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Article

A Quantitative Analysis of the Proliferation of Microplastics in Williamston's Waterways

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SUMMARY

One of the most concerning aspects of human progress is the spread of pollution. Microplastic pollution is only a small part of this issue, but a relevant one nevertheless. Plastic debris can disrupt marine ecosystems, spread contaminants, and take years to naturally degrade. Our aim for this study was to establish an understanding of the scope of Williamston, Michigan's microplastics problem, as well as to attempt to find the source of these plastics. We sampled four sites from the Red Cedar River in the Williamston School District. Sites were chosen due to their proximity in relation to the boundary of the school district, with samples collected both upstream and downstream of the wastewater treatment plant. In analyzing our samples, we used an aspirator vacuum to filter the water we collected, left the filters in an incubator to dry for 48 hours, and then counted microplastics under a microscope by systematically scanning through gridded filter paper. We found a general trend of increasing concentrations of microplastics from upstream to downstream, but we were not able to locate the source of Williamston's microplastics pollution. Originally, we hypothesized that the Williamston Wastewater Treatment Plant was the primary contributor to Williamston's microplastics pollution, but we could not find statistically sufficient evidence to confirm this theory. Further research is needed to determine whether the Wastewater Treatment Plant or another source is responsible for the microplastics pollution.

INTRODUCTION

Currently, 8 million tons of plastic waste enters the marine ecosystem each year, much of which is in the form of tiny microplastics, which are plastic particles smaller than 5mm (1). If these plastics are allowed to build up at their current rate, without any reduction, an ecological disaster may be inevitable. Even now we may have already reached a tipping point. The United Nations estimates that there are 51 trillion microplastics in the ocean (2). Microplastics are incredibly harmful to the environment (3). When found in high concentrations, they can spread contaminants, endanger wildlife, and damage marine ecosystems. In April 2018, a dead sperm whale discovered by Spanish researchers was found to contain 64 pounds of plastics in its digestive system (2). Plastic is a substance that is made to last, and as a result, microplastic particles can take decades to degrade, allowing for their buildup. This fact further emphasizes the necessity for action, as the global microplastic threat is frequently overlooked (4). A major reason for this could be the "out of sight, out of mind" mentality that many people subscribe to, seeing as most people do not come face-to-face with extensive buildup of microplastics on a daily basis.

The purpose of this study was to gain an understanding of Williamston's microplastics accumulation and the extent of the problem in the Red Cedar River. We focused on two aspects of this issue: the origin of Williamston's microplastics and their concentration. If we have detailed knowledge of the concentration of microplastics, it will allow us to gauge the scope of Williamston's microplastics problem and more importantly its potential impact. In addition, knowing the origin of microplastics will aid in containing the issue at hand.

In this situation, there were clear independent and dependent variables. The independent variable was the location that our team decided to collect our water samples. These sample locations were located near notable sites on the Red Cedar that fall within the Williamston School District's boundaries (**Figure 1**). The dependent variable, or the focus of our observations, was the concentration of microplastics in our samples. We assumed that the number of microplastics per liter that we found would be dependent upon where we sampled.

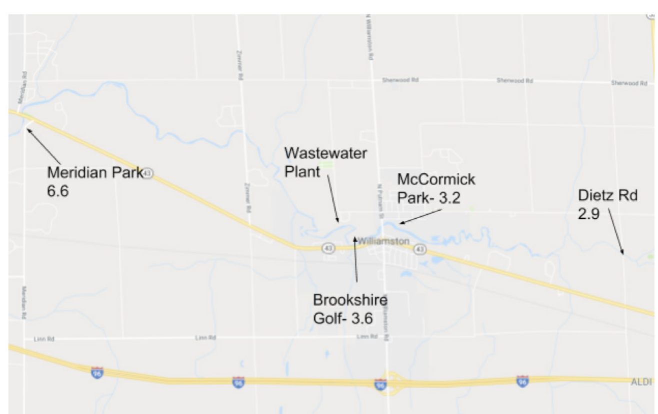

Figure 1. Map of testing sites. All values are concentration, in microplastics per liter, at each site. We did not test the water directly outside of the wastewater plant, but included it to show where it is in correlation to the other sites.

The results of our study were difficult to predict, but based on other studies, we expected relatively low microplastic levels. For example, Baldwin et al. (2016) found as little as 0.002 particles per liter in rivers in the Great Lakes Basin (5). However, Castañeda et al. (2014) found over 87 particles per liter (6). Based on this disparity between the studies, it was hard to predict the amount of microplastics that we would find. However, our location is relatively rural and therefore similar to the area analyzed by the first study, which led us to hypothesize that Williamston would have a low concentration of microplastics, somewhere in the range of 0-10 particles per liter. We also hypothesized that the Williamston Wastewater Treatment Plant would be the primary contributor of microplastics into the river. This hypothesis was derived from research that occurred at the University of Leeds in the United Kingdom that linked wastewater treatment plants to microplastics pollution (7).

According to the US Geological Survey, microplastics can generally be categorized into five main types: fibers, beads, films, foams, and fragments (8). A fiber is a long strand of microplastic that is found in synthetic clothing and cigarette butts. A bead is a small pebble-like microplastic that is found in toothpaste and shampoo. A film is a small fragment of wrappers and plastic bags. A foam comes from styrofoam. Fragments are small pieces of plastic that have been chipped off plastic litter such as water bottles. We hypothesized that the majority of the plastics we found would be fibers. This is because a 2016 study of the Great Lakes found that around 70% of microplastics were fibers (8).

RESULTS

The foundation of this study was built on the hypothesis that we would see increases in microplastic levels as we looked further downstream, if we discovered any at all. Starting at 2.9 microplastics per liter by the Dietz Road Bridge (n=3, SE=1.1) the number of microplastics per liter increased steadily, peaking at Meridian Park (6.6 microplastics per liter, n=2, SE 5.8), the furthest site that we sampled downstream. At the other sites that we sampled, we found 3.2 microplastics per liter at McCormick Park (n=3, SE=0.512) and 3.6 microplastics per liter at Brookshire Golf Course (n=3, SE=0.864) (**Figure 2**).

One of our overarching research questions was "Does Williamston add microplastics pollution to the Red Cedar?" Based on our data it seems as though Williamston does add a substantial amount of plastics to the Red Cedar, but according to the Mann Whitney tests we performed, that does not seem to be the case. We found that the microplastics concentration was not significantly increased in the river as it passed through Williamston because our calculated U Value was 1.

Another aspect of our research dealt with the individual types of microplastics. In our samples from the river, we found fibers (91%), fragments (7%), beads (1%), foams (0.5%), and films (0.5%) (**Figure 3**). We also tracked the types of plastics that we found from the wastewater plant, where we found fibers (90%), fragments (6%), foams (2%), films (2%), and zero beads (**Figure 4**). Given this similarity in concentrations of the microplastic type, we decided to conduct a statistical analysis to see if the wastewater plant was the main emitter of microplastics into the Red Cedar River.

As previously mentioned, one of our initial hypotheses was that the Williamston Wastewater Treatment Plant would be the main reason for the increase in microplastics throughout Williamston. With the aid of Dr. Kurt Guter, an expert in water research, we used a dilution equation in order to test this. After calculating the dilution by taking into account the average stream flow of the Red Cedar River and the average daily effluent of the wastewater treatment plant, we got a dilution factor of 127.4. This means that for every increase of 1 microplastic per liter downriver of the wastewater treatment plant, there should be 127.4 microplastics per liter in our wastewater samples if the wastewater treatment plant is the sole contributor of plastics. This poses a challenge to our initial hypothesis because we found only 6 microplastics per liter (n=4, SE=0.38) in the wastewater samples (**Figure 4**).

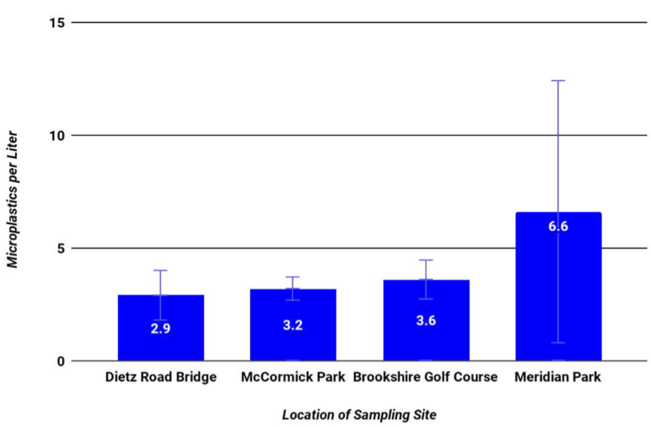

Figure 2. Microplastics concentrations in the Red Cedar: upstream to downstream. Column values are mean concentration of microplastics. Error bars are standard error.

