

**The Effect of Wastewater Treatment Plant Effluent on Water Temperature,
Macroinvertebrate Community, and Functional Feeding Groups Structure in the Lower
Rouge River, Michigan**

by

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**A thesis submitted in partial fulfillment
of the requirements for the degree of
Master of Science
(Environmental Science)
in the University of Michigan-Dearborn
2016**

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DEDICATION

I dedicate this Master's thesis to my husband Joel Deussen who has encouraged me and supported me through the whole research process. He is the pillar of my life and gave me the strength for perseverance and determination.

I also dedicate this thesis to my parents Selma and Hidayet Tuncay and my siblings Fatma and Ahmet in Germany. Even though, we are separated by the Atlantic Ocean and a vast distance, they always were at my side and supported me.

Thanks to all for their huge support.

Istanbul, 9 Dec 2016.

ACKNOWLEDGMENTS

First, I would like to thank my thesis advisor Larissa Sano, PhD of the College of Literature, Science, and the Arts at the University of Michigan. Prof. Sano was always available when I had questions regarding my research and writing and guided me in the whole research process. Additionally, I would like to thank her for help during the summer sampling of macroinvertebrates for my research and providing all the necessary equipment for my research.

I would also thank Orin Gelderloos, PhD of the Natural Science Department at the University of Michigan-Dearborn. The door to Prof. Gelderloos office was always open for additional questions regarding my thesis. I am grateful for his very valuable comments and guidance throughout the thesis process.

I would also like to acknowledge Sonia M. Tiquia-Arashiro of the Natural Science Department at the University of Michigan-Dearborn and Sally Petrella from Friends of the Rouge as the thesis committee member for their expertise and I am gratefully indebted to both of them for their very valuable comments on this thesis.

Finally, I would like to thank Robert Muller and Philip Kukulski for designing the PVC temperature sensor holders. I would like to thank Robert Muller for building the PVC temperature sensor holders, for helping me setting the temperature sensors and regularly checking on them. I also thank all the volunteers at Friends of the Rouge and Wayne County Department of Public Works Water Quality Management Division, especially Susan Thompson for helping me sample and picking the sampled macroinvertebrates from trays for identification.

Additionally, I would like to thank 4th Street Auto located at 303 E. Fourth Street in Royal Oak, MI. 48067 for donating the break discs which were used to weigh down the temperature sensors and secure them at place.

Thank you,

Esma Tuncay

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LIST OF ABBREVIATIONS

CPOM	Coarse particulate organic matter
FPOM	Fine particulate organic matter
DOM	Dissolved organic matter
AOC	Area of Concern
YCUA	Ypsilanti Community Utilities Authority
NPDES	National Pollutant Discharge Elimination System
WWTP	wastewater treatment plant
WTUA	Western Township Utilities Authority
ML/d	million liters per day
Mgd	million gallons per day
MDNR	Michigan Department of Natural Resources
MDEQ	Michigan Department of Environmental Quality
CBOD₅	Carbonaceous biochemical oxygen demand
N	Nitrogen
FOTR	Friends of the Rouge
NOAA	National Oceanic and Atmospheric Administration

USGS United States Geological Survey

stdev Standard deviation

m³/sec Cubic meter per sec

cft/sec Cubic feet per sec

ABSTRACT

The release of effluent from wastewater treatment plants can impact receiving water bodies by altering water temperatures. The Ypsilanti Community Utilities Authority (YCUA) has been discharging its wastewater effluent into the Lower Rouge River since 1996. To understand the impact of these discharges on water temperature in the Rouge River, this study measured the water temperature hourly from April 13 to October 26, 2015 at five different locations: one at the discharge site (LR-2), three below the effluent discharge (LR-12, LR-6, and LR-10), and one upstream of the discharge (Fowl2, control site) at the Lower Rouge River, MI. Additionally, benthic macroinvertebrates were sampled during spring, summer, and fall to analyze the impacts of changes in water temperatures on the macroinvertebrate fauna. Water temperatures at the discharge site (LR-2) showed significantly different temperatures than the upstream control site (Fowl2) during summer 2015. The sites further downstream significantly differed in water temperature compared to LR-2 during summer 2015 for LR-12, LR-6, and LR-10 and during fall 2015 for LR-12 and LR-6. However, water temperature at the Lower Rouge River below the YCUA discharge did not contribute to changes in macroinvertebrate family richness and diversity. Additionally, functional feeding groups were analyzed. Fowl2 had higher numbers of the functional feeding group collector-gatherers compared to collector-filterers, which suggests that the fine organic particulate matter (FOPM) is distributed on the river bottom. Yet, the functional feeding group collector-filterer were higher below the YCUA discharge compared to Fowl2, which indicates a shift in FOPM from the river bottom into the water column which could be caused by the YCUA discharge flow. My results suggest that the YCUA discharge temperature did not have

any influence on the downstream sites LR-12, LR-6, and LR-10. Additionally, different family richness and diversity of macroinvertebrates are most likely caused by a shift in nutrient distribution (FPOM) rather than a change in water temperature.

CHAPTER I

1. INTRODUCTION

Urban watersheds are water catchment areas of waterbodies in urban areas. They have multiple stressors on the watershed affecting the watersheds biological, chemical, and physical conditions resulting from urbanization compared to rural watersheds. They are characterized by impervious surfaces created through residential and commercial structures, as well as roads. For certain reasons such as flooding control, infrastructural issues, or water transport, the river is shaped, straightened, concrete channeled or closed underground to fit municipal planning. The natural shape of the watershed is lost and subject to many anthropogenic influences. Point and non-point source pollution result in biological and chemical pollution. In addition, impervious surfaces result in higher surface run-off which increases the river discharge rapidly and result in extreme river bank erosion and flooding. Thermal pollution can also occur through municipal or industrial discharges into the river which can change the natural water temperature and affect the aquatic ecosystem (Caissie 2006). The Rouge River watershed in Michigan, USA is an example of an urban watershed.

1.1. Thermal regime of streams and rivers

The thermal regime of a river reflects the daily and annual water temperature variation in a watershed. These fluctuations affect the quality and health of the aquatic ecosystem and stream productivity (Caissie 2006; Verones et al. 2010) because water temperature is an important abiotic

factor, which influences biochemical and physiological activities of aquatic organisms (Verones et al. 2010). Temperature is a physical factor that can also influence chemical reactions, the properties of chemicals, and microbial activity (Dallas 2008).

1.1.1. Factors influencing the thermal regime of streams and rivers

The thermal regime of a river is influenced by atmospheric conditions, topography, stream discharge, and streambed (Caissie 2006; Dallas 2008). Atmospheric conditions such as solar radiation, air temperature, cloud cover, vapor pressure, wind speed, humidity, precipitation, evaporation, condensation, and phase changes of water which are responsible for heat exchange processes at the water surface (Caissie 2006; Dallas 2008). Upland shading, riparian vegetation, geology, stream orientation, channel form, slope, water depth, turbidity, percentage of pool habitat, as well as latitude and altitude define the topographical and structural influences on the thermal regime of rivers surface (Caissie 2006; Dallas 2008). Therefore, topography also determines the intensity of the influences of atmospheric conditions on the thermal regime, for example high canopy cover during spring and summer reduces the input of solar radiation to the stream and therefore protects the stream from getting too warm for some fauna and flora (Caissie 2006). Stream discharge determines the volume of water based on inflow and outflow (Caissie 2006). The heat capacity of river water depends on the amount of water and the mixing by turbulence, slopes, waterfalls, and friction on the streambed (Caissie 2006). The streambed is important for groundwater input and hyporheic exchange, which is also important for the volume of water and the heat exchange capacity (Figure 1).

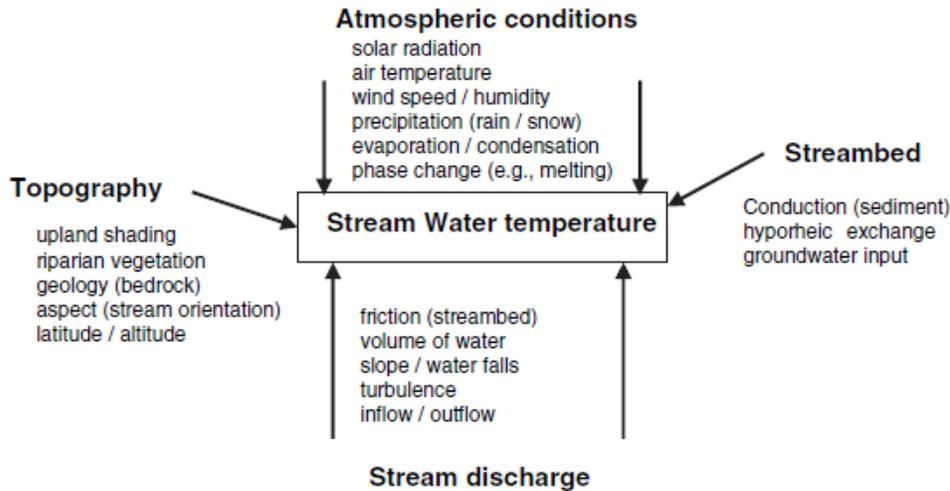


Figure 1. Factors influencing the thermal regime of rivers (Caissie 2006).

1.1.2. Thermal regime heat exchange processes

The thermal regime heat exchange processes take part at the air/water interface and also streambed/water interface (Caissie 2006). The air/water interface heat exchange contributes up to 82% to the thermal regime and therefore atmospheric conditions are the most important factor in regulating thermal regimes of streams. The streambed/water interface contributes around 15% to the thermal regime. The heat exchange processes at the air/water surface mainly consist of net short wave solar radiation, net long wave solar radiation, evaporation, and convective heat flux. Other components such as precipitation and friction on the water surface have a very small input to the heat exchange process and therefore can be considered negligible. Solar radiation, or lack of it, is crucial for heat-gain and -loss that is influenced by riparian vegetation cover. The heat exchange at streambed/water interface consists of geothermal conduction, groundwater contribution and hyporheic exchange. Groundwater temperatures are also influenced by air temperature which again shows the high importance of atmospheric conditions on the water temperature. Even though atmospheric conditions have the highest influence on the thermal

regime, riparian vegetation and the size of the river must be considered in determining the influence of the different heat exchange processes.

1.1.3. Longitudinal water temperature variation of streams and rivers

Streams and rivers begin as small tributaries at the headwater. They increase in size and water volume in downstream direction as they connect with other tributaries. The downstream direction or river length from the headwaters to the mouth is called the longitudinal direction of streams and rivers. Water temperature at the source of a river is close to groundwater temperature and increases both in a longitudinal direction from headwaters to the mouth with increasing stream order (Caissie 2006; Vannote and Sweeney 1980). Small streams usually increase 0.6 °C per km, intermediate streams increase 0.2 °C per km, and larger rivers increase 0.09 °C per km (Caissie 2006). Therefore, water temperatures changes with river size, but temperature change is usually non-linear and depends on many factors (Caissie 2006). Different habitat types such as pool, run, and riffle habitats show small scale temperature variation because they differ in size, depth and stream velocity (Dallas 2008; Vannote and Sweeney 1980). Also, different types of rivers and streams have different thermal regimes (Caissie 2006). For example, braided rivers have small and shallow channels that are prone to faster temperature changes.

1.1.4. Annual and daily variation of thermal regime

In temperate areas, the thermal regimes of streams vary both daily and seasonally (Caissie 2006; Dallas 2008). Daily minimum temperature is reached at sunrise and daily maximum temperature is reached in the late afternoon and early evening (Caissie 2006; Dallas 2008). Daily water temperature fluctuations vary with location, whereas headwater streams show the lowest temperature fluctuation due to the influence of groundwater sources (Caissie 2006; Vannote and Sweeney 1980). In a downstream direction, rivers are more affected by atmospheric conditions

and show a higher diel variation because the river increases in width, which the riparian cover cannot completely shade anymore and protect it from solar radiation (Figure 2; Caissie 2006). For stream orders greater than 4, the influence of atmospheric conditions is reduced because more energy input is required to heat up the large volume of water (Caissie 2006; Dallas 2008; Vannote and Sweeney 1980). These rivers demonstrate low diel variability (Caissie 2006; Dallas 2008; Vannote and Sweeney 1980). The thermal regime of rivers also varies seasonally for temperate regions, with a cold phase during winter and a warmer period between spring and fall, reaching the maximum annual temperature during summer (Caissie 2006).

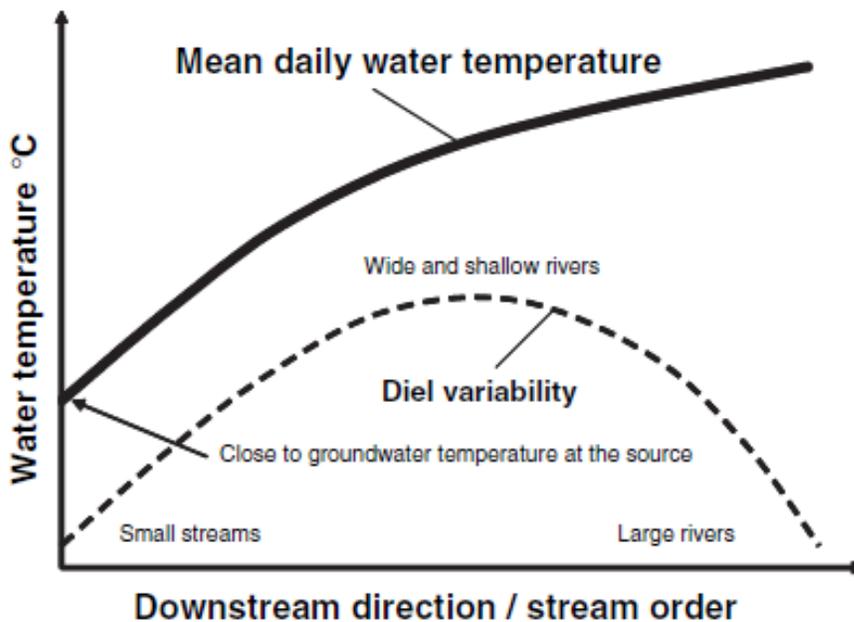


Figure 2. Mean daily and diel variability of water temperatures as a function of stream order/downstream direction (Caissie 2006).

1.1.5. Analyzing thermal regime

1.1.5.1. Scaling area of study

In analyzing the thermal regime of rivers, it is important to consider the scale of the area of study. Three different scales are typically used to compare the thermal regime of rivers (Dallas 2008). The first and largest is the catchment scale, which compares various thermal regimes of

individual rivers with each other, thereby accounting for the change in climate, geography, topography, and vegetation (Dallas 2008). The second scale is at the river level, which compares the thermal regime within a river system (Dallas 2008). Analysis at the river scale focuses on water temperature changes longitudinally from the headwaters to the mouth (Dallas 2008, Figure 2). Water temperatures usually increase in a downstream direction and reach maximum values in the middle reaches (Dallas 2008). The third scale is the site scale, which compares the thermal regime of a small section of a river with different habitat that consists of different depths and therefore different water temperatures (Dallas 2008; Langford 1990).

1.1.5.2. Water temperature of streams and rivers

The thermal regime of lotic waterbodies can be divided into water column temperature, water and streambed substrate interface temperature, and substrate temperature (Langford 1990). Most aquatic animals live at the water and streambed substrate interface (Langford 1990). At the water and streambed substrate interface the temperature is generally the same as that of the water column (Langford 1990).

1.1.6. Influence of water temperature on aquatic fauna

Many aquatic animals require a specific temperature range for optimal distribution, growth, reproduction, and fitness (Caissie 2006; Dallas 2008; Vannote and Sweeney 1980). The optimal temperature range for aquatic fauna is defined as the temperature that leads to maximum body weight and fecundity without causing physiological stress (Vannote and Sweeney 1980). It differs between organisms (Vannote and Sweeney 1980). For example, the carp (*Cyprinus carpio*) grows best at temperatures between 23 - 29 °C whereas the brown trout (*Salmon trutta*) requires lower temperature between 7 - 17 °C for optimal growth (Figure 3). Macroinvertebrates also have optimal water temperature preference such as 9.1 – 10.6 °C for *Drunella cryptomenria*, 10.3 – 11.6 °C for

Stenelmis sp., and 10.8 – 12 °C for *Asellus* sp. (Li et al. 2013). However, most freshwater macroinvertebrates reach thermal death for water temperatures between 30 to 40°C (Wallace and Anderson 1996). In general, aquatic fauna are more sensitive to high temperature than aquatic flora (Verones et al. 2010).

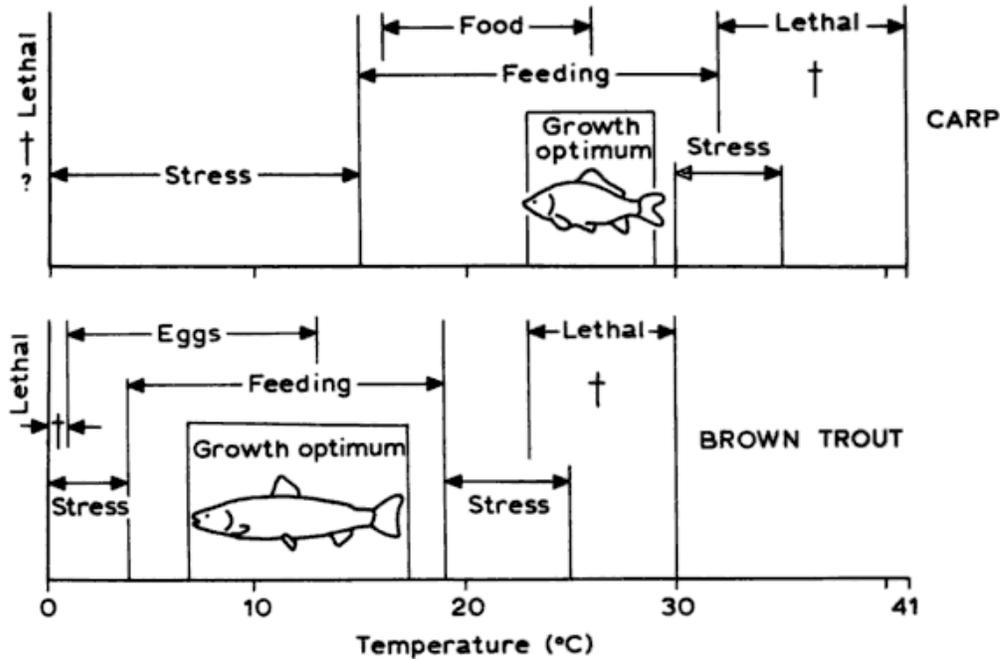


Figure 3. Comparison of thermal requirements of carp (*Cyprinus carpio*) and brown trout (*Salmon trutta*) (Langford 1990).

Aquatic animals are ectothermic organisms and therefore water temperature can have a direct effect on their growth, development, respiration, excretion, and general fitness (Dallas 2008; De Stasio, Golemgski, and Livingstone 2009). Because of their sensitivity to water temperature, water temperature can affect their abundance and diversity (Dallas 2008; Vannote and Sweeney 1980). Temperature is also a physical factor that can influence other chemical reactions, the properties of chemicals, and microbial activity (Dallas 2008). For instance, the toxicity of chemicals increases with increasing temperature (e.g. Ammonia increases in toxicity by a factor of 1.3 to 1.6 in relation to pH for temperature increases from 10°C to 20°C) (Cairns, Heath, and

Parker 1975) while the dissolved oxygen concentration decreases with higher temperatures (e.g. Concentration of O₂ (mg/L) in pure water at 0°C is 14.2 mg/L and at 30°C is 7.5 mg/L) (Allan and Castillo 2007a). With higher water temperature, the metabolic activity of aquatic animals increases as well, which leads to faster depletion of dissolved oxygen concentration and lower food resources (Dallas 2008). Thus spatial and temporal changes in water temperature influences the behavior of aquatic animals and therefore are important factors to understanding the responses of aquatic fauna to water temperature changes (Dallas 2008).

According to the river continuum concept (Vannote et al. 1980), daily and seasonal stream water temperature variation is responsible for the distribution of aquatic animals (Caissie 2006). Fishes are able to detect temperature changes of 0.05 °C and their neuronal system selects the optimal growth temperature (Langford 1990). Fish thus tend to avoid waters around thermal discharge sites and move to water with a temperature closer to their optimal growth temperature (Langford 1990). Benthic macroinvertebrates are also impacted by temperature. They show higher drifting rates with higher temperatures and therefore avoid the thermal discharge by moving downstream (Langford 1990). Individual species show different tolerance to temperature and the movement rate will vary from species to species (Langford 1990). Li et al. (2013) researched the distribution of macroinvertebrates species according to different water temperatures in South Korean streams. In their study, the most sensitive taxa to increasing water temperature were Ephemeroptera, Plecoptera, and Trichoptera (Li et al. 2013).

1.2. Thermal discharge and thermal pollution

1.2.1. Anthropogenic influences on thermal regime

Thermal discharges generate a point source of water with an elevated temperature into a receiving waterbody (Gooch 2007). Thermal discharges can be divided into natural and artificial

ones. Geothermal discharges such as geysers and hot springs are natural and discharge their hot waters (up to 100°C) to adjacent lentic and lotic habitats (Langford 1990). These hot temperatures support a unique flora and fauna for these areas (Langford 1983). Artificial thermal discharges results from anthropogenic activities that affect the thermal regime of lotic ecosystems both directly and indirectly (Dallas 2008). The magnitude of direct and indirect effects on thermal regimes can vary in regard to the river water volume and geographical location of the river. Rivers and streams can be affected by a variety of point- and non-point sources, which can have a cumulative effect on the change of their thermal regime (Poole, Risley, and Hicks 2001).

Artificial thermal discharges range from point to non-point sources. Thermal discharges from power plants, industrial processes, municipal waste water treatment plants, air conditioning and refrigerator plants, impoundments and dams are classified as point source discharges because they are discharged directly to the waterbody next to their location (Langford 1990). The most prominent thermal discharges come from power plants and may show higher water temperatures (up to 42°C) (Coutant 1962) than the other discharge types and, therefore, could have a larger influence on thermal regimes over a section of streams and rivers (Langford 1990).

Non-point sources are often scattered around the watershed. These non-point sources range from impervious surface run-off to agricultural drainage (Dallas 2008; Langford 1990; Schueler 1994). Other activities such as afforestation and deforestation, change of riparian vegetation, and global warming can change the water temperature as well (Dallas 2008; Langford 1990). The degree of effects depends on the number of point and non-point sources (Langford 1983).

1.2.2. History of thermal discharge

Humans start using water for cooling purposes since ancient time. Historically, thermal

discharge became more prevalent and larger with human activities during the Industrial Revolution which resulted in mass production and increased the usage of water for cooling purposes (Langford 1990). Factories were built adjacent to waterbodies in order to use the water for cooling processes. This water was then discharged back to the waterbody after use. This large amount of thermal discharge can change the thermal regime of receiving waterbodies and affect their aquatic ecosystem (Dallas 2008; Langford 1990).

Some of the first concerns about the ecological effects of thermal discharge arose from exotic snails appearing in cooling ponds for steam engines (Langford 1990). These snails (*Menetus dilatatus*) were first discovered in cooling ponds in Manchester, UK in 1869, having been introduced from North America (Macan 1960). The “Electricity Act of 1919” in the United Kingdom noted that thermal discharges may have an effect on the aquatic ecosystem and the “Ministry of Health” of the United Kingdom noted in 1949 that 1°C could have an adverse effect on the aquatic ecosystem (Langford 1990). But the first public awareness of thermal discharges as thermal pollution and their effects on the native aquatic ecosystem arose in 1952 from the lawsuit against the Spondon Power Station. Many studies focused on the effects of thermal discharges on the aquatic ecosystem from the mid-1950s to the 1960s and started the “thermal pollution era”. Research concerning thermal discharges began to decline in the late 1970s, even though the amount of thermal discharges continued to increase.

1.2.3. Thermal discharge properties

Thermal discharges can be divided into rapid jet discharges, which have a higher turbulence and mixing rate with the receiving water, and low turbulence and velocity discharges, which usually go into the top layer of the receiving waters (Langford 1990). Thermal discharge has the ability to change the thermal regime of rivers by increasing the water temperature and flow

regime of these waterbodies. Changes in flow regime may lead to change in direction and velocity and therefore in sediment deposition. In addition, warm water has a lower density and higher buoyancy, which can change the density and buoyancy properties of the receiving water.

1.2.4. The effects of thermal discharges on thermal regime

1.2.4.1. Thermal regime

The effects of thermal discharges on the receiving waterbodies usually depends on the size, volume of water, water depth, rate of mixing, and velocity of the receiving waters as well as on the volume of discharged water (Langford 1990). Therefore, smaller receiving rivers will be more affected than larger ones (Vannote and Sweeney 1980). In addition, in temperate regions, the seasonal variation of lotic water temperatures further determines the effect of the thermal discharge on the thermal regime (Caissie 2006) because the temperature of thermal discharge also fluctuates with seasons (Dallas 2008; Langford 1990). Temperature fluctuations of thermal discharges can lead to unstable thermal regime of the receiving waters, but an unstable thermal regime of receiving streams could be stabilized by stable thermal discharges (Langford 1990). The impact area of the thermal discharge depends on the channel width and depth of the stream. Thermal discharge usually mixes well with small streams, but large rivers could concentrate the effect of the thermal discharge on a narrow path of flow along on the bank at the discharge and lead to lateral stratification of the river. Usually, thermal discharges are transported downstream, but during low flow and high upstream wind periods, the discharge could be transported a small distance upstream. The high upstream wind periods could move the thermal discharge over a short distance upstream during low flow periods and also alter the thermal regime in the immediate upstream region.

1.2.4.2. Effects of thermal discharge on aquatic animals

The effects of temperature on aquatic animals can be either lethal or sub-lethal (Langford 1990). Lethal temperatures are temperatures that are high enough to cause direct death to the animal (Caissie 2006; Langford 1990). Sub-lethal temperatures are temperatures that do not directly kill an individual, but that can cause changes in behavior such as movement and migration and can impact physiological and biochemical processes such as higher metabolic activity and respiration and/or impairments in growth and reproduction (Langford 1990). Metabolic activity increases with temperature and reaches a maximum level, which is followed by death (Langford 1990; Verones et al. 2010; Voshell 2002). Van't Hoff's rule implies a doubling in metabolic activity with every 10 °C increase in water temperature (Caissie 2006; Dallas 2008). Therefore, increased metabolic activity leads to indirect effects resulting in oxygen depletion (Caissie 2006).

Other indirect effects of thermal discharges are the change in chemical toxicity and a higher demand of food (Caissie 2006; Langford 1990). The increase in metabolic activity of aquatic animals' result in a higher demand of food. The higher demand for food will in turn have an effect on predators, competitors, and prey and therefore could change the trophic level composition (Dallas 2008; Langford 1990). The impaired metabolic activity can also lead to impaired reproduction (Verones et al. 2010) which will result in lower off spring and less competition (Vannote and Sweeney 1980). Other sub-lethal effects can influence growth (Hogg et al. 1995; Hogg and Williams 1996), behavior, food and feeding habitats (Dallas 2008), life history (Hogg and Williams 1996), geographical distribution (Li et al. 2013), community structure (Li et al. 2013), movements (Durrett and Pearson 1975), migration (Durrett and Pearson 1975), and the tolerance to parasites, diseases and other pollution (Dallas 2008). Therefore, a single impairment has the ability to cause a chain reaction of other effects that can have impacts on the individual

species, and at subpopulation, population, and community levels. Lethal and sub-lethal temperatures are specific to each species, but in general the tolerance to temperature changes is higher in physiologically and morphologically less complex taxa (Langford 1990).

1.2.4.3. Other effects of thermal discharges

Thermal discharges can also influence the chemical properties of water such as dissolved oxygen concentrations as well as the toxicity of chemicals (Caissie 2006; Dallas 2008; Langford 1990). Dissolved oxygen concentrations are inversely related to water temperature and decrease with increasing water temperatures (Caissie 2006; Dallas 2008; Langford 1990). However, high velocity and mixing rate can create turbulence that mitigates the effect of the thermal discharge on dissolved oxygen concentrations (Langford 1990). On the other hand, the toxicity of chemicals increases with higher temperatures (Langford 1990). Chlorine is one of the chemicals that is added to the thermal discharge to prevent biological fouling of pipes and culverts (Langford 1983) and is the main culprit for impaired aquatic ecology rather than temperature alone (Langford 1990).

1.2.5. Previous research on thermal discharges

Most studies of the effects of thermal discharges on river ecology are from power plant releases and generally were conducted between the 1960s and the 1980s (Langford and Aston 1972; Osborne and Davies 1987; Alston et al. 1978; Benda and Proffitt 1974; Coutant 1962; Howell and Gentry 1974; Massengill 1976; LeRoy Poff and Matthews 1986; Wurtz and Skinner 1984). Studies from the 1990s shifted to focus on climate warming and its effect on the macroinvertebrate fauna (Hogg and Williams 1996; Hogg et al. 1995), but research on thermal discharges continued (Wellborn and Robinson 1996; Worthington et al. 2015).

1.2.5.1. Thermal discharge and its effects on macroinvertebrates

Coutant (1962) is one of the earliest researchers to analyze the effects of thermal discharge

on macroinvertebrate fauna in riffle habitats. Several subsequent studies found that thermal discharge has the most deleterious effect during summer months, with water temperature ranging between 40 and 42 °C causing decreases in the abundance of macroinvertebrates (Coutant 1962; Durrett and Pearson 1975; Wellborn and Robinson 1996). Sensitive taxa such as Trichoptera, Ephemeroptera and Plecoptera occurred in lower numbers and demonstrated lower species diversity in thermally disturbed areas (Howell and Gentry 1974; Li et al. 2013), whereas tolerant taxa dominated these areas including Chironomidae (Benda and Proffitt 1974; Coutant 1962; Coutant and Brook 1970; Howell and Gentry 1974) and Oligochaeta (Osborne and Davies 1987). Unlike these other studies, elevated water temperatures in winter were associated with an increase of up to 10 to 40 % in benthic fauna biomass (Coutant and Brook 1970). In contrast to the other studies, Langford (1972) reported no significant negative effects associated with thermal discharges in British rivers. Therefore, a generalized assumption about the effects of thermal discharge on macroinvertebrates fauna is not possible.

Other studies compared the effects of thermal discharges on invertebrate fauna with considering the chemical pollution level of the river. Langford and Aston (1972) compared a chemical polluted and non-polluted river receiving thermal discharge in Britain. The water temperature was measured below the outfall between 1965 to 1970 for the non-polluted river and between 1965 to 1966 for the polluted river. The maximum water temperature below the thermal discharge was up to 6°C above average in the non-polluted river and 10°C above average in the polluted river. Neither river showed any difference in their upstream and downstream fauna diversity of invertebrates. The non-polluted river consisted of many diverse invertebrate taxa with many intolerant species, however, in comparison, the polluted river had low diversity. The polluted river showed lower macroinvertebrate diversity mainly due to the chemical pollution from

domestic and industrial effluents because macroinvertebrate diversity was drastically reduced at River Trent below the confluence with the highly chemically polluted River Tame. There were differences between rivers in terms of life history of certain species, specifically the Oligochaete community. They showed a shift towards the species *Limnodrilus hoffmeisteri* with a peak cocoon production in October below the discharge of the polluted river compared to May for population in the upstream reaches. Also, below the outfall in non-polluted reaches, the species *Heptagenia sulphurea* emerged earlier, which show that the life history of some species seems to be affected by higher water temperature. However, Hogg and Williams (1996) observed faster adult emergence and faster growth of some species due to moderate high water temperatures of 2°C in spring, summer and fall, and 3.5°C in winter. In the higher temperature sites, some species also showed smaller size at maturity and altered sex ratios in comparison to thermally non-affected areas. These findings indicate that the effects of thermal pollution can have different effects on species in terms of life history.

Thermal discharges do not always result in high elevated water temperatures in the receiving waterbodies and macroinvertebrates show different behavior compared to higher water temperature increases. Regarding moderate water temperature increases, studies have reported different outcomes in macroinvertebrates in comparison to higher elevated water temperatures. LeRoy Poff and Matthews (1986) observed an increase in macroinvertebrate abundance and taxa at the confluence site of the stream with the Savannah River in North Carolina which was slightly thermally affected. Dahlberg and Conyers (1974) also observed an increase in taxa and individual numbers during winter for water temperatures between 9.5 and 17 °C. However, Hogg et al. (1995) observed a decrease in total density of benthic macroinvertebrates with an average increase of 2°C in spring, summer and fall, and 3.5°C in winter. Therefore, a general assumption about the fate of

macroinvertebrate fauna for moderate water temperature increases is not possible.

1.2.5.2. Longitudinal effects of thermal discharges on macroinvertebrates

Thermal discharge effects are often limited to the immediate downstream areas (Coulter et al. 2014; Osborne and Davies 1987). For example, Worthington et al. (2015) measured 4.5 °C elevated water temperatures 2 km downstream of a power station outfall. However, abundance and taxa richness were only affected at sites 0.5 km downstream of the outfall. There were no significant changes in the benthic macroinvertebrate community greater than 2 km downstream. Similarly, Massengill (1976) observed a recovery of benthic fauna approximately 1 km below the discharge. Benda and Proffitt (1974) observed effects on benthic macroinvertebrate abundance and taxa diversity 152 m to 274 m below the discharge, which was on average 6 °C higher than control stations.

1.2.5.3. Thermal discharge and their effects on functional feeding groups

Few studies have researched the effects of thermal discharge on the functional feeding groups (functional feeding groups are described in detail in section 1.3.2) of benthic macroinvertebrates. LeRoy Poff and Matthews (1986) was the only study that reported the effects of thermal discharge on the response of functional feeding groups. They reported a change in functional feeding groups from collector-gatherers at control areas to scrapers at thermally-affected areas. However, areas affected only moderately by thermal release showed a different response with a change in community composition from collector-gatherers to collector-filterers (LeRoy Poff and Matthews 1986). These data were collected over a 48-day period during winter and did not provide information for other seasons. Given this, it appears that additional research is needed to understand effects of thermal discharges on the functional feeding groups of benthic macroinvertebrates at different seasons.

1.2.6. Future research

One opportunity for future research is to measure the long-term stream temperatures in relation to macroinvertebrate fauna. Most of the previously cited studies did not describe temperature measurement methods and lacked detail in terms of temperature reporting such as how frequently and with what instruments they measured the water temperature (Benda and Proffitt 1974; Coutant 1962; Dahlberg and Conyers 1974; Langford and Aston 1972; Massengill 1976; LeRoy Poff and Matthews 1986; Wellborn and Robinson 1996; Wurtz and Skinner 1984). Also, some researchers reported that water temperature was just measured during macroinvertebrate sampling (Coutant 1962) or measured once a month (Osborne and Davies 1987). This lack of daily water temperature reporting over the study time frame makes it difficult to relate water temperature with macroinvertebrate fauna and therefore can miss short term pulses of water temperature, which can eliminate all fauna. Therefore, a more detailed analysis regarding temperature measurements would help to compare different research outcomes for long-term studies. In addition, research regarding thermal discharge and their effects on functional feeding groups is limited in the literature and future research is needed to assess if impairments in the stream food-web are caused by thermal discharge.

1.3. Benthic macroinvertebrates

Benthic macroinvertebrates are aquatic animals that includes aquatic insects, clams, snails, worms, and crayfish (USEPA 1997). These organisms are used for biological monitoring in streams to assess long- and short-term effects of certain pollutants and to determine the habitat and water quality of the stream (Nedeau, Merritt, and Kaufman 2003; USEPA 1997). Macroinvertebrates are considered a useful way of monitoring biological conditions in a stream because they cannot escape pollution due to their limited mobility (Nedeau, Merritt, and Kaufman

2003; USEPA 1997). Thus, they are good biological indicators of long-term chemical, physical, and biological pollution. Different orders of macroinvertebrates show different tolerance to pollution and are classified into sensitive, somewhat sensitive, and tolerant taxonomic orders (MCWC 2005). Macroinvertebrates also play an important part in nutrient cycling, primary production, decomposition, and translocation (Wallace and Webster 1996) and are therefore food sources for numerous fish species and important in the stream-food web (USEPA 1997; Wallace and Webster 1996). Macroinvertebrates link many trophic levels. Therefore, impairments on macroinvertebrates could have both a top-down and a bottom-up effect on the food chain because they are consumers at the intermediate trophic level (Wallace and Webster 1996). Given their importance in the food web, changes in the thermal regime could lead to a reduction in size of macroinvertebrates with low fecundity and therefore lower their ability to compete (Vannote and Sweeney 1980).

1.3.1. Habitat

Benthic macroinvertebrates are found in all types of stream habitat such as riffles, runs, pools, and undercut banks, but usually prefer slow water habitats (Hershey et al. 2010). Macroinvertebrates live mostly at and in the water-streambed substrate interface (Hershey et al. 2010; Voshell 2002). They are also found on submerged wood and leaf packages. The substrate they inhabit can be organic or inorganic (Hershey et al. 2010). Organic substrates consist of leaf material and wood, while inorganic substrates range from silty sediments to boulders (Hershey et al. 2010).

1.3.2. Functional feeding groups

Most macroinvertebrates consume a range of foods including wood, algae, live vascular plants, detritus, and other animals (Voshell 2002). Because they have specific ways of obtaining

food, they are divided into functional feeding groups (Huryn 2009; Voshell 2002). The functional feeding groups include shredders, scrapers (grazers), collectors, engulfing predators, and piercers (Hershey et al. 2010; Huryn 2009; Voshell 2002). Shredders, scrapers, collectors, and piercer-herbivores are plant material eating macroinvertebrates (Voshell 2002). Engulfing-predator and piercer-predator macroinvertebrates are carnivores (Voshell 2002).

Shredders consume coarse particulate organic matter (CPOM) resulting from decomposing terrestrial litter and living macrophyte tissue (Hershey et al. 2010; Wallace and Webster 1996). They use COPM to produce fine particulate organic matter (FPOM) and dissolved organic matter (DOM), which is food for other macroinvertebrates (Huryn 2009; Wallace and Webster 1996). Grazers or scrapers feed on microbial biofilms attached to substrate including periphyton, diatoms, and other heterotrophic prokaryotes and eukaryotes (Hershey et al. 2010; Huryn 2009). They scrape the biofilm from the substrate surface (Wallace and Webster 1996). Grazer and algae abundance are interrelated and a lack in grazer population can lead to an increase of algae and is another example of macroinvertebrates having a top-down effect on the trophic level (Wallace and Webster 1996). Collectors are subdivided into collector-gatherers and collector-filterers (Huryn 2009). Collector-gatherers feed on FPOM on the streambed and collector-filterers feed on FPOM suspended in the water column (Huryn 2009). Piercers are divided into piercer-herbivores and piercer-predators (Voshell 2002). Piercer-herbivores feed on plant fluid by penetrating the plant tissue (Voshell 2002). Piercer-predators feed on animal fluids by sucking the fluids out of their prey (Voshell 2002). Their prey are usually larger than the predators and consist mostly of other invertebrates, although some can also prey on vertebrates such as fish and tadpoles (Voshell 2002). Engulfing predators eat living animals and forage on benthic and water-column habitat (Hershey et al. 2010; Huryn 2009).

1.4. Study area: Lower Rouge River

The Rouge River is a small urban watershed of up to 1210 km² catchment area with 203 km of stream (LOSAG 2001a; LTSAG 2001a). It is located in Southeast Michigan and a tributary to the Detroit River. The watershed encompasses the Washtenaw, Wayne, and Oakland Counties and contains 48 communities with 1.5 million residences (Figure 4). The Rouge River is divided into the four major branches: Main, Upper, Middle, and Lower Branch. The four major branches are divided into seven subwatersheds. The main branch consists of the subwatersheds Main 1-2 and 3-4, the Middle Branch of Middle 1 and 3, and the Lower Branch of Lower 1 and 2. As much as 50 % of the urbanization in the Rouge River watershed is concentrated along the Lower 2 and Main Branch in Dearborn, Melvindale, and Inