Black Spot Disease on Fish Species in the French Creek Watershed

By

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Table of Contents

Figures and Tables.....3

Abstract.....4

Introduction.....5

Methods.....10

Data Analysis.....13

Results.....14

Discussion.....18

Acknowledgements.....23

References.....24

Figures and Tables

(Figure 1: Life Cycle of parasite *Uvulifer ambloplitis*)

(Figure 2: Major Tributaries and Subwatersheds of French Creek)

(Figure 3: Fish with Black Spot Disease)

(Figure 4: BSD and Water Chemistry Data)

(Figure 5: Abundance and Severity of BSD per Location)

(Figure 6: Abundance of BSD compared to water temperature)

(Figure 7: Abundance of BSD compared to pH of the stream)

(Figure 8: Abundance of BSD compared to TDS in the streams)

Abstract:

Freshwater habitats like French Creek are home to rich biodiversity and various species of plants and animals. In today's day and age human activity has caused degradation to freshwater ecosystems. These negative externalities that are caused by humans are being experienced first hand by these fish and plant species. The objective of this study is to assess the relationship between stream health and Black Spot Disease parasite loads in fish. Fish were sampled at various sites along French Creek on 3 dates in the fall of 2022. Areas of the stream were electroshocked, fish were collected, and data gathered. Location, species name, and severity of BSD found on each fish was recorded. Additionally, water chemistry data including water temperature, pH and total dissolved solids (TDS) was collected. Data was analyzed to determine if water chemistry metrics were related to the abundance of BSD. It was determined that water temperature, pH, and TDS did not impact the abundance of BSD found on fish species. Future studies should look at snail populations around the same stream locations to see if they are carrying BSD and if so, do any water chemistry metrics affect their abundance of BSD. This in all could draw a correlation between human environmental degradation and disease in French Creek watershed.

Introduction:

Globally, in our world today freshwater habitat health is declining and we need to begin to stress the importance of studying these ecosystems in order to improve them (Poff, 2009). Freshwater habitats such as French Creek are home to various species of animals and plant life that help support rich biodiversity (McAllister, 1997). French Creek provides benefits to humans in many forms. For example, it gives people access to fresh water and food, along with many leisure and recreational activities (Bowden & Pallant, 2004). French Creek meanders through New York and Pennsylvania, and is located near many residential areas which makes it very accessible for human interaction and unfortunately degradation (Pennsylvania Rivers Conservancy, 2022). Humans have influenced our environments since early times and with increased technologies and industrialization, have caused climate change (Hegerl et al, 2019). Naturally freshwater environments contain natural systems like buffers in which water quality and health are provided through natural systems. (Carroll & Thornburgh, 1997). Overtime human interaction has caused these freshwater environments to be the host of habitat degradation, fisheries overexploitation, pollution, and alien species introductions (Capps, 2017). Also, by releasing greenhouse gasses at the rate in which we currently do, which causes climate change, we warm our planet and in return warm our waters (Althor et al, 2016).

Surface waters over the last one hundred years have been warming by 0.13 degrees celsius (Water, 2018). These warmer waters offer up new challenges to these ecosystems. One way increased temperatures affect freshwater ecosystems is that warm water contains less dissolved oxygen which is required by most species that live in these

ecosystems (Zhang et al, 2022). In freshwater ecosystems the warmer the water the more the rate of chemical reactions increases. These reactions create compounds and bacteria that are sometimes toxic and cause negative consequences on many different species. Different from climate change, exploitation is another way freshwater streams are being negatively affected today. When freshwater streams get exploited it leads to loss of biodiversity and habitats (Hancock, 2002). Studies have shown that when you have increased biodiversity in freshwater ecosystems there is a relationship to having decreased numbers of infection risks for local species that are able to host parasites. (Lagrue & Poulin 2015). One way to begin the effort to preserve these ecosystems is to better understand them and document findings. Human degradation of freshwater ecosystems can be assessed by measuring fish species diversity and abundance (Turner, 1999). Fish are favorable species to study when looking at changes in water chemistry due to human degradation. This is because fish are sensitive to a wide variety of direct stresses, like changes in the water chemistry in which they live. Also they have influence on other species in the aquatic ecosystems and their longevity (Fausch, 1990). Therefore, I have selected to look at fish in the French Creek watershed, specifically common darter and common sucker fish species for this study. Common darter and sucker fish species are not any more particularly sensitive to stream degradation effects but are found in abundance in the French Creek Watershed.

Stream health is commonly assessed using a biomonitoring tool called the Index of Biotic Integrity (IBI) metrics that were developed by Karr (1981). IBI is a well known tool for determining ecosystem health. This tool uses 13 metrics that discerns habitat and species, type and quality. In the United States IBI is considered one of the best ways

to evaluate ecosystem health and has been used in numerous studies (Dauwalter and Jackson, 2004; Tejerina-Garro et al., 2006). The IBI system takes into account species richness, community structure, abundance, health of individual fish, and more to determine impact of human disturbance (Harris & Silveira, 1999). The objective of this study is to determine if water chemistry is associated with the abundance of BSD on fish species. If correlation is proven then scientists in the future could add this metric to the fish IBI tool, which would be the severity of infection of black spot disease. This new metric would further support the integrity of the stream's health and quality knowing that BSD has correlations to poor water quality. Looking at metrics like number of species and species diversity along with habitat assessment which would include variables such as temperature, dissolved oxygen, pH, turbidity, and conductivity and comparing this with the prevalence of black spot disease will help better understand the occurrence of this disease in French Creek.

Uvulifer ambloplitis (Black Spot Disease) is a schistosome-like parasite that falls under the category of Digenean, which is a flatworm (Kirse, 2010). These parasites, also called digenetic trematodes, infect many different freshwater fish species (Lane & Morris, 2000). Diagenetic trematodes are also referred to as grubs and they infect many different species throughout its life cycle (Kelly, 2011). The first host black spot disease uses are snails (Schaaf, 2017). Snails live along moist environments like streams and carry high populations which in return pass along the parasite. Thus, increased snail population directly correlates with increased trematode populations, which in return increases the prevalence of BSD. Once the parasite is on the snail host, it begins to asexually reproduce into cercariae, which are free swimming and infect fish species

(Berra, 1978). Fish become infected by the parasite by entering their body through their scales. Once in the fish the parasite begins to form an encysted metacercariae which then gets at the tissue of the fish and in the end turns into the black spots that are a visual sign of Black Spot Disease (Kurochkin and Biserova, 1996). When a fish is infected with BSD it results in deformities that occur by slow growth and in severe cases mortality of the fish (Harrison et al, 1982). After the fish are infected the parasite continues its journey when the Kingfisher (Megaceryle a. alcyon), consumes the fish (Achatz, 2019). When this happens the parasite passes through the Kingfishers digestive system and is released due to defecation which allows for the parasite to find a new snail host or to be reintroduced to a water ecosystem.

Black Spot Disease can result in mortality of its host if they hold an abundant amount of BSD. As for BSD effects on humans directly there is not any (Kirse, 2010). If fish contain BSD you are still able to eat them and will not become sick due to it. The prevalence of this study is to take BSD as an example of disease in the French Creek watershed and draw correlations with water chemistry metrics to in all correlate human influence on degradation of water systems.

