NIWA
Taihoro Nukurangi

Updating nitrate toxicity effects on freshwater aquatic species

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Authors/Contributors:
C.W. Hickey

For any information regarding this report please contact:

Dr C.W. Hickey
Principal Scientist

+64-7-856 1713
chris.hickey@niwa.co.nz

National Institute of Water & Atmospheric Research Ltd
Gate 10, Silverdale Road
Hillcrest, Hamilton 3216
PO Box 11115, Hillcrest
Hamilton 3251
New Zealand

Phone +64-7-856 7026
Fax +64-7-856 0151

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Reviewed by

C. Depree

Approved for release by

T. Hume

Formatting checked by

Executive summary

The National Policy Statement on Freshwater (NPS-FW) on freshwater management requires regional councils to identify water quality limits to manage values of our aquatic ecosystems. Recognition of appropriate contaminant toxicity thresholds is an integral part of this process. This report provides an updated derivation on the freshwater nitrate toxicity guidelines and guidance on their application to New Zealand's river and lakes.

This report updates the revised nitrate-N guidelines derived in 2009 for Environment Canterbury. This review includes a substantial number of new acute and chronic studies which have been undertaken in the interim period. Those studies include chronic data for two New Zealand native species, including a whitebait species (juvenile inanga) and the widespread mayfly species (*Deleatidium*). The additional data has resulted in revised chronic guideline values for nitrate-N using the ANZECC (2000) methodology. This database and NOEC-based guideline derivation will form the basis of the interim ANZECC nitrate guideline, which is expected to be promulgated in 2013.

The proposed numeric nitrate guideline values are based on the statistically-derived no observed effect concentration (NOEC) and threshold effect concentration (TEC) effect thresholds for 22 species. The two-number guideline and management framework is based on, and consistent with, the ANZECC (2000) guidelines risk-based methodology to provide various levels of ecosystem protection. The compliance monitoring basis provides species protection for both average long-term exposure and seasonal maximum concentrations. The guidelines are summarised together with narrative descriptors in the table below. The terms "Grading" and "Surveillance" are derived from the compliance descriptors applied to microbiological standards. The Grading values are derived from the species NOEC values and recommended for compliance assessment based on the annual median concentrations. The Grading values are equivalent to trigger values as derived using the ANZECC (2000) procedure. The Surveillance values are derived from the species TEC values and recommended for compliance assessment based on the annual 95th percentile of the monitoring data.

A number of studies have identified water hardness as a factor affecting both acute and chronic nitrate toxicity in some species. A hardness-related response was also found for chronic growth in juvenile inanga, but not for acute inanga exposures. The mechanism for this effect is presently unknown so no algorithms were included in the 2012 Environment Canada nitrate toxicity guidelines to adjust for water quality factors. However, the guidelines reported here should be considered conservative as the most sensitive species in the database used to derive the nitrate guidelines is for long-term tests undertaken in very low hardness water – which would be expected to result in the highest nitrate toxicity.

Some information gaps were identified in undertaking this review. These included: (i) the adequacy of native fish and invertebrate data for surface waters; (ii) absence of hyporheic species; and (iii) toxicity modification in relation to water mineral content (measured by hardness). Nine native species (4 fish and 5 invertebrate) are recommended as potential candidates for both acute and chronic testing. A multi-criteria assessment is applied to these species to provide a basis for selection of priority species for chronic testing. Priority species would include the common bully and eel elvas, which are both widespread and major inhabitants of lowland streams where nitrate concentrations tend to be highest. The invertebrate species include crustaceans (koura and amphipod), which tend to be highly sensitive to contaminants and the bivalve Sphaeriid, which is common in lowland streams and has been shown to be particularly sensitive to ammoniacal-N. Additional testing is also recommended to establish the key water quality factor(s) affecting nitrate toxicity.

The recommended freshwater nitrate toxicity guidelines are:

Guideline Type	Grading Nitrate concentration (mg NO_3-N /L)	Surveillance Nitrate concentration (mg NO_3-N /L)	Description of Management Class
Chronic – high conservation value systems (99% protection)	1.0	1.5	Pristine environment with high biodiversity and conservation values.
Chronic – slightly to moderately disturbed systems (95% protection)	2.4	3.5	Environments which are subject to a range of disturbances from human activities, but with minor effects.
Chronic – highly disturbed systems (90% protection)	3.8	5.6	Environments which have naturally seasonally elevated concentrations for significant periods of the year (1-3 months).
Chronic – highly disturbed systems (80% protection)	6.9	9.8	Environment which are measurably degraded and which have seasonally elevated concentrations for significant periods of the year (1-3 months).
Acute	20	30	Environments which are significantly degraded. Probable chronic effects on multiple species.
Method of comparison	Annual median	Annual 95^{th} percentile	

1 Introduction

The National Policy Statement on Freshwater (NPS-FW) on freshwater management requires regional councils to identify water quality limits to manage values of our aquatic ecosystems. Recognition of appropriate contaminant toxicity thresholds is an integral part of this process. This report provides an updated derivation on the freshwater nitrate toxicity guidelines and guidance on their application to New Zealand's river and lakes.

A review of the ANZECC (2000) [1] nitrate guidelines was undertaken in 2009 [2], however, since that time significant additional international data and some New Zealand native species data has become available on nitrate sensitivity. The chronic toxicity thresholds derived in the 2009 review largely included aquatic species from overseas, though some of the species were applicable in a New Zealand context. There was, however, no native New Zealand species included in the 2009 review. Native species data has recently become available as part of the Hawke's Bay Regional Council's (HBRC) studies to provide information to incorporate into a Plan Change for the Tukituki River catchment. This native species data included chronic nitrate toxicity thresholds for a whitebait species (Inanga) and the widespread mayfly species (*Deleatidium*). Both these species are widespread throughout New Zealand and have high relevance on a national scale with Inanga having strong cultural and recreational values and *Deleatidium* mayflies as important food source for trout.

The objectives for this study were to:

1. incorporate results of nitrate toxicity trials being carried out by HBRC into the guideline values published in 2009 [2]

2. review recent overseas studies on nitrate toxicity effects on aquatic species. This should include a critique of recent Canadian studies on rainbow trout which is likely provide a better estimate of toxicity thresholds for this species, and

3. update the Hickey & Martin (2009) guideline values for aquatic species based on the information collected in the first two stages. The update should also include summaries of NOEC and LOEC concentration thresholds as well as investigating the applicability of 'Ecologically Significant Effects Concentrations' (ESEC). Also included will be a gap analysis that will:

    1) review environmental factors that may influence nitrate toxicity, such as temperature, and

    2) identify 'target' New Zealand species for future trials that would allow for robust national guidelines to be developed for New Zealand.

The native species data included in this report are reported in detail elsewhere [3, 4]. The chronic guideline values are consistent with the site-specific approach applied to the HBRC Tukituki River catchment [5]. Additional acute toxicity data for nitrate are included in this report and used for an acute guideline derivation to update the 2009 review [2].

The proposed numeric nitrate guideline values are based on a two number approach for "Grading" and "Surveillance" – relating to annual median and 95th percentile of nitrate monitoring concentrations.

2 Background

Nitrate occurs naturally in the environment and is produced and consumed through the processes of the nitrogen cycle, and anthropogenically produced for agricultural use as a fertilizer. The major anthropogenic sources of nitrate to surface waters are agricultural runoff, municipal and industrial wastewaters, urban runoff and groundwater inputs. Nitrate concentrations are an important indicator of agricultural enrichment and ecological health. As such, they form an important component of the management of freshwaters, requiring robust guideline values to support environmental planning and management.

Since the last revision of the freshwater nitrate trigger values (TVs) for toxicity in 2000 [1], errors were identified in the derivation and new data have become available. The revised 2009 guideline trigger value is significantly lowered from the 2000 value (changing from 7.2 mg NO_3-N /L [6] to 1.7 mg NO_3-N /L for 95% species protection [2]). Subsequent to the 2009 review, Environment Canada has released a revised freshwater nitrate water quality guideline [7], which included new acute and chronic data for several freshwater species. Recent chronic nitrate toxicity studies have also been undertaken for two New Zealand freshwater species (sub-adult and juvenile inanga, *Galaxias maculatus*; mayfly, *Deleatidium* sp.; [3, 4]) which further expanded the database for local species. The chronic nitrate guideline values reported here are based on 22 species, including the additional New Zealand data. This updated database and guideline derivation will form the basis for the ANZECC interim revised nitrate guidelines scheduled to be completed in 2013.

Background information on nitrate as a toxicant can still be sourced from ANZECC/ARMCANZ (2000) [1] (Volume 2, Section 8.3.7.2) and Environment Canada (2012) [8].

The National Policy Statement for Freshwater Management, 2011, requires the development of management plans for freshwater lakes and rivers. This report proposes effects-based thresholds for life-supporting capacity which could be applicable to a range of management classes.

The approach uses the risk-based approach as used in the ANZECC (2000) [1] guidelines to derive numeric threshold for potential management classes. The basis for these calculations is effects-based data which use concentration-response relationships to derive critical thresholds for contaminants of concern. The limitations of the databases are noted in relation to native species incorporation, water quality modifiers and site-specific issues for regional derivations.

3 General information

The nitrate TVs are based on toxicity data for sodium nitrate ($NaNO_3$; CAS 7631-99-4) only. Data based on tests using potassium nitrate (KNO_3) were omitted from the final dataset due to the possibility that potassium was contributing to toxicity.

It should be noted that the nitrate concentrations are reported in this document as nitrate nitrogen (NO_3-N), rather than the nitrate ion (NO_3^-), which is the basis for guidance derived by some other jurisdictions (e.g., Environment Canada [7, 8]). This difference in reporting convention makes no difference to the toxic sensitivity, and a nitrate ion concentration may be converted to nitrate nitrogen by multiplying by 0.23. Conversely, nitrate nitrogen concentrations can be converted to nitrate ion equivalent by multiplying by 4.43.

ANZECC guideline approach

The ANZECC (2000) guidelines provide a methodology to derive risk-based trigger values (TVs) for surface waters using no observed effect concentration (NOEC) sensitivity values for aquatic species. The TVs are based on a dataset describing chronic (long term) effect (i.e., growth, reproduction) for a range of species groups (e.g., fish, invertebrates, amphibians) which are representative of the ecosystem.

The ANZECC (2000) guidelines provide a statistical derivation procedure for differing levels of ecosystem protection. The ANZECC descriptors recognise three broad ecosystem conditions:

1. *High conservation/ecological value systems (99% species protection[1]) — effectively unmodified or other highly-valued ecosystems, typically (but not always) occurring in national parks, conservation reserves or in remote and/or inaccessible locations. While there are no aquatic ecosystems in Australia and New Zealand that are entirely without some human influence, the ecological integrity of high conservation/ecological value systems is regarded as intact.*

2. *Slightly to moderately disturbed systems (95% species protection) — ecosystems in which aquatic biological diversity may have been adversely affected to a relatively small but measurable degree by human activity. The biological communities remain in a healthy condition and ecosystem integrity is largely retained. Typically, freshwater systems would have slightly to moderately cleared catchments and/or reasonably intact riparian vegetation; marine systems would have largely intact habitats and associated biological communities. Slightly to moderately disturbed systems could include rural streams receiving runoff from land disturbed to varying degrees by grazing or pastoralism, or marine ecosystems lying immediately adjacent to metropolitan areas.*

3. *Highly disturbed systems (80-90% species protection). These are measurably degraded ecosystems of lower ecological value. Examples of highly disturbed systems would be some shipping ports and sections of harbours serving coastal cities, urban streams receiving road and stormwater runoff, or rural streams receiving runoff from intensive horticulture.*

[1] The percentage species protection refers to a statistically-derived numeric value which is calculated from the chronic toxicity sensitivity data for the species groups. These species are assumed to be representative of the "ecosystem" of species in the receiving environment.

A site-specific framework would replace these general descriptors with regional descriptors applicable to nitrate management within the catchments and sub-catchment areas.

The chronic freshwater TVs reported here differ from those of Hickey and Martin [2] due to: (i) data included in the recently published Environment Canada nitrate guidelines [8]; (ii) a toxicity value for the tropical prawn species (*Macrobrachium rosenbergii*, 21 d NOEC, 32.0 mg/L)[2]; (iii) inclusion of native species data for inanga (sub-adult and juvenile *Galaxias maculatus*) and a mayfly (*Deleatidium* sp.) [3, 4]; (iv) inclusion early life-stage data for the New Zealand strain of rainbow trout (*Oncorhynchus mykiss*) [3]; and (v) in provision of a two number basis for management and compliance monitoring.

An acute guideline value was calculated based on the species sensitivity distribution approach, as used in the ANZECC (2000) guidelines, to calculate a community 5 percentile effect threshold based on LC_{50}/EC_{50} effects data [3]. This acute 5th percentile effects value then has an application factor (AF) of 2 applied to generate a final acute guideline following the U.S. EPA standard procedure [9, 10] and is termed the Acute Grading guideline value. The Acute Surveillance guideline value is the median of the Acute Grading guideline value and the acute 5th percentile effects value. Though the recent Environment Canada protocol [11] includes short-term exposure guidelines, which are: “meant to estimate severe effects and to protect most species against lethality during intermittent and transient events (e.g., spill events to aquatic-receiving environments, infrequent releases of short-lived/non-persistent substances)”, their acceptance of a 50% effect for some species at the guideline level is probably inconsistent with the New Zealand Resource Management Act legislation [12], as this would potentially constitute a significant adverse effect on aquatic life. Because of the large number of acute guideline derivations following the U.S. EPA procedure, we consider that this is the preferred approach for use in this study to benchmark the acute nitrate toxicity relative to the available data. The recent Environment Canada nitrate guidelines include both short-term (acute) and long-term (chronic) guideline values [7, 8].

The species sensitivity distribution model used for all guideline derivations was the BUR III model referred to in the ANZECC (2000) procedures. Further details on the nitrate guideline derivations can be sourced from Hickey and Martin (2009) [2].

[2] Hickey and Martin [2] omitted the prawn value as it was from a tropical species and they were deriving site-specific TVs for the cool temperate waters of New Zealand. The inclusion of the single tropical species and other species data makes a small difference to the derived freshwater TVs and provides a more comprehensive generic guideline derivation.

[3] LC_{50} = Median lethal concentration; EC_{50} = 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.

4 Aquatic toxicology

Chronic data

A summary of the 22 chronic no observed effect concentration (NOEC) and threshold effect concentration (TEC) results are provided in Table 2 with the sensitivity distribution of NOEC values shown in Figure 4-1. The details are contained in Appendix B. These include data for 9 fish, 8 invertebrate, 4 amphibians and 1 algal species. The dataset spans a 391-fold range in sensitivity, with lake trout (*Salvelinus namaycush*) the most sensitive, with a NOEC of 1.6 mg NO_3-N/L for both growth and development endpoints measured after a 146-day exposure. In general, the chronic data indicate higher exposure sensitivity for fish, although both fish and invertebrates show wide ranges in sensitivity. The most sensitive invertebrate NOEC (a freshwater crayfish, *Astacus astacus*) was 9x less sensitive than the most sensitive fish NOEC. Rainbow trout, the mayfly *Deleatidium* sp., and juvenile inanga were all markedly less sensitive than the most sensitive species, lake trout (by a factor of 16x, 13x and 7x respectively). These sensitivities corresponding to: 57th, 48th and 29th percentiles (Table 4-1).

The key chronic data are the recent long-term (126–146-day) chronic studies of fish sensitivity by McGurk et al. [13], who measured acute and chronic sensitivity of embryo-alevins-swim-up fry life stages of lake trout (*Salvelinus namaycush*) and lake whitefish (*Coregonus clupeaformis*) under laboratory conditions in low hardness water. The lake trout were the most sensitive species with a NOEC of 1.6 mg NO_3-N/L, and LOEC values of 6.25 mg NO_3-N/L for both growth and development endpoints. Growth showed a progressive concentration response with a 12% reduction in wet weight at the LOEC value and a 22% reduction at 25 mg NO_3-N/L. The delayed development endpoint (>90% fry) had a comparable sensitivity.

The rainbow trout data included two concurrent tests undertaken for fry of resident and anadromous ("Steelhead"[4]) rainbow trout by Kinchloe et al*.* [14] and recent chronic studies with embryo-alevin-fry life stages, including the New Zealand strain, for a range of water hardness values [3, 15, 16]. The Kinchloe et al*.* study measured mortality effects on eggs and fry after a 30-day exposure period. The egg sensitivity data in this study were compromised by the mortalities associated with *Saprolegnia* fungal infestations so were not included. There is no indication that the fry were adversely affected by fungal infestation, with good control survival (>95%) and a partial concentration response for the "non-anadromous" rainbow trout. However, Environment Canada did not include this data because of the uncertain health status of the test organisms [8]. The NOEC values for the two trout types were 1.1 mg NO_3-N/L and >4.5 mg NO_3-N/L were included in the data summary (Appendix B). More recent data has measured an embryo-alevin-fry reduction in chronic toxicity to rainbow trout with increasing water hardness, with NOEC values ranging from 15 mg NO_3-N/L to 405 mg NO_3-N/L for hardness values from 10 mg $CaCO_3$/L to 176 mg $CaCO_3$/L [15], and 115 mg NO_3-N/L for a hardness value of 310 mg $CaCO_3$/L [16] (Appendix B). The New Zealand rainbow trout strain was tested for 42 days in moderate hardness water (40 mg $CaCO_3$/L) and had a NOEC value of 99 mg NO_3-N/L. This NOEC value for a New Zealand strain is markedly higher (7-fold), indicating lower sensitivity, than the recent study with the Canadian rainbow trout strain (15 mg NO_3-N/L) for a similar hardness value (Appendix B). This indicates either strain sensitivity differences or that the nominal "hardness" measure is

[4] Steelhead are rainbow trout which ascend rivers from the sea for breeding.

not an adequate modifier parameter for sensitivity adjustment (see discussion below on Toxicity Modifying Factors). A geometric mean value of 26.3 mg NO_3-N/L for the reported NOEC concentrations (368-fold range, 1.5–405 mg NO_3-N/L, Appendix B) was used in the guideline derivation following the ANZECC (2000) [1] derivation protocol.

These revised guideline calculations now incorporate chronic nitrate toxicity data for two New Zealand native species – a mayfly (20 day test with *Deleatidium* sp.) and juvenile and sub-adult whitebait (40 d and 31 d tests with inanga, *Galaxias maculatus*; called common jollytail in Australia). The chronic survival NOEC for the mayflies was 20.3 mg NO_3-N/L in moderate hardness water (40 mg $CaCO_3$/L) (Appendix B). The mayfly sensitivity was 11th of the 22 species (48th percentile, Table 4-1). The growth of juvenile inanga was the most sensitive chronic measure, with results showing significant sensitivity differences between the two hardness concentrations tested. Juvenile inanga in the low hardness water were 3.5-times more sensitive to nitrate than in moderate hardness water (6.0 mg NO_3-N/L c.f. 20.8 mg NO_3-N/L), indicating that site-specific guidelines for low hardness waters may require use of appropriately selected data. Growth reduction in the soft water test averaged 11% at the NOEC concentration of 6.0 mg NO_3-N/L and 35% at the LOEC concentration of 20 mg NO_3-N/L. A 20% growth reduction would be expected to occur at the TEC concentration of 11.0 mg NO_3-N/L.

A geometric mean value of 11.2 mg NO_3-N/L was calculated for inanga from the soft and moderately hard water NOEC values and used for guideline derivation.

The only tropical data used in the guideline derivation were for the freshwater prawn *Macrobrachium rosenbergii* [17]. The NOEC value for this species was ranked 15th of the 22 species (67th percentile) and was therefore not a significant driver of the toxicity thresholds. It was included in the greater dataset to provide a more comprehensive database for the assessment of relative species and group sensitivities.

Environment Canada [8] has provided additional review comments on nitrate publications.

Acute data

The acute toxicity data for nitrate has been updated with data published since 2009. The data is shown in Figure 4-1 with details in Table B-2. The total acute dataset is for 29 species including 11 fish, 16 invertebrate and 2 amphibian species. The new data includes rainbow trout sensitivity for a range of water hardness values, and New Zealand data for a mayfly larvae and juvenile inanga.

The inclusion of additional species has not changed the acute guideline value of 20 mg NO_3-N/L [2], but provides additional data on the relative sensitivity of species and their susceptibility to water quality factors affecting toxicity. The acute toxicity sensitivity ranged 35-fold, with invertebrates generally being more sensitive than fish (Table B-2). The mayfly *Deleatidium* sp. was the most sensitive species and comparable with sensitive amphipod species. The most sensitive fish species, Siberian sturgeon, were 7-times less sensitive than the mayfly. With respect to NZ relevant species, rainbow trout and native juvenile inanga were 24- and 27-times less sensitive, respectively than the mayfly. These sensitivities correspond to the 75th and 86th percentile of the dataset respectively (Figure 4-1, Table B-2).

Acute nitrate toxicity to an amphipod (invertebrate) species and rainbow trout has been shown to be sensitive to water hardness, with increasing toxicity at low hardness [18]. However, acute toxicity of juvenile inanga was not affected by water hardness [3]. The modifying factors affecting nitrate toxicity are discussed in the following section.

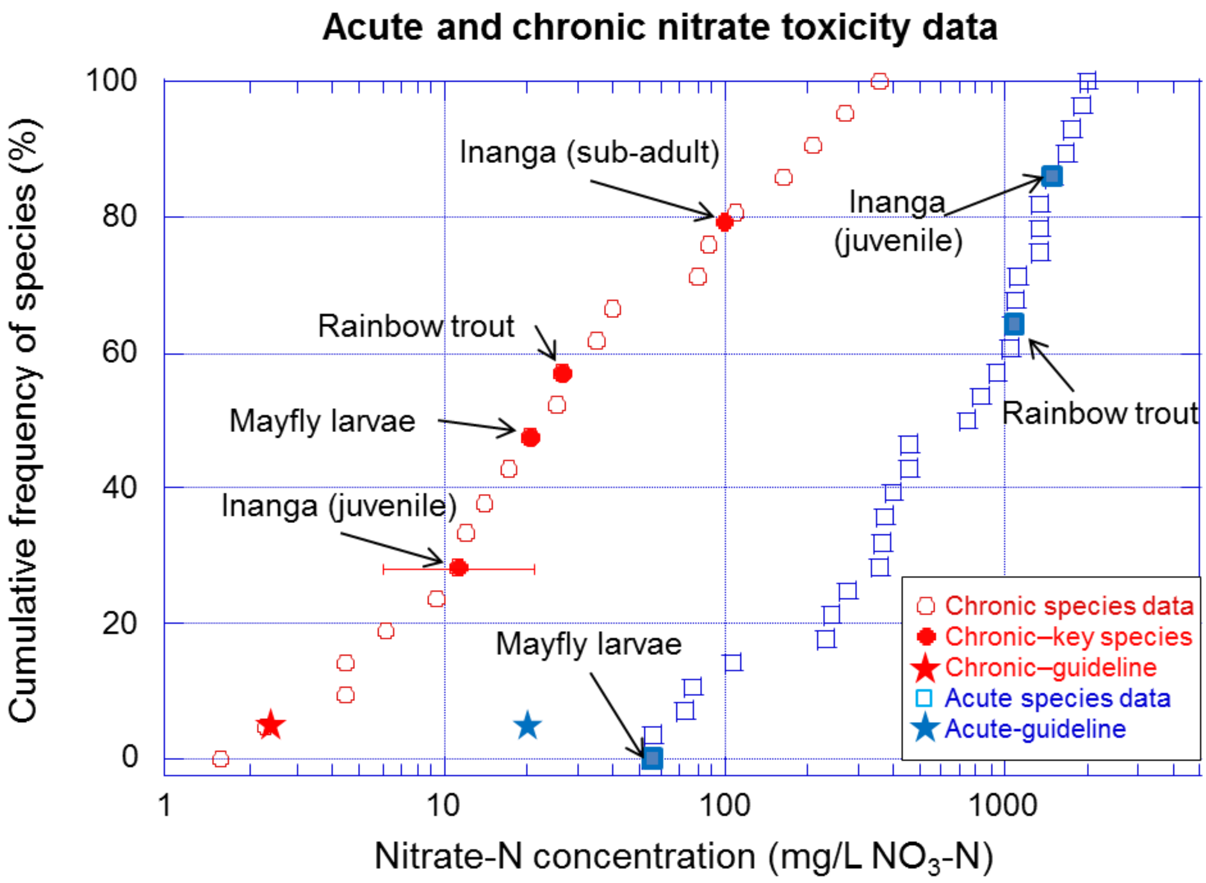

Figure 4-1: Species sensitivity distribution for acute and chronic nitrate-N exposure. Chronic NOEC data from Table 4-1 and calculated NOEC 95th percentile value are shown. Acute survival data from Table B-2. Juvenile inanga data range shown for tests at two hardness concentrations.

Table 4-1: Summary of data used for chronic nitrate guideline derivation. See Appendix B for detailed data.

Group	Common name	Scientific name	Life Stage	Duration (d)	Effect	Temp (°C)	Hardness of exposure water (mg/L as $CaCO_3$)	NOEC (mg/L NO_3^--N)	TEC (mg/L NO_3^--N)	Rank	Cum %
Fish	**Lake trout**	***Salvelinus namaycush***	**Embryo-Alevin-I**	**146d**	**DVP, GRO**	**7.5**	**10-16**	**1.6**	**3.2**	1	0
Fish	**Chinook salmon**	***Oncorhynchus tshawytscha***	**Fry**	**30d**	**MOR**	**10**	**8-10**	**2.3**	**3.2**	2	5
Fish	Lahontan cutthroat trout	*Salmo clarki*	Fry	30d	MOR	13	6-9	4.5	5.9	3	10
Fish	Coho salmon	*Oncorhynchus kisutch*	Fry	30d	MOR	10	8-10	>4.5	>4.5	4	14
Fish	Lake whitefish	*Coregonus clupeaformis*	Embryo-Alevin-Fry	126d	DVP	7.5	10-16	6.25	12.5	5	19
Amphibian	American Toad	*Bufo americanus*	Egg	23d	HAT	5-10	ND	>9.3	>9.3	6	24
Fish	**Inanga**	***Galaxias maculatus***	**Juveniles**	**40d**	**GRO**	**15**	**40**	**11.2***	**46.5***	7	29
Amphibian	Pacific treefrog	*Pseudacris regilla*	Tadpoles	10d	GRO	22	75	12.0	19.0	8	33
Invertebrate	Freshwater crayfish	*Astacus astacus*	NR	7d	MOR	15	ND	>14.0	14.0	9	38
Invertebrate	**Water flea**	***Ceriodaphnia dubia***	**Neonate**	**7d**	**REP**	**25**	**44-172**	**17.1***	**31.1***	10	43
Invertebrate	**Mayfly**	***Deleatidium* sp**	**Larvae**	**20d**	**MOR**	**15**	**40**	**20.3**	**35.0**	11	48
Invertebrate	Florida apple snail	*Pomacea paludosa*	Juveniles	14d	MOR	21-24	ND	25.3*	29.0*	12	52
Fish	**Rainbow trout**	***Oncorhynchus mykiss***	**Embryo-Alevin-Fry**	**30d**	**DVP, GRO**	**10**	**8-310**	**26.3***	**56.7***	13	57
Invertebrate	Freshwater prawn	*Macrobrachium rosenbergii*	Juveniles	21d	GRO	28	ND	35.0	62.0	14	62
Amphibian	African clawed frog	*Xenopus laevis*	Embryo	10d	GRO	22	21-24	40.4	58.5	15	67
Invertebrate	Midge	*Chironomus dilutus*	Larvae	10d	GRO	23	46-172	80.0*	113*	16	71
Invertebrate	Crustacean	*Hyalella azteca*	Juveniles	10d	GRO	23	46-310	88.1*	136*	17	76
Fish	**Inanga**	***Galaxias maculatus***	**Sub-adults**	**31d**	**MOR**	**15**	**40**	**>103**	**>103**	[a]	
Fish	Fathead minnows	*Pimephales promelas*	Embryos and larvae	11d	MOR, GRO	25	12-230	111*	159*	18	81
Amphibian	Red-eared frog	*Rana aurora*	Embryo	10d	GRO	15	26	162	195	19	86
Algae	**Green algae**	***Pseudokirchneriella subcapitata***	**Exponential**	**3d**	**GRO**	**24**	**10**	**206**	**289**	20	90
Fish	Topeka shiner	*Notropis topeka*	Juveniles	30d	GRO	24.5	210-230	268	361	21	95
Invertebrate	Water flea	*Daphnia magna*	Neonates	7d	REP	25	156-172	358	507	22	100

[a] Sub-adult inanga data not used in guideline derivation as juvenile growth a more sensitive endpoint; * indicates geometric mean of data from multiple studies or measurements over a range of hardness values.

