

12 GRASSLAND BYPASS PROJECT PEER REVIEW

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Compiled by the San Francisco Estuary Institute

OVERALL INTRODUCTION

The Grassland Bypass Project (GBP) manages and reduces the volume of agricultural drainwater and floodwater discharged from the 97,000-acre (44,000-hectare) Grassland Drainage Area (GDA) to the lower San Joaquin River and adjacent wetlands water supply channels. The Project is located in the Central Valley of California (Figure 1). The GBP includes a compliance monitoring project for tracking selenium, salt, boron, and other constituents in water, sediment, and biota throughout the study area (Figure 2). Data are reviewed by the interagency Data Collection and Review Team (DCRT) that consists of technical experts from the US Bureau of Reclamation (USBR), US Fish and Wildlife Service, US Environmental Protection Agency (USEPA), US Geological Survey, California Department of Fish and Wildlife, and the California Regional Water Quality Control Board.

The GBP has been regulated by waste discharge requirements (WDRs) since 1998. The objective of the Project was to consolidate agricultural drainwater and floodwater generated in the GDA and divert these flows through the San Luis Drain, thus drastically reducing discharges into more sensitive areas. Various sites along the waterways now receiving the agricultural drainage, as well as several reference sites, are monitored regularly. The ultimate goal of the Project is to completely eliminate discharges of agricultural drainwater from the GDA by 2019 through highly conservative irrigation methods, reuse of water on salt-tolerant crops, and drainwater treatment within the GDA.

More detailed information, regulatory documents, monitoring plans, use agreements, biological opinions, and other documents can be found at the USBR website: <http://www.usbr.gov/mp/grassland/>. Monitoring results are published by the San Francisco Estuary Institute (SFEI) and are available at <http://www.sfei.org/projects/grassland-bypass-project>.

To ensure the quality and suitability of these monitoring efforts relative to the objectives of the GBP, this peer review was conducted to evaluate and possibly suggest ways to improve performance, and credibility of the long-term monitoring. The peer review for the GBP monitoring efforts was coordinated by SFEI and conducted by three experts in the field who were not associated with the Project. These reviewers, with expertise in environmental health science, fish and wildlife ecology, and toxicology, were Dr. Tom Grieb (TetraTech), Dr. Harry Ohlendorf (CH2M HILL), and Dr. David Janz (University of Saskatchewan).

Peer review and publication are critical elements in determining the contribution and credibility of scientific findings and monitoring results. The three external experts were asked to review the monitoring program and to evaluate how well the GBP is meeting the environmental commitments outlined in the WDRs. The peer reviewers were asked to identify the GBP's strengths and weaknesses, and to suggest strategies for improving the monitoring program where feasible for the remaining duration of the GBP through 2020.

To structure and guide this peer review, the following five key questions were developed:

1. Is the GBP monitoring program effectively tracking the consequences (i.e., biological effects) of salt and selenium discharges to Mud Slough (north) (i.e., Stations D, I [I2], and E) and the lower San Joaquin River (Stations H [R] and N)? Is it effectively tracking potential biological effects downstream of the Merced River confluence (i.e., Station N)?
2. Is the scope and frequency of monitoring of wetland channels and critical habitat (designated habitat for listed species but critical habitat has not been designated for giant garter snake or San Joaquin kit fox) effective in tracking regulatory commitments?
3. Is the Project design based on the current understanding of the environmental science of selenium?
4. What are the key data that contribute to the better scientific understanding of the behavior of selenium in the system? How can GBP datasets be modified or improved to be more helpful to other programs (e.g., San Joaquin River Restoration Program, Bay-Delta Restoration Program, and the development of site-specific or tissue-based selenium standards by USEPA)?
5. Does the Biological Effects Monitoring Program achieve the following? If not, how can it be improved?
 - a. Monitor selenium concentrations across all important media (water, particulates, tissue of different food web species) that contribute to selenium bioaccumulation in ecosystems.
 - b. Identify at-risk species and their food webs to protect communities from selenium exposure.
 - c. Determine the environmental risk occurring to fish and wildlife (including protected species such as the giant garter snake and San Joaquin kit fox) in Mud Slough (north) and lower San Joaquin River.
 - d. Are there environmental benefits of diminished selenium in other parts of the Grasslands wetland water supply channels and the San Joaquin River? Biological monitoring in potentially improved channels has occurred at Mud Slough (north), upstream of the SLD discharge (Site C), Salt Slough (Sites F and F2), and at San Joaquin River at Fremont Ford (Site G).
 - e. Is the GBP supporting healthy fish communities within the study area? Does the GBP monitor fish communities and abundance to be effectively integrated within other projects?
 - f. Does the Biological Effects monitoring accomplish an assessment of the risk to human health from consumption of fish from affected channels?

The review focused on the areas directly affected by the GBP and did not include other regional drainage management projects. It evaluated the comprehensive environmental monitoring program and how well it provides results for the monitored locations and parameters.

The Basin Plan (http://www.waterboards.ca.gov/centralvalley/water_issues/basin_plans/sacsjr.pdf), which identifies beneficial uses for the waters of the Sacramento and San Joaquin River basins, states in summary that discharges from the San Luis Drain shall not cause or contribute to pollution or nuisance and cannot be in exceedance of applicable water quality objectives resulting in degradation of beneficial uses or harm to humans, plants, animals, or aquatic life. Based on this goal, the 19 years of operation of the GBP were evaluated by the reviewers, but with a stronger focus on the recent years of the program.

SUMMARY OF REVIEWS

The reviewers agreed that the GBP is in compliance with the WDRs and that the scope and frequency of monitoring at the designated sampling sites are consistent with regulatory commitments. They recognized that the GBP has established a comprehensive monitoring program resulting in extensive datasets to assess changes in a variety of water quality parameters over time, but they focused their review on selenium, which was identified for the review as the primary constituent of concern for potential toxicological effects in biota downstream of the agricultural discharges. However, they agree that the toxicity as measured by laboratory bioassays is not likely related to selenium. The long-term dataset provides monitoring data of drainage-affected sites in comparison to reference sites, and the inclusion of historic data in the annual reports facilitates the understanding of temporal changes and their potential consequences to aquatic and terrestrial organisms. Additionally, seasonal changes are tracked through monthly and quarterly reporting. This is important because precipitation can influence loading of selenium and other constituents from agricultural discharges.

The reviewers also agreed that the sampling locations overall are appropriate to evaluate spatial trends, following a gradient approach for assessing potential biological effects, and representing the following distinct impact zones: (1) high exposure (Sites D, I2, and E), receiving direct discharge from the San Luis Drain; (2) medium exposure (Site R), ecologically relevant because of the mixing of Mud Slough and San Joaquin River water; and (3) low exposure (Site N), downstream of the Merced River confluence with the San Joaquin River, representing the effects to areas downstream of the watershed including the Delta and North San Francisco Bay. Additionally, Site I2 is also representative of an ecosystem particularly favorable for selenium bioaccumulation due to its minimal flows and lentic nature.

The reviewers included several suggestions for improvements. They recommended that for reporting of results from sampling conducted under the draft 2015 Environmental Monitoring Program more emphasis be placed on describing the rationale for the monitoring effort and using the existing dataset to assess the expected performance of the continued sampling effort. Knowledge for key parameters of the magnitude and source of variability, seasonal differences, and differences among sampling locations should provide the basis for future sampling. Incorporation of information gained from previous sampling will allow refinements to the 2015 Monitoring Program in response to the lessons learned and will provide for improved understanding of the effects of the GBP.

A comprehensive summary of the existing data that describes the variability, trends, cycles, and correlations in previous measurements would help with the overall goal of understanding whether the GBP has caused or contributed to pollution or nuisance conditions that exceed applicable water quality objectives, thereby resulting in degradation of beneficial uses or harm to humans, plants, animals, or aquatic life. That summary should be used to optimize future monitoring. It is extremely important that the probability for the monitoring program to yield statistically significant results is considered. With this in mind, the statistical power of the tests used in the data analyses should be determined. The power would predict the probability of correctly detecting existing trends or specified levels of differences among sampling locations.

Reviewers also indicated that statistical approaches quantifying the differences between exposure sites and hydrologically and ecologically similar reference sites are a key aspect for the evaluation of biological responses. These site comparisons would allow for verification of recovery from exposure. In particular, comparing Mud Slough to a reference site (e.g., Salt Slough) could provide a dose-response relationship and a useful effects assessment if, for example, fish egg selenium concentrations would be compared to larval deformities at these sites. Adding more reference sites could provide improved understanding of the natural background levels of selenium in water and biota, which would be helpful for the GBP and other, longer-term recovery programs outside the GBP.

While tissue analysis for selenium in fish, bird eggs, and invertebrates is very useful to characterize their exposure and evaluate potential effects in certain reaches within the Project area, and with that meet the requirements for the monitoring program, the biological effects monitoring could be improved. It was recommended that in addition to monitoring potential effects based on concentrations in biota, it would be beneficial to more directly evaluate biological effects as well. Additional information could be obtained by collecting eggs and sperm from spawning fish at high-exposure sites receiving selenium and similarly at reference sites. Eggs could then be fertilized and raised under controlled conditions. Selenium incorporated into yolk is used by fish larvae right after hatching, and effects are manifested as deformities or death. Dose-response relationships in the developing larvae can indicate reproductive effects through dietary exposure of the adult female fish (which transfer selenium to their eggs). With this additional test, the ecological risk assessment would be based on measurement of both exposure and actual effects. Furthermore, the conversion of whole-body selenium to egg/ovary selenium for comparison to the hazard scale developed by Lemly is outdated, and underscores the major improvement that additional fish egg collection could provide.

Aqueous exposure of invertebrates and fish to selenium in the currently conducted toxicity tests does not represent the most useful information, because dietary exposure is the most important pathways for effects. Additionally, the observed toxicity may be related to any number or combination of other chemicals in the surface water samples, which can confound the test results. An improvement would be to conduct longer-term exposure testing of caged fish and invertebrates with reallocated resources, allowing evaluation of multiple dietary pathways in addition to aqueous exposure. Additionally, the selenium concentrations in fish exhibit large variation, probably because the sites are open systems with fish movement to and away from the collection sites. Caged fish testing would provide conditions that better evaluate site-specific exposures and effects.

The fish community sampling has not shown any significant differences in community composition or occurrence of selenium-related anomalies in fish among site. This is a strong indicator of similar fish assemblages at exposure and reference sites with decreases over time in the occurrence of abnormalities in individual fish at sites receiving drainage water. If continued, the numbers of abnormalities should be standardized to the total numbers of captured fish. However, reducing the sampling sites for fish communities and dropping sites that have recovered well was recommended, and this would allow reallocation of money toward caged fish testing and fish egg collection.

Another budgetary reduction could be achieved through the focus on Mud Slough as the main sampling site and reduced sampling at other sites. Sampling a greater number of sites within Mud Slough could describe transport, mixing, and deposition characteristics of selenium. Considering the reduction of biota data collection to possibly twice a year and focusing on seasons when egg development is occurring would still meet the monitoring goals for detecting spatial and temporal trends. Additionally, targeting a smaller subset of invertebrate taxa and fish species should be considered.

Similar to fish eggs, eggs of reptiles, amphibians, and birds from reference and exposure sites are also the better approach for a thorough exposure assessment. Even though tadpoles have been collected and analyzed previously, they may represent a subset of amphibians that received a smaller selenium dose from their mothers and thus survived. Again, a much more sensitive approach would include the collection of eggs of amphibians and other organisms to assess the biological risk.

Furthermore, the determination of trophic transfer of selenium through the food web could include measurements at the very bottom of the food web, specifically in biofilm at the sediment-water interface and suspended particulates (including phytoplankton, etc.) in the water column. This would be beneficial at sites downstream of the discharge area to better understand the assimilation of selenium and the potential dietary pathway to consumers. This information would help describe food web dynamics and could contribute significantly to creation of a conceptual model to describe overall selenium bioaccumulation. The reviewers understand that the development of a conceptual model is outside the scope of this monitoring program but the collection of biofilm and suspended particulates samples at Sites N, R, and G would better inform the ecological risk assessment for the downstream system and could additionally provide valuable data for other ecosystem-scale studies. Especially within Mud Slough, the area with the greatest impact of selenium discharges, a bioaccumulation model could help make predictions about future risks to organisms and recovery of the area.

It was pointed out that waterborne selenium concentrations at Site N represent potential effects in the lower San Joaquin River, downstream of the Grassland area, well and monitoring is in accordance with requirements. Concentrations have stayed well below the 5.0 µg/L water quality objective and should not pose adverse effects to fishes. However, this objective causes some uncertainty for the ecological risk assessment since it does not consider selenium biodynamics and differences in fish feeding preferences. Selenium has complex speciation and a complex biogeochemical cycle. A better approach would be to use a tissue-based concentration from fish eggs or ovaries for Site N that can be compared to a newly developed USEPA aquatic life water quality criterion for freshwater (which is now in draft form). Alternatively, the GBP could consider lowering the water thresholds to 1.2 µg/L for lentic portions and 3.1 µg/L for lotic portions of the study area, according the USEPA draft criterion.

Even though the primary emphasis of the monitoring is to demonstrate compliance with the selenium loading criterion, the reviewers recognized that many aspects of the program are highly relevant to improving the current understanding of selenium behavior in the environment, such as bioaccumulation, loading, and concentrations in abiotic and biotic components in the system. The focus on exposure and potential effects is consistent with the environmental science of selenium. However, the recommendations for more detailed ecotoxicological studies (e.g., fish and amphibian egg collection and selenium biofilm and water-column particulates concentrations) could improve the understanding of the effect of the operation of the San Luis Drain within the region and the potential effects to downstream sections of the San Joaquin River, Delta, and North San Francisco Bay.

Consistent improvements in water quality with regard to selenium concentrations and most other measured parameters downstream of agricultural drainwater inputs were acknowledged by all three reviewers. A significant reduction in selenium loading overall has been an important benefit to the ecology of the San Joaquin River system and associated wetlands. The San Joaquin River is a complex setting in which cumulative effects from human activities and natural occurrences upstream and downstream of the study area can make cause-and-effect assessments difficult. To provide better context on a larger landscape scale, it was suggested that selenium and other contributing stressors could be evaluated through a cumulative effects assessment.

Overall, the GBP provides a unique dataset that greatly facilitates the understanding of selenium sources, transport, and bioaccumulation. The monitoring program has met the requirements of the use agreement and the WDRs. Some future modifications, mentioned above and in more detail in the individual chapters of the reviewers, could be incorporated in the draft 2015 monitoring plan to enhance the understanding of selenium behavior and the prediction of ecological risk in the San Luis Drain, the wetland channels, and San Joaquin River.

CHAPTER 1 - REVIEW BY TOM GRIEB

Introduction

This review used the Grassland Bypass Project (GBP) document entitled 2015 Environmental Monitoring Program (2015 EMP; Bureau of Reclamation, 2015) as the primary description of the GBP monitoring program. In the course of the review, the requirements of the Monitoring and Reporting Program for the U.S. Bureau of Reclamation and the San Luis & Delta-Mendota Water Authority described in the Waste Discharge Requirements (WDR; Regional Water Quality Control Board, 2015) were considered. The GBP 2010-2011 Annual Monitoring Report (Grassland Bypass Project Oversight Committee, 2013) and the Grassland Bypass Project, 2010–2019 EIS/EIR, Final August 2009 (2009 EIR/EIS; ENTRIX, 2009) were also reviewed to better understand the results of previous sampling and analyses. The focus of this review is on the water quality sampling and analyses. The habitat and toxicity elements of the biological monitoring are not addressed.

General Observations

The sampling requirements from the WDR, the sampling locations, frequency of sampling, and analytical techniques are described in adequate detail to show compliance with the WDR. However, more emphasis should be placed on describing the rationale and basis for the monitoring effort “to assess environmental conditions”. It is critical to the development of an effective monitoring effort to assess the expected performance of the sampling effort. This information should be developed by using existing data from the proposed sampling locations or data from comparable sites. Based on the statistical distribution characteristics of the parameters to be measured, the expected performance of the monitoring program should be described in terms of the expected magnitude and sources of variability associated with the measured parameters as well as the minimum differences between parameter values over time and/or between sampling locations that can be detected with the proposed level of sampling effort. The 2015 EMP descriptions for each section, e.g., Water Quality Monitoring, should look something like:

- Summary of the existing data, number of samples, summary statistics, distributional characteristics of key parameters, indication of seasonal effects, differences between sample locations. Most of this information is available elsewhere, e.g., in the 2009 EIS/EIR. A summary of the relevant information from that report would make the 2015 EMP more complete and should be used to provide the basis for the sampling design.
- Synthesis of the existing data and a statement of how the data characteristics influenced the design of the monitoring program.
- Description of the sampling design. The sampling elements, station locations, parameters, sampling frequency are described in the 2015 EMP.
- Description of the expected sampling results and how the data will be analyzed and used.

Here are some specific comments and questions about the 2015 EMP Report that was reviewed:

- **P. 12, ¶ 5:** “The GBP 2015 Environmental Monitoring Program includes water quality parameters ... plus several other sites ... to assess the restoration of the area with the removal of agricultural drainwater under the GBP.” It’s not clear if these assessments of the restoration efforts will be conducted as part of the 2015 EMP or as part of other programs. In either case, how these assessments will be conducted and what is expected to be achieved is of interest to a better understanding of the GBP’s environmental effects and should be described in detail.
- **P. 14, Section 3.2 Integration with Research/Investigations Activities.** Here is the opportunity to describe relevant research activities and how the data will be used to advance the understanding of the effect of the GBP. The collection of additional measurements without a plan for using the data in this program (or in collaboration with other ongoing efforts) could result in a missed opportunity.
- **P. 18 ¶ 2:** “Reclamation will incorporate data collected for other programs in the GBP monthly and quarterly reports.” Identify these other programs. Describe these data, and how the information will be used.
- **P. 19, San Luis Drain.** “The concentration of selenium in sediment will be measured in the ten places Why 10 locations? How will the data be used? It seems that a plan for the collection of sediment data, TSS, flow, water column Se concentrations could be used to characterize the transport and behavior of Se in the drain system.

- **P. 19, Mud Slough (north).** Has Site C been dropped because it will be monitored by other programs? How will the monitoring results in Mud Slough be used to assess the effects of the GBP without information from the upstream station? Will the data from the Westside Coalition be comparable?
- **P. 20, San Joaquin River.** “The GBP 2015 Environmental Monitoring Program will continue to evaluate the impacts of the GBP on the San Joaquin River at three places.” What has been learned from the previous sampling, and how has this information been incorporated into the 2015 EMP? How will these data be used to assess flows and contaminant concentrations in water? Is the goal to determine the effects of the GBP at specific distances? What are the mixing characteristics of the GBP discharge?
- **P. 20 Stormwater Monitoring.** The description of the stormwater monitoring is incomplete. Yet this seems like a great opportunity to describe the effects of large, episodic discharges. Are there some critical factors that should be measured? Critical timing? Critical locations?
- **P. 21 “5. Reduce the frequency of sampling nutrients”** ...Why? What’s the rationale?
- **P. 21, Section 6.0 Flow Monitoring.** “Flow in the San Luis Drain must be managed to prevent erosion of sediments”. What data support this, what are the critical flow characteristics?
- **P. 22, 6.6 Load Calculations.** How are the load estimates from these calculations influenced by the sampling frequencies for Se? Here is a specific example to show the existing data, describe what has been learned from previous sampling efforts, and to explain how the 2015 EMP has been developed in response to this information.
- It appears that the data will be collected by several entities (multiple agency responsibility), and reported or saved in different databases, e.g., the flow data stored in USGS database. All data from the GBP should be made readily available/accessible.
- **P. 24, Section 7.0 Water Quality Monitoring.** “The historic data have been used to develop regulatory programs for the control of agricultural drainage discharges.” What have these data shown, and how have these results been utilized in the design of this program? What are the critical data issues (e.g., the importance of a continuous record to characterize data distributions and the ability to define statistical ranges on parameters)?
- **P. 27, 7.3 Water Quality Parameters:**
 - » What has been the performance of the monitoring efforts to date?
 - » Nutrients: why these parameters and sampling frequencies? What have we learned and what do we expect to get out of the program moving forward?
 - » “Mercury in water will also be measured at four places that convey GBP water.” Why 4 places? What’s been observed in the past? In general, there isn’t much data available to characterize mercury or methyl mercury loads from irrigated agriculture in California. There may be an opportunity to contribute to this understanding but the level of sampling would have to be increased, other parameters such as DOC, EC, TSS, total and MeHg would have to be measured simultaneously, and the sampling strategy would have to be thought through. It’s not at all clear what the contribution of these measurements at four places will be.

Responses to Key Questions

1. **Is the GBP monitoring program effectively tracking the consequences (i.e., biological effects) of salt and selenium discharges to Mud Slough (north) (i.e., Stations D, I [I2], and E) and the lower San Joaquin River (Station H [R] and N)? Is it effectively tracking potential biological effects downstream of the Merced River confluence (i.e., Station N)?**

Considering the sampling of the water quality parameters, e.g., elevated salt, selenium and mercury, there appears to be several opportunities for examining the link with biological effects downstream. But it is not clear that the existing sampling effort will take full advantage of these opportunities. Items to consider:

- » It is not clear what sampling will be conducted at Site C (that will be monitored by the Westside Coalition for the Irrigated Lands Regulatory Program). Coordinated sampling should be conducted at locations both upstream and downstream of the discharge to better understand the magnitude and a real extent of discharge effects. (Note: After these comments were submitted, the revised 2015 Environmental Monitoring Plan includes sampling at Site C again.)

- » Station D is located about a quarter mile below the SLD discharge (2015 EMP, p. 27). This distance from the discharge is most likely not adequate to characterize local effects and the areal extent of effects of the discharge. Sampling a greater number of locations near the discharge could be conducted to describe the mixing of the discharge in Mud Slough and the attenuation of effects with distance. Linking the water column sampling to sediment sampling could also be done to characterize the mixing of the discharge and the deposition of suspended material.

2. Is the scope and frequency of monitoring of wetland channels and critical habitat (designated habitat for listed species but critical habitat has not been designated for giant garter snake or San Joaquin kit fox) effective in tracking regulatory commitments?

Not addressed by this reviewer.

3. Is the project design based on the current understanding of the environmental science of selenium?

The primary emphasis of the monitoring program is on demonstrating compliance with the selenium loading criterion. The primary ecological concern of selenium is the bioaccumulation of selenium and the effects on wildlife. A key to understanding the potential effects on wildlife is the ability to understand the form of selenium, the level of exposure to prey items, and the bioaccumulation in the food web. A large effort has been made by the USGS in the development of ecosystem-scale selenium modeling in support of fish and wildlife criteria development (Presser and Luoma. 2010 a,b). Their work has shown that one of the keys to understanding selenium bioaccumulation is to characterize the form of selenium exposure to organisms. In the 2015 EMP there is little emphasis on expanding the current understanding of the form of selenium discharged or the transformations that take place that might affect bioaccumulation potential. The measurement of dissolved selenium and selenium in suspended particulates is limited to quarterly samples. The efficacy of quarterly sampling to characterize the importance of dissolved and particulate forms of selenium on potential biological effects is not addressed in the 2015 EMP. Understanding dissolved and particulate material selenium speciation is extremely important in assessing the assimilation efficiency of Se by prey items. However, speciation is not part of the monitoring program.

4. What are the key data that contribute to the better scientific understanding of the behavior of selenium in the system? How can GBP datasets be modified or improved to be more helpful to other programs (e.g., San Joaquin River Restoration Program, Bay-Delta Restoration Program, and the development of site-specific or tissue-based selenium standards by US EPA)?

An important issue associated with the July 2015 Draft Bay Delta Conservation Plan/California WaterFix EIR/EIS is the effect of an increase in the relative proportion of San Joaquin River flow compared to Sacramento River flow on selenium concentrations in North San Francisco Bay (listed on the 2010 303(d) list for selenium impairment). The modeling analysis in the 2015 EIR/EIS shows, for example, that there is an increase in the load to North San Francisco Bay from the existing conditions by 6 – 11 %, e.g., for the different alternatives considered as part of Alternative 4. It is of interest of how the data from the 2015 EMP can be used to quantify the effect of the operation of the SLD on selenium concentrations on the San Joaquin River at Vernalis and the potential contribution of selenium loading to North San Francisco Bay.

5. Does the Biological Effects Monitoring Program achieve the following? If not, how can it be improved?

- a. Monitor selenium concentrations across all important media (water, particulates, tissue of different food web species) that contribute to selenium bioaccumulation in ecosystems. As noted above, the infrequent sampling of particulate and dissolved Se and the lack of Se speciation limit the use of the expected data in assessing the contribution to Se bioaccumulation. However, fully meeting that objective would require a greatly expanded sampling effort. It is important to meet fewer monitoring objectives successfully than to unsuccessfully address multiple objectives.
- b. Identify at-risk species and their food webs to protect communities from selenium exposure. Not addressed by this reviewer

- c. Determine the environmental risk occurring to fish and wildlife (including protected species such as the giant garter snake and San Joaquin kit fox) in Mud Slough (north) and lower San Joaquin River. Not addressed by this reviewer
- d. Are there environmental benefits of diminished selenium in other parts of the Grasslands wetland water supply channels and the San Joaquin River? Biological monitoring in potentially improved channels has occurred at Mud Slough (north), upstream of the SLD discharge (Site C), Salt Slough (Site F and F2), and at San Joaquin River at Fremont Ford (Site G). Not addressed by this reviewer
- e. Is the GBP supporting healthy fish communities within the study area? Does the GBP monitor fish communities and abundance to be effectively integrated within other projects? Not addressed by this reviewer
- f. Does the Biological Effects monitoring accomplish an assessment of the risk to human health from consumption of fish from affected channels? Not addressed by this reviewer

Conclusions and Recommendations

The 2015 Environmental Monitoring Program demonstrates compliance with sampling requirements from the WDR. The sampling locations, frequency of sampling, and analytical techniques are described in adequate detail. However, more detailed information on the nature of existing conditions and the projected effectiveness of the 2015 EMP is needed if a goal is to show how the program will contribute to the understanding of the environmental conditions in the San Luis Drain and the receiving water (Mud Slough). The demonstration of how the program will contribute to the understanding of the environmental system can be developed by describing existing conditions and presenting how the results of the monitoring program will enhance the existing knowledge.

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CHAPTER 2 - REVIEW BY DAVID JANZ

Introduction

The Grasslands Bypass Project (GBP) began in 1996 with the goal of diverting unusable sub-surface agricultural drain water from the approximately 97,000 acre Grasslands Drainage Area (GDA) to avoid potential ecological impacts to biota inhabiting surrounding grasslands habitats. These habitats include (1) wetlands utilized by waterbirds and certain other wildlife (mammals, reptiles and amphibians) that are susceptible because they prey upon aquatic organisms, and (2) the San Joaquin River and its tributaries utilized by fishes for feeding, migration, and reproduction. A major impetus for development of this project was observations in the early 1980s of early life stage developmental toxicities (deformities) and mortality of aquatic birds feeding in the Kesterson Reservoir, part of the Kesterson National Wildlife Refuge, which received agricultural drain water from areas similar to the GDA. Although the drain water contained elevated levels of several trace elements, including selenium, boron, molybdenum, arsenic and chromium, and associated ions, selenium was identified as the causal agent responsible for these effects in birds. At that time, scientific investigations into the aquatic ecotoxicology of selenium were first being reported, and the ecotoxicological work at Kesterson Reservoir represents one of the classic case studies on this topic (Skorupa 1998; Ohlendorf 2002; Young et al. 2010).

Other than mercury, selenium contamination of aquatic ecosystems represents the greatest ecotoxicological hazard of all trace elements to biota that feed in such systems (Luoma and Presser 2009). This is especially true for oviparous (yolk-bearing) vertebrate animals: fishes, birds, amphibians and reptiles (Janz et al. 2010). During oogenesis in oviparous vertebrates, synthesis and transport of yolk to eggs (i.e., vitellogenesis) by reproductively mature females results in excess selenium obtained by females via their diet to be maternally transferred to eggs in an unregulated, dose-dependent manner. When yolk and albumin are utilized by embryos and/or hatched offspring as an energy source during these sensitive early life stages, excess selenium is biotransformed to reactive metabolites that are believed to cause toxicity (Spallholz 1994; Janz et al. 2010). These early life stage toxicities are characteristic and diagnostic of elevated selenium exposure, and include skeletal, craniofacial, limb and eye deformities, edema, and mortality. Such toxic effects have been shown to have significant negative impacts on fish populations by reducing recruitment of new individuals into populations, and provide one of the clearest cause-effect relationships in ecotoxicology between exposure to a toxic substance and impaired population dynamics (Lemly 1993; Skorupa 1998; Janz et al. 2010). Although selenium is an essential trace micronutrient, there is a very narrow range between essentiality and toxicity, particularly in oviparous vertebrates, which further exemplifies the hazard posed by this trace element to susceptible aquatic-dependent vertebrates.

From a regulatory perspective, there has been much debate in recent years concerning revisions of water quality criteria (WQC) for selenium aimed at protecting aquatic biota. The present United States Environmental Protection Agency chronic WQC of 5 $\mu\text{g Se/L}$ causes great uncertainty in the ecological risk assessment of selenium. This is because differences in feeding preferences, geochemistry, hydrology and ecology among aquatic ecosystems dictate the speciation, bioavailability and toxicity of Se to aquatic vertebrates (Luoma and Presser 2009; Stewart et al. 2010; Janz 2012). In other words, an ecosystem with 1 $\mu\text{g Se/L}$ in the water column can potentially pose a greater risk to aquatic-dependent species than an ecosystem with 10 $\mu\text{g Se/L}$. There is now consensus among scientists and regulators that (1) dietary exposure is the dominant route of uptake in aquatic vertebrates, (2) fishes are the most sensitive vertebrate class to selenium toxicity, (3) measurement of selenium in eggs of oviparous vertebrates, particularly fishes, provides the most accurate assessment of ecological risk, and (4) there often needs to be some form of site-specific assessment of selenium biodynamics in selenium-contaminated aquatic ecosystems. Thus, the current draft USEPA aquatic life ambient water quality criterion for freshwater is tissue-based, with selenium concentration in fish eggs or ovary proposed as the most accurate means to assess ecological risk to all aquatic biota.

Selenium is an insidious aquatic contaminant due to its persistence in abiotic and biotic compartments of freshwater ecosystems; indeed it behaves more like notorious persistent organic pollutants such as organochlorine compounds. Selenium has one of the most complex patterns of speciation among trace elements, and biogeochemical cycling of these different chemical forms in aquatic systems is equally complex (Maher et al. 2010; Janz et al. 2014). Combined with the fact it is an essential trace element actively taken up by organisms, it is unknown how long selenium will persist in the food web of a given contaminated ecosystem even after loading has ceased. Thus, monitoring programs that assess the recovery of aquatic systems historically receiving large selenium loading must be conducted over the long term (years to decades).

Due to the above rationale, and given my expertise in the aquatic ecotoxicology of selenium, the focus of my comments will be on selenium. When appropriate, I will offer comments on other constituents (e.g., other trace elements, ions, pesticides) but these comments will be brief and hopefully other invited contributors will expand on these aspects of the GBP monitoring program. It is my belief that selenium loading from the GDA represents the primary ecotoxicological concern in the San Joaquin River and associated wetlands, and if selenium is maintained at levels in biota that do not pose ecotoxicological risks, then other constituents will likely follow suit and also not pose such risks.

General Observations

The GBP has recently completed its 19th year of operation, and to date there have been consistent improvements in water quality associated with loading of selenium and most other measured constituents at monitoring stations downstream of agricultural drain water inputs. This is particularly true for the past 3-4 years, although this may be confounded by the relative lack of annual precipitation over this period. By 2014, selenium loading from the GDA has been reduced to less than 5% of the pre-project (1986-1995) average. Overall, based on data available from the comprehensive GBP monitoring program it appears that achieving the water quality objective (WQO) of 5 µg Se/L by 2019 is achievable in all downstream environments with the possible exception of within Mud Slough. However, regulations change as science progresses, and therefore the GBP should anticipate a lowering of this WQO prior to the end of this project; refer to the draft USEPA criterion based on aqueous selenium concentration (1.2 µg Se/L for lentic systems and 3.1 µg Se/L for lotic systems, monthly average). Of note, and I imagine some concern, is the fairly consistent increase in invertebrate and fish selenium concentrations that occurred in 2014 in Mud Slough (sites D and I2).

Responses to Key Questions

1. Is the GBP monitoring program effectively tracking the consequences (i.e., biological effects) of salt and selenium discharges to Mud Slough (north) (i.e., Stations D, I [I2], and E) and the lower San Joaquin River (Station H [R] and N)? Is it effectively tracking potential biological effects downstream of the Merced River confluence (i.e., Station N)?

General comments

The GBP has established a comprehensive monitoring program to assess temporal changes in a variety of water quality parameters and potential toxicological effects in biota at several locations downgradient of agricultural drain water inputs. This long-term dataset provides useful information to assess changes in water quality and biological effects over almost two decades on an annual basis, but also importantly changes that occur within years on a seasonal basis. This is important because of distinct seasonal differences in precipitation in this area, which can influence loading of selenium, salts, and other constituents of agricultural drain water. In general, the GBP monitoring program is effectively tracking water quality parameters in both time and space, but is lacking somewhat on biological effects monitoring, as will be discussed below. In addition, certain aspects of the monitoring program are not essential, and these resources could potentially be directed more towards focused studies on biological effects associated with selenium exposure in vertebrate animals.

Selection of monitoring sites

The monitoring sites listed above are appropriate locations to collect samples, as they fall into three distinct impact zones: (1) within Mud Slough (sites D, I2 and E), which currently receives the majority of constituents from the GDA and can be considered a high exposure area, particularly the furthest downstream site (E), (2) site R, perhaps the most ecologically relevant monitoring station since it is upstream of the Merced River confluence on the San Joaquin River and represents the full influence of the GBP loading after mixing with the San Joaquin River (medium exposure area), and (3) Site N, located on the San Joaquin River downstream of the Merced River confluence (low exposure area). These sites follow a classic gradient approach for assessing potential biological effects of aquatic pollution (Fox 1991). As discussed below, Mud Slough represents a key monitoring area for future investigations since it appears to be the zone of most influence as the GBP has evolved to minimize GDA inputs arising from wetland water supply channels to areas such as Salt Slough. Site I2, a periodically flooded backwater of Mud Slough, is an important monitoring site since it provides an ideal environment for selenium assimilation into the food web due to its lentic nature and potentially important habitat for fish reproduction.

Site G, located on the San Joaquin River upstream of the Mud Slough confluence at Fremont Ford, now represents an appropriate reference site for sessile organisms (e.g., invertebrates and perhaps fishes with very small home ranges), but not fishes that migrate to any extent since it will be unknown whether they have spent time in downgradient exposure areas, including Mud Slough. It is surprising that the 2015 monitoring program was modified to exclude site G. Without an appropriate reference site to compare data collected from exposure sites, it is unclear how recovery of the exposure sites can be verified statistically. It is recommended that site G be retained as a reference site, and it is also suggested that an appropriate reference site located further (>10 miles) upstream of site G be incorporated into the monitoring program to avoid potential issues with migrating organisms, particularly small-bodied (“bait”) fishes. If such a site is chosen, it will be important to select a location that is hydrologically and ecologically similar to sites G, R and N. This raises a key point about the importance of appropriate reference sites in monitoring programs. The GBP monitoring program has many exposure sites, but relatively few reference sites, in fact none that I am aware of located upstream of all GDA inputs or in an ecologically and hydrologically similar area outside the immediate vicinity. It is recommended that the GBP include an additional reference site(s) outside the immediate zone of GDA influence to provide further comparison of the many parameters being determined. This may be especially important over the next 5 years as the GBP comes to an end. It is also important because it is not apparent that the current monitoring program addresses the issue of natural background levels of selenium in water or biota in this region. In my opinion this approach would also be useful for longer term recovery initiatives such as the San Joaquin River Water Quality Improvement Project (SJRIP). (Note: After these comments were submitted, the revised 2015 Environmental Monitoring Plan includes sampling at Site G again.)

Based on the rationale provided in the preceding paragraph, my discussion will focus on monitoring of the “core area”: Mud Slough and the sites immediately above (G) and below (H, R) its confluence with the San Joaquin River. I have reviewed the long term data available from monitoring Salt Slough, and it is evident that diversions of agricultural drain water from this waterbody have successfully reduced selenium loading to the San Joaquin River, leaving Mud Slough as the area of most concern with respect to the 2019 end-date of the GBP. There are also data I have reviewed from other sampling sites, some of which have included bird eggs and tadpoles, but for the sake of brevity I will focus on the aforementioned monitoring sites. Many of the comments I will provide are applicable to other monitoring sites beyond the “core area” mentioned above.

