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PROVINCE OF BRITISH COLUMBIA

AMBIENT WATER QUALITY GUIDELINES FOR IRON

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Prepared By:

Burke Phippen, Cheryl Horvath,
Rick Nordin and Narender Nagpal

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TABLE OF CONTENTS

LIST OF TABLES AND FIGURES

1.0 INTRODUCTION

Iron is an absolute requirement for all forms of life. Importance of iron is especially notable in biogeochemical processes because of its unique ability to serve as both an electron donor and acceptor and thus can play an important role in metabolic processes of many organisms.

Iron can also be potentially toxic at high concentrations. Iron's ability to donate and accept electrons means that if iron is free within the cell, it can catalyze the conversion of hydrogen peroxide into free radicals. Free radicals can cause damage to a wide variety of cellular structures, and ultimately kill the cell (Crichton *et al*. 2002). To prevent that kind of damage, life forms have evolved a biochemical protection mechanism by binding the iron atoms to proteins. This allows the cells to use the benefits of iron, but also limit its ability to do harm (Andrews 1999). The most important group of iron-binding proteins is the heme molecules, all of which contain iron at their centers. Organisms use variants of heme to carry out redox reactions and electron transport processes. Iron is required for oxidative phosphorylation, the process that is the principal source of energy for cells; without it, cells would die. In higher organisms, iron is also an essential component of myoglobin which stores oxygen in muscle cells.

1.1 Occurrence and Behaviour in the Environment

Iron is the fourth most abundant element by weight in the earth's crust, and is often a major constituent of soils (especially clays). It has an atomic number of 26 and atomic weight of 55.85 and exists primarily (91.7%) as ^{56}Fe but has one radioactive isotope (^{54}Fe, 5.8% of total mass) and two stable isotopes ^{57}Fe (2.2%) and ^{58}Fe (0.3%). Moore (1991) reviews iron as an inorganic contaminant in water and summarizes relevant production, chemistry and toxicity.

Solubility of iron in water varies with compound and temperature (as well as pH and other physical factors). For example, iron (III) chloride ($FeCl_3 \cdot 6H_20$) is very soluble with solubility ranging from 919 g/l at 20 ºC to 5251 g/L at 80 ºC. Iron (II) sulfate ($FeSO_4 \cdot 7H_2O$) is also very soluble at 266 g/L at 20 ºC, whereas the solubility of iron (II)

bicarbonate is relatively much less soluble (0.77 g/L at 18 ºC). Of primary biological importance in freshwater aquatic systems are iron (III) hydroxide ($Fe(OH)_3$) with very low solubility of 4.8×10^{-8} g/L (0.048 µg/L) at 18 ºC, and in contrast, iron (II) hydroxide $Fe(OH)_2$, being relatively more soluble at 1.42×10^{-3} g/L (1,420 µg/L) at 20 ºC.

The relationship between the relative proportions of the almost insoluble ferric Fe^{3+} iron and the bioavailable and bioactive ferrous (Fe^{2+}) (II) iron varies with a wide range of factors including pH, dissolved oxygen, dissolved and total organic carbon (DOC/TOC) ratio, color, humic and other organic acids, exposure to sunlight and chloride concentration (Davison and DeVitre 1982, Stumm and Morgan 1981). It is, therefore, extremely important when discussing iron concentrations in water to distinguish between iron in an ionic state (dissolved iron) and iron in a suspended particulate state (measured typically as total iron). In general, dissolved (Fe^{2+}) iron occurs at low concentrations in well-oxidized waters with near-neutral pH (Den Dooren de Jong 1965).

1.2 Sources in the Environment and uses

Anthropogenic sources of iron in surface water are often related to mining activities. Iron pyrites (FeS_2), common in coal seams, are exposed to weathering and bacterial action by mining. The oxidation of these pyrites results in the production of sulphuric acid and the release of soluble ferrous (Fe^{2+}) iron (Smith *et al.* 1973; cited in Smith and Sykora 1976).

As noted above, ferrous iron is generally chemically unstable in water and exists in this state only between pH 4.0 and 5.0 in low oxygen conditions (Freda 1991). As pH and the partial pressure of oxygen (pO_2) increase, ferrous iron oxidizes to its ferric (Fe^{3+}) forms (e.g. ferric hydroxide, $Fe(OH)_3$) (Smith and Sykora 1976). In these forms it is insoluble and may precipitate from solution, sometimes producing a thick sludge on the bottom of streams (Smith and Sykora 1976). However, Fe^{3+} that is complexed with organic compounds can be photo-reduced by UV light to the ferrous or soluble state, which can cause large diurnal fluctuations in the speciation and concentration of iron (Francko and Heath 1982 and Sulzberger *et al.* 1990 cited in Vuori 1995). The following equations

represent the simplified oxidation reaction for ferrous and ferric iron (Sykora *et al.* 1972a):

$2\ FeS_2 + 7\ O_2 \rightarrow 2\ FeSO_4 + H_2SO_4$ (ferrous)

$4\ FeSO_4 + O_2 + 10\ H_2O \rightarrow 4\ Fe(OH)_3 + 4\ H_2SO_4$ (ferric)

Industrial uses of iron compounds are many as identified in the table below:

Industrial Uses of iron compounds (U.S.EPA, 1993)

Form of Iron	Use
Iron (III) acetate: $(Fe(C_2H_3O_2)_3$	Dyeing of cloth
Iron (III) ammonium oxalate $(Fe(NH_4)_3(C_2O_4)_4)$	Blueprints
Iron (III) arsenate ($FeAsO_4$)	Insecticides
Iron (III) chloride ($FeCl_3$)	Water purification and sewage treatment, in the dyeing of cloth, as an additive in animal feed, and as an etching material for engraving, photography and printed circuits
Iron (III) chromate ($Fe_2(CrO_4)_3$)	Yellow pigment for paints and ceramics
Iron (III) hydroxide ($Fe(OH)_3$)	Brown pigment for rubber and in water purification systems
Iron (III) phosphate ($FePO_4$)	Fertilizer and additive in human and animal food
Iron (II) acetate $(Fe(C_2H_3O_2)_2$	Dyeing of fabrics and leather, and as a wood preservative
Iron (II) oxalate (FeC_2O_4)	Yellow pigment for paints, plastics, glass and ceramics, and in photography
Iron (III) sulfate, iron (II) sulfate, and iron (II) sulfate heptahydrate	In water purification and sewage treatment systems; catalyst for ammonia production; an ingredient in fertilizer, moss control and herbicide; additive in animal feed, wood preservative, and flour to increase iron levels (University of Sheffield 2007)

1.3 Levels in the Environment

Ambient concentrations of dissolved iron in the environment are typically very low. At depth in the ocean, dissolved iron (Fe^{+2} + less than 0.45µm iron) concentrations are generally constant at 0.6 nM (1.0 nM = 55.85 ng/L iron) or 33.5 x 10^{-9} mg/L (Haese 2006). In surface Antarctic marine waters where iron is considered to be limiting to biological productivity, Sedwick *et al*. (2000) reported dissolved iron concentrations of 0.23 nM to 1.0 nM (12.8 x 10^{-9} mg/L to 55.8 x 10^{-9} mg/L) and considered 0.5nM (27.9 x 10^{-9} mg/L) to be a high concentration for this ecosystem. To deal with the low concentrations of this essential element, phytoplankton and bacteria have evolved the ability to synthesize chelators commonly called siderophores. Siderophores, organic in nature, consist of low molecular weight compounds that have a high affinity for ferric iron. They are secreted out of the microorganism where they form a complex with ferric iron. After transport into the cell, the chelated ferric iron is enzymatically reduced and released (thus become available as ferrous ion) from the siderphores, which is secreted again for further complexation (Haese 2006).

In freshwater, in undisturbed areas, dissolved iron is generally less than normal analytical detection limits (5 µg/L - ICP), the only usual exception is the hypolimnia of productive lakes where oxygen depletion results in iron release from anoxic sediments where iron is typically bound to phosphorus in its ferric (Fe^{3+}) form. Low dissolved oxygen allows the conversion from ferric to the more soluble ferrous form and releases the iron from the phosphorus. In effluents where oxygen is low and pH acidic, dissolved iron (Fe^{2+}) may be present in concentrations of concern. In storm water runoff where erosion of soil is a factor, total iron concentrations may be high – and generally correlated with suspended sediments, but dissolved iron is very low (Brandstetter 1998). For dissolved iron (dissolved being able to pass through a 0.45µm filter – but may contain colloidal material as well as truly dissolved ions) ambient concentrations can be surprisingly high depending on circumstances. In a river in Colorado, the dissolved (presumably largely ferrous) iron concentration up to 2.6 mg/L was measured (Fey *et al*. 2002). These investigators found that the dissolved iron showed an inverse response to turbidity and

total iron concentrations. They felt that photoreduction of ferric iron from colloidal material to dissolved ferrous iron might have been occurring in this situation – related to shading caused by diurnal turbidity patterns. Total iron concentrations in freshwater can be very high (100 mg/L or more) since the typical analytical techniques would include any suspended soil particles.

Groundwater can have high concentrations of dissolved iron (20 mg/L) (Iowa 2005). Sediment pore water concentrations in mining areas can have very high concentrations of dissolved iron (Fe II) – up to 2000 mg/L (Nordstrom *et al*. 1999).

Iron hydroxides produced in water and iron-humus colloids can affect fish by clogging gills and reducing respiratory potential and subsequent survival (Dalzell and MacFarlane 1999, Lehtinen and Kingstedt 1983 and Peuranen *et al*. 1994), food availability by benthos (Gerhardt 1992, Randal et al 1999), altering the structure and quality of aquatic benthic habitats (McKnight and Feder 1984, Letterman and Mitsch 1978, Scullion and Edwards 1980). Reduced abundance and species diversity of periphyton, benthic invertebrates and fish are typically reported as consequences of high iron concentration in freshwater (McKnight and Feder 1984, Letterman and Mitsch 1978, Scullion and Edwards 1980, Koryak et al 1972, Greenfield and Ireland 1978, Rasmussen and Lindegaard 1988).

2.0 Aquatic Toxicity of Iron

2.1 Fish

2.1.1 Mechanisms of Toxicity

One of the difficulties associated with trying to measure iron toxicity in a laboratory environment is that the addition of free iron to solutions results in the formation of iron hydroxide and a subsequent drop in pH. For example, when Havas and Hutchinson (1982) added 30 mg/L of Fe^{3+} to water, the pH of water decreased from 8.2 to 4.5 in 50 hours. Therefore, especially in early studies, it was difficult to carry out toxicological tests with a stable form of iron and maintain control over pH and P_{O2} (Maltby *et al.* 1987).

Doudoroff and Katz (1953) prepared a review of toxicity studies on many metals including iron to various fish species. In their report, they discuss inconsistencies in reported toxicity values for iron in studies prior to 1953 (Table 1). They questioned the validity of many of these studies since pH was not reported, and suggested that the increase in acidity resulting from the addition of iron may have actually been the cause of mortality. Therefore it would appear that iron toxicity might, in some cases, be linked to the toxicity of low pH.

A possible mechanism for dissolved iron toxicity involves a disruption of the sodium balance. Gonzalez *et al.* (1990) describe a large decrease in body sodium concentrations in brook charr (*Salvelinus fontinalis*) exposed for two days to iron concentrations near the estimated LC_{50} (18 μM or 1.0 mg/L). This effect was similar to that caused by exposure to manganese in the same experiment. However, concentrations of sodium in the plasma were not affected by iron, and a doubling of iron concentrations did not result in a greater loss of body sodium. Further, increasing calcium concentrations in the water had no consistent influence on the LC_{50}. Further study is needed to determine the actual mechanisms of iron toxicity based on sodium balance.

Table 1: A compilation of the reported toxicity values expressed in a literature review by Doudoroff and Katz (1953).

Species	Iron concentration (mg/L)	Iron salt used	Test Duration to death	Other variables	Reference *
Eels	4.9	$FeCl_3$	50 hours	T=20°C –22°C	Oshima (1931)
Eels	12.7	$FeCl_2$	50 hours	T=20°C –22°C	Oshima (1931)
Eels	14.3	$KFe(SO_4)_2$	25 hours	T=20°C –22°C	Oshima (1931)
Various spp.	0.2 mg/L	$FeCl_3$		pH 7.2-7.4	Minkina (1946)
Various spp.	0.1	Ferric sulphate	24 hours		Clark & Adams (1913)
Various spp.	1.28	Ferrous sulphate	24 hours		Clark & Adams (1913)
Goldfish	100	$FeCl_3$	4 days did not kill	pH 5.5, hard H_2O	Ellis (1937)
Goldfish	10	$FeCl_3$	4 days did not kill	pH 5.0, soft H_2O	Ellis (1937)
Goldfish	1000	Ferrous sulphate	2-10 hours	pH 6.4, hard H_2O	Ellis (1937)
Black bass & bluegill sunfish	100	Ferrous sulphate	2.5-7 days	Tap water, pH 6.4	Sanborn (1945)
Goldfish	100	Ferrous sulphate	7 days = no kill	pH 6.7-6.4	Sanborn (1945)
Black bass & bluegill sunfish	50	Ferrous sulphate	7 days = no kill	pH 6.6	Sanborn (1945)
Very young carp	1000	Ferrous sulphate	48 hours		Dyk (1942)
Brook trout	133	Ferrous sulphate	24 hours		Belding (1927)
Various spp.	0.9	Dissolved iron		pH 6.5-7.5, well aerated	Bandt (1938)
Pike & tench	1.9	Dissolved iron		pH 6.7	Schaeperclaus (1941)

*as cited in Doudoroff and Katz (1953)

At higher pH values (above about pH 5.0, where iron is less soluble), one possible mechanism of iron toxicity is the precipitation of ferric hydroxide ($Fe(OH)_3$) directly onto the gills of fish in the form of a brown slime (Doudoroff and Katz 1953; Schaeperclaus 1954 cited in Sykora *et al.* 1972b). Damage can result either from the clogging action of the precipitate (which would interfere directly with respiration) or from possible injury to the tissues by corrosive action (Larson and Olsen 1950, cited in Brenner *et al.* 1976). Peuranen *et al.* (1994) found that surviving brown trout (*Salmo trutta*) exposed to 2 mg/L iron ($FeCl_3$:$FeSO_4$ in 1:1 ratio, with and without humic acid) in water of pH 5.0 and 6.0 suffered fusion of gill lamellae, separation of the outer epithelial layer, hypertrophy and necrosis of the lamellar epithelium. Iron was detected only at the gill epithelium, not inside, which indicated that the acute metal toxicity was mediated through action on the

gill surface. Dalzell and MacFarlane (1999) used 96-h LC_{50} bioassays of brown trout (*Salmo trutta*) to examine the toxicity of a commercial iron (III) sulfate liquor used for treating reservoirs to reduce algal growth and the analytical grade of iron sulfate. They found that the 96-h LC_{50} for the iron sulfate liquor was 28 mg/L total iron (0.05 mg soluble iron) and the 96-h LC_{50} for the analytical grade iron sulfate was 47 mg/L total iron (0.24 mg/L soluble iron). They felt that the mechanism of toxicity to the fish was through respiratory disruption due to physical clogging of the gills. Wepener *et al*. (2001) reported that iron can damage gill surfaces and effect gill cytosol. Iron has also been shown to cause cell membrane damage (Dandapat *et al*. 1999), and damage DNA of fish (Payne *et al*. 1998).

The precipitation of ferric hydroxide can also affect fish according to their life stage. For example, Smith *et al.* (1973) found that low iron concentrations (about 1.5 mg/L iron) reduced the hatchability of fathead minnows (*Pimephales promelas*) more so than higher concentrations, and suggest that this may be due to the difference in particle sizes of ferric hydroxide between the various concentrations of total iron. They found a decreasing geometric mean of particle size with lower iron concentration, and suggest that the smaller particles are more likely to clog the pores of the fathead minnow egg chorion, resulting in reduced dissolved oxygen diffusion and therefore increased mortality. In contrast, higher concentrations of iron (up to 52.9 mg/L) can reduce visibility in the water and cause impaired food perception to fry and juvenile stages, resulting in prolonged stress and reduced growth (Smith *et al.* 1973). Finally, the precipitation of ferric hydroxide onto stream or lake bottoms can reduce light penetration and decrease productivity (thus decreasing food sources for the fish), as well as cover potential spawning grounds (Sykora *et al.* 1972b).

2.1.2 Ameliorating Effects

Peuranen *et al.* (1994) found that the toxicity of iron to brown trout was reduced by presence of humic acids. They found that organic material in the water (a concentration of 15 mg/L humic acids) had an ameliorating effect on ion regulation and facilitated oxygen uptake by preventing most of the accumulation of iron on the gills.

2.1.3 Acclimation

Brenner *et al.* (1976) found that common shiners (*Notropus cornutus*) were able to adapt to ferric hydroxide concentrations of 3.0 mg/L (1.54 mg/L iron) after four to six weeks exposure. Blood sugar decreased following a two-week exposure to ferric hydroxide, and remained low for the next two weeks. The concentration of blood sugar subsequently increased and stabilized at a level significantly less than in control fish. However, this did not affect their activity levels compared with fish maintained in conditions with no iron. Blood serum potassium decreased and sodium ions increased after a two-week exposure. After a four-week exposure, potassium then increased and remained significantly higher than that of controls for the remainder of the study (eight weeks), while sodium concentrations decreased to approximately the same level as control fish.

Grobler-van Heerden *et al.* (1991) found that approximately the same level of bioconcentration of iron occurred in the blood of *Tilapia sparrmanii* exposed to concentrations of total iron ranging from 1.8 mg/L to 18.6 mg/L for 72 hours. The bioconcentration decreased considerably (to below that of fish maintained in water with no iron) after a period of four weeks, which indicates that these fish can acclimatize to iron and regulate toxic concentrations.

2.1.4 Life Stage

The life stage of fish exposed to iron is very important in terms of long-term impact. In a number of studies, different life stages of three species of fish (fathead minnow, coho salmon and brook trout) were examined for sensitivity to lime-neutralized iron hydroxide. The safe upper limit of lime-neutralized iron in suspension for survival, growth, and reproduction of the fathead minnow was between 0.29 and 1.87 mg/L iron and the initial deleterious effect occurred during the egg incubation stage (Smith *et al.* 1973). For coho, the safe upper limit lay between 0.97 and 1.27 mg/L (lime-neutralized suspended) iron, with initial deleterious effect occurring during the early alevin development stage (Smith and Sykora 1976). Finally, the safe upper limit for brook trout was between 7.5 and 12.5

mg/L (lime-neutralized suspended) iron, with deleterious effects occurring during the juvenile development stage (Sykora *et al.* 1972a, 1972b, 1975 cited in Smith and Sykora 1976). Highly sensitive fish appear to be affected by lime-neutralized iron hydroxide suspensions earlier in their life history than species of lower sensitivity (Smith and Sykora 1976).

2.1.5 Toxicity to Fish

Smith and Sykora (1976) exposed brook trout and coho salmon at various concentrations (12, 6, 3, 1.5 and 0.75 mg/L) of lime-neutralized suspended iron. They found that none of the concentrations had an effect on the hatching success of brook trout or coho salmon. Also, brook trout alevin survival and size at 30, 60 and 90 days post-hatch did not change in response to an increase in lime-neutralized suspended iron concentration (Sykora *et al.* 1975). However, Sykora *et al.* (1972a) found that brook trout had retarded growth at 12 mg/L suspended iron and higher, and fish exposed to levels exceeding 6 mg/L were more susceptible to injury and disease. At 50 mg/L suspended iron, viability of brook trout eggs was greatly reduced (Sykora *et al.* 1972a). Coho salmon alevin survival at 30, 60, and 90 days post-hatch was similar in the 3, 1.5 and 0.75 mg/L suspended iron and control solutions, but declined sharply in 6 and 12 mg/L suspended iron water. They suggest that the safe upper limit for prolonged exposure of coho salmon egg and alevin stages may lie between 0.75 and 1.5 mg/L suspended iron.

Sykora *et al.* (1972b) suggested that ferric iron was not particularly dangerous to fish, although the soluble iron species may have been considerably more toxic. Sprague (1966, unpublished report cited in Sykora *et al.* 1972b) stated that concentrations of 1.0 to 3.5 mg/L Fe^{3+} caused little or no mortality to small salmon in a seven-day exposure. Later studies revealed a lethal threshold of 300 mg/L of a concentrate containing high quantities of suspended solids and a high percentage of precipitated iron. Mortality seemed more closely related to suspended solids concentrations that to total iron content.

