Cadmium Resistance in *Limnodrilus hoffmeisteri* in Foundry Cove Following a Super Fund Cleanup

A Final Report of the Tibor T. Polgar Fellowship Program

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Abstract

I investigated the effects of the recent restoration of Foundry Cove on an evolved population of the tubificid oligochaete, *Limnodrilus hoffmeisteri*. The return of *L. hoffmeisteri* to the pre-contamination state in terms of population densities, resistance to cadmium, and body burdens of cadmium will indicate a successful restoration. The effects of the restoration on population densities, resistance to cadmium and cadmium body burdens were determined. Sediment concentrations of cadmium are greatly reduced compared to 1984 levels and are not significantly different from cadmium concentrations in the sediment of a control site, South Cove. Foundry Cove *L. hoffmeisteri* no longer differ in resistance to cadmium as compared with South Cove *L. hoffmeisteri*, and no longer exhibit higher cadmium body burdens. The return of cadmium resistance levels to pre-contamination levels is indicative of a Darwinian recovery. However, current population densities of Foundry Cove are slightly reduced as compared to 1984, while the population densities of the control site have increased significantly during the same time period.

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Introduction

Environmental contaminants can often have large detrimental effects on ecosystems. Populations in metal-polluted ecosystems, however, often undergo a variety of evolutionary adaptations allowing them to survive in metal-rich conditions. Cadmium, in particular, has been shown to be an effective agent of natural selection (Klerks and Levinton, 1989b). In the event of an environmental restoration, however, selection pressure is removed. In response to this removal of selection pressure, organisms that previously adapted to contamination may adapt again to new conditions. There are a number of possible responses to an environmental restoration, owing to the complexity of ecosystems. Therefore, different measures of success of environmental restorations are needed. One possible measure of success may be to examine the evolutionary response of organisms to the restoration. The return of organisms to their pre-contamination state of adaptation is one indication of a successful restoration. Such a response to a restoration would be an example of a Darwinian recovery.

The recent restoration of Foundry Cove, a freshwater tidal bay, located 86 kilometers north of Battery Park, on the Hudson River, provides a unique opportunity for evaluating the response of an evolved population to the reversal of the conditions under which natural selection took place. Foundry Cove was once the site of the Marathon Battery Factory, which produced nickel-cadmium batteries from 1953 until the factory closed in 1979. During this time, nickel cadmium waste was discharged into the Hudson River, and in particular into Foundry Cove. In total 179 metric tons of waste were released, 55 metric tons of which were discharged directly into the cove. This resulted in cadmium concentrations in Foundry Cove that ranged from 500 to 225,000 ppm (Knutson et al. 1987), making Foundry Cove the most highly metal-polluted cove at the time. In 1983, the EPA declared that Foundry Cove would be the site of a Super Fund cleanup. This cleanup began in 1994 and consisted of draining and dredging the entire cove, as well as the surrounding marsh, and removing sediment to a depth of 10-30 cm. Following the cleanup, cadmium concentrations in the sediment ranged from 10 to 100 ppm, a huge reduction compared to previous levels.

Prior to the cleanup, it had been found that population densities of *Limnodrilus hoffmeisteri*, the dominant oligochaete in Foundry Cove, were similar to those found in South Cove (Klerks and Levinton, 1989a). The high population densities found in Foundry Cover led researchers to believe that *L. hofffmeisteri* had evolved resistance to cadmium (Klerks, 1987). It was found that Foundry Cove worms were able to absorb more cadmium that South Cove worms, especially in the form of metal-rich granules and metals bound to protein (Klerks and Bartholomew, 1991). This mechanism of resistance has been shown to evolve over a relatively short period of time (Klerks and Levinton, 1989b). Currently, levels of cadmium are comparable to those of South Cove, where the evolution of resistance did not take place. According to evolutionary theory, when selective pressure is weakened, adaptations to special conditions may be lost. Thus, it is thought that after the restoration of Foundry Cove, there might be a decrease in cadmium resistance as compared to previous to the cleanup (Suatoni and Levinton, 1997). A return of resistance levels to those found in South Cove would indicate that a Darwinian recovery had taken place.

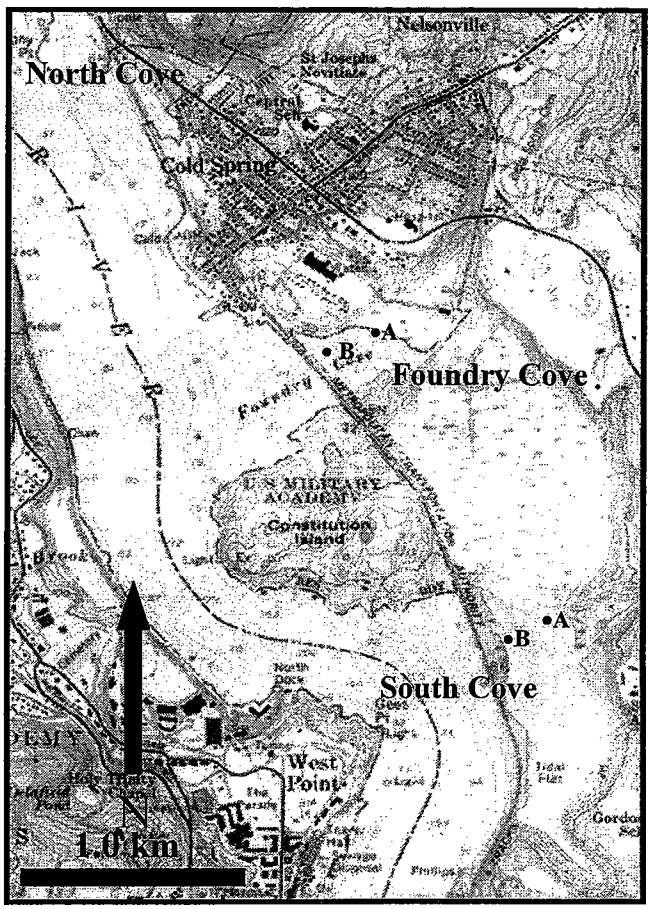
This project concludes a seven year study of the effects of the restoration on *L. hoffmeisteri,* and is one of the first to examine the loss of resistance as a measure of restoration success. We hypothesized that there would be a convergence between resistance levels of *L. hoffmeisteri* living in Foundry and South Coves and that there would be a reduction in body burdens of cadmium in Foundry Cove *L. hoffmeisteri* as compared to worms prior to the restoration.

Methods

Study site

Both sampling sites in Foundry Cove were chosen to roughly correspond with sites sampled in previous years to determine the change of resistance following the restoration (Suatoni and Levinton, 1997). However, previously only one South Cove site was sampled. This year two South Cove sites were sampled. These two sites correspond to those sampled in 2001 by Kelaher and Levinton in order to determine the relative species diversity and abundance in South Cove and Foundry Cove. The South Cove and Foundry Cove sites are indicated in Figure 1.

Figure 1. Map of Foundry Cove and South Cove, and the Locations Within these Sites.



Cadmium concentrations in sediment

Sediment was collected from Foundry and South Coves to a depth of approximately 5 cm. One sample of sediment from Foundry Cove and one sample from South Cove were analyzed for cadmium, using an atomic absorption spectrophotometer.

Resistance to cadmium

Toxicity bioassays were performed in order to determine resistance to cadmium in *L. hoffmeisteri*. Worms that were collected from two sites in Foundry Cove and two sites in South Cove were acclimated for 24 hours to reconstituted deionized fresh water and exposed to dissolved cadmium (8.9μM). Starting with a population of 96 worms, each kept in individual wells of a plastic fraction dish, the time to 50% survival was determined, and the survivorship curves of the worms was compared using Gehan's Generalized Wilcoxon test. The regression line indicating the change in time to 50% survival over the past seven years was determined.

Cadmium concentrations in Limnodrilus hoffmeisteri

Cadmium concentrations in *L. hoffmeisteri* were determined by direct measures of cadmium in an atomic absorption spectrophotometer. Worms were collected from Foundry and South Cove sediment to a depth of approximately 5 cm and were stored in the lab in their own sediment. Upon collection of worms from the sediment, they were placed in reconstituted deionized fresh water, and allowed to empty their gut contents for approximately two days. They were then frozen and stored at -80°C. Approximately 100 worms, or 0.1 gram/wet weight of tissues, were digested in hot nitric acid for analysis.

Population densities

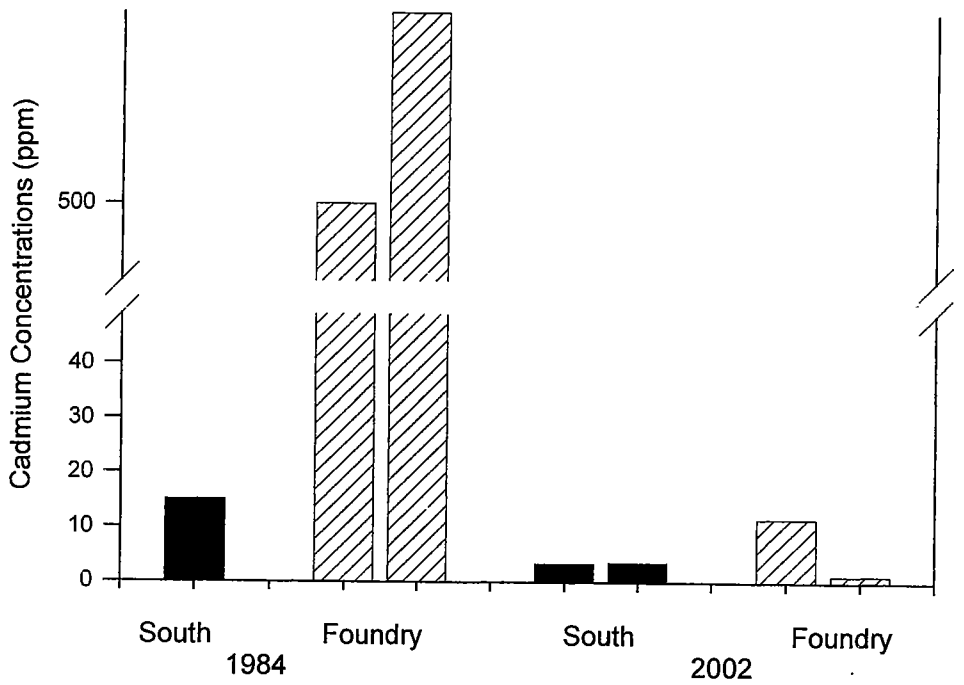
L. hoffmeisteri population densities were determined using data collected in 2001, which examined the total benthic assemblages of Foundry Cove and South Cove following the restoration (Kelaher and Levinton, in preparation). Worms were collected from Foundry Cove and South Cove sediment using a 10 cm diameter core, to a depth of 5 cm, and were washed on a 500 micron sieve to separate them from the sediment. They were then preserved in formalin, tinted with Rose Bengal, and population densities were determined. These data were compared to those collected previous to the cleanup (Klerks, 1987). Comparisons of population densities were performed using a multi-way ANOVA.

Results

Sediment concentrations of cadmium

Cadmium concentrations in Foundry Cove sediment were found to be similar to that of South Cove. In South Cove sites A and B, sediment concentrations of cadmium were 3.3 ppm and 3.5 ppm, respectively, and in Foundry Cove concentrations in sites A and B were 11.6 ppm and 1.1 ppm (Figure 6).

Figure 6. Cadmium Concentrations in Sediment Before and After the Cleanup



Resistance to cadmium.

The time to 50% survival was similar in South cove and Foundry Cove *L. hoffmeisteri*. The times to 50% survival for the Foundry Cove A and B worms were 20 and 22 hours, respectively. The times to 50% survival for the South Cove A and B worms were 19 and 21 hours, respectively. Resistance declined steadily in Foundry Cove (Figure 3, Table 1) since the cleanup (ANOVA of regression: F=31.52, p~0.0003), whereas South Cove resistance has not changed (F=0.97, p~0.36). The survival curves, when compared using a Wilcoxon test, did not differ significantly (Figure 4, Table 2).

Figure 3. Change in Time to 50% Survival 1994 to 2002.

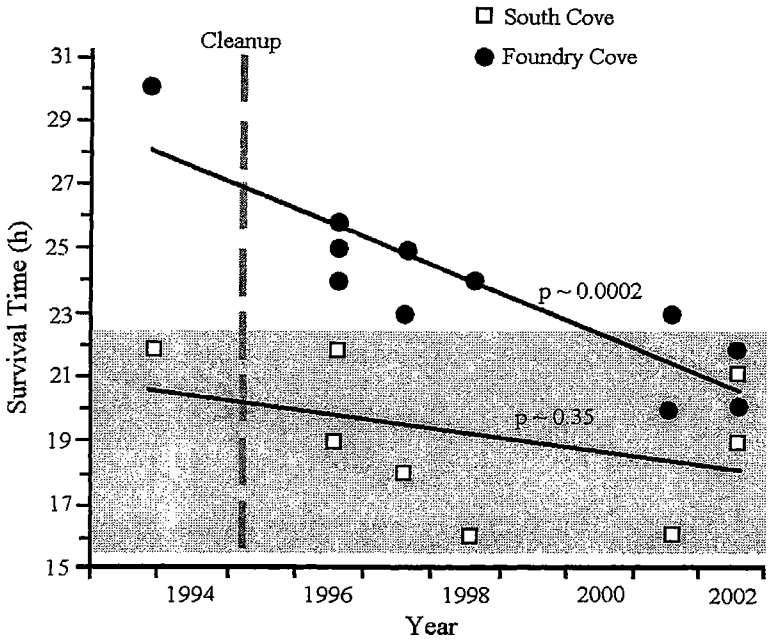


Table 1. Regression equations for change in cadmium resistance of South Cove and Foundry Cove *L. hoffmeisteri* populations, with ANOVAs of regression analyses.

South Cove: Time to 50% survival = -0.2819*year + 582.573					
	Sum of Sqs.	DF	MS	F-Ratio	*p*
Regression	**5.71**	**1**	**5.71**	**0.97**	**0.36**
Residual	**35.16**	**6**	**5.86**		
Foundry Cove: Time to 50% survival = -0.8490*year + 1720+.815					
	Sum of Sqs.	DF	MS	F-Ratio	*p*
Regression	**61.95**	**1**	**61.95**	**31.52**	**0.00033**
Residual	**17.69**	**9**	**1.97**		

Figure 4. Percent Survivors After Exposure to Cadmium.

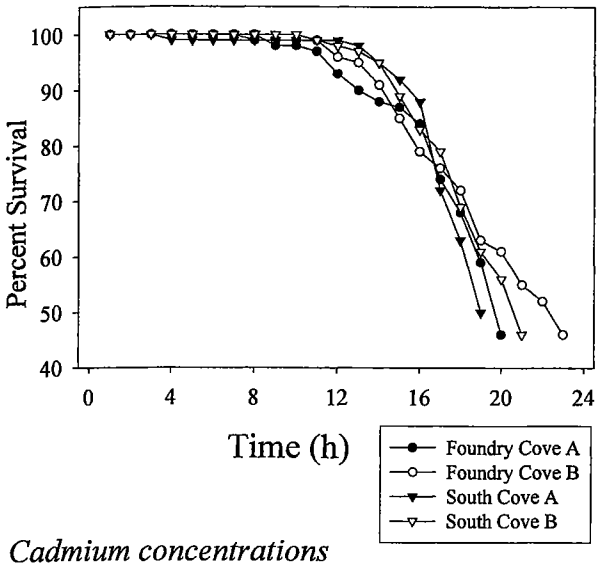


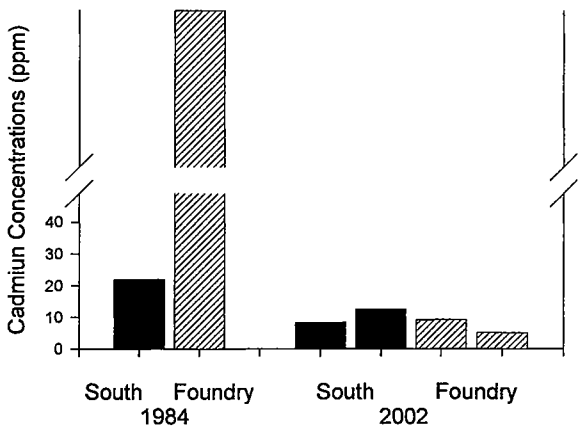
Table 2. Survival Curve Comparisons

	Test	χ^2	DF	*p*
Foundry Avs.	Log-Rank	3.46	1	0.063
	Wilcoxon	2.24	1	0.13
Foundry A	Log-Rank	0.57	1	0.45
	Wilcoxon	0.30	1	0.58
Foundry A	Log-Rank	1.83	1	0.18
	Wilcoxon	1.42	1	0.23
Foundry B	Log-Rank	1.86	1	0.17
	Wilcoxon	1.11	1	0.29
Foundry B	Log-Rank	0.91	1	0.34
	Wilcoxon	0.39	1	0.53
South A vs.	Log-Rank	1.57	1	0.21
	Wilcoxon	1.23	1	0.27

Cadmium concentrations

Cadmium concentrations in *L. hoffmeisteri* from South Cove and Foundry Cove were found to be similar. Prior to the cleanup, cadmium concentrations in Foundry Cove worms were as high as 1100 ppm, 50 times the concentration of cadmium in South Cove worms (Figure 5). Following the cleanup, cadmium concentrations in *L. hoffmeisteri* from the two Foundry Cove sites, A and B, were 9.1 and 5.0 ppm, respectively. These concentrations were similar to those found in *L. hoffmeisteri* in South Cove sites A and B, which were 8.3 and 12.4 ppm (Figure 5).

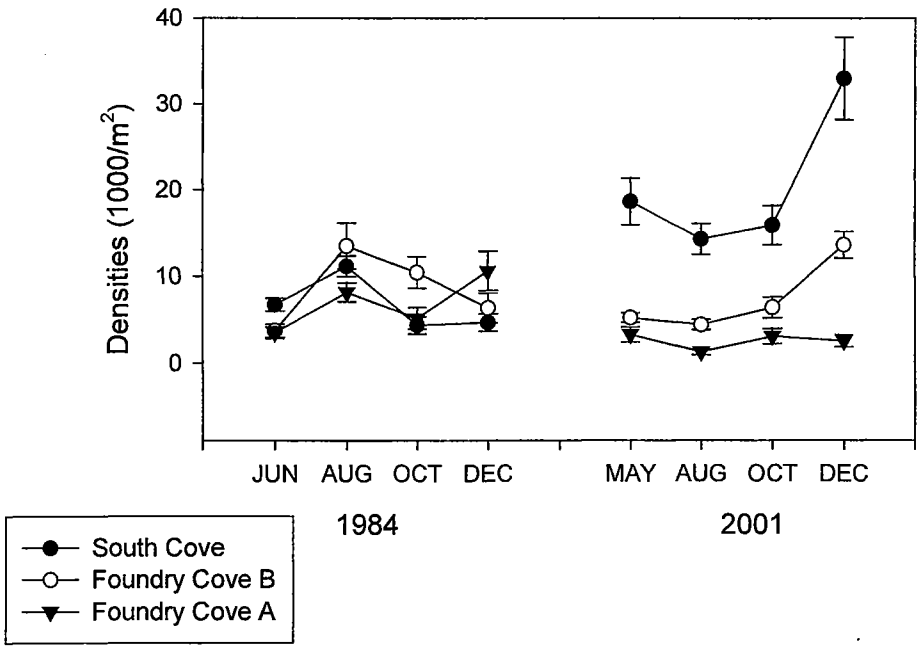
Figure 5. Cadmium Concentrations in *L. hoffmeisteri* Before and After the Cleanup.



Population densities

Whereas prior to the cleanup, population densities of *L. hoffmeisteri* in South Cove were similar to those in Foundry Cove, following the cleanup, population densities in South Cove greatly exceeded those in Foundry Cove. On average, between the year 1984 and 2001 the population densities in South Cove increased by more than 200% (Figure 2). During the same time period, population densities in the first and second Foundry Cove sites decreased by 13% and 63%, respectively (Figure 2).

Figure 2. Population Densities of *L. hoffmeisteri* in Foundry Cove and South Cove Before and After the Cleanup.



Discussion

Cadmium concentrations in sediment

The data indicate that current sediment concentrations of cadmium are drastically reduced as compared to prior to the restoration. The reduction in cadmium in the sediment was as large as 1000-fold in Foundry Cove. The EPA goal for cadmium concentrations in sediment was 10 ppm. My samples and analyses support the conclusion that the goal was reached.

Resistance to cadmium

The convergence of time to 50% survival in South Cove and Foundry Cove *L. hoffmeisteri* indicates a loss of resistance (resistance being relative to South Cove worms) in Foundry Cove worms. Whereas the time to 50% survival for South Cove worms has fluctuated randomly throughout the years following the restoration, the time to 50% survival for Foundry Cove has steadily declined (Table 1, Figure 3).

There are two possible explanations for the loss of resistance in *L. hoffmeisteri.* First, the loss of resistance to cadmium could be due to an evolutionary cost associated with the resistance. The nature and presence of this cost have yet to be established. Alternatively, the loss of resistance could be due to mixing of resistant and non-resistant phenotypes, as the removal of the selective pressure associated with cadmium may lead to the opportunity for non-resistant worms to migrate into the cove. Further studies will identify to what extent *L. hoffmeisteri* is able to disperse, in order to verify these two hypotheses.

Cadmium body burdens

The reduction of cadmium in Foundry Cove *L. hoffmeisteri* following the cleanup indicates a reduction in the uptake of cadmium. Cadmium resistant *L. hoffmeisteri* have been shown to bind cadmium, leading to increased body burdens of cadmium (Klerks and Bartholomew, 1991). Following the cleanup, however, cadmium concentrations of Foundry Cove worms were similar to those of South Cove worms. This reduction in cadmium concentrations could be a result of a loss of the mechanism of cadmium

resistance. Alternatively, the reduction in levels of cadmium in *L. hoffmeisteri* could reflect the reduced concentration of cadmium in the sediment. Cadmium concentrations in sediment are similar in South Cove and Foundry Cove, thus there is less cadmium available for accumulation. Further studies could examine the ability of current Foundry Cove worms to take up cadmium when exposed to cadmium-rich sediment.

Population densities - what was the effect of the restoration?

Before the cleanup, population densities were similar in Foundry and South Coves. However, the population densities in Foundry Cove are now significantly lower than those of South Cove. Although population densities appear to change naturally throughout the years (for example, since 1984, population densities in South Cove have increased greatly), it appears as though the high population densities found in South Cove in 2001 exemplify a trend across coves. This is supported by the fact that population densities in nearby North Cove were also relatively high in 2001. Population densities in Foundry Cove did not increase from 1984 to 2001, but decreased during this time period.

The reduction in population densities is presumably a result of the restoration process in which *L. hoffmeisteri* contained in the sediment to be dredged were removed from the cove, reducing the numbers of worms in the Foundry Cove population. Further, dredging removed much of the nutrient-rich, soft upper sediment, leaving the hard, nutrient-poor soil from underneath. The reestablishment of healthy sediment may take many years.

Conclusion

In summary, it was found that following the restoration of Foundry Cove, sediment concentrations of cadmium were reduced to near non-toxic levels. This reduction in cadmium has allowed a population of cadmium-resistant oligochaete to loose their resistance. Further, these worms no longer exhibit the high body burdens of cadmium which are associated with metal binding mechanisms. However, population densities remain low as compared to the control site, indicating that although a Darwinian Recovery has indeed occurred, the recovery of Foundry Cove following the restoration is not yet complete.

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