Cumulative effects assessment

It is important to note here that the San Joaquin River represents a “confounded” system where cumulative effects are present from human activities, and natural phenomena, occurring upstream throughout the watershed. It may be worth thinking to some extent about the GBP in terms of larger scale cumulative effects assessment (CEA) in order to provide more context into the relative contributions of other stressors to the San Joaquin River, both upstream and downstream of the study area. Cumulative effects assessment uses an effects-driven approach to assess environmental impacts on multiple landscape scales. Several recent publications are available to provide guidance on CEA, including an entire issue of the journal *Integrated Environmental Assessment and Management* devoted to this issue (volume 9(3), 2013). Although it is realized that the GBP monitoring program is focused primarily, and correctly, on potential biological effects of the GDA on immediate, smaller scale receiving environments, it may be worthwhile to include discussion of larger scale aspects in future reports. This may include referring to the recently completed Total Maximum Daily Load for Selenium in the North San Francisco Bay report. This will provide a context for the role of the GDA within the entire San Joaquin River watershed.

Specific comments

THE IMPORTANCE OF EGG SELENIUM DETERMINATIONS

Since the GBP monitoring program has evolved to some extent since its inception, I will be referring mainly to the most recent documents available, provided to me as the draft “2012-13-14 report” (zipped file), the document dated August 14, 2015 titled “GBP 2015 Environmental Monitoring Program”, and to some extent the most recently available GBP Annual Report (2010-2011). From 2012 to 2014 (and presumably 2015, but data may not yet be available), the California Department of Fish and Wildlife (CDFW) has conducted sampling of fishes, invertebrates, bulrush (*Typha*) seed heads, and “plankton” from sites E, G, and H on a quarterly basis. Site R replaced site H in July 2013 to avoid possible dilution issues associated with the Merced River; I completely agree with this change to the monitoring program as mentioned above. Samples were analyzed for total selenium (using hydride generation AAS) and boron (using ICP-MS). It was unclear from the information provided whether “plankton” included zooplankton and/or phytoplankton, and I did not see any selenium data in the most recent draft report (I am assuming based on the description of methods by CDFW, that this is zooplankton). It is suggested that this be specified. The CDFW monitoring has also included a fish community assessment at sites G, E, and H (replaced by R in 2013) since before implementation of the GBP monitoring program (1993). Overall, this is a robust monitoring program aimed at assessing exposure to selenium and boron, but has limited value in assessing the effects of exposure to drain water from the GDA. Potential effects assessment ideas will be brought forth later in this section.

As mentioned previously, it is now agreed by scientists and regulators that measurement of selenium in eggs or ovary of fishes provides the most important (accurate) data to inform ecological risk assessment. Although the GBP includes collection of bird eggs in the Grasslands area for selenium analysis and observations of mortality of embryo malformations, a major gap is the lack of fish egg collections. Thus, one of my major recommendations is that the GBP monitoring program include collection of resident fishes during the prespawning or spawning period in order to obtain eggs for selenium analysis. Importantly, for many fishes this can be achieved nonlethally, allowing release of fish after sampling. In small-bodied fishes, it will be necessary to obtain composite samples from 5-10 fish to obtain sufficient egg mass for selenium analysis, but for larger-bodied fishes there will likely be sufficient mass from a single individual. (It is worth noting here that switching the selenium analyses from HG-AAS to ICP-MS will likely reduce the mass of sample required to reliably quantify total selenium, although it is realized that there is rationale for using the same laboratory for the remainder of the GBP for consistency). In many cases eggs can be expressed manually from fishes by gentle pressure on the abdomen. If this does not work, mature female (and male) fish can be induced to ovulate by injection of gonadotropin-releasing hormone (GnRH) analogues such as Ovaprim®, left in net pens overnight, and then sampled the following day. Egg collection can also be conducted lethally by dissecting out eggs/ovary after euthanasia. The life history of each fish species will need to be consulted to know when spawning occurs. It is important to note that there are two main patterns of oogenesis in fishes: (1) synchronous spawners, where all eggs are at the same developmental stage during maturation and spawning occurs in a single (or in some species several closely-timed) event (e.g., salmonids, suckers), and (2) asynchronous spawners, where eggs are at different stages of oogenesis and repeat spawning occurs over an extended period of weeks to months (e.g., cyprinids such as minnows). Asynchronously spawning fish species provide better logistics since the timing of fish sampling, and thus availability of mature eggs, occurs over a greater window of time. The current monitoring timelines for quarterly biological sampling may need to be more flexible to match the timing of fish spawning.

Chinook salmon and steelhead trout are iconic fish species and important ecological and cultural components of the San Joaquin River, especially given the recent re-introduction of chinook into the river and long-term goals of enhancing both species' presence. Both of these salmonid species are synchronous spawners and eggs are easily collected nonlethally during the spawning period. If any opportunities arise to collaborate with fisheries biologists or hatchery staff in order to obtain eggs from either species, it is suggested that these samples be collected for selenium analysis.

Similar to fishes, collection of amphibian, reptile or bird eggs from exposure and reference areas are the ultimate samples to obtain for selenium exposure assessment. Frog egg masses are collected during the breeding period, and only a fraction of the egg mass is usually required for selenium analysis. Although tadpoles have been collected sporadically during the GBP monitoring program for selenium analysis, frog eggs are a better option. This is because there will be significant “growth dilution” of selenium in tadpoles compared to eggs, and also potentially that tadpoles may represent the survivors that received lower selenium doses from their mothers compared to embryos with greater selenium doses that perished prior to the tadpole stage. Waterbirds usually build nests on the ground near waterbodies, particularly on islands or other refuges, and for many bird species if a single egg is removed from the nest the female will replace it. This is definitely true for ospreys as well. It appears the GBP monitoring will include bird egg collections from the Grasslands area. Reptile eggs, notably those laid by the giant garter snake, will be hard to come by and given this species’ conservation status it is not likely permits will be issued for collection. Since Mud Slough appears to be the area of most concern with respect to the long term goals of the GBP, it is recommended that a more intensive monitoring program be conducted at this high exposure area for the duration of the project, focusing on collection of eggs from fishes, amphibians, birds, and potentially reptiles.

In the absence of egg selenium data, whole-body or muscle selenium concentrations provide the next best information for risk assessment. The current GBP monitoring program is effectively tracking such tissue selenium concentrations in a variety of fish species, and occasionally amphibians (tadpoles) at sites outside the core area. The reason for uncertainty in using whole-body or muscle selenium for risk assessment is that there is significant variability among species in relationships to egg/ovary selenium concentrations. However, in the absence of egg/ovary selenium data then the current monitoring of whole-body selenium in smaller fishes and muscle selenium in larger fishes is an effective approach. It should be noted that muscle samples can be collected nonlethally from large-bodied fishes through the use of muscle plugs, which provide sufficient mass for selenium analysis.

CAGED FISH AS BIOMONITORS

Another monitoring approach that might be considered is the use of caged small-bodied fish for assessing exposure to selenium and other constituents of the GDA discharges. This approach has been used successfully for exposure assessment in many scenarios where more controlled conditions are needed, especially with respect to fish mobility (Palace et al. 2005; Allert et al. 2006; Oikari et al. 2006; Phibbs et al. 2011a,b). Fish migration in and out of exposure areas, such as Mud Slough, may be responsible for the large variation observed in tissue selenium concentrations of fishes collected at this and other GBP monitoring sites. In caging studies, relatively small sinking cages (0.5 m³) are deployed with 10-20 fishes (commonly minnows) in each cage. Fish are able to feed on available benthic and pelagic invertebrates, and graze on periphyton/biofilm, depending on their feeding niche. For selenium bioaccumulation, it has been established that 30-60 days are required for fishes to reach steady state equilibrium with respect to selenium bioaccumulation (Allert et al. 2006; Phibbs et al 2011a). Detailed guidance for conducting such caging studies is available (Palace et al. 2005). For the GBP monitoring program, fathead minnow would be an ideal fish species to use, since it is readily available commercially, is resident in the area (although introduced), has a short life cycle, is omnivorous, and is an asynchronous spawner. It is highly recommended to use fathead minnow caged for 60 days at sites G (reference), N, R and E (or D or I2) to assess bioaccumulation of selenium in eggs. It may also be a consideration to cage minnows at site F or F2 in Salt Slough, since these sites have recovered significantly in the past decade with respect to selenium inputs, and they are likely more ecologically and hydrologically similar to Mud Slough, thus a valid reference site. It should also be noted that simple approaches are also available to use caged benthic invertebrates at study sites (Franz et al. 2013). As discussed below, these approaches are far superior to standardized laboratory toxicity tests for selenium and other constituents of the GDA discharge into Mud Slough.

THE IMPORTANCE OF PRIMARY PRODUCERS

Another major recommendation is to include a more comprehensive determination of selenium concentration at the base of the food web at sites G, N, R and E. At these sites, assimilation of dissolved inorganic selenium by the biofilm/periphyton (hereafter referred to as biofilm) layer at the sediment-water interface will be a critical process dictating further trophic transfer through the food web, although algae in the water column may also be important (Fan et al. 2002; Stewart et al. 2010; Janz et al. 2014). It appears that there will be monitoring of dissolved and “particulate” selenium, but I could find no details on this approach. It is assumed that water samples will be filtered to distinguish between dissolved selenium and selenium bound to particulate matter. However I could find no data on dissolved vs. particulate selenium; perhaps this is a new aspect of the GBP monitoring program initiated in 2015. Regardless, selenium incorporated into biofilm will represent a key step in the assimilation of selenium into the food web and should be considered as an addition to the monitoring program.

Biofilm is a complex matrix composed of algae, bacteria and fungi as the primary biotic components. Many organisms within biofilm are able to actively take up inorganic selenium and convert it to more toxic organoselenium species that are then trophically transferred through dietary pathways to consumers (i.e., benthic invertebrates, fishes, birds, and other vertebrates). Although trophic transfer of organoselenium via diet is relatively consistent among food webs, assimilation or “enrichment” by biofilm is highly variable due to a number of abiotic and biotic factors. The GBP monitoring program already has important selenium data from consumers in food webs. Including selenium analyses of biofilm at different sites will allow creation of conceptual models at each site that better describe the overall selenium bioaccumulation. This is the only major data gap I see in the GBP monitoring program, other than the lack of fish egg selenium data. Thus, it is highly recommended that the GBP monitoring program include selenium determinations in biofilm. Biofilm is easily collected by scraping rocks along shorelines and/or deploying tiles into waterbodies to allow colonization (the latter technique is likely preferable). Although the GBP includes sediment monitoring for selenium concentration, it is the top 5-10 mm layer of sediment (the biofilm layer) that is the critical component, since this is where all the “action” occurs with respect to selenium assimilation and incorporation into food webs. As stated above, this site-specific information on food web dynamics will better inform the ecological risk assessment of selenium in these systems. A further step would be to develop a biodynamic model of selenium behavior at tMud Slough (Presser and Luoma 2010), which is discussed later in this chapter.

FISH COMMUNITY SAMPLING

Unlike human health risk assessment, which is focused on the toxicological risk to individual humans, the goal of ecological risk assessment is to protect populations and communities of non-human organisms. In other words, the ultimate question is whether anthropogenic activities threaten the sustainability of individual species at impacted sites. Fish (and invertebrate) community assessments are thus important approaches to address this question. Monitoring of fish communities has been conducted since 1993 at several sites, with no clear differences over time or among sites; this may be due to lack of statistical power to detect biologically significant differences. In addition, fish mobility likely plays a role since these monitoring sites represent an open system where fish are free to move among sites. Another problem with traditional fish community assessments is that they often involve lethal sampling of fishes, usually by gillnetting. I could not find details about the methodology, but if this is the case it raises the question of whether the fish community monitoring causes a greater impact on community structure than the constituents within drainage water that are of concern. Alternatively, if beach seining or other nonlethal techniques were used to sample fish communities then this should have a relatively minor impact. Overall, this long-term fish community assessment is a strong aspect of the monitoring program, indicating that somewhat similar fish assemblages are present at sites representing a gradient of selenium exposure. It also appears that the CDFW has examined each fish captured during the community assessments and provided data on observed anomalies, which appear to be decreasing over the past 6-8 years (GBP 2010-2011 Annual Report, chapter 7, Figs 14-16). I was not able to evaluate these Figures since the y-axis is not labeled, and it appears that these are the actual number of observations without standardizing to the total number of fish captured (i.e., frequency or percentage).

