

Heavy Metal Presence in the Connecticut River as a Product of Socioeconomic Status

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Abstract

For decades, communities of lower socioeconomic status have been disproportionately affected by hazardous waste and other harmful environmental toxins due to phenomena like redlining and white flight. Specifically, locations of lower socioeconomic status experience greater instances of heavy metals in the environment and in drinking water. In this study, we placed samples of ribbed mussels (*Geukensia demissa*) in the Connecticut River in four locations of varying socioeconomic status based on median household income: Hartford, Middletown, Haddam, and Glastonbury, CT. Using these mussels as a bioindicator, we sought to determine whether heavy metal concentrations varied based on location socioeconomic status. After collecting samples of the mussels weekly over a five week period, we analyzed the heavy metal concentration of lead, chromium, and cadmium, as well as other metals, using a graphite furnace atomic absorption spectrometer (AAS). Using one-sample t tests, we found that lead, chromium, and cadmium were reliably present in each sample location (with the exception of lead in Haddam and Hartford). Using independent-groups t tests, we found that the concentration of lead did not differ based on socioeconomic status, but that chromium and cadmium existed in higher concentrations in locations of higher socioeconomic status. These results indicate that some factor other than socioeconomic status may be responsible for heavy metal concentration differences. Based on the trends in our data, we posit that relative location upriver/downriver may play a bigger role in determining these concentrations than socioeconomic status.

It is broadly understood that minorities and people in lower income brackets are disproportionately affected by environmental hazards, but practices beginning in the early 20th century have perpetuated the disparity between resource-rich neighborhoods and economically disadvantaged neighborhoods (Tyrrell et al, 2013). Specifically, because of practices like redlining which, decades ago, began when the Home Owners' Loan Corporation deemed neighborhoods inhabited by people of color "hazardous," the land in these neighborhoods is less expensive and therefore more easily exploited by governing bodies and corporations. Thus, these neighborhoods are exposed to hazardous waste and superfund sites that leach toxins into their environment, but are allocated fewer resources than other, more advantaged communities to combat environmental challenges.

Lead Poisoning in Flint, Michigan

Environmental racism and injustices, as described above, manifest in higher rates of exposure to toxic substances and heavy metals, such as lead. For example, in 2014, the city of Flint, Michigan was confronted with lead-poisoned water due to degrading infrastructure and pollution from industrial facilities (Pulido 2016). In 2010, 56.6% of the Flint population was African-American, and the city had a poverty rate of 41.6%. After an "emergency manager" switched the city's water supply from the Detroit River to the Flint River in an effort to save money, residents began complaining of the stench, discoloration, and bad taste from their water. Their concerns were ignored by authorities, and even after water tests revealed lead levels seven times higher than the federal limit, the tests were hidden and "business as usual" carried on. Even though city officials knew that public infrastructure was not upkept, that the water from the Flint River was literally "corrosive to car parts," and that their residents were exposed to extremely potent neurotoxins (thousands of which later were determined to have been poisoned by these

toxins), they did nothing to combat the issue (Pulido 2016). The residents of Flint, as “poor people of color” with the “least value and power,” are victims of environmental racism, leaving them vulnerable to environmental harms in spaces where they should feel safe. The story of Flint, Michigan is just one example of the consequences of environmental injustice, and it helped inspire the present research into unjust environmental heavy metal exposure.

Connecticut Socioeconomics

Despite its reputation as an overly White, wealthy state, Connecticut has a very wide income disparity. Hartford, although it is the state’s capital, is a city more economically disenfranchised than most other Connecticut communities. Its population is diverse in race and socioeconomics, with very little greenspace or river access compared to communities with more wealth. Conversely, Connecticut is home to very affluent, White communities that reside on beaches and riverbanks. These communities have easy access to local beaches on the Connecticut River, and residents often engage in river activities such as fishing and boating during the warmer months. Wealthy communities are likely allocated more resources to treat their local bodies of water, ensuring they are safe for residents to engage in recreational activities. Because of the precedent set by national environmental injustices, it is likely that lower-income communities are more exposed to pollutants, including heavy metals, in their environment (Huang & Sehgal, 2022). Lower-income areas also have far fewer policies and agencies to regulate the health and safety of the community or quality of land and water, which increases their chances of heavy metal absorption.

Biological Effects of Heavy Metals

Heavy metals are toxic for the human brain and body, causing neurological disabilities, muscular pain and seizures, and immune system dysfunction (Jaisankar et al., 2014). We focus

on lead, chromium, and cadmium, all of which can have devastating effects on the human body. Lead, for example, has been known to cause permanent brain and reproductive damage, as well as severely impact speech and neurological development in children. Chromium exposure increases the risk of lung, nasal, and sinus cancer, as well as causing severe dermatitis and liver abnormalities. Cadmium, the third heavy metal we focus on, has the tendency to replace calcium in human (and animal) bones, causing severe osteoporosis. Cadmium also causes renal and pulmonary dysfunction.

The problem lies in the ease with which heavy metals can enter the body. They can be consumed by eating or drinking, or they can be absorbed through the skin when the body comes in contact with a contaminated substance. Heavy metal contamination in any facet of the environment is detrimental to human health, and minorities are likely most susceptible to exposure because of the aforementioned instances of environmental injustices. Also, in all of our sample locations, residents were observed boating and fishing on many occasions, which are activities that would leave them vulnerable to heavy metal contamination via consumption or absorption through the skin.

***Geukensia demissa* as a Bioindicator**

Humans are not the only organisms vulnerable to heavy metal poisoning. In addition to most living things, shellfish are extremely sensitive to contaminants in their habitat, and will absorb any heavy metals (or other contaminants) that they are exposed to (Kapranov et al., 2021). Freshwater mussels, which were planted in the Connecticut River near communities of differing socioeconomic status, served as a marker to determine the concentration of heavy metals in these different communities.

Since the goal of the present research is to apply the findings to the human body, we used a living organism as a vessel to absorb heavy metals in the Connecticut River rather than solely collecting soil and water samples. We used invertebrates, and specifically shellfish, to avoid any ethical concerns with collection and analysis. We elected to use *Geukensia demissa* (ribbed mussels), because they can easily survive in both freshwater and saltwater environments. *G. demissa* are native to the East Coast of the United States, and are a eurythermal bivalve, allowing them to survive in extreme water temperatures between -7.6 to 104 degrees Fahrenheit (Reynolds, 2019). They are also photosensitive, enabling them to open and close their shells when threatened by predators or during periods of tidal change. *G. demissa*, like their bivalvia cousins, are filter feeders, which means they feed by opening their mouth and taking in water, along with any nutrients or microorganisms that might be in the water as well. They then filter out anything in the water that they do not need, such as particulate matter and bacteria, at a rate of 6.8 liters per hour.

