an increase of pH for algae results in an increase in toxicity while for invertebrates/fish a decrease in toxicity is observed. These findings on algae seem to be contradictory with earlier observations that metal toxicity is reduced at increased pH because of the reduction of the free metal ion fraction which is considered to be the most bioavailable (and toxic) metal species (Figure 2-2). The results indicate that for algae, copper become less toxic at lower pH. There is growing evidence that competition between hydrogen ions and the free metal ions for binding at the cell surface ligands could be responsible for reduced metal uptake (and toxicity) at lower pH levels [15].



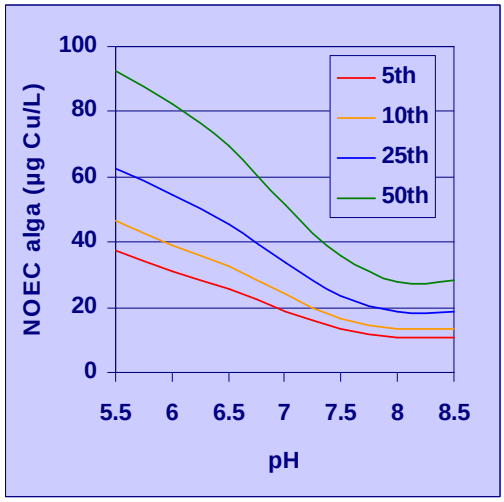


Figure 2-3: Differences in pH sensitivity between algae and invertebrates for different DOC percentiles in EU surface waters. Figure 3.7 from [15].

The incorporation of multiple BLM equations in the Cu-BLM and Bio-met results in a complex pH sensitivity behaviour depending on the more sensitive biological species in the region of the WQC value. Predictions from these models for Australian and New Zealand water quality GV ranges shows marked differences in the predicted GVs. For example, Bio-met predictions for median New Zealand water quality showed a 3x range for pH ranging from 7.1 to 8.4, with the lowest GV at pH 8.4 and the highest at pH 7.7 (Appendix B); while Australian predictions show a 4x range, with the lowest GV at pH 6 and the highest at pH 7.6. These BLM model predictions indicate the important role of pH for establishing site-specific guidelines for copper in natural waters.

The guidance for the updating of the ANZECC GVs did not indicate that pH adjustment should be incorporated into the species sensitivity normalisation prior to GV derivation. This reflects that lack of validation of relevant BLM equations for pH sensitivity of key native Australian and New Zealand species. In the absence of this validation we recommend that, for effectively comparing toxicity data, toxicity test results should include sufficient data to enable calculation of a dissolved free metal ion concentration and, at a later stage, a computed (or measured) bioavailable metal concentration, when the biotic ligand or other appropriate models are more fully developed and validated.

2.2.1 Recommendations for freshwater Cu GV derivations for pH incorporation:

1. That the toxicity database be restricted to:

    a. Data with measured pH values reported for the tests.
    b. Toxicity endpoints in the pH range from 6.5 to 8.0.

Table 2-3: Cu-BLM predictions of HC5 (hazardous concentration for 5% of species) for representative water quality modifier concentrations. (Cu PNEC estimator version 1.3.2).

Input			required	required	required										Output			
Site Name	Sample Name	Temp.	pH	DOC	Ca	Mg	Na	K	SO4	Cl	Alkalinity	Cu	HA	S	HC5(5%)	HC5(50%)	HC5(95%)	BioF
		°C		mg C/L	mg/L	mg/L	mg/L	mg/L	mg/L	mg/L	mg/L CaCO3	µg/L	%	mg/L	µg Cu/L	µg Cu/L	µg Cu/L	
Median	NZ median_all parameters		7.7	2.1	7.70E+00										7.272234	10.87484	14.56332	1.39421
5 percentile	NZ 5 percentile_ all parameters		7.1	0.2	7.10E+00										0.520384	0.882043	1.293659	0.113082
95 percentile	NZ percentile_ all parameters		8.4	12	22.9										15.52166	26.5468	39.19027	3.403436
pH range	NZ_median		7.7	2.1	7.7										7.272234	10.87484	14.56332	1.39421
pH range	NZ_5 percentile		7.1	2.1	7.7										6.933959	10.25071	13.61368	1.314193
pH range	NZ_95 percentile		8.4	2.1	7.7										2.494211	4.478279	6.848484	0.574138
Calcium range	NZ_median		7.7	2.1	7.70E+00										7.272234	10.87484	14.56332	1.39421
Calcium range	NZ_5 percentile		7.7	2.1	7.10E+00										7.371891	10.99404	14.69405	1.409493
Calcium range	NZ_95 percentile		7.7	2.1	22.9										6.003162	9.384966	12.98007	1.203201
DOC range	NZ_median		8.4	2.1	7.7										2.494211	4.478279	6.848484	0.574138
DOC range	NZ_5 percentile		8.4	0.2	7.7										0.221054	0.501056	0.907462	0.064238
DOC range	NZ_95 percentile		8.4	12	7.7										18.76928	30.75518	44.01311	3.942972
DOC range_low calcium	NZ_median		7.7	0.2	7.1										0.489991	0.89909	1.396805	0.115268
DOC range_low calcium	NZ_5 percentile		7.7	2.1	7.1										6.003162	9.384966	12.98007	1.203201
DOC range_low calcium	NZ_95 percentile		7.7	12	7.1										41.77292	61.28996	80.95276	7.857687
DOC range_high calcium	NZ_median		7.7	0.2	22.9										0.514969	0.875282	1.28632	0.112216
DOC range_high calcium	NZ_5 percentile		7.7	2.1	22.9										6.933959	10.25071	13.61368	1.314193
DOC range_high calcium	NZ_95 percentile		7.7	12	22.9										44.59439	64.05363	83.30791	8.212004
Calcium range_low pH	NZ_median		7.1	0.2	7.7										0.221054	0.501056	0.907462	0.064238
Calcium range_low pH	NZ_5 percentile		7.1	2.1	7.7										-2.1E+09	-2.1E+09	-2.1E+09	-2.8E+08
Calcium range_low pH	NZ_95 percentile		7.1	12	7.7										-2.1E+09	-2.1E+09	-2.1E+09	-2.8E+08
Calcium range_high pH	NZ_median		8.4	0.2	7.7										-2.1E+09	-2.1E+09	-2.1E+09	-2.8E+08
Calcium range_high pH	NZ_5 percentile		8.4	2.1	7.7										2.494211	4.478279	6.848484	0.574138
Calcium range_high pH	NZ_95 percentile		8.4	12	7.7										18.76928	30.75518	44.01311	3.942972
Test_2	NZ large DOC range_pH 8		8	0.2	7.7										0.350618	0.710051	1.184994	0.091032
Test_2	NZ large DOC range_pH 8		8	0.5	7.7										0.969387	1.754863	2.699704	0.224982
Test_2	NZ large DOC range_pH 8		8	1	7.7										2.173102	3.666446	5.359481	0.470057
Test_2	NZ large DOC range_pH 8		8	2	7.7										4.835312	7.744024	10.89994	0.992824
Test_2	NZ large DOC range_pH 8		8	4	7.7										10.59653	16.38132	22.47296	2.100169
Test_2	NZ large DOC range_pH 8		8	6	7.7										16.75116	25.49057	34.57155	3.268022
Test_2	NZ large DOC range_pH 8		8	8	7.7										23.2335	35.00575	47.13575	4.487916
Test_2	NZ large DOC range_pH 8		8	10	7.7										30.01832	44.8967	60.13196	5.755987
Test_2	NZ large DOC range_pH 8		8	12	7.7										37.08151	55.13412	73.52747	7.068477
Test_2	NZ large DOC range_pH 8		8	14	7.7										44.40508	65.69597	87.29762	8.42256
Test_2	NZ large DOC range_pH 8		8	16	7.7										51.98938	76.58249	101.4426	9.818268
Test_2	NZ large DOC range_pH 8		8	20	7.7										67.88911	99.25306	130.7557	12.72475

Table 2-4: Bio-met evaluation for copper and zinc for representative NZ waters. Software version: v3.03 [10]. Note: 'Y' indicates that a parameter is out of the calibration range for the respective BLM so the default GV is used as the predicted EQS value. Red boxed areas indicates calcium range simulations for New Zealand (upper box) and Australian waters (lower box). Median and percentile ranges of water quality variables from Table 2-1, with updated DOC data for NZ from Scott et al. (2006) [12].