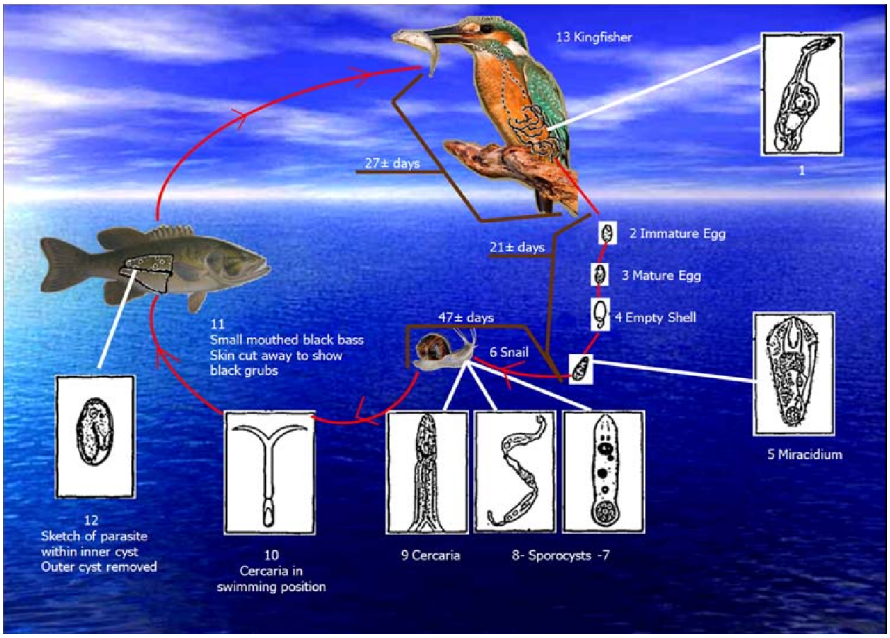


Figure 1: Life Cycle of parasite *Uvulifer ambloplitis*

The objective of this study is to determine if water chemistry data is associated with abundance of BSD. This would then be a possible additional metric for fish health based on severity of infection of black spot disease and could improve the relationship between existing IBI tailored to this region. Looking at metrics like number of species and species diversity along with habitat assessment which would include variables such as temperature, dissolved oxygen, pH, turbidity, and conductivity and comparing this with the prevalence black spot disease will help better understand the occurrence of this disease in French Creek.

Previous studies have found inconclusive evidence to support the hypothesis that Black Spot Disease should be used as a bioindicator (MacKnight, 2016; Nigro, 2017). MacKnights results were found inconclusive due to the lack of variation when it came to

stream location. Nigro's results were found inconclusive due to lack of variation when it came down to the species of fish. In my study I will be adopting similar methods of both studies but introducing more locations and more species of fish to the metrics in order to determine parasite load in fish and how that correlates to stream health that will be determined from the FIBI's. My hope is that adding these new metrics to existing ones will help correlate BSD and stream health and improve the IBI or Fish Index of Biological Integrity (IBI). With that it can help me support the idea and build an argument that human degradation and influence has led to declining environmental health and unhealthy freshwater ecosystems.

Methods:

Scientists at the Allegheny College Watershed Conservation Research Center and Mark Kirk, recorded data from 2015 through 2022. In October, 2022 the WCRC research team and I collected data on three dates, September 23rd, October 7th and October 10th. BSD and water chemistry data was collected at over 20 different sites across the French Creek watershed. The water chemistry data included water temperature, pH, dissolved oxygen, total dissolved solids, alkalinity and hardness.