Notes: Bold highlight indicates species resident in New Zealand.

Abbreviations: NOEC = no observed effect concentration; LOEC = lowest observed effect concentration DVP = development; GRO = growth (length or weight); MOR = mortality; HAT = hatching; REP = reproduction.

Toxicity modifying factors

The mechanisms regulating nitrate uptake and toxicity are not fully understood. Recent work has investigated the effect of water hardness on the toxicity of nitrate using both short-term and long-term toxicity tests. Short-term exposures were conducted with several fish and invertebrate species and showed linear relationships between the logarithm of toxicant concentration and logarithm of hardness for each of the test species [18]. A long-term study evaluating the relationship between water hardness and nitrate toxicity was also conducted using a 40-day embryo-alevin-fry test with rainbow trout (*O. mykiss*) [15]. However, the results did not definitively demonstrate the relationship between increasing hardness and nitrate toxicity. In some cases, sensitivity appeared greater in the moderately hard water (92 mg $CaCO_3$/L) compared to the soft water (50 mg $CaCO_3$/L). Chronic test results for native inanga juveniles showed they were 3.5-times more sensitive to nitrate in low hardness water compared to moderate hardness water, but that no acute toxicity response was observed (Table B-1, Table B-2). Overall, the trend for all species in both short-term and long-term exposures was one of decreasing toxicity with increasing hardness. However, because of the uncertainty in hardness-dependent short-term or long-term equations no hardness-modifying relationships were incorporated into the recent Environment Canada nitrate guideline derivation [8].

Nitrite toxicity and the potential for ameliorating effects of increased chloride ion concentrations is well documented for fish [19] and more recently for freshwater macroinvertebrates [20]. Because nitrite and chloride ions compete for the same transport site, elevated chloride concentrations in the environment have the potential of reducing nitrite toxicity. For this mechanism to occur the nitrate ions would require initial reduction to the nitrite ion, which would occur in the digestive tract prior to uptake from the digestive system. Because this is a less direct mechanism than for nitrite toxicity modification, where the primary interaction is at a gill surface, any chloride effects with nitrate would be most significant for chronic rather than acute exposures. Further studies will be required to demonstrate that chloride is a significant modifier of chronic nitrate toxicity.

5 Guideline derivation

The guideline values used in this report use the ANZECC methodology and the updated species database (Appendix B). The ANZECC TV derivation procedure is based on chronic NOEC values for all species and forms the basis of the Grading values shown in Table 5-1. The two-number guideline and management framework is based on: (i) guideline derivation using NOEC values to provide ecosystem protection for average long-term exposure – termed “Grading”; and (ii) threshold effect concentration (TEC)[5] values for management of seasonal maximum concentrations – termed “Surveillance”.

The proposed numeric chronic nitrate guideline values are based on NOEC and TEC effect thresholds. These are summarised together with narrative descriptors in Table 5-1. Background to the thresholds derivation procedure is provided in 0. The terms “Grading” and “Surveillance” are derived from the compliance descriptors applied to microbiological standards. The Grading values are derived from the species NOEC values and it is recommended that compliance should be based on the annual median concentration. The Surveillance values are derived from the species TEC values and it is recommended that compliance should be based on the annual 95th percentile of the monitoring data. The TEC value is a conservative sub-lethal measure of species tolerance to protect from seasonally high concentrations which is less than the statistically derived lowest observed effect concentration (LOEC) for each species.

The guideline and management framework is based on, and consistent with, the ANZECC (2000) guidelines [1] risk-based methodology to provide various levels of ecosystem protection. The compliance monitoring basis provides species protection for both average long-term exposure and seasonal maximum concentrations. This approach is considered suitable for protection from chronic nitrate exposure and differs from the monitoring regime that would be considered appropriate for other toxicants (e.g., metals). Generically, a higher level of conservatism is generally recommended for compliance monitoring for more highly toxic contaminants, such as metals, where adverse effects can occur in relatively short exposure periods. ANZECC (2000) recommends an annual 95th percentile concentration value should be compared with the TVs as a Tier I assessment approach.

[5] The threshold effect concentration (TEC) is the geometric mean of the no observed effect concentration (NOEC) and the lowest observed effect concentration (LOEC). The TEC value is below the lowest statistically significant effect concentration.

Table 5-1: Guideline derivations for nitrate-N. Grading guidelines are based on species NOEC values and Surveillance guidelines based on TEC values (see Table 4-1). Bold indicates default guideline values applicable to most waters.

Guideline Type	Grading Nitrate concentration (mg NO_3-N /L)	Surveillance Nitrate concentration (mg NO_3-N /L)	Description of Management Class
Chronic – high conservation value systems (99% protection)	1.0	1.5	Pristine environment with high biodiversity and conservation values.
Chronic – slightly to moderately disturbed systems (95% protection)	**2.4**	**3.5**	**Environments which are subject to a range of disturbances from human activities, but with minor effects.**
Chronic – highly disturbed systems (90% protection)	3.8	5.6	Environments which have naturally seasonally elevated concentrations for significant periods of the year (1-3 months).
Chronic – highly disturbed systems (80% protection)	6.9	9.8	Environment which are measurably degraded and which have seasonally elevated concentrations for significant periods of the year (1-3 months).
Acute	20	30	Environments which are significantly degraded. Probable chronic effects on multiple species.
Method of comparison	Annual median	Annual 95th percentile	

5.1 Adequacy of environmental protection

An evaluation of the environmental risk posed by a particular contaminant of concern involves consideration of multiple factors relating to an exposure and effects assessment. These include: the nature of the chemical contaminant; the exposure concentration and duration; factors affecting chemical toxicity and bioavailability; environmental fate of the chemical (including dilution/dispersion, degradation); the biodiversity and uniqueness of the receiving water communities; and the adequacy of the toxicological database used to derive the water quality guidelines.

Determining the adequacy of protection of a water quality guideline is ultimately a site-specific consideration, which involves an assessment of both the environmental monitoring for exposure (to characterise the spatial and temporal concentrations and durations) and the quality of the effects database. A tiered approach is generally applied to this assessment process. For example, a tiered assessment scheme is provided in the ANZECC (2000) guidelines for metal speciation guidelines (Figure 3.4.2, [1]). A risk-based approach is generally used to determine the need for further investigative assessments. Thus conservative 'worst case' assessments are generally made as the primary screening level in order to provide an initial measure of potential contaminant exposure. Subsequent investigations would be initiated if unacceptable risks were identified in the initial assessment.

5.1.1 Adequacy of international guidelines

The protocols for the derivation of numeric water quality guidelines are not consistent between international jurisdictions.

The ANZECC (2000) approach uses the statistically-derived chronic NOEC value for all available species, and calculates trigger values (TVs) statistically derived from the species sensitivity distribution for all species tested to calculated 95% protection and other thresholds [1]. This approach is considered to be more conservative than the approaches used in other jurisdictions [21, 22].

The U.S. EPA procedures use only data for species resident in North America, assembled a comprehensive acute and chronic databases. Both short-term and long-term criteria values are derived. The chronic criteria are derived using acute-to-chronic ratios to derive a chronic species sensitivity distribution. A chronic criteria value is calculated from the lower 5th percentile value (i.e., 95% protection) of the derived chronic dataset [9]. More recently, the new U.S. EPA ammonia guidelines [23] have been derived from chronic data using regression approaches to calculate the 20% effect value for each species and the 5th percentile value derived from the chronic distribution. The recently updated copper guideline uses the biotic ligand model to derive guideline values which incorporate water quality factors which affect copper bioavailability to organisms [24].

The Canadian approach uses the statistically-derived LOEC values for species and a guideline value statistically derived from the species sensitivity distribution for 95% protection based on this dataset [11]. This approach has been used for the recent nitrate guideline derivation [7]. Thus numeric guideline values will be expected differ between jurisdictions, even if the same species toxicity database is used.

The suitability of using NOECs for toxicity thresholds for deriving water quality guidelines has been a recent area of concern [25, 26]. The primary reasons are that NOEC and LOEC values are arbitrary values, set by the choice of exposure concentrations and calculated by statistical procedures, such as analysis of variance (ANOVA), which are affected by the variability of organism responses at a given exposure concentration. As such, the ability to detect a specific level of adverse effect may differ greatly between tests. Improved statistical methods are available using regression-based approaches with specific low-effect thresholds (e.g., EC_{10} or EC_{20}), however, the incorporation of solely regression-based techniques are not generally practical because these analyses have not been applied to historic test data. Consideration is being given to revising the statistical procedures in the current revision of the ANZECC guidelines.

5.1.2 Risk assessment to key species

The ANZECC (2000) guideline methodology has been applied to derive a range of nominal species protection levels based on the nitrate species sensitivity database (Table B-1). These will generally be conservative because NOEC and TEC values used in the derivation of the numeric guidelines that are below significant effects thresholds for the species and the most sensitive species in the database will determine the guideline values for the various statistically-derived levels of protection. However, lower levels of nominal protection may include some species which experience some chronic effects. Information on the level of protection for groups which are not well represented in the database may have higher uncertainty.

Table 5-2 provides an assessment of risks and potential impacts on species groups at the various protection levels in moderate hardness waters. All effects considered are sub-lethal (i.e., development, growth, reproduction, refer to Table 4-1) and occur at concentrations markedly lower than the lethality thresholds (Figure 4-1). This assessment is based on the narrative criteria provided in Table 5-2 (see footnotes) and the species sensitivity data from Table 5-1.

The relative acute lethality and sub-lethal chronic sensitivity distribution for all species is shown in Figure 4-1.The analysis indicates that most species groups have a negligible risk and that any impacts resulting from exceedance would be insignificant. The indication of low risk with moderate effects for Lake Trout, which are generally not resident in New Zealand rivers or lakes, would equate to a 10% growth reduction for that species and be 16-times below the lethality threshold for the fry.

The “minor” impact threshold for rainbow trout relates to concentrations below the LOEC values for this species in moderate hardness waters. No information is available for the early life-stage sensitivity of the New Zealand strain of rainbow trout in soft waters.

The toxicity data indicates that a revised site-specific assessment would need to be undertaken for low hardness (i.e., soft; hardness values <15 mg$CaCO_3$/L) waters. The limited native species data indicates that chronic growth and survival thresholds for inanga are markedly lower in soft water (Table B-1), with potentially ecologically significant effects occurring for the 80% level of protection. The very limited availability of native species data for low hardness waters prevents a risk assessment being undertaken for may key species.

5.1.3 Ecological effects thresholds

A toxicological concentration-response relationship provides a quantitative basis to determine effects thresholds for potential adverse ecological effects. The ‘Ecologically Significant Effects Concentrations’ (ESEC) values differ from the statistically-derived values in that they require a value judgement as to the level of effect for a given effect which would constitute an “ecologically significant effect” for a given species and effect.

International guidelines derived using a regression-based approach are limited and include the U.S. EPA freshwater ammonia guideline [23]. This uses a 20% response as a measure of adverse effect for survival, growth and reproduction effects. This level of effect represents a standardised approach to provide a measure which approximates both the detection limit of many toxicity tests and a threshold level for potential ecological concern. Guidelines derived using this procedure can be used for risk assessment applications and more detailed studies undertaken if high risks are indicated.

The appropriate ESEC values will differ between species and effects measures. While decisions on effects such as reduced survival and growth may be relatively uncontroversial, other measures; such as developmental delay, reduced hatching and physiological effects, are more subjective, largely because the linkages to population effects are less certain. Additionally, the time exposure of a contaminant is an important consideration in making these assessments. For example, an exposure concentration which exceeds a growth-effect threshold may not be relevant if occurring during a winter period when growth rates are minimal.

Thus the determination of ESEC values will largely be a site-specific consideration. This will require an assessment of both exposure and effects on a site or catchment basis. Key exposure factors are concentration variability, seasonal and spatial concentrations and fate in the receiving environment (including dilution/dispersion, biodegradation and bioaccumulation). Effects may require consideration of multiple species with the assessment including: key species of concern, relative toxicant sensitivity and the level of effect which is considered and ecologically adverse threshold. The location of various species life-stages would also consider breeding areas, juvenile and adult habitats. Generally, the toxicity data is derived for more sensitive early life-stages and are thus conservative for providing protection for adult life-stages.

Table 5-2: Qualitative risk summary for revised nitrate toxicity species protection levels in moderate hardness waters.

Target group	Level of protection - Level of risk / Impacts [1]			
	99%	95%	90%	80%
Fish eggs [2]	Negligible/Insignificant	Negligible/Insignificant	Negligible/Insignificant	Negligible/Insignificant
Native fish larvae/fry	Negligible/Insignificant	Negligible/Insignificant	Negligible(?)/Insignificant(?)	Negligible(?)/Insignificant(?)
Mature native fish	Negligible/Insignificant	Negligible/Insignificant	Negligible/Insignificant	Negligible(?)/Insignificant(?)
Lake Trout fry/fingerlings [3]	Negligible/Insignificant	Very low / Minor	Low / Moderate	Low / Moderate
Rainbow trout fry/fingerlings	Negligible/Insignificant	Negligible/Insignificant	Very low / Minor	Very low / Minor
Mature rainbow trout	Negligible/Insignificant	Negligible/Insignificant	Negligible/Insignificant	Negligible/Insignificant
Aquatic invertebrates	Negligible/Insignificant	Negligible/Insignificant	Negligible/Insignificant	Negligible/Insignificant

[1] Descriptors:

Risk (relates to the number of species exposed): Negligible = all target group protected for all conditions; Very low = only most sensitive species affected; Low = several sensitive species affected; Moderate = several sensitive species from multiple target groups; High = several species affected, including key local environment species; ? = minimal or insufficient data for this group.

Impacts (relates to the level of effect from the exposure): Insignificant = below all effects thresholds; Minor =chronic sensitivity threshold for the most sensitive species exceeded occasionally; Moderate = chronic sensitivity threshold for most sensitive species exceeded under average conditions; Major = value approaches acute sensitivity threshold.

See Table 5-1 for numeric values for protection levels for Grading and Surveillance.

[2] Based on 37-64d early life-stage (ELS) rainbow trout exposure (eggs, alevin, fry life-stages). Includes ELS data for New Zealand strain of rainbow trout.

[3] Most sensitive species. Only recorded as resident in Lake Pearson, Canterbury.

5.2 Site-specific guideline derivation

A site-specific guideline can be calculated by three alternative processes: (i) selection of local resident species from the acute or chronic datasets; (ii) recalculation using specific endpoints (e.g., recalculated from original data in publication); or (ii) selection of the most sensitive acute species and application of an acute-to-chronic ratio (ACR) to provide an estimated NOEC value. All these selection processes would include consideration of the water quality factors which may affect contaminant bioavailability and toxicity (e.g., temperature, hardness, pH).

The factors which affect nitrate toxicity are poorly known and have not been incorporated into the guidelines. General water quality factors, such as temperature, will affect organism sensitivity and the fate of nitrate in the receiving environment (e.g., affecting plant uptake rates and denitrification) but would not be specifically included as a site-specific modifier for toxicity thresholds. Both international and New Zealand species data indicate that water hardness significantly affects nitrate toxicity [3, 8], however, there is insufficient data to incorporate a hardness algorithm modifier in the guidelines [7]. Additional studies need to be undertaken to determine the factors responsible for the toxicity modification and to develop a modifier algorithm.

The site-specific datasets generally require information on key species, or genera directly related to key species relevant to a specific site. The chronic nitrate dataset contains only two native species, mayfly and inanga, with a total of eight of the 22 test species being resident in New Zealand freshwaters (Table 4-1). The very limited data on native species sensitivity restricts the ability to undertake site-specific guideline derivations based on local species. No site-specific assessment can be undertaken for hyporheic invertebrate communities which inhabit gravel-bed aquifers [27, 28], and potentially have elevated nitrate concentrations. Potential native species which may be suitable for providing additional data are addressed in the following section.

5.3 Recommended native species for additional testing

The ANZECC guidelines currently recommend that chronic data for at least five species are used for deriving risk-based guidelines using the statistical distribution approach [1]. However, an increased minimum number to eight species is currently being considered for the revised ANZECC guidelines in order to provide a more reliable statistical derivation.

A number of native freshwater invertebrate species have been evaluated for suitability for laboratory toxicity testing using internationally recognised standard protocols, and their sensitivity established for reference toxicants [29, 30]. Invertebrate community responses have also been measured for ammoniacal-nitrogen [31] and metals [32] using mesocosm techniques, and contamination gradients in natural streams for metals [33].The relative sensitivity of fish and invertebrate species differs between contaminants, making it difficult to make generalisations about the species which are most likely to be highly sensitive to toxicants.

Selection of potential native freshwater species for additional nitrate toxicity testing should include both invertebrate and fish species in order to cover a representative range of freshwater inhabitants. A summary of nine species is provided in Table 5-3, Priority species would include the common bully and eel elvas, which are both widespread and major inhabitants of lowland streams where nitrate concentrations tend to be highest. The invertebrate species include crustaceans (koura and amphipod), which tend to be highly sensitive to contaminants and the bivalve Sphaeriid, which is common in lowland streams and has been shown to be particularly sensitive to ammoniacal-N [34]. While sensitivity information for a large number of test species is desirable, selection criteria may need to be applied to prioritise species for chronic testing because of the expense of undertaking such tests.

A number of criteria are applicable to the selection of species for toxicity testing and are applied in Table 5-4 to the species recommended in Table 5-3. These include being widespread in the environment and of economical or cultural significance. While some species are highly culturally significant, such as eels and koura, their suitability for chronic laboratory testing is currently limited because the standard procedures have not been developed. These species are suitable for acute laboratory testing, however, the feeding and culture requirements to satisfy good growth during chronic tests have not yet been established. Such requirements are often difficult to develop, particularly to be able to supply sufficiently large quantities of food for early life-stages of many species.

Table 5-3: Suggested suite of native species for additional nitrate toxicity testing.

Group	Common name	Scientific name	Reference
Fish	Common bully	*Gobiomorphus cotidianus*	1
	Banded kokopu	*Galaxias fasciatus*	ND
	Smelt	*Retropinna retropinna*	1
	Eel (short and longfin)	*Anguilla australis, A. dieffenbachii*	1
Invertebrate	Crayfish, koura	*Paranephrops planifrons*	ND
	Amphipod	*Parcalliope fluviatalus*	1
	Bivalve	*Sphaerium novaezelandiae*	1
	Caddis	*Pycnocentria evecta*	1
	Mudsnail	*Potamopyrgus antipodarum*	1

[1] Hickey (2000) [29]; ND = no data

Table 5-4: Summary of criteria for selection of native species for chronic nitrate testing.

Suitability characteristic	Fish					Invertebrate				
	Common bully	Banded kokopu	Smelt	Eel		Crayfish	Amphipod	Bivalve	Caddis	Mudsnail
Native species	Y	Y	Y	Y		Y	Y	Y	Y	Y
Ecological importance	Y	Y	Y	Y		Y	Y	?	Y	Y
Wide distribution and abundance	Y	R	R	Y		Y	Y	R	Y	Y
Relevance to New Zealand culturally, recreationally or economically	N	Y	N	Y		Y	N	N	N	N
Potentially culturable or available for field collection year-round	Y	N	N	N		N	Y	Y	Y	Y
Taxonomically distinct and well described	Y	Y	Y	Y		Y	Y	Y	Y	Y
Toxicity testing protocols for similar species existing elsewhere	Y	N	N	N		Y	?	Y	Y	Y
Sensitivity to a wide range of contaminants	Y	?	?	N		?	Y	Y	?	?
Suitable for laboratory handling	Y	Y	Y	Y?		Y?	Y	Y	Y	Y

Abbreviations: Y = yes; N = no; R = restricted distribution; ? = uncertainty about this component; Y? = known suitability for acute laboratory procedures but chronic protocols not yet developed.
See Table 5-3 for further species details.

6 Discussion

This report updates the revised nitrate-N guidelines derived in 2009 [2]. This review includes a substantial number of new acute and chronic studies which have been undertaken in the interim period. Those studies include chronic data for two New Zealand native species. The additional data has resulted in revised chronic guideline values for nitrate-N using the ANZECC (2000) methodology. This database and NOEC-based guideline derivation will form the basis of the interim ANZECC nitrate guideline, which is expected to be promulgated in 2013. There was no change to the acute guideline value.

The proposed numeric chronic nitrate guideline values are based on NOEC and TEC effect thresholds. These are summarised together with narrative descriptors in Table 5-1. The terms "Grading" and "Surveillance" are derived from the compliance descriptors applied to microbiological standards. The Grading values are derived from the species NOEC values and it is recommended that compliance should be based on the annual median concentrations. These Grading values are equivalent to trigger values (TV) derived using the ANZECC (2000) methodology. The Surveillance values are derived from the species TEC values and recommended that compliance should be based on the annual 95th percentile of the monitoring data.

The two-number guideline approach differs from the ANZECC (2000) guidance which is based solely on the NOEC effect thresholds, with the general guidance for compliance based on the 95th percentile of the environmental monitoring data. The two-number guideline approach provides protection from chronic nature of nitrate toxicity based on annual median concentrations and addresses the need to provide robust limits for seasonally elevated nitrate concentrations in rivers. The TEC-derived guideline values are conservative in being derived from effect thresholds which are less than the statistically-derived LOEC values for the individual species.

The Environment Canada nitrate guidelines are derived from chronic LOEC data for both long-term and short-term-exposures [7]. The Canadian long-term guideline value of 3.0 mg NO_3-N/L is derived for 95% species protection and may be compared with the Grading and Surveillance values for 95% protection of 2.4 mg NO_3-N/L and 3.5 mg NO_3-N/L respectively (Table 5-1). Thus while there are methodological differences in derivation procedures, the final guideline values are similar.

Factors affecting nitrate toxicity:

A number of studies have identified water hardness as a factor affecting both acute and chronic toxicity in some species [8]. A hardness-related response was also found for chronic growth in juvenile inanga, but not for acute inanga exposures [3]. The mechanism for this effect is presently unknown so no algorithms were included in the Environment Canada guidelines to adjust for water quality factors [7]. However, the guidelines reported here should be considered conservative in that the most sensitive species in the database used to derive the nitrate guidelines is for long-term tests undertaken in very low hardness water – which would be expected to result in the highest nitrate toxicity.

Further chronic studies will be required to establish the water quality factor which results in the nitrate toxicity modification. The present use of water hardness provides a measure of indicative measure of the mineral content of the water, but cations are unlikely to be the modifying factor. Rather, chloride ion, which is known to modify nitrite toxicity [19, 20, 35] is the most likely factor. For nitrate the likely mechanism of effect would be for reduction of nitrate to occur in the gut and that chloride concentration would modify toxicity from a gut-mediated pathway. This differs from nitrite toxicity where the gill is the primary site of interaction with the toxicity modifier [19, 20].

Limitations of nitrate database:

The database is relatively limited in native species, with only seven species resident in New Zealand and two native species (mayfly and inanga). The results are conservative in that the most sensitive species, Lake Trout, were tested in very low hardness water. Higher hardness, as occurs in many New Zealand freshwaters [29], will reduce the toxicity of nitrate to freshwater species. Site-specific investigations may be required for sites with high nitrate concentrations present in waters with low mineral content.

There are no hyporheic-dwelling invertebrate species in the database. The most appropriate surrogate species would be amphipods, which are among the most acutely sensitive to nitrate but no chronic data is available. Further chronic studies with appropriate hyporheic species will be required to derive numeric guidelines which are appropriate for the protection of groundwater-dwelling species.

Proposed nitrate application:

The nitrate toxicity guidelines would be applicable to river and lake environments.

7 Recommendations

Some information gaps were identified in undertaking this review. These include: (i) the adequacy of native fish and invertebrate data for surface waters; (ii) absence of hyporheic species; and (iii) toxicity modification in relation to water mineral content (measured by hardness).

Nine native species (4 fish and 5 invertebrate) are recommended as potential candidates for both acute and chronic testing. A multi-criteria assessment was applied to these species to provide a basis for selection of priority species for chronic testing. Priority species would include the common bully and eel elvas, which are both widespread and major inhabitants of lowland streams where nitrate concentrations tend to be highest. However, there is presently no standard procedure for chronic testing with elvas, with no information on the quality and quantity of live feed required for early life-stage rearing. The priority invertebrate species would include crustaceans (koura and amphipod), which tend to be highly sensitive to contaminants and the bivalve Sphaeriid, which is common in lowland streams and has been shown to be particularly sensitive to ammoniacal-N.

Additional testing is also recommended to establish the key water quality factor affecting nitrate toxicity. This could be done for chronic test procedures using juvenile inanga and a range of chloride concentrations in low hardness water. It would also be useful if comparable measurements were undertaken for one or more macroinvertebrate species in order to determine the generality of the response relationship.

Field validation studies for nitrate toxicity should also be undertaken. These should be designed to measure species and community effects along nitrate concentration gradients in comparable waters with low mineral content. Such studies should include the ability to resolve sub-lethal effects on target species.

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9 Glossary of abbreviations and terms

ANZECC	Australian and New Zealand Environment and Conservation Council.
ARMCANZ	Agricultural and Resource Management Council of Australia and New Zealand
CAS	Chemical Abstracts Service.
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.
EC_{50} (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 EC_{50} is usually expressed as a time-dependent value (e.g., 24 hour or 96 hour LC_{50}).
Endpoint	Measured attainment response, typically applied to ecotoxicity or management goals.
ESEC	Ecologically Significant Effects Concentrations.
Guideline (water quality)	Numerical concentration limit or narrative statement recommended to support and maintain a designated water use.
HBRC	Hawke's Bay Regional Council.
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 ($CaCO_3$).
LC_{50}	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.
NPS-FW	National Policy Statement on Freshwater.
$NaNO_3$	Sodium nitrate.
NO_3^-	Nitrate ion.
NO_3-N	Nitrate-nitrogen.
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.
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.
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 or other test material 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.
Water quality criteria	Scientific data evaluated to derive the recommended quality of water for various uses.

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Appendix A Adequacy of proposed protection levels for management classes in relation to the Resource Management Act

Adequacy of proposed protection levels for management classes in relation to the Resource Management Act

This section provides a background to the nitrate-N guideline values developed for the range of management classes.

The ANZECC [1] numeric guideline protection levels are inherently precautionary and use no observed effect concentration (NOEC) data for sub-lethal chronic (long-term) effects measures (e.g., growth reproduction) for the derivation procedures, which in many cases are likely to be result in more conservative guideline values compared with the sensitivity of field monitoring approaches (e.g., reduced species diversity).

The ANZECC (2000) guidelines use a statistical approach and a risk-based methodology to determine numeric protection guidelines which are applicable to the narrative guidelines in the New Zealand Resource Management Act [12]. The use of the 99% protection guideline values was recommended for waters to be protected from ‘adverse effects’ [21]. This would correspond to pristine environmental with high conservation values and an ‘Excellent’ management classification.

For receiving waters requiring protection from ‘significant adverse effects’, the statistical approach can be used to vary the level of protection according to the values to be protected. The ANZECC (2000) descriptor: ‘slightly disturbed systems’ would correspond to this class and would be compared with a 95% protection guideline value. This equates to environments which are subject to a range of disturbances from human activities, but with minor human effects.

A management classification of ‘Fair’ would equate to environments which are measurably degraded and which have seasonally elevated nitrate concentrations for significant periods of the year. The ANZECC (2000) descriptor: ‘highly disturbed systems’ would correspond to this class and monitoring data would be compared with an 80% protection value.

The nitrate guidelines based on NOECs would be recommended to be compared with annual median nitrate concentrations measured in the receiving environments. The NOEC-based guideline values are termed ‘Grading values’ in Table 5-1.Seasonal variation in nitrate concentration in streams is often significant, with elevated concentrations occurring for 1 to 3 month periods, requiring some additional consideration of protective guidelines for species protection during these exposure periods. The statistical approach may also be used to derive guideline values based on the threshold effect concentration (TEC) values for each species to provide a protective threshold for elevated concentrations. Environment Canada uses the TEC values to derive their water quality guidelines and have recently published guidelines for nitrate in marine and freshwaters [7]. The TEC value is calculated as the geometric mean of the NOEC and the lowest observed effect concentration (LOEC) value for all species. As such, the TEC value is a concentration lower than the lowest statistically significant effect concentration.