Billard and Roubaud (1985) found that dissolved iron at a concentration of 0.005 mg/L had an unfavorable effect on rainbow trout (*Salmo gairdneri*) spermatozoa when they

were highly diluted with water (one part per hundred, 10^{-2}), and at 0.73 mg/L at a dilution of one part per ten (10^{-1}). Ova were similarly affected at concentrations of about 0.73 mg/L, and fertilization was affected at levels as low as 0.08 mg/L. The methods used in this study were experimental and do not appear to have been commonly adopted for measuring the toxicity of other compounds. For this reason, and because the value of 0.005 mg/L (5 μg/L) is much lower than other concentrations of iron found to be toxic in the literature, this value is considered an outlier and was not used in the derivation of the guideline. Sykora *et al.* (1975, cited in Billard and Roubaud 1985) found that a much higher chronic concentration of (lime-neutralized suspended) iron (12 mg/L) was necessary to affect the survival and growth of *Salmo fontinalis*.

In replicate tests, Alam and Maughan (1995) found that 96-hour LC_{50} values for carp (*Cyprinus carpio*) ranged from 0.56 to 1.36 mg/L for 3.5 cm fish and 1.22 to 2.25 mg/L for 6.0 cm fish. For this species, toxicity of iron decreased with the size of the fish. The pH for these tests was 7.1, and D.O. was 6.4 mg/L. No specific mention was made in the paper of whether the iron was measured as total or dissolved – but given the pH and dissolved oxygen, it would likely have been total iron.

Smith *et al.* (1973) found that there was a significant difference in egg hatchability, fry survival and juvenile growth of fathead minnows between a control concentration of 0.24 mg/L and a test concentration of 1.5 mg/L (lime-neutralized suspended) iron, suggesting that the critical concentration lies somewhere within this range. Sykora *et al.* (1972a) had similar findings, and also found that no fathead minnows survived beyond 22 days in concentrations of 12, 25, or 50 mg/L (lime-neutralized suspended) iron.

Wepener *et al.* (1992) exposed a minimum of ten banded tilapia (*Tilapia sparrmanii*) to 1.57 mg/L ferric chloride (0.54 mg/L iron) at pH 5 and also at pH 7.4. Significant mortalities occurred at both pH levels but they were three times higher at pH 5 (9 fish vs. 3 fish at pH 7.4). Significant increases in red blood cells and white blood cells occurred at pH 5. Decreases in hemoglobin and hematocrit occurred at both pH levels but were more extreme at pH 7.4, and mean corpuscular volume increased at pH 5.4 but decreased at pH 7.4. The increase in red blood cells was a reaction to hypoxic conditions that were

caused by an epithelial lifting of the gill lamellae (Wepener 1990 cited in Wepener *et al.* 1992). Mukhopadhyay and Konar (1984) performed LC_5, LC_{50}, and LC_{95} tests on *Tilapia mossambica* at pH 6.5, 7.0, and 8.5: the LC_5 was 110.8 mg/L total iron at pH 6.5, 73.6 mg/L total iron at pH 7.0 and 99.4 mg/L total iron at pH 8.5; the LC_{50} was 119.6 mg/L total iron at pH 6.5, 83.2 mg/L total iron at pH 7.0, and 118 mg/L total iron at pH 8.5; and the LC_{95} was 128.5 mg/L total iron at pH 6.5, 92.8 mg/L total iron at 7.0, and 136.8 mg/L total iron at pH 8.5. They also found that when mixtures of 1:1:1, iron:copper:zinc were tested, the toxicity of the mixture was many times greater than the toxicity of the single metal.

du Preez *et al.* (1993) found some evidence of bioaccumulation of iron in tissues of *Tilapia sparrmanii*. The order of bioconcentration changed slightly from liver > ovary > heart > muscle > testis > brain after acute exposure (72 hours) to liver > muscle > heart > ovary > brain > testis after chronic exposure (four weeks).

In-situ experiments were performed with brown trout in four high-altitude mountain streams in central Sweden during the melt of high-acidity snow (Andersson and Nyberg 1984). The pH in these streams decreased from between 6.1 and 7.2 to a minimum value of 4.5 in three of the streams. Mortality was high, especially during the beginning of the period when the pH was still above 5.5. The concentration of iron, manganese and aluminum were in the range of 0.55-1.2 mg/L (iron), 0.08-0.18 mg/L (manganese) and 0.09-0.16 mg/L (aluminum) during periods of trout mortality. The gills were the target organs for all metals, and iron accumulated ten times faster than the other two metals. It is expected that iron accumulation was probably the primary cause of mortality. In another stream, concentrations of 52, 3.9 and 0.45 mg/L of iron, manganese and aluminum respectively were not fatal, perhaps because they were present as non-toxic compounds. It appears likely that the change in pH and subsequent precipitation of the metals onto the gills of the fish was responsible for the high mortality.

Dalzell and MacFarlane (1999) used 96-h LC_{50} bioassays of brown trout (*Salmo trutta*) to examine the toxicity of commercial iron (III) sulfate liquor (presumably primarily iron sulfate but certainly with other metals contained as well) and the analytical grade of iron

sulfate. They found that the 96-h LC_{50} for the iron sulfate liquor was 28 mg/L total iron (0.05 mg soluble iron) and the 96-h LC_{50} for the analytical iron sulfate was 47 mg/L total iron and 0.24 mg/L soluble iron). The mechanism of toxicity to the fish was believed to be through respiratory disruption due to physical clogging of the gills. Inferred from this work is that the fish bioassay response is to the particulate iron on the gills (28 or 47 mg/L) rather than the dissolved iron threshold concentration reported (0.05 or 0.24 mg/L) but it is difficult to confirm from the paper.

Field observations by Myllynen *et al*. (1997) of lampreys in a Finnish river system with high total iron (1.5 - 3 mg/L) suggested that reproductive failure in populations were due to the high iron concentrations. The observations of Payne *et al*. (1998) of genetic damage to fish in a lake in Labrador that received iron mine waste at generally low concentrations may be indicative that the effects of iron on fish, previously believed to be benign, are not completely understood.

2.2 Invertebrates

At a concentration of 7.48 mg/L total iron, *Daphnia magna* experienced a 77% decrease in weight over 3 weeks, a 48% increase in protein, and a 13% decrease in L-aspartate: 2-oxo-glutarate aminotransferase activity (a metabolic enzyme). The 3-week chronic LC_{50} value (5.9 mg/L iron; Table 2) reported by Biesinger and Christensen (1972) is similar to the 64-hour apparent threshold concentration (< 6.2 mg/L iron) reported by Anderson (1950, cited in Biesinger and Christensen 1972). However, Biesinger and Christensen (1972) reported that the apparent threshold level for iron should be closer to their 16% reproductive impairment concentration of 4.38 mg/L total iron. It was also noted that adding organic matter as food for the Daphnids decreased the toxicity of many metals; therefore the values generated in these tests are not necessarily universal across water bodies, since organic matter obviously ameliorated metal toxicity.

Table 2: Chronic, acute and reproductive impairment (decrease in the number of young born) concentrations to *Daphnia magna* in Lake Superior water at pH approx. 7.5 for Fe^{3+} (from Biesinger and Christensen 1972).

Metal ion	LC_{50} (48 hr = acute)	LC_{50} (3 week = chronic)	Reproductive impairment=50%	Reproductive impairment=16%
Fe (III)	9.6 mg/L food	5.9 mg/L	5.2 mg/L	4.38 mg/L

Martin and Holdich (1986) found that the higher oxidation state for iron (Fe^{3+}) is less toxic than the lower oxidation state (Fe^{2+}), probably due to the lower stability of the Fe^{2+}. Toxicity appeared to be enhanced (or occurs at lower concentration) when the metal ions were capable of oxidizing or reducing molecules or ions in an organism (see Table 3). Table 3 summarizes the results of toxicity tests conducted by Martin and Holdich (1986) on *Asellus aquaticus* and *Crangonyx pseudogracilis*.

Table 3: Toxicity of Fe^{2+} (as $FeSO_4.7H_2O$) and Fe^{3+} (as $FeCl_3.6H_2O$) salts to *Asellus aquaticus* (an isopod) and *Crangonyx pseudogracilis* (an amphipod found in both North America and Europe)

Organism	pH	Hardness	48 hr LC_{50}	96 hr LC_{50}	Author
Asellus aquaticus	6.75	50	183 mg/L Fe^{3+}	124 mg/L Fe^{3+}	Martin and Holdich 1986
Asellus aquaticus			81.1 mg/L Fe^{3+}		Furmanska cited in Martin and Holdich 1986
Crangonyx pseudogracilis	6.75	50	160 mg/L Fe^{3+}	120 mg/L Fe^{3+}	Martin and Holdich 1986
Crangonyx pseudogracilis	6.75	50	143 mg/L Fe^{2+}	95 mg/L Fe^{2+}	Martin and Holdich 1986

Warnick and Bell (1969) conducted a study in which *Acroneuria lycorias* (a stonefly), *Ephemerella subvaria* (a mayfly), and *Hydropsyche betteni* (a caddisfly), were subjected to filtered water from Lake Superior. The water quality characteristics were: temperature 18.5°C; dissolved oxygen 8.0 mg/L; pH ~ 7.25; alkalinity ~ 50.0 mg/L; acidity ~ 6.0 - 12.0 mg/L; and hardness 53.0 mg/L. The 9-day LC_{50} for *Acroneuria lycorias* was 16.0 mg/L Fe^{2+} (from $FeSO_4$). Iron was the most toxic to *Ephemerella subvaria* with a 96-hour TL_m (median tolerance limit, equivalent to LC_{50}) of 0.32 mg/L Fe^{2+}. The 7-day LC_{50} for *Hydropsyche betteni* was 16.0 mg/L.