Due to their filter-feeding mechanisms, tissue of *G. demissa* serves as an excellent marker of water quality. During the filtration process, pollutants (such as heavy metals), tend to accumulate in the mussels' tissue because they have a very low ability to metabolize them (Saleh et al., 2021). As such, the presence of heavy metals in mussel tissue would indicate the presence of those contaminants in the mussel's habitat—in this case, the locations they were planted in along the Connecticut River. We selected *G. demissa* as our bioindicator due to its hardiness and its ability to filter, and accumulate, contaminants including heavy metals.

Atomic Absorption Spectroscopy

To analyze the presence of heavy metals in the mussel tissue samples, we used an Atomic Absorption Spectrometer (AAS), an instrument that was developed in 1952 as one of the earliest

elemental analysis techniques (*Atomic Absorption Spectroscopy, How Does AAS Work, AAS FAQs*, n.d.). The AAS is based on the theory that atoms or ions of specific elements will absorb electromagnetic energy at a specific wavelength. The AAS produces this wavelength, allowing the atom to absorb the light, which then stimulates electrons in the atom to move from the ground state to the excited state (Figure 1). Once this occurs, the AAS measures the amount of light that has been absorbed, which it uses to calculate the concentration of the element in a sample. This method works for most metals on the periodic table, enabling us to efficiently test for any heavy metals we suspect may be present in the Connecticut River.

Background and Hypothesis

Minority and lower income communities are less likely to receive adequate funding for disaster mitigation and are more likely to be exposed to hazardous waste and superfund sites. These communities experience higher rates of health issues and emergency room visits, as well as shorter lifespans and lower quality of life, overall. We suspect to see these trends mimicked in the health of the reintroduced mussels. Therefore, we hypothesize that mussels planted in areas with lower socioeconomic status (Hartford and Middletown) will have greater concentrations of various heavy metals, including lead and cadmium, compared to areas with higher socioeconomic status (Haddam and Glastonbury), which will negatively affect the health of community residents.

Method

Description of the Study Area

The sampling (Baseline) site from which we originally collected our *G. demissa* specimens, is located in Madison, CT, along the southern shore of the state in New Haven County. As of 2020, there were 18,036 inhabitants in Madison, and its median household income

was \$108,231 (*Madison, Connecticut*, n.d.). Madison is home to multiple beaches and parks, including Hammonasset Beach State Park, which is the state's largest shoreline park attracting an estimated one million visitors per year (*Beach & Recreation Department*, n.d.). The majority of the population of Madison, CT is White, at 96.62% (*Madison, Connecticut*, n.d.). We collected our mussels at a local boat launch, where the salinity of the water was 19.6 parts per thousand (ppt), the pH of the water was 7.74, and the temperature of the water was 17.7°C when we first collected our specimens..

The mussels were reintroduced in four Connecticut towns: Hartford, Middletown, Haddam, and Glastonbury (in order of increasing socioeconomic status). Hartford, the state capitol, is the northernmost reintroduction location along the Connecticut River, less than thirty miles south of the Massachusetts border (Figure 2). Its population in 2022 was 120,686 inhabitants, 45.5% of which are Hispanic or Latino, 36.4% of which are Black or African American, and 27.8% of which are White (*U.S. Census Bureau QuickFacts: Hartford City, Connecticut*, n.d.). Its median household income was \$37,477 in 2021. We reintroduced the mussels at Riverside Park, where the public has access to the river for recreational or boating purposes. The salinity of the river at the time of reintroduction was 73 parts per million (ppm), the pH was 7.60, and the temperature of the water was 16.7°C.

Another site is located in Middletown, CT, which is home to Wesleyan University (Figure 2). Middletown's population in 2022 was 48,729 inhabitants, 70.7% of which are White, 15.7% of which are Black or African American, and 10.9% of which are Hispanic or Latino (*U.S. Census Bureau QuickFacts: Middletown City, Connecticut*, n.d.). The median household income in Middletown is \$67,485. At this location, we reintroduced the mussels near a local marina,

where we observed inhabitants engaging in various boating activities. The salinity of the river at the time of reintroduction was 82ppm, the pH was 7.92, and the water temperature was 19.2°C.

Our southernmost site is Haddam, CT, which is less than 30 miles from the Long Island Sound (Figure 2). Haddam's population in 2022 was 8,670 inhabitants (*Haddam Town, Lower Connecticut River Valley Planning Region, Connecticut*, n.d.). The median household income in Haddam is \$120,247, while 88.1% of the population is White, 5.6% Asian, and 3.2% Hispanic or Latino (*Haddam Demographics - Get Current Census Data for Haddam, CT*, n.d.). We reintroduced the mussels at Eagle Landing State Park, from where river cruises often embark and where we observed many local residents fishing and boating. At the time of reintroduction, the salinity of the river at this site was 77ppm, the pH was 7.55, and the water temperature was 19.6°C.

The last reintroduction location was Glastonbury, CT, which is our site of highest socioeconomic status (Figure 2). The population in 2022 was 35,061 inhabitants, 79% of which are White, 9% of which are Asian, 2% of which are Black or African American, and 8% of which are Hispanic or Latino. Glastonbury's median household income is \$130,294 (*Glastonbury Town, Hartford County, CT - Profile Data*, n.d.). We introduced the mussels at Riverfront Park, which is home to the Crew Boat Memorial. We observed many people fishing and boating around the boathouse. The salinity of the water at the time of reintroduction was 73ppm, the pH was 7.68, and the water temperature was 17.0°C.

Sampling and Chemical Analysis

Field Collection

From Madison, CT, we collected roughly 120 ribbed mussels, *Geukensia demissa*, and distributed them in mesh bags along the Connecticut River in Middletown, Haddam,

Glastonbury, and Hartford, CT. After distributing the mussels at each of the four collection locations (Week 0) along the Connecticut River in late May, 2023, we returned to each location weekly for six weeks. Upon each site visit, we collected a sample of five live mussels, when possible, and when not possible, we collected all live mussels and the remaining deceased specimens. We also collected information on water salinity, temperature, and pH using a handheld probe monitor *in situ*. After each sample collection, we immediately exposed the sample to dry ice to flash freeze the tissue, then stored the tissue in a freezer. During Week 3, after mass mussel casualties at the Middletown and Haddam sites during Week 2, we replaced the mussels at all four locations with a newly collected batch of roughly 20 mollusks, and continued with the aforementioned collection procedures for the remaining three weeks. On Weeks 0, 2, and 6, we also took a water sample from each site, (except for our Haddam location, from which we took the water sample on Weeks 0, 2, and 5). Additionally, on the same Weeks, except for Week 0, we took a soil sample from each site. Water and soil samples were exposed to dry ice and frozen according to the same procedures as the mussel tissue, itself.

Instances of Mass Death. During Week 2, mass death events were observed at two sites: Middletown and Haddam. In both of these instances, between 60%-70% of our reintroduced population had died. Their remains were found in an open-shelled state, and in some cases, with individual mussel tissue having been completely washed away. The same events were observed during Week 3 in Glastonbury and Hartford, and again during Week 5 in Middletown and Glastonbury, with mortality rates of over 90%. For each mass death event, we collected the remains of the dead mussels. After separating from the survivors, they were frozen with dry ice and stored in a freezer until analysis.

Laboratory Analysis

Following the six weeks of sample collection, we transported the samples to the Hamilton College analytical chemistry laboratory to assume sample analysis procedures. After partially thawing the samples, we shucked the mollusks by removing the soft tissue from the shell; in some cases, we were able to pry the shells open, in others, we cut the adductor muscle with a plastic spoon, and in yet others, we smashed open the shells with a rubber mallet. The soft tissue of the samples was dried at 60°C until completely dry and brittle, then ground samples as an aggregate from each Week/Location into a fine, homogenous powder with a coffee grinder. To digest the samples, approximately 100mg of dried mollusk was combined with 1.5mL 70% nitric acid and boiled in a 70°C water bath until the sample was completely dissolved. Then, we added 2.5mL 30% hydrogen peroxide to the mixture and allowed it to boil in a 70°C water bath for two hours. We then loaded roughly 1mL of each sample into the graphite furnace AAS for heavy metal analysis.

To prepare the soil samples for AAS analysis, we thawed the samples and dried them at 60°C until completely dry. We then ground them with a ceramic mortar and pestle to break apart any clumps and sifted the samples through a 0.8mm sieve to remove any larger particles and debris. To digest the soil samples, we combined 1g of soil with 2mL of water, then sonicated them for approximately 5 minutes to ensure that samples were fully hydrated. Then, we added 2mL of 30% nitric acid to the samples and heated the samples in an 80°C water bath for roughly 15 minutes. After bubbling ceased, we removed the samples from the water bath and added 0.5mL of 30% nitric acid, 0.5mL of 30% hydrogen peroxide, and 0.1mL 38% hydrochloric acid, then filtered the samples through filter paper using a vacuum to pull the samples through filter paper. The samples were then diluted with water until the total volume of each sample was equal

to 10mL. We then loaded approximately 1mL of the fully digested and filtered soil samples into the AAS for heavy metal analysis.

We prepared the water samples by simply thawing them, then loading roughly 1mL of each sample into the AAS for heavy metal analysis.

Data Analysis

To determine which heavy metals were present in our samples and showed trends of interest, we ran an exploratory qualitative analysis of many different heavy metals—including lead, chromium, cadmium, copper, arsenic, nickel, molybdenum, manganese, aluminum, and zinc—present in the alive samples taken from Collection Week 1. Based on this analysis, we decided to perform a more in depth qualitative analysis on lead, chromium, cadmium, and copper on all of our alive, dead, soil, and water samples.

Using Microsoft Excel (2023), we computed descriptive statistics for lead, chromium, and cadmium concentrations from each collection location. To determine whether the concentration of these heavy metals was statistically significant, we conducted one-sample t tests for each location for lead, chromium, and cadmium. Additionally, to determine if there was a reliable difference in heavy metal concentration (the dependent variable) as location socioeconomic status varied, we conducted independent-samples t tests for lead, chromium, and cadmium in the locations of high socioeconomic status (Haddam and Glastonbury) compared to the locations of low socioeconomic status (Hartford and Middletown). If our data support our hypothesis, the concentration of lead, chromium, and cadmium should be significantly greater in the locations with a low socioeconomic status compared to the locations with a high socioeconomic status.

Results

In this study, we sought to examine the relationship between socioeconomic status and heavy metal concentrations in the Connecticut River, using ribbed mussels (*Geukensia demissa*) as a bioindicator. After relocating the mussels from their native habitat on the Connecticut shoreline to four locations of varying socioeconomic status along the Connecticut River—Haddam, Middletown, Glastonbury, and Hartford—we collected samples of mussels from each location for five weeks. We then analyzed the samples' lead, chromium, and cadmium concentrations (by way of peak absorbance area values) as a product of location, socioeconomic status, and time using AAS analysis.

Pb, Cr, and Cd Concentration

One-sample t tests were conducted to determine whether lead concentrations in the mussel tissue from each location were significantly greater than zero. The mean lead peak absorbance area values for Middletown ($M = 0.03$, $SD = 0.05$) and Glastonbury ($M = 0.02$, $SD = 0.02$) were statistically significantly greater than zero, $t(8) = 1.93$, $p = .045$; $t(5) = 2.78$, $p = .020$, respectively (Figure 3). The mean lead peak absorbance area value for Hartford ($M = 0.008$, $SD = 0.01$) was not significantly greater than zero, but it did approach significance, $t(6) = 1.78$, $p = .063$. The mean lead peak absorbance values for Haddam ($M = 0.04$, $SD = 0.08$) and our Baseline location ($M = 0.02$, $SD = 0.04$) were not statistically significant, $t(5) = 1.27$, $p = .129$; $t(3) = 1.02$, $p = .192$, respectively.

Additionally, one-sample t tests were conducted to determine whether chromium concentrations in the mussel tissue from each location were significantly greater than zero. The mean chromium peak absorbance area values for Middletown ($M = 0.32$, $SD = 0.11$), Glastonbury ($M = 0.50$, $SD = 0.08$), Hartford ($M = 0.41$, $SD = 0.23$), Haddam ($M = 0.44$, $SD =$

0.09), and the Baseline location ($M = 0.18$, $SD = 0.05$) were all significantly greater than zero, $t(7) = 8.03$, $p < .001$; $t(5) = 15.09$, $p < .001$; $t(6) = 4.64$, $p = .002$; $t(5) = 12.20$, $p < .001$; $t(3) = 6.83$, $p = .003$, respectively (Figure 4).