LABORATORY TOXICITY TESTING

It is well established that aqueous exposure of invertebrates and fishes to selenium is not a relevant exposure pathway in aquatic ecosystems, since toxicity occurs at dissolved selenium levels orders of magnitude greater than during dietary exposure. Thus, the quarterly chronic three-species toxicity testing currently in place is irrelevant to selenium exposures that would occur at any of the sites. However, other constituents entering exposure sites from the GDA (e.g., boron) may cause toxicity in laboratory tests, so this may be the rationale for continuing these tests. It is suggested that the current laboratory toxicity testing be re-evaluated, since these resources could be allocated to other, more relevant activities. An example would be the use of caged fish and invertebrates mentioned previously, since this would allow for exposures from multiple dietary pathways, in addition to aqueous exposure to other constituents of the GDA discharge.

EFFECTS ASSESSMENT

As mentioned previously, the GBP has established a comprehensive long-term monitoring program that effectively assesses exposure of biota to selenium and boron. This is summarized for selenium nicely in Table 4a of the GBP 2010-2011 annual report, which shows changes in the selenium hazard scale developed by Dennis Lemly. This Table shows clearly that the aquatic hazard to fishes has decreased dramatically over the first 15 years of the GBP. However there is significant uncertainty in Table 4a because a generic formula from 20 years ago was used for converting whole-body selenium to egg/ovary selenium. This further illustrates the need to collect fish eggs during future monitoring activities.

Ecological risk assessment is based on both exposure and effects assessment. Unfortunately the GBP monitoring program has collected relatively little effects-based data. In my review of the significant amount of information available, I did not see even basic morphometric data (body length, body weight, Fulton's condition factor) for collected fish. Perhaps these data are available but I could not find them. As stated previously, the observed anomalies reported in chapter 7 of the 2010-2011 annual report are meaningless without some standardization to the total number of fish captures.

The ideal approach to conduct an effects assessment for selenium is to collect gametes (eggs and sperm) from spawning fish inhabiting exposure and reference areas, fertilize the eggs in the laboratory or in the field, and raise the embryos to the swim-up (fry) stage under controlled conditions in a laboratory. Selenium incorporated into yolk proteins during vitellogenesis is utilized by larval fish following hatch and just prior to swim-up, and this is when the fish receive their ultimate dose of maternally-transferred selenium, and when characteristic deformities manifest. The frequency of total deformities in fry is plotted as a function of egg selenium concentration (determined in a different subset of eggs from the same adult female) across all sites to describe a dose-response relationship. This is the approach used for several of the data points in the species sensitivity distribution (SSD) used by the USEPA in their derivation of the draft water quality criterion for selenium in freshwater. It is realized that this field-based effects assessment may be beyond the scope of the GBP monitoring program, but nonetheless it is worth mentioning here since it provides both exposure and effects assessment from fish exposed chronically to dietary selenium in situ.

2. Is the scope and frequency of monitoring of wetland channels and critical habitat (designated habitat for listed species but critical habitat has not been designated for giant garter snake or San Joaquin kit fox) effective in tracking regulatory commitments?

I am not familiar with regulations in California associated with definitions of critical habitat, but can offer the following comments. The ongoing fish community assessments provide detailed lists of species captured, their abundances at monitoring sites, and their current conservation status. As mentioned previously, the GBP monitoring program is comprehensive in terms of the number of exposure sites, frequency of sampling, and the variety of parameters being assessed. At this point in time, it appears that Mud Slough is the area of greatest concern with respect to selenium and other constituents discharged from the GDA. The aquatic habitats and surrounding terrestrial habitats are thus of greatest concern with respect to providing a healthy environment for fish and wildlife to meet their ecological requirements. If resources need to be reallocated, I suggest that a more focused monitoring program be conducted in and around Mud Slough for the duration of the GBP, with perhaps less resources allocated towards other exposure sites that have recovered substantially in the past decade. It may also be worth developing a site-specific model for selenium bioaccumulation in Mud Slough that can provide future predictions of an aqueous selenium concentration that will protect aquatic life.

With respect to the giant garter snake, previous attempts at sampling this rare species have been fruitless. In my opinion Mud Slough and surrounding terrestrial habitat represent the greatest, and perhaps only, toxicological risk associated with GDA discharges to this reptile in the study area, and should be the focus of remediation efforts to provide critical habitat. However there are many other factors other than selenium, boron and salts that are likely more important, such as adequate prey, breeding sites, cover, etc., that are not necessarily impacted by toxic agents.

As far as the kit fox, mammals are far less sensitive to selenium exposure than oviparous vertebrates. Although this species may forage on aquatic species to some extent, it likely includes terrestrial organisms in its diet as well. In my opinion, selenium, boron and other constituents discharged from the GDA are the least of this species' problems with respect to conservation. Adequate terrestrial habitat in this highly human-influenced landscape is likely the key factor limiting its recovery.

3. Is the project design based on the current understanding of the environmental science of selenium?

Overall, the GBP monitoring program includes many aspects relevant to our current understanding of selenium bioaccumulation, such as estimates of selenium loading, and selenium concentrations in abiotic (water, sediments) and biotic (invertebrates and vertebrates) components of the various monitoring sites. The notable exceptions are monitoring selenium concentrations in biofilm and eggs as mentioned previously. With all of these data, the GBP has an opportunity to establish a site-specific selenium model for Mud Slough that will allow prediction of a water column selenium level that is protective of aquatic life in this waterbody, which would thus be protective of aquatic life at downstream sites in the San Joaquin River. Excellent guidance is available for taking this next step in monitoring the future recovery of this system (Presser and Luoma 2010). It is also recommended that the GBP personnel read the draft USEPA selenium criterion document, as this contains current state-of-the-science information regarding the aquatic ecotoxicology of selenium.

4. What are the key data that contribute to the better scientific understanding of the behavior of selenium in the system? How can GBP datasets be modified or improved to be more helpful to other programs (e.g., San Joaquin River Restoration Program, Bay-Delta Restoration Program, and the development of site-specific or tissue-based selenium standards by US EPA)?

Most of the answers to this question have been addressed in my comments previously. In summary, it is recommended that an ecosystem-scale, site-specific selenium bioaccumulation model be developed for Mud Slough, which will include determinations of selenium in biofilm and fish eggs in addition to the already established monitoring activities. If and when this model predicts no ecological risk to resident biota in Mud Slough, then it can be inferred that there would be no risk to biota inhabiting downstream sites on the San Joaquin River.

5. Does the Biological Effects Monitoring Program achieve the following? If not, how can it be improved?

a. Monitor selenium concentrations across all important media (water, particulates, tissue of different food web species) that contribute to selenium bioaccumulation in ecosystems.

Again, fish and amphibian egg selenium concentrations and biofilm selenium concentrations are notable gaps in the monitoring program that should be included.

b. Identify at-risk species and their food webs to protect communities from selenium exposure.

Several of the native fish species collected during community assessments appear to be listed (although I am not familiar with the designations). It is recommended that certain of these fish species be collected during spawning in order to collect eggs for selenium analysis. If the egg selenium concentration for a certain species is lower than the draft USEPA criterion of 15.8 µg Se/g egg dry weight, then this provides the most accurate prediction of no ecological risk to this species, and by extension to the fish community.

c. Determine the environmental risk occurring to fish and wildlife (including protected species such as the giant garter snake and San Joaquin kit fox) in Mud Slough (north) and lower San Joaquin River.

The answer to this question has been addressed previously. More focused research on Mud Slough will be critical in assessing ecological risk to protected species. When this waterway recovers to the extent that it does not pose ecological risk, then the problem is solved as long as current engineering practices to manage loading of selenium and other constituents of the GDA continue to be in place.

- d. **Are there environmental benefits of diminished selenium in other parts of the Grasslands wetland water supply channels and the San Joaquin River? Biological monitoring in potentially improved channels has occurred at Mud Slough (north), upstream of the SLD discharge (Site C), Salt Slough (Site F and F2), and at San Joaquin River at Fremont Ford (Site G).**

The answer to this question is related to larger scale benefits to the entire San Joaquin watershed. Reduced loading of selenium from the GDA has environmental benefits to downstream areas of the watershed, including San Francisco Bay. See my comments above related to cumulative effects assessment for further information. Since inception of the GBP, there have been significant reductions in selenium loading overall, which has been an important benefit to the ecology of the San Joaquin River and associated wetland ecosystems.

- e. **Is the GBP supporting healthy fish communities within the study area? Does the GBP monitor fish communities and abundance to be effectively integrated within other projects?**

As mentioned previously, the fish community assessments conducted by the CDFW are an important aspect of the monitoring program. Although potential differences among exposure and reference sites are difficult to assess statistically, it is apparent that diverse fish communities are present at even the high exposure area (Mud Slough). However it cannot be determined whether fish migration among study sites is occurring. It is possible that if fish populations decline in Mud Slough due to toxicity, then this area provides an attractive “sink” for fish migration due to lower fish densities and thus less competition for food and other resources, and reduced predation risk. Expanded population ecology approaches, such as mark-recapture studies using passive integrated transponder (PIT) tags would be required to get a better assessment of the “health” of fish communities (such as abundances, diversity, and presence of sensitive species and species-at-risk). Another approach that might be worth considering is the Index of Biotic Integrity (IBI; Karr 1981; Karr 1997), which allows for quantitative assessment of fish community ecology in combination with habitat assessment.

- f. **Does the Biological Effects monitoring accomplish an assessment of the risk to human health from consumption of fish from affected channels?**

Yes, there is a component of the GBP monitoring program that includes determination of selenium in skinless fish muscle (filets) of fish consumed by sport fishers at sites including Mud Slough. These species include catfish, sunfish and carp. It is unlikely that concentrations of selenium (or other contaminants, but I did not see any data) would pose any threat to human health, even at Mud Slough. Humans, as with other mammals, are relatively tolerant to elevated selenium exposure in the range that would occur if consuming these fish.

Conclusions and Recommendations

In conclusion, the GBP has established an effective, comprehensive long-term monitoring program to assess exposure of biota to selenium and other constituents discharged from the GDA. The monitoring program has shown that significant reductions in selenium loading and exposure have occurred over the first 19 years of the project, particularly in the past decade. Importantly, discharges of selenium and other constituents have been greatly reduced to surrounding wetlands and the San Joaquin River inhabited by fish and wildlife, and potential ecological risks to animals in these areas are now likely minimal. However, Mud Slough remains the area of greatest impact and it is uncertain whether in the final five years of the GBP that this waterbody can recover to the extent that other areas have with respect to selenium ecorisk. My major recommendations, as discussed previously, are summarized below.

1. Scientific attention and financial resources should be re-allocated to focus primarily on biological monitoring of Mud Slough for the remainder of the GBP. Other sites monitored by the GBP should continue, but perhaps at less frequent intervals.
2. Selenium quantification in biofilm, and eggs collected from fishes and amphibians, should be added to the biological monitoring at Mud Slough. These sample analyses represent major data gaps in the GBP monitoring program.

3. A site-specific selenium bioaccumulation model should be developed for Mud Slough. This will be useful for making predictions about future recovery of this waterbody and the ecological risks to organisms that utilize it for habitat.
4. Continued monitoring of Mud Slough should be anticipated beyond the 2019 end-date of the GBP. A site-specific model will aid in simplifying this potential monitoring phase, since aqueous selenium concentrations, with the possibility of occasional (e.g., annual) selenium concentrations in fish eggs, should be sufficient data to predict ecological risk.
5. The GBP monitoring program should consider the use of caged fish experiments as a relevant semi-controlled approach to assess selenium exposure and effects in Mud Slough. A major advantage of such experiments is that the confounding factor of fish migration is eliminated.
6. The GBP monitoring program should focus more attention on statistical approaches to quantify differences, or lack of differences, in biological responses among sites. The importance of appropriate reference site(s) and sufficient sample sizes are key aspects of this recommendation, as discussed previously.
7. Although fish migration cannot be accounted for, the fish community assessment is a strong aspect of the GBP monitoring program as it addresses the ultimate question in ecological risk assessment. Similar to (6) above, attempts at better quantifying community dynamics among sites should be a priority in order to statistically compare sites.
8. The current laboratory toxicity testing is irrelevant to selenium toxicity. Unless the rationale for continuing this testing is related to other constituents of the GDA discharges (e.g., boron, salts, other trace elements), then these financial resources should be allocated to other initiatives as outlined above.
9. If possible, the GBP monitoring program should consider a classical selenium effects assessment comparing Mud Slough to an appropriate reference site (e.g., Salt Slough). This assessment would quantify dose-response relationships between egg selenium concentrations and the frequency of characteristic selenium-induced larval deformities in fish inhabiting each site. This assessment could potentially be integrated with 60-day caging studies using fathead minnow.

