



Charge for Peer Review of “Analysis of the Wild Rice Sulfate Standard Study”

Introduction

The Minnesota Pollution Control Agency (MPCA) has contracted with Eastern Research Group, Inc. (ERG) to convene and facilitate a scientific peer review of the Analysis of the Wild Rice Sulfate Standard Study: Draft for Scientific Peer Review (Analysis). This peer review is the next step in MPCA’s ongoing efforts to enhance scientific understanding of the effects of sulfate on wild rice. This enhanced scientific understanding will inform MPCA’s review of the wild rice sulfate standard and the development of a rulemaking proposal, if warranted, regarding that standard.

Background and Current Standard

Water quality standards are fundamental tools under the federal Clean Water Act (CWA) and Minnesota Statutes, designed to help protect and improve the quality of the state’s lakes, streams, wetlands and groundwater. Minnesota water quality standards consist of three components:

1. The beneficial use(s) for which a water body is to be protected,
2. The narrative and/or numeric criteria that specify what conditions in the water are protective of the beneficial uses, and
3. Antidegradation provisions to minimize the lowering of water quality that is better than the minimum level needed to protect beneficial uses.

Under the CWA, states and federally authorized Indian Tribes are required to identify the beneficial uses for which their waters are to be protected, then to adopt criteria and antidegradation provisions to protect those beneficial uses. The CWA also requires that water quality standards be regularly reviewed to solicit public input and incorporate new science. MPCA is the state agency responsible for this work in Minnesota.

One of the beneficial uses identified in Minnesota’s water quality standards rules is “water used for production of wild rice” (Minnesota Rules, Part 7050.0224, subpart 2). Wild rice is an important plant species in aquatic environments in parts of Minnesota, particularly northern Minnesota. It provides food for waterfowl, is economically important to those who harvest and market wild rice for human consumption, and is also a very important cultural resource for many Minnesotans.

This recognition of the importance of wild rice in Minnesota, and an observed relationship between the presence of wild rice in waters with lower sulfate levels (and its absence in waters with elevated sulfate), led to the adoption of a 10 milligrams per liter (mg/L) sulfate standard in 1973 applicable to water used for production of wild rice. Based on testimony presented at public hearings leading to the adoption of the sulfate standard, it was intended to apply both to waters with naturally occurring wild rice and to waters used for paddy rice production.

The standard was based on field observations and water chemistry correlations made by Dr. John Moyle primarily in the late 1930s and early 1940s, who concluded that “No large stands of rice occur in water having sulfate content greater than 10 ppm [parts per million, or mg/L], and rice generally is absent from water with more than 50 ppm. ”

The existing wild rice sulfate standard was developed based on correlations of Dr. Moyle’s observations and water chemistry data. However, the specific mechanism by which sulfate appears to be impacting wild rice was not the subject of Dr. Moyle’s study. This, along with questions that have arisen regarding the implementation of the current standard, led to interest in further understanding the effects of sulfate on wild rice to inform a review of the wild rice sulfate standard.

Overview of the Wild Rice Sulfate Standard Study

In 2010, MPCA initiated a multi-year effort to clarify implementation of the state’s wild rice sulfate standard. As part of this effort, the state legislature funded a study to gather additional information about the effects of sulfate and other substances on the growth of wild rice. This research was intended to inform an evaluation of the existing wild rice sulfate standard.

Following the development of a detailed research protocol in 2011, in 2012 the MPCA contracted with groups of scientists at the University of Minnesota Duluth and Twin Cities to undertake the Wild Rice Sulfate Standard Study. The Study’s main hypothesis is that wild rice is impacted by sulfate via the conversion of sulfate to sulfide dissolved in the water in the sediment, known as the sediment porewater. The Study consists of five components:

1. **Controlled Laboratory Hydroponic Experiments:** Controlled laboratory hydroponic experiments determined the effect of elevated sulfate and sulfide on seed germination and early stages of wild rice growth and development.
2. **Field Survey of Wild Rice Habitats:** A field survey of wild rice habitats was conducted over two field seasons (2012-2013) to investigate physical and chemical conditions correlated with the presence or absence of wild rice, including sulfate in surface water and sulfide in the sediment porewater of the rooting zone. The survey crews also sampled the mesocosms (below) on a monthly basis during 2013. This field survey was supplemented by the data collected during a pilot field survey in 2011.
3. **Mesocosm Experiment:** An outdoor container wild rice growth experiment conducted in 2013, using natural sediments, determined the response of wild rice to a range of sulfate concentrations in the surface water, and associated sediment porewater sulfide concentrations in the rooting zone. This mesocosm experiment continued an experiment conducted by the principal investigator during 2011-2012 under separate funding.
4. **Collection and Analysis of Rooting Zone Depth Profiles:** Depth profiles of dissolved chemicals were collected in the mesocosms and at two field sites to characterize sulfate, sulfide, and iron.
5. **Sediment Incubation Laboratory Experiment:** Sediment was incubated in the laboratory, without wild rice, to explore the difference that ambient temperature has on the rate at which elevated sulfate concentrations in water enter underlying sediment and convert to sulfide, and to what degree sulfate is later released back into the overlying water.

Each of the Study components has a specific purpose and associated strengths and limitations. Data collection was completed in December 2013 and is documented in individual reports from the researchers.

Overview of MPCA Analysis

During the first half of 2014, MPCA staff integrated the Study results; analyzed the data as a whole; gained input from the Wild Rice Standards Study Advisory Committee; and reviewed existing monitoring data, other relevant scientific studies/information, and the original basis for the wild rice sulfate standard to develop an analysis of the Study results (the Analysis). The MPCA first developed a preliminary analysis and then refined the analysis based on feedback received and MPCA scientists' continued data analysis and interpretation. MPCA's Analysis draws most heavily from the data collected in the hydroponics experiments, the field survey of wild rice habitats, and the mesocosm experiment. The sediment incubation experiment provides some insight into sulfate-sulfide conversion dynamics, and results from the rooting zone depth profiles can be used to characterize the effect of elevated sulfate concentrations on rooting zone geochemistry and to define seasonal differences in the rooting zone geochemistry from both mesocosm and field sites. However, though the results from the rooting zone depth profiles and sediment incubation experiment are briefly summarized, further analysis and possibly additional study are needed before general findings can be drawn from these two Study components.

Purpose of the Peer Review

Independent scientific peer review is the next step to ensure that MPCA's scientific work is technically sound, and therefore can be relied upon to inform future work or decision-making. Any change to the water quality standard must be based on a sound scientific rationale; any change may also be controversial. Normally, MPCA relies on published peer-reviewed scientific studies during the development or revision of water quality standards. In the case of the Study, MPCA is analyzing the Study results prior to publication of those results in scientific journals. Therefore, MPCA is undertaking this peer review to receive independent scientific feedback on MPCA's Analysis.

It is important to note that this is not a review of the existing wild rice sulfate standard, or any recommendations for changes to the standard. MPCA has not yet developed policy recommendations or a proposal regarding any changes to the wild rice sulfate standard. Rather, MPCA will consider the peer reviewers' comments as the agency further refines MPCA's Analysis of the effects of sulfate on wild rice. If warranted, a Technical Support Document will be developed to describe the scientific basis for any proposed changes to Minnesota's water quality standards.

This peer review is a step in the larger process in which MPCA will consider available information to determine if changes to the wild rice sulfate standard are needed. MPCA will also seek informal and formal public comment on any recommendations and rulemaking proposal that are developed. Any proposed change to the wild rice sulfate standard would be adopted into Minnesota's water quality standard rule (Minnesota Rules Chapter 7050) in accordance with the procedural requirements of the Minnesota Administrative Procedures Act and would require the approval of the US Environmental Protection Agency.

Charge to Reviewers

ERG is conducting this peer review under contract to MPCA. The review questions consist of 10 specific questions, followed by three general questions. In conducting the peer review, reviewers are asked to refer particularly to the following sources:

- MPCA Analysis, and associated references.
- The individual report for each Study component, as needed, to further understand and evaluate the MPCA Analysis.
- The MPCA's wild rice sulfate standard web page <http://www.pca.state.mn.us/ktqh1083> for more background information about the Study and Analysis. A link to an FTP site with all the Study reports and data is also available on this page.

Laboratory Hydroponic Experiments (see Analysis, pp. 13-16, 38-39)

The hydroponic experiments involved a series of aquatic toxicity tests designed to evaluate the relationship between a controlled exposure of wild rice plants to a dilution series of sulfate or sulfide concentrations, and the biological responses observed in the plant's growth and development. The sulfide seedling experiment involved immersing photosynthesizing seedlings in an anoxic sulfide solution. Over time, sulfide was oxidized by the oxygen the seedlings produced. This led to a decrease in the sulfide exposure concentration between each renewal of the test solution. MPCA staff relied on the initial sulfide concentration, rather than the lower sulfide concentrations that developed between renewals, as the operative exposure concentration in this Analysis, as this was the highest, and presumably most toxic, concentration to which the plants were exposed. It is uncertain if the leaves of seedlings would ever be exposed to sulfide in a natural setting (see pages 38-39 of the Analysis).

Charge Question 1: Discuss the appropriateness of the sulfide seedling hydroponic test method and performance in evaluating the hypothesis that elevated sulfide in the sediment porewater can be toxic to wild rice.

Charge Question 2: Is it reasonable to use the initial exposure solutions as the operative exposure concentration for the test? Why or why not? If not, what approach do you suggest?

Charge Question 3: Is regression analysis to derive EC20 and EC50 values an appropriate way to analyze the sulfide seedling hydroponic data to identify effect levels? Why or why not? Is there an alternative approach to evaluate the data for effect levels that you would suggest the MPCA pursue?

Utility of the Field Survey Data (see Analysis, pp. 21-25, 35-36, 41-47)

Statisticians recommend that surveys be probability-based when the point of the survey is to characterize the population being sampled. Probability-based surveys allow the survey results to be extrapolated back to the larger population. The field survey site selection was purposefully not probability based, in that the point was not to characterize the population of wild rice production waters but rather to explore the effect of elevated sulfate on the chemistry of the porewater of actual and potential wild rice habitat.