Furthermore, one-sample t tests were conducted to determine whether cadmium concentrations in the mussel tissue from each location were significantly greater than zero. The mean cadmium peak absorbance values for Middletown ($M = 0.15$, $SD = 0.05$), Glastonbury ($M = 0.24$, $SD = 0.04$), Hartford ($M = 0.19$, $SD = 0.03$), Haddam ($M = 0.17$, $SD = 0.02$), and the Baseline location ($M = 0.19$, $SD = 0.08$) were all significantly greater than zero, $t(8) = 9.61$, $p < .001$; $t(5) = 16.89$, $p < .001$; $t(6) = 17.96$, $p < .001$; $t(5) = 17.54$, $p < .001$; $t(3) = 4.83$, $p = .008$, respectively (Figure 5).

Pb, Cr, and Cd Concentration and Socioeconomic Status

To assess the relationship between socioeconomic status and heavy metal concentrations, we performed independent-groups t tests (Figure 6). Specifically, an independent-groups t test compared whether the mussel tissue from locations of high socioeconomic status (Haddam and Glastonbury) and the locations of low socioeconomic status (Hartford and Middletown) differed in overall lead peak absorption area values. The test was not statistically significant, $t(15) = 0.74$, $p = 0.236$, indicating that mussel tissue from locations of high socioeconomic status ($M = 0.04$, $SD = 0.06$) and locations of low socioeconomic status ($M = 0.02$, $SD = 0.04$) did not differ in overall lead concentrations.

Another independent-groups t test compared whether mussel tissue from locations of high and low socioeconomic status differed in overall chromium peak absorption area values. The test was statistically significant, $t(22) = 1.92$, $p = .034$, indicating that mussel tissue from

locations of high socioeconomic status had significantly greater levels of chromium ($M = 0.47$, $SD = 0.10$) compared to locations of low socioeconomic status ($M = 0.36$, $SD = 0.18$).

A final independent-groups t test compared whether mussel tissue from locations of high and low socioeconomic status differed in overall cadmium peak absorption area values. The test was statistically significant, $t(18) = 2.34$, $p = .031$, indicating that mussel tissue from locations of high socioeconomic status had significantly greater levels of cadmium ($M = 0.21$, $SD = 0.05$) compared to locations of low socioeconomic status ($M = 0.17$, $SD = 0.05$).

Dead Sample Analysis

After collecting a plethora of dead samples and observing the differences in raw peak absorption area values between heavy metals of living versus dead mussel tissue, we decided to run a series of *post-hoc* independent-groups t tests. Specifically, an independent-groups t test compared whether mussel tissue that was living upon sample collection and tissue from specimens that were dead when sampled differed in overall lead peak absorption area values. The test was statistically significant, $t(18) = -9.67$, $p < .001$, indicating that living mussel tissue had significantly lower levels of lead ($M = 0.03$, $SD = 0.05$) compared to dead mussel tissue ($M = 0.22$, $SD = 0.06$).

Another independent-groups t test compared whether living and dead mussel tissue differed in overall chromium peak absorption area values. The test was statistically significant, $t(15) = -6.84$, $p < .001$, indicating that living mussel tissue had significantly lower levels of chromium ($M = 0.37$, $SD = 0.17$) compared to dead mussel tissue ($M = 0.98$, $SD = 0.30$).

A final independent-groups t test compared whether living and dead mussel tissue differed in overall cadmium peak absorption area values. The test was statistically significant,

$t(28) = 2.24, p = .017$, indicating that living mussel tissue had significantly greater levels of cadmium ($M = 0.19, SD = 0.06$) compared to dead mussel tissue ($M = 0.15, SD = 0.05$).

Soil and Water Samples

We also performed AAS analysis on our soil and water samples and found that the soil samples had relatively high peak absorption area values for all metals compared to that of the living tissue, with lead values ranging from 0.08 to 0.77, chromium values ranging from 0.86 to 2.03, and cadmium values ranging from 0.16 to 0.61. Alternatively, water samples had relatively low peak absorption area values for all metals compared to that of the living tissue, with lead values ranging from 0.00024 to 0.0054, chromium values ranging from 0.0038 to 0.011, and cadmium values ranging from 0.00 to 0.39.

Copper and Other Metals

In addition to lead, chromium, and cadmium, we also ran a preliminary qualitative investigation of several other metals, including copper, arsenic, nickel, molybdenum, manganese, aluminum, and zinc. Analysis by AAS revealed that the peak absorption area values for copper were completely saturated for all sample locations. This result remained true even after a ten-fold sample dilution. Alternatively, the other six metals had extremely low peak absorption area values.

Discussion

Pb, Cr, and Cd Concentration

The purpose of this study was to investigate any possible disparities in heavy metal presence in the Connecticut River between locations of high and low socioeconomic status—using ribbed mussels as a bioindicator—with a hypothesis that towns of lower socioeconomic status would have higher concentrations of heavy metals. We first found that

average lead concentrations of mussels collected throughout the collection period were significantly greater than zero in Middletown (low socioeconomic status) and Glastonbury (high socioeconomic status; Figure 3). Additionally, Hartford (low socioeconomic status) approached significance, whereas Haddam (high socioeconomic status) and the Baseline location did not. We suggest that the small sample size for Haddam (4, compared to 8 for Middletown) resulted in a lack of statistical power, and thus the failure of statistical significance despite its apparent relatively high concentration. Qualitatively, Haddam and Middletown, where the mussels experienced the earliest mass death events at Week 2, appear to have some of the highest lead concentrations. Conversely, Hartford and Glastonbury, where the mussels survived beyond Week 2, have some of the most consistently low concentrations of lead.

We also found that chromium concentrations were significantly greater than zero at all sampling locations and the Baseline location (Figure 4). Of all the metals we investigated, chromium appeared to have the most variability over time and across locations, with Hartford's chromium peak absorption area value jumping from 0.14 at Week 1 to 0.71 at Week 2. Specifically, Hartford and Middletown (both low socioeconomic status) showed an overall increasing trend of chromium concentrations over time, whereas Haddam and Glastonbury (both high socioeconomic status) displayed a slight downward, mostly constant chromium concentration trend. In addition to variability, chromium peak absorption area values were relatively greater than both lead and cadmium values, with Hartford and Glastonbury having the highest chromium concentrations. Given that these two locations are where the mussels survived beyond Week 2, it is surprising that they have the highest relative chromium concentrations.

