United States Environmental Protection Agency
Office of Water 4304
EPA-822-D-04-001 November 2004

Draft Aquatic Life Water Quality Criteria for Selenium - 2004

Draft

Aquatic Life Water Quality Criteria for

Selenium

2004

November 2004

U.S. Environmental Protection Agency
Office of Water
Office of Science And Technology
Washington, D.C.

NOTICES

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ACKNOWLEDGMENTS

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Introduction

This document provides guidance to States and Tribes authorized to establish water quality standards under the Clean Water Act (CWA) to protect aquatic life from toxic effects of selenium. Under the CWA, States and Tribes are to establish water quality standards to protect designated uses. While this document constitutes the U.S. Environmental Protection Agency's (U.S. EPA) scientific recommendations regarding ambient concentrations of selenium, this document does not substitute for the CWA or U.S. EPA's regulations; nor is it a regulation itself. Thus, it cannot impose legally binding requirements on the U.S. EPA, States, Tribes or the regulated community, and might not apply to a particular situation based upon the circumstances. Interested parties are free to raise questions and objections about the substance of this guidance and the appropriateness of the application of this guidance to a particular situation. State and Tribal decision-makers retain the discretion to adopt approaches on a case-by-case basis that differ from this guidance when appropriate. The U.S. EPA may change this guidance in the future.

This document establishes water quality criteria for protection of aquatic life for selenium. Under Section 304(a) of the CWA, U.S. EPA is to periodically revise water quality criteria to accurately reflect the latest scientific knowledge. Toward this end, a U.S. EPA-sponsored Peer Consultation Workshop on Selenium Aquatic Toxicity and Bioaccumulation on May 27-28, 1998 brought together experts in selenium research to discuss issues related to the chronic criterion for selenium. As a result of findings from the workshop and the fact that a substantial body of literature on the chronic toxicity of selenium has accumulated since the 1987 document was published, U.S. EPA has decided to update the acute and chronic criteria for selenium.

The criteria presented herein supersede all previous national aquatic life water quality criteria for selenium (U.S. EPA 1976, 1980a, 1987a, 1995).

Selenium Chemistry

Water quality criteria are being derived for total selenium measured as selenite-Se plus selenate-Se, but a variety of forms of selenium can occur in water and tissue. Selenium in aquatic ecosystems exists in a broad range of oxidation states: (+ VI) in selenates ($HSeO_4^-$, SeO_4^{2-}) and selenic acid (H_2SeO_4), (+ IV) in selenites ($HSeO_3^-$, SeO_3^{2-}) and selenous acid (H_2SeO_3), 0 in elemental selenium, and (-II) in selenides (Se^{2-}, HSe^-), hydrogen selenide (H_2Se), and organic selenides (R_2Se). Selenium also shows some

tendency to form catenated species like organic diselenides (RseSeR). Within the normal physiological pH range and the reduction potential range permitted by water, only Se, SeO_3^{2-}, $HSeO_3^-$, and SeO_4^{2-} can exist at thermodynamic equilibrium (Milne 1998). While ionic reactions are expected to be rapid in water, oxidation-reduction reactions may be slow, and the possibility exists for the formation of HSe^- in living systems and some environments where anoxic conditions arise. The parallel behavior of comparable species of sulfur and selenium in living systems has often been observed, but it is important to recognize that their chemical characteristics are different in many ways. For instance, selenate is comparable to chromate in oxidizing strength and far stronger than sulfate [$E^0(SeO_4^{2-}/H_2SeO_3)$ = 1.15 V; $E^0(Cr_2O_7^{2-}/Cr^{3+})$ = 1.33V; $E^0(SO_4^{2-}/H_2SO_3)$ = 0.200V (standard potentials in acid solution: Weast 1969)], whereas selenide is a much stronger reducing agent than sulfide [$E^0(Se/H_2Se)$ = -0.36 V; $E^0[S/H_2S$]= 0.14V)].

Inorganic Selenium

Selenate usually predominates in well-aerated surface waters, especially those with alkaline conditions. In spite of its oxidizing strength, selenate (SeO_4^{2-}) exhibits considerable kinetic stability in the presence of reducing agents (Cotton and Wilkinson 1988). The radius of SeO_4^{2-} is comparable to that of SO_4^{2-} (Frausto da Silva and Williams 1991), and uptake by cells is expected to take place via the same ion channels or permeases for both anions. Competition between sulfate and selenate uptake has been observed in many species: algae (Riedel and Sanders 1996), aquatic plants (Bailey et al., 1995), crustacea (Olge and Knight 1996), fungi (Gharieb et al. 1995), HeLa cells (Yan and Frenkel 1994), and wheat (Richter and Bergmann 1993). Reduced selenate bioconcentration with increasing sulfate concentration has been demonstrated in *Daphnia magna* (Hansen et al. 1993). A significant relationship was shown to exist between acute selenate toxicity to aquatic organisms and ambient sulfate concentrations (Brix et al. 2001a). Competition with selenate has also been observed for phosphate in green algae (Riedel and Sanders 1996), and with chromate and tungstate in anaerobic bacteria (Oremland et al. 1989).

Selenous acid species ($HSeO_3^-$ and SeO_3^{2-}) can predominate in solution under the moderately oxidizing conditions encountered in oxygenated waters. Between pH 3.5 and 9.0 biselenite ion is the predominant ion in water, and at pH values below 7.0, selenites are rapidly reduced to elemental selenium under mildly reducing conditions (Faust 1981), situations that are common in bottom sediments.

Most selenite salts are less soluble than the corresponding selenates. The extremely low solubility of ferric selenite $Fe_2(SeO_3)_3$ ($K_s = 2.0 \pm 1.7 \times 10^{-31}$), and of the basic ferric selenite $Fe_2(OH)_4SeO_3$ ($K_s = 10^{-61.7}$), is important to the environmental cycling of selenium. Selenites also form stable adsorption complexes with ferric oxides, forming complexes of even lower solubility than the ferric selenites. Under certain conditions, selenite (in contrast to selenate) seems to be completely adsorbed in high amounts by ferric hydroxide and, to a lesser extent, by aluminum hydroxide (Faust 1981). Coprecipitation techniques have been applied for preconcentration of selenium in natural waters, using iron (III) hydroxides, which coprecipitates selectively the selenite, but not the selenate, species in river and sea waters (Yoshii et al. 1977). Alum and iron coagulation precipitation can be used in water treatment processes to remove selenite (Clifford et al. 1986). The low levels of selenium in ocean waters have been attributed to the adsorption of selenite by the oxides of metals, such as iron and manganese (National Academy of Sciences 1976).

Relative to selenate, selenite is more reactive because of its polar character, resulting from the asymmetric electron density of the ion, its basicity (attraction to bond with proton), and its nucleophilicity (attraction to bond to a nucleus using the lone pair electrons of the ion). No evidence has yet been presented to show that $HSeO_3^-$ or SeO_3^{2-} is taken up intact into the cell interior. Evidence indicates that selenite is reduced rapidly, even before uptake in some cases, making it difficult to distinguish between uptake and metabolic processes (Milne 1998). Freshwater phytoplankton process selenate and selenite by different mechanisms, leading to different concentrations within the cell, and the concentrations attained are affected by various chemical and biological factors in the environment (Riedel et al. 1991). These authors suggested that selenate is transported into the cell by a biological process with low affinity, whereas selenite appears to be largely physically adsorbed. Contradictory evidence suggesting that selenite uptake is enzymatically mediated was found with marine phytoplankton (Baines and Fisher 2001). Experimental results supporting the hypothesis that separate accumulation mechanisms for selenate and selenite are present in *D. magna* have been published (Maier et al. 1993). However, while some organisms appear to absorb selenite nonspecifically, specific transport systems exist in other species. Sulfate competition is insignificant in *Ruppia maritima* (Bailey et al. 1995), and specific uptake systems have been demonstrated in some microorganisms (Heider and Boeck 1993). Selenite uptake in green algae, unlike selenate, is increased substantially at lower pH values, a property that represents another difference between these two anions (Riedel and Sanders 1996). The uptake of

inorganic selenium species, selenate and selenite, by the green alga *Chlamydomonas reinhardtii* (Dang) was examined as a function of pH over the range 5 to 9, and in media with varying concentrations of major ions and nutrients using ^{75}Se as a radiotracer. Little difference was noted in the uptake of selenate as a function of pH, with the maximum uptake found at pH 8; however, selenite uptake increased substantially at the lower pH values. Differences in speciation are suggested to be the cause of these differences. Selenate exists as the divalent ion SeO_4^{2-} over the range of pH tested; whereas monovalent biselenite ion $HSeO_3^-$ is prevelant at these pH values. At the low end of the pH range, neutral selenous acid may also play a role.

Elemental selenium is not measurably soluble in water. It has been reported that elemental selenium is slowly metabolized by several bacteria (Bacon and Ingledew 1989), and the translocation of elemental selenium into the soft tissue of *Macoma balthica* has been reported (Luoma et al. 1992). The bioavailability of elemental selenium to *M. balthica* was assessed by feeding the organisms ^{75}Se-labeled sediments in which the elemental selenium was precipitated by microbial dissimilatory reduction. A 22% absorption efficiency of particulate elemental selenium was observed. In view of the insolubility of elemental selenium, uptake may be preceeded by air oxidation, or in reducing environments thiols may facilitate the solubilization (Amaratunga and Milne 1994). Elemental selenium can be the dominant fraction in sediments (Zawislanski and McGrath 1998).

Selenium is reduced to hydrogen selenide, H_2Se, or other selenides at relatively low redox potentials. Hydrogen selenide by itself is not expected to exist in the aquatic environment since the Se^0/H_2Se couple falls even below the H^+/H_2 couple. Aqueous solutions of H_2Se are actually unstable in air due to its decomposition into elemental selenium and water. Under moderately reducing conditions, heavy metals are precipitated as the selenides, which have extremely low solubilities. The following are log K_s values of some heavy metal selenides of environmental interest: -11.5 (Mn^{2+}), -26.0 (Fe^{2+}), -60.8 (Cu^+), -48.1 (Cu^{2+}), -29.4 (Zn^{2+}), -35.2 (Cd^{2+}), and -64.5 (Hg^{2+}). The precipitation of selenium as heavy metal selenides can be an important factor affecting the cycling of the element in soils and natural waters.

Organoselenium

Organic selenides (conventionally treated as Se(-II) species) in variable concentrations, usually in the form of free and combined selenomethionine and selenocysteine, are also present in natural surface

waters (Fisher and Reinfelder 1991). Dissolved organic selenides may be an important source of selenium for phytoplankton cells, because they can account for ~80% of the dissolved selenium in open ocean surface waters, and for a significant fraction in many other environments as well (Cutter 1989; Cutter and Cutter 1995). Dissolved organoselenium levels of 14.2%, 65% and 66% were measured in samples (one meter depth) from Hyco Reservoir, NC; Robinson Impoundment, SC; and Catfish Lake, NC; respectively (Cutter 1986). The Hyco Reservoir organoselenium was identified as being protein bound. Organoselenium concentrations were found to range from 10.4% (58.7 µg/L) to 53.7% (1.02 µg/L) of the total selenium present in Lake Creek and Benton Lake, MT surface waters (Zhang and Moore 1996). Organoselenium quite often is measured as the difference between total dissolved selenium and the sum of selenite plus selenate, and is therefore not typically characterized. Much more work is needed in the area of specific identification and characterization of the nature of the organic selenides present in aquatic ecosystems. Organoselenium form(s) are much more bioavailable and probably play a very important role in selenium ecotoxic effects (e.g. Besser et al., 1993; Rosetta and Knight 1995).

Departure from Thermodynamic Equilibrium

In the highly dynamic natural waters, there is often a departure from thermodynamic equilibrium. In the thermodymanic models, kinetic barriers to equilibrium and biological processes are not adequately considered, and the speciation of selenium in oxidized natural waters is not accurately predicted. Selenate is usually the predominate form in solution; however, selenite and organoselenium can both exist at concentrations higher than predicted (Faust 1981; Luoma et al. 1992). Bioaccumulation by microorganisms, bioproduction and release of organoselenium, and mineralization of particulate selenium forms contribute to the disequilibrium.

Physical Distribution of Species in Surface Water

The physical distribution of various selenium species in surface waters is regulated by:

- sorption to or incorporation in suspended particulate matter (SPM), and
- complexation with inorganic and/or organic colloidal material, such as $(FeO \bullet OH)_n$ and humic substances (dissolved organic matter, DOM).

Both sorption to SPM and complexation with colloidal matter reduces the bioavailability of the selenium species. The average fraction of selenium associated with the particulate phase (0.45µm filtration) as determined from eleven different studies of various surface waters was found to be 16% (0-39% range) of the total selenium, i.e., an average operationally defined dissolved selenium level of 84% (Text Table

A). In the James River, VA, the dissolved inorganic and organic selenium was found to be 77% and 70% associated with colloidal matter, respectively (Takayangi and Wong 1984). A study of lake ecosystems in Finland (Wang et al. 1995) found that 52% of the dissolved selenium was associated with humic substances, and in a similar speciation study of Finnish stream waters, Lahermo et al. (1998) determined that 36% of the selenium was complexed with humic matter. Hence, in various waterbodies physical distribution as well as chemical speciation of selenium must be considered in relationship to bioavailability and aquatic toxicity.

Up until recently, the organic selenium fraction has been routinely measured as the difference between total dissolved selenium and the sum of selenite and selenate. Unfortunately, the calculation of this important selenium fraction in water as the difference between the total and measurable inorganic fractions has not permitted this fraction to be fully characterized. New techniques are currently being developed which should help the specific identification and characterization of the nature of the organic selenides present in aquatic systems. This work is particularly important because portions of the organic selenium fraction (e.g., selenomethionine) of total dissolved selenium in water have been shown to be much more bioavailable than the other forms of selenium, and therefore this work is also important for understanding the manifestation of selenium ecotoxic effects.

Sources of Selenium to Aquatic Systems

Selenium occurs in many soil types and enters ground and surface waters through natural weathering process such as erosion, leaching and runoff. The national average concentration of selenium in non-seleniferous surface water ranges from 0.1 to 0.4 µg Se/L (Maier and Knight 1993). Elevated levels of selenium occur in surface waters when substantial quantities of selenium enter surface waters from both natural and anthropogenic sources. It is abundant in the alkaline soils of North America from the Great Plains. Some ground waters in California, Colorado, Kansas, Oklahoma, South Dakota and Wyoming contain elevated concentrations of selenium due to weathering of and leaching from rocks and soils. Ecological impacts have been observed where selenium is concentrated through irrigation practices in areas with seleniferous soils. Selenium also occurs in sulfide deposits of copper, lead, mercury, silver and zinc and can be released during the mining and smelting of these ores. In addition, selenium occurs naturally in coal and fuel oil and is emitted in flue gas and in fly ash during combustion. Some selenium then enters surface waters in drainage from fly-ash ponds and in runoff from fly-ash deposits on land. Notable examples of systems that have been affected by selenium originating from coal ash include

Belews Lake, NC, where 16 of the 20 species originally present were eliminated within a few years after discharge began, and Hyco Reservoir, NC, where selenium toxicity was associated with fish larval mortality (Gillespie and Baumann 1986).

Text Table A. Particulate and dissolved selenium as a function of total selenium in freshwater and marine aquatic ecoystems.

Reference	Waterbody	Particulate Se (% of Total)	Fraction dissolved, fd
Cutter 1989	Carquinezitist, CA	20 - 40	0.6 - 0.8
Cutter 1986	Hyco Reservoir, NC	0	1
Tanizaki et al. 1992	Japanese Rivers	16	0.84
Luoma et al. 1992	San Francisco Bay, CA	22 - 31	0.69-0.78
Cumbie and VanHorn, 1978	Belews Lake, NC	8	0.92
GLEC 1997	Unnamed Stream, Albright, WV	4	0.96
Wang et al. 1995	Finnish Lakes	10	0.9
Lahermo et al. 1998	Finnish Streams	8	0.92
Hamilton et al. 2001ab	Adobe Creek, Fruita, CO	18	0.82
Hamilton et al. 2001a,b	North Pond, Fruita, CO	0	1
Hamilton et al. 2001a,b	Fish Ponds, Fruita, CO	7	0.93
Nakamoto and Hassler 1992	Merced River, CA	0	1
Nakamoto and Hassler 1992	Salt Slough, CA	4	0.96
Welsh and Maughan 1994	Cibola Lake, CA	39	0.62
Welsh and Maughan 1994	Hart Mine Marsh, Blythe, CA	6	0.94
Welsh and Maughan 1994	Colorado River, Blythe, CA	11	0.89
Welsh and Maughan 1994	Palo Verda Oxbow Lake, CA	33	0.67
Welsh and Maughan 1994	Palo Verda Oufall Drain, CA	0	1
Welsh and Maughan 1994	Pretty Water Lake, CA	21	0.79

Selenium Biogeochemistry

The current understanding of the biogeochemistry of selenium has recently been reviewed by Fan et al. (2002). Their review clearly shows the extreme complexity of selenium biogeochemistry in aquatic environments. Fan et al. describe the selenium biogeochemical cycle as follows: dissolved selenium oxyanions are primarily absorbed by aquatic producers, including microphytes and bacteria, and biotransformed into organoselenium form(s) and selenium element (Se^{O}). These, together with other particle-bound selenium sources, constitute the particulate selenium fraction of the water column, and they are poorly understood (Zawislanski and McGrath, 1998). Once accumulated in the aquatic primary and secondary producers, selenium can be transferred through various aquatic consumers (e.g.

zooplankton, insect larvae, larval fish, bivalves) into the top predators, including aquatic birds and piscivorous fish. Selenium can be further chemically transformed through the food chain transfer process.

The microscopic planktonic organisms, including microphytes (cyanobacteria and phytoplankton), bacteria, protozoa, and zooplankton are major components of the particulate matter in the water column. The particulate matter, in turn, forms the basis for detrital materials which can settle onto the sediment, and become the food source for sediment organisms. Suspended particulate matter can also be mineralized in the water column. In addition to this selenium input into the sediment, waterborne selenite and selenate can be physically adsorbed onto the sediment particles, ingested, absorbed, and transformed by the sediment organisms. Sediment-bound selenate and selenite can be reduced to insoluble Se^0 by anaerobic microbial activities. This and water column-derived Se^0 can be reduced further to inorganic and organic selenides (-II form), and/or reoxidized to selenite and selenate by microorganisms in the sediment and/or in the digestive tracts of sediment macroinvertebrates. Selenides can enter the food chain via absorption and/or ingestion (by chironomids or tubificid worms, for example) into sediment organisms, or be oxidized to selenite and selenate. Selenium of different oxidation states can be further biotransformed by sediment organisms and transferred up the food chain. Selenium biotransformation, bioaccumulation, and transfer through both sediment and water column foodwebs constitute the major biogeochemical pathways in aquatic ecosystems.

In addition to accumulating selenium into the biomass, the aquatic producers are the main factors controlling the volatilization of selenium via the production of methylated selenides including, dimethylselenide (DMSe) and dimethyldiselenide (DMDSe). These methylated selenides can be oxidized to selenite, or can exit the water column into the atmosphere. Selenium volatilization into the atmosphere may represent an important process responsible for significant loss of selenium in some aquatic systems. Methylated selenides can also be generated from dissolved selenonium precursor(s) released by aquatic producers into the water. Moreover, other organoselenium forms can be released into the water by aquatic producers, and are reoxidized to selenite and/or reabsorbed by aquatic producers.

Narrow Margin Between Sufficiency and Toxicity

Of all the priority and non-priority pollutants, selenium has the narrowest range of what is beneficial for biota and what is detrimental. Selenium is an essential element required as a mineral cofactor in the biosynthesis of glutathione peroxidases. All of the classic glutathione peroxidases contain selenium and

are found to be involved in the catalytic reaction of these many enzymes (Allan 1999). The major function of the glutathione peroxidases was found to involve the reduction of hydrogen peroxide to water at the expense of the oxidation of glutathione, the enzyme's cofactor. Aquatic and terrestrial organisms require 0.5 µg/g dry weight (dw) of selenium in their diet to sustain metabolic processes, whereas concentrations of selenium that are only an order of magnitude greater than the required level have been shown to be toxic to fish. Selenium deficiency has been found to affect humans (U.S. EPA 1987a), sheep and cattle (U.S. EPA 1987a), deer (Oliver et al. 1990) fish (Thorarinsson et al. 1994; Wang and Lovell 1997; Wilson et al. 1997; U.S. EPA 1987a), aquatic invertebrates (Audas et al. 1995; Caffrey 1989; Cooney et al. 1992; Cowgill 1987; Cowgill and Milazzo 1989; Elendt 1990; Elendt and Bais 1990; Harrison et al. 1988; Hyne et al. 1993; Keating and Caffrey 1989; Larsen and Bjerregaard 1995; Lim and Akiyama 1995; Lindstrom 1991; U.S. EPA 1987a; Winner 1989; Winner and Whitford 1987), and algae (Doucette et al. 1987; Keller et al. 1987; Price 1987; Price et al. 1987; Thompson and Hosja 1996; U.S. EPA 1987a; Wehr and Brown 1985).

Selenium has been shown to mitigate the toxic effects of arsenic, cadmium, copper, inorganic and organic mercury, silver, ofloxacin, methyl parathion and the herbicide paraquat to biota in both aquatic and terrestrial environments (Bjerregaard 1988a, b; Cuvin and Furness 1988; Ding et al. 1988; Krizkova et al. 1996; Malarvizhi and Usharani 1994; Micallef and Tyler 1987; Patel et al. 1988; Paulsson and Lundbergh 1991; Pelletier 1986b, 1988; Phillips et al. 1987; Ramakrishna et al. 1988; Rouleau et al. 1992; Salte et al. 1988; Siegel et al. 1991; Szilagyi et al. 1993; U.S. EPA 1987a). Selenium pretreatment resulted in reduced effects in 128-hr old, but not 6-hr old, embryos of *Oryzias latipes* from cadmium and mercury, whereas prior exposure to selenium did not affect the sensitivity of white suckers to cadmium (U.S. EPA 1987a). In contrast, Birge et al. and Huckabee and Griffith reported that selenium and mercury acted synergistically in producing toxic effects to fish embryos (U.S. EPA 1987a). Selenium is reported to reduce the uptake of mercury by some aquatic species (Southworth et al. 1994; U.S. EPA 1987a), to have no effect on uptake of mercury by a mussel, and to increase the uptake of mercury by mammals and some fish (U.S. EPA 1987a). Selenium augmented accumulation of cadmium in some tissues of the shore crab, *Carcinus maenas* (U.S. EPA 1987a). The available data do not show whether the various inorganic and organic compounds and oxidation states of selenium are equally effective sources of selenium as a trace nutrient, or as reducing the toxic effects of various pollutants.

Selenium Document Information

All concentrations reported herein are expressed as selenium, not as the chemical tested. Although Se(VI) is expected to be the predominant oxidation state at chemical equilibrium in oxygenated alkaline waters, the rate of conversion of Se(IV) to Se(VI) seems to be slow in most natural waters. Therefore, it was assumed that when Se(IV) was introduced into stock or test solutions, it would persist as the predominate state throughout the test, even if no analyses specific for the Se(IV) oxidation state were performed. Similarly, it was assumed that when Se(VI) was introduced into stock or test solutions, it would persist as the predominant state throughout the test, even if no analyses specific for Se(VI) were performed.

An understanding of the "Guidelines for Deriving Numerical National Water Quality Criteria for the Protection of Aquatic Organisms and Their Uses" (Stephan et al. 1985), hereafter referred to as the Guidelines, and the response to public comments (U.S. EPA 1985a) is helpful for understanding the derivation of the acute criteria for selenium. Briefly, the Guidelines procedure involves the following steps: (1) Acute toxicity test data is gathered from all suitably conducted studies. Data are to be available for species in a minimum of eight families representing a diverse assemblage of taxa. (2) The Final Acute Value (FAV) is derived by extrapolation or interpolation to a hypothetical genus more sensitive than 95 percent of a diverse assemblage of taxa. The FAV, which represents an LC_{50} or EC_{50}, is divided by two in order to obtain an acute criterion protective of nearly all individuals in such a genus. (3) Chronic toxicity test data (longer-term survival, growth, or reproduction) are needed for at least three taxa. Most often the chronic criterion is set by determining an appropriate acute-chronic ratio (the ratio of acutely toxic concentrations to the chronically toxic concentrations) and applying that ratio to the FAV from the previous step. (4) When necessary, the acute and/or chronic criterion may be lowered to protect critically important species.

The chronic criteria procedure explicitly set forth in the Guidelines (Step 3 above) is not well suited to bioaccumulative contaminants for which diet is the primary route of aquatic life exposure. Consequently, that procedure was not used for deriving the chronic criterion for selenium either in the original 1987 criteria document or in this update. Rather, to accord with other provisions of the Guidelines, it was necessary to apply what the Guidelines refer to as “appropriate modifications” of the procedures in order to obtain a criterion “consistent with sound scientific evidence”, as will be described in a later section.

Results of such intermediate calculations as recalculated LC_{50} values and Species Mean Acute Values are given to four significant figures to prevent roundoff error in subsequent calculations, not to reflect the precision of the value. The latest comprehensive literature search for information for this document was conducted in August 2001; some more recent information was included.

The body of this document contains only the information on acute and chronic toxicity of selenium that is relevant to the derivation of the acute and chronic criteria. Supporting information on the toxicity and bioaccumulation of selenium, and the data that were reviewed and not used in deriving the criteria are provided in appendices and include: sulfate correction of selenate acute toxicity (Appendix A); toxicity to aquatic plants (Appendix B); bioconcentration and bioaccumulation (Appendix C); environmental factors affecting selenium toxicity and bioaccumulation (Appendix D); site-specific considerations (Appendix E); other data (Appendix F); unused data (Appendix G); tissue relationships (Appendix H); chronic data summaries (Appendix I); and background Se levels (Appendix J).

Acute Toxicity of Selenite

Data that may be used, according to the Guidelines, in the derivation of Final Acute Values for selenite are presented in Tables 1a and 1b. The following text presents a brief overview of the acceptable data obtained for selenite, followed by a discussion of the more sensitive, and commercially and recreationally important species. A ranking of the relative sensitivity of selenite to selenate for each genera is listed in Tables 2a and 2b.

Acute Toxicity of Se(IV) to Freshwater Animals

Acceptable data on the acute effects of selenite in freshwater are available for 14 species of invertebrates and 20 species of fish (Table 1a). These 34 species satisfy the eight family provision specified in the Guidelines. Invertebrates are both the most sensitive and the most tolerant freshwater species to selenite with Species Mean Acute Values (SMAV) ranging from 440 µg/L for the crustacean, *Ceriodaphnia dubia*, to 203,000 µg/L for the leech, *Nephelopsis obscura*. The selenite SMAVs for fishes range from 1,783 µg/L for the striped bass, *Morone saxatilis*, to 35,000 µg/L for the common carp, *Cyprinus carpio*. The following text presents a species-by-species discussion of the eight most sensitive genera, plus all commercially and recreationally important species.

Hyalella (amphipod)

The most sensitive freshwater genus is the amphipod, *Hyalella*, with a Genus Mean Acute Value (GMAV) of 461.4 µg Se/L. The GMAV is derived from five 96-hr acute flow-through measured tests where the LC_{50} values ranged from 340 to 670 µg Se/L (GLEC 1998; Halter et al. 1980). A sixth test conducted under non flow-through conditions is also listed in Table 1a (Brasher and Ogle 1993), but the Guidelines recommend using flow-through measured data in preference to static or renewal data.

Ceriodaphnia (cladoceran)

The second most sensitive freshwater genus is *Ceriodaphnia*, with a GMAV of <515.3 µg Se/L that is derived from the geometric mean of the *C. affinis* (<603.6 µg Se/L) and *C. dubia* (440 µg Se/L) SMAVs. Four static unmeasured 48-hr studies are available for *C. affinis* where the LC_{50} values ranged from <480 to 720 µg Se/L (Owsley 1984; Owsley and McCauley 1986). The one available *C. dubia* acute study was conducted by GLEC (1999) that exposed <24-hr old neonates to sodium selenite for 48 hours under flow-through measured conditions. The resultant 48-hr LC_{50} value was 440 µg Se/L, which is the most sensitive SMAV for selenite in the database.

Daphnia (cladoceran)

The eleven available acute values are used to calculate the *Daphnia magna* SMAV of 905.3 µg Se/L (acute LC_{50} values ranged from 215 to 3,020 µg Se/L), but only one flow-through measured acute LC_{50} test value of 1,987 µg Se/L is used for the for *D. pulex* SMAV (a second static measured test conducted by Reading (1979) is listed, but not used to calculate the SMAV) . The resultant GMAV of 1,341 µg Se/L for *Daphnia* is the third most sensitive for selenite.

Hydra

The fourth most sensitive freshwater genus is *Hydra*, with a GMAV of 1,700 µg Se/L. The GMAV is derived from the one available static-measured test conducted by Brooke et al. (1985).

Morone (striped bass)

Two 96-hr static unmeasured tests are available for the striped bass, *Morone saxatilis,* and the LC_{50} values were 1,325 and 2,400 µg Se/L (Palawski et al. 1985). The geometric mean of the two values yield the GMAV of 1,783 µg Se/L.

Pimephales (fathead minnow)

A total of 16 fathead minnow acute studies are presented in Table 1a, but only the eight flow-through measured LC_{50} values are used to derive the GMAV of 2,209 µg Se/L. The eight flow-through LC_{50} values ranged from 620 to 5,200 µg Se/L (Cardwell et al. 1976a,b; GLEC 1998; Kimball manuscript).

Gammarus (amphipod)

The seventh most sensitive freshwater genus is *Gammarus*, with a GMAV of 3,489 µg Se/L that is derived from the geometric mean of five flow-through measured studies (GLEC 1998, 1999) where the LC_{50} values ranged from 1,800 to 10,950 µg Se/L. Two static measured acute studies were conduced by Brooke et al. (1985) and Brooke (1987), but as recommended by the Guidelines, were not used to calculate the SMAV for this species.

Jordanella (flagfish)

The eighth most sensitive freshwater genus is *Jordanella*, with a GMAV of 6,500 µg Se/L. The GMAV is derived from the one available 96-hr flow-through measured test conducted by Cardwell et al. (1976a,b) that exposed *Jordanella floridae* to selenium dioxide.

Oncorhynchus (salmonid)

The GMAV of 10,580 µg Se/L for the commercially important salmonid *Oncorhynchus* is derived from the geometric mean of the coho salmon (O. *kisutch*; 7,240 µg Se/L), chinook salmon (*O. tshawytscha*; 15,596 µg Se/L) and rainbow trout (*O. mykiss*; 10,488 µg Se/L) SMAVs. Three static unmeasured 96-hr studies are used to calculate the coho salmon SMAV where the LC_{50} values ranged from 3,578 to 13,600 µg Se/L (Hamilton and Buhl 1990b; Buhl and Hamilton 1991). A fourth coho salmon LC_{50} value is available for an acute test initiated with the tolerant alevin life stage (Buhl and Hamilton 1991), but based on Guideline recommendations this value is not used when data are available from a more sensitive life stage.

Six acute chinook salmon static unmeasured 96-hr acute studies conducted with the more sensitive post-alevin life stage of the fish are used to determine the 15,596 µg Se/L SMAV for the species and the LC_{50} values ranged from 8,150 to 23,400 µg Se/L (Hamilton and Buhl 1990b). The two acute studies conducted with the tolerant eyed egg and alevin life stages by the same authors are not used in the SMAV determination as recommended by the Guidelines. Hamilton and Buhl (1990b) noted that chinook salmon fry were consistently more sensitive than either the embryos or alevin to selenite.

A total of seven rainbow trout acute studies are presented in Table 1a, but only the two flow-through measured LC_{50} values are used to derive the SMAV of 10,488 µg Se/L as recommended by the Guidelines. The two 96-hr flow-through test LC_{50} values are 8,800 and 12,500 µg Se/L (Goettl and Davies 1976; Hodson et al. 1980). As with the coho and chinook salmon, the alevin life stage was less sensitive to selenite.

Lepomis (bluegill)

The GMAV of 28,500 µg Se/L for the recreationally important bluegill sunfish, *Lepomis macrochirus*, is derived from the 96-hr flow-through measured test conducted by Cardwell et al. (1976a,b). The static measured acute study conduced by Brooke et al. (1985) was not used to calculate the SMAV for this species, as recommended by the Guidelines.

Se(IV) Freshwater Final Acute Value Determination

Freshwater Species Mean Acute Values (Table 1a) were calculated as geometric means of the available acute values for selenite, and Genus Mean Acute Values (Table 2a) were then calculated as geometric

means of the Species Mean Acute Values. Of the 28 genera for which freshwater mean acute values are available, the most sensitive genus, *Hyalella*, is 440 times more sensitive than the most tolerant, *Nephelopsis*. The range of sensitivities of the four most sensitive genera spans a factor of 3.7. The freshwater Final Acute Value (FAV), representing the most sensitive 5th percentile genus, is calculated to be 514.9 µg/L for selenite using the procedure described in the Guidelines and the Genus Mean Acute Values in Table 2a. The Final Acute Value is higher than the lowest Species Mean Acute Value (Figure 1).

Acute Toxicity of Se(IV) to Saltwater Animals

Acute toxicity data that can be used to derive a saltwater criterion for selenite are available for 10 species of invertebrates and eight species of fish that are resident in North America (Table 1b). These 18 species satisfy the eight family provision specified in the Guidelines. The range of SMAVs for saltwater invertebrates extends from 255 µg Se/L for juveniles of the bay scallop, *Argopecten irradians* (Nelson et al. 1988) to greater than 10,000 µg Se/L for embryos of the blue mussel, *Mytilus edulis* (Martin et al. 1981) and embryos of the Pacific oyster, *Crassostrea gigas* (Glickstein 1978; Martin et al. 1981). The range of SMAVs for fish is slightly wider than that for invertebrates, extending from 599 µg Se/L for larvae of the haddock, *Melanogrammus aeglefinus*, to 17,350 µg Se/L for adults of the fourspine stickleback, *Apeltes quadracus* (Cardin 1986). No consistent relationship was detected between life stage of invertebrates or fish and their sensitivity to selenite, and few data are available concerning the influence of temperature or salinity on the toxicity of selenite to saltwater animals. Acute tests with the copepod, *Acartia tonsa*, at 5 and 10°C gave similar results (Lussier 1986). The following text presents a species-by-species discussion of the eight most sensitive genera, plus all commercially and recreationally important species. The genera sensitivity ranking is listed in Table 2b.

<u>*Argopecten* (bay scallop)</u>

The most sensitive saltwater genus is *Argopecten*, with a GMAV of 255 µg Se/L. The GMAV is derived from the one available bay scallop (*Argopecten irradians)* static-renewal unmeasured test conducted by Nelson et al. (1988) at a salinity of 25 g/kg.

Melanogrammus (haddock)

The second most sensitive saltwater genus is *Melanogrammus*, with a GMAV of 599 µg Se/L. The GMAV is derived from the one available haddock (*Melanogrammus aeglefinus*) static unmeasured test conducted by Cardin (1986) at a salinity of 30 g/kg.

Cancer (dungeness crab)

The third most sensitive saltwater genus is *Cancer*, with a GMAV of 1,040 µg Se/L. The GMAV is derived from the one available static unmeasured test conducted by Glickstein (1978) that exposed *Cancer magister* to selenium oxide at a salinity of 33.8 g/kg.

Penaeus (brown shrimp)

The fourth most sensitive saltwater genus is *Penaeus*, with a GMAV of 1,200 µg Se/L. The GMAV is derived from the one available static unmeasured test conducted by Ward et al. (1981) that exposed *Penaeus aztecus* to sodium selenite at a salinity of 30 g/kg.

Acartia (copepod)

The fifth most sensitive saltwater genus is *Acartia*, with a GMAV of 1,331 µg Se/L that is derived from the geometric mean of the *A. clausi* (2,110 µg Se/L) and *A. tonsa* (839 µg Se/L) SMAVs. Each of the SMAVs is derived from one static unmeasured acute test conducted by Lussier (1986) that exposed each species to selenious acid at a salinity of 30 g/kg.

Americamysis (*Mysidopsis*) (mysid)

The GMAV of 1,500 µg Se/L for the mysid *Americamysis* (formerly *Mysidopsis*) is derived from the one *Americamysis bahia* 96-hr flow-through measured test conducted by Ward et al. (1981). The static unmeasured acute study conduced by U.S. EPA (1978) was not used to calculate the SMAV for this species as recommended by the Guidelines. The flow-through measured test was conducted with selenious acid at a salinity of 15-20 g/kg.

Spisula (surf clam)

The seventh most sensitive saltwater genus is *Spisula*, with a GMAV of 1,900 µg Se/L. The GMAV is derived from the one available static-renewal unmeasured test conducted by Nelson et al. (1988) that exposed *Spisula solidissima* to sodium selenite at a salinity of 25 g/kg.

Morone (striped bass)

Five 96-hr static unmeasured tests are available for the striped bass, *Morone saxatilis,* and the LC_{50} values ranged from 1,550 to 3,900 µg Se/L (Chapman 1992; Palawski et al. 1985). The geometric mean of the five values yielded the GMAV of 3,036 µg Se/L. All the tests were conducted with sodium selenite at a salinity of 1-5 g/kg.

Paralichthys (summer flounder)

The GMAV of 3,497 µg Se/L for the commercially important summer flounder, *Paralichthys dentatus,* is derived from one 96-hr static unmeasured acute test conducted by Cardin (1986) that exposed embryos to selenious acid at a salinity of 30.2 g/kg.

Callinectes (blue crab)

The GMAV of 4,600 µg Se/L for the commercially important blue crab, *Callinectes sapidus,* is derived from one static unmeasured acute test conducted by Ward et al. (1981) that exposed juveniles to sodium selenite at a salinity of 30 g/kg.

Crassostrea (Pacific oyster)

Two static unmeasured tests are available for the commercially important Pacific oyster, *Crassostrea gigas*, and the LC_{50} values were both >10,000 µg Se/L (Glickstein 1978; Martin et al. 1981). The geometric mean of the two values yielded the GMAV of >10,000 µg Se/L. The tests were conducted with selenium oxide and sodium selenite at a salinity of 33.8 g/kg.

Mytilus (blue mussel)

The GMAV for the commercially important blue mussel, *Mytilus edulis*, is also >10,000 µg Se/L, and is derived from the one static unmeasured acute test conducted by Martin et al. (1981) that exposed embryos to selenium oxide at a salinity of 33.8 g/kg.

Pseudopleuronectes (winter flounder)

The GMAV of 14,649 µg Se/L for the commercially important winter flounder, *Pseudopleuronectes americanus,* is derived from two 96-hr static unmeasured acute tests conducted by Cardin (1986) that exposed larvae to selenious acid at a salinity of 28-30 g/kg.

Se(IV) Saltwater Final Acute Value Determination

Of the 17 genera for which saltwater mean acute values are available for selenite (Table 2b), the most sensitive genus, *Argopectin*, is 68 times more sensitive than the most tolerant, *Apeltes*. The sensitivities of the four most sensitive genera differ by a factor of 4.7, and these four include three invertebrates and one fish, of which an invertebrate is the most sensitive of the four. The saltwater Final Acute Value, representing the most sensitive 5th percentile genus, is 253.4 µg/L for selenite, which is slightly lower than the lowest Species Mean Acute Value (Figure 2).

Acute Toxicity of Selenate

Data that may be used, according to the Guidelines, in the derivation of Final Acute Values for selenate are presented in Tables 1a and 1b. The following text presents a brief overview of the acceptable data obtained for selenate, and includes a discussion of the more sensitive and important species. The genera sensitivity ranking is listed in Tables 2a and 2b.

Sulfate-dependent Toxicity of Selenate

The toxicity of a number of metals (e.g., copper and cadmium) to aquatic organisms is related to the concentration of hardness in the water. The toxicity of these metals to many different aquatic species has been shown to decrease as the hardness concentration increases. A similar relationship also has been recognized between selenate and dissolved sulfate in freshwater (a similar relationship is not evident between selenite and sulfate or between either form of selenium and hardness). The studies reviewed in this document indicate that, as the concentration of sulfate increases, the acute toxicity of selenate is reduced (less toxic). Selenate acute toxicity tests conducted at different levels of dissolved sulfate are available with *C. dubia, D. magna, H. azteca, G. pseudolimnaeus,* chinook salmon and fathead minnows (Table 1a). These data indicate that, in general, selenate is more toxic to these species in low sulfate water than in higher sulfate water.

Sulfate Correction

As discussed in the introduction of this document, sulfate has been shown to compete with selenate in their uptake into aquatic organisms (Olge and Knight 1996; Riedel and Sanders 1996; Bailey et al. 1995; Hansen et al. 1993) and affect the acute toxicity of selenate (Brix et al. 2001a). Sulfate is used here as a correction to the toxicity of selenate. However, it should be emphasized that the sulfate adjustment is not a precise measure, but an estimation. The variability associated with different life stages, clones and test conditions of the studies used to determine the sulfate slope all contribute to the uncertainty of the sulfate correction. In selected cases, insensitive life stages were not used in the analysis (e.g., the eyed-egg and alevin test results were not used for the chinook salmon).

Following recommendations in the guidelines (Stephan et al. 1985), an analysis of covariance (Sokal and Rohlf 1981) was implemented in Microsoft Excel to calculate a common slope for regression lines projecting the natural logarithm of selenate LC_{50}s as a function of the natural logarithm of sulfate concentrations. The common regression line is the best estimate of the collective relationship between

toxicity and sulfate concentration. With analysis of covariance, different species will be weighted relative to the number of data points they have. In this case, the fathead minnow has 18 data points out of the total of 57, the next most frequent species, *C. dubia,* has 13 data points, and the four remaining species have eight or fewer data points.

This analysis of covariance model was fit to the selenate data in Table 1a for the six species for which definite acute values ("less than" or "greater than" values were not used) were available over a range of sulfate levels, such that the highest sulfate value was at least three times the lowest, and the highest was also at least 100 mg/L higher than the lowest (other species in Table 1a either did not meet these criteria or did not show any sulfate-toxicity trend due to differences in exposure methods, species, age, etc.). A list of the species and acute toxicity-sulfate values used to estimate the acute sulfate slope is provided in Appendix A.

Regression analysis revealed significant, positive slopes for five of six species that had acute values precisely determined. The slopes for all six species ranged from 0.19 to 0.87, and the common slope for these six species was 0.5812. An F-test was used to test the null hypothesis that slopes of all species were equal. This test revealed that the null hypothesis could not be rejected ($F_{5,45} = 2.82$, $P>0.05$). Individual slopes were not significantly different than the overall pooled slope (Tukey test, all $|q| <3.3$, $q_{0.05,(2),47,7} = 4.39$). Analysis of covariance thus confirmed that it is correct to assume that there is no significant variation in slopes among species, and that the overall slope is a reasonable estimate of the relationship between sulfate concentration and selenate toxicity.

The pooled slope of 0.5812 was used to adjust the freshwater selenate acute values in Table 1a to a sulfate level of 100 mg/L, except where it was not possible because no sulfate value was reported. Species Mean Acute Values (SMAV) were calculated as geometric means of the adjusted acute values (only the underlined EC50/LC50 species values were used to calculate the respective SMAV). As stated in the Guidelines (Stephen et al. 1985), flow-through measured study data are normally given preference over non-flow-through data for a particular species. In certain cases flow-through measured results were available, yet preference was given to the sensitive life stage for certain species in calculating SMAVs. Genus Mean Acute Values (GMAV) at a sulfate level of 100 mg/L were then calculated (Table 1a) as geometric means of the available freshwater Species Mean Acute Values and ranked (Table 2a).

Acute Toxicity of Se(VI) to Freshwater Animals (Sulfate Adjusted Values)

Acceptable data on the acute effects of selenate in freshwater are available for 12 invertebrate species and 11 species of fish (Table 1a). These 23 species satisfy the eight family provision of the Guidelines. Invertebrates are both the most sensitive and the most tolerant freshwater species to selenate with sulfate adjusted SMAVs ranging from 593 µg/L for the crustacean, *Daphnia pulicaria*, to 1,515,616 µg/L for the leech, *Nephelopsis obscura*. The selenate SMAVs for fishes range from 10,305 µg/L for the razorback sucker, *Xyrauchen texanus*, to 226,320 µg/L for channel catfish, *Ictalurus punctatus*. The following text presents a species-by-species discussion of the eight most sensitive genera, plus all commercially and recreationally important species.

Ceriodaphnia (cladoceran)

The most sensitive freshwater genus is the cladeceran, *Ceriodaphnia*, with a sulfate adjusted GMAV of 842 µg Se/L. The GMAV is derived from one 48-hr acute flow-through measured test (GLEC 1999). Twelve additional tests conducted under non flow-through conditions are also listed in Table 1a (Brix et al. 2001a,b), but the Guidelines recommend using flow-through measured data in preference to static or renewal data.

Hyalella (amphipod)

The second most sensitive freshwater genus is the amphipod, *Hyalella*, with a sulfate adjusted GMAV of 1,397 µg Se/L. The GMAV is derived from four 96-hr acute flow-through measured tests where the LC_{50} values ranged from 723 to 4,224 µg Se/L (GLEC 1998). Three tests conducted under non flow-through conditions are also listed in Table 1a (Adams 1976; Brasher and Ogle 1993; Brix et al. 2001a,b), but are not used to calculate the SMAV as recommended by the Guidelines.

Daphnia (cladoceran)

The third most sensitive freshwater genus is *Daphnia*, with a sulfate adjusted GMAV of 1,887 µg Se/L that is derived from the geometric mean of the *D. magna* (3,314 µg Se/L), *D. pulex* (3,420 µg Se/L) and *D. pulicaria* (593 µg Se/L) SMAVs. Five static and one static-renewal measured 48-hr studies are available for *D. magna* where the LC_{50} values ranged from 1,955 to 5,093 µg Se/L (Boyum 1984; Brooke et al. 1985; Dunbar et al. 1983; Ingersol et al. 1990; Maier et al. 1993).

The *D. pulex* SMAV of 3,420 µg Se/L is based on the 48-hr flow-through measured test conducted by GLEC (1999) that exposed <24-hr old neonates to sodium selenate. Two static measured tests conducted by Brix et al. (2001a,b), are not used to calculate the SMAV as recommend by the Guidelines.

The one available *D. pulicaria* acute study was conducted by Boyum (1984) that exposed neonates to sodium selenate for 48 hours under static measured conditions. The resultant 48-hr LC_{50} value was 593 µg Se/L, which is the most sensitive SMAV for selenate in the database.

Gammarus (amphipod)

The fourth most sensitive freshwater genus is *Gammarus*, with a sulfate adjusted GMAV of 2,522 µg Se/L that is derived from the geometric mean of the *G. lacustris* (2,747 µg Se/L) and *G. pseudolimnaeus* (2,315 µg Se/L) SMAVs. The static measured acute test conduced by Brix et al. (2001a,b) is the only LC_{50} value available for *G. lacustris*.

The *G. pseudolimnaeus* SMAV of 2,315 µg Se/L is based on five 96-hr flow-through measured tests conducted by GLEC (1998, 1999). Two static measured acute studies were conduced by Brooke et al. (1985) and Brooke (1987), but as recommended by the Guidelines, were not used to calculate the SMAV for this species.

Xyrauchen (razorback sucker)

Six 96-hr static unmeasured tests are available for the razorback sucker, *Xyrauchen texanus*, and the LC_{50} values ranged from 7,839 to 16,184 µg Se/L (Buhl and Hamilton 1996; Hamilton 1995; Hamilton and Buhl 1997a). The geometric mean of the six values yield the GMAV of 10,309 µg Se/L.

Gila (bonytail)

The sixth most sensitive freshwater genus is *Gila*, with a sulfate adjusted GMAV of 10,560 µg Se/L. The GMAV is derived from the one static-unmeasured test conducted with the more sensitive larval stage (Buhl and Hamilton 1996). Four other static-unmeasured tests were conducted with less sensitive life stages, but as recommended by the Guidelines, the results were not used to calculate the SMAV for this species.

Pimephales (fathead minnow)

A total of nine fathead minnow acute studies are presented in Table 1a, but only the five flow-through measured LC_{50} values are used to derive the sulfate adjusted GMAV of 11,346 µg Se/L. The five flow-through LC_{50} values ranged from 7,286 to 18,860 µg Se/L (Spehar 1986; GLEC 1998). The four static tests are not used to calculate the SMAV as recommended by the Guidelines.

Ptychocheilus (Colorado squawfish)

The eighth most sensitive freshwater genus is *Ptychocheilus* with a sulfate adjusted GMAV of 18,484 µg Se/L. The GMAV is derived from the three static-unmeasured test conducted with the sensitive life stage of *Ptychocheilus lucius* (Buhl and Hamilton 1996; Hamilton 1995). Three other static-unmeasured tests were conducted with less sensitive life stages, but as recommended by the Guidelines, the results were not used to calculate the SMAV for this species.

Oncorhynchus (salmonid)

The sulfate adjusted GMAV of 47,164 µg Se/L for the commercially important salmonid *Oncorhynchus* is derived from the geometric mean of the coho salmon (*O. kisutch*; 29,141 µg Se/L), chinook salmon (*O. tshawytscha*; 83,353 µg Se/L) and rainbow trout (*O. mykiss*; 43,192 µg Se/L) SMAVs. Three static unmeasured 96-hr studies are used to calculate the coho salmon SMAV where the LC_{50} values ranged from 20,963 to 51,935 µg Se/L (Buhl and Hamilton 1991; Hamilton and Buhl 1990b). A fourth coho salmon LC_{50} value is available for an acute test initiated with the tolerant alevin life stage (Buhl and Hamilton 1991), but based on Guideline recommendations this value is not used when data are available from a more sensitive life stage.

Five acute chinook salmon static unmeasured 96-hr acute studies conducted with the more sensitive life stage of the fish are used to determine the sulfate adjusted 83,353 µg Se/L SMAV for the species with LC_{50} values ranging from 69,939 to 97,550 µg Se/L (Hamilton and Buhl 1990b). The two acute studies conducted with the tolerant eyed egg and alevin life stages by the same authors are not used in the SMAV determination as recommended by the Guidelines.

A total of four rainbow trout acute studies are presented in Table 1a, but only the results from the two static tests conducted with the sensitive juvenile life stage were used to calculate the SMAV of 43,192 µg Se/L (Brooke et al. 1985; Buhl and Hamilton 1991). The two test results obtained with less sensitive life stages were not used as recommended by the Guidelines.

Lepomis (bluegill)

The sulfate adjusted GMAV of 216,033 µg Se/L for the recreationally important bluegill sunfish, *Lepomis macrochirus*, is derived from the 96-hr static measured test conducted by Brooke et al. (1985) that exposed juvenile bluegill to sodium selenate.

Ictalurus (channel catfish)

The sulfate adjusted GMAV of 226,320 µg Se/L for the commercially important channel catfish, *Ictalurus punctatus*, is derived from the 96-hr static measured test conducted by Brooke et al. (1985) that exposed juvenile catfish to sodium selenate.

Se(VI) Freshwater Final Acute Value Determination

Of the 18 freshwater genera for which mean sulfate adjusted acute values are available for selenate, the most sensitive, *Ceriodaphnia*, is 1,800 times more sensitive than the most tolerant, *Nephelopsis*. The range of sensitivities of the four most sensitive genera, all invertebrates, spans a factor of 3.0.

At a sulfate level of 100 mg/L, the freshwater Final Acute Value, representing the most sensitive 5th percentile genus, was calculated to be 834.4 µg/L for selenate. This Final Acute Value is lower than the acute value of the most sensitive freshwater species (Table 2a and Figure 3). The resultant freshwater Criterion Maximum Concentration (CMC) for selenate (in µg/L) = $e^{(0.5812[\ln(\text{sulfate})]+3.357)}$. At a sulfate level of 100 mg/L this yields 417.2 µg/L, or one-half the FAV.

Acute Toxicity of Se(VI) to Saltwater Animals

The only species with which acute tests have been conducted on selenate in salt water is the striped bass (Table 1b). Klauda (1985a, b) obtained 96-hr selenate LC_{50} values of 9,790 and 85,840 µg/L using flow-through measured methodology with prolarvae and juvenile striped bass, respectively. In static unmeasured tests, Chapman (1992) determined selenate 96-hr LC_{50} values that ranged from 23,700 to 29,000 µg/L using 24 to 32 day posthatch striped bass larvae. The more sensitive prolarvae life stage test conducted under flow-through conditions is used to yield the SMAV and GMAV of 9,790 µg Se/L for the striped bass.

Se(VI) Saltwater Final Acute Value Determination

The one saltwater species available for selenate does not satisfy the eight family provision specified in the Guidelines. Therefore, a saltwater Final Acute Value for selenate cannot be determined.

Comparison of Selenite and Selenate Acute Toxicity

Species Mean Acute Values have been determined for both selenite and selenate with 20 freshwater species (Table 3a) and one saltwater species (Table 3b). Of these 21 species, 20 are more sensitive to Se(IV). Only the amphipod, *Gammarus pseudolimnaeus*, is more sensitive to Se (VI), and is in the sensitive portion of the Table 3a distribution. Consistent with the acute toxicity sensitivity pattern, the FAV for Se(VI) is higher than the FAV for Se (IV).

Table 1a. Acute Toxicity of Selenium to Freshwater Animals

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
FRESHWATER SPECIES						
Selenite						
Hydra (adult), *Hydra sp.*	S, M	Sodium selenite	-	**1,700**	1,700	Brooke et al. 1985
Worm, *Tubifex tubifex*	R, U	Sodium selenite	245	**7,710**	7,710	Khangarot 1991
Leech (adult), *Nephelopsis obscura*	S, M	Sodium selenite	49.8	**203,000**	203,000	Brooke et al. 1985
Snail (adult), *Aplexa hypnorum*	S, M	Sodium selenite	50.6	**53,000**	-	Brooke et al. 1985
Snail (adult), *Aplexa hypnorum*	S, M	Sodium selenite	49.8	**23,000**	34,914	Brooke et al. 1985
Snail, *Physa sp.*	S, U	Sodium selenite	45.7	**24,100**	24,100	Reading 1979
Cladoceran (<24 hr), *Ceriodaphnia dubia*	F, M	Sodium selenite	127 (sulfate=25)	**440**	440	GLEC 1999
Cladoceran (<24 hr), *Ceriodaphnia affinis*	S, U	Sodium selenite	100.8	**600**	-	Owsley 1984; Owsley and McCauley 1986
Cladoceran (36-60 hr), *Ceriodaphnia affinis*	S, U	Sodium selenite	100.8	**720**	-	Owsley 1984
Cladoceran (84-108 hr), *Ceriodaphnia affinis*	S, U	Sodium selenite	100.8	**640**	-	Owsley 1984
Cladoceran (72-120 hr), *Ceriodaphnia affinis*	S, U	Sodium selenite	100.8	**<480**	<603.6	Owsley 1984
Cladoceran, *Daphnia magna*	S, U	Sodium selenite	214	**2,500**	-	Bringmann and Kuhn 1959a
Cladoceran, *Daphnia magna*	S, U	Selenious acid[c]	72	**430**	-	LeBlanc 1980
Cladoceran, *Daphnia magna*	S, M	Sodium selenite	129.5	**1,100**	-	Dunbar et al. 1983
Cladoceran, *Daphnia magna*	S, M	Sodium selenite	138	**450**	-	Boyum 1984
Cladoceran (<24 hr), *Daphnia magna*	S, U	Sodium selenite	-	**215**	-	Adams and Heidolph 1985
Cladoceran (<24 hr), *Daphnia magna*	S, U	Sodium selenite	40	**870**	-	Mayer and Ellersieck 1986
Cladoceran (<24 hr), *Daphnia magna*	S, U	Sodium selenite	280	**2,370**	-	Mayer and Ellersieck 1986

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
Cladoceran, *Daphnia magna*	S, M	Sodium selenite	45.5	**700**	-	Ingersoll et al. 1990
Cladoceran, *Daphnia magna*	S, M	Sodium selenite	136	**3,020**	-	Ingersoll et al. 1990
Cladoceran (<24 hr), *Daphnia magna*	R, M	Sodium selenite	80-100	**550**	-	Maier et al. 1993
Cladoceran, *Daphnia magna*	S, M	Selenious acid	220[d]	**1,220**	9,05.3	Kimball, Manuscript
Cladoceran, *Daphnia pulex*	S, M	Sodium selenite	46.4	3,870	-	Reading 1979; Reading and Buikema 1983
Cladoceran (<24 hr), *Daphnia pulex*	F, M	Sodium selenite	128 (sulfate=25)	**1,987**	1,987	GLEC 1999
Amphipod (adult), *Gammarus pseudolimnaeus*	S, M	Sodium selenite	48.3	4,300	-	Brooke et al. 1985
Amphipod (adult), *Gammarus pseudolimnaeus*	S, M	Sodium selenite	53.6	1,700	-	Brooke 1987
Amphipod, *Gammarus pseudolimnaeus*	F, M	Sodium selenite	139 (sulfate=24)	**2,260**	--	GLEC 1998
Amphipod, *Gammarus pseudolimnaeus*	F, M	Sodium selenite	137 (sulfate=138)	**3,130**	--	GLEC 1998
Amphipod, *Gammarus pseudolimnaeus*	F, M	Sodium selenite	144 (sulfate=326)	**1,800**	--	GLEC 1998
Amphipod, *Gammarus pseudolimnaeus*	F, M	Sodium selenite	138 (sulfate=758)	**3,710**	--	GLEC 1998
Amphipod (adult), *Gammarus pseudolimnaeus*	F, M	Sodium selenite	128 (sulfate=25)	**10,950**	3,489	GLEC 1999
Amphipod (2 mm length), *Hyalella azteca*	R, M	Sodium selenite	133	420	-	Brasher and Ogle 1993
Amphipod, *Hyalella azteca*	F, M	Sodium selenite	329	**340**	-	Halter et al. 1980
Amphipod, *Hyalella azteca*	F, M	Sodium selenite	132 (sulfate=64)	**670**	--	GLEC 1998

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
Amphipod, *Hyalella azteca*	F, M	Sodium selenite	132 (sulfate=138)	**<350**	--	GLEC 1998
Amphipod, *Hyalella azteca*	F, M	Sodium selenite	138 (sulfate=359)	**<460**	--	GLEC 1998
Amphipod, *Hyalella azteca*	F, M	Sodium selenite	138 (sulfate=642)	**570**	461.4	GLEC 1998
Midge (4th instar), *Chironomus decorus*	R, M	Sodium selenite	85	**48,200**	48,200	Maier and Knight 1993
Midge, *Chironomus plumosus*	S, U	Sodium selenite	39	**24,150**	-	Mayer and Ellersieck 1986
Midge, *Chironomus plumosus*	S, U	Sodium selenite	280	**27,850**	25,934	Mayer and Ellersieck 1986
Midge, *Tanytarsus dissimilis*	F, M	Selenium dioxide	48	**42,500**	42,500	Call et al. 1983
Coho salmon (0.5 g), *Oncorhynchus kisutch*	S, U	Sodium selenite	211	**7,800**	-	Hamilton and Buhl 1990b
Coho salmon (2.6 g), *Oncorhynchus kisutch*	S, U	Sodium selenite	333	**13,600**	-	Hamilton and Buhl 1990b
Coho salmon (alevin), *Oncorhynchus kisutch*	S, U	Sodium selenite	41	35,560[f]	-	Buhl and Hamilton 1991
Coho salmon (juvenile), *Oncorhynchus kisutch*	S, U	Sodium selenite	41	**3,578**	7,240	Buhl and Hamilton 1991
Chinook salmon (0.7 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenite	211	**14,800**	-	Hamilton and Buhl 1990b
Chinook salmon (0.5 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenite	211	**13,000**	-	Hamilton and Buhl 1990b
Chinook salmon (1.6 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenite	333	**23,100**	-	Hamilton and Buhl 1990b
Chinook salmon (1.6 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenite	333	**23,400**	-	Hamilton and Buhl 1990b
Chinook salmon (eyed egg), *Oncorhynchus tshawytscha*	S, U	Sodium selenite	41.7	>348,320[f]	-	Hamilton and Buhl 1990b
Chinook salmon (alevin), *Oncorhynchus tshawytscha*	S, U	Sodium selenite	41.7	64,690[f]	-	Hamilton and Buhl 1990b

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
Chinook salmon (0.31 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenite	41.7	**16,980**	-	Hamilton and Buhl 1990b
Chinook salmon (0.46 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenite	41.7	**8,150**	15,596	Hamilton and Buhl 1990b
Rainbow trout, *Oncorhynchus mykiss*	S, U	Sodium selenite	330	4,500	-	Adams 1976
Rainbow trout, *Oncorhynchus mykiss*	S, U	Sodium selenite	330	4,200	-	Adams 1976
Rainbow trout, *Oncorhynchus mykiss*	S, U	Sodium selenite	272	1,800	-	Hunn et al. 1987
Rainbow trout (alevin), *Oncorhynchus mykiss*	S, U	Sodium selenite	41	118,000	-	Buhl and Hamilton 1991
Rainbow trout (juvenile), *Oncorhynchus mykiss*	S, U	Sodium selenite	41	9,000	-	Buhl and Hamilton 1991
Rainbow trout, *Oncorhynchus mykiss*	F, M	Sodium selenite	30	**12,500**	-	Goettl and Davies 1976
Rainbow trout, *Oncorhynchus mykiss*	F, M	Sodium selenite	135	**8,800**	10,488	Hodson et al. 1980
Brook trout (adult), *Salvelinus fontinalis*	F, M	Selenium dioxide	157	**10,200**	10,200	Cardwell et al. 1976a,b
Arctic grayling (alevin), *Thymallus arcticus*	S, U	Sodium selenite	41	34,732[f]	-	Buhl and Hamilton 1991
Arctic grayling (juvenile), *Thymallus arcticus*	S, U	Sodium selenite	41	**15,675**	15,675	Buhl and Hamilton 1991
Goldfish, *Carassius auratus*	F, M	Selenium dioxide	157	**26,100**	26,100	Cardwell et al. 1976a,b
Common carp, *Cyprinus carpio*	R, U	-	-	**35,000**	35,000	Sato et al. 1980
Golden shiner, *Notemigonus crysoleucas*	F, M	Sodium selenite	72.2	**11,200**	11,200	Hartwell et al. 1989
Fathead minnow, *Pimephales promelas*	S, U	Sodium selenite	312 (13°C)	10,500	-	Adams 1976
Fathead minnow, *Pimephales promelas*	S, U	Sodium selenite	312 (13°C)	11,300	-	Adams 1976

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
Fathead minnow, *Pimephales promelas*	S, U	Sodium selenite	303 (20°C)	6,000	-	Adams 1976
Fathead minnow, *Pimephales promelas*	S, U	Sodium selenite	303 (20°C)	7,400	-	Adams 1976
Fathead minnow, *Pimephales promelas*	S, U	Sodium selenite	292 (25°C)	3,400	-	Adams 1976
Fathead minnow, *Pimephales promelas*	S, U	Sodium selenite	292 (25°C)	2,200	-	Adams 1976
Fathead minnow (30 days), *Pimephales promelas*	S, M	Sodium selenite	51.1	1,700	-	Brooke et al. 1985
Fathead minnow (juvenile), *Pimephales promelas*	S, U	Sodium selenite	40	7,760	-	Mayer and Ellersieck 1986
Fathead minnow (fry), *Pimephales promelas*	F, M	Selenium dioxide	157	**2,100**	-	Cardwell et al. 1976a,b
Fathead minnow (juvenile), *Pimephales promelas*	F, M	Selenium dioxide	157	**5,200**	-	Cardwell et al. 1976a,b
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenite	131 (sulfate=24)	**3,670**	--	GLEC 1998
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenite	131 (sulfate=160)	**2,920**	--	GLEC 1998
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenite	145 (sulfate=214)	**3,390**	--	GLEC 1998
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenite	140 (sulfate=870)	**2,380**	-	GLEC 1998
Fathead minnow, *Pimephales promelas*	F, M	Selenious acid	220[d]	**620**	-	Kimball, Manuscript
Fathead minnow, *Pimephales promelas*	F, M	Selenious acid	220[d]	**970**	2,209	Kimball, Manuscript
Colorado squawfish (fry), *Ptychocheilus lucius*	S, U	Sodium selenite	197	**6,398**	-	Hamilton 1995
Colorado squawfish (0.4-1.1 g juvenile), *Ptychocheilus lucius*	S, U	Sodium selenite	197	**16,452**	-	Hamilton 1995
Colorado squawfish (1.7 g juvenile), *Ptychocheilus lucius*	S, U	Sodium selenite	197	**14,624**	-	Hamilton 1995

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
Colorado squawfish (larva), *Ptychocheilus lucius*	S, U	Sodium selenite	199	**7,960**	-	Buhl and Hamilton 1996
Colorado squawfish (juvenile), *Ptychocheilus lucius*	S, U	Sodium selenite	199	**17,350**	-	Buhl and Hamilton 1996
Colorado squawfish (0.024-0.047 g), *Ptychocheilus lucius*	S, U	Sodium selenite	144	**20,700**	12,801	Hamilton and Buhl 1997a
Bonytail (fry), *Gila elegans*	S, U	Sodium selenite	197	**8,680**	-	Hamilton 1995
Bonytail (1.1 g juvenile), *Gila elegans*	S, U	Sodium selenite	197	**7,769**	-	Hamilton 1995
Bonytail (2.6 g juvenile), *Gila elegans*	S, U	Sodium selenite	197	**6,855**	-	Hamilton 1995
Bonytail (larva), *Gila elegans*	S, U	Sodium selenite	199	**14,490**	-	Buhl and Hamilton 1996
Bonytail (juvenile), *Gila elegans*	S, U	Sodium selenite	199	**12,870**	9,708	Buhl and Hamilton 1996
Razorback sucker (fry), *Xyrauchen texanus*	S, U	Sodium selenite	197	**6,855**	-	Hamilton 1995
Razorback sucker (0.9 g juvenile), *Xyrauchen texanus*	S, U	Sodium selenite	197	**4,067**	-	Hamilton 1995
Razorback sucker (2.0 g juvenile), *Xyrauchen texanus*	S, U	Sodium selenite	197	**7,312**	-	Hamilton 1995
Razorback sucker (larva), *Xyrauchen texanus*	S, U	Sodium selenite	199	**10,450**	-	Buhl and Hamilton 1996
Razorback sucker (juvenile), *Xyrauchen texanus*	S, U	Sodium selenite	199	**8,520**	-	Buhl and Hamilton 1996
Razorback sucker (0.006-0.042 g), *Xyrauchen texanus*	S, U	Sodium selenite	144	**11,300**	7,679	Hamilton and Buhl 1997a
White sucker, *Catostomus commersoni*	F, M	Sodium selenite	10.2	**29,000**	-	Klaverkamp et al. 1983a
White sucker, *Catostomus commersoni*	F, M	Sodium selenite	18	**31,400**	30,176	Duncan and Klaverkamp 1983

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
Flannelmouth sucker (12-13 days), *Catostomus latipinnis*	S, U	Sodium selenite	144	**19,100**	19,100	Hamilton and Buhl 1997b
Striped bass (63 days), *Morone saxatilis*	S, U	Sodium selenite	40	**1,325**	-	Palawski et al. 1985
Striped bass (63 days), *Morone saxatilis*	S, U	Sodium selenite	285	**2,400**	1,783	Palawski et al. 1985
Channel catfish (juvenile), *Ictalurus punctatus*	S, M	Sodium selenite	49.8	16,000	-	Brooke et al. 1985
Channel catfish (juvenile), *Ictalurus punctatus*	S, U	Sodium selenite	41	4,110	-	Mayer and Ellersieck 1986
Channel catfish, *Ictalurus punctatus*	F, M	Selenium dioxide	157	**13,600**	13,600	Cardwell et al. 1976a,b
Flagfish, *Jordanella floridae*	F, M	Selenium dioxide	157	**6,500**	6,500	Cardwell et al. 1976a,b
Mosquitofish, *Gambusia affinis*	S, U	Sodium selenite	45.7	**12,600**	12,600	Reading 1979
Bluegill (juvenile), *Lepomis macrochirus*	S, M	Sodium selenite	50.5	12,000	-	Brooke et al. 1985
Bluegill, *Lepomis macrochirus*	F, M	Selenium dioxide	157	**28,500**	28,500	Cardwell et al. 1976a,b
Yellow perch, *Perca flavescens*	F, M	Sodium selenite	10.2	**11,700**	11,700	Klaverkamp et al. 1983a

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	LC50 or EC50 Adj. To Sulfate = 100 (µg/L)	Species Mean Acute Value at Sulfate = 100 (µg/L)	Reference
FRESHWATER SPECIES							
Selenate							
Hydra (adult), *Hydra sp.*	S, M	Sodium selenate	53.6 (sulfate=12)	7300	**25,032**	25,032	Brooke et al. 1985
Leech (adult), *Nephelopsis obscura*	S, M	Sodium selenate	49.3 (sulfate=12)	442000	**1,515,661**	1,515,661	Brooke et al. 1985
Snail, *Aplexa hypnorum*	S, M	Sodium selenate	51.0 (sulfate=12)	193000	**661,816**	661,816	Brooke et al. 1985

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	LC50 or EC50 Adj. To Sulfate = 100 (µg/L)	Species Mean Acute Value at Sulfate = 100 (µg/L)	Reference
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=52)	1967	2,877	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=55)	1864	2,638	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=31)	1078	2,129	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=38)	580	1,018	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=98)	1822	1,844	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=98)	1728	1,748	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=213)	1453	936	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=217)	2812	1,793	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=378)	5553	2,564	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=378)	5481	2,531	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=926)	9157	2,512	--	Brix et al. 2001a,b

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	LC50 or EC50 Adj. To Sulfate = 100 (µg/L)	Species Mean Acute Value at Sulfate = 100 (µg/L)	Reference
Cladoceran (<24 hr), *Ceriodaphnia dubia*	S, M	Sodium selenate	52 (sulfate=1205)	9311	2,191	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Ceriodaphnia dubia*	F, M	Sodium selenate	127 (sulfate=25)	376	**842**	842	GLEC 1999
Cladoceran, *Daphnia magna*	S, M	Sodium selenate	129.5 (sulfate=163)	5300	**3,990**	--	Dunbar et al. 1983
Cladoceran, *Daphnia magna*	S, M	Sodium selenate	138 (sulfate=22)	1010	**2,435**	--	Boyum 1984
Cladoceran, *Daphnia magna*	S, M	Sodium selenate	48.1 (sulfate=12)	570	**1,955**	--	Brooke et al. 1985
Cladoceran, *Daphnia magna*	S, M	Sodium selenate	45.5 (sulfate=41)	2560	**4,298**	--	Ingersoll et al. 1990
Cladoceran, *Daphnia magna*	S, M	Sodium selenate	136 (sulfate=68)	4070	**5,093**	--	Ingersoll et al. 1990
Cladoceran (<24 hr), *Daphnia magna*	R, M	Sodium selenate	80-100 (sulfate=82)	2840	**3,187**	3,314	Maier et al. 1993
Cladoceran (<24 hr), *Daphnia pulex*	S, M	Sodium selenate	52 (sulfate=54)	10123	14,482	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Daphnia pulex*	S, M	Sodium selenate	52 (sulfate=38)	8126	14,233	--	Brix et al. 2001a,b
Cladoceran (<24 hr), *Daphnia pulex*	F, M	Sodium selenate	147 (sulfate=25)	1528	**3,420**	3,420	GLEC 1999
Cladoceran, *Daphnia pulicaria*	S, M	Sodium selenate	138 (sulfate=22)	246	**593**	593	Boyum 1984
Amphipod (8-12 mm), *Gammarus lacustris*	S, M	Sodium selenate	116 (sulfate=120)	3054	**2,747**	2,747	Brix et al. 2001a,b
Amphipod (adult), *Gammarus pseudolimnaeus*	S, M	Sodium selenate	46.1 (sulfate=12)	75	257	--	Brooke et al. 1985
Amphipod (adult), *Gammarus pseudolimnaeus*	S, M	Sodium selenate	51.0 (sulfate=12)	57	196	--	Brooke 1987

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	LC50 or EC50 Adj. To Sulfate = 100 (µg/L)	Species Mean Acute Value at Sulfate = 100 (µg/L)	Reference
Amphipod, *Gammarus pseudolimnaeus*	F, M	Sodium selenate	139 (sulfate=25)	1180	**2,641**	--	GLEC 1998
Amphipod, *Gammarus pseudolimnaeus*	F, M	Sodium selenate	132 (sulfate=125)	2870	**2,521**	--	GLEC 1998
Amphipod, *Gammarus pseudolimnaeus*	F, M	Sodium selenate	137 (sulfate=367)	3710	**1,743**	--	GLEC 1998
Amphipod, *Gammarus pseudolimnaeus*	F, M	Sodium selenate	134 (sulfate=635)	3270	**1,167**	--	GLEC 1998
Amphipod (adult), *Gammarus pseudolimnaeus*	F, M	Sodium selenate	131 (sulfate=25)	2191	**4,904**	2,315	GLEC 1999
Amphipod, *Hyalella azteca*	F, U	Sodium selenate	336.8 (sulfate NA)	760	--	--	Adams 1976
Amphipod (2 mm length), *Hyalella azteca*	R, M	Sodium selenate	133 (sulfate=13)	1031	3,375	-	Brasher and Ogle 1993
Amphipod (7-10 days), *Hyalella azteca*	S, M	Sodium selenate	52 (sulfate=55)	1424	2,021	--	Brix et al. 2001a,b
Amphipod, *Hyalella azteca*	F, M	Sodium selenate	143 (sulfate=40)	2480	**4,224**	--	GLEC 1998
Amphipod, *Hyalella azteca*	F, M	Sodium selenate	132 (sulfate=125)	1350	**1,186**	--	GLEC 1998
Amphipod, *Hyalella azteca*	F, M	Sodium selenate	137 (sulfate=367)	1540	**723**	--	GLEC 1998
Amphipod, *Hyalella azteca*	F, M	Sodium selenate	133 (sulfate=822)	3580	**1,052**	1,397	GLEC 1998
Midge (4th instar), *Chironomus decorus*	R, M	Sodium selenate	85 (sulfate=27)	23700	**50,727**	50,727	Maier and Knight 1993
Midge (3rd instar), *Paratanytarsus parthenogeneticus*	S, M	Sodium selenate	49.4 (sulfate=12)	20000	**68,582**	68,582	Brooke et al. 1985
Coho salmon (0.5 g), *Oncorhynchus kisutch*	S, U	Sodium selenate	211 (sulfate=185)	32500	**22,730**	--	Hamilton and Buhl 1990b

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	LC50 or EC50 Adj. To Sulfate = 100 (µg/L)	Species Mean Acute Value at Sulfate = 100 (µg/L)	Reference
Coho salmon (1.7 g), *Oncorhynchus kisutch*	S, U	Sodium selenate	333 (sulfate=291)	39000	**20,963**	--	Hamilton and Buhl 1990b
Coho salmon (alevin), *Oncorhynchus kisutch*	S, U	Sodium selenate	41 (sulfate=41)	158,422[f]	265,990[f]	--	Buhl and Hamilton 1991
Coho salmon (juvenile), *Oncorhynchus kisutch*	S, U	Sodium selenate	41 (sulfate=41)	30932	**51,935**	29,141	Buhl and Hamilton 1991
Chinook salmon (0.7 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenate	211 (sulfate=185)	121000	**84,626**	--	Hamilton and Buhl 1990b
Chinook salmon (0.5 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenate	211 (sulfate=185)	100000	**69,939**	--	Hamilton and Buhl 1990b
Chinook salmon (1.6 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenate	333 (sulfate=291)	180000	**96,752**	--	Hamilton and Buhl 1990b
Chinook salmon (1.6 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenate	333 (sulfate=291)	134000	**72,026**	--	Hamilton and Buhl 1990b
Chinook salmon (eyed egg), *Oncorhynchus tshawytscha*	S, U	Sodium selenate	41.7 (sulfate=47)	>552,000[f]	>856,083[f]	--	Hamilton and Buhl 1990b
Chinook salmon (alevin), *Oncorhynchus tshawytscha*	S, U	Sodium selenate	41.7 (sulfate=47)	>176,640[f]	>273,947[f]	--	Hamilton and Buhl 1990b
Chinook salmon (0.31 g), *Oncorhynchus tshawytscha*	S, U	Sodium selenate	41.7 (sulfate=47)	62900	**97,550**	83,353	Hamilton and Buhl 1990b
Rainbow trout (juvenile), *Oncorhynchus mykiss*	S, M	Sodium selenate	51.0 (sulfate=12)	24000	**82,298**	--	Brooke et al. 1985

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	LC50 or EC50 Adj. To Sulfate = 100 (µg/L)	Species Mean Acute Value at Sulfate = 100 (µg/L)	Reference
Rainbow trout (alevin), *Oncorhynchus mykiss*	S, U	Sodium selenate	41 (sulfate=41)	196460	329,856[f]	--	Buhl and Hamilton 1991
Rainbow trout, *Oncorhynchus mykiss*	F, M	Sodium selenate	45 (sulfate=12)	47000	161,168[f]	--	Spehar 1986
Rainbow trout (juvenile), *Oncorhynchus mykiss*	S, U	Sodium selenate	41 (sulfate=41)	13501	**22,668**	43,192	Buhl and Hamilton 1991
Arctic grayling (alevin), *Thymallus arcticus*	S, U	Sodium selenate	41 (sulfate=41)	41800	**70,182**	--	Buhl and Hamilton 1991
Arctic grayling (juvenile), *Thymallus arcticus*	S, U	Sodium selenate	41 (sulfate=41)	75240	**126,328**	94,159	Buhl and Hamilton 1991
Fathead minnow, *Pimephales promeles*	S, U	Sodium selenate	323 (sulfate NA)	11800	--	--	Adams 1976
Fathead minnow, *Pimephales promeles*	S, U	Sodium selenate	323 (sulfate NA)	11000	--	--	Adams 1976
Fathead minnow, *Pimephales promeles*	S, U	Sodium selenate	323 (sulfate NA)	12500	--	--	Adams 1976
Fathead minnow (juvenile), *Pimephales promelas*	S, M	Sodium selenate	47.9 (sulfate =12)	2300	7,887	--	Brooke et al. 1985
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenate	46 (sulfate =12)	5500	**18,860**	--	Spehar 1986
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenate	136 (sulfate=24)	6210	**14,236**	--	GLEC 1998
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenate	127 (sulfate=160)	10800	**8,218**	--	GLEC 1998
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenate	131 (sulfate=474)	18000	**7,286**	--	GLEC 1998

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	LC50 or EC50 Adj. To Sulfate = 100 (µg/L)	Species Mean Acute Value at Sulfate = 100 (µg/L)	Reference
Fathead minnow, *Pimephales promelas*	F, M	Sodium selenate	147 (sulfate=906)	42100	**11,695**	11,346	GLEC 1998
Colorado squawfish (fry), *Ptychocheilus lucius*	S, U	Sodium selenate	196 (sulfate=164)	27588	**20,694**	--	Hamilton 1995
Colorado squawfish (0.4-1.1 g juvenile), *Ptychocheilus lucius*	S, U	Sodium selenate	196 (sulfate=164)	119548	89,676[f]	--	Hamilton 1995
Colorado squawfish (1.7 g juvenile), *Ptychocheilus lucius*	S, U	Sodium selenate	196 (sulfate=164)	138358	103,786[f]	--	Hamilton 1995
Colorado squawfish (larva), *Ptychocheilus lucius*	S, U	Sodium selenate	199 (sulfate=174)	13580	**9,842**	--	Buhl and Hamilton 1996
Colorado squawfish (0.024-0.047 g), *Ptychocheilus lucius*	S, U	Sodium selenate	144 (sulfate=97)	88000	89,572[f]	--	Hamilton and Buhl 1997a
Colorado squawfish (juvenile), *Ptychocheilus lucius*	S, U	Sodium selenate	199 (sulfate=174)	42780	**31,005**	18,484	Buhl and Hamilton 1996
Bonytail (fry), *Gila elegans*	S, U	Sodium selenate	196 (sulfate=164)	22990	17,245[f]	--	Hamilton 1995
Bonytail (1.1 g juvenile), *Gila elegans*	S, U	Sodium selenate	196 (sulfate=164)	102828	77,134[f]	--	Hamilton 1995
Bonytail (2.6 g juvenile), *Gila elegans*	S, U	Sodium selenate	196 (sulfate=164)	90706	68,041[f]	--	Hamilton 1995
Bonytail (juvenile), *Gila elegans*	S, U	Sodium selenate	199 (sulfate=174)	24010	17,401[f]	--	Buhl and Hamilton 1996

Table 1a. Acute Toxicity of Selenium to Freshwater Animals (continued)

Species	Method[a]	Chemical	Hardness (mg/L as $CaCO_3$)	LC50 or EC50 (µg/L)[b]	LC50 or EC50 Adj. To Sulfate = 100 (µg/L)	Species Mean Acute Value at Sulfate = 100 (µg/L)	Reference
Bonytail (larva), *Gila elegans*	S, U	Sodium selenate	199 (sulfate=174)	14570	**10,560**	10,560	Buhl and Hamilton 1996
Razorback sucker (fry), *Xyrauchen texanus*	S, U	Sodium selenate	196 (sulfate=164)	20064	**15,051**	--	Hamilton 1995
Razorback sucker (0.9 g juvenile), *Xyrauchen texanus*	S, U	Sodium selenate	196 (sulfate=164)	15048	**11,288**	- -	Hamilton 1995
Razorback sucker (2.0 g juvenile), *Xyrauchen texanus*	S, U	Sodium selenate	196 (sulfate=164)	10450	**7,839**	--	Hamilton 1995
Razorback sucker (larva), *Xyrauchen texanus*	S, U	Sodium selenate	199 (sulfate=174)	13910	**10,081**	--	Buhl and Hamilton 1996
Razorback sucker (juvenile), *Xyrauchen texanus*	S, U	Sodium selenate	199 (sulfate=174)	7620	**5,523**	--	Buhl and Hamilton 1996
Razorback sucker (0.006-0.042 g), *Xyrauchen texanus*	S, U	Sodium selenate	144 (sulfate=97)	15900	**16,184**	10,309	Hamilton and Buhl 1997a
Flannelmouth sucker (12-13 days), *Catostomus latipinnis*	S, U	Sodium selenate	144 (sulfate=97)	26900	**27,380**	27,380	Hamilton and Buhl 1997b
Channel catfish (juvenile), *Ictalurus punctatus*	S, M	Sodium selenate	51.0 (sulfate=12)	66000	**226,320**	226,320	Brooke et al. 1985
Bluegill (juvenile), *Lepomis macrochirus*	S, M	Sodium selenate	50.4 (sulfate=12)	63000	**216,033**	216,033	Brooke et al. 1985

[a] S = static; R = renewal; F = flow-through; M = measured; U = unmeasured.
[b] Concentration of selenium, not the chemical. **Note:** The values underlined in this column were used to calculate the SMAV for the respective species.
[c] Reported by Barrows et al. (1980) in work performed in the same laboratory under the same contract.
[d] From Smith et al. (1976).
[e] Calculated from regression equation.
[f] Not used in calculation of Species Mean Acute Value because data are available for a more sensitive life stage.

Table 1b. Acute Toxicity of Selenium to Saltwater Animals

Species	Method[a]	Chemical	Salinity (g/kg)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
SALTWATER SPECIES						
Selenite						
Blue mussel (embryo), *Mytilus edulis*	S, U	Selenium oxide	33.79	**>10,000**	>10,000	Martin et al. 1981
Bay scallop (juvenile), *Argopecten irradians*	R, U	Sodium selenite	25	**255**	255	Nelson et al. 1988
Pacific oyster (embryo), *Crassostrea gigas*	S, U	Selenium oxide	33.79	**>10,000**	-	Glickstein 1978; Martin et al. 1981
Pacific oyster (embryo), *Crassostrea gigas*	S, U	Sodium selenite	33.79	**>10,000**	>10,000	Glickstein 1978
Surf clam (juvenile), *Spisula solidissima*	R, U	Sodium selenite	25	**1,900**	1,900	Nelson et al. 1988
Copepod (adult), *Acartia clausi*	S, U	Selenious acid	30	**2,110**	2,110	Lussier 1986
Copepod (adult), *Acartia tonsa*	S, U	Selenious acid	30	**839**	839	Lussier 1986
Mysid (juvenile), *Americamysis bahia*	S, U	Selenious acid	-	600	-	U.S. EPA 1978
Mysid (juvenile), *Americamysis bahia*	F, M	Selenious acid	15-20	**1,500**	1,500	Ward et al. 1981
Brown shrimp (juvenile), *Penaeus aztecus*	S, U	Sodium selenite	30	**1,200**	1,200	Ward et al. 1981
Dungeness crab (zoea 1arva), *Cancer magister*	S, U	Selenium oxide	33.79	**1,040**	1,040	Glickstein 1978
Blue crab (juvenile), *Callinectes sapidus*	S, U	Sodium selenite	30	**4,600**	4,600	Ward et al. 1981

Table 1b. Acute Toxicity of Selenium to Saltwater Animals (continued).

Species	Method[a]	Chemical	Salinity (g/kg)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
Haddock (larva), *Melanogrammus aeglefinus*	S, U	Selenious acid	30	**599**	599	Cardin 1986
Sheepshead minnow (juvenile), *Cyrinodon variegatus*	S, U	Selenious acid	-	6,700	-	Heitmuller et al. 1981
Sheepshead minnow (juvenile), *Cyrinodon variegatus*	F, M	Sodium selenite	30	**7,400**	7,400	Ward et al. 1981
Atlantic silverside (juvenile), *Menidia menidia*	S, U	Selenious acid	30	**9,725**	9,725	Cardin 1986
Fourspine stickleback (adult), *Apeltes quadracus*	S, U	Selenious acid	30	**17,350**	17,350	Cardin 1986
Striped bass, *Morone saxatilis*	S, U	Sodium selenite	1	**1,550**	-	Palawski et al. 1985
Striped bass (24 d posthatch), *Morone saxatilis*	S, U	Sodium selenite	5	**3,400**	-	Chapman 1992
Striped bass (25 d posthatch), *Morone saxatilis*	S, U	Sodium selenite	5	**3,300**	-	Chapman 1992
Striped bass (31 d posthatch), *Morone saxatilis*	S, U	Sodium selenite	5	**3,800**	-	Chapman 1992
Striped bass (32 d posthatch), *Morone saxatilis*	S, U	Sodium selenite	5	**3,900**	3,036	Chapman 1992
Pinfish (juvenile), *Lagodon rhomboides*	S, U	Sodium selenite	30	**4,400**	4,400	Ward et al. 1981
Summer flounder (embryo), *Paralichthys dentatus*	S, U	Selenious acid	30.2	**3,497**	3,497	Cardin 1986
Winter flounder (larva), *Pseudopleuronectes americanus*	S, U	Selenious acid	30	**14,240**	-	Cardin 1986
Winter flounder (larva), *Pseudopleuronectes americanus*	S, U	Selenious acid	28	**15,070**	14,649	Cardin 1986

Table 1b. Acute Toxicity of Selenium to Saltwater Animals (continued).

Species	Method[a]	Chemical	Salinity (g/kg)	LC50 or EC50 (µg/L)[b]	Species Mean Acute Value (µg/L)	Reference
			Selenate			
Striped bass (24 d posthatch), *Morone saxatilis*	S, U	Sodium selenate	5	26,300[c]	-	Chapman 1992
Striped bass (25 d posthatch), *Morone saxatilis*	S, U	Sodium selenate	5	23,700[c]	-	Chapman 1992
Striped bass (31 d posthatch), *Morone saxatilis*	S, U	Sodium selenate	5	26,300[c]	-	Chapman 1992
Striped bass (32 d posthatch), *Morone saxatilis*	S, U	Sodium selenate	5	29,000[c]	-	Chapman 1992
Striped bass (juvenile), *Morone saxatilis*	F, M	Sodium selenate	6.0-6.5	85,840[c]	-	Klauda 1985a,b
Striped bass (prolarvae), *Morone saxatilis*	F, M	Sodium selenate	3.5-4.2	**9,790**	9,790	Klauda 1985a,b

[a] S = static; R = renewal; F = flow-through; M = measured; U = unmeasured.
[b] Concentration of selenium, not the chemical. **Note:** The values underlined in this column were used to calculate the SMAV for the respective species.
[c] Not used in calculation of Species Mean Acute Value because data are available for a more sensitive life stage.

Table 2a. Ranked Freshwater Genus Mean Acute Values

Rank[a]	Genus Mean Acute Value (µg/L)	Species	Species Mean Acute Value (µg/L)[b]	Number of Acute Values used to Calculate Species Mean Value[b]
		FRESHWATER SPECIES		
		Selenite		
28	203,000	Leech, *Nephelopsis obscura*	203,000	1
27	42,500	Midge, *Tanytarsus dissimilis*	42,500	1
26	35,356	Midge, *Chironomus decorus*	48,200	1
		Midge, *Chironomus plumosus*	25,934	2
25	35,000	Common carp, *Cyprinus carpio*	35,000	1
24	34,914	Snail, *Aplexa hypnorum*	34,914	2
23	28,500	Bluegill, *Lepomis macrochirus*	28,500	1
22	26,100	Goldfish, *Carassius auratus*	26,100	1
21	24,100	Snail, *Physa sp.*	24,100	1
20	24,008	White sucker, *Catostomus commersoni*	30,176	2
		Flannelmouth sucker *Catostomus latipinnis*	19,100	1
19	15,675	Arctic grayling *Thymallus arcticus*	15,675	1
18	13,600	Channel catfish, *Ictalurus punctatus*	13,600	1
17	12,801	Colorado squawfish, *Ptychocheilus lucias*	12,801	6
16	12,600	Mosquitofish, *Gambusia affinis*	12,600	1
15	11,700	Yellow perch, *Perca flavescens*	11,700	1
14	11,200	Golden shiner, *Notemigonus crysoleucas*	11,200	1
13	10,580	Chinook salmon, *Oncorhynchus tshawytscha*	15,596	6

Table 2a. Ranked Freshwater Genus Mean Acute Values (continued)

Rank[a]	Genus Mean Acute Value (µg/L)	Species	Species Mean Acute Value (µg/L)[b]	Number of Acute Values used to Calculate Species Mean Value[b]
		Coho salmon, *Oncorhynchus kisutch*	7,240	3
		Rainbow trout, *Oncorhynchus mykiss*	10,488	2
12	10,200	Brook trout *Salvelinus fontinalis*	10,200	1
11	9,708	Bonytail *Gilas elegans*	9,708	5
10	7,710	Worm, *Tubifex tubifex*	7,710	1
9	7,679	Razorback sucker, *Xyrauchen texanus*	7,679	6
8	6,500	Flagfish, *Jordanella floridae*	6,500	1
7	3,489	Amphipod, *Gammarus pseudolimnaeus*	3,489	5
6	2,209	Fathead minnow, *Pimephales promelas*	2,209	8
5	1,783	Striped bass, *Morone saxatilis*	1,783	2
4	1,700	Hydra, *Hydra sp.*	1,700	1
3	1,341	Cladoceran, *Daphnia magna*	905.3	11
		Cladoceran, *Daphnia pulex*	1,987	1
2	<515.3	Cladoceran, *Ceriodaphnia affinis*	<603.6	4
		Cladoceran, *Ceriodaphnia dubia*	440	1
1	461.4	Amphipod, *Hyalella azteca*	461.4	5

Table 2a. Ranked Freshwater Genus Mean Acute Values (continued)

Rank[a]	Genus Mean Acute Value (µg/L)	Species	Species Mean Acute Value (µg/L)[b]	Number of Acute Values used to Calculate Species Mean Value[b]
		Selenate (at sulfate = 100 mg/L)		
18	1,515,661	Leech, *Nephelopsis obscura*	1,515,661	1
17	661,816	Snail, *Aplexa hypnorum*	661,816	1
16	226,320	Channel catfish, *Ictalurus punctatus*	226,320	1
15	216,033	Bluegill, *Lepomis macrochirus*	216,033	1
14	94,159	Arctic grayling, *Thymallus arcticus*	94,159	2
13	68,582	Midge, *Paratanytarsus parthenogeneticus*	68,582	1
12	50,727	Midge, *Chironomus decorus*	50,727	1
11	47,164	Chinook salmon, *Oncorhynchus tshawytscha*	83,353	5
		Coho salmon, *Oncorhynchus kisutch*	29,141	3
		Rainbow trout, *Oncorhynchus mykiss*	43,192	2
10	27,380	Flannelmouth sucker *Catostomus latipinnis*	27,380	1
9	25,032	Hydra, *Hydra sp.*	25,032	1
8	18,484	Colorado squawfish, *Ptychocheilus lucius*	18,484	3
7	11,346	Fathead minnow, *Pimephales promelas*	11,346	5
6	10,560	Bonytail, *Gila elegans*	10,560	1
5	10,309	Razorback sucker, *Xyrauchen texanus*	10,309	6
4	2,522	Amphipod, *Gammarus lacustris*	2,747	1
		Amphipod, *Gammarus pseudolimnaeus*	2,315	5

Table 2a. Ranked Freshwater Genus Mean Acute Values (continued)

Rank[a]	Genus Mean Acute Value (µg/L)	Species	Species Mean Acute Value (µg/L)[b]	Number of Acute Values used to Calculate Species Mean Value[b]
3	1,887	Cladoceran, *Daphnia magna*	3,314	6
		Cladoceran, *Daphnia pulex*	3,420	1
		Cladoceran, *Daphnia pulicaria*	593	1
2	1,397	Amphipod, *Hyalella azteca*	1,397	4
1	842	Cladoceran, *Ceriodaphnia dubia*	842	1

[a] Ranked from most resistant to most sensitive based on Genus Mean Acute Value. Inclusion of "greater than" and "less than" values does not necessarily imply a true ranking, but does allow use of all genera for which data are available so that the Final Acute Value is not unnecessarily lowered.
[b] From Table 1a.

Table 2b. Ranked Saltwater Genus Mean Acute Values

Rank[a]	Genus Mean Acute Value (µg/L)	Species	Species Mean Acute Value (µg/L)[b]	Number of Acute Values used to Calculate Species Mean Value[b]
		SALTWATER SPECIES		
		Selenite		
17	17,350	Fourspine stickleback, *Apeltes quadracus*	17,350	1
16	14,649	Winter flounder, *Pseudopleuronectes americanus*	14,649	2
15	>10,000	Blue mussel, *Mytilus edulis*	>10,000	1
14	>10,000	Pacific oyster, *Crassostrea gigas*	>10,000	2
13	9,725	Atlantic silverside, *Menidia menidia*	9,725	1
12	7,400	Sheepshead minnow, *Cyprinodon variegatus*	7,400	1
11	4,600	Blue crab, *Callinectes sapidus*	4,600	1
10	4,400	Pinfish, *Lagodon rhomboides*	4,400	1
9	3,497	Summer flounder, *Paralichthys dentatus*	3,497	1
8	3,036	Striped bass, *Morone saxatilis*	3,036	5
7	1,900	Surf clam, *Spisula solidissima*	1,900	1
6	1,500	Mysid, *Americamysis bahia*	1,500	1
5	1,331	Copepod, *Acartia clausi*	2,110	1
		Copepod, *Acartia tonsa*	839	1
4	1,200	Brown shrimp, *Penaeus aztecus*	1,200	1
3	1,040	Dungeness crab, *Cancer magister*	1,040	1
2	599	Haddock, *Melanogrammus aeglefinus*	599	1

Table 2b. Ranked Saltwater Genus Mean Acute Values

Rank[a]	Genus Mean Acute Value (µg/L)	Species	Species Mean Acute Value (µg/L)[b]	Number of Acute Values used to Calculate Species Mean Value[b]
1	255	Bay scallop, *Argopecten irradians*	255	1
		Selenate		
1	9,790	Striped bass, *Morone saxatilis*	9,790	1

[a] Ranked from most resistant to most sensitive based on Genus Mean Acute Value. Inclusion of "greater than" and "less than" values does not necessarily imply a true ranking, but does allow use of all genera for which data are available so that the Final Acute Value is not unnecessarily lowered.
[b] From Table 1b.

Selenite

Fresh Water

Final Acute Value = 514.9 µg/L

Criterion Maximum Concentration = (514.9 µg/L) ÷ 2 = 257 µg/L

Salt Water

Final Acute Value = 253.4 µg/L

Criterion Maximum Concentration = (253.4 µg/L) ÷ 2 = 127 µg/L

Selenate

Fresh Water

Final Acute Value = 834.4 µg/L (calculated at a sulfate level of 100 mg/L from GMAVs)

Criterion Maximum Concentration = (834.4 µg/L) ÷ 2 = 417 µg/L (at a sulfate level of 100 mg/L)

Pooled Slope = 0.5812 (see Appendix A)

ln (Criterion Maximum Intercept) = ln(417.2) - [slope x ln(100)]

= 6.0335 - (0.5812 x 4.605) = 3.357

Criterion Maximum Concentration for Selenate (at a sulfate level of 100 mg/L) = $e^{(0.5812[\ln(\text{sulfate})]+3.357)}$

Table 3a. Ratios of Freshwater Species Mean Acute Values for Selenite and Selenate.

Selenite Sensitivity Rank from Table 2a[a]	Species	Selenite Species Mean Acute Value (µg/L)[b]	Selenate Species Mean Acute Value at Sulfate = 100 (µg/L)[b]	Ratio
	FRESHWATER SPECIES			
28	Leech, *Nephelopsis obscura*	203,000	1,515,661	0.134
27	Midge, *Tanytarsus dissimilis*	42,500	NA[c]	NA
26	Midge, *Chironomus decorus*	48,200	50,727	0.95
	Midge, *Chironomus plumosus*	25,934	NA	NA
25	Common carp, *Cyprinus carpio*	35,000	NA	NA
24	Snail, *Aplexa hypnorum*	34,914	616,816	0.057
23	Bluegill, *Lepomis macrochirus*	28,500	216,033	0.132
22	Goldfish, *Carassius auratus*	26,100	NA	NA
21	Snail, *Physa sp.*	24,100	NA	NA
20	White sucker, *Catostomus commersoni*	30,176	NA	NA
	Flannelmouth sucker *Catostomus latipinnis*	19,100	27,380	0.698
19	Arctic grayling *Thymallus articus*	15,675	94,159	0.166
18	Channel catfish, *Ictalurus punctatus*	13,600	226,320	0.06
17	Colorado squawfish, *Ptychocheilus lucias*	12,801	18,484	0.693
16	Mosquitofish, *Gambusia affinis*	12,600	NA	NA
15	Yellow perch, *Perca flavescens*	11,700	NA	NA
14	Golden shiner, *Notoemigonus crysoleucas*	11,200	NA	NA

Table 3a. Ratios of Freshwater Species Mean Acute Values for Selenite and Selenate (continued).

Selenite Sensitivity Rank from Table 2a[a]	Species	Selenite Species Mean Acute Value (µg/L)[b]	Selenate Species Mean Acute Value at Sulfate = 100 (µg/L)[b]	Ratio
13	Chinook salmon, *Oncorhynchus tshawytscha*	15,596	83,353	0.187
	Coho salmon, *Oncorhynchus kisutch*	7,240	29,141	0.248
	Rainbow trout, *Oncorhynchus mykiss*	10,488	43,192	0.243
12	Brook trout *Salvelinus fontinalis*	10,200	NA	NA
11	Bonytail *Gilas elegans*	9,708	10,560	0.919
10	Worm, *Tubifex tubifex*	7,710	NA	NA
9	Razorback sucker, *Xyrauchen texanus*	7,679	10,309	0.745
8	Flagfish, *Jordanella floridae*	6,500	NA	NA
7	Amphipod, *Gammarus pseudolimnaeus*	3,489	2,315	1.507
6	Fathead minnow, *Pimephales promelas*	2,209	11,346	0.195
5	Striped bass, *Morone saxatilis*	1,783	NA	NA
4	Hydra, *Hydra sp.*	1,700	25,032	0.068
3	Cladoceran, *Daphnia magna*	905.3	3,314	0.273
	Cladoceran, *Daphnia pulex*	1,987	3,420	0.581
2	Cladoceran, *Ceriodaphnia affinis*	<603.6	NA	NA
	Cladoceran, *Ceriodaphnia dubia*	440	842	0.523
1	Amphipod, *Hyalella azteca*	461.4	1,397	0.33

[a] Ranked from most resistant to most sensitive based on selenite Genus Mean Acute Value (from Table 2a).
[b] From Table 1a.
[c] NA = Not Available

Table 3b. Ratios of Saltwater Species Mean Acute Values for Selenite and Selenate.

Sensitivity Rank from Table 2b[a]	Species	Selenite Species Mean Acute Value (µg/L)[b]	Selenate Species Mean Acute Value (µg/L)[b]	Ratio
	SALTWATER SPECIES			
8	Striped bass, *Morone saxatilis*	3,036	9,790	0.31

[a] Ranked from most resistant to most sensitive based on Genus Mean Acute Value (from Table 2b).
[b] From Table 1b.

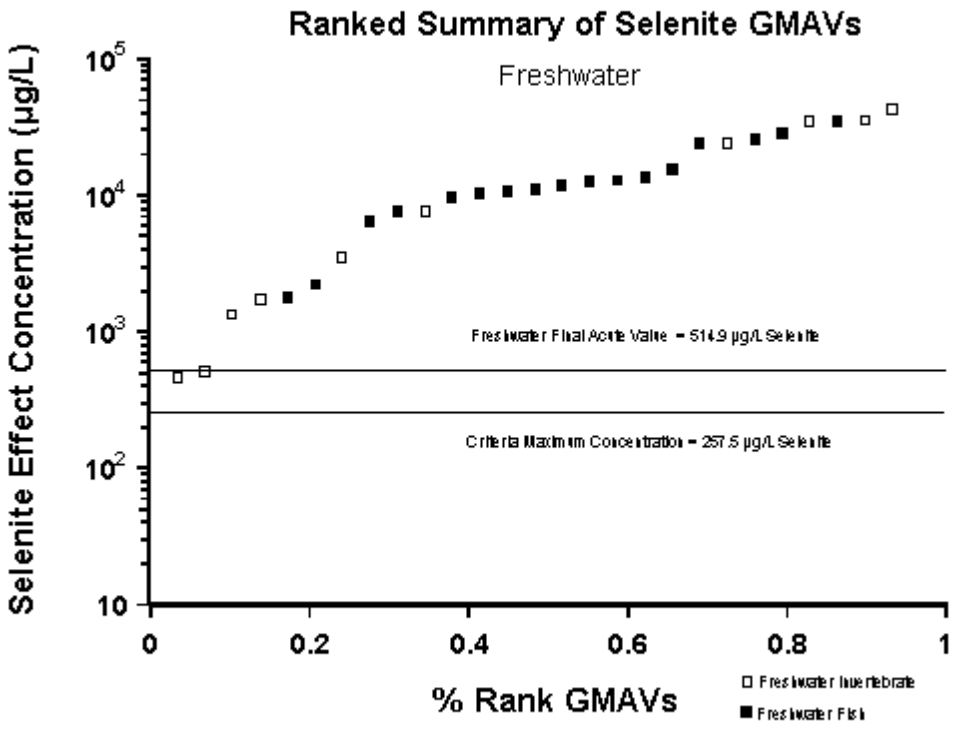

Figure 1. Ranked summary of selenite GMAVs (freshwater).

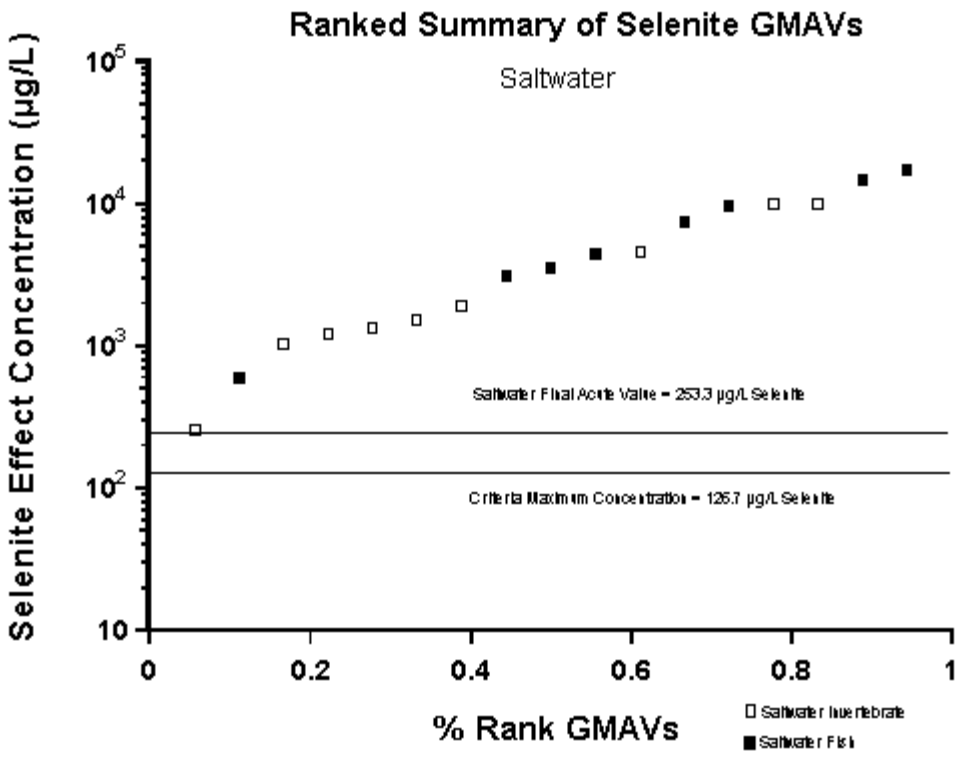

Figure 2. Ranked summary of selenate GMAVs (saltwater).

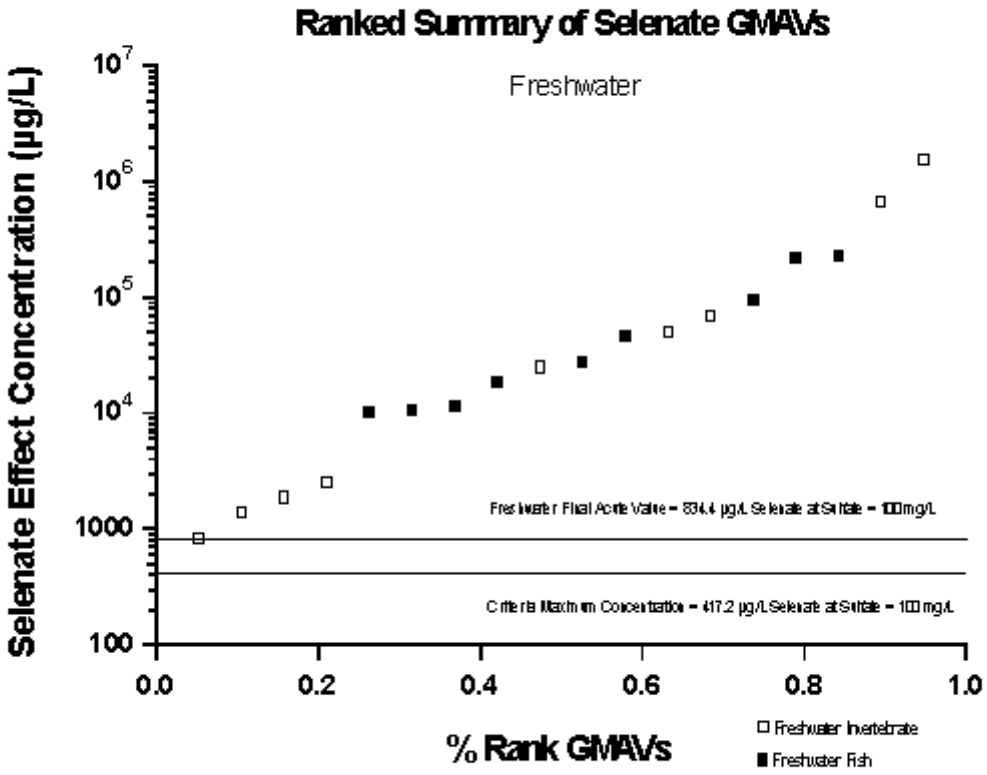

Figure 3. Ranked summary of selenate GMAVs (freshwater) at a sulfate level of 100 mg/L.

Review and Analysis of Chronic Data

Since the issuance of the 1987 chronic criterion of 5 µg/L, considerable information has come forth regarding the route of exposure of selenium to aquatic organisms. Studies have shown that diet is the primary route of exposure that controls chronic toxicity to fish, the group considered to be the most sensitive to chronic selenium exposure (Coyle et al. 1993; Hamilton et al. 1990; Hermanutz et al. 1996). Chronic tests in which test organisms were exposed to selenium only through water and which have measured selenium in the tissue of the test species have produced questionably low chronic values based on the tissue concentrations. Some of these water-only exposures have required aqueous concentrations of selenium of greater than 300 µg/L to attain body burdens sufficient to achieve a chronic response that would have been reached in the real world at aqueous concentrations approximately 30 times lower (Cleveland et al. 1993; Gissel-Nielsen and Gissel-Nielsen 1978).

Because diet controls selenium chronic toxicity in the environment and water-only exposures require unrealistic aqueous concentrations in order to elicit a chronic response, only studies in which test organisms were exposed to selenium in their diet alone or in their diet and water were considered in the derivation of a chronic value. To be able to use the chronic study results, the measurements had to include selenium in the test species tissue. Both laboratory and field studies were considered in the review process. Chronic studies reviewed were obtained through a literature search extending back to the last revision review, from information supplied to U.S. EPA through the Notice of Data Availability, and using the references cited in previous selenium criteria documents.

Selection of Medium for Expressing Chronic Criterion

Whole-body tissue concentration of selenium on a dry weight basis, for species eliciting the chronic response, was selected as the medium from which to base the chronic criterion value. As discussed above, a water-based criterion is not appropriate for selenium because diet is the most important route of exposure for chronic toxicity. The option of basing the chronic criterion on the concentration of selenium in prey species (that is, in the diet of the target species), was considered inappropriate for two reasons: 1) the concentration of selenium in the diet is an indirect measure of effects observed in the test species and is dependent on feeding behavior of the target species, and 2) selection of what organism to sample to assess attainment of a criterion based on diet is problematic in the implementation of such a criterion. Sediment has also been proposed as a medium upon which to base the selenium chronic criterion (Canton and Van Derveer 1997; Van Derveer and Canton 1997), but because of the patchiness

of selenium in sediment and an insufficient amount of data to support a causal link between concentrations of selenium in sediment and chronic effects observed in fish (see Hamilton and Lemly 1999, for a review), a sediment-based criterion was rejected.

Besides being a direct link to chronic endpoints, a tissue-based criterion has the positive attributes of integrating many site-specific factors, such as chemical speciation and rates of transformation, large variations in temporal concentrations in water, types of organisms constituting the food chain, and rates of exchange between water, sediment, and organisms (Hamilton, in preparation; U.S. EPA 1998). Whole-body tissue was selected over specific tissue types, such as ovary, liver, kidney or muscle because of practical reasons of sampling and because a sufficient data base containing chronic effects based on whole-body tissue is present in the literature. Ovaries may be the best tissue to link selenium to reproductive effects because of its role in the maternal transfer of selenium to eggs, and embryo-larval development being one of the most sensitive endpoint for chronic effects. However, ovarian tissue is also only available seasonally and sometimes difficult to extract in quantities sufficient for analysis, especially in smaller fish species. Whole-body larval tissue is also not practical due to sampling and seasonal constraints.

To increase the number of studies in which chronic effects could be compared with selenium concentrations in whole-body tissue, the relationships between selenium concentrations in whole-body and selenium concentrations in ovary, liver, and muscle tissues were estimated. Data from 4 dietary exposure studies that sampled whole-body as well as muscles, ovary, or liver allowed the projection of whole-body concentrations as a function of concentrations in these individual tissues. It was not possible to estimate such relationship for kidneys and carcass because of insufficient data. One species (bluegill sunfish) comprised over 90 percent of the data evaluated for these relationships.

Median concentrations of selenium in the whole-body were projected as a linear function of selenium concentrations in ovaries and liver, or as an exponential function of the natural logarithm of selenium concentrations in muscles (Figure 4; Appendix H). When selenium concentration in more than one organ or tissue was available, muscle tissue was used preferentially for converting into an equivalent whole-body value. Where appropriate, whole-body selenium concentrations were estimated from selenium concentrations in muscle, ovary and liver according to the following equations (see Appendix H for details on statistical analyses):

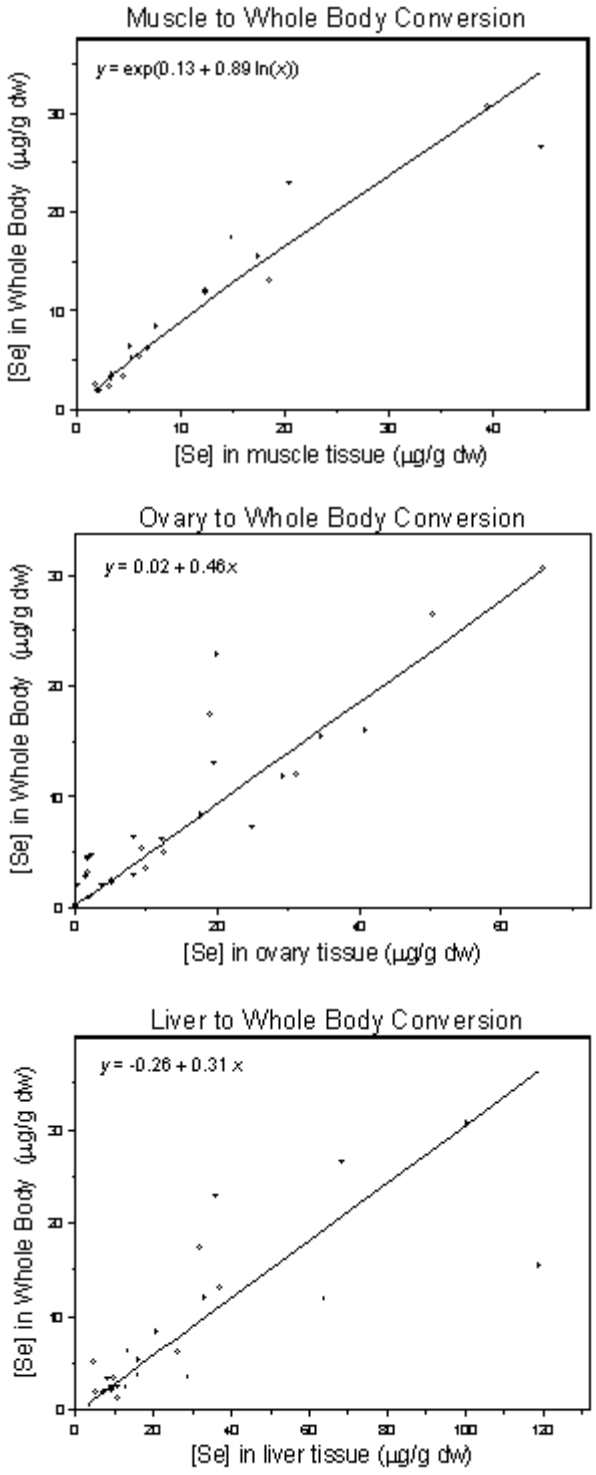

Figure 4. The quantile regression curves project median selenium concentrations in the whole body of bluegill, largemouth bass, tilapia and carp as a function of selenium concentrations in their tissues. Most data are from bluegill. Estimates of model parameters minimize the sum of weighted absolute deviations (see Appendix H for details about statistical analyses).

$$[Se_{whole\text{-}body}] = \exp(0.1331 + (0.8937 \times \ln[Se_{muscle}])) \quad \text{(I)}$$

$$[Se_{whole\text{-}body}] = 0.0173 + (0.4634 \times [Se_{ovary}]) \quad \text{(II)}$$

$$[Se_{whole\text{-}body}] = -0.2609 + (0.3071 \times [Se_{liver}]) \quad \text{(III)}$$

Chronic studies that reported selenium concentrations in tissues based on wet weight were converted to dry weight using a moisture content of 0.80 (U.S. EPA 1985b), unless specified otherwise. Note that because conversion from wet to dry weight and from tissue to whole-body selenium concentration can increase uncertainty in the estimate, site-data analysts should develop their own conversion factors whenever possible to improve accuracy. The basis for such factors can be obtained from local historical data or from newly acquired data specific for that site and species.

Calculation of Chronic Values

In aquatic toxicity tests, chronic values have usually been defined as the geometric mean of the highest concentration of a toxic substance at which no adverse effect is observed (highest no observed adverse effect concentration, NOAEC) and the lowest concentration of the toxic substance that causes an adverse effect (lowest observed adverse effect concentration, LOAEC). The significance of observed effects is determined by statistical tests comparing responses of organisms exposed to natural concentrations of the toxic substance (control) against responses of organisms exposed to elevated concentrations. Analysis of variance is the most common test employed for such comparisons. This approach however, has its limitations. Since neither NOAEC or LOAEC are known in advance and the number of concentrations that can be tested is constrained by logistic and financial resources, observed effects of elevated concentrations may not permit accurate estimates of chronic values. For instance, if all elevated concentrations had high adverse effects or if the difference in concentrations between two significantly different treatments was large, it would not be possible to define either the NOAEC or LOAEC with precision. Furthermore, as the concentration of some substances (e.g., selenium) naturally varies among ecosystems, a concentration that is above the normal range at one site, maybe within the normal range at a different location. In this approach to calculate chronic values, natural variation in concentrations of a substance implies that controls are site specific, and thus multiple tests are needed to define the chronic value at different locations.

An alternative approach to calculate chronic values focuses on the use of regression analysis to define the dose-response relationship. With a regression equation, which defines the level of adverse effects as a function of increasing concentrations of the toxic substance, it is possible to determine the concentration that causes a relatively small effect, for example a 5 to 30 percent reduction in response. A reduction of 20 percent in the response observed at control (EC_{20}) was used as the chronic value because it represents a low level of effect that is generally significantly different from the control (U.S. EPA 1999). Smaller reductions in growth, survival, or other endpoints only rarely can be detected statistically. Effect concentrations associated with such small reductions have wide uncertainty bands, making them unreliable for criteria derivation. Adverse effects are generally modeled as a sigmoid function of increasing concentrations of the toxic substance (Figure 5).

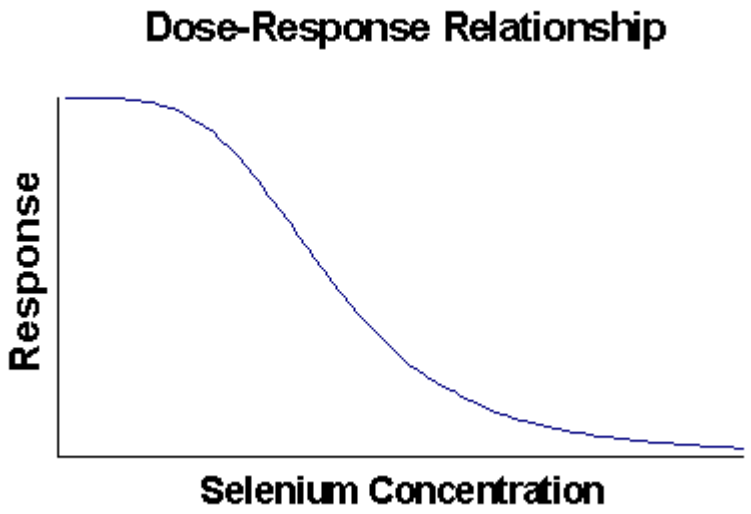

Figure 5. Reductions in survival, growth or other responses of organisms are often modeled as a sigmoid function of increasing concentrations of selenium, or any other toxic substance.

A logistic regression was used to model negative effects of increasing concentrations of selenium on growth, survival, or percent of normal individuals (without deformities) of several aquatic species. The equations that described such functions were then used to estimate the concentration that promoted a 20 percent reduction in response observed at control levels (EC_{20}). These analyses were performed using the Toxic Effects Analysis Model software (version 0.02; R. Erickson, U.S. EPA Duluth).

Only data sets that met the following conditions were included in the analysis: (1) the experiment had a control treatment, which made it possible to define response levels at natural concentrations of selenium, (2) and at least four concentrations of selenium. (3) The highest tested concentration of selenium caused >50 percent reduction relative to the control treatment, and (4) at least one tested concentration of selenium caused <20 percent reduction relative to the control treatment to ensure that the EC_{20} was bracketed by tested concentrations of selenium. When the response was expressed as percentages (e.g., percent survival), transformed values (arcsin of the square root) were used to homogenize the variance.

Logistic regression assumes that a logistic model describes the log dose-response curve. For a visual display of such model, a logistic curve with three parameters was fitted to each data set using nonlinear least-squares regression analysis (Draper and Smith 1981). The logistic model was

$$y = \frac{y_0}{1 + ax^b}$$

where x symbolizes the selenium concentration in the organism's tissues, y is the response of interest (survival, growth, or reproduction), and y_0, a and b are model parameters estimated by the regression analysis. The y_0 parameter represents the response of interest at background levels of selenium. The graphs also include the 95 percent confidence interval for projections of the logistic model. These tasks were performed in S-Plus version 6.0 (Insightful 2001).

When the data from an acceptable chronic test met the conditions for of the logistic regression analysis, the EC_{20} was the preferred chronic value. When data did not meet the conditions, best scientific judgment was used to determine the chronic value. In this case the chronic value is the geometric mean of the NOAEC and LOAEC and termed the maximum allowable toxicant concentration (MATC). But when no treatment concentration was an NOAEC, the chronic value is less than the lowest tested concentration. And when no treatment concentration was a LOAEC, the chronic value is greater than the highest tested concentration.

Evaluation of Freshwater Chronic Data for Each Species

Acceptable freshwater chronic toxicity data are currently available for an aquatic invertebrate (*Brachionus calyciflorous*), eight different fish species, and a mix of fish species from the family Centrarchidae in a total of 21 distinct studies (Table 4). Detailed summaries of each study are included in Appendix I. Collectively, only these data were considered for the derivation of a final tissue residue criterion for selenium. Below is a brief synopsis of the experimental design, test duration, relevant test

endpoints, and other critical information regarding the derivation of each specific chronic value. The chronic toxicity values for other chronic selenium toxicity values and endpoints are included in Appendix I.

Brachionus calyciflorus (freshwater rotifer)

This study reported by Dobbs et al. (1996) is one of two laboratory-based experiments (also see Bennett et al. 1986) that involved exposing algae to selenium (in this case as sodium selenate) in water, and subsequently feeding the algae to rotifers which were in turn fed to fish (fathead minnows). In this particular study, the rotifers and fish were exposed to the same concentrations of sodium selenate in the water as the algae, but received additional selenium from their diet (i.e., the algae fed to rotifers and the rotifers fed to fish). The overall exposure lasted for 25 days. Rotifers did not grow well at concentrations exceeding 108.1 µg Se/L in water, and the population survived only 6 days at selenium concentrations equal to or greater than 202.4 µg Se/L in the water (40 µg Se/g dw in the algae). Regression analysis of untransformed growth data (dry weight) determined 4 day post-test initiation resulted in a calculated EC_{20} of 42.36 µg Se/g dw tissue (Table 4).

Oncorhynchus tshawytscha (chinook salmon)

Hamilton et al. (1990) conducted a 90-day growth and survival study with swim-up larvae fed one of two different diets. The first diet consisted of Oregon moist pellets where over half of the salmon meal was replaced with meal from selenium-laden mosquitofish (*Gambusia affinis*) collected from the San Luis Drain, CA (SLD diet). The second diet was prepared by replacing half the salmon meal in the Oregon moist pellets with meal from low-selenium mosquitofish (i.e., the same relatively uncontaminated mosquitofish that were used in the control diet) and spiked with seleno-DL-methionine (SeMe diet). Analysis of the trace element composition in the two different diets indicated that while selenium was the most toxic element in the SLD diet, concentrations of boron, chromium, iron and strontium in the high-selenium mosquitofish replacement diet (SLD diet type) were slightly elevated compared to the replacement diet composed of uncontaminated control mosquitofish that were spiked with organic selenium (SeMe diet type). These trace elements were, however, only 1.2 (e.g., iron) to 2.0 times (e.g., chromium) higher in the SLD diet than the SeMe diet, which contained the following measured concentrations (dry weight basis) in the food: boron- 10 µg/g; chromium- 2.8 µg/g, iron- 776 µg/g, and strontium- 48.9 µg/g.

During the test, the survival of control chinook salmon larvae and larvae fed the lowest dietary selenium concentrations in either dietary exposure type (SLD and SeMe, respectively, consuming food at approximately 3 µg Se/g dw) exceeded ≥97 percent up to 60 days post-test initiation. Between 60 and 90 days of exposure, however, the control survival declined significantly. Therefore, only data collected up to 60 days post-test initiation was considered for analysis. Regression analysis of untransformed growth data after 60 days of exposure resulted in a calculated EC_{20} of 15.74 µg Se/g dw tissue for fish fed the SLD diet type, and 10.47 µg Se/g dw tissue for fish fed the SeMe diet type (Table 4). Note: The mosquitofish from San Luis Drain were not tested for contaminants other than certain key elements suspected to be present in these fish. The San Luis Drain receives irrigation drainage from the greater San Joaquin Valley; and therefore, there is the possibility that the mosquitofish used in this study may have contained elevated levels of pesticides. The use of the SLD diet results assumes that selenium, and not these other possible contaminants, was the cause of any adverse chronic effects.

Oncorhynchus mykiss (rainbow trout)

Hilton and Hodson (1983) reared juvenile rainbow trout on either a high (25 percent) or low (11 percent) available carbohydrate diet supplemented with sodium selenite for 16 weeks. Body weights, feed:gain ratios, and total mortalities were followed throughout the exposure every 28 days. Tissues (livers and kidneys) were extracted for selenium analysis after 16 weeks. Fish fed the diets (low carbohydrate and high carbohydrate) with the highest selenium concentration (11.4 and 11.8 µg Se/g dw food, respectively) exhibited a 45 to 48 percent reduction in body weight (expressed as kg per 100 fish) compared to control fish by the end of the exposure, which the authors attributed to food avoidance. With only two dietary exposure concentrations and a control, these data were not amenable to regression analysis. The MATC for growth of juvenile rainbow trout relative to the final concentrations of selenium in liver tissue of trout reared on the high carbohydrate seleniferous dietary type is the geometric mean (GM) of 21.0 µg Se/g dw (NOAEC) and 71.7 µg Se/g dw (LOAEC), or 38.80 µg Se/g dw. Using the equation III to convert the selenium concentration in liver tissue to a concentration of selenium in the whole-body, the MATC becomes 11.65 µg Se/g dw (Table 4). The calculated MATC for the same group of experimental fish exposed to selenium in the low carbohydrate diet becomes 13.08 µg Se/g dw tissue, which is the same MATC for trout exposed for an additional 4 weeks based on the occurrence of nephrocalcinosis in kidneys (see Hicks et al. 1984; Appendix I).

Hilton et al. (1980) employed a similar test design as Hilton and Hodson (1983) in a later experiment to examine the narrow window at which selenium changes from an essential nutrient to a toxicant affecting

juvenile rainbow trout. The food consisted of a casein-torula yeast diet supplemented with selenium as sodium selenite. The experiment lasted for 20 weeks. During this time, the trout were fed to satiation 3 to 4 times per day, 6 days per week, with one feeding on the seventh day. Organs (liver and kidney) and carcasses were analyzed for selenium from fish sacrificed at 4 and 16 weeks. No gross histopathological or physiological effects were detected in the fish, although trout raised on the highest dietary level of selenium (13.06 µg Se/g dw) had a significantly lower body weight (wet basis), a higher feed:gain ratio, and higher number of mortalities (10.7; expressed as number per 10,000 fish days). The MATC for growth and survival of juvenile rainbow trout relative to the final concentrations of selenium in liver tissue is the GM of the NOAEC (40 µg Se/g dw tissue) and the LOAEC (100 µg Se/g dw tissue), or 63.25 µg Se/g dw. Using equation III to convert selenium concentrations in the liver to selenium concentrations in the whole body, the MATC becomes 19.16 µg/g dw (Table 4).

Eggs and milt were obtained from ripe rainbow trout collected from reference streams and streams containing elevated selenium from an active coal mine in Alberta, Canada (Holm 2002; Holm et al. 2003). Eggs were fertilized and monitored in the laboratory until swim-up stage for percent fertilization, deformities (craniofacial, finfold, and spinal malformations), edema, and mortality. Similar investigations were conducted in 2000 and in 2001. The effort in 2001 added a stream with an intermediate level of selenium contamination and another reference stream. The only other notable difference between 2000 and 2001 was the temperature at which the embryos were incubated; 8°C in 2000 and 5°C in 2001. The author stated 5°C more closely approximated actual incubation temperatures for rainbow trout eggs. No differences were observed for percent fertilization or mortality between the reference and contaminated sites in both the 2000 and 2001 investigations. The frequencies of embryonic deformities and edema were significantly greater in the stream affected by coal mining than in the reference stream in the 2000 study. The average frequencies of embryonic craniofacial, skeletal and finfold deformities in the contaminated stream were 7.7, 13.8 and 3.2 percent, respectively; the average frequency of edematous embryos was 30.8 percent. The effect level for selenium was determined to be the average selenium concentration in rainbow trout muscle tissue, 1.50 µg Se/g ww. Muscle ww was converted to dw using 75.84 percent moisture derived for rainbow trout and equation 1 was used to convert selenium muscle dw to selenium in whole body dw. The chronic value determined for embryonic abnormalities in rainbow trout (2000 study) was 5.79 µg Se/g adult whole body dw. A comparison of the frequency of embryonic deformities or edema between selenium contaminated and reference streams with the 2001 data indicated there were no significant differences. An EC_{20} value, however, was computed for the relationship between craniofacial deformities and the concentration of

selenium in eggs, 10.4 µg Se/g eggs ww. Quantile regression was used to convert selenium in egg ww to muscle ww using the rainbow data reported by Holm et al. (2003). The remaining conversion to the whole body dw value of 5.85 µg Se/g was made using 75.84 percent moisture and equation 1. See Appendix I for details on these studies.

Oncorhynchus clarki (cutthroat trout)

No significant effects of bioaccumulated selenium on mortalities and deformities in the eggs, larvae, and fry from wild-caught cutthroat trout from a reference and exposed site (Fording River, British Columbia, Canada) were observed by Kennedy et al. (2000). The observations were made on eggs reared in well water from spawning age females collected from the two locations (N = 17 and 20, respectively) and fertilized by one male collected at each site. The mean selenium content in muscle tissue from adult fish was 2.4 µg/g dw tissue for fish collected from the reference site, and 12.5 µg/g dw tissue for fish collected from the Fording River. Using Equation I to convert the selenium concentration in muscle tissue to a selenium concentration in the whole-body, the chronic value for this study was estimated to be >10.92 µg/g dw parental fish tissue (see Table 4).

Hardy (2002) fed cutthroat trout experimental diets containing a range of selenomethionine (0-10 µg/g dw) for 124 weeks. No significant growth or survival effects were observed in the adult fish over the 124 weeks which reached a whole body concentration of 12.5 µg/g dw selenium after 44 weeks. Embryo-larval observations (percent hatch and percent deformed) were not related to whole body selenium concentrations in the spawning females (9.37 µg/g dw) fed the selenium-laden diet for 124 weeks. The chronic value for this study was determined to be >9.37 µg Se/g dw.

Salvelinus fontinalis (brook trout)

Spawning brook trout were collected from streams with elevated selenium contaminated by coal mining activity and from reference streams in 2000 and again in 2001 (Holm 2002; Holm et al. 2003) . Similar to that described for rainbow trout above, fertilized eggs were monitored in the laboratory for percent fertilization, deformities (craniofacial, finfold, and spinal malformations), edema, and mortality. The only abnormality observed in the embryos spawned from the brook trout collected in 2000 at the contaminated stream that had a frequency greater than the reference stream was craniofacial deformity (13.6 percent for the contaminated stream compared to 3.0 percent in the reference stream). The effect level for craniofacial deformity in brook trout for the 2000 data was determined to be the average selenium concentration in adult muscle tissue, 3.79 µg Se/g ww or 13.2 µg Se/g whole body dw using

conversion factors (75.84 percent moisture and equation 1). The only significant difference observed in 2001 brook trout was a greater frequency of finfold deformities in brook trout collected from Gregg Creek (intermediate selenium levels) relative to the reference stream (4.1 percent in Gregg Creek compared to 0.1 percent in the reference stream). The effect level for finfold deformites in the 2001 study was estimated to be the concentration of selenium in brook trout eggs from Gregg Creek, 6.88 µg Se/g ww. Using the same conversion factors used for rainbow trout in the Holm study described above, the chronic value in adult whole body dw is 12.4 µg Se/g. See Appendix I for more details.

Salmonidae summary

Four of the studies with salmonids discussed above evaluated the effects of selenium directly on growth of juvenile fish (Hamilton et al. 1990; Hilton and Hodson 1983; Hilton et al. 1980; Hicks et al. 1984), while three of the studies evaluated the effects of selenium on embryo/larval survival and deformity where exposure was through the parents (Hardy et al. 2002; Holm 2000; Holm et al. 2003; Kennedy et al. 2000). Of the studies based on embryo/larval survival and deformity where exposure was through the parents, fry from hatchery brood fish were fed a selenium-spiked diet, grown to sexual maturity, and spawned for the effects determination in the Hardy et al. study, and wild-caught adults from selenium contaminated streams were spawned for the effects determination in the Holm studies and in the Kennedy et al. study. Significant effects due to selenium exposure in these field exposed studies were not observed for cutthroat trout (Hardy et al. 2002; Kennedy et al. 2000). Significant effects were observed for rainbow trout and brook trout, albeit relatively minor effects in the latter species (Holm 2002; Holm et al. 2003). Although significant effects were not observed in the Hardy et al. and Kennedy et al. studies, the data are meaningful with respect to the effect levels obtained for embryo-larval development in *Oncorhynchus*, and thus retained for GMCV (10.66 µg Se/g dw) calculation (Table 4).

Pimephales promelas (fathead minnows)

Chronic values for fathead minnows were derived from three laboratory-based studies and one mesocosm study (Table 4). Two of the laboratory studies (Bennett et al. 1986 and Dobbs et al. 1996) involved exposing algae to selenium (either as sodium selenite or sodium selenate) in water, and subsequently feeding the algae to rotifers which were in turn fed to fathead minnows. In the Bennett et al. (1986) study, larval fathead minnows were fed control (cultured in chambers without selenium containing algae) or selenium-contaminated rotifers (cultured in chambers with selenium containing algae previously exposed to sodium selenite in the water) in three separate experiments lasting 9 to 30 days. The different experiments were distinguished by: 1) the day selenium-laden rotifers were first fed, 2) the day selenium-

laden rotifers were last fed, and 3) the age of larvae at experiment termination. The results from the three experiments reported by Bennett et al. (1986) were conflicting. Larval growth was significantly reduced at whole-body selenium concentrations ranging from 43.0 to 51.7 µg/g dw tissue in the first two experiments (see Appendix I for conditions), but growth was not significantly reduced in larvae that had accumulated 61.1 µg/g dw tissue in the third experiment (Table 4). The geometric mean of these three values, 51.40 µg/g dw, was considered the chronic value for selenium for this test.

A similar test system was used by Dobbs et al. (1996), in which larval fathead minnows were exposed to the same concentrations of sodium selenate in the water as their prey (rotifers), but also received additional selenium from the consumption of the selenium-contaminated rotifers. In this study, the fathead minnows did not grow well at concentrations exceeding 108.1 µg Se/L in water, and they survived only to 11 days at selenium concentrations equal to or greater than 393.0 µg/L in the water (75 µg Se/g dw in the diet, i.e., rotifers). The LOAEC for retarded growth (larval fish dry weight) in this study was <73 µg Se/g dw tissue (Table 4).

In contrast to the above laboratory-based food chain studies, Ogle and Knight (1989) examined the chronic effects of only elevated foodborne selenium on growth and reproduction of fathead minnows. Juvenile fathead minnows were fed a purified diet mix spiked with inorganic and organic selenium in the following percentages: 25 percent selenate, 50 percent selenite, and 25 percent seleno-L-methionine. The pre-spawning exposure lasted 105 days using progeny of adult fathead minnows originally obtained from the Columbia National Fishery Research Laboratory, and those obtained from a commercial fish supplier. After the 105 day exposure period, a single male and female pair from each of the respective treatment replicates were isolated and inspected for spawning activity for 30 days following the first spawning event of that pair. There was no effect from selenium on any of the reproductive parameters measured, including larval survival, at the dietary concentrations tested (5.2 to 29.5 µg Se/g dw food). Sub-samples of larvae from each brood were maintained for 14 days post-hatch and exhibited >87.4 percent survival. The pre-spawning adult fish fed a mean dietary level of 20.3 µg Se/g dw did exhibit a significant reduction in growth compared to controls (16 percent reduction), whereas no effect on growth occurred in the fish fed 15.2 µg Se/g dw. The whole-body chronic value, as determined by the GM of the NOAEC and the LOAEC measured at 98 days post-test initiation, was 5.961 µg/g dw tissue (Table 4).

The chronic value of 5.961 µg/g dw determined for growth after 98 days of exposure to pre-spawning fathead minnow adults (Ogle and Knight, 1989) was approximately an order of magnitude lower than the

growth effects to fathead minnow observed in Bennett et al. (1986) and Dobbs et al (1996). The length of exposure in the Ogle and Knight test was more than twice as long as either Bennett et al. or Dobbs et al., suggesting a longer duration was needed in order to detect any growth effects from selenium. However, in addition to the absence of effects observed for the reproductive parameters measured, survival of larvae hatched from parents exposed to each of the five selenium treatments (including those in which growth was affected) was not affected.

Other studies (Bryson et al. 1984; Bryson et al. 1985a; Coyle et al. 1993; Hermanutz et al. 1996) have found larval deformities and larval survival to be the most sensitive endpoint to fish. This also appears true for fathead minnows. Schultz and Hermanutz (1990) examined the effects of selenium in fathead minnow larvae transferred from parental fish (females). The parental fathead minnows were originally exposed to selenite which was added to artificial streams in a mesocosm study. The selenite entered the food web which contributed to exposure from the diet. Spawning platforms were submerged into treated and control streams. The embryo samples that were collected from the streams were brought into the laboratory and reared in incubation cups which received stream water dosed with sodium selenite via a proportional diluter. Edema and lordosis were observed in approximately 25 percent of the larvae spawned and reared in natural water containing 10 µg Se/L. Selenium residues in the ovaries of females from the treated stream averaged 39.27 µg/g dw. Using equation II to convert the selenium concentration in the ovaries to a concentration of selenium in the whole-body, the chronic value for this species was estimated to be <18.21 µg Se/g dw (Table 4).

Since Ogle and Knight reported that food in the higher selenium concentrations remained uneaten and fish were observed to reject the food containing the higher selenium concentrations, the authors suggested that the decreased growth was caused by a reduced palatability of the seleniferous food items. This is a common observation also noted by Hilton and Hodson (1983) and Hilton et al. (1980) and apparent in Coughlan and Velte (1989). Given the no observed effect to larval survival and the apparent non-toxicological effect on growth in the Ogle and Knight study, the SMCV for fathead minnows does not include the 5.961 µg/g dw chronic value.

Also excluded from the SMCV calculation for fathead minnows were the chronic value and LOAEC estimated from the laboratory food-chain experiments of Bennett et al. (1986) and Dobbs et al. (1996). In both of these studies, the effect concentrations based on larval growth appear to be less sensitive than the effect on larval edema and deformity observed in Schultz and Hermanutz (1990). The greater

sensitivity of larval fathead minnows to selenium as measured by edema and deformity (lordosis) in the Schultz and Hermanutz (1990) study is consistent with other studies using bluegill (Table 4); and thus, the SMCV for fathead minnows of <18.21 µg/g dw was based on this endpoint.

Catostomus latipinnis (flannelmouth sucker)

Beyers and Sodergren (2001a) exposed flannelmouth sucker larvae to a range of aqueous selenate concentrations (<1, 25.4, 50.6, 98.9, and 190.6 µg/L) and respectively fed them a range of selenium in their diet (rotifers containing <0.702, 1.35, 2.02, 4.63, and 8.24 µg/g dw). There were no survival or growth effects observed after the 28 day exposure. The chronic value based on the concentration of selenium measured in the larvae exposed to the highest test concentration was >10.2 µg Se/g dw.

Xyrauchen texanus (razorback sucker)

Two laboratory exposure studies have been done with the endangered razorback sucker. In the first study, Beyers and Sodergren (2001a) exposed larval razorback suckers to the same aqueous and diet concentrations as described above for the flannelmouth sucker. Similar to the results found for the flannelmouth sucker, survival and growth of the razorback sucker larvae were not reduced after the 28 day exposure. The chronic value for this study based on selenium measured in the larvae at the end of the test is > 12.9 µg Se/g dw. In a second study, Beyers and Sodergren (2001b) exposed larval razorback suckers to a control water and three different site waters containing varying concentrations of selenium. Two treatments were tested within each water type, fish fed rotifers cultured in the same water type (site diet) and fish fed rotifers cultured in control water. There were no reductions in survival or growth in fish exposed to both the site water and site diet compared to fish exposed to control water and control diet. There were, however, reductions in growth in fish exposed to site water/site food compared to the same site water and control food. The authors did not attribute the effect on larval growth by the diet to selenium and cited several lines of evidence, including: (1) there was not a dose-response relationship in the concentration of selenium in the food (rotifers) and growth, nor in the concentration of selenium in the fish larvae and growth across the three water types; and (2) the site water type, identified as De Beque, showed a significant reduction in the growth of fish exposed to site water/site food relative to site water/control food, but contained levels of selenium in the water (< 1µg/L) and food (2.10 µg/g dw) typically lower than those that have been found to elicit effects. The chronic value for this study is > 42 µg Se/g dw based on the whole body concentration of selenium in the larval razorback suckers exposed to North Pond site water.

Lepomis macrochirus (bluegill sunfish)

Applicable chronic data for bluegill sunfish can be grouped according to field exposure versus laboratory exposure. In some field studies, chronic tolerance to selenium appears to be much higher than in laboratory studies (Bryson et al. 1985a). In the Bryson et al. (1984, 1985a) and Gillespie and Baumann (1986) studies, the progeny of females collected from a selenium contaminated reservoir, Hyco Reservoir, Person County, NC and artificially crossed did not survive to swim-up stage, irrespective of the origin of milt used for fertilization. Measured waterborne selenium concentrations prior to the experiments ranged from 35 to 80 µg/L. The whole-body tissue selenium concentration in the female parent associated with this high occurrence of mortality of hatched larvae was <43.70 µg/g dw tissue, as reported by Bryson et al. (1985a), and <21.47 µg/g dw tissue, as reported by Gillespie and Baumann (1986) (Table 4). In the case of the latter, nearly all swim-up larvae from the Hyco Reservoir females were edematous, none of which survived to swim-up. These chronic effect tissue values are in line with the EC_{20} calculated for the occurrence of deformities among juvenile and adult fishes from the family Centrarchidae collected from Belews Lake, NC, i.e., 44.57 µg Se/g dw (see Lemly 1993b, Table 4).

Bryson et al. (1985b) conducted juvenile survival toxicity tests using hatchery bluegill and various forms of selenium spiked to an artificial diet as well as a diet consisting of zooplankton collected from Hyco Reservoir. There was no effect on length or weight of the juvenile bluegill after 60 days of exposure. The highest concentration of selenium measured in whole body fish tissues in these tests was in the seleno-DL-cysteine-2X treatment (3.74 µg Se/g dw). Bryson et al. (1985b) also examined percent hatch and percent swim-up larvae from spawns using fish collected from Hyco Reservoir and a control site. There were no differences in the Hyco measurements relative to the control. The concentration of selenium in the liver of the parental Hyco bluegill was 18.6 µg/g dw or 5.45 µg Se/g dw whole body using equation III for conversion. The chronic values for the juvenile bluegill test and the embryo-larval development tests were >3.74 and >5.45 µg Se/g dw whole body, respectively.

In contrast, the chronic effects threshold for larval survival in a combination laboratory waterborne and dietary selenium exposure (Coyle et al. 1993), or even a long-term mesocosm exposure (Hermanutz et al. 1996), occurs at concentrations approximately 3 times lower than those recorded above (Table 4). In the Coyle et al. (1993) study, two-year old pond reared bluegill sunfish were exposed in the laboratory to a nominal 10 µg Se/L in water (measured concentrations in respective dietary treatments ranging from 8.4 to 11 µg/L) and fed (twice daily *ad libitum*) Oregon moist pellets containing increasing concentrations of seleno-L-methionine. The fish were grown under these test conditions for 140 days. Spawning

frequency, fecundity, and percentage hatch were monitored after 60 days when spawning began to occur. There was no effect of the combination of the highest dietary selenium concentration (33.3 µg Se/g dw) in conjunction with waterborne selenium concentrations averaging 11 µg/L on adult growth, condition factor, gonadal somatic index, or the various reproductive endpoints (Appendix I). The survival of newly hatched larvae, however, was markedly reduced; only about 7 percent survived to 5 days post-hatch. Regression analysis on arcsin square root transformed fry survival data 5 days post-hatch resulted in a calculated EC_{20} of 8.954 µg Se/g dw tissue (Table 4).

Hermanutz et al. (1996), as corrected by Tao et al. (1999), and peer reviewed in Versar (2000), exposed bluegill sunfish to sodium selenite spiked into artificial streams (nominal test concentrations: 0, 2.5, 10, and 30 µg Se/L) which entered the food web, thus providing a simulated field-type exposure (waterborne and dietary selenium exposure). A series of three studies were conducted over a 3 year period lasting anywhere from 8 to 11 months. All three studies began exposure to adult bluegill sunfish in the fall and ended the respective study in the summer of the following year. Winter temperatures averaged 4.1 and 4.5°C and spawning months (June-July) averaged 23.9 and 22.4°C, respectively for Studies II and III. The Hermanutz et al. (1996) report contains the data presented in the Hermanutz et al. (1992) article (Study I, 10 and 30 µg/L exposures) as well as Studies II and III (2.5 and 10 µg/L and recovering mesocosms). Spawning activity was monitored in the stream, and embryo and larval observations were made *in situ* and from fertilized eggs taken from the streams and incubated in egg cups in the laboratory. None of the adult bluegill exposed to the highest concentration of selenium in the water (Study I, mean measured concentration equal to 29.4 µg/L) survived. Incidence of edema, hemorrhage, and lordosis in the larvae incubated in egg cups and spawned from fish exposed to 10 µg Se/L were 100, 45 and 15 percent, respectively (see Hermanutz 1996 in Appendix I). Such health problems were not observed in larvae from fish that were not exposed to elevated concentrations of selenium (control treatment). Rates of edema, hemorrhage, and lordosis occurrence in larvae (egg cup data) from fish exposed to 2.5 µg Se/L were 0, 3.6 and 0 percent, respectively. Mean concentrations of selenium in fish tissues (whole body) of the control, 2.5 and 10 µg Se/L treatments were 1.95, 5.55, and 26.46 µg Se/g dw, respectively. Except for the 2.5 µg Se/L treatment, each value is a geometric mean of 2 replicates.

Results of this experiment were not suitable for regression analysis. Exposure of adult fish to 10 µg Se/L caused a small reduction in larval survival (in their first three days), from 75 to 57 percent. However, responses lower than half of the values observed in control treatments are needed to adequately characterize the slope of decline in survival (or growth, reproduction...) with increasing concentrations of

selenium. It is not sufficient to have only extremely low and high responses. Intermediate effects are necessary to properly estimate the shape of the dose-response curve. The percent of larvae with edema increased from 0 percent at the control and 2.5 µg Se/L treatments to 100 percent in streams that received 10 µg Se/L. With these data, it is not possible to accurately estimate the lowest concentration with adverse effects (LOAEC) nor the rate at which incidence of edema increases with higher tissue concentrations of selenium.

The chronic value for this study was estimated from results of analysis of variance (ANOVA) reported by Tao et al. (1999). ANOVA was utilized to evaluate effects of elevated concentrations of selenium on percent hatch, percent survival, maximum percent edema, lordosis, and hemorrhage, and minimum percent healthy (egg cup data). Treatment effects were only significant for maximum percent edema and minimum percent healthy (see their Table 4-19), and in no instance were differences between the 2.5 µg Se/L and control treatments significant (Dunnett's Means test, all probabilities > 0.1, see their Table 4-20). These results clearly suggest that the 2.5 µg Se/L treatment had no adverse impact on bluegill larvae. They are further supported by analysis of the field nest data (see Hermanutz 1996 in Appendix I). In this experiment, treatment had a significant effect on maximum percent edema (raw data and ranks) and maximum percent hemorrhage (ranks only). Probabilities of differences between the 2.5 µg Se/L and control treatments were >0.2 for all response variables except maximum percent hemorrhage, which had an estimated probability of 0.05 (raw data, P=0.022 for ranks; Dunnett's means test). Such values, though, were well above the adjusted experiment-wise error rate for multiple comparisons (α'=0.0085, $\alpha'=1-(1-\alpha)^{1/k}$; α=0.05, k=6 comparisons; Sokal and Rohlf 1981), which takes into account the fact that selenium effects were tested on six different response variables. Therefore, the chronic value for this study, 12.12 µg Se/g dry weight, was calculated as the geometric mean of tissue concentrations of selenium in the 2.5 (NOAEC) and 10 µg Se/L (LOAEC) treatments (5.55 and 26.46 µg/g dw, respectively).

The importance of diet in the bioaccumulation of selenium was demonstrated in one additional experiment. Study III consisted of the addition of new adult bluegill to the same streams that received the 2.5, 10 and 30 µg/L sodium selenite during previous studies, but with all dosing of selenite halted. The adult bluegills exposed only to dietary selenium present in the food web accumulated selenium to levels very near to the levels accumulated during Study II in which aqueous selenium was also present demonstrating the importance of diet on selenium accumulation. There were no effects (no effect on larval survival, 0 percent deformities, 0 percent hemorrhaging), on the bluegill progeny in Study III even

from fish that accumulated 11.7 and 14.5 µg/g dw in the recovering 10 µg/L streams, and 17.35 µg/g dw in the recovering 30 µg/L stream. The lack of any effect on the Study III larvae suggests that although dietary exposure would have been the predominant exposure route in both Study II and Study III, environmental differences influenced the toxicological significance of the tissue concentrations.

A 90-day diet-only laboratory exposure in which juvenile bluegill sunfish were fed a range of selenomethionine concentrations added to Oregon moist did not have any significant effects on survival (Cleveland et al. 1993). The authors did report a significant decrease in the condition factor (K) at the diet treatment where bluegill whole body tissue concentrations were measured at 7.7 µg Se/g dw. The condition factor (weight x 10^5/length^3) is reflective of the weight of the fish, and as discussed earlier, the avoidance of food at similar dietary concentrations in other fish studies (Ogle and Knight 1989; Hilton and Hodson 1983; Hilton et al. 1989; Coughlan and Velte 1989) suggests the reduction in K is possibly a non-toxicological effect. Given the very slight reduction in K (1.3 to 1.2) and the uncertain relevance of growth data, the chronic value for this study was estimated at > 13.4 µg Se/g dw.

Data from Lemly (1993a) indicate that over-wintering fish may be more susceptible to the effects of waterborne and dietary selenium due to increased sensitivity at low temperature. The authors exposed juvenile bluegill sunfish in the laboratory to waterborne (1:1 selenite:selenate; nominal 5 µg Se/L) and foodborne (seleno-L-methionine in TetraMin; nominal 5 µg Se/g dw food) selenium for 180 days. Tests with a control and treated fish were run at 4^{O}C and 20^{O}C with biological and selenium measurements made every 60 days. Survival and whole-body lipid content were unaffected at 20^{O}C (whole-body selenium concentrations equal to 6 µg/g dw) when compared to control fish. Fish exposed to the combination low-level waterborne and dietary selenium at 4^{O}C exhibited significantly elevated mortality (40.4 percent) relative to controls (2.9 percent), and exhibited significantly greater oxygen consumption and reduced lipid content, which are all indicative of an additional stress load. The chronic value for juvenile bluegill sunfish exposed to waterborne and dietary selenium at 4^{O}C was <7.9 µg/g dw tissue, whereas the chronic value for juvenile bluegill sunfish exposed to waterborne and dietary selenium based on survival at 20^{O}C was >6 µg/g dw whole-body tissue.

Five of the studies discussed above evaluated the effects of selenium on fish larvae to which exposure was through the parents. Three of these studies collected adult fish from Hyco Reservoir to which the bluegill population had been exposed to elevated selenium concentrations for multiple generations (Bryson et al. 1984; Bryson et al. 1985a; Gillespie and Baumann 1986), whereas the other two studies

exposed bluegill parents obtained from an uncontaminated source (Coyle et al. 1993; Hermanutz et al. 1996). The average of the chronic values reported for the Hyco studies was four times greater than the value in the latter two studies. This difference may simply be the inability of the field tests to detect a lower effect concentration than that which was observed at the site. However, Bryson et al. (1985a) found no effects to larval survival from Hyco Reservoir females collected in an "unaffected area" containing 19.18 µg Se/g dw suggesting the possibility of tolerance through physiological or genetic adaptation of the previous exposed bluegill population at Hyco Reservoir.

Acquisition of tolerance to selenium has also been implied in the literature for other fish species. For example, Kennedy et al. (2000) suggested tolerance at the cellular level as an explanation for the normal development of early life stages for cutthroat trout collected from a stream containing 13.3 to 14.5 µg Se/L in the water column. These authors reported that the overall frequency of larval deformities in the exposed population was less than 1 percent, and in one fish containing eggs with 81.3 µg Se/g dw, there were 0.04 percent pre-ponding deformities and 3.3 percent larval mortalities. It should be noted that the acquisition of tolerance to selenium has been hypothesized (Kennedy et al. 2000), but has not yet been substantiated. Other than the Kennedy et al. study, tolerance to selenium in one of the endpoints consistently sensitive to fish (embryo-larval development) has not been reported in the literature and its reality is uncertain at this time. However, given the need to protect sensitive populations of species, the chronic values for the studies in which eggs and larvae were obtained from bluegill adults that were exposed to elevated selenium for multiple generations (i.e., Bryson et al. 1984; Bryson et al. 1985a; Gillespie and Baumann, 1986) were not included in the SMCV calculation.

Morone saxitilis (Striped bass)

The only remaining applicable chronic value for selenium was determined from a laboratory dietary exposure conducted using yearling striped bass (Coughlan and Velte 1989). During the experiment, the bass were fed contaminated red shiners (38.6 µg Se/g dw tissue) from Belews Lake, NC (treated fish) or golden shiners with low levels of selenium (1.3 µg/g dw tissue) purchased from a commercial supplier (control fish). The test was conducted in soft well water and lasted up to 80 days. During the experiment, all fish were fed to satiation 3 times per day. Control fish grew well and behaved normally. Treated fish behaved lethargically, grew poorly due to a significant reduction in appetite, and showed histological damage, all eventually leading to the death of the animal. The final selenium concentration in muscle of treated striped bass averaged from 17.50 to 20.00 µg/g dw tissue (assuming 80 percent moisture content), which was 3.2 to 3.6 times higher than the final selenium concentrations in control

striped bass, which averaged 5.500 µg/g dw tissue. Using equation I to convert the selenium concentration in muscle tissue to a selenium concentration in the whole-body, the chronic value for this species was determined to be <14.75 µg/g dw (Table 4).

Formulation of the Final Chronic Value (FCV) for Selenium

The lowest GMCV in Table 4 is for bluegill, 9.500 µg/g dw whole body, which is the geometric mean of chronic values from the laboratory study of Coyle et al. (1993), the laboratory study of Lemly (1993a) and the macrocosm exposure study of Hermanutz et al. (1996). Several of the chronic values listed in Table 4 were not used in the calculation of this GMCV. These values fall under several categories. The "less than" values tabulated for Bryson et al. (1984) and Gillespie and Baumann (1986) for Hyco Reservoir bluegill were not used to because they only indicate a chronic value in a range that includes 9.500 µg/g dw. The "greater than" values for Bryson et al (1985b) were not used because similar studies with bluegill sunfish provided more meaningful information on effect levels. The "greater than" value for the recovering systems in Study III from Hermanutz et al. (1996) was not used in the mean calculation because, as previously discussed in the *Lepomis* section, less tolerance was observed in the freshly exposed systems of Study II. The Table 4 results for Bryson et al. (1985a) and Lemly (1993b) were also not used in calculating the bluegill GMCV. Bryson et al. (1985a) indicated a chronic value for Hyco Reservoir bluegill somewhere between 20.29 and 43.70 µg/g dw. Lemly (1993b), appearing in Table 4 under the category Centrarchidae, the family to which bluegill belong, yielded a chronic EC_{20} of 44.57 µg/g dw specific for fish from Belews Lake, NC, again substantially above the GMCV of 9.500 µg/g dw. It is not known whether historical exposure to elevated selenium concentrations, such as occurred at Belews Lake and Hyco Reservoir, will dependably lead to this magnitude of increase in the chronic tolerance of resident fish.

The Lemly (1993a) laboratory results, indicating a chronic value for over-wintering juvenile bluegill sunfish of <7.91 µg/g dw, are not completely comparable to the other values used to calculate the bluegill GMCV. This study involved an additional natural stress, exposure to a simulated winter low temperature of 4°C. In this study, juvenile bluegill sunfish exposed to the over-wintering temperature 4°C appeared to accumulate more selenium in whole-body tissues (7-8 µg Se/g dw tissue) relative to those exposed at 20°C (5-6 µg Se/g dw tissue), but also exhibited increased signs of chronic toxicity. Because this stress occurs annually to one degree or another in nearly all the country, the FCV was lowered to 7.91 µg/g dw to protect sensitive fish species Although the literature contains little information on the temperature-dependence of selenium toxicity, Lemly's study (further summarized in Appendix I) was judged to be

sufficiently definitive to merit lowering the FCV. The study showed a clear effect on juvenile bluegill survivorship when tissue concentrations reach 7.91 µg Se/g dw under extended cold temperature conditions.

In the Lemly (1993a) study, the author relates the selenium induced hematological changes to gill lamellar damage (possible reasons cited were the collection of cell parts in capillaries restricting blood flow increasing pressure and rupturing or swelling lamellar vessels, and smaller red cells becoming tightly packed in vessels). The author postulates that an imbalance between respiratory demands (i.e., Se-exposed fish used more O_2 at both 4°C and 20°C) and decreased respiratory capacity could have constituted a stress that resulted in reduced body condition and lipid content of fish in the cold treatment. The condition of the combination of selenium-induced elevation in energy demand and reductions in feeding due to cold temperature and short photoperiod, leading to severe depletion of stored body lipid was termed, Winter Stress Syndrome.

The Guidelines indicate that the chronic criterion (in this case the FCV) is intended to be a good estimate of the threshold for unacceptable effect. The Guidelines point out that the threshold for unacceptable effect does not equate with a threshold for any adverse effect. For example, some adverse effects, possibly even a small reduction in survival, growth, or reproduction may occur at this threshold. If over-wintering bluegill are as sensitive as indicated by the Lemly (1993a) results, some reduction in survival (compared to populations accumulating lesser concentrations of selenium or exposed to less severe winter temperatures) would occur at the FCV. Nevertheless, other studies, those of Lemly (1993b) and Bryson et al. (1985a), suggest that historically exposed populations may not be as sensitive as the organisms studied by Lemly (1993a).

The bluegill exposed to selenium at 4°C in the Lemly (1993a) study accumulated 7.91 µg/g dw, whereas those exposed to Se at 20°C accumulated only 5.74 µg/g dw. The increase in the concentration of selenium in whole body tissue at 4°C was apparently due to reductions in lipid and body weight caused by decreased feeding by the juvenile bluegill resulting in a concentration of selenium in their tissues. If this concentration of selenium in tissues occurs in sensitive overwintering fish in nature, a criterion of 5.85 µg/g dw (the selenium tissue concentration in the 4°C exposure after 60 days) in for fish collected during the summer or fall months might be warranted to protect the selenium-sensitive fish during the winter months. However, it is not understood at this time whether fish in nature do concentrate selenium during the winter. The Lemly (1993a) study used an artificial diet spiked with seleno-methionine.

Although the 20°C fish did not show signs of food avoidance to the Se-spiked food, as discussed earlier in this section, other studies did observe decreased feeding and effects on growth.

If sensitive juvenile fish are indeed adversely affected during winter months, field studies should indicate an altered age structure relative to selenium whole body tissue levels. May et al. (2001) reported that an analysis of the size structure of bluegill populations in the Republican River and in 7 reservoirs within this river's basin, where mean tissue concentrations ranged from 2.85 to 8.84 mg Se/g dw, revealed large numbers of small fishes. Similar patterns in the size structure of fish populations were observed for 7 additional species: common carp, green sunfish, channel catfish, largemouth bass, gizzard shad, black bullhead, and river carpsucker.

Given the uncertainty of juvenile fish concentrating selenium over the winter, an FCV of 7.91 µg Se/g dw is recommended. However, if the concentration of selenium in whole body fish tissues approaches 5.85 µg Se/g dw during the summer or fall months, it is recommended fish be sampled during the winter to determine if they exceed the FCV of 7.91 µg Se/g dw.

The FCV may not necessarily protect fish in a hypothetical environment where they are exposed only via water and not via diet. If the organisms are provided with an uncontaminated diet, then exceedingly high water concentrations, possibly above the acute criterion, are needed to elicit effects, but such effects may occur at tissue concentrations below the FCV (Cleveland et al. 1993; Gissel-Nielsen and Gissel-Nielsen 1978). This is not a practical limitation, however, since water-only exposure of selenium is not representative of the actual exposure of selenium to aquatic organisms in the environment.

The FCV of 7.91 µg/g dw was based on a scientific interpretation of the data presented in Table 4. Although the FCV is derived from a limited number of species (9 species/7 genera), it is intended to be protective of aquatic organisms across the United States. There may be aquatic communities whose fish assemblage may contain species with different sensitivities to selenium compared to those listed in Table 4. Furthermore, even within the Table 4 bluegill data, there is a range of reported tissue NOAECs from various sites. Consequently, results from appropriate site-specific studies could be used to modify the criterion.

A comparison of the FCV to tissue values measured in U.S. Fish and Wildlife Service's National Contaminant Biomonitoring Program and U.S. Geological Survey's National Water Quality Assessment (NAWQA) program is provided in Appendix J.

Table 4. Freshwater Chronic Values from Acceptable Tests

Species	Reference	Exposure route	Selenium form	Toxicological endpoint	Chronic value, µg/g dw[a]	SMCV µg/g dw	GMCV µg/g dw
Brachionus calyciflorus rotifer	Dobbs et al. 1996	dietary and waterborne (lab)	algae exposed to SeVI in water, algae then fed to rotifers	EC_{20} for rotifer dry weight after 4 d	42.36	42.36	42.36
Oncorhynchus tshawytscha chinook salmon	Hamilton et al. 1990	dietary (lab)	Se-laden mosquitofish from San Luis Drain, CA	EC_{20} for juvenile growth	15.74 (juvenile tissue)	12.84	10.66
Oncorhynchus tshawytscha chinook salmon	Hamilton et al. 1990	dietary (lab)	Mosquitofish spiked with seleno-DL-methionine	EC_{20} for juvenile growth	10.47 (juvenile tissue)		
Oncorhynchus mykiss rainbow trout	Hilton and Hodson 1983: Hicks et al. 1984	dietary (lab)	sodium selenite in food preparation	MATC for juvenile growth; nephrocalcinosis	11.65[b] (juvenile tissue)	9.32	
Oncorhynchus mykiss rainbow trout	Hilton et al. 1980	dietary (lab)	sodium selenite in food preparation	MATC for juvenile survival and growth	19.16[b] (juvenile tissue)		
Oncorhynchus mykiss rainbow trout	Holm 2000; Holm et al. 2003	dietary and waterborne (field Luscar River, Alberta)	not determined	2000 study: chronic value for embryo larval deformities 2001 study: EC_{20} for craniofacial deformities	5.79[c] (parent tissue) 5.85[c] (parent tissue)		
Oncorhynchus clarki cutthroat trout	Kennedy et al. 2000	dietary and waterborne (field - Fording River, BC)	not determined	NOAEC for embryo/larval deformities and mortality	>10.92[c] (parent tissue)	>10.12	
Oncorhynchus clarki cutthroat trout	Hardy, R.W. 2002	dietary (lab)	selenomethionine in food preparation	NOAEC for embryo/larval deformities	>9.37 (parent tissue)		

Species	Reference	Exposure route	Selenium form	Toxicological endpoint	Chronic value, µg/g dw[a]	SMCV µg/g dw	GMCV µg/g dw
Salvelinus fontinalis brook trout	Holm 2002; Holm et al. 2003	dietary and waterborne (field Luscar River, Alberta)	not determined	2000 study: chronic value for craniofacial deformities 2001 study: chronic value for finfold deformities	13.2[c] (parent tissue) 12.4[c] (parent tissue)	12.8	12.8
Pimephales promelas fathead minnow	Bennett et al. 1986	dietary (lab)	algae exposed to selenite then fed to rotifers which were fed to fish	Chronic value for larval growth	51.40[d] (larval tissue)	<18.21	<18.21
Pimephales promelas fathead minnow	Ogle and Knight 1989	dietary (lab)	mix of 25, 50, and 25 percent selenate, selenite, and seleno-L-methionine in food preparation	MATC for pre-spawning adult growth	5.961[d] (pre-spawning adult tissue)		
Pimephales promelas fathead minnow	Dobbs et al. 1996	dietary and waterborne (lab)	algae exposed to selenate in water then fed to rotifers which were fed to fish	LOAEC for larval fish dry weight after 8 d	<73[d] (larval tissue)		
Pimephales promelas fathead minnow	Schultz and Hermanutz 1990	dietary and waterborne (mesocosm - Monticello)	selenite added to artificial streams which entered food web and provided dietary exposure	LOAEC for larval edema and lordosis	<18.21[e] (parent tissue)		
Catostomus latipinnis flannelmouth sucker	Beyers and Sodegren 2001a	dietary and waterborne (lab)	water: selenate; diet: algae exposed to selenate in water then fed to rotifers which were fed to fish	NOAEC for survival and growth	>10.2 (larval tissue)	>10.2	>10.2
Xyrauchen texanus razorback sucker	Beyers and Sodegren 2001a	dietary and waterborne (lab)	water: selenate; diet: algae exposed to selenate in water then fed to rotifers which were fed to fish	NOAEC for survival and growth	>12.9 (larval tissue)	>23.28	>23.28

Species	Reference	Exposure route	Selenium form	Toxicological endpoint	Chronic value, µg/g dw[a]	SMCV µg/g dw	GMCV µg/g dw
Xyrauchen texanus razorback sucker	Beyers and Sodegren 2001b	dietary and waterborne (lab)	water: site waters; diet: algae exposed to site water then fed to rotifers which were fed to fish	NOAEC for survival and growth	>42 (larval tissue)		
Lepomis macrochirus bluegill	Bryson et al. 1984	dietary and waterborne (field - Hyco Reservoir, NC)	not determined	LOAEC for larval mortality	<59.92[c,d] (parent tissue)	9.50	9.50
Lepomis macrochirus bluegill	Bryson et al. 1985a	dietary and waterborne (field - Hyco Reservoir, NC)	not determined	Chronic value for swim-up larvae	<43.70[c,d] >20.29[c,d] (parent tissue)		
Lepomis macrochirus bluegill	Bryson et al. 1985b	dietary and waterborne (field - Hyco Reservoir, NC)	not determined	NOAEC for swim-up larvae	>5.45[c,d] (parent tissue)		
Lepomis macrochirus bluegill	Gillespie and Baumann 1986	dietary and waterborne (field - Hyco Reservoir, NC)	not determined	Chronic value for larval survival	<28.20[d] (larval tissue); or <21.47[d,e] (parent tissue)		
Lepomis macrochirus bluegill	Coyle et al. 1993	dietary and waterborne (lab)	diet: seleno-L-methionine water: 6:1 selenate:selenite	EC_{20} for larval survival	8.954 (parent tissue - females only)		
Lepomis macrochirus bluegill	Lemly 1993a	dietary and waterborne (lab)	diet: seleno-L-methionine water: 1:1 selenate:selenite	LOAEC for juvenile mortality at 4°C	<7.91 (juvenile tissue)		
Lepomis macrochirus bluegill	Lemly 1993a	dietary and waterborne (lab)	diet: seleno-L-methionine water: 1:1 selenate:selenite	NOAEC for juvenile mortality at 20°C	>6.0[d] (juvenile tissue)		

Species	Reference	Exposure route	Selenium form	Toxicological endpoint	Chronic value, µg/g dw[a]	SMCV µg/g dw	GMCV µg/g dw
Lepomis macrochirus bluegill	Hermanutz et al. 1996	dietary and waterborne (mesocosm - Monticello)	selenite added to artificial streams which entered food web and provided dietary exposure	MATC for larval survival, edema, lordosis and hemorrhaging Study II	12.12 (parent tissue)		
Lepomis macrochirus bluegill	Bryson et al. 1985b	dietary	seleno-DL-cysteine	NOAEC for juvenile growth	>3.74[d] (juvenile tissue)		
Lepomis macrochirus bluegill	Cleveland et al. 1993	dietary	seleno-L-methionine	NOAEC for juvenile survival	>13.4[d] (juvenile tissue)		
Lepomis macrochirus bluegill	Hermanutz et al. 1996	dietary (mesocosm - Monticello)	selenite originally added to artificial streams which entered food web and provided dietary exposure	NOAEC for larval survival, edema, lordosis and hemorrhaging Study III	>17.35[d] (parent tissue)		
Centrarchidae (9 species)	Lemly 1993b	dietary and waterborne (field - Belews Lake, NC)	not determined	EC_{20} for deformities among juveniles and adults	44.57 (juvenile and adult tissue)	NA	NA
Morone saxitilis striped bass	Coughlan and Velte 1989	dietary (lab)	Se-laden shiners from Belews Lake, NC	LOAEC for survival of yearling bass	<14.75[c] (juvenile tissue)	<14.75	<14.75

[a] All chronic values reported in this table are based on the measured or estimated (see footnotes below) concentration of selenium in whole body tissue.
[b] Estimated using the equation III.
[c] Estimated using the equation I.
[d] Chronic value not used in SMCV calculation (see text).
[e] Estimated using the equation II.

National Criteria

The available data for selenium, evaluated in accordance with EPA's guidelines for deriving aquatic life criteria (Stephan et al. 1985) indicate that, except possibly where an unusually sensitive species is important at a site, freshwater aquatic life should be protected if the following conditions are satisfied.

A. The concentration of selenium in whole-body fish tissue does not exceed 7.91 µg/g dw (dry weight). This is the chronic exposure criterion. In addition, if whole-body fish tissue concentrations exceed 5.85 µg/g dw during summer or fall, fish tissue should be monitored during the winter to determine whether the selenium concentration exceeds 7.91 µg/g dw.

B. The 24-hour average concentration of total recoverable selenium in water seldom (e.g., not more than once in three years) exceeds 258 µg/L for selenite, and likewise seldom exceeds the numerical value given by exp(0.5812[ln(sulfate)]+3.357) for selenate. These are the acute exposure criteria. At an example sulfate concentration of 100 mg/L, the 24-hour average selenate concentration should not exceed 417 µg/L.

The available data for selenium, evaluated as above, indicate that saltwater aquatic life should likewise be protected from acute effects of selenium if the 24-hour average concentration of selenite seldom exceeds 127 µg/L. Because selenium might be as chronically toxic to saltwater fishes as it is to freshwater fishes, the status of the fish community should be monitored if selenium exceeds 5.85 µg/g dw in summer or fall or 7.91 µg/g dw during any season in the whole-body tissue of salt water fishes.

Implementation

As discussed in the Water Quality Standards Regulation (U.S. EPA 1983b), a water quality criterion for aquatic life has regulatory force only after it as been adopted in a State water quality standard. Such a standard specifies a criterion for a pollutant that is consistent with a particular designated use. With the concurrence of the U.S. EPA, States designate one or more uses for each body of water or segment thereof and adopt criteria that are consistent with the uses (U.S. EPA 1983c, 1987b). In each standard, a State may adopt the national criterion (if one exists), or an adequately justified state-specific or site-specific criterion.

Criterion concentrations, durations of averaging periods, and frequencies of allowed excursions may be established on a state-specific or site-specific basis (U.S. EPA 1983c, U.S. EPA 1985c). Because the

chronic criterion is tissue-based for selenium, the averaging period only applies to the acute criterion, which is defined as a 24-hour average, based on the speed at which effects may occur in the toxicity tests used for its derivation. Implementation guidance on using criteria to derive water quality-based effluent limits is available in U.S. EPA (1985c and 1987b).

APPENDIX A

INFORMATION USED IN THE SULFATE CORRECTION OF SELENATE ACUTE TOXICITY

Table A-1. Degrees of freedom (df), coefficients of determination (r^2), slopes and respective confidence intervals (CI) for regressions of the natural logarithm of selenate LC50% on the natural logarithm of sulfate concentration. The "Common regression" combines regression lines for individual species into a single model (Zar 1999), its slope is computed as in analysis of covariance. The "Total regression" estimates a linear function for all points, irrespective of taxa.

Species	df	r^2	Slope	95% CI
Fathead Minnow	14	0.83	0.48	[0.35, 0.60]
Chinook Salmon	3	0.87	0.49	[0.14, 0.83]
Gammarus pseudolimnaeus	5	0.61	0.86	[0.07, 1.66]
Hyalella azteca	4	0.39	0.19	[-0.14, 0.51]
Daphnia magna	4	0.92	0.87	[0.52, 1.22]
Ceriodaphnia dubia	11	0.84	0.70	[0.50, 0.91]
"Common regression"	46	0.65	0.58	[0.45, 0.71]
"Total regression"	51	0.36	0.76	[0.48, 1.04]

Table A-2. Data used in the regressions of the natural logarithm of selenate acute values on the natural logarithm of sulfate concentrations.

Species	Age/ Size	Data Source	[Sulfate] (µg/L)	LC50 or EC50 (µg/L)	Adjusted LC50
Hydra					
Hydra sp.	adult	Brooke et al. 1985	12000	7,300	25031.02
Leech					
Nephelopsis obscura	adult	Brooke et al. 1985	12000	442,000	1515577
Snail					
Aplexa hypnorum		Brooke et al. 1985	12000	193,000	661779.1
Cladoceran					
Ceriodaphnia dubia	<24 hr	Brix et al. 2001a,b	52000	1,969	2879.368
Ceriodaphnia dubia		Brix et al. 2001a,b	55000	1,864	2638.398
Ceriodaphnia dubia		GLEC 1999	25000	376	841.5682
Cladoceran					
Daphnia magna		Dunbar et al. 1983	163000	5,300	3989.863
Daphnia magna		Boyum 1984	22000	1,010	2434.939
Daphnia magna		Brooke et al. 1985	12000	570	1954.477
Daphnia magna		Ingersoll et al. 1990	41000	2,560	4298.133
Daphnia magna		Ingersoll et al. 1990	68000	4,070	5092.556
Daphnia magna		Maier et al. 1993	82000	2,840	3187.186
Daphnia pulex	<24 hr	Brix et al. 2001a,b	54000	10,123	14482.21
Daphnia pulex		Brix et al. 2001a,b	38000	8,111	14232.89
Daphnia pulex		GLEC 1999	25000	1,528	3419.99
Daphnia pulicaria		Boyum 1984	22000	246	593.0643
Amphipod					
Gammarus lacustris	8-12 mm	Brix et al. 2001a,b	120000	3,054	2746.951
Gammarus pseudolimnaeus	adult	Brooke et al. 1985	12000	75	257.168
Gammarus pseudolimnaeus		Brooke 1987	12000	57	195.4477
Gammarus pseudolimnaeus		GLEC 1998	25000	1,180	2641.092

Species	Age/ Size	Data Source	[Sulfate] (µg/L)	LC50 or EC50 (µg/L)	Adjusted LC50
Gammarus pseudolimnaeus		GLEC 1998	125000	2,870	2520.927
Gammarus pseudolimnaeus		GLEC 1998	367000	3,710	1742.628
Gammarus pseudolimnaeus		GLEC 1998	635000	3,270	1116.855
Gammarus pseudolimnaeus		GLEC 1999	25000	2,191	4903.925
Amphipod					
Hyalella azteca		Adams 1976	-	760	-
Hyalella azteca		Brasher and Ogle 1993	13000	1,031	3374.516
Hyalella azteca		Brix et al. 2001a,b	55000	1,428	2021.262
Hyalella azteca		GLEC 1998	40000	2,480	4224.001
Hyalella azteca		GLEC 1998	125000	1,350	1185.802
Hyalella azteca		GLEC 1998	367000	1,540	723.3552
Hyalella azteca		GLEC 1998	822000	3,580	1052.407
Midge					
Chironomus decorus	4th instar	Maier and Knight 1993	27000	23,700	50725.32
Midge					
Paratanytarsus parthenogeneticus	3rd instar	Brooke et al. 1985	12000	20,000	68578.14
Coho salmon					
Oncorhynchus kisutch	0.5 g	Hamilton and Buhl 1990b	185000	32,500	22730.56
Oncorhynchus kisutch	1.7 g	Hamilton and Buhl 1990b	291000	39,000	20963.42
Oncorhynchus kisutch	alevin	Buhl and Hamilton 1991	41000	158,422	265983.9
Oncorhynchus kisutch	juvenile	Buhl and Hamilton 1991	41000	30,932	51933.53
Oncorhynchus tshawytscha	(0.7 g	Hamilton and Buhl 1990b	185000	121,000	84627.63
Oncorhynchus tshawytscha	0.5 g	Hamilton and Buhl 1990b	185000	100,000	69940.19
Oncorhynchus tshawytscha	1.6 g	Hamilton and Buhl 1990b	291000	180,000	96754.25
Oncorhynchus tshawytscha		Hamilton and Buhl 1990b	291000	134,000	72028.17
Oncorhynchus tshawytscha	eyed egg	Hamilton and Buhl 1990b	47000>552,000		-
Oncorhynchus tshawytscha	alevin	Hamilton and Buhl 1990b	47000>176,640		-

Species	Age/ Size	Data Source	[Sulfate] (µg/L)	LC50 or EC50 (µg/L)	Adjusted LC50
Oncorhynchus tshawytscha	0.31 g	Hamilton and Buhl 1990b	47000	62,900	97548.09
Rainbow trout					
Oncorhynchus mykiss	juvenile	Brooke et al. 1985	12000	24,000	82293.77
Oncorhynchus mykiss	alevin	Buhl and Hamilton 1991	41000	196,460	329848.1
Oncorhynchus mykiss	juvenile	Buhl and Hamilton 1991	41000	13,501	22667.61
Oncorhynchus mykiss		Spehar 1986	12000	47,000	161158.6
Arctic grayling					
Thymallus arcticus	alevin	Buhl and Hamilton 1991	41000	41,800	70180.45
Thymallus arcticus	juvenile	Buhl and Hamilton 1991	41000	75,240	126324.8
Fathead minnow					
Pimephales promeles		Adams 1976	-	11,800	-
Pimephales promeles		Adams 1976	-	11,000	-
Pimephales promeles		Adams 1976	-	12,500	-
Pimephales promelas	juvenile	Brooke et al. 1985	12000	2,300	7886.486
Pimephales promelas		Spehar 1986	12000	5,500	18858.99
Pimephales promelas		GLEC 1998	24000	6,210	14233
Pimephales promelas		GLEC 1998	160000	10,800	8218.538
Pimephales promelas		GLEC 1998	474000	18,000	7286.649
Pimephales promelas		GLEC 1998	906000	42,100	11695.65
Colorado squawfish					
Ptychocheilus lucius	fry	Hamilton 1995	164000	27,588	20694.67
Ptychocheilus lucius	0.4-1.1 g juvenile	Hamilton 1995	164000	119,548	89676.92
Ptychocheilus lucius	1.7 g juvenile	Hamilton 1995	164000	138,358	103786.9
Ptychocheilus lucius	larva	Buhl and Hamilton 1996	174000	13,580	9842.351
Ptychocheilus lucius	juvenile	Buhl and Hamilton 1996	174000	42,780	31005.58
Ptychocheilus lucius	0.024-0.047 g	Hamilton and Buhl 1997a	97000	88,000	89571.65

Species	Age/ Size	Data Source	[Sulfate] (µg/L)	LC50 or EC50 (µg/L)	Adjusted LC50
Bonytail					
Gila elegans	fry	Hamilton 1995	164000	22,990	17245.56
Gila elegans	1.1 g juvenile	Hamilton 1995	164000	102,828	77134.7
Gila elegans	2.6 g juvenile	Hamilton 1995	164000	90,706	68041.58
Gila elegans	larva	Buhl and Hamilton 1996	174000	14,570	10559.87
Gila elegans	juvenile	Buhl and Hamilton 1996	174000	24,010	17401.68
Razorback sucker					
Xyrauchen texanus	fry	Hamilton 1995	164000	20,064	15050.67
Xyrauchen texanus	0.9 g juvenile	Hamilton 1995	164000	15,048	11288.00
Xyrauchen texanus	2.0 g juvenile	Hamilton 1995	164000	10,450	7838.892
Xyrauchen texanus	larva	Buhl and Hamilton 1996	174000	13,910	10081.52
Xyrauchen texanus	juvenile	Buhl and Hamilton 1996	174000	7,620	5522.733
Xyrauchen texanus	0.006-0.042 g	Hamilton and Buhl 1997a	97000	15,900	16183.97
Flannelmouth sucker					
Catostomus latipinnis	12-13 days	Hamilton and Buhl 1997b	97000	26,900	27380.43
Channel catfish					
Ictalurus punctatus	juvenile	Brooke et al. 1985	12000	66,000	226307.9
Bluegill					
Lepomis macrochirus	juvenile	Brooke et al. 1985	12000	63,000	216021.1

APPENDIX B

TOXICITY OF SELENIUM TO AQUATIC PLANTS

Toxicity to Aquatic Plants

Selenite

Data are available on the toxicity of selenite to 13 species of freshwater algae and plants (Table B-1). Results ranged from an LC_{50} of 70,000 µg/L for the green alga, *Chlorella ellipsoidea* (Shabana and El-Attar 1995) to 522 µg/L for incipient inhibition of the green alga,*Scenedesmus quadricauda* (Bringmann and Kuhn 1977a, 1978a,b, 1979, 1980b). Foe and Knight (Manuscript) found that 75 µg/L decreased the dry weight of *Selenastrum capricornutum* (Table F-1). Wehr and Brown (1985) reported that 320 µg/L increased the growth of the alga *Chrysochromulina breviturrita.* Thus, the sensitivities of freshwater algae to selenite cover about the same range as the acute and chronic sensitivities of freshwater animals.

The 96-hr EC_{50} for the saltwater diatom, *Skeletonema costatum*, is 7,930 µg/L, based on reduction in chlorophyll a (Table B-1). Growth of *Chlorella* sp., *Platymonas subcordiformis*, and *Fucus spiralis* increased at selenite concentrations from 2.6 to 10,000 µg/L (Table F-1). Other marine algae exposed to selenite from 14 to 60 days had no observed effect concentrations (NOAEC) that ranged from 1,076 to 107,606 µg/L. These data suggest that saltwater plants will not be adversely affected by concentrations of selenite that do not affect saltwater animals.

Selenate

Growth of several species of green algae were affected by concentrations ranging from 100 to 40,000 µg/L (Table B-1). Blue-green algae appear to be more tolerant to selenate with 1,866 µg/L being the lowest concentration reported to affect growth (Kiffney and Knight 1990). Kumar (1964) found that a blue-green alga developed and lost resistance to selenate. The difference in the sensitivities of green and blue-green algae to selenate might be of ecological significance, particularly in bodies of water susceptible to nuisance algal blooms. For example, Patrick et al. (1975) reported that a concentration of 1,000 µg/L caused a natural assemblage of algae to shift to a community dominated by blue-green algae.

The saltwater coccolithophore, *Cricosphaera elongata*, had reduced growth when exposed to 41,800 µg/L selenate for 14 days (Boisson et al. 1995). Seven other saltwater algal species investigated by Wong and

Oliveira (1991a) exhibited NOEC growth values that ranged from 1,043 to 104,328 µg/L. At 10,000 µg/L, selenate is lethal to four species of saltwater phytoplankton and lower concentrations increase or decrease growth (Table F-1). Wheeler et al. (1982) reported that concentrations as low as 10 µg/L reduced growth of *Porphyridium cruentum* (Table F-1).

Although selenite appears to be more acutely and chronically toxic than selenate to most aquatic animals, this does not seem to be true for aquatic plants. Selenite and selenate are about equally toxic to the freshwater algae *Anabaena cylindrica*, *Anabaena flos-aquae*, *Anabaena variabilis, Anacystis nidulans,* and *Scenedesmus dimorphus* (Kiffney and Knight 1990; Kumar and Prakash 1971; Moede et al. 1980) and the saltwater algae *Agemenellum quadroplicatum*, *Chaetoceros vixvisibilis* and *Amphidinium carterae* (Wong and Oliveira 1991a). The two oxidation states equally stimulated growth of*Chrysochromulina breviturrita* (Wehr and Brown 1985). On the other hand, selenate is more toxic than selenite to the freshwater *Selenastrum capricornutum* (Richter 1982; Ibrahim and Spacie 1990) and the saltwater *Chorella* sp., *Platymonas subcordiformis* and *Nannochloropsis oculata* (Wheeler et al. 1982; Wong and Oliveira 1991a). In addition, Fries (1982) found that growth of thalli of the brown macroalga,*Fucus spiralis*, was stimulated more by exposure to selenite at 2.605 µg/L than to the same concentration of selenate.

A Final Plant Value, as defined in the Guidelines, cannot be obtained because no test in which the concentrations of selenite or selenate were measured and the endpoint was biologically relevant has been conducted with an important aquatic plant species.

Table B-1. Toxicity of Selenium to Aquatic Plants

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration (days)	Effect	Concentration (µg/L)[a]	Reference
			FRESHWATER SPECIES			
			Selenium (IV)			
Green alga, *Chlorella vulgaris*	Sodium selenite	-	90-120	Reduced growth	5,480	De Jong 1965
Green alga, *Chlorella ellipsoidea*	Sodium selenite	-	7	EC50	70,000	Shabana and El-Attar 1995
Green alga, *Scenedesmus dimorphus*	Sodium selenite	-	14	Reduced growth	24,000	Moede et al. 1980
Green alga, *Scenedesmus quadricauda*	Sodium selenite	-	8	Incipient inhibition	522	Bringmann and Kuhn 1977a; 1978a,b; 1979; 1980b
Green alga, *Scenedesmus quadricauda*	Sodium selenite	-	8	Incipient inhibition	2,500	Bringmann and Kuhn 1959a
Green alga, *Selenastrum capricornutum*	Sodium selenite	-	4	EC50	2,900	Richter 1982
Green alga, *Selenastrum capricornutum*	Sodium selenite	-	6	EC50	65,000	Ibrahim and Spacie 1990
Blue-green alga, *Anabaena constricta*	Sodium selenite	-	7	EC50	67,000	Shabana and El-Attar 1995
Blue-green alga, *Anabaena cylindrica*	Sodium selenite	-	14	Reduced growth	24,000	Moede et al. 1980
Blue-green alga, *Anabaena flos-aquae*	Sodium selenite	-	10	Reduced chlorophyll a	1,866	Kiffney and Knight 1990
Blue-green alga, *Anabaena variabilis*	Sodium selenite	-	6-18	LC50	15,000[b]	Kumar and Prakash 1971
Blue-green alga, *Anacystis nidulans*	Sodium selenite	-	10-18	LC50	30,000[b]	Kumar and Prakash 1971
Blue-green alga, *Microcystis aeruginisa*	Sodium selenite	-	8	Incipient inhibition	9,400 (9,300)	Bringmann and Kuhn 1976; 1978a,b

Table B-1. Toxicity of Selenium to Aquatic Plants (cont.)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration (days)	Effect	Concentration (µg/L)[a]	Reference
Alga, *Euglena gracilis*	-	-	15	Reduced growth	5,920	Bariaud and Mestre 1984
Duckweed, *Lemna minor*	-	-	4	EC50	2,400	Wang 1986
Duckweed, *Lemna minor*	Sodium selenite	-	14	EC50 (mult. rate)	3,500	Jenner and Janssen-Mommen 1993
Duckweed, *Lemna minor*	Sodium selenite	-	14	NOEC (mult. rate)	800	Jenner and Janssen-Mommen 1993
			Selenium (VI)			
Green alga, *Ankistrodesmus falcatus*	Sodium selenate	-	14	Did not reduce growth	10	Vocke et al. 1980
Green alga, *Scenedesmus dimorphus*	Sodium selenate	-	14	Reduced growth	22,100	Moede et al. 1980
Green alga, *Scenedesmus obliquus*	Sodium selenate	-	14	Reduced growth	100	Vocke et al. 1980
Green alga, *Selenastrum capricornutum*	Sodium selenate	-	14	Reduced growth	300	Vocke et al. 1980
Green alga, *Selenastrum capricornutum*	Sodium selenate	-	4	EC50	199	Richter 1982
Green alga, *Selenastrum capricornutum*	Sodium selenate	-	6	EC50	<40,000	Ibrahim and Spacie 1990
Blue-green alga, *Anabaena cylindrica*	Sodium selenate	-	14	Reduced growth	22,100	Moede et al. 1980
Blue-green alga, *Anabaena flos-aquae*	Sodium selenate	-	10	Reduced chlorophyll a	1,866	Kiffney and Knight 1990
Blue-green alga, *Anacystis nidulans*	Sodium selenate	-	6-18	EC50	39,000[b]	Kumar and Prakash 1971

Table B-1. Toxicity of Selenium to Aquatic Plants (cont.)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration (days)	Effect	Concentration (µg/L)[a]	Reference
Blue-green alga, *Anabaena viriabilis*	Sodium selenate	-	10-18	EC50	17,000[b]	Kumar and Prakash 1971
Blue-green alga, *Microcoleus vaginatus*	Sodium selenate	-	14	Reduced growth	10,000	Vocke et al. 1980
Duckweed, *Lemna minor*	Sodium selenate	-	14	EC50 (mult. rate)	11,500	Jenner and Janssen-Mommen 1993
Duckweed, *Lemna minor*	Sodium selenate	-	14	NOEC (mult. Rate)	>2,400	Jenner and Janssen-Mommen 1993

Table B-1. Toxicity of Selenium to Aquatic Plants (cont.)

Species	Chemical	Salinity (g/kg)	Duration (days)	Effect	Concentration (µg/L)[a]	Reference
			SALTWATER SPECIES			
			Selenium (IV)			
Green alga, *Dunaliella tertiolecta*	Sodium selenite	-	60	NOEC growth	1,076	Wong and Oliveira 1991a
Cyanophyceae alga, *Agemenellum quadruplicatum*	Sodium selenite	-	60	NOEC growth	10,761	Wong and Oliveira 1991a
Diatom, *Chaetoceros vixvisibilis*	Sodium selenite	-	60	NOEC growth	1,076	Wong and Oliveira 1991a
Diatom, *Skeletonema costatum*	Selenious acid[c]	-	4	EC50 (reduction in chlorophyll a)	7,930	U.S. EPA 1978
Coccolithophore, *Cricosphaera elongata*	Sodium selenite	-	14	Reduced growth	4,570	Boisson et al. 1995
Dinoflagellate, *Amphidinium carterae*	Sodium selenite	-	60	NOEC growth	10,761	Wong and Oliveira 1991a
Dinoflagellate, *Peridinopsis borgei*	Selenium oxide	-	70-75	Maximum growth	0.01-0.05	Lindstrom 1985
Eustigmatophyceae alga, *Nannochloropsis oculata*	Sodium selenite	-	60	NOEC growth	107,606	Wong and Oliveira 1991a
Pyrmnesiophyceae alga, *Isochrysis galbana*	Sodium selenite	-	60	NOEC growth	1,076	Wong and Oliveira 1991a
Pyrmnesiophyceae alga, *Pavlova lutheri*	Sodiun selenite	-	60	NOEC growth	1,076	Wong and Oliveira 1991a

Table B-1. Toxicity of Selenium to Aquatic Plants (cont.)

Species	Chemical	Salinity (g/kg)	Duration (days)	Effect	Concentration (µg/L)[a]	Reference
			Selenium (VI)			
Green alga, *Dunaliella tertiolecta*	Sodium selenate	-	60	NOEC growth	104,328	Wong and Oliveira 1991a
Cyanophyceae alga, *Agemenellum quadruplicatum*	Sodium selenate	-	60	NOEC growth	10,433	Wong and Oliveira 1991a
Diatom, *Chaetoceros vixvisibilis*	Sodium selenate	-	60	NOEC growth	1,043	Wong and Oliveira 1991a
Coccolithophore, *Cricosphaera elongata*	Sodium selenate	-	14	Reduced growth	41,800	Boisson et al. 1995
Dinoflagellate, *Amphidinium carterae*	Sodium selenate	-	60	NOEC growth	10,433	Wong and Oliveira 1991a
Eustigmatophyceae alga, *Nannochloropsis oculata*	Sodium selenate	-	60	NOEC growth	10,433	Wong and Oliveira 1991a
Pyrmnesiophyceae alga, *Isochrysis galbana*	Sodium selenate	-	60	NOEC growth	10,433	Wong and Oliveira 1991a
Pyrmnesiophyceae alga, *Pavlova lutheri*	Sodium selenate	-	60	NOEC growth	104,328	Wong and Oliveira 1991a

[a] Concentration of selenium, not the chemical.
[b] Estimated from published graph.
[c] Reported by Barrows et al. (1980) in work performed under the same contract.

APPENDIX C

BIOCONCENTRATION AND BIOACCUMUALTION OF SELENIUM

Bioconcentration and Bioaccumulation

Laboratory-Derived

Bioconcentration factors (BCFs) for selenium(IV) that have been obtained with freshwater species range from a low of 2 for the muscle of rainbow trout to 470 for the bluegill sunfish (Table C-1). Adams (1976) studied both uptake and elimination of selenium ^{75} by fathead minnows exposed to mean concentration of 12, 24, and 50 µg/L in the water. He found that concentrations in whole fish and in individual tissues increased at a rapid rate during the first 8 days and then at a slower rate for the next 88 days. Steady-state was approached, but not reached, after 96 days. The highest concentrations were found in viscera. Elimination of selenium was curvilinear and became asymptotic with the time axis after 96 days. Elimination was most rapid from the viscera with a half-life of 5.1 days, but the half-life of selenium in other tissues was greater than 50 days.

Adams (1976) also conducted uptake studies with rainbow trout exposed for 48 days to selenium(IV) at water concentrations ranging from 310 to 950 µg/L. Some of the trout died, and concentrations were somewhat higher in dead fish than in survivors. As with the fathead minnow, the viscera contained more selenium than gill or muscle. Based on his tests with the two fish species, Adams (1976) concluded that there was an inverse relationship between BCF and the concentration of selenium(IV) in water.

Gissel-Nielsen and Gissel-Nielsen (1978) exposed juvenile rainbow trout (*Oncorhynchus mykiss*) to waterborne selenium(IV) over a four week period. Exposure to selenium at 100 µg/L raised the selenium concentration in fish to 2.3 ± 0.02 µg/g dw, without increasing mortality, and steady-state conditions were shown to have been achieved.

Hodson et al. (1980) exposed rainbow trout to selenium(IV) from fertilization until 44 weeks post-hatch. At 53 µg/L selenium in the water, the BCF ranged from 8 L/kg for whole-body to 240 L/kg for liver. They concluded that selenium in tissues did not increase in proportion to selenium(IV) in water.

Hunn et al. (1987) exposed rainbow trout in a flow-through system to waterborne selenium(IV) for 90 days. The selenium concentration in the water where significant effects were not observed was 21 µg/L and the corresponding whole-body tissue level was 0.64 µg/g dw, the data yielding a BCF value of 30.5 L/kg.

Barrows et al. (1980) exposed bluegills to selenious acid for 28 days. They reported a maximum BCF in the whole fish of 20 L/kg and a half-life of between 1 and 7 days. If bluegills bioconcentrate selenium in the same manner as the rainbow trout used by Adams (1976), the 28-day exposure might not have been long enough to reach steady-state.

Lemly (1982) exposed bluegills and largemouth bass to 10 µg of selenium/L for 120 days to determine the effect of hardness and temperature on uptake and elimination. For bluegills, the geometric mean whole-body BCF at 20° and 30°C was 452 L/kg. For largemouth bass in similar tests, the BCF was 295 L/kg. For both species, the spleen, liver, kidney, and heart had higher concentrations than the whole-body. Neither water temperature nor hardness had a significant effect on the amount of selenium accumulated in the tissues after 90 days, although earlier values were influenced. After 30 days in clean water, selenium concentrations remained unchanged in spleen, liver, kidney, and white muscle, but the half-life for selenium in gills and erythrocytes was less than 15 days.

Besser et al. (1993) measured the aqueous bioaccumulation of both waterborne selenium(IV) and selenium(VI) by bluegill over a 30-day period. Selenium concentrations were monitored radiometrically with ^{75}Se- labeled compounds. Bluegills concentrated selenium about equally from both inorganic species and demonstrated similar aqueous selenium uptake rate constants (about 3 per day at 10 µg of selenium/L). A kinetic uptake-depuration model was used to estimate BCFs. Estimated BCFs for both selenium(IV) and selenium(VI) derived from the data were 56 L/kg.

Bertram and Brooks (1986) exposed adult fathead minnows to sodium selenate in water, in food, and in food and water together. The food was specially prepared by raising algae in a medium containing selenium(VI), feeding the algae to daphnids, mixing the exposed daphnids with unexposed daphnids, dewatering to form a "cake", and freezing for storage. Uptake of selenium(VI) from water (without the additional selenium in food) reached steady-state within 28 days. The whole-body BCFs ranged from 21 to 52 L/kg and decreased as the concentration in water increased (Table C-1). Uptake of selenium(VI) from food alone or from food and water together did not reach steady-state in 8 and 11 weeks, respectively. The uptake of selenium from food and water were additive.

Besser et al. (1993) also determined BCF values for algae and *Daphnia magna* exposed separately to waterborne selenium(IV) and selenium(VI). At 10 µg of selenium/L, the BCFs calculated for algae were

1440 L/kg for selenium(IV) and 428 L/kg for selenium(VI). In these laboratory simulated food web studies (waterborne selenium to algae; algae to *Daphnia*; and *Daphnia* to bluegills) concentration factors (CFs) for the transfer of selenium from algae to *Daphnia* and *Daphnia* to bluegill (0.61 and 0.51 L/kg, respectively) were also determined (Table C-2). Using the BCF and CF data, one can calculate an estimated BAF for bluegill for this laboratory food chain. An estimated BAF value of 550 L/kg was calculated for a waterborne exposure of 10 µg/L of 1:1 selenite:selenate to the algae- *Daphnia* - bluegill web.

A three-trophic level food chain experiment consisting of the alga, *Chlorella vulgaris*, the rotifer, *Brachionus calyciflorus*, and the fathead minnow, *Pimephales promelas* was conducted by Dobbs et al. (1996). The three species were exposed to selenium(VI) for 25 days in a three-trophic level system whereby the organisms were linked in a continuous flow-through system in separate vessels, with each organism feeding on the trophic level below it. These organisms were continuously exposed for 25 days to either 0, 110.3, 207.7 or 396.1 µg of total recoverable selenium/L from selenium(VI) in natural creek water supplemented with nutrients to sustain algal growth. Algal population growth, rotifer standing crop, and fathead minnow growth were reduced at 207.7, 110.3 and 110.3 µg/L, respectively, after the 25-day exposure. Bioconcentration factors were found to be dependent on the species, treatment level and length of exposure, and they ranged between 100 and 1,000 L/kg.

Hamilton et al. (2000) exposed, separately, swim-up larvae of razorback sucker (*Xyrauchen texanus*) and bonytail (*Gila elegans*) to waterborne selenium in a simulated Green River, Utah water formulation. The selenium was 6:1 selenate:selenite, and the measured ambient or base level was 76 µg/L in the razorback exposure and 73 µg/L in the bonytail exposure. A flow-through system was utilized, and a 90-day partial life-cycle chronic toxicity study monitoring growth, behavior and mortality was conducted. No chronic effects were observed in tests conducted at base level. Higher than ambient concentrations were studied also, but were not selected for use in the BCF derivation due to either observed chronic effects or abnormally high concentrations of selenium and other metals in the test waters. At 90 days, the whole-body tissue levels of selenium were 3.2 µg/g dw in the razorback and 2.2 µg/g dw in the boneytail, reflecting BCF values of 42 and 30 L/kg, respectively.

Field-Derived

Hermanutz et al. (1996) exposed bluegills to selenium(IV) over 221 days in outdoor experimental streams at Monticello, MN which contained a natural food web. At the end of the 221 days in waters maintained at a nominal selenium concentration of 2.5 µg/L, the average whole-body fish tissue level of selenium was 4.825 µg/g Se dw (based on a factor of 0.8 moisture content in fish tissue). The resulting BAF value was 1,930 L/kg.

Garcia-Hernandez et al. (2000) collected fish samples form October 1996 to March 1997 in a Sonora, Mexico wetland. Dissolved selenium concentrations in the water ranged from 5 to 19 µg/l (median of 11 µg/l). Median whole-body concentration of selenium was measured in *Tilapia* (3.0 µg/g dw), carp (3.3 µg/g dw), and largemouth bass (5.1 µg/g dw). Resulting BAF values were 273, 300, and 464 L/kg, respectively.

Kennedy et al. (2000) collected spawning age (3-6 years) cutthroat trout from the Fording River, British Columbia in 1998. The waters of the river had an average selenium level of 13.9 µg/L at the time of collection. The tissue (muscle) of the trout contained 12.5 ± 7.7 µg of selenium/g dw. Utilization of these values provides a field derived muscle BAF of 899 L/kg.

Mason et al. (2000) collected biota in two streams (Blacklick Run and Herrington Creek) in western Maryland in October 1997, April 1998, and July 1998. Water samples were collected for analysis monthly over the duration of the study. Numerous fish species, among other organisms, were collected during each of the sampling periods, and whole-body tissue levels of selenium were measured. In Herrington Creek, the average water concentration of selenium was found to be 0.33 µg/L, and the average tissue levels of selenium in the fish were: bullhead (1.35 µg/g dw); sucker (1.55 µg/g dw), trout (1.94 µg/g Se dw), and chub (1.50 µg/g Se dw). The resulting calculated BAF values were 4,091, 4,697, 5,879, and 4,545 L/kg, respectively. In Blacklick Creek the average water concentration was 0.39 µg/L, and the average tissue levels of selenium in fish were: dace (1.79 µg/g dw), trout (1.94 µg/g dw), and sculpin (2.55 µg/g dw). Resulting BAF values were 4,590, 4,974, and 6,538 L/kg, respectively. Dry weight values were obtained from the published wet weight data employing a 0.8 factor for fish moisture content.

Table C-1. Bioconcentration and Bioaccumulation of selenium by fish.

Fish Species	Selenium Species	Concentration in Water (µg/L)[a]	Duration (days)	Tissue (Concentration)	BCF[b] (L/kg)	BAF[b] (L/kg)	Reference
			LABORATORY DERIVED				
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	-	48	Muscle	2		Adams 1976
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	-	48	Whole-body	10[c]		Adams 1976
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	100	28	Whole-body (2.3 µg/g)	23		Gissel-Nielsen and Gissel-Nielsen 1978
Rainbow trout (embryo), *Oncorhynchus mykiss*	Sodium selenite	-	308 (post-hatch)	Whole-body (estimate)	42		Hodson et al. 1980
Rainbow trout *Oncorhorynchus mykiss*	Sodium selenite	21	90	Whole-body (0.64 µg/g)	30.5		Hunn et al. 1987
Fathead minnow, *Pimephales promelas*	Sodium selenite	-	96	Muscle	11.6		Adams 1976
Fathead minnow, *Pimephales promelas*	Sodium selenite	-	96	Whole-body	17.6		Adams 1976
Fathead minnow (6-9 mo.), *Pimephales promelas*	Sodium selenate	10.7	56	Whole-body	52[d]		Bertram and Brooks 1986
Fathead minnow (6-9 mo.), *Pimephales promelas*	Sodium selenate	21.5	56	Whole-body	26[d]		Bertram and Brooks 1986
Fathead minnow (6-9 mo.), *Pimephales promelas*	Sodium selenate	43.5	56	Whole-body	21[d]		Bertram and Brooks 1986
Bluegill, *Lepomis macrochirus*	Selenious acid	-	28	Whole-body	20		Barrows et al. 1980

Table C-1 continued.

Fish Species	Selenium Species	Concentration in Water (µg/L)[a]	Duration (days)	Tissue (Concentration)	BCF[b] (L/kg)	BAF[b] (L/kg)	Reference
Bluegill, *Lepomis macrochirus*	Sodium selenite	10 10 10 10	120 120 120 120	Whole-body Whole-body Whole-body Whole-body	450 470 430 460		Lemly 1982
Bluegill, *Lepomis macrochirus*	Selenate	10	30	Whole-body	56		Besser et al. 1993
Largemouth bass, *Micropterus salmoides*	Sodium selenite	10 10 10 10	120 120 120 120	Whole-body Whole-body Whole-body Whole-body	310 300 300 270		Lemly 1982
Bluegill, *Lepomis macrochirus*	Selenite	10	30	Whole-body	56		Besser et al. 1993
Bluegill, *Lepomis macrochirus*	selenite: selenate 1:1	10	30	Whole-body		550[e]	Besser et al. 1993
Razorback suker, *Xyrauchen texanus*	selenate/ selenite[f]	76	90	Whole-body (3.2 µg/g)	42		Hamilton et al. 2000
Bonytail, *Gila elegans*	selenate/ selenite[f]	73	90	Whole-body (2.2 µg/g)	30		Hamilton et al. 2000
			FIELD DERIVED				
Bluegill *Lepomis macrochirus*	Selenite	2.5	221	Whole-body (4.825 µg/g)		1,930	Hermanutz et al. 1996
Tilapia sp.	Natural[f]	11	Field	Whole-body (3.0 µg/g)		273	Garcia-Hernandez et al. 2000
Carp, *Cyprinus carpio*	Natural[f]	11	Field	Whole-body (3.3 µg/g)		300	Garcia-Hernandez et al. 2000
Largemouth bass, *Micropterus salmoides*	Natural[f]	11	Field	Whole-body (5.1 µg/g)		464	Garcia-Hernandez et al. 2000
Cutthroat trout, *Oncorhynchus clarki*	Natural[f]	13.9	Field	Muscle (12.5 µg/g)		899	Kennedy et al. 2000
Brown bullhead, *Ictalurus nebulosus*	Natural[f] (Herrington Creek, MD)	0.33	N/A[g] (10 month study)	Whole-body (1.35 µg/g)		4,091	Mason et al. 2000

Table C-1 continued.

Fish Species	Selenium Species	Concentration in Water (µg/L)[a]	Duration (days)	Tissue (Concentration)	BCF[b] (L/kg)	BAF[b] (L/kg)	Reference
White sucker, *Catostomus commersoni*	Natural[f] (Herrington Creek, MD)	0.33	N/A (10 month study)	Whole-body (1.55 µg/g)		4,697	Mason et al. 2000
Brook Trout, *Salvelinus fonticnalis*	Natural[f] (Herrington Creek, MD)	0.33	N/A (10 months study)	Whole-body (1.94 µg/g)		5,879	Mason et al. 2000
Creek Chub, *Semotilus arromaculatus*	Natural[f] (Herrington Creek, MD)	0.33	N/A (10 months study)	Whole-body (1.50 µg/g)		4,545	Mason et al. 2000
Mottled Sculpin, *Cottus bairdi*	Natural[f] (Blacklick Run, MD)	0.39	N/A (10 months study)	Whole-body (2.55 µg/g)		6,538	Mason et al. 2000
Blacknose Dace *Rhinchthus atratulus*	Natural[f] (Blacklick Run, MD)	0.39	N/A (10 months study)	Whole-body (1.79 µg/g)		4,590	Mason et al. 2000
Brook Trout *Salvelins fortinalus*	Natural[f] (Blacklick Run, MD)	0.39	N/A (10 months study)	Whole-body (1.94 µg/g)		4,974	Mason et al. 2000

[a] Measured concentration of selenium.
[b] Bioconcentration factors (BCFs) and bioaccumulation factors (BAFs) are based on measured concentrations of selenium in water and in tissue (dry weight).
[c] Estimated from graph.
[d] Calculated by dividing the reported equilibrium concentration in tissue (steady-state body burden) by the average measured concentration in water.
[e] Laboratory food chain: water–> algae –> daphnia –> bluegill.
[f] Not speciated.
[g] N/A not applicable.

Table C-2. Bioconcentration and Bioaccumulation of selenium by other aquatic organisms.

Other Species	Selenium Form	Concentration in Water (µg/L)[a]	Duration (days)	Tissue (Concentration)	BCF[b] (L/kg)	BAF[b] (L/kg)	Reference
			LABORATORY DERIVED				
Algae, *Chlamydomonas reinhardtii*	Selenite Selenate	10 10	4 4		1440 428		Besser et al. 1993
Cladoceran, *Daphnia magna*	Selenate: Selenite 1:1	156	21	Whole-body (14.7 µg/g)	94		Ingersoll et al. 1990
	Selenate: Selenite 1:1	348	21	Whole-body (31.7 µg/g)	91		
Cladoceran, *Daphnia magna*	Selenite Selenate	10 10	4 4		570[e] 293[e]		Besser et at. 1993
			FIELD DERIVED				
Ephemeroptera	Selenite	2.5	221	Whole-body (5.05 µg/g)		1,957	Hermanutz et al. 1996
Heptageniidae	Selenite	10	221	Whole-body (17.30 µg/g)		1,787	
Ephemeroptera	Natural[f] (Herrington Creek, MD)	0.33	N/A (10 mo. study)	Whole-body (5.05 µg/g)		17,600	Mason et al. 2000
Heptageniidae	Natural[f] (Blacklick Run, MD)	0.39	N/A (10 mo. study)	Whole-body (5.8 µg/g)		14,900	
Chironomidae	Natural[f]	14.5	N/A (3 yr. study)	Wholebody (24.7 µg/g)		1703	Zhang and Moore 1996
	Natural[f]	1.58	N/A (3 yr. study)	Wholebody (10.4 µg/g)		6582	
Chironomidae	Selenite	2.5	221	Wholebody (3.61 µg/g)		1399	Hermanutz et al. 1996
	Selenite	10	221	Wholebody (13.60 µg/g)		1405	
Hydropsychidae	Natural (ite/ate 9:1)	32	N/A	Wholebody (3.1 µg/g)		969	Reash et al. 1999

Table C-2 Continued.

Other Species	Selenium Form	Concentration in Water (µg/L)[a]	Duration (days)	Tissue (Concentration)	BCF[b] (L/kg)	BAF[b] (L/kg)	Reference
Hydropsychidae	Natural[f] (Herrington Creek, MD)	0.33	N/A (10 mo. study)	Wholebody (10.5 µg/g)		31,800	Mason et al. 2000
	Natural[f] (Blacklick Run, MD)	0.39	N/A (10 mo. study)	Wholebody (4.6 µg/g)		11,800	
Astacidae	Natural[f] (Herrington Creek, MD)	0.33	N/A (10 mo. study)	Wholebody (1.275 µg/g)		3864	Mason et al. 2000
	Natural[f] (Blacklick Run, MD)	0.39	N/A (10 mo. study)	Wholebody (0.405 µg/g)		1038	
Periphyton	Natural[f] (Herrington Creek, MD)	0.33	N/A (10 mo. study)	Whole (2.860 µg/g)	8667		Mason et al. 2000
	Natural[f] (Blacklick Run, MD)	0.39	N/A (10 mo. study)	Whole (0.245 µg/g)	628		
Bryophytes	Natural[f] (Herrington Creek, MD)	0.33	N/A (10 mo. study)	Whole (1.860 µg/g)	5636		Mason et al. 2000
	Natural[f] (Blacklick Run, MD)	0.39	N/A (10 mo. study)	Whole (0.780 µg/g)	2000		

[a] Measured concentration of selenium.
[b] Bioconcentration factors (BCFs) and bioaccumulation factors (BAFs) are based on measured concentrations of selenium in water and in tissue (dry weight).
[c] Estimated from graph.
[d] Calculated by dividing the reported equilibrium concentration in tissue (steady-state body burden) by the average measured concentration in water.
[e] Laboratory food chain: water–> algae –> daphnia –> bluegill.
[f] Not speciated.
[g] N/A not applicable.

APPENDIX D

ENVIRONMENTAL FACTORS AFFECTING SELENIUM TOXICITY AND BIOACCUMULATION

Environmental Factors Affecting Selenium Toxicity and Bioaccumulation

A variety of environmental factors have been shown to influence the toxicity/bioaccumulation of selenium. A brief summary of the influence of sulfate, hardness, heavy metals, pH, temperature and day length on selenium toxicity/bioaccumulation is presented below.

Sulfate

In acute toxicity tests and uptake experiments with selenium, sulfate has been shown to antagonize Se toxicity and Se uptake in plants and animals, frequently with a major effect on Se action. Where multiple Se forms are used in joint action experiments, Se(VI) is antagonized most by SO_4 with Se(IV) and Se(II) affected to a lesser extent. Sulfate has reduced Se mortality responses by 90 percent and Se uptake to 10 percent of controls or less. Thus, sulfate is a major co-factor in a number of Se toxicity and Se uptake experiments.

In four acute toxicity tests, sulfate antagonized selenate toxicity in three algae species and the cladoceran *Daphnia*. The LC_{50} values of two desmids (*Cosmerium* spp.) exposed to selenate plus sulfate were 4x and 8x the LC_{50} values of selenate only (Sarma and Jayaraman 1984). The growth of *Selenastrum capricornutum* increased by 50 percent when 11 or 107 µg/L Se(VI) were combined with 3.3or 33 µg/L sulfate (Williams et al. 1994). The toxicity of 490 µg/L selenate to *D. magna* was reduced by 90 percent mortality by combining it with either 10 or 308 mg/L sulfate. Uptake studies, with one exception, document sulfate as antagonistic to uptake of selenium. In many cases, Se uptake rates are reduced to 40 to 50 percent of controls (Se alone or lowest SO_4 concentration), but there are examples of sulfate reducing uptake to 20 percent of controls. These examples include a rooted plant (six percent of control rate), an alga (7 percent), *Daphnia* (20 percent) and a midge (20 percent).

Of the two algal species investigated, *Chlamydomonas reinhardtii* responded less to sulfate and Se(VI) co-exposure (Williams et al. 1994) than *Selenastrum capricornutum* (7 percent low SO_4 rate) (Riedel and Sanders 1996). Widgeon grass (*Ruppia moritima*) uptake reductions (Se uptake, high or low sulfate) occurred most for Se(VI) (6 percent), then Se(IV) (44 percent) and Se(II) (56 percent) (Bailey et al. 1995).

Experiments with *Daphnia* show no interaction of Se and SO_4 in a microcosm experiment (Besser et al. 1989). However, other experiments with Se and SO_4 show a 43 percent reduction of Se uptake by sulfate (Hansen et al.1993) and uptake reductions ranging from 20 to 65 percent among three Se(VI) exposures and two sulfate levels (Ogle and Knight 1996). Se uptake by a midge, *Chironomus decorus*, was reduced to 20 to 65 percent of controls in a 48 hour exposure to 6 mg/L Se(VI) and 3 levels of SO_4 (Hansen et al. 1993).

Hardness

Acute toxicity tests of selenium forms with hardness as a variable were conducted with an invertebrate and three fish species. In all cases, water hardness variations did not cause major changes in the acute toxicity of selenium. LC_{50} value differences due to hardness were no less than half or more than double the LC_{50} of the standard of comparison.

D. magna were exposed to three forms of selenium and one Se mixture in acute toxicity tests (48h LC_{50}) to determine the effect of soft (46 mg/L $CaCO_3$) and hard (134 mg/L $CaCO_3$) water on selenium toxicity. Water hardness did not affect the toxicity of Se(VI) and Se(II), but Se(IV) was slightly more toxic in hard than soft water (LC_{50}, hard/soft = 0.5), as was the 1:1 mixture of Se(IV) and Se(VI) (LC_{50}, hard/soft = 0.6) (Ingersoll et al. 1990). *Mytilus edulis* were exposed to selenite in sea water with salinities of 15, 20, 27 and 30‰ (27‰ was close to the mussel's natural habitat). Se(IV) influx measured during 2 hours of exposure demonstrated an effect on uptake as follows: maximum influx at 20‰ ; greatest influx difference = 0.7 max (34‰) (Wang et al. 1996a).

Fry of chinook salmon and coho salmon were exposed for 96 hr to selenate, selenite and a 1:1 mixture in soft (42 mg/L $CaCO_3$) and hard (211 mg/L $CaCO_3$) water. Advanced fry of chinook salmon were exposed to Se(II) in brackish water (333 mg/L $CaCO_3$). In all cases, variable hardness had no effect on the toxicity of three forms of selenium or the mixture (Hamilton and Buhl 1990b).

Young striped bass (*Morone saxatilis*) exhibited some differential susceptibility to selenite in hard (285 mg/L $CaCO_3$) vs. soft (40 mg/L $CaCO_3$) water (LC_{50} hard/soft = 1.8) with Se(IV) in soft water being more toxic. The LC_{50} of Se(IV) in 1‰ saline (455 mg/L $CaCO_3$) was not significantly different than Se in soft or hard water (Palawski et al. 1985).

The Se BCF values for young salmon (*Onchorhynchus tsawytscha*) exposed for 90 days to a Se(VI):Se(IV) mixture (6:1) were no different in fresh water (371 mg/L $CaCO_3$) or well water (612 mg/L $CaCO_3$). Exposure for 60 days to Se in 1‰ saline reduced the Se BCF to approximately 50% of BCFs for well water and fresh water (Hamilton and Wiedmeyer 1990).

Heavy Metals

Joint action studies with selenium and metals were conducted with cadmium and mercury, which have been investigated frequently in this regard, and arsenic and molybdenum. The latter two chemicals were investigated in a chronic test (at least 3 broods) with *Ceriodaphnia dubia* at exposure concentrations of the three chemicals that alone caused chronic mortality and reproductive effects. As^{+5} or Mo^{+6}, combined with Se(VI) in a chronic test, reduced reproduction and increased cumulative mortality (Naddy et al. 1995).

Mercury uptake experiments with selenite had opposite results in two separate studies with the marine mussel, *Mytilus edulis*. In 30 - 50-day tests, Se(IV) uptake was doubled (Se alone = 0.8 ng/g/d) by joint exposure to Se (30 µg/L) and $HgCl_2$ (5 µg/L) (Pelletier 1986a). Uptake of Se in a 96-hr study (Se(IV), 2 µg/L; $HgCl_2$, 0.1- 1.0 mg/L) decreased as a function of Hg concentration (r^2 = - 0.93) (Micallef and Tyler 1987).

The toxicity of Se(IV) and Se(VI) to a fresh water snail (*Lymnaea*) was reduced by 55 to 66 percent mortality by 0.1 mg/L cadmium in an 11-day water exposure. Using growth to evaluate toxicity of selenium-cadmium pairs in two species of marine phytoplankton (*Crypthecodinium* sp., *Procentrum* sp.), Prevot and Sayer-Gobillard (1986) demonstrated in both species that the toxicity of the higher Se doses was reduced by cadmium. Cadmium slightly elevated Se(IV) uptake in gill tissues of *Carcinas maenas* (marine shore crabs) but Se levels in two other tissues and carapace were no different than Se exposure in a 29-day experiment.

In summary, cadmium mortality effects were consistent in antagonizing the toxicity of selenium, although the level of antagonism was low to moderate in these two cases. Mercury effects on Se uptake by *Mytilus* were not in agreement, i.e. in a 96-hour study, selenium uptake decreased as mercury increased, but in 30 - 50-day tests, mercury enhanced selenium uptake. Both metals are generally toxic which complicates Se-

metal investigations. For example, Se interaction with arsenic or molybdenum were conducted with metal concentrations that were toxic.

pH, Temperature and Day Length

Except for Se(IV) at acidic pH, pH changes in the range associated with natural waters do not have an appreciable effect on uptake of selenium. Temperature is a major modifying influence on the interaction of chemicals and aquatic organisms as shown by sediment storage and *Paramecium* experiments. Interaction by low temperature and day length dramatically enhanced the toxicity of Se in fish chronically exposed under laboratory conditions.

As presented in the chronic section, Lemly (1993b) investigated the effect of temperature and day-length effects with selenium on juvenile bluegills exposed for 180 days. Selenium exposures included 4.8 µg/L in water (SeVI:SeIV = 1:1) and Se(II) in food (5.1 µg/g) and simulation of summer conditions and winter conditions. Functions monitored during the study were percent lipid content of fish (energy reserve), cumulative mortality, body condition factor, Q_{O2} and gill pathology and blood abnormalities. All of these major functions were significantly affected by winter simulation plus selenium in experiments designed to chronically expose bluegills to a combination of selenium and environmental factors that would reflect actual exposure of natural fish populations to selenium during seasonal change.

APPENDIX E

SITE-SPECIFIC CONSIDERATIONS

Site-specific Considerations

Aquatic organism uptake of selenium by both water column exposure and dietary pathways has prompted a number of researchers to investigate the toxicity of selenium under site-specific conditions. Previous site-specific studies have addressed the water-based chronic criterion of 5 µg/L through examination of environmental variables that could potentially influence the availability and/or accumulation of selenium within the aquatic ecosystem under consideration, thereby either increasing or decreasing the toxic impact of selenium on the aquatic community (Adams et al. 1998; Canton and VanDerveer 1997; VanDerveer and Canton 1997).

Now that the recommended chronic criterion is tissue-based, site-specific factors that affect the bioaccumulation of selenium are not relevant in the modification of the criterion. Recent studies on the effects of selenium on bluegill in streams receiving wastewater from a coal ash effluent suggest fish exposed to Se-laden effluents may exhibit tolerance (Lohner et al. 2001a,b,c). The authors found the bluegill population receiving the coal ash effluent to have an age class structure and condition indices similar to reference locations despite having selenium concentration in the ovary and whole-body tissues twice the level of the FCV. Hematological and biochemical assays using samples from exposed bluegill have shown a reduced response relative to reference fish,but the authors contend that they are not always related to selenium. The authors hypothesize that selenium speciation, metabolism, bioavailability and antagonism are possible reasons for the decreased sensitivity of the resident bluegill population in the ash stream. To date, no experiments on the success on embryo-larval development have been conducted.

In an effort to determine if a proposed multiple-use water development project (Animas La Plata) would adversely affect aquatic biota in Colorado and New Mexico, Lemly (1997c) conducted a hazard assessment of selenium using the Protocol Method (Lemly 1995). Using existing environmental monitoring data, the hazard assessment indicated that selenium poses a siginificant toxic threat to aquatic biota in the Animas La Plata Project. Incorporating this information into the proposed water development will substantially reduce the chances of experiencing significant environmental problems similar to those encountered at Belews Lake and Kesterson National Wildlife Refuge. Once an aquatic system is impacted with selenium, it could take several to many years before the biological health of the system can be returned to the original condition prior to perturbation. The Grassland Water District in central California is an example of an

aquatic system that was contaminated with selenium as a result of subsurface agricultural drainwater used for wetland management since 1954 (Paveglio et al. 1997). Selenium contamination of aquatic bird food chains prompted the California State Water Resources Board to mandate the Grassland Water District to reduce selenium concentration starting in 1985 by essentially filling the wetlands with freshwater only. Selenium concentrations in a number of aquatic birds have gradually declined since 1985 (1985 to 1994), but selenium concentrations in some wintering birds still were above concentrations associated with impaired reproduction in laboratory and field studies. The authors estimated under the current management strategy, an additional 1 to 13 years from 1994 are needed for selected species to reach background selenium levels in liver. Thus, approximately 10 to 20 years are needed at this site to reduce the elevated levels of selenium in avian species and restore normal reproductive success.

APPENDIX F

OTHER DATA

Other Data

Selenite

Additional data on the lethal and sublethal effects of selenium on aquatic species are presented in Table F-1. Bringmann and Kuhn (1959a,b, 1976, 1977a, 1979, 1980b, 1981), Jakubczak et al. (1981), and Patrick et al. (1975) reported the concentrations of selenite that caused incipient inhibition (defined variously, such as the concentration resulting in a 3% reduction in growth) for algae, bacteria, and protozoans (Table F-1). Although incipient inhibition might be statistically significant, its ecological importance is unknown. Albertano and Pinto (1986) found the growth of three red algal species was inhibited at selenite concentrations that ranged from 790 to 3,958 µg/L.

Selenate

Dunbar et al. (1983) exposed fed *D. magna* to selenate for seven days and obtained an LC_{50} of 1,870 µg/L. This value is in the range of the 48-hr EC_{50}s in Table F-1.

Watenpaugh and Beitinger (1985a) found that fathead minnows did not avoid 11,200 µg/L selenate during 30-minute exposures (Table F-1). These authors also reported (1985b) a 24-hr LC_{50} of 82,000 µg/L for the same species and they found (1985c) that the thermal tolerance of the species was reduced by 22,200 µg/L. Westerman and Birge (1978) exposed channel catfish embryos and newly hatched fry for 8.5 to 9 days to an unspecified concentration of selenate. Albinism was observed in 12.1 to 36.9% of the fry during the five years of such exposures. Pyron and Beitinger (1989) also investigated fathead minnows, and after a 24-hr exposure, no effect on reproductive behavior was found at 36,000 µg/L, but when adults were exposed to 20,000 µg/L selenate for 24-hr, edema was observed for their larvae.

The respiratory rate of the eastern oyster, *Crassostrea virginica*, was unaffected by exposure to selenate at 400 µg/L for 14 days (Fowler et al. 1981). Embryos of the striped bass were quite tolerant to selenate in dilute salt water (Klauda 1985a, b). There was a 93% successful hatch of embryos at 200,000 µg/L, but 50% of 72-day-old juveniles died after four days at 87,000 µg/L. Exposure of juvenile fish for up to 65 days to concentrations of selenate between 39 and 1,360 µg/L caused developmental anomalies and pathological lesions.

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
			FRESHWATER SPECIES			
			Selenium (IV)			
Green alga, *Scenedesmus quadricauda*	Sodium selenite	-	96 hr	Incipient inhibition (river water)	2,500	Bringmann and Kuhn 1959a,b
Green alga, *Selenastrum capricornutum*	Sodium selenite	-	72 hr	Decreased dry weight and chlorophyll a	75	Foe and Knight, Manuscript
Green alga, *Selenastrum capricornutum*	Sodium selenite	-	72 hr	BCF = 12-21[b]	10-100	Foe and Knight, Manuscript
Green alga, *Selenastrum capricornutum*	Sodium selenite	-	72 hr	BCF = 11,164[c]	150	Foe and Knight, Manuscript
Alga, *Chrysochromulina breviturrita*	Selenious acid	-	30 days	Increased growth	320	Wehr and Brown 1985
Red alga, *Cyanidium caldarium*	Selenious acid	-	20 days	Inhibited growth	3,958	Albertano and Pinto 1986
Red alga, *Cyanidioschyzon merolae*	Selenious acid	-	20 days	Inhibited growth	3,140	Albertano and Pinto 1986
Red alga, *Galdieria sulphuraria*	Selenious acid	-	20 days	Inhibited growth	790	Albertano and Pinto 1986
Algae (diatoms), Mixed population	Sodium selenite	-	18 days	Inhibited growth	11,000	Patrick et al. 1975
Bacterium, *Escherichia coli*	Sodium selenite	-	-	Incipient inhibition	90,000	Bringmann and Kuhn 1959a
Bacterium, *Pseudomonus putida*	Sodium selenite	-	16 hr	Incipient inhibition	11,400 (11,200)	Bringmann and Kuhn 1976; 1977a; 1979; 1980b
Protozoan, *Entosiphon sulcatum*	Sodium selenite	-	72 hr	Incipient inhibition	1.8 (1.9)	Bringmann 1978; Bringmann and Kuhn 1979; 1980b; 1981
Protozoan, *Microreqma heterostoma*	Sodium selenite	-	28 hr	Incipient inhibition	183,000	Bringmann and Kuhn 1959b

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
Protozoan, *Chilomonas paramecium*	Sodium selenite	-	48 hr	Incipient inhibition	62	Bringmann and Kuhn 1981; Bringmann et al. 1980
Protozoan, *Uronema parduezi*	Sodium selenite	-	20 hr	Incipient inhibition	118	Bringmann and Kuhn 1980a; 1981
Snail, *Lymnaea stagnalis*	Sodium selenite	-	7.5 days	LT50	3,000	Van Puymbroeck et al. 1982
Cladoceran, *Daphnia magna*	Sodium selenite	-	48 hr	EC50 (river water)	2,500	Bringmann and Kuhn 1959a,b
Cladoceran, *Daphnia magna*	Sodium selenite	214	24 hr	LC50	16,000	Bringmann and Kuhn 1977a
Cladoceran, *Daphnia magna*	Sodium selenite	214	24 hr	EC50 (swimming)	9.9	Bringmann and Kuhn 1977b
Cladoceran, *Daphnia magna*	Sodium selenite	329	48 hr 96 hr 14 days	EC50 (fed)	710 430 430	Halter et al. 1980
Cladoceran (<24 hr), *Daphnia magna*	Sodium selenite	-	48 hr 21 days	EC50 (fed)	685 160	Adams and Heidolph 1985
Cladoceran (5th instar), *Daphnia magna*	Sodium selenite	-	48 hr	LC50 (fed)	680	Johnston 1987
Cladoceran, *Daphnia magna*	Selenious acid	220[d]	48 hr	LC50 (fed)	1,200	Kimball, Manuscript
Cladoceran (preadult), *Daphnia pulex*	Sodium selenite	42	24 hr	Did not reduce oxygen consumption or filtering rate	>498	Reading and Buikema 1980
Ostracod, *Cyclocypris* sp.	Sodium selenite	100.8	48 hr	LC50	130,000	Owsley 1984
Amphipod, *Hyalella azteca*	Sodium selenite	329	14 days	LC50 (fed)	70	Halter et al. 1980
Amphipod (2 mm length), *Hyalella azteca*	Sodium selenite	133	48 hr	LC50	623	Brasher and Ogle 1993
Amphipod (2 mm length), *Hyalella azteca*	Sodium selenite	133	10 days	LC50 (fed)	312	Brasher and Ogle 1993

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
Amphipod (2 mm length), *Hyalella azteca*	Sodium selenite	133	24 days	LOEC reproduction (static-renewal)	200	Brasher and Ogle 1993
Midge (first instar), *Chironomus riparius*	Sodium selenite	134	48 h	LC50	7,950	Ingersoll et al. 1990
Midge (first instar), *Chironomus riparius*	Sodium selenite	40-48	48 h	LC50	14,600	Ingersoll et al. 1990
Coho salmon (fry), *Oncorhynchus kisutch*	Sodium selenite	325	43 days	LC50	160	Adams 1976
Rainbow trout (fry), *Oncorhynchus mykiss*	Sodium selenite	334	21 days	LC50	460	Adams 1976
Rainbow trout (fry), *Oncorhynchus mykiss*	Sodium selenite	334	21 days	Reduced growth	250	Adams 1976
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	330	5 days	LC50	2,700 2,750	Adams 1976
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	325	48 days	LC50	500	Adams 1976
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	325	96 days	LC50	280	Adams 1976
Rainbow trout (juvenile), *Oncorhynchus mykiss*	Sodium selenite	-	4 wk	MATC survival	200	Gissel-Nielsen and Gissel-Nielsen 1978
Rainbow trout (juvenile), *Oncorhynchus mykiss*	Sodium selenite	-	4 wk	MATC survival	4.7 µg/g dw (whole-body)	Gissel-Nielsen and Gissel-Nielsen 1978
Rainbow trout (juvenile), *Oncorhynchus mykiss*	Sodium selenite	-	4 wk	BCF = 23	100	Gissel-Nielsen and Gissel-Nielsen 1978
Rainbow trout (juvenile), *Oncorhynchus mykiss*	Sodium selenite	-	42 wk	MATC growth (dietary only exposure)	>9.96 µg Se/g dw (food)	Goettl and Davies 1978

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
Rainbow trout (juvenile), *Oncorhynchus mykiss*	Sodium selenite	-	42 wk	MATC survival (dietary only exposure)	5.34 µg Se/g dw (food)	Goettl and Davies 1978
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	135	9 days	LC50	7,020	Hodson et al. 1980
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	135	96 hr 9 days	LC50 (fed)	7,200 5,410	Hodson et al. 1980
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	135	96 hr 9 days	LC50 (fed)	8,200 6,920	Hodson et al. 1980
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	135	41 days	LOAEC (Reduced hatch of eyed embryos)	26	Hodson et al. 1980
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	135	50 wk	Decreased iron in blood and red cell volume	53	Hodson et al. 1980
Rainbow trout (fertilized egg), *Oncorhynchus mykiss*	Sodium selenite	135	44 wk	BCF = 33.2 BCF = 21.1	53	Hodson et al. 1980
Rainbow trout (embryo), *Oncorhynchus mykiss*	Sodium selenite	-	120 hr	Did not reduce survival or time to hatch	10,000	Klaverkamp et al. 1983b
Rainbow trout, *Oncorhynchus mykiss*	Sodium selenite	-	90 days	Chronic value for survival	14	Mayer et al. 1986
Rainbow trout (sac fry), *Oncorhynchus mykiss*	Sodium selenite	272	90 days	LC50	55.2[e]	Hunn et al. 1987
Rainbow trout (sac fry), *Oncorhynchus mykiss*	Sodium selenite	272	90 days	MATC survival	31.48	Hunn et al. 1987
Rainbow trout (egg), *Oncorhynchus mykiss*	Sodium selenite	-	96 hr	BCF = 17.5 BCF = 3.5	0.4 45.6	Hodson et al. 1986

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
Rainbow trout (embryo), *Oncorhynchus mykiss*	Sodium selenite	-	96 hr	BCF = 3.1 BCF = 3.0	0.4 45.6	Hodson et al. 1986
Rainbow trout (sac-fry), *Oncorhynchus mykiss*	Sodium selenite	-	96 hr	BCF = 13.1 BCF = 1.6	0.4 45.6	Hodson et al. 1986
Rainbow trout (swim-up fry) *Oncorhynchus mykiss*	Sodium selenite	-	96 hr	BCF = 80.3 BCF = 20.2	0.4 45.6	Hodson et al. 1986
Northern pike, *Esox lucius*	Sodium selenite	10.2	76 hr	LC50	11,100	Klaverkamp et al. 1983a
Goldfish, *Carassius auratus*	Selenium dioxide	157	14 days	LC50	6,300	Cardwell et al. 1976a,b
Goldfish, *Carassius auratus*	Sodium selenite	-	10 days	Mortality	5,000	Ellis 1937; Ellis et al. 1937
Goldfish, *Carassius auratus*	Sodium selenite	-	46 days	Gradual anorexia and mortality	2,000	Ellis et al. 1937
Goldfish, *Carassius auratus*	Selenium dioxide	-	7 days	LC50	12,000	Weir and Hine 1970
Goldfish, *Carassius auratus*	Selenium dioxide	-	48 hr	Conditional avoidance	250	Weir and Hine 1970
Fathead minnow, *Pimephales promelas*	Selenium dioxide	157	9 days	LC50	2,100	Cardwell et al. 1976a,b
Fathead minnow, *Pimephales promelas*	Sodium selenite	329	96 hr	LC50 (fed)	1,000	Halter et al. 1980
Fathead minnow, *Pimephales promelas*	Sodium selenite	329	14 days	LC50 (fed)	600	Halter et al. 1980
Fathead minnow, *Pimephales promelas*	Selenious acid	220[d]	8 days	LC50 (fed)	420	Kimball, Manuscript
Creek chub, *Semotilus atromaculatus*	Selenium dioxide	-	48 hr	Mortality	▾12,000	Kim et al. 1977

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
Bluegill, *Lepomis macrochirus*	Sodium selenite	318	48 days	LC50	400	Adams 1976
Bluegill, *Lepomis macrochirus*	Selenium dioxide	157	14 days	LC50	12,500	Cardwell et al. 1976a,b
Bluegill (juvenile), *Lepomis macrochirus*	Sodium selenite	16	323 days	MATC larval survival (dietary only exposure)	19.75 µg Se/g dw (food)	Woock et al. 1987
Bluegill (juvenile), *Lepomis macrochirus*	Sodium selenite	25 and 200	120 days	No mortality	>10	Lemly 1982
Largemouth bass (juvenile), *Micropterus salmoides*	Sodium selenite	25 and 200	120 days	No mortality	10	Lemly 1982
Yellow perch, *Perca flavescens*	Sodium selenite	10.2	10 days	LC50	4,800	Klaverkamp et al. 1983a,b
African clawed frog, *Xenopus laevis*	Sodium selenite	-	7 days	LC50	1,520	Browne and Dumont 1980
African clawed frog, *Xenopus laevis*	Sodium selenite	-	1-7 days	Cellular damage	2,000	Browne and Dumont 1980
			Selenium (VI)			
Alga, *Chrysochromulina breviturrita*	-	-	30 days	Increased growth	50	Wehr and Brown 1985
Snail, *Lymnaea stagnalis*	Sodium selenate	-	6 days	LT50	15,000	Van Puymbroeck et al. 1982
Cladoceran, *Daphnia magna*	Sodium selenate	129.5	7 days	LC50 (fed)	1,870	Dunbar et al. 1983
Cladoceran (juvenile), *Daphnia magna*	Sodium selenate	-	48 hr	LC50 (fed)	550	Johnston 1987
Cladoceran (5th instar), *Daphnia magna*	Sodium selenate	-	48 hr	LC50 (fed)	750	Johnston 1987

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
Cladoceran (5th instar), *Daphnia magna*	Sodium selenate	-	90 hr	42% of organ-isms had visible changes in gut morphology	250	Johnston 1989
Amphipod (2 mm length), *Hyalella azteca*	Sodium selenate	133	48 hr	LC50	2378	Brasher and Ogle 1993
Amphipod (2 mm length), *Hyalella azteca*	Sodium selenate	133	10 days	LC50 (fed)	627	Brasher and Ogle 1993
Amphipod (2 mm length), *Hyalella azteca*	Sodium selenate	133	24 days	LOEC reproduction (static renewal)	>700	Brasher and Ogle 1993
Midge (first instar), *Chironomus riparius*	Sodium selenate	134	48 h	LC50	16,200	Ingersoll et al. 1990
Midge (first instar), *Chironomus riparius*	Sodium selenate	40-48	48 h	LC50	10,500	Ingersoll et al. 1990
Rainbow trout (embryo, larva), *Oncorhynchus mykiss*	Sodium selenate	104 (92-110)	28 days	EC50 (death and deformity)	5,000 (4,180) (5,170)	Birge 1978; Birge and Black 1977; Birge et al. 1980
Goldfish (embryo, larva), *Carrassius auratus*	Sodium selenate	195	7 days	EC50 (death and deformity)	8,780	Birge 1978
Goldfish, *Carassius auratus*	Sodium selenate	-	24 hr	BCF = 1.42 BCF = 1.15 BCF = 1.47 BCF = 0.88 BCF = 1.54	0.45 0.9 1.35 2.25 4.5	Sharma and Davis 1980
Fathead minnow, *Pimephales promelas*	Sodium selenate	337.9	48 days	LC50	2,000	Adams 1976
Fathead minnow, *Pimephales promelas*	Sodium selenate	338	48 days	LC50	1,100	Adams 1976
Fathead minnow, *Pimephales promelas*	-	51	30 min	No avoidance	11,200	Watenpaugh and Beitinger 1985a
Fathead minnow, *Pimephales promelas*	-	-	24 hr	LC50	82,000	Watenpaugh and Beitinger 1985b

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
Fathead minnow, *Pimephales promelas*	-	-	24 hr	Reduced thermal tolerance	22,200	Watenpaugh and Beitinger 1985c
Fathead minnow, *Pimephales promelas*	Sodium selenate	44-49	7 days	Chronic value - growth Chronic value-growth Chronic value-survival	1,739 561 2,000	Norberg-King 1989
Fathead minnow, *Pimephales promelas*	Sodium selenate	160-180	24 hr	No effect on reproductive behavior	36,000	Pyron and Beitinger 1989
Fathead minnow, *Pimephales promelas*	Sodium selenate	160-180	24 hr	Edema in larvae produced from adults exposed to Selenium VI	20,000	Pyron and Beitinger 1989
Channel catfish (embryo, fry), *Ictalurus punctatus*	Sodium selenate	90	8.5-9 days	Induced albinism	-	Westerman and Birge 1978
Narrow-mouthed toad (embryo, larva), *Gastrophryne carolinensis*	Sodium selenate	195	7 days	EC50 (death and deformity)	90	Birge 1978; Birge and Black 1977; Birge et al. 1979a
			Organo-selenium			
Bluegill (juvenile), *Lepomis macrochirus*	Seleno-L-methionine	16	323 days	MATC larval survival (dietary only exposure)	20.83 µg Se/g dw (food)	Woock et al. 1987
Bluegill (juvenile), *Lepomis macrochirus*	Seleno-L-methionine	283	90 days	EC20 survival (dietary only exposure)	>13.4 µg/g dw (food)	Cleveland et al. 1993
Bluegill (2 yr and adult), *Lepomis macrochirus*	Selenium	-	field	NOEC deformities	53.83 µg Se/g dw (liver)	Reash et al. 1999
Bluegill (2 yr and adult), *Lepomis macrochirus*	Selenium	-	field	NOEC deformities	23.38 µg Se/g dw (ovaries)	Reash et al. 1999

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Hardness (mg/L as $CaCO_3$)	Duration	Effect	Concentration[a]	Reference
Redear sunfish (adult), *Lepomis microlophus*	Selenium	-	field	LOEC Adverse histopathological alterations	<38.15 µg Se/g dw	Sorensen 1988
			Selenium Mixtures			
Phytoplankton, Mixed population	Selenium	-	field	Reduced growth rates	18	Riedel et al. 1991
Cladoceran (<24 hr), *Daphnia magna*	Selenite-Selenate mixture	138	21 days	MATC growth	115.2 µg Se/L	Ingersoll et al. 1990
Cladoceran (<24 hr), *Daphnia magna*	Selenite-Selenate mixture	138	21 days	MATC productivity	21.59 Og/g dw (whole-body)	Ingersoll et al. 1990
Midge (<24-hr), *Chironomus riparius*	Selenite-Selenate mixture	138	30 days	MATC emergence	503.6	Ingersoll et al. 1990
Bluegill (juvenile), *Lepomis macrochirus*	Selenite-Selenate mixture	283	60 days	NOEC survival	340	Cleveland et al. 1993
Bluegill (juvenile), *Lepomis macrochirus*	Selenite-Selenate mixture	283	60 days	EC20 survival	4.07 µg/g dw (whole body)	Cleveland et al. 1993

Species	Chemical	Salinity (g/kg)	Duration	Effect	Concentration (ug/L)[a]	Reference
			SALTWATER SPECIES			
			Selenium (IV)			
Anaerobic bacterium, *Methanococcus vannielli*	Sodium selenite	-	110 hr	Stimulated growth	79.01	Jones and Stadtman 1977
Bacterium, *Vibrio fisheri*	Sodium selenite	-	5 min	50% decrease in light output (Microtox®)	68,420	Yu et al. 1997
Green alga, *Chlorella* sp.	Sodium selenite	32	14 days	5-12% increase in growth	10-10,000	Wheeler et al. 1982

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Salinity (g/kg)	Duration	Effect	Concentration (ug/L)[a]	Reference
Green alga, *Platymonas subcordiformis*	Sodium selenite	32	14 days	23% increase in growth	100-10,000	Wheeler et al. 1982
Green alga, *Dunaliella primolecta*	Sodium selenite	32	20 days	Increased growth; induced glutathione peroxidase	4,600	Gennity et al. 1985a,b
Diatom, *Skeletonema costatum*	Selenium dioxide	-	5 days	BCF = 18,000 BCF = 16,000 BCF = 10,000	0.06 0.79 3.6	Zhang et al. 1990
Diatom, *Chaetoceros muelleri*	Selenium dioxide	-	6 days	BCF = 337,000 BCF = 65,000 BCF = 5,000	0.06 0.79 3.6	Zhang et al. 1990
Diatom, *Phaeodactylum tricornutum*	Selenium dioxide	-	8 days	BCF = 109,000 BCF = 27,000 BCF = 7,000	0.06 0.79 3.6	Zhang et al. 1990
Diatom, *Thallassiosira aestivalis*	Selenium oxide	29-30	72 hr	No effect on cell morphology	78.96	Thomas et al. 1980a
Brown alga, *Fucus spiralis*	Sodium selenite	-	60 days	1355% increase in growth of thalli	2.605	Fries 1982
Red alga, *Porphyridium cruentum*	Sodium selenite	32	27 days	Increase growth; induced glutathione peroxidase	4,600	Gennity et al. 1985a,b
			Selenium (VI)			
Bacterium, *Vibrio fisheri*	Sodium selenate	-	15 min	50% decrease in light output (Microtox®)	3,129,288	Yu et al. 1997
Green alga, *Chlorella* sp.	Sodium selenate	32	14 days	No effect on rate of cell	10-1,000	Wheeler et al. 1982
Green alga, *Chlorella* sp.	Sodium selenate	32	4-5 days	100% mortality	10,000	Wheeler et al. 1982
Green alga, *Dunaliella primolecta*	Sodium selenate	32	14 days	No effect on rate of cell population growth	10-100	Wheeler et al. 1982
Green alga, *Dunaliella primolecta*	Sodium selenate	32	14 days	71% reduction in rate of cell population growth	1,000	Wheeler et al. 1982
Green alga, *Dunaliella primolecta*	Sodium selenate	32	4-5 days	100% mortality	10,000	Wheeler et al. 1982

Table F-1. Other Data on Effects of Selenium on Aquatic Organisms (continued)

Species	Chemical	Salinity (g/kg)	Duration	Effect	Concentration (ug/L)[a]	Reference
Green alga, *Platymonas subcordiformis*	Sodium selenate	32	14 days	No effect on rate of cell population growth	10	Wheeler et al. 1982
Green alga, *Platymonas subcordiformis*	Sodium selenate	32	14 days	16% decrease in rate of cell population growth	100	Wheeler et al. 1982
Green alga, *Platymonas subcordiformis*	Sodium selenate	32	14 days	50% decrease in rate of cell population growth	1,000	Wheeler et al. 1982
Green alga, *Platymonas subcordiformis*	Sodium selenate	32	4-5 days	100% mortality	10,000	Wheeler et al. 1982
Brown alga, *Fucus spiralis*	Sodium selenate	-	60 days	160% increase in growth rate of thalli	2.605	Fries 1982
Red alga, *Porphridium cruentum*	Sodium selenate	32	14 days	23-35% reduction in rate of cell population growth	10-1,000	Wheeler et al. 1982
Red alga, *Porphyridium cruentum*	Sodium selenate	32	4-5 days	100% mortality	10,000	Wheeler et al. 1982
Eastern oyster (adult), *Crassostrea virginica*	Sodium selenate	34	14 days	No significant effect on respiration rate of gill tissue	400	Fowler et al. 1981
Striped bass (embryo), *Morone saxatilis*	Sodium selenate	7.2-7.5	4 days	93% successful hatch and survive	200,000	Klauda 1985a,b
Striped bass (larva), *Morone saxatilis*	Sodium selenate	4.0-5.0	4 days	LC50 (control survival= 77%)	13,020	Klauda 1985a,b
Striped bass (juvenile), *Morone saxatilis*	Sodium selenate	3.5-5.5	9-65 days	Significant incidence of development anomalies of lower jaw	39-1,360	Klauda 1985a,b
Striped bass (juvenile), *Morone saxatilis*	Sodium selenate	3.5-5.5	45 days	Significant incidence of severe blood cytopathology	1,290	Klauda 1985a,b

[a] Concentration of selenium, not the chemical. Units are µg selenium/L of water unless noted otherwise.
[b] Converted from dry weight to wet weight basis (see Guidelines)
[c] Growth of algae was inhibited
[d] From Smith et al. (1976).
[e] Calculated from the published data using probit analysis and allowing for 8.9% spontaneous mortality.

Other Data - Endangered Species

Two similar studies were conducted in subsequent years, 1996 and 1997, to determine the effects of site water and site food contaminated with selenium on the endangered species, razorback sucker, *Xyrauchen texanus* (Hamilton et al. 2001a,b). Both studies show marked effects on the survival of razorback sucker larvae exposed to contaminated food and to a lesser extent, contaminated water. Although the data convincingly demonstrate effects to larval survival from exposure to contaminated food, it was not considered acceptable data for use in the derivation of the chronic criterion because of inconsistencies between levels of selenium in the food and larvae and degree and time to response. A summary of each of these two studies is presented below.

Evaluation of Contaminant Impacts on Razorback Sucker held in Flooded Bottomland Sites Near Grand Junction , Colorado - 1996 (Hamilton et al. 2001a)

This study was initiated with 5-day old razorback sucker larvae spawned from adults which were previously held (9 months) in three different location along the Colorado River that contained varying levels of selenium: Horsethief (the designated reference site which receives water pumped directly from the Colorado River near Fruita, CO), Adobe Creek (low level selenium contamination), and North Pond (high level selenium contamination). The selenium content in the eggs from three Horsethief females ranged from 5.8 to 6.6 µg Se/g dw, and the selenium content in adult muscle plugs at spawning was from 3.4 to 5.0 µg Se/g dw. The selenium content in the eggs from three Adobe Creek females ranged from 38.0 to 54.5 µg Se/g dw, and the selenium content in adult muscle plugs at spawning was from 11.5 to 12.9 µg Se/g dw. The selenium content in the eggs from three North Pond females ranged from 34.3 to 37.2 µg Se/g dw, and the selenium content in adult muscle plugs at spawning was from 14.1 to 17.3 µg Se/g dw. The selenium content in the eggs from a hatchery brood stock female was 7.1 µg Se/g dw, and the selenium content in adult muscle plugs at spawning ranged from 2.6 to 13.8 µg Se/g dw. The razorback sucker larvae spawned from fish hatchery brood stock and held in Colorado River (Horsethief) water were used as an additional reference group of test fish.

The experimental groups were subdivided into those receiving reference water (hatchery water; 24-Road Fish Hatchery) or site water. They were further subdivided into those receiving a daily ration of reference food (brine shrimp) or zooplankton collected from each site where their parents were exposed for the previous 9 months. A total of 60 larvae from each of the four adult sources (Horsethief, Adobe Creek, North Pond,

Brood Stock) were exposed to each treatment (2 replicates x 3 spawns x 10 fish/beaker). The larvae were held in beakers containing 800 mls of test water. Fifty percent of the test water was renewed daily.

Treatment conditions during the 30-day larval study:

Source of Larvae	**Treatments**	**Se in food (µg/g dw)**	**Se in water (µg/L)**
Horsethief Adults	Reference food: Reference water	2.7	< 1
	Reference food: Site water	2.7	0.9
	Site food: Reference water	5.6	< 1
	Site food: Site water	5.6	0.9
Adobe Creek Adults	Reference food: Reference water	2.7	< 1
	Reference food: Site water	2.7	5.5
	Site food: Reference water	20	< 1
	Site food: Site water	20	5.5
North Pond Adults	Reference food: Reference water	2.7	< 1
	Reference food: Site water	2.7	10.7
	Site food: Reference water	39	<1
	Site food: Site water	39	10.7
Hatchery raised Adults	Reference food: Reference water	2.7	< 1
	Reference food: Site water	2.7	0.9
	Site food: Reference water	5.6	< 1
	Site food: Site water	5.6	0.9

Growth, survival and development were evaluated amongst treatment groups for up to 30 days in the treatment conditions. Each treatment group was fed once daily after renewal. Test waters were collected every day from each site as grab samples for the renewal. A small portion of this water was retained at 3- and 7-day intervals for an analysis of total and dissolved selenium concentrations. At approximately 2-day intervals, aquatic invertebrates and brine shrimp not used for feeding were sieved from the media for selenium analysis. The number of live fish were recorded daily. After the 30-day exposure period, the surviving fish were

sacrificed and measured for total length. At this same time, approximately four fish from each treatment, when available, were collected as a composite sample and analyzed for selenium. Specific treatment conditions were as those described above.

After 30 days of exposure in the reference food-reference water treatment, survival of razorback sucker larvae from brood stock and Horsethief adults (89 and 87 percent, respectively) was slightly higher than those from Adobe Creek adults (84 percent) and North Pond adults (75 percent). Corresponding selenium concentrations in larval whole-body tissue were 3.6, 3.3, 7.7 and 9.7 µg Se/g dw, respectively. Survival was similar or slightly reduced in larvae from all four sources after 30 days of exposure in the reference food-site water treatments; corresponding selenium concentrations in larval whole-body tissue were 5.2, 5.1, 12.7 and 15.2 µg Se/g dw, respectively. In contrast, none of the larvae spawned from parents from Horsethief, Adobe Creek, or North Pond survived to 30 days when fed zooplankton collected from the three sites, irrespective of the water type they were exposed to (i.e., reference or site). Only the larvae from brood stock adults, which were fed zooplankton from the Horsethief site for this treatment, survived, and even these larvae suffered substantial mortality (40 and 60 percent respectively). The mean selenium concentrations in whole-body tissue of larvae from brood stock adults after the 30-day exposures were 5.4 µg Se/g dw (site food-reference water treatment) and 6.9 µg Se/g dw (site food-site water treatment).

Several inconsistencies were observed that suggest selenium may not be solely responsible for the effect on larval survival. Larval survival in the Adobe Creek treatment group exposed to reference water and reference food was 84 percent, similar to control survival (86-89 percent). The selenium concentration in the larvae from this Adobe Creek treatment group after 30 days was higher (7.7 µg/g dw) than brood stock fish (5.4 µg Se/g dw) which had a lower 30-day survival (62 percent). Also, the time to 50 percent mortality between the site food treatments, where most mortality occurred, was not related to selenium concentration in the diet or in the larvae.

Evaluation of Contaminant Impacts on Razorback Sucker held in Flooded Bottomland Sites Near Grand Junction , Colorado - 1997 (Hamilton et al. 2001b)

In a similar 30-day larval study conducted by the authors thein following year (1997), razorback sucker larvae from a single hatchery brood stock female (11 µg Se/g dw muscle) were subjected to one of the sixteen different combined water and dietary exposure conditions described in the earlier (1996) study. The female parent was held at Horsethief Canyon State Wildlife Area before spawning. The larvae were held in beakers containing 800 mls of test water as before, fifty percent of the test water was renewed daily. Specific treatment conditions for the 1997 30-day larval study were as follows:

Treatment conditions during the 30-day larval study:

Water Treatments	**Se in food (µg/g dw)**	**Se in water (µg/L)**
Reference food (brine shrimp): Reference water (24-Road Hatchery)	3.2	< 1
Reference food: Site water (Horsethief)	6.0	1.6
Reference food: Site water (Adobe Creek)	32.4	3.4
Reference food: Site water (North Pond)	52.5	13.3
Horsethief food: Reference water	3.2	< 1
Horsethief food: Site water (Horsethief)	6.0	1.6
Horsethief food: Site water (Adobe Creek)	32.4	3.4
Horsethief food: Site water (North Pond)	52.5	13.3
Adobe Creek food: Reference water	3.2	< 1
Adobe Creek food: Site water (Horsethief)	6.0	1.6
Adobe Creek food: Site water (Adobe Creek)	32.4	3.4
Adobe Creek food: Site water (North Pond)	52.5	13.3
North Pond food: Reference water	3.2	< 1
North Pond food: Site water (Horsethief)	6.0	1.6
North Pond food: Site water (Adobe Creek)	32.4	3.4
North Pond food: Site water (North Pond)	52.5	13.3

In this year's study, after 30 days of exposure, there was also good survival of razorback sucker larvae fed reference food (brine shrimp) and held in reference water or water from Horsethief (83 and 81 percent, respectively). The survival of these larvae was significantly greater than survival of larvae fed brine shrimp and held in water from North Pond (only 52 percent). Corresponding selenium concentrations in larval whole-body tissue after 10 days were 6.3, 6.7, and 11 µg Se/g dw, respectively. The average concentrations of selenium in the water for the three treatments were <1, 1.6, and 13.3 µg Se/L. After 30 days the mean selenium concentrations in these larvae were 5.2, 5.2, and 16 µg Se/g dw, respectively. Survival was markedly reduced (0 to 30 percent survival) in the remainder treatments where larvae were fed zooplankton from the various sites. Complete mortality was experienced by larvae exposed to Horsethief food and reference water treatment after 30 days.

Similar to the previous study, there are several inconsistencies in the results that suggested selenium may not be solely responsible for the effect on larval survival. The most notable inconsistency was that the greatest effect on larval survival (percent survival or time to 50 percent mortality) was from exposure to Horsethief food, the food with the lowest selenium contamination.

APPENDIX G

UNUSED DATA

Unused Data

Based on the requirements set forth in the guidelines (Stephen et al. 1985) the following studies are not acceptable for the following reasons and are classified as unused data.

Studies Were Conducted with Species That Are Not Resident in North America

Ahsanullah and Brand (1985)
Ahsanullah and Palmer (1980)
Baker and Davies (1997)
Barghigiani et al. (1993)
Chidambaram and Sastry (1991a,b)
Congiu et al. (1989)
Cuvin and Furness (1988)
Fowler and Benayoun (1976a,b)
Gaikwad (1989)
Gotsis (1982)
Hiraika et al. (1985)
Juhnke and Ludemann (1978)
Kitamura (1990)
Manoharan and Prabakaran (1994)
Minganti et al. (1994, 1995)
Niimi and LaHam (1975, 1976)
Regoli (1998)
Regoli and Principato (1995)
Rhodes et al. (1994)
Ringdal and Julshamn (1985)
Rouleau et al. (1992)
Sastry and Shukla (1994)
Savant and Nilkanth (1991)
Shultz and Ito (1979)
Srivastava and Tyagi (1985)
Takayanagi (2001)
Tomasik et al. (1995b)
Tian and Liu (1993)
Wrench (1978)

Deelstra et al. (1989), Forsythe and Klaine (1994), Okasako and Siegel (1980) and Petrucci et al. (1995) conducted tests with brine shrimp species that are too atypical to be used in derving national criteria.

These Reviews Only Contain Data That Have Been Published Elsewhere

Adams and Johnson (1981)
Biddinger and Gloss (1984)
Bowie et al. (1996)
Brandao et al. (1992)
Brooks (1984)
Burton and Stemmer (1988)
Chapman et al. (1986)
Davies (1978)
Devillers et al. (1988)
Eisler (1985)
Hall and Burton (1982)
Hodson and Hilton (1983)
Hodson et al. (1984)
Jenkins (1980)
Kaiser et al. (1997)
Kay (1984)
LeBlanc (1984)
Lemly (1993c, 1996ab, 1997d)
Lemly and Smith (1987)
McKee and Wolf (1963)
National Research Council (1976)
Neuhold (1987)
NCDNR&CD (1986)
Peterson and Nebeker (1992)
Phillips and Russo (1978)
Presser (1994)
Roux et al. (1996)
Thompson et al. (1972)
Versar (1975)

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Authors Did Not Specify the Oxidation State of Selenium Used in Study

Greenberg and Kopec (1986)
Hutchinson and Stokes (1975)
Kapu and Schaeffer (1991)
Kramer et al. (1989)
Mahan et al. (1989)
Rauscher (1988)
Snell et al. (1991b)

Selenium Was a Component of an Effluent, Fly Ash, Formulation, Mixture, Sediment or Sludge

Apte et al. (1987)
Baer et al. (1995)
Baker et al. (1991)
Berg et al. (1995)
Besser et al. (1989)
Biedlingmaier and Schmidt (1989)
Bjoernberg (1989)
Bjoernberg et al. (1988)
Blockmann et al. (1995)
Boisson et al. (1989)
Bondavalli et al. (1996)
Bowmer et al. (1994)
Brieger et al. (1992)
Burton and Pinkney (1984)
Burton et al. (1983, 1987)
Cherry et al. (1987)
Cieminski and Flake (1995)
Clark et al. (1989)
Cooke and Lee (1993)
Cossu et al. (1997)
Coyle et al. (1993)
Crane et al. (1992)
Crock et al. (1992)
Cushman et al. (1977)
Davies and Russell (1988)
de Peyster et al. (1993)
Dickman and Rygiel (1996)
Dierenfeld et al. (1993)
Drndarski et al. (1990)
Eriksson and Forsberg (1992)
Eriksson and Pedros-Alio (1990)
Fairbrother et al. (1994)
Fava et al. (1985a,b)
Feroci et al. (1997)
Finger and Bulak (1988)
Finley (1985)
Fisher and Wente (1993)
Fjeld and Rognerud (1993)
Fletcher et al. (1994)
Follett (1991)
Gerhardt (1990)
Gerhardt et al. (1991)
Gibbs and Miskiewicz (1995)
Graham et al. (1992)
Gunderson et al. (1997)
Hall (1988)
Hall et al. (1984, 1987, 1988,1992)
Hamilton et al. (1986, 2000)
Harrison et al. (1990)
Hartwell et al. (1987ab, 1988, 1997)
Hatcher et al. (1992)
Haynes et al. (1997)
Hayward et al. (1996)
Hellou et al. (1996)
Henebry and Ross (1989)
Henry et al. (1989, 1990, 1995)
Hildebrand et al. (1976)
Hjeltner and Julshman (1992)
Hockett and Mount (1996)
Hodson (1990)
Hoffman et al. (1988, 1991)
Homziak et al. (1993)
Hopkins et al. (2000)
Hothem and Welsh (1994a)
Jackson (1988)
Jackson et al. (1990)
Jacquez et al. (1987)
Jay and Muncy (1979)
Jayasekera (1994)
Jayasekera and Rossbach (1996)
Jenner and Bowmer (1990) (1992)
Jenner and Janssen-Mommen (1989)
Jin et al. (1997)
Jorgensen and Heisinger (1987)
Karlson and Frankenberger (1990)
Kemble et al. (1994)
Kenned (1986)
Kersten et al. (1991)
King and Cromartie (1986)
King et al. (1991, 1994)
Klusek et al. (1993)
Koh and Harper (1988)
Koike et al. (1993)
Krishnaja et al. (1987)
Kruuk and Conroy (1991)
Kuehl and Haebler (1995)
Kuehl et al. (1994)
Kuss et al. (1995)
Landau et al. (1985)
Livingstone et al. (1991)
Lobel et al. (1990)

Luoma and Phillips (1988)
Lundquist et al. (1994)
Lyle (1986)
MacFarlane et al. (1986)
Mann and Fyfe (1988)
Marcogliese et al. (1987)
Marvin et al. (1997)
Maurer et al (1999)
McCloskey and Newman (1995)
McCloskey et al. (1995)
McCrea and Fischer (1986)
McLean et al. (1991)
Mehrle et al. (1987)
Metcalf-Smith (1994)
Micallef and Tyler (1989)
Mikac et al. (1985)
Miles and Tome (1997)
Miller et al. (1996)
Misitano and Schiewe (1990)
Moore (1988)
Munawar and Legner (1993)
Muskett et al. (1985)
Naddy et al. (1995)
Nielsen and Bjerregaard (1991)
Norman et al. (1992)
Nuutinen & Kukkonen (1998)
Oberbach and Hartfield (1987, 1988)
Oberbach et al. (1989)
Ohlendorf et al. (1989, 1990, 1991)
Olsen and Welsh (1993)
Peters et al.(1999)
Phillips and Gregory (1980)
Pratt and Bowers (1990)
Presser and Ohlendorf (1987)
Prevot and Sayer-Gobillard (1986)
Pritchard (1997)
Pyleet al. (2001)
Reash et al. (1988, in press)
Rhodes and Burke (1996)
Ribeyre et al. (1995)
Rice et al. (1995)
Riggs and Esch (1987)
Riggs et al. (1987)
Robertson et al. (1991)
Roper et al. (1997)
Russell et al. (1994)
Ryther et al. (1979)
Saiki and Jenings (1992)
Saiki and Ogle (1995)
Saleh et al. (1988)
Seelye et al. (1982)
Sevareid and Ichikawa (1983)
Skinner (1985)
Somerville et al. (1987)
Sorenson and Bauer (1983)
Specht et al. (1984)
Steele et al. (1992)
Stemmer et al. (1990)
Summers et al. (1995)
Thomas et al. (1980b)
Timothy et al. (2001)
Trieff et al. (1995)
Turgeon and O'Conner (1991)
Twerdok et al. (1997)
Ursal (1987)
Van Metre and Gray (1992)
Wahl et al. (1994)
Wandan and Zabik (1996)
Wang et al. (1992, 1995)
Welsh (1992)
Weres et al. (1990)
White and Geitner (1996)
Wiemeyer et al. (1986)
Wildhaber and Schmitt (1996)
Williams et al. (1989)
Wolfe et al. (1996)
Wolfenberger (1987)
Wong and Chau (1988)
Wong et al. (1982)
Wu et al. (1997)
Yamaoka et al. (1994)
Zagatto et al. (1987)
Zaidi et al. (1995)
Zhang et al. (1996)

Exposed enzymes, excised tissue or tissue extractor

Tripathi and Pandey (1985) and Heinz (1993b) used test organisms that had been previously exposed to pollutants in food or water.

Albers et al. (1996)
Al-Sabti (1994, 1995)
Arvy et al. (1995)
Augier et al. (1993)
Avery et al. (1996)
Baatrup (1989)
Baatrup and Dansher (1987)
Baatrup et al. (1986)
Babich et al. (1986, 1989)

Barrington et al. (1997)
Becker et al. (1995a,b)
Bell et al. (1984, 1985, 1986a,b, 1987ab)
Berges and Harrison (1995)
Blondin et al. (1988)
Boisson et al. (1996)
Bottino et al. (1984)
Braddon (1982)
Braddon-Galloway and Balthrop (1985)
Bradford et al. (1994a,b)
Brandt et al. (1990)
Byl et al. (1994)
Chandy and Patel (1985)
Chen et al. (1997)
Cheng et al. (1993)
Christensen and Tucker (1976)
Dabbert and Powell (1993)
DeQuiroga et al. (1989)
Dierickx (1993)
Dietrich et al. (1987)
DiIlio et al. (1986)
Doyotte et al. (1997)
Drotar et al. (1987)
Dubois and Callard (1993)
Ebringer et al. (1996)
Engberg and Borsting (1994)
Engberg et al. (1993)
Eun et al. (1993)
Foltinova and Gajdosova (1993)
Foltinova et al. (1994)
Freeman and Sanglang (1977)
Grubor-Lajsic et al. (1995)
Hait and Sinha (1987)
Hanson (1997)
Heisinger and Scott (1985)
Heisinger and Wail (1989)
Henderson et al. (1987)
Henny and Bennett (1990)
Hoffman and Heinz (1988, 1998)
Hoffman et al. (1989, 1998)
Hontela et al. (1995)
Hoglund (1991)
Hsu et al. (1995)
Hsu and Goetz (1992)
Ishikawa et al. (1987)
James et al. (1993)
Jovanovic et al. (1995, 1997)
Kai et al. (1995)
Kedziroski et al. (1996)
Kelley et al. (1987)
Kralj and Stunja (1994)
Lalitha and Rani (1995)
Lan et al. (1995)
Lemaire et al. (1993)
Livingstone et al. (1992)
Low and Sin (1995, 1996)
Micallef and Tyler (1990)
Montagnese et al. (1993)
Murata et al. (1996)
Nakonieczny (1993)
Neuhierl and Boeck (1996)
Nigro et al (1992, 1994)
Norheim and Borch-Iohnsen (1990)
Norheim et al. (1991)
O'Brien et al. (1995)
Olson and Christensen (1980)
Overbaugh and Fall (1985)
Palmisano et al. (1995)
Patel et al. (1990)
Patel and Chandy (1987)
Perez et al. (1990)
Perez-Trigo et al. (1995)
Phadnis et al. (1988)
Price and Harrison (1988)
Rady et al. (1992)
Rani and Lalitha (1996)
Regoli et al. (1997)
Schmidt et al. (1985)
Schmitt et al. (1993)
Segner et al. (1994)
Sen et al. (1995)
Shigeoka et al. (1990, 1991)
Siwicki et al. (1994)
Srivastava and Srivastava (1995)
Sun et al. (1995)
Takeda et al. (1992a,b,(1993, 1997)
Treuhardt (1992)
Vazquez et al. (1994)
Veena et al. (1997)
Wise et al. (1993a,b)
Wong and Oliveira (1991)
Yokota et al. (1988)

Test procedures test material or results were not adequately described by Botsford (1997), Botsford et al. (1997, 1998), Bovee (1978), Gissel-Nielsen and Gissel-Nielsen (1973, 1978), Greenberg and Kopec (1986), Mauk (2001), and Nassos et al. (1980) or when the test media contained an excessive amount (>200 Og/L) of EDTA (Riedel and Sanders (1996).

Some data obtained from tests conducted with just one exposure concentration to evaluate acute or chronic toxicity were not used (e.g., Bennett 1988; Heinz and Hoffman 1998; Munawar et al. 1987; Pagano et al. 1986; Wolfenberger 1986).

Kaiser (1980) calculated the toxicities of selenium(IV) and selenium(VI) to *Daphnia magna* based on physiochemical parameters. Kumar (1964) did not include a control treatment in the toxicity tests. The daphnids were probably stressed by crowding in the tests reported be Schultz et al. (1980). Siebers and Ehlers (1979) exposed too few test organisms as did Owsley (1984) in some tests.

Data Were Not Used When the Organisms Were Exposed to Selenium by Food or by Gavage or Injection

Frankenberger and Engberg (in press)
Hamilton (1999)
Hamilton and Lemly (1999)
Heinz and Sanderson (1990)
Heinz et al. (1990, 1996)
Hilton et al. (1982)
Hoffman and Heinz (1988)
Hoffman et al. (1991, 1992a,b,1996)
Huerkamp et al. (1988)
Julshamn et al. (1990)
Kleinow (1984)
Kleinow and Brooks (1986a,b)
Lemly (1996, 1997, 1999)
Lorentzen et al. (1994)
Maage and Waagboe (1990)
Malchow et al. (1995)
Paripatananontand Lovell(1997)
Sheline and Schmidt-Nielson (1977)
Stanley et al. (1994, 1996)
Wiemeyer and Hoffman (1996)
Wilson et al. (1997)

BCFs and BAFs from laboratory tests were not used when the tests were static or when the concentration of selenium in the test solution was not adequately measured or varied too much (Nassos et al. 1980; Ornes et al. 1991; Riedel et al. 1991; Sharma and Davis 1980; Vandermeulen and Foda 1988).

Selenium Concentrations Reported in Wild Aquatic Organisms Were Insufficient to Calculate BAF

Abdel-Moati and Atta (1991)
Adeloju and Young (1994)
Aguirre et al. (1994)
Akesson and Srikumar (1994)
Aksnes et al. (1983)
Allen and Wilson (1990)
Ambulkar et al. (1995)
Amiard et al. (1991, 1993)
Andersen and Depledge (1997)
Andreev and Simeonov (1992)
Angulo (1996)
Arrula et al. (1996)
Arway (1988)
Ashton (1991)
Augier et al. (1991, 1993, 1995a,b)
Augspurger et al. (1998)
Avery et al. (1996)
Badsha and Goldspink (1988)

Baines and Fisher (2001)
Baldwin and Maher (1997)
Baldwin et al. (1996)
Barghigiani (1993)
Barghigiani et al. (1991)
Baron et al. (1997)
Batley (1987)
Baumann and Gillespie (1986)
Baumann and May (1984)
Beal (1974)
Beck et al. (1997)
Beland et al. (1993)
Beliaeff et al. (1997)
Bell and Cowey (1989)
Benemariya et al. (1991)
Berry et al. (1997)
Bertram et al. (1986)
Besser et al. (1994, 1993)
Birkner (1978)
Boisson and Romeo (1996)
Bowerman et al. (1994)
Braune et a. (1991)
Brezina and Arnold (1977)
Brugmann and Hennings (1994)
Brugmann and Lange (1988)
Brumbaugh and Walther (1991)
Burger (1992, 1994, 1995, 1996, 1997a,b)
Burger and Gochfeld (1992a,b, 1993, 1995 ab, 1996, 1997)
Burger et al. (1992a,b,c,1993, 1994a,b)
Byrne and DeLeon (1986)
Byrne et al. (1985)
Cantillo et al. (1997)
Capar and Yess (1996)
Capelli et al. (1987, 1991)
Cappon (1984)
Cappon and Smith (1981)
(1982a,b)
Cardellicchio (1995)
Carell et al. (1987)
Carter and Porter (1997)
Caurant et al. (1994, 1996)
Chau and Riley (1965)
Chiang et al. (1994)
Chou and Uthe (1991)
Chvojka (1988)
Chvojka et al. (1990)
Clifford and Harrison (1988)
Collins (1992)
Combs et al. (1996)
Cosson et al. (1988)
Courtney et al. (1994)
Crowys et al. (1994)
Crutchfield (2000)
Cumbie and Van Horn (1978)
Currey et al. (1992)
Custer and Hohman (1994)
Custer and Mitchell (1991, 1993)
Custer et al. (1997)
Dabeka and McKenzie (1991)
Davoren (1986)
Deaker and Maher (1997)
Demon et al. (1988)
Dietz et al. (1995, 1996)
Doherty et al. (1993)
Elliott and Scheuhammer (1997)
Eriksson et al. (1989)
Evans et al. (1993)
Felton and Mathews (1990)
Felton et al. (1994)
Fitzsimmons et al. (1995)
Focardi et al. (1985, 1988)
Fowler (1986)
Fowler et al. (1975, 1985)
France (1987)
Friberg (1988)
Froslie et al. (1985, 1987)
Gabrashanske and Daskalova
(1985)
Gabrashanska and Nedeva (1994)
Galgan and Frank (1995)
Garcia - Hermandez et al. (2000)
Giardina et al. (1997)
Gillespie and Baumann (1986)
Gochfeld (1997)
Goede (1985, 1991, 1993a,b)
Goede et al. (1989, 1993)
Goede and DeBruin (1984, 1985)
Goede and Wolterbeek (1993, 1994a,b)
Gras et al. (1992)
Greig and Jones (1976)
Gutenmann et al. (1988)
Gutierrez-Galindo et al. (1994)
Guven et al. (1992)
Halbrook et al. (1996)
Hall and Fisher (1985)
Hamilton and Waddell (1994)
Hamilton and Wiedmeyer (1990)
Hansen et al. (1990)
Hardiman and Pearson (1995)
Hargrave et al. (1992)
Harrison and Klaverkamp (1990)
Hasunuma et al. (1993)
Haynes et al. (1995)
Hein et al. (1994)
Heiny and Tate (1997)
Heinz (1993a)
Heinz and Fitzgerald (1993a,b)
Heit (1985)
Heit and Klusek (1985)
Heit et al. (1980, 1989)

Hellou et al. (1992a,b) (1996a,b)
Henny and Herron (1989)
Hodge et al. (1996)
Hilton et al. (1982)
Honda et al. (1986)
Hothem and Ohlendorf (1989)
Hothem and Welsh (1994b)
Hothem and Zador (1995)
Hothem et al. (1995)
Houpt et al. (1988)
Hunter et al. (1995, 1997)
Ibrahim and Farrag (1992)
Ibrahim and Mat (1995)
Ishikawa et al. (1993)
Itano et al. (1984, 1985a,b)
Jarman et al. (1996)
Johns et al. (1988)
Johnson (1987)
Jop et al. (1997)
Jorhem et al. (1994)
Julshamn et al. (1987)
Kai et al. (1986a,b, 1988, 1992a,b, 1996)
Kaiser et al. (1979)
Kalas et al. (1995)
Kidwell et al. (1995)
Koeman et al. (1973)
Kovacs et al. (1984)
Krogh and Scanes (1997)
Krushevska et al. (1996)
Lakshmanan and Stephen (1994)
Lalitha et al. (1994)
LamLeung et al. (1991)
Lan et al. (1994a,b)
Langlois and Langis (1995)
Larsen and Stuerup (1994)
Larsen et al. (1997)
Lauchli (1993)
Law et al. (1996)
Lee and Fisher (1992a,b, 1993)
Leighton and Wobeser (1994)
Leland and Scudder (1990)
Lemly (1985a, 1994)
Leonzio et al. (1986, 1989, 1992)
Leskinen et al. (1986)
Li et al. (1996)
Lie et al. (1994)
Liu et al. (1987)
Lizama et al. (1989)
Lobel et al. (1989, 1991, 1992a,b)
Lonzarich et al. (1992)
Lourdes et al. (1990)
Lowe et al. (1985)
Lucas et al. (1970)
Lytle and Lytle (1982)
Mackey et al. (1996)
Maher (1987)
Maher et al. (1992, 1997)
Mann et al. (1988)
Mason et al. (2000)
Masuzawa et al. (1988)
Matsumoto (1991)
Maven et al. (1995)
May and McKinney (1981)
Mcdowell et al. (1995)
McKenzie-Parnell et al. (1988)
Meador et al. (1993)
Mehrle et al. (1982)
Meltzer et al. (1993)
Metcalfe-Smith et al. (1992, 1996)
Michot et al. (1994)
Mills et al. (1993)
Moharram et al. (1987)
Moller (1996)
Mora and Anderson (1995)
Morera et al. (1997)
Muir et al. (1988)
Mutanen et al. (1986)
Nadkarni and Primack (1993)
Nakamoto and Hassler (1992)
Narasaki and Cao (1996)
Navarrete et al. (1990)
Nettleton et al. (1990)
Nicola et al. (1987)
Nielsen and Dietz (1990)
Norheim (1987)
Norheim et al. (1992)
Norrgren et al. (1993)
Norstrom et al. (1986)
O'Conner (1996)
O'Shea et al. (1984)
Ober et al. (1987)
Oehlenschlager (1997)
Ohlendorf (1986)
Ohlendorf and Harrison (1986)
Ohlendorf and Maron (1990)
Ohlendorf et al. (1986a,b, 1987, 1988a,b)
Okazaki and Panietz (1981)
Ostapczuk et al. (1997)
Pakkala et al. (1972)
Pal et al. (1997)
Palawski et al. (1991)
Palmer-Locarnini and Presley (1995)
Paludan-Miller et al. (1993)
Papadopoulou and Andreotis (1985)
Park and Presley (1997)
Park et al. (1994)
Paveglio et al. (1994)
Payer and Runkel (1978)
Payer et al. (1976)
Pennington et al. (1982)

Presley et al. (1990)
Quevauviller et al. (1993a,b)
Ramos et al. (1992)
Rao et al. (1996)
Reinfelder and Fisher (1991)
Reinfelder et al. (1993, 1998)
Renzoni et al. (1986)
Riget et al. (1996)
Risenhoover (1989)
Roditi (2000)
Roux et al. (1994)
Ruelle and Keenlyne (1993)
Sager and Cofield (1984)
Saiki (1986 ab, 1987, 1990)
Saiki and Lowe (1987)
Saiki and May (1988)
Saiki and Palawski (1990)
Saiki et al. (1992, 1993)
Sanders and Gilmour (1994)
Scanes (1997)
Scheuhammer et al. (1988)
Schantz et al. (1997)
Schmitt and Brumbaugh (1990)
Schramel and Xu (1991)
Schuler et al. (1990)
Scott and Latshaw (1993)
Secor et al. (1993)
Seelye et al. (1982)
Sharif et al. (1993)
Shen et al. (1997)
Shirasaki et al. (1996)
Shultz and Ito (1979)
Simopoulos (1997)
Skaare et al. (1990, 1994)
Smith and Flegal (1989)
Smith et al. (1992)
Sorensen (1988)
Sorensen and Bauer (1984a,b)
Sorensen and Bjerregaard (1991)
Sorensen et al. (1982, 1983, 1984)
Southworth et al. (2000)
Sparling and Lowe (1996)
Speyer (1980)
Steimle et al. (1994)
Stoeppler et al. (1988)
Stone et al. (1988)
Stripp et al. (1990)
Sundarrao et al. (1991) (1992)
Svensson et al. (1992)
Tabaka et al. (1996)
Talbot and Chang (1987)
Tallandini et al. (1996)
Tan and Marshall (1997)
Tang et al. (1997)
Tao et al. (1993)
Teherani (1987)
Teigen et al. (1993)
Thomas et al. (1999)
Tilbury et al. (1997)
Topcuoglu et al. (1990)
TranVan and Teherani (1988)
Trocine and Trefry (1996)
Uthe and Bigh (1971)
Vanderstoep et al. (1990)
Varanasi et al. (1993, 1994)
Vitaliano and Zdanowicz (1992)
Vlieg (1990)
Vlieg et al. (1993)
Vos et al. (1986)
Waddell and May (1995)
Wagemann (1988)
Wagemann and Stewart (1994)
Wagemann et al. (1988) (1996)
Walsh et al. (1977)
Wang (1996)
Ward and Flick (1990)
Warren et al. (1990)
Weber (1985)
Welsh and Maughan (1994)
Wen et al. (1997)
Wenzel and Gabrielsen (1995)
Whyte and Boutillier (1991)
Williams et al. (1994)
Wilson et al. (1992, 1997)
Winger and Andreasen (1985)
Winger et al. (1984, 1990)
Woock and Summers (1984)
Wren et al. (1987)
Wu and Huang (1991)
Yamaoka et al. (1996)
Yamazaki et al. (1996)
Yoshida and Yasumoto (1987)
Zatta et al. (1985)
Zeisler et al. (1988, 1993)
Zhou and Liu (1997)

APPENDIX H

DATA USED IN REGRESSION ANALYSIS OF SELENIUM IN WHOLE-BODY FISH TISSUE WITH SELENIUM IN MUSCLE, OVARY AND LIVER TISSUES

DATA USED IN REGRESSION ANALYSIS OF SELENIUM IN WHOLE-BODY FISH TISSUE WITH SELENIUM IN MUSCLE, OVARY AND LIVER TISSUES

Quantile regression was used to estimate median concentrations of selenium in the whole body as a function of selenium concentration in selected tissues (Tables H-2, H-3, H-4). Only data where organisms were exposed to selenium in water and in diet or in only diet were considered for analysis. Quantile regression fits a curve to the data such that a selected proportion τ (the quantile) of observations are below and the complementary fraction 1- τ is above it (Koenker and Basset 1978). Estimates of model parameters minimize the sum of weighted absolute deviations. In contrast, ordinary least squares minimize the sum of squared deviations. Least absolute deviation is less sensitive to outliers than least squares (Birkes and Dodge 1993). Other desired properties of quantile regression include: it is equivariant to scale changes, location shift, and monotonic transformations (Koenker and Basset 1978, Cade et al. 1999). Furthermore, with rank-score statistics it is possible to test hypotheses and build confidence intervals for parameters of linear models fit to data with heteroscedastic errors (Koenker 1994, Koenker and Machado 1999). The rank-score test does not have to assume homogeneous error distributions because the statistic is based on signs of residuals and not their size (Koenker and Machado 1999). For introductory presentations of quantile regression see Cade et al. (1999), Koenker and Hallock (2001), and Cade & Noon (2003). All quantile regressions reported here were performed using the R software (Ihaka & Gentleman 1996) version 1.8.0.

As the exact form of the relationship between selenium concentrations in the whole body ($[Se]_{WB}$) and in tissues ($[Se]_{Tissue}$) is not known, we considered three candidate models:

I) $[Se]_{WB} = a$.

II) $[Se]_{WB} = a + b\,[Se]_{Tissue}$ and

III) $[Se]_{WB} = \exp(a + b \ln([Se]_{Tissue}))$

where a and b are the model parameters we wish to estimate. Model (I) implicitly assumes that selenium concentrations in the whole body are independent of selenium concentrations in liver, muscle, or ovary tissues. Model (II) projects selenium concentrations in the whole body as a linear function of selenium concentrations in a tissue. Model (III) estimates selenium concentrations in the whole body as an exponential function of the logarithm of selenium concentrations in a tissue. This model is derived from the assumption of a linear relationship between the natural logarithms of $[Se]_{WB}$ and $[Se]_{Tissue}$.

Selection of the best model(s) considers both the fit and number of parameters. Models with greater number of parameters generally fit the data better, but such reduction in bias is invariably associated with an increase in variance of parameter estimates (Burnham and Anderson 2002). Model selection methods attempt to find a parsimonious model with the proper tradeoff between bias and variance. We apply the information theoretic approach for model selection (Burnham and Anderson 2002). It is based on the Kullback-Leibler information, $I(f,g)$, which expresses the information lost when model g is used to estimate the full reality f. Obviously, the full reality is never known, but an estimate of the relative distance from reality can be estimated by the Akaike Information Criterion (AIC, Akaike 1973)

$$AIC = -2 \ln(\mathcal{L}(\text{parameters}|\text{data})) + 2k$$

where k is the number of parameters in the model and $\mathcal{L}$(parameters|data) is the maximized likelihood of parameter estimates for the available data.. The AIC is a poor estimator of $I(f,g)$ when $n/k < 40$ (n is the sample size). In such instances, a second-order version of AIC, AIC_c, is recommended (Hurvich and Tsai 1989):

$$AIC_c = -2 \ln(\mathcal{L}(\text{parameters}|\text{data})) + 2k\left(\frac{n}{n-k-1}\right)$$

Hurvich and Tsai (1990) demonstrated that the modified version of AIC_c for least absolute deviation($L1AIC_c$) provides an unbiased estimator for the Kullback-Leibler information, but the small sample criterion for normal least squares regression, which is less computationally demanding, performs equally well

$$\mathbf{AIC} = n\ln(\sigma^2) + 2k\left(\frac{n}{n-k-1}\right)$$

where σ^2 is estimated as the sum of squared residuals divided by n. For the least absolute deviation regression, σ^2 is estimated as $(SWAD/n)^2$, thus AICc is computed by the expression

$$\mathbf{AIC} = 2n\ln(\mathbf{SWAD}/n) + 2k\left(\frac{n}{n-k-1}\right)$$

The AIC and AIC_c are used to rank candidate models. Comparisons among the M ranked candidates are based on the Akaike weight (w), which represents the likelihood of a model given the data

$$w_i = \frac{\exp\left(\frac{-\Delta_i}{2}\right)}{\sum_{i=1}^{M}\exp\left(\frac{-\Delta_i}{2}\right)}$$

where Δ_i is the difference in AIC (AIC_c) between model i and the model with the lowest AIC (AIC_c) value. Weights for all candidate models sum to 1. For each model, we computed the sum of weighted absolute deviations (SWAD), AIC_c and the Akaike weight (Table H-1).

The linear model (II) was selected the best among the three candidate functions for projecting concentrations of selenium in the whole body as a function of selenium concentrations in the liver (Table H-1). The exponential model (III) was selected the best for projections based on concentrations of selenium in muscles and ovaries. However, fits of models II and III to ovary data had similar weights. As the best model may not explain much of the observed variation in the data, we calculated coefficients of determination (R^1), defined as

$$R^1 = 1 - (SAF/SAR)$$

where SAF and SAR are the sum of weighted absolute deviations for the full and reduced models, respectively (Cade and Richards 1996). Coefficients of determination for models II and III were also very similar, suggesting that both models are equally effective in predicting concentrations of selenium in the whole body as a function of selenium concentrations in ovaries. With such knowledge, we opted to use the linear model (II) because it is easier to compute. The exponential model for muscle presented the highest coefficient of determination (0.77), indicating that samples of selenium concentrations from this tissue are more effective predictors than samples from liver and ovaries. The fitted quantile regression curves are shown in figure 5 of the selenium document.

TableH- 1. Number of parameters (k), sum of weighted absolute deviations (SWAD), second-order Akaike Information Criterion (AIC_c), differences between the model AIC_c and the lowest AIC_c of all candidate models (Delta), weight (*w*), rank (by weight), and coefficient of determination (R^1) for three candidate models to project selenium concentrations in the whole body as a function of selenium concentrations in a selected tissue.

Tissue: Muscle (*n* = 21)

Model	k	SWAD	AICc	Delta	Weight	Rank	R^1
$[Se]_{WB} = a$	2	66.00	52.76	59.20	1.27e-13	3	
$[Se]_{WB} = a + b\,[Se]_{Tissue}$	3	16.84	-1.85	4.59	9.17e-02	2	0.74
$[Se]_{WB} = \exp(a + b*\ln([Se]_{Tissue}))$	3	15.10	-6.43	0.00	9.08e-01	1	0.77

Tissue: Ovary (*n* = 23)

Model	k	SWAD	AICc	Delta	Weight	Rank	R^1
$[Se]_{WB} = a$	2	73.95	58.32	46.89	3.31e-11	3	
$[Se]_{WB} = a + b\,[Se]_{Tissue}$	3	25.20	11.46	0.03	4.97e-01	2	0.66
$[Se]_{WB} = \exp(a + b*\ln([Se]_{Tissue}))$	3	25.18	11.43	0.00	5.03e-01	1	0.66

Tissue: Liver (*n* = 26)

Model	k	SWAD	AICc	Delta	Weight	Rank	R^1
$[Se]_{WB} = a$	2	41.05	28.27	22.81	1.11e-05	3	
$[Se]_{WB} = a + b\,[Se]_{Tissue}$	3	25.20	5.46	0.00	9.99e-01	1	0.39
$[Se]_{WB} = \exp(a + b\ln([Se]_{Tissue}))$	3	40.83	30.56	25.10	3.54e-06	2	0.01

Table H-1. Whole body vs muscle

reference	species	site/treatment	Se in tissue, µg/g dw muscle	whole body
Hermanutz *et al*. 1996	bluegill	I control-down	2.05	1.95
	bluegill	I 10 µg/L-down	20.55	22.85
	bluegill	II control-up	1.9	2.45
	bluegill	II control-down	2.25	1.95
	bluegill	II 2.5 µg/L-up	3.5	3.5
	bluegill	II 2.5 µg/L-down	6.9	6.15
	bluegill	II 10 µg/L-up	17.55	15.45
	bluegill	II 10 µg/L-down	44.7	26.45
	bluegill	II rec 30-up	12.45	11.85
	bluegill	II rec 30-down	39.6	30.6
	bluegill	III control-up	3.35	3.35
	bluegill	III control-down	3.2	2.3
	bluegill	III rec 2.5-up	5.25	6.3
	bluegill	III rec 2.5-down	6.1	5.3
	bluegill	III rec 10-up	12.45	12
	bluegill	III rec 10-down	18.6	13
	bluegill	III rec 30-up	7.75	8.35
	bluegill	III rec 30-down	15.05	17.35
Garcia-Hernandez 2000	tilapia	Cienega de Santa Cl	3.5	3
	carp	Cienega de Santa Cl	4.6	3.3
	LM bass	Cienega de Santa Cl	5.4	5.1

Table H-2. Whole Body vs Ovary

reference	species	site/treatment	Se in tissue, µg/g dw ovary	whole body
Coyle 1993	bluegill	control	2.1	0.9
	bluegill	control + water Se	2.1	0.9
	bluegill	4.6 µg/g diet	8.3	2.9
	bluegill	8.4 µg/g diet	12.5	4.9
	bluegill	16.8 µg/g diet	25	7.2
	bluegill	33.3 µg/g diet	41	16
Hermanutz *et al*. 1996	bluegill	I control-down	0.35	1.95
	bluegill	I 10 µg/L-down	20.05	22.85
	bluegill	II control-up	5.25	2.45
	bluegill	II control-down	3.85	1.95
	bluegill	II 2.5 µg/L-up	10.1	3.5
	bluegill	II 2.5 µg/L-down	12.35	6.15
	bluegill	II 10 µg/L-up	34.8	15.45
	bluegill	II 10 µg/L-down	50.5	26.45
	bluegill	II rec 30-up	29.35	11.85
	bluegill	II rec 30-down	66	30.6
	bluegill	III control-down	5.3	2.3
	bluegill	III rec 2.5-up	8.4	6.3
	bluegill	III rec 2.5-down	9.5	5.3
	bluegill	III rec 10-up	31.15	12
	bluegill	III rec 10-down	19.55	13
	bluegill	III rec 30-up	17.85	8.35
	bluegill	III rec 30-down	19.1	17.35

Table H-3. Whole body vs liver

reference	species	site/treatment	Se in tissue, µg/g dw liver	whole body
Bryson 1985-84	bluegill	control	3.9	0.45
	bluegill	Se-plankton diet	9.1	2.35
	bluegill	Selenite diet	11	1.21
	bluegill	Se-cysteine diet	9.23	2.16
	bluegill	Se-cysteine 2X diet	16.33	3.74
	bluegill	Se-methionine diet	10.85	2.46
Hermanutz *et al*. 1996	bluegill	I control-down	5.4	1.95
	bluegill	I 10 µg/L-down	36.05	22.85
	bluegill	II control-up	13.2	2.45
	bluegill	II control-down	7.2	1.95
	bluegill	II 2.5 µg/L-up	29.2	3.5
	bluegill	II 2.5 µg/L-down	26.45	6.15
	bluegill	II 10 µg/L-up	119	15.45
	bluegill	II 10 µg/L-down	68.5	26.45
	bluegill	II rec 30-up	64	11.85
	bluegill	II rec 30-down	100.5	30.6
	bluegill	III control-up	9.95	3.35
	bluegill	III control-down	9.4	2.3
	bluegill	III rec 2.5-up	13.85	6.3
	bluegill	III rec 2.5-down	16.3	5.3
	bluegill	III rec 10-up	33.25	12
	bluegill	III rec 10-down	37.15	13
	bluegill	III rec 30-up	21	8.35
	bluegill	III rec 30-down	31.9	17.35
Garcia-Hernandez 2000	carp	Cienega de Santa Cl	8.2	3.3
	LM bass	Cienega de Santa Cl	4.7	5.1

APPENDIX I

SUMMARIES OF CHRONIC STUDIES CONSIDERED FOR FCV DERIVATION

Dobbs, M.G., D.S. Cherry, and J. Cairns, Jr. 1996. Toxicity and bioaccumulation of selenium to a three-trophic level food chain. Environ. Toxicol. Chem. 15:340-347.

Test Organism: Rotifer (*Brachionus calyciflorus*), and fathead minnow (*Pimephales promelas*) 12 to 24 hr-old at start.

Exposure Route: Dietary and waterborne

Water
Filtered and sterilized natural creek water supplemented with nutrients (Modified Guillard's Woods Hole Marine Biological Laboratory algal culture medium) for algal growth. Sodium selenate (Na_2SeO_4) was added to test water to obtain nominal concentrations of 100, 200, or 400 µg Se/L. Concentrations remained stable and equal in each trophic level.

Control Diet
No selenium was added to the water medium for the alga; green alga was free of selenium for the rotifer; and rotifers were free of selenium for the fathead minnow.

Selenium Diet
Sodium selenate was added to the culture medium for the alga; green alga thereby contained a body burden for the rotifer; and rotifers thereby contained a body burden for the fathead minnow.

Dietary Treatments: Each trophic level had a different treatment. The green alga was exposed directly from the water (1, 108.1, 204.9, 397.6 µg total Se/L); rotifers were exposed from the water (1, 108.1, 204.9, 393.0 µg total Se/L) and the green alga as food (2.5, 33, 40, 50 µg Se/g dry wt.); and the fathead minnow were exposed from water (1, 108.1, 204.9, 393.0 µg total Se/L) and the rotifer as food (2.5, 47, 53, 60 µg Se/g dry wt.).

Test Duration: 25 days

Study Design: A flow-through system utilizing a stock solution of filtered and sterilized creek water controlled at 25°C was used to expose three trophic levels of organisms. Approximately one liter of media was pumped from the algal chamber into the rotifer chamber each day. A cell density between 3 and 6×10^6 cells/ml was delivered to the rotifer chambers. Rotifers were started at a density of 151.4 ± 7.7 females/ml and one liter/day of rotifers containing culture water was intermittently pumped into the minnow chamber. (*B. calyciflorus* have a life span of about 7 days at 25°C.) The pump was necessary to overcome the swimming ability of rotifers to avoid an overflow tube. Larval fathead minnows (35/chamber) were prevented from escaping by a screened overflow. Chambers were cleaned daily and aeration was provided. All chambers were duplicated for test replication and water was measured for selenium on days 0, 2, 6, 7, 11, 14, 17, 20, and 24. All algal and rotifer biomass and selenium samples were made

on these days. Fathead minnow chambers were measured for biomass, dissolved selenium, and tissue selenium concentrations of days 0, 7, 11, 14, 20, and 24. Additional measurements were made in the 200 µg Se/L test chambers on the fathead minnow on day 16. Selenium concentrations were maintained near the nominal concentrations and the standard deviation of mean concentrations was less than 4 percent.

Effects Data:

Rotifers. Rotifers did not grow well and demonstrated reduced survival at all selenium exposure concentrations during the 25 day test. By test day 7 only the lowest test concentration (108.1 µg/L) had surviving rotifers which showed a decrease in selenium content from test days 18 through 25. A reduction in rotifer biomass was discernable by test day 4 in the selenium treatments and since all test concentrations had viable rotifer populations present, the effect level was calculated using these data.

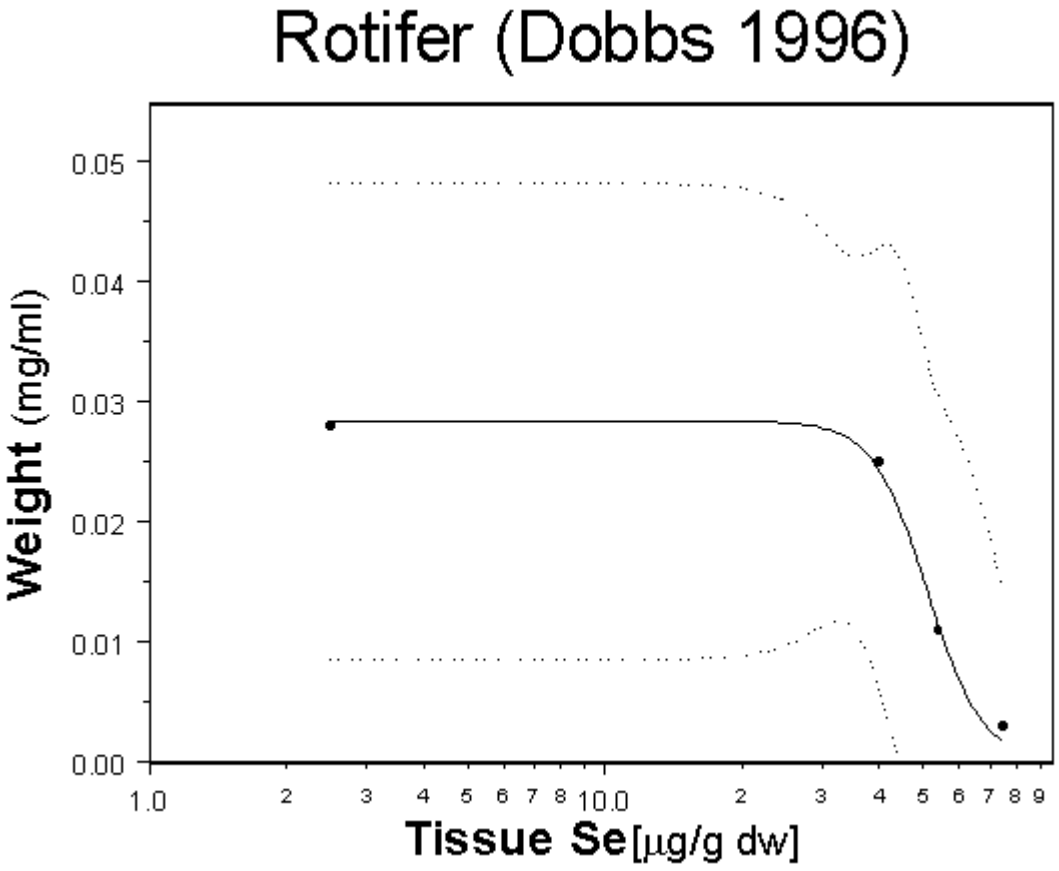
Effect of Dietary and Waterborne Selenium on Rotifers after 4 Days Exposure			
Se in water, µg/L	Se in diet, µg/g dw	Se in rotifer tissue, µg/g dw	rotifer biomass, mg/ml dw
1	2.5	2.5	0.028
108.1	33	40	0.025
202.4	40	54	0.011
393	50	75	0.003

Fathead minnows. Due to the reduction of rotifer biomass in the higher test concentrations, fish mortality and reduction in fish growth observed in the latter days of the test was difficult to discern between effects from starvation and selenium toxicity. The data from test day 8 was selected for determining the effect of selenium on fathead minnows because starvation could be excluded as a variable.

Effect of Dietary and Waterborne Selenium on Larval Fathead Minnows after 8 Days Exposure			
Se in water, µg/L	Se in diet, µg/g dw	Se in fathead minnow tissue, µg/g dw	average fish weight, mg dw
1	2.5	2.5	0.8
108.1	47	45	0.7
202.4	53	75	0.4
393	60	73	0.2

Chronic Value:

Rotifers	42.36 µg Se/g dw (EC_{20})
Fish	< 73 µg Se/g dw (LOAEC)-not amenable to statistical treatment; the LOAEC was based on the observation that a >50 percent reduction in mean fish weight occurred at this tissue concentration.



Hamilton, S.J., K.J. Buhl, N.L. Faerber, R.H. Wiedermeyer and F.A. Bullard. 1990. Toxicity of organic selenium in the diet of chinook salmon. Environ. Toxicol. Chem. 9:347-358.

Test Organism: Chinook salmon (*Oncorhynchus tshawytscha* Walbaum; swim-up larvae)

Exposure Route: Dietary only

Control Diet

Oregon moist pellet diet where over half of the salmon meal was replaced with meal from low-selenium mosquitofish (1.0 µg Se/g dw) collected from a reference site.

Selenium Diet #1

Oregon moist pellet diet where over half of the salmon meal was replaced with meal from high-selenium mosquitofish (35.4 µg Se/g dw) collected from the San Luis Drain, CA, termed SLD diet.

Selenium Diet #2

Oregon moist pellet diet where over half of the salmon meal was replaced with meal from low-selenium mosquitofish same as in the control diet, but fortified with seleno-DL-methionine (35.5 µg Se/g dw), termed SeMet diet.

Dietary Treatments: Each selenium diet was formulated to contain about 36 µg Se/g dw as the high exposure treatment. The remaining treatments were achieved by thoroughly mixing appropriate amounts of high-exposure treatment diet with control diet to yield the following nominal concentrations (3, 5, 10, and 18 µg Se/g dw).

Test Duration: 90 days

Study Design: Each dietary treatment was fed twice each day to swim-up larvae (n=100) in each of two replicate aquaria that received 1 L of replacement water (a reconstituted experimental water that simulated in quality a 1:37 dilution of water from the San Luis Drain, CA minus the trace elements) every 15 minutes (flow-through design). Mortality was recorded daily. Growth was evaluated at 30-day intervals by measuring the total lengths and wet weights of two subsets of individual fish (n=10x2) held in separate 11.5 L growth chambers within each replicate aquarium. Tissue samples were collected for whole-body selenium determinations (dry wt. basis) at 30-day intervals throughout the study; 10, 5, and 2 fish were sampled from each duplicate treatment after 30, 60, and 90 days of exposure, respectively. Concentrations of selenium measured in water were below the limit of detection (1.5-3.1 µg/L) in all dietary selenium exposure concentrations.

Effects Data: The magnitude of reduced growth was most evident in the weight of the fish, although total length was significantly reduced in fish fed high Se-laden diets as well. The effect of increasing dietary selenium on mean larval weight was similar in both the SLD and seleno-methionine diets.

Effect of San Luis Drain Diet on Growth and Survival of Chinook Salmon Larvae after 60 Days			
Se in diet, µg/g dw	Se in chinook salmon, µg/g dw	mean larval weight, g	survival, %
1	0.9	3.35	99
3.2	3.3	2.68	97.3
5.3	4.5	2.76	93
9.6	8.4	2.8	95
18.2	13.3	2.62	92.4
35.4	29.4	1.4	89

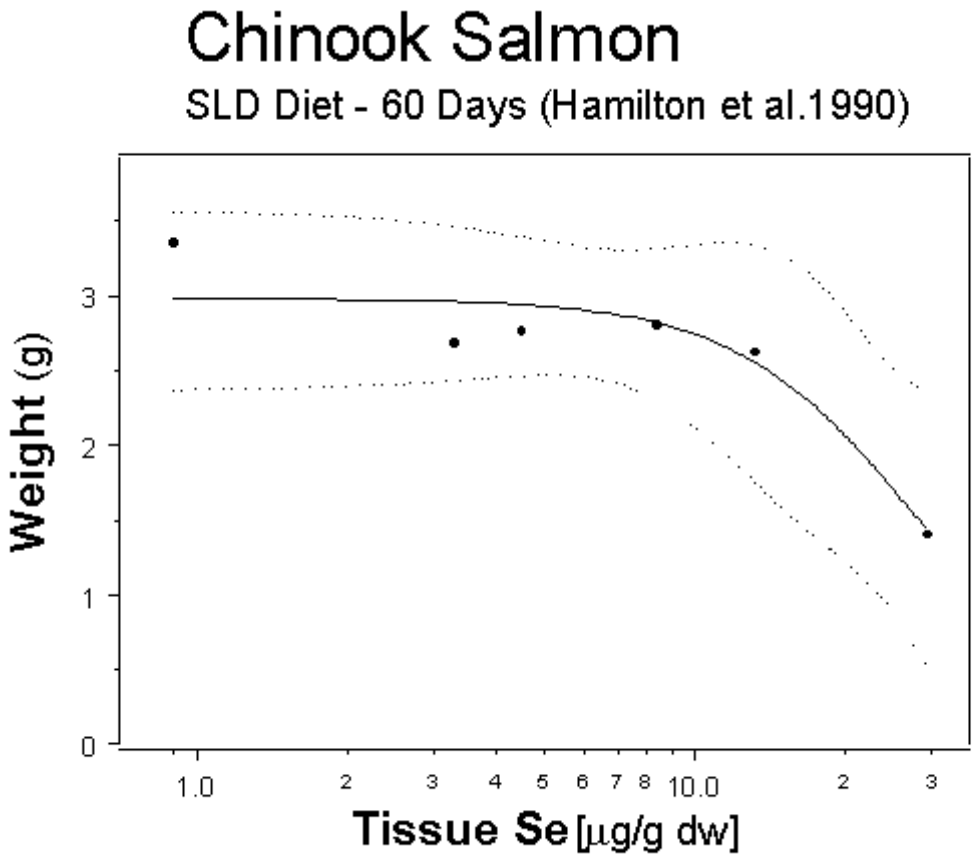
Effect of Seleno-methionine Diet on Growth and Survival of Chinook Salmon Larvae after 60 Days			
Se in diet, µg/g dw	Se in chinook salmon, µg/g dw	mean larval weight, g	survival, %
1	0.9	3.35	99
3.2	2	3.08	100
5.3	3.1	3.22	95
9.6	5.3	3.07	94.1
18.2	10.4	2.61	92.4
35.4	23.4	1.25	62.5

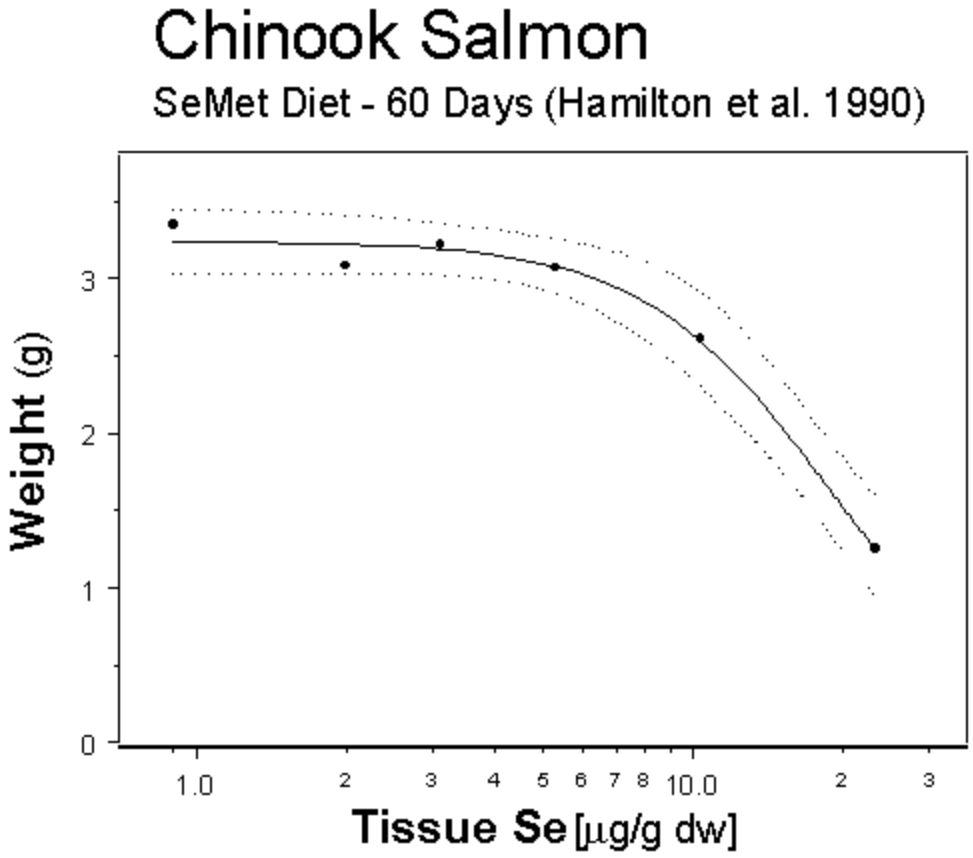
Chronic Value: Due to unacceptable control mortality of swim-up larvae in control treatments after 90 days (33.3 percent - SLD diet; 27.5 percent - SeMet diet), chronic values had to be determined from respective values reported after 60 days (tables above).

Analysis of the elemental composition of the SLD diet indicated that B, Cr, Fe, Mg, Ni and Sr were slightly elevated compared to the control and SeMet diets. No additional analyses were performed to determine the presence of other possible contaminants, i.e., pesticides.

	EC_{20} values	
	Survival (after 60 d of exposure)	Growth (after 60 d of exposure)
Diet type	Tissue Se (µg/g dw)	Tissue Se (µg/g dw)
SLD	NA[a]	15.74
SeMet	NA[a]	10.47

[a] The EC_{20} values for survival of swim-up larvae versus levels of selenium for the SLD and SeMet dietary exposure could not be estimated using non-linear regression.


Chinook Salmon
SLD Diet - 60 Days (Hamilton et al.1990)
Weight (g)
0
1
2
3
1.0
2
3
4
5
6
7
8
10.0
2
3
Tissue Se [µg/g dw]


Chinook Salmon
SeMet Diet - 60 Days (Hamilton et al. 1990)
Weight (g)
0
1
2
3
1.0
2
3
4
5
6
7
8
10.0
2
3
Tissue Se [µg/g dw]

Hilton, J.W. and P.V. Hodson. 1983. Effect of increased dietary carbohydrate on selenium metabolism and toxicity in rainbow trout (*Salmo gairdneri*). J. Nutr. 113:1241-1248.

Test Organism: Rainbow trout (*Oncorhynchus mykiss*; juvenile; approx. 0.6 g each)

Exposure Route: Dietary only

Low carbohydrate diet (LCD)

This diet contained capelin oil at 11 percent of the diet with cellulose as the filler.

High carbohydrate diet (HCD)

This diet contained cerelose at 25 percent of the diet with cellulose as the filler.

For both diets, the selenium was supplemented as sodium selenite which was mixed with cellulose and then added to the diet as a selenium premix.

Test Treatments: The two diets were supplemented with selenium (as sodium selenite) at the rate of 0, 5, or 10 µg/g dw to make up the six different dietary selenium treatments (n = 3 low carbohydrate diet; n= 3 high carbohydrate diet). The six diets were fed to duplicate groups of 100 fish. The trout were fed to satiation 3-6 times per day. Measured concentrations of selenium in the low carbohydrate diet were: 0.6 (control), 6.6, and 11.4 µg/g dw, and the measured concentrations of selenium in the high carbohydrate diet were: 0.7 (control), 6.6, and 11.8 µg/g dw. The tanks received a continuous flow of water with a flow rate of 3-4 Liters per minute.

Test Duration: 16 weeks

Study Design: Body weights, feed:gain ratios, and total mortalities were determined after each 28-day interval. After 16 weeks, approximately 20 fish were randomly removed from each tank, weighed, and blood was collected for hemoglobin, hematocrit, and plasma glucose, protein, and calcium determination. The livers and kidneys were then dissected. The livers were assayed for glycogen content, and samples of both liver and kidney were assayed for selenium content. Additional subsamples of fish were sacrificed and assayed for selenium content and for ash, crude protein, and moisture content (n=6 per treatment). Finally, 30 fish were killed, their livers and kidneys dissected, and analyzed for Ca, Cu, Fe, Mg, P, and Zn content.

Effects Data: The only overt sign of selenium toxicity was food avoidance observed in trout fed the highest selenium content in both low and high carbohydrate diets, which led to significantly reduced body weight after 16 weeks. There were no significant differences detected between treatment groups in hematological parameters. Kidney, liver, and carcass selenium levels increased with increasing selenium content of the diet, however, only the liver selenium concentrations were significantly affected by dietary selenium level, dietary carbohydrate level, and the interaction between the two treatments. Mineral analysis of the kidney

showed significantly higher levels of calcium and phosphorous in trout reared on the two highest levels of dietary selenium. Concentrations of copper in the liver increased significantly with increasing dietary selenium levels and decreasing dietary carbohydrate levels.

Effect of Selenium in Low carbohydrate Diet to Rainbow Trout		
Se in diet, µg/g dw	Se in trout liver, µg/g dw	trout weight, kg/100 fish
0.6	0.8	3.3
6.6	38.3	3.3
11.4	49.3	1.8

Effect of Selenium in High carbohydrate Diet to Rainbow Trout		
Se in diet, µg/g dw	Se in trout liver, µg/g dw	trout weight, kg/100 fish
0.7	0.6	2.7
6.6	21.0	2.3
11.8	71.7	1.4

Chronic Value: The MATC estimated for growth of rainbow trout relative to final concentration of selenium in liver tissue of trout reared on the low carbohydrate diet is the GM of 38.3 (NOAEC) and 49.3 (LOAEC) µg/g dw, or 43.45 µg/g dw. The MATC estimated for growth of rainbow trout relative to final concentration of selenium in liver tissue of trout reared on the high carbohydrate diet is the GM of 21.0 (NOAEC) and 71.7 (LOAEC) µg/g dw, or 38.80 µg/g dw. Using equation III in the text to convert this selenium concentration in liver tissue to a concentration of selenium in whole-body, the MATC for rainbow trout exposed to selenium in food with low carbohydrate content becomes 13.08 µg Se/g dw., whereas the MATC for rainbow trout exposed to selenium in food with high carbohydrate content becomes 11.65 µg Se/g dw. The latter value is selected as the chronic value for the study.

EC_{20} values could not be determined for this study. Data did not meet minimum requirements for analysis.

Hicks, B.D., J.W. Hilton, and H.W. Ferguson. 1984. Influence of dietary selenium on the occurrence of nephrocalcinosis in the rainbow trout, *Salmo gairdneri* Richardson. J. Fish Diseases. 7:379-389.

(Note: These data are the exact same as reported for the low carbohydrate diet in Hilton and Hodson 1983, with the addition of prevalence of nephrocalcinosis occurring in trout after 16 to 20 weeks of consuming the contaminated test diets).

Test Organism: Rainbow trout (*Oncorhynchus mykiss*; juvenile; approx. 0.6 g each)

Exposure Route: Dietary only
This diet contained capelin oil at 11 percent of the diet with cellulose as the filler. The selenium was supplemented as sodium selenite which was mixed with cellulose and then added to the diet as a selenium premix.

Test Treatments: The test diet was supplemented with selenium (as sodium selenite) at the rate of 0, 5, or 10 µg/g dw to make up the three different dietary selenium treatments. The three diets were fed to duplicate groups of 100 fish. The trout were fed to satiation 3-6 times per day. Measured concentrations of selenium in the low carbohydrate diet were: 0.6 (control), 6.6, and 11.4 µg/g dw. The tanks received a continuous flow of water with a flow rate of 3-4 Liters per minute.

Test Duration: 16 to 20 weeks

Study Design: See Hilton and Hodson (1983). After 20 weeks on the test diets, ten fish were randomly removed from each treatment. Tissues for histopathological examination included the stomach, intestine and pyloric ceca (including pancreas), spleen, liver, heart, kidney, skin, muscle, and gills.

Effects Data: Only effects of selenium on kidney tissue are included in the article. The kidneys of the 10 trout fed the highest selenium content in the diet exhibited normal appearance. Five of these trout exhibited precipitation of calcium in the tubules with some epithelial necrosis, but no loss of epithelial continuity. Extensive mineralized deposition of Ca within the tubules, tubular dilation and necrosis of tubular epithelium, ulceration of tubules, and intestinal Ca mineralization was observed in four of the ten fish.

Chronic Value: Same as for growth of rainbow trout reported by Hilton and Hodson (1983). The MATC estimated for growth of rainbow trout relative to final concentration of selenium in liver tissue of trout reared on the low carbohydrate diet is the GM of 38.3 (NOAEC) and 49.3 (LOAEC) µg/g dw, or 43.45 µg/g dw. Using equation III to convert the selenium concentration in liver tissue to a concentration of selenium in whole-body, the MATC becomes 13.08 µg/g dw.

EC_{20} values could not be determined for this study. Data did not meet minimum requirements for analysis.

Hilton, J.W., P.V. Hodson, and S.J. Slinger. 1980. The requirements and toxicity of selenium in rainbow trout (*Salmo gairdneri*). J. Nutr. 110:2527-2535.

Test Organism: Rainbow trout (*Oncorhynchus mykiss*; juvenile; approx. 1.28 g each)

Exposure Route: Dietary only
A casien-torula yeast diet was formulated to contain geometrically increasing levels of selenium from 0 to 15 µg/g dw. The selenium was supplemented as sodium selenite which was mixed with cellulose and then added to the diet as a selenium premix.

Test Duration: 20 weeks

Study Design: Six test diets were fed to triplicate groups of 75 fish. The trout were fed to satiation 3-4 times per day, 6 days per week, with one feeding on the seventh day. Measured concentrations of selenium in the diet were: 0.07 (control), 0.15, 0.38, 1.25, 3.67, and 13.06 µg/g dw. The tanks received a continuous flow of dechlorinated tap water from the City of Burlinton, Ontario municipal water supply. The waterborne selenium content of this water was 0.4 µg/L. During the experiment, the fish were weighed every 2 weeks with the feeding level adjusted accordingly. Mortalities were noted daily and the feed consumption for each treatment was recorded weekly. After 4 and 16 weeks, three to six fish were randomly removed from each tank, sacrificed, and their livers and kidneys removed and weighed. An additional three to six fish were then obtained from each treatment, killed, and prepared for tissue analysis. Organs and carcasses were freeze-dried for determination of selenium concentration. After 16 weeks, three more fish were removed. Kidney, liver, spleen and dorsal muscle tissue was dissected for examination of histopathology. At the end of 8 and 16 weeks, four to five fish were removed, sacrificed, and a blood sample was taken for hematological measurements (hematocrit, red blood cell count, and blood iron concentration). After 20 weeks, three to four more fish were removed, sacrificed, and a blood sample was taken for measurement of glutathione peroxidase activity.

Effects Data: There were no significant differences detected between treatment groups in histopathology, hematology, or plasma glutathione peroxidase activity. Trout raised on the highest dietary level of selenium (13.06 µg/g dw) had a significantly lower body weight and a higher number of mortalities (10.7; expressed as number per 10,000 fish days) than trout from the other treatments levels after 20 weeks of exposure.

Effects on Juvenile Rainbow Trout			
Se in diet, µg/g dw	Se in Liver, µg/g dw	weight, g/fish	mortality*
0.07	0.6	3.2	0
0.15	0.95	3.5	0
0.38	2.4	3.7	0.6
1.25	11	4.1	0.6
3.67	40	4.1	0
13.06	100	1.4	10.7

*expressed as number per 10,000 fish-days

Chronic Value: An MATC was preferred over regression analysis because of the large standard error associated with the EC_{20} value. The MATC for the growth and survival of juvenile trout based on selenium in liver tissue is the GM of the NOAEC (40 µg/g dw) and the LOAEC (100 µg/g dw), or 63.25 µg Se/g dw. Using the equation III in the text to convert the selenium concentration in liver tissue to a concentration of selenium in whole-body tissue, the MATC becomes 19.16 µg/g dw.

Holm, J. 2002. Sublethal effects of selenium on rainbow trout (*Oncorhynchus mykiss*) and brook trout (*Salvelinus fontinalis*). Masters Thesis. Department of Zoology, University of Manitoba, Winnipeg, MB.

Holm, J., V.P. Palace, K. Wautier, R.E. Evans, C.L. Baron, C. Podemski, P. Siwik and G. Sterling. 2003. An assessment of the development and survival of rainbow trout (*Oncorhynchus mykiss*) and brook trout (*Salvelinus fontinalis*) exposed to elevated selenium in an area of active coal mining. Proceedings of the 26th Annual Larval Fish Conference 2003, Bergen, Norway. ISBN 82-7461-059-B.

Test Organism: Rainbow trout (*Oncorhynchus mykiss*; spawning adults) and brook trout (*Salvelinus fontinalis*; spawning adults)

Exposure Route: dietary and waterborne - field exposure
Total selenium concentrations measured at the high selenium site ranged from 6 to 32 µg/L. Selenium was not measured at the reference streams; selenium concentrations at reference locations in the area ranged from <0.5 to 2.2 µg/L.

Study Design: Spawning fish were collected at low selenium or reference streams (Deerlick Creek and Cold Creek), a slightly elevated selenium stream (Gregg Creek), and an elevated selenium stream (Luscar Creek) in the Northeastern slopes region of Alberta, Canada. An active coal mine is the source of selenium in the elevated streams. Eggs and milt from the spawning trout were expressed by light pressure from abdomen. Individual clutches of eggs were fertilized from a composite volume of milt derived from 3-5 males. Fertilized eggs from individual females were reared to swim-up stage and examined for a number of parameters including percent fertilization, mortality, edema, and deformities (craniofacial, finfold, and spinal malformations). Similar studies were conducted in both 2000 and 2001. One notable difference is that the embryos were incubated at 8°C in 2000 and at 5°C in 2001. The authors noted that 5°C is a better representation of the actual stream temperature during embryo development..

Effects Data : Other than selenium, there were no significant differences in the concentrations of other elements (Al, As, Sb, Ba, Be, Ni, B, Cd, Ca, Cr, Co, Cu, Fe, Pb, Li, Mg, Mn, Hg, Mo, Ag, Sr, Tl, Th, Sn, Ti, U, V, Zn) in trout eggs between the low level and elevated selenium streams. There are two ways to approach determination of effects due to selenium in this study and both are presented here. The first approach determines effects based on a comparison of average conditions between streams (*between streams approach*). For example, if there is a significant difference between the average frequency of deformities in a contaminated stream and reference stream, the effect level for the *between streams approach* would be the average concentration of selenium in the tissue from the contaminated stream. The second approach evaluates individual response variables (e.g., edema, deformities) against the individual selenium tissue concentrations for the combined contaminated and reference stream data set with each year (*within streams approach*). This approach, which results in an EC_{20} value if the data meet the model assumptions, is explained in the *Calculations of Chronic Values* section of the text.

Between streams approach: For both rainbow and brook trout embryos, there were no significant differences in fertilization, time to hatch and mortality between the streams with elevated selenium and the reference streams in both 2000 and 2001. The frequency of embryonic effects were significantly greater in the high selenium stream (Luscar Creek) in 2000. Rainbow trout embryos from Luscar Creek had a greater frequency of craniofacial, skeletal and finfold deformities and edema; whereas brook trout from Luscar Creek had a greater frequency of only craniofacial deformities (see Holm Tables 1 and 2 below). In 2001, however, there were no significant differences in embryonic deformities between Luscar Creek and reference streams for both species of trout. The only difference observed in 2001 was a greater frequency of finfold deformities in brook trout collected from Gregg Creek (intermediate selenium levels) relative to the reference stream (see Holm table 2 below). All other embryonic measurements in 2001 were not significantly different between streams with elevated selenium and reference streams. When the data for both years were pooled, no significant effects were observed in embryos obtained from rainbow and brook trout collected in Luscar Creek relative to reference streams (see Holm Table 3).

Within streams approach: EC_{20} values could not be calculated for total deformities or edema for the 2000 rainbow trout data because a logistic curve could not be fitted to the data (see Holm Figures 1 and 2). For the 2001 data, EC_{20} values could not be computed for edema and skeletal and finfold deformities for rainbow trout data because a logistic curve could not be fitted to the data (see Holm Figures 3 and 4). Craniofacial deformities in the rainbow embryo as a function of selenium in egg ww (2001 data) was fitted to a logistic curve from which an EC_{20} value was calculated (see Holm Figure 5). The brook trout data for 2000 and 2001 were not suitable for fitting logistic curves (see Holm Figure 6).

Holm Table 1

Mean embryo-larval parameters for rainbow trout collected from a high Se site (Luscar Creek), an intermediate Se site (Gregg River), and reference sites (Deerlick Creek and Wampus Creek) in northeastern Alberta over two consecutive years (mean ± SE). Values that are significantly different at $\alpha = 0.05$ are marked with different letters. (Table modified from Holm 2002)

	2000		2001			
Measurement	Luscar	Deerlick	Luscar	Gregg	Deerlick	Wampus
Se, egg, µg/g ww	8.37 ± 1.62	2.05 ± 1.06	6.49 ± 0.89	6.65 ± 1.83	2.77 ± 0.20	2.35 ± 0.31
Se, adult muscle, µg/g ww	1.50 ± 0.28	0.48 ± 0.15	NT	NT	NT	NT
n[a]	297	261	2021	720	1342	209
% fertilization	79.8 ± 4.3	51.5 ± 10.9	81.5 ± 5.0	79.4 ± 5.2	88.0 ± 2.1	94.0 ± 4.8
% mortality	3.3 ± 1.0	0.7 ± 0.4	27.8 ± 7.3	38.3 ±13.7	26.5 ± 4.7	4.2 ± 0.8
% CR	7.7 ± 3.7[b]	0.2 ± 0.2[c]	14.7 ± 3.4	11.7 ± 2.7	10.6 ± 1.9	12.0 ± 4.1
% SK	13.8 ± 5.6[b]	0.7 ± 0.5[c]	19.4 ± 8.2	11.1 ± 2.3	15.6 ± 4.7	4.9 ± 4.9
% FF	3.2 ± 2.0[b]	0.2 ± 0.2[c]	6.8 ± 3.0	15.5 ± 6.6	4.0 ± 0.9	1.5 ± 0.2
% ED	30.8 ± 27.4[b]	0.2 ± 0.2[c]	19.9 ± 8.5	13.9 ± 5.3	10.8 ± 2.5	7.5 ± 0.4
% TD	38.9 ± 25.6[b]	0.7 ± 0.5[c]	ND	ND	ND	ND

[a] number of fry to reach the swim-up stage

[b and c] statistically different values

CR = craniofacial defects, SK = skeletal defects, FF = finfold defects, ED = edema, TD = total defects, NT = not tested, ND = not done

Holm Table 2

Mean embryo-larval parameters for brook trout collected from a high Se site (Luscar Creek), an intermediate Se site (Gregg River), and reference sites (Cold Creek) in northeastern Alberta over two consecutive years (mean ± SE). Values that are significantly different at $\alpha = 0.05$ are marked with different letters. (Table modified from Holm 2002)

Site	2000		2001		
	Luscar	Cold	Luscar	Gregg	Cold
Se, egg, µg/g ww	6.37 ± 0.78	1.35 ± 0.24	8.02 ± 0.77	6.88 ± 0.51	1.25 ± 0.15
Se, adult muscle, µg/g ww	3.79 ± 0.51	0.55 ± 0.10	NT	NT	NT
n[a]	4904	1560	3440	1892	1440
% fertilization	97.4 ± 0.8	96.1 ± 1.2	87.2 ± 2.6	85.2 ± 5.4	77.8 ± 14.2
% mortality	12.6 ± 3.8	9.3 ± 2.4	2.9 ± 0.8	2.9 ± 0.9	3.7 ± 1.6
% CR	13.6 ± 3.5[b]	3.0 ± 0.5[c]	5.6 ± 3.2	2.12 ± 1.0	0.7 ± 0.3
% SK	1.9 ± 0.8	1.3 ± 0.8	2.1 ± 1.1	0.81 ± 0.3	0.6 ± 0.4
% FF	1.1 ± 0.6	1.2 ± 0.8	3.7 ± 1.8	4.1 ± 2.4[c]	0.1 ± 0.1[b]
% ED	0.6 ± 0.4	0.3 ± 0.1	0.4 ± 0.1	0.3 ± 0.2	1.7 ± 1.2
% TD	14.4 ± 3.6[b]	4.0 ± 2.3[c]	ND	ND	ND

[a] number of fry to reach the swim-up stage

[b and c] statistically different values

CR = craniofacial defects, SK = skeletal defects, FF = finfold defects, ED = edema, TD = total defects, ND = not done

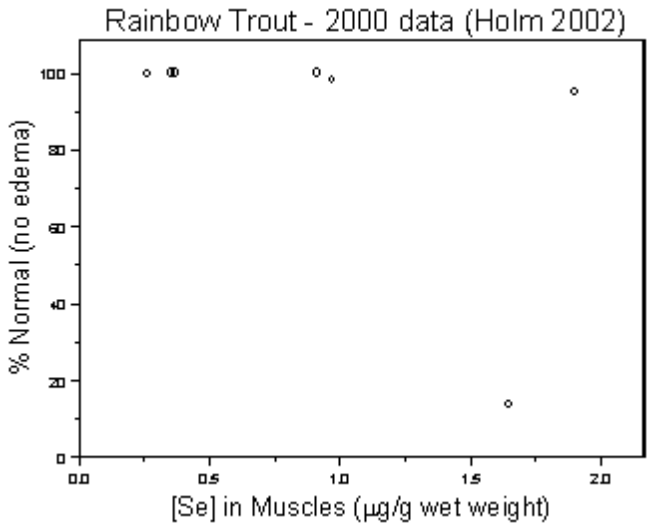
Holm Table 3

Mean embryo-larval parameters for rainbow trout and brook trout collected from a high Se site (Luscar Creek) and reference sites (Deerlick Creek and Cold Creek) in northeastern Alberta over two consecutive years, combined over both years of the study by site (mean ± SE). Values that are significantly different at $\alpha = 0.05$ are marked with different letters. (Table modified from Holm 2002)

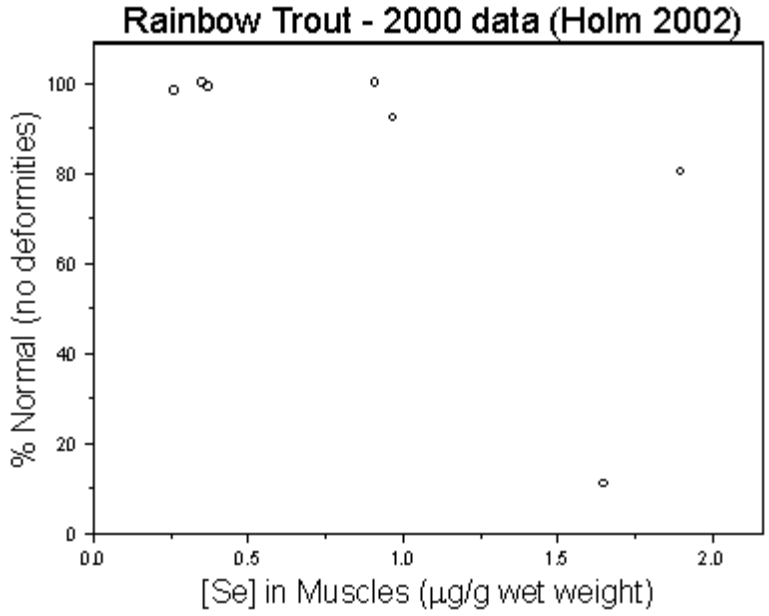
Measurement	Rainbow Trout		Brook Trout	
	Luscar	Deerlick	Luscar	Cold
Se, egg, µg/g ww	6.92 ± 0.78	2.56 ± 0.32	7.20 ± 0.56	1.30 ± 0.14
n[a]	2318	1603	8344	3000
% fertilization	81.1 ± 3.9	77.6 ± 5.6	92.3 ± 17.7	88.5 ± 6.2
% mortality	22.2 ± 6.3	19.1 ± 4.6	7.7 ± 2.1	6.9 ± 1.7
% CR	13.1 ± 3.2	7.6 ± 7.1	9.9 ± 2.4	2.7 ± 0.6
% SK	18.1 ± 6.3	11.4 ± 3.8	2.0 ± 0.6	1.0 ± 0.4
% FF	6.0 ± 2.4	2.9 ± 0.8	2.6 ± 1.0	1.2 ± 0.5
% ED	22.4 ± 8.5	7.8 ± 2.2	1.3 ± 0.7	0.9 ± 0.5

[a] number of fry to reach the swim-up stage

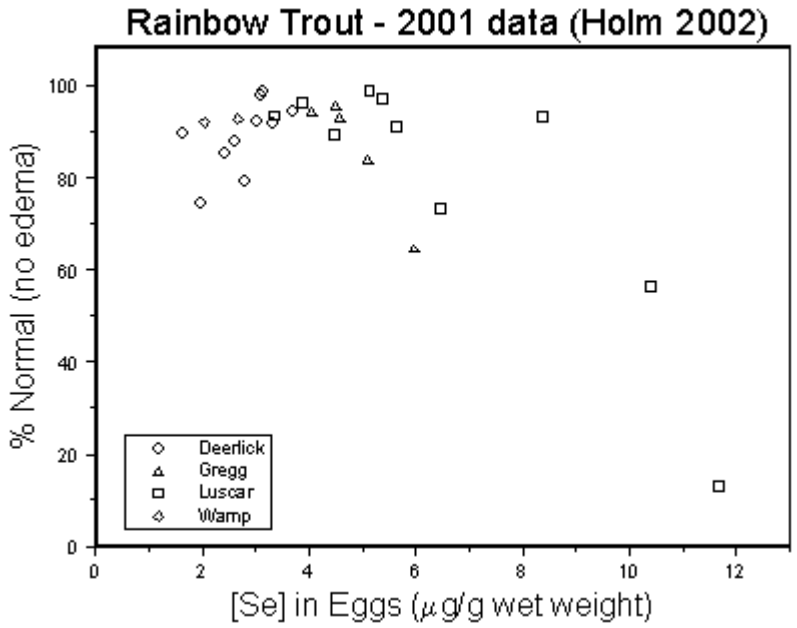
CR = craniofacial defects, SK = skeletal defects, FF = finfold defects, ED = edema,



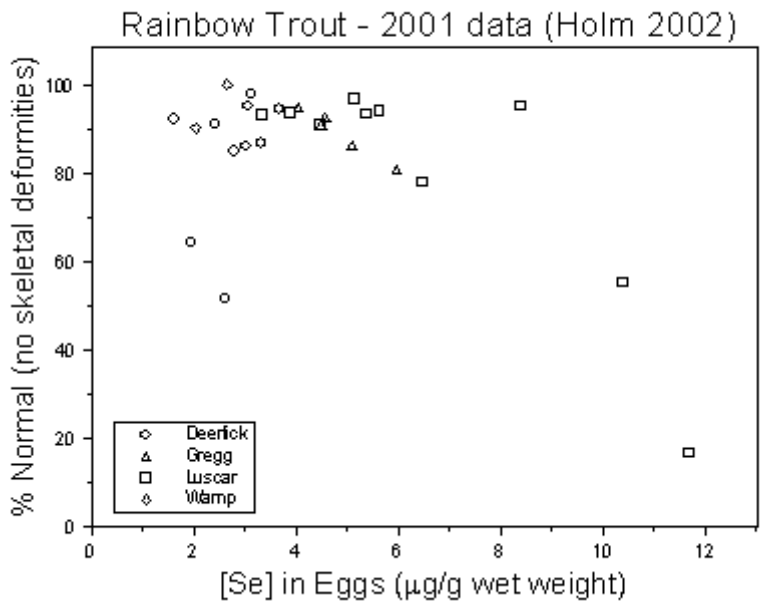
Holm Figure 1. Plot of percent normal (100 - percent edematous) against selenium concentration in adult rainbow trout muscle ww.



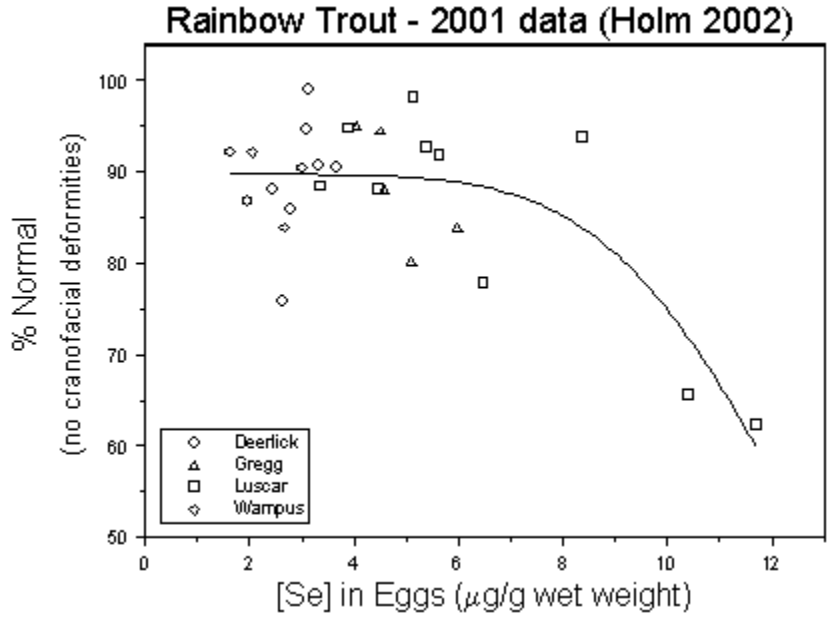
Holm Figure 2. Plot of percent normal (100 - percent total deformities) against selenium concentration in adult rainbow trout muscle ww, 2000 data.



Holm Figure 3. Plot of percent normal (100 - percent total deformities) against selenium concentration in rainbow trout eggs ww, 2001 data.

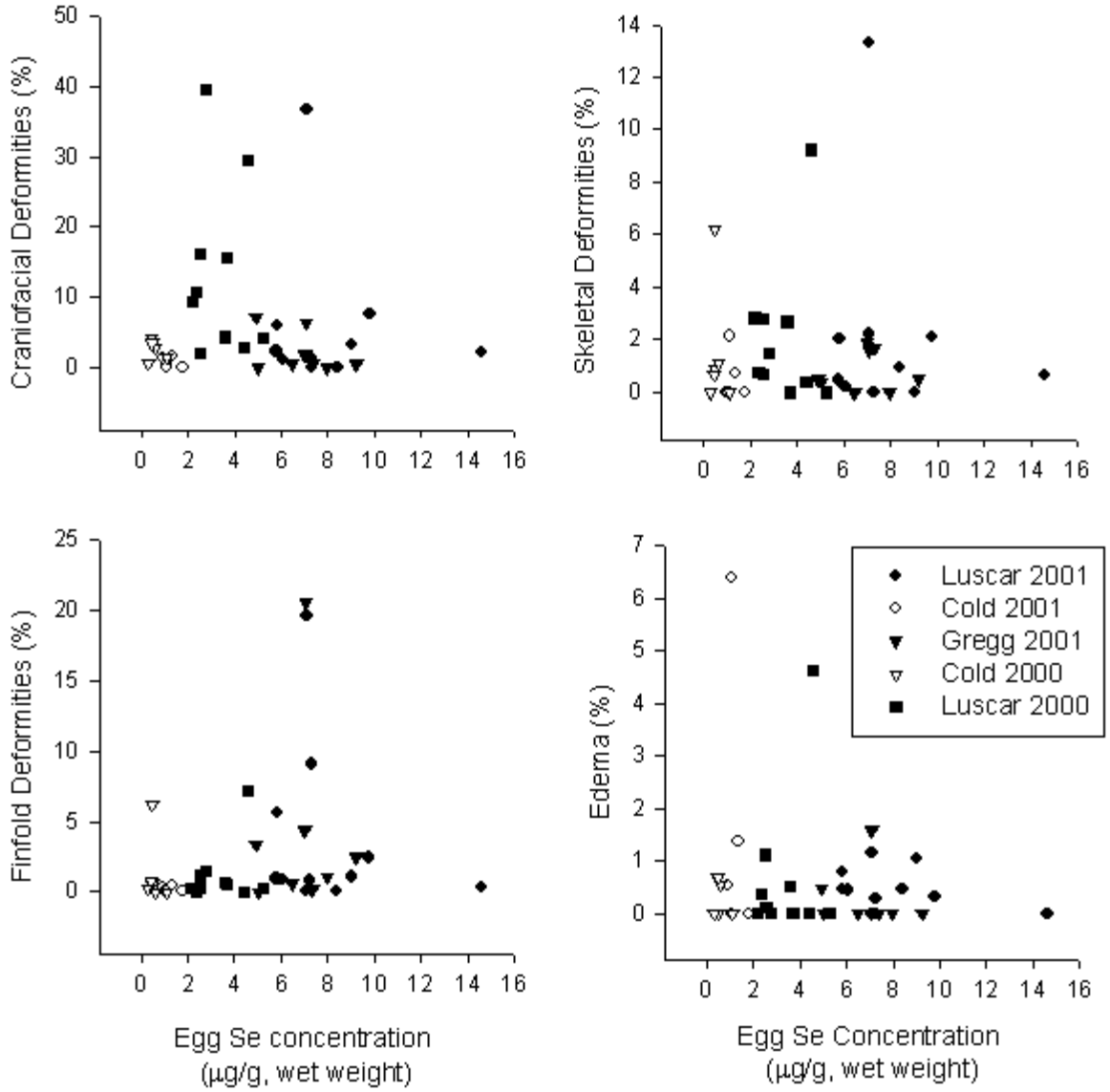


Holm Figure 4. Plot of percent normal (100 - percent skeletal deformities) against selenium concentration in rainbow trout eggs ww, 2001 data.



Holm Figure 5. Plot of percent normal (100 - percent total deformities) against selenium concentration in rainbow trout eggs ww, 2001data. EC_{20} value at 10.4 µg Se/g egg ww.

Brook Trout - 2000 and 2001



Holm Figure 6. Plot of percent normal (100 - total abnormalities) for craniofacial, skeletal and finfold deformities and edema against selenium concentration in brook trout eggs ww, 2000 and 2001 data.

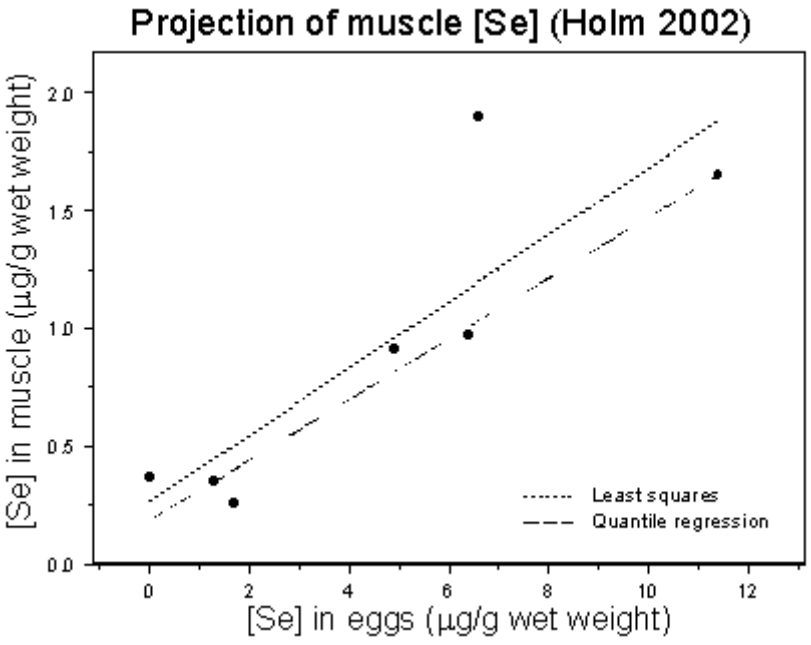
The effect levels determined using the *between streams* or *within streams* approach resulted in values based on ww in eggs or muscle. Several conversions were necessary to transform a selenium concentration in egg ww to whole body dw. Using data reported by Holm et al., quantile regression was used to estimate selenium in adult muscle (ww) from selenium in egg (ww) (see Projection of Muscle Selenium Concentrations below). A percent moisture of 75.84% derived from rainbow trout data was used to convert ww to dw and equation 1 was used to convert muscle dw to the whole body dw values listed below (under Chronic Values).

Projection of Muscle Selenium Concentrations

Median concentrations of selenium in rainbow trout muscles were projected from selenium concentrations in rainbow trout eggs according to an empirical equation:

$$[Se_{muscle}] = 0.1827 + 0.1287[Se_{egg}] \qquad (R = 0.6244, 5\ df)$$

Parameters of the linear model were estimated by quantile regression, which minimizes the sum of weighted absolute deviations. Such method is less sensitive to outliers than ordinary least squares (Koenker and Portnoy 1996). This difference is clearly illustrated in Holm Figure 7: projections of selenium concentrations in muscles of rainbow trout by the least squares regression line are consistently greater than projections by the quantile regression line ($[Se_{muscle}] = 0.2613 + 0.1418[Se_{egg}]$) due to the disproportional influence of one data point (6.6,1.9).



Holm Figure 7. Regression lines projecting selenium concentrations in muscles of rainbow trout as a function of selenium concentrations in rainbow trout eggs.

Chronic Values:

Between streams approach

Rainbow trout 2000: effects (craniofacial, skeletal and finfold deformities and edema) at 1.50 µg Se/g muscle ww or 5.79 µg Se/g dw whole body using conversion factors listed above; **chronic value is 5.79 µg Se/g dw whole body**

Brook trout 2000: effects (craniofacial deformities) at 3.79 µg Se/g muscle ww or 13.2 µg Se/g dw whole body using conversion factors listed above; **chronic value is 13.2 µg Se/g dw whole body**

Rainbow trout 2001: no effects at 6.65 µg Se/g egg ww or 4.14µg Se/g dw whole body using conversion factors listed above; **chronic value is >4.14 µg Se/g dw whole body**

Brook trout 2001: effects (finfold deformities) at 6.88 µg Se/g egg ww or 12.4 µg Se/g dw whole body using conversion factors listed above; **chronic value is 12.4 µg Se/g dw whole body**

Within streams approach

Rainbow trout 2000: **no value available**; EC_{20} analysis not appropriate for data sets

Brook trout 2000: **no value available**; EC_{20} analysis not appropriate for data sets

Rainbow trout 2001: EC_{20} value (craniofacial deformities) at 10.4 µg Se/g egg ww or 5.85 µg Se/g whole body dw; **chronic value is 5.85 µg Se/g whole body dw**

Brook trout 2001: **no value available**; EC_{20} analysis not appropriate for data set

Kennedy, C.J., L.E. McDonald, R. Loveridge, M.M. Strosher. 2000. The effect of bioaccumulated selenium on mortalities and deformities in the eggs, larvae, and fry of a wild population of cutthroat trout (*Oncorhynchus clarki lewisi*). Arch. Environ. Contam. Toxicol. 39:46-52.

Test Organism: Cutthroat trout (*Oncorhynchus clarki lewisi*; spawning adults, 3-6 years)

Exposure Route: dietary and waterborne - field exposure
Total selenium concentrations measured at the time the eggs were taken were <0.1 µg/L from the reference site and 13.3 to 14.5 µg/L at the exposed site.

Study Design: At reference and exposed site (Fording River, BC, Canada which receives drainage from open-pit coal mining), eggs were stripped from females (n=20 from reference site; n=17 from exposed site) and fertilized from milt from one male collected at each site. Fertilized eggs were reared in well water and examined for time to hatch, deformities (craniofacial, finfold, skeletal and yolk sac malformations), and mortalities. Inspection of deformities in eggs were performed using 40X magnification.

Effects Data : No significant correlations between the selenium concentrations in the eggs from either site and: hatching time (reference, 25.5-26.5 days; exposed, 22-25.5 days); percent deformities preponding (reference, 0-2.4%; exposed, 0-0.34%); percent deformities after ponding (reference, 0-0.26%; exposed, 0-0.09%); percent mortalities preponding (reference, 1.5-70.3%; exposed, 1-100%); percent mortalities after ponding (reference, 0.3-4.3%; exposed, 1.5-43.7%); total percent mortalities (reference, 2.8-55.8%; exposed, 3.7-100%). The average selenium residue in tissues were as follows:

Site	Adult fish liver, µg Se/g dw	Adult fish muscle, µg Se/g dw	eggs, µg Se/g dw
reference	8.2; Range: 3.4-14.6	2.4; 1.4-3.8	4.6
exposed	36.6; Range:18.3-114	12.5; Range: 6.7-41	21.2

Effects >12.5 µg Se/g dw in muscle

Chronic Value: >10.92 µg Se/g dw estimated using the equation I to convert the selenium concentration in muscle tissue (>12.5 µg Se/g dw) of adult fish to a selenium concentration in whole-body.

Hardy, R.W. 2002. Effects of dietary selenium on cutthroat trout (*Oncorhynchus clarki*) growth and reproductive performance. Annual report for Montgomery Watson Harza. April 30, 2002.

Test Organism: Cutthroat trout (*Oncorhynchus clarki*, 0.9 g)

Exposure Route: Dietary only
Six experimental dietary treatments were produced by cold extrusion. The formulation of the diet was designed to be similar to commercial trout diets and had a proximate composition of 45% protein and 16% lipid. Seleno-methionine diluted in distilled water (100 ug/L) was added in appropriate volumes to each batch of feed to facilitate pelleting. Measured dietary selenium concentrations were 1.2 (control), 3.8, 6.4, 9.0, 11.5, and 12 ug Se/g dw. Fry were fed initially at a rate of 10 times per day 6 days a week to apparent saturation. Feeding frequency decreased as fish grew.

Test Duration: 124 weeks (865 days, 2.5 yrs)

Study Design: Groups of 50 fish were placed into triplicate tanks (145 L) receiving 4-15 L/min of hatchery water at 14.5°C and fed one of the six experimental diets. The fish in each tank were bulk-weighed and counted every 14 days for the first 12 weeks of the experiment, and then every 4 weeks until 48 weeks. Samples of fish for whole-body selenium analysis were taken at each sampling date for the first 12 weeks followed by every 3 months thereafter. After six months of feeding, the fish were transferred to 575 L tanks and the number of replicate tanks per dietary treatment was reduced to two. After 80 weeks of feeding, the fish were transferred to 1050 L outdoor tanks each supplied with 70 L/min of constant temperature (14.5°C) spring (hatchery) water. After 2.5 years of the feeding trial, fish were spawned and whole body selenium level , egg selenium level, % eyed eggs, % hatched eggs, and % deformed larvae were examined.

Effects Data : No signs of toxicity (reduced growth or survival relative to controls) were observed in fish fed the highest dietary selenium treatment (12 ug Se/g dw) after the first 80 weeks of exposure just prior to transfer outdoors. No signs of clinical disease were evident, and no relationship was found between feed conversion ratios and the level of selenium added to the feed. Whole body selenium levels were approximately 6.8, 10, 12 and 12.5 ug Se/g dw in the four highest dietary treatments. Nine months later, whole body selenium levels at spawning had decreased somewhat to 5.21, 8.80, 9.37 and 6.66 ug Se/g dw in these four highest dietary treatment groups, respectively. Percent survival from the eyed stage to hatching varied among treatment groups, with the control having the highest survival (97%) and the fifth dietary treatment group the second highest (93%). Percent deformed larvae ranged from a low of 3.4% in controls to a high of 30% in the 9.0 ug Se/g dw dietary treatment group; larvae in the two highest dietary treatment groups only exhibited 7 and 6.8 %, respectively.

Chronic Value: The chronic value for this study is a NOAEC of >9.37 ug Se/g dw whole-body parent tissue based on embryo/larval deformity.

Bennett, William N., Arthur S. Brooks, and Martin E. Boraas. 1986. Selenium uptake and transfer in an aquatic food chain and its effects on fathead minnow larvae. Arch. Environ. Contam. Toxicol. 15:513-517.

Test Organism: Fathead minnow (*Pimephales promelas*; 2 to 8 day-old larvae).

Exposure Route: Dietary only
Green alga, *Chlorella pyrenoidosa* were exposed to Se ($H_2{}^{75}SeO_4$) in culture water for 3 days. Rotifers, *Brachionus calyciflorus*, were cultured in chambers with selenium containing green algae at the ratio of 25 µg algae/ml to 50 µg rotifer/ml for 5 hr. The rotifers were filtered to separate them from the algae and immediately heat-killed. The Se concentration in the rotifers was measured for ^{75}Se activity.

Test Duration: 9 to 30 days

Study Design: Selenium uptake by larval fathead minnows was measured in three experiments. Se-contaminated and control rotifers for feeding to larval fish were prepared in advance using the low algae:rotifer ratio. Daily equal volumes of rotifers were divided among five 800 mL polypropylene larval chambers. Three chambers received Se-contaminated rotifers and two received control rotifers. The rotifers were dead at the time of feeding (heat killed).

Larval fish were hatched from eggs spawned in the laboratory. After hatching, active larvae were divided equally among the larval test chambers (daily renewal exposures using declorinated Lake Michigan water). Larvae were initially fed rotifers raised on control algae (no selenium). The age of the larvae when first fed Se-contaminated rotifers was 4, 9, and 3 days post-hatch for experiments 1, 2, and 3, respectively. Larval fish were fed Se-contaminated rotifers for 7, 9, and 7 days in the 3 experiments. A post-exposure observation period of 19 and 2 days was used for experiments 1 and 2, respectively. During this time the larvae were fed control rotifers. Daily, larvae from a replicate were removed from the test chamber, washed, placed in a 20 ml vial, and counted for ^{75}Se activity for 20 min. All larvae were then placed in test chambers with fresh food rations. At the end of the study all fish were individually dried and weighed.

	Experiment 1	Experiment 2	Experiment 3
Initial feeding of control diet (days)	3	8	2
Day Se diet first fed	4	9	3
Day Se diet last fed	11	17	9
Observation days on control diet	19	2	0
Age at study termination (days)	30	19	9

Effects Data:

	Experiment 1	Experiment 2	Experiment 3
Mean food Se concentration (µg/g)	>70	68	55
Food intake (µg rotifers/larva)	50	1330	1190
Initial larvae mean dry wt. at start of Se-laden food (µg)	90	400	100
Final larvae mean dry wt. (µg) at end of test	1470 (Control) 800 (Treatment)[a]	1888 (Control) 1354 (Treatment)[a]	475 (Control) 416 (Treatment)
Final mean larval Se content (µg Se/larva)[b]	0.0062	0.0700	0.0248
Final mean larval Se concentrations (µg Se/g dw)	43.0	51.7	61.1

[a] Significantly different from the control.
[b] Values when Se-laden feeding was ended.

Selenium was measured in the test water during the feeding exposures, but the concentrations were insignificant (0.84 µg/L). Survival was not affected by the selenium exposures. Preliminary tests showed that fathead minnow larvae would reach plateau concentrations of selenium within the 7- to 9-day exposure periods. The food supply was sufficient to sustain growth of the larvae during the study, according to the authors. The authors state that selenium uptake and higher selenium content in experiment 2 larvae was due to their larger size and ability to consume more rotifers/unit time. Se-exposed larvae were significantly smaller ($p<0.05$) in mass than controls for experiments 1 and 2.

Chronic Value: The estimated whole-body chronic value for this study, determined as the geometric mean of the final mean larval selenium concentrations measured in the three experiments, i.e., 43.0, 51.7, and 61.1 µg/g dw, respectively, is 51.40 µg Se/g dw.

Ogle, R.S. and A.W. Knight. 1989. Effects of elevated foodborne selenium on growth and reproduction of the fathead minnow (*Pimephales promelas*). Arch. Environ. Contam. Toxicol. 18:795-803.

Test Organism: Fathead minnows (*Pimephales promelas*; juvenile, 59 to 61 d old)

Exposure Route: Dietary only
Purified diet mix spiked with inorganic and organic selenium: 25 percent selenate, 50 percent selenite, and 25 percent seleno-L-methionine, homogenized in dextrin.

Test Treatments: Completely randomized block design (2 blocks); 4 replicates per block (n = 8 replicates total per treatment). Actual mean total selenium levels in each exposure treatment were: 0.4 (control), 5.2, 10.2, 15.2, 20.3, and 29.5 µg/g dw. Fish used in the first randomized block (F_2 generation fish) were progeny from F_1 generation originally obtained from the Columbia National Fishery Research Laboratory, some of which were used in an initial range-finding experiment. Fish obtained from a commercial supplier were used in the second randomized block. The prepared diet was extruded into 1.5 mm pellets which were air-blow dried to 5 percent moisture content and crushed and sieved so that only particles retained by an 11.8 mesh/cm sieve were used in the study. The amount of selenium in water that leached from the food during the experiment averaged only 0.8 µg/L.

Test Duration: 105 days, F_2 generation (block one) and commercial fish (block two);
14 days F_3 generation

Study Design: Ten fish were randomly placed in each cell per block (n = 8x10, or 80 fish total per treatment). Fish were fed twice daily at 6 percent body weight per day, with wastes and uneaten food removed 30 min. after each feeding. Test tanks were flushed with two tank volumes of fresh test water after each feeding (solution renewal). Growth (as wet weight) was determined every two weeks by bulk weighing, and one fish from two of the cells per treatment in a given block (n = 4 total per treatment) was removed for selenium (whole-body) analysis. After 105 days of exposure, a single male and female fish from each treatment replicate (n = 4 breeding pairs per treatment in a given block, or 8 breeding pairs per treatment total) were placed in 250 ml beakers and inspected for spawning activity for 30 days following the first spawning event for that pair (each pair being one replicate). Gonads and muscle tissue were dissected for selenium analysis from these fish at the end of the 30 days spawning period. The spawning substrates were inspected daily for eggs to determine fertility and viability. Samples of not more than 50 eggs from each spawn were incubated in flowing, aerated water and inspected for percent hatch determination. Ten larvae from each incubated brood were transferred to separate glass test chambers and maintained (48 h renewal; fed brine shrimp twice daily) for 14 days to determine percent larval survival.

Effects Data: There was no effect of selenium on any of the reproductive parameters measured at the dietary concentrations tested. Percent hatch and percent larval survival were very high (>87.4 percent) and essentially equal for all of the treatments.

Growth of pre-spawning adults was affected by the selenium exposure. Growth data are given in the following table:

Effects on Fathead Minnow Growth after 98 days of Exposure to Dietary Selenium		
Measured mean selenium in diet, µg/g dw	Whole-body selenium, µg/g dw	Mean fish weight, g ww
0.4	1.76	1.30
5.2	2.78	1.24
10.2	3.42	1.20
15.2	5.40	1.21
20.3	6.58	1.09
29.5	7.46	0.94

Chronic Value: An EC_{20} value could not be calculated for these data because the data did not meet the minimum requirements for analysis. The MATC for growth of pre-spawning fathead minnows versus levels of selenium found in whole-body tissue was the GM of 5.40 and 6.58 µg/g dw, or 5.961 µg Se/g dw.

Schultz, R. and R. Hermanutz. 1990. Transfer of toxic concentrations of selenium from parent to progeny in the fathead minnow (*Pimephales promelas*). Bull. Environ. Contam. Toxicol. 45:568-573.

Test Organism: Fathead minnow (*Pimephales promelas*; Adults)

Exposure Route: Dietary and waterborne
Selenite was added to artificial streams which entered the food web; thus, fish were also exposed to selenium in the diet.

Study Design: Four Monticello artificial streams were used for the study which lasted from September 1987 to September 1988. For each study, two streams (treated) were dosed continuously to achieve 10 µg/L and two streams served as controls. Mean selenium concentrations at the head of the treated streams were 9.8 ± 1.2 and 10.3 ± 1.7 µg/L, respectively. The concentrations of selenium measured in the water from controls streams were all less than the detection limit, i.e., 2 µg/L. Spawning platforms were submerged into each stream. One subset of six embryo samples (n = 2000 embryos per sample) were collected from the streams for selenium analysis. Another subset of ten embryo samples were reared in incubation cups receiving the same streamwater dosed with sodium selenite via a proportional diluter. The treated embryos in egg cups received an average 9.7 ± 2.6 µg of selenium/L. Samples of hatched larvae were analyzed for selenium content while others were inspected for occurrence of edema and lordosis. Prior to test termination, female parents were seined. The mean selenium content in the ovaries of seven to eight females from the treated and control streams was reported.

Effects Data : Edema and lordosis occurred in approximately 25 percent of the fish spawned and reared in 10 µg of selenium/L. Corresponding occurrence in control fish incubated in the egg cups was only 1 and 6 percent, respectively. Selenium residues in the ovaries of females from the control and treated streams were 0.77 and 5.89 µg/g ww. Assuming 85 percent moisture content in the ovaries (see Gillespie and Baumann below), these concentrations equate to 5.133 and 39.27 µg Se/g dw.

Chronic Value: <18.21 µg Se/g dw estimated using equation II to convert the selenium concentration in adult female ovaries (39.27 µg Se/g dw) to a selenium concentration in whole-body.

Beyers, D.W. and Sodergren, C. 2001a. Evaluation of interspecific sensitivity to selenium exposure: Larval razorback sucker versus flannelmouth sucker. Larval Fish Laboratory. Department of Fishery and Wildlife Biology, Colorado State University, Fort Collins, Colorado.

Test Organism: Larval flannelmouth sucker (*Catostomus latipinnis*) and larval razorback sucker *(Xyrauchen texanus)*

Exposure Route: Dietary and waterborne - laboratory exposure (28-d early life stage) Continuous flow diluter supplied a range of aqueous test concentrations <1, 25.4, 50.6, 98.9, and 190.6 µg/L selenate. Well water was used as the dilution water. Across the range of aqueous exposure concentrations, each test chamber was fed the same daily ration of living rotifers containing selenium at <0.702, 1.35, 2.02, 4.63, and 8.24 µg/g dw, respectively. Rotifers accumulated selenium from algae (*Chlorella vulgaris*) exposed to 0, 25, 50, 100, and 200 µg/L selenate.

Study Design: Replicated (n=4) exposure beakers using a randomized, balanced 5x2 factorial design (1st factor - selenium; 2nd factor - species). Survival was monitored daily and growth measured at the end of the 28-day exposure. Selenium was measured in the larvae at the end of the 28-day exposure.

Effects Data : No survival effects were observed and there were no decreases in fish weight or length. Fish mass was found to increase as a function of selenium concentration.

Chronic Value: The chronic values for the flannelmouth sucker and razorback sucker were >10.2 and >12.9 µg Se/g dw, respectively, based on the concentrations of selenium measured in whole-body tissue of larval fish at the highest water and dietary selenium concentrations.

Beyers, D.W. and Sodergren, C. 2001b. Assessment of exposure of larval razorback sucker to selenium in natural waters and evaluation of laboratory-based predictions. Larval Fish Laboratory. Department of Fishery and Wildlife Biology, Colorado State University, Fort Collins, Colorado.

Test Organism: Larval razorback sucker *(Xyrauchen texanus)*

Exposure Route: Dietary and waterborne - laboratory exposure (28-d early life stage)
Larvae were exposed in a daily static-renewal system to control water (reconstituted very hard) and site waters: De Beque, Orchard Mesa, North Pond diluted 50%, and North Pond. Each water type received either a control diet (rotifers) or a diet previously exposed to the site water (site food: rotifers fed algae exposed to respective site water).

Study Design: Replicated (n=4) exposure beakers using a randomized, balanced 5x2 factorial design (1st factor - test water type; 2nd factor - rotifers cultured in control water or in site water). Survival was monitored daily and growth measured at the end of the 28-day exposure. Selenium was measured in the larvae at the end of the 28-day exposure.

Effects Data : No survival effects were observed. There were no significant decreases in growth of fish exposed to both site water and site food compared to fish exposed to control water and control food. There was a significant increase in growth of fish exposed to site water and control food relative to fish exposed to control water and control food ($p<0.0001$). There were reductions in the growth of fish (14%) exposed to site water and site food compared to site water and control food ($p<0.0001$). Due to the lack of a dose-response relationship in both the concentration of selenium in the food (rotifers) and growth, and the concentration of selenium in the fish larvae and growth, the authors did not attribute the effect of site food on the growth of fish to selenium.

Chronic Value: The NOAEC for the razorback sucker larvae in the four site water types based on selenium in whole-body tissue were: De Beque >5.45 µg Se/g dw; Orchard Mesa >11 µg Se/g dw; North Pond 50% dilution >41.1 µg Se/g dw; North Pond >42 µg Se/g dw. Because no significant effects were observed in larvae exposed to North Pond water at >42 µg Se/g dw whole-body tissue, this value was selected as the chronic value for the study.

Bryson, W.T., W.R.Garrett, M.A. Mallin, K.A. MacPherson, W.E. Partin, and S.E. Woock. 1984. Roxboro Steam Electric Plant 1982 Environmental Monitoring Studies, Volume II, Hyco Reservoir Bioassay Studies. Environmental Technology Section. Carolina Power & Light Company.

28-day Embryo/Larval Study

Test Organism: Bluegill sunfish (*Lepomis macrochirus;* embryos and larvae)

Exposure Route: dietary and waterborne - field exposure
Native adult bluegill were collected from Hyco Reservoir in Person County, North Carolina and from a nearby control lake (Roxboro City Lake). Hyco Reservoir is a cooling lake for Carolina Power & Light and receives the discharge from the ash storage pond. No selenium values were given for Hyco Reservoir, total selenium was not detected in the control lake (< 1 µg/L). A mean selenium for the ash pond effluent from a previous study was 53 µg/L (N=59; range 35-80 µg/L).

Study Design: All combinations of crosses between the Hyco and control fish were made using gametes from the collected fish. Fertilized eggs were exposed in egg cups to 0, 20 and 50 percent ash pond effluent under flow-through conditions. Percent hatch and swim-up success were measured. Swim-up larvae were released to exposure tanks where there were fed zooplankton collected from Hyco and the control lake. Larvae were observed for 28 days at which time survival and weight were measured.

Effects Data : Survival to the swim-up stage was different between larvae from Hyco females fertilized with either male type and those larvae from control females fertilized with either male type. All crosses involving a Hyco female resulted in larvae exhibiting 100 percent mortality prior to reaching swim-up. Percent survival from hatch to 28 days for larvae from control females exposed to control water and fed control lake zooplankton was only 5 and 12 percent for the two replicates so no meaningful comparisons can be made to the different dilution exposures or diet exposure. The mean concentrations of selenium in the ovaries, female liver and female muscle were 49, 130, and 84 µg/g dw, respectively.

Effect level: < 49, <130 and < 84 µg Se/g dw in adult ovaries, liver and muscle, respectively

Chronic Value: <59.92 µg Se/g dw estimated using the equation I to convert the selenium concentration in the muscle of Hyco females (84 µg Se/g dw) to a selenium concentration in whole-body.

Ingestion Study

Test Organism: Bluegill sunfish (*Lepomis macrochirus*; 30-day old larvae)

Exposure Route: Dietary and waterborne - field exposed adults
Juvenile bluegill from crosses with females in 0, 20 and 50 percent ash pond effluent were transferred to control water and fed zooplankton from either Hyco or the control lake. Selenium in Hyco and control zooplankton was 45 and 1.9 µg/g dw, respectively. Duration was not given.

Study Design: Survival and observations on pathology and morphology were made in the two diet treatments.

Effects Data: Mortality in larvae fed control zooplankton was 23.7 percent, whereas mortality in larvae fed Hyco zooplankton was 97.3 percent. There were no differences in survival (for two diet treatments) in larvae that were raised for the 30 days prior to the test in different effluent concentrations (0, 20 50 percent). The average selenium concentrations in the larvae fed control and Hyco zooplankton were 1.9 and 24.7 µg/g dw, respectively.

Effect level for larval survival: <24.7 µg Se/g dw in larvae

Chronic Value: None recommended for larval tissue.

Bryson, W.T., W.R.Garrett, M.A. Mallin, K.A. MacPherson, W.E. Partin, and S.E. Woock. 1985a. Roxboro Steam Electric Plant Hyco Reservoir 1983 Bioassay Report. Environmental Services Section. Carolina Power & Light Company. September 1985.

28-day Embryo/Larval Study

Test Organism: Bluegill sunfish (*Lepomis macrochirus*; embryos and larvae)

Exposure Route: dietary and waterborne - field exposed
Resident adult bluegill were collected from Hyco Reservoir in Person County, North Carolina and from a nearby control lake (Roxboro City Lake). Hyco Reservoir is a cooling lake for Carolina Power & Light and receives the discharge from the ash storage pond. For embryo/larval study up to swim-up stage, control fish were collected from the unaffected portion of Hyco.

Study Design: Repeat of 1982 28-day Embryo/Larval Study. Three crosses between: Hyco female and Hyco male; control female with Hyco male; and control female with control male. Gametes were fertilized and maintained for the 28-day test in ash pond effluent dilutions of 0, 20 and 50 percent. Percent hatch, percent swim-up success and survival were measured to 28 days post hatch. Two treatments were replicated and fed zooplankton collected from Hyco-affected and Hyco-unaffected (control). Larvae were observed for 28 days at which time survival and weight were measured.

Embryo/Larval Study up to Swim-up Stage. Five crosses were made between fish collected from the affected and unaffected areas. Percent hatch, percent swim-up and survival were measured until swim-up (approximately 3-4 days after hatch).

Effects Data : 28-day Embryo/Larval Study. All larvae that hatched from eggs obtained from Hyco females died prior to completing swim-up (see table below).

Effect level (larval survival): < 30, < 33 and < 59 µg Se/g dw for adult female bluegill in ovaries, liver and muscle, respectively

Summary of 28-day embryo larval study										
% effluent	parent source in cross M X F	% hatch	% swim-up	% survival, 28-days	adult tissue, µg Se/g dw					
					gonad		liver		muscle	
					M	F	M	F	M	F
0	H X H	92	0	0	33	30	43	33	62	59
20	H X H	98	0	0	33	30	43	33	62	59
20	H X H	92	0	0	33	30	43	33	62	59
50	H X H	97	0	0	33	30	43	33	62	59
0	H X C	89	87	18	33	2.2	43	4.4	62	2.7
20	H X C	96	96	34	33	2.2	43	4.4	62	2.7
50	H X C	60	84	58	33	2.2	43	4.4	62	2.7
0	C X C	79	95	40	nd	2.2	37	4.4	27	2.7
20	C X C	90	96	36	nd	2.2	37	4.4	27	2.7
20	C X C	88	97	25	nd	2.2	37	4.4	27	2.7
50	C X C	72	92	42	nd	2.2	37	4.4	27	2.7

Chronic Value: <43.70 µg Se/g dw estimated using equation I to convert the selenium concentration in the muscle of Hyco females (59 µg Se/g dw) to a selenium concentration in whole-body.

Embryo/larval study to swim-up. Percent swim-up of larvae from parents collected in non-affected Hyco averaged 93 percent, whereas percent swim-up from larvae collected from affected Hyco was 12 percent. Effect levels were determined for adult female and larval tissues. Larval tissues were averaged across effluent concentrations (geometric mean).

Effect level (percent swim-up):

Adult female ovaries: >9.1 µg/g dw; <30 µg/g dw
Adult female liver: >26 µg/g dw, <33 µg/g dw
Adult female muscle: >25 µg/g dw, <59 µg/g dw
Larvae: >12.8 µg/g dw; < 165 µg/g dw

Summary of Embyo/Larval Study up to Swim-up - Affected vs Unaffected Hyco											
date of fert.	Parents' capture location in Hyco	percent hatch			percent swim-up			selenium in tissue, µg/g dw			
		at % effluent			at % effluent			adult female			
		0	20	50	0	20	50	ovary	liver	musc	larvae
6-24	affected	93	98	94	0	0	0	30	33	59	0: 130 20: 120
6-27	affected	99	88	77	0	0	0	30	33	59	0: 130 20: 120
6-28	affected	29	34	35	25	14	3	30	33	59	0: 130 20: 120
6-28	affected	98	86	91	5	0	0	30	33	59	0: 130 20: 120
6-29	affected	88	93	85	59	42	25	30	33	59	0: 130 20: 120
7-14	unaffected	92	80	84	79	92	89	9.1	26	25	0: 19 20: 11 50: 10
7-26	unaffected	99	94	93	100	98	98	9.1	26	25	0: 19 20: 11 50: 10
7-27	unaffected	76	84	86	100	89	91	9.1	26	25	0: 19 20: 11 50: 10

Chronic Value: The chronic value estimated for the percentage larvae reaching the swim-up stage is presented as a range >25 µg Se/g dw in muscle tissue of Hyco females from the unaffacted area and >59 µg Se/g dw in muscle tissue of Hyco females from the affected area. Using equation I to convert the selenium concentration in the muscle of Hyco females to a selenium concentration in whole-body these values become >20.29 µg Se/g dw and <43.70 µg Se/g dw, respectively.

Bryson, W.T., K.A. MacPherson, M.A. Mallin, W.E. Partin, and S.E. Woock. 1985b. Roxboro Steam Electric Plant Hyco Reservoir 1984 Bioassay Report. Environmental Services Section. Carolina Power & Light Company

Ingestion Study

Test Organism: Bluegill sunfish (*Lepomis macrochirus;* juvenile- hatchery raised)

Exposure Route: Dietary only

Test Treatments: 5 diets: Se form (nominal selenium concentration in base diet)
seleno-DL-cystine (5 µg/g)
seleno-DL-cystine (10 µg/g)
seleno-DL-methionine (5 µg/g)
sodium selenite (5 µg/g)
Hyco zooplankton (5 µg/g)

Test Duration: 60 days

Study Design: Each treatment contained 40 fish which were maintained in a flow-through system. Fish were fed at 3 percent of their body weight. Length and weight were measured on days 30 and 60. Total selenium was measured in liver and whole-body.

Effects Data: No decreased length or weight in any of the Se-diets relative to the control.

Chronic Value: all values are whole-body
seleno-DL-cysteine: >2.16 µg Se/g dw
seleno-DL-cysteine-2X: >3.74 µg Se/g dw
seleno-DL-methionine: >2.46 µg Se/g dw
sodium selenite : >1.21 µg Se/g dw
Hyco zooplankton: >2.35 µg Se/g dw

Because none of the selenium-spiked diet formulations affected growth of juvenile fish at the concentrations tested, the chronic value selected for this study is >3.74 µg Se/g dw for the seleno-DL-cysteine-2X formulation.

Source and Exposure Embryo-Larval Study

Test Organism: Bluegill sunfish (*Lepomis macrochirus;* Adults from Hyco and a control lake)

Exposure Route: dietary and waterborne - field exposure

Test Treatments: Four treatments:
Hyco–collected fish exposed to Hyco water in flow through spawning tanks.
Hyco-collected fish in control water in flow through spawning tanks.

Control fish exposed to Hyco water in flow through spawning tanks.
Hyco-collected fish in control water in flow through spawning tanks.

Test Duration: Adult fish were in spawning tanks 4-7 months

Study Design: Eggs from each treatment were observed for percent hatch and percent swim-up.

Effects Data: Fish collected from the control lake did not spawn. Percent hatch and percent swim-up from Hyco fish in Hyco and control water are given in the table below. The percent hatch and percent swim-up were >83 and >83 for all the Hyco fish suggesting no effect for these endpoints.

Source of parents	Se in parental liver tissue, µg/g dw	water type for eggs and larvae	N	percent hatch	percent swim-up
Hyco	18.6	Hyco	16	86.6	91.1
Hyco	18.6	well water	10	83.8	95.5
Control	13.8	Hyco	a	a	83.3
Control	13.8	well water	12	86.0	97.4

a percent hatch unknown.

Chronic Value: The chronic value for this study is >18.6 µg Se/g dw liver tissue, or >5.45 µg of Se/g dw whole body tissue using equation III.

Gillespie, R.B. and P.C. Baumann. 1986. Effects of high tissue concentrations of selenium on reproduction by bluegills. Trans. Am. Fish. Soc. 115:208-213.

Test Organism: Bluegill sunfish, wild-caught (*Lepomis macrochirus*; adults; embryos and larvae)

Exposure Route: dietary and waterborne - field exposure

Test Treatments: High selenium adult fish were collected (electrofishing and with Fyke nets) from Hyco Reservoir. Low selenium adult fish were collected from Roxboro City Lake, Roxboro, NC.

Study Design: All possible combinations of bluegill parents from Hyco Reservoir and Roxboro City Lake were artificially crossed in June and July, 1982 and 1983, respectively. Fertilization success was assessed by stripping subsamples of 100 to 500 eggs per female and combining them with 2 ml of sperm. All zygotes were reared in Roxboro City Lake water and percent fertilization was estimated 2-3 hours later as the proportion of mitotically active zygotes. To estimate hatching success, gametes were combined as before and subsamples of 100 to 300 embryos per cross were transferred to egg cups and maintained in closed aquaria receiving recirculated Roxboro City Lake water. Percent hatch (approx. 2d at 22 to 25°C) was based on the number of yolk-sac larvae.

In 1982, about 200 embryos from 8 crosses were observed and preserved at intervals up to 40 h afer fertilization, and about 450 larvae were preserved at intervals of 40 to 180 h after fertilization. In 1983, about 1,800 larvae were observed and preserved from 40 to 150 hr from crosses involving females from Hyco Reservoir, and about 40-300 hr for crosses involving females from Roxboro City Lake (10 crosses total).

Effects Data: No significant differences were found in percent fertilization or in percent hatch among parent combinations from the 18 crosses made in June 1982 and July 1983. In contrast, larvae from all crosses involving a Hyco female were edematous; 100 percent of the larvae were abnormal in 7 of 8 crosses. Note: This outcome was observed when the same female from Hyco Reservoir was crossed with males from either Hyco Reservoir or Roxboro City Lake. The range of selenium concentrations in the ovaries of Hyco Reservoir females used for the cross experiments was from 5.79 to 8.00 (GM = 6.945 µg/g wet weight; n=7). The reported concentrations of selenium in ovaries and carcasses of females collected from Hyco Reservoir in 1982 and 1983 were 6.96 and 5.91 µg/g wet weight (n=22 and 28, respectively). The reported concentrations of selenium in ovaries and carcasses of females collected from Roxboro City Lake in 1982 and 1983 were 0.66 and 0.37 µg/g wet weight (n=14 and 19, respectively). The mean selenium concentration in bluegill larvae (n=222) from artificial crosses of parents from Hyco Reservoir was 28.20 µg Se/g dw.

Chronic Value: <21.47 µg Se/g dw estimated using equation II to convert the selenium concentration in ovaries of Hyco females (46.30 µg Se/g dw; assuming 85 percent moisture content) to a selenium concentration in whole-body.

Coyle, J.J., D.R. Buckler and C.G. Ingersoll. 1993. Effect of dietary selenium on the reproductive success of bluegills (*Lepomis macrochirus*). Environ. Toxicol. Chem. 12:551-565.

Test Organism: Bluegill sunfish (*Lepomis macrochirus*; two-year old pond-reared adult fish and resultant fry)

Exposure Route: Dietary and waterborne
Dietary
Seleno-L-methionine added in an aqueous solution to Oregon moist pellets; moisture content of diet was 25 percent.
Waterborne
Flow through,10 µg Se/L nominal, 6:1 ratio of selenate:selenite, 98 percent purity, adjusted to pH 2 with HCl to prevent bacterial growth and change in oxidation states of Se(IV) and Se(VI).

Test Duration: 140 days

Study Design: The experiment consisted of a test control and food control (see Test Treatment table below) with fish (n=28 initially) in the four remaining treatments fed one of the four seleno-methionine diets in combination with 10 µg Se/L in water. Spawning frequency, fecundity, and percentage hatch were monitored during the last 80 days of the exposure period. Survival of resulting fry (n=20) was monitored for 30 days after hatch. Adults and fry were exposed in separate, modified proportional flow-through diluters. Fry were exposed to the same waterborne selenium concentrations as their parents. Adults were fed twice daily *ad libitum*. Whole-body selenium concentrations in adult fish were measured at days 0, 60, and were calculated from individually analyzed carcass and gonadal tissue (ovaries and testes) at day 140. Eggs not used in percentage of hatch determinations were frozen and analyzed for total selenium.

Measured Se in:	Test Treatments					
	1 (test control)	2 (food control)	3	4	5	6
water (µg Se/L)	0.56	8.4	10.5	10.5	10.1	11.0
diet (µg Se/g dw)	0.76	0.76	4.63	8.45	16.8	33.3

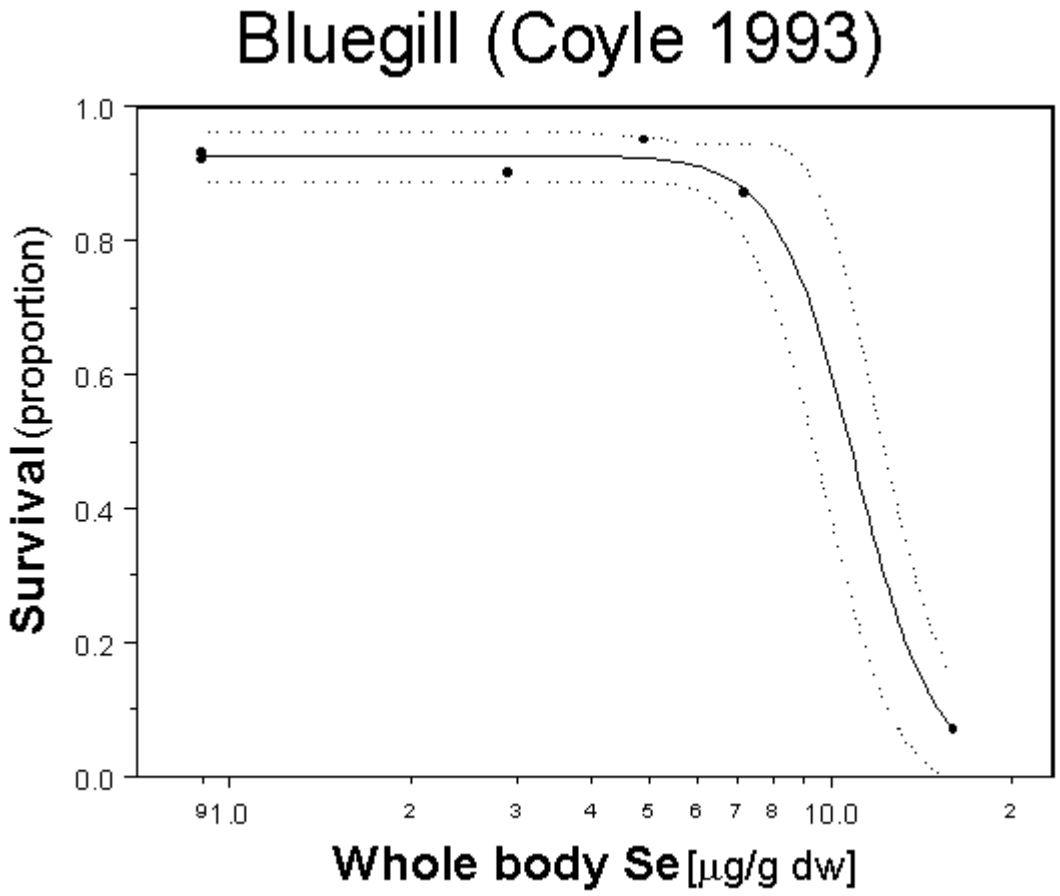
Effects Data: There was no effect of the combination of highest dietary selenium concentration (33.3 µg/g dw) in conjunction with exposure to a waterborne selenium concentration of 11.0 µg/L on adult growth (length and weight), condition factor, gonad weight, gonadal somatic index, or reproductive endpoints (i.e., spawning frequency, number of eggs per spawn, percentage hatch) during the 140-day exposure. The mean corresponding whole-body selenium concentration in adults

exposed to this waterborne and dietary selenium combination was 19 µg/g dw. Survival of fry from the exposed adults was affected by 5 days post-hatch. Concentrations of whole-body selenium in adult tissue at day 60 were used to determine effects in the fry because eggs were taken for the larval tests beginning at day 60 of the adult exposure.

Effects on Adults						
Se in diet, µg/g dw	Se in water, µg/L	whole-body Se (140 d), µg/g dw	replicate	total no. spawns	eggs/spawn	hatchability, %
0.8	0.5	0.8	A	15	14,099	94.5
			B	10	5,961	90.5
0.8	7.9	1.0	A	12	9,267	89.5
			B	11	9,255	84.5
4.6	10.5	3.4	A	20	9,782	86.5
			B	12	13,032	96.5
8.4	10.5	6.0	A	2	10,614	96.5
			B	9	7,995	90
16.8	10.1	10	A	13	10,797	83
			B	13	9,147	91.5
33.3	10.1	19	A	14	8,850	80
			B	4	8,850	80

Effects on Larvae			
Se in diet, µg/g dw	Se in water, µg/L	adult whole-body (60 d), µg/g dw	mean survival, %
0.8	0.5	0.9	92
0.8	7.9	0.9	93
4.6	10.5	2.9	90
8.4	10.5	4.9	95
16.8	10.1	7.2	87
33.3	10.1	16	7

Chronic Value: The EC_{20} value calculated for survival of fry versus levels of selenium found in the eggs and whole-body tissue of adults after 60 d of exposure is 8.954 µg Se/g dw.


Bluegill (Coyle 1993)
Survival (proportion)
1.0
0.8
0.6
0.4
0.2
0.0
9
1.0
2
3
4
5
6
7
8
10.0
2
Whole body Se [µg/g dw]

Cleveland, L. et al. 1993. Toxicity and bioaccumulation of waterborne and dietary selenium in juvenile bluegill sunfish (*Lepomis macrochirus*). Aquatic Toxicol. 27:265-280.

Test Organism: Bluegill sunfish (*Lepomis macrochirus*)
Life Stage: juvenile (5 months - waterborne exposure; 3 months - dietary exposure)

Exposure Route: waterborne (60-d) and dietary (90-d) - separate exposures
waterborne - 6:1 selenate:selenite at 0.17, 0.34, 0.68, 1.38, 2.73 mg/L; dietary - seleno-L-methionine in Oregon moist at 1.63, 3.25, 6.5, 13, 26 µg Se/g dw)

Study Design: Fish were exposed using a flow-through diluter. Each test consisted of an exposure and a depuration phase. Whole body tissue measurements were made at 31 and 60 days of waterborne exposure and at 31, 59 and 90 days of dietary exposure. Mortality and condition factor, K (weight x 10^5/length^3), were measured at selected intervals.

Effects Data : The waterborne exposure (see table below) was determined to have an EC_{20} = 4.07 µg Se/g dw (1.96-8.44 µg/g 95% CL). However, because it was a water-only exposure, it was not considered in the derivation of the FCV.
A mortality effect level for the dietary exposure could not be calculated because the highest selenium whole body concentration (13.4 µg Se/g dw) only had 17.5% mortality. The middle selenium concentration did have 22.5% mortality.
Cleveland et al. reported a significant decrease in K between 4.7 and 7.7 µg/g dw (see table below).

Waterborne Exposure Study

measured selenium in water (µg/L)	60-d measured selenium in whole body (µg/g dw)	60-d mortality (%)	condition factor (K)
20 (control)	1.1	10	1.5
160	2.8	12.5	1.5
330	4	22.5	1.6
640	5.3	52.5	1.5
1120	9.8	70	1.6
2800	14.7*	97.5	NA

*A 30-d measurement because all fish were dead at 60 days in this concentration.

Dietary Exposure Study

measured selenium in food (µg/g ww)	60-d measured selenium in whole body (µg/g dw)	60-d mortality (%)	condition factor (K)

0.68 (control)	1	5	1.3
2.3	2.1	7.5	1.3
3.5	3.3	10	1.3
6.6	4.7	22.5	1.3
12.7	7.7	15	1.2
25	13.4	17.5	1.2

Chronic Value: Given the very slight reduction in K (1.3 to 1.2) and the uncertain relevance of growth data, the NOAEC is interpreted to be 13.4 µg Se/g dw for this study; and the chronic value is >13.4 µg Se/g dw.

Lemly, A.D. 1993a. Metabolic stress during winter increases the toxicity of selenium to fish. Aquatic Toxicol. 27:133-158.

Test Organism: Bluegill sunfish (*Lepomis macrochirus;* juvenile 50-70 mm)

Exposure Route: Waterborne and dietary
Water
1:1 selenite:selenate in stock at pH 2; metered in to reach 5 µg/L
Diet
seleno-L-methionine in TetraMin (5 µg/g dw)

Test Duration: 180 days

Study Design: Fish were exposed (treatment and control) under intermittent flow-through conditions for 180 days. Tests were run at 4° and 20°C with biological (histological, hematological, metabolic and survival) and selenium measurements made at 0, 60, 120 and 180 days. Fish were fed at a rate of 3% body weight per day. All treatments were initiated at 20°C and then decreased in the cold treatment at a rate of 2°C per week for 8 weeks to reach 4°C and then maintained at that temperature for the remainder of the 180 days.

Effects Data : In the 20°C test, fish accumulated 6 µg/g dw selenium (whole-body) with no significant effect on survival (4.3% and 7.4% mortality in control and treatment, respectively). In the 4°C test, fish exposed to selenium accumulated 7.9 µg/g dw (whole-body) selenium and had significant mortality after 120 (33.6%) and 180 days (40.4%) relative to control (3.9%). Several hematological measurements were significantly different in both the warm and cold selenium exposures relative to controls. Both warm and cold selenium treatments also had greater O_2 consumption than controls. Fish lipid content in the cold Se treatment decreased more than the cold control; lipid content did not decrease in either the warm control or the warm Se treatment (see summary tables below). The results suggest significant mortality occurs in juvenile bluegill during winter months when tissue concentrations reach 7.91 µg/g dw and lipid levels decrease to 6 percent.

Chronic Value: 20°C, > 6 µg/g Se whole-body; 4°C, < 7.91 µg/g dw Se whole body

Mean Concentration of Selenium in Tissues, Cumulative Survival*, Percent Lipid Content and Oxygen Consumption in Juvenile Bluegill

day	cold - Se control				cold + Se				warm - Se control				warm + Se			
	Se[a]	Surv. %	lipid, %	O_2[b]	Se[a]	Surv. %	lipid, %	O_2[b]	Se[a]	Surv. %	lipid, %	O_2[b]	Se[a]	Surv. %	lipid, %	O_2[b]
0	1	100	13.2	98	1	100	13.2	98	1	100	13.2	98	1	100	13.2	98
60	1	97.1	12.5	58	5.8	92.9	10	63	1.2	95.7	13.3	98	5.8	100	13.3	103
120	1.1	97.1	11.5	57	7.9	66.4	6	81	1.1	95.7	13.4	100	6	96.7	13.4	120
180	1.4	97.1	10.5	57	7.9	59.6	6	78	1.2	95.7	13.6	100	6	92.6	13.5	120

a whole body Se tissue concentration, µg/g dw
b oxygen consumption, mg/kg/hr

* Cumulative Survival: In this experiment, 240 juvenile bluegill were placed in three 400-L fiberglass tanks, 80 in each, and exposed to each control and treatment for a period of 180 days. Ten fish were removed at random from each treatment replicate on days 0, 60, 120, and 180 for selenium, histological, hematological, and metabolic measurements.

Replicate and Average Whole-body concentrations (µg/g dry weight) of selenium in juvenile bluegill*

	day 0				day 60				day 120				day 180			
replicate	1	2	3	mean	1	2	3	mean	1	2	3	mean	1	2	3	mean
c+Se	0.87	1.21	0.95	1.01	6.30	5.49	5.76	5.85	8.36	7.31	7.85	7.84	7.53	8.01	8.19	7.91
w+Se	1.17	0.96	0.90	1.01	5.61	6.19	5.43	5.74	6.37	5.92	5.50	5.93	5.48	5.72	6.02	5.74
c-Se	0.89			0.89	0.97			0.97	1.01			1.01	1.10			1.10
w-Se	0.99			0.99	1.12			1.12	0.99			0.99	0.96			0.96

* Each value is for a composite sample made from 5 fish.

The Kaplan-Meier estimator was used to calculate survival at time *t*

$$\hat{S}(t) = \prod \frac{r(t_i) - d_i}{r(t_i)}$$

where $r(t_i)$ is the number of fish alive just before time t_i, i.e. the number at risk, and d_i is the number of deaths in the interval $I_i = [t_i, t_{i+1}]$. The 95% confidence interval for such estimate (Venables and Ripley 2002) was computed as

$$\exp\left\{ - \hat{H}(t) \exp\left[\pm k_\alpha \frac{\text{s.e.}\left(\hat{H}(t)\right)}{\hat{H}(t)} \right] \right\}$$

where

$$\hat{H}(t) = \sum \frac{d_j}{r(t_j)} \qquad \text{and } j \le i$$

The following table lists the estimates of survival in the cold + Se treatment at 60, 120 and 180 days. The term n.event is the number of deaths at a given interval; n.risk is the number of organisms alive at the beginning of the interval; survival is computed by the Kaplan-Meier estimator.

Time	n.risk	n.event	survival	std.err	lower 95% CI	upper 95% CI
60	210	15	0.929	0.0178	0.884	0.956
120	165	47	0.664	0.0350	0.590	0.728
180	88	9	0.596	0.0381	0.517	0.666

Hematological Measurements in Juvenile Bluegill Sunfish (*indicates significantly different from control)

Warm Exposure	day 0		day 60		day 120		day 180	
blood parameter	warm-Se	warm+Se	warm-Se	warm+Se	warm-Se	warm+Se	warm-Se	warm+Se
total erythrocyte, 10^6/ml	2.95	2.92	2.96	2.93	2.99	2.95	2.96	2.89
% mature	85	86	86	93*	86	94*	85	94*
nuclear shadows, 10^4/ml	0.95	0.86	0.97	2.05*	0.83	2.38*	0.91	2.30*
total leucocytes, 10^4/ml	17.22	17.41	16.90	17.55	16.73	17.62	17.05	17.36
% lymphocytes	23	25	20	23	19	26	21	22
% neutrophils	15	13	14	15	17	19	17	16
hematocrit, %	37	36	37	29*	36	29*	38	28*
MCHC (mean corpuscular hemoglobin conc.)	23	25	25	19*	25	18*	25	17*
Cold Exposure	day 0		day 60		day 120		day 180	
blood parameter	cold-Se	cold+Se	cold-Se	cold+Se	cold-Se	cold+Se	cold-Se	cold+Se
total erythrocyte, 10^6/ml	2.91	2.93	2.97	2.90	3.01	2.95	3.00	2.99
% mature	84	82	87	95*	85	96*	85	97*
nuclear shadows, 10^4/ml	0.86	0.84	0.83	2.30*	0.89	2.49*	0.90	2.36
total leucocytes, 10^4/ml	16.48	16.88	16.79	16.91	16.80	16.74	16.96	16.63
% lymphocytes	17	16	16	17	19	15	19	18
% neutrophils	13	12	15	11	15	12	12	14
hematocrit, %	39	37	40	30*	41	28*	39	27*
MCHC (mean corpuscular hemoglobin conc.)	26	25	25	18*	22	17*	23	17*
MCV (mean corpuscular volume)	182	171	188	146*	180	135*	185	130*

Hermanutz et al. 1996. Exposure of bluegill (*Lepomis macrochirus*) to selenium in outdoor experimental streams. U.S. EPA Report. Mid-Continent Ecology Division. Duluth, MN.

Test Organism: Bluegill (*Lepomis macrochirus*; 3 to 4-year old adults)

Exposure Route: Dietary and waterborne followed by dietary only

Dietary and waterborne

Selenite was added to artificial streams which entered the food web; thus, fish were also exposed to selenium in the diet.

Dietary only

Recovering streams exposed bluegill to selenium in prey organisms. Selenite addition to water was ceased (selenium in water was below detection level).

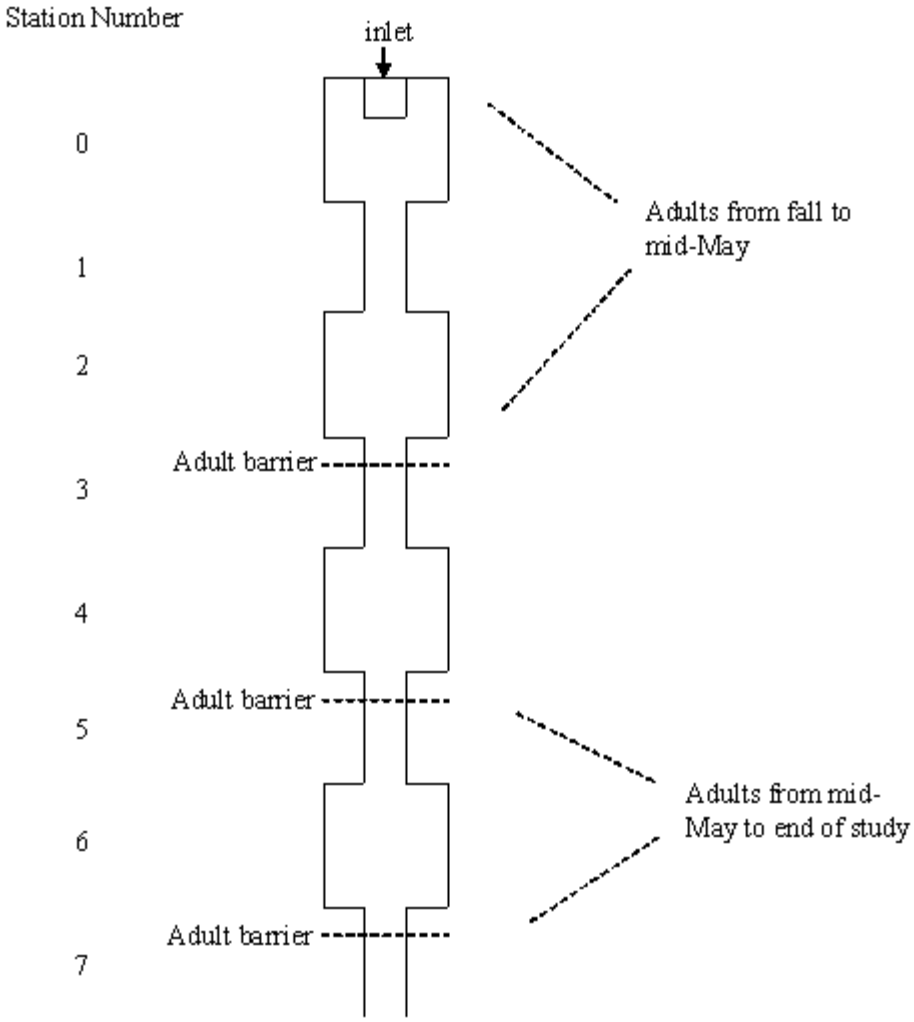
Study Design: Eight Monticello artificial streams were used for three separate studies between 1987 and 1990.

Stream	Study I	Study II	Study III
Dates BG[a] put in station 0-2 BG transferred to sta. 6 End of study	 9-1-87 5-16-88 8-22-88	 10-88 5-89 8-89	 11-89 5-90 7-90
1	Unused	Control	Control
2	Unused	2.5 µg/L	Recovering
3	10 µg/L	10 µg/L	Recovering
4	30 µg/L	Recovering	Recovering
5	Control	Control	Control
6	30 µg/L	Recovering	Recovering
7	Control	2.5 µg/L	Recovering
8	10 µg/L	10 µg/L	Recovering

[a] BG = Bluegill.

A schematic diagram of an artificial stream is provided below. For each study, a random sample of 22-50 adult bluegill were transferred from stations 0-2 (provided temperatures above 4°C during winter) to station 6 (most suitable for nests) during mid-May for spawning. Spawning activity was monitored in the streams. Embryo and larval observations were made *in situ* and in the laboratory from fertilized eggs taken from the streams and incubated in the lab.

Schematic Design of One of the Artificial Streams in the Monticello Study



Effects Data : Adult survival in Studies II and III was very low and will not be considered in the effects analysis. The percent hatch, percent larval survival, percent edema, percent lordosis and percent hemorrhaging in the 2.5 and 10 µg/L streams for Study II are provided in the table below. The values presented in this table are corrected values for Study II as reported by Tao et al. (1999). The data from Study II (both egg cup and field nest) were not amenable for regression analysis. As reported by Tao et al. (1999), ANOVA was utilized to evaluate effects of elevated concentrations of selenium on percent hatch, percent survival, maximum percent edema, lordosis, and hemorrhage, and minimum percent healthy (egg cup data). Treatment effects were only significant for maximum percent edema and minimum percent healthy (see their Table 4-19), and in no instance were

differences between the 2.5 µg Se/L and control treatments significant (Dunnett's Means test, all probabilities > 0.1, see their Table 4-20). These results clearly suggest that the 2.5 µg Se/L treatment had no adverse impact on bluegill larvae. They are further supported by analysis of the field nest data (see table below). In this experiment, treatment had a significant effect on maximum percent edema (raw data and ranks) and maximum percent hemorrhage (ranks only). Probabilities of differences between the 2.5 µg Se/L and control treatments were >0.2 for all response variables except maximum percent hemorrhage, which had an estimated probability of 0.05 (raw data, P=0.022 for ranks; Dunnett's means test). Such values, though, were well above the adjusted experiment-wise error rate for multiple comparison (α'=0.0085, $\alpha'=1-(1-\alpha)^{1/k}$; α=0.05, k=6 comparisons; Sokal and Rohlf 1981), which takes into account the fact that selenium effects were tested on six different response variables. Therefore, the chronic value for this study, 12.12 µg Se/g dry weight, was calculated as the geometric mean of tissue concentrations of selenium in the 2.5 (NOAEC) and 10 µg Se/L (LOAEC) treatments (5.55 and 26.46 µg Se/g dw whole body tissue, respectively).

Chronic Value: 12.12 µg Se/g dw whole-body tissue, calculated as the GM of the NOAEC, 5.55 µg Se/g dw, and LOAEC, 26.46 µg Se/g dw, based on percent larval survival and percent larvae exhibiting edema in the egg cup exposures. Note: the NOAEC value of >17.35 µg Se/g dw was selected as the chronic value for Study III based on percent larval survival in egg cup exposures and percent larvae exhibiting edema in nest observations.

Effects on Progeny - Study II[a,b]

Egg cup observations								
treatment	stream	number of trials	% hatch	% survival to 3rd day	% edema	% lordosis	% hemorr	whole-body Se (µg/g dw)
control	1	6	93.0	75.2	0	0	0	2.05
control	5	5	96.4	71.5	0	0	0	1.85
2.5 µg/L	2	0	NA	NA	NA	NA	NA	6.8
2.5 µg/L	7	4	81.4	71.6	0	0	3.6	5.55
10 µg/L	3	3	83.3	57.7	100	11.1	49.3	20.75
10 µg/L	8	2	91.1	57.1	100	18.2	41.1	33.75
rec 30 µg/L	4	0	NA	NA	NA	NA	NA	NA
rec 30 µg/L	6	6	92.9	73.0	17.4	0	11.5	30.6

Nest Observations											
treatment	stream	# active nests	# embryos collected	% dead embryos	# larvae collected	% dead larvae	#samples w larvae	% edema	% lordosis	% hemorr	whole-body Se (µg/g dw)
control	1	6	2458	0.94	3252	0.03	7	0	0	0	2.05
control	5	9	1329	0	3435	1.05	13	0	0	0	1.85
2.5 µg/L	2	1	0		2497	0.20	3	4.1	25	77.6	6.8
2.5 µg/L	7	5	1462	0	4717	0.08	8	0	0	52	5.55
10 µg/L	3	2	672	0	5376	0.50	9	81.4	5.0	55.5	20.75
10 µg/L	8	3	931	0.32	750	0.40	4	50	14.7	26.7	33.75
rec 30 µg/L	4	0	NA	NA	NA	NA	NA	NA	NA	NA	NA
rec 30 µg/L	6	8	646	0	6782	7.8	16	27.3	0	17.1	30.6

a Values in table were taken from Tao et al. (1999).

b The chronic value for the study was calculated as the GM of whole-body selenium concentrations in the 2.5 (NOAEC 5.55 µg Se/g dw; stream 7 only) and 10 µg Se/L (LOAEC of 26.46 µg Se/g dw; GM of streams 3 and 8, respectively) treatments in the egg cup exposures.

Effects on Progeny - Study III[a]

Egg cup observations								
treatment	Stream	number of trials	% hatch	% survival to 3rd day	% edema	% lordosis	% hemorr	whole- body Se (µg/g dw)
control	1	2	92	58.6	0	0	0	1.6
control	5	3	76.7	69.2	0	0.9	0.8	3.35
rec 2.5 µg/L	2	3	87.3	66	0	0	0	5.25
rec 2.5 µg/L	7	6	87.2	76.5	0	0	0	5.35
rec 10 µg/L	3							14.5
rec 10 µg/L	8	3	75.3	74.5	0	0	0	11.7
rec 30 µg/L	4	5	92	78				17.35
rec 30 µg/L	6							

Nest observations							
treatment	stream	# active nests	# samples with larvae	% edema	% lordosis	% hemorr	whole-body Se (µg/g dw)
control	1	2	5	0	0	0	1.6
control	5	2	3	0	0	0	3.35
rec 2.5 µg/L	2	5	5	0	0	0	5.25
rec 2.5 µg/L	7	5	2	0	0	0	5.35
rec 10 µg/L	3	2	4	0	0	0	14.5
rec 10 µg/L	8	4	4	0	0	0	11.7
rec 30 µg/L	4	9	13	0	0	0	17.35
rec 30 µg/L	6						

a The chronic value for the study was selected as the NOAEC of >17.35 µg Se/g dw from the recovering 30 µg Se/L treatment.

Coughlan, D.J. and J.S. Velte. 1989. Dietary toxicity of selenium-contaminated red shiners to striped bass. Trans. Am. Fish Soc. 118:400-408.

Test Organism: Striped bass (*Morone saxitilis*; adults from Lake Norman, NC, approximately 250 g each)

Exposure Route: dietary only
Treated fish were fed selenium contaminated red shiners (1 g) from Belews Lake, NC (9.6 µg Se/g ww or 38.6 µg Se/g dw based on a mean reported moisture content of 75.1 percent). Control fish were fed golden shiners from a local bait dealer (0.3 µg Se/g ww or 1.3 µg Se/g dw based on a mean reported moisture content of 76.3 percent).

Test Treatments: Test treatments were as described above. Two tanks contained treated fish (n = 20 fish total), and one tank of fish served as the control (n = 10 fish). Each tank received a continuous flow of soft well water (hardness and alkalinity approx. 30 mg/L as $CaCO_3$) throughout the exposure.

Test Duration: 80 days

Study Design: During the experiment, all striped bass (n = 10 per tank) were fed to satiation three times per day. Pre-weighed rations of live red shiners (treated fish) and golden shiners (controls) were added to the tanks and allowed 5 hours to feed. Uneaten prey was removed and weighed. Composite whole-body samples of each prey fish were collected at regular intervals throughout the study for whole-body tissue selenium analysis. The final selenium concentration in epaxial white muscle was determined for surviving striped bass at the end of the test. Moribund striped bass were sacrificed so as to obtain muscle tissue samples for selenium analysis. Samples of liver and trunk kidney of these and the surviving striped bass were dissected for observations of histopathology.

Effects Data: Striped bass fed selenium-laden red shiners exhibited changes in behavior (lethargy, reduced appetite), negligible weight gain, elevated selenium concentrations in muscle, histological damage, and death. Control fish ate and grew well, and behaved normally. Average selenium ingestion was between 60 and 140 µg Se/fish per day until day 30. Appetite of the treated fish appeared to be significantly reduced beyond this point compared to the appetite of the control group. By day 78, all striped bass fed the Se-laden red shiners either had died or were moribund and sacrificed for analysis. The final selenium concentration in muscle of treated striped bass averaged from 3.5 (tank 1) and 4.0 (tank 2) µg/g ww, or 17.5 and 20.0 µg/g dw, respectively, assuming 80 percent moisture content in muscle tissue. The final selenium concentration in muscle of control striped bass fed uncontaminated golden shiners averaged 1.1 µg/g ww, or 5.50 µg/g dw (assuming 80 percent moisture content in muscle tissue).

Chronic Value: The chronic value for percent survival of striped bass relative to final selenium in muscle tissue after being fed Se-laden red shiners is <17.50 µg/g dw, or 14.75 µg/g dw whole body tissue converted using equation I.

An EC_{20} value could not be calculated for this data set because the data did not meet the assumptions required for analysis.

Lemly, A.D. 1993b. Teratogenic effects of selenium in natural populations of freshwater fish. Ecotoxicol. Environ. Safety. 26: 181-204.

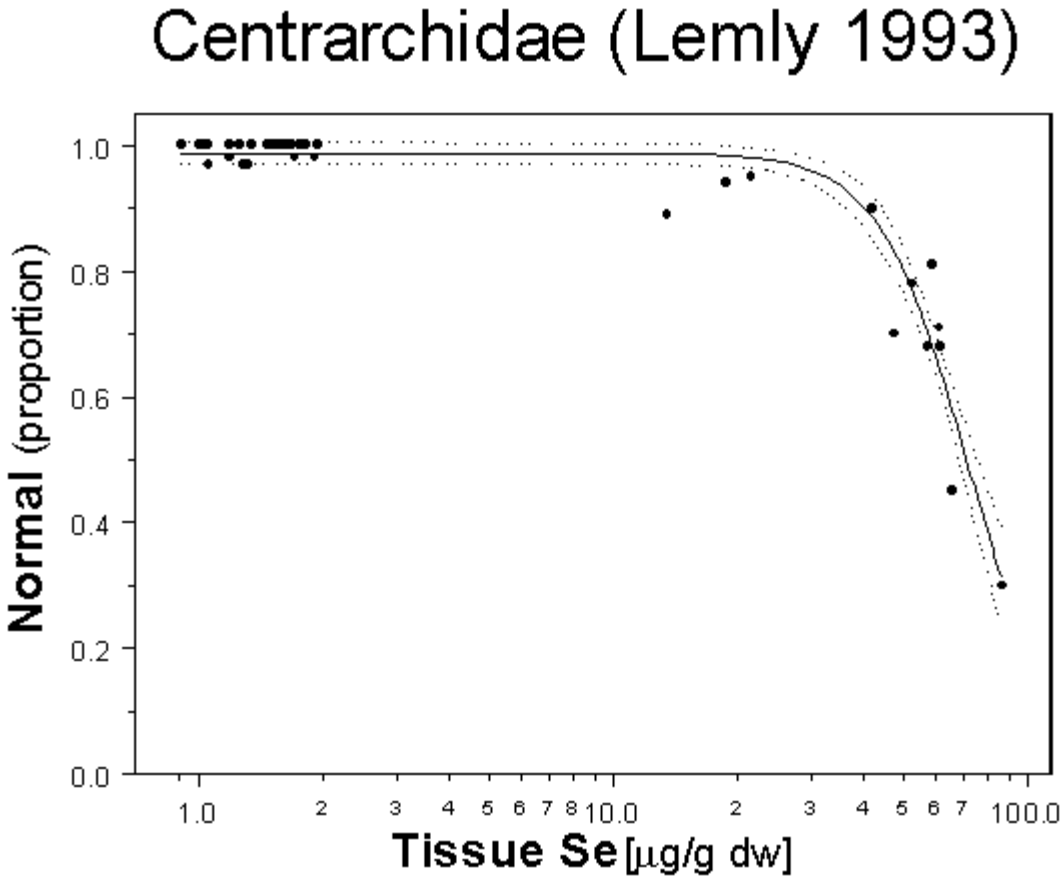
Test Organism: All possible fish species collected from Belews Lake and a reference site.

Exposure Route: dietary and waterborne - field exposed

Study Design: Surveys of external abnormalities in fish collected from Belews Lake and two reference lakes were done in 1975, 1978, 1982, and 1992. Five classifications of abnormalities were reported: (1) spinal deformities (lordosis, scoliosis, kyphosis); (2) accumulation of body fluid (edema, expothalmus or popeye); (3) missing or abnormal fins; (4) abnormally shaped head or mouth; and (5) cloudy eye lens or cornea (cataracts). Whole-body selenium was measured in each fish. The relationship between whole-body selenium and malformations was examined.

Effects Data : The relationship between whole-body selenium and the frequency of malformations in all the fish species collected at Belews (n=22) did not follow a clear trend. When evaluating only fish from the family Centrarchidae using a polynomial regression (cubic model) an R^2 value of 0.881 was obtained. Lemly reported that the inflection point where a rapid rise in deformities occurred was between 40 and 50 µg Se/g dw in whole-body tissue. The EC_{20} value determined by regression analysis of percent normal fish versus whole-body tissue selenium concentration for the family Centrarchidae (most sensitive family or group of families) was 44.57 µg Se/g dw. Centrarchidae was the most sensitive family or group of families of those collected during the survey.

Chronic Value: The EC_{20} value determined by regression analysis of percent normal fish versus whole-body tissue selenium concentration for the family Centrarchidae was 44.57 µg Se/g dw.


Centrarchidae (Lemly 1993)
Normal (proportion)
1.0
0.8
0.6
0.4
0.2
0.0
1.0
2
3
4
5
6
7
8
10.0
2
3
4
5
6
7
100.0
Tissue Se [µg/g dw]

APPENDIX J

SELENIUM (µg/g dw WHOLE-BODY) IN FISH SAMPLES COLLECTED FROM 112 SITES AS PART OF U.S. FISH AND WILDLIFE'S NATIONAL BIOMONITORING PROGRAM, 1978-1981 (LOWE ET AL. 1985).

AND

SELENIUM (µg/g dw WHOLE-BODY) IN 322 AQUATIC LIFE TISSUE SAMPLES COLLECTED FROM 264 SITES AS PART OF USGS NATIONAL WATER QUALITY ASSESSMENT (NAWQA) PROGRAM (http://water.usgs.gov/nawqa/ as of May 11, 2004).

FCV Relative to Natural Background Levels of Selenium in Fish

As an essential element, selenium naturally occurs in all living things. Since selenium is found in all fish, two questions arise. 1) How close is the FCV of 7.91 µg/g dw to natural background levels in fish, and 2) how frequently do natural selenium tissue concentrations exceed the FCV. The latter situation would pose problems in the implementation of the FCV as an ambient water quality criterion.

As part of the National Contaminant Biomonitoring Program, the U.S. Fish and Wildlife Service collected fish from 112 sites distributed evenly across the U.S. during 1979 through 1981, and measured several contaminants including selenium (Lowe et al. 1985). Selenium, measured in 591 fish samples representing 60 different species, ranged from 0.3 to 10.5 µg/g dw and had an overall average and standard deviation of 1.9 ± 1.4 µg/g dw.

A separate data set of selenium levels in 231 macroinvertebrate samples, 90 fish samples, and one plant sample collected from 25 different states across the United States was generated by USGS's National Water Quality Assessment (NAWQA) program. NAWQA is intended to measure water quality in a sampling of smaller watersheds having known land use. Among these sites, whole body tissue concentrations ranged from 0.3 to 22.37 µg/g dw and had an overall average and standard deviation of 3.22 ± 2.29 µg/g dw. The distribution of both these data sets indicates that the FCV would not be exceeded by over 97 percent of aquatic tissue samples collected across the United States (Figure J-1). The FCV thus appears to be sufficiently greater than natural selenium levels that unavoidable exceedances of the criterion are unlikely.

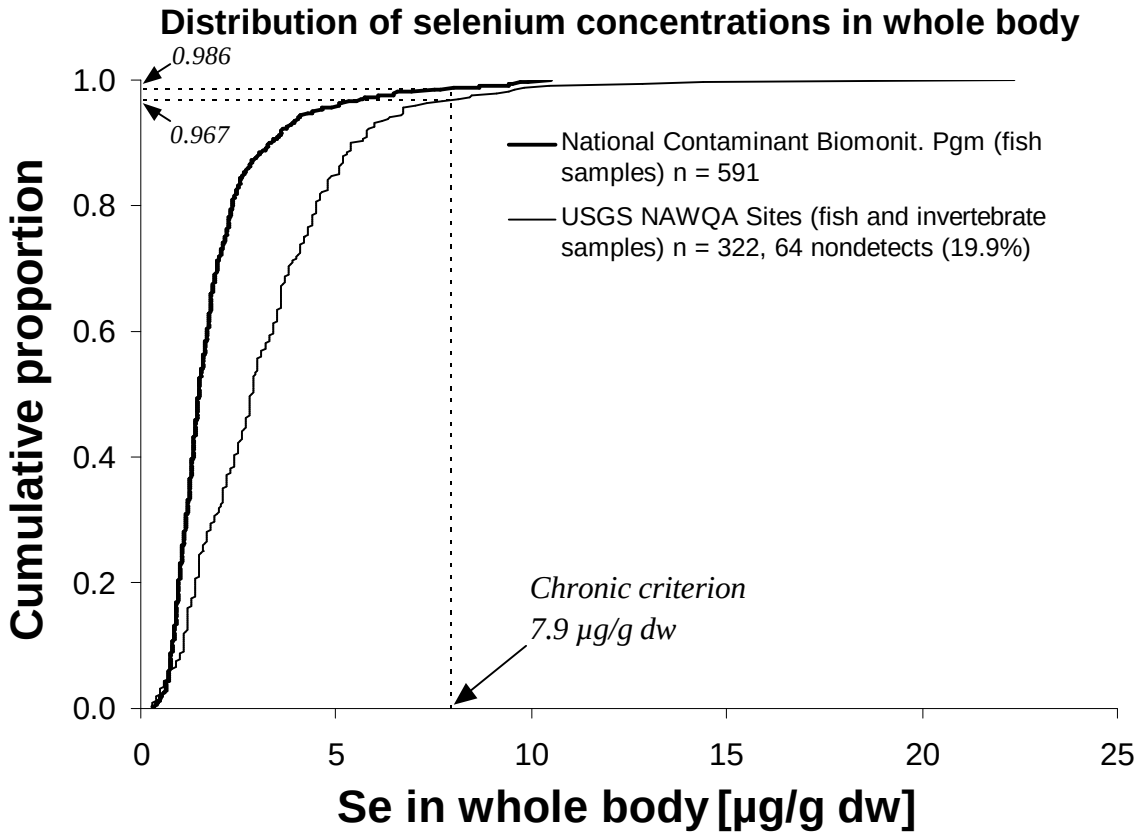


Figure J-1. Cumulative distribution of selenium concentrations in aquatic organisms (whole-body, µg/g dw) collected by the National Contaminant Biomonitoring Program (NCBP) and the U.S. Geological Service National Water-Quality Assessment (NAWQA) Program. NCBP and NAWQA data from Lowe et al. (1985) and query results from NAWQA's database on contaminant concentrations in animal tissues (http://water.usgs.gov/nawqa/), respectively.

Table J-1. Selenium (µg/g dw whole-body) in fish samples collected from 112 sites as part of U.S. Fish and Wildlife's National Biomonitoring Program, 1978-1981. From Lowe et al. 1985

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
		Station 1, Penobscot River at Old Town, MA		
78	Smallmouth bass	12.9	1.2	0.8562
78	White sucker	13.7	1.1	1.2227
78	White sucker	14.4	1.3	0.9292
80	Smallmouth bass	12.8	1.1	0.6513
80	White sucker	15.2	1.3	0.8261
80	White sucker	15.3	1.4	0.7634
		Station 2, Connecticut River at Windsor Locks, Conn.		
78	White catfish	16.6	2.3	0.4651
78	White catfish	16.5	2.3	0.6818
78	Yellow perch	8	0.3	0.9934
80	White catfish	14.5	0.9	0.6007
80	White catfish	13.3	0.9	0.9738
80	Yellow perch	9.5	0.4	0.9811
		Station 3, Hudson River at Poughkeepsie, NY		
78	Goldfish	11	1	0.9353
78	Goldfish	11.4	1.1	0.6545
78	Largemouth bass	11.1	0.8	1.0676
80	Goldfish	10.9	1	1.2333
80	Largemouth bass	14.8	2.2	1.0701
		Station 4, Delaware River at Trenton, NY- Yardley, Pa.		
79	White perch	7.3	0.2	4.6429
79	White sucker	12.8	0.8	1.1438
79	White sucker	14.3	1.2	0.8389
81	Largemouth bass	9.5	0.4	2.4206
81	White sucker	15	1.3	1.1864
81	White sucker	14.4	1.1	1.4423
		Station 5, Susquehanna River at Conowingo Dam, Md.		
79	Common carp	12.9	2	2.0690
79	Common carp	16.9	2.3	2.2381
79	White perch	7.6	0.3	5.5401
81	Common carp	14.4	1.6	2.5431
81	Common carp	14.1	1.7	1.5358
81	White perch	7.9	0.3	3.4951
		Station 6, Potomac River at Little Falls Md.- McLean Va		
79	Common carp	18.7	3.1	1.5248
79	Common carp	17	2.5	1.1628
79	Smallmouth bass	10	0.5	2.6587
81	Largemouth bass	11.5	8	1.8474
81	Redhorse	17.2	2	1.2963
81	Redhorse	17.5	2.1	1.3208
		Station 7, Roanoke river at Roanoke Rapids, N.C.		

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
78	White catfish	12.7	0.7	1.2134
80	Striped bass	14.5	1.4	1.3665
80	White catfish	11.7	0.6	1.0473
80	White catfish	10.1	0.4	1.0164
		Station 8, Cape Fear River at Elizabethtown, NC		
78	Spotted sucker	16.3	2	2.5177
78	Spotted sucker	16	2	2.5263
80	Flathead catfish	19	2	1.0656
80	Quillback	15	1.7	1.6719
80	Quillback	14.9	1.1	1.6558
		Station 9, Cooper River at Lake Moultrie, Monck;s Coner, S.C		
78	Channel catfish	16.3	1.3	1.6078
78	Channel catfish	14.6	1	1.4563
80	Channel catfish	14.5	1	1.4497
80	Channel catfish	13.6	0.6	1.4917
80	Striped bass	20.6	3.3	1.4894
		Station 10, Savannah River at Savannah, Ga		
78	Channel catfish	11	0.4	3.2444
78	White catfish	12.7	1	2.0248
80	White catfish	11.3	0.7	1.4592
80	White catfish	7.9	0.2	1.2319
80	Bowfin	21	3.6	2.2568
		Station 12, St. Lucie Canal at Indiantowm, Fla		
78	Largemouth bass			1.0954
78	White catfish			1.0580
78	White catfish			0.7931
80	Largemouth bass			1.1837
80	White catfish			1.3208
80	White catfish			0.9690
		Satation 13, Appalachicola River at J. Woodruff Dam, Fla.		
79	Largemouth bass			0.8803
79	Spotted sucker			1.8219
79	Spotted sucker			1.0980
81	Largemouth bass			0.9402
81	Spotted sucker			1.3060
81	Spotted sucker			1.5600
		Station 14, Tombigbee Tiver at McIntosh, Ala.		
79	Smallmouth buffalo			0.7325
79	Smallmouth buffalo			1.1513
81	Black crappie			1.2545
81	Blue catfish			0.8765
81	Blue catfish			0.7782
		Station 15, Mississippi Tiver at Luling, La.		

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
79	Common carp			1.6667
79	Common carp			1.7162
79	Largemouth bass			1.7200
81	Channel catfish			0.6599
81	Channel catfish			0.7561
81	Largemouth bass			1.8147
		Station 16, Rio Grande at Mission, Tex		
80	Gizzard shad			2.4638
80	Gizzard shad			2.4719
80	Largemouth bass			2.2800
81	Common carp			2.1858
81	Gizzard shad			2.6190
81	Gizzard shad			2.8125
		Station 17, Genessee River at Scottsville, NY		
80	Pumpkinseed			0.9901
80	Redhorse			0.7692
80	Redhorse			0.7328
81	Pumpkinseed			2.1186
81	Redhorse			1.2450
81	Redhorse			1.3853
		Station 18,, Lake Ontario at Prot Ontario, NY		
78	Rock bass			1.1355
79	Yellow perch			1.3306
79	Yellow perch			1.1719
81	Rock bass			1.4886
81	Yellow perch			1.7293
81	Yellow perch			1.3383
		Station 19, Lake Erie at Erie, Pa		
80	Redhorse			1.7625
80	Redhorse			1.7241
80	Yellow perch			2.4576
		Station 20, Lake Huron (Saginaw Bay) at Bay port, Mich.		
79	Common carp			1.8237
79	Common carp			1.9113
79	Yellow perch			1.9196
81	Common carp			2.3355
81	Common carp			2.5776
81	Yellow perch			2.1723
		Station 21, Lake Michigan at Sheboygan, Wis.		
79	Bloater			0.8060
79	Bloater			0.6897
79	Lake trout			1.1730
81	Bloater			0.7104

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
81	Bloater			0.9687
81	Lake trout			1.2828
		Station 22, Lake Superior at Bayfield, Wis.		
79	Lake trout			0.8911
79	Lake whitefish			1.3278
79	Lake whitefish			1.5058
81	Bloater			1.1304
81	Bloater			1.3419
81	Lake trout			1.4741
		Station 23, Kanawha River at Winfield, W.VA		
78	Channel catfish			0.9091
78	Channel catfish			0.8841
78	Sauger			1.4334
80	Channel catfish			0.9508
80	Channel catfish			1.2635
80	Sauger			2.3651
		Station 24, Ohio River at marietta, Ohio- Williamstown, W VA		
78	Channel catfish			1.1871
78	Sauger			1.4716
80	Common carp			2.2819
80	Common carp			1.7687
80	Sauger			2.2511
		Station 25, Cumberland River at Clarksville, Tenn.		
78	Common carp			1.3793
78	common carp			1.8077
78	White catfish			1.2203
80	Common carp			1.3514
80	Common carp			1.5909
80	Largemouth bass			1.7669
		Station 26, Illinois River at Beardstown, Ill.		
78	Black crappie			0.8638
78	Common carp			2.0438
78	Common carp			2.6766
80	Black crappie			1.5751
80	Common carp			1.8051
80	Common carp			2.1687
		Station 27, Mississippi River at Gutenburg, Iowa- Glen Haven, Wis.		
78	Common carp			1.7628
78	Common carp			1.3907
78	Largemouth bass			2.2742
80	Common carp			1.3231
80	Common carp			0.9064
80	Largemouth bass			1.1885

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
		Station 28, Arkansas River at Pine Bluff, Ark.		
79	Bluegill			0.8936
79	Common carp			1.2453
79	Common carp			1.0357
81	Common carp			1.7931
81	Common carp			1.3937
81	Largemouth bass			1.3011
		Station 29, Arkansas River at Keystone Reservoir, Okla.		
79	Common carp			1.3974
79	Common carp			1.6509
79	White bass			2.2167
81	Common carp			2.2394
81	Common carp			1.3410
81	White crappie			1.0738
		Station 30, White River at De Valls Bluff, Ark.		
79	Freshwater drum			0.8874
79	Freshwater drum			0.9091
79	Largemouth bass			0.8696
81	Common carp			2.2857
81	Common carp			1.7472
		Station 31, Missouri River at Nebraska City, Nebr.- Hamburg, Iowa		
79	Common carp			1.8774
79	Common carp			2.8163
79	Goldeye			1.2712
81	Common carp			3.0189
81	Common carp			3.2051
81	Goldeye			3.1803
		Station 32, Missouri River at Garrison Dam, N. Dak.		
79	Northern pike			1.4884
79	Redhorse			0.9600
81	Walleye			1.6041
81	White sucker			2.4883
81	White sucker			3.9252
		Station 33, Missouri River at Great Falls Mont.		
79	Brown trout			2.3432
79	White sucker			1.3333
79	White sucker			1.3158
81	Brown trout			2.1591
81	White sucker			1.9617
		Station 34, Red River of the Norh at Noyes, Minn. _ Pembina, N. Dak.		
78	Common carp			2.3166
78	Common carp			2.0629
78	Sauger			0.4682
80	Mooneye			3.3754

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
80	Sauger			0.9328
80	Sauger			0.8117
		Station 35, Green River at Vernal, Utah		
78	Common carp			3.7410
78	Common carp			3.9286
78	Smallmouth bass			3.6076
80	Common carp			3.2537
80	Common carp			2.7811
80	Smallmouth bass			3.1500
		Station 36, Colorado River at Imperial Reservoir, Ariz.- Calif.		
78	Common carp			6.5552
78	Common carp			8.0364
78	Largemouth bass			10.5204
80	Common carp			7.5210
80	Common carp			6.4783
80	Largemouth bass			8.6531
		Station 37, Truckee River at Fernley, Nev.		
80	Green sunfish			1.0794
80	Tahoe sucker			0.9211
80	Tahoe sucker			1.1401
81	Green sunfish			0.8835
		Station 38, Utah lake at Provo, Utah		
78	Common carp			2.9333
78	Common carp			3.1741
78	White bass			3.4799
80	Common carp			9.6863
80	Common carp			2.1633
80	White bass			3.5246
		Station 39, Sacramento River at Sacramento, Calif.		
79	Brown bullhead			0.7035
79	Largemouth bass			1.2644
79	largescale sucker			1.0811
81	largescale sucker			1.2454
81	Largemouth bass			1.4286
		Station 40, San Joaquin River at Los Banos, Calif.		
79	Black bullhead			3.3333
79	Black bullhead			3.3871
79	Green sunfish			6.0748
81	Sacramento blackfish			5.3425
81	Sacramento blackfish			5.7407
		Station 41, Snake River at Hagerman, Idaho		
78	Largescale sucker			1.2431
78	Largescale sucker			1.4126

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
78	Rainbow trout			2.3630
80	Largescale sucker			1.3913
80	Largescale sucker			1.5447
80	Rainbow trout			1.9495
		Station 42, Snake River at Lewiston, Idaho- Clarkston, Wash.		
78	Largescale sucker			0.9325
78	Largescale sucker			1.3636
78	Smallmouth bass			1.4765
80	Largescale sucker			0.8861
80	Largescale sucker			0.9746
80	White crappie			1.0870
		Station 43, Salmon River at Riggins, Idaho		
78	Bridgelip sucker			1.5719
78	Bridgelip sucker			0.8494
78	Northern squawfish			1.1930
80	Bridgelip sucker			0.9016
80	Bridgelip sucker			0.8475
80	Northern squawfish			2.9897
		Station 44, Yakima River at Granger, Wash.		
78	Common carp			2.3026
78	Common carp			1.4047
80	Black crappie			1.6716
80	Largescale sucker			1.7742
80	Largescale sucker			1.6508
		Station 45, Willamette River at Oregon City, Oreg		
78	Northern squawfish			0.5078
78	Chiselmouth			0.6615
78	Chiselmouth			0.4082
80	Largescale sucker			0.5479
80	Largescale sucker			0.6907
80	Northern squawfish			1.4286
		Station 46, Columbia River at Cascade Locks, Wash. -Oreg.		
78	Largescale sucker			1.2684
78	Largescale sucker			1.3712
78	Northern squawfish			1.7818
80	Largescale sucker			0.9236
80	Largescale sucker			0.6765
80	Northern squawfish			0.7025
		Station 47, Klamath River at br k, Calif.		
79	Klamath largescale sucker			0.3409
79	Klamath largescale sucker			0.3774
79	Yellow perch			0.6693
81	Klamath largescale sucker			1.0121

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
81	Yellow perch			0.9016
		Station 48, Rogue River at G)Id, y Da Oreg.		
79	Black crappie			0.3836
79	Redside shine			0.4887
81	Black crappie			0.3158
81	Brown bullhead			0.7805
81	Brown bullhead			0.7692
		Station 49, (,hena River at rks, aska		
79	Burbot			2.3005
79	Longnose sucker			1.2903
81	Longnose sucker			1.7757
81	Longnose sucker			1.8519
81	Northern pike			1.8026
		Station 50, Kenai River at SDidatna, laska		
78	Rainbow trout			2.0391
78	Round whitefish			2.9538
78	Dolly Varden			1.6992
80	Rainbow trout			1.8910
80	Round whitefish			1.8954
80	Dolly Varden			1.6910
		Station 51, Kennebec Rive at iiic y, Maine		
78	White sucker			1.1060
78	White sucker			0.9692
78	Yellow perch			1.2549
80	White sucker			1.0046
80	White sucker			0.9459
80	Yellow perch			0.7011
		Station 52 Lake Champlain Burlic gton, Vt.		
78	Northern pike			0.7451
78	White sucker			0.8400
78	White sucker			0.8676
80	Northern pike			1.1163
		Station 53, Menimack River t Low@ll, Mass.		
78	Largemouth bass			0.8070
78	White sucker			1.0357
78	White sucker			1.0676
80	Smallmouth bass			0.7343
80	White sucker			0.8230
80	White sucker			1.2389
		Station 54, Raritan River at Highland Park, N.J.		
78	Largemouth bass			1.8060
78	White sucker			1.9454

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
78	White sucker			1.8301
80	White sucker			1.5126
80	White sucker			2.3348
80	Redfin pickerel			1.6450
		Station 55, James River at Richmond, Va.		
79	Redhorse			1.2877
79	Redhorse			1.4194
79	Smallnouth bass			1.8657
81	Redhorse			1.9243
81	Redhorse			1.0658
81	Smallnouth bass			1.0359
		Station 56, Pee Dee River at Johnsonville, S.C.		
80	Gizzard shad			1.1170
80	Gizzard shad			1.0000
80	Largemouth bass			1.5235
		Station 57, Altamaha River at Doctortown, Ga.		
78	Black crappie			1.2857
78	Carpsucker			1.2342
80	Largemouth bass			1.7094
80	Spotted sucker			2.0408
80	Spotted sucker			1.5574
		Station 59, Alabama Rive'r at Chrysler, Ala.		
79	Smallmouth buffalo			1.0035
79	Smallmouth buffalo			1.0175
79	Bowfin			1.2203
81	Largemouth bass			1.1538
81	Blue catfish			0.6716
81	Blue catfish			0.7326
		Station 60, Brazos River a@ Richmond, Tex.		
79	Longnose gar			1.2681
79	Smallmouth buffalo			1.0320
79	Smallmouth buffalo			1.3693
		Station 61, Colorado River at Wharton, Tex.		
79	Channel catfish			0.9662
79	Freshwater drum			1.7844
79	Freshwater drum			1.4943
		Station 63, Rio Grande at Elephant Butte, N. Mex.		
78	Common carp			2.1514
78	Common carp			1.9028
78	Largemouth bass			1.9310
80	Common carp			1.7597
80	Common carp			1.5830

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
80	Largemouth bass			1.5709
		Station 64, Rio Grande at Alamosa, Colo.		
78	White sucker			0.7442
78	White sucker			0.9231
80	Brown trout			1.2775
80	White sucker			0.6911
80	White sucker			0.8036
		Station 65, Pecos River at Red Bltiff Lake, Tex.		
78	Gizzard shad			4.2715
78	White bass			9.5016
80	Gizzard shad			3.8559
80	Gizzard shad			5.0673
80	White bass			6.0681
		Station 66, St. Lawrence River at Massena, N.Y.		
79	Smallmouth bass			1.1765
79	White sucker			1.0280
79	White sucker			1.4414
81	Northern pike			1.3592
		Station 67, Allegheny River at Natrona, Pa.		
78	Redhorse			2.0155
78	Redhorse			1.4232
78	Smallmouth bass			2.2794
79	Largemouth bass			1.3693
79	Redhorse			1.9005
79	Redhorse			1.5789
80	Redhorse			2.7511
80	Redhorse			2.8139
80	Smallmouth bass			2.2656
		Station 68, Wabash River at New Harmony, -Crossville, III		
78	Common carp			2.0505
78	Common carp			2.2302
78	Largemouth bass			2.3413
80	Common carp			1.3043
80	Common carp			1.4873
80	Largemouth bass			1.5175
		Station 69, Ohio River at Cincinnati, Ohio		
78	Common carp			2.5890
78	Common carp			4.0333
78	Sauger			1.5031
80	Common carp			2.1071
80	Common carp			2.6070
80	Sauger			1.5113

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
		Station 70, Ohio River at Metropolis Ill.-Paducah, Ky.		
78	Common carp			2.2222
78	Common carp			1.4067
78	Largemouth bass			1.3514
80	Common carp			2.2118
80	Common carp			2.0599
80	Largemouth bass			1.7770
		Station 71, Tennessee iver at Savannah, Tenn.		
78	White bass			1.4610
79	Common carp			2.3790
79	Carpsucker			1.3333
80	Common carp			2.1973
80	Common carp			1.8103
80	Largemouth bass			1.9178
		Station 72, Wisconsin River at Woodman, Wis.		
80	Common carp			1.4107
80	Common carp			1.1688
80	Largemouth bass			1.1538
		Station 73, Des Moines River at Keosauqua, Iowa		
78	Common carp			3.5986
78	Common carp			4.0956
78	Sauger			2.4706
80	Common carp			3.8462
80	Common carp			2.3221
80	Channel catfish			1.7883
		Station 74, Mississippi River at Little Falls, Minn.		
78	White sucker			1.4340
78	Yellow perch			1.5825
80	rock bass			1.6207
80	Yellow bullhead			2.0155
80	Yellow bullhead			1.9149
		Station 75, Mississippi River at Cape Girardeau, Mo.-Ill		
78	Common carp			2.3511
78	Common carp			2.1019
78	White crappie			1.2360
80	Common carp			1.4449
80	Common carp			1.8077
80	White bass			2.8839
		Station 76, Mississippi River		
79	Bluegill			1.8000
79	Smallmouth buffalo			0.6818
79	Smallmouth buffalo			0.6140
81	Smallmouth buffalo			1.0979
81	Smallmouth buffalo			0.9884

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
81	White crappie			0.9343
		Station 78, Verdigris River at Oologah Okla.		
79	Common carp			1.0268
79	Common carp			1.4218
79	White bass			1.9907
81	Bluegill			1.7063
81	Common carp			2.1223
81	Common carp			2.1456
		Station 79, Canadian River at Eufaula, Okl.		
79	Common carp			2.4324
79	Common carp			1.4113
79	Largemouth bass			1.6318
81	Common carp			1.8382
81	Common carp			1.9403
81	Largemouth bass			2.3789
		Station 80, Yazoo River at Redwood, Miss		
79	Common carp			1.1620
79	Common carp			2.7356
81	Smallmouth buffalo			1.1875
81	Smallmouth buffalo			1.3559
81	White crappie			1.4444
		Station 81, Red River at Alexandria.		
79	Smallmouth buffalo			0.8929
79	Smallmouth buffalo			0.9667
79	White bass			1.3043
81	Freshwater drum			1.4103
81	Freshwater drum			1.2500
81	Spotted gar			0.7353
		Station 82, Red River at Lake Texoma, Okla.-Tex.		
79	Black crappie			1.2955
79	River carpsucker			1.3366
79	River carpsucker			1.7617
81	Common carp			1.8280
81	Common carp			2.4627
81	Largemouth bass			2.3043
		Station 83, Missoun River at Hermann, Mo.		
79	River carpsucker			0.9121
79	River carpsucker			1.1350
79	Smallmouth buffalo			1.4706
		Station 84, Bighorn River at Hardin, Mont.		
79	Common carp			5.6522
79	Goldeye			9.4118
79	White sucker			6.9466

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
81	Brown trout			5.0896
81	Longnose sucker			3.0717
		Station 85, Yellowstone River at Sidney,Mont.		
79	Common carp			1.4773
79	Common carp			1.9277
79	Sauger			1.7257
81	Redhorse			1.8919
81	Redhorse			2.4832
81	Sauger			1.6832
		Station 86, James River at Olivet, S. Dak	vet, S. ak.	
79	Carpsucker			1.7188
79	Carpsucker			1.9184
79	Goldeye			1.7302
81	Carpsucker			1.0154
81	Carpsucker			1.2805
81	Goldeye			2.5185
		Station 87, North Platte River at Lak McConaughy, Nebr.		
79	Common carp			3.7288
79	Common carp			4.8918
79	Walleye			1.4907
81	Common carp			3.0488
81	Common carp			2.6601
81	Walleye			2.0077
79	Black crappie			2.7881
79	Common carp			4.3902
79	Common carp			4.3590
		Station 88, South Platte River at Brule, Nebr.		
81	White sucker			4.6538
81	Orangespotted sunfish			8.6786
		Station 89, Platte River at Lduisville Nebr.		
79	Carpsucker			2.2549
79	Carpsucker			1.6514
79	Goldeye			3.2335
81	Carpsucker			2.5207
81	Carpsucker			2.8270
81	Goldeye			4.0972
		Station 90, Kansas River at	onner prings, Kans.	
79	Common carp			2.4615
79	River carpsucker			1.0676
81	Common carp			1.5858
81	Channel catfish			2.1635
81	River carpsucker			0.9859
		Station 91 Colorado River at Lake Havasu, Ariz.-Calif.		

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
78	Common carp			7.6490
78	Largemouth bass			5.1449
80	Common carp			3.6494
80	Common carp			5.6944
80	Largemouth bass			2.7666
		Station 92, Colorado River at Lake Mead Ariz.-Nev.		
79	Channel catfish			4.6441
79	Channel catfish			3.0169
79	Striped bass			0.8182
81	Common carp			3.3735
81	Channel catfish			3.2707
81	Striped bass			3.7109
		Station 93, Colorado v" t Lake 11, Ariz.		
78	Common carp			9.4218
78	Largemouth bass			9.8990
80	Common carp			4.3922
80	Common carp			3.9574
80	Largemouth bass			2.3759
		Station 94, Gfla River at Sa "IIDI servoir, Ariz.		
78	Common carp			1.9231
78	Common carp			1.5918
78	Largemouth bass			1.7466
80	Common carp			1.9588
80	Common carp			1.4079
80	Largemouth bass			1.1524
		Station 96, Snake River at I H @b, 'D.@'oWash		
78	Largescale sucker			1.0000
78	Largescale sucker			1.2625
78	Northern squawfish			0.8970
80	Largescale sucker			0.8765
80	Largescale sucker			0.8456
80	Northern squawfish			1.7316
		Station 97, Columbia River Pa,,O, sh.		
78	Yellow perch			3.8667
78	Chiselmouth			1.6242
78	Chiselmouth			1.2375
80	Common carp			4.0244
80	Common carp			2.6923
80	Yellow perch			3.5662
		Station 98, Columbia i,er G @,n 'C,uloee, Wash.		
78	Largescale sucker			0.8300
78	Largescale sucker			0.9266
78	Yellow perch			1.1847
80	Largescale sucker			0.7692

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
80	Largescale sucker			0.7519
80	Walleye			0.8108
		tation 99, Waikele Stea, Waipah Hawaii		
79	Cuban limia			4.0755
		Station 100, Manoa Stream at Honol Hawai		
79	Cuban limia			3.5577
79	Mazambique tilapia			1.6502
		Station 101, Androscoggin iver at wiston, Main		
78	White sucker			0.9426
78	White sucker			0.7059
78	Yellow perch			0.7042
80	White sucker			0.6299
80	White sucker			0.5691
80	Yellow perch			0.8961
		Station 102, Laice Superior at@Keewe@naw Point, Mich.		
79	Bloater			1.2798
79	Bloater			0.9385
79	Lake trout			0.7339
81	Bloater			1.3354
81	Bloater			1.6961
81	Lake trout			1.4286
		Station 103, Lake Superior at Whitefish Point, Mich.		
79	Lake trout			0.7427
79	Lake whitefish			1.1379
79	Lake whitefish			1.8051
81	Lake trout			1.0617
81	Lake whitefish			1.4947
		104, Lake Michigan at Beavei,lsland,Mich.		
79	Bloater			0.8537
79	Bloater			0.4963
79	Lake trout			1.3483
		Station 105. Lake Michigan at Saugat@ck, Mich		
79	Bloater			0.4948
79	Bloater			0.6651
79	Lake trout			0.8310
81	Bloater			0.8696
81	Bloater			0.8939
81	Lake trout			1.1480
		Station 106, Lake Huron at Alpena Mich.		
79	White sucker			1.6596
79	White sucker			1.3793
79	Yellow perch			2.5556

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
81	Lake trout			1.2808
81	White sucker			2.3293
81	White sucker			2.2403
		Station 107, Lake St. Clair at Mount Clements. Mich.		
79	Common carp			1.2030
79	Common carp			1.6418
79	Walleye			1.7731
81	Common carp			1.8182
81	Common carp			2.0861
81	Walleye			1.9101
		Station 108, Lake Erie at Port Clinton. Ohio		
79	Common carp			1.4696
79	Common carp			1.4483
79	Walleye			0.6329
81	Common carp			2.1951
81	Common carp			1.6992
81	Walleye			1.0811
		Station 109, Lake Ontario at Roosevelt Beach, N.Y.		
79	Brown trout			1.1184
79	Rock bass			1.7626
79	Rock bass			1.4179
81	Lake trout			1.1310
81	Rock bass			1.6988
81	Rock bass			2.0949
		Station I I 1. Mississippi River at Lake City, Minn.-Pepin. Wis.		
78	White sucker			1.6867
78	White sucker			1.5203
80	Walleye			1.2162
80	White sucker			1.4231
80	White sucker			1.1020
		Station 112, Mississippi River at Dubuque, Iowa		
78	Common carp	12.9	1.1	1.8868
78	Common carp	14.4	1.5	1.4462
78	Largemouth bass	11.2	1.1	1.5901
80	Black crappic	10.2	0.7	1.6000
80	Common carp	17	2.6	2.2018
80	Common carp	17.8	3	1.4789
		Station 113, San Antonio River at McFadden, Tex.		
79	Longnose gar	28.5	2.3	0.7767
		Station 114, Bear River at Brigham City, Utah		
78	Common carp	11.7	0.8	1.5625
78	Common carp	10.2	0.6	0.9957
78	Channel catfish	17.9	2.3	1.3109

Year	Species	Mean total length, cm	Mean total weight, Kg	Se, ug/g dry wt.
80	Common carp	17	2.3	1.6667
80	Common carp	12	0.9	1.4583
80	Channel catfish	22.7	4.7	1.2551
		Station 115, Colorado River at Yuma, Ariz.- Winterhaven, Calif.		
78	Common carp	16	2.7	6.4815
78	Striped mullet	20.3	3.4	3.5958
		Station 116, Souris River at Upham, N. Dak.		
78	White sucker	16.5	2.3	1.0526
80	Northem pike	19.6	1.9	0.9709
80	White sucker	15.3	1.7	1.1161
80	White sucker	13.8	1.3	1.0331
		Station 117, Flathead River at Creston, Mont.		
78	Northem squawfish	19.1	2.7	0.7925
80	Largescale sucker	15.4	1.3	0.7589
80	Largescale sucker	15.5	1.3	0.9237
80	Northem squawfish	15.8	1.3	0.9091
			Average	1.8836
			Std	1.4373
			max	10.5204
			min	0.3158
			count	591

Table J-2. Selenium concentration (µg/g dw whole-body) in fish and invertebrate samples collected at sites of the USGS National Water Quality Assessment (NAWQA) Program, 1992-1997.

[Se]	Scientific Name	Common Name	Place Name
0.30	Odonata	-	GOOSE LAKE WMA
0.30	Hydropsyche	-	SF PALOUSE R. AT ARMSTRONG RD NR PULLMAN, WA
0.30	Hydropsyche	-	PALOUSE R. AT ENDICOTT-ST. JOHN RD NR COLFAX, WA
0.40	Odonata	-	JOHNSON WPA
0.40	Odonata	-	WOOD DUCK WMA
0.40	Corbicula	-	SALUDA RIVER NEAR COLUMBIA, SC
0.50	Pacifastacus leniusculus	signal crayfish	TRUCKEE R AT FARAD, CA
0.50	Odonata	-	DEPARTMENT OF ROADS - ONEILL
0.50	Odonata	-	TODD VALLEY - MEDUNA SITE
0.50	Micropterus salmoides	largemouth bass	TRINITY RV BL DALLAS, TX
0.60	Hydropsyche	-	ROCK CREEK BLW US HWY 30/93 AT TWIN FALLS ID
0.60	Pacifastacus leniusculus	signal crayfish	TRUCKEE R AT CLARK, NV
0.60	Hydropsyche	-	WOLF RIVER AT TURTLE LAKE ROAD AT POST LAKE, WI
0.60	Potamogeton pectinatus	sago pondweed	NORTH BRANCH MILWAUKEE RIVER NR RANDOM LAKE, WI
0.69	Tilapia melanotheron	blackchin tilapia	ALA WAI CANAL AT HONOLULU, HI
0.70	Hydropsyche	-	CRAB CREEK AT MORGAN LAKE ROAD NEAR OTHELLO, WA
0.70	Cottus sp.	freshwater sculpins	MILLER CREEK NEAR DES MOINES, WA
0.70	Cheumatopsyche	-	WOLF RIVER NEAR POST LAKE, WI
0.70	Hydropsyche	-	WOLF RIVER NEAR POST LAKE, WI
0.80	Hydropsyche	-	PESHEKEE RIVER NEAR MARTINS LANDING, MI
0.90	Catostomus clarki	desert sucker	PINTO CREEK NEAR MIAMI, AZ.
0.90	Odonata	-	SABATKA SALINE WETLAND
0.90	Corbicula	-	SYCAMORE CK AT SYCAMORE PK, FT WORTH, TX
0.90	Hydropsychidae	net-spinning caddisflies	WOLF RIVER AT HIGHWAY M NEAR LANGLADE, WI
1.00	Hydropsyche	-	SNAKE RIVER AT KING HILL ID
1.00	Cottus sp.	freshwater sculpins	BIG SOOS CREEK ABOVE HATCHERY NEAR AUBURN, WA
1.00	Orconectes	-	EAST RIVER AT MIDWAY ROAD NEAR DE PERE, WI
1.00	Hydropsyche	-	PENSAUKEE RIVER NEAR KRAKOW, WI
1.08	Poecilia sphenops	black molly	KANEOHE STR BLW KAMEHAMEHA HWY, OAHU, HI
1.10	Acroneuria	-	WEST BRANCH WHITEFISH RIVER NEAR DIFFIN, MI
1.10	Pacifastacus leniusculus	signal crayfish	EAST FORK CARSON RIVER NEAR GARDNERVILLE, NV
1.10	Pacifastacus leniusculus	signal crayfish	EAST FORK CARSON RIVER NEAR DRESSLERVILLE, NV
1.10	Cottus sp.	freshwater sculpins	SANDY RIVER NEAR TROUTDALE, OR
1.10	Cottus sp.	freshwater sculpins	GALES CREEK NEAR GLENWOOD, OR
1.10	Cottus sp.	freshwater sculpins	GALES CREEK NEAR GLENWOOD, OR
1.10	Hydropsyche	-	PALOUSE RIVER AT HOOPER, WA
1.10	Hydropsyche	-	SNAKE RIVER AB JACKSON LAKE AT FLAGG RANCH WY
1.10	Hydropsyche	-	SNAKE RIVER AB JACKSON LAKE AT FLAGG RANCH WY
1.15	Cheumatopsyche	-	DUCK CREEK AT SEMINARY ROAD NEAR ONEIDA, WI
1.20	Cyprinella lutrensis	red shiner	GRANITE CREEK AT PRESCOTT, AZ.
1.20	Orconectes causeyi	-	GRANITE CREEK AT PRESCOTT, AZ.
1.20	Catostomus occidentalis	Sacramento sucker	COTTONWOOD C NR COTTONWOOD CA
1.20	Xiphophorus helleri	green swordtail	WAIHEE STR NR KAHALUU, OAHU, HI
1.20	Hydropsyche	-	HENRYS FORK NR REXBURG ID
1.20	Hydropsyche	-	ROCK CREEK AB HWY 30/93 XING AT TWIN FALLS ID
1.20	Cottus sp.	freshwater sculpins	TUALATIN RIVER AT WEST LINN, OR

[Se]	Scientific Name	Common Name	Place Name
1.20	Cottus sp.	freshwater sculpins	DENNIS C BL BLACK BUTTE MINE, NR COTTAGE GROVE LK
1.20	Cottus sp.	freshwater sculpins	WEST BRANCH KELSEY CREEK AT BELLEVUE, WA
1.20	Cottus sp.	freshwater sculpins	BERTRAND CREEK NEAR LYNDEN, WA
1.20	Cheumatopsyche	-	TOMORROW RIVER NEAR NELSONVILLE, WI
1.20	Hydropsyche	-	SNAKE RIVER AB JACKSON LAKE AT FLAGG RANCH WY
1.30	Pacifastacus leniusculus	signal crayfish	TRUCKEE R AT FARAD, CA
1.30	Cottidae	sculpins	JOHNSON CREEK AT MILWAUKIE, OR
1.30	Micropterus salmoides	largemouth bass	WHITE ROCK LK IN DALLAS, TX
1.30	Cottus sp.	freshwater sculpins	DUWAMISH RIVER AT GOLF COURSE AT TUKWILA, WA
1.30	Hydropsyche	-	DUCK CREEK AT SEMINARY ROAD NEAR ONEIDA, WI
1.39	Cottus cognatus	slimy sculpin	NINILCHIK R AT NINILCHIK AK
1.40	Hydropsyche	-	PORTNEUF RIVER AT POCATELLO ID
1.40	Odonata	-	TRUST - WILD ROSE SLOUGH
1.40	Pacifastacus leniusculus	signal crayfish	TRUCKEE R AT HWY 447 AT NIXON, NV
1.40	Cottus sp.	freshwater sculpins	MARYS RIVER AT CORVALLIS, OR
1.40	Cottus sp.	freshwater sculpins	FIR CREEK NEAR BRIGHTWOOD, OR
1.40	Cottus sp.	freshwater sculpins	FANNO CREEK AT DURHAM, OR
1.40	Cottus sp.	freshwater sculpins	FANNO CREEK AT DURHAM, OR
1.40	Cheumatopsyche	-	DUCK CREEK AT SEMINARY ROAD NEAR ONEIDA, WI
1.40	Hydropsyche	-	NORTH BRANCH MILWAUKEE RIVER NR RANDOM LAKE, WI
1.50	Catostomus clarki	desert sucker	SAN PEDRO RIVER AT CHARLESTON, AZ.
1.50	Hydropsyche	-	ROCK CREEK AB DAYDREAM RANCH NR TWIN FALLS ID
1.50	Brachycentrus	-	ROCK CREEK AB HWY 30/93 XING AT TWIN FALLS ID
1.50	Odonata	-	TRUST - MORMON ISLAND CRANE MEADOW, EAST SLOUGH
1.50	Pacifastacus leniusculus	signal crayfish	CARSON RIVER AT DEER RUN ROAD NEAR CARSON CITY, NV
1.50	Pacifastacus leniusculus	signal crayfish	CARSON RIVER NEAR FORT CHURCHILL, NV
1.50	Cottidae	sculpins	MUDDY CREEK NEAR PEORIA, OR
1.50	Hydropsyche	-	PALOUSE RIVER NEAR COLFAX, WA
1.50	Cottus sp.	freshwater sculpins	THORNTON CREEK NEAR SEATTLE, WA
1.50	Hydropsyche	-	TOMORROW RIVER NEAR NELSONVILLE, WI
1.50	Hydropsyche	-	SALT RIVER AB RESERVOIR NR ETNA WY
1.50	Hydropsyche	-	SALT RIVER AB RESERVOIR NR ETNA WY
1.53	Cyprinus carpio	common carp	BEAR RIVER NEAR CORINNE, UT
1.60	Catostomus clarki	desert sucker	GILA RIVER AT KELVIN, AZ.
1.60	Hydropsyche	-	BARK RIVER NEAR BARK RIVER, MI
1.60	Cottidae	sculpins	FANNO CREEK AT DURHAM, OR
1.60	Elliptio	-	CEDAR CREEK BELOW MYERS CREEK NR HOPKINS, SC
1.60	Hydropsyche	-	PINE CREEK AT PINE CITY ROAD AT PINE CITY, WA
1.62	Corbicula	-	TRUCKEE R AT CLARK, NV
1.70	Hydropsyche	-	PORTNEUF RIVER AT TOPAZ ID
1.70	Hydropsyche	-	SNAKE RIVER AT KING HILL ID
1.70	Corbicula	-	TRENT RIVER NEAR TRENTON, NC
1.70	Elliptio	-	MCTIER CREEK (RD 209) NEAR MONETTA, SC
1.70	Corbicula	-	GILLS CREEK NEAR HOPKINS,SC
1.70	Hydropsyche	-	SHEBOYGAN RIVER AT DOTYVILLE, WI
1.79	Cottidae	sculpins	LITTLE ABIQUA CREEK NEAR SCOTTS MILLS, OR
1.80	Hydropsychidae	net-spinning caddisflies	TRUCKEE R AT FARAD, CA
1.80	Hydropsyche	-	TETON RIVER NR ST ANTHONY ID
1.80	Hydropsyche	-	SNAKE R NR MINIDOKA ID (AT HOWELLS FERRY)
1.90	Corbicula manilensis	asian clam	CHATTAHOOCHEE R AT SR 253 NEAR CHATTAHOOCHEE, FL
1.90	Hydropsyche	-	PALOUSE RIVER AT LAIRD PARK NR HARVARD, ID
1.90	Brachycentrus	-	ROCK CREEK AB HWY 30/93 XING AT TWIN FALLS ID
1.90	Hydropsyche	-	DUCK CREEK AT SEMINARY ROAD NEAR ONEIDA, WI
1.95	Poecilia sphenops	black molly	NUUANU STR ABV WAOLANI ST. AT HONOLULU, OAHU, HI
2.00	Hydropsyche	-	SPRING CREEK AT SHEEPSKIN RD NR FORT HALL ID
2.00	Acroneuria	-	PESHEKEE RIVER NEAR MARTINS LANDING, MI
2.00	Cheumatopsyche	-	DUCK CREEK AT SEMINARY ROAD NEAR ONEIDA, WI

[Se]	Scientific Name	Common Name	Place Name
2.03	Ameiurus natalis	yellow bullhead	SANTA ANA R A HAMNER RD NR NORCO CA
2.05	Richardsonius balteatus	redside shiner	BEAR RIVER ABOVE RESERVOIR, NEAR WOODRUFF, UT
2.10	Catostomus clarki	desert sucker	VERDE RIVER ABV W. CLEAR CREEK, NR CAMP VERDE, AZ
2.10	Hydropsyche	-	SNAKE R NR MINIDOKA ID (AT HOWELLS FERRY)
2.10	Hydropsyche	-	ROCK CK AT USFS FOOTBRIDGE, NR ROCK CREEK
2.10	Hydropsyche	-	ROCK CREEK AB HWY 30/93 XING AT TWIN FALLS ID
2.10	Pacifastacus leniusculus	signal crayfish	TRUCKEE R AT LOCKWOOD, NV
2.10	Hydropsyche	-	TUALATIN RIVER AT WEST LINN, OR
2.10	Hydropsyche	-	ESQUATZEL COULEE AT MESA, WA
2.10	Cottus sp.	freshwater sculpins	FISHTRAP CREEK AT FLYNN ROAD AT LYNDEN, WA
2.17	Corbicula fluminea	Asian clam	CONTENTNEA CREEK AT HOOKERTON, NC
2.18	Corbicula	-	TENNESSEE RIVER AT CHATTANOOGA, TN
2.20	Brachycentrus	-	BITCH CREEK NR LAMONT ID
2.20	Hydropsyche	-	SOUTH BRANCH PAINT RIVER NEAR ELMWOOD, MI
2.20	Cottus sp.	freshwater sculpins	LITTLE ABIQUA CREEK NEAR SCOTTS MILLS, OR
2.20	Ceratopsyche	-	EAST RIVER @ CTH PP IN BROWN COUNTY NR DE PERE, WI
2.20	Brachycentrus	-	SALT RIVERV NR FISH CK ABOVE SMOOT
2.30	Agosia chrysogaster	longfin dace	SANTA CRUZ RIVER AT TUBAC, AZ.
2.30	Catostomus clarki	desert sucker	WEST CLEAR CREEK NEAR CAMP VERDE, AZ.
2.30	Hydropsyche	-	PORTNEUF RIVER AT TOPAZ ID
2.37	Ameiurus natalis	yellow bullhead	SANTA ANA R A MWD CROSSING CA
2.40	Xiphophorus helleri	green swordtail	WAIKELE STR AT WAIPAHU, OAHU, HI
2.40	Hydropsyche	-	MALAD RIVER NR GOODING ID
2.40	Corbicula	-	CRABTREE CREEK AT US 1 AT RALEIGH, NC
2.40	Corbicula	-	BLACKWATER RIVER NEAR FRANKLIN, VA
2.40	Cottus sp.	freshwater sculpins	ROCK CREEK AT CEDAR FALLS ROAD NEAR LANDSBURG, WA
2.40	Cottus sp.	freshwater sculpins	JUANITA CREEK AT JUANITA, WA
2.50	Agosia chrysogaster	longfin dace	SALT RIVER NEAR ROOSEVELT, AZ.
2.50	Hydropsyche	-	SNAKE RIVER NR BLACKFOOT ID
2.50	Hydropsyche	-	SNAKE RIVER NR BLACKFOOT ID
2.50	Anaspidacea	-	EAST FORK CARSON RIVER NEAR GARDNERVILLE, NV
2.50	Elliptio	-	COOSAWHATCHIE RIVER NR EARLY BRANCH, SC
2.50	Corbicula	-	TAYLOR FLAT CREEK ABV BIRCH RD NR PASCO, WA
2.52	Xiphophorus helleri	green swordtail	MANOA STR AT KANEWAI FIELD, HONOLULU, OAHU, HI
2.60	Cottus sp.	freshwater sculpins	PALMER C AT DAYTON, OR
2.60	Elliptio	-	SHAWS CREEK NR TRENTON, SC ON CNTY RD 149
2.60	Corbicula	-	PIGEON RIVER AT NEWPORT, TN
2.60	Corbicula	-	RUSH CK AT WOODLAND PARK BLVD, ARLINGTON, TX
2.60	Cottus sp.	freshwater sculpins	NEWAUKUM CREEK NEAR BLACK DIAMOND, WA
2.64	Cyprinus carpio	common carp	SAN JACINTO R NR ELSINORE CA
2.70	Corbicula	-	EMORY RIVER AT OAKDALE, TN
2.70	Corbicula	-	GUADALUPE RV AT GONZALES, TX
2.70	Corbicula	-	NORTH MEHERRIN RIVER NEAR LUNENBURG, VA
2.70	Cottus sp.	freshwater sculpins	GREEN RIVER ABOVE TWIN CAMP CREEK NEAR LESTER, WA
2.70	Cottus sp.	freshwater sculpins	LEACH CREEK NEAR STEILACOOM, WA
2.70	Cottus sp.	freshwater sculpins	NORTH CREEK BELOW PENNY CREEK NEAR BOTHELL, WA
2.77	Gambusia affinis	western mosquitofish	MANOA STR AT KANEWAI FIELD, HONOLULU, OAHU, HI
2.79	Cottus sp.	freshwater sculpins	WEBER RIVER NEAR COALVILLE, UT
2.80	Corbicula manilensis	asian clam	MUCKALEE CREEK AT GA 195, NEAR LEESBURG, GA
2.80	Corbicula	-	TAR RIVER NEAR TAR RIVER, NC
2.80	Decapoda	crabs	PLATTE RIVER AT BRADY, NE (TOTFLO)
2.80	Corbicula	-	COPPER CREEK NEAR GATE CITY, VA
2.80	Brachycentrus	-	SECOND SOUTH BRANCH OCONTO RIVER NR MOUNTAIN, WI
2.80	Hydropsyche	-	TOMORROW RIVER NEAR NELSONVILLE, WI
2.80	Hydropsyche	-	NORTH BRANCH MILWAUKEE RIVER NR RANDOM LAKE, WI
2.80	Hydropsyche	-	SNAKE RIVER AB JACKSON LAKE AT FLAGG RANCH WY
2.81	Salvelinus fontinalis	brook trout	WOOD RIVER ABOVE MIDDLE FORK NEAR MEETEETSE, WY

[Se]	Scientific Name	Common Name	Place Name
2.85	Cottus cognatus	slimy sculpin	KENAI R AT JIMS LANDING NR COOPER LANDING AK
2.88	Salvelinus fontinalis	brook trout	CROW CREEK AT MOUTH, AT PAHASKA, WY
2.90	Corbicula manilensis	asian clam	APALACHICOLA RIVER AT CHATTAHOOCHEE FLA
2.90	Corbicula manilensis	asian clam	FLINT RIVER AT NEWTON, GA
2.90	Corbicula manilensis	asian clam	ICHAWAYNOCHAWAY CREEK BELOW NEWTON, GA
2.90	Corbicula manilensis	asian clam	PEACHTREE CREEK AT ATLANTA, GA
2.90	Corbicula manilensis	asian clam	FLINT RIVER AT LAKE BLACKSHEAR NEAR WARWICK, GA.
2.90	Corbicula manilensis	asian clam	CHATTAHOOCHEE RIVER AT COLUMBUS, GA
2.90	Hydropsyche	-	SNAKE RIVER AT KING HILL ID
2.90	Corbicula	-	TRUCKEE R AT CLARK, NV
2.96	Cottus cognatus	slimy sculpin	CHESTER C AT ARCTIC BOULEVARD AT ANCHORAGE AK
2.96	Gambusia affinis	western mosquitofish	KANEOHE STR BLW KAMEHAMEHA HWY, OAHU, HI
3.00	Cottus cognatus	slimy sculpin	KENAI R BL RUSSIAN R NR COOPER LANDING AK
3.00	Hydropsyche	-	PORTNEUF RIVER AT TOPAZ ID
3.00	Brachycentrus	-	TETON RIVER AB SOUTH LEIGH CREEK NR DRIGGS ID
3.00	Corbicula fluminea	Asian clam	BIG BLUE RIVER AT SHELBYVILLE, IN
3.00	Corbicula	-	TAR RIVER AT TARBORO, NC
3.00	Corbicula	-	NORTH FLAT RIVER AT TIMBERLAKE, NC
3.00	Corbicula	-	GILLS CREEK AT COLUMBIA, SC
3.10	Perlidae	common stoneflies	BIG WOOD RIVER BLW BOULDER CK NR KETCHUM
3.10	Corbicula	-	NEUSE RIVER NEAR COX MILL, NC
3.10	Corbicula	-	SOUTH FORK CATAWBA RIVER AT MCADENVILLE, NC
3.10	Corbicula	-	INDIAN CREEK NEAR LABORATORY, NC
3.18	Corbicula manilensis	asian clam	SNAKE CREEK NEAR WHITESBURG, GA
3.20	Corbicula manilensis	asian clam	SPRING CREEK NEAR IRON CITY, GA.
3.20	Corbicula	-	ROANOKE RIVER AT ROANOKE RAPIDS, NC
3.20	Corbicula	-	NOTTOWAY RIVER NEAR SEBRELL, VA
3.30	Corbicula manilensis	asian clam	BULL CREEK AT US 27 AT COLUMBUS, GEORGIA
3.30	Corbicula	-	CONOCOCHEAGUE CREEK AT FAIRVIEW, MD
3.30	Decapoda	crabs	WOOD RIVER NEAR GRAND ISLAND NEBR
3.30	Corbicula	-	NOLICHUCKY RIVER NEAR LOWLAND
3.40	Corbicula fluminea	Asian clam	SUGAR CREEK AT CO RD 400 S AT NEW PALESTINE, IN
3.40	Ceratopsyche	-	WEST BRANCH WHITEFISH RIVER NEAR DIFFIN, MI
3.40	Cheumatopsyche	-	JOHNSON CREEK AT MILWAUKIE, OR
3.40	Corbicula	-	POWELL RIVER NEAR ARTHUR, TN
3.40	Hydropsyche	-	PARADISE CREEK AT PULLMAN, WA
3.40	Hydropsyche	-	SALT RIVER AB RESERVOIR NR ETNA WY
3.50	Corbicula manilensis	asian clam	FLINT R @ 10-MI STILL LANDING NR CHATTAHOOCHEE, FL
3.50	Corbicula manilensis	asian clam	PEACHTREE CREEK AT ATLANTA, GA
3.50	Corbicula	-	NEUSE RIVER AT KINSTON, NC
3.50	Corbicula fluminea	Asian clam	NEUSE RIVER AT KINSTON, NC
3.50	Corbicula	-	SANTEE R AT TREZESVANTS LANDING NR FT MOTTE, SC
3.50	Corbicula	-	NOLICHUCKY RIVER AT EMBREEVILLE, TN
3.50	Corbicula	-	SAN MARCOS RV ABV BLANCO RV BL SAN MARCOS, TX
3.60	Cyprinella lutrensis	red shiner	SALT RIVER NEAR ROOSEVELT, AZ.
3.60	Agosia chrysogaster	longfin dace	AGUA FRIA RIVER NEAR ROCK SPRINGS, AZ.
3.60	Carpiodes carpio	river carpsucker	BUCKEYE CANAL NR HASSAYAMPA
3.60	Corbicula	-	48TH STREET DRAIN NR INTERSTATE 10
3.60	Corbicula manilensis	asian clam	APALACHICOLA RIVER NR BLOUNTSTOWN,FLORIDA
3.60	Corbicula manilensis	asian clam	CHATTAHOOCHEE R AT SR 369 NR FLOWERY BRANCH, GA.
3.60	Corbicula	-	SWIFT CREEK AT HILLIARDSTON, NC
3.60	Corbicula	-	CHICOD CR AT SR1760 NEAR SIMPSON, NC
3.60	Corbicula	-	LITTLE RIVER NEAR MARYVILLE, TN
3.60	Hydropsyche	-	CRAB CREEK AT ROCKY FORD ROAD NEAR RITZVILLE, WA
3.60	Cottus sp.	freshwater sculpins	ROCK CREEK NEAR MAPLE VALLEY, WA
3.60	Cottus sp.	freshwater sculpins	NOOKSACK RIVER AT BRENNAN, WA

[Se]	Scientific Name	Common Name	Place Name
3.66	Corbicula	-	CONGAREE RIVER AT COLUMBIA, SC
3.70	Corbicula manilensis	asian clam	KINCHAFOONEE CREEK NEAR DAWSON, GA
3.70	Hydropsyche	-	BLACKFOOT RIVER AB RESERVOIR NR HENRY ID
3.70	Corbicula fluminea	Asian clam	WHITE RIVER AT RAYMOND STREET AT INDIANAPOLIS, IN
3.70	Corbicula manilensis	asian clam	CURRENT RIVER AT VAN BUREN, MO
3.77	Corbicula	-	HOLSTON RIVER AT SURGOINSVILLE, TN
3.80	Corbicula manilensis	asian clam	APALACHICOLA RIVER NR SUMATRA,FLA.
3.80	Corbicula	-	SWIFT CREEK NEAR APEX, NC
3.80	Cottus sp.	freshwater sculpins	LUCKIAMUTE RIVER NEAR SUVER, OR
3.80	Corbicula	-	CATOCTIN CREEK AT TAYLORSTOWN, VA
3.85	Corbicula manilensis	asian clam	SOPE CREEK NEAR MARIETTA, GA
3.90	Corbicula manilensis	asian clam	NICKAJACK CR AT COOPER LAKE DR NR MABLETON, GA.
3.90	Corbicula manilensis	asian clam	CHATTAHOOCHEE RIVER NEAR COLUMBIA, ALA.
4.00	Elliptio	-	GEORGES CREEK NEAR OLAR, SC ON SC 64
4.00	Corbicula	-	WATEREE RIVER NR. CAMDEN, SC
4.01	Cyprinus carpio	common carp	LEON CK AT IH 35 AT SAN ANTONIO, TX
4.10	Corbicula manilensis	asian clam	WILLEO CREEK AT ST RT 120 NEAR ROSWELL, GA.
4.10	Hydropsyche	-	MALAD RIVER NR GOODING ID
4.10	Corbicula	-	CARSON RIVER AT TARZYN ROAD NR FALLON, NV
4.10	Corbicula	-	FRENCH BROAD RIVER NEAR NEWPORT, TN
4.10	Corbicula	-	MIDDLE FORK HOLSTON RIVER AT SEVEN MILE FORD, VA
4.16	Corbicula	-	OBED RIVER NEAR LANCING, TN
4.20	Corbicula manilensis	asian clam	SEWELL MILL CR AT SEWELL MILL RD NEAR MARIETTA
4.20	Arctopsyche	-	BIG LOST RIVER AT HOWELL RANCH NR CHILLY ID
4.20	Corbicula	-	CONGAREE RIVER AT U.S. HWY 601 NR. FORT MOTTE, SC
4.20	Corbicula	-	GUADALUPE RV NR SPRING BRANCH, TX
4.30	Agosia chrysogaster	longfin dace	GILA RIVER AT KELVIN, AZ.
4.30	Corbicula manilensis	asian clam	SOPE CREEK NEAR MARIETTA, GA
4.30	Corbicula manilensis	asian clam	AYCOCKS CREEK NEAR BOYKIN, GA.
4.30	Elliptio	-	COW CASTLE CREEK NEAR BOWMAN, SC
4.30	Corbicula	-	NORTH FORK HOLSTON RIVER NEAR CLOUD FORD, TN
4.40	Cottus cognatus	slimy sculpin	TALKEETNA R NR TALKEETNA AK
4.40	Corbicula manilensis	asian clam	COOLEEWAHEE CREEK NEAR NEWTON, GA.
4.40	Corbicula manilensis	asian clam	SOPE CREEK NEAR MARIETTA, GA
4.40	Corbicula manilensis	asian clam	FLINT RIVER NEAR CULLODEN, GA
4.40	Corbicula fluminea	Asian clam	CLIFTY CREEK AT HARTSVILLE, IN
4.40	Pacifastacus leniusculus	signal crayfish	CARSON RIVER NEAR FORT CHURCHILL, NV
4.47	Corbicula manilensis	asian clam	CHATTAHOOCHEE RIVER NEAR WHITESBURG, GA
4.50	Agosia chrysogaster	longfin dace	SAN PEDRO RIVER AT CHARLESTON, AZ.
4.50	Corbicula manilensis	asian clam	CHATTAHOOCHEE RIVER NEAR NORCROSS, GA
4.50	Arctopsyche	-	BIG LOST RIVER AT HOWELL RANCH NR CHILLY ID
4.50	Corbicula fluminea	Asian clam	LOST RIVER NEAR LEIPSIC, IN
4.50	Corbicula	-	CLINCH RIVER ABOVE TAZEWELL, TN
4.50	Corbicula	-	ESQUATZEL COULEE AT SAGEMOOR RD NEAR PASCO, WA
4.57	Corbicula	-	CHAMBERS CK NR RICE, TX
4.60	Corbicula manilensis	asian clam	FLAT SHOAL CREEK AT STOVALL RD NEAR STOVALL, GA
4.60	Corbicula	-	BIG LIMESTONE CREEK NEAR LIMESTONE, TN
4.60	Corbicula	-	SALADO CK AT LOOP 13 AT SAN ANTONIO, TX
4.64	Corbicula	-	CONGAREE RIVER AT COLUMBIA, SC
4.76	Catostomus commersoni	white sucker	SADDLE RIVER AT RIDGEWOOD NJ
4.80	Agosia chrysogaster	longfin dace	PINTO CREEK NEAR MIAMI, AZ.
4.80	Corbicula manilensis	asian clam	SNAKE CREEK NEAR WHITESBURG, GA
4.80	Corbicula	-	BEAVER CREEK BELOW LIBERTY HILL, SC
4.80	Cottus sp.	freshwater sculpins	NF SKOKOMISH R BL STAIRCASE RPDS NR HOODSPORT, WA
4.80	Cottus sp.	freshwater sculpins	NOOKSACK RIVER AT NORTH CEDARVILLE, WA
4.81	Corbicula	-	NORTH FORK HOLSTON RIVER NEAR HAYTER GAP, VA
4.86	Cottus cognatus	slimy sculpin	CHESTER C AT ARCTIC BOULEVARD AT ANCHORAGE AK
5.09	Corbicula	-	MENARD CK NR FUQUA, TX

[Se]	Scientific Name	Common Name	Place Name
5.10	Corbicula manilensis	asian clam	LIME CREEK NEAR COBB, GA
5.10	Corbicula fluminea	Asian clam	KESSINGER DITCH NEAR MONROE CITY, IN
5.10	Corbicula fluminea	Asian clam	SALT CREEK AT HOOSIER AVENUE AT OOLITIC, IN
5.10	Cottus sp.	freshwater sculpins	SKOKOMISH RIVER NEAR POTLATCH, WA
5.13	Cottus sp.	freshwater sculpins	BEAR RIVER BELOW SMITHS FORK, NEAR COKEVILLE, WY
5.19	Cottus cognatus	slimy sculpin	KAMISHAK R NR KAMISHAK AK
5.20	Cyprinella lutrensis	red shiner	AGUA FRIA RIVER NEAR ROCK SPRINGS, AZ.
5.20	Corbicula fluminea	Asian clam	SUGAR CREEK AT CO RD 400 S AT NEW PALESTINE, IN
5.20	Corbicula	-	SOUTH BRANCH POTOMAC RIVER NEAR SPRINGFIELD, WV
5.30	Corbicula fluminea	Asian clam	EAST FORK WHITE RIVER AT SHOALS, IN
5.30	Corbicula	-	EDISTO RIVER NEAR COTTAGEVILLE,SC
5.40	Corbicula manilensis	asian clam	CHATTAHOOCHEE RIVER NEAR CORNELIA, GA
5.40	Corbicula manilensis	asian clam	WEST FORK LITTLE RIVER NEAR GAINESVILLE, GA.
5.40	Corbicula	-	TRUCKEE R AT LOCKWOOD, NV
5.40	Corbicula	-	BRUSHY CREEK NEAR PELHAM, SC
5.40	Corbicula	-	BIG CREEK ABOVE SALUDA, SC
5.70	Corbicula	-	AHOSKIE CR NEAR POORTOWN, NC
5.70	Corbicula	-	CLINCH RIVER AT SPEERS FERRY, VA
5.78	Hemichromis	jewelfishes	POAMOHO STREAM NR WAIALUA, OAHU, HI
5.79	Corbicula	-	KNOB CREEK AT AUSTIN SPRINGS
5.80	Corbicula manilensis	asian clam	FLINT RIVER NEAR LOVEJOY, GA
5.80	Corbicula	-	SABINAL RV NR SABINAL, TX
5.81	Cottus cognatus	slimy sculpin	MOOSE C NR PALMER AK
6.00	Corbicula fluminea	Asian clam	MUSCATATUCK RIVER NEAR DEPUTY, IN
6.00	Corbicula	-	LICK CREEK NEAR HOLLAND MILL, TN
6.00	Hydropsyche	-	CHAFFEE CREEK AT NESHKORO, WI
6.20	Corbicula	-	MEDINA RV AT LA COSTE, TX
6.30	Corbicula	-	INDIAN CREEK ABOVE NEWBERRY, SC
6.35	Cottus cognatus	slimy sculpin	SF CAMPBELL C NR ANCHORAGE AK
6.68	Cottus cognatus	slimy sculpin	COSTELLO C AB CAMP C NR COLORADO AK
6.70	Agosia chrysogaster	longfin dace	AGUA FRIA RIVER NEAR MAYER, AZ.
6.70	Agosia chrysogaster	longfin dace	AGUA FRIA RIVER AT BLOODY BASIN ROAD
6.70	Corbicula manilensis	asian clam	CHATTAHOOCHEE RIVER NEAR WHITESBURG, GA
6.70	Corbicula	-	BLANCO RV AT WIMBERLEY, TX
7.00	Corbicula	-	LONG CREEK ON SPENCER MTN RD NR SPENCER MTN, NC
7.30	Corbicula	-	GUEST RIVER AT COEBURN, VA
7.70	Corbicula	-	FRIO RV AT CONCAN, TX
8.10	Corbicula	-	VERDE R BLW TANGLE CREEK, ABV HORSESHOE DAM, AZ.
8.40	Corbicula	-	COMAL RV AT NEW BRAUNFELS, TX
8.47	Cottus cognatus	slimy sculpin	COSTELLO C NR COLORADO AK
9.10	Corbicula	-	NUECES RV BL UVALDE, TX
9.40	Cyprinella lutrensis	red shiner	VERDE R BLW TANGLE CREEK, ABV HORSESHOE DAM, AZ.
9.56	Cottus cognatus	slimy sculpin	CAMP C AT MOUTH NR COLORADO AK
9.83	Ictalurus punctatus	channel catfish	SABINAL RV NR SABINAL, TX
10.47	Cottus cognatus	slimy sculpin	COSTELLO C BL CAMP C NR COLORADO AK
12.83	Corbicula	-	GERONIMO CK AT HWY 90A NR SEGUIN, TX
14.40	Hydropsyche	-	GREEN CREEK NEAR PALMER, MI
22.37	Salmo trutta	brown trout	TONGUE RIVER NEAR DAYTON, WY

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