External Peer Review of the Draft Aquatic Life Ambient Water Quality Criterion for Selenium – Freshwater 2014

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PEER REVIEW COMMENTS FROM

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Responses to Charge Questions by Dr. Kevin V. Brix

PART I: OVERARCHING QUESTIONS

1. Please comment on the overall clarity of the document and construction of the criterion statement with its multiple elements.

I found the overall clarity of the document to be good, although there are several specific areas that require clarification (detailed in comments to specific charge questions). I also found the construction of the criterion statement to be quite clear and logical.

2. EPA has developed a tiered selenium criterion with four elements, with the fish tissue elements having primacy over the water-column elements, and the egg-ovary element having primacy over any other element. Inclusion of the fish whole-body or fish muscle element into the selenium criterion ensures the protection of aquatic life when fish egg or ovary tissue measurements are not available, and inclusion of the water column elements ensures protection when fish tissue measurements are not available

    a. Please comment on the tiered construction of the selenium chronic criterion; is it logical, and scientifically defensible as it applies to protection of freshwater aquatic life:

        i. That is, is the primacy of the egg-ovary element over the other elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Yes, primacy of the egg-ovary element is sound and well supported by the scientific literature. EPA has cited all of the key references for support of this approach.

        ii. Is the primacy of the whole-body/fish muscle element over the water column elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Yes, in general a tissue-based criterion should have primacy over a water-based criterion for Se due to the complex site-specific nature for Se bioaccumulation. This is well documented in the literature. As discussed by EPA, an egg-ovary based criterion is highly desirable but may not always be achievable due to logistical constraints or the potentially significant impacts on populations of terminal sampling of ovaries for some threatened or endangered species. In such cases, whole body or muscle plugs provide a reasonable surrogate for the egg-ovary element. One item lacking from the WQC is guidance on when use of whole body or muscle elements is acceptable. Some questions that come to mind:

1.) Can WB or muscle elements be used instead of EO even when collection of EO samples is considered logistically and environmentally feasible?

2.) Are there seasonal considerations to use of WB and muscle samples? For example, is it acceptable to use WB or muscle samples collected in the Fall for a species that spawns in the Spring?

iii. Please comment on the scientific uncertainty that may be associated with this tiered approach? Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

There are of course a number of uncertainties in the tiered approach proposed by EPA. I provide specific comments on these uncertainties throughout this review. Overall though, I do not believe there are any currently available data sources, models or alternative approaches that EPA has not considered that would significantly reduce the uncertainty.

iv. Are the draft recommended magnitude, duration, and frequency for each criterion element scientifically sound and appropriate? Please provide detailed comments.

I have provided specific comments on these issues in response to the questions below.

PART II: FISH TISSUE CRITERION ELEMENTS DERIVATION: DERIVATION OF FISH EGG-OVARY, WHOLE BODY AND MUSCLE CRITERION ELEMENT(S)

EPA is requesting a technical review of the methods and procedures used to derive a chronic selenium criterion based on an egg-ovary concentration, as well as its translation to a criterion element applicable to whole-body and muscle tissue. Please address the following questions:

1. Please comment on EPA's use of the effects concentration 10^{th} centile (EC_{10}) as the measurement endpoint for the fish reproductive toxicity studies used to derive the egg-ovary element.

It is unclear to me why EPA has selected the EC10 as the measurement endpoint for these studies. EPA argues because it is a tissue-based criterion, the measure of exposure is less variable than might occur for a water-based criterion. I understand the point EPA is making and agree that a tissue-based criterion is more integrative of exposure than a water-based criterion. However, following this logic, EPA is then stating that for a chemical with a water-based criterion in a system where the exposure concentration is consistently above the EC10 (e.g., very stable at a concentration equivalent to the EC15) that it is not sufficiently protective.

It seems to me that the ECx selected should be based on the level of protection EPA intends to provide and that this is independent of variability in exposure. Variability in exposure is more appropriately addressed via averaging periods as EPA has done with the intermittent exposure element of the criterion. In fact, by considering both an intermittent exposure element and using an EC10, EPA is addressing the same issue twice.

Given the above, I do not believe EPA has provided a scientific rationale for use of the EC10 in a tissue-based criterion as providing an equivalent level of protection as an EC20 in a water-based criterion. I recommend EPA evaluate how use of the EC20 would affect the final criteria calculations. I suspect given the sharp dose-response relationships for Se, it will not dramatically change the final criteria calculations. Alternatively, if EPA now believes the EC10 is an appropriate level of protection for WQC, then this should be applied across chemicals.

2. Data used to derive the final chronic egg-ovary criterion element were differentiated based on the type of effect (reproductive vs. non-reproductive effects). Acceptable chronic toxicity data on fish reproductive effects are available for a total of nine fish genera. The genus Sensitivity Distribution (SD) is predominantly populated with data on fish genera because field evidence demonstrates that fish communities can be affected by selenium even when there is no observable change in the invertebrate community diversity and abundance. As a result, decades of aquatic toxicity research have focused primarily on fish. Available field and laboratory studies indicate that invertebrates are more tolerant to selenium than most of the tested fish species (Criteria document, Table 6c, Section 4.1.2). The data set used to derive the selenium criterion marks a change from the traditional method used to derive water quality criteria that requires toxicity tests with aquatic organisms from 8 phylogenetically distinct taxa (including three vertebrate and five invertebrate genera) in order to derive aquatic life criteria (Stephan et al., 1985).

    a. Given selenium's more taxon-specific and life stage-specific toxicity, please comment on EPA's use of the available data to derive the egg-ovary tissue element.

Overall, I found EPA's use of the available data to derive the egg-ovary element to be scientifically sound. However, see caveats in b and c below. I did find EPA's use of the data for *Gambusia* to be questionable. Given the variability in the EO:WB ratio across species and the complete lack of data on this ratio for ovovivaprous fish, the EO-based threshold for this genus is highly questionable. Given this uncertainty and that these are the only data used in the WQC calculation in which EO Se was not directly measured, in my opinion, data for this genus should not be used in the WQC calculation.

    b. Given the greater general sensitivity of oviparous fish to selenium compared to aquatic invertebrates, please comment on the appropriateness of EPA's fish tissue-based criterion for affording protection to the aquatic community as a whole (e.g., including invertebrates).

I agree with EPA that currently available data indicates oviparous fish are more sensitive than aquatic invertebrates to Se. However, it is important to note that there is a paucity of data for invertebrates. I agree with EPA's approach to translate available invertebrate data to an EO threshold for purposes of developing a species sensitivity distribution (SSD). However, I strongly disagree with the addition of 2 hypothetical crustaceans to the SSD. This is scientifically indefensible (just making up data) and the WQC calculation should be based only on taxa for which there are actually data available. By this logic, why add only 2 crustacean taxa, why not 3 or 5?

Note, EPA needs to include the data from Conley et al. (2011, 2013, and 2014) in its assessment of Se toxicity and trophic transfer to mayflies.

Overall, given the limited data, I think EPA has overstated the certainty with which we can conclude fish are more sensitive than invertebrates. All we can really say is that based on a relatively small data set, available data suggests the tissue based WQC will be protective of invertebrates.

    c. With respect to the tests that quantified non-reproductive effects, did the EPA use that data to the best extent possible given its limitations (e.g., relevance compared to reproductive tests, and data quality concerns which increased uncertainty (e.g., Hamilton et al., 1990)?

I agree with EPA, that generally, the reproductive endpoint is more sensitive than other endpoints such as juvenile growth. However, in the case of salmonids, there is at least some evidence (e.g., Hamilton et al.,

1990) that juvenile growth is comparable in sensitivity to reproduction. It is also worth pointing out that these studies did not include pre-exposure of the parents and subsequent maternal transfer, so it is possible that exposure and subsequent effects on juvenile growth have been underestimated. Further, juvenile salmonids have a much more limited home range and potentially higher intensity of exposure if they rear in Se contaminated areas compared to adult salmonids (particularly migratory species). Given this, it is unclear to me that placing primacy on the egg-ovary element will necessarily be protective of these species. EPA should consider the potential that juvenile whole body Se concentrations for migratory salmonids may need primacy or at least concurrent compliance monitoring to ensure the protection of these important species.

d. EPA also rejected studies that used the injection route of exposure for selenium due to uncertainty related to uptake, distribution and metabolism/transformation kinetics when compared with the dietary and/or maternal transfer routes of exposure. Was this reasonable? Does the panel envision an appropriate and scientifically defensible use for this type of data? Please provide detailed comments.

Yes, it was reasonable to reject these studies for the reasons stated by EPA. In my opinion, there is currently insufficient information to have confidence that injection studies replicate realistic environmental exposures with respect to Se homeostasis. Indeed, the fact that the catfish study resulted in such an unusually low effect level suggests there may be different processes occurring in these types of studies. EPA has adequately documented that catfish do no appear to be uniquely sensitive based on available field abundance data in Se-impacted systems, counter to the lab-based injection study.

3. Was the method (Section 4.1.5, 7.1.7) used to translate the fish egg-ovary criterion element into muscle and whole body criterions elements understandable, transparent and scientifically defensible? Was there sufficient data for making the translations for each element?

Yes, I found the egg-ovary to muscle and whole body translations to be understandable and scientifically defensible.

PART III: EVALUATION OF THE TRANSLATION PROCEDURE TO DERIVE THE WATER COLUMN ELEMENT(S)

EPA is also requesting a technical review of the methods and procedures used to translate the egg-ovary element of the chronic selenium criterion to water-column elements. Relevant sections of the document include:

- A description of the method used to derive an equation to translate the egg-ovary element to a monthly water-column element in perennial (lentic and lotic) waters and an equation that can be used to convert the monthly water-column element to an intermittent water column element (Sections 3.8.3, 3.8.4, 4.2.1, 4.3, and Appendix G).

- An analysis of the translation equation precision using data obtained from published literature (Sections 7.2.1, 7.2.2, and Appendix H).

- A description of the method and data sources used to derive the translation equation parameters (Sections 4.2.2, 4.2.3, and Appendix B).

- A description of the method and data sources used to categorize waterbody types where a single water-column chronic criterion concentration value would be adequately protective in most circumstances (Section 4.2.4).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for established categories of waters (Section 4.2.5).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for intermittent discharges that may occur in lentic and lotic waterbodies (Section 4.3).

Please address the following questions:

1. Please comment on the scientific defensibility of EPA's translation equation method for translating the concentration of selenium in fish tissue to a concentration of selenium in the water-column. Please comment on major sources of uncertainty in applying the translation equation to different types of waterbodies (e.g., with differing retention times, water chemistries, and/or species present). Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

I appreciate that EPA is dealing with a very difficult issue in terms of translating a tissue-based criteria to water for routine monitoring and screening purposes. I agree with the general conceptual model EPA has developed for making this translation. Having said that, the details of how EPA has implemented this conceptual model I think are very problematic. My concerns center on two major themes – compounding multiple uncertain values in the food chain transfer models and lack of transparency on what level of protection the proposed water elements provide.

I am very concerned that EPA is placing too much value on extrapolated and modeled values. The translation approach involves building food chain models for 69 sites that in many cases have significant data gaps (e.g., dietary composition, extrapolated TTFs, extrapolated CFs, etc.). To address these uncertainties, EPA developed a series of protocols for filling in the data gaps (e.g., using TTFs for species in the same order). While I appreciate the logic and largely agree with these protocols, ultimately, information derived in this manner is not measured data. This approximated information is then used in a very quantitative manner for setting the water-based WQC. Figure 11 in particular I find very misleading. How many of the data points in those two distributions (lotic and lentic) are based on sites where all parameters in the food chain models were actually measured? I did not take the time to calculate this, but EPA must explicitly provide this information. I suspect the percentage will be quite low. What do these distributions in Figure 11 look like if based on only studies where all parameters were directly measured? In my view, use of such data provides a potentially very inaccurate picture of what we actually know about the distribution of waterborne Se concentrations associated with the tissue-based WQC. This seems to be a significant departure from previous WQC criteria derivation processes where if data for a particular study were insufficient, the study was simply excluded and the resulting uncertainty from having relatively few complete data sets was reflected in a lower WQC (e.g., a WQC less than the most sensitive taxa tested if $n<20$).

An important element of previous WQC was transparency in the level of protection being provided (e.g., 95 % of taxa) and the assumptions underlying that protection (e.g., that tested taxa were representative of aquatic communities in the US). It is entirely unclear to me what level of protection is being provided by

the water element of proposed WQC. The proposed water-based WQC is based on the 20th percentile for lotic and lentic sites that were modeled (see concerns about this in the previous paragraph). But even this is not correct, because for some sites, multiple fish species were modeled per site. This raises numerous questions regarding independence of values in the distribution, whether the sites evaluated are biased towards those with known Se issues, etc. EPA has also not made it clear why protection of 80% of sites is a desirable regulatory objective. Why not 70%, 90%, or 95%? I appreciate that EPA has undertaken a ground truthing exercise to evaluate the proposed water element WQC. However, it is unclear exactly how EPA undertook this analysis. Were there truly over 3,000 independent sites that EPA evaluated? If this exercise concluded that <10% of sites would result in false negatives, then what does this say about the representativeness of the 69 sites and what is the real level of protection being provided?

2. Regarding the trophic transfer factor (*TTF)* values, did EPA use a scientifically defensible method to derive the *TTF* values (p. 71-77 of the criteria document)? Were the exclusion criteria, (pp. 71-77 of the criteria document) developed by EPA to screen the available data applied in a consistent and scientifically defensible manner? In particular, EPA noticed that application of the exclusion criteria resulted in *TTF* values for aquatic insect larvae that differ from other published values. Given this, are you aware of any other methods of screening data that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included, if appropriate? Please provide detailed comments.

In general, EPA has used a scientifically defensible method to derived TTFs. However, I am concerned that the TTFs derived from field data by EPA are biased low and potentially not protective. I note that the data in Figure 16 appear to show a rather significant bias towards underprediction of EO selenium concentrations, consistent with this concern. As recognized by EPA, there is typically an inverse relationship between the exposure concentration and the TTF such that low dietary Se will result in relatively high TTFs for a given predator-prey species pair. Many of the field data sets used by EPA are from sites with high levels of Se contamination (10's to 100's $\mu g\ l^{-1}$ waterborne Se). Conversely, a number of the data sets are from extremely low Se environments (e.g., mayfly). Perhaps, for TTF derivation purposes, EPA should constrain calculation of the median TTF to conditions that approximate the range of WQC (e.g., 0.5-10 $\mu g\ l^{-1}$ in water) that EPA might consider on a site-specific basis, or the range range of concentrations typically associated with the EC10 for sensitive fish species. Otherwise, individual TTFs have the potential to be biased either low or high depending on the site(s) from which they were collected. EPA should carefully review the biokinetic data using similar criteria.

3. Regarding the conversion factor (*CF*) values used, did EPA use an appropriate and scientifically defensible method to derive those values (p. 78-79 of the criteria document and Appendix B)? Are you aware of any other methods that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included? Please provide detailed comments.

EPA has used a scientifically defensible method for deriving CFs. I am not aware of any other data EPA should consider. It could be argued that a regression based approach be used instead of the ratio approach EPA has adopted. In some cases, it appears that residuals are structured, suggesting that assumptions of the CF approach may be violated. At least for the 4-5 most sensitive taxa, EPA should conduct a sensitivity analysis of the regression-based approach versus the ratio approach and particularly consider confidence in the CF at concentrations that approximate the EC10.

4. Regarding the derivation of enrichment factor (*EF*) values, was the method EPA used to screen data from the literature applied appropriately and consistently (see inclusion/exclusion criteria on p. 71-77 of the criteria document)? Was the method for deriving *EF* values applied to those data in a consistent manner so as to derive *EF* values for selected waters in a scientifically defensible manner? Is the method that EPA used to establish the lentic and lotic categories for *EF* values reasonable given the available data? Are you aware of other methods or relevant data the EPA should consider? Please provide detailed comments.

Yes, the method for deriving EFs was scientifically defensible and appears to have been applied in a consistent manner. However, similar to my comments regarding TTFs, there is frequently an inverse relationship between water Se and EF. EPA should carefully examine the distribution of EFs as a function of water Se and assess whether their data set is unduly biased by EFs measured in systems with unusually low or high waterborne Se. It would be helpful if Table 12 included the mean or median water Se concentration at the site. Note, in the section on calculation of EFs, there is no reference to where the EFs for the 69 individual sites can be found (i.e., Appendix L).

5. Please comment on the scientific defensibility of EPA's conversion of the selenium fish tissue – water translation equation into an equation that allows for calculation of a criterion for waters that may be subject to intermittent discharges of selenium. Please comment on major sources of uncertainty in this approach. Is this method appropriate, given the bioaccumulative nature of selenium? Please comment on the uncertainty associated with the application of this conversion equation to intermittent discharges that may occur in different types of waterbodies and/or in different locations, particularly with respect to loads transported to potentially more sensitive aquatic systems. Does the method employed result in criteria that are similarly protective to the 30-day chronic criterion? Are there any other models or approaches that EPA should consider that would reduce this uncertainty? Please provide detailed comments.

EPA's proposed method for addressing intermittent and time-varying discharges appears reasonable given available data. Ideally, intermittent criteria would be based on a biokinetic modeling approach and EPA's effort to evaluate their proposed approach using biokinetic modeling is encouraging. However, given the limited biokinetic data currently available, it is probably premature to implement such an approach for setting WQC. Further use of such an approach may be unnecessarily complicated if the simpler approach proposed by EPA continues to achieve the same objective as the biokinetic approach. A major uncertainty in the approach and subsequent biokinetic evaluation is the near complete lack of kinetic data for EF. If depuration kinetics are slower than EPA has assumed for primary producers, then this will have significant impacts on the validity of this approach.

The issue of generating pulse loads of Se that may ultimately result in Se accumulation in sensitive downstream systems (e.g., pulse loads in a river that discharges to a wetland) is a legitimate concern. However, in my opinion, this is a site-specific issue and it is not reasonable to establish national WQC that ensure protection of these sites without dramatically increasing the false positive rate for the WQC. However, it would be useful for EPA to provide specific language on the need to consider loading to downstream environments when regulating intermittent discharges or developing site-specific WQC.

PART IV: SIGNIFICANCE OF SCIENTIFIC VIEWS FROM THE PUBLIC/STAKEHOLDERS

EPA will also be providing scientific views and other comments from stakeholders and the public received via the public docket to the peer review panel. Although EPA will be providing the full contents from the docket, EPA is only requesting a review of any scientific views/public comments that may be of technical significance to the selenium criterion.

1. Has the peer review panel identified any scientific views from the public or stakeholders as being technically significant to the draft of the selenium criterion going forward; that is, has information or data been introduced during the comment period that would change the scientific direction of the criterion? Is there any information or data that may refine or enhance the scientific defensibility of this criterion that EPA should consider further? Please provide detailed comments on specific issues of technical significance or refinement.

After reviewing the public/stakeholder comments, I highlight the following comments which I would also make above and beyond responses to specific review questions EPA has asked:

1.) Because some states will continue to use an acute WQC for Se, I agree EPA needs to clarify its position on the scientific credibility of the existing acute WQC.

2.) There were a number of comments indicating that use of an instantaneous averaging period and "never to exceed" for the tissue element is inappropriate and inconsistent with the Guidelines. I disagree with these comments and support EPA's decision.

3.) I agree with several commenters that EPA must develop rigorous definitions of lentic and lotic as guidance for regulators.

4.) EPA needs to provide some guidance on how small first order and ephemeral streams that naturally do not support fish populations should be regulated. There are a large number of these streams in the western US that have Se issues. Note, in these types of systems or in small wetland systems without fish, aquatic-dependent birds may be the most sensitive receptor. These leads to the obvious comment that if this WQC is intended to protect all US surface waters, EPA must develop guidance on the protection of aquatic-dependent wildlife.

PEER REVIEW COMMENTS FROM

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External Peer Review of the Draft Aquatic Life Ambient Water Quality Criterion for Selenium – Freshwater 2014

Responses to Charge Questions by Dr. Gregory A. Cutter

PART I: OVERARCHING QUESTIONS

1. Please comment on the overall clarity of the document and construction of the criterion statement with its multiple elements.

In general the document is clearly written, but there are numerous typographical errors, missing references (e.g., EPRI, 2006 cited first on p. 16), and incorrect citations (e.g., Table 12 cites Appendix L, but the Appendices only go to K). Some key words are poorly chosen (the freshwater criterion parts are called "elements"), especially considering that this document concerns an aquatic trace ELEMENT, and other elements such as mercury are also discussed; I recommend selecting another key word for this. The use of acronyms and abbreviations are unavoidable in a document like this, and while they provided a table listing them all (which should be numbered Table 1 on page xi), it would make the document more readable to those only looking for some specific details to periodically redefine these in the text, for example the first time it is used extensively in a new section. The criterion statement (largely in Section 3.8) is clearly written and presented, although I have serious scientific problems with parts of it to be elaborated below. While this was not directly requested in our charge, but has direct bearing on the problems in this document, the review section on the aquatic biogeochemistry of selenium (pp. 9-17) has factual errors that may reflect on the authors understanding of the selenium or on some biases. First, in Section 3.2 the statement that "…the effects are integrated across forms of selenium; thus water column values are based on total selenium exposure." is an oversimplification that leads to conceptual errors later. The amount of dissolved selenium that enters the food web through the first trophic level is strongly linked to the speciation of dissolved selenium (e.g., Reidel et al., 1991; Baines and Fisher, 2001; Baines et al., 2001; Baines et al., 2004), which for freshwater and marine/brackish species is: selenite=organic selenide>>selenate. So for a lotic or lentic water body that is dominated by selenate, the incorporation of selenate into the phytoplankton biomass is much lower than that if the selenium was in the +4 oxidation state. In the next section 3.2.1, it starts off with serious errors, in particular "organo-selenide" being selenomethionine. Data on the speciation of dissolved organic selenide show it to be in soluble peptides and proteins, not free amino acids (e.g., Cutter, 1982; Cutter and Cutter, 1995), so phytoplankton uptake studies using free selenomethionine are not using the actual dissolved forms and likely overestimating uptake.

A following sentence says that selenite tends to dominate in "slow moving waters", presumably lentic environments. However, there are no data in the literature to support this statement (e.g., see compilations in Cutter, 1989a); selenite is only dominant when there is a large, fossil fuel-derived input, regardless of water residence time (e.g., Cutter, 1989a, 1989b). In this respect, on p. 14, 2nd complete sentence, they state that geologic AND anthropogenic sources often release mostly selenate, but most anthropogenic sources produce selenite (e.g., Cutter, 1989a, 1989b; Cutter and Church, 1986), only geological sources (weathered or irrigated) yield selenate; the presence of selenite in surface waters can in fact be used as a fossil fuel-combustion source indicator (e.g, Cutter, 1989a, 1989b). Interestingly, the last paragraph on p. 14 is largely correct in stating that the concentration of particulate selenium in the first trophic level (algae) is highly dependent on the dissolved speciation; this begs the question of why the authors later ignore speciation and calculate EF on total (presumably dissolved) selenium in the water column and particles; see later comments.

In the Bioaccumulation section (3.2.2), the major error, and this is significant in terms of bioavailability, is that dissolved selenium uptake results in elemental selenium and organoselenium (2nd to last sentence on p. 15). Elemental selenium is only produced by dissimilatory (heterotrophic) reduction under low oxygen conditions (many works of Oremland, but they correctly cite Oremland et al., 1989); autotrophs perform assimilatory reduction to selenide that is then coupled with acetyl CoA, serine, etc to produce seleno amino acids. Also, the use of the term "absorbed" is poorly chosen in that it implies simple exchange with no chemical reactions; dissolved selenium is assimilated (or incorporated) into autotrophic organic matter, which in the case of selenite uptake/assimilation/incorporation involves a change in oxidation state and chemical form (i.e., selenite is reduced to selenide and bonded with carbon to produce seleno amino acids like selenocysteine).

2. EPA has developed a tiered selenium criterion with four elements, with the fish tissue elements having primacy over the water-column elements, and the egg-ovary element having primacy over any other element. Inclusion of the fish whole-body or fish muscle element into the selenium criterion ensures the protection of aquatic life when fish egg or ovary tissue measurements are not available, and inclusion of the water column elements ensures protection when fish tissue measurements are not available

    a. Please comment on the tiered construction of the selenium chronic criterion; is it logical, and scientifically defensible as it applies to protection of freshwater aquatic life:

        i. That is, is the primacy of the egg-ovary element over the other elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Given the known, well documented, and published in the peer-reviewed literature information, choosing the egg-ovary compartment/vector/whatever (not element) is very well justified. The accuracy of then selecting a suitable value for various fish species depends on a critical evaluation of the literature, or new experiments.

        ii. Is the primacy of the whole-body/fish muscle element over the water column elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Again, this is well documented and the only proviso would be the choice/selection of the CF value

        iii. Please comment on the scientific uncertainty that may be associated with this tiered approach? Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

While the approach is scientifically justifiable, the propagation of errors that combine to make the total uncertainty is a bit daunting. Indeed, their frequent use of r or r^2 values for log/log plots completely masks the overall uncertainty; what are the correlations for direct concentration comparisons? I suspect they are much less than 0.4 and the p values would make them far less significant. Having said this, the trophic level transfers between higher levels (1 and above) are well described and parameterized in the literature, so the authors really should do a complete error/sensitivity analysis to quantify the overall error/uncertainty.

iv. Are the draft recommended magnitude, duration, and frequency for each criterion element scientifically sound and appropriate? Please provide detailed comments.

I found the time and frequency evaluations of the factors (not elements) well justified, with the exception of the EF, to be explained below.

PART II: FISH TISSUE CRITERION ELEMENTS DERIVATION: DERIVATION OF FISH EGG-OVARY, WHOLE BODY AND MUSCLE CRITERION ELEMENT(S)

EPA is requesting a technical review of the methods and procedures used to derive a chronic selenium criterion based on an egg-ovary concentration, as well as its translation to a criterion element applicable to whole-body and muscle tissue. Please address the following questions:

1. Please comment on EPA's use of the effects concentration 10^{th} centile (EC_{10}) as the measurement endpoint for the fish reproductive toxicity studies used to derive the egg-ovary element.