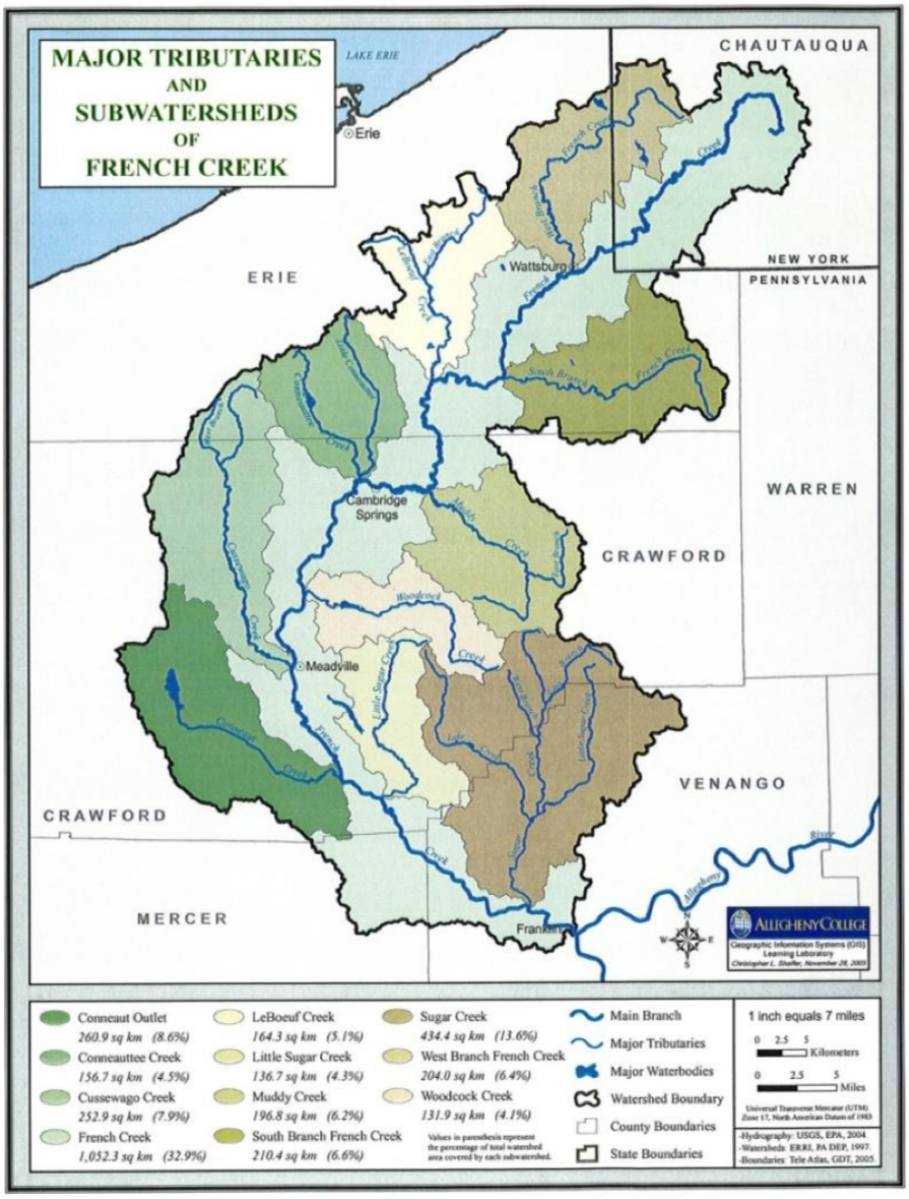


Figure 2: Major Tributaries and Subwatersheds of French Creek

Specific sites within the stream were selected as areas with favorable fish habitat. Thirty to fifty yard transects were laid out in the middle of each stream. We began at one end of the transect and traveled through the water to the farthest point in the transect. A backpack electro shocker was used to electrically pulse the water and momentarily shock any fish that were in that area of the water. We had success in catching fish in high energy parts of the stream like riffles and low energy portions where fish would be located near a calm bank. I walked behind Mark and collected the momentarily stunned fish with a net. The shocked fish were put into a bucket with oxygenated water to minimize the amount of stress put on the fish. We then transported the fish from the bucket to viewing chambers, which was similar to a bucket but has clear walls to be able to view the fish, and recorded the species. Additionally, we looked for evidence of BSD.



Figure 3: Fish with Black Spot Disease

The indicator for black spot disease is black spots usually located on the dorsal fins of fish and on the body. If the fish had black spots present, they were counted. The fish were then put into categories of mild (0-10 spots), moderate (11-20 spots), and severe (>20) infection. Once that data was recorded, the fish were released back into

the stream. Water temperature, total dissolved solids, pH, hardness, alkalinity, and dissolved oxygen were also recorded using a water quality meter and other various devices. Water chemistry data was obtained at the beginning, middle, and end of each transect in order to record the stream in its entirety and to strengthen our data. The water chemistry data and BSD data was then entered into a master excel spreadsheet where it has been compiled since 2015.

Data Analysis:

The raw data that was collected during this study was analyzed through statistical means. It was determined that using regressions for this study would help analyze and show correlations between the water chemistry data and BSD prevalence. The r squared value is derived when you input a Y value, abundance of BSD, and an X value, water chemistry values, into a regression equation which gives a product. The regression can prove statistically if the X and Y values are correlated if the r squared value is or is near 1.

