A Process-Oriented Approach to the Biological Removal Of Selenium

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Abstract

As more is learned about selenium toxicity, increasingly stringent discharge levels make selenium another potential concern for mine closures. Biological treatment of selenium has been used successfully and offers the possibility of a low-cost, low-input means of treatment. However, a thorough understanding of the processes involved is crucial to an effective design. There are several biological processes which can remove selenium from a water column, including volatilization, sequestration, and bacterial reduction. Bacterial reduction of selenate and selenite oxyanions appears to be the key process in removing selenium from aquatic systems. Two case studies of biological treatment are presented, with an emphasis on the design approach and how preliminary benchscale tests contributed to an understanding of the processes being designed for. Removal rates of 88% and 96% were obtained in the laboratory tests.

Introduction

The past two decades have seen an extraordinary increase in concern about the cycling of selenium as a result of industrial activity. Concerns regarding selenium toxicity were galvanized in the early 1980s as a result of field studies on declining migratory bird populations at the Kesterson Reservoir in California. The studies specifically attributed the decline in reproduction of waterfowl, shorebirds and other aquatic birds to selenium[i]. Subsequent studies of other reservoirs exhibiting impacts from Se revealed that this was not an isolated incident[ii]. These cases involved high selenium concentrations in agricultural drainage or runoff from coal ash. Many mines throughout North America are now being forced to address selenium as an effluent problem.

Selenium is widely distributed in the environment, and is typically present at sub- to low part per billion (ppb - µg/L) concentrations. It is an essential micronutrient for mammals, birds, fish, algae and many bacteria[iii], but can be toxic at concentrations only slightly higher than typical ambient concentrations[iv]. A complex chemistry, the vanishingly low concentrations at which toxicity can occur, and a poor understanding of the exact circumstances leading to toxicity have caused a tremendous state of flux in this field. Certain species are highly tolerant, while others show chronic effects with intake concentrations as low as 8 ppb[v]. Particularly at risk are certain species offish and waterfowl.

The apparent species-specific nature of selenium toxicity" has led many to question the suitability of a single discharge criteria. At any rate, regulatory bodies ranging from the World Health Organization to provincial ministries of the environment have felt the need to set guidelines.

The CCREM guideline for the protection of aquatic life for selenium is 1 µg/L. The IJC recommended 1 µg/L in unfiltered water, 5 µg/g in sediments, and 3 µg/g (wet) in aquatic biota. EPA discharge standards are *5* ppb "chronic" and 20 ppb "acute". The EPA is currently evaluating a lower chronic criterion of 2.5 ppb.

Se Chemistry

Selenium (Se) is physically and chemically similar to sulfur, and is present in the environment in both inorganic and organic forms. The inorganic forms are elemental Se (Se°), selenite (Se[IV], SeO_3^{2-}), selenate (Se[VI], SeO_4^{2-}), and hydrogen selenide (Se[-II], H_2Se). In aquatic systems, the most important forms of Se in terms of bioavailability and toxicity are the oxyanions selenite [Se(IV)] and selenate [Se(VI)]. These forms can be transformed into organoselenium by living organisms. Organoselenium is both highly toxic and easily transferred through the food chain.

Se cycling in aquatic ecosystems is dominated by biological transformations. Bacteria, plants and animals can assimilate dissolved Selenium oxyanions and either incorporate them into organoselenium compounds, detoxify them as volatile methylselenides, or return them to the water (depuration or elimination). Se bioaccumulates (i.e. it accumulates in the tissues of living organisms), and thereby reaches; toxic levels in aquatic ecosystems. Selenium retained in plants and animals will either be taken up by predators or cycle through detritus upon death of the organism.

When Selenium concentrations in water exceed typical ambient concentrations, Se begins to be substituted for sulfur in living organisms. Specifically, it is incorporated into the sulfur-containing amino acids cysteine and methionine. Because selenium and sulfur behave differently, these seleno-amino acids alter the proteins into which they have been incorporated. When seleno-amino acids reach high enough concentrations in the body, they impair the normal function of the organism, resulting in toxic effects .

Treatment options

Although many different chemical, physical, and biological processes have been examined for the removal of selenium, few engineered solutions have been shown to be successful on a commercial scale. In the past, the most widely used approach involved chemical co-precipitation with iron salts at an acidic pH. While this process is effective with selenite, selenate is left untouched. Additionally, chemical co-precipitation is not capable of reducing selenium concentrations to currently existing discharge criteria. The biological nature of the transformations leading to toxicity suggests that the most straightforward solution to toxicity may also be biological. The recent need to address selenium toxicity has resulted in developments focused on the exploitation of biological systems which are known to remove selenium to low ambient concentrations. Bioreactors, amended soil beds, evaporation ponds and wetlands have been used with some success in treating selenium. However, because the science is still in a state of infancy, it is still a hit-and-miss proposition in many cases.

To date, the typical approach for biological remediation has been a "black box" one, with a design focus on mass loading and removal relationships. What is lacking in this approach is an appreciation and understanding of the *processes* responsible for removal. These processes and their applicability are outlined below:

1. Adsorption - this makes a poor treatment option as surfaces quickly become saturated and the selenium retained is still biologically available and therefore a toxicological risk.

2. Bacterial, algal and macrophyte volatilization - can be a useful process component, but reported volatilization rates are low, difficult to measure, and may require saturation concentrations to occur.

3. Plant uptake and sequestration - this can effectively tie up selenium for as long as the plants are alive. Selenium is still biologically available to organisms feeding from the plants.

4. Bacterial reduction - occurs under reduced conditions (such as in a sediment layer) in the presence of selenate reducing bacteria. Oxidized selenium is reduced to elemental selenium and is biologically unavailable as long as it remains in the sediment. The bulk of removed selenium in wetlands is found in the sediment, indicating that bacterial reduction is a crucial process.

An understanding of these processes and how they cam be incorporated into a treatment design requires a broad review of literature from the various studies and treatment attempts of the past two decades. As bacterial reduction is the dominant mechanism for long-term removal, an understanding of microbiology is crucial to an understanding of the process.

Microbial Technologies, Inc. has been involved in the development of cost-effective biological systems to treat a number of pollutants, including selenium. One of the keys to the success of our solutions is the *process-oriented* approach given our biological treatment systems. It is the natural result of combining microbiology and ecology with an understanding of process control. Following are descriptions of how treatments were developed for two clients with selenium effluent problems.

Case Study #1: Gold mine tailings water

Microbial technologies joined an engineering firm contracted for the closure of a gold mine in the western U.S. The site has a positive water balance and a large tailings pond which is nearly full. The water that needs to be discharged will first require treatment. We were tasked with the design of a treatment system for a large tailings pond containing 700 ppb selenium (an approximately even split of selenate, selenite and selenocyanate), as well as high concentrations of a few metals (Pb, Zn, Fe, Cu, Ni). The mandate was to create an inexpensive, low-maintenance treatment capable of reducing these concentrations to meet discharge standards.

Approach

Bacterial transformation was agreed upon as the process best suited for removing selenium from aquatic systems. With the client's high water volume, positive water balance, and desire for a fast treatment, an in-pond treatment was selected.

The bacteria responsible for reducing selenium oxyanions require anoxic conditions, carbon, nitrogen and phosphorus. The treatment concept was to provide the optimal environment for the bacteria in as simple a manner as possible. The design consists of an engineered sediment capable of hosting the necessary bacteria consisting of organic matter (to provide a carbon and nutrient source) on top of a sediment layer. The sediment layer must provide a reduced environment, as selenium-reducing bacteria require anoxic conditions. The presence of selenate- and selenite reducing bacteria is ensured through lab tests. Selenium is reduced at the sediment/water interface by the bacteria, and will remain in its reduced, elemental form as long as it is buried in a reducing sediment. Inputs of fertilizer provide additional nitrogen and phosphorus for the bacteria and also promote algal growth, which in turn revitalizes the layer of organic matter. This is a low-input, low-maintenance approach with much room for flexibility. It has easily controlled and measurable inputs and outputs.

In order to test the validity of this approach, Microbial Technologies performed a set of bench scale tests. These tests were designed to provide a proof-of-concept, and to yield design and operational parameters. Specifically, we wanted to determine which conditions best stimulate microbial biotransformation and volatilization of dissolved selenium. The following questions were addressed:

- Are there native bacteria that can effectively remove selenate and selenite from the tailings water?
- If not, will bacteria from another source work effectively in mine water?
- How are other metals (copper, lead, zinc, nicked) affected by the treatment?
- Are there selenium-removing algae currently present in the tailings pond, or will it be necessary to introduce them from other sources?
- What level of nutrient addition is required to sustain an algal bloom and promote selenium removal in the water column?
- Can rooted aquatic macrophytes thrive in the tailings pond water with deposited tailings as the substrate? Can they remove selenium from the system?

Test Design

Tailings and mine water were added to eight different test tanks. Mine water alone was added to a ninth tank to serve as a control. The tanks were amended variously with the following:

Organic matter - to determine its necessity for supporting algal and microbial populations. Sediments from an algal pond in Tulare County, California were used. It was previously determined that these sediments contain some selenate-reducing bacteria.

Fertilizer - to supply nutrients for algal and microbial growth. Both high and low levels were tested.

Algae - some tanks were inoculated with an algal strain known to remove selenium. This strain was previously collected and propagated by Microbial Technologies.

Bacteria - some tanks were inoculated with bacteria from a different source, also known to reduce selenate.

Plants - two different types of aquatic macrophytes were planted in tailings.

The test design was such that effects of individual components could be determined.

Florescent lighting and aeration were provided to the tanks on twelve-hour cycles to simulate daylight and natural (convective) mixing. Plexiglass covers were placed on each tank to *minimize* evaporation, and water lost to evaporation was made up with distilled water.

Monitoring

Water samples collected on Days 25, 56 and 83 were analyzed for Selenium and metals.

Filtrate was sampled for chlorophyll-A as a possible measure of algae population.
Weekly tests of temperature, dissolved oxygen, conductivity, and pH were performed on each tank to ensure operational parameters were being met, and to monitor changes in the various ecosystems.
Nitrate levels were also monitored to determine the effectiveness of the tank ecosystems in removing nitrate, since these anions can inhibit microbial reduction of selenate.

Results and Discussion

As much as 88% of the selenium was removed after 56 days. The effectiveness of selenium removal varied with the different amendments.

The greatest removal of selenium was seen in those tanks which received organic matter. Introducing selenate-reducing bacteria from outside sources improved selenium removal, but was not significantly more effective than bacteria already present in the organic matter. Most likely, the benefits of introducing additional bacteria lie primarily in the initial removal rates, when the ecosystem in the sediment is still establishing itself.

Algae was grown in tanks with and without the addition of fertilizer. A high fertilizer dose inhibited algal growth. The addition of foreign algae did not significantly aid removal, and may in fact have inhibited native algal growth.

The development of algal blooms also resulted in removal of other metals (particularly Cu, Pb, Zn_5 Ni, and Hg). Final concentrations were near or below detection limits. Again, removal was enhanced by the presence of organic matter.

The high levels of selenium removal gave the required proof of concept for the proposed approach. The fact that over 80% of the soluble selenium was removed in the tanks indicates that selenate;, selenite, and selenocyanate were removed from the water column in the process. This is a crucial point, as the difficulty of removing selenate and selenocyanate is a stumbling block for many treatments.

The tests yielded a better understanding of the processes involved, methods of monitoring the process, and some operating guidelines. With a confirmation of the process viability, an idea of removal rates and those conditions which promoted microbial activity, the designer is well armed to proceed with a treatment plan.

We have used this information to design a pilot scale test pond, including a supplemental bacteria growing tank.

Case Study #2 Coal ash landfill runoff

A coal power generation plant in the southern U.S. has selenium concentrations of over 50 ppb in runoff from their coal ash landfill. The current NPDES permit limit for Selenium is 29 ppb. A new facility is being planned with the desire to treat runoff of approximately 250 gpm for at least twenty years. A biological treatment was desired because of its ability to treat selenate, and because of the low operating costs.

Microbial Technologies was retained to provide a design for the new treatment facility, and to provide assistance with previously unsuccessful tests of a wetland treatment concept. The previously unsuccessful tests used wetland mesocosms to attempt selenium removal, and offer a contrast between a "black box" approach, and a process-oriented one.

Formerly, the approach in. the mesocosm was to put together soil and wetland plants, circulate runoff through it and expect results. When selenium was not removed from the water, there was neither explanation nor an approach for finding one. Inheriting this mystery, our first question was, "What is the process responsible for removing selenium and is it active here?". Tests performed by Microbial Technologies revealed that no selenium-reducing bacteria were present in the test mesocosm.

By sampling from likely locations on the property (a volunteer wetland in a runoff ditch, and the detritus layer in the collection pond), a source of selenium-reducing bacteria was located. The presence of the bacteria was a clue that the desired process was already at work in the runoff collection pond, even if in limited amounts. Previously, lower summertime selenium concentrations had been attributed to analytical error. It was more likely a result of bacterial activity - crucial organic matter may have been supplied by algae that grew in the warmer summertime temperatures.

When selenium reducing bacteria were introduced to the mesocosm, selenium was immediately removed from the water. This was a relatively simple, yet fundamental adjustment. With the process now functioning, the next step was to determine what elements were limiting optimization. These could include the following:

- organic carbon supply
- inorganic nutrient supply
- water chemistry

A fourth possibility is that the selenium concentration in the runoff water was too low to sustain a healthy population of selenium reducing bacteria.

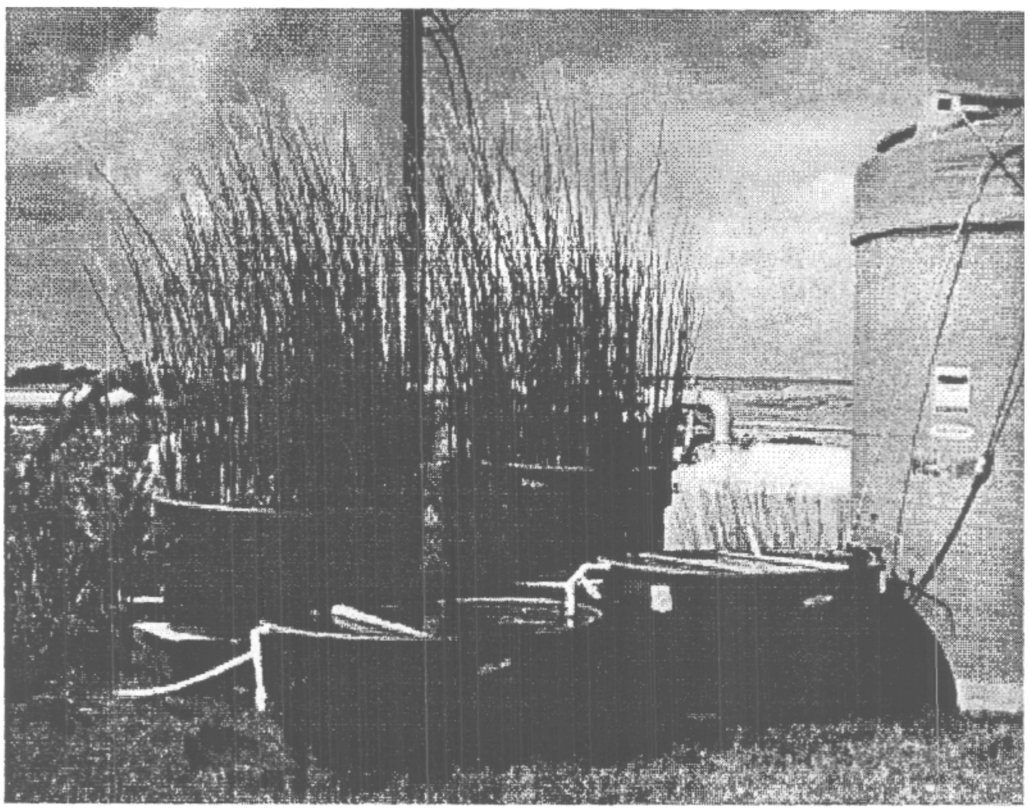

Figure 1, Test mesocosm for wetland removal of selenium

Test Design

With the removal process now working, the next step was to test adjustments that could maximize removal. A series of jar tests were performed to determine the effects on selenium removal of adding different amounts of fertilizer and of increasing the selenium concentration. The latter was achieved by adding fly ash - some blended with the sediment, and some layered underneath.

The results of the tests (see Table 1) showed that selenium removal was aided by both fertilizer addition and fly ash addition. Removal was greatest in sediments receiving a high dose of fertilizer and in sediments with fly ash layered below, combined with a fertilizer supplement. This finding is somewhat counterintuitive since we increased the concentration of contaminant in order to increase its removal. Yet it is a logical approach when examined from a process point of view: low Se concentrations can support only a low number of selenium-reducing bacteria. If the number of these bacteria is a performance-limiting element of the process, their numbers must be increased. More than just a logical supposition, it proved to be a low-cost improvement of treatment performance.

Table 1. Selenium concentrations in Jar Tests.

Sample	Wetland Seds	Fly ash Blended	Fly ash Layered	Fertilizer	[Se] (ppb) after 7 days	[Se] (ppb) after 21 days
1					**137**	116
2	*				60	
3	*			*	36	
4	*			** (High)	9.1	
5	*	*		*	19	39
6	*	*			108	35
7	*		*	*	9.9	5.7
8	*		*		72	7.6

As in the first case study, the designer now has a positive proof-of-concept, an increased understanding of the process involved, and some data on how to optimize that process. The information is specific to the client's site. Only with such information could an effective biological treatment process be designed. The water balance, time scale and topography involved indicated that a wetland is a desirable treatment option, and our approach ensured the design was rigorous, A wetland was designed for incorporation into the new treatment facility.

Conclusion

Two different scenarios dictated two different treatment schemes. In one case, a sediment was engineered for an in-pond treatment of selenium. In the other case, a treatment wetland was designed. In both cases, the design was based on the processes responsible for removing selenium. Relatively simple benchscale tests offered a proof-of-concept, as well as design and operational parameters.

Despite the site- and design-specific nature of biological treatment processes, an understanding of microbiology and ecology can ensure effective system designs. These designs must rely on bench-scale and/or pilot-scale tests. Biological treatment systems will continue to win favour so long as they are designed to support and sustain the desired treatment processes.

References

[i] Ohlendorf, H. M., in Jacobs, L.M. (Ed.), "Selenium in Agriculture and the Environment", Soil Science Society of America, Inc. and American Society of Agronomy, Inc, 1989

[ii] Skorupa, J.P. 1998. Selenium poisoning offish and wildlife in nature: lessons from twelve real-world examples. In: Environmental chemistry of selenium. Chapter 29. Frankenberger, W. T., Jr. and R.A. Engberg (Eds.). Marcel Dekker, New York. pp. 315-354.

[iii] Stadtman, T.C. 1979. Some selenium-dependent biochemical processes. J. Adv.Enzymol. Rdat. Areas MoI. Biol. *48* 1-28.

[iv] Ohlendorf, H.M. and G.M. Santolo. 1994. Kesterson reservoir - Past, present and future: An ecological risk assessment. In Selenium in the Environment. Frankenberger, W.T., Jr., and S.M. Benson (Eds.), Marcel Dekker, New York. pp. 69-117.

[v] Heinz, G.H., DJ. Hoffman, and L. G. Gold. 1989. Impaired reproduction of mallards fed an organic form of selenium. J. Wild. Manage. *53,* 418-428.

[vi] Ecological planning and toxicology, inc. 1997. Ecological risk assessment Southshore wetlands. Final Report. Prepared for Kennecott Utah Copper, Salt Lake City, UT. January 1997.