This seems like a statistically-valid approach to setting the threshold, but toxicology is not my field of expertise.

2. Data used to derive the final chronic egg-ovary criterion element were differentiated based on the type of effect (reproductive vs. non-reproductive effects). Acceptable chronic toxicity data on fish reproductive effects are available for a total of nine fish genera. The genus Sensitivity Distribution (SD) is predominantly populated with data on fish genera because field evidence demonstrates that fish communities can be affected by selenium even when there is no observable change in the invertebrate community diversity and abundance. As a result, decades of aquatic toxicity research have focused primarily on fish. Available field and laboratory studies indicate that invertebrates are more tolerant to selenium than most of the tested fish species (Criteria document, Table 6c, Section 4.1.2). The data set used to derive the selenium criterion marks a change from the traditional method used to derive water quality criteria that requires toxicity tests with aquatic organisms from 8 phylogenetically distinct taxa (including three vertebrate and five invertebrate genera) in order to derive aquatic life criteria (Stephan et al., 1985).

    a. Given selenium's more taxon-specific and life stage-specific toxicity, please comment on EPA's use of the available data to derive the egg-ovary tissue element.

In as much as fish are the most vulnerable to Se toxicity, and it is manifested primarily at reproduction, the egg-ovary focus is justified. The availability of data that passed the EPA criteria is somewhat limited, but statistically valid. Having said this, I am not well-versed in fish toxicity literature, so I rely on the other reviewers to point out data sets that may have been overlooked (e.g., I know they missed many water column data).

b. Given the greater general sensitivity of oviparous fish to selenium compared to aquatic invertebrates, please comment on the appropriateness of EPA's fish tissue-based criterion for affording protection to the aquatic community as a whole (e.g., including invertebrates).

In the aquatic systems in which I have worked with selenium, we have never encountered Se problems with invertebrates, and the literature seems to bear this out. So it seems to me that setting the criteria for the most at risk population is the best approach.

c. With respect to the tests that quantified non-reproductive effects, did the EPA use that data to the best extent possible given its limitations (e.g., relevance compared to reproductive tests, and data quality concerns which increased uncertainty (e.g., Hamilton et al., 1990)?

Again, fish toxicity is not my expertise, so I cannot adequately respond to this question.

d. EPA also rejected studies that used the injection route of exposure for selenium due to uncertainty related to uptake, distribution and metabolism/transformation kinetics when compared with the dietary and/or maternal transfer routes of exposure. Was this reasonable? Does the panel envision an appropriate and scientifically defensible use for this type of data? Please provide detailed comments.

I cannot recommend using any artificial means of introducing selenium to tissues; exposure must be through food and the assimilation pathways it follows for a given species. In this respect, chemical speciation is very important, so the exact form of organic selenide (peptide vs free amino acid, seleno methionine vs seleno cysteine; cytosol vs proteins) is critical to its uptake and eventual assimilation (e.g., Reinfelder and Fisher, 1994; Luoma et al., 1992).

3. Was the method (Section 4.1.5, 7.1.7) used to translate the fish egg-ovary criterion element into muscle and whole body criterions elements understandable, transparent and scientifically defensible? Was there sufficient data for making the translations for each element?

The methodology is well described and documented, but as above I would like to see a more thorough error analysis for the resulting CFs.

PART III: EVALUATION OF THE TRANSLATION PROCEDURE TO DERIVE THE WATER COLUMN ELEMENT(S)

EPA is also requesting a technical review of the methods and procedures used to translate the egg-ovary element of the chronic selenium criterion to water-column elements. Relevant sections of the document include:

- A description of the method used to derive an equation to translate the egg-ovary element to a monthly water-column element in perennial (lentic and lotic) waters and an equation that can be used to convert the monthly water-column element to an intermittent water column element (Sections 3.8.3, 3.8.4, 4.2.1, 4.3, and Appendix G).

- An analysis of the translation equation precision using data obtained from published literature (Sections 7.2.1, 7.2.2, and Appendix H).

- A description of the method and data sources used to derive the translation equation parameters (Sections 4.2.2, 4.2.3, and Appendix B).

- A description of the method and data sources used to categorize waterbody types where a single water-column chronic criterion concentration value would be adequately protective in most circumstances (Section 4.2.4).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for established categories of waters (Section 4.2.5).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for intermittent discharges that may occur in lentic and lotic waterbodies (Section 4.3).

Please address the following questions:

1. Please comment on the scientific defensibility of EPA's translation equation method for translating the concentration of selenium in fish tissue to a concentration of selenium in the water-column. Please comment on major sources of uncertainty in applying the translation equation to different types of waterbodies (e.g., with differing retention times, water chemistries, and/or species present). Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

The overall approach of considering selenium's pathway from the water column, dissolved state, through trophic levels, and into tissues such as reproductive organs is well justified, particularly the trophic transfer model that is dynamic and rate/kinetically based (uptake rate * assimilation efficiency/elimination rate); the trophic transfer approach largely developed by Nick Fisher and collaborators. However, the water to first trophic level approach is completely unacceptable in that it is not dynamic or rate-based (actually assumes equilibrium) and completely ignores the effects of speciation. The latter is curious in that they seem to be relying on the Chapman et al (2009 and 2010) recommendations from the SETAC Pellston workshop which specifically states, "Understanding Se speciation is critical to understanding its mobility, transformation, partitioning in the environment, and potential risk to aquatic ecosystems." and "The single largest step in the bioaccumulation of Se occurs at the base of food webs, characterized by an "enrichment function"; thermodynamic or equilibrium-based principles are not appropriate for predicting Se bioaccumulation at the base of food webs." The choice of the Presser and Luoma model used in this EPA

document is completely contrary to these recommendations since the water/particle ratio called the Enrichment Factor (EF) is only a renamed equilibrium distribution coefficient (K_d) that was used long ago for metal cations. Dissolved and particulate selenium speciation cannot be modeled with equilibrium approaches, it must consider the kinetics of the transfers/transformations (e.g., Cutter, 1992). Since the transfer of dissolved selenium in any of its chemical forms to the particulate state (largely assimilation by phytoplankton and conversion to organic selenide – seleno amino acids in proteins) changes the chemical forms, how does one calculate a distribution coefficient (EF)? For selenium, dissolved selenite or selenate are not what are in the particulate state (organic selenides), so which dissolved species and which particulate species do you use to calculate EF? And, they are certainly not reversible (selenite uptake followed by regeneration does not return selenite, but rather organic selenide…which may later oxidize back to selenite and selenate; Cutter, 1982; Cutter and Bruland, 1984). In this EPA document, they "solve" this issue by only considering total dissolved selenium, in contradiction to the recommendations at the Pellston workshop.

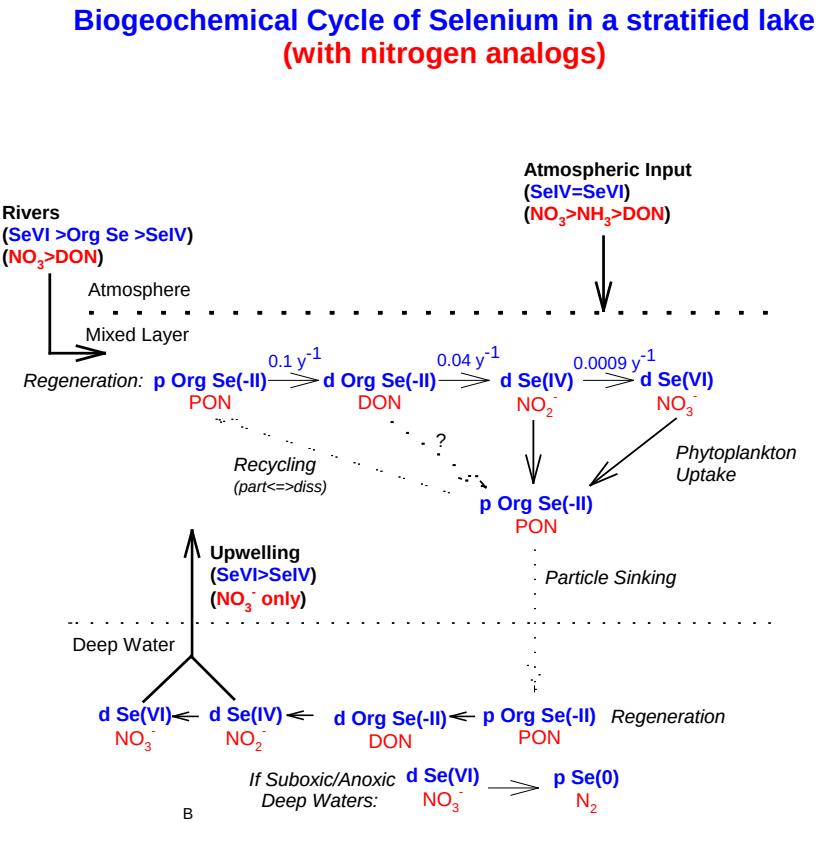
The use of the Presser and Luoma (2006, 2010) model for any aquatic ecosystem to predict dissolved or particulate concentrations is questionable for the simple reason that while it acknowledges the importance of chemical speciation, and the rates of processes (kinetics as opposed to equilibrium thermodynamics), it largely ignores them in application. It is a totally empirical model designed for the San Francisco Bay-Delta system, so its application to other systems may not work. To reiterate the preceding paragraph in detail, the primary problem with this model is the exchange between the dissolved and particulate phases, in this case the first trophic level (autotrophs/primary producers). While there is some adsorption of dissolved selenite and selenate to suspended particles (e.g., Doblin et al., 2006), most particulate selenium in organic matter is organic selenide in the form of seleno-amino acids in proteins (Wrench, 1978). In other words, the uptake of dissolved selenite and selenate from the water column by phytoplankton changes their chemical forms, it is reductively incorporated (Cutter, 1982; Cutter and Bruland, 1984).

Biological uptake of dissolved nutrients such as nitrogen, and metals, is best (most accurately) modeled using Michaelis-Menten kinetics, or at least pseudo-first order rate expressions. The release of this particulate organic selenide back into the water column as dissolved organic selenide is coupled to oxic (or anoxic) respiration (Cutter, 1982; Cutter and Bruland, 1984), which is also modeled using an appropriate rate expression (e.g., first order; see discussion in Meseck and Cutter, 2006). The critical point here is that the speciation of particulate selenium has no relation to that in the water column – reductive incorporation and subsequent regeneration obliterates this relationship and only a rate-based (kinetic) approach can accurately quantify it. However, the Presser and Luoma (2006, 2010) model uses equilibrium distribution coefficients (K_d or in this EPA document EF) to quantify how particulate selenium in the first trophic level reflects the dissolved concentration in the water column. The distribution coefficient approach works well for divalent metal cations where no oxidation state change occurs. For a given K_d value, if the dissolved concentration goes up, more adsorbs to the particles (to maintain equilibrium), and when the dissolved concentration drops, the particulate-bound metal desorbs. But, when there is a redox change between dissolved and particulate conversions, the equilibrium concept is violated. For example, if the concentration of selenite goes up, the rate of uptake increases, and the concentration of particulate organic selenide increases; in a crude fashion, the use of a K_d could mimic this biochemical process. But, when the concentration of dissolved selenite goes down, particulate organic selenide doesn't desorb to balance it; they are different chemical species. Particulate organic selenide is only released through respiration/regeneration, not adsorption/desorption (for which the K_d concept was created). So in this scenario, the Presser and Luoma (2006) cannot accurately predict the response to a change in dissolved concentration, and more importantly cannot predict the speciation of selenium.

Interestingly, Presser and Luoma (2006) note that as more recycling (i.e., the regeneration part of the selenium cycle depicted in Cutter and Bruland, 1984) occurs, organic selenide concentrations increase. Indeed, they do, but their model cannot reproduce this**, a problem if you "reverse" their model to predict water column dissolved concentrations of selenium for a given particulate concentration in**

the food web (e.g., 11.8 ppm Se in fish muscle; this document). This latter (highlighted) point is exactly what Section 4.2 is doing. On a related matter, the Presser and Luoma model suggests that it handles selenium speciation, but only in the dissolved phase, and then rather than using separate K_ds for each species, and presumably summing the contributions from each from to derive the particulate selenium concentration, they simply average the K_ds to one value and omit speciation.

To put this modeling approach into another perspective, it has been observed (Cutter, 2005) that the aquatic selenium and nitrogen cycles are very similar/parallel. Adding N cycling to the Se cycle depicted in Cutter and Bruland (1984) gives:



Thus, I ask those who wrote this document if they would use the Presser and Luoma (2006, 2010) approach to model nitrogen cycling and therefore set N discharge, etc limits? I suspect the answer would be no, and my response then would be, why use it for selenium?

To be constructive, what modeling approach should be used? In Cutter (1992) it was argued that a kinetic/rate approach, and not an equilibrium thermodynamic one (EFs are an equilibrium concept) is the only way to quantify the selenium cycle. There are at least two existing kinetic models for the selenium cycle: for lakes there is the one described in Porcella et al. (1991) and Bowie et al. (1996), and one for estuaries, Meseck and Cutter (2006). The Meseck and Cutter model focuses on the dissolved to first trophic level dynamics and includes the full speciation of selenium in the dissolved and particulate states in an estuary (San Francisco Bay/Delta). The Bowie et al. (1996) model uses a kinetic approach to modeling selenium speciation and dynamics from the dissolved state to all trophic levels in freshwaters, and was designed to assist in mitigation/restoration efforts. The Meseck and Cutter (2006) model also has direct applications to mitigation via scenario modeling (what if…). However, this model includes components to simulate sediment resuspension, mixing and dispersion, and primary production (light-

limited in this case), so it may be too complicated for the application needed here. Indeed, all that is needed is a model that covers dissolved to first trophic level interactions, and from there the existing biodynamic part of the Presser and Luoma (2006; 2010) could be employed. In this case, using Equations 4-6, and 7, in the Meseck and Cutter (2006) paper (and related equations in the Appendices) could suffice. Or, use simple Michealis-Menten equations and values in the literature (e.g., Riedel et al., 1991), and simple first order rate equations (and values) described in the literature (e.g., Cutter, 1982; Cutter and Bruland, 1984; Reinfelder et al., 1993).

2. Regarding the trophic transfer factor (*TTF)* values, did EPA use a scientifically defensible method to derive the *TTF* values (p. 71-77 of the criteria document)? Were the exclusion criteria, (pp. 71-77 of the criteria document) developed by EPA to screen the available data applied in a consistent and scientifically defensible manner? In particular, EPA noticed that application of the exclusion criteria resulted in *TTF* values for aquatic insect larvae that differ from other published values. Given this, are you aware of any other methods of screening data that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included, if appropriate? Please provide detailed comments.

After the dissolved to first trophic level particulate selenium part of the model that I am criticizing above, the rest of the Presser and Luoma (2006) model (including the derivation of TTFs) is excellent and accurately predicts bioaccumulation through the various parts of the food web (and earlier documented in the Luoma and Rainbow (2005) peer-reviewed paper). The reason here is that once into the first trophic level, the primary speciation of particulate selenium is organic selenide, and the concepts of assimilation efficiency, trophic transfer factors, ingestion and depuration (egestion) work well for selenium (and any other metal or nutrient).

The screening of data followed well-set protocols and are quite defensible. I am not aware of additional data to be included, but I'm sure there must be some in the grey literature.

3. Regarding the conversion factor (*CF*) values used, did EPA use an appropriate and scientifically defensible method to derive those values (p. 78-79 of the criteria document and Appendix B)? Are you aware of any other methods that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included? Please provide detailed comments.

The calculation of the CF values was rather straightforward, with my only concern, as noted above, being a thorough quantification of the resulting errors in the CF values. As an overall statement, error propagation seems to have been largely ignored in this document.

4. Regarding the derivation of enrichment factor (*EF*) values, was the method EPA used to screen data from the literature applied appropriately and consistently (see inclusion/exclusion criteria on p. 71-77 of the criteria document)? Was the method for deriving *EF* values applied to those data in a consistent manner so as to derive *EF* values for selected waters in a scientifically defensible manner? Is the method that EPA used to establish the lentic and lotic categories for *EF* values reasonable given the available data? Are you aware of other methods or relevant data the EPA should consider? Please provide detailed comments.

See above comments; I feel the EF values are completely useless and in fact incapable of being calculated given that they really need to include the chemical speciation of dissolved selenium. They did however

miss lots of dissolved and particulate data, many examples including: Cutter, 1989a; Cutter, G. A. 1991., Riedel and Cole, 2001 in their reference list, and river data in Cutter, 1989b and Cutter and San Diego-McGlone that are also in their reference list.

5. Please comment on the scientific defensibility of EPA's conversion of the selenium fish tissue – water translation equation into an equation that allows for calculation of a criterion for waters that may be subject to intermittent discharges of selenium. Please comment on major sources of uncertainty in this approach. Is this method appropriate, given the bioaccumulative nature of selenium? Please comment on the uncertainty associated with the application of this conversion equation to intermittent discharges that may occur in different types of waterbodies and/or in different locations, particularly with respect to loads transported to potentially more sensitive aquatic systems. Does the method employed result in criteria that are similarly protective to the 30-day chronic criterion? Are there any other models or approaches that EPA should consider that would reduce this uncertainty? Please provide detailed comments.

If a realistic concentration can be established using a more appropriate modeling approach (as above), then the calculation for intermittent discharges is fine. However, the propagation of errors must be carefully evaluated.

PART IV: SIGNIFICANCE OF SCIENTIFIC VIEWS FROM THE PUBLIC/STAKEHOLDERS

EPA will also be providing scientific views and other comments from stakeholders and the public received via the public docket to the peer review panel. Although EPA will be providing the full contents from the docket, EPA is only requesting a review of any scientific views/public comments that may be of technical significance to the selenium criterion.

1. Has the peer review panel identified any scientific views from the public or stakeholders as being technically significant to the draft of the selenium criterion going forward; that is, has information or data been introduced during the comment period that would change the scientific direction of the criterion? Is there any information or data that may refine or enhance the scientific defensibility of this criterion that EPA should consider further? Please provide detailed comments on specific issues of technical significance or refinement.

I examined the public comments AFTER I had reviewed the document and written the above comments, so as to not bias my own evaluation. The comments (by my count, 429) ranged from editorial ones, to simple criticisms, to detailed scientific evaluations and suggestions. Of the later, the most common concerned "implementation" (16% of total), followed by "translation" (to water column criteria; 14%), and site specific criteria (13%). If we combine all the "criteria" comments (site, tiered, tissue, intermittent), these received the most comments (30%). Of these, most dealt with the details of developing the criteria (justifying the calculation methods, literature missed, apparent oversights or conflicts with existing procedures). Thus, the peer-review community (it seems that most of these comments came from consulting companies, municipal and state agency scientists, and some from the academic sector) feels the document needs considerable attention to reformulating the criteria. The next most important topic was then implementing the criteria (16% by itself) and in this respect most comments (actually criticisms) were directed to the water column formulation. Related to this was the "translation" of the tissues (all)-based criteria to the water column (14% of comments), and most of these comments were directed to the inappropriate use of the Presser and Luoma model. Considering my review above and the community

response, it would seem that the EPA needs to reformulate their methodology for setting water column criteria.

References for Cutter evaluation that are not in the existing EPA reference list:

Baines, S.B., N.S. Fisher, M.A. Doblin, and G.A. Cutter. 2001. Uptake of dissolved organic selenides by marine phytoplankton. Limnol. Oceanogr., 46: 1936-1944.

Baines, S. B., N.S. Fisher, M.A. Doblin, G.A. Cutter, L.S. Cutter, and B. Cole. 2004. Light dependence of selenium uptake by phytoplankton and implications for predicting selenium incorporation into food-webs. Limnol. Oceanogr., 49: 566-578..

Cutter, G.A. 1982. Selenium in reducing waters. Science 217: 829-831.

Cutter, G.A. and T.M. Church. 1986. Selenium in Western Atlantic precipitation. Nature 322: 720-722.

Cutter, G.A. 1989a. Selenium in fresh water systems. In: *Occurrence and Distribution of Selenium* (M. Ihnat, ed.). CRC Press, Florida, Chap. 10.

Cutter, G. A. 1991. Selenium biogeochemistry in reservoirs. Volume 1: Time series and mass balance results. Electric Power Research Institute, EPRI EN-7281, 97 pp.

Cutter, G.A. 1992. Kinetic controls on the speciation of metalloids in seawater. Mar. Chem., 40: 65-80.

Cutter, G.A. 2005. Biogeochemistry: now and into the future. Palaeogeogr. Palaeoclimatol. Palaeoecol. 219: 191-198.

Meseck, S.C. and G.A. Cutter. 2006. Evaluating the biogeochemistry of selenium in San Francisco Bay through modeling. Limnol. Oceanogr., 51:2018-2032.

Porcella, D.B., G.L. Bowie, J.G. Sanders, and G.A. Cutter. 1991. Assessing Se cycling and toxicity in aquatic ecosystems. Water Air Soil Pollut., 57-58: 3-11.

PEER REVIEW COMMENTS FROM

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External Peer Review of the Draft Aquatic Life Ambient Water Quality Criterion for Selenium – Freshwater 2014

Responses to Charge Questions by Mr. David DeForest

PART I: OVERARCHING QUESTIONS

1. Please comment on the overall clarity of the document and construction of the criterion statement with its multiple elements.

There is a lot of information to digest and it may be difficult for non-technical readers to follow, but I feel that the document was organized in a logical manner and that the approaches were adequately described. Although I have technical comments relative to the criterion statement, I feel that format for presenting the selenium criteria based on multiple elements is clearly presented and easily digestible to the reader.

I have included here a few miscellaneous typos and editorial suggestions that I noted during my review:

p. 59, Table 7a: Correct spelling of "Onchyrhynchus" to " Oncorhynchus "

p. 60, paragraph below Table 7b: Correct spelling of "Leopmis" to "Lepomis"

p. 62, 1st paragraph: Correct spelling of "Oncorhyncus" to " Oncorhynchus "

p. 89, footnote a in Table 12: Appendix L should be Appendix K

p. 114, 1st paragraph, last sentence: Correct spelling of "criteirion" to "criterion"

2. EPA has developed a tiered selenium criterion with four elements, with the fish tissue elements having primacy over the water-column elements, and the egg-ovary element having primacy over any other element. Inclusion of the fish whole-body or fish muscle element into the selenium criterion ensures the protection of aquatic life when fish egg or ovary tissue measurements are not available, and inclusion of the water column elements ensures protection when fish tissue measurements are not available

    a. Please comment on the tiered construction of the selenium chronic criterion; is it logical, and scientifically defensible as it applies to protection of freshwater aquatic life:

        i. That is, is the primacy of the egg-ovary element over the other elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Yes, in my opinion the tiered construction of the chronic selenium criterion is logical and scientifically defensible. First, the critical exposure route for fish is dietary organic selenium (Janz et al. 2010), which is the basis for all of the studies in which egg or ovary selenium concentrations are linked to toxicity in offspring. Dietary organic selenium exposures are implicit in those studies in which adult females were exposed in the field and explicit in those studies in which adult females were exposed in the laboratory (primarily through the use of diets enriched with organic selenium, such as selenomethionine). Second, the critical toxicity endpoint for fish exposed to selenium is larval mortality, deformities, and/or edema following exposure to selenium during absorption of the yolk-sac. The selenium concentration in the egg or ovaries is the most relevant exposure metric for this exposure route and toxicity endpoint. Third, and related to the second point, is that fish species partition varying amounts of their total selenium burden to

the ovaries and eggs (deBruyn et al., 2008). Direct measurement of the selenium concentration in the eggs or ovaries addresses this between-species variability in selenium partitioning within tissues. Fourth, fish egg- or ovary-based selenium toxicity values (e.g., EC10s) are not highly variable among fish species, regardless of whether adult females were exposed to dietary organic selenium in the field or in the laboratory or whether species may be considered "warm-water" or "cold-water" species.

Some studies have also shown that juvenile fish survival and growth can be relatively sensitive to dietary organic selenium. For this toxicity endpoint, of course, an egg or ovary selenium criterion would not be applicable (but a whole-body selenium criterion would be). An important question, therefore, is whether compliance with an egg or ovary selenium criterion would be protective of juvenile fish. DeForest (2008) evaluated this question by comparing dietary Se toxicity data for juvenile growth and effects on larvae via maternal transfer. Although data were limited to bluegill sunfish (*Lepomis macrochirus*) for that evaluation, it was concluded that juvenile bluegill are not more sensitive than bluegill larvae exposed to selenium via maternal transfer. This would indicate that an egg or ovary selenium criterion should be protective of effects on juvenile survival and growth (if the observations for bluegill are translatable across fish species).

Although I agree that the primacy of each criterion element is logical, it is not clearly stated whether a water Se criterion could be adopted into a permit limit. For example, if compliance with the lotic or lentic Se criterion is demonstrated, is measurement of fish tissue Se concentrations necessary? If a water body meets a fish tissue-based Se criterion, but not a surface water criterion, would the water body be considered in compliance? I believe the answer to the latter is "yes", but this does not seem to be clearly stated in the draft AWQC document.

Literature cited:

deBruyn A, Hodaly A, Chapman P. 2008. Tissue selection criteria: Selection of tissue types for the development of a meaningful selenium tissue threshold in fish. Tissue Selection Criteria, Threshold Development Endpoints, and Potential to Predict Population or Community Effects in the Field Prepared for the North American Metals Council - Selenium Working Group, Washington, DC.

DeForest D. 2008. Threshold development endpoints: Review of selenium tissue thresholds for fish: Evaluation of the appropriate endpoint, life stage, and effect level and recommendation for a tissue-based criterion. Tissue Selection Criteria, Threshold Development Endpoints, and Potential to Predict Population or Community Effects in the Field Prepared for the North American Metals Council - Selenium Working Group, Washington, DC.

Janz DM, DeForest DK, Brooks ML, Chapman PM, Gilron G, Hoff D, Hopkins WA, McIntyre DO, Mebane CA, Palace VP, Skorupa JP, Wayland M. 2010. Selenium toxicity to aquatic organisms. 141-231 in Chapman PM, Adams WJ, Brooks ML, Delos CG, Luoma SN, Maher WA, Ohlendorf HM, Presser TS, Shaw DP, eds. Ecological assessment of selenium in the aquatic environment. SETAC Press, Pensacola, FL, USA.

ii. Is the primacy of the whole-body/ fish muscle element over the water column elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Yes, in my opinion the primacy of the whole-body or muscle selenium criterion over the water column criterion is scientifically sound. Selenium bioaccumulation potential from water to fish is highly site-specific (Brix et al., 2005; Presser and Luoma 2010; Stewart et al., 2010), so it is appropriate that a whole-body or muscle selenium criterion is given a priority over a water column selenium criterion.

Consideration of only a water column selenium criterion (or a water column selenium criterion that is given priority over a fish tissue-based selenium criterion) would necessarily have to be very low to ensure protection of the sites with the greatest selenium bioaccumulation potential. However, this would potentially be problematic because it would trigger concerns (i.e., selenium criterion exceedances) at locations where selenium bioaccumulation potential is lower and not of ecological concern.

Literature cited:

Brix KV, Toll JE, Tear LM, DeForest DK, Adams WJ. 2005. Setting site-specific water-quality standards by using tissue residue thresholds and bioaccumulation data. Part 2. Calculating site-specific selenium water-quality standards for protecting fish and birds. Environ Toxicol Chem 24:231-237.

Presser TS, Luoma SN. 2010. A methodology for ecosystem-scale modeling of selenium. Integr Environ Assess Manag 6:685-710.

Stewart R, Grosell M, Buchwalter D, Fisher N, Luoma S, Mathews T, Orr P, Wang W-X. 2010. Bioaccumulation and trophic transfer of selenium. 93-139 in Chapman PM, Adams WJ, Brooks ML, Delos CG, Luoma SN, Maher WA, Ohlendorf HM, Presser TS, Shaw DP, eds. Ecological assessment of selenium in the aquatic environment. SETAC Press, Pensacola, FL, USA.

iii. Please comment on the scientific uncertainty that may be associated with this tiered approach? Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

Overall, I believe that the tiered approach is scientifically appropriate. I do have specific comments on the actual selenium criteria at each tier, which are provided under specific charge questions below.

iv. Are the draft recommended magnitude, duration, and frequency for each criterion element scientifically sound and appropriate? Please provide detailed comments.

The comments below are organized first by magnitude, duration, and frequency, and then by criterion element (i.e., fish egg or ovary, fish whole-body or muscle, and water column) within each of these categories.

Magnitude

Fish Egg/Ovary Se Criterion

<u>Brown Trout</u>

The draft fish egg/ovary selenium criterion is 15.2 mg/kg dw. This draft criterion is driven by brown trout (*Salmo trutta*), which had an EC10 of 15.91 mg/kg dw in the EPA's draft AWQC document. This study, conducted by Formation Environmental (2011a), has received tremendous scrutiny in how to best interpret the results and derive a defensible EC10. In my earlier review of that study on behalf of the Eastern Research Group (ERG) and EPA, I had concluded that the most relevant egg selenium EC10s that could be derived from that study ranged from 20.70-21.60 mg/kg dw. In that same review, however, I concluded that an egg selenium EC10 of 16.76 mg/kg dw was on the lower end of the range of possible EC10s that could be derived from the study. Accordingly, in my opinion, the EC10 of 15.2 mg/kg dw used by the EPA is an overly conservative interpretation of the brown trout Se toxicity study.

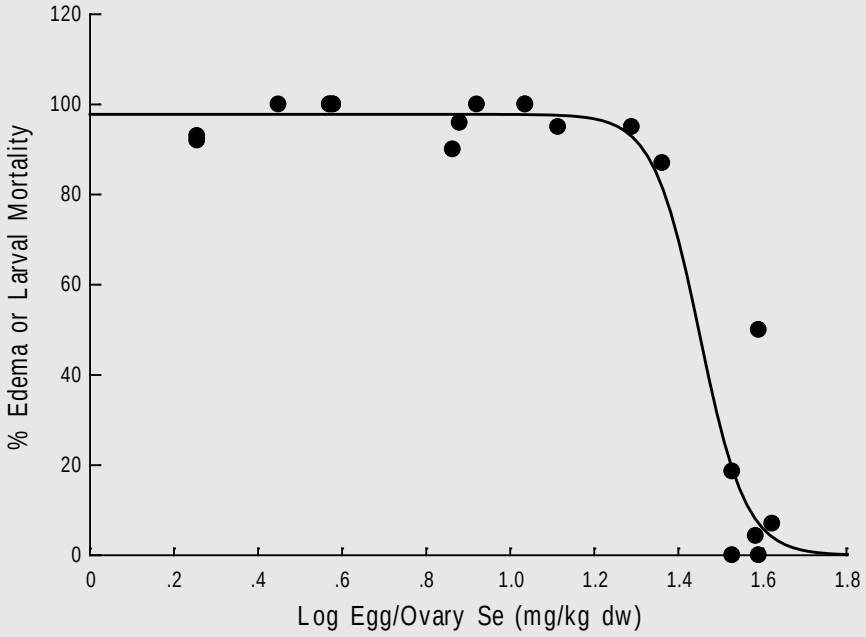
Bluegill

The second lowest species mean chronic value (SMCV) was 18.41 mg/kg dw for bluegill sunfish (*Lepomis macrochirus*). This SMCV was based on the geometric of EC10s from three studies: (1) an EC10 of 20.05 mg/kg dw from Doroshov et al. (1992); (2) an EC10 of 24.55 mg/kg dw from Coyle et al. (1993); (3) an EC10 of 12.68 mg/kg dw from Hermanutz et al. (1992, 1996). The latter EC10 is much less than the other two EC10s for bluegill and less than even a very conservative interpretation of the EC10 for brown trout. I agree with the interpretations of the Doroshov et al. (1992) and Coyle et al. (1993) studies, but disagree with the interpretation of the Hermanutz et al. (1992, 1996) study. The EC10 of 12.68 mg/kg dw from Hermanutz et al. (1992, 1996) is driven by two treatments from Study 1: these were Streams 3 and 8 which had an ovary Se concentration of 17.71 mg/kg dw and 80% edema was observed and Steam 4 which had an ovary Se concentration of 15.46 mg/kg dw and 50.3% edema was observed. At first glance, there are three issues that stand out:

- First, the water Se treatment concentration that resulted in an ovary Se concentration of 17.71 mg/kg dw in Study I was 10 µg/L—in the 10 µg Se/L treatment in Study II the ovary Se concentrations averaged 36.39 mg/kg dw and the average rate of edema was 83%. Thus, the rates of edema were consistent between the 10 µg Se/L treatments in Study I and II, on average, but the ovary Se concentrations were widely different. The mean macroinvertebrate Se concentrations in the 10 µg Se/L treatments in Study I and II were similar (grand means among all invertebrate taxa were 21.6 and 22.8 mg/kg dw for Study I and Study II, respectively [Hermanutz et al., 1996]). The relatively large difference in the bluegill ovary Se concentrations in Study I compared to Study II, therefore, is unexpected.
- Second, in Study I, the ovary Se concentration of 17.71 mg/kg dw in the 10 µg Se/L treatment was greater than the ovary Se concentration of 15.46 mg/kg dw in the 30 µg Se/L treatment. This is also unexpected because the grand mean Se concentration in invertebrate taxa collected from the 10 and 30 µg Se/L streams were 21.6 and 44.7 mg/kg dw, respectively. Thus, a higher ovary Se concentration in the 30 µg Se/L stream would be expected. This basis for this discrepancy is not clear, although the ovary Se concentration measured in the 30 µg Se/L stream was based on a single fish, which may have randomly had a lower ovary Se concentration.
- Third, a potentially more important source of uncertainty is that the ovary Se concentrations in the Hermanutz et al. (1992, 1996) study were reported on a wet weight basis. Dry weight ovary Se concentrations were estimated assuming a moisture content of 76%, which was based on the average from Gillespie and Baumann (1986), 85%, and Nakamoto and Hassler (1992), 67%. If the true moisture content was 85%, the bluegill Se EC10 from Hermanutz et al. (1992, 1996) would be 20.3 mg/kg dw (almost identical to the EC10 derived from Doroshov et al. [1992]). In contrast, if the true moisture content was 67%, the bluegill Se EC10 from Hermanutz et al. (1992, 1996) would be 9.2 mg/kg dw.

In my opinion, the uncertainty in the moisture content of the bluegill ovaries in the Hermanutz et al. (1992, 1996), along with uncertainties in the ovary Se concentrations in Study I, are sufficiently great that this study should not be included in the SMCV for bluegill, as there are two other studies (Doroshov et al. [1992] and Coyle et al. [1993]) for which dry weight ovary Se concentrations were reported and the EC10s from those two studies were very comparable. The SMCV for bluegill based on those two studies would be 22.2 mg/kg dw. Alternatively, if data from Study I of Hermanutz et al. (1992, 1996) are pooled with data from Doroshov et al. (1992) and Coyle et al. (1993), the consistency in the concentration-response data is apparent and an EC10 of 21.4 mg/kg dw can be derived (Fig.1).

Fig. 1. Concentration-response relationship for bluegill based on data pooled from Study I of Hermanutz et al. (1992, 1996), Doroshov et al. (1992), and Coyle et al. (1993). EC10 = 21.4 mg/kg dw based on logistic regression analysis in TRAP.



Other Fish Species in the SSD

The draft fish egg/ovary Se criterion derived following EPA guidelines is based on the four lowest GMCVs and the total number of GMCVs. The two lowest GMCVs in the EPA's draft document are for *Salmo* (represented by brown trout) and *Lepomis* (represented by bluegill), which were both discussed above. The 3rd and 4th lowest GMCVs are for *Micropterus* (represented by largemouth bass) and *Oncorhynchus* (represented by cutthroat trout and rainbow trout). I do not disagree with EPA's interpretation of the studies for those genera.

The *Esox* GMCV of <34 mg/kg dw, represented by northern pike, is an EC24 because the data were not amenable to derivation of an EC10 using TRAP. The EPA compared this EC24 to the EC24 that could be derived for rainbow trout and noted that the two species appear to be similar in sensitivity, with northern pike perhaps slightly less tolerant. In contrast, the original study authors for the northern pike study, Muscatello et al. (2006), reported an EC10 of 20.38 mg/kg dw based on linear regression. The EC10 of 20.38 mg/kg dw would make the *Esox* GMCV the 4th lowest in the EPA's dataset. This change alone, however, would have a negligible influence on the draft fish egg/ovary Se criterion—it would raise it slightly from 15.2 mg/kg dw to 15.6 mg/kg dw (lowering the 4th lowest GMCV steepens the slope of the line through the four lowest GMCVs, which increases the 5th percentile).

Number of GMCVs Assumed in Fish Egg/Ovary Se Criterion Calculation

The logic for setting the number of GMCVs to 14 is flawed in my opinion. This number is based on 9 fish genera, 3 invertebrate genera with tissue-based toxicity data available, and 2 crustacean genera that were waived. In my opinion, a genus sensitivity distribution based on Se toxicity values for fish eggs/ovaries, and for which the resulting criterion will be a Se concentration in fish eggs/ovaries, and for which compliance will be determined by measuring Se concentrations in fish eggs/ovaries, cannot include data for non-fish taxa. It must be remembered that a criterion based on an internal tissue concentration is not the same as a criterion based on an external concentration to which the entire aquatic community may be

exposed. One will not be able to measure Se concentrations in invertebrates in order to determine compliance with the fish tissue-based Se criterion, so they should not be included in the SSD. Further, if I understand correctly, the three whole body Se EC10s for invertebrates (37.84 mg/kg dw for *B. calyciflorus*, >140 mg/kg dw for *L. variegatus*, and 24.2 mg/kg dw for *C. triangulifer*) were multiplied by a (1) diet-to-whole body fish TTF and (2) a whole body-to-egg/ovary conversion factor in order to estimate the Se concentrations in fish eggs/ovaries that may result from the toxicity thresholds for invertebrates. These values were then used as "SMCV & GMCV as estimated EO concentration in an accompanying fish assemblage (mg Se/kg dw EO)" in Table 6b of the draft AWQC document. However, these are simply predicted concentrations in fish eggs/ovary and are not effect concentrations for fish. I believe that n should equal the number of fish genera, which is 9 based on the draft AWQC document.

Additional Genera that Could be Added to the Total N

Although the EPA did not include the egg/ovary Se toxicity data for white suckers (*Catostomus commersonii*; de Rosemond et al. 2005) and razorback suckers (*Xyrauchen texanus*; Hamilton et al. 2005a,b) because reliable toxicity thresholds (EC10s or other) could not be derived, there does appear to be sufficient evidence that they would be among the four most sensitive genera. Thus, the number of GMCVs used in the criterion calculation could be increased from 9 fish genera to 11 fish genera.

Toxicity Data for an Additional Fish Species

Nautilus Environmental in Burnaby, British Columbia has conducted a Se maternal transfer toxicity study with mountain whitefish (*Prosopium williamsoni*). This species does not appear to be especially sensitive (i.e., it would not be among the four lowest GMCVs), but it would added another genus to the sensitivity distribution. I recommend that the EPA investigate whether this study is publically available and, if so, whether it meets the EPA guideline for test acceptability and inclusion in the sensitivity distribution. The Se toxicity study with Yellowstone cutthroat trout (Formation Environmental 2011b) should also be considered.

Influence of Potential Changes to GMCVs and N

As summarized above, in my opinion, the most conservative and reasonable EC10 that can be derived for brown trout is 16.76 mg/kg dw (although the weight-of-evidence suggest to me that the EC10 falls between about 20.7-21.6 mg/kg dw) and that the bluegill SMCV should be 22.2 mg/kg dw. If the four lowest GMCVs were 16.76 mg/kg dw for *Salmo*, 20.35 mg/kg dw for *Micropterus*, 22.2 mg/kg dw for *Lepomis*, and 22.53 mg/kg dw for *Oncorhynchus*, and the total number of fish genera was set equal to 11 (with inclusion of the two sucker genera), the resulting criterion would be 16.0 mg/kg dw. Alternatively, if the *Esox* (northern pike) GMCV was adjusted from <34 mg/kg dw to 20.4 mg/kg dw, the resulting criterion would change slightly to 16.1 mg/kg dw.

Fish Whole-body and Muscle Se Criteria

The draft fish whole-body and muscle selenium criteria are 8.1 and 11.8 mg/kg dw, respectively. In general, I believe that the approach for deriving these draft criteria is reasonable and that the magnitudes of these criteria are consistent with the toxicological literature. My only suggestion is that the EPA consider using empirically measured whole-body Se (or muscle Se) data for those species where it is available, rather than applying CFs to egg/ovary Se data. It would be interesting to see whether that has a significant influence on the draft whole-body or muscle Se criteria. And of course if any modifications are made to the egg/ovary Se GMCVs, this would influence the draft whole-body and muscle Se criteria, as would a change to the number of genera, if my suggestions above are considered.

Surface Water Se Criteria - Monthly Average

The draft water column selenium criteria are 4.8 and 1.3 µg/L for lotic and lentic waters, respectively. In general, I do not agree with the approach used by the EPA in deriving these water column criteria.

Although I do not agree with the approach, I do believe that the draft criterion of 4.8 µg/L for lotic waters is reasonable and consistent with our understanding of the range of Se bioaccumulation potential into fish across a wide range of lotic sites. However, for the draft lentic Se criterion of 1.3 µg/L, the approach used by the EPA results in this criterion being almost exclusively driven by data for two reference locations. This in turn is mostly due to what I perceive as a flaw in the approach, where site-specific Se data in invertebrates and fish are ignored and instead non-site-specific TTFs and CFs are applied that are inconsistent with the site-specific data. This resulted in cases where erroneously high modeled Se concentrations in fish tissue are linked with low water Se concentrations (i.e., reference site concentrations), and then these become the "drivers" for the draft lentic criterion of 1.3 µg/L. Please see my detailed comments on this issue in Part III.

Surface Water Se Criteria - Intermittent Exposure

The draft intermittent exposure Se criteria represent a mathematical manipulation of the monthly average criteria in order to derive values that would still result in 30-day average concentrations of 4.8 and 1.3 µg/L for lotic and lentic waters, even if those were exceeded for *x* number of days. A limitation of this approach is that it does not consider the uptake and elimination kinetics of Se in aquatic food chains and the influence of exposure duration and magnitude on these biokinetic parameters. In my opinion, a biokinetic modeling-based approach would be more appropriate for deriving intermittent, or acute, criteria that are protective against exceeding fish tissue-based Se criteria. More details are provided in my comments in Part III below.

Duration

Fish Egg/Ovary, Whole-body, and Muscle Se Criteria

The draft fish tissue-based selenium criteria (eggs, ovaries, whole-body, muscle) are "instantaneous measurements" as "Fish tissue data provide point measurements that reflect integrative accumulation of selenium over time and space in the fish at a given site" and "Selenium concentrations in fish tissue are expected to change only gradually over time in response to environmental fluctuations." I agree with the EPA's decision that the duration for fish tissue Se measurements should be an instantaneous measurement since, for most scenarios and fish species, the Se concentrations in fish tissue will be reflective of a longer term exposure.

Surface Water Se Criteria - Monthly Average and Intermittent Exposures

In my opinion, 30 days for an average exposure duration is reasonable, especially since an intermittent criterion is being considered (although, as noted, I believe the intermittent criterion would best be derived using a biokinetic modeling approach). Biokinetic data for algae and several freshwater invertebrates indicate that steady-state Se concentrations in the food chain may be achieved within this time frame.

Frequency

Fish Egg/Ovary, Whole-body, and Muscle Se Criteria

Although the EPA's AWQC, including the draft water Se criteria, are not to be exceeded more than once in three years, the fish tissue-based Se criteria are "never to be exceeded." To my knowledge, the "frequency" component of AWQC is rarely incorporated into permit limitations, so the implications of fish tissue-based Se criteria "never to be exceeded" are not entirely clear to me. The "frequency" component was initially incorporated into AWQC based on the premise that ecosystems will not be harmed if the number of criterion excursions is limited and/or there are compensating periods of time below the criterion over which the ecosystem can recover. As far as I can tell, the draft AWQC document for Se does not explain the basis for the "never to be exceeded" frequency decision for fish tissue. It seems that there should be some level of consistency between the allowable "frequency" for fish tissue-based and water-based Se criteria.

Surface Water Se Criteria - Monthly Average and Intermittent Exposures

The "frequencies" of "not more than once in three years on average" are consistent with the EPA guidelines and AWQC for other chemicals. As noted above, however, I am not aware of the "frequency" component of AWQC being incorporated into most effluent limitation so am unsure of the significance of this component. The fixed monitoring benchmark (FMB) approach, which has initially been developed for copper and biotic ligand model (BLM)-based criteria, represents a method that does explicitly account for exceedance frequency (USEPA 2012). However, this approach is for use under a site-specific context and would not apply to the national (non-site-specific) Se criteria. A reasonable excursion frequency for Se in water should be determined carefully, however, as Se is bioaccumulative and has variable persistence depending on receiving water conditions. For example, more frequent excursion frequencies may not be consequential in lotic systems with low biological productivity and short resident times, while an excursion frequency greater than once every three years may be warranted for lentic systems with high biological productivity and long residence times. In summary, I think the "frequency" decisions should be evaluated and explained in more detail.

Literature cited:

Coyle JJ, Buckler DR, Ingersoll CG, Fairchild JF, May TW. 1993. Effect of dietary selenium on the reproductive success of bluegills (Lepomis macrochirus). Environ Toxicol Chem 12:551-565.

de Rosemond SC, Liber K, Rosaasen A. 2005. Relationship between embryo selenium concentration and early life stage development in a white sucker (Catostomus commersoni) from a northern Canadian lake. Bull Environ Contam Toxicol 74:1134-1142.

Doroshov S, Van Eenennaam J, Alexander C, Hallen E, Bailey H, Kroll K, Restrepo C. 1992. Development of water quality criteria for resident aquatic species of the San Joaquin River. University of California, Davis.

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

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

Gillespie RB, Baumann PC. 1986. Effects of high tissue concentrations of selenium on reproduction by bluegills. Trans Am Fish Soc 115:208-213.

Hamilton SJ, Holley KM, Buhl KJ, Bullard FA. 2005a. Selenium impacts on razorback sucker, Colorado River, Colorado. II. Eggs. Ecotoxicol Environ Saf 61:32-43.

Hamilton SJ, Holley KM, Buhl KJ, Bullard FA. 2005. Selenium impacts on razorback sucker, Colorado River, Colorado. III. Larvae. Ecotoxicol Environ Saf 61:168-189.

Hermanutz RO, Allen KN, Roush TH, Hedtke SF. 1992. Effects of elevated selenium concentrations on bluegills (*Lepomis macrochirus*) in outdoor experimental streams. Environ Toxicol Chem 11:217-224.

Hermanutz RO, Allen KN, Detenbeck NE, Stephan CE. 1996. Exposure of bluegills (*Lepomis macrochirus*) to selenium in outdoor experimental streams. U.S. Environmental Protection Agency, Duluth, MN, USA.

Nakamoto RJH, T.J. 1992. Selenium and other trace elements in bluegills from agricultural return flows in the San Joaquin Valley, California. Arch Environ Contam Toxicol 22:88-98.

USEPA. 2012. Calculation of BLM fixed monitoring benchmarks for copper at selected monitoring sites in Colorado. Office of Water, USEPA. 820R12009.

PART II: FISH TISSUE CRITERION ELEMENTS DERIVATION: DERIVATION OF FISH EGG-OVARY, WHOLE BODY AND MUSCLE CRITERION ELEMENT(S)

EPA is requesting a technical review of the methods and procedures used to derive a chronic selenium criterion based on an egg-ovary concentration, as well as its translation to a criterion element applicable to whole-body and muscle tissue. Please address the following questions:

1. Please comment on EPA's use of the effects concentration 10^{th} centile (EC_{10}) as the measurement endpoint for the fish reproductive toxicity studies used to derive the egg-ovary element.

The draft AWQC document notes that "an EC10 was determined to be a more appropriate endpoint for tissue-based criteria given the nature of exposure and effects for this bioaccumulative chemical. EC20s have historically been used in the derivation of EPA criteria applicable to the water medium. While water concentrations may vary rapidly over time, tissue concentrations of bioaccumulative chemicals are expected to vary gradually. Thus, where concentrations of selenium in fish tissue approach an effect threshold, there is potential for sustained impacts on aquatic systems, relative to chemicals that are not as bioaccumulative."

I agree with this logic for using the EC10 as the measurement endpoint for tissue-based toxicity values, where this effects statistic can be derived. I also agree with the use of an EC10 rather than a no-observed-effect concentration (NOEC), lowest-observed-effect concentration (LOEC), or geometric mean of the two, for the reasons discussed in the draft AWQC document.

2. Data used to derive the final chronic egg-ovary criterion element were differentiated based on the type of effect (reproductive vs. non-reproductive effects). Acceptable chronic toxicity data on fish reproductive effects are available for a total of nine fish genera. The genus Sensitivity Distribution (SD) is predominantly populated with data on fish genera because field evidence demonstrates that fish communities can be affected by selenium even when there is no observable change in the invertebrate community diversity and abundance. As a result, decades of aquatic toxicity research have focused primarily on fish. Available field and laboratory studies indicate that invertebrates are more tolerant to selenium than most of the tested fish species (Criteria document, Table 6c, Section 4.1.2). The data set used to derive the selenium criterion marks a change from the traditional method used to derive water quality criteria that requires toxicity tests with aquatic organisms from 8 phylogenetically distinct taxa (including three vertebrate and five invertebrate genera) in order to derive aquatic life criteria (Stephan et al., 1985).

    a. Given selenium's more taxon-specific and life stage-specific toxicity, please comment on EPA's use of the available data to derive the egg-ovary tissue element.

I agree with the EPA's approach of only considering fish data in the genus sensitivity distribution as fish are the most sensitive aquatic taxa (although the sensitivity of amphibians relative to fish is still uncertain).

There is a fundamental difference in a criterion that is based on an internal organism concentration versus an external environmental concentration (such as a water concentration). If fish are accepted to be the most sensitive taxa, and if selenium criteria are to be based on the selenium concentration in fish tissue (either eggs/ovaries or whole body), then the toxicity data and genus sensitivity distribution need to necessarily be based only on selenium concentrations in fish tissue. Development of a tissue-based genus sensitivity distribution that includes toxicity data for other taxa would not be relevant to the application of any criterion that could be derived using such an approach.

b. Given the greater general sensitivity of oviparous fish to selenium compared to aquatic invertebrates, please comment on the appropriateness of EPA's fish tissue-based criterion for affording protection to the aquatic community as a whole (e.g., including invertebrates).

Although it has perhaps not been rigorously evaluated at all levels of food chain structure and function, field data indicates that adverse Se-related effects on fish can occur when there is no evidence of effects to food chain organism communities, including invertebrates. Selenium trophic transfer factors (TTFs) for invertebrates-to-fish typically average about 1 for whole body Se concentrations in fish and ≥2 for egg/ovary Se concentrations in fish (with the latter being more variable). Thus, a whole body Se criterion of 8.1 mg/kg dw and an egg/ovary Se criterion of 15.2 mg/kg dw may, on average, both be associated with an invertebrate Se concentration of about 8 mg/kg dw.

Based on a review of Se toxicity to invertebrate taxa, deBruyn and Chapman (2007) identified two studies in which whole body invertebrate Se concentrations of <8 mg/kg dw were associated with adverse effects. Both of these studies were based on growth effects in larval midges (*Chironomus decorus*). deBruyn and Chapman (2007) reported an EC40 of 1.0 mg/kg dw from Alaimo et al. (1994) and an EC15 and EC46 of 2.6 and 4.1 mg/kg dw, respectively, from Malchow et al. (1995). However, in Alaimo et al. (1994), Se was below the detection limit in the treatment with a 40% reduction in growth relative to the control, which suggests the growth reduction was due to other factors. In Malchow et al. (1995), whole-body Se LOECs of 2.6 and 4.1 mg/kg dw in midges were observed after 96-hr exposures. It is unclear whether growth effects would be related to tissue concentrations under such a short exposure period, but perhaps the water concentrations themselves (10 µg/L of either selenate or selenite) were directly responsible for the reduced growth. More recent data for a mayfly (*C. triangulifer*) suggest that the whole-body Se toxicity threshold for this species is also >8 mg/kg dw (Conley et al. 2009, 2011, 2013).

Overall, in my opinion, the above provides support that a fish tissue-based Se criterion should ensure protection of the aquatic community as a whole, including invertebrates.

Literature cited:

Alaimo J, Ogle RS, Knight AW. 1994. Selenium uptake by larval *Chironomus decorus* from a *Ruppia maritima*-based benthic/detrital substrate. Arch Environ Contam Toxicol 27:441-448.

Conley JM, Funk DH, Buchwalter DB. 2009. Selenium bioaccumulation and maternal transfer in the mayfly *Centroptilum triangulifer* in a life-cycle, periphyton-biofilm trophic assay. Environ Sci Technol 43:7952-7957.

Conley JM, Funk DH, Cariello NJ, Buchwalter DB. 2011. Food rationing affects dietary selenium bioaccumulation and life cycle performance in the mayfly *Centroptilum triangulifer*. Ecotoxicology 20:1840-1851.

Conley JM, Funk DH, Hesterberg DH, Hsu L-C, Kan J, Liu Y-T, Buchwalter DB. 2013. Bioconcentration and biotransformation of selenite versus selenate exposed periphyton and subsequent toxicity to the

mayfly *Centroptilum triangulifer*. Environ Sci Technol 47:7965-7973.

deBruyn AMH, Chapman PM. 2007. Selenium toxicity to invertebrates: Will proposed thresholds for toxicity to fish and birds also protect their prey? Environ Sci Technol 41:1766-1770.

Malchow DE, Knight AW, Maier KJ. 1995. Bioaccumulation and toxicity of selenium in *Chironomus decorus* larvae fed a diet of seleniferous *Selenastrum capricornutum*. Arch Environ Contam Toxicol 29:104-109.

c. With respect to the tests that quantified non-reproductive effects, did the EPA use that data to the best extent possible given its limitations (e.g., relevance compared to reproductive tests, and data quality concerns which increased uncertainty (e.g., Hamilton et al, 1990)?

Overall, I generally agree with the EPA's interpretation of the non-reproductive effects data and the draft whole-body Se criterion appears to be protective of the toxicity endpoints evaluated in those studies (at least the GMCVs reported in Table 17 of the draft AWQC document certainly are). The one study that could be interpreted somewhat differently is the juvenile Chinook salmon study conducted by Hamilton et al. (1990). The EPA derived whole-body Se EC10s of 7.355 and 11.14 mg/kg dw for juvenile growth based on a seleno-DL-methionine spiked diet and San Luis Drain (SLD)-spiked diet. For comparison, DeForest and Adams (2011) had derived a whole-body Se EC10 of 6.4 mg/kg dw based on the seleno-DL-methionine spiked diet, using a different concentration-response model (they excluded the SLD-spiked diet due to concerns associated with other contaminants). Overall, the model fit by the EPA to the data using TRAP appears to be quite good and the greater EC10 that they derived based on SLD-diet provides support that other contaminants did not adversely affect growth in the juvenile Chinook. Accordingly, I do not disagree with the SMCV (and GMCV) of 9.052 mg/kg dw that the EPA derived from juvenile Chinook salmon. This would also support that the draft whole-body Se criterion of 8.1 mg/kg dw based on reproductive effects would be protective against growth effects in juvenile Chinook.

Literature cited:

DeForest DK, Adams WJ. 2011. Selenium accumulation and toxicity in freshwater fishes. 193-229 in Beyer WN, Meador JP, eds. Environmental contaminants in biota: Interpreting tissue concentrations Second edition. CRC Press, Boca Raton, FL, USA.

Hamilton SJ, Buhl KJ, Faerber NL, Wiedmeyer RH, Bullard FA. 1990. Toxicity of organic selenium in the diet to chinook salmon. Environ Toxicol Chem 9:347-358.

d. EPA also rejected studies that used the injection route of exposure for selenium due to uncertainty related to uptake, distribution and metabolism/transformation kinetics when compared with the dietary and/or maternal transfer routes of exposure. Was this reasonable? Does the panel envision an appropriate and scientifically defensible use for this type of data? Please provide detailed comments.

In my opinion it was reasonable to exclude microinjection studies because there are sufficient questions as the environmental relevance of the exposure. For example, Linville (2006) exposed white sturgeon larvae to selenium using two different approaches: (1) by microinjection of L-selenomethionine into larval yolk sacs immediately after hatching and (2) by exposing parent females to dietary selenium (as selenized yeast) for up to six months before they deposited eggs (i.e., maternal transfer exposure). In larvae that received L-selenomethionine microinjections, mortality was a more sensitive endpoint than developmental-

related effects. In contrast, in the maternal transfer test, larval developmental effects was a more sensitive endpoint than larval mortality. Further the egg Se EC10 for white sturgeon was 15.8 mg/kg dw in the maternal transfer study versus 6.77 mg/kg dw in the microinjection study (as derived by Beckon [2012]). The microinjection methodology has not been validated in other studies and the results from Linville (2006) suggest that it is not an appropriate substitute for maternal transfer. Further, to my knowledge, studies on injection of Se into muscle tissues and subsequent maternal transfer of Se to the ovaries and eggs, and comparison to maternal transfer data following dietary Se exposures, have not been conducted.

(Although the data from Linville [2006] are sufficient to make some comparisons between maternal transfer and microinjection studies, the concentration-response data are too limited to derive an EC10 that would be considered reliable in a sensitivity distribution for criteria development. Further, the egg Se EC10 from the maternal transfer test was estimated from the larval Se EC10 using a regression relationship between egg and larval Se concentrations from a microinjection test.)

Literature cited:

Beckon WN. 2012. Evaluation of the toxicity of selenium to white and green sturgeon. U.S. Fish and Wildlife Service, Sacramento, CA.

Linville RG. 2006. Effects of excess selenium on the health and reproduction of white sturgeon (*Acipenser transmontanus*): Implications for San Francisco Bay-delta. Ph.D. Thesis, University of California, Davis. 232 pp.

3. Was the method (Section 4.1.5, 7.1.7) used to translate the fish egg-ovary criterion element into muscle and whole body criterions elements understandable, transparent and scientifically defensible? Was there sufficient data for making the translations for each element?

In general, I am hesitant about considering tissue-to-tissue Se relationships in order to estimate toxicity thresholds for one tissue based on measured concentrations in another tissue. However, the "EO/WB" ratios shown in Table 7a appear bracket the ratios typically observed, while still reflecting the variability observed between different species and families. The resulting draft whole-body Se criterion of 8.1 mg/kg dw is not inconsistent with other whole-body fish Se guidelines that have been recommended based on direct whole-body Se measurements. DeForest and Adams (2011), for example, recommended a whole-body fish Se guideline of 8.1 mg/kg dw following a different approach. However, per my above comment, I believe that the number of GMCVs should be 11 rather than 14 (or 12 if a recently conducted study for mountain whitefish were added to the sensitivity distribution.

In addition, for those species with measured Se concentrations in whole-body tissue or muscle, why not use the empirical measurements? For example, for Dolly Varden, McDonald et al. (2010) reported a whole body Se EC10 of 44 mg/kg dw based on the site-specific relationship between egg and WB Se in their study (this would not influence the draft whole-body Se criterion because *Salvelinus* is not among the four most sensitive genera, but it would be more accurate). Likewise, Coyle et al. (1993) and Hermanutz et al. (1992, 1996) report whole body Se concentrations in bluegills. This could be checked for other species as well.

Finally, perhaps it should be noted that, if possible or desired, site- and species-specific relationships between egg/ovary Se and whole-body or muscle Se could be derived and used in place of the draft criteria of 8.1 and 11.8 mg/kg dw.

Literature cited:

Coyle JJ, Buckler DR, Ingersoll CG, Fairchild JF, May TW. 1993. Effect of dietary selenium on the

reproductive success of bluegills (*Lepomis macrochirus*). Environ Toxicol Chem 12:551-565.

DeForest DK, Adams WJ. 2011. Selenium accumulation and toxicity in freshwater fishes. 193-229 in Beyer WN, Meador JP, eds Environmental contaminants in biota: Interpreting tissue concentrations Second edition. CRC Press, Boca Raton, FL, USA.

Hermanutz RO, Allen KN, Roush TH, Hedtke SF. 1992. Effects of elevated selenium concentrations on bluegills (*Lepomis macrochirus*) in outdoor experimental streams. Environ Toxicol Chem 11:217-224.

Hermanutz RO, Allen KN, Detenbeck NE, Stephan CE. 1996. Exposure of bluegills (*Lepomis macrochirus*) to selenium in outdoor experimental streams. U.S. Environmental Protection Agency, Duluth, MN, USA.

McDonald BG, deBruyn AMH, Elphick JRF, Davies M, Bustard D, Chapman PM. 2010. Developmental toxicity of selenium to Dolly Varden char (*Salvelinus malma*). Environ Toxicol Chem 29:2800-2805.

PART III: EVALUATION OF THE TRANSLATION PROCEDURE TO DERIVE THE WATER COLUMN ELEMENT(S)

EPA is also requesting a technical review of the methods and procedures used to translate the egg-ovary element of the chronic selenium criterion to water-column elements. Relevant sections of the document include:

- A description of the method used to derive an equation to translate the egg-ovary element to a monthly water-column element in perennial (lentic and lotic) waters and an equation that can be used to convert the monthly water-column element to an intermittent water column element (Sections 3.8.3, 3.8.4, 4.2.1, 4.3, and Appendix G).

- An analysis of the translation equation precision using data obtained from published literature (Sections 7.2.1, 7.2.2, and Appendix H).

- A description of the method and data sources used to derive the translation equation parameters (Sections 4.2.2, 4.2.3, and Appendix B).

- A description of the method and data sources used to categorize waterbody types where a single water-column chronic criterion concentration value would be adequately protective in most circumstances (Section 4.2.4).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for established categories of waters (Section 4.2.5).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for intermittent discharges that may occur in lentic and lotic waterbodies (Section 4.3).

Please address the following questions:

1. Please comment on the scientific defensibility of EPA's translation equation method for translating the concentration of selenium in fish tissue to a concentration of selenium in the water-column. Please comment on major sources of uncertainty in applying the translation equation to different types of waterbodies (e.g., with differing retention times, water chemistries, and/or species present). Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

I believe that the EPA's translation method is not unreasonable, but I have three primary concerns: (1) TTFs and CFs derived for taxa from other studies are applied to sites regardless of whether those TTFs and CFs are reflective of site-specific trophic transfer data; (2) the EFs and TTFs are treated as constants regardless of exposure concentrations; and (3) the level of protection associated with the draft criteria is unclear. These are discussed further below (in response to questions 1 and 2).

Model for translating fish egg/ovary Se criterion to lentic and lotic water Se criteria is not always consistent with site-specific information:

The EPA identified sites where Se EFs could be calculated based on reported co-located Se concentrations in surface water and particulates (algae, detritus, sediment). Information on the fish species present at those sites was then used to develop food web models, which determined the CFs and TTFs that were then applied in translating from the draft fish egg/ovary Se criterion back to corresponding water Se concentrations. Site-specific food web information was used where reported, but the EPA mostly relied on the NatureServe database (http://www.natureserve.org) for information on the typical diet and/or eating habits of the fish at each site.

A limitation of this modeling approach is that it ignored site-specific information on Se bioaccumulation in fish and their diets. The EFs used were site-specific, but Se modeling up the rest of the food chain and into fish was based on assumed model parameters. This becomes particularly important when considering the data "drivers" for the draft lentic Se criterion of 1.3 µg/L. This value is driven almost exclusively by data for two reference lakes (Badin Lake and High Rock Lake, NC, USA). Badin Lake was reported to have a water Se concentration of 0.32 µg/L and High Rock Lake a water Se concentration of 0.67 µg/L (Lemly 1985). For comparison, the mean water Se concentrations translated from a fish egg/ovary Se criterion of 15.2 mg/kg dw were 0.54 µg/L for Badin Lake and 1.2 µg/L for High Rock Lake. The former falls between the water Se concentrations reported for these two reference lakes and the latter almost equals the draft lentic criterion of 1.3 µg/L. Since six fish species were assumed to represent each of these two sites, these two reference sites are the drivers for the draft lentic Se criterion of 1.3 µg/L.

In addition to two reference sites being the drivers for the draft lentic Se criterion of 1.3 µg/L, the model for translating a fish egg/ovary Se criterion of 15.2 µg/L to a water Se concentration does not appear to be correct for these two sites. Although fish egg/ovary Se concentrations were not reported for Badin Lake and High Rock Lake, muscle Se concentrations were. Those muscle Se concentrations were reported on a wet weight basis and converted to a dry weight basis by assuming a moisture content of 75%. The muscle-to-egg CFs reported in Table 12 of the draft AWQC document were then used to estimate fish egg Se concentrations. These estimated fish egg Se concentrations for the two reference sites were, on average, less than one-half of the draft fish egg/ovary Se criterion of 15.2 mg/kg dw. Further, the muscle Se concentrations at the references sites ranged from 2.3 to 5.8 mg/kg dw, which are well below the draft muscle Se criterion of 11.8 mg/kg dw. The above demonstrates that the food web model for these two reference sites does not accurately reflect Se bioaccumulation potential at these two sites and in fact greatly overestimates Se bioaccumulation potential.

Overall opinion on method for translating from a fish tissue criterion to water Se criteria:

In my opinion, the approach should rely more on empirical data in order to eliminate cases where the food web models do not reflect the site-specific data. One alternative approach is that described in DeForest et al. (2014). That approach was also based on multi-step Se partitioning, but rather than using EFs and TTFs, the empirical relationships between (1) water and particulate Se; (2) particulate and invertebrate Se; and (3) invertebrate and fish egg/ovary Se were used. Quantile regression was used to work backward from an egg/ovary Se threshold to conservative Se concentrations in lentic and lotic water bodies. This regression-based approach accounts for the breadth of data on Se enrichment and trophic transfer potential, which can essentially represent the bounds of Se bioaccumulation potential from water to fish eggs/ovaries. The regression-based approach also accounts for the slopes of the relationships between water and particulate Se, particulate and invertebrate Se, and invertebrate and fish Se. This would be one example of an alternative model that could be considered.

Level of protection associated with draft water selenium criteria unclear:

The draft lentic and lotic criteria are based on the 20th percentiles of the data points plotted in Fig. 11 of the draft AWQC document. Those data points in Fig. 11 are for individual fish species at a given site. For example, 18 of the 51 data points for lentic systems (35%) are for just three water bodies (six fish species per water body). It is unclear what the 20th percentiles of those lentic and lotic distributions are protective of, as they do not represent 20% protection of sites or 20% protection of fish species. The latter was presumably not the intent, as those levels of protection would not be acceptable for national AWQC recommendations.

Literature cited:

DeForest DK, Brix KV, Gilron G, Hughes SA, Tear LM, Elphick JR, Rickwood CJ, DeBruyn AMH, Adams WJ. 2014. Selenium partitioning between water and fish tissue in freshwater systems: Development of water-based selenium screening guidelines. http://www.namc.org/docs/Selenium%20Integrated%20Report%20-%20Final%20(2014-05-20).pdf

Lemly AD. 1985. Toxicology of selenium in a freshwater reservoir: Implications for environmental hazard evaluation and safety. Ecotoxicol Environ Saf 10:314-338.

2. Regarding the trophic transfer factor (TTF) values, did EPA use a scientifically defensible method to derive the TTF values (p. 71-77 of the criteria document)? Were the exclusion criteria, (pp. 71-77 of the criteria document) developed by EPA to screen the available data applied in a consistent and scientifically defensible manner? In particular, EPA noticed that application of the exclusion criteria resulted in TTF values for aquatic insect larvae that differ from other published values. Given this, are you aware of any other methods of screening data that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included, if appropriate? Please provide detailed comments.

Overall, I generally agree with how the EPA derived TTFs from either physiological coefficients or from field data. Following are specific comments.

TTFs from empirical measurements in laboratory studies:

Laboratory-based TTFs were calculated from physiological coefficients (AE, IR, ke), but it does not appear that TTFs were calculated from laboratory data in which Se concentrations were empirically measured in invertebrates or fish and their diets. This approach is analogous to the field-based TTFs calculated by the EPA, but there is less uncertainty in the dietary Se concentration because the dietary Se

concentration is known in laboratory studies. Is there a reason why these studies were not considered?

TTFs are not constants across exposure concentrations:

As previously noted, one potential limitation of the modeling approach is that TTFs tend to be inversely related to exposure concentration (i.e., TTFs are inversely related to the corresponding dietary Se concentration). However, the TTFs in the model used by the EPA are constants that are specific to the exposure concentration in the test from which they were derived. The EPA did note, on p. 74, that the "distribution of ratios could be biased high toward larger values if the data are obtained from aquatic systems with low selenium concentrations" and on p. 75 a regression-based approach was considered. EPA ultimately used what was described as a hybrid approach, in which ordinary least squares (OLS) linear regression was used to confirm that a significant ($p \leq 0.05$) and positive relationship was observed, and then the median of individual ratios was used to estimate central tendency and avoid bias from systems with very low or very high selenium concentrations. This helps to partially address the issue, but a regression-based approach may still be more appropriate (see previous comment).

TTFs for insect larvae:

The draft AWQC document includes Se TTFs of 1.97 for a dragonfly (Anisoptera), 2.88 for a damselfly (Coenagrionidae), 1.28 for a mayfly (*Centroptilum triangulifer*), 1.90 for a midge (Chironomidae), and 1.48 for a corixid (Corixidae).

- **Dragonflies and damselflies:** The dragonfly and damselfly TTFs do not always appear to be calculated as described. On p. B-63 it is noted that the Se concentration in dragonfly and damselfly food is the median selenium concentration in all invertebrate tissues that co-occur with an Odonate species. For Site 29 in Birkner (1978), however, only corixids are considered in the damselfly diet, even though data for chironomids are available. The damselfly Se concentration at this site was 55.0 mg/kg dw and the corixid Se concentration was 29.4 mg/kg dw, which resulted in a TTF of 1.87. However, if chironomids were also considered part of the diet, which had a Se concentration of 58.2 mg/kg dw, the median Se concentration in the damselfly diet would be 43.8 mg/kg dw and the TTF would be 1.26. I recommend that the EPA double-check the dietary data used to calculated the TTFs for these taxa.
- **Mayfly (*C. triangulifer*):** The Se TTF of 1.28 for this species may be too low. This value was based on biokinetic data from Riedel and Cole (2001). However, empirical laboratory data from Conley et al. (2009, 2011, 2013) indicate that the Se TTF may range from about 1-3, with a mean of about 2 depending on exposure and test conditions. I recommend that the EPA consider these studies, which may result in a higher Se TTF for *C. triangulifer*.
- **Midges (Chironomidae):** The Se TTF of 1.90 for this taxa may be high when considering laboratory-based TTFs, for which the dietary Se concentration is known. Based data for chironomids from Malchow et al. (1995) and Rickwood and Jatar (2013), mean and maximum Se TTFs are 0.3 and 1.4. The chironomid Se TTFs derived from field data by the EPA include dietary Se assumptions that may underestimate the dietary Se concentration and result in relatively high Se TTFs. For example, the TTFs from Saiki et al. (1993) average 1.0 when a detritus-based food chain is assumed, as suggested by the study authors. I recommend that the EPA consider the dietary assumptions in the field studies in light of the laboratory data.
- **Corixids (Corixidae):** Additional Se TTF data for corixids are available from a laboratory study with *Trichorixa reticulata* (water boatman). In this study, the TTF was very high (32.6) in the control with a low dietary Se concentration of <0.1 mg/kg dw, but then TTFs were <1 at dietary Se concentrations of about 6 to 86 mg/kg dw. It is recommended that this laboratory study be included in deriving the corixid and be used to check the dietary assumptions in the field studies.

Additional potentially relevant TTF data sources:

Laboratory data:

- Conley et al. (2009, 2011, 2013) - *Centroptilum triangulifer* (mayfly)
- Malchow et al. (1995) - *Chironomus decorus* (chironomid)
- Rickwood and Jatar (2013) - *Chironomus dilutus* (chironomid)
- Besser et al. (1989) - *Daphnia magna* (cladoceran)
- Besser et al. (1993) - *Daphnia magna* (cladoceran)
- Guan and Wang (2004) - *Daphnia magna* (cladoceran)
- Thomas et al. (1999) - *Trichorixa reticulata* (water boatman)

Literature cited:

Besser JM, Huckins JN, Little EE, La Point TW. 1989. Distribution and bioaccumulation of selenium in aquatic microcosms. Environ Pollut 62:1-12.

Besser JM, Canfield TJ, La Point TW. 1993. Bioaccumulation of organic and inorganic selenium in a laboratory food chain. Environ Toxicol Chem 12:57-72.

Conley JM, Funk DH, Buchwalter DB. 2009. Selenium bioaccumulation and maternal transfer in the mayfly *Centroptilum triangulifer* in a life-cycle, periphyton-biofilm trophic assay. Environ Sci Technol 43:7952-7957.

Conley JM, Funk DH, Cariello NJ, Buchwalter DB. 2011. Food rationing affects dietary selenium bioaccumulation and life cycle performance in the mayfly *Centroptilum triangulifer*. Ecotoxicology 20:1840-1851.

Conley JM, Funk DH, Hesterberg DH, Hsu L-C, Kan J, Liu Y-T, Buchwalter DB. 2013. Bioconcentration and biotransformation of selenite versus selenate exposed periphyton and subsequent toxicity to the mayfly *Centroptilum triangulifer*. Environ Sci Technol 47:7965-7973.

Guan R, Wang W-X. 2004. Dietary assimilation and elimination of Cd, Se, and Zn by *Daphnia magna* at different metal concentrations. Environ Toxicol Chem 23:2689-2698.

Malchow DE, Knight AW, Maier KJ. 1995. Bioaccumulation and toxicity of selenium in *Chironomus decorus* larvae fed a diet of seleniferous *Selenastrum capricornutum*. Arch Environ Contam Toxicol 29:104-109.

Rickwood CJ, Jatar M. 2013. Investigation into the fate and effects of selenium on the life-cycle of a benthic invertebrate (*Chironomus dilutus*). CanmetMINING, Project: 603994. Natural Resources Canada (NRCan), Ottawa, Canada.

Riedel GF, Cole L. 2001. Selenium cycling and impact in aquatic ecosystems: Defining trophic transfer and water-borne exposure pathways. Chapter 3 in EPRI Report 2001. EPRI, Palo Alto, CA.

Thomas BV, Knight AW, Maier KJ. 1999. Selenium bioaccumulation by the water boatman *Trichocorixa reticulata* (Guerin-Meneville). Arch Environ Contam Toxicol 36:295-300.

3. Regarding the conversion factor (*CF*) values used, did EPA use an appropriate and scientifically defensible method to derive those values (p. 78-79 of the criteria document and Appendix B)? Are you aware of any other methods that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included? Please provide detailed comments.

I think the EPA used a reasonable approach for deriving CFs. As a partial confirmation of those values, fish species for which diet-to-egg TTFs can be derived could be compared to the combined CFs and TTFs values.

4. Regarding the derivation of enrichment factor (*EF*) values, was the method EPA used to screen data from the literature applied appropriately and consistently (see inclusion/exclusion criteria on p. 71-77 of the criteria document)? Was the method for deriving *EF* values applied to those data in a consistent manner so as to derive *EF* values for selected waters in a scientifically defensible manner? Is the method that EPA used to establish the lentic and lotic categories for *EF* values reasonable given the available data? Are you aware of other methods or relevant data the EPA should consider? Please provide detailed comments.

Overall, I believe that the EPA used a reasonable approach in calculating EF values. However, I do not necessarily agree that Se concentrations should be available for at least two particulate types in order to derive an EF. Periphyton, for example, may be the dominant particulate in certain lotic systems and in my opinion such data should be included. I do agree that Se concentrations in sediment alone is insufficient for deriving EF values. I have greater reservations in how the EFs (and CFs and TTFs) were ultimately used to translate from the draft fish egg/ovary Se criterion to water Se criteria.

Potential sources of additional EF data may include:

Bowie GL, Sanders JG, Riedel GF, Gilmour CC, Breitburg DL, Cutter GA, Porcella DB. 1996. Assessing selenium cycling and accumulation in aquatic ecosystems. Water Air Soil Pollut 90:93-104.

Casey R. 2005. Results of aquatic studies in the McLeod and Upper Smoky River systems. Alberta Environment. 64 pp.

Fan TW-M, Swee JT, Hinton DE, Higashi RM. 2002. Selenium biotransformations into proteinaceous forms by foodweb organisms of selenium-laden drainage waters in California. Aquat Toxicol 57:65-84.

Greater Yellowstone Coalition. 2005. Technical Reports on selenium concentrations in water, macrophytes, macroinvertebrates, and fish.

Hamilton SJ, Buhl KJ. 2003a. Selenium and other trace elements in water, sediment, aquatic plants, aquatic invertebrates, and fish from streams in southeastern Idaho near phosphate mining operations: September 2000. US Geological Survey. 64 pp.

Hamilton SJ, Buhl KJ. 2003b. Selenium and other trace elements in water, sediment, aquatic plants, aquatic invertebrates, and fish from streams in southeastern Idaho near phosphate mining operations: May 2001. US Geological Survey. 61 pp.

Hamilton SJ, Buhl KJ, Lamothe PJ. 2002. Selenium and other trace elements in water, sediment, aquatic plants, aquatic invertebrates, and fish from streams in southeastern Idaho near phosphate mining operations: June 2000. USGS, Yankton, SD and Denver, CO. 72 pp.

McDonald LE, Strosher MM. 1998. Selenium mobilization from surface coal mining in the Elk River basin, British Columbia: A survey of water, sediment and biota. Ministry of Environment, Land and Parks, Cranbrook, BC. 46 pp. + appendices.

Orr PL, Guiguer KP, Russel CK. 2006. Food chain transfer of selenium in lentic and lotic habitats of a western Canadian watershed. Ecotoxicol Environ Saf 63:175-188.

Orr PL, Wiramanaden CIE, Paine MD, Franklin W, Fraser C. 2012. Food chain model based on field data to predict westslope cutthroat trout (*Oncorhynchus clarkii lewisi*) ovary selenium concentrations from water selenium concentrations in the Elk Valley, British Columbia. Environ Toxicol Chem 31:672-680.

Presser TS, Luoma SN. 2009. Modeling of selenium for the San Diego Creek watershed and Newport Bay, California. US Geological Survey, Open-File Report 2009-1114. 48 pp.

Zhang Y, Moore JN. 1996. Selenium fractionation and speciation in a wetland system. Environ Sci Technol 30:2613-2619.

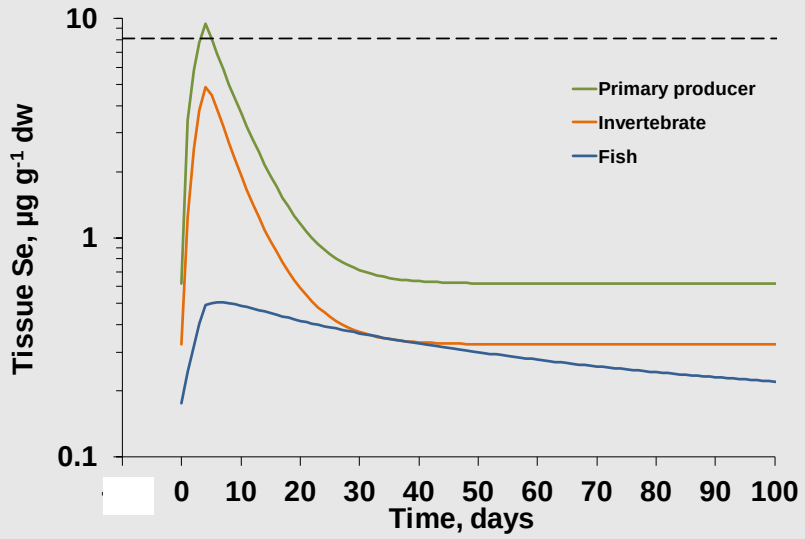
5. Please comment on the scientific defensibility of EPA's conversion of the selenium fish tissue – water translation equation into an equation that allows for calculation of a criterion for waters that may be subject to intermittent discharges of selenium. Please comment on major sources of uncertainty in this approach. Is this method appropriate, given the bioaccumulative nature of selenium? Please comment on the uncertainty associated with the application of this conversion equation to intermittent discharges that may occur in different types of waterbodies and/or in different locations, particularly with respect to loads transported to potentially more sensitive aquatic systems. Does the method employed result in criteria that are similarly protective to the 30-day chronic criterion? Are there any other models or approaches that EPA should consider that would reduce this uncertainty? Please provide detailed comments.

I am not sure that the criterion equation for intermittent dischargers is meaningful, as it is basically a mathematical manipulation and does not in any way account for selenium uptake and elimination kinetics. An alternative approach that the EPA may want to consider is based on biokinetic modeling, such as that described in Brix and DeForest (2008). The method they described was based on modeling of a food chain comprised of periphyton, an invertebrate (mayfly), and a fish (fathead minnow). Inputs to the model include the background water Se concentration, the magnitude of an intermittent Se pulse, and the duration of the Se pulse. This provides a tool for evaluating whether a Se pulse of a given magnitude and duration could result in exceedance of a whole-body fish Se criterion, or short-term Se criteria could be derived for given short-term durations.

For a comparison of the biokinetic-based approach to the intermittent criterion equation in the draft AWQC document, I assumed that the background water Se concentration is 1 µg/L, the lotic criterion is 4.8 µg/L, and the number of days elevated is 4. The intermittent criterion would be 29.5 µg/L. Just as an example, if a lotic food chain consisting of periphyton→mayflies→fathead minnows were assumed, a 4-d pulse of 29.5 µg Se/L would not be nearly sufficient to reach a whole body Se concentration of 8.1 mg/kg dw (Fig. 2). There is a rapid increase in predicted Se concentrations in periphyton and mayflies and then a rapid elimination, but uptake is slower in fathead minnows.

In my opinion, a biokinetic-based modeling approach would be more appropriate for deriving acute or intermittent water Se criteria.

Fig. 2. Example of modeled selenium uptake and elimination in a model food chain exposed to a 4-d pulse of 29.5 µg Se/L.



Literature cited:

Brix KV, DeForest DK. 2008. Selenium. Pages 123-172 in Gensemer RW, Meyerhoff RD, Ramage KJ, Curely EF, eds. Relevance of ambient water quality criteria in ephemeral and effluent-dependent watercourses of the arid western US. SETAC Press, Pensacola, FL, USA.

PART IV: SIGNIFICANCE OF SCIENTIFIC VIEWS FROM THE PUBLIC/STAKEHOLDERS

EPA will also be providing scientific views and other comments from stakeholders and the public received via the public docket to the peer review panel. Although EPA will be providing the full contents from the docket, EPA is only requesting a review of any scientific views/public comments that may be of technical significance to the selenium criterion.

1. Has the peer review panel identified any scientific views from the public or stakeholders as being technically significant to the draft of the selenium criterion going forward; that is, has information or data been introduced during the comment period that would change the scientific direction of the criterion? Is there any information or data that may refine or enhance the scientific defensibility of this criterion that EPA should consider further? Please provide detailed comments on specific issues of technical significance or refinement.

A substantial number of comments from stakeholders and the public were provided. These comments covered a large variety of topics and were often conflicting. I did not identify any comments that would lead me to think that the scientific direction of the criterion should be changed. The comments relative to interpretation of toxicity studies and derivation of EC10 values should all be carefully reviewed by the EPA, as some suggested that certain EC10 values should be lowered and other suggested they should be raised (although I personally believe that the GMCVs values derived by the EPA were generally conservative, especially for *Salmo* and *Lepomis*). Aside from the technical comments and disagreements that are related to magnitudes of the various Se criterion elements, it appears that there is a desire (or need) for the EPA to more clearly define how the draft Se criteria should be implemented by the states. Perhaps

case studies could be provided as examples? It is also apparent that the basis of the intermittent criterion, and its relationship to an acute criterion (if there is a relationship), needs to be more clearly explained. Although some comments seem to agree that an acute Se criteria is not necessary any longer, there does still appear to be a need for acute Se criteria from the perspectives of certain states. Finally, again related to implementation, is the question of whether the lotic and lentic water Se criteria can be replaced by a different metric, such as residence time. In my opinion, the latter would be worthy of further consideration by the EPA, although I wonder whether more reliable categories could be developed based on existing datasets.

PEER REVIEW COMMENTS FROM

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External Peer Review of the Draft Aquatic Life Ambient Water Quality Criterion for Selenium – Freshwater 2014

Responses to Charge Questions by Dr. Nicholas S. Fisher

PART I: OVERARCHING QUESTIONS

1. Please comment on the overall clarity of the document and construction of the criterion statement with its multiple elements.

Reasonably clear, although some phrases and terms need further clarification.

2. EPA has developed a tiered selenium criterion with four elements, with the fish tissue elements having primacy over the water-column elements, and the egg-ovary element having primacy over any other element. Inclusion of the fish whole-body or fish muscle element into the selenium criterion ensures the protection of aquatic life when fish egg or ovary tissue measurements are not available, and inclusion of the water column elements ensures protection when fish tissue measurements are not available

    a. Please comment on the tiered construction of the selenium chronic criterion; is it logical, and scientifically defensible as it applies to protection of freshwater aquatic life:

        i. That is, is the primacy of the egg-ovary element over the other elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

The tiered construction makes sense for most natural conditions, but not when acutely high Se levels are present (e.g., Kesterson reservoir). But for most sublethal concentrations this approach makes sense as a general approach for the EPA to adopt.

        ii. Is the primacy of the whole-body/fish muscle element over the water column elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

This approach is wholly justifiable because Se is accumulated by animals almost exclusively through diet rather than directly from the dissolved phase in ambient water. In fact, Se and perhaps methylmercury would be extreme examples in which this approach is appropriate.

        iii. Please comment on the scientific uncertainty that may be associated with this tiered approach? Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

The EPA can provide further levels of uncertainty with regard to toxicity associated with fish egg/ovary contamination. How many studies is this approach ultimately reliant upon? The report is based on a limited number of studies, but more studies are warranted before we can be assured that this approach is rock-solid.

iv. Are the draft recommended magnitude, duration, and frequency for each criterion element scientifically sound and appropriate? Please provide detailed comments.

I do not see obvious errors in their approach.

PART II: FISH TISSUE CRITERION ELEMENTS DERIVATION: DERIVATION OF FISH EGG-OVARY, WHOLE BODY AND MUSCLE CRITERION ELEMENT(S)

EPA is requesting a technical review of the methods and procedures used to derive a chronic selenium criterion based on an egg-ovary concentration, as well as its translation to a criterion element applicable to whole-body and muscle tissue. Please address the following questions:

1. Please comment on EPA's use of the effects concentration 10^{th} centile (EC_{10}) as the measurement endpoint for the fish reproductive toxicity studies used to derive the egg-ovary element.

Strikes me as rather arbitrary.

2. Data used to derive the final chronic egg-ovary criterion element were differentiated based on the type of effect (reproductive vs. non-reproductive effects). Acceptable chronic toxicity data on fish reproductive effects are available for a total of nine fish genera. The genus Sensitivity Distribution (SD) is predominantly populated with data on fish genera because field evidence demonstrates that fish communities can be affected by selenium even when there is no observable change in the invertebrate community diversity and abundance. As a result, decades of aquatic toxicity research have focused primarily on fish. Available field and laboratory studies indicate that invertebrates are more tolerant to selenium than most of the tested fish species (Criteria document, Table 6c, Section 4.1.2). The data set used to derive the selenium criterion marks a change from the traditional method used to derive water quality criteria that requires toxicity tests with aquatic organisms from 8 phylogenetically distinct taxa (including three vertebrate and five invertebrate genera) in order to derive aquatic life criteria (Stephan et al., 1985).

    a. Given selenium's more taxon-specific and life stage-specific toxicity, please comment on EPA's use of the available data to derive the egg-ovary tissue element.

I have no particular insight on this issue.

    b. Given the greater general sensitivity of oviparous fish to selenium compared to aquatic invertebrates, please comment on the appropriateness of EPA's fish tissue-based criterion for affording protection to the aquatic community as a whole (e.g., including invertebrates).

Until we find more Se-sensitive groups of freshwater animals than fish, the fish tissue-burden approach seems warranted.

c. With respect to the tests that quantified non-reproductive effects, did the EPA use that data to the best extent possible given its limitations (e.g., relevance compared to reproductive tests, and data quality concerns which increased uncertainty (e.g., Hamilton et al., 1990)?

I'm not sure.

d. EPA also rejected studies that used the injection route of exposure for selenium due to uncertainty related to uptake, distribution and metabolism/transformation kinetics when compared with the dietary and/or maternal transfer routes of exposure. Was this reasonable? Does the panel envision an appropriate and scientifically defensible use for this type of data? Please provide detailed comments.

It is hard to argue on behalf of egg injection studies in favor of dietary uptake (the obviously more natural process) studies. This is particularly the case if the Se contents of the tissues and eggs are measured during the dietary exposure.

3. Was the method (Section 4.1.5, 7.1.7) used to translate the fish egg-ovary criterion element into muscle and whole body criterions elements understandable, transparent and scientifically defensible? Was there sufficient data for making the translations for each element?

It seemed reasonably clear to me.

PART III: EVALUATION OF THE TRANSLATION PROCEDURE TO DERIVE THE WATER COLUMN ELEMENT(S)

EPA is also requesting a technical review of the methods and procedures used to translate the egg-ovary element of the chronic selenium criterion to water-column elements. Relevant sections of the document include:

- A description of the method used to derive an equation to translate the egg-ovary element to a monthly water-column element in perennial (lentic and lotic) waters and an equation that can be used to convert the monthly water-column element to an intermittent water column element (Sections 3.8.3, 3.8.4, 4.2.1, 4.3, and Appendix G).

- An analysis of the translation equation precision using data obtained from published literature (Sections 7.2.1, 7.2.2, and Appendix H).

- A description of the method and data sources used to derive the translation equation parameters (Sections 4.2.2, 4.2.3, and Appendix B).

- A description of the method and data sources used to categorize waterbody types where a single water-column chronic criterion concentration value would be adequately protective in most circumstances (Section 4.2.4).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for established categories of waters (Section 4.2.5).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for intermittent discharges that may occur in lentic and lotic waterbodies (Section 4.3).

Please address the following questions:

1. Please comment on the scientific defensibility of EPA's translation equation method for translating the concentration of selenium in fish tissue to a concentration of selenium in the water-column. Please comment on major sources of uncertainty in applying the translation equation to different types of waterbodies (e.g., with differing retention times, water chemistries, and/or species present). Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

The EPA is justified in simplifying the bioaccumulation equations by eliminating the growth rate constant (g) because it is negligible compared to the loss rate constant of Se from aquatic animals. This is generally the case for most metals and metalloids, with some notable exceptions where the loss rate constants are very low (e.g., methylmercury). Their equations 2 and 3 (pages 64-65) have already been published, and the reference for this should be cited. (Reinfelder, J.R., N.S. Fisher, S.N. Luoma, J.W. Nichols, and W.-X. Wang. 1998. Trace element trophic transfer in aquatic organisms: a critique of the kinetic model approach. Science of the Total Environment 219: 117-135.) The authors should note that the loss rate constant of some contaminants can differ following uptake from the aqueous phase and uptake from diet---this is because the contaminant may deposit in different tissues from these two uptake routes. As such, the term ke should be converted to kef and kew (following uptake from food and uptake from water). For Se, fortunately, this correction is unlikely to be an important one because uptake from the aqueous phase (water) is negligible compared to dietary uptake. But strictly speaking, the mathematical expression (Eq. 2) should reflect two different loss rate constants.

By using tissue concentrations of Se in fish to calculate dissolved Se concentrations in ambient water, one must ultimately calculate the Se concentration in organisms at the base of the food chain, namely phytoplankton. This is because none of the animals in the food chain appreciably take up Se from the aqueous phase. The problem of inferring Se concentrations in water from phytoplankton Se concentrations is that the enrichment factors (or bioconcentration factors) of Se in phytoplankton can vary by up 2 or 3 orders of magnitude, depending on the type of phytoplankton that happen to be dominant in the water. Chlorophyceae (green algae), for example, bioconcentrate Se far less than diatoms, and so the variability in these calculations would depend heavily on which types of phytoplankton happen to be dominating the community, and this can change temporally and geographically.

2. Regarding the trophic transfer factor (*TTF)* values, did EPA use a scientifically defensible method to derive the *TTF* values (p. 71-77 of the criteria document)? Were the exclusion criteria, (pp. 71-77 of the criteria document) developed by EPA to screen the available data applied in a consistent and scientifically defensible manner? In particular, EPA noticed that application of the exclusion criteria resulted in *TTF* values for aquatic insect larvae that differ from other published values. Given this, are you aware of any other methods of screening data that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included, if appropriate? Please provide detailed comments.

I am more familiar with the marine literature and am not well-versed in the freshwater literature regarding Se TTF values.

3. Regarding the conversion factor (*CF*) values used, did EPA use an appropriate and scientifically defensible method to derive those values (p. 78-79 of the criteria document and Appendix B)? Are you aware of any other methods that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included? Please provide detailed comments.

See my response to question 2.

4. Regarding the derivation of enrichment factor (*EF*) values, was the method EPA used to screen data from the literature applied appropriately and consistently (see inclusion/exclusion criteria on p. 71-77 of the criteria document)? Was the method for deriving *EF* values applied to those data in a consistent manner so as to derive *EF* values for selected waters in a scientifically defensible manner? Is the method that EPA used to establish the lentic and lotic categories for *EF* values reasonable given the available data? Are you aware of other methods or relevant data the EPA should consider? Please provide detailed comments.

See my response to question 2.

5. Please comment on the scientific defensibility of EPA's conversion of the selenium fish tissue – water translation equation into an equation that allows for calculation of a criterion for waters that may be subject to intermittent discharges of selenium. Please comment on major sources of uncertainty in this approach. Is this method appropriate, given the bioaccumulative nature of selenium? Please comment on the uncertainty associated with the application of this conversion equation to intermittent discharges that may occur in different types of waterbodies and/or in different locations, particularly with respect to loads transported to potentially more sensitive aquatic systems. Does the method employed result in criteria that are similarly protective to the 30-day chronic criterion? Are there any other models or approaches that EPA should consider that would reduce this uncertainty? Please provide detailed comments.

See my response to question 1.

PART IV: SIGNIFICANCE OF SCIENTIFIC VIEWS FROM THE PUBLIC/STAKEHOLDERS

EPA will also be providing scientific views and other comments from stakeholders and the public received via the public docket to the peer review panel. Although EPA will be providing the full contents from the docket, EPA is only requesting a review of any scientific views/public comments that may be of technical significance to the selenium criterion.

1. Has the peer review panel identified any scientific views from the public or stakeholders as being technically significant to the draft of the selenium criterion going forward; that is, has information or data been introduced during the comment period that would change the scientific direction of the criterion? Is there any information or data that may refine or enhance the scientific defensibility of this criterion that EPA should consider further? Please provide detailed comments on specific issues of technical significance or refinement.

Some of the comments made about acute toxicity are valid, but are unlikely to be relevant to most real-

world situations. Note that acute toxicity can affect other than reproduction, but such effects are rarely seen (I think).

PEER REVIEW COMMENTS FROM

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External Peer Review of the Draft Aquatic Life Ambient Water Quality Criterion for Selenium – Freshwater 2014

Responses to Charge Questions by Dr. David M. Janz

PART I: OVERARCHING QUESTIONS

1. Please comment on the overall clarity of the document and construction of the criterion statement with its multiple elements.

The document is generally well-written and is based on a comprehensive evaluation of the extensive body of freshwater Se literature. This said, I found many typographical and other errors throughout the document, which I will address in a marked-up copy (Adobe would not let me use the edit text functions so I simply highlighted the text in yellow and provided a comment if necessary). There were also several areas that I believe require significant clarification, which I will address in my subsequent review comments found below.

I agree with the concept of the tiered criterion approach, particularly that tissue (i.e., ovary, egg, muscle, or whole-body)-based Se concentrations ([Se]) are key to accurately assess the toxicological risk posed to fishes, and that egg/ovary [Se] overrides/supersedes whole-body or muscle [Se]. However, I do not fully agree with the approach, in the absence of tissue [Se] data, that a water-column criterion will be protective of aquatic species. There are many examples of aquatic systems, due to their specific biogeochemistry, ecology, and physiology, where very low dissolved [Se] (i.e., less than the proposed criteria for lentic or lotic systems) results in toxicologically significant bioaccumulation in fishes and their prey, and elevated frequencies of larval abnormalities. I suggest that dissolved [Se] be used as a "trigger" to initiate further monitoring (i.e., collection of fishes to determine tissue [Se]). I also do not agree with the intermittent exposure criterion; it is unclear why it was developed, how it could be implemented consistently and reliably, and in general I think it just adds too much complexity to an already complex (indeed perhaps the most complex) water quality criterion.

These are my general comments, and more specific details can be found in my subsequent review comments.

2. EPA has developed a tiered selenium criterion with four elements, with the fish tissue elements having primacy over the water-column elements, and the egg-ovary element having primacy over any other element. Inclusion of the fish whole-body or fish muscle element into the selenium criterion ensures the protection of aquatic life when fish egg or ovary tissue measurements are not available, and inclusion of the water column elements ensures protection when fish tissue measurements are not available

    a. Please comment on the tiered construction of the selenium chronic criterion; is it logical, and scientifically defensible as it applies to protection of freshwater aquatic life:

        i. That is, is the primacy of the egg-ovary element over the other elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Yes, it has been clearly shown in the scientific literature that egg/ovary [Se] provides the greatest certainty in predicting the toxicological risk associated with Se exposure in fishes. This is because (a) embryo-larval

abnormalities are the most sensitive toxicological response, and (b) maternal transfer of Se to the eggs by adult female fishes provides the ultimate dose received by their offspring (i.e., during yolk resorption prior to swim-up). In addition, the frequency and severity of early life stage abnormalities caused by Se has clear ramifications for population dynamics; impaired recruitment of individuals into fish populations can alter demographics and ultimately result in extirpation. This is Ecotoxicology 101. Indeed, documented Se poisoning events (e.g., Belews Lake) provide some of the most convincing evidence of a cause-effect relationship between exposure to a toxic substance and resulting negative impacts on fish populations and communities. This is the goal of aquatic ecotoxicology: to protect populations and communities of organisms, not individuals.

ii. Is the primacy of the whole-body/fish muscle element over the water column elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Yes, in the absence of egg/ovary [Se], the next best thing is whole-body or muscle [Se]. Practically, whole-body or muscle samples are more reliably collected throughout the year since most adult female fishes do not have appreciable ovarian tissue mass during non-reproductive periods. This is especially true in small-bodied fishes. In addition, muscle tissue can be collected non-lethally in larger fishes, which may be particularly relevant to threatened species.

It is important to note that [Se] in ovarian tissue containing only primary oocytes or pre-vitellogenic ovarian follicles (i.e., during the non-reproductive period spanning most of the year in many fishes) will likely provide similar information on Se risk as whole-body or muscle [Se]. This is because the ultimate Se dose is maternally delivered to eggs during the period of vitellogenesis in fishes. Eggs will not be present in the ovary of most fish species for much of the year. During vitellogenesis (the period of egg "growth"), adult females synthesize the yolk precursor protein, vitellogenin, in their liver, where it is transported via the bloodstream to the ovary and taken up by growing (vitellogenic) ovarian follicles (eggs). Thus, the [Se] in the <u>liver</u> of adult female fishes may provide a better predictor of Se risk than whole-body or muscle [Se]. To be even more scientifically correct, it is the concentration of the seleno-amino acid, selenomethionine, in the liver of adult female fishes that is incorporated into vitellogenin in a non-specific, dose-dependent manner (replacing the amino acid methionine) that defines the ultimate dose of Se received by their offspring. For more details see the following paper, which was not cited in the EPA document:

Janz, D.M. 2012. Selenium. Pp. 327-374 In: C.W. Wood, A.P. Farrell and C.J. Brauner (Eds.) *Fish Physiology Vol 31A, Homeostasis and Toxicology of Essential Metals*. Elsevier, San Diego, CA.

Thus, I do not agree with the statement on page 27 (line 4) that "concentrations of Se in ovaries are considered equivalent to concentrations of Se in eggs…" because fish ovarian tissue during the non-reproductive phase contains somatic cells responsible for ovarian maturation processes (i.e., steroidogenic cells), and gametes (primary oocytes and pre-vitellogenic follicles), and the [Se] in these cells do not necessarily reflect the dose of Se that will be received by the eggs (i.e., in the yolk) during vitellogenesis. Further studies are needed to examine the relationship between [Se] in ovarian tissue vs. eggs. It is strongly suggested that the EPA inspect the ovary and egg data carefully and attempt to derive the potential relationship between [Se] in ovarian tissue vs. eggs.

iii. Please comment on the scientific uncertainty that may be associated with this tiered approach? Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

One major source of uncertainty is the translation of whole-body or muscle [Se] to egg/ovary [Se]. This relationship has been documented for 10 fish taxa in the document (Tables 7a and 8a). These ratios vary about two-fold among taxa (1.21-2.44 for EO:WB and 0.95-1.92 for EO:M). Not all fish taxa have been studied, and more work is needed in this area. Importantly, in a given fish species these ratios may vary considerably among aquatic ecosystems due to differences in the food web, biogeochemistry of Se, and other factors. These ratios may also vary across seasons. Nonetheless, the data sources, models and approaches used by the EPA to derive these ratios are valid; we simply need more data to more accurately define these conversion factors.

The major source of uncertainty in the tiered approach is the conversion of tissue (egg, ovary, muscle or whole-body) [Se] to water column [Se]. The approach used by the EPA is appropriate and uses, for the most part, the recent biodynamic modeling approach to derive water column [Se] from tissue [Se]. However, to use water column [Se] as a criterion in of itself in the absence of tissue [Se] data is a recipe for inappropriate conclusions, which may penalize industry (i.e., false positives) or cause harm to certain fish populations (i.e., false negatives). I strongly believe that water column [Se] should be used more as a "trigger" to initiate further monitoring that includes collection of fish for tissue [Se] determinations. I also think that a safety factor should be applied to the proposed 1.3 ug/L and 4.8 ug/L criteria for lentic and lotic systems, respectively, which would reduce these values as triggers for further ecosystem monitoring. There are many examples of lentic systems with < 1 ug/L dissolved [Se] where negative effects of Se on early life stage development of fishes have been demonstrated.