Additionally, we found that cadmium concentrations were significantly greater than zero at all sampling locations and the Baseline location (Figure 5). Of all the metals we investigated,

cadmium appeared to remain the most constant over time and across location, with concentrations at the sampling locations almost mirroring those at the Baseline location. The lack of apparent correlation between cadmium concentration and the mass death events given the overall consistency in values indicates that cadmium was likely not a decisive factor in the overall health of the mussels.

Pb, Cr, and Cd Concentration and Socioeconomic Status

We next explicitly investigated our hypothesis about the relationship between socioeconomic status and heavy metal concentrations (Figure 6). Specifically, we found that mussel tissue from locations of high socioeconomic status (Haddam and Glastonbury) did not significantly differ in lead peak absorption area values from locations of low socioeconomic status (Hartford and Middletown). This lack of statistical significance indicates that socioeconomic status does not play a role in the levels of lead present in the Connecticut River.

Alternatively, we found that both chromium and cadmium peak absorption area values were significantly greater in the locations of high socioeconomic status (Haddam and Glastonbury) than in the locations of low socioeconomic status (Hartford and Middletown). This finding is contrary to our hypothesis that areas of low socioeconomic status would have higher concentrations of heavy metals. While we cannot be certain based on our current data, we theorize that this finding may be because of relative location along the river rather than socioeconomic status. For example, Hartford (low socioeconomic status), which was our most upriver location, had some of the lowest heavy metal concentrations, whereas Haddam (high socioeconomic status), which was our most downriver location, had some of the highest heavy metal concentrations. Since our sampling locations, from upriver to downriver, go Hartford (low socioeconomic status), Glastonbury (high socioeconomic status), Middletown (low

socioeconomic status, and Haddam (high socioeconomic status), locations of varying socioeconomic status are intermixed, with the towns of higher socioeconomic status being generally more downriver than towns of lower socioeconomic status (Figure 2). While heavy metals tend to sink and accumulate in the riverbed soil, some metals may be adsorbed to particulate matter suspended in the river water and thereby carried downriver until eventually deposited in the riverbed (Cui et al., 2019). As such, it is plausible that heavy metal contaminants from sources including the landfill located in Hartford, CT and Wesleyan University located in Middletown, CT, traveled downriver and were detected in the mussel tissue of more downriver locations, thereby conflating any potential relationship with socioeconomic status.

Soil, Water, and Dead Sample Analysis

Our analysis of the dead tissue samples revealed significant differences in lead, chromium, and cadmium between mussels that were alive versus those that were dead upon collection (Figure 7). Specifically, lead and chromium concentrations in the dead mussels were significantly greater than those of living mussels, with dead samples collected from the same week having up to three times greater lead peak absorption area values and up to twice as great chromium peak absorption area values than living samples. Alternatively, cadmium peak absorption area values were significantly greater in the living tissue than the dead tissue, though comparatively, these values were relatively similar. Interestingly, analysis of the dead samples and the soil samples revealed similar values for all metals, indicating that once mussels die, they begin to act as a similar contaminant sink as riverbed soil (Cui et al., 2019).

Conversely, the water samples revealed extremely low peak absorption area values compared to living, dead, and soil samples, indicating that the water itself is a much more diluted source for heavy metal contaminants. This finding makes sense considering that heavy metals

often tend to be pulled by gravity toward the riverbed, and only some particles remain in the water (Cui et al., 2019).

Copper and Other Metals

The oversaturation readings for copper from the AAS indicate relatively high concentrations of copper at all locations. Unfortunately, these saturated readings do not provide specific information on concentration trends across locations. However, these extremely high copper peak absorbance area values (compared to the other heavy metals tested) are of interest because of copper's toxic effect on marine invertebrates (MacKenzie, 1961). A discussion of copper's potential role in the mass death events witnessed during this study can be found in the appropriate section below.

Conversely, the lack of any detectable concentrations of arsenic, nickel, molybdenum, manganese, and aluminum is also interesting to note. These results indicate that either these metals were not present in our samples at all, or that they were present, but in concentrations that were far too low for the AAS to detect. However, given the AAS's extremely low limits of detection (0.05ppb for arsenic, 0.07ppb for nickel, 0.05ppb for molybdenum, 0.005ppb for manganese, 0.1ppb for aluminum, and 0.02ppb for zinc), these metals would have to exist in very low concentrations. Thus, these metals were not of particular interest to this study.

Mass Death Events

Instances of mass death were observed during Weeks 2, 3 and 5. Over the course of the sampling period, at least one mass death event was observed at every reintroduction site, with mortality rates ranging from 60% to 100%. No correlation between mortality rate and socioeconomic status was observed. These instances of mass death were unexpected, considering our research into *G. demissa* as a resilient species of bivalvia. To rule out shock after

reintroduction as a potential cause of death, we reinvestigated *G. demissa*'s preferential living conditions. The mussels were transferred from a high salinity location to a freshwater environment, so we theorized that the dramatic shift in salinity may have been too drastic a difference in the mussel's environment. However, we confirmed that *G. demissa* is able to survive even in the most extreme temperatures and salinities (Leinbach, 2020). Furthermore, our research confirmed that *G. demissa* is in fact found thriving in freshwater lakes, streams, and riverbeds (Reynolds, 2019). After thorough investigation, we were able to rule out complications with reintroduction as a possible cause of death. We instead propose three alternative theories: drought, effects from wildfires in Canada, and copper poisoning.

Theory 1: Drought

Over the course of the sampling period, the State of Connecticut slid further and further into an abnormally dry season, according to the United States Geological Survey (USGS). Starting May 30th, 2023 (Week 1 of sampling), approximately half of Middlesex County was recorded as “abnormally dry” (*Current Map | U.S. Drought Monitor*, n.d.). Both Middletown and Haddam were located in this drought area, and they experienced mass death events the following week. We noticed the same trend for the mass death events during Week 5. The two weeks prior, nearly the entire state was categorized as abnormally dry by USGS (*Current Map | U.S. Drought Monitor*, n.d.). This would explain the mass deaths in Middletown and Glastonbury during Week 5. However, this theory does not explain the mass death events during Week 3, when mass death was observed in Hartford and Glastonbury. Neither of these sites are located in the area categorized as “abnormally dry” during the weeks prior (*Current Map | U.S. Drought Monitor*, n.d.). Drought has been shown to impact the abundance of freshwater mussels, specifically *Corbicula fluminea* (Haag & Warren, Jr., 2008). *C. fluminea* was especially affected by low rates

of dissolved oxygen in the water, which is characteristic of drought areas. Low rates of dissolved oxygen are caused by low stream flow, warmer water temperatures, and high oxygen demand, all of which are exaggerated during periods of drought. Considering this information, we believe that it is possible that the incidences of mass death were caused, at least in part, by the state's abnormally dry conditions.