							RESULTS (Copper)					RESULTS (Nickel)					RESULTS (Zinc)				
	Optional	Optional	Required	Required	Required	Optional															
ID	Sample Name	Sample Number	pH	DOC [mg/L]	Ca [mg/L]	Zinc ABC Conc (dissolved) [µg/L]	Local EQS (dissolved) [µg/L]	BioF	Bioavailable Copper Conc [µg/L]	RCR	Notes	Local EQS (dissolved) [µg/L]	BioF	Bioavailable Nickel Conc (µg/L)	RCR	Notes	Local EQS (dissolved) [µg/L]	BioF	Bioavailable Zinc Conc (µg/L)	RCR	Notes
1	Median	NZ_median	7.7	2.1	7.7	1	10.88	0.09				5.56	0.72				20.45	0.53	0.00		
2	5 percentile	NZ_5 percentile	7.1	0.2	7.1	1	**1.00**	**1.00**			Y	6.30	0.64				11.30	0.32	0.00		
3	95 percentile	NZ_95 percentile	8.4	12	22.9	1	24.67	0.04				11.57	0.35				34.55	0.12	0.00		
4	pH range	NZ_median	7.7	2.1	7.7	1	10.88	0.09				5.56	0.72				20.45	0.53	0.00		
5	pH range	NZ_5 percentile	7.1	2.1	7.7	1	10.17	0.10				3.10	0.44				15.84	0.69	0.00		
6	pH range	NZ_95 percentile	8.4	2.1	7.7	1	4.14	0.24				**4.00**	**1.00**			Y	26.18	0.42	0.00		
7	DOC range	NZ_median	7.7	2.1	7.7	1	10.88	0.09				5.56	0.72				20.45	0.53	0.00		
8	DOC range	NZ_5 percentile	7.7	0.2	7.7	1	**1.00**	**1.00**			Y	**4.00**	**1.00**			Y	11.30	0.32	0.00		
9	DOC range	NZ_95 percentile	7.7	12	7.7	1	70.33	0.01				20.53	0.19				108.44	0.10	0.00		
10	Calcium range	NZ_median	7.7	2.1	7.7	1	10.88	0.09				5.56	0.72				20.45	0.53	0.00		
11	Calcium range	NZ_5 percentile	7.7	2.1	7.1	1	10.88	0.09				5.56	0.72				20.45	0.53	0.00		
12	Calcium range	NZ_95 percentile	7.7	2.1	22.9	1	10.88	0.09				5.56	0.72				20.69	0.53	0.00		
13	Median_Au	Australia_median	7	5	30	1	17.56	0.06				12.65	0.32				26.35	0.40	0.00		
14	10 percentile	Australia_10 percentile	6	2	1	1	**1.00**	**1.00**			Y	**11.83**	**0.34**			Y	**17.75**	**0.61**	**0.00**		Y
15	90 percentile	Australia_90 percentile	7.6	12	60	1	51.81	0.02				21.25	0.19				72.43	0.15	0.00		
16	pH range	Australia_median	7	5	30	1	17.56	0.06				12.65	0.32				26.35	0.40	0.00		
17	pH range	Australia_10 percentile	6	5	30	1	4.56	0.22				**14.26**	**0.28**			Y	27.12	0.40	0.00		
18	pH range	Australia_90 percentile	7.6	5	30	1	19.26	0.05				3.30	0.43				31.38	0.35	0.00		
19	DOC range	Australia_median	7	5	30	1	17.56	0.06				12.65	0.32				26.35	0.40	0.00		
20	DOC range	Australia_10 percentile	7	2	30	1	5.97	0.17				3.67	0.41				20.47	0.53	0.00		
21	DOC range	Australia_90 percentile	7	12	30	1	36.18	0.03				23.76	0.17				48.48	0.22	0.00		
22	Calcium range	Australia_median	7	5	30	1	17.56	0.06				12.65	0.32				26.35	0.40	0.00		
23	Calcium range	Australia_10 percentile	7	5	1	1	**1.00**	**1.00**			Y	**12.65**	**0.32**			Y	**37.41**	**0.29**	**0.00**		Y
24	Calcium range	Australia_90 percentile	7	5	60	1	16.31	0.06				12.65	0.32				31.36	0.35	0.00		

2.3 Whether corrections for water hardness and other parameters is appropriate for zinc in freshwaters

The Zn^{2+} species is considered the most important form for toxicity. The concentration of Zn^{2+} in a waterbody is affected by the physico-chemical water quality, including pH, hardness, alkalinity, dissolved organic matter and suspended particulate matter, all of which affect the speciation of zinc. In addition to effects on speciation, water chemistry can also affect bioavailability and toxicity through competition between zinc and other cations with biotic ligands of organisms (see discussion on BLM below).

The influence of water hardness (due to calcium and magnesium) on zinc toxicity is probably the best studied. The toxicity of zinc is generally decreases as water hardness increases and is attributed to competition between zinc and calcium cations for binding sites on biological tissues [16, 17]. This occurs for algae, invertebrates and fish and in both acute and chronic toxicity tests, though there are far more data documenting this relationship in acute tests.

The pH of a waterbody can affect zinc toxicity in antagonistic ways: firstly, by influencing zinc speciation with higher concentrations of Zn^{2+} at low pH (increasing toxicity); and secondly by influencing binding to biotic ligands through direct competition (decreasing toxicity at low pH) and through modifying the affinity between zinc and membrane binding sites (decreasing toxicity at low pH). Many studies conducted with varying pH and no ligands (used artificial or filtered water) have indeed shown the expected increasing toxicity with increasing pH, including studies on *Oncorhynchus mykiss* [18, 19]; *Pimephales promelas* [20, 21]; *Daphnia magna* [22], *Ceriodaphnia dubia* [21, 23]; two green algal species: *Pseudokirchneriella subcapitata* [24] and *Chlorella* sp. [25]; however in other studies the reverse or no effect has been found [26, 27].

Alkalinity (usually due to carbonate) affects zinc toxicity by reducing the concentrations of Zn^{2+} in water. In general, studies have shown lower toxicity at higher alkalinity, however, in most cases the hardness of the water and also the pH vary alongside the alkalinity, and changes in toxicity may be attributable to these characteristics. Of the few studies that compared alkalinity while maintaining constant water hardness, two showed alkalinity had no influence on toxicity to rainbow trout at or below pH 7 [16, 28], whilst a third study suggested that both hardness and alkalinity were important influences on the toxicity to rainbow trout and brook trout [29].

Dissolved organic matter (DOM) affects zinc toxicity primarily through formation of zinc complexes which are of low bioavailability, thus reducing the toxicity of zinc in waters with high DOM. This has been observed in acute toxicity studies using various cladoceran species [30-33], fathead minnow larvae [34] and for the green algae *P. subcapitata* [24]. DOM also reduces toxicity in chronic tests as shown for *Daphnia* sp. [27, 35]. Its effect appears to be strongest in waters of soft to moderate hardness (< 200 mg L^{-1} as $CaCO_3$)[35] and at concentrations > 5-10 mg L^{-1} of DOM, though some protective effect of DOM at 1.5 mg L^{-1} of humic acids has been shown [30].

Biotic ligand models (BLMs) have been developed to predict the combined effect of these water quality characteristics (hardness, pH, DOC) on zinc bioavailability and toxicity. The US EPA was the first to incorporate a BLM into their water quality criteria, for the 2007 ambient freshwater water quality criteria for copper [6], based on a BLM derived from acute toxicity data. This has been followed by the European Union in their risk assessments for copper [36], nickel [37] and zinc [38], all of which use chronic BLMs to derive chronic "Predicted No Effect Concentration" (PNEC) guidelines.

BLM models have been developed for zinc and a comprehensive risk assessment completed for zinc in European waters [39]. The Bio-met model [10] provides EQS predictions for Zn based on input of key water quality modifiers (pH, DOC, Ca). Assessment of the Bio-met model for a range of Australian and New Zealand water quality conditions showed a strong sensitivity to DOC, moderate sensitivity to pH and no sensitivity to hardness (Table 2-4). Thus, like copper, the Bio-met model makes no hardness-related GV adjustment. This is not the case for the most recently updated Environment Canada GV.

Environment Canada has recently developed draft updated guidelines for zinc in freshwaters [40]. The short-term GV incorporates adjustment for hardness and DOC, while the long-term GV incorporates hardness and pH adjustment. The hardness slopes used differ between the short- and long-term GVs. Their long-term guideline incorporates hardness and pH adjustment based on multiple linear regression analyses for different aquatic species[40]. Using this approach the Canadian guidelines generate a matrix table for each GV. The relationship for their long-term GV and hardness and pH is shown in Figure 2-4.

For the ANZECC guideline revision, data was collated from studies with multiple toxicity values derived from tests in differing concentrations of hardness and DOC and pH [41]. EC50 values were used as these are more rigorous statistical estimates of toxicity than EC10 values or NOEC, which are greatly affected by the test concentrations used in the toxicity study. Data was available for six species from 10 different studies (see Appendix C). Multiple linear regression was used to examine the influence of hardness, pH and DOC for each of the 12 combination of species, endpoint and study (Table 2-5). The slopes of these models (where statistically significant) ranged from -0.49 to -3.0 for pH and 0.64 to 1.4 for ln(hardness). The slopes were pooled using a geometric mean to provide an overall slope of -1.02 for pH and 0.77 for ln(hardness). This leads to equation as follows [41]:

$$\text{Standardised EC10/NOEC} = \exp[\ln(\text{EC10/NOEC}_{meas}) - 0.81(\ln[\text{hardness}_{meas}] - \ln[30]) + 1.29(\text{pH}_{meas} - 8.0)$$

All toxicity data was adjusted to a consistent hardness of 30 mg L^{-1} and pH of 8.0, based on exponential slopes for hardness of 0.766 and for pH of -1.02. These slopes were derived from multiple linear regression analyses of chronic toxicity data (EC50s) and water quality factors for six species.

2.3.1 Recommendations for freshwater Zn GV derivations for hardness incorporation:

1. That the toxicity database be restricted to:

    a. Data with measured zinc concentrations in exposure solutions (i.e., all nominal data excluded).
    b. Maximum zinc concentrations of <10.0 mg L^{-1} for chronic endpoints.
    c. Hardness measurements must be included.
    d. Toxicity data restricted to hardness values <200 mg $CaCO_3$ L^{-1} (i.e., values below the practical upper limit for Australian and New Zealand waters).
    e. Laboratory data only with low ($\leq$ 1 mg L^{-1}) DOC concentrations) (i.e., excluding elevated DOC testing data and natural waters testing where DOC may have been elevated).
    f. Test data within the pH range of 6.5 to 8.0.

2. That selected **acute** toxicity data be **hardness adjusted to 30 mg $CaCO_3$ L^{-1}** based on the US EPA hardness slope for low DOC (i.e., 0.5 mg L^{-1}).

3. That selected **chronic** toxicity data be **hardness adjusted to 30 mg $CaCO_3$ L^{-1}** prior to GV derivation based on a regression-derived hardness slope for low DOC (i.e., 0.5 mg L^{-1}).