This is an appropriate place to discuss the problems with a crude classification of systems as lentic vs lotic. Many rivers in the USA are impounded, essentially creating lentic systems for a significant portion of their river-miles, although they would still be classified as lotic. I think the EPA needs to more clearly define these terms. One suggestion is to use water residence time and/or mean annual flow velocity as more quantitative descriptors. Many of the studies that have shown lower Se bioaccumulation in lotic systems have been conducted in fast-flowing mountain streams, creeks and rivers. To classify a river in the southern USA that has numerous dams as a lotic system does not make sense.

iv. Are the draft recommended magnitude, duration, and frequency for each criterion element scientifically sound and appropriate? Please provide detailed comments.

The egg/ovary criterion of 15.2 mg/kg relies strongly on the reassessment of brown trout data, in particular the Formation study. It seems that much of the issue is related to the lab accident where larval trout were removed from an aquarium due to a faulty standpipe. The EPA has chosen to assume the worst-case, that 100% of the fish that escaped were dead and/or deformed, resulting in an EC10 of 15.91 mg/kg egg. However it is plausible that certain of these fish were not dead or deformed, as discussed in certain public comment documents. The EPA has reanalyzed these data to account for different scenarios, and shown that the EC10 varies from 15.91 to 21.16 mg/kg egg. It seems to me that the 15.91 mg/kg EC10 may be overly conservative. Due to the lack of knowledge regarding the status of these escaped fish (dead, deformed, or healthy), perhaps the assumption could be made that 50% of the escaped fish were dead/deformed, and 50% were normal. This would only slightly increase the EC10 value from which the 15.2 mg/kg egg/ovary criterion is being largely driven. This is only a suggestion of a reasonable compromise given the diverse opinions on this lab occurrence.

For the egg/ovary criterion, the timing of fish sampling is absolutely critical, and the EPA provides no

guidance on sampling design for determining egg/ovary [Se] in the document. As discussed above in 2a(ii), it is the [Se] in eggs that drives early life stage toxicity, so adult female fish absolutely must be collected during the late vitellogenic or preovulatory periods of oogenesis for this criterion to be scientifically and toxicologically meaningful. Measuring [Se] in ovarian tissue during other periods of oogenesis will be much less informative (i.e., about as informative as muscle or whole-body [Se]). The EPA must provide guidance for specific times of the year to collect adult female fish for egg [Se] determinations. For synchronous spawning species (e.g., salmonids, esocids, catostomids, ictalurids), this will be a defined period of 1-2 months on average (usually spring). For asynchronous (batch) spawning species (e.g., cyprinids), this period will be less defined and will usually be 3-6 months (usually spring to late summer or early fall).

For the whole-body and muscle criteria, the EPA has used best available knowledge and approaches to derive these values, and they are of appropriate magnitude, duration and frequency. Collecting fish at any time of the year and determining whole-body or muscle [Se] will provide sufficient information on Se bioaccumulation. Although there will likely be some variation across seasons, due to prey availability, temperature and other factors, this approach should work.

PART II: FISH TISSUE CRITERION ELEMENTS DERIVATION: DERIVATION OF FISH EGG-OVARY, WHOLE BODY AND MUSCLE CRITERION ELEMENT(S)

EPA is requesting a technical review of the methods and procedures used to derive a chronic selenium criterion based on an egg-ovary concentration, as well as its translation to a criterion element applicable to whole-body and muscle tissue. Please address the following questions:

1. Please comment on EPA's use of the effects concentration 10^{th} centile (EC_{10}) as the measurement endpoint for the fish reproductive toxicity studies used to derive the egg-ovary element.

The EC10 is absolutely the appropriate endpoint for early life stage toxicity in fish to be used to derive the egg/ovary criterion. This is due to the very steep dose-response relationships observed for larval abnormalities/mortality as a function of egg [Se]. Thus, EC10 provides a toxicologically relevant threshold for appearance of such toxicities, that is, only a marginal increase in egg [Se] will result in a much greater frequency of toxicity. In addition, the main alternative endpoint (EC20) will not differ greatly from EC10 for a given species due to this steep dose-response relationship.

Something the EPA should consider when developing the genus sensitivity distribution is the nature of the experiment for each taxa (lab- vs. field-based). In lab studies, adult female fish are most commonly exposed to selenomethionine (SeMet), which is valid because it is the dominant Se species (60-80% of total Se) found in organisms throughout food webs, particularly at higher trophic levels. In field studies, fish are exposed to SeMet and several other selenium species that likely vary in their toxicity, and in fact are likely less toxic than SeMet. Thus, lab exposures using pure SeMet may overestimate toxicity (i.e., generate lower EC10 values) compared to real-world exposures.

2. Data used to derive the final chronic egg-ovary criterion element were differentiated based on the type of effect (reproductive vs. non-reproductive effects). Acceptable chronic toxicity data on fish reproductive effects are available for a total of nine fish genera. The genus Sensitivity Distribution (SD) is predominantly populated with data on fish genera because field evidence demonstrates that fish communities can be affected by selenium even when there is no observable change in the invertebrate community diversity and abundance. As a result, decades of aquatic toxicity research have focused

primarily on fish. Available field and laboratory studies indicate that invertebrates are more tolerant to selenium than most of the tested fish species (Criteria document, Table 6c, Section 4.1.2). The data set used to derive the selenium criterion marks a change from the traditional method used to derive water quality criteria that requires toxicity tests with aquatic organisms from 8 phylogenetically distinct taxa (including three vertebrate and five invertebrate genera) in order to derive aquatic life criteria (Stephan et al., 1985).

a. Given selenium's more taxon-specific and life stage-specific toxicity, please comment on EPA's use of the available data to derive the egg-ovary tissue element.

This certainly makes the regulator's job easier due to the exquisite sensitivity of oviparous fish species to Se, and the well-established, characteristic and diagnostic response pattern in fishes (larval deformities and edema) that have clear links to population-level impacts. So yes, the egg/ovary tissue element is appropriate. However, it is important to note that we have limited data for all species, whether vertebrate or invertebrate. Recent work in David Buchwalter's lab at NC State U has observed a certain invertebrate taxon (Ephemeroptera I think) to be very sensitive to Se, and should be considered by EPA in the future criterion document. Nevertheless, in my opinion protecting fish based of an egg/ovary criterion will be protective of aquatic ecosystem sustainability.

To my knowledge, the EPA has used a scientifically sound procedure to use available data on 9 fish species to derive the egg/ovary criterion.

b. Given the greater general sensitivity of oviparous fish to selenium compared to aquatic invertebrates, please comment on the appropriateness of EPA's fish tissue-based criterion for affording protection to the aquatic community as a whole (e.g., including invertebrates).

See previous comment regarding aquatic insects. In my opinion the tissue-based criteria in fish will protect freshwater aquatic communities.

c. With respect to the tests that quantified non-reproductive effects, did the EPA use that data to the best extent possible given its limitations (e.g., relevance compared to reproductive tests, and data quality concerns which increased uncertainty (e.g., Hamilton et al., 1990)?

Since the non-reproductive effects occur at tissue [Se] equal to or more commonly greater than reproductive effects, and since reproductive effects have clearer links to population-level impacts than non-reproductive effects such as reduced growth or altered behavior, the EPA has appropriately chosen not to use non-reproductive effects in their derivation of tissue-based criteria.

d. EPA also rejected studies that used the injection route of exposure for selenium due to uncertainty related to uptake, distribution and metabolism/transformation kinetics when compared with the dietary and/or maternal transfer routes of exposure. Was this reasonable? Does the panel envision an appropriate and scientifically defensible use for this type of data? Please provide detailed comments.

I think the EPA should use studies that use maternal injection of Se as the route of exposure (e.g., the Doroshov et al. (1992) study in catfish). Whether Se is absorbed from the gut or injected into adult female fish, it will reach the systemic circulation and become part of the Se pool, some of which will be

incorporated into vitellogenin in the liver and transported/deposited into eggs. Including the Doroshov et al. (1992) study is thus scientifically sound, and will add an additional fish taxon (ictalurids) into the species sensitivity distribution.

3. Was the method (Section 4.1.5, 7.1.7) used to translate the fish egg-ovary criterion element into muscle and whole body criterions elements understandable, transparent and scientifically defensible? Was there sufficient data for making the translations for each element?

The EPA used an appropriate approach to translate the egg/ovary element to whole-body and muscle elements. Unfortunately, data are limited to few fish species. As discussed above in 2a(iii), conversion ratios vary by about two-fold for both EO:WB and EO:M. In addition, within-species ratios may vary throughout the year. These aspects all create uncertainty, but these are the data we have and this is the best approach. It is suggested that as more studies measure [Se] in egg/ovary, whole-body and muscle, that these data be used to update criteria through time.

One thing that was not clear. In certain cases it appears that [Se] in egg/ovary and whole-body were determined in the same fish. If eggs were removed for [Se] determination prior to determination of whole-body [Se], then how did the removal of eggs influence the whole-body [Se]? Was the absolute quantity of Se removed by subsampling eggs added back into the whole-body quantity, and was the mass of eggs removed added back to the whole-body?

PART III: EVALUATION OF THE TRANSLATION PROCEDURE TO DERIVE THE WATER COLUMN ELEMENT(S)

EPA is also requesting a technical review of the methods and procedures used to translate the egg-ovary element of the chronic selenium criterion to water-column elements. Relevant sections of the document include:

- A description of the method used to derive an equation to translate the egg-ovary element to a monthly water-column element in perennial (lentic and lotic) waters and an equation that can be used to convert the monthly water-column element to an intermittent water column element (Sections 3.8.3, 3.8.4, 4.2.1, 4.3, and Appendix G).

- An analysis of the translation equation precision using data obtained from published literature (Sections 7.2.1, 7.2.2, and Appendix H).

- A description of the method and data sources used to derive the translation equation parameters (Sections 4.2.2, 4.2.3, and Appendix B).

- A description of the method and data sources used to categorize waterbody types where a single water-column chronic criterion concentration value would be adequately protective in most circumstances (Section 4.2.4).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for established categories of waters (Section 4.2.5).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for intermittent discharges that may occur in lentic and lotic waterbodies (Section 4.3).

Please address the following questions:

1. Please comment on the scientific defensibility of EPA's translation equation method for translating the concentration of selenium in fish tissue to a concentration of selenium in the water-column. Please comment on major sources of uncertainty in applying the translation equation to different types of waterbodies (e.g., with differing retention times, water chemistries, and/or species present). Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

The EPA has used the modern and scientifically valid biodynamic model approach to derive water quality elements from tissue-based elements. I am not aware of other data sources, models or approaches that would reduce the inherent uncertainty. However, based on comments provided above (in 1 and especially 2a(iii)), relying on water column dissolved [Se] has a high likelihood of generating both false positive and false negative results with respect to regulatory action. I think the proposed water column criteria (a) should be used as triggers to initiate further monitoring of fish tissue [Se], (b) should be made more conservative (reduced) by application of a safety factor to avoid false negatives, and (c) that the simple classification of a water body as lentic or lotic should be modified to include more quantitative measures of flow such as water residence time and/or mean annual water velocity. Given that many impounded riverine systems in the USA are essentially lentic systems for much of their river-miles, perhaps a water column trigger [Se] could be set at 1 ug/L (same as the current Canadian [CCME] water quality guideline for Se). If exceeded, this trigger value would result in further action in terms of fish collections for tissue [Se].

2. Regarding the trophic transfer factor (*TTF)* values, did EPA use a scientifically defensible method to derive the *TTF* values (p. 71-77 of the criteria document)? Were the exclusion criteria, (pp. 71-77 of the criteria document) developed by EPA to screen the available data applied in a consistent and scientifically defensible manner? In particular, EPA noticed that application of the exclusion criteria resulted in *TTF* values for aquatic insect larvae that differ from other published values. Given this, are you aware of any other methods of screening data that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included, if appropriate? Please provide detailed comments.

The method used to derive TTF values is scientifically sound by using the widely accepted biodynamic modeling approach, which is particularly appropriate for Se. The EPA also demonstrated that temporal changes in TTF are for the most part not a factor that may cause large data discrepancies. Since the EPA used a large dataset to derive TTF values for insects, any differences between the EPA-derived values and values reported from individual studies are not of concern to this reviewer. I am not aware of any other data, other than the recent work by Buchwalter mentioned in II2a above. It is suggested the EPA include an updated literature search for this and other supporting data prior to the next revision of the document.

3. Regarding the conversion factor (*CF*) values used, did EPA use an appropriate and scientifically defensible method to derive those values (p. 78-79 of the criteria document and Appendix B)? Are you aware of any other methods that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included? Please provide detailed comments.

EO:WB conversion factors ranged from 1.38 to 7.39 with a median value of 1.27. As mentioned in II3 above, it was unclear how determination of [Se] in both whole-body and egg were determined in the same fish, and this should be clarified in the document. Similarly when muscle and whole body were determined in the same fish.

Overall, this is a simple method and I am not aware of any alternative methods nor data sources for these analyses.

4. Regarding the derivation of enrichment factor (*EF*) values, was the method EPA used to screen data from the literature applied appropriately and consistently (see inclusion/exclusion criteria on p. 71-77 of the criteria document)? Was the method for deriving *EF* values applied to those data in a consistent manner so as to derive *EF* values for selected waters in a scientifically defensible manner? Is the method that EPA used to establish the lentic and lotic categories for *EF* values reasonable given the available data? Are you aware of other methods or relevant data the EPA should consider? Please provide detailed comments.

EF values were derived from all available data that I am aware of and used scientifically valid approaches, including inclusion/exclusion criteria. See comments above regarding the simple distinction used for lentic vs. lotic systems.

5. Please comment on the scientific defensibility of EPA's conversion of the selenium fish tissue – water translation equation into an equation that allows for calculation of a criterion for waters that may be subject to intermittent discharges of selenium. Please comment on major sources of uncertainty in this approach. Is this method appropriate, given the bioaccumulative nature of selenium? Please comment on the uncertainty associated with the application of this conversion equation to intermittent discharges that may occur in different types of waterbodies and/or in different locations, particularly with respect to loads transported to potentially more sensitive aquatic systems. Does the method employed result in criteria that are similarly protective to the 30-day chronic criterion? Are there any other models or approaches that EPA should consider that would reduce this uncertainty? Please provide detailed comments.

As mentioned above, I am not in favor of the intermittent water column criterion. If the EPA decides to go ahead with it, then (a) the rationale for such a criterion should be clarified in the document, and (b) clear guidance on the practical use of the criterion should be provided. In my opinion, the intermittent criterion makes the complex issue of Se aquatic life criteria unnecessarily more complicated, and may be manipulated to either underestimate or overestimate the actual risk posed by Se to fish and other aquatic life.

PART IV: SIGNIFICANCE OF SCIENTIFIC VIEWS FROM THE PUBLIC/STAKEHOLDERS

EPA will also be providing scientific views and other comments from stakeholders and the public received via the public docket to the peer review panel. Although EPA will be providing the full contents from the docket, EPA is only requesting a review of any scientific views/public comments that may be of technical significance to the selenium criterion.

1. Has the peer review panel identified any scientific views from the public or stakeholders as being technically significant to the draft of the selenium criterion going forward; that is, has information or data been introduced during the comment period that would change the scientific direction of the criterion? Is there any information or data that may refine or enhance the scientific defensibility of this criterion that EPA should consider further? Please provide detailed comments on specific issues of technical significance or refinement.

I have read through the entire package of views from public and stakeholders, not just the summarized Excel file but the actual documents, some of which are >100 pages. The EPA should pay close attention to these documents, since some excellent scientific issues are raised in many of them. It is good to see that there presently exists such good knowledge of the aquatic ecotoxicology of Se among stakeholders; 10 years ago this would not be true.

The public/stakeholder views represent the classic range, from industry-based opinions that the proposed criteria are too conservative, to conservation group-based opinions that the proposed criteria will not protect all aquatic life. Both sides of the argument present many good points that should be considered carefully by the EPA. I will provide my views on each category of public/stakeholder comments at the end of this section.

The bottom line is that industry would prefer the egg/ovary criterion to be about 20 mg/kg egg (or greater), whereas conservation groups would prefer it to be about 10 mg/kg egg (or lower). Perhaps the 15.2 mg/kg criterion represents a workable compromise between these two extremes? I believe the EPA document for the most part has used current, scientifically sound approaches without significant bias in either direction (but see my comments regarding the Formation brown trout study). Since the proposed EPA criteria would still allow some aspect of site-specific assessment at the State level, then there could be modifications based on site specific issues such as relatively high background [Se] in certain areas, fish species not included in derivation of the egg/ovary criterion, lack of fish species ("fishless" waters), high aqueous sulfate, the presence of listed/threatened/endangered fish species, the presence of critical aquatic-dependent wildlife such as birds, or other biological/chemical/physical factors.

Specific comments on public/stakeholder documents:

An acute criterion is not needed and is not relevant. If you are releasing Se into the aquatic environment at levels that cause acute toxicity to fish, then you have a big problem!

Lentic and lotic systems must be clearly defined and perhaps a more quantitative approach should be used as I have discussed above.

The EPA should read the public/stakeholder input carefully and use these suggestions to come to a final decision on the Formation brown trout study. This is of critical importance since brown trout was found to be the most sensitive fish species and the egg/ovary criteria is driven largely by the brown trout EC10.

Elevated sulfate ion in aquatic systems may reduce Se bioaccumulation in food webs by competing with selenate for uptake by primary producers, particularly algae. However, if regulatory limits are based on fish tissue [Se] then any modification of Se uptake by primary producers will be reflected in fish tissue

[Se]. In my opinion sulfate is not really a regulatory issue when fish tissue [Se] is used.

Ideally freshwater criteria for Se should include aquatic-dependent wildlife such as birds. However this makes the Se criteria more complicated than perhaps it needs to be. The issue of birds could be considered on a site-specific basis in certain ecosystems inhabited by ecologically significant avian populations and migrating water birds.

The EPA must provide guidance on several aspects related to implementation of the tiered criteria approach, at the very least including (a) when to sample fish so that females are in vitellogenic or pre-ovulatory stages of oogenesis, (b) what sample size of fish to collect for tissue [Se] determinations (I suggest a minimum of n=10 female fish per site), (c) recommended analytical procedures for quantification of Se, (d) guidelines for implementation of the 30-day average water column criterion element (how, when, where), and (e) guidelines for implementation of the intermittent water column criterion, if the EPA chooses to keep it in the tiered criterion.

An interesting comment made in one of the public/stakeholder documents (US Fish and Wildlife Service, document 354-A2)) regards the use of recently published studies in zebrafish, a non-native cyprinid, in the species sensitivity distribution for larval deformities as a function of egg [Se]. They present a compelling argument to consider these data in the criterion development.

PEER REVIEW COMMENTS FROM

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External Peer Review of the Draft Aquatic Life Ambient Water Quality Criterion for Selenium – Freshwater 2014

Responses to Charge Questions by Dr. Gregory Möller

PART I: OVERARCHING QUESTIONS

1. Please comment on the overall clarity of the document and construction of the criterion statement with its multiple elements.

On an overall basis, the 2014 Selenium Criterion is well-organized and well-written. The major sections of the document serve to critically analyze the scientific and regulatory background of the issue, and to develop and rigorously justify a tiered criterion. Overall, the writing is clear and communicative, with key details, data and background information appropriately appended to the main document. The included tables and figures act to support the analysis of cause for a substantially different approach to risk management and furthermore this information serves to validate this criterion approach by critically evaluating decades worth of peer-reviewed laboratory and field observations in a fair and scientifically valid manner. The concordance observed in many tables exploring and ground-truthing modeled approaches, available data, and a broad array of published study results yields exceptional weight and justification for this new approach developed for the protection of aquatic life.

Importantly, the criterion statement on p. 96 does indicate dry weight basis for tissue analyses, and this is discussed in the text, however Table 15 and the tabular Summary on p. 4 do not carry the dry weight basis notation and this should be included. With the advantage of subsequent key published selenium research targeting trophic transfer and reproductive endpoints in fish, as well as the expert panel contributions published in Chapman et al., 2009, this current document is a significant improvement over the 2004 AWQC draft. In its presentation and treatment of a broad and diverse study and data set, the draft criterion document can be characterized as exhaustive in its attempt to quantitatively and qualitatively address the myriad issues related to this task under the CWA. Furthermore the draft criterion document addresses that task in a manner that synthesizes a new tiered criterion approach well-grounded in our current understanding of selenium risks in aquatic ecosystem and best available peer-reviewed knowledge. The draft approach balances knowns and unknowns, data and data gaps, simplicity and complexity in an overall sound attempt to address the time-value requirement of regulatory science. Although additional implementation guidance for this new tiered approach may be necessary, and observing that the discussion of background science, data and methods used in the intermittent exposure tier of the present criterion needs significant improvement, the draft document is overall remarkable for its clarity and completeness, in a scientifically driven and defendable analysis of a complex risk management challenge.

2. EPA has developed a tiered selenium criterion with four elements, with the fish tissue elements having primacy over the water-column elements, and the egg-ovary element having primacy over any other element. Inclusion of the fish whole-body or fish muscle element into the selenium criterion ensures the protection of aquatic life when fish egg or ovary tissue measurements are not available, and inclusion of the water column elements ensures protection when fish tissue measurements are not available

    a. Please comment on the tiered construction of the selenium chronic criterion; is it logical, and scientifically defensible as it applies to protection of freshwater aquatic life:

        i. That is, is the primacy of the egg-ovary element over the other elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

The primacy of the egg-ovary element over other elements of the selenium chronic criterion is logical and broadly scientifically defensible. As identified in the document, numerous published studies outline the major aquatic ecosystem impact of selenium, beyond its nutritional requirement, as a reproductive toxicant. While the specific bio-molecular mechanisms of reproductive toxicity and teratogenesis still require further work, it is well-established from controlled laboratory studies and field studies that the best indicator of the potential for reproductive end effects from selenium is in tissue concentrations, and specifically in egg-ovary concentrations. While the relationships of tissue and water concentrations can be studied, quantified, modeled, and tasked to risk assessment, the now well-established relationship of egg-ovary Se levels to toxicity endpoints fully justifies this primacy of this indicator.

        ii. Is the primacy of the whole-body/ fish muscle element over the water column elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

With regards to many chemical exposures in aquatic ecosystems, tissue levels in resident or migratory aquatic animals often help to assess toxic risk by integrating the exposure and revealing the storage, distribution, metabolism, and excretion of the toxicant, regardless of the geography, hydrograph or acute-to-chronic exposure dynamic of the chemical. The bimodal nutrient-toxicant behavior of selenium adds to the complexity of evaluating approaches to risk management. Metabolic and environmental conditions can also add complexity and uncertainty to a full understanding of risk in selenium impacts aquatic ecosystems. It is clear from many studies that the physiological homeostasis (uptake/efflux) of Se is not well controlled and the biochemical metabolic co-relationship of Se and S pathways *in vivo* allows for chronic Se exposure to advance to toxic endpoints. A recurring issue in aquatic ecosystem Se management has been co-exposure to high levels of sulfur, typically as sulfate. While high sulfate co-exposure may impact Se toxicosis, tissue Se levels yield a high quality, aquatic Se toxic impact potential metric regardless of sulfate co-exposure or other co-factors in Se reproductive toxicity (e.g., synergists or antagonists), known or unknown. This Se fish tissue approach, including eggs, ovary, muscle or whole body, is robust with respect to the findings of several decades of peer-reviewed studies. Selenium levels in fish tissue are broadly accepted in the scientific community as a high quality indicator suitable for risk management of aquatic life. The document supports the tissue approach and key toxicological endpoints with a critical review of the peer-reviewed literature and inclusion or rejection of specific studies and the data or findings therein, in an overall transparent, logical and defendable manner.

The tier placement of whole-body/fish muscle is appropriate since egg/ovary assessment may have practical challenges with some ecosystems, with some species, the size of the target fish, and with some aspects of the life-cycle of the target fish. The inclusion and tier level of fish tissue selenium gives

flexibility in aquatic ecosystem risk assessment.

Inclusion of water column Se levels in the tiered criteria will no doubt help screen for potential Se impacts in aquatic ecosystems that have not had a history or occurrence of selenium contamination, and in the prevention of discharges or other anthropogenic activities that present an unacceptable risk to water quality.

iii. Please comment on the scientific uncertainty that may be associated with this tiered approach? Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

The tiered approach presented in the criterion embodies the best available scientific knowledge of selenium in aquatic ecosystems actively studied by a broad range of investigators, disciplines and institutions, across a diverse range of water environments and potentially impacted organisms, over more than three decades of focused effort. While all science has uncertainty, the magnitude and diversity of the research effort in the environmental toxicology and regulatory science community to understand the complex risk dynamic of selenium in aquatic ecosystems is unprecedented in the history of U.S. environmental law. The 2014 Aquatic Life Ambient Water Quality Criterion for Selenium balances the available data, models, and approaches to risk management. The document and the tiered criteria within represent a balanced approach where assumptions and data uncertainties are clearly laid out and discussed. Published data or results that were not included in criterion determination were adequately and satisfyingly discussed and defended for exclusion. The data and peer-reviewed studies used in the quantitative and qualitative development of the criterion are sufficiently robust, sufficiently concordant in their conclusions, and sufficiently broad in their scope and number to result in a criterion that can protect aquatic organisms under the requirements of the Clean Water Act.

iv. Are the draft recommended magnitude, duration, and frequency for each criterion element scientifically sound and appropriate? Please provide detailed comments.

The recommended magnitude, duration, and frequency for each criterion element are scientifically sound and appropriate. The derivation of the tissue based criteria are well-supported by including the major published works in the related fields and by rejecting with transparent cause and inclusion/rejection standards those studies that do not attain the stated benchmark for quality and reproducibility (e.g., NOECs). The criterion development satisfyingly addresses a diverse range of major fish types indicative of aquatic ecosystem health in geographically diverse lentic and lotic systems. With chronic exposure, fish egg-ovaries are now recognized as the best indicator of toxic selenium risk, however practical monitoring may require whole body-muscle tissue analysis. Water column selenium values fill the need for screening and analysis of potential for risk, abatement of new contamination pathways, and managing discharge, as well as other activities that may impact water quality.