Results:

ID	Year	Water Temp	TDS (ppm)	pH	Mild	Moderate	Severe	Total
MAK226	2016	14	110	8.2	82	7	2	91
MAK114	2016	14	182	8.4	60	17	5	82
MAK139	2016	22	167	8.6	27	10	5	42
MAK268	2016	18	100	7.9	24	18	20	62
MAK276	2016	17	150	8.2	81	5	1	87
MAK272	2016	25	70	8	85	13	3	101
MAK253	2016	21	60	8.1	65	4	0	69
MAK271	2016	16	100	8	43	1	0	44
MAK280	2016	15	20	7.9	22	2	0	24
MAK265	2016	12	50	8.8	21	0	0	21
MAK285	2016	17	130	8.1	5	0	0	5
MAK274	2016	20	100	8	5	5	0	10
MAK260	2016	13	80	7.9	11	3	5	19
MAK254	2016	10	20	8	48	3	0	51
MAK269	2016	14	100	7.9	94	2	4	100
MAK83	2016	16	100	7.8	110	16	12	138
MAK278	2016	19	70	8.1	21	3	3	27
MAK279	2016	17	150	8.4	5	6	1	12
MAK263	2016	20	90	7.7	41	12	4	57
MAK267	2016	16	90	7.7	86	17	10	113
MAK295	2016	18	120	8	41	0	0	41
MAK283	2016	20	30	8	42	14	24	80
MAK290	2016	18	30	7.3	52	3	2	57
MAK442	2022	18	81	8.4	121	55	24	200
MAK488	2022	11	145	8.3	2	0	0	2
MAK469	2022	19	193	8.6	10	3	4	17
MAK491	2022	19	123	8.1	26	30	42	98
MAK482	2022	19	166	8.4	91	11	0	102
MAK486	2022	10	112	8.3	122	27	2	151
MAK492	2022	13	97	8	5	1	0	6

Figure 4: BSD and Water Chemistry Data

Figure 4 is a data chart representing all of the raw data that was collected for use in this study. Each MAK corresponds to a specific site location of the French Creek watershed. The year was also recorded because this study accounted for data from 2016 and 2022. Water chemistry data which includes water temperature, total dissolved solids, and pH are also represented. Lastly, the number of fish that had BSD present are recorded along with values of the specific cases of mild, moderate, and severe cases of BSD.

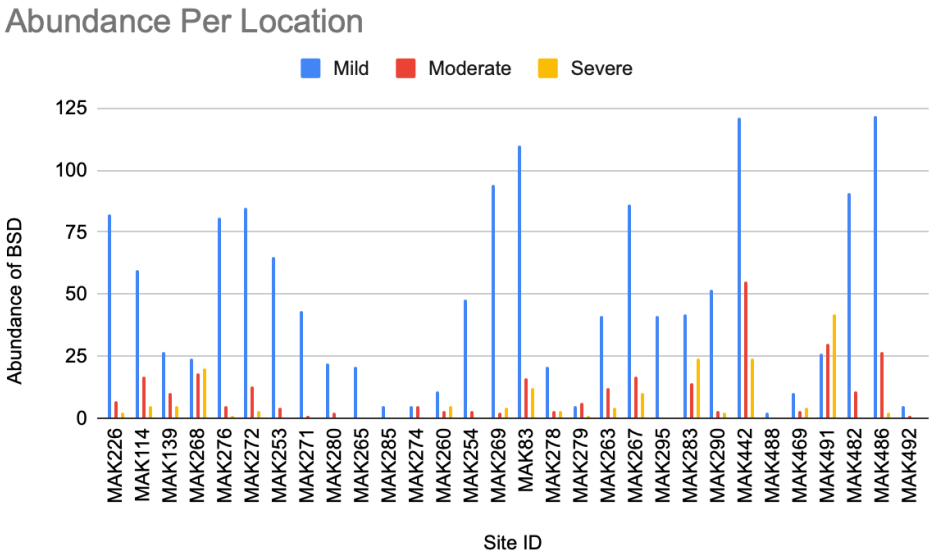


Figure 5: Abundance and Severity of BSD per Location

Abundance of BSD at each site is displayed in figure 5 and is split into 3 different categories. The blue bar represents mild or 0-10 black spots, red is moderate or 10-20 black spots, and yellow is greater than 20 black spots. The Y axis or abundance of BSD is showing the total number of fish that had BSD at each site. Figure 5 shows at most sites the BSD cases were mild. Many different species were collected in this study. Species include blacknose dace, creek chub, longnose dace, pearl dace, white sucker, blue gill, bluntnose minnow, stone roller, fantail darter, redside dace, fathead minnow, and tonguetied minnow.

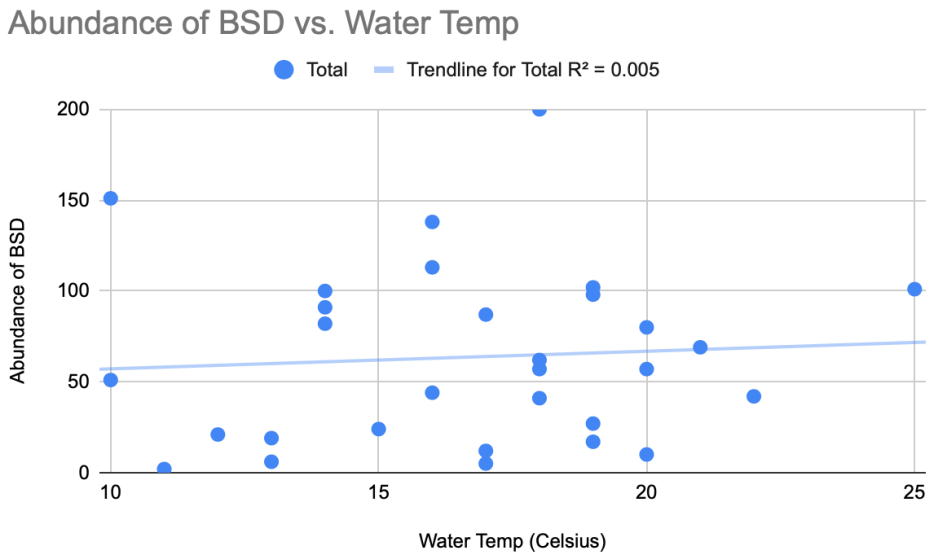


Figure 6: Abundance of BSD compared to water temperature

Figure 6 demonstrates the abundance of BSD and water temperature . In this study water temperatures from the French Creek watershed streams ranged from 10 degrees celsius to 25 degrees celsius. The line of best fit is also present to show the trend line and relationship between abundance of BSD and water temperature. Aside from the line of best fit this graph also represents outlier data points which are distant from the line. The R squared value for this figure is 0.005.

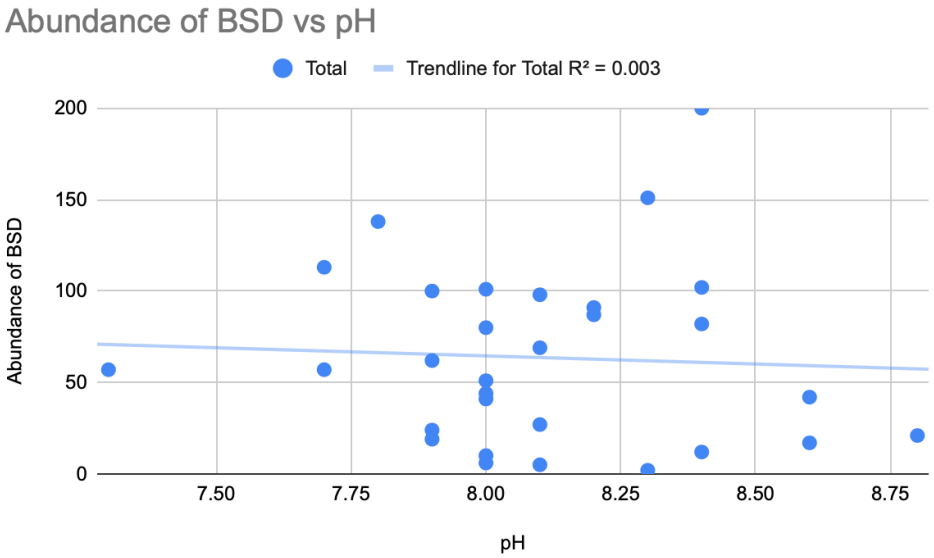


Figure 7: Abundance of BSD and stream pH.

