GEI Consultants

Consulting
Engineers and
Scientists

Updated Freshwater Aquatic Life Criteria for Selenium

March 2015

Updated Freshwater Aquatic Life Criteria for Selenium

Consulting
Engineers and
Scientists

Submitted to:
Henthorn Environmental Services, LLC
517 Sixth Avenue
St Albans, WV 25177

Submitted by:
GEI Consultants, Inc.
Ecological Division
4601 DTC Boulevard, Suite 900
Denver, CO 80237

March 2015
Project 1324860

Table of Contents

List of Tables

List of Appendices

1. Introduction

At the request of Henthorn Environmental Services, LLC, GEI Consultants, Inc. (GEI) has prepared this analysis in support of potential updates to state-wide water quality standards for selenium (Se).

Selenium is an essential micronutrient required by most aquatic and terrestrial species in order to maintain metabolic function (U.S. Environmental Protection Agency [EPA] 2004). It occurs in virtually all environmental media at trace concentrations, including rocks, soils, water, and living organisms. Anthropogenic activities, such as irrigating seleniferous soils, coal and phosphorus mining, operation of coal-fired power plants, and oil refining, have increased Se beyond background concentrations in many aquatic ecosystems (Lemly 1997).

Given the role of Se as an essential micronutrient, aquatic organisms readily bioaccumulate organic forms of Se (e.g., selenomethionine), yet frequently are not able to excrete Se at the same rate of consumption at elevated concentrations. This imbalance of intake and excretion can lead to elevated tissue concentrations that can be toxic to the organism. Direct toxic effects have been measured in adult organisms via decreased survival or growth and in young by decreased survival, growth, or increased occurrences of larval deformities (together, considered measures of reproductive success). The margin between required concentrations and those that may become toxic is narrow; perhaps as low as one order of magnitude for some vertebrate species, and highly variable within and between species. Furthermore, it has been difficult to differentiate the toxicity of different species of Se that occur due to varying hydrological and redox (reduction-oxidation) conditions (Payne 2013).

Chronic Se toxicity is directly related to dietary exposure and bioaccumulative properties of Se in aquatic biota rather than water column concentrations. As such, recent focus has been on development of fish tissue-based criterion (Payne 2013, EPA 2014). Tissue-based criteria are the most ecologically relevant for Se, as they are based on the chronic toxicity pathway which includes bioaccumulation of Se through dietary exposure (Brix and DeForest 2008, Chapman et al. 2009).

While the EPA is in the process of updating the national criteria for Se, it is unknown when the document will be finalized. The document is scheduled to be rereleased in draft form for a second round of public comment prior to being finalized, so it could still be a significant amount of time before it is complete. Therefore, in the meantime, interested states may develop their own updated criteria instead of relying on EPA's outdated and inappropriate criteria from 1987.

Regardless of when a new EPA criteria document may become available, derivation of an updated Se standard for an individual state is scientifically defensible, using approaches and analyses provided herein, due to new toxicity data made available since the current criterion

(EPA 1987) and the last draft criterion (EPA 2004) were released. This document provides a review of all available data, including that found in the EPA 2014 draft Se criteria document. Reanalysis of these data allow derivation of updated acute water column-based Se criteria and chronic fish tissue-based criteria for West Virginia.

2. Summary of Existing Criteria

2.1 National Ambient Water Quality Criteria for Selenium

The first national ambient water quality criteria (AWQC) for Se for the protection of aquatic life were published in 1976 (EPA 1976), updated in 1980 (EPA 1980), and then partially updated in 1987, 1995, and 1996 (EPA 1987 and 1995). These criteria were recommendations of water column limits for Se for the protection of aquatic life as required in the Clean Water Act (CWA). Under Section 304(a) of the CWA, the EPA must also periodically revise AWQC to incorporate the latest scientific knowledge on the kind and extent of all identifiable effects of pollutants on aquatic communities and human health. National AWQC are recommendations to states that must adopt water quality standards. Respective criteria can be modified to best reflect each state’s unique aquatic communities and environmental conditions.

The current acute (CMC) national AWQC (EPA 2012) for Se is:

$$\mathrm{CMC} = \frac{1}{[\mathrm{f1/CMC1}) + (\mathrm{f2/CMC2})]}$$

where f1 and f2 are the fraction of total Se that are comprised as selenite (Se^{+4}) and selenate (Se^{+6}), respectively, and CMC1 and CMC2 (acute values) are 185.9 and 12.82 micrograms per liter (µg/L), respectively, based on acute toxicity data and calculations from the 1987 criteria document (EPA 2012). The current chronic national AWQC for Se is 5 µg/L.

In 2002 and later in 2004, the EPA published draft criteria documents that recognize the differential modes of Se toxicity – primarily water column exposure for acute toxicity and mixed water column and dietary exposure followed by bioaccumulation into tissues for chronic (Canton 1999, Brix et al. 2001a,b, EPA 2002 and 2004). The document also acknowledged the different acute toxicity of selenite and selenate and the relationship between selenate toxicity and ambient sulfate concentration (EPA 2004). Se speciation is important in determining potential exposure routes and biogeochemical cycling in aquatic environments (Ralston et al. 2008). Elemental Se and most metallic selenides have relatively low toxicities because of their low bioavailability. By contrast, selenate and selenite are very bioavailable. At pH values below 7.0, selenites are rapidly reduced to elemental Se under mildly reducing conditions (Faust and Aly 1981) that are common in most aquatic sediments. Selenate usually predominates in well-aerated surface waters, especially those with alkaline conditions (Faust and Aly 1981, Luoma et al. 1992). Selenite is more reactive than selenate because of its polarity and high attraction to other molecules (EPA 2004), making selenite more bioavailable, increasing exposure and potential toxicity to aquatic organisms.

The EPA (2004, 2012) derived two separate acute criteria for selenite and selenate. The draft selenite criterion (258 µg/L) was derived using the established 5th percentile criteria derivation methodology (Stephan et al. 1985) based on an updated selenite acute toxicity database. The selenate criterion was derived using the same 5th percentile methodology on an updated acute toxicity database. Additionally, the acute selenate values were normalized based on sulfate concentrations in the test, as data indicate sulfate has a significant influence on selenate acute toxicity (Brix et al. 2001a,b, EPA 2004). The result is a sulfate-based acute toxicity water quality criteria equation for selenate:

$$Acute\ selenate = e^{(0.5812[ln\,(sulfate)]\,+\,3.357)}$$

Chronic Se toxicity, on the other hand, is related to dietary exposure and bioaccumulative properties of Se in aquatic biota rather than water column concentrations. Therefore, the draft criteria document (EPA 2004) proposed a national tissue-based chronic criterion. Fish are considered particularly sensitive to chronic Se exposure (Coyle et al. 1993, GEI et al. 2008, Hamilton et al. 1990, Hermanutz et al. 1996), with early life history stages of fish development being most affected. Due to the bioaccumulative properties of Se, exposure routes in embryonic and larval fish can be from maternally derived yolk absorption or directly from the environment. Selective early life stage sensitivities in fish can create a scenario where significant population mortality occurs in Se affected waters, despite the presence of seemingly healthy adult populations (Lemly 2002).

The most recent 2014 EPA draft Se criteria document focuses on chronic criteria only and recommends two fish-tissue based (egg/ovary and whole-body) and two water-based criterion (lotic and lentic) elements (EPA 2014) (Table 1). The most recent EPA document does not include recommendations for acute water column-based criteria, noting that values such as those proposed in their earlier 2004 document would likely not be protective of bioaccumulation and could result in exceedence of the chronic tissue values.

For the 2014 draft, the criteria are based solely on studies that included reproductive (i.e., maternal transfer) effects on larval survival/deformity/etc. The draft egg/ovary criterion was developed using 19 reproductive studies with nine fish genera, and the whole-body and muscle criterion were translated from the egg/ovary number using tissue-to-tissue conversion factors (CF). The monthly average water column criteria were developed using enrichment factors (EF), CFs and composite trophic transfer functions ($TTF^{composite}$), with different values for lentic and lotic systems. As no acute criterion is proposed, the 2014 draft includes an intermittent exposure component as an attempt to address pulses of elevated Se concentration that could contribute to chronic effects. The equation includes background concentrations and the fraction of the month during which elevated concentrations occur.

Table 1: Summary table of the EPA 2014 draft Se chronic criterion (from p. 4, EPA 2014).

Media Type	**Fish Tissue**		**Water Column**[3]	
Criterion Element	**Egg/Ovary**[1]	**Fish Whole-Body or Muscle**[2]	**Monthly Average Exposure**	**Intermittent Exposure**[4]
Magnitude[5]	15.2 µg/g	8.1 µg/g whole-body or 11.8 µg/g muscle (skinless, boneless filet)	1.3 µg/L in lentic aquatic systems 4.8 µg/L in lotic aquatic systems	$WQC_{int} = \frac{WQC_{30\text{-day}} - C_{bkgrnd}(1-f_{int})}{f_{int}}$
Duration	Instantaneous measurement[5]	Instantaneous measurement[5]	30 days	Number of days/month with an elevated concentration
Frequency	Never to be exceeded	Never to be exceeded	Not more than once in three years on average	Not more than once in three years on average

[1] Overrides any whole-body, muscle, or water column elements when fish egg/ovary concentrations are measured.

[2] Overrides any water column element when both fish tissue and water concentrations are measured.

[3] Water column values are based on dissolved total selenium in water.

[4] Where $WQC_{30\text{-day}}$ is the water column monthly element, for either a lentic or lotic system, as appropriate. C_{bkgrnd} is the average background selenium concentration, and f_{int} is the fraction of any 30-day period during which elevated selenium concentrations occur, with f_{int} assigned a value ≥0.033 (corresponding to 1 day).

[5] Instantaneous measurement. Fish tissue data provide point measurements that reflect integrative accumulation of selenium over time and space in the fish at a given site. Selenium concentrations in fish tissue are expected to change only gradually over time in response to environmental fluctuations.

2.2 West Virginia State Surface Water Quality Standards

West Virginia’s current surface water quality standards are presented in Rule 47CSR2 (WVDEP 2011). The acute and chronic Se standards for the protection of aquatic life, based on the EPA’s 1987 criteria (EPA 1987), are 20 µg/L and 5 µg/L total Se, respectively.

3. Updates to West Virginia State Se Standards

As discussed above (Section 2.1), the EPA supports a tissue-based criterion for Se in its draft document (EPA 2014) because it incorporates site-specific factors such as chemical speciation and rates of transformation, as well as variations in temporal concentrations in water, and types of organisms constituting the food chain.

While EPA is in the process of finalizing the national Se criteria, interested states would benefit from developing their own updated criteria and not relying on the outdated and inappropriate criteria from the 1987 criteria document – now over 25 years old.

3.1 Acute Se

As mentioned previously, acute criteria are not proposed in the 2014 EPA draft document. The proposed acute freshwater criteria in the 2004 AWQC draft document are greatly improved from the 1987 Se criteria, and represent a significant increase from the current West Virginia acute total Se standard of 20 µg/L. West Virginia water quality criteria for total Se cannot differentiate between the markedly different reported selenate and selenite toxicity. Substantial improvements over current criteria include:

1. recognition of the differential modes of toxicity between acute (water column) and chronic (dietary and bioaccumulation) Se exposure (Canton 1999),
2. developing the relationship between selenate toxicity and sulfate concentration (Brix et al. 2001a, b), and
3. development of separate acute criteria for selenite and selenate.

Given the considerable difference in the acute Se criteria values proposed by the EPA in the 2004 draft Se AWQC document (EPA 2004) compared to their previous criteria (EPA 1987), there is substantial evidence that the current West Virginia acute standard of 20 µg/L is not relevant and adoption of updated acute standards is warranted. EPA's updated draft acute Se criteria for selenite and selenate would provide a strong, scientifically defensible update to acute Se standards for West Virginia.

Based on this analysis, we would strongly recommended acute Se standards for West Virginia be replaced with the current acute (CMC) national AWQC (EPA 2012) equation for total Se and that the values for selenite and selenate currently based on the 1987 criteria document (EPA 1987) be replaced with the more scientifically-defensible values from the 2004 draft:

$$\mathrm{CMC} = \frac{1}{[\mathrm{f1}/\mathrm{CMC1}) + (\mathrm{f2}/\mathrm{CMC2})]}$$

where f1 and f2 are again the fraction of total Se that are comprised as selenite (Se^{+4}) and selenate (Se^{+6}), respectively, and CMC1 and CMC2 are now 258 µg/L for selenite and the equation $e^{(0.5812[\ln (\text{sulfate})] + 3.357)}$ for selenate (EPA 2004). It is understood that many water quality programs do not include monitoring of the various species of Se. Thus, if Se speciation analyses are not conducted on water column Se samples, we would recommend use of the more restrictive of the two values, 258 µg/L, as a conservative acute total Se standard, assuming sulfate concentrations greater than approximately 44 mg/L. If sulfate values are less than approximately 44 mg/L (Table 2), then speciation may be warranted to develop acute standards that are fully protective, as this equation may result in values lower than 258 µg/L.

Table 2: Freshwater selenate values (µg/L dissolved) for varying concentrations of sulfate.

	Mean Sulfate (mg/L)									
	5	10	15	20	25	30	35	40	45	50
Selenate (acute)	73	109	138	164	186	207	227	245	262	279

3.2 Chronic Se

Prior to proposing an updated chronic water quality standard using tissue-based criteria for West Virginia, EPA’s 2014 draft criteria were evaluated. We reviewed the available chronic toxicity Se tissue data at the family level specific to the families of fish that occur (or would be expected to occur) in West Virginia waters (WVDEP 2015). The most species-rich families in West Virginia include Centrarchidae, Cyprinidae, Catostomidae, Ictaluridae, and Percidae. The 2014 draft criteria document (EPA 2014) included chronic tissue endpoints for three of these families: Centrarchidae, Cyprinidae, and Catostomidae. In addition, chronic data were available for Esocidae, Poeciliidae, and Salmonidae, which are also present in West Virginia.

The number and scope of available toxicity studies addressing tissue-based effects of chronic Se exposure remain limited. Previously, 24 studies were evaluated in the 2004 Se draft document (EPA 2004) resulting in Se tissue thresholds for nine species in seven genera and one general family tissue threshold. After their evaluation of all acceptable studies, the EPA proposed the chronic criterion of 7.9 micrograms per gram (µg/g) Se whole-body (wb) dry weight (dw), which was derived from a single study that investigated juvenile bluegill mortality during winter months (Lemly 1993).

The EPA approach in the 2014 draft Se criteria document, as well as the Se standards adopted in Kentucky (Payne 2013) are more in line with standard water quality criteria development methodology (Stephan et al. 1985). EPA 2014 includes a critical evaluation of 37 studies on various fish species and results in Se tissue thresholds for eleven fish species in nine genera. Criteria calculations follow recommendations by Stephan et al. (1985) and use the 5th percentile calculation accounting for the relative sensitivities of all species in the data set. This approach results in more scientifically defensible criteria than the previous draft tissue criterion based on a single study.

The 2014 draft Se criteria document only uses EC_{10} values from studies with reproductive endpoints to derive tissue-based criteria. EPA prefers tissue-based criteria that focus on the reproductive tissues, represented by egg/ovary tissue, and the primary criterion in the draft 2014 Se criterion document is an egg/ovary number (EPA 2014). However, we believe the use of whole-body tissue thresholds will also be helpful in the implementation of any tissue-based fish tissue numeric target, given the difficulties of field-collection of egg/ovary tissue.

3.2.1 *Comments on Studies Deemed Acceptable in EPA 2014*

The 2014 draft Se criteria document includes reproductive toxicity study data for nine fish genera and discussion on use of the various studies' data to develop their criteria can be found in Appendices of their document (EPA 2014). In our review of the EPA data for development of tissue-based criteria for West Virginia, we have excluded two genera not found in the state, Dolly Varden (*Salvelinus malma*) and the desert pupfish (*Cyprinodon macularius*), and one Oncorhynchus species, cutthroat trout (*O. clarki*). There were two species used to develop the genus mean chronic value (GMCV) for Gambusia, the western mosquitofish (*Gambusia affinis*) and the eastern mosquitofish (*Gambusia holbrooki*). While *G. affinis* are not found in West Virginia, we included this species in the calculations because the effect concentrations for these species are all "greater than" values, indicating they are not highly sensitive to Se, but it is unknown precisely how sensitive they are. Therefore, using values for both species provides a more conservative approach for this genus.

Overall, we concur with most of the data usage decisions made by EPA in the 2014 draft. We have provided comments and suggestions on some of the data decisions that were used to develop EPA's draft egg/ovary chronic criterion and subsequently, the whole-body criteria (GEI 2014a and GEI 2014b). We believe incorporation of these suggested changes would result in tissue-based criteria for West Virginia that are more scientifically defensible and consistent with EPA's other data-usage decisions.

3.2.1.1 Fathead Minnow Data

One of our recommendations results in a recalculation of the number used for fathead minnows. EPA omitted the data from the fathead maternal transfer study conducted by GEI (2008) citing high variability and insufficient response as the reasons for excluding this study. However the results of this study are consistent with other studies used by EPA (GEI 2014a and GEI 2014b). We recommend including the chronic whole-body value of 42.067 µg/g calculated in the GEI study along with the Schultz and Hermanutz (1990) values in the derivation of a fathead minnow GMCV.

As the Schultz and Hermanutz (1990) study results in an egg/ovary value, and the GEI study (2008) results in a whole-body value, a conversion factor (CF) is needed to translate the values.

EPA (2014) used a generic median Cyprinidae value of 2.00 to convert between these tissues. However, we believe that species-specific CFs and/or actual regression equations should be used when possible (GEI 2014a). Using matched tissue data from the study conducted by GEI to supplement the EPA CF database, it was possible to calculate a new egg/ovary to whole-body CF for the fathead minnow of 1.4. When this species-specific CF is used, the Schultz and Hermanutz (1990) egg/ovary value of <23.85 µg/g would be translated to a whole-body value of 17.04 µg/g. Using this value with the GEI whole-body value of 42.067 µg/g would result in a fathead minnow whole-body GMCV of 26.77 µg/g (Table 3 and Table 4).

3.2.1.2 Bluegill Data

In the 2014 draft Se criteria document, EPA utilized three bluegill studies in the derivation of the tissue-based criteria: Doroshov et al. (1992), Coyle et al. (1993), and Hermanutz et al. (1992, 1996). While the data in the Doroshov et al. (1992) and Coyle et al. (1993) studies are useable as is, we recommend revising the egg/ovary EC_{10} of 12.68 µg/g derived from the Hermanutz et al. (1992, 1996) studies.

For reasons described in detail in GEI 2014a, we recommend excluding Study I and only using data from Study II from EPA 2014. Using only Study II data, we used the same methods and parameters as EPA to calculate an egg/ovary EC_{10} of 23.15 µg/g. This value is much closer to the other two chronic values for bluegill reported by EPA (20.05 µg/g [Doroshov et al. 1992] and 24.55 µg/g [Coyle et al. 1993]) in their maternal transfer toxicity database (Table 5 in EPA 2014), indicating that combination of Studies I and II by EPA was producing a potentially unrealistic value for this species. Use of this updated chronic egg/ovary value of 23.15 µg/g results in an updated bluegill egg/ovary GMCV of 22.50 µg/g (Table 3 and Table 4). This is the value we are recommending for this genus.

To develop whole-body chronic values, EPA translated the egg/ovary chronic values using median-based egg/ovary to whole-body CFs. In the past, EPA had used regression-based CFs (EPA 2004). We believe EPA should use regression-based egg/ovary to whole-body translators when appropriate (i.e., when the regression relationship had an R^2 value >0.70) to translate the egg/ovary values to whole-body. For bluegill, use of the regression-based CF results in a whole-body chronic value of 10.78 µg/g (Table 3 and Table 4).

3.2.1.3 Brown Trout Data

In the 2014 draft Se criteria document, EPA utilized brown trout data from Formation Environmental (2011). During this study, a tank overflow accident occurred which resulted in the inadvertent loss of several study fish. EPA presented two approaches for dealing with this loss of these study organisms: (1) A“worst case” assumption that all fry lost were dead or deformed and (2) An “optimistic” assumption that fry lost had the same rates of mortality and deformities as those not lost. EPA chose to assume the “worst case” scenario and derived an egg/ovary EC_{10} of 15.91 µg/g.

We believe the scenario where the fry lost had the same mortality and deformity rates as those not lost would be a more “realistic” assumption, as it reflects what was observed in the remaining population (i.e., the fish not lost to overflow) (GEI 2014a). Using this “optimistic”/realistic approach, the reported egg/ovary EC_{10} is 18.36 µg/g, which is considerably more comparable to data for other Salmonids (Table 5 in EPA 2014) than the EC_{10} of 15.91 µg/g from EPA’s “worst case” approach.

Additionally, there is no valid reason to use the deformities endpoint alone when the combined survival and deformities endpoint is available. In fact, this is more in line with the EPA’s previous approach in the 1999 ammonia criteria document where they used the combined survival and growth endpoint, termed “biomass”. When these combined data are used, the EC_{10} for the “optimistic” assumption is 21.16 µg/g.

Based on these analyses, we believe that the egg/ovary EC_{10} of 21.16 µg/g for combined endpoints (i.e., survival and deformities) is the most appropriate for the brown trout study (GEI 2014b). The egg/ovary value can then be translated using the median brown trout CF provided by EPA to a whole-body chronic value of 14.59 µg/g (Table 3 and Table 4).

Table 3: Selenium tissue threshold values for fish. MT = maternal transfer, WB = whole-body, CF = conversion factor, LOAEC = lowest observable adverse effect concentration, LOEC = lowest observed effect concentration, NOEC = no observed effect concentration, EC = effect concentration.

Species	Reference	Notes	Test Type	Toxicological Endpoint	Chronic Value µg/g dw	
					Egg/Ovary	WB
Pimephales promelas Fathead minnow	Schultz and Hermanutz 1990	EPA 2014 used 75.3% moisture for eggs/ovaries; translated from ovary using CF of 1.4 (GEI 2014a)	Dietary and waterborne (mesocosm; Monticello)	LOAEC for larval edema and lordosis	Ovary LOAEC: <23.85	WB LOAEC: <17.04
	GEI 2008	Translated from WB using CF of 1.4 (GEI 2014a)	Dietary and waterborne (field; Denver, CO)	EC_{10} larval skeletal and edema abnormality	Egg/Ovary EC_{10}: 58.89	WB EC_{10}: 42.07
Esox lucius Northern pike	Muscatello et al. 2006	EPA notes EC10 cannot be estimated; translated from egg (Eq. A)	Dietary and waterborne (field Saskatoon, Sask.)	EC24 larval deformities	Egg EC24: 34.0	WB EC_{10}: 30.55
Gambusia holbrooki Eastern mosquitofish	Staub et al. 2004	Translated from WB using EPA CF of 1.71	Field MT	NOEC for brood size/offspring viability	Egg NOEC: >20.26	WB NOEC: >11.85
Gambusia affinis Western mosquitofish	Saiki et al. 2004	Translated from WB using EPA CF of 1.71	Field MT	NOEC for fry mortality and deformities	Egg NOEC: >25.82	WB NOEC: >15.1
Micropterus salmoides Largemouth bass	CP&L 1997	Translated from ovary (Eq. B)	Lab MT	EC_{10} for larval mortality and deformity	Ovary EC_{10}: 20.35	WB EC_{10}: 11.08
Lepomis macrochirus Bluegill	Doroshov et al. 1992	Translated from egg (Eq. C)	Dietary (lab)	EC_{10} for larval edema	Egg EC_{10}: 20.05	WB EC_{10}: 9.74
	Coyle et al. 1993	Translated from egg (Eq. C)	Dietary and waterborne (lab)	EC_{10} for larval survival	Egg EC_{10}: 24.55	WB EC_{10}: 11.65
	Hermanutz et al. 1992; Hermanutz et al. 1996; Tao et al. 1999.; EC_{10} calculated by GEI	Translated from ovary (Eq. C)	Dietary and waterborne (mesocosm; Monticello)	EC_{10} for larval edema	Ovary EC_{10}: 23.15	WB EC_{10}: 11.05
Oncorhynchus mykiss Rainbow trout	Holm 2002; Holm et al. 2003; Holm et al. 2005	EPA 2014 used 61.2% moisture for eggs; translated from egg (Eq. D)	Dietary and waterborne (field; Luscar River, AB)	EC_{10} for skeletal deformities	Egg EC_{10}: 21.1	WB EC_{10}: 13.79
Salmo trutta Brown trout	Formation Environmental 2011; AECOM 2012; EC10 calculated by GEI	Translated from egg using EPA CF of 1.45	Dietary and waterborne (field; Lower Sage Creek and Crow Creek, ID)	EC_{10} for larval survival and deformities	Egg EC_{10}: 21.16	WB EC_{10}: 14.59

Equations used to translate between whole-body and egg/ovary:

$^{\text{Eq. A}}$ NP [Se] dw WB = 0.9426*(NP [Se] dw egg/ovary) - 1.4953

$^{\text{Eq. B}}$ CENTRARCHIDAE [Se] dw WB = 0.4384*(CENTRARCHIDAE [Se] dw egg/ovary) + 2.161

$^{\text{Eq. C}}$ BG [Se] dw WB = 0.4239*(BG [Se] dw egg/ovary) +1.2392

$^{\text{Eq. D}}$ RBT [Se] dw WB = 0.6582*(RBT [Se] dw egg/ovary) - 0.0949

Table 4: Ranked selenium toxicity data available for West Virginia fish species used to calculate GMCVs. CV = Chronic Value, GMCV = Genus Mean Chronic Value, WB = whole-body. *Asterisks indicate GMCV values that differ from those reported in the 2014 draft Se criteria document.

			Whole-body			**Egg/Ovary**		
Species	**Endpoint**	**Reference**	**CV µg/g**	**GMCV µg/g**	**WB Rank**	**CV µg/g**	**GMCV µg/g**	**Egg/Ovary Rank**
Lepomis macrochirus Bluegill	Larval edema EC_{10}	Doroshov et al. 1992	9.74	10.78*	1	20.05	22.50*	4
	Larval edema EC_{10}	Hermanutz et al. 1992, 1996	11.05			23.15		
	Larval survival EC_{10}	Coyle et al. 1993	11.65			24.55		
Micropterus salmoides Largemouth bass	Larval mortality and deformities EC_{10}	CP&L 1997	11.08	11.08*	**2**	20.35	20.35	**1**
Oncorhynchus mykiss Rainbow trout	Skeletal deformities EC_{10}	Holm 2002; Holm et al. 2003; Holm et al. 2005	13.79	13.79*	**3**	21.1	21.1	**2**
Salmo trutta Brown trout	Larval survival and deformities EC_{10}	Formation Environmental 2011; AECOM 2012	14.59	14.59*	**4**	21.16	21.16*	**3**
Gambusia holbrooki Eastern mosquitofish	Brood size/offspring viability NOEC	Staub et al. 2004	>11.85	>15.1	**5**	>20.26	>25.82*	**5**
Gambusia affinis Western mosquitofish	Fry mortality and deformities NOEC	Saiki et al. 2004	>15.1			>25.82		
Pimephales promelas Fathead minnow	Larval edema/lordosis LOEC	Schultz and Hermanutz 1990	<17.04	26.77*	**6**	<23.85	37.48*	**6**
	Larval skeletal and edema abnormality EC_{10}	GEI 2008	42.07			58.89		
Esox lucius Northern pike	Larval deformities EC_{24}	Muscatello et al. 2006	30.55	30.55*	**7**	34.0	34.0	**7**

3.2.2 *Conversion Factors*

As mentioned previously, we believe regression-based CFs using the matched egg/ovary, whole-body, and muscle Se data provided in the 2014 draft Se criteria document is preferable to median CFs, if appropriate data are available (GEI 2014a). As part of our evaluation of the conversion factors (CFs) developed by EPA in the 2014 draft Se criteria document, we reviewed all of the data used and corrected values where mistakes were found. In addition, we calculated geometric means of the tissue-to-tissue ratios to determine how CF outcomes might vary under different statistical methods. A detailed evaluation of this issue is also presented by NAMC-SWG (2014). The corrected/updated values specific to this analysis for West Virginia are found in Table 5.

In addition to reviewing EPA's data and calculations, we also compiled matched tissue data from studies conducted by GEI to supplement their CF database (Appendix A). As a result of these data additions, it was possible to calculate new egg/ovary to whole-body CFs for creek chub and fathead minnow (Table 5). Without these species-specific CFs, it would be necessary to use surrogate CFs for similar species or families to convert between tissues, which introduces uncertainty into the translation. For instance, data for the fathead minnow, which were included in the tissue-based criteria calculations, were translated from egg/ovary to whole-body concentrations by EPA using a generic conversion factor of 2.00 (based on the median for Cyprinidae). However, sufficient data are available to calculate a fathead minnow-specific conversion factor. Using 45 matched datapoints from GEI (2008), we calculated the median of the 45 individual matched egg/ovary to whole-body ratios to develop an egg/ovary to whole-body CF of 1.4 for the fathead minnow (Table 5). When translating between fathead minnow tissues, this species-specific CF for fathead minnow is more relevant than a generic Cyprinidae CF.

In addition, using the matched egg/ovary, whole-body, and muscle Se data provided in the 2014 draft Se criteria document, which was further updated by GEI as described above, we developed regression-based CFs (Appendix A; Table 5). When the regression has a relatively high goodness of fit (i.e., when R^2 is at least 0.70), we recommend using the regression equation in place of the median (or geometric mean) ratios, as the regression better predicts tissue concentrations, particularly at the high and low ends of the spectrum. Where the strength of the regression is not as high (e.g., fathead minnow), it may be more appropriate to use the median or geometric mean CF to represent the central tendency of the relationship. As shown in Section 3.2.1, we used the regression-based CFs for northern pike, bluegill, rainbow trout, and Centrarchidae to translate the updated egg/ovary criterion database for these species to whole-body for the purposes of deriving the updated whole-body criterion. For the remaining species, we used the updated and new median ratio-based CFs (Table 5).

Table 5: Egg/ovary to whole-body CF median ratios from Table 11 in the 2014 draft Se criteria document, calculated geomean ratios, and regression equations. Strikeouts indicate EPA calculation errors. Revised values and species added to the database are shown in bold.

Common Name	Scientific Name	CF Median Ratio	CF Geomean	CF Regression
Fathead minnow	*Pimephales promelas*	**1.40**	**1.48**	y=0.6995x+1.0477 (R^2 = 0.54)
Northern pike	*Esox lucius*	~~2.39~~ **2.41**	2.55	y=0.9426x-1.4953 (R^2 = 0.8307)
Bluegill	*Lepomis macrochirus*	2.13	1.86	y=0.4239x+1.2392 (R^2 = 0.816)
Green sunfish	*Lepomis cyanellus*	1.45	1.41	y=0.7301x - 0.1638 (R^2 = 0.8696)
Smallmouth bass	*Micropterus dolomieu*	1.42	1.44	y=0.5721x + 0.9636 (R^2 = 0.7254)
Brown trout	*Salmo trutta*	1.45	1.15	y=0.272x+4.2255 (R^2 = 0.4652)
Rainbow trout	*Oncorhynchus mykiss*	~~2.44~~ **2.46**	2.32	y=0.6582x-0.0949 (R^2 = 0.9565)
Centrarchidae	*Lepomis macrochirus*, *L. cyanellus*, *Micropterus salmoides* and *M. dolomieu*	~~1.45~~ **1.53**	1.57	y=0.4384x + 2.161 R^2 = 0.7555

3.2.3 *Invertebrate Data*

EPA used data from chronic invertebrate toxicity studies and translated them to predicted fish tissue concentrations that would result from consuming invertebrates containing Se at these chronic values. The resulting egg/ovary values were then considered to be GMCVs and were incorporated with the fish GMCVs into the species sensitivity distributions and used in criteria calculations (see Figure 5 in EPA 2014).

While we appreciate EPA's effort to follow criteria derivation protocols and meet the eight-family rule, this approach is not toxicologically valid. The cited invertebrate studies were conducted to assess the toxicity of Se to invertebrates. Simply translating these values to expected fish tissue concentrations does not make them equivalent to fish tissue chronic effects values. Specifically, the translated values are not linked to fish toxicity in any way—rather, they only reflect what expected fish tissue concentrations would be if a fish consumed invertebrates containing Se concentrations found to elicit effects in invertebrates. The fish tissue criteria should only be based on fish tissue data, not invertebrate data.

In EPA's analysis of invertebrate data, the mayfly, *Centroptilum triangulifer*, was found to be the most sensitive, with a GMCV of 24.2 mg Se/kg dw wb. Effect concentrations for the other invertebrates for which chronic data were available were substantially higher than

24.2 µg/g. The EC_{10} calculated for rotifer growth was 37.84 µg/g dw wb, and effects were seen with oligochaetes at levels greater than 140 µg/g dw wb. Therefore, based on these data, an invertebrate Se tissue concentration of 24.2 µg/g dw would be protective of these invertebrates.

3.3 Recommended West Virginia Chronic Tissue Criterion

3.3.1 *Whole-body Criterion*

Incorporation of the data decisions described above also results in changes to the calculated whole-body chronic values for many of the species from those in EPA (2014). Using the data presented in Table 6 and Table 7, an updated whole-body criterion of 9.0 µg/g can be derived using EPA criteria calculation methodology (Stephan et al. 1985). This whole-body criterion is scientifically defensible and consistent with EPA's other data-usage decisions in their draft (EPA 2014).

Table 6: Ranked genus mean chronic values for calculated fish WB endpoints *Asterisks indicate values that differ from those reported in the 2014 draft Se criteria document.

Rank	GMCV (µg Se/g dw WB)	Species	SMCV (µg Se/g dw WB)
7	30.55*	Northern pike, *Esox lucius*	30.55
6	26.77*	Fathead minnow, *Pimephales promelas*	26.77
5	> 15.1	Eastern mosquitofish, *Gambusia holbrooki*	>11.85
		Western mosquitofish, *Gambusia affinis*	>15.1
4	14.59*	Brown trout, *Salmo trutta*	14.59
3	13.79*	Rainbow trout, *Oncorhynchus mykiss*	13.79
2	11.08*	Largemouth bass, *Micropterus salmoides*	11.08
1	10.78*	Bluegill sunfish, *Lepomis macrochirus*	10.78

Table 7: Calculation of whole-body fish tissue-based Se criterion (N = 7 genera, R = sensitivity rank in database).

Rank	Genus	GMCV	ln GMCV	(ln GMCV)^2	P = R/(N+1)	√P
1	*Lepomis*	10.78	2.3777	5.6534	0.1250	0.3536
2	*Micropterus*	11.08	2.4051	5.7847	0.2500	0.5000
3	*Oncorhynchus*	13.79	2.6239	6.8851	0.3750	0.6124
4	*Salmo*	14.59	2.6803	7.1842	0.5000	0.7071
Sum			10.0871	25.5074	1.2500	2.1730

Calculations:
Chronic Whole-body Criterion

$$S^2 = \frac{\sum(\ln GMCV)^2 - (\sum \ln GMCV)^2/4}{\sum P - (\sum\sqrt{P})^2/4} = \frac{25.5074 - (10.0871)^2/4}{1.2500 - (2.1730)^2/4} = 1.0066 \qquad S = 1.0033$$

$$L = [\sum \ln GMCV - S(\sum\sqrt{P})]/4 = [10.0871 - 1.0033\ (2.1730]/4 = 1.9767$$

$$A = S(\sqrt{0.05}) + L = (1.0033)(0.2236) + 1.9767 = 2.2011$$

Final Chronic Value = FCV = e^A = **9.0347**

3.3.2 *Egg/ovary Criterion*

Implementing the data decisions discussed above (Section 3.2) results in changes to the criteria calculations from those in EPA (2014). The order and chronic values for the top four most sensitive species change as a result of the modifications to the bluegill and brown trout GMCVs (Table 8). Using the revised brown trout and bluegill values and the recommendation to use only fish data relevant to West Virginia in the calculation (i.e., N=7, not N=14), results in an egg/ovary criterion of 19.5 µg/g (Table 9).

Table 8: Ranked genus mean chronic values for fish reproductive endpoints *Asterisks indicate values that differ from those reported in the 2014 draft Se criteria document.

Rank	GMCV (µg Se/g dw EO)	Species	SMCV (µg Se/g dw EO)
7	37.48*	Fathead minnow, *Pimephales promelas*	37.48*
6	<34	Northern pike, *Esox lucius*	<34
5	> 25.82 estim. EO* (> 15.1 meas. WB)	Eastern mosquitofish, *Gambusia holbrooki*	> 20.26 estim. EO* (> 11.85 meas. WB)
		Western mosquitofish, *Gambusia affinis*	> 25.82 estim. EO* (> 15.1 meas. WB)
4	22.50*	Bluegill sunfish, *Lepomis macrochirus*	22.50*
3	21.16*	Brown trout, *Salmo trutta*	21.16*
2	21.1	Rainbow trout, *Oncorhynchus mykiss*	21.1
1	20.35	Largemouth bass, *Micropterus salmoides*	20.35

Table 9: Calculation of egg/ovary fish tissue-based Se criterion based on values in Table 8 (N = 7 genera, R = sensitivity rank in database).

Rank	Genus	GMCV	ln GMCV	(ln GMCV)^2	P = R/(N+1)	√P
1	*Micropterus*	20.35	3.0131	9.0787	0.1250	0.3536
2	*Oncorhynchus*	21.10	3.0493	9.2981	0.2500	0.5000
3	*Salmo*	21.16	3.0521	9.3154	0.3750	0.6124
4	*Lepomis*	22.50	3.1135	9.6940	0.5000	0.7071
		Sum	12.2280	37.3861	1.2500	2.1730

Calculations:
Chronic Egg/Ovary Criterion

$$S^2 = \frac{\sum(\ln GMCV)^2 - (\sum \ln GMCV)^2/4}{\sum P - (\sum\sqrt{P})^2/4} = \frac{37.3861 - (12.2280)^2/4}{1.2500 - (2.1730)^2/4} = 0.0749 \qquad S = 0.2737$$

$L = [\sum \ln GMCV - S(\sum\sqrt{P})]/4 = [12.2280 - 0.2737(2.1730]/4 = 2.9083$

$A = S(\sqrt{0.05}) + L = (0.2737)(0.2236) + 2.9083 = 2.9695$

Final Chronic Value = FCV = e^A = **19.4821**

4. Implementation Recommendations

4.1 Acute Se

We would recommend deleting the current acute Se criterion of 20 µg/L and replacing it with the EPA footnote equation (EPA 2012) for Se:

$$CMC = 1/[f1/CMC1)+(f2/CMC2)],$$

where f1 and f2 are the fraction of total Se that are comprised as selenite (Se^{+4}) and selenate (Se^{+6}), respectively. In addition, we would recommend use of the updated values for sulfate, where CMC1 = 258 µg/L for selenite and CMC2 = $e^{(0.5812[\ln (\text{sulfate})] + 3.357)}$, consistent with EPA (2004) updates – not the values currently cited in EPA (2012), which are based on the outdated 1987 criteria document. If Se speciation is not conducted, the more conservative value of 258 µg/L would be applied (if sulfate is greater than 44 mg/L at a site – see Table 2 for example values at varying sulfate concentrations).

4.2 Chronic Se

Based on the current science, it is known that tissue Se concentrations better represent actual Se exposure and uptake by aquatic life. However, implementation of a tissue-based threshold is potentially difficult for regulators and the regulated community, as attainment assessments would require collection of fish tissue data on a regular basis in a wide variety of aquatic habitats – and potentially collection during the reproductive cycle of multiple resident fish species, given potential use of egg/ovary Se criteria.

Thus, we would recommend a tiered approach that would retain the 5 µg/L total Se value as the primary standard for initial assessment of attainment of the Se chronic standard, and employ a tissue-based standard as needed in a tiered approach. Such an approach would require determination of compliance/non-compliance with the default water column value first. If the water column value is met, no further assessment is needed. If the water column value is exceeded, fish tissue (whole-body or egg/ovary tissue) can be collected and compared to the thresholds calculated above. The suggested tiered approach is as follows:

Step 1. Determine whether site is in attainment of the 5 µg/L water column-based standard.

- If water quality is below 5 µg/L, the analysis is complete and water-body is considered in attainment.
- If water quality is greater than 5 µg/L, proceed to Step 2.

Step 2. Determine whether the site is in attainment of the tissue threshold (whole-body [9.0 µg/g], or egg/ovary [19.5 µg/g]).

- If tissue Se concentrations are less than the appropriate tissue-based standard, the analysis is complete and the water-body is considered in attainment.
- If the site tissue Se concentrations exceed the tissue-based standard, the site is considered in non-attainment, and evaluation of Se sources and effects is necessary.
- Results of this step may include an analysis of whether the Se source is natural or anthropogenic, and also if Se is negatively impacting aquatic life populations. These analyses could be accomplished through detailed sampling and analysis of fish populations, as well as determination of sources/fate of Se in the affected waterbody.

It is important to note that West Virginia’s method for determining attainment is generally based on the percentage of individual samples indicating impairment (WVDEP 2012). If the total number of samples is 20 or fewer, then three or more samples with Se concentrations greater than 5 µg/L is indicative of impairment of the reach. If the total number of samples is greater than 20, then a reach is considered to be impaired when at least ten percent of the samples have Se concentrations greater than 5 µg/L. Therefore, no single sample can be used to assess attainment status; evaluation of multiple data points is imperative for accurate attainment status.

5. Summary

West Virginia's current aquatic life standards for Se are based on recommendations from the EPA's 1987 Se criteria document (EPA 1987). These standards are not based on laboratory-derived toxicity data and do not represent the current state of the science. Tissue-based Se criteria are the most toxicologically and ecologically relevant, and represent the best science. Tissue-based Se criteria are protective of aquatic life and should be implemented in West Virginia.

Thus, we recommend that West Virginia adopt the following acute and chronic Se standards:

Acute

Acute = 1/[f1/CMC1)+(f2/CMC2)]

Where:

f1 and f2 are the fraction of total Se that are comprised as selenite (Se^{+4}) and selenate (Se^{+6}), respectively

CMC1 = 258 µg/L for selenite
CMC2 = $e^{(0.5812[\ln(\text{sulfate})] + 3.357)}$ for selenate

Chronic

Chronic Tiered Standards:

Initial screening: 5 µg/L water column
Follow-up screening: 9.0 µg/g (dw) whole-body tissue
19.5 µg/g (dw) egg/ovary tissue

6. References

Brix K.V., J.S. Volosin, W.J. Adams, R.J. Reash, R.G. Carlton, and D.O. McIntyre. 2001a. Effects of sulfate on the acute toxicity of selenate to freshwater organisms. *Environmental Toxicology and Chemistry* 20:1037-1045.

Brix, K.V., W.J. Adams, R.J. Reash, R.C. Carlton, and D.O. McIntyre. 2001b. Acute toxicity of selenate on two daphnids and three gammarid amphipods. *Environmental Toxicology and Chemistry* 16: 142-150.

Brix, K and D. De Forest. 2008. Selenium. In: Relevance of ambient water quality criteria in ephemeral and effluent-dependent watercourses of the arid western US (Gensemer RW, Meyerhoff RD, Ramage KJ, Curely EF, eds.). Pensacola, FL:SETAC Press, pp. 123-172. Canton, S.P. 1999. Acute aquatic life criteria for selenium. *Environmental Toxicology and Chemistry* 18:1425-1432.

Canton, S.P. 1999. Acute aquatic life criteria for selenium. *Environmental Toxicology and Chemistry* 18:1425-1432.

Carolina Power & Light (CP&L). 1997. Largemouth Bass Selenium Bioassay- Report. Carolina Power & Light Company, Environmental Services Section, NC. December 1997.

Chapman P.M., W.J. Adams, M.L. Brooks, C.G. Delos, S.N. Luoma, W.A Maher, H.M. Ohlendorf, T.S. Presser, and D.P. Shaw. 2009. Ecological Assessment of Selenium in the Aquatic Environment: Summary of a SETAC Pellston Workshop. Pensacola FL (USA): Society of Environmental Toxicology and Chemistry (SETAC).Coyle, J.J., D.R. Buckler, C.G. Ingesoll, J.F. Fairchild, and T.W. May. 1993. Effect of dietary selenium on the reproductive success of bluegills *Lepomis macrochirus*. *Environmental Toxicology and Chemistry* 12(3):551-565.

Coyle, J.J., D.R. Buckler, C.G. Ingersoll, J.F. Fairchild, and T.W. May. 1993. Effect of dietary selenium on the reproductive success of bluegills *Lepomis macrochirus*. *Environmental Toxicology and Chemistry* 12(3):551-565.

Doroshov, S., J.V. Eenennaam, C. Alexander, E. Hallen, H. Bailey, K. Kroll, and C. Restreppo. 1992. Development of water quality criteria for resident aquatic species of the San Joaquin River. Draft final report to the California State Water Resources Control Board for Contract No. 7-197-250-0. Department of Animal Science, University of California, Davis, CA.

Faust, S.D., and O.M. Aly. 1981. *Chemistry of Natural Waters*. Ann Arbor Science, Ann Arbor, MI. pp. 359-371.

Formation Environmental. 2011. Yellowstone cutthroat trout laboratory reproduction studies conducted in support of development of a site-specific selenium criterion. Draft. Prepared for J.R. Simplot Company. Pocatello (ID):Smoky Canyon Mine.

GEI Consultants, Inc. (GEI). 2008. Maternal transfer of selenium in fathead minnows, with modeling of ovary tissue-to-whole body concentrations. Technical Report prepared on behalf of Conoco-Phillips.

GEI Consultants, Inc. (GEI). 2014a. Review of EPA 2014 draft Se criteria document EPA 822-P-14-001. Submitted to EPA Docket ID No. EPA-HQ-OW-2004-0019 on behalf of the National Mining Association.

GEI Consultants, Inc. (GEI). 2014b. Supplemental comments on EPA 2014 draft Se criteria document EPA 822-P-14-001. Submitted to EPA Docket ID No. EPA-HQ-OW-2004-0019 on behalf of the National Mining Association.

GEI Consultants, Inc., Golder Associates, Parametrix, and University of Saskatchewan Toxicology Centre. 2008. Selenium tissue thresholds: tissue selection criteria, threshold development endpoints, and field application of tissue thresholds. Report to: North American Metals Council-Selenium Working Group, Washington, DC. http://namc.org/docs/00043675.PDF.

Hamilton, S.J., K.J. Buhl, N.L. Faerber, R.H. Wiedmeyer, and F.A. Bullard. 1990. Toxicity of organic selenium in the diet to Chinook salmon. *Environmental Toxicology and Chemistry* 9(3):347-358.

Hermanutz, R.O., K.N. Allen, T.H. Roush, and S.F. Hedtke. 1992. Effects of elevated Se concentrations on bluegills (*Lepomis macrochirus*) in outdoor experimental streams. *Environmental Toxicology and Chemistry* 11:217-224.

Hermanutz, R.O., K.N. Allen, N.E. Detenbeck, and C.E. Stephan. 1996. Exposure of bluegill (*Lepomis macrochirus*) to selenium in outdoor experimental streams. U.S. EPA Report. Mid-Continent Ecology Division. Duluth, MN.

Holm, J. 2002. Sublethal effects of selenium in rainbow trout (*Oncorhynchus mykiss*) and brook trout (*Salvelinus fontinalis*) [MA thesis]. Winnipeg (MB):Univ Manitoba.

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.

Holm J., V. Place, P. Siwik, G. Sterling, R. Evans, C. Baron, J. Werner, and K. Wautier. 2005. Developmental effects of bioaccumulated selenium in eggs and larvae of two salmonid species. *Environmental Toxicology and Chemistry* 24(9):2373-2381.

Lemly, A.D. 1993. Metabolic stress during winter increases the toxicity of selenium to fish. *Aquatic Toxicology* (Amsterdam) 27(1-2):133-158.

Lemly, A.D. 1997. Ecosystem recovery following selenium contamination in a freshwater reservoir. *Ecotoxicology and Environmental Safety* 36:275-281.

Lemly, A.D. 2002. *Selenium Assessment in Aquatic Ecosystems: A Guide for Hazard Evaluation and Water Quality Criteria.* Springer-Verlag, NY.

Luoma, S.N., C. Johns, N.S. Fisher, N.A. Steinberg, R.S. Oremland, and J.R. Reinfelder. 1992. Determination of selenium bioavailability to a benthic bivalve from particulate and solute pathways. *Environmental Science and Technology* 26:485-491.

Muscatello, J.R., P.M. Bennett, K.T. Himbeault, A.M. Belknap, and D.M. Janz. 2006. Larval deformities associated with selenium accumulation in northern pike (*Esox lucius*) exposed to metal mining effluent. *Environmental Science and Technology* 40:6506-6512.

North American Metals Council-Selenium Working Group (NAMC-SWG). 2014. Comments of the North American Metals Council-Selenium Working Group in Response to EPA's Draft National Recommended Aquatic Life Criterion for the Pollutant Selenium. Submitted to Docket ID EPA-HQ-OW-2004-0019.

Payne, R.G. 2013. Update to Kentucky Water Quality Standards for Protection of Aquatic Life: Acute Selenium Criterion and Tissue-Based Selenium Chronic Criteria. Kentucky Cabinet for Energy and Environment, Department of Environmental Protection, Division of Water, Frankfort, KY.

Ralston, N.V.C., Unrine, J. and D. Wallschlager. 2008. Biogeochemisty and Analysis of Selenium and its Species. Report Prepared for the North American Metals Council – Selenium Working Group, Washington, DC. http://namc.org/docs/00043673.PDF.

Saiki, M.K., B.A. Martin, and T.M. May. 2004. Reproductive status of western mosquitofish inhabiting selenium-contaminated waters in the grassland water district, Merced County, California. *Archives of Environmental Contamination and Toxicology* 47:363-369.

Schultz, R., and R. Hermanutz. 1990. Transfer of toxic concentrations of selenium form parent to progeny in the fathead minnow. *Bulletin of Environmental Contamination and Toxicology* 45:568-573.

Staub, B.P., W.A. Hopkins, J. Novak, and J.D. Congdon. 2004. Respiratory and reproductive characteristics of eastern mosquitofish (*Gambusia holbrooki*) inhabiting a coal ash settling basin. *Archives of Environmental Contamination and Toxicology* 46:96-101.

Stephan, C.E., D.I. Mount, D.J. Hansen, J.H. Gentile, G.A. Chapman, and W.A. Brungs. 1985. Guidelines for deriving numerical national water quality criteria for the protection of aquatic organisms and their uses. PB85-227049. National Technical Information Service, Springfield, VA.

United States Environmental Protection Agency (EPA). 1976. Quality Criteria for Water. PB-263943 or EPA-440/9-76-023. National Technical Information Service, Springfield, VA.

United States Environmental Protection Agency (EPA). 1980. Ambient Water Quality Criteria for Selenium. EPA-440/5-80-070. National Technical Information Service, Springfield, VA.

United States Environmental Protection Agency (EPA). 1987. Ambient Water Quality Criteria for Selenium. EPA-440/5-87-006. National Technical Information Service.

United States Environmental Protection Agency (EPA). 1995. Ambient Water Quality Criteria for Selenium.

United States Environmental Protection Agency (EPA). 2002. Draft Aquatic Life Water Quality Criteria for Selenium. February 2002. Office of Water, Washington, DC.

U.S. Environmental Protection Agency (EPA). 2004. Draft Aquatic Life Water Quality Criteria for Selenium - 2004. EPA-822-D-04-001. Office of Water, Washington, DC.

U.S. Environmental Protection Agency (EPA). 2012. National Recommended Water Quality Criteria. Office of Water. http://water.epa.gov/scitech/swguidance/standards/criteria/current/index.cfm#L.

United States Environmental Protection Agency (EPA). 2014. External peer review draft aquatic life ambient water quality criterion for selenium—freshwater 2014. Federal Register, Vol. 79, No. 93. pp. 27601-27604. http://www.gpo.gov/fdsys/pkg/FR-2014-05-14/html/2014-11080.htm.

West Virginia Department of Environmental Protection (WVDEP). 2011. Title 47 Legislative Rule Series 2 Requirements Governing Water Quality Standards. Effective June 27, 2011. West Virginia Department of Environmental Protection, Charleston, WV.

West Virginia Department of Environmental Protection (WVDEP). 2012. 2012 Draft West Virginia Integrated Water Quality Monitoring and Assessment Report. West Virginia Department of Environmental Protection, Charleston, WV.

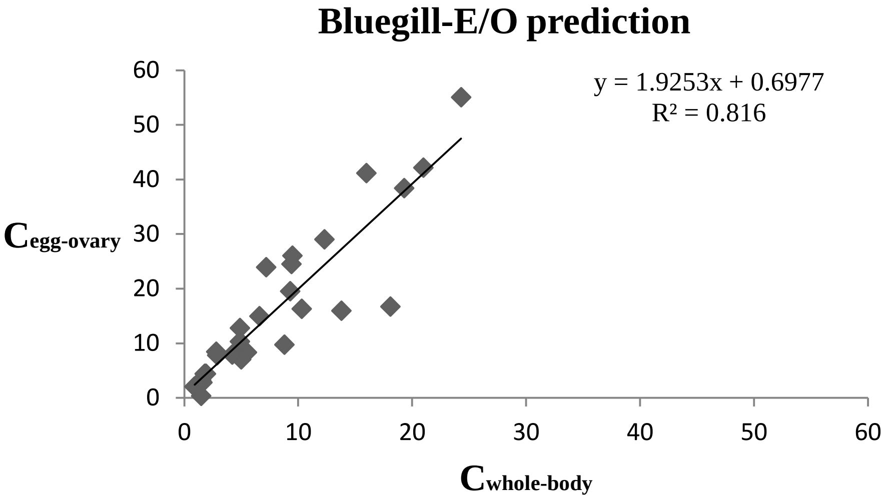
West Virginia Department of Environmental Protection (WVDEP). 2015. Fishes of West Virginia. West Virginia Department of Environmental Protection, Charleston, WV. http://www.dep.wv.gov/WWE/getinvolved/sos/Pages/Fishes.aspx.