PART II: FISH TISSUE CRITERION ELEMENTS DERIVATION: DERIVATION OF FISH EGG-OVARY, WHOLE BODY AND MUSCLE CRITERION ELEMENT(S)

EPA is requesting a technical review of the methods and procedures used to derive a chronic selenium criterion based on an egg-ovary concentration, as well as its translation to a criterion element applicable to whole-body and muscle tissue. Please address the following questions:

1. Please comment on EPA's use of the effects concentration 10th centile (EC_{10}) as the measurement endpoint for the fish reproductive toxicity studies used to derive the egg-ovary element.

The EC10 is an appropriate endpoint to use in the development of the egg-ovary element of the tiered criterion. Egg-ovary Se concentration is well recognized in the peer-reviewed scientific literature as a high quality indicator of reproductive toxic risk in fish. Because selenium is a reproductive toxicant, special considerations in risk management are warranted. For precedent, the Food Quality Protection Act of 1996 which manages risk of chemical exposure in the human food system, uses an extra ten-fold safety factor for chemicals used in food production that have reproductive toxicology or neurotoxic endpoints. This extra safety factor results from our common understanding in toxicology that those chemicals with repro- or neuro-toxic activity represent an exceptional risk and thus require exceptional safeguards. Reproductive toxicity is a significant threat to the population of the impacted aquatic organisms and thus to the aquatic food-web. There are valid questions whether the EC10 is sufficiently protective of endangered aquatic species and the criterion document should address these concerns more thoroughly. Overall the EC10 egg-ovary endpoint is scientifically consistent and defendable with the intent and required actions of the CWA.

2. Data used to derive the final chronic egg-ovary criterion element were differentiated based on the type of effect (reproductive vs. non-reproductive effects). Acceptable chronic toxicity data on fish reproductive effects are available for a total of nine fish genera. The genus Sensitivity Distribution (SD) is predominantly populated with data on fish genera because field evidence demonstrates that fish communities can be affected by selenium even when there is no observable change in the invertebrate community diversity and abundance. As a result, decades of aquatic toxicity research have focused primarily on fish. Available field and laboratory studies indicate that invertebrates are more tolerant to selenium than most of the tested fish species (Criteria document, Table 6c, Section 4.1.2). The data set used to derive the selenium criterion marks a change from the traditional method used to derive water quality criteria that requires toxicity tests with aquatic organisms from 8 phylogenetically distinct taxa (including three vertebrate and five invertebrate genera) in order to derive aquatic life criteria (Stephan et al., 1985).

    a. Given selenium's more taxon-specific and life stage-specific toxicity, please comment on EPA's use of the available data to derive the egg-ovary tissue element.

The use of fish data to drive the tiered criteria, and specifically the egg-ovary tissue element is fully justified and well-supported in the criterion document and the relevant scientific literature. While the sources, pathways, receptors and controls of chemicals impacting water quality have inherent diversity, selenium demonstrates significant trophic transfer potential and potential for fish reproductive effects in aquatic ecosystems. The reproductive endpoints observed in peer-reviewed, published controlled and field studies strongly suggest the potential for accumulation, magnification, and trophic transfer, and thus population level effects in a higher tropic level organism such as fish. The concomitant food-web impacts and observed impacts to aquatic birds support the criterion approach. The guidelines of Stephan et al., 1985 pre-date much of the knowledge base of Se in aquatic ecosystems, and the somewhat unique behavior and impact potential of this toxicant across trophic levels did not come into a more complete

understanding for nearly two decades since that work. Hence, deviation from prior risk assessment approaches that pre-date our current knowledge base and the evolution of understanding of Se behavior in aquatic ecosystems is broadly justified in the risk management of selenium. Stephan et al., 1985 pre-date much of the knowledge base of Se in aquatic ecosystems, and the somewhat unique behavior and impact potential of this toxicant across trophic levels did not come into a more complete understanding for nearly two decades since that work. Hence, deviation from prior risk assessment approaches that pre-date our current knowledge base and the evolution of understanding of Se behavior in aquatic ecosystems is broadly justified in the risk management of selenium.

b. Given the greater general sensitivity of oviparous fish to selenium compared to aquatic invertebrates, please comment on the appropriateness of EPA's fish tissue-based criterion for affording protection to the aquatic community as a whole (e.g., including invertebrates).

The fish tissue-based criterion affords protection to the aquatic community as a whole and is appropriately placed in the tiered criterion. Since tissue Se integrates chronic and intermittent acute aquatic Se exposure, it provides a good quality indicator of impacts and potential impacts to the broader aquatic community. The complex interactions of predator-prey relationships in these environments rely on nominal stability in each tropic level and the food-web as a whole. In field practice and in published controlled studies, fish tissue Se has been shown to provide a valuable assessment and management tool for Se impacted aquatic ecosystems. Except where fish populations are absent, very low, endangered or otherwise insufficient, tissue monitoring is a high quality indicator of water quality with regards to selenium.

c. With respect to the tests that quantified non-reproductive effects, did the EPA use that data to the best extent possible given its limitations (e.g., relevance compared to reproductive tests, and data quality concerns which increased uncertainty (e.g., Hamilton et al., 1990)?

The non-reproductive fish data, limited in scope and diversity, were adequately explored and treated in the development of the tiered criterion. The increased concerns over reproductive effects from a risk management perspective, study diversity (e.g., species, geography, lentic/lotic), in addition to the quality and quantity of reproductive toxicity endpoint data and studies reproductive toxic risk the superior driver of selenium risk management in aquatic ecosystems. The summary statement that the non-reproductive data were less reproducible (p. 57) suggests that including them would have added uncertainty to the final criterion values. It is reasonable, acceptable, and scientifically defensible to have reproductive toxicity as the driving endpoint for criterion development, as these criteria appear to afford protection from non-reproductive toxic effects.

d. EPA also rejected studies that used the injection route of exposure for selenium due to uncertainty related to uptake, distribution and metabolism/transformation kinetics when compared with the dietary and/or maternal transfer routes of exposure. Was this reasonable? Does the panel envision an appropriate and scientifically defensible use for this type of data? Please provide detailed comments.

The rejection of injection exposure route studies is reasonable. Injection based toxicology studies have their place in understanding the interface of chemistry and biology. They are of significant value when metabolism of the toxicant is of interest or when digestive and absorption processes (i.e., bioavailability) confound or complicate study goals. Since controlled feed/water laboratory exposure trials, and field observation data and published studies are available in overall sufficient quantity, diversity, and quality for

establishment of the criterion, the rejection of injection-based trials results yield a data set more amenable to generalization of aquatic ecosystem exposure and dose, as well as the subsequent analysis of trophic transfer and potential for toxic end effects. Although injection route studies have scientific value, they are not necessary or required for a qualitative and quantitative understanding of Se aquatic ecosystem risk potential given the other peer-reviewed resources presently available.

3. Was the method (Section 4.1.5, 7.1.7) used to translate the fish egg-ovary criterion element into muscle and whole body criterions elements understandable, transparent and scientifically defensible? Was there sufficient data for making the translations for each element?

The approach and method of translating the fish egg-ovary criterion into muscle/whole body is transparent and broadly scientifically defensible, and there appears to be sufficient data to make the translation. Although there is some variability in the calculated results of whole body and muscle calculations, the relative consistency across taxon gives significant support to the modeling approach and in the data used to derive the values. The Figure 5 references to Table 10 and 11 should be introduced and explained in the body text prior to using them in a Figure caption since the reader has not seen that data. Some editing in this regard would improve clarity and help the reader understand and follow the approach. The body text of paragraph 1 of page 59 needs to be rewritten for clarity; statements of "it can be seen" assume much and explain little. Because the paragraph references a subsequent Section 4.2, editing page 59 to introduce and summarize the detail of 4.2 would be an improvement in clarity for the reader. Table 7a and 8a would be improved with units (mg Se/kg DW) for tissue concentrations. Footnotes on these important tables cross-referencing the specific source, table or appendix where the data originated would be helpful and aid in reader understanding and transparency.

PART III: EVALUATION OF THE TRANSLATION PROCEDURE TO DERIVE THE WATER COLUMN ELEMENT(S)

EPA is also requesting a technical review of the methods and procedures used to translate the egg-ovary element of the chronic selenium criterion to water-column elements. Relevant sections of the document include:

- A description of the method used to derive an equation to translate the egg-ovary element to a monthly water-column element in perennial (lentic and lotic) waters and an equation that can be used to convert the monthly water-column element to an intermittent water column element (Sections 3.8.3, 3.8.4, 4.2.1, 4.3, and Appendix G).

- An analysis of the translation equation precision using data obtained from published literature (Sections 7.2.1, 7.2.2, and Appendix H).

- A description of the method and data sources used to derive the translation equation parameters (Sections 4.2.2, 4.2.3, and Appendix B).

- A description of the method and data sources used to categorize waterbody types where a single water-column chronic criterion concentration value would be adequately protective in most circumstances (Section 4.2.4).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for established categories of waters (Section 4.2.5).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for intermittent discharges that may occur in lentic and lotic waterbodies (Section 4.3).

Please address the following questions:

1. Please comment on the scientific defensibility of EPA's translation equation method for translating the concentration of selenium in fish tissue to a concentration of selenium in the water-column. Please comment on major sources of uncertainty in applying the translation equation to different types of waterbodies (e.g., with differing retention times, water chemistries, and/or species present). Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

The translation equation approach used to convert toxicologically relevant fish tissue concentrations to water-column concentrations is broadly scientifically sound and defensible, and represents our best available understanding of these relationships across trophic levels in an aquatic ecosystem food web. This may be especially true because the approach is based on a straightforward model, and alternative approaches that introduce complexity can also introduce uncertainty from the requirements of additional data beyond that currently available. Risk estimation rarely has perfection due to situational variability and uncertainty involving the integration of exposure, uptake, and biokinetics. The draft criterion approach uses qualified data and reasonable analysis to reduce complexity and increase the transparency of criterion. Modeling dynamic relationships in complex multi-level systems with innate variability is a significant environmental management challenge, however the effort can yield a valuable management tool. Figure 8 (p. 73) graphically demonstrates "hysteresis" with regards to aquatic food chain selenium levels and potential for toxic impact as well as the temporal relationship to periodic sampling. Any challenges in application of this approach across diverse aquatic ecosystem types with variable water chemistries and annual variability (e.g., flow and flux), are equally met by the challenges of sufficiently devising specific criteria to address every subset of variables with less or equal uncertainty in the protection of aquatic life. The duration and frequency requirements of the water column selenium criterion address the potential for system variability (e.g., year to year weather/hydrograph changes) and propagation of system uncertainty (e.g., non-selenium related chemical or biological changes) in this risk management.

2. Regarding the trophic transfer factor (*TTF)* values, did EPA use a scientifically defensible method to derive the *TTF* values (p. 71-77 of the criteria document)? Were the exclusion criteria, (pp. 71-77 of the criteria document) developed by EPA to screen the available data applied in a consistent and scientifically defensible manner? In particular, EPA noticed that application of the exclusion criteria resulted in *TTF* values for aquatic insect larvae that differ from other published values. Given this, are you aware of any other methods of screening data that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included, if appropriate? Please provide detailed comments.

The trophic transfer factor (TTF) values were developed as an application of a peer-reviewed, published approach that represents our best available scientific information. The method and data used are adequately described, and the approach is satisfyingly direct. The confounding dynamic to this approach could be the bi-modal essential-toxic behavior of selenium where low-level exposure has different metabolic and storage behavior that non-essential metals and therefore different toxicodynamics across a broad range of exposures. This dynamic is adequately discussed (p. 74). The screening criteria for data used in TTF calculations appear defensible and reasonable, and complete with regard to major published works.

3. Regarding the conversion factor (*CF*) values used, did EPA use an appropriate and scientifically defensible method to derive those values (p. 78-79 of the criteria document and Appendix B)? Are you aware of any other methods that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included? Please provide detailed comments.

There is inherent uncertainty and variability in deriving conversion factors given the diversity of fish types, lifecycle stage, and environmental conditions. The single 1.27 conversion factor approach appears to be a straightforward and reasonable approach given the limitations of data and species data sets. This is especially true in practice where a criterion will be applied to fish types including those not subjected to controlled studies. While species specific CFs are desirable, this would require considerably more data that currently available especially in regards to life cycle of the target fish analyzed. The conversion factor (CF) method and input data appear to be a reasonable and defendable approach to addressing data limitations and practical application of the criterion. Other numerical approaches can also rise to developing CFs however it is unclear if the absence of data would bias those results or create similar uncertainties as well. The calculation approach in the current draft is straightforward and robust. Appendix B appears to have most freshwater fish data used in the CF analyses addressed in multiple published scientific papers or agency reports. Because of the critical nature of this calculation to criterion development, updating literature searches for new research data is important.

4. Regarding the derivation of enrichment factor (*EF*) values, was the method EPA used to screen data from the literature applied appropriately and consistently (see inclusion/exclusion criteria on p. 71-77 of the criteria document)? Was the method for deriving *EF* values applied to those data in a consistent manner so as to derive *EF* values for selected waters in a scientifically defensible manner? Is the method that EPA used to establish the lentic and lotic categories for *EF* values reasonable given the available data? Are you aware of other methods or relevant data the EPA should consider? Please provide detailed comments.

The enrichment factor (EF) approach and method is scientifically defensible and represents our best understanding of selenium dynamics in aquatic ecosystems. While all modeling approaches have uncertainties and limits in application, the approach is reasonable, transparent, appropriately applied and representative of the present selenium knowledge base. The criterion document uses available data in a consistent manner, and extending the water system terminology used by study authors for data used in EF value determinations is a best practice. The evaluation of categories of aquatic systems is well treated in the analysis. The grouping of streams, drains, washes and creeks into a common category is reasonable. The results of Figure 9 and 10, and furthermore in Figure 11, help to validate the EF approach of the criterion document when measured against our cumulative knowledge base of selenium behavior in different aquatic systems. The use of a 20^{th} percentile approach for water column values accommodates system variability and system uncertainty that is inherent in all modeling approaches. Whereas tissue levels of Se can more reliably predict toxic risk, a 20^{th} percentile affords adequate protection in many risk management situations such as water quality-based effluent limits, especially in light of the primacy of the tissue based components of the criterion.

5. Please comment on the scientific defensibility of EPA's conversion of the selenium fish tissue – water translation equation into an equation that allows for calculation of a criterion for waters that may be subject to intermittent discharges of selenium. Please comment on major sources of uncertainty in this approach. Is this method appropriate, given the bioaccumulative nature of selenium? Please comment on the uncertainty associated with the application of this conversion equation to intermittent discharges that may occur in different types of waterbodies and/or in different locations, particularly with respect to loads transported to potentially more sensitive aquatic systems. Does the method employed result in criteria that are similarly protective to the 30-day chronic criterion? Are there any other models or approaches that EPA should consider that would reduce this uncertainty? Please provide detailed comments.

While the need for a criterion tier that addresses intermittent discharges is clear, this part of the document is not well documented for scientific support as evidenced in the main document by no citations in this section beyond that of the general Chapman et al. 2009 reference. Appendix G Part 3.0 documents the modeling approach, however a list of references is missing. Since this is original work, further description of methods, key data inputs, and model run output may be useful for potential replication of the results by others. A citation on page G-6 (EPA 1986; should be USEPA 1986) may be important to sourcing this modeling approach, but it is unclear in the writing whether this is so; without references to Appendix G, validation of scientific defensibility of the intermittent water-column criterion is not possible. Infrequently, some of the writing in Appendix G is informal or tech-speak and should be edited for clarity. Figure captions should contain a short description of all relevant model inputs to increase communication value and transparency. The modeling approach and the results of Appendix G appear to be a reasonable and defendable approach to developing a criterion for intermittent water column selenium values, although the polished execution of this important part of the tiered criterion is lacking in comparison to the other criterion elements. Thus, there appears to be sufficient support for the criterion approach in Appendix G and this information should be summarized and referenced in the main document body. This part of the tiered criterion is the most difficult to study in the field, although our practical and experiential knowledge of Se bioaccumulation in aquatic ecosystems suggests it has high importance in protecting aquatic life. The practical implementation of this tier of the criterion will require enhanced guidance and regulatory sensitivity to the cost of monitoring.

PART IV: SIGNIFICANCE OF SCIENTIFIC VIEWS FROM THE PUBLIC/STAKEHOLDERS

EPA will also be providing scientific views and other comments from stakeholders and the public received via the public docket to the peer review panel. Although EPA will be providing the full contents from the docket, EPA is only requesting a review of any scientific views/public comments that may be of technical significance to the selenium criterion.

1. Has the peer review panel identified any scientific views from the public or stakeholders as being technically significant to the draft of the selenium criterion going forward; that is, has information or data been introduced during the comment period that would change the scientific direction of the criterion? Is there any information or data that may refine or enhance the scientific defensibility of this criterion that EPA should consider further? Please provide detailed comments on specific issues of technical significance or refinement.

Acute criterion: The comments largely support or request guidance concerning abandonment of an acute criterion. The intermittent water column tier of the draft selenium criterion does much to address potential ecosystem impact potential from discharge concentrations historically regarded as having "acute" toxic

potential.

Alternative more sensitive endpoint: Comment lacks clarity and method/approach publication or peer-review to fully consider the point being made.

Aquatic dependent wildlife: Sound points are made concerning the potential for impact to aquatic birds. The author overstates that the criterion set a *de facto* limit for invertebrates. While the comments are broadly valid and demonstrate the complexity of the Se aquatic impact issue, equal concerns should be weighed on the relative balance of over- or under- protection of the draft criteria if deployed. The rigor of this present document to address aquatic life ambient water quality is significant, broadly inclusive and broadly defendable. The tier approach may be expected to have significant impact in overall water quality and aquatic dependent wildlife because of the integrative exposure nature of the tissue criterion.

Averaging period: Comment reasonably addresses the need for clearer implementation guidance of the intermittent water column criterion.

Bioaccumulation factors: The context of this question is addressed in the document, however additional clarification may be useful.

Biphasic modeling: The comment author expresses an opinion regarding modeling approach. The available peer-reviewed published studies supporting this approach for selenium in fish/aquatic ecosystems is limited and thus of less value in setting the criterion. The author may have a good point however the availability of published work limits its practical consideration. The Atlantic salmon graph referenced appears to be a Wikipedia selenium entry without attribution.

Bluegill Hermanutz: The conclusion that the Hermanutz data are outliers is not supported in the comment by any numerical/statistical analysis and thus must be treated as opinion, unless otherwise verified. Data variability in biological systems can be tested to determine outliers however it is unlikely the data count would support exclusion, thus inclusion is more defendable.

Brown trout study: The presentation and role of the brown trout study, related serial reviews, and re-reviews in the draft criterion document and supporting resources raises questions in the public comments. While some of the questions addressed in public comments are broadly addressed in the draft document, additional effort should be made by EPA to specifically address concerns outlined in these comments. The use of the study data is confounded by unfortunate experimental system failure encountered during the study.

Clarification: The comment authors state reasonable requests for clarification that can be addressed in the main text body.

Conversion factors: Several of the public comments regarding conversion factors represent valid concerns. Some of the issues are addressed in the draft document and thus additional explanation could be useful. The suggested approach of using species specific CFs and determining a 80 or 90^{th} percentile cut is a solid suggested for an alternative approach.

Correction: These should be validated and corrected.

Criteria are over-protective: these are speculative comments.

Criteria are under-protective: There are valid concerns expressed, especially in the apparent disconnection between agencies working towards similar goals. Concerns over the water column tier of the criteria are adequately addressed by the primacy of the tissue tiers. The risk differentiation argued between 4-6 mg/kg and 8.1 whole body/muscle tissue selenium, in light of the egg-ovary tissue primacy in the draft criterion, is moot.

Data analysis: This comment should be explored for its validity.

Data paucity affecting criterion: This comment appears to somewhat understate the available data. An additional literature search may yield new studies that increase egg-ovary data counts.

Define terms in document: Solid points are made to enhance clarity.

Dietary requirements of Se in fish: The identified citations are of value.

Document process: No comment.

EC10 clarification: Editing error identified; requires correction.

Endangered species protection: This process observation should be considered.

Exclude invertebrates: Risk assessment using extrapolations from animal models is a keystone of toxicology. The approach in the document is a modeling effort based on a similar extrapolation of available data. While not perfect, the data have value.

General comments: Many opinions expressed. Sulfate impacts can be argued to be adequately incorporated into the primacy of the egg-ovary criterion.

GMCV alternative: There are several useful comments, including apparently revised data that should be addressed.

Human health: This comment contains information useful in addressing human health implications of the draft criterion.

Implementation: The public comments express thoughtful concerns and practical implementation questions that can serve as prompts to draft additional guidance.

Importance of Se speciation: The comment expresses academically valid concerns however the practicality and data quality issues of speciated Se analyses for routine sampling and monitoring discount this concern. There are additional confounding issues of analytical sensitivity and result uncertainty at the criterion levels. Total dissolved Se sampling will filter out selenite that is readily adsorbed to suspended sediment particles.

Intermittent criterion: Several good points are raised in the public comments. Suggestions to abandon one model for another do not provide adequate support for the suggestion. Practical implementation concerns are valid and should be addressed.

Lentic lotic clarification: The public comments express thoughtful concerns and practical questions that can serve as prompts to draft additional guidance and supporting information.

Mayfly toxicity: This study should be reviewed for inclusion.

Mercury interaction: This observation is not unequivocal in the scientific literature and thus does not require significant consideration in criterion development.

Misunderstanding of MDRs: Some points are valid, however the practice of extrapolating and translating data is commonplace in toxicology.

Mode of action: The authors correctly identify an oversimplification of the wording in the draft criterion document.

Natural background: The public comments correctly identify concerns of naturally occurring selenium contamination of waters and impacted aquatic life. The draft criterion should explicitly address these concerns in regards to implementation of the draft criterion.

New information: Some of the submitted information has value and should be considered for inclusion. Sulfate modification to selenium impacts are addressed in the primacy of the egg-ovary criterion which

reasonably characterizes endpoint risk regardless of modified uptake.

Number of GMCVs in data set: Draft text should be modified to address clarification.

Other comments: Most labs report 2 significant figures for water Se analysis at these levels.

Rainbow trout study clarification: Clarifying language should be added to the draft text.

Recommend other studies: These studies should be reviewed for inclusion in the data set.

Recommended modifications: This is a summary state of previous suggestions in the list. Data updates once validated are reasonable requests.

Recommended muscle criterion: The approach should be critically reviewed.

Recommended research: While interesting, the method is not used in all studies. Citable references are absent from the comment.

Recommended whole body criterion: The approach should be critically reviewed.

Recommends alternative analysis of Hardy cutthroat trout: The commenter's calculation lacks peer review and detail.

Recommends alternative statistical analysis for Hermanutz bluegill: The commenter's calculation lacks peer review and detail.

Recommends alternatives to Guidelines SSD: Several practical comments are contained in this collection that can assist in drafting clarifying language and guidance.

Recommends including catfish study: The comments are well developed but not necessarily compelling for inclusion, especially in light of previous comments directed at lowering the outcome of the criterion development.

Recommends including zebrafish in data set: A sound argument is forwarded to include this new dataset.

Requests clarification of GEI fathead minnow analysis and its exclusion: This request can be reasonably addressed in the draft document.

Salinity freshwater distinction: Guidance should be included to address these concerns.

Se speciation: The comments addressing plant Se speciation are correct in that the draft text is overly simplified and dated in its discussion of plant Se. Mesocosm studies will also adopt a test water that will influence Se speciation and thus similar Se species exposure concerns will be present as will transferability or differential sediment/particulate/container reactivity of Se species in the test system.

Site-specific criteria: There are numerous public comments that should be addressed in guidance for implementation.

Tiered criteria: There are numerous public comments that should be addressed in guidance for implementation.

Tissue criterion: There are numerous public comments that should be addressed in guidance for implementation.

Translation: There are numerous public comments that should be addressed in the draft document.

Update data set: If practical and possible, this is always a consideration.

Water column values: The concerns should be addressed in the draft document text.

Wildlife criterion: It is apparent from FWS comments that there is significant concern with the draft

criteria potential for protection of aquatic dependent wildlife and fish as well. The pathway for further consideration and development of protection proposed in the draft document appear reasonable to move CWA requirements forward.

Winter stress: Comments opine on winter stress exclusion.

Ww to dw conversions: The comment should be addressed in the draft criterion text as best as possible. It is unlikely that the variability of WW-DW can be uniformly captured in a standardized approach.

PEER REVIEW COMMENTS FROM

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External Peer Review of the Draft Aquatic Life Ambient Water Quality Criterion for Selenium – Freshwater 2014

Responses to Charge Questions by Dr. Vince Palace

PART I: OVERARCHING QUESTIONS

1. Please comment on the overall clarity of the document and construction of the criterion statement with its multiple elements.

The *Draft Aquatic Life Ambient Water Quality Criterion for Selenium in Freshwater* is generally clearly written and logically organized. While there are some issues which require clarification in the document, these generally arise as technical issues (identified in subsequent sections of this review) rather than writing clarity within the document. In contrast to some of the public comments, this reviewer believes that the document clearly states the order of preference for criterion (e.g., egg/ovary over muscle and whole body over water column concentrations) and the ultimate primacy of the egg/ovary criterion. The lone issue of clarity in the document concerns the water column values of selenium. Table 15 (page 97) specifies that water column selenium concentrations are based on "dissolved total selenium in water" however, elsewhere in the document the criterion is described as including "all oxidation states (e.g., selenite, selenate, organic selenium, and any other form)". While clarity regarding the species and analytical methods for assessing water column selenium are provided in Appendix J (Analytical Methods for measuring Selenium), a more precise definition of water column Se is warranted within the body of the document.