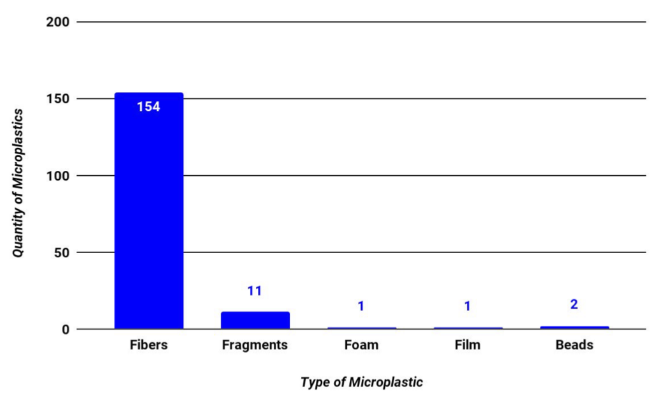

Figure 3. Total microplastics by type. Columns are totals of each microplastic found across all four river testing sites. These represent the total amount across all of our testing days and are not an average.

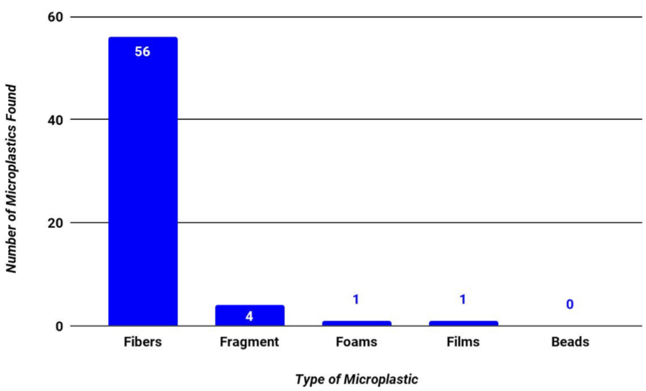

Figure 4. Wastewater microplastic totals by type. The columns represent the total amount of each microplastic found at the Williamston Wastewater Treatment Plant. These columns represent total amount across all of our wastewater samples and do not represent averages.

A final aspect of our research pertained to microplastics in drinking fountain water at Williamston High School. Midway through our testing period, Williamston experienced a 100-year flood that prevented us from sampling from the Red Cedar River. We used this time to sample water from the high school drinking fountains. We found plastics in all four drinking fountains that we sampled. We found our highest concentration of plastics at the 20-year-old D Hallway Fountain, where there were 4.4 plastics per liter (SE=1.54). The lowest concentration of plastics that we found was at the 20-year-old Lunch Room Fountain, where we found 1.8 plastics per liter (SE=1.476). This seems to be a very large difference, but it was not statistically significant because the ranges of the respective standard errors overlap (**Figure 5**).

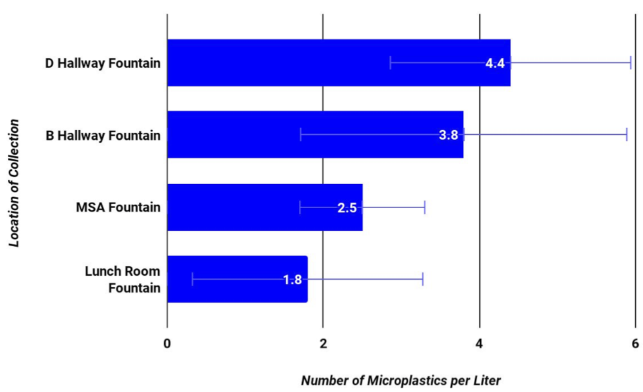

Figure 5. Microplastics in Williamston High School Drinking Fountains. The column values are means and the error bars show standard error.

DISCUSSION

Initially, most of our research questions were quantitative, concerning the concentration of microplastics in the Red Cedar within the Williamston School District. We found that throughout the river, levels of plastic debris increased from upstream to downstream. From our results, we see an increase in microplastics concentrations between the Brookshire Golf Course and Meridian Park. However, a word of caution must be added to this conclusion. We did not have a large enough sample size to indicate significant results and there could be other factors at play that would cause an increase. Our results indicate an increase, but more testing must be done in order to achieve more conclusive evidence.