4. That selected **chronic** toxicity data be **pH adjusted to 8.0** prior to GV derivation.

Table 2-5: Slopes for zinc toxicity for hardness from multiple linear regression models for each species / study combination.

	Species	Study	Slopes for ln(EC50 conc) vs		
			pH	Ln (Hardness)	Ln (DOC)
Fish	*O. mykiss*	253	-0.741 °	0.540 **	NA
	O. mykiss	254	-0.936 **	1.41 ***	0.353 **
	P. promelas	121	0.486	1.96 °	No data
Invertebrates	*D. magna*	234	0.155	0.0671	0.514 ***
	D. magna	254	-0.487 *	0.639 **	0.231 *
	D. magna	128	-0.433 ***	0.508 ***	No data
	C. dubia	101	0.596	No data	No data
	C. dubia	107	0.705	No data	No data
Algae	*P. subcapitata*	223	-1.45 ***	0.119	NA
	P. subcapitata	233	-3.00	0.747 °	-0.391
	P. subcapitata	254	-2.49 *	1.07 *	0.960 **
	Chlorella sp.	271	-1.56 **	No data	No data
Geomean (of significant slopes)			**-1.2**	**0.85**	**0.45**
Geomean (of significant slopes, including additional D. magna study)			**-1.02**	**0.766**	**0.448**

Note: Level of statistical significance shown by symbol after slope as follows:
p-value 0 < *** < 0.001 ** < 0.01 * < 0.05 ° < 0.1.

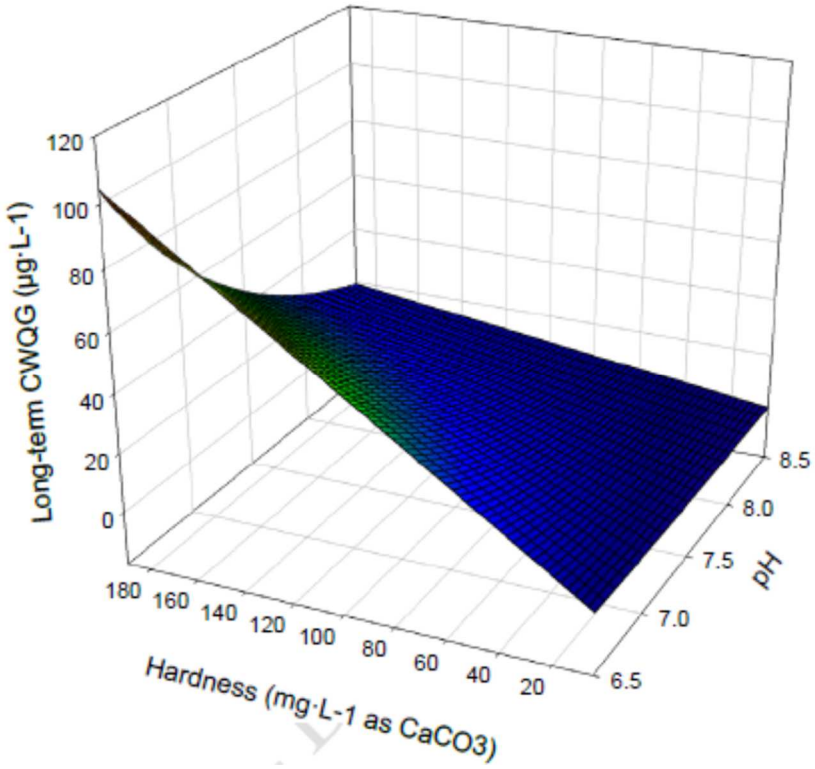


Figure 2-4: Canadian long-term chronic water quality guidelines for zinc as a function of hardness and pH. (from [40]).

2.4 Maximum metals concentrations which should be acceptable for data acceptability

Appropriate quality assurance and quality control procedures are considered an essential for component of acceptability testing both from a chemical and a biological perspective [14]. Specific chemical considerations include: validation of chemical concentrations; solubility maxima and choice of test containers.

It is important to recognize that most metal salt solutions are weakly acidic; however, at the pH of most natural waters (6.0-8.5) hydrolysis occurs and, depending on both the pH and alkalinity, usually leads to the formation of either hydroxyl- or carbonate species [14]. When the solubility limit is exceeded, colloidal and precipitated species may form. The soluble metal concentrations could be limited to values as low as 500 µg L^{-1} (Table 2-6). These will be particularly influences in natural waters by the presence, due to the very low solubility of dissolved iron, of colloidal iron (III) hydroxides which will adsorb dissolved copper in solution.

Table 2-6: Speciation of copper calculated from MINITEQA2. (from [14]).

pH	Hardness (mg L^{-1} $CaCO_3$)	Total Cu (µg L^{-1})	Soluble Cu (% of total)	Cu^{2+} (% of soluble)
7.0	100	500	100	37.1
7.0	30	500	100	58.7
7.5	100	500	80.8	13.6
7.5	30	500	39.8	25.5
7.5	100	5000	8.1	13.6
7.5	30	5000	4.0	19.3

We are not aware of any rigorous basis for determining the upper concentration limit for copper and zinc in marine and freshwaters. Short-term (acute) toxicity data may have higher acceptable maximum concentrations because of the time required for some metals to precipitate as hydroxy complexes at natural water pH and alkalinity levels. As part of the recommendations in the revised guidelines [2, 3], data at concentrations more than twice the solubility limit should be excluded from the GV derivation, although in some cases particulate forms can themselves exert toxicity so the wisdom of this is questionable (Table 2-7).

Table 2-7: Recommended upper concentration limits for data acceptability for chronic marine and freshwater toxicity tests.

Element	Freshwater	Marine
Copper	500 µg L^{-1}	700 µg L^{-1}
Zinc	10 mg L^{-1}	9 mg L^{-1}

2.4.1 Recommendations for freshwater Cu and Zn GV derivations for maximum concentrations:

1. That the toxicity database be restricted to endpoints with the:

    a. Maximum freshwater copper concentrations of <0.5 mg L^{-1} for chronic endpoints.

    b. Maximum freshwater zinc concentrations of <10 mg L^{-1} for chronic endpoints.

2.6 The suitability of the use of the Biotic Ligand Model (BLM) for use in derivation of copper guideline values for marine and freshwaters

2.6.1 Freshwater BLMs

The BLM approach has matured to the extent of being incorporated in regulatory freshwater criteria derivation in two international jurisdictions for copper, and in Europe for both copper and zinc. The development of the BLM approach is important both to facilitate normalisation of toxicity data to a standard water quality prior to guideline derivation, and to allow robust site-specific applications of the GV incorporating the effects of the key water quality modifiers.

The science behind the BLM derivation has improved the mechanistic understanding for metal toxicity in many of the species which have been studied. Most of the early studies to derive the hardness-dependent algorithms have confounded the effects of true water hardness (calcium and/or magnesium) with alkalinity (carbonate concentration), since an increase in calcium or magnesium is often associated with an increase in alkalinity [42]. While the effect of calcium and magnesium is accepted as directly affecting copper uptake, and hence toxicity, alkalinity affects copper speciation in solution, which will impact copper bioavailability [43]. These mechanisms have been uncoupled for copper in order to derive the BLM models for regulatory application. However, because of the different basis for the derivation of the US and European BLM models for copper (i.e., based on acute and chronic data respectively, as addressed in Section 2.1) there are differences in chronic guideline values predicted for differing 'true' hardness levels. Differences also exist in the toxicological responses of organisms to metals at differing pH levels (as discussed in Section 2.2). The adjustment of organism sensitivity to a standard pH has been incorporated in the European procedures but has not been adopted in other jurisdictions. This is important.

Canada is presently updating their freshwater guidelines for zinc and have published a draft document for discussion [44]. Their approach does not use a Zn BLM but does incorporate water quality modifier relationships for hardness, pH and DOC (see Section 2.3).

A major study was undertaken in the UK in 2008 to evaluate the adequacy of environmental quality standards (EQS) based on total metal concentrations and laboratory toxicity data for providing protection for field communities [8]. This study undertook field sampling of metal-contaminated streams to establish quantitative relationships between chemical and biological variables. The study concluded that: '*By showing that metal toxicity operates in the field, with dose–response relationships, and in line with bioavailability and chemical speciation concepts, the study provides support for metals regulation through EQSs. It further shows that targeted fieldwork can provide the information necessary for EQS evaluation and modification*.' The findings demonstrated that metals do affect freshwater ecosystems, and that these effects can be at least partially quantified in terms of bioavailability and chemical speciation.

The complexity of the BLM models, requiring the application of differing species-specific BLM binding coefficients, and calibrations over a wide range of water quality modifiers (i.e., DOC, Ca, Mg, pH) requires an intensive calibration effort and multi-parameter mathematical calculations for implementation. Presently the BLM models are limited to single metals and do not handle metal mixtures which are commonly present at contaminated sites.

We consider that a critical consideration of the incorporation of water quality modifiers in the guideline procedure is essential. That these modifiers may have different response relationships for different taxonomic groups and with relationships depending on the duration of test exposures (i.e.,

whether acute or chronic) adds significant complexity to normalising toxicity data to a standard water quality condition. The alternatives to application of a BLM modifier approach are to censor the toxicity data in relation to the modifiers – in order to bound variability which might be associated with that parameter – or in some cases to exclude data where there is a high likelihood that strong effects will be occurring (e.g., DOC).

The uncertainty of the BLM model predictions for copper do not favour implementation for GV derivation at this time. Rather, the database for GV derivation should be based on bounded data for key water quality modifiers so as to limit variability attributable to water quality modifiers.