Maltby *et al.* (1987) conducted a study to determine the LC_{50} values for *Asellus aquaticus* exposed to Fe^{2+} in a 198 hour acute test; the organisms were collected from two different sites (one polluted and one non-polluted) at both pH 4.5 and pH 6.0. They used two methods to calculate the LC_{50} values: the first involved using the regression of the log (number surviving) against the concentration of Fe^{2+}, and the second used a probit analysis, i.e. probit (proportion responding) against log Fe^{2+} concentrations. Results were as follows:

Method used	Water Source	LC_{50} at pH 4.5 - mg/L Fe^{2+} -	LC_{50} at pH 6.0 - mg/L Fe^{2+} -
Log Regression	Polluted	428.5	466.7
	Un-polluted	299.8	419.2
Probit Analysis	Polluted	383.2	466.9
	Un-polluted	255.9	430.5

These results show that the low pH (4.5) solution was much more toxic than the pH 6.0 solution. Maltby *et al.* (1987) suggest that this could be due to either a synergistic effect between iron and hydrogen ions at the low pH, or a reaction with the buffer (0.0025M sodium potassium tartrate) which caused the iron to become less toxic (possibly by complexing with it) at the higher pH. Also, since organisms from the polluted site were more tolerant of iron at both pHs, there appears to be a resistance to iron due to genetic variation attributable to differential selection pressures, and/or to phenotypical differences due to differences in acclimation.

Sykora *et al* (1972a) found that the highest concentration of iron (lime-neutralized suspended) tolerated by *Gammarus minus* (freshwater shrimp) was less than 3 mg/L. At concentrations above 3 mg/L iron, reproduction was impaired and there was increased mortality in both adults and juveniles with increasing iron concentration. They also found that caddis fly larvae, *Cheumatopsyche*, were prevented from emerging into adults with iron concentrations as low as 3 mg/L.

Rousch *et al*. (1997) studied the effects of iron on the water mite *Arrenurus manubriator*, and the insect *Chironomus riparius*. Survival of midge larvae, mite deutonymphs and male adult mites was reduced at 400, 200, and 1,000 mg/L total iron (dissolved iron concentrations was within 5% of the total iron in theses tests) respectively. Female adult mites and larvae were unaffected by iron.

Gerhardt (1992) experimented with the mayfly *Leptophlebia marginata* to determine the toxicity of Fe^{3+} and Fe^{2+} at various concentrations (0, 10, 20 and 50 mg/L iron) at pH 4.5 and 7. Survival was greater than 90% in all treatments after 30 days, except the 50 mg/L iron - pH 4.5 treatment where mortality increased to 20% near the end of the experiment. Ferrous iron (Fe^{2+}) appeared to be the more toxic iron species. The cause of death in the pH 4.5/50 mg/L iron treatments appeared to be constipation. The larvae could not pass their food and so stopped feeding, and 20% starved after two weeks. At 10 and 20 mg/L iron and pH 4.5, feeding activity and motility decreased temporarily, but resumed near the end of the experiment. In a follow-up paper Gerhardt (1994) reported 96 hour LC_{50} for iron were 106.3 mg/L at pH7 and 89.5 mg/L at pH 4.5. In 96 hour EC_{50} bioassays, he found 70 mg/L at pH 7 and 63.9 mg/L at pH 4.5. A change he found when exposed to metals was the loss of escape behavior.

Milam and Farris (1998) examined the response of the freshwater mussel *Corbicula fluminea* and found enzyme activity was effected at concentrations as low as 0.19 mg/L measured as dissolved Fe^{2+}. In conjunction with other bioassays using the crustacean zooplankon *Daphnia, Ceriodaphnia* and the fish *Pimephales promelas*, they recommended a guideline of 0.37 mg/L dissolved iron.

Mukhopadhyay and Konar (1984) tested toxicity of iron and mixed metals to the plankton *Cyclops viridis* and the worm *Branchiura soerbyi.* They found that a mixture of iron, copper and zinc in a 1:1:1 ratio was far more toxic than when the metals were tested individually (Table 4).

Dandapat *et al*. (1999) found that $FeCl_3$ at 100uM (5.6 mg/L Fe^{3+}) resulted in lipid damage in gills and hepatopancreas of the prawn *Macrobrachium rosenbergii*.

Table 4: Toxicity of iron (mg/L) to *Cyclops viridis* and *Branchiura soerbyi* at various pHs. (from Mukhopadhyay and Konar 1984).

Metal	Species	pH	LC_5 mg/L	LC_{50} mg/L	LC_{95} mg/L
Iron (only)	*Cyclops viridis*	6.5	11.8	35.2	54.0
Iron (only)	*Cyclops viridis*	7.0	15.6	33.2	50.6
Iron (only)	*Cyclops viridis*	8.5	17.4	36.0	63.4
Iron (only)	*Branchiura soerbyi*	6.5	250	580	920
Iron (only)	*Branchiura soerbyi*	7.0	290	560	830
Iron (only)	*Branchiura soerbyi*	8.5	55	446	835
Fe, Cu, Zn mix	*Cyclops viridis*	6.5	0.546	3.186	6.843
Fe, Cu, Zn mix	*Cyclops viridis*	7.0	2.276	2.959	8.138
Fe, Cu, Zn mix	*Cyclops viridis*	8.5	0.086	0.173	0.428
Fe, Cu, Zn mix	*Branchiura soerbyi*	6.5	0.859	1.957	3.096
Fe, Cu, Zn mix	*Branchiura soerbyi*	7.0	0.136	0.394	0.660
Fe, Cu, Zn mix	*Branchiura soerbyi*	8.5	0.068	0.530	0.894

Randall *et al*. (1999) reported an LC_{50} of 11.5 mg/L Fe^{3+} for *Daphnia* from a reservoir dosed with iron. The mechanism of toxicity was the iron hydroxide floc interfering with the filtering mechanism of the organism. They suggested a safe concentration of 1.7 mg/L Fe^{3+} on the basis of their work.

In addition to the direct toxic effects of iron to invertebrates, iron-hydroxide precipitates can also cause drastic changes in habitat, thereby displacing some species and favoring others (Vuori 1995). Boult *et al.* (1994, cited in Vuori 1995) describe a situation in which a tributary entered an acid river with extremely high iron concentrations (up to 260 mg/L). Downstream from the point of entry, the neutralizing effect of the tributary resulted in the precipitation of ferrihydrite and the co-precipitation of other metals, producing an orange blanket over the streambed. In a similar situation, an iron concentration of 0.66 mg/L, along with high aluminum concentration (2.36 mg/L), in low

pH water (3.75) was sufficient to cause a thick flocculent, brownish-white precipitate of hydrous aluminum and iron oxides co-precipitated with aquatic humic substances in a small Rocky Mountain stream (McKnight and Feder 1984). As in the situation described by Boult *et al.* above, this precipitation also occurred downstream from the confluence with a tributary containing higher-pH (7.3), lower iron (0.06 mg/L), and lower aluminum (0.04 mg/L) water.

2.3 Amphibians

Porter and Hakanson (cited in Freda 1991) found that the lethal limit of iron for *Bufo boreas* tadpoles was between 20 and 30 mg/L.

2.4 Algae

At 6 mg/L of iron ($FeCl_3$), the growth of *Chlorella vulgaris* was inhibited (Den Dooren de Jong 1965).

2.5 Plants

A number of studies have reported iron toxicity as a problem in aquatic plants, particularly rice (Howeler 1973, Snowden and Wheeler 1993). Batty and Younger (2003) examined the effects of total iron (added as $FeSO_4.7H_2O$) on a species of aquatic reed (*Phragmites australis*) and they found an inhibition of growth above a concentration of 1 mg/L total iron. However they felt that the iron might not directly explain the reduction in growth. Wang (1986) reported that the EC_{50} (Effective Concentration) for duckweed *Lemna minor* was 3.7 mg/L and suggested a maximum permissible concentration of 0.37 mg/L.

Sinha *et al*. (1999) found that relatively low concentrations of iron (0.5 to 5.0 mg/L) resulted in damage to the membranes by lipid oxidation in the aquatic plant *Hydrilla*. They also noted effects on other physiological indicators.

2.6 Drinking Water

Cohen *et al.* (1960) reported that the threshold level for 50% of the human population to taste ferrous sulfate was 3.4 mg/L when the iron was added to distilled water. In spring water, the 50% taste threshold was only 1.8 mg/L. Hydrous ferric oxide could be present in higher concentrations before it was tasted: in distilled water, the threshold concentration for 50% of the subjects was 8.8 mg/L, and in spring water it was only 1.8 mg/L.

Riddick *et al.* (1958) reported that groundwater containing levels as high as 30-50 mg/L of iron appeared clear when first sampled. However, when the water stood for a few hours, the uptake of oxygen and the release of carbon dioxide resulted in a precipitation of iron as it was oxidized from ferrous to ferric hydrate. Thus, a fresh water sample may have an acceptable level of turbidity, only to become highly unacceptable within a few hours.

3.0 B.C. MINISTRY OF ENVIRONMENT TOXICITY TESTING

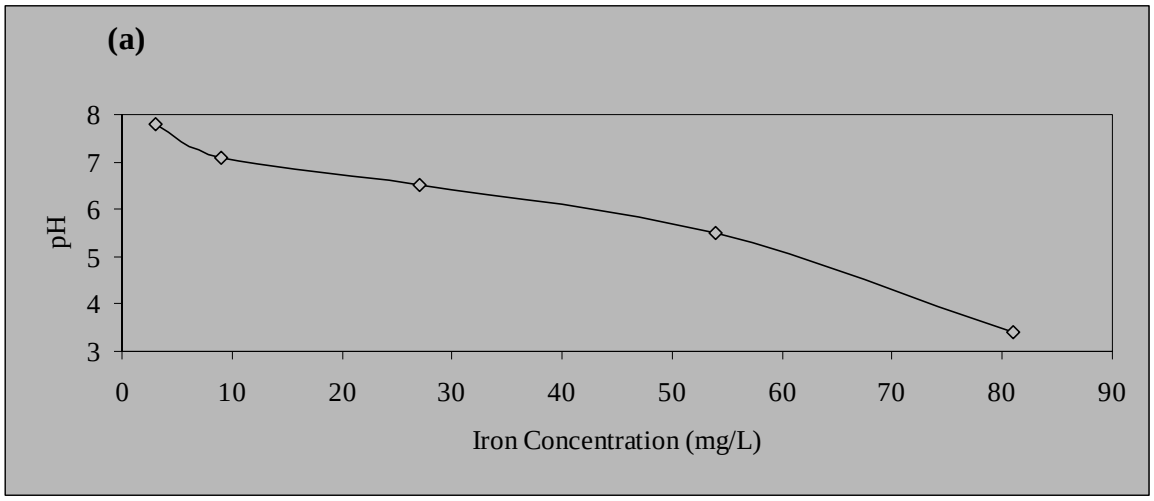
In 1997 the B.C. Ministry of Environment undertook a study to determine the toxic effects of iron on various aquatic species. Species used in the freshwater tests were: rainbow trout, the amphipod *Hyalella azteca*, the chironomid *Chironomus tentans*, the crustacean zooplankter *Daphnia magna*, and the green alga *Selanastrum capricornutum*. For tests conducted in salt waters, the following species were used: Chinook salmon, the amphipod *Euhaustorius washingtonianus*, sand dollars (*Dendraster excentricus*) and topsmelts (*Atherinops affinis*). A micro-toxicology test involving bacteria was conducted in both salt and fresh waters. Finally, an early-life-stage salmonid test (e-test) was conducted to determine the effects of iron on the development of newly fertilized rainbow trout eggs.