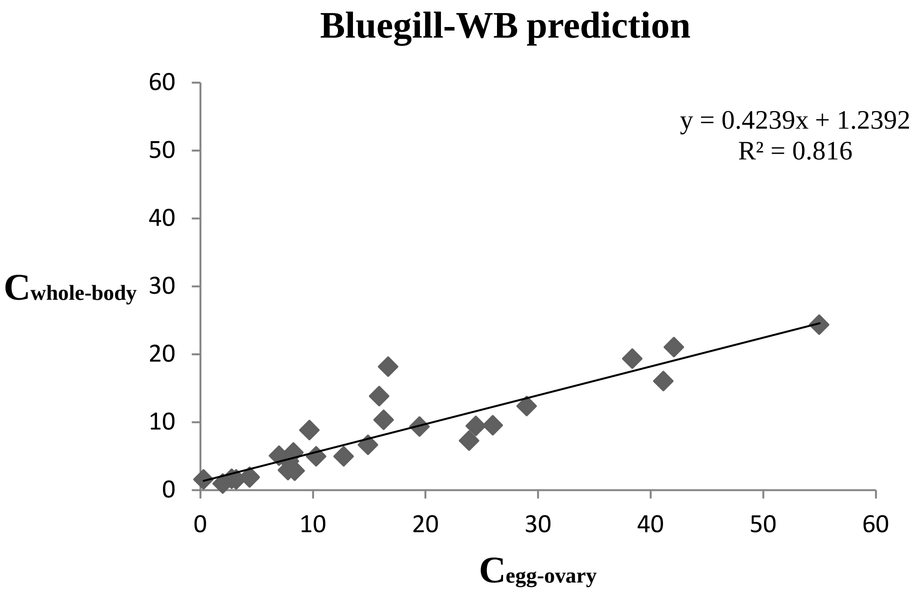
Appendix A Conversion Factor Calculations

Bluegill (*Lepomis macrochirus*)

Study	$C_{whole\text{-}body}$	C_{egg}	C_{ovary}	$C_{egg\text{-}ovary}$	Ratio
Coyle et al. 1993	0.90	1.90	2.10	2.00	2.22
Coyle et al. 1993	2.90	7.30	8.30	7.80	2.69
Coyle et al. 1993	4.90	13.00	12.50	12.75	2.60
Coyle et al. 1993	7.20	22.80	25.00	23.90	3.32
Coyle et al. 1993	16.00	41.30	41.00	41.15	2.57
Doroshov et al. 1992	1.60	2.80	-	2.80	1.75
Doroshov et al. 1992	5.50	8.30	-	8.30	1.51
Doroshov et al. 1992	9.30	19.50	-	19.50	2.10
Doroshov et al. 1992	19.30	38.40	-	38.40	1.99
Hermanutz et al. 1996	1.50	-	0.30	0.30	0.20
Hermanutz et al. 1996	18.10	-	16.70	16.70	0.92
Hermanutz et al. 1996	1.90	-	4.40	4.40	2.32
Hermanutz et al. 1996	2.80	-	8.40	8.40	3.00
Hermanutz et al. 1996	12.30	-	29.00	29.00	2.36
Hermanutz et al. 1996	9.40	-	24.50	24.50	2.61
Hermanutz et al. 1996	1.50	-	3.20	3.20	2.13
Hermanutz et al. 1996	4.90	-	10.30	10.30	2.10
Hermanutz et al. 1996	21.00	-	42.10	42.10	2.00
Hermanutz et al. 1996	24.30	-	55.00	55.00	2.26
Hermanutz et al. 1996	5.00	-	7.00	7.00	1.40
Hermanutz et al. 1996	9.50	-	26.00	26.00	2.74
Hermanutz et al. 1996	6.60	-	14.90	14.90	2.26
Hermanutz et al. 1996	1.80	-	4.40	4.40	2.44
Hermanutz et al. 1996	4.20	-	7.90	7.90	1.88
Hermanutz et al. 1996	10.30	-	16.30	16.30	1.58
Hermanutz et al. 1996	13.80	-	15.90	15.90	1.15
Osmundson et al. 2007	8.80	-	9.70	9.70	1.10

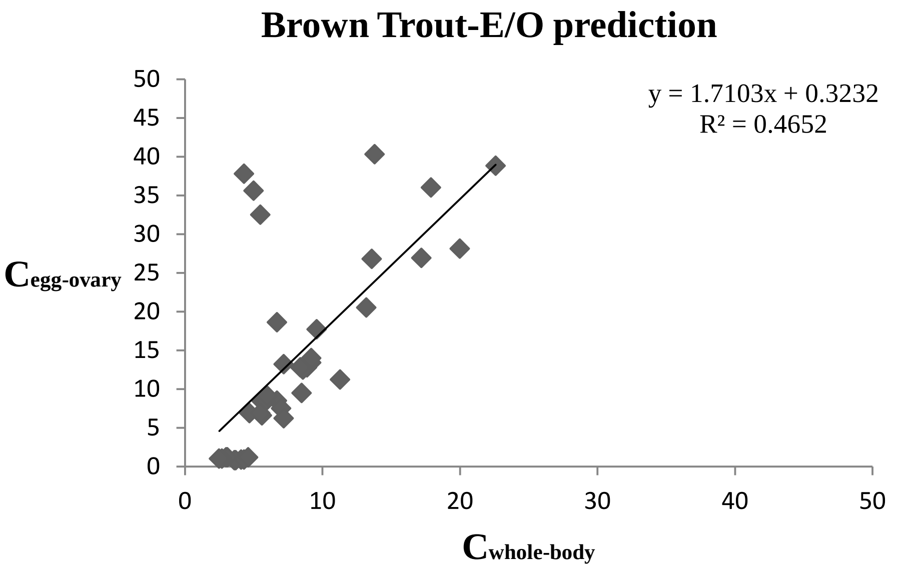
Median Ratio:	2.13
Geomean:	1.86



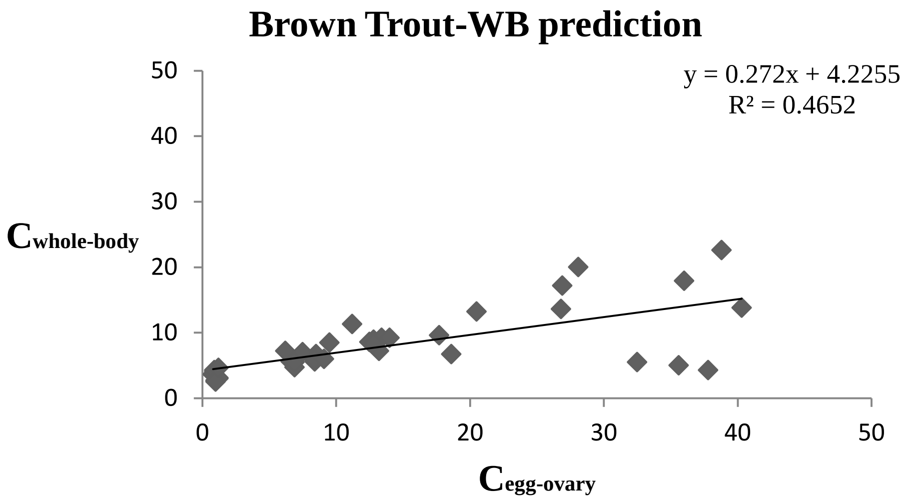

Bluegill-WB prediction
y = 0.4239x + 1.2392
R² = 0.816
$C_{whole\text{-}body}$
$C_{egg\text{-}ovary}$
0
10
20
30
40
50
60
0
10
20
30
40
50
60

Brown trout (*Salmo trutta*)

Study	$C_{whole-body}$	C_{egg}	C_{ovary}	$C_{egg-ovary}$	Ratio
NewFields 2009	3.60	0.80	-	0.80	0.22
NewFields 2009	4.10	0.90	-	0.90	0.22
NewFields 2009	3.70	0.80	-	0.80	0.22
NewFields 2009	4.30	0.90	-	0.90	0.21
NewFields 2009	3.00	1.20	-	1.20	0.40
NewFields 2009	3.10	1.20	-	1.20	0.39
NewFields 2009	2.70	1.00	-	1.00	0.37
NewFields 2009	2.50	1.00	-	1.00	0.40
NewFields 2009	8.90	12.80	-	12.80	1.44
NewFields 2009	13.80	40.30	-	40.30	2.92
NewFields 2009	17.90	36.00	-	36.00	2.01
NewFields 2009	13.60	26.80	-	26.80	1.97
NewFields 2009	17.20	26.90	-	26.90	1.56
NewFields 2009	6.70	18.60	-	18.60	2.78
NewFields 2009	9.60	17.70	-	17.70	1.84
NewFields 2009	22.60	38.80	-	38.80	1.72
NewFields 2009	7.20	13.20	-	13.20	1.83
NewFields 2009	9.20	13.40	-	13.40	1.46
NewFields 2009	13.20	20.50	-	20.50	1.55
NewFields 2009	8.60	12.50	-	12.50	1.45
NewFields 2009	11.30	11.20	-	11.20	0.99
NewFields 2009	20.00	28.10	-	28.10	1.41
NewFields 2009	8.40	12.80	-	12.80	1.52
NewFields 2009	5.60	8.40	-	8.40	1.50
NewFields 2009	6.70	8.50	-	8.50	1.27
NewFields 2009	5.90	8.40	-	8.40	1.42
NewFields 2009	6.00	9.10	-	9.10	1.52
NewFields 2009	7.00	7.50	-	7.50	1.07
NewFields 2009	5.60	6.60	-	6.60	1.18
NewFields 2009	4.70	6.90	-	6.90	1.47
NewFields 2009	7.20	6.20	-	6.20	0.86
NewFields 2009	9.20	14.00	-	14.00	1.52
NewFields 2009	5.50	6.90	-	6.90	1.25
NewFields 2009	8.50	9.50	-	9.50	1.12
Osmundson et al. 2007	4.60	-	1.20	1.20	0.26
Osmundson et al. 2007	4.30	-	37.80	37.80	8.79
Osmundson et al. 2007	5.00	-	35.60	35.60	7.12
Osmundson et al. 2007	5.50	-	32.50	32.50	5.91

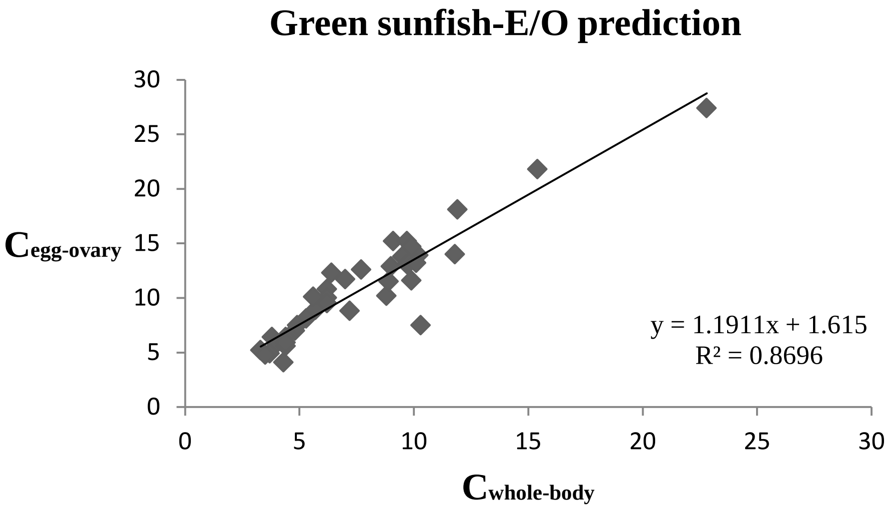


Median ratio:	1.45
Geomean:	1.15

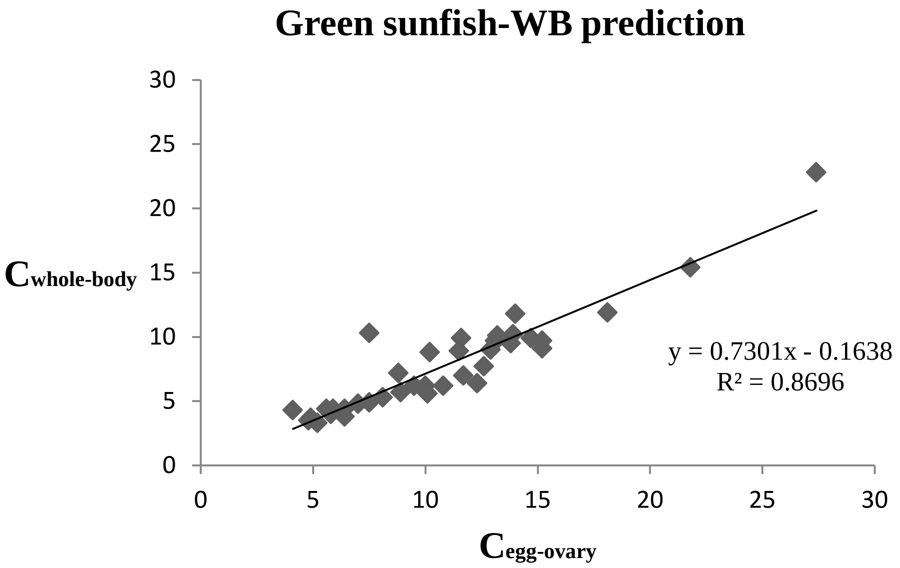


Green sunfish (*Lepomis cyanellus*)

Study	**$C_{whole\text{-}body}$**	**C_{egg}**	**C_{ovary}**	**$C_{egg\text{-}ovary}$**	**Ratio**
Osmundson et al. 2007	22.80	-	27.40	27.40	1.20
Osmundson et al. 2007	8.80	-	10.20	10.20	1.16
Osmundson et al. 2007	15.40	-	21.80	21.80	1.42
Osmundson et al. 2007	4.80	-	7.00	7.00	1.46
Osmundson et al. 2007	5.70	-	8.90	8.90	1.56
Osmundson et al. 2007	4.40	-	6.40	6.40	1.45
Osmundson et al. 2007	3.80	-	6.40	6.40	1.68
Osmundson et al. 2007	11.90	-	18.10	18.10	1.52
Osmundson et al. 2007	6.40	-	12.30	12.30	1.92
Osmundson et al. 2007	9.50	-	13.80	13.80	1.45
Osmundson et al. 2007	9.10	-	15.20	15.20	1.67
Osmundson et al. 2007	6.20	-	10.80	10.80	1.74
Osmundson et al. 2007	7.00	-	11.70	11.70	1.67
Osmundson et al. 2007	7.70	-	12.60	12.60	1.64
Osmundson et al. 2007	6.20	-	10.00	10.00	1.61
Osmundson et al. 2007	10.20	-	13.90	13.90	1.36
Osmundson et al. 2007	9.70	-	15.20	15.20	1.57
Osmundson et al. 2007	9.90	-	14.70	14.70	1.48
Osmundson et al. 2007	7.20	-	8.80	8.80	1.22
Osmundson et al. 2007	9.00	-	12.90	12.90	1.43
Osmundson et al. 2007	9.70	-	13.10	13.10	1.35
Osmundson et al. 2007	8.90	-	11.50	11.50	1.29
Osmundson et al. 2007	9.80	-	13.20	13.20	1.35
Osmundson et al. 2007	9.90	-	11.60	11.60	1.17
Osmundson et al. 2007	10.30	-	7.50	7.50	0.73
Osmundson et al. 2007	5.30	-	8.10	8.10	1.53
Osmundson et al. 2007	10.10	-	13.20	13.20	1.31
Osmundson et al. 2007	11.80	-	14.00	14.00	1.19
Osmundson et al. 2007	3.30	-	5.20	5.20	1.58
Osmundson et al. 2007	4.00	-	5.80	5.80	1.45
Osmundson et al. 2007	4.30	-	4.10	4.10	0.95
Osmundson et al. 2007	3.70	-	4.90	4.90	1.32
Osmundson et al. 2007	6.20	-	9.50	9.50	1.53
Osmundson et al. 2007	3.50	-	4.80	4.80	1.37
Osmundson et al. 2007	4.40	-	5.60	5.60	1.27
Osmundson et al. 2007	5.60	-	10.10	10.10	1.80
Osmundson et al. 2007	4.90	-	7.50	7.50	1.53
Osmundson et al. 2007	4.40	-	5.90	5.90	1.34

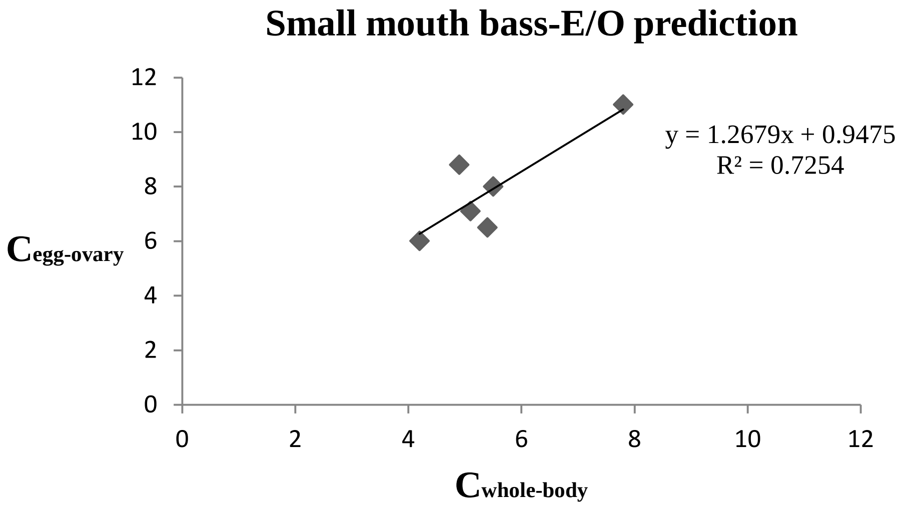


Median ratio:	1.45
Geomean	1.41

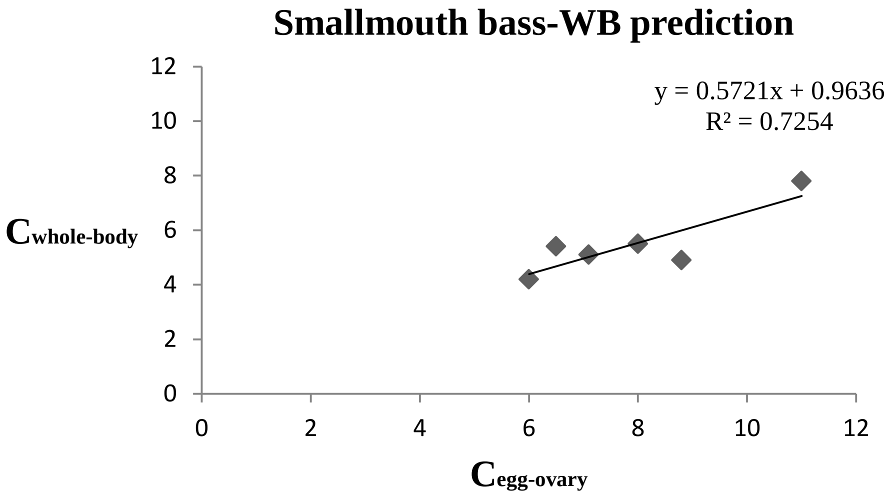


Smallmouth bass (*Micropterus dolomieu*)

Study	**$C_{whole-body}$**	**C_{egg}**	**C_{ovary}**	**$C_{egg-ovary}$**	**Ratio**
Osmundson et al. 2007	4.20	-	6.00	6.00	1.43
Osmundson et al. 2007	5.50	-	8.00	8.00	1.45
Osmundson et al. 2007	5.40	-	6.50	6.50	1.20
Osmundson et al. 2007	7.80	-	11.00	11.00	1.41
Osmundson et al. 2007	5.10	-	7.10	7.10	1.39
Osmundson et al. 2007	4.90	-	8.80	8.80	1.80


Small mouth bass-E/O prediction

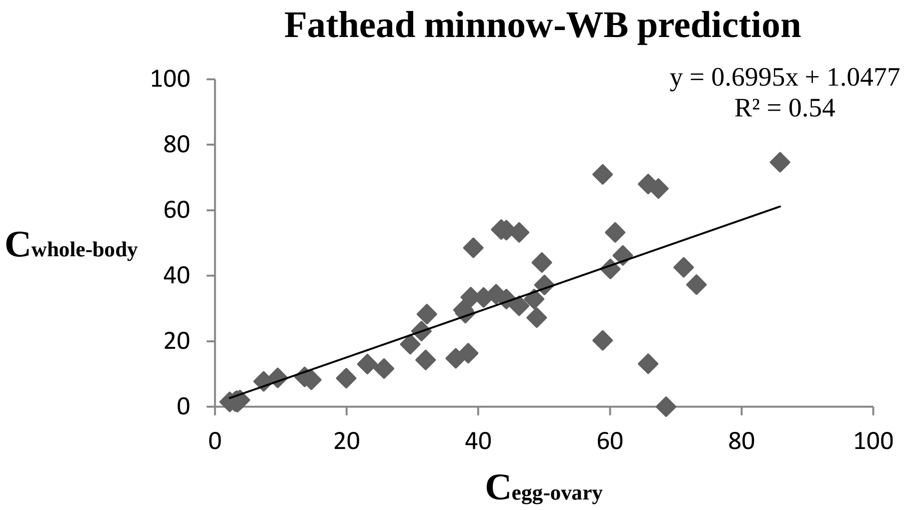
Median ratio: 1.42
Geomean: 1.44


Smallmouth bass-WB prediction

Fathead minnow (*Pimephales promelas*)

Study	$C_{whole\text{-}body}$	C_{egg}	C_{ovary}	$C_{egg\text{-}ovary}$	Ratio
GEI 2008	2.04	-	3.81	3.81	1.87
GEI 2008	1.39	-	2.23	2.23	1.60
GEI 2008	1.85	-	3.31	3.31	1.79
GEI 2008	1.32	-	3.43	3.43	2.60
GEI 2008	1.55	-	3.08	3.08	1.99
GEI 2008	37.13	-	50.06	50.06	1.35
GEI 2008	29.54	-	37.77	37.77	1.28
GEI 2008	33.32	-	40.82	40.82	1.23
GEI 2008	28.26	-	32.23	32.23	1.14
GEI 2008	30.74	-	46.21	46.21	1.50
GEI 2008	53.17	-	60.84	60.84	1.14
GEI 2008	48.52	-	39.28	39.28	0.81
GEI 2008	53.81	-	44.28	44.28	0.82
GEI 2008	53.20	-	46.21	46.21	0.87
GEI 2008	54.01	-	43.51	43.51	0.81
GEI 2008	12.93	-	23.18	23.18	1.79
GEI 2008	8.19	-	14.67	14.67	1.79
GEI 2008	14.25	-	32.04	32.04	2.25
GEI 2008	8.65	-	19.95	19.95	2.31
GEI 2008	16.33	-	38.51	38.51	2.36
GEI 2008	7.69	-	7.39	7.39	0.96
GEI 2008	19.05	-	29.69	29.69	1.56
GEI 2008	8.78	-	9.55	9.55	1.09
GEI 2008	14.68	-	36.58	36.58	2.49
GEI 2008	9.02	-	13.63	13.63	1.51
GEI 2008	46.17	-	61.99	61.99	1.34
GEI 2008	41.97	-	60.07	60.07	1.43
GEI 2008	34.33	-	42.74	42.74	1.24
GEI 2008	33.40	-	38.89	38.89	1.16
GEI 2008	42.53	-	71.24	71.24	1.68
GEI 2008	74.56	-	85.87	85.87	1.15
GEI 2008	67.94	-	65.85	65.85	0.97
GEI 2008	70.85	-	58.91	58.91	0.83
GEI 2008	43.93	-	49.67	49.67	1.13
GEI 2008	66.57	-	67.39	67.39	1.01
GEI 2008	20.21	-	58.91	58.91	2.91
GEI 2008	13.08	-	65.85	65.85	5.03
GEI 2008	23.02	-	31.38	31.38	1.36
GEI 2008	11.55	-	25.72	25.72	2.23
GEI 2008	NA	-	68.54	68.54	
GEI 2008	32.80	-	48.52	48.52	1.48
GEI 2008	27.17	-	48.90	48.90	1.80
GEI 2008	28.54	-	38.04	38.04	1.33

GEI 2008	*37.20*	-	*73.16*	*73.16*	*1.97*
GEI 2008	*32.79*	-	*44.28*	*44.28*	*1.35*

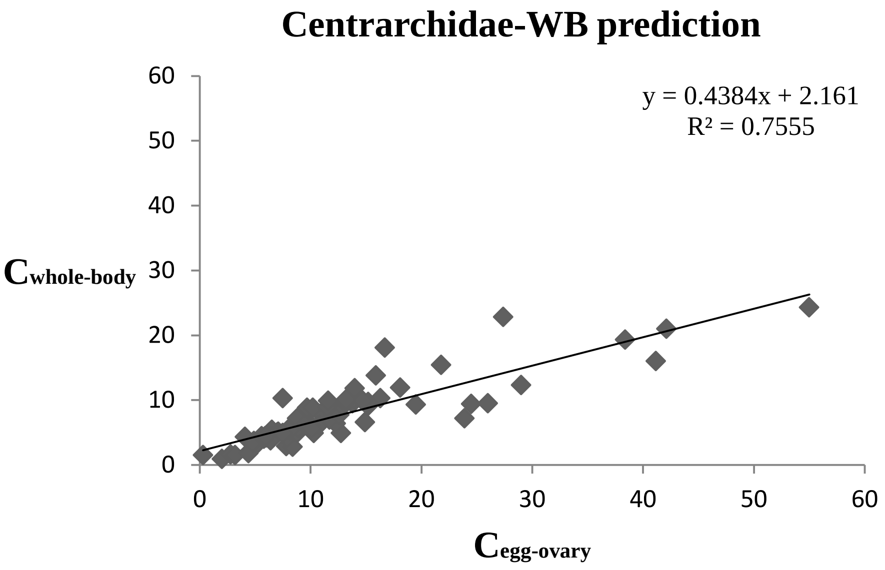


Median ratio:	1.3972
Geomean:	1.4751

Centrarchidae

Study	$C_{whole\text{-}body}$	C_{egg}	C_{ovary}	$C_{egg\text{-}ovary}$	Ratio
Coyle et al. 1993	0.90	1.90	2.10	2.00	2.22
Coyle et al. 1993	2.90	7.30	8.30	7.80	2.69
Coyle et al. 1993	4.90	13.00	12.50	12.75	2.60
Coyle et al. 1993	7.20	22.80	25.00	23.90	3.32
Coyle et al. 1993	16.00	41.30	41.00	41.15	2.57
Doroshov et al. 1992	1.60	2.80	-	2.80	1.75
Doroshov et al. 1992	5.50	8.30	-	8.30	1.51
Doroshov et al. 1992	9.30	19.50	-	19.50	2.10
Doroshov et al. 1992	19.30	38.40	-	38.40	1.99
Hermanutz et al. 1996	1.50	-	0.30	0.30	0.20
Hermanutz et al. 1996	18.10	-	16.70	16.70	0.92
Hermanutz et al. 1996	1.90	-	4.40	4.40	2.32
Hermanutz et al. 1996	2.80	-	8.40	8.40	3.00
Hermanutz et al. 1996	12.30	-	29.00	29.00	2.36
Hermanutz et al. 1996	9.40	-	24.50	24.50	2.61
Hermanutz et al. 1996	1.50	-	3.20	3.20	2.13
Hermanutz et al. 1996	4.90	-	10.30	10.30	2.10
Hermanutz et al. 1996	21.00	-	42.10	42.10	2.00
Hermanutz et al. 1996	24.30	-	55.00	55.00	2.26
Hermanutz et al. 1996	5.00	-	7.00	7.00	1.40
Hermanutz et al. 1996	9.50	-	26.00	26.00	2.74
Hermanutz et al. 1996	6.60	-	14.90	14.90	2.26
Hermanutz et al. 1996	1.80	-	4.40	4.40	2.44
Hermanutz et al. 1996	4.20	-	7.90	7.90	1.88
Hermanutz et al. 1996	10.30	-	16.30	16.30	1.58
Hermanutz et al. 1996	13.80	-	15.90	15.90	1.15
Osmundson et al. 2007	8.80	-	9.70	9.70	1.10
Osmundson et al. 2007	4.20	-	6.00	6.00	1.43
Osmundson et al. 2007	5.50	-	8.00	8.00	1.45
Osmundson et al. 2007	5.40	-	6.50	6.50	1.20
Osmundson et al. 2007	7.80	-	11.00	11.00	1.41
Osmundson et al. 2007	5.10	-	7.10	7.10	1.39
Osmundson et al. 2007	4.90	-	8.80	8.80	1.80
Osmundson et al. 2007	22.80	-	27.40	27.40	1.20
Osmundson et al. 2007	8.80	-	10.20	10.20	1.16
Osmundson et al. 2007	15.40	-	21.80	21.80	1.42
Osmundson et al. 2007	4.80	-	7.00	7.00	1.46
Osmundson et al. 2007	5.70	-	8.90	8.90	1.56
Osmundson et al. 2007	4.40	-	6.40	6.40	1.45
Osmundson et al. 2007	3.80	-	6.40	6.40	1.68
Osmundson et al. 2007	11.90	-	18.10	18.10	1.52
Osmundson et al. 2007	6.40	-	12.30	12.30	1.92
Osmundson et al. 2007	9.50	-	13.80	13.80	1.45

Osmundson et al. 2007	9.10	-	15.20	15.20	1.67
Osmundson et al. 2007	6.20	-	10.80	10.80	1.74
Osmundson et al. 2007	7.00	-	11.70	11.70	1.67
Osmundson et al. 2007	7.70	-	12.60	12.60	1.64
Osmundson et al. 2007	6.20	-	10.00	10.00	1.61
Osmundson et al. 2007	10.20	-	13.90	13.90	1.36
Osmundson et al. 2007	9.70	-	15.20	15.20	1.57
Osmundson et al. 2007	9.90	-	14.70	14.70	1.48
Osmundson et al. 2007	7.20	-	8.80	8.80	1.22
Osmundson et al. 2007	9.00	-	12.90	12.90	1.43
Osmundson et al. 2007	9.70	-	13.10	13.10	1.35
Osmundson et al. 2007	8.90	-	11.50	11.50	1.29
Osmundson et al. 2007	9.80	-	13.20	13.20	1.35
Osmundson et al. 2007	9.90	-	11.60	11.60	1.17
Osmundson et al. 2007	10.30	-	7.50	7.50	0.73
Osmundson et al. 2007	5.30	-	8.10	8.10	1.53
Osmundson et al. 2007	10.10	-	13.20	13.20	1.31
Osmundson et al. 2007	11.80	-	14.00	14.00	1.19
Osmundson et al. 2007	3.30	-	5.20	5.20	1.58
Osmundson et al. 2007	4.00	-	5.80	5.80	1.45
Osmundson et al. 2007	4.30	-	4.10	4.10	0.95
Osmundson et al. 2007	3.70	-	4.90	4.90	1.32
Osmundson et al. 2007	6.20	-	9.50	9.50	1.53
Osmundson et al. 2007	3.50	-	4.80	4.80	1.37
Osmundson et al. 2007	4.40	-	5.60	5.60	1.27
Osmundson et al. 2007	5.60	-	10.10	10.10	1.80
Osmundson et al. 2007	4.90	-	7.50	7.50	1.53
Osmundson et al. 2007	4.40	-	5.90	5.90	1.34



Median ratio:	1.53
Geomean:	1.57