2.6.2 Marine BLMs

The recent draft US EPA aquatic life estuarine/marine water quality criteria (WQC) for copper uses a BLM to calculate a WQC at site-specific concentrations of DOC, pH, salinity and temperature [9]. Of these four factors, DOC has the most influence in determining the WQC, and has an approximately 1:1 relationship whereby the WQC approximately doubles at DOC doubles. This and other studies have highlighted the importance of incorporating DOC as a major bioavailability modifier for toxicity of copper in marine waters.

Maycock et al. (2011) [45] developed a relationship between copper ecotoxicity to marine organisms and DOC in order to give a biologically relevant environmental metric of copper exposure. Using the DOC correction relationship each individual NOEC/L(E)C10 value was normalised to a predefined DOC concentration of 0.5 mg L^{-1} active DOC (equivalent to 1 mg L^{-1} measured DOC in natural seawater) and used to construct a reference species sensitivity distribution. An HC5 of 2.64 µg Cu L^{-1} was generated their reassessment of the available data.

Based on the review of the available data we recommend that all data used for copper guideline derivation be limited to DOC concentrations of less than 1 mg L^{-1}. Field data should be adjusted for DOC concentration prior to comparison with the GVs. The US EPA copper marine guideline values vary with DOC and are calculated using a linear slope of 1.24 in their BLM [9]. This DOC adjustment procedure is incorporated into the guideline derivation procedure used for this update for Cu in marine waters[46].

Recommendation

1. That the selected **chronic** toxicity data be DOC adjusted to 0.5 mg L^{-1} prior to GV derivation based on a regression-derived DOC slope of 1.24
2. The final GVs should provide GVs for a range of potential receiving water concentrations and the equation to permit site-specific GV calculation

2.7 The use of time-response data for establishing stormwater effects guidelines

The recommended procedures for updating the ANZECC guidelines involve categorising species into test durations for acute and chronic exposure periods – with the durations being dependent on test species and their life-stages [3]. The ANZECC guidelines are derived using long-term (chronic) exposure durations, however, other jurisdictions derive both short-term (acute) and long-term (chronic) GVs or criteria (e.g., US EPA Cu, [6]; Environment Canada Zn, [40]).

The updated database being prepared for this project follows the new procedures [3] and will have toxicity data categorised into acute and chronic data. A separate guideline calculation procedure can be undertaken to provide risk-based acute GV values. These acute GVs would be adjusted to a normalised water quality using appropriate adjustment algorithms.

The database will contain a limited number of species which have been tested for a wide range of exposure times. Analysis of these data can provide toxicity time-response thresholds for exposure times more typical of stormwater peak concentration events (e.g., 8 to 24 h), less than the typical acute guideline exposure period (96 h). If suitable time-response data is available, mathematical models for adjusting the acute guideline values to a shorter duration exposure for representative species will be evaluated. Analysis of these data will indicate whether a conservative default acute guideline might be acceptable for management or whether short-term adjustment should be incorporated.

For stormwaters, a metals risk assessment needs to be undertaken to establish the relative importance of water quality modifiers and elevated dissolved metal concentrations in the first flush time period of storm events. As the initial runoff period is likely to have both the highest metal concentrations and also the most elevated DOC concentrations, the combined influence of these components may serve to negate a potential requirement for more complex assessments of event-related stormwater toxicity. Rather, it may be appropriate for stormwaters to base management on chronic guidelines to provide a suitably precautionary approach. An initial comprehensive analysis of metals in New Zealand urban stormwaters has been completed [47]. The gaps analysis from that study identified the need for state of the environment monitoring programmes to include measurements of both total and dissolved metals, together with water quality modifiers (i.e., pH, DOC, Ca, Mg), and to include intensive event monitoring studies. Monitoring programmes undertaken by Auckland Council and Greater Wellington Regional Council have been expanded to provide some of this additional information.

2.8 General guidelines approach

The generic guidance provided for the updating of the ANZECC GVs leaves scope for the application of best professional judgement for inclusion or exclusion of specific data in the derivation database. The large size of the toxicity databases for copper and zinc in marine and freshwaters increases the requirement for implementing a rigorous decision-making process in the guideline derivation procedure.

The process being applied to the copper and zinc derivations is provided in Table 2-8. Of necessity, this process may be refined as the process is progressed and other publications or guidance incorporated in the procedure.

Various exceptions to the decision criteria were made for toxicity data using New Zealand and Australian species, as outlined in Table 2-8. New Zealand species were rigorously identified by comparison to the New Zealand Inventory of Biodiversity [48-50] whilst Australian species were identified either in the text of the scientific paper, or through AlgaeBase [51], WoRMS [52], or a google search.

Table 2-8: Decision criteria summary for data inclusion for freshwater and marine copper and zinc guideline derivation. Base guidance from [3] with specific additional references noted in table.

	Selection criteria	Notes	Reference
1	Data must be from chronic studies		[3]
2	Antarctic studies excluded	Will only be applicable to marine data. Studies remain in database.	
3	Other exclusions: (i) Natural water tests with high DOC; (ii) Marine tests with salinity <5‰ (iii) Publication year exclusions (i.e., >1980)	(i) As DOC concentration strongly affects both copper and zinc toxicity tests with >2 mg L^{-1} DOC are excluded; except for NZ/Australian species where included by best professional judgement (BPJ); (ii) Updated guidance indicates <0.5 ‰ which we consider too low to be considered marine or estuarine; (iii) Guidance is subjected to critical evaluation of specific earlier publications if those include key species or provide defining data for metal toxicity or modifier effects.	[3]
4	Data must have measured concentrations	(i) General: measured concentrations in test chambers, not stock solutions (EU requires this, Canada requires it for primary data but not secondary (both primary and secondary can be used in guideline derivation); (ii) For Australian/NZ data measurement in stock solutions is accepted; (iii) Accept L(E)C10 type data with measurement of stock solutions (based on good concentration/response required to generate data).	
5	Use EC5-20 data preferentially, with NZ/Aus data for other endpoints (NOEC, L(E)C50)	Allow converted NOECs from EC50 for Australian/NZ data to increase inclusion.	
6	If insufficient EC5-20 data, NOEC data included	For copper marine we can derive a GV using chronic, measured, EC5-20 + NZ/Aus data; For zinc marine we can derive a GV using chronic, measured, EC5-20 + NOEC + NZ/Aus data.	
7	If insufficient EC5-20 + NOEC data, converted LOEC /EC50 data included (provided it meets above requirements)		
8	Key species selection	Special consideration for inclusion of high economic, recreational or ecologically important species. Some species groups may be included, if appropriate, with a separate guideline derivation (e.g., corals).	
9	Water quality modifiers: (i) Selected data must report hardness and should report pH and DOC ;	(i) Note that Environment Canada use a default value for DOC for laboratory tests with no measured DOC [44];	This report

	Selection criteria	Notes	Reference
	(ii) No freshwater hardness adjustment for copper for chronic data. Bound selection of toxicity data to <200 mg $CaCO_3$ L^{-1}; (iii) Hardness adjustment of freshwater Zn data to follow Environment Canada procedure; (iv) No pH adjustment for Cu or Zn data. Bound toxicity data selection to range 6.5 to 8.0; (v) Only DOC data for <2 mg L^{-1} acceptable for marine or freshwater (following EU RAR for Zn)	(ii) Review of available information provided in this report; (iii) Hardness slope derived for this study; (iv) pH – no specific guidance; (v) As DOC concentration strongly affects both copper and zinc toxicity tests with >2 mg L^{-1} DOC are excluded; except for NZ/Australian species where included by BPJ (Clearwater et al. glochidia paper where DOC was 2.5 mg/L) Haven't included P. antipodarum where DOC = 4.3 & hardness probably > 200 (Ca based hardness =158 mg/L).	
9	Specific considerations: (i) Outliers (ii) Natural background concentrations for essential nutrients; (iii) Special environments (iv) Study QC reviews	(i) Specific consideration may be required for 'outlier' values, either for single tests or for data included in geometric means. Presently no regulatory decision rules exist for derivation procedures. Chapman (2015, [53]) provided 7 guidelines for developing geomeans for guidelines and includes guidance for outliers; (ii) Specific consideration should be included for background concentrations for essential elements – such as Cu and Zn. Note that Recommended approach is to measure background concentrations and adopt an 80th percentile as the default GV for areas naturally elevated in metal concentrations [54]; (iii) Toxicity data for species inhabiting a wide range of environments included (e.g., freshwater rivers, lakes, groundwaters); (iv) For the large databases, such as for Cu and Zn, the primary quality screening relies on the primary database (e.g., from AQUIRE or Environment Canada), with specific screening being undertaken for the most sensitive 25th percentile of data and for included Australian and New Zealand studies.	[53]

Changed DOC cut-off to be 2 mg L^{-1}, consistent with EU RAR for zinc. This resulted in the inclusion of additional species, as follows:

Species	Author	EC10/NOEC for this study	Effect on GV derivation
Zinc			
Pseudokirchneriella subcapitata	[55]	1.0-12	Additional data for species, from new study, contributes to geomean used in species sensitivity distribution (SSD) but no overall change.
Mountain whitefish (*Prosopium williamsoni*)	[56]	131-265	New species included, new NOEC for SSD of 131
Brown trout (*Salmo trutta*)	[57]	416 (not hardness adjusted)	New species included, new NOEC for SSD of 416 (not hardness adjusted).
Daphnia magna	[27]	23	Additional data for species, from new study, contributes to geomean used in SSD but no overall change.
Ceriodaphnia dubia	[58]	37	Additional data for species from new study, no overall change.
Copper			
Mountain whitefish (*Prosopium williamsoni*)	[56]	3.9 – 8.3	New species included, new NOEC for SSD of 3.9.
Brown trout (*Salmo trutta*)	[57]	5.8	New species included, new NOEC for SSD of 5.8.
Pseudokirchneriella subcapitata	[59]	19, 16	Additional data for species, from new study, contributes to geomean used in SSD but no overall change.