Charge Question 4: Discuss whether the Analysis demonstrates that the lake and stream field survey data and results are sufficiently representative of Minnesota lakes and streams with wild rice to 1) examine the chemical relationships between sulfate in surface water and acid-extractable iron, acid-volatile sulfide, and porewater concentrations of sulfide and iron, and 2) inform protection of wild rice from elevated sulfate. Please note any specific questions or concerns.

Mesocosm Experiment (see Analysis, pp. 26-32)

Wild rice was grown from seed in 100-gallon polyethylene containers containing sediment from a natural wild rice bed. Concentrations of sulfate in the overlying water were maintained at desired experimental treatment levels, and wild rice was allowed to grow and self-propagate for three seasons (2011-2013). An unusual rate of wild rice mortality occurred in all the containers at the beginning of the 2013 growing season. The mortality may be related to an unusually cold spring and subsequent late start to the growing season. Statistical analysis of the mesocosm experiment data shows significant differences in biological endpoints, such as seed weight, associated with increased sulfate exposure concentrations. The mesocosm experiment provides data on the interactive effects of sulfate, sulfide, and iron on wild rice in a system that is less controlled but more natural than the hydroponic experiments, and more controlled but less natural than the field survey.

Charge Question 5: Does the MPCA Analysis make appropriate use of the mesocosm experiment data? Please describe any suggestions you have about how the data could be further analyzed, or any cautions about the existing or potential use of these data.

Wild Rice in Relation to Sulfate, Sulfide, and Iron (see Analysis, pp. 35-39)

The MPCA Analysis uses the results of the hydroponic experiments to demonstrate that elevated sulfide concentrations are toxic to wild rice seedlings. These data showed deleterious effects of sulfide on seedling plant growth when sulfide exceeded the range of 150 to 300 micrograms per liter ($\mu\text{g/L}$). In addition, this effect is corroborated by data from the field survey and the mesocosm experiment. A histogram of field survey data (Figure 17) shows that 69 to 80% of the sites had wild rice present (above a threshold of 5% cover) when porewater sulfide was less than 75 $\mu\text{g/L}$, and, as porewater sulfide increased, a more-or-less continuous decline in the percent of sites with wild rice present was observed. The Analysis also draws on data from the mesocosm experiment, in which 300 mg/L sulfate treatments exhibited a median sulfide concentration of 778 $\mu\text{g/L}$ that would be predicted to impair wild rice growth based on the EC50 of 383 $\mu\text{g/L}$ estimated from the hydroponic experiments. Seed weights were significantly lower in the highest sulfate treatment.

Charge Question 6: Do you agree or disagree with the MPCA's assertion that the field survey and mesocosm experiment data further support the hypothesis that elevated sulfide in the sediment porewater above 300 $\mu\text{g/L}$ can be toxic to wild rice? Why or why not?

Control of Porewater Sulfide by the Availability of Sulfate and Iron (see Analysis, pp. 21-25, 45-47)

The MPCA Analysis suggests that a quantile regression analysis of the field survey data is an appropriate way to estimate the relationship between sulfate in surface water and the potential concentration of sulfide in porewater, and that the uncertainty in the estimate can be reduced by incorporating the concentration of available iron in the sediment (as measured by an acid extraction).

Charge Question 7: Is the use of multiple quantile regression an appropriate tool for predicting porewater sulfide concentrations? Why or why not? If not, what other options for predicting porewater sulfide would be suitable?

Charge Question 8: In the multiple quantile regression, MPCA relied on the acid-extractable iron rather than the porewater iron to predict porewater sulfide concentrations based on surface water sulfate concentrations. Do you agree or disagree with this approach? Why or why not?

Synthesis: How Sulfate, Sulfide, and Iron Interact to Affect Wild Rice (see Analysis, pp. 51-52)

Elevated sulfate concentrations in surface water are a concern because bacteria convert sulfate to sulfide in the sediment where wild rice seeds germinate and develop roots and leaves. The Analysis demonstrates that the availability of iron mediates the accumulation of sulfide in the porewater of the sediment, and hence the exposure of wild rice seedlings to potentially toxic levels of sulfide. Therefore, based on the Analysis, the potential toxicity of sulfide is jointly controlled by sulfate in surface water and iron in the sediment.

Charge Question 9: The MPCA Analysis focuses on sulfide in the porewater as the sulfur parameter impacting wild rice, and the role of sulfate and iron as key variables controlling sulfide concentrations in porewater. Was this focus appropriate to inform understanding of the effects of sulfate on wild rice? Why or why not? If not, what other variables do you suggest the MPCA explore?

Charge Question 10: Please identify any concerns you have about the Synthesis, particularly any key omissions or assumptions in the logic that should be further evaluated.

General Questions

Charge Question 11: Please state your overall assessment of the five Study components. Did MPCA choose appropriate Study components to meet Study objectives and to support the Analysis? Why or why not?

Charge Question 12: Please provide any other comments you may have on the Study data collection and interpretation, or on the Analysis.

Charge Question 13: Please identify any other issues or critical data gaps for further research that should be considered when evaluating the relationship between wild rice and sulfate.