Theory 2: Wildfires

Beginning in March, 2023, wildfires began burning throughout much of Quebec and Nova Scotia, Canada, and are still burning as of late July, 2023. Since June, the fires have significantly grown in intensity. Southwesterly winds blew smoke from these fires to much of the northeastern United States, including Connecticut. The fires began to grow in intensity between sampling Week 1 and Week 2, and therefore just before the first instance of mass mussel death. The air quality in Connecticut, as well as much of New England quickly plummeted, reaching the classification of “unhealthy” by June 6th, 2023 (*Connecticut Air Quality*, n.d.). Public service announcements regarding the dangerous air quality were put out by the state, so we theorized that the contaminants in the air caused by the Canadian wildfires may be dangerous to the mussels. Research showed that wildfires are often correlated with excess concentrations of nitrate, nitrite, and ammonia, mostly due to excess sediment runoff (*Wildfire Impacts on Surface Water Quality*, n.d.). High concentrations of any of these compounds are fatal to many aquatic organisms, including mussels. With this in mind, we tested the river water for these three compounds during the weeks of mass death. At every reintroduction site, the concentrations of nitrate, nitrite and ammonia were all safely within normal limits. With no evidence to suggest that the incidences of mass death were caused by the Canadian wildfires, we ruled that the correlation was likely due to coincidental timing.

Theory 3: Copper Poisoning

Exposure to toxic levels of copper in the river water was our third and final theory to explain the mass death events that occurred with the samples during our collection period. In a notable 1961 experiment, copper, and specifically copper sulfate solutions between 2.5% and 5.0%, were shown to cause nearly 100% mortality of the experimental mussels (MacKenzie). More recently, this information has been used to guide marine invertebrate restoration efforts. Specifically, in 2017, water authorities in Lewisville, Texas began using copper ion generators to inject copper into the local waterways with the intention of killing invasive zebra mussels (Perez, 2019). Given copper's toxic effects on sensitive marine invertebrates, these water treatments have proven successful in removing unwanted zebra mussels.

While the subjects of this research were ribbed mussels rather than zebra mussels, copper's toxic effects extend to many invertebrate species, so it is more than likely that high concentrations of copper are toxic to ribbed mussels (Yanong, 2010). As previously stated, our samples displayed relatively very high levels of copper, and thus chronic copper poisoning could plausibly have been a factor in these mass death events. However, given the saturated AAS readings, we cannot say definitively how much copper each sample was exposed to, nor make any causal claims about copper's role in these mass death events. Despite this lack of clarity, we are most confident in this theoretical explanation of the mass death events we encountered with this field research.

Conclusion

The data we collected does not support our hypothesis that locations of low socioeconomic status would have higher concentrations of heavy metals. However, we found that various heavy metals, including lead, chromium, copper, and cadmium, are present in the

Connecticut River both in areas of high and low socioeconomic status. The results of this study warrant further investigation, specifically into quantifications of copper in the Connecticut River. Since the peak absorption area values for copper were completely saturated for all sample locations, we were not able to record specific quantities of copper. Running a quantitative analysis of the data would help to determine the causes of the mass death events as well as determine if the levels of lead, chromium, copper and cadmium in the Connecticut River exist in a safe concentration to humans.

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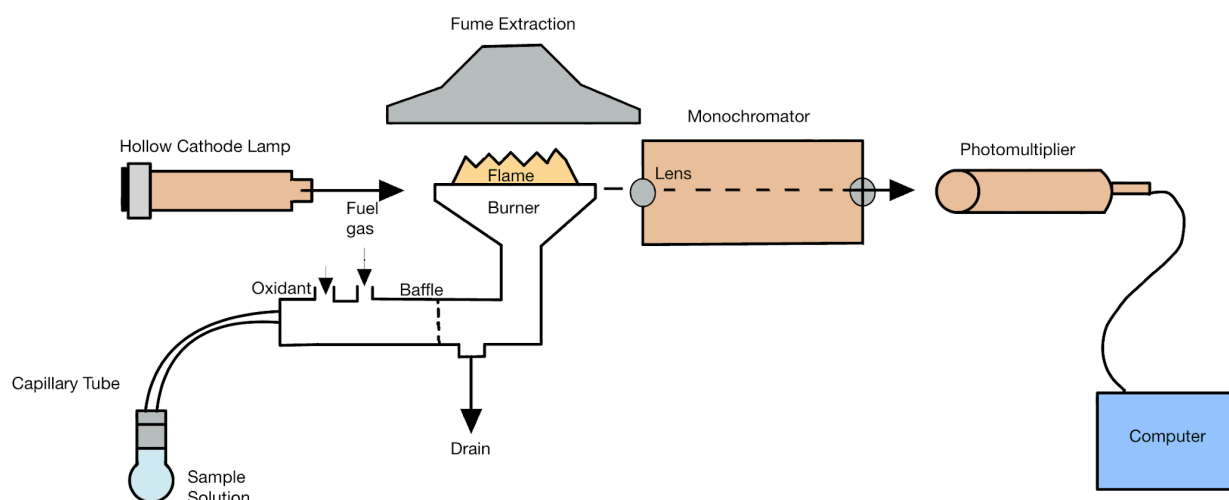
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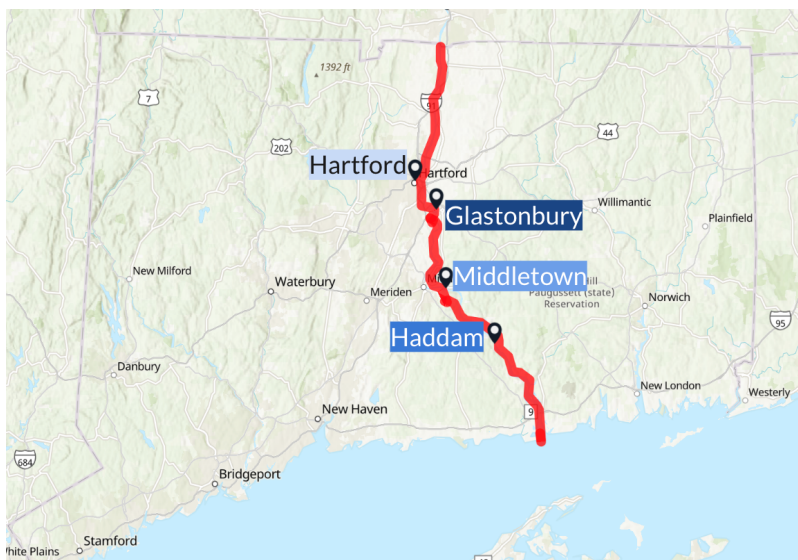
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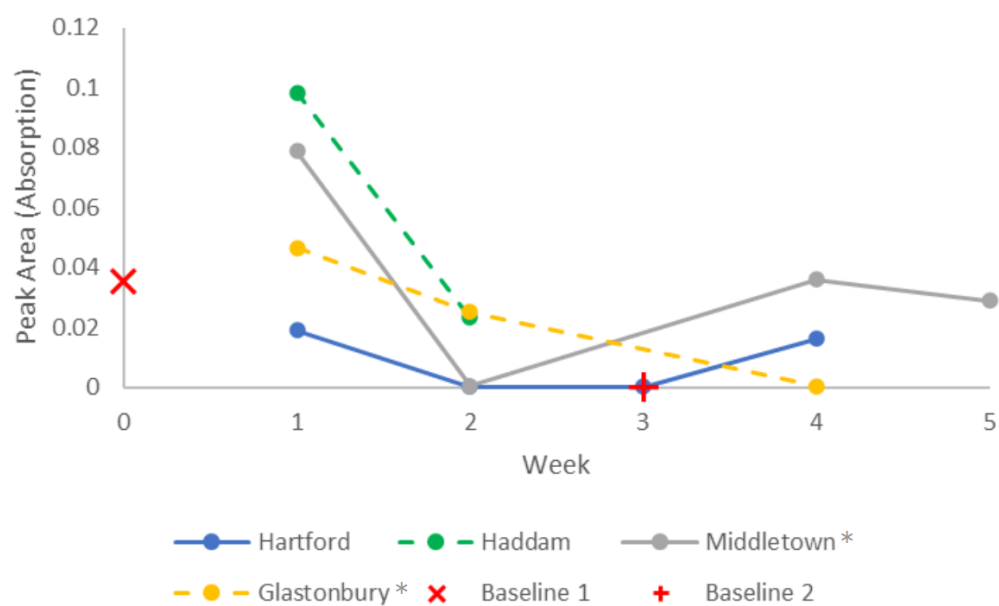
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Figure 1*Atomic Absorption Spectroscopy*