2.9 New Zealand water quality relationships

Representative New Zealand water quality data were summarised in the revision documents [2] as shown in Table 2-1. These data provide the basis for simulations in the European Cu-BLM models.

Exploratory data analysis for the national rivers dataset is shown in Appendix E. This shows the generally high correlation between water hardness and alkalinity for New Zealand rivers. However, some sites, characterised by elevated chloride concentrations, show a different hardness/alkalinity slope.

The preliminary calculation estimates of DOC from a published relationship with spectrophotometric absorption indicate a wide range in hardness for a limited range in DOC. New measured data on DOC have subsequently become available which indicate that these estimates are too high. Lower DOC concentrations were used for the Cu-BLM simulations.

Generally, these relationships indicate that changes in hardness or alkalinity will co-vary for New Zealand rivers. Thus, changes in metal bioavailability for either of these parameters will result in conditions which warrant site-specific metal guidelines.

2.10 Implication for regulatory policy of a bioavailability-driven guidelines approach

The development of BLM modelling approaches for metals aims to include key water quality modifiers in the guideline derivation process. The US implementation of the BLM for copper requires that water quality information be available for a site prior to being able to calculate a numeric guideline [6]. This differs from the European approach where a default PNEC is derived and the BLM provides a modification of that default guideline to provide a site-specific assessment for that metal. The derivation of the PNEC uses the BLM to provide normalised toxicity data as part of the derivation process.

The Environment Canada approach generates relationships for their updated zinc guidelines and provides look-up tables for categorised water quality modifiers [44].

All of the updated guideline derivation approaches indicate that water quality modifiers are important and that some level of categorisation of sites may be required as part of the general implementation of national standards for metals. Additionally, some further level of site-specific assessment may be appropriate to incorporate specific locations (e.g., those with high DOC) or for specific applications (e.g., management of acute toxicity for storm-waters).

3 Acknowledgements

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4 Glossary of abbreviations and terms

ANZECC	Australian and New Zealand Environment and Conservation Council.
ARMCANZ	Agriculture and Resource Management Council of Australia and New Zealand.
BLM	Biotic Ligand Model.
Chronic toxicity	Lingering or continuing for a long time; often for periods from several weeks to years. Can be used to define either the exposure of an aquatic species or its response to an exposure (effect). Chronic exposure typically includes a biological response of relatively slow progress and long continuance, often affecting a life stage.
Cu	Copper.
Default guideline value (Default GV)	A guideline value recommended for generic application in the absence of a more specific guideline value (e.g., site-specific), in the Australian and New Zealand Water Quality Guidelines.
DOC	Dissolved organic carbon.
EC50 (median effective concentration)	The concentration of material in water that is estimated to be effective in producing some lethal or growth response in 50% of the test organisms. The EC50 is usually expressed as a time-dependent value (e.g., 24-hour or 96-hour LC50).
Endpoint	Measured attainment response, typically applied to ecotoxicity or management goals.
Guideline (water quality) value (GV)	Numerical concentration limit or narrative statement recommended to support and maintain a designated water use. GV has been adopted as new terminology for ANZECC guidelines.
Hardness	Hard water is water that has high mineral content. Water hardness is generally determined by the concentration of the common cations calcium and magnesium and expressed as equivalent calcium carbonate ($CaCO_3$).
LC50	Median lethal concentration.
LOEC (Lowest observed effect concentration)	The lowest concentration of a material used in a toxicity test that has a statistically significant adverse effect on the exposed population of test organisms as compared with the controls.
NO[A]EL	No observed [adverse] effects level.
NOEC (No observed effect concentration)	The highest concentration of a toxicant at which no statistically significant effect is observable, compared to the controls; the statistical significance is measured at the 95% confidence level.
PNEC	Predicted No Effect Concentration.
Site-specific	Relating to something that is confined to, or valid for, a particular place. Site-specific trigger values are relevant to the location or conditions that are the focus of a given assessment.

Species	A group of organisms that resemble each other to a greater degree than members of other groups and that form a reproductively isolated group that will not produce viable offspring if bred with members of another group.
SSD	Species Sensitivity Distribution. A method that plots the cumulative frequency of species sensitivity and fits the best possible statistical distribution to the data. From the distribution the concentration that should theoretically protect a selected percentage of species can be determined.
Standard (water quality)	An objective that is recognised in enforceable environmental control laws of a level of government.
Toxicity	The inherent potential or capacity of a material to cause adverse effects in a living organism.
Toxicity test	The means by which the toxicity of a chemical is determined. A toxicity test is used to measure the degree of response produced by exposure to a specific level of stimulus (or concentration of chemical).
Trigger value (TV)	These are the concentrations (or loads) of the key performance indicators measured for the ecosystem, below which there exists a low risk that adverse biological (ecological) effects will occur. They indicate a risk of impact if exceeded and should 'trigger' some action, either further ecosystem specific investigations or implementation of management/remedial actions. Terminology used in ANZECC (2000) guidelines and not adopted for this revision.
Water quality criteria	Scientific data evaluated to derive the recommended quality of water for various uses.
Zn	Zinc

5 References

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Appendix A Comparison of US EPA BLM derivation and European Union BLM for copper

US EPA approach (from [8])

The use of the Biotic Ligand Model in considering acute toxicity and the derivation of the CMC entailed some significant changes to previous data acceptability and screening procedures. In particular, requirements for water chemistry parameters are more stringent due to the requirements of the BLM. Some tests previously considered suitable are no longer so due to a lack of sufficiently detailed water chemistry. Some previously considered unsuitable due to extreme water chemistry (e.g., high DOC concentrations in the test waters) are now considered acceptable.

An important feature of the recommended process is that the BLM parameters are taken to be invariant for different organisms. The actual parameters are those based on the results of acute toxicity studies using fathead minnows (*Pimephales promelas*), which have been shown to adequately predict toxicity of copper to other organisms. The steps for calculating a WQC are essentially the same as those given above; the concepts of Species and Genus Mean Acute (Chronic) Values and the Final Acute (Chronic) Value are retained.

The steps to calculate the Final Acute Value are as follows:

1. Water chemistry information for the available toxicity data is collated and suitable data selected.
2. The endpoint copper concentration is converted from total recoverable to dissolved if necessary, using a conversion factor.
3. The reported L(E)C50 and water chemistry for each suitable toxicity test are input to the BLM to calculate the BL-bound copper at the endpoint (the L(E)A50).
4. The L(E)C50 at a standard water composition (Table A1.2) is calculated, by predicting the dissolved copper concentration in equilibrium with this BL-bound amount.
5. The Final Acute Value is calculated for the standard water composition using the normalised toxicity data, following steps 2 and 3 above.
6. The L(E)A50 corresponding to the standard water Final Acute Value is calculated using the BLM. This criterion is termed the L(E)A50 and is the single invariant toxicity parameter across different water chemistries.
7. Site-specific Final Acute Values are calculated as the dissolved copper in equilibrium with the criterion L(E)A50. The site-specific CMC is set to half the Final Acute Value.

Chronic toxicity data were calculated as EC_{20} values. The chronic toxicity dataset was not considered sufficient for direct calculation of a Final Chronic Value. Therefore an Acute–Chronic Ratio was calculated and this is used to calculate a site-specific Final Chronic Value from the site-specific Final Acute Value. The site-specific CCC is equal to the Final Chronic Value.

Table A1.2 Standard test water composition for BLM-normalisation of acute Cu toxicity data.

Temp	pH	DOC	Na	Mg	K	Ca	Cl	SO_4	HCO_3	S
20.0	7.50	0.5	26.3	12.1	2.1	14.0	81.4	1.9	65	0.0003

Notes: Temperature in °C, DOC and solution ions in mg l^{-1}, DOC modelled as 90% fulvic acid and 10% humic acid.

EU approach (from [8])

Consideration of the effects of copper speciation on its toxicity is thoroughly covered in the copper risk assessment. It is proposed that models describing these effects on Science Report – Environmental Quality Standards for trace metals in the aquatic environment 135 specific organisms be used in the assessment. The proposed methodology is as follows: 1. NOEC data were collected, assessed and selected according to the guidelines in the EU Technical Guidance Document for Risk Assessment. In the latest version of the draft report [dated July 2007] the Total Risk approach only is applied. 2. For the generic case (large-scale risk assessment), two scenarios were considered, 'reasonable worst case' and 'typical'. Reasonable worst case refers to chemical conditions of high bioavailability, typical refers to median chemical conditions of EU waters. For each scenario, the individual NOECs were normalised accordingly. Normalisation was done using two models: for invertebrates a chronic BLM for *Daphnia magna* (De Schamphelaere and Janssen, 2004) was used, for algae an empirical model for *Pseudokirchneriella subcapitata* (De Schamphelaere et al. 2003), and for fish a chronic BLM based on data for *Pimephales promelas* and *Oncorhynchus mykiss*. NOECs were normalised to the chosen conditions, aggregated by taking species geometric means, and PNECs were calculated by statistical extrapolation using the best fit from a large number of tested statistical distributions. 3. It was suggested that for site-specific risk assessment, normalisation to a sitespecific chemistry could be carried out on the same principles outlined in Step 2. The power of this approach was shown by the reduction in the variability of NOECs for the same species after normalisation to standard water chemistry. Of 13 species (four invertebrate, six fish and three algae) with multiple NOECs in the test database, normalisation reduced the inter-test variability by up to 83%, and variability was reduced by 40% or more in nine cases. Only for one species, *Onchrhynchus kitusch* (coho salmon), did inter-test variability increase.

Appendix B Copper Biotic Ligand Model (Cu-BLM)

Explanatory note on the chronic copper BLM - ARCHE Excel version 1.3.2

The ARCHE BLM version 1.3.2 is an excel version of the original Hydroqual model used for the copper RA. The model includes the model parameters of the original Hydroqual model and thus calculates:

- the BLM -normalized NOEC values.
- HC5-50 from the SSD.

Some modifications were introduced compared to the Hydroqual model:

- The HC5-50 from the SSD - with the RIVM ETX model (instead of the original statistical package used by Hydroqual - as recommended by the EU Member States during the discussions of the copper RAR).
- Calculation of alkalinity are based on pH-alkalinity regression (on test media) instead of using a default value.
- Change of some parameters data files in accordance due to small to changes between 2005 reports and the final publications.

To run the model, please install the file in a folder/subfolder that has NO SPACES IN THE FOLDERS NAMES.

Please note that the model requires the basic parameters: pH, DOC, Ca. Preferably, all parameters should be filled. However, if no data are provided for the less critical parameters (Mg, Na...), these are calculated from regressions - as done in Bio-met.

Prior to any further distribution, we would appreciate your comments, and please do not hesitate to contact us of you have problems or need further information.

Please contact us if you have problems running the model.

Friendly greetings.

Katrien Delbeke
ECI
Tel : +3227777086

Appendix C Relationships between zinc chronic toxicity data and hardness and pH in freshwaters

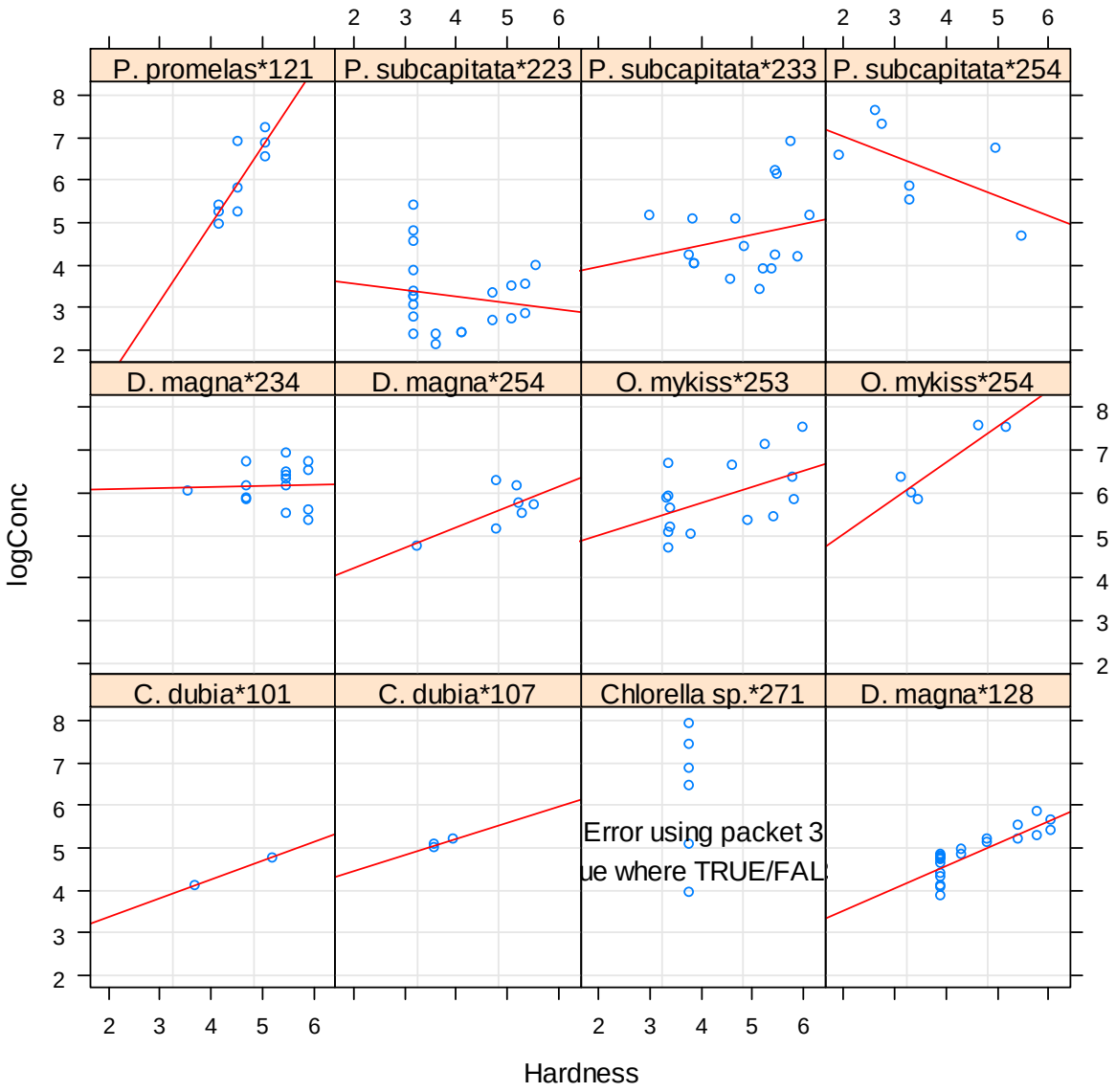

Figure C-1: Regression slope derivation for chronic Zn data and water hardness.

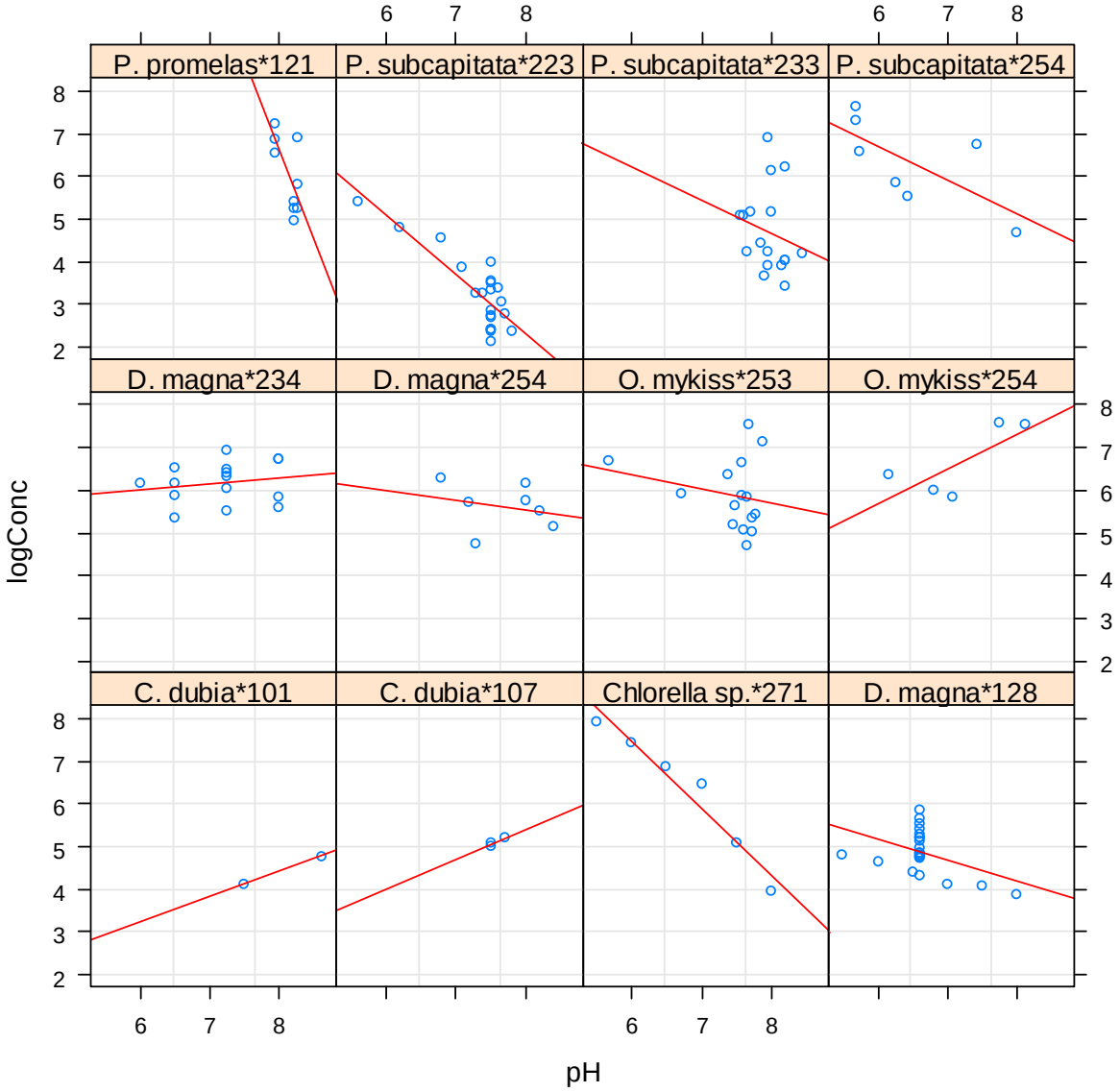

Figure C-2: Regression slope derivation for chronic Zn data and water pH.

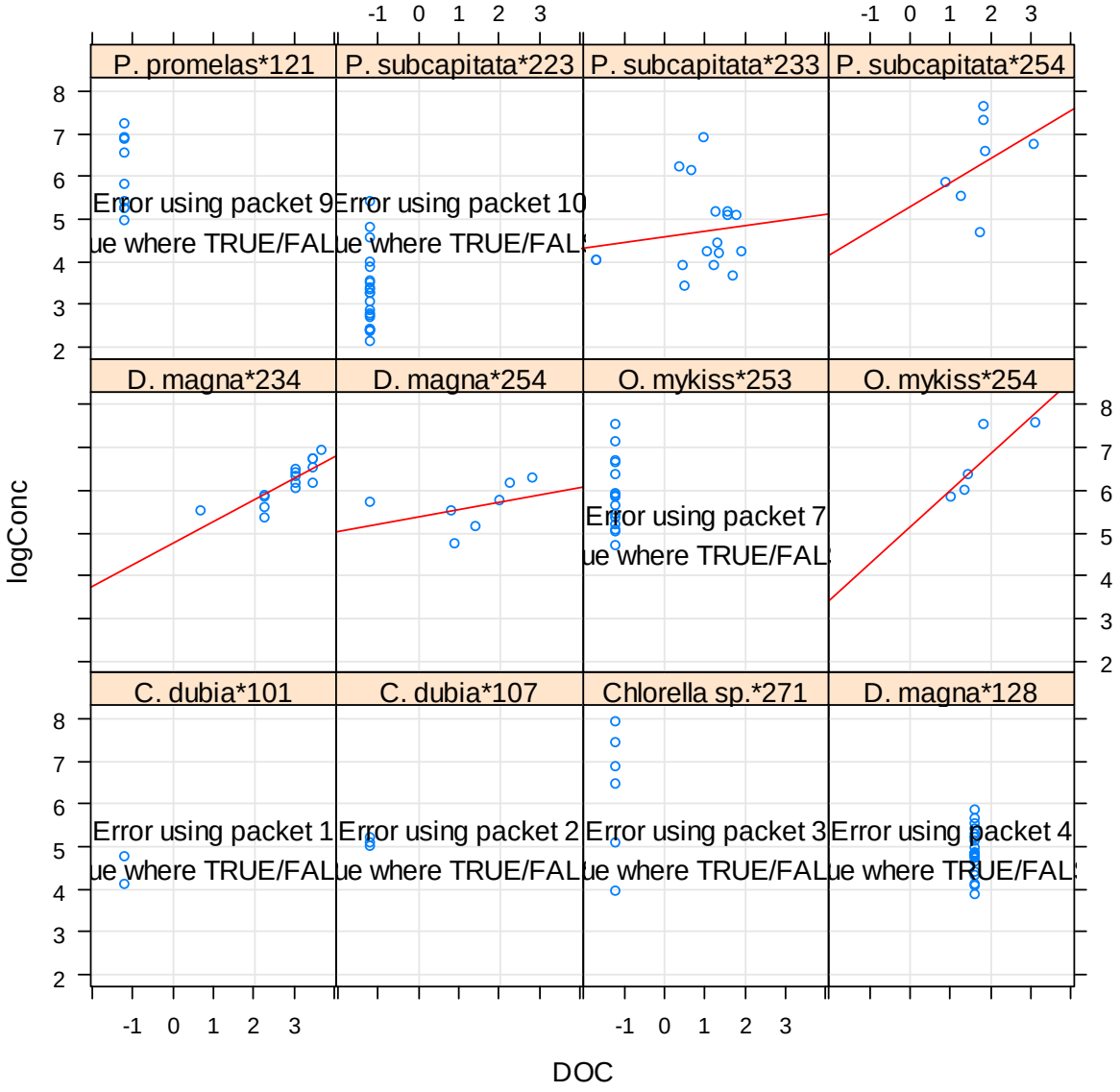

Figure C-3: Regression slope derivation for chronic Zn data and DOC.

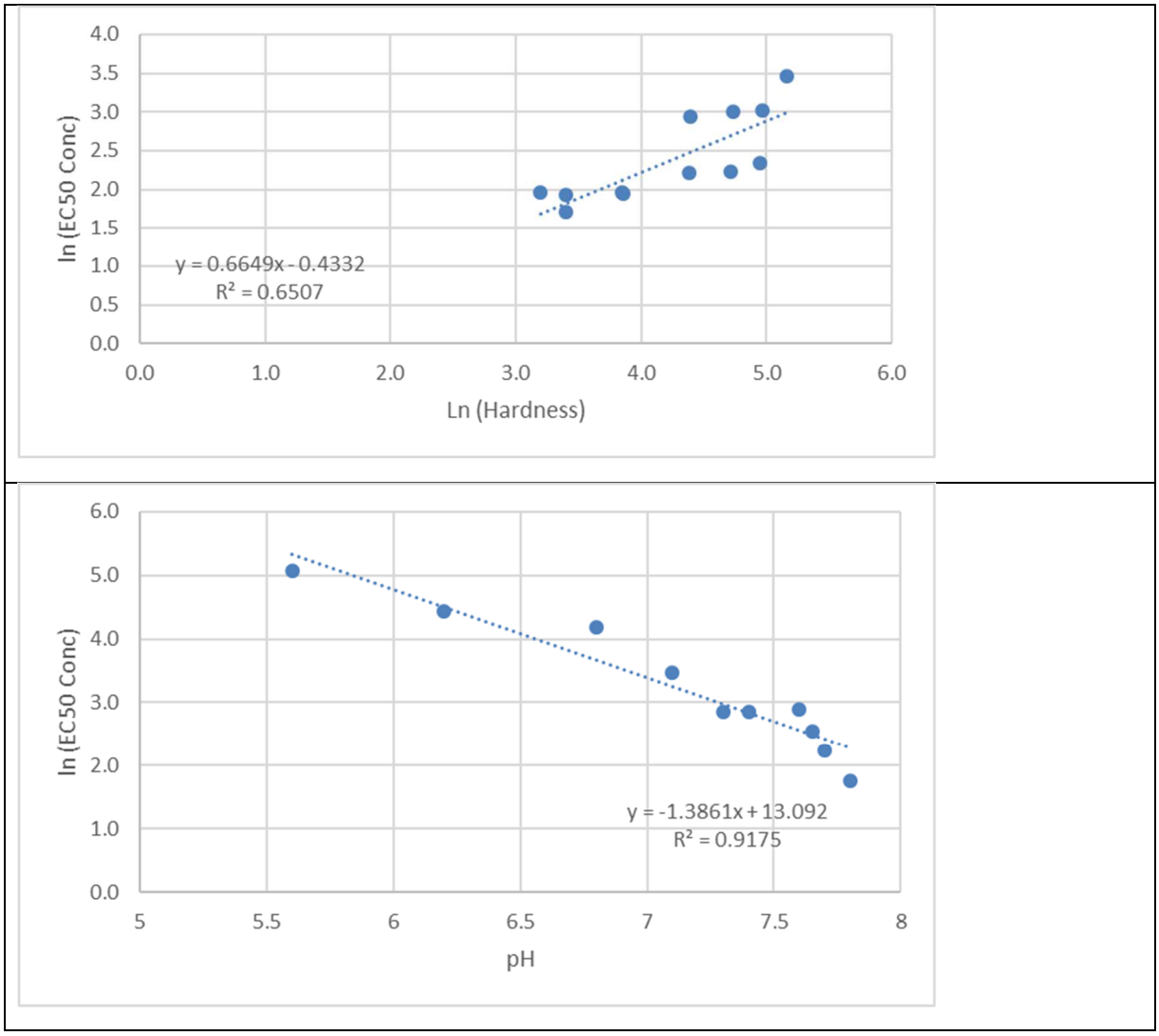

Figure C-4: Chronic zinc data from Heijerick et al. (2002) [60] for *P. subcapitata*, standardising by pH & Na. Note that hardness varies by varying Ca & Mg hence some variability due to which is changed.

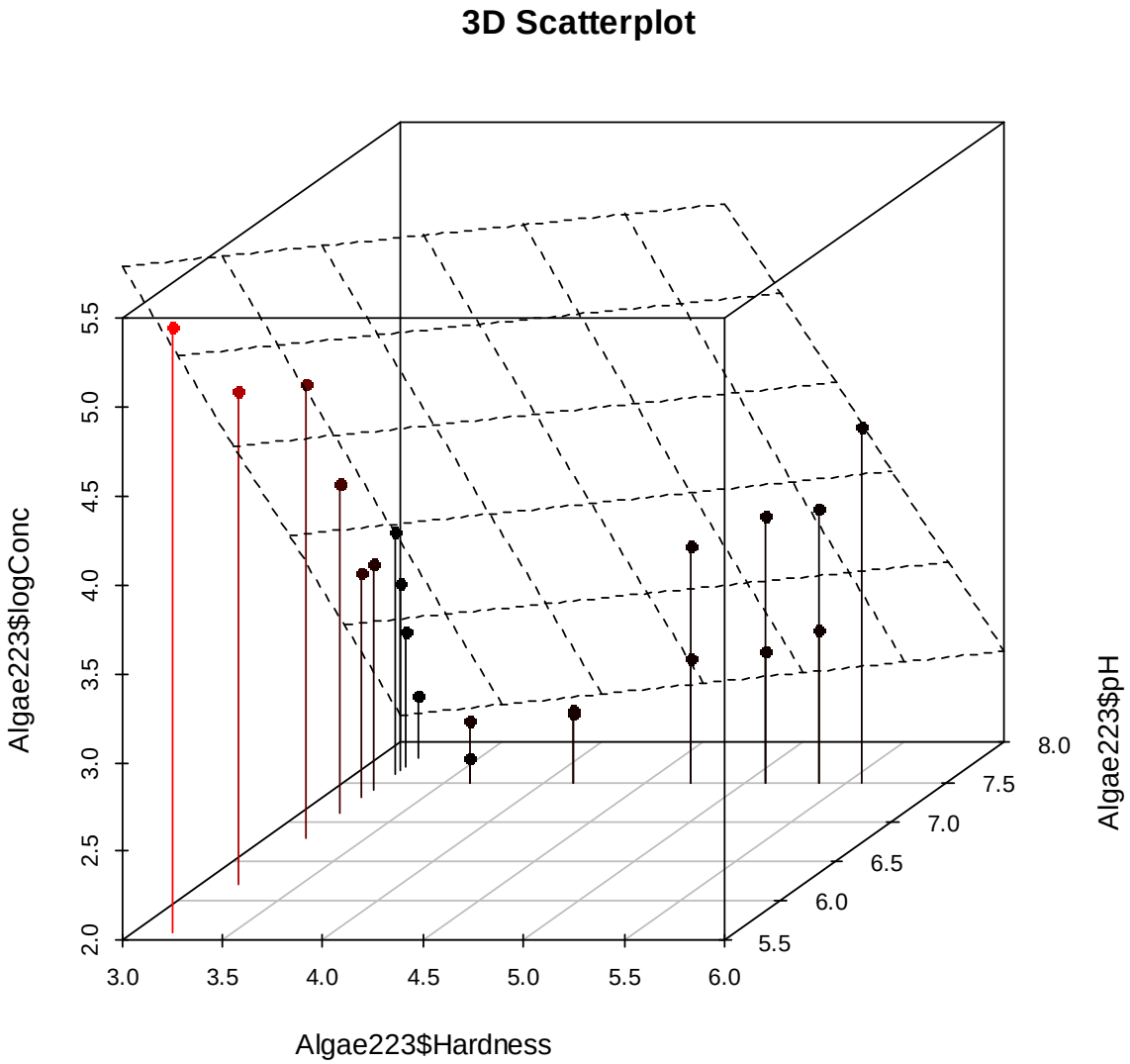

Figure C-5: Chronic zinc data from Heijerick et al. (2002) [60] for *P. subcapitata*, standardising by pH & Na, as 3D-scatterplot.

Appendix D Generalised classification procedure for chronic test selection for guideline derivation

Table D-1: Generalised classification of acute and chronic toxicity tests for temperate species, based on test duration and, where applicable, endpoint, for the purposes of water quality guideline value derivation (from [2, 3]). Note: This table has been updated in May 2017: (i) The content of the rows for acute microinvertebrates, chronic macroinvertebrates and chronic microinvertebrates were changed; (ii) Changes were made to the footnotes, particularly the definitions of macroinvertebrates and microinvertebrates (included below); (iii) There are now rows for three early life stage endpoints (lethality, development and fertilisation) each with their own definition of the minimum exposure duration to be considered chronic.

TOXICITY TEST	LIFE STAGE[a]	RELEVANT ENDPOINTS[b]	TEST DURATION
Acute			
Fish and amphibians	Adults/juveniles	All[c]	<21 d
	Embryos/larvae	All	<7 d
Macroinvertebrates[d]	Adults/juveniles	All	<14 d
	Embryos/larvae	All (except fertilisation, larval development/ metamorphosis)	<7 d
	Embryos/larvae	Larval development/ metamorphosis	<48 h
Microinvertebrates[e]	Adults/juveniles/larvae	All (except fertilisation and larval development – see microinvertebrate chronic)	<7 d
Macrophytes	Mature	All	<7 d
Macroalgae	Mature	Lethality, growth and photosynthesis and biochemical endpoints	<7 d
Microalgae	Not applicable	All	≤24 h
Microorganisms	Not applicable	All	≤24 h
Chronic			
Fish and amphibians	Adults/ juveniles	All[f]	≥21 d
	Embryos/larvae/eggs	All	≥7 d
Macroinvertebrates	Adults/juveniles/larvae	All (except reproduction, larval development/metamorphosis)	≥14 d
	Adults/juveniles/larvae	Reproduction	≥14 d (or at least 3 broods for large cladocerans)
	Larvae	Larval development/ metamorphosis	≥48 h
	Embryos	Fertilisation	≥1 h
Microinvertebrates	Adults/juveniles/larvae	Reproduction	≥7 d (or at least 3 broods for small cladocerans)
	Adults/juveniles/larvae	Lethality/immobilisation	≥7 d
	Larvae	Development	≥48 h
	Embryo	Fertilisation	≥1 h
Macrophytes	Mature	All	≥7d
Macroalgae	Mature	All	≥7 d
	Early life stages	Lethality	≥7 d

TOXICITY TEST	LIFE STAGE[a]	RELEVANT ENDPOINTS[b]	TEST DURATION
	Early life stages	Development	≥48 h
	Early life stages	Fertilisation	≥1 h
Microalgae	Not applicable	All	>24 h
Microorganisms	Not applicable	All	>24 h

[a] The life stage at the start of the toxicity test. [b] Endpoints need to be ecologically relevant – see the section 3.2 of Warne et al. (2015)[3] - Acceptable test endpoints. [c] For acute tests, "All" refers to all ecologically relevant endpoints for a particular life stage of a particular species. [d] Macroinvertebrates include invertebrates where adults are ≥2 mm long (e.g., decapods, echinoderms, molluscs, annelids, corals, amphipods, larger cladocerans (such as *Daphnia magna*, *Daphnia carinata* and *Daphnia pulex*) and insect larvae of similar sizes with life cycles markedly longer than most microinvertebrates. [e] Microinvertebrates are operationally defined here as invertebrate species where full grown adults are typically <2 mm in length with relatively short life cycles. Examples of invertebrates that meet this criterion are some cladocerans (e.g., *Ceriodaphnia dubia* and *Moina australiensis*), copepods, conchostracans, rotifer, acari, bryozoa, and hydra. Large cladocerans such as *Daphnia magna* or *Daphnia pulex* are macroinvertebrates. [f] For chronic tests, "All" encompasses all ecologically relevant endpoints measured in both single and multi-generation tests.

Appendix E Relationships between water quality parameters for New Zealand rivers

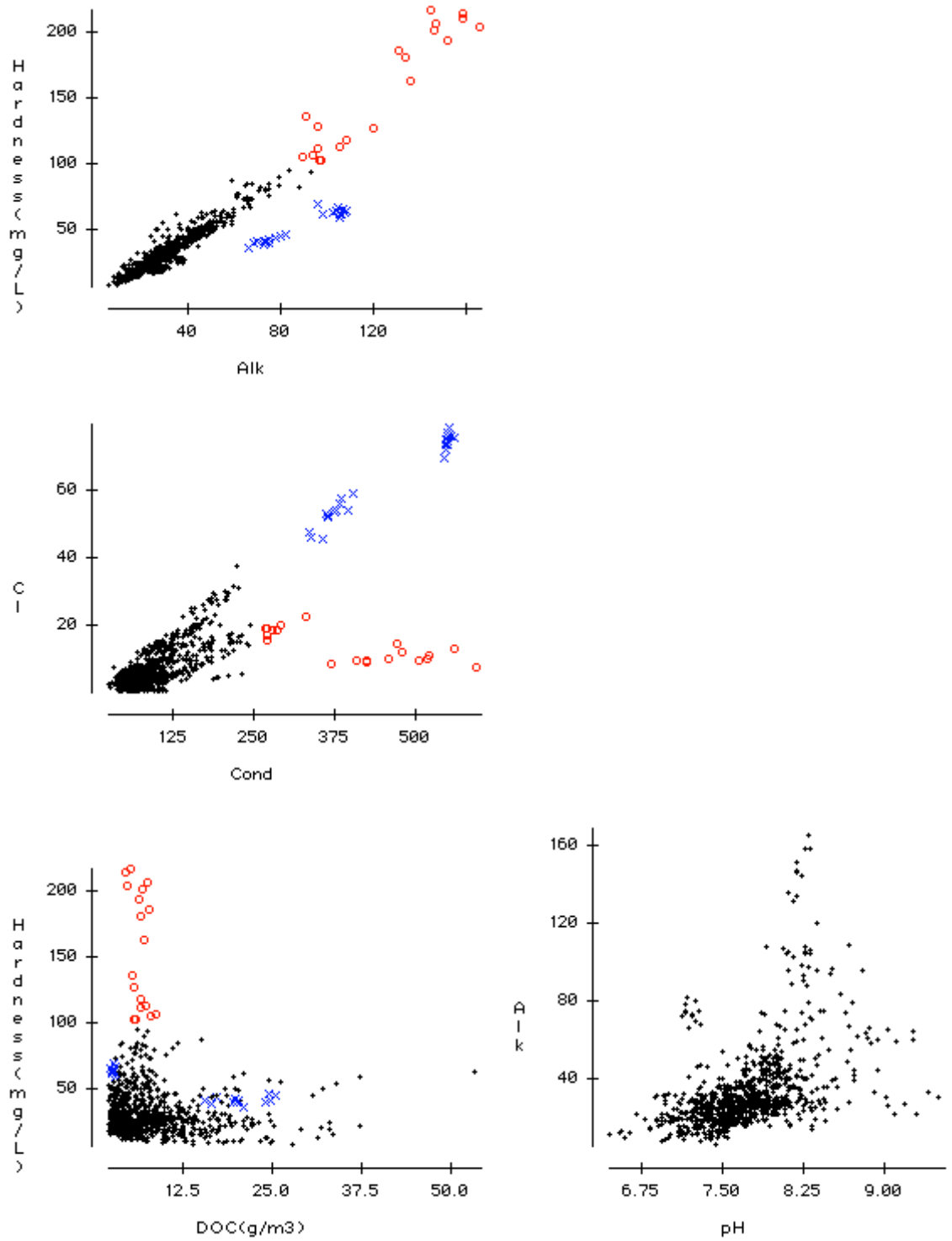

Figure E-1: Exploratory data analysis for water quality relationships in New Zealand rivers. Red circles are hardness above 100 mg $CaCO_3$ L^{-1}. Blue + are lower relationship for hardness alkalinity plot. Data from Smith and Maasdam [11] with DOC calculated using Collier relationship [61].