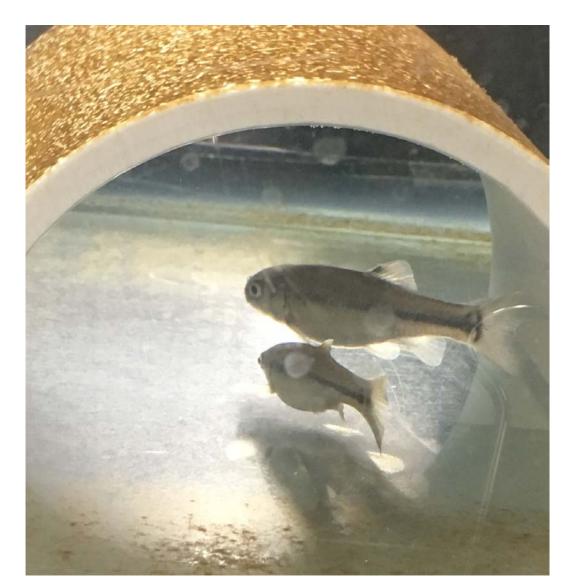
# Effect of Lead Nitrate on Fathead Minnow Reproductive Behavior

### Abstract

The purpose of our experiment was to test the effects of lead nitrate on fathead minnow reproductive behavior. Four tanks were used for the experiment, and each contained one type of fish: a control with no lead and no DMSA, lead exposed and no DMSA, lead exposed with the DMSA, and a control with no lead and given DMSA. Males with lead in their bodies exhibited less reproductive and more aggressive behaviors along with significantly less secondary sex characteristics. As the issue of lead-contaminated water becomes a spotlight issue due to cases such as in Flint, there are increasing concerns about the exact effects of lead on development and biochemical processes in humans.

### Introduction

Lead is a gray metal with many desirable qualities such as electrical conductivity, malleability, density and lowcorrosivity, and it has been mined for centuries [1]. Children absorb more lead than adults and, consequently, are more susceptible to the effects of such exposure, particularly with symptoms like cognitive impairment [2]. Low-level exposure to adult males may result in low sperm count and impotence [3]. Annually, the costs of childhood lead exposure are estimated around \$50 billion in the US alone [4]; however, for each dollar spent mitigating lead poisoning in housing alone, society's benefit is \$17 to as much as \$220 [5]. Studies conducted in the United States linked childhood lead exposure to a higher levels of conduct disorder, delinquency, and crime [6, 7, 8]. DMSA is a drug that chelates with lead, nickel, arsenic, iron, mercury, and cadmium [9], and it is used for treatment of lead toxicity in children [10]. DMSA can not penetrate the blood-brain barrier, so removal of lead from the nervous system occurs because of the lead concentration between the brain and blood [9]. The aim of our research is to study the effects of chronic lead poisoning on the reproductive behavior of male fathead minnows. We tested the hypothesis that fathead minnows exposed to lead would result in a significant decrease in the number of their reproductive behaviors and an increase in aggressive chasing behaviors.



### Methods & Materials

There were 4 different tanks that each had a male and a female minnow in it. Minnows in the lead tanks were exposed to 1 ppm of lead nitrate for two weeks. The DMSA tanks were exposed to 20mL/day of 10mM DMSA in 0.1M NaOH in 20 gallons. We documented the amount of times the male performed a behavior, and the times they were viewed. Then we calculated the SEM.

## Results

In the blind study, male fathead minnows in four different tanks were observed for two and a half weeks for five minute observation intervals. Two control tanks (one with no exposure and one with only DMSA) were compared to the experimental tanks (one with lead exposure and one with lead and DMSA exposure). The independent variables were the exposure to lead and the treatment of DMSA. The dependent variables were the presence of male secondary sex characteristics and behaviors. For the secondary sex characteristics all of the tanks included fish with an observed dorsal fin spot. The head pad and tubercles were viewed 20% more often in the control and control DMSA tanks than the other two tanks. Side bars were only visible in the control DMSA exposed group. The average number of male behaviors (nest preps, hovers, patrols, chases, spawns) were observed and calculated. Nest prepping and hovering averages were significantly higher in the control tanks. Patrolling and chasing male behaviors were significantly greater in tanks that were exposed to lead and lead with the DMSA treatment. Spawning was only viewed in the DMSA control tank; however, eggs were spotted in the control tank, indicating that spawning occured outside of the observation periods. Also, spawning likely did not occur in lead exposed and lead with the DMSA treatment tanks due to the absence of eggs.

The fish treated with lead and DMSA exhibited the most chasing behaviors.