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CHAPTER 3 - REVIEW BY HARRY OHLENDORF

Introduction

The Grassland Bypass Project (GBP) has operated since 1996 pursuant to a series of agreements that have allowed the San Luis and Delta-Mendota Water Authority (Authority) to use about 28 miles of the lower San Luis Drain (Drain) to convey subsurface agricultural drainwater through adjacent federal and state wildlife management areas to Mud Slough, a tributary to the San Joaquin River. This drainage previously flowed through a variety of channels to wetland habitats in the wildlife areas and the Grassland Water District before discharging to the San Joaquin River. The history of the GBP as well as purposes and objectives of the third Use Agreement for continuation of the GBP in 2010 through 2019 are described in the Final GBP Environmental Impact Statement and Environmental Impact Report (FEIS/EIR; Entrix 2009). The current purposes and objectives are to:

- extend the San Luis Drain Use Agreement to allow the Grassland Basin Drainers time to acquire funds and develop feasible drainwater treatment technology to meet revised Basin Plan objectives and Waste Discharge Requirements (WDRs) by December 31, 2019;
- continue the separation of unusable agricultural drainage water discharged from the Grassland Drainage Area (GDA) from wetland water supply conveyance channels for the period 2010–2019; and
- facilitate drainage management that maintains the viability of agriculture in the Project Area and promotes continuous improvement in water quality in the San Joaquin River.

Section 15.2 of the FEIS/EIR (Entrix 2009) describes the monitoring and reporting program for the GBP. This program is designed to meet the objectives of the comprehensive monitoring program required in Section V of the Use Agreement (Reclamation and Authority 2009; see below).

In association with approval of the proposed continuation of the GBP, the U.S. Bureau of Reclamation (Reclamation) and the Authority requested formal consultation with the U.S. Fish and Wildlife Service (USFWS) on the potential effects of the project to listed species. The request focused primarily on potential effects to the endangered San Joaquin kit fox (*Vulpes macrotis mutica*) and the threatened giant garter snake (*Thamnophis gigas*). The USFWS provided its response in its Biological Opinion of December 2009 (USFWS 2009). Critical habitat has not been designated for either of those species, which are discussed below in response to Question 2. USFWS also provided comments relative to project effects on the threatened Delta smelt (*Hypomesus transpacificus*), which also is discussed below under Question 2.

The California Regional Water Quality Control Board, Central Valley Region (Regional Board) issued updated WDRs in 2015 for the Authority and Reclamation surface water discharges from the GBP as Order R5-2015-0094 (Regional Board 2015). Requirements in the Order implement the Water Quality Control Plan (Basin Plan) for the Sacramento River and San Joaquin River Basins (Regional Board 2011) with respect to the GBP. The Basin Plan designates beneficial uses, establishes water quality objectives, and contains implementation plans and policies for waters of the Sacramento and San Joaquin Basins. The beneficial uses of Mud Slough (north), as identified in the Basin Plan, include limited irrigation supply, stock watering, water contact recreation and noncontact water recreation, sports fishing, shellfish harvesting, warm water aquatic habitat, warm water spawning and wildlife habitat.

According to the Order (Regional Board 2015), p. 13: “The discharge from the San Luis Drain shall not cause or contribute to the following in Mud Slough (north) or the San Joaquin River. a. In surface water, an exceedance of applicable water quality objectives or a trend of degradation that may threaten applicable beneficial uses, or cause or contribute to a condition of pollution or nuisance.” . . . and “g. Toxic pollutants to be present in the water column, sediments or biota in concentrations that adversely affect beneficial uses; that produce detrimental physiological response in human, plant, animal, or aquatic life; or that bioaccumulate in aquatic resources at levels which are harmful to human health.” Attachment B of the Order describes the monitoring and reporting requirements for surface water discharges from the GBP as components of the WDRs. On p. 12 of Attachment B, with reference to the sediment monitoring plan, the text states “Within six months of this Order’s approval, the Dischargers shall submit a sediment monitoring plan for Executive Officer approval. The plan shall include the constituents to be analyzed in the annual sampling event and the schedule for sampling. At a minimum, sediment analysis shall include total selenium.” That plan was not available for review (because it was not due to the Regional Board until the end of January 2016), but the review considered other information relative to sediment monitoring that was available (e.g., sediment selenium concentrations, sediment toxicity testing). (Note: After these comments were submitted, the Plan has been submitted to the Regional Board and is available for review.)

The current (2009) Use Agreement (No. 10-WC-20-3975; Reclamation and Authority 2009) and the previous one (No. 01-WC-20-2075; Reclamation and Authority 2001) established the terms and conditions for using the Drain and operation of the GBP. They include monthly and annual load objectives, plus significant fines for exceeding these limits, and have monitoring requirements for the Authority to continue using the Drain. According to Section V of the current Agreement (Reclamation and Authority 2009) and the Record of Decision (Reclamation 2009), the objectives of the comprehensive monitoring program are to provide the following information:

- water quality data for purposes of determining the Draining Parties’ compliance with Selenium Load Values and Salinity Load Values as set forth in this Agreement;
- biological data to allow an assessment of whether any environmental impacts constitute Unacceptable Adverse Environmental Effects that have resulted from this Agreement; and
- data on sediment levels, distribution, and selenium content.

Specifics of the monitoring and reporting program (which includes contaminant monitoring in the Drain, Mud Slough, and the San Joaquin River; acute and chronic toxicity monitoring in Mud Slough; sediment monitoring; and storm water monitoring) are provided in a series of monitoring plans (e.g., Reclamation et al. 2002, 2013, 2015; Reclamation 2011) and annual reports (e.g., San Francisco Estuary Institute [SFEI] 2010, 2011, 2015; Grassland Bypass Project Oversight Committee 2013). The recent reports (e.g., Grassland Bypass Project Oversight Committee 2013; SFEI 2015) provide helpful compilations of data and comparisons toward meeting the environmental commitments defined for continued use of the Drain and the associated monitoring program.

This chapter provides my review of the GBP environmental monitoring program specifically focused on the control and reduction of selenium in the Grasslands wetlands water supply channels and the lower San Joaquin River. It encompasses surface water, biological and sediment monitoring, but excludes groundwater monitoring that is required under the various monitoring programs. The purpose of this review is to evaluate the current environmental monitoring program and the data collected to date, and to provide recommendations for improvements in monitoring for the duration of the GBP through 2020. The scope of this review includes the areas directly affected by the GBP (the Drain between Russell Avenue and the terminus, Mud Slough (north) from Kesterson Reservoir to the San Joaquin River, and the San Joaquin River between Fremont Ford and Crows Landing). It does not include regional drainage management projects in the GDA, such as the San Joaquin River Improvement Project (SJRIP) and the San Luis Demonstration Treatment Facility (DTF). Although implementation of those projects will affect the success of the GBP, they are regulated by the 2009 Record of Decision and Biological Opinion for the GBP (Reclamation 2009; USFWS 2009), 2007 Record of Decision (Reclamation 2007) and Biological Opinion (USFWS 2006) for the San Luis Unit Feature Re-evaluation, and the 2012 Biological Memorandum for the DTF (USFWS 2012).

The chapter provides my general observations, responses to key questions provided by SFEI to guide feedback in regard to important management issues the GBP seeks to answer, and conclusions and recommendations from the review.

General Observations

The GBP has operated since 1996 under a series of agreements for use of the Drain to discharge subsurface drainage from the GDA through Mud Slough (north) while removing discharges from wetland channels supplying water to state and federal wildlife areas and wetlands in the Grassland Water District. Effects of the GBP have been monitored under comprehensive environmental programs that have provided data on selenium concentrations in surface water, biota, and sediment for evaluation of the Project. Monitoring locations and specifics of the monitoring/sampling have varied somewhat through time on the basis of changing requirements and evaluation of monitoring results. Overall, the available datasets for the locations affected by the GBP and comparable data for upstream reference locations are very comprehensive. Inclusion of historic data in each of the annual reports facilitates understanding of the comprehensive data. Some of the monitoring data (such as tissue analyses for fish and invertebrates) are particularly useful, while other parts of the monitoring program (such as fish community assessment and toxicity testing) are less useful with respect to selenium. These observations are described in more detail in response to the posed questions below.

Responses to Key Questions

Responses to key questions provided by SFEI to guide feedback in regard to important management issues the GBP seeks to answer are provided below.

1. **Is the GBP monitoring program effectively tracking the consequences (i.e., biological effects) of salt and selenium discharges to Mud Slough (north) (i.e., Stations D, I [I2], and E) and the lower San Joaquin River (Station H [R] and N)? Is it effectively tracking potential biological effects downstream of the Merced River confluence (i.e., Station N)?**

The monitoring program includes several components that indicate the potential for biological effects of selenium discharges to these reaches, as reported for surface water, biota, and sediment selenium concentrations in various annual monitoring reports (e.g., SFEI 2010, 2011, 2015; Grassland Bypass Project Oversight Committee 2013). The reporting of selenium concentrations in these media is consistent with requirements of the monitoring plans discussed above. Selenium concentrations in biota give a comprehensive characterization of fish and bird exposure to selenium through the food web and of bioaccumulation in their tissues (whole-body fish or bird eggs); those results of the monitoring program are most useful in assessing the potential for effects in the exposed biota. The potential effects of the selenium concentrations in biota can be assessed by comparisons of concentrations in diet (e.g., invertebrates, plant seeds) and the receptors (e.g., as measured in whole-body fish or bird eggs) to levels of concern or toxicity thresholds for fish or birds (as they have been in the annual reports), but actual effects of selenium have generally not been determined. When monitoring/sampling results indicate a potential for effects in fish but actual effects are uncertain, following a phased approach that includes laboratory studies is generally appropriate for reducing those uncertainties (Janz et al. 2010; Ohlendorf et al. 2011). For example, eggs of selected fish species that are common in the fish community at Station D could be fertilized in the field and then transported to the laboratory, where embryos and larvae would be monitored for effects.

Selenium concentrations in surface water and sediment also are useful for monitoring purposes and the concentrations can similarly be compared to literature-derived effect benchmarks (as they have been in the reports), but assessment of the consequences of those exposures is less direct than for biota selenium. This is because dietary exposure is the most significant exposure pathway for selenium, and tissue concentrations are more interpretable with respect to biological effect concentrations than are those in abiotic media.

Comparisons of recent results for selenium concentrations in surface water, biota, and sediment to previous results for each station in each annual report are very useful for tracking temporal changes in selenium levels and evaluating their potential consequences. In contrast, results for the fish community assessments and examinations of fish for anomalies at Stations E and H (as well as G) have shown no trends in composition of the fish community or occurrence of selenium-related anomalies in fish (Grassland Bypass Project Oversight Committee 2013). This is not surprising, given the high variability in community metrics and low probability of finding selenium-related anomalies under site conditions.

Short-term toxicity testing of surface water with invertebrates or algae (as has been conducted historically or required by the WDRs) is not very useful for evaluating actual or potential biological effects of selenium discharges to fish, birds, or their food webs. As noted in Chapter 12 of the 2010-2011 report (Grassland Bypass Project Oversight Committee 2013), “The current toxicity test regime was not designed to characterize potential impacts of selenium to aquatic organisms and wildlife. It was designed to document whether the GDA discharges would result in increased aquatic toxicity in receiving waters.” And “The observed toxicity may be related to any number or combination of pesticides and other chemicals that entered the surface water system, which consists predominantly of subsurface drain discharges.” Similarly, although toxicity testing of sediment may be useful for general monitoring of trends, results likely will be confounded by the presence of other chemicals, an association of effects with selenium concentrations will be uncertain. Although *Hyaella azteca* (the test organism used for GBP sediment toxicity testing) are more sensitive to waterborne selenium than some other species (e.g., *Chironomus* sp.), selenium concentrations causing lethality in acute and chronic literature-reported bioassays were $>100 \mu\text{g/L}$ (deBruyn and Chapman 2007). I did not find toxicity threshold values for selenium in sediment for *Hyaella*, and tissue concentrations were neither measured in the GBP test organisms nor did I find threshold values for them in the literature.

2. Is the scope and frequency of monitoring of wetland channels and critical habitat (designated habitat for listed species but critical habitat has not been designated for giant garter snake or San Joaquin kit fox) effective in tracking regulatory commitments?

Water quality monitoring of wetland channels has occurred in Salt Slough (Station F, where sediment also is monitored although not required by applicable WDRs), Camp 13 Canal (Station J), Agatha Canal (Station K), San Luis Canal (Station L2), and Santa Fe Canal (Station M2) (see, e.g., Grassland Bypass Project Oversight Committee 2013). Salt Slough is the principal wetland water supply channel for wildlife areas from which drainwater has been removed by the GBP. Station F, located where State Highway 165 crosses Salt Slough, is the monitoring location that reflects selenium concentrations in water for the upstream state and federal wildlife areas, while Stations L2 and M2 characterize inflows to the North Grasslands and Stations J and K do so for inflows to the South Grasslands. Monthly mean selenium concentrations in Salt Slough and the Grassland wetland channels have decreased and usually have met the (monthly mean) objective for fish and wildlife since construction of the bypass. Biota sampling for Salt Slough occurs on the San Luis National Wildlife Refuge at Station F, about 2 miles upstream of where Highway 140 crosses the stream. Selenium concentrations in fish and invertebrates from Salt Slough declined markedly soon after implementation of the GBP and have remained below the toxicity level and mostly below the level of concern. The scope and frequency of monitoring at those locations has been consistent with regulatory commitments.

Critical habitat has not been designated for either the giant garter snake or San Joaquin kit fox (USFWS 2009). Giant garter snakes may occur in permanent aquatic habitat or habitats seasonally flooded during the snakes' active season (early-spring through mid-fall), such as marshes, sloughs, ponds, low-gradient streams, irrigation and drainage canals, and rice fields. Giant garter snakes have historically been found in areas potentially affected by GDA discharges (e.g., along Santa Fe Grade), but surveys in 2003 through 2007 (California Department of Fish and Game [CDFG] 2003, 2004, 2007; Hansen 2007, 2008) did not find any occurrences of the species in the potentially affected wetland channels or downstream of discharges to Mud Slough (north). Thus, it is unlikely the GBP would cause significant exposure or adverse effects in the species.

However, in its Biological Opinion, USFWS (2009) stated "Through requirements of the Service's biological opinion on interim water contract renewals (USFWS 2000a), Reclamation will support studies on selenium impacts to giant garter snakes (USBR 2009)." Little is known about the effects of selenium on the giant garter snake. Bioaccumulation of selenium was measured in common garter snakes (*Thamnophis sirtalis*) exposed in a laboratory study (Sheffield 2006). Snakes receiving a diet of fish that had been fed a selenium-fortified diet accumulated selenium in their blood, but adverse effects on reproduction (typically the most sensitive biological endpoint for egg-laying vertebrates) were not included in the study.

In its Biological Opinion, USFWS (2009) mentioned concern about effects of construction activities and selenium bioaccumulation by salt-tolerant crops in the GDA and SJRIP with respect to the San Joaquin kit fox. Monitoring of selenium concentrations in vegetation and small mammals designed to detect potential selenium exposure by kit foxes in those areas began in 2008, and monitoring of blood and hair in coyote was initiated in 2010. Levels measured in 2010 and 2011 showed concentrations below concern levels (Grassland Bypass Project Oversight Committee 2013). As mentioned above, the GDA and SJRIP are outside the area considered in this review, and those results were not reviewed in detail.

Depending on availability, waterfowl and/or shorebird eggs were collected from areas adjacent to Mud Slough and the Drain in the spring of each year from 1996 through 2011 (Grassland Bypass Project Oversight Committee 2013). The data were evaluated for potential effects on bird reproduction (discussed under other questions) but not as food items for kit foxes. The monitoring program also includes sediment quality and quantity in the Drain. The USFWS (2009) mentioned its objection to disposal of sediments containing selenium at concentrations exceeding 2 mg/kg dry weight to nearby upland open areas because of risk to wildlife foraging in those areas. Those risks may exist for terrestrial species, including the kit fox, but a risk assessment for the sediment application to land was beyond the scope of this review.

The Reclamation and Authority request for consultation with the USFWS on potential effects of the GBP to listed species indicated that the Delta smelt would not be affected by the proposed action. In its Biological Opinion, the USFWS (2009) reiterated from a 2001 biological opinion that, although Delta smelt do not reach Mud Slough or the San Joaquin River above the Merced River, GBP discharges travel downstream via the San Joaquin River to the Delta and Delta smelt critical habitat, and that the species should be listed under the "may affect" category. This review considered potential effects in the lower San Joaquin River as reflected by data from Station N, but not into the Delta, as agreed during scoping discussions for the review. Thus, effects on the Delta smelt are similar to those for other species at that location, where recent monthly mean selenium concentrations have been well below the 5.0 µg/L water quality objective (Grassland Bypass Project Oversight Committee 2013). It seems unlikely the species would be adversely affected in its critical habitat.

Critical habitat for the California Central Valley Steelhead Distinct Population Segment is located in the San Joaquin River upstream to the Merced River and up that river, but not in the San Joaquin River upstream of that confluence (National Oceanic and Atmospheric Administration [NOAA] 2005). Thus, exposure of steelhead (*Oncorhynchus mykiss*) would occur mainly downstream of the Merced River confluence and can be evaluated by the data for Station N, where water quality is monitored but selenium concentrations in biota are not. Recent monthly mean selenium concentrations at Station N have been well below the 5.0 µg/L water quality objective (Grassland Bypass Project Oversight Committee 2013), and should not adversely affect the steelhead.

Also, adult steelhead do not feed extensively when they return to fresh water to spawn (NOAA 2005), so the threshold for effects on growth of juvenile salmonids is more relevant than thresholds based on reproductive effects. The monitoring reports (e.g., Grassland Bypass Project Oversight Committee 2013) describe derivation of conservative threshold concentrations for effects in juvenile salmonids, but tissue data are not available from the steelheads' critical habitat for evaluation. Also, USEPA (2015) derived a less conservative threshold for reduced growth of juvenile Chinook salmon (*Oncorhynchus tshawytscha*) fed a selenomethionine-dosed diet (whole-body selenium concentration of 7.36 mg/kg), based on the same study by Hamilton et al. (1990) that was used to derive lower effect concentrations in the monitoring report (Chapter 7).

3. Is the project design based on the current understanding of the environmental science of selenium?

The location of monitoring stations (upstream and downstream of discharges; wetland channels) and the frequency of monitoring (more frequent for variable concentrations in water, less frequent for biota and sediment that change more slowly) are consistent with the understanding of selenium behavior in the environment. The monitoring program has included a comprehensive evaluation of selenium concentrations in different invertebrate taxa (including "zooplankton" at Station I/I2, as shown in Figure 5F of Grassland Bypass Project Oversight Committee 2013, although it is difficult to distinguish different taxa in the figure) and fish species in the affected drainages and established patterns among the trophic levels sampled. Knowing the selenium concentrations in these diverse biota allows for direct interpretation of the exposure levels and potential for effects in different species and trophic levels and for monitoring of temporal trends. The focus on exposures of fish and birds as well their diet is consistent with the environmental science of selenium. Monitoring of particulates to make the linkage between waterborne selenium concentrations and those in the lowest trophic levels of invertebrates would provide more information about the bioaccumulation of selenium (consistent with Presser and Luoma 2010, for example), but is not necessary for evaluating potential effects, because of the direct interpretability of the monitoring data to assess risk.

Fish community assessments and examinations of fish for anomalies are sometimes useful for selenium assessment if reproductive effects are expected or they are severe (Janz et al. 2010; Ohlendorf et al. 2011), and they have been included in the comprehensive monitoring program. However, monitoring at Stations E, G, and H has shown no trends in composition of the fish community or occurrence of selenium-related anomalies in fish (Grassland Bypass Project Oversight Committee 2013). This is not surprising, given the high variability in community metrics and low probability of finding selenium-related anomalies under site conditions, as noted above in response to Question 1.

4. What are the key data that contribute to the better scientific understanding of the behavior of selenium in the system? How can GBP datasets be modified or improved to be more helpful to other programs (e.g., San Joaquin River Restoration Program, Bay-Delta Restoration Program, and the development of site-specific or tissue-based selenium standards by USEPA)?

The key data are the multi-media monitoring results (i.e., in water, biota, and sediment) from multiple stations since the GBP was implemented in 1996. The comprehensive, long-term monitoring program supports understanding of selenium sources, transport through the Drain and wetland channels, and bioaccumulation in fish, birds, and their food webs. The unique dataset available from the GBP monitoring program also facilitates understanding temporal associations of selenium concentrations among different trophic levels of biota and those in water (e.g., Beckon 2014), which contributes substantially to understanding of selenium behavior in this system. Levels of concern and toxicity thresholds for interpretation of results and prediction of risk (i.e., potential effects) are fairly well established for fish and birds. Information also is available for amphibians (bullfrog tadpoles; *Lithobates catesbeianus*) at selected monitoring stations and the data are useful for monitoring temporal trends, but effect levels are less well established for amphibians than for fish or birds (Janz et al. 2010), so the monitoring data are not readily assessed for prediction of risk to amphibians.

The available monitoring data (e.g., as reported and summarized in Grassland Bypass Project Oversight Committee 2013) should be useful to other programs having an interest in the spatial patterns or temporal trends of selenium concentrations associated with the GBP (such as San Joaquin River Restoration Program and Bay-Delta Restoration Program). They can be used to evaluate the potential for adverse effects in fish and birds inhabiting the areas receiving GDA drainage or in areas from which the drainage has been removed (as reflected by data for wetland channels and associated biota from Station F) and similarly for humans eating fish from the drainages. Selenium concentrations in invertebrates (as dietary items for fish or birds), whole-body fish, or bird eggs in Mud Slough (including the backwater at Station I/I2) often exceed levels of concern and indicate the potential for adverse effects in fish and birds, whereas the risks of effects in other wetlands have been greatly reduced. Samples of carp (*Cyprinus carpio*) muscle collected at Station E, but not at Stations G or H, have sometimes exceeded the 2 mg/kg (wet weight) screening level for human health protection in recent years (based on California Office of Environmental Health Hazard Assessment [OEHHA] 2008; SFEI 2011). These muscle samples from Stations E and H are considered to be representative of fish that would be eaten by people fishing in areas downstream of the GBP discharge. The frequency of exceedance has been about 10 percent in the carp samples from Station E since implementation of the GBP (SFEI 2011; Grassland Bypass Project Oversight Committee 2013).

If there is a concern about actual effects (e.g., to satisfy the receiving water limitation on p. 13 in the Order [Regional Board 2015] that states “The discharge from the San Luis Drain shall not cause or contribute to the following in Mud Slough (north) or the San Joaquin River . . . Toxic pollutants to be present in the water column, sediments or biota in concentrations that adversely affect beneficial uses; that produce detrimental physiological response in human, plant, animal, or aquatic life (emphasis added); or that bioaccumulate in aquatic resources at levels which are harmful to human health.”), more detailed toxicological/effect studies are needed.

Similarly, if more detailed information is needed for development of site-specific or tissue-based selenium standards by USEPA, it may be useful to include analyses of particulate selenium in the water column concurrently with water and biota sampling at locations where both water and biota are sampled for selenium analysis. Although bioaccumulation modeling can be done without including particulates, they were included in the USEPA (2015) modeling conducted for development of the draft aquatic life ambient water quality criterion for selenium and likely would be used by USEPA for development or future modification of site-specific or tissue-based selenium standards.

5. **Does the Biological Effects Monitoring Program achieve the following? If not, how can it be improved?**
- a. **Monitor selenium concentrations across all important media (water, particulates, tissue of different food web species) that contribute to selenium bioaccumulation in ecosystems.**

Although particulates (e.g., suspended particulates filtered from the water column, algae, or phytoplankton) are not sampled, the inclusion of this matrix in the monitoring program is not necessary to understand exposure and potential risk of selenium effects in fish, birds, or humans. This is because concentrations measured in fish (whole-body or muscle) and birds (eggs) as well as their food webs (invertebrates and plant seeds) allow for direct comparisons to levels of concern or toxicity thresholds for effects in the important endpoint receptors. If there is an interest in modeling selenium bioaccumulation in the ecosystem (i.e., from water to endpoint receptors or the reverse thereof, such as in Presser and Luoma 2010), monitoring of particulates would be essential.