The guidelines based on TEC values would be recommended to be compared with the annual 95th percentile nitrate concentrations measured in the receiving environments. The TEC-based guideline values are termed 'Surveillance values' in Table 5-1.

Appendix B Toxicity data used for guideline derivations

Table B-1: Chronic data used for guideline derivations.

Group	Common name	Scientific name	Life Stage	Duration (d)	Toxicity measure	Test endpoint	Temp (ºC)	Hardness (mg/L $CaCO_3$)	NOEC (mg/L NO_3-N)	LOEC (mg/L NO_3-N)	Reference
Fish	Lake trout	*Salvelinus namaycush*	Embryo-Alevin-Fry	146	NOEC [1]	GRO	7.5	10-16	1.6	6.25	1
Fish	Chinook salmon	*Oncorhynchus tshawytscha*	Fry	30	NOEC	MOR	10	8-10	2.3	4.5	2
Fish	Lahontan cutthroat trout	*Salmo clarki*	Fry	30	NOEC	MOR	13	6-9	4.5	7.6	2
Fish	Coho salmon	*Oncorhynchus kisutch*	Fry	30	NOEC	MOR	10	8-10	>4.5	>4.5	2
Fish	Lake whitefish	*Coregonus clupeaformis*	Embryo-Alevin-Fry	126	NOEC	DVP	7.5	10-16	6.25	25	1
Amphibian	American toad	*Bufo americanus*	Egg	23	NOEC	HAT	5-10	ND	>9.3	>9.3	6
Fish	Inanga (Common jollytail)	*Galaxias maculatus*	Juvenile	40	NOEC	GRO	15	14	6.0	20	5
Fish	Inanga (Common jollytail)	*Galaxias maculatus*	Juvenile	40	NOEC	GRO	15	40	20.8	108	5
								14-40	*11.2*	*Geometric mean*	
Amphibian	Pacific treefrog	*Pseudacris regilla*	Tadpole	10	NOEC	GRO	22	75	12.0	30.1	7
Invertebrate	Freshwater crayfish	*Astacus astacus*	Juvenile	7	NOAEL [1]	MOR	15	ND	>14.0	>14.0	8
Invertebrate	Water flea	*Ceriodaphnia dubia*	Neonates	7	NOEC	REP	25	156-172	7.1	14.1	9
Invertebrate	Water flea	*Ceriodaphnia dubia*	Neonates	7	NOEC	REP	25	156-172	56.5	113	9
Invertebrate	Water flea	*Ceriodaphnia dubia*	Neonates	7	NOEC	REP	25	156-172	7.1	14.1	9
Invertebrate	Water flea	*Ceriodaphnia dubia*	Neonates	7	NOEC	REP	25	156-172	17.9	35.9	9
Invertebrate	Water flea	*Ceriodaphnia dubia*	Neonates	7	NOEC	REP	25	156-172	17.9	35.9	9
Invertebrate	Water flea	*Ceriodaphnia dubia*	Neonates	7	NOEC [3]	REP	25	44	10.0	20	10
Invertebrate	Water flea	*Ceriodaphnia dubia*	Neonates	7	NOEC [3]	REP	25	98	20.0	40	10
Invertebrate	Water flea	*Ceriodaphnia dubia*	Neonates	7	NOEC [3]	REP	25	166	40.0	80	10
								44-172	*17.1*	*Geometric mean*	
Invertebrate	Mayfly	*Deleatidium* sp.	Larvae	20	NOEC [3]	MOR	15	40	20.3	60.1	11
Invertebrate	Florida apple snail	*Pomacea paludosa*	Juvenile	14	NOEC [2]	MOR	21-24	ND	23.2		12

Group	Common name	Scientific name	Life Stage	Duration (d)	Toxicity measure	Test endpoint	Temp (ºC)	Hardness (mg/L $CaCO_3$)	NOEC (mg/L NO_3-N)	LOEC (mg/L NO_3-N)	Reference
Invertebrate	Florida apple snail	*Pomacea paludosa*	Juvenile	14	NOEC [2]	MOR	21-24	ND	28.6		12
									25.3	*Geometric mean*	
Fish	Rainbow trout	*Oncorhynchus mykiss*	Fry	30	NOEC	MOR	10	8-10	1.1	2.3	2
Fish	Rainbow trout	*Oncorhynchus mykiss*	Fry	30	NOEC	MOR	10	8-10	>4.5	>4.5	2
Fish	Rainbow trout	*Oncorhynchus mykiss*	Embryo-Alevin-Fry	37-40	NOEC [3]	GRO	14	10	15.0	4.5	3
Fish	Rainbow trout	*Oncorhynchus mykiss*	Embryo-Alevin-Fry	37-40	NOEC [3]	GRO	14	50	15.0	4.5	3
Fish	Rainbow trout	*Oncorhynchus mykiss*	Embryo-Alevin-Fry	37-40	NOEC [3]	GRO	14	92	45.0	13.5	3
Fish	Rainbow trout	*Oncorhynchus mykiss*	Embryo-Alevin-Fry	37-40	NOEC [3]	GRO	14	176	405	>405	3
Fish	Rainbow trout	*Oncorhynchus mykiss*	Embryo-Alevin-Fry	64	NOEC [3]	GRO	14	310	115	240	4
Fish	Rainbow trout	*Oncorhynchus mykiss*	Embryo-Alevin-Fry	42	NOEC	DEV	16	40	99	389	5
								8-310	*26.3*	*Geometric mean*	
Invertebrate	Freshwater prawn	*Macrobrachium rosenbergii*	Juvenile	21	NOEC [2,3]	GRO	28	ND	35.0	62.0	13
Amphibian	African clawed frog	*Xenopus laevis*	Tadpole	5-10	NOEC [4]	GRO	22	21-36	40.4	58.5	7,14
Invertebrate	Midge	*Chironomus dilutus*	Larvae	10	NOEC [3]	GRO	23	46	40.0	80	10
Invertebrate	Midge	*Chironomus dilutus*	Larvae	10	NOEC [3]	GRO	23	86	80.0	160	10
Invertebrate	Midge	*Chironomus dilutus*	Larvae	10	NOEC [3]	GRO	23	172	160	320	10
								46-172	*80.0*	*Geometric mean*	
Invertebrate	Amphipod	*Hyalella azteca*	Juvenile	14	NOEC	GRO	23	46	10.0	20	10
Invertebrate	Amphipod	*Hyalella azteca*	Juvenile	14	NOEC	GRO	23	86	80.0	160	10
Invertebrate	Amphipod	*Hyalella azteca*	Juvenile	14	NOEC	GRO	23	172	160	320	10
Invertebrate	Amphipod	*Hyalella azteca*	Juvenile	10	NOEC	GRO	23	310	470	965	4
								46-310	*88.1*	*Geometric mean*	

Group	Common name	Scientific name	Life Stage	Duration (d)	Toxicity measure	Test endpoint	Temp (ºC)	Hardness (mg/L $CaCO_3$)	NOEC (mg/L NO_3-N)	LOEC (mg/L NO_3-N)	Reference
Fish	Inanga (Common jollytail)	*Galaxias maculatus*	Sub-adult	31	NOEC [3,5]	MOR	15	40	>103	>103	11
Fish	Fathead minnows	*Pimephales promelas*	Embryo-larvae	7	NOEC	MOR, GRO	25	140-170	358	717	9
Fish	Fathead minnows	*Pimephales promelas*	Larvae	7	NOEC [3]	MOR,GRO	25	12	50	100	10
Fish	Fathead minnows	*Pimephales promelas*	Larvae	7	NOEC [3]	MOR	25	50	100	200	10
Fish	Fathead minnows	*Pimephales promelas*	Larvae	7	NOEC [3]	MOR,GRO	25	94	200	400	10
Fish	Fathead minnows	*Pimephales promelas*	Larvae	7	NOEC [3]	MOR	25	168	200	400	10
Fish	Fathead minnows	*Pimephales promelas*	Juvenile	30	NOEC [3]	MOR	23.1	210-230	58	121	15
Fish	Fathead minnows	*Pimephales promelas*	Embryo-larvae	32	NOEC [3]	MOR,GRO	25	132-180	49	109	16
								12-230	*111*	*Geometric mean*	
Amphibian	Red-legged tree frog	*Rana aurora*	Embryo	10	NOEC	GRO	25.5	25.5	162	235	17
Fish	Topeka shiner	*Notropis topeka*	Juvenile	30	NOEC [3]	GRO	24.5	210-230	268	486	15
Invertebrate	Water flea	*Daphnia magna*	Neonates	7	NOEC	REP	25	156-172	358	717	9
Alga	Green alga	*Pseudokirchneriella subcapitata*	Exponential	3	NOEC [3]	GRO	24	~10	206	406	10

[1] NOEC: No observed effect concentration; NOAEL: No observed adverse effect level.
[2] Converted from Chronic EC_{50} (NOEC = EC_{50}/5).
[3] Not included in Hickey and Martin (2009) [2] guideline derivation.
[4] Data corrected from Hickey and Martin (2009) [2].
[5] Sub-adult live stage not included in inanga data as juvenile data available and more sensitive endpoint.
Abbreviations: DVP = development; GRO = growth (length or weight); MOR = mortality; HAT = hatching; REP = reproduction.
References: 1, McGurk et al. (2006) [13]; 2, Kinchloe et al. (1979) [14]; 3, Nautilus Environmental (2011a) [15]; 4, Stantec (2006) [16]; 5, Hickey et al. (2013) [3]; 6, Laposata & Dunson (1998) [36]; 7, Schuytema & Nebeker (1999c) [37]; 8, Jensen (1996) [38] ; 9, Scott & Crunkilton (2000) [39]; 10, Nautilus Environmental (2011b) [18]; 11, Martin & Thompson (2012) [4]; 12, Corrao et al. (2006) [40]; 13, Wickins (1976) [17]; 14, Schuytema & Nebeker (1999) [37, 41]; 15, Adelman et al. (2009) [42]; 16, US EPA (2010) [43];17, Schuytema & Nebeker (1999b).