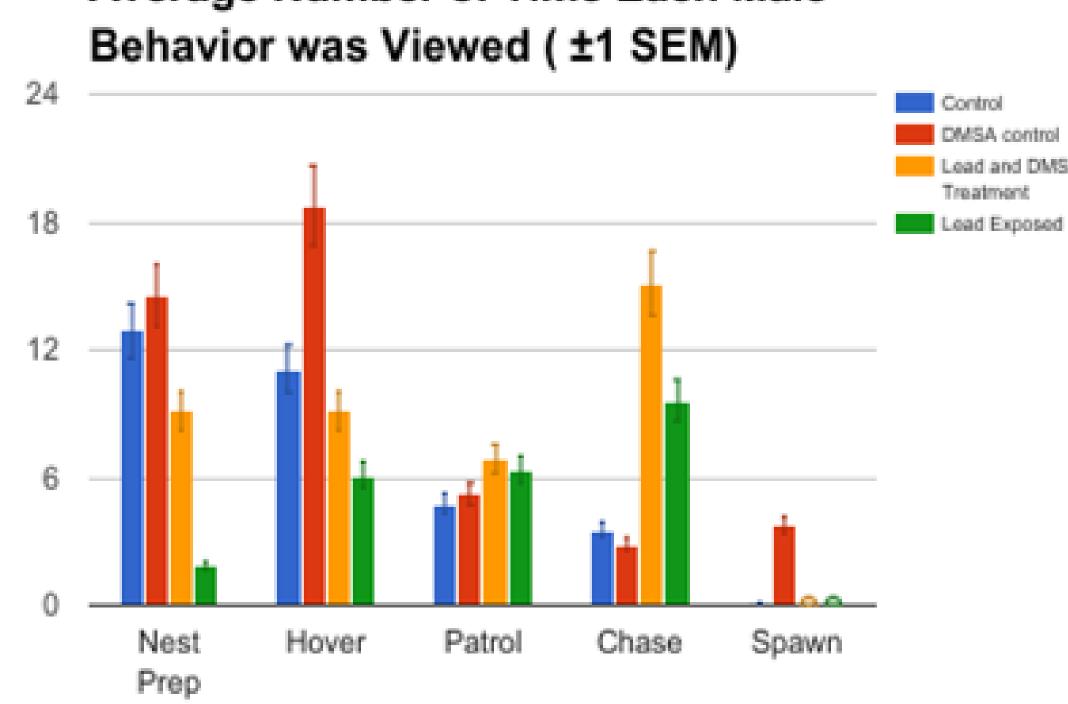
### By: Alexa Schnittka

1. Four tanks were set up: a control, one with

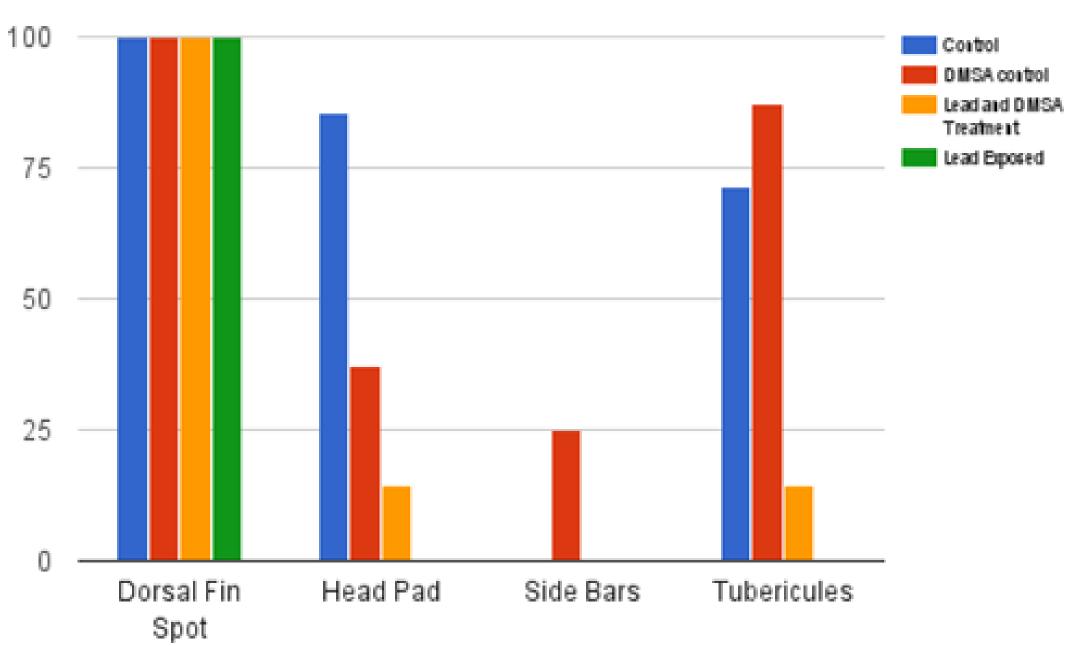
DMSA(control), one with lead and DMSA, and one with just lead. The tanks containing lead, were exposed to this element 2 weeks prior to arriving to our class. 2. Each day, we were assigned tanks to observe their behaviors: hovering, spawning, nest prep, chase, and

patrol. We referenced a module that helped us see which behaviors were being performed.

3. We observed and recorded the male's behaviors for 6 days for 2 weeks with five minute intervals.



#### Presence of Male Secondary Sex Characteristics Seen



Male Secondary Sex Characteristics



# Average Number of Time Each Male

Male Beahvior

In the DMSA control tank, the minnows are spawning.

### Discussion

In our experiment, the lead fish had a reduced number of reproductive behaviors, which supports our hypothesis. The fish struggled to reproduce or swim due to the accumulation of lead in their bones. The results of the study can be extrapolated to humans, since humans and minnows have similar biochemical processes and anatomical structures. Furthermore, our results match current research about the effects of lead on human populations including lethargy and reduced fertility. In addition, the lead exposed fish were far more aggressive than the control fish who were not exposed, which supports our hypothesis that the lead will cause the fathead minnows to become more aggressive.

There were many limitations in our experiment. One limitation encountered was the small sample size. There were only two tanks exposed to lead and one of which also had the DMSA to combat the poisoning. Each tank had six fish that were separated into three different groups. If there was a larger sample size in a more natural habitat, that could cause variations in our result. Another restriction was the limited time spent observing the minnows. We only watched the fathead minnows during five minute intervals during the class period. It could be possible that the minnows acted differently when we were not watching. Finally, there could be varying interpretations of certain behaviors, especially for hovers. A hover involves the fish floating to the upper half of the PVC pipe, but the observer has to interpret whether or not the fish actually moved above the imaginary middle line: sometimes deciding whether a behavior was a hover or not is just a judgement call.



In the lead exposed tank the male fathead minnow lacks a head pad, tubercles, and side bars.

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