Note. Schematic diagram of a working graphite furnace atomic absorption spectrometer.

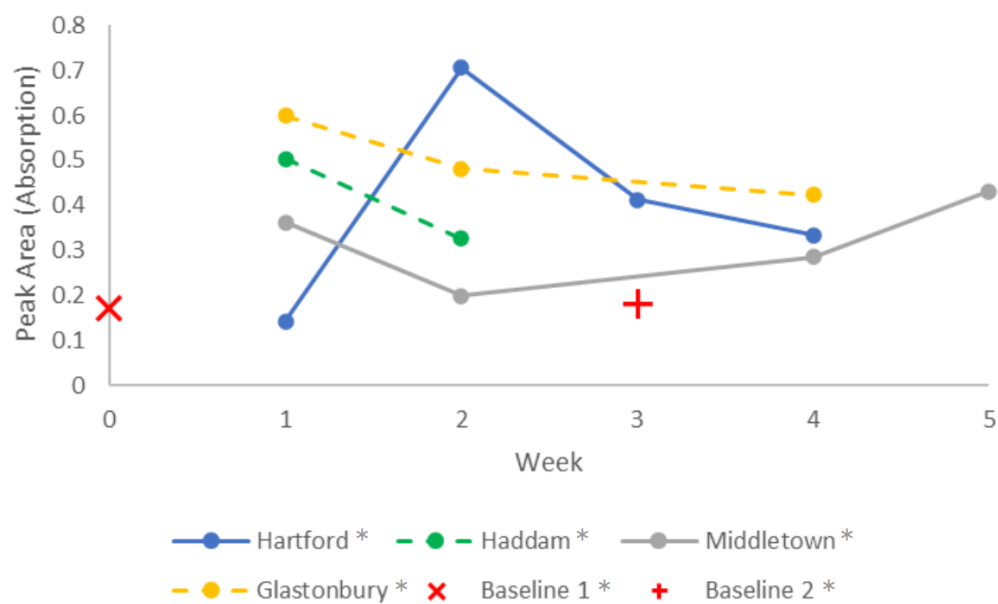
Figure 2*Site Map*

Note. Map of the four reintroduction sites and their orientation along the Connecticut River. The darker the blue highlight, the higher the median household income of that location.

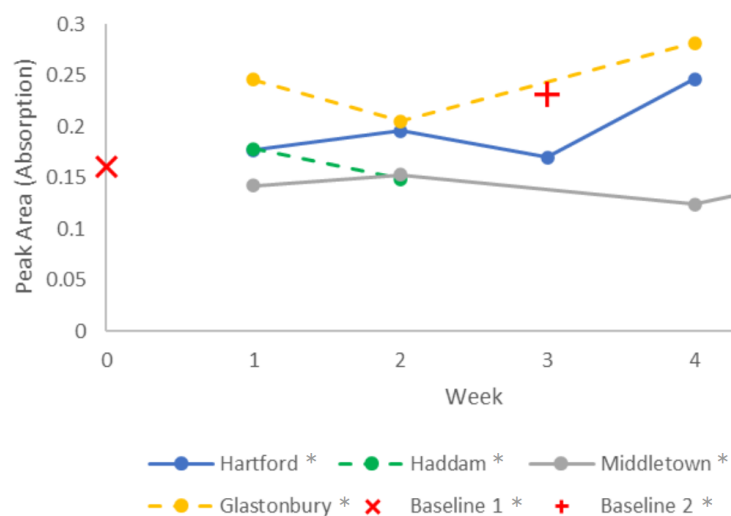
Figure 3*Peak Area vs. Week (Pb)*

Note. Lead peak absorption area values over time at each of the four collection locations

(Hartford, Hadam, Middletown, and Glastonbury), as well as the Baseline location. $*p < .05$.

Figure 4*Peak Area vs. Week (Cr)*

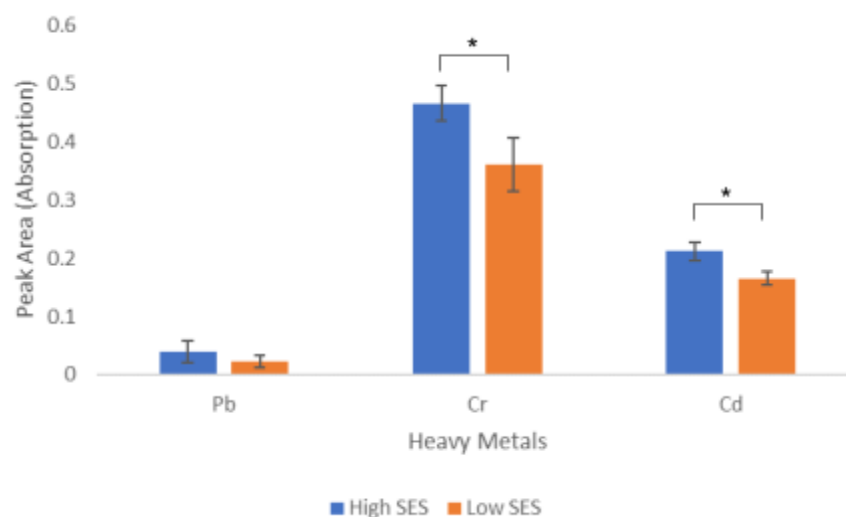
Note. Chromium peak absorption area values over time at each of the four collection locations (Hartford, Hadam, Middletown, and Glastonbury), as well as the Baseline location. $*p < .05$.

Figure 5*Peak Area vs. Week (Cd)*

Note. Cadmium peak absorption area values over time at each of the four collection locations (Hartford, Hadam, Middletown, and Glastonbury), as well as the Baseline location. $*p < .05$.

Figure 6

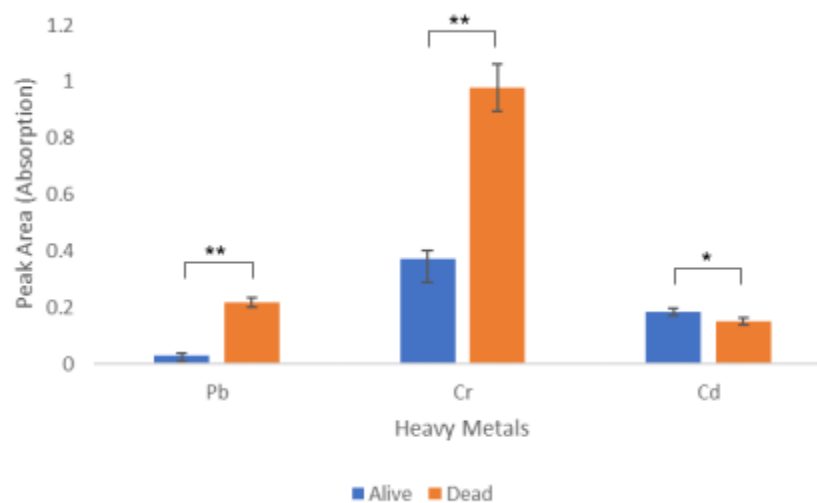
Peak Absorption in Locations of High vs. Low Socioeconomic Status (SES)



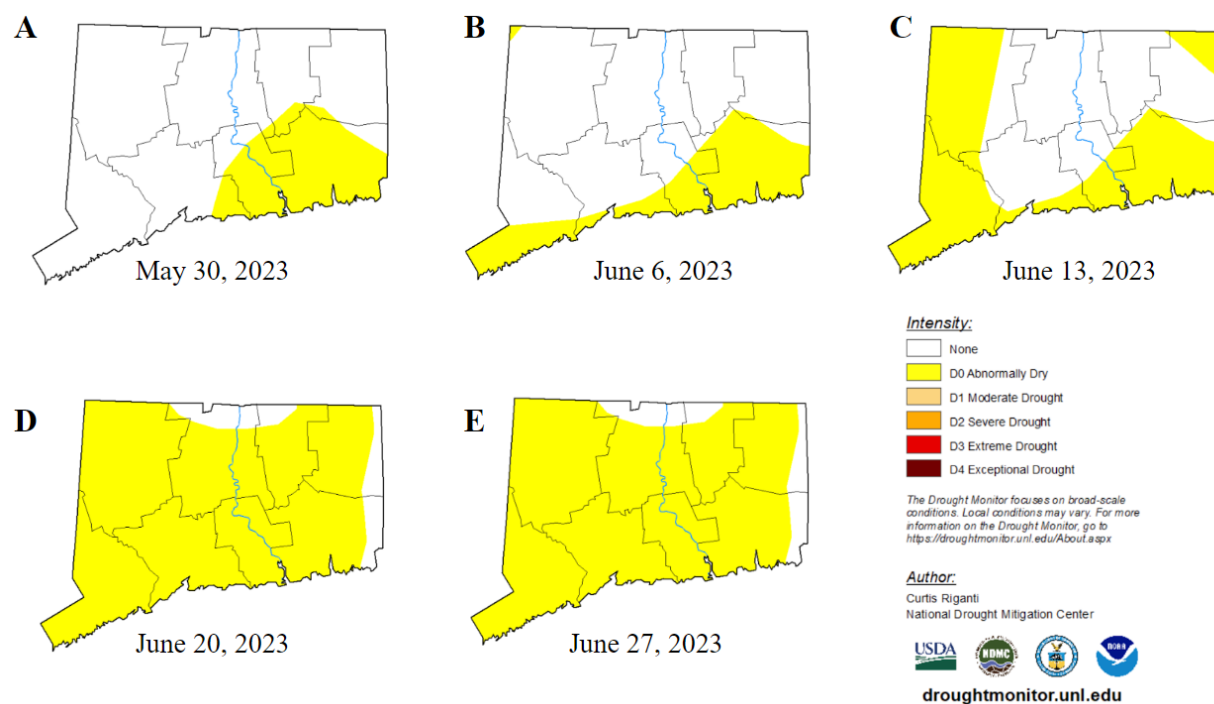
Note. Comparison of mean peak absorption area values for lead, chromium, and cadmium between locations of low and high socioeconomic status. Error bars represent the standard error of the mean. * $p < .05$.

Figure 7

Peak Absorption in Dead and Living Tissue



Note. Comparison of mean peak absorption area values for lead, chromium, and cadmium between mussels that were collected while they were either living or dead. Error bars represent the standard error of the mean. * $p < .05$, ** $p < .001$.

Figure 8*Drought in Connecticut in June, 2023*

Note. United States Geological Survey (USGS) map depicting areas of drought in Connecticut between May 30, 2023 and June 27, 2023.