2. EPA has developed a tiered selenium criterion with four elements, with the fish tissue elements having primacy over the water-column elements, and the egg-ovary element having primacy over any other element. Inclusion of the fish whole-body or fish muscle element into the selenium criterion ensures the protection of aquatic life when fish egg or ovary tissue measurements are not available, and inclusion of the water column elements ensures protection when fish tissue measurements are not available

    a. Please comment on the tiered construction of the selenium chronic criterion; is it logical, and scientifically defensible as it applies to protection of freshwater aquatic life:

        i. That is, is the primacy of the egg-ovary element over the other elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

The tiered construction of the chronic criterion is logical and scientifically defensible, and the primacy of the egg/ovary element over all other elements is also defensible. In fact, the primacy of the egg/ovary criterion was also recognized by a multidisciplinary and international group of selenium experts convened at a workshop in 2009. Proceedings from that workshop were published (Chapman et al. 2009) and in the executive summary it was noted "Selenium concentrations in eggs are the best predictors of effects in sensitive egg-laying vertebrates". Additional sections of that volume further supports the USEPA's Draft Document approach by recommending that measurement endpoints for risk assessment should be as closely associated with reproductive endpoints in egg laying vertebrates as possible and that measurements in eggs or ovaries, or in the absence of these measures, selenium concentrations in muscle or whole body are required. The scientific evidence supporting these conclusions has not changed substantively since the

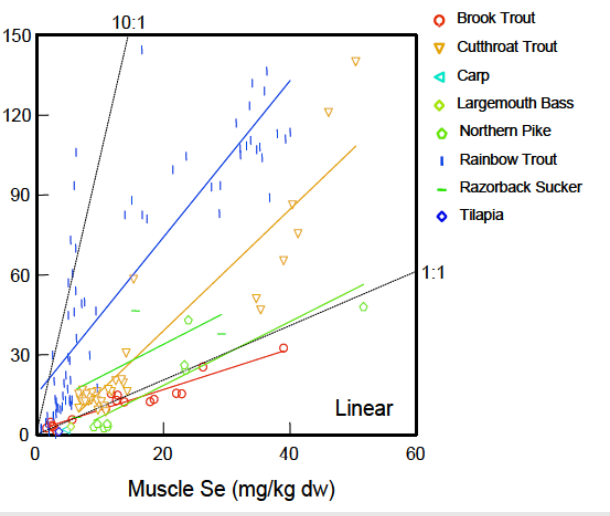
time of that volume's publication and the approach remains the most valid scientifically. In fact, this general approach was also recently adopted by the British Columbia Ministry of the Environment (BC MoE 2014) after an extensive, and peer reviewed, analysis of the literature relevant to the ecotoxicology of selenium.

It is unclear however, how the USEPA will interpret the "never to be exceeded" criteria. Biological variability, coupled with uncertainty regarding the residence of mobile fish species, will make it likely that some fish in a given collection may exceed the guidelines. It is unclear if a result from one fish (i.e. a single exceedance) will render a given management area in non-compliance, or if some average value is intended as the trigger.

ii. Is the primacy of the whole-body/ fish muscle element over the water column elements scientifically sound, given the prevailing toxicological knowledge and the data and supporting scientific information currently available to the Agency? Please provide detailed comments.

Affording primacy to the measurement of selenium in tissues over measurements in the water column is scientifically sound. While egg/ovary are recognized as the best predictors of potential impacts of selenium in oviparous vertebrates, there may be situations where these tissues are not available or where technical expertise is not sufficient to allow collection. In this instance, muscle or whole body measures are the next best alternative to egg/ovary as a risk assessment tool. The use of water column concentrations of selenium as environmental assessment tools or as triggers for additional assessment is fraught with uncertainty from several sources, which are discussed in subsequent sections of this review.

However, it is important to recognize that the use of these tissues for monitoring purposes introduces a layer of uncertainty with regard to potential reproductive toxicity assessments. This uncertainty arises because selenium partitions between egg/ovary and muscle/whole body differently in different species. For example, regression plots of selenium concentrations in eggs versus those in muscle of 8 fish species revealed vastly different slopes and strengths of regression between species (see figure below reproduced directly from North America Metal Council ([NAMC] 2008), y axis scale is Egg Se (mg/kg dry weight (dw)). Due to this divergence, in order for muscle to be used as an effective surrogate for concentrations of selenium in egg/ovary, the specific regression for the fish species in question will have to be documented.



With regard to whole body as a criterion, it is unclear whether the USEPA intended to include visceral tissues (e.g., liver, kidney, gonads, gastrointestinal tract) with the carcass for whole body measurements of

selenium. Because these tissues can account for a significant amount of the whole body pool of selenium, when Se concentrations in liver and gonads are elevated (especially during oogenesis [i.e. egg formation]), this requires clarification.

iii. Please comment on the scientific uncertainty that may be associated with this tiered approach? Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

Uncertainty associated with the US-EPA's tiered approach arises from the species specific disposition of selenium into egg/ovary versus muscle and whole body (noted above) and in the sampling methods used to obtain these tissues. In terms of sampling methods, timing may contribute to variability. A recent study showed that some fish may partition selenium to the eggs/ovaries more immediately from the diet than from their tissue stores (Conley 2014). In these species of fish, muscle/whole body might be less reflective of egg/ovary selenium concentrations than concentrations in the diet. However, the authors noted that spawning strategy may play a role in determining the importance of tissue reserves versus dietary sources accounting for selenium partitioned to egg/ovary. Specifically, for species with longer periods of oogenesis and which spawn only once annually, tissue stores may be better predictors of egg/ovary selenium concentrations than dietary sources. However, for multiple spawners, the diet may be a more important determinant. This has relevance to the both the egg/ovary and muscle/whole body criteria recommended by the US-EPA and the variability inherent in each. If muscle/whole body were used as a measure of compliance the timing of sampling within the fishes' reproductive cycle could have an influence on the concentration of selenium in the tissue, especially among single spawners with extended oogenesis periods. Therefore, if muscle/whole body were sampled immediately following the spawning period lower concentrations of selenium might be expected than if the tissues were sampled prior to oogenesis.

Another source of variability concerning the application of muscle and whole body as a criterion concerns a precedent that USEPA has established with regard to conversion of concentrations in one of these tissue types to another. While the Draft Document acknowledges that matched pairs of muscle and whole body concentrations of selenium were assessed for each species, only a few fish species provided data for assessing the conversion (Page 78). As a result, USEPA used the median ratio for all species (i.e., 1.27) to convert muscle selenium to whole body concentrations. In the absence of additional species specific conversion ratios, continued use of this generic ratio would be expected to introduce additional variability. For example, and with reference to derivation of the egg/ovary criterion for the Draft Document, variability would be expected to have arisen from the fact that almost half (i.e., 7 of 16) of the Conversion Factor (CF) values for egg/ovary to whole body were derived using the generic muscle to whole body conversion ratio.

iv. Are the draft recommended magnitude, duration, and frequency for each criterion element scientifically sound and appropriate? Please provide detailed comments.

As noted in our response above, there is some confusion regarding how "never to be exceeded" concentrations of selenium in the tissue based criterion will be applied (i.e., is this applied to analysis of single fish or to arithmetic or geometric means from sampled populations?). Clarification on this question is required before the scientific defensibility of the duration and frequency can be assessed for the two tissue based criterion.

With regard to the magnitude of the tissue based elements, it would appear that at least two issues may challenge the scientific defensibility of these criteria. First, it is our understanding that the egg/ovary

criterion was developed from EC10 values derived from the literature. Where multiple results of acceptable quality for a given species were available, a geometric mean was calculated. In the case of the EC10 for bluegill (*Lepomis macrochirus*), the mean EC10 resulted from 4 studies, published by three authors: Hermanutz et al., 1992 and 1996, Doroshov et al. (1992) and Coyle et al. (1993). However, the EC10 value calculated from the Hermanutz et al. studies (=12.7mg/kg) is quite different from the values rom the other two studies (20 and 24.6 mg/kg respectively), indicating cause for investigation of the reasons for the difference, especially in light of their importance for determining the egg/ovary tissue based criterion. One of the supplemental comments provided as additional information with this package (Docket ID EPA-HQ-OW-2004-0019-0331) indicates that the TRAP model plot of the Hermanutz et al. data provide a poor fit. While we were not afforded access to figure 1, which cited in that docket submission, if the data are indeed poorly fit, it is appropriate to consider them questionable and eliminate them from the geometric mean calculation for this species.

A second, and potentially more serious issue with regard to the magnitude of the egg/ovary tissue based criterion, is the reliance on the reanalysis of data from the brown trout (*Salmo trutta)* study (Formation 2011). Uncertainty in this study arises because some fry escaped from their respective incubation chambers and could not be assigned to a given treatment. As a result, several scenarios were calculated based on whether the escaped fry had similar deformity rates relative to the retained fry, were all deformed, or were all normal. While this cannot be resolved, the criterion was calculated based upon the most conservative approach: that all fry were dead or deformed. This conservative approach to calculating an EC10 value for brown trout result in it being the most sensitive species, thereby affecting the overall egg/ovary criterion. Subsequently, because other criterion (i.e., muscle/whole body and the water based criterion) are back calculated based on the egg/ovary value, conservatism is compounded in the values for these criterion as well.

For the water column based criterion, two separate elements are prescribed in the Draft Document: a monthly average and a separate element for intermittent (discontinuous) exposures. Each of these is further delineated to apply to either lentic or lotic systems. Presumably the definitions for lotic and lentic systems would be based on residence time of water or some related criteria, but the Draft Document does not contain an explicit definition of either type of system. Back calculating from egg/ovary to muscle/whole body and then down through trophic levels to derive allowable water column criterion for each of these types of aquatic systems is not scientifically valid, because of the use of generic conversion factors and broadly based trophic transfer factors. These generic terms do not incorporate site specific information, including concentration dependent uptake kinetics and consideration for important influencing factors (e.g., sulfate). The water based criterion is therefore, conservative and variable. As evidence for this, the monthly average exposure value for lentic systems is 1.3 μg/L. This value is at the upper end of background values for freshwater and may be exceeded even in the absence of industrial inputs in areas receiving runoff from seleniferous soils. The value is also lower than recently recommended lentic values based on similar analysis (2 and 2.1 μg/L respectively (Deforest et al., 2104, BC MoE 2014).

PART II: FISH TISSUE CRITERION ELEMENTS DERIVATION: DERIVATION OF FISH EGG-OVARY, WHOLE BODY AND MUSCLE CRITERION ELEMENT(S)

EPA is requesting a technical review of the methods and procedures used to derive a chronic selenium criterion based on an egg-ovary concentration, as well as its translation to a criterion element applicable to whole-body and muscle tissue. Please address the following questions:

1. Please comment on EPA's use of the effects concentration 10th centile (EC_{10}) as the measurement endpoint for the fish reproductive toxicity studies used to derive the egg-ovary element.

The slope of the response curve for selenium rates of deformities plotted against selenium concentrations in eggs/ovaries rises rapidly above the EC10 value. Therefore, use of the 10th percentile as the measurement endpoint is scientifically defensible, appropriate and consistent with USEPA's assessment of toxicity of other compounds as well as the assessment of reproductive toxicity in other jurisdictions (BC MoE 2014).

2. Data used to derive the final chronic egg-ovary criterion element were differentiated based on the type of effect (reproductive vs. non-reproductive effects). Acceptable chronic toxicity data on fish reproductive effects are available for a total of nine fish genera. The genus Sensitivity Distribution (SD) is predominantly populated with data on fish genera because field evidence demonstrates that fish communities can be affected by selenium even when there is no observable change in the invertebrate community diversity and abundance. As a result, decades of aquatic toxicity research have focused primarily on fish. Available field and laboratory studies indicate that invertebrates are more tolerant to selenium than most of the tested fish species (Criteria document, Table 6c, Section 4.1.2). The data set used to derive the selenium criterion marks a change from the traditional method used to derive water quality criteria that requires toxicity tests with aquatic organisms from 8 phylogenetically distinct taxa (including three vertebrate and five invertebrate genera) in order to derive aquatic life criteria (Stephan et al., 1985).

    a. Given selenium's more taxon-specific and life stage-specific toxicity, please comment on EPA's use of the available data to derive the egg-ovary tissue element.

The use of reproductive effects in fish to derive the sensitivity distribution is appropriate because non-reproductive effects may arise from mechanisms that are not central to the primary ecological effects of selenium; reproductive toxicity in oviparous vertebrates manifested by maternal transfer of selenium to eggs. Additionally, as noted in the Draft Document, non-reproductive effects thresholds are highly variable and provide less confidence for deriving threshold values for selenium. The use of data from fish as the most sensitive organisms is appropriate and likely to be protective of invertebrates. However, it should be noted that sensitivity among invertebrates is highly variable and that some invertebrate taxa do exhibit sensitivity at low μg/L concentrations (see BC MoE 2014 for a review of this data).

While we agree that the Draft Document predominantly uses data from fish generally sensitivity, the approach in the Draft Document is not a complete departure from the principles surrounding the use of eight phylogenetically distinct taxa. The US-EPA has attempted to increase taxonomic coverage of the sensitivity distribution by converting results from studies of three invertebrate taxa into fish reproductive endpoints. Specifically, threshold concentrations of selenium in the invertebrates were converted to predicted fish concentrations of selenium in egg/ovary based on consumption of the invertebrates by fish. These values were then included in the fish distribution (Figure 5, page 58). The variability inherent in this calculation is large because a generic trophic transfer factor of 1.27 was applied to convert invertebrate

concentrations to fish whole body concentrations and then a generic conversion factor of 1.71 was applied to convert whole-body concentrations to egg/ovary. The result is a highly variable, and scientifically questionable, series of three additional data points that were added to the distribution of reproductive effects for fish.

b. Given the greater general sensitivity of oviparous fish to selenium compared to aquatic invertebrates, please comment on the appropriateness of EPA's fish tissue-based criterion for affording protection to the aquatic community as a whole (e.g., including invertebrates).

As noted above, the use of data from oviparous fish as the most sensitive aquatic organisms to derive criterion is appropriate and likely to be protective of invertebrates. However, the USEPA may wish to consider sensitivity data for some invertebrate taxa that do exhibit sensitivity at low μg/L concentrations (see BC MoE 2014 for a review of this data).

c. With respect to the tests that quantified non-reproductive effects, did the EPA use that data to the best extent possible given its limitations (e.g., relevance compared to reproductive tests, and data quality concerns which increased uncertainty (e.g., Hamilton et al., 1990)?

Because non reproductive tests do not evaluate the most sensitive measure of selenium ecotoxicology, their use as regulatory criteria are questionable. However, the USEPA has provided summaries of non-reproductive tests and compared the results from these studies with the criterion derived using reproductive data. In most cases, the studies have evaluated growth or survival of fish. The species mean chronic values (SMCV) and genus mean chronic values (GMCV) from the non-reproductive tests are generally greater than the egg/ovary criterion and, therefore, it is expected that the criteria derived from the reproductive studies (e.g., Egg/ovary) will be protective of non-reproductive endpoints as well.

d. EPA also rejected studies that used the injection route of exposure for selenium due to uncertainty related to uptake, distribution and metabolism/transformation kinetics when compared with the dietary and/or maternal transfer routes of exposure. Was this reasonable? Does the panel envision an appropriate and scientifically defensible use for this type of data? Please provide detailed comments.

The US-EPA rejected the Doroshov et al. (1992) study in which female catfish were injected intramuscularly with seleno-methionine and effects were determined in their offspring. The chemical form of selenium was appropriate for injection into these fish, but it could be argued that injection circumvents dietary uptake, tissue partitioning and timing of muscular uptake with respect to reproductive cycle of the fish. Some may therefore consider this injection study to be invalid. However, relating selenium concentrations in egg/ovary to reproductive effects was the primary focus of the USEPA's assessment. While several compromises have been established to allow data to be included in the development of the criterion (see discussion of the bluegill and brown trout data from earlier comments), the exclusion of the data from the Doroshov et al. (1992) study appears arbitrary. Moreover, citing abundance of Ictalurids in the Hyco Reservoir (Crutchfield (2000) and at Belews Lake (Young et al. 2010) at selenium concentrations that may have affected abundance of other fish species is not sufficient evidence to dismiss the data from the Doroshov et al. (1992) study. A reexamination of the data and consideration to include them in the egg/ovary criterion is warranted.

3. Was the method (Section 4.1.5, 7.1.7) used to translate the fish egg-ovary criterion element into muscle and whole body criterions elements understandable, transparent and scientifically defensible? Was there sufficient data for making the translations for each element?

The methods used to translate egg/ovary to muscle and whole body criteria are understandable and transparent, but as we noted in our earlier comments, there are scientific issues with some of the transformations. The USEPA attempts to use matched pairs of muscle and whole body concentrations of selenium for each species, but only a few fish species provided data for directly assessing the conversion (Page 78). As a result, US-EPA used the median conversion value for all species (i.e., 1.27) to convert muscle selenium to whole body concentrations where species specific data were not available. Continued use of this generic ratio would be expected to introduce additional variability and uncertainty, particularly for the conversion from egg/ovary to whole body because in many cases this requires a two step conversion (i.e., from egg/ovary to muscle and then from muscle to whole body). More specifically, almost half (i.e., 7 of 16) of the Conversion Factor (CF) values for egg/ovary to whole body were derived by including the generic muscle to whole body conversion ratio. The issue is less important for conversion of egg/ovary to the muscle criteria because for most species (other than desert pupfish) there were data available to calculate the conversion directly.

PART III: EVALUATION OF THE TRANSLATION PROCEDURE TO DERIVE THE WATER COLUMN ELEMENT(S)

EPA is also requesting a technical review of the methods and procedures used to translate the egg-ovary element of the chronic selenium criterion to water-column elements. Relevant sections of the document include:

- A description of the method used to derive an equation to translate the egg-ovary element to a monthly water-column element in perennial (lentic and lotic) waters and an equation that can be used to convert the monthly water-column element to an intermittent water column element (Sections 3.8.3, 3.8.4, 4.2.1, 4.3, and Appendix G).

- An analysis of the translation equation precision using data obtained from published literature (Sections 7.2.1, 7.2.2, and Appendix H).

- A description of the method and data sources used to derive the translation equation parameters (Sections 4.2.2, 4.2.3, and Appendix B).

- A description of the method and data sources used to categorize waterbody types where a single water-column chronic criterion concentration value would be adequately protective in most circumstances (Section 4.2.4).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for established categories of waters (Section 4.2.5).

- A description of the method and data sources used to derive water-column chronic criterion concentration values for intermittent discharges that may occur in lentic and lotic waterbodies (Section 4.3).

Please address the following questions:

1. Please comment on the scientific defensibility of EPA's translation equation method for translating the concentration of selenium in fish tissue to a concentration of selenium in the water-column. Please comment on major sources of uncertainty in applying the translation equation to different types of waterbodies (e.g., with differing retention times, water chemistries, and/or species present). Are there other data sources, models, or approaches that EPA should consider that would reduce uncertainty? Please provide detailed comments.

The scientific method for translating concentrations of selenium in fish tissues to allowable concentrations in the water column is clearly written and understandable. However, while we understand the regulatory need for triggers to initiate site investigation where selenium is suspected of being an issue, the derivation of allowable water column concentrations from eggs or ovaries is oversimplified and likely to need site specific inputs for refinement. Back calculating from egg/ovary to muscle/whole body and then down through trophic levels to derive allowable water column criterion for each of these types of aquatic systems is not scientifically valid, because of the use of generic CF, assumptions regarding proportions of prey items consumed by resident fish and broadly applied trophic transfer factors. These generic terms do not incorporate site specific information, including concentration dependent uptake kinetics and consideration for important influencing factors (e.g., sulfate, organic carbon, temperature,etc.). The water based criterion developed in the Draft Document are therefore, necessarily conservative. As evidence for this, the monthly average exposure value for lentic systems is 1.3 μg/L. This value is at the upper end of background values for freshwater and may be exceeded even in the absence of industrial inputs in areas receiving runoff from seleniferous soils. The value is also lower than recently recommended lentic values based on similar analysis (Deforest et al. 2104, BC MoE 2014).

2. Regarding the trophic transfer factor (*TTF)* values, did EPA use a scientifically defensible method to derive the *TTF* values (p. 71-77 of the criteria document)? Were the exclusion criteria, (pp. 71-77 of the criteria document) developed by EPA to screen the available data applied in a consistent and scientifically defensible manner? In particular, EPA noticed that application of the exclusion criteria resulted in *TTF* values for aquatic insect larvae that differ from other published values. Given this, are you aware of any other methods of screening data that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included, if appropriate? Please provide detailed comments.

The derivation of Trophic Transfer Factors (TTF) by the US-EPA in the Draft Document is clearly outlined and presented. However there are several issues which, again, result in the introduction of error and therefore an element of conservatism in the data that was derived. For example, the USEPA matched selenium concentrations in consumers and their likely prey items from a thorough investigation of the available data. However, where matched data from more than one prey item was identified from a site, the median of lower trophic organisms was used to calculate a TTF. While we understand the rationale for this practice from a data handling perspective, by not acknowledging that prey items may comprise different proportions of the diet ultimately introduces variability in the calculated TTF, with the potential for an influence in either direction. Additionally, while the US-EPA presents a statistical argument for the validity of matching pairs of samples taken from an aquatic site over a year, it is also acknowledged that some sites may present selenium loads or bioaccumulation kinetics that require different collection time criteria. Recognizing that the Draft Document will largely be applied to impacted receiving environments that are influenced by industrial activity and which present dynamic ranges in selenium loading, it appears likely that establishing a precedent to allow matching concentrations of selenium in aquatic compartments collected a year apart will, in most cases, not be appropriate. Finally, the USEPA designated single TTF

based on the median value of only those regressions that were significant (Page 75). While this is a conservative approach, it does not fully incorporate consideration for differential uptake among lower trophic organisms at varying concentrations of selenium exposure.

3. Regarding the conversion factor (*CF*) values used, did EPA use an appropriate and scientifically defensible method to derive those values (p. 78-79 of the criteria document and Appendix B)? Are you aware of any other methods that EPA should consider? Also, are you aware of any data that was not considered in this effort and should be screened and included? Please provide detailed comments.

As noted in our response to Charge Question #2, almost half (i.e., 7 of 16) of the Conversion Factor (CF) values for egg/ovary to whole body were derived using a generic (i.e., not species specific) muscle to whole body conversion ratio that was calculated as the median value of the available data for all fish species. This practice will have likely contributed to the variability in the dataset.

4. Regarding the derivation of enrichment factor (*EF*) values, was the method EPA used to screen data from the literature applied appropriately and consistently (see inclusion/exclusion criteria on p. 71-77 of the criteria document)? Was the method for deriving *EF* values applied to those data in a consistent manner so as to derive *EF* values for selected waters in a scientifically defensible manner? Is the method that EPA used to establish the lentic and lotic categories for *EF* values reasonable given the available data? Are you aware of other methods or relevant data the EPA should consider? Please provide detailed comments.

Derivation of Enrichment Factors (EF) based on paired concentrations of selenium determined in water and particulate would have been influenced by the practice of allowing data to be paired if they were collected up to a year apart. In terms of application of EF to categories for lentic and lotic systems it is difficult to judge because of the lack of specific criteria to distinguish between the two types of systems in the Draft Document. While the US-EPA acknowledges the importance of residence time for defining aquatic systems as either lentic or lotic, the criterion for their initial assignment to each category is not apparent (Page 82). Despite statistical comparisons that support their aggregation, it is very likely that lakes, reservoirs, ponds and marshes will have vastly different selenium kinetics, and yet they are all designated as lentic systems. Likewise, selenium uptake into aquatic food-webs of creeks, drains, washes, rivers and streams may differ markedly. The wide range of variability in the aggregated categories (Figure 10, page 84) is compelling evidence in support of this point. Additional specific guidance is required to distinguish between the two types of aquatic systems and the applicability of EFs for each.

5. Please comment on the scientific defensibility of EPA's conversion of the selenium fish tissue – water translation equation into an equation that allows for calculation of a criterion for waters that may be subject to intermittent discharges of selenium. Please comment on major sources of uncertainty in this approach. Is this method appropriate, given the bioaccumulative nature of selenium? Please comment on the uncertainty associated with the application of this conversion equation to intermittent discharges that may occur in different types of waterbodies and/or in different locations, particularly with respect to loads transported to potentially more sensitive aquatic systems. Does the method employed result in criteria that are similarly protective to the 30-day chronic criterion? Are there any other models or approaches that EPA should consider that would reduce this uncertainty? Please provide detailed comments.

It is not clear how the intermittent criterion outlined in the Draft Document will be applied. The mathematical expression of the criteria on page 93 is clear but the terms surrounding the application of the

criterion are not. For example, the criterion is not intended to apply to "ordinary smoothly varying concentrations" (Page 94). However, what specifically will constitute a discharge curve that is not "smooth" has not been defined. It is also not clear what magnitude of selenium concentration spikes would designate a discharge as having to be regulated as an intermittent discharge. Finally, designation of an intermittent criterion appears to contradict the data in Appendix G and the statement on page 94 that "kinetics of selenium accumulation and depuration are sufficiently slow that attainment of the water criterion concentration element by ambient 30 day averages will protect sensitive aquatic life species even where concentrations exhibit a high degree of variability. While outside the area of our expertise it is noted that several comments in the public registry suggest that a biokinetic model may be more appropriate than the application of an expansion of the 30-day average calculation for determining intermittent criterion.

PART IV: SIGNIFICANCE OF SCIENTIFIC VIEWS FROM THE PUBLIC/STAKEHOLDERS

EPA will also be providing scientific views and other comments from stakeholders and the public received via the public docket to the peer review panel. Although EPA will be providing the full contents from the docket, EPA is only requesting a review of any scientific views/public comments that may be of technical significance to the selenium criterion.

1. Has the peer review panel identified any scientific views from the public or stakeholders as being technically significant to the draft of the selenium criterion going forward; that is, has information or data been introduced during the comment period that would change the scientific direction of the criterion? Is there any information or data that may refine or enhance the scientific defensibility of this criterion that EPA should consider further? Please provide detailed comments on specific issues of technical significance or refinement.

Relevant comments from the public or stakeholders have been acknowledged where they are relevant to the other charge questions above. No further specific issues arising from our review of the public comments are noted.

References

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