Table B-2: Acute data used for guideline derivations.

Group	Common name	Latin Name	Life Stage	Duration (h)	Endpoint	Effect	Temp (°C)	Hardness (mg/L $CaCO_3$)	EC_{50}/ LC_{50} (mg NO_3-N/L)	EC_{50}/ LC_{50} (mg NO_3-N/L)	Rank	Cumulative %	Ref
Invertebrate	**Mayfly**	***Deleatidium* sp.**	**Larvae**	**240h [a]**	**LC50**	**MOR**	**15**	**40**		**54.9**	**1**	0	**1**
Invertebrate	Amphipod	*Echinogammarus echinosetosus*	Adults	120h	LC50	MOR	17.9			56.2	2	3.5	2
Invertebrate	Amphipod	*Eulimnogammarus toletanus*	Adults	120h	LC50	MOR	17.9			73.1	3	7.1	2
Invertebrate	Caddisfly	*Hydropsyche accidentalis*	Last instar larvae	120h	EC50	MOR	18			77.2	4	11	3
Invertebrate	Caddisfly	*Cheumatopsyche pettiti*	Early instar larvae	120h	LC50	MOR	18			107	5	14	3
Invertebrate	Caddisfly	*Hydropsyche exocellata*	Last instar larvae	120h	LC50	MOR	17.9			230	6	18	2
Invertebrate	Amphipod	*Hyalella azteca*	Juveniles	96h	LC50	MOR	23	44	168				4
Invertebrate	Amphipod	*Hyalella azteca*	Juveniles	96h	LC50	MOR	23	100	485				4
Invertebrate	Amphipod	*Hyalella azteca*	Juveniles	96h	LC50	MOR	23	164	921				4
Invertebrate	Amphipod	*Hyalella azteca*	Juveniles	96h	LC50	MOR	22.5	117	667				5
Invertebrate	Amphipod	*Hyalella azteca*	Juveniles	96h	LC50	MOR	22	80-84	16.4				6
Invertebrate	Amphipod	*Hyalella azteca*	Juveniles	96h	LC50	MOR		*44-164*		*241*	7	21	
Invertebrate	Midge	*Chironomus dilutis*	Juveniles	48h	LC50	MOR	22.0	84-136		278	8	25	6
Invertebrate	Fatmucket mussel	*Lampsilis siliquoidea*	Juveniles	96h	LC50	MOR	20.0	90-92		357	9	29	6, 5
Invertebrate	Fingernail clam	*Sphaerium simile*	Juveniles	96h	LC50	MOR	22.8	90-92		371	10	32	6, 5
Invertebrate	**Water flea**	***Ceriodaphnia dubia***	**Neonates**	**48h**	**LC50**	**MOR**	**25**			**374**	**11**	36	**7**
Fish	Siberian sturgeon	*Acipenser baeri*	Adults	96h	LC50	MOR	22.5	260		397	12	39	8
Invertebrate	Stonefly	*Amhinemura delosa*	Juveniles	96h	LC50	MOR	12.5	88-92		456	13	43	6, 5
Invertebrate	Water flea	*Daphnia magna*	Neonates	48h	EC50	MOR	25	156-172		462	14	46	7
Fish	Fathead minnows	*Pimephales promelas*	Larvae	96h	LC50	MOR	25.0	90-92	415				6
Fish	Fathead minnows	*Pimephales promelas*	Larvae	96h	LC50	MOR	25.0	156-172	1341				7
Fish	Fathead minnows	*Pimephales promelas*	Larvae	96h	LC50	MOR	25.0	*90-172*		*746*	15	50	

Group	Common name	Latin Name	Life Stage	Duration (h)	Endpoint	Effect	Temp (°C)	Hardness (mg/L $CaCO_3$)	EC_{50}/ LC_{50} (mg NO_3-N/L)	EC_{50}/ LC_{50} (mg NO_3-N/L)	Rank	Cumulative %	Ref
Invertebrate	Stonefly	*Allocapnia vivipara*	Juveniles	96h	LC50	MOR	11.0	99		836	16	54	5
Invertebrate	Washboard mussel	*Megalonaias nervosa*	Juveniles	96h	LC50	MOR	20.9	90-92		937	17	57	6, 5
Invertebrate	**Snail**	***Potamopyrgus antipodarum***	**Adults**	**96h**	**LC50**	**MOR**	**20.4**			**1042**	**18**	61	**8**
Fish	**Chinook salmon**	***Oncorhynchus tshawytscha***	**fingerlings**	**168h**	**LC50**	**MOR**	**13-14**			**1084**	**19**	64	**9**
Fish	Eastern mosquitofish	*Gambusia holbrooki*	Juveniles	96h	LC50	MOR				1095	20	68	10
Fish	**Lake Trout**	***Salvelinus namaycush***	**Fry**	**96h**	**LC50**	**MOR**	**7.5**	**10**		**1121**	**21**	71	**11**
Fish	**Rainbow trout**	***Oncorhynchus mykiss***	**Fingerlings**	**96h**	**LC50**	**MOR**	**15**	**11**	**691**				**4**
Fish	**Rainbow trout**	***Oncorhynchus mykiss***	**Fingerlings**	**96h**	**LC50**	**MOR**	**15**	**54**	**1436**				**4**
Fish	**Rainbow trout**	***Oncorhynchus mykiss***	**Fingerlings**	**96h**	**LC50**	**MOR**	**15**	**90**	**1768**				**4**
Fish	**Rainbow trout**	***Oncorhynchus mykiss***	**Fingerlings**	**96h**	**LC50**	**MOR**	**15**	**164**	**1768**				**4**
Fish	**Rainbow trout**	***Oncorhynchus mykiss***	**Fingerlings**	**168h**	**LC50**	**MOR**	**13-14**	**ND**	**1061**				**9**
Fish	**Rainbow trout**	***Oncorhynchus mykiss***	**Fingerlings**	**168h**	**LC50**	**MOR**	**12**	**40-42**	**1658**				**12**
Fish	**Rainbow trout**	***Oncorhynchus mykiss***	**Fingerlings**	**96-168h**	**LC50**	**MOR**	***7.5-15***	***10-164***		***1327***	**22**	75	
Fish	Topeka shiner	*Notropis topeka*	Juveniles	96h	LC50	MOR	24.5	210-230		1354	23	79	13
Fish	Catfish	*Ictalurus punctatus*	Fingerlings	96h	LC50	MOR	22			1355	24	82	14
Fish	**Whitebait, Inanga**	***Galaxias maculatus***	**Juveniles**	**96h**	**LC50**	**MOR**	**15**	**14**	**1549**				**15**
Fish	**Whitebait, Inanga**	***Galaxias maculatus***	**Juveniles**	**96h**	**LC50**	**MOR**	**15**	**41**	**1427**				**15**
Fish	**Whitebait, Inanga**	***Galaxias maculatus***	**Juveniles**	**96h**	**LC50**	**MOR**	**15**	**100**	**1482**				**15**
Fish	**Whitebait, Inanga**	***Galaxias maculatus***	**Juveniles**	**96h**	**LC50**	**MOR**	**15**	***14-100***		**1485**	**25**	86	

Group	Common name	Latin Name	Life Stage	Duration (h)	Endpoint	Effect	Temp (ºC)	Hardness (mg/L $CaCO_3$)	EC_{50}/ LC_{50} (mg NO_3-N/L)	EC_{50}/ LC_{50} (mg NO_3-N/L)	Rank	Cumulative %	Ref
Amphibian	African clawed frog	*Xenopus laevis*	Tadpoles	96h	LC50	MOR	22	21		1656	26	89	16
Amphibian	Pacific Treefrog	*Pseudacris regilla*	Tadpoles	96h	LC50	MOR	22	70-80		1750	27	93	16
Fish	Lake Whitefish	*Coregonus clupeaformis*	Fry	96h	LC50	MOR	7.5	10		1903	28	96	11
Fish	Bluegill	*Lepomis macrochirus*	Fingerlings	96h	LC50	MOR	22	45-46		1974	29	100	17

[a] Longer duration than normal acute response. Data included provide a conservative acute response measure for key native species.

Abbreviations: LC_{50} = Lethal concentration causing 50% response; EC_{50}= Concentration which is effective in causing measured response; MOR = Mortality.

Notes: Bold highlight indicates species resident in New Zealand freshwaters.

References: 1, Martin & Thompson (2012) [4]; 2, Camargo et al. (2005) [44]; 3, Camargo & Ward (1995) [45]; 4, Nautilus (2012) [18]; 5, Soucek & Dickinson (2012) [46]; 6, US EPA (2010) [43]; 7, Scott & Crunkilton (2000) [39]; 8, Hamlin (2006) [47]; 9, Westin (1974) [48] ; 10, Wallen et al. (1957) [49]; 11, McGurk et al. (2006) [13]; 12, Buhl & Hamilton (2000) [50]; 13, Adelman et al. (2009) [42]; 14, Colt & Tchobanoglous (1976) [51]; 15, Hickey et al. (2013) [5]; 16, Schuytema & Nebeker (1999c) [37]; 17, Trama (1954) [52]