- b. **Identify at-risk species and their food webs to protect communities from selenium exposure.**

Yes, the monitoring program is very comprehensive and identifies all of these for evaluation of potential effects/risk. In fact, as noted in Conclusions and Recommendations, the number of invertebrate taxa and fish species that are analyzed could be reduced somewhat without substantially decreasing the ability to detect spatial patterns and temporal changes associated with GBP discharges. If it is necessary to understand actual effects on the at-risk species, additional studies would be required, as noted elsewhere in this chapter.

- c. **Determine the environmental risk occurring to fish and wildlife (including protected species such as the giant garter snake and San Joaquin kit fox) in Mud Slough (north) and lower San Joaquin River.**

Yes, direct measurement of selenium concentrations in whole-body fish and in bird eggs as well as fish and bird food webs (invertebrates and plant seeds) allows for direct comparisons to levels of concern or toxicity thresholds for effects in the important endpoint receptors. In this way environmental risk can be predicted and changes through time can be determined by differences in the frequency of exceedance of the benchmarks used for evaluation. The levels of concern and toxicity thresholds used in the monitoring program are conservative, so they should be protective for the receptor species.

Please see response to Question 2 about giant garter snake, San Joaquin kit fox, and other protected species.

- d. **Are there environmental benefits of diminished selenium in other parts of the Grasslands wetland water supply channels and the San Joaquin River? Biological monitoring in potentially improved channels has occurred at Mud Slough (north), upstream of the SLD discharge (Site C), Salt Slough (Site F and F2), and San Joaquin River at Fremont Ford (Site G).**

Yes, the environmental benefits of diminished selenium concentrations are reflected by monitoring data from wetland channels and the San Joaquin River. Selenium concentrations in water at all locations from which high-selenium drainage water was removed, in fish and invertebrates from Salt Slough (Station F), as well as in fish from the San Joaquin River (Station G) declined rapidly after implementation of the GBP and have remained low, only occasionally exceeding the level of concern (Grassland Bypass Project Oversight Committee 2013). Please see also response to Question 2 for other comments about monitoring of the wetland channels.

- e. **Is the GBP supporting healthy fish communities within the study area? Does the GBP monitor fish communities and abundance to be effectively integrated within other projects?**

The “health” of fish communities within the study area is uncertain. Fish community assessments have been conducted since 1993 in Mud Slough at Highway 140 (Station E) and in the San Joaquin River at Fremont Ford (Station G) and below Mud Slough (Station H) to describe species richness, abundance and community structure (Grassland Bypass Project Oversight Committee 2013). Fish assemblages from these sites were compared both spatially and temporally to determine whether conditions for fish species in the San Joaquin River improved and conditions in Mud Slough degraded. Because the GBP began operation in 1996, this sampling schedule was intended to provide a before-and-after characterization of the fish communities at these sites. No time trends are apparent in fish species assemblages during 1993 to 2011 at these locations, and no time trend is evident in total anomalies for the various groups of fishes at each site. After 15 years of Project operation, no clear pattern of temporal or geographic variation in fish community structure attributable to the Project has emerged. However, as noted in the report, “current methods of assessing fish species assemblages may lack the power to detect all but the most pronounced alterations in community structure.”

- f. **Does the Biological Effects monitoring accomplish an assessment of the risk to human health from consumption of fish from affected channels?**

Adequacy of monitoring data to assess risk to human health from consumption of fish from affected channels should be determined by OEHHA, because that agency has particular requirements for adequacy of data. About 10 percent of the more than 120 samples of carp muscle collected from Mud Slough at Highway 140 (Station E) have exceeded the 2 mg/kg (wet weight) screening level for human health protection in recent years. In contrast, selenium concentrations in carp muscle samples from the San Joaquin River at Fremont Ford upstream of Mud Slough (north) (Station G) and San Joaquin River at Hills Ferry (Station H) have not exceeded the screening level. The carp muscle samples from Stations E and H are considered to be representative of fish that would be eaten by people fishing in areas downstream of the GBP discharge.

Conclusions and Recommendations

Comprehensive monitoring programs have provided large datasets for evaluation of the environmental effects of the GBP since it was implemented in 1996. Selenium concentrations in surface water, biota, and sediment have been measured through time, and they provide a basis for comparing locations affected by the GBP to conditions in upstream reference locations. Sampling of water and sediment for selenium analyses is more straightforward and less effort than that for biota sampling. Continuing monitoring of those media in accordance with the 2015 monitoring plan (Reclamation et al. 2015) should meet the requirements of the Use Agreement (Reclamation and Authority 2009) and WDRs (Regional Board 2015). I recommend that approach.

Monitoring data for invertebrates, fish, and bird eggs are particularly useful for evaluating potential adverse effects from selenium. Available data provide a good understanding of trophic level and inter-taxa (invertebrate) or inter-species (fish) differences. Those data should be evaluated toward streamlining the analyses for selenium. I recommend consideration of reducing the frequency of sampling to less than quarterly by focusing on the most important season, which, for selenium, would be the time when egg development is occurring. It seems possible that sampling twice a year would meet the monitoring goals for detecting spatial patterns and temporal trends.

Available data also should be reviewed to determine if a subset of invertebrate taxa and fish species would provide sufficient data for monitoring spatial patterns and temporal trends to meet the objectives of monitoring. It seems likely that representative taxa/species could be identified for analyses, and the other samples could be archived for possible future analyses, if they were warranted.

Monitoring of bird eggs also should be continued, consistent with the 2015 plan (Reclamation et al. 2015).

If more detailed information is needed for development or future modification of site-specific or tissue-based selenium standards by USEPA, I recommend including analyses of particulate selenium in the water column concurrently with water and biota sampling at locations where those media are sampled for selenium analysis. Although bioaccumulation modeling can be done without including that trophic level, it likely would be used for development or modification of site-specific or tissue-based selenium standards by USEPA in the longer timeframe (it would not be helpful in the shorter timeframe, if USEPA develops standards within the next year, as expected). Cost savings realized from streamlining the biota monitoring effort (as suggested above) could be used to support the analyses of particulates.

The *potential effects* of the selenium concentrations found in biota can be assessed by comparisons of concentrations in diet (e.g., invertebrates, plant seeds) and the higher trophic-level receptors (e.g., as measured in whole-body fish or bird eggs) to levels of concern or toxicity thresholds for fish or birds (as they have been in the annual reports), but actual effects of selenium have generally not been determined. Selenium concentrations in fish collected at Station D (Mud Slough (north) below the GBP discharge from the Drain) often exceed the level of concern, and sometimes the toxicity threshold. If assessment of potential effects is adequate for meeting goals and objectives for the monitoring, comparisons to the threshold levels should be continued. However, if it is desirable to determine actual effects, I recommend considering laboratory studies with a selected fish species as an approach for reducing uncertainties about effects. For example, eggs of a fish species that is common at Station D and for which adequate husbandry information is available could be fertilized in the field and then transported to the laboratory, where embryos and larvae would be monitored for effects.

Because results for the fish community assessments conducted since 1993 have shown no trends in composition of the fish community, I recommend discontinuing fish community assessments, the requirements of the WDRs (Regional Board 2015) notwithstanding. Given the high variability in community metrics and low probability of detecting differences/changes, it seems unlikely that community assessments will be worth the monitoring effort. Although examinations of fish for selenium-related anomalies in fish since 1993 also have shown no trends, fish that are collected for selenium analyses should be examined for anomalies, even though the incidence is likely to be low.

Toxicity testing of water with invertebrates, such as has been included in the monitoring program, may be useful for other purposes, but such tests are not very relevant for selenium, in part because of the relative insensitivity of invertebrates. As noted above, the results also are likely to be confounded because of the presence of other chemicals in the water.

Similarly, although toxicity testing of sediment may be useful for general monitoring of trends, results likely will be confounded by the presence of other chemicals, and an association of effects with selenium concentrations will be uncertain.

Inclusion of historic data in each of the annual reports facilitates understanding of the comprehensive data. This should be continued in future reports.

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About the Reviewers



Dr. Thomas Grieb is a principal scientist and director of the Research & Development Group at Tetra Tech, Inc. with 40 years' experience. He received his B.A. in Zoology from the University of California, Berkeley and M.A. degrees in Marine Biology from San Francisco State University and Biostatistics from the University of California Berkeley. He received his Ph.D. from the University of California, Berkeley in Environmental Health Science. He recently served as project manager for technical efforts that supported the preparation of the Selenium TMDL by the California Regional Water Quality Control Board. This work included source characterization and selenium loading assessment, assessment of the toxicological effects of selenium on biota, the development of a conceptual model of the processes that affect selenium biogeochemistry, and the modeling of selenium fate and transport in the Bay-Delta. His primary research interests include the behavior of metals in the aquatic environment and the application of statistical methods to characterize uncertainty in environmental datasets and simulation models.



Prof. Dr. David Janz grew up in Vancouver, BC, Canada and was educated at Simon Fraser University (B.Sc. Ecology, 1987), Trent University (M.Sc. Watershed Ecosystems, 1991) and the University of British Columbia (Ph.D. Pharmacology and Toxicology, 1995). After postdoctoral training at the University of Guelph, he was an Assistant Professor in the Department of Zoology, Oklahoma State University from 1997-2002. In 2002, he joined the faculty in Veterinary Biomedical Sciences, University of Saskatchewan, where he was promoted to Professor in 2008. His academic position is closely associated with the Toxicology Centre at the University of Saskatchewan, where he is currently Associate Director (Academic) and Chair of the Toxicology Graduate Program. Prof Janz's research program focuses on how environmental stressors such as toxicants interact with physiological processes in vertebrate animals, primarily aspects of developmental biology and reproductive endocrinology. Although fish have always been his primary animal model, he has also published research conducted in amphibians, reptiles, birds, and mammals. Since 2005, a major focus of his research program has been on the aquatic ecotoxicology of selenium, where he has published over 30 peer-reviewed journal articles and book chapters on this topic, including three Highlighted Articles in the journal *Ecotoxicology and Environmental Safety*, and the 2014 Best Paper Award in the SETAC journal *Integrated Environmental Assessment and Management*.



Dr. Harry Ohlendorf has been employed at CH2M HILL since 1990 and is a Certified Wildlife Biologist. He received his undergraduate and graduate degrees from Texas A&M University, including a B.S. in Wildlife Management (Fisheries Option) in 1962, an M.S. in Wildlife Science in 1969, and his Ph.D. in Wildlife Science in 1971. At CH2M HILL, he manages or provides technical oversight for a wide variety of environmental projects, including the planning, implementation, and reporting of site ecological characterizations and surveys, contaminant exposure and effect analyses, risk characterization, and project impact evaluations. He provides firm-wide technical guidance in the area of ecological risk assessment and risk management. Recent projects include the application of wildlife toxicology knowledge to projects for government (e.g., California Department of Water Resources; USFWS; USEPA; USDOE; U.S. Army, Air Force, and Navy) and private-sector (e.g., oil refinery, coal and metal mining, chemical manufacturing) clients in a wide variety of environmental settings.

Dr. Ohlendorf began his career with the U.S. Fish and Wildlife Service's Patuxent Wildlife Research Center in Laurel, Maryland, where he served for 7 years as assistant director of the Research Center and was actively involved in pollution ecology research. Subsequently, he was leader of the Center's Pacific Coast Research Station in Davis, California, and studied the pollution ecology of wildlife. For 18 years, Dr. Ohlendorf's research for the USFWS focused on the occurrence and impacts of contaminants in aquatic and terrestrial ecosystems.

Ecotoxicology of selenium is one of Dr. Ohlendorf's specialties. He has been recognized as one of the "Pioneers of Selenium Research" in a book, *Environmental Chemistry of Selenium* (edited by W.T. Frankenberger, Jr., and R.A. Engberg; Marcel Dekker, Inc., 1998), and was a co-editor of *Ecological Assessment of Selenium in the Aquatic Environment* (along with P.M. Chapman, et al; CRC Press, 2010). He has authored numerous related technical papers and book chapters. His current interests also focus particularly on the importance of doing well-planned, relevant studies of the effects of environmental contaminants in fish and wildlife, and application of the results of those studies to environmental decision-making. Dr. Ohlendorf has been a member of The Wildlife Society's Wildlife Toxicology Working Group since it was formed, and has served as Chair of the Group.