The correlation between abundance of BSD and pH of the stream is displayed on figure 7. In this study the French Creek watershed streams ranged in pH from 7.3 to 8.8. A line of best fit is also displayed to show the relationship between the abundance of BSD and pH. This graph also shows data points that are outliers and distant from the line of best fit. The R squared value for this figure is 0.003.

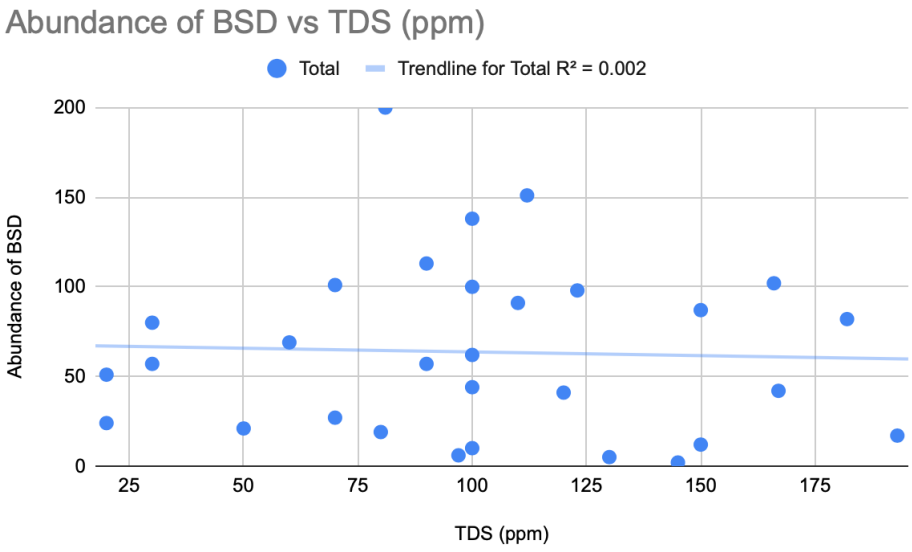


Figure 8: Abundance of BSD and TDS in the streams

The correlation between abundance of BSD and TDS in the streams is displayed on figure 8. In this study the French Creek watershed streams ranged in TDS from 20 parts per million to 193 parts per million. A line of best fit is also displayed to show the relationship between the abundance of BSD and TDS in the streams. The R squared value for this figure is 0.002.

Discussion:

This study demonstrates that BSD is thriving and is found in many different streams in the French Creek watershed. Water chemistry factors such as water temperature, TDS, and pH are being affected by human activity and are responsible for negative effects in aquatic ecosystems like BSD.

Figure 5 shows that BSD is abundant at all of these sites. In most cases the abundance of BSD on fish species at each site was found to be mild. When looking at

the three water chemistry graphs, water temperature, TDS and pH when compared to abundance of BSD they all tell a similar story.

BSD does increase with rising temperatures; however, the R-squared value does not show a strong correlation. All data points in this figure are not uniform or located near the trendline. This means that even though there is a trend the trend is not strong enough to argue correlation due to the abundant outliers not positioned near the line. The R squared value also helps prove no correlation due to its value being 0.005 and correlation occurs at 1. Water temperature was used in this study because warmer waters are more likely to host waterborne diseases (Funari et al, 2019). In this study we can see that there is not enough evidence to support an argument that temperature affects BSD abundance in streams.