An important secondary aspect of our project concerned the source of these plastics. As suspected, the Williamston Wastewater Treatment Plant fits the criteria for a major source of microplastics in the Red Cedar. It lies between our testing sites of Brookshire and Meridian where we saw the most significant increase in pollution (**Figure 1**). Our hypothesis was that we would see extremely high levels of plastics in the outflow of the plant. However, according to our dilution calculation in the results, since the wastewater treatment plant is emitting only 6 plastics per liter, and not 127, it is not contributing nearly enough microplastics to the Red Cedar to fully explain the increase in microplastics from Brookshire to Meridian Park. Therefore, we concluded that the wastewater treatment plant may be contributing microplastics into the Red Cedar, but it is more likely that there are other sources contributing more plastics. A plausible explanation is that many of the microplastics are coming from the individual homes along the Red Cedar River in between Brookshire and Meridian Park. Each use of a washing machine can create around 700,000 microplastic fibers (9). We hypothesize that since many of the homes between Brookshire and Meridian Park do not utilize a city sewer system, instead relying upon septic tanks, there may be infiltration into the Red Cedar from those septic tanks causing the greater values. This is merely speculation and more testing would be required in order to confirm this hypothesis. What we do know is that the levels of microplastics, and more specifically fibers, are higher at Meridian Park than they are at the Brookshire site and that these plastics are coming from somewhere. Further testing is required to determine a definitive source.

An important component of our research dealt with the types of microplastics. We tracked the number of each type because we felt this might help us locate the source of the plastics and we wanted to compare our numbers to those found by other studies. In our research, we found that 91% of our plastics were fibers. This elevated level of fibers was similar to another analysis of microplastics in the Great Lakes Basin, in which the US Geological Survey found that in Great Lakes Tributaries, 70% of collected microplastics were fibers (8). Another noteworthy result that the categorization of microplastics revealed was the near absence of beads. Only 1% of our collected plastics were beads. We find this interesting since there was a nationwide ban on the use of microbeads that was implemented in 2016 (8).

If it is indeed true that Williamston is adding microplastics to the Red Cedar River, the future may be concerning. Currently, the EPA's range of regulation in regard to microplastics has been minimal, meaning that if the Williamston Wastewater

Treatment Plant is a source of a majority of the Red Cedar's microplastics, it has no economic incentive to attempt to reduce its output. Another worrying aspect of this issue is the prospect of public health concerns resulting from the proliferation of plastic fibers in areas of commercial fishing, in this case the Great Lakes. If fish are consuming these microplastics, it is only a matter of time until the microplastics work their way up the food chain (10).

Another aspect of our research dealt with microplastics in drinking water supplies. It was interesting to note that in our samples, we found no correlation between age of a drinking fountain and the concentration of microplastics. This suggests that the plastics are coming from other places such as the pipes leading to the fountains, not the fountains themselves. Again, since we only took two samples, more research is needed in order to establish the veracity of these results. What is alarming about our result is how it is mirrored in other studies of US drinking water. A recent study that tested sites such as the EPA Headquarters and Trump Tower found that 94% of US drinking water samples contained microplastics (11). In this study, the researchers found an average of 9.6 plastics per liter in the drinking water, which was higher than our average of 3.14 plastics per liter. Despite no research to date being conducted on the effects of microplastic consumption on humans, it is definitely something that should be avoided. Even bottled water, which is considered by many to be safer than tap water, has been found to contain microplastics. A study of US bottled water found that 90% of bottles contained microplastics (11).

While our research was conducted to the best of our available resources, there is room for improvement in terms of methodology and design. One issue is that we collected our samples in plastic buckets. While we ran blanks to ensure that there was no exfoliation, we would recommend that future samples be collected in glass jars. Another area that could be improved relates to the filters. We used the best paper filters available to us and checked 50% of them before use for microplastics, only finding plastics on 2 occasions. However, we would recommend individually wrapped cellulose membrane filters in the future since these are the filters used by Dr. McNeish at Loyola University. Our final recommendation would be to collect more samples. We were not able to obtain statistically significant evidence due to our small sample size. We feel that such an issue could be averted with a larger sample size.

Additionally, our study faced certain limitations. The applications of our findings are limited to gaining an understanding of the microplastics problem in our community. Williamston's microplastics levels will not be representative of the microplastics levels in other areas due to certain factors such as population and industry. Additionally, our team sampled a relatively narrow section of the Red Cedar, seeing as we sampled within the Williamston school district, and only sampled four spots. Therefore, our results are only applicable to the portion of the Red Cedar River as it passes through Williamston.

In the interest of time and due to limited materials, we decided to confine our research to four sites in the Williamston area. This did not in any way hurt our overall research goal; in fact, these guidelines helped our research by providing a clear focus upon microplastics in Williamston. Another boundary of limitation of our research was that we only sampled water. We did this in the interest of time, since sediment separation (another method of sampling) takes far longer to analyze than bulk water separation. Another limitation of our research is time. We sampled in a 5-month window, and had we been able to sample over a longer span of time, we would be able to have a better idea of the average microplastics concentration in the Red Cedar River.

Overall, we had some surprising results and some expected results. The quantity of microplastics that we saw was expected since it fell within the 0-10 microplastics per liter range that we hypothesized that we would see. However, we were surprised that the data we collected suggested that we were incorrect in our assumption that the wastewater treatment plant would be the primary emitter of microplastics into the Red Cedar. Since we know the wastewater plant is not the source of the pollution, and using this information we could develop another hypothesis that states that septic systems and washing machines are the primary polluters of microplastics into the river. Our findings pose an interesting challenge to our community, both in regards to how to deal with the pollution, but also how to find the definitive source and how to end the pollution.

MATERIALS AND METHODS

1. Site Selection

In our study, we analyzed the levels of microplastic concentration in Williamston's waterways. We primarily tested the Red Cedar River, but we also tested the final effluent from the Williamston Wastewater Treatment Plant. We tested the microplastics levels by collecting bulk water samples from the Red Cedar. The bulk water was collected by submerging a five-gallon bucket into the river. We filled the buckets with approximately 2.5 gallons of river water of each sample.

We collected samples from four sites along the Red Cedar. These sites included the location where the Red Cedar enters the Williamston School District at Dietz Road, McCormick Park, the 12th hole at Brookshire Golf Course, and the location where it leaves the school district at Meridian Road. These locations were chosen to show how much the Williamston area as a whole is contributing to the microplastics problem. At each location, we sampled in the exact same spot in order to maintain consistent results. We also sampled each site two to four times (November 2017- April 2018) in order to have a more representative set of data.

We also took samples from the Williamston Wastewater Treatment Plant. The manner in which we sampled from the wastewater treatment plant was slightly different than the manner in which we sampled from the river. We collected a two-gallon sample from a composite 24-hour sample from the wastewater treatment plant. We used a composite sample of the final effluent because we wanted an accurate representation of how many microplastics the plant was emitting, and if we used a sample from a specific point in time, it would be subject to variation in the quantity of microplastics at that given time.

We also sampled from one off-river site; this was the Williamston High School building. While scanning for our literature review, we came upon an article that stated that over 90% of tap water samples in the United States contained microplastics. We tested this in our high school by collecting two separate 1 liter samples from four drinking fountains throughout the school. We then ran them through the same process as the river samples.

2. Sample Processing

We used a systematic process to gather data from our samples. Once we collected the bulk water samples, we brought them to the lab in order to filter them. We used gridded filter paper to filter the water from the microplastics. We attempted to control the infiltration of microplastics by examining 50% of the filters for potentially misleading particles prior to using them. We then used an aspirator vacuum in order to suck the water through the filter. The filtered water was placed into a beaker to determine the exact quantity of water that passed through the filter. Once the sample was filtered, we placed the filter paper into an incubator for 48 hours at 60°C to dehydrate the sample and remove all the water from the filter, thus making it easier to analyze under a microscope. After a 48-hour period, we put the filter into a petri dish and scanned for microplastics under 10x magnification (12). The grid on the filter paper helped us to scan the sample systematically. After both group members agreed on the number of microplastics on a given filter, we organized the data by splitting the different particles into categories based on the type of microplastic.

There were a few materials that we needed in order to accurately complete our research. The primary materials that we needed were an aspirator vacuum, paper filters, an incubator, a dissecting microscope, and glass beakers in a variety of sizes. We were provided with a materials list from Dr. Rachel McNeish (an expert in microplastics at the University of Loyola Chicago) and found that no other tools were required.

3. Statistical Analyses

Finally, after we conducted all of our sample processing, we performed a variety of statistical analyses, namely dilution equations and Mann-Whitney U Tests. We used a one-tailed Mann Whitney test because this test does not make assumptions about standard deviation and is more applicable to our research findings. To determine if there was statistical evidence that Williamston adds microplastics, we used a null hypothesis that the concentration of microplastics at Dietz Road would be the same as at Meridian Park. Our alternate hypothesis was that the concentration at Meridian Park would be higher. We also conducted a dilution calculation when we were evaluating our hypothesis that the Williamston Wastewater Treatment Plant was the source of Williamston's microplastics pollution.

ACKNOWLEDGEMENTS

In order to complete our research, we needed some help from outside sources. Dr. Kurt Guter provided us access to the wastewater treatment plant's final effluent. He was also instrumental in focusing the scope of our research. Under his guidance, we were able to overcome several obstacles, including minimizing the risk of allowing unwanted microplastics to get onto our filters. We also consulted Dr. Rachel McNeish from Loyola University whose input we relied upon throughout the early stages of the research process. She provided us with our methodology, and we couldn't have conducted our research without her.

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