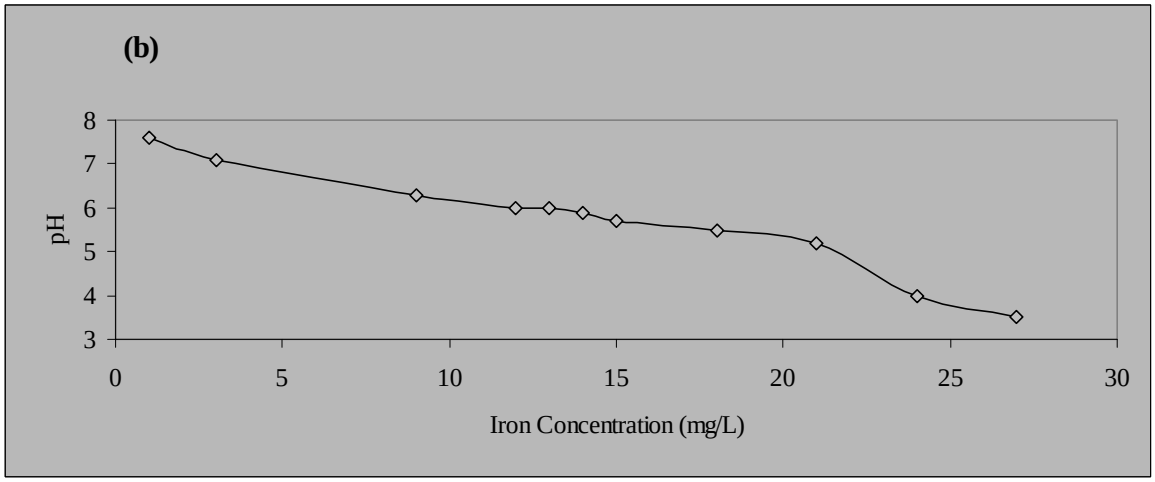
The LC_{50} for each species was determined in three different waters with varying degrees of hardness. The three waters were termed soft (50 mg/L hardness), medium (well water - 100 mg/L hardness) and hard water (250 mg/L hardness). Exceptions to this were the *Selenastrum* and the chronic *Daphnia*, which were tested in well water only. The salt water was collected in Burrard Inlet (Vancouver, B.C.), with a typical salinity of 26-28 parts per thousand. Dissolved oxygen, pH and temperature were measured at the beginning and end of each experiment. The purpose of these studies was to augment existing toxicological data, and provide a consistent basis for comparison of methodologies and results for all of the water quality guidelines developed by the Ministry of Environment.

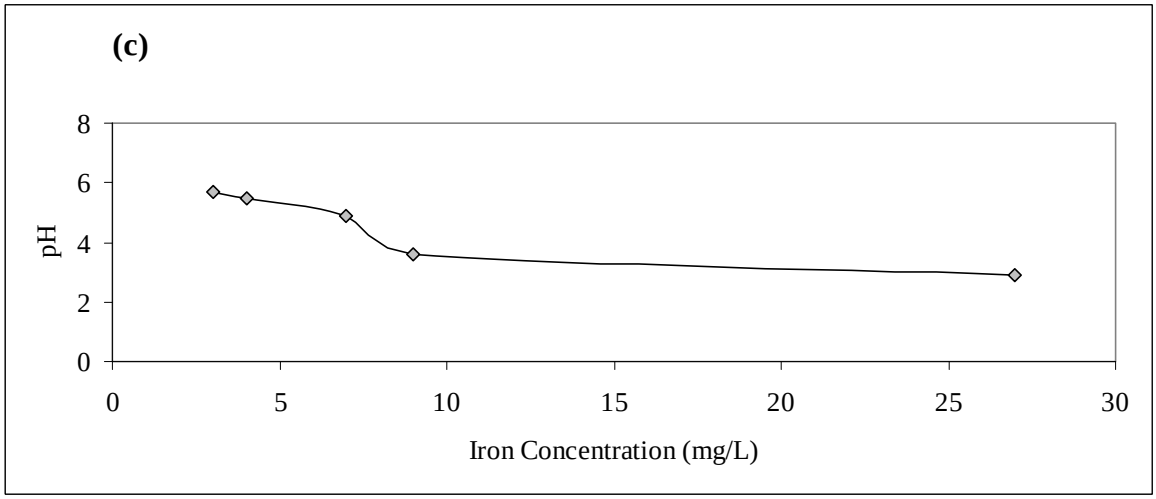
3.1 Effect of Hardness on pH and Iron Toxicity

A common difficulty arose while conducting the toxicity tests on all organisms. The addition of ferric chloride to test water to increase the iron concentration also resulted in a dramatic decrease in pH (see Figure 1; Section 2.1.1). This decrease in pH varied with the buffering capacity of the dilution water. Hard water was able to dissolve a greater concentration of ferric chloride before the pH depression occurred. In addition, the

Figure 1. The relationship between iron concentration and water pH with the addition of ferric chloride to water with varying degrees of hardness (from Pacific Environmental Science Center, unpublished data). (a) Hard water, (b) well water and (c) soft water







solubility of iron changed with the hardness of the water, with a higher degree of solubility occurring in harder water. In order to discern between the toxicity of iron and toxic effects of low pH, a minimum pH threshold of 5 was arbitrarily assigned to the tests. This factor resulted in a limitation on the amount of ferric chloride that could be added to each solution. In hard water, the maximum iron concentration that could be achieved with a pH of at least 5 was 54 mg/L. This value decreased to 36 mg/L in saltwater, to 21 mg/L in well water, and to 7 mg/L in soft water. However, even at these levels, iron precipitate was observed on the bottom of the test vessels over the course of the bioassays. This precipitation was probably due to O_2 uptake and CO_2 release resulting in the oxidation of iron from its ferrous to ferric forms.

The precipitate formed large enough particles that it was relatively easily avoided by motile organisms such as the fish and *Daphnia*. While iron precipitate was observed on the gills of fish, it did not appear to hamper either respiration or mobility (PESC, unpublished notes). It is possible that a direct examination of the gill tissue would reveal structural damage that did not visibly affect the activity of the fish (see Section 2.1.1).

One potential problem introduced by the effect of iron on pH is that the controls utilized for the test were conducted at a near-neutral pH, while the pH for the various other concentrations decreased directly with the concentration of iron used. This in turn would not allow the potentially toxic effects of pH alone to be separated from those of iron and pH. Therefore, it is recommended that for future studies the pH of all solutions (including the control) be decreased such that they are equal to that of the solution with the highest concentration of iron to be tested.

3.2 Materials and Methods

Static tests were used for all aquatic organisms except the chronic *Daphnia*, which had replacement of the sample water on Mondays, Wednesdays and Fridays, and the e-test, which had daily replacement of the solution. The number of replicates used for each concentration varied with the species as follows: one for the micro-toxicology; two for the marine amphipod and acute *Daphnia*; three for the Rainbow trout, coho, *Hyalella*,

chironomid, e-test, and sand dollars; five for topsmelts and *Selenastrum*; and ten for the chronic *Daphnia*.

Ferric chloride was added in the appropriate concentrations to each of the four types of water, and a control was established with no ferric chloride. Iron concentrations were measured in the solutions at the initiation and completion of the exposures. All tests were conducted to determine 96-hour LC_{50} values except for the *Daphnia* which were tested for both a 48-hour acute LC_{50} and a 21-day chronic no-observed-effects-level (NOEL) and lowest-observed-effects-level (LOEL). As well, the topsmelts and the e-test were tested for a 7-day LC_{50}. Both salmonid species were tested in 15°C water, the chironomids and *Hyalella* were maintained at 23°C, and the daphnids were maintained at 20°C.

Silica sand was used as the substrate for the chironomids, and cotton gauze was used as the substrate for the *Hyalella*. Though the tests were static, measurement of the various water characteristics before and after testing (including iron concentration) allows this data to be considered primary pursuant to the criteria outlined by CCME (1991).

3.3 Results

For the majority of the organisms tested, the maximum levels of iron possible in each water type were below lethal limits (Table 5). In the soft water, the sole exception to this was for *Hyalella*, with a 96-hour LC_{50} of 3.5 mg/L iron. In the well water, organisms for which an iron LC_{50} could be determined were rainbow trout with a 96-hour LC_{50} of 15.2 mg/L, *Selenastrum* with a 96-hour LC_{50} of 3.6 mg/L, the 21-day chronic *Daphnia* with a NOEC of 5.3 mg/L and an LOEC of 10.7 mg/L, and the 5-minute micro-toxicology IC_{50} of 14.5 mg/L. In hard water, the 5-minute micro-toxicology IC_{50} was 41.8 mg/L and the 15-minute IC_{50} was 49.1 mg/L. Finally, in salt water, a 96-hour LC_{50} of 37.7 mg/L was determined for Chinook, the sand dollars had an EC_{50} of 8.1 mg/L, and a 96-hour LC_{50} of 31.3 mg/L was determined for the amphipods.

Table 5. A summary of iron toxicity tests conducted by PESC for the B.C. Ministry of Environment in 1997 and 1998.

	LC_{50}			
Test Organism	**Soft Water**	**Well Water**	**Hard Water**	**Salt Water**
Rainbow Trout	> 6.4 mg/L	15.2 mg/L	> 53.6 mg/L	
Hyalella	3.5 mg/L	> 19.8 mg/L	> 52.4 mg/L	
Chironomid	> 5.2 mg/L	> 19.1 mg/L	> 49.8 mg/L	
Daphnia	> 6.2 mg/L	> 21.5 mg/L	> 50.1 mg/L	
Chronic *Daphnia* NOEC		5.3 mg/L		
Chronic *Daphnia* LOEC		10.7 mg/L		
Selenastrum		3.6 mg/L		
Microtox 5-minute IC_{50}	> 7 mg/L	14.5 mg/L	41.8 mg/L	
Microtox 15-minute IC_{50}	> 7 mg/L	> 21 mg/L	49.1 mg/L	
Chinook				37.7 mg/L
Amphipods				31.3 mg/L
Topsmelts				>34 mg/L
Sand Dollars				8.1 mg/L
Microtox 5-minute IC_{50}				> 36 mg/L
Microtox 15-minute IC_{50}				> 36 mg/L
E-test	> 6.7	> 19.6	> 50.7	

4.0 EXISTING GUIDELINES

Loeffelman *et al.* (1985) suggested that total iron concentrations of over 1.0 mg/L were required to adversely affect *Pimephales promelas* and *Salmo gairdneri*, while much lower concentrations of bathophenathroline (BPA) reactive ferrous iron (Fe^{2+}) could have similar effects. They suggested that regulating only this ferrous iron form should be considered for protecting aquatic life. They recommended a guideline of 0.37 mg/L for BPA-reactive ferrous iron, with the justification that it was: 1) 70% lower than the lowest measured effect concentration of 1.2 mg/L on rainbow trout post-hatch larvae; 2) derived from an LC_{50} lower than that for rainbow trout, which is often a sensitive species; and 3) similar in magnitude to measured levels of BPA-reactive ferrous iron (0.01 to 0.246 mg/L) believed to have no significant adverse effect on fish populations of the Ohio River.

The existing U.S. Environmental Protection Agency guidelines for iron are 0.3 mg/L for domestic water supplies and 1.0 mg/L for freshwater aquatic life. The drinking water guideline is for aesthetic rather than toxicological significance, since this amount constitutes only a small fraction of the iron normally consumed. Diets generally contain 7 to 35 mg of iron per day, with an average level of 16 mg iron. They suggest that the guideline of 1 mg/L iron for freshwater aquatic life is adequately protective, and this is the same value recommended by the European Fisheries Advisory Commission (1964, cited in the USEPA (1976) document).

The Ministry of Environment in Saskatchewan currently has a water quality guideline of 1.0 mg/L total iron for freshwater aquatic life, while the Ministries of Environment in both Ontario and Alberta have adopted a guideline of 0.3 mg/L for the protection of aquatic life (Appendix IV). The Quebec government (Guay *et al*. 2002) reviewed the literature related to iron and protection of aquatic life and recommended concentrations of 6.9 mg/l for effluent, 1 mg/L for acute toxicity and less than 0.3 mg/L to protect against chronic effects. No indication was made if these guidelines were for total or dissolved but presumably the former.

A recommendation was made for the state of Iowa for a surface water criterion of 1 mg/L total iron (Iowa 2005) and many other US states have adopted the USEPA national guideline.

5.0 RECOMMENDED GUIDELINES

In order to establish a safe upper limit for iron concentrations to protect freshwater aquatic life, two crucial factors that contribute to iron toxicity must be considered. These factors are 1) the direct toxic effects of iron, generally in its dissolved ferrous (Fe^{2+}) state, on aquatic organisms, and 2) the indirect effects on habitat and species displacement by iron precipitates, generally related to total iron concentrations of the ferric (Fe^{3+}) forms which tend to precipitate out of solution at all but the lowest pH levels or in anoxic conditions. Therefore, a separate guideline for dissolved and total iron is recommended.

The review of the scientific data around the topic of iron toxicity shows that there are a number of problems that exist. The original criterion of 1 mg/L to protect aquatic life was published by EIFAC (1964); the same criterion was, with some additional rationale, used again by the National Academy of Sciences (1974) and then again in the EPA Redbook in 1976 but has not been revised since then. Thurston *et al*. (1979) were critical of many of the USEPA criteria. The acceptance of what seems to have been poor rationale and not very strong science is a major weakness in this particular criterion. Once established, the 1 mg/L criterion seems to have been accepted over a long period without appropriate critical examination.

The initial criterion was 'based on field observations principally" (USEPA 1976) with some early bioassay data being discounted. For instance, Warnick and Bell (1969), albeit with a static bioassay, obtained 96-hour LC_{50} values of 0.32 mg/L. Much of the effort in assessing iron toxicity since then has used laboratory bioassays. Due to the complexity of iron chemistry including pH shifts, transformations from Fe^{2+} to Fe^{3+} and changes from dissolved to particulate phase, it has been difficult to assess much of this laboratory work. As noted in the PESC bioassay work commissioned by the B.C. Ministry of Environment reported here, there were problems with precipitation and pH shift. This is likely a

problem with most of the bioassay work that has been done previously and has to be taken into account in interpreting all previously published work. The CCME protocols puts primary importance on laboratory bioassays as the acceptable form of data for calculating new guidelines and discounts the value of field data. However, in the case of iron, it would seem useful to consider field observations in arriving at an appropriate guideline. Given the present emphasis on bioassay data, it would seem appropriate to at least consider more of the field data to establish guidelines for problematic substances like iron. This is difficult, however, since the protocols for the derivation of guidelines have been established to consider and evaluate bioassay data, but not the field data.

Augusto *et al*. (2005) have proposed a field-based process using biotic response to evaluate and improve chemical criteria. Using a quantile regression of contaminant concentrations against numbers of sensitive species, criteria values were derived for copper and zinc for Ohio fresh waters. One of their findings in using this approach was that zinc criteria derived in this way seemed to indicate a lower acceptable concentration for zinc than had been derived from the standard bioassay approach. The investigators also suggested that such a field-based effect concentrations (FECs) process will be useful to develop water quality criteria for chemicals whose toxic effects are not feasible to test in laboratory assays. Iron was quoted to be an ideal example, since most of its negative effects results from the deposition of iron colloids on organisms and substrates, disturbing normal metabolism and osmoregulation, and changing the structure and quality of benthic habitats and food resources.

In subsequent work by the same group (Linton *et al*. 2007 in press) iron criteria were also proposed based on this method. They used the most sensitive species in the Ohio area (the mayfly family Leptophlebiidae) as an indicator and statistical analysis of concentration and population response to propose a total iron criterion of 0.21 mg/L as being required to protect the biological integrity of the aquatic ecosystem (as defined under the US Clean Water Act). They also proposed a second criterion to protect uses under the Clean Water Act section 101(a) of 1.74 mg/L. This concentration allowed more of a biological change than the 0.21 mg/L criterion which was established at the threshold of noticeable community structure change. This approach should be examined as an

option to be used more widely to establish water quality guidelines with problematical characteristics like iron.

5.1 Dissolved Iron: Freshwater

The lack of data characterizing the concentration of dissolved iron in the environment makes it difficult to link environmental consequences with a specific dissolved iron concentration. The lack of clear data from bioassay work reported in the literature, specifically differentiating the effects of dissolved and total iron make the proposal of a guideline for dissolved iron difficult. However it is felt that a guideline using dissolved iron is useful and that by establishing a guideline for dissolved iron would require and encourage the sampling and analysis of dissolved iron in the aquatic environment.

There seems to be some other suggestions as to the value of a guideline using dissolved iron. From the initial suggestion by Loeffelman *et al*. 1985, there have been a number of investigators who have suggested that the dissolved fraction should be the basis of a guideline. The Ohio EPA (1998) have indicated that along with their approach using aquatic biological communities, additional effort should be made to collect dissolved iron data in order to establish a future criterion.

The guideline proposed for the maximum concentration of dissolved iron (being able to pass through 0.45µ filter) (most dissolved iron is usually is in the ferrous state) in freshwater is 0.35 mg/L, to protect aquatic life from the toxic effects of iron. This value is slightly more conservative than the 0.37 mg/L guideline for BPA-reactive ferrous iron suggested by Loeffelman *et al.* (1985; see Section 3.0), and is considerably lower than all but a few of the acute LC_{50} values reported in Appendix I. Those LC_{50} values listed in Appendix I near or below 0.35 mg/L are from earlier, more unreliable studies that did not report pH, and did not specify whether the iron concentrations presented were dissolved or total values. As such, the data generated by these studies is considered to be secondary under the CCME protocol for scoring toxicity-related references.

Unfortunately, the majority of the results summarized in Appendix I is also considered to be secondary data. The reason for this, in the majority of the cases, was because studies

were conducted in static conditions without replacement of the iron solution and/or did not measure iron concentrations at the end of the test. Therefore, the actual concentrations of iron in solution are uncertain because iron can be lost from the water due to precipitation. Another possible reason that studies might be considered secondary is because toxicity values were based on nominal rather than measured concentrations. In almost all of the literature examined it was difficult to confirm basic information about the details of the bioassays so little of these data could be used with confidence.

The guideline recommended in this document is based primarily on the lowest 96-hour LC_{50} value reported in the testing conducted by the B.C. Ministry of Environment (Table 5). This value (3.5 mg/L for *Hyalella* in soft water, supported by the LC_{50} value of 3.6 mg/L for *Selenastrum, deemed to be a chronic test*) was divided by a safety factor of 10 to arrive at the recommended guideline. However, the group of organisms that have been reported as being most sensitive to iron in other studies (stream immature insects, particularly mayflies) (Linton *et al*. 2007; Gerhardt 1992, 1994; Rasmussen and Lindegard 1988; Warnick and Bell 1969) were not included in this suite of bioassay organisms that were tested by B. C. Environment, thereby justifying the use of a safety factor of ten. The other factor in support of the safety factory stems from the B.C. Environment's bioassay work; it indicated that there was some precipitation of the added iron in the test water as a very fine floc that settled in the bottom of the bioassay tanks. The implication of this is that the effective iron concentration (the concentration responsible for the toxicity) is likely lower than was calculated.

There are a number of studies that also support a proposed guideline in the relative range suggested above. Milam and Farris (1998) suggested on the basis of their bioassays with clams, that a no effect level of 0.4 mg/L (as Fe^{2+}) would be appropriate. They cite a recommendation from AEPSC (1983) for a criterion of 0.37 mg/L. There seem to be a number of field studies that indicate that negative environmental impacts occur at lower concentrations than are shown by laboratory bioassays (Vouri 1995). Interaction of other factors and mixtures of contaminants is a possible reason for this difference. Warnick and Bell (1969) suggested a protective guideline of 0.32 mg/L. Wang (1986) suggested 0.37 mg/L was appropriate to protect aquatic plants. Warnick and Bell (1969) based on

their fish bioassays, suggested 0.32 mg/L. A study by Linton *et al*. (2007) suggests a water quality criterion of 0.21 mg/L is necessary to protect sensitive species (mayflies) in Ohio waters. Their criterion is expressed as total iron but uses the response of biological communities as an indicator and so may be more functionally equivalent to dissolved iron.

Due to the arbitrary nature of the term "dissolved" (usually designating any material that passes through a 0.45 μm membrane), this guideline designation becomes slightly more complicated since colloidal iron is capable of passing through such a membrane (McKnight and Feder 1984). However, this serves only to increase the sensitivity (and therefore decrease the risk) of the dissolved iron guideline.

Although hardness is a factor in the toxicity of many other metals, the factor of hardness is not presently incorporated into the derivation of the guideline for a number of reasons. Although a significant increase occurred between the Microtox IC_{50} values reported for the well water (14.5 mg/L at hardness 100 mg/L $CaCO_3$) and the hard water (41.8 mg/L at hardness 250 mg/L $CaCO_3$), there was no real change in the minimum 96-h LC_{50} values between the soft water (3.5 for *Hyalella* at hardness 25 mg/L $CaCO_3$) and the well water (3.6 for *Selenastrum* at hardness 100 mg/L $CaCO_3$). This would indicate that the ameliorating effect of hardness on iron toxicity is not linear, and at this stage we have insufficient data to determine the true relationship between iron toxicity and hardness.

This observation is supported by the findings of Gonzales *et al.* (1990), who found no consistent influence of increased calcium concentration on the LC_{50} of brook trout (see Section 2.1.1). Finally, the fact that the toxicity tests could not be conducted in hard water for either the *Selenastrum* or the chronic *Daphnia* has resulted in an incomplete data set, and makes a proper comparison between the varying degrees of water hardness impossible. If future studies are undertaken which clearly illustrate the relationship between hardness and iron toxicity, such a guideline may be adapted to reflect the relationship at that time.

The lack of reliable chronic toxicity data for either dissolved or total iron (see Appendix II) makes the establishment of a guideline for the 30-day mean concentration difficult. The low mean tolerance limit found by Warnick and Bell (1969) of 0.32 mg/L iron suggests that the guideline for the maximum concentration of dissolved iron (0.35 mg/L) would be protective from the chronic effects of iron toxicity. However, Warnick and Bell conducted static tests without adequate replacement of the stock solutions, and therefore the accuracy of their findings is somewhat questionable. As there are no data to indicate that a chronic, 30-day mean guideline is necessary, none is recommended at this time. However, if future studies indicate that concentrations of dissolved iron lower than 0.35 mg/L can have toxic effects on some species of aquatic organisms after a long-term exposure, a guideline for the 30-day mean concentration may be proposed at that time.

5.2 Total Iron: Freshwater

A number of studies described in this report address the potential toxicity of relatively high concentrations of suspended ferric iron (*e.g.*, Sykora *et al.* 1972a, 1972b, Smith and Sykora 1976, Brenner *et al.* 1976). As previously discussed, iron in near-neutral water, or in well-oxygenated water, will tend to convert from the soluble, ferrous state to the insoluble, ferric state. It is possible for this insoluble iron to remain suspended in solution, especially in moving waters such as rivers and streams. The potential toxic effects from this suspended iron generally occur either as:

- damage to the gills of fish from the corrosive effects of the ferric iron;
- from smothering of eggs or organisms which live in the sediment where the iron is deposited, or
- from decreased visibility in the water, which can affect feeding success and other behaviour.

Of these three possibilities, it appears that the egg and early alevin stages are most susceptible to the ferric form of iron. A safe upper limit of approximately 1.0 mg/L has been derived for a number of fish species (see Sections 2.1.4 and 2.1.5). However, there is also some evidence that in many circumstances 1 mg/L might be overprotective.

Determining a critical threshold to use as a guideline for total iron is difficult. A concentration of total iron might represent a range of the directly biologically important dissolved iron (generally in the Fe^{2+} form) from a negligible percentage to a high percentage as part of the measurement of total iron. Total iron concentration and the relative proportion of ferrous iron may have a biological consequence depending on pH temperature, dissolved oxygen concentration, humic acid concentration, chloride concentration or whether the sun is shining. These reasons are some of why a dissolved guideline was proposed.

There are multiple factors that influence the toxicity of total iron and as such, it would appear that in many circumstances, the use of the existing value of 1 mg/L may be overprotective. There are a variety of factors that have been discussed previously that need, to be not only taken into account, but the interactions also need to be understood that make proposing a guideline for total iron inadvisable. For example, the factor of hardness which is generally considered for evaluation of metal toxicity has not been felt to have a significant effect on suspended iron toxicity – but few investigations of this factor have been published. While humic acids have been shown to have an ameliorating effect on the toxicity of total iron (Peuranen *et al.* 1994), there are insufficient studies to determine the actual relationship between humic acids and iron toxicity.

In light of the contradictory data, the recommendation is to use what seems to be the most recent and best field-based research of Linton *et al*. (2007) as the basis of a guideline for total iron. Linton *et al*. used two benchmarks of change in community structure to establish guidelines. The first (0.21 mg/L) corresponds to no or minimal changes in aquatic community structure and function. A second benchmark that allowed for a slight to moderate changes in community population structure because of loss of some rare species and/or replacement of sensitive ubiquitous taxa with more tolerant taxa generated a guideline of 1.74 mg/L. In the context of environmental protection whether some change in sensitive species is acceptable is an open question. In the spirit of the precautionary principle, a lower concentration seems appropriate and the existing if poorly justifiable 1.0 mg/L seems to be an acceptable value to end up at. This is possible that, as discussed earlier, the 1 mg/L may be overly protective but with the present data, it

is difficult to rationalize. Other recent research, for example Randall *et al*. 1999 have coincidentally recommended 1.7 mg/L.

It is, therefore, recommended that the maximum total iron concentration should not exceed 1.0 mg/L to protect aquatic systems from detrimental effects of iron.

5.3 Application of Guidelines

With guidelines for both dissolved and total iron proposed, there needs to be some discussion of the how the two guidelines are to be applied. The guideline for dissolved iron is of primary importance and if monitoring for iron toxicity is appropriate, dissolved iron should be sampled and analyzed and should be the focus of any evaluation. If measurements for both dissolved and total are taken, the possibility exists that one guideline might be met and the other exceeded. In this case, if the dissolved is exceeded and the total is below the limit, there would be more reason for concern than if the opposite (total iron guideline exceeded and dissolved iron guideline met) were to occur. There is value in the two guidelines when sampling and when it is obvious that iron precipitation is occurring, the dissolved iron concentration should be of primary importance.
In certain circumstance, total iron concentration in water may exceed the recommended guideline of 1.0 mg/L due to natural cases (This may be true for total iron but not for dissolved iron.). This is often caused by high load of suspended material in water during high flow conditions and the association of total iron content with the suspended materials. In such cases, it is suggested that the back ground total iron concentration be used as a guideline. This is consistent with anti-degradation policy of the CCME where substance/contaminant concentration is naturally high and the background procedure employed by BCMOE for setting water quality objectives.

5.4 Marine Guideline

To date, there have been very few studies that have examined the toxicity of iron in the marine environment. As such, there are insufficient data to establish an appropriate guideline at the current time. Due to the relatively high mean pH of marine waters

(approximately pH 8.2), very little iron would remain in solution, and it is not anticipated that iron toxicity would therefore be a concern. If, in the future, studies are conducted which suggest that iron toxicity in the marine environment may be a concern, a guideline for marine iron levels may be considered at that time.

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APPENDIX I: SUMMARY OF ACUTE IRON LC_{50} VALUES (IN ASCENDING ORDER)

Species	Iron conc. (mg/L)	Form of iron used	Test Duration	Other variables	Reference *	CCME reference score
Various fish spp.	0.1	Ferric sulphate	24 hours		Clark & Adams 1913 cited in Doudoroff and Katz 1953	2°
Various fish spp.	0.2	$FeCl_3$		pH 7.2-7.4	Minkina 1946 cited in Doudoroff and Katz 1953	2°
Cyprinus carpio (3.5 cm long)	0.56-1.36	Not mentioned	96 hour LC_{50}	pH 7.1, D.O. 6.4	Alam and Maughan 1995	2°
Various fish spp.	0.9	Dissolved		pH 6.5-7.5, well aerated	Bandt 1938 cited in Doudoroff and Katz 1953	
Cyprinus carpio (6.0 cm long)	1.22-2.25	Not mentioned	96 hour LC_{50}	pH 7.1, D.O. 6.4	Alam and Maughan 1995	2°
Various fish spp.	1.28	Ferrous sulphate	24 hours		Clark & Adams 1913 cited in Doudoroff and Katz 1953	
Eels	4.9	$FeCl_3$	50 hours	T=20°C–22°C	Oshima 1931 cited in Doudoroff and Katz 1953	
Tilapia mossambica	6.5	Dissolved	96 hour LC_5	pH 6.5	Mukhopadhyay and Konar 1984	2°
Daphnia magna	9.6	Fe (III)	48 hr LC_{50}		Biesinger and Christensen 1972	2°
Cyclops viridis	11.8	Dissolved	96 hour LC_{50}	pH 6.5	Mukhopadhyay and Konar 1984	2°
Daphnia	11.5	Fe sulphate	96 hour LC_{50}		Randall at al 1999	2°
Eels	12.7	$FeCl_2$	50 hours	T=20°C–22°C	Oshima 1931 cited in Doudoroff and Katz 1953	
Eels	14.3	$KFe(SO_4)_2$	25 hours	T=20°C–22°C	Oshima 1931 cited in Doudoroff and Katz 1953	
Cyclops viridis	15.6	Dissolved	96 hour LC_5	pH 7.0	Mukhopadhyay and Konar 1984	2°
Cyclops viridis	17.4	Dissolved	96 hour LC_5	pH 8.5	Mukhopadhyay and Konar 1984	2°
Bufo boreas	20-30		Lethal limit		Porter and Hakanson 1976; cited in Freda 1991	
Salmo trutta	28	Total	96 hr LC_{50}		Dalzell and Macfarlane 1999	2°
Cyclops viridis	33.2	Dissolved	96 hr LC_{50}	pH 7.0	Mukhopadhyay and Konar 1984	2°
Cyclops viridis	35.2	Dissolved	96 hr LC_{50}	pH 6.5	Mukhopadhyay and Konar 1984	2°
Cyclops viridis	36.0	Dissolved	96 hr LC_{50}	pH 8.5	Mukhopadhyay and Konar 1984	2°
Cyclops viridis	50.6	Dissolved	96 hour LC_{95}	pH 7.0	Mukhopadhyay and Konar 1984	2°
Cyclops viridis	54.0	Dissolved	96 hour LC_{95}	pH 6.5	Mukhopadhyay and Konar 1984	2°
Branchiura	55	Dissolved	96 hour LC_5	pH 8.5	Mukhopadhyay and Konar 1984	2°
Cyclops viridis	63.4	Dissolved	96 hour LC_{95}	pH 8.5	Mukhopadhyay and Konar 1984	2°
Tilapia mossambica	73.6	Dissolved Fe	96 hour LC_5	pH 7.0	Mukhopadhyay and Konar 1984	2°
Asellus aquaticus	81.1	Fe^{3+}	48 hr LC_{50}		Furmanska cited in Martin and Holdich 1986	
Tilapia	83.2	Dissolved	96 hour LC_{50}	pH 7.0	Mukhopadhyay and Konar 1984	2°
Tilapia	92.8	Dissolved	96 hour LC_{95}	pH 7.0	Mukhopadhyay and Konar 1984	2°
Crangonyx pseudogracilis	95	Fe^{2+}	96 hr LC_{50}	pH 6.75, hardn 50 mg/L	Martin and Holdich 1986	2°
Tilapia mossambica	99.4	Dissolved Fe	96 hour LC_5	pH 8.0	Mukhopadhyay and Konar 1984	2°
Leptophiebia (mayfly)	106.3	Fe^{2+}	96 hour LC_{50}	pH 7	Gerhardt 1994	2°
Tilapia mossambica	118	Dissolved Fe	96 hour LC_{50}	pH 8.0	Mukhopadhyay and Konar 1984	2°
Tilapia mossambica	119.6	Dissolved Fe	96 hour LC_{50}	pH 6.5	Mukhopadhyay and Konar 1984	2°

APPENDIX I (CONTINUED)

Species	Iron conc. (mg/L)	Form of iron used	Test Duration	Other variables	Reference *	CCME reference score
Crangonyx pseudogracilis	120	Fe^{3+}	96 hr LC_{50}	pH 6.75, hardness 50 mg/L	Martin and Holdich 1986	2°
Asellus aquaticus	124	Fe^{3+}	96 hr LC_{50}		Martin and Holdich 1986	2°
Tilapia mossambica	128.5	Fe^{2+}	96 hour LC_{95}	pH 6.5	Mukhopadhyay and Konar 1984	2°
Brook trout	133	Ferrous sulphate	24 hours		Belding 1927 cited in Doudoroff and Katz 1953	
Tilapia mossambica	136.8	Fe^{2+}	96 hour LC_{95}	pH 8.0	Mukhopadhyay and Konar 1984	2°
Crangonyx pseudogracilis	143	Fe^{2+}	48 hr LC_{50}	pH 6.75, hardness 50 mg/L	Martin and Holdich 1986	2°
Crangonyx pseudogracilis	160	Fe^{3+}	48 hr LC_{50}	pH 6.75, hardness 50 mg/L	Martin and Holdich 1986	2°
Asellus aquaticus	183	Fe^{3+}	48 hr LC_{50}	pH 6.75, hardness 50 mg/L	Martin and Holdich 1986	2°
Branchiura soerbyi	250	Fe^{2+}	96 hour LC_5	pH 6.5	Mukhopadhyay and Konar 1984	2°
Asellus aquaticus	255.9	Fe^{2+}	50 hour LC_{50}	pH 4.5, organisms from non-polluted site	Maltby *et al*. 1987	2°
Branchiura soerbyi	290	Fe^{2+}	96 hour LC_5	pH 7.0	Mukhopadhyay and Konar 1984	2°
Asellus aquaticus	383.2	Fe^{2+}	50 hour LC_{50}	pH 4.5, from polluted site	Maltby *et al*. (1987)	2°
Asellus aquaticus	430.5	Fe^{2+}	50 hour LC_{50}	pH 6.0, from non-polluted site	Maltby *et al*. (1987)	2°
Branchiura	446	Fe^{2+}	96 hour LC_{50}	pH 8.5	Mukhopadhyay and Konar	2°
Asellus aquaticus	466.9	Fe^{2+}	50 hour LC_{50}	pH 6.0, from polluted site	Maltby *et al*. 1987	2°
Branchiura	560	Fe^{2+}	96 hour LC_{50}	pH 7.0	Mukhopadhyay and Konar	2°
Branchiura soerbyi	580	Fe^{2+}	96 hour LC_{50}	pH 6.5	Mukhopadhyay and Konar 1984	2°
Branchiura soerbyi	830	Fe^{2+}	96 hour LC_{95}	pH 7.0	Mukhopadhyay and Konar 1984	2°
Branchiura soerbyi	835	Fe^{2+}	96 hour LC_{95}	pH 8.5	Mukhopadhyay and Konar 1984	2°
Branchiura soerbyi	920	Fe^{2+}	96 hour LC_{95}	pH 6.5	Mukhopadhyay and Konar 1984	2°
Goldfish	1000	Ferrous sulphate	2-10 hours	pH 6.4, hard H_2O	Ellis 1937 cited in Doudoroff and Katz 1953	
Very young carp	1000	Ferrous sulphate	48 hours		Dyk 1942 cited in Doudoroff and Katz 1953	

2° - secondary by the CCME definition

APPENDIX II: SUMMARY OF CHRONIC IRON LC_{50} VALUES (IN ASCENDING ORDER)

Species	Iron conc. (mg/L)	Form of iron used	Test Duration	Other variables	Reference *	CCME reference score
Rainbow trout spermatozoa	0.005	Fe^{2+}	40 minutes – reduced fertility		Billard and Roubaud 1985	1°
Ephemerella subvaria (a mayfly)	0.32	Fe^{2+}	Mean tolerance limit	temp 18.5 °C; D.O. 8.0 mg/L; pH ~7.25; alkalinity ~50.0 mg/L; acidity ~6.0-12.0 mg/L; hardness 53.0 mg/L	Warnick and Bell 1969	2°
Brook char	1.0	Fe^{2+}	48-hrs - decrease in body [Na]	14°C	Gonzalez *et al.* 1990	1°
Fathead minnows	1.5	Fe^{3+}	Reduced hatchability of eggs		Smith *et al.* 1973	1°
Common shiners	1.54	Fe^{3+}	4-6 weeks – reduced blood sugar		Brenner *et al.* 1976	1°
Pike & tench	1.9	Dissolved iron	"Harmful"	pH 6.7	Schaeperclaus 1941 cited in Doudoroff and Katz 1953	
Brown trout	2.0	Fe^{2+}	72-hours: gill damage	pH 5.0-6.0	Peuranen *et al.* 1994	1°
Daphnia magna	4.38	Fe (III)	16% reproductive impairment		Biesinger and Christensen 1972	2°
Daphnia	4.5	Fe (III)			Randall *et al.* 1999	
Daphnia magna	5.2	Fe (III)	50% reproductive impairment		Biesinger and Christensen 1972	2°
Daphnia magna	5.9	Fe (III)	3 week LC_{50}		Biesinger and Christensen 1972	2°
Goldfish	10	$FeCl_3$	4 days did not kill	pH 5.0, soft H_2O	Ellis 1937 cited in Doudoroff and Katz 1953	
Acroneuria lycorias (a stonefly)	16.0	Fe^{2+}	9-day LC_{50}	temp 18.5 °C; D.O. 8.0 mg/L; pH ~7.25; alkalinity ~50.0 mg/L; acidity ~6.0-12.0 mg/L; hardness 53.0 mg/L	Warnick and Bell 1969	2°
Hydropsyche betteni (a caddisfly)	16.0	Fe^{2+}	7-day LC_{50}	temp 18.5 °C; D.O. 8.0 mg/L; pH ~7.25; alkalinity ~50.0 mg/L; acidity ~6.0-12.0 mg/L; hardness 53.0 mg/L	Warnick and Bell 1969	2°
Black bass & bluegill sunfish	50	Ferrous sulphate	7 days = no kill	pH 6.6	Sanborn 1945 cited in Doudoroff and Katz 1953	
Leptophiebia (mayfly)	70	Fe^{2+}	Lost escape behaviour	pH 7	Gerhardt 1994	
Goldfish	100	$FeCl_3$	4 days did not kill	pH 5.5, hard H_2O	Ellis 1937 cited in Doudoroff and Katz 1953	
Black bass & bluegill sunfish	100	Ferrous sulphate	2.5-7 days	Tap water, pH 6.4	Sanborn 1945 cited in Doudoroff and Katz 1953	
Goldfish	100	Ferrous sulphate	7 days = no kill	pH 6.7-6.4	Sanborn 1945 cited in Doudoroff and Katz 1953	

APPENDIX III: SUMMARY OF EXISTING IRON CRITERIA IN NORTH AMERICA

Water Use	Criteria Values	Reference
Drinking Water (aesthetic)	0.3 mg/L	U.S. EPA 1986
Freshwater Aquatic Life	1.0 mg/L	U.S. EPA 1986
Freshwater Aquatic Life	1.0 mg/L total iron	Sam Ferris, MOE Saskatchewan
Drinking Water	0.3 mg/L	Sam Ferris, MOE Saskatchewan
Drinking Water	0.3 mg/L	EPA 1997
Freshwater Aquatic Life	0.3 mg/L	Ontario MOE 1979 (cited from Bernie Neary)
Freshwater Aquatic Life	0.3 mg/L	Karen Saffron, MOE Alberta