As the stream becomes more basic or pH increases, the abundance of BSD decreases. In figure 7, the data again is not closely fit to the trend line therefore there is not a strong relationship between abundance of BSD and pH. The r squared value is 0.003 which also supports the argument towards no correlation. Most stream pH tested in this study fell between the range of 7.3 and 8.7. This range of pH is suitable for growth of waterborne pathogens or disease (Leclerc et al, 2002). Although the pH range of the stream is suitable for diseases and is proven to host BSD there is no correlation in my study between pH values and abundance of BSD.

The last water chemistry factor I tested for correlation was TDS compared to abundance of BSD. Figure 7 shows close similarities to figure 8 in which the trend line is negative and the data points are not closely situated to the trend line. Therefore, once again it is not enough to establish a correlation between levels of TDS in the stream and

its effect on abundance of BSD on fish species. The R squared value also helps prove no correlation due to its value being 0.002 and correlation occurs at 1. High levels of TDS are often found in environments that have pollution or bacteria which contributes to disease in fish and other species. (Hill et al, 2005). Again, even though there is past evidence of this, using the data I recorded for this study there is no such correlation between abundance of BSD and TDS.

During this study water chemistry data varied a significant amount. Multiple reasons could and are behind this offering a lot of factors to be thought of. These factors include rates of precipitation and severe weather events. Proximity of these streams to nearby industry, farms, or any other possible sources of pollution. These two factors combined could account for polluted runoff that could change the water chemistry of the streams along with many other factors.

Although my study was not able to find correlations between water chemistry data which included water temperature, pH and TDS and BSD, it does serve a purpose when thinking about how we can understand BSD. In this study BSD is looked at through one of its hosts throughout its lifecycle, fish. After performing this research project I believe that an in depth look at snails would be very interesting. Fish receive the parasite from snails that live near the streams. I think it would be very beneficial to understand and perform a study that evaluates the snail populations that host BSD and find the abundance of BSD on each snail. This could be performed in the French Creek watershed so that preexisting data from this study and others could contribute. This in all would help tell the story of how BSD gets into stream ecosystems and how it affects certain species. Once that data is gathered then you could look at water chemistry again to see if there are any correlations with the abundance of BSD in snail populations.

At the root of any problem like BSD in water ecosystems you want to try and figure out why these diseases are around. Since the 1900's climate change is something people in the scientific community have had to come face to face with (Trenberth, 2011). Climate change is the changing of weather patterns and climates driven by human activity (Karl et al, 2003). Climate change is happening and humans are playing a role in creating these climate issues. The largest way humans are contributing to climate change is by burning fossil fuels. By burning fossil fuels we are releasing greenhouse gasses which absorb light that's trying to escape the atmosphere but instead is being trapped (Hansen & Silveria, 1999). This causes higher temperatures and has many effects on our climate like warming our waters. Besides burning fossil fuels, humans pollute waterways with plastics, metals, and other non decomposable materials found in today's popular consumer goods (Weis, 2015). Burning fossil fuels and polluting plastics and other materials are ways that humans are having negative impacts on our aquatic ecosystems. Human activity has caused negative impacts that are causing the irreversible loss of biodiversity and aquatic ecosystems. Water ecosystems like streams are at the forefront of these issues and are leading to issues such as BSD. We know from previous studies that waterborne diseases flourish in warm waters and environments with high levels of TDS (Nwidu et al, 2008). Human influence is causing warmer waters, higher levels of TDS, and more extreme pH levels. Human practices like agriculture, water use, industry processes, and mining are ways that humans are causing higher levels of TDS in our waterways (Olsen and Hawkins, 2017). Similarly pH levels in aquatic ecosystems are also being affected due to certain human activity that is responsible for increased hydrogen ion inputs. These include agriculture or animal feedlots, urbanization or vehicles, industry or coal fired power plants, and mining or acid mine drainage (Eissa and Zaki, 2011). The relationship between water temperature,

TDS, and pH are not bound to one another but are bound to human activity resulting in unnatural values of these being represented in our aquatic ecosystems today. Instead of focusing on which one of these might cause the other to change, we need to address the root of the problem, humans' effect on our environment. BSD is just one example of the issues we are seeing as a result of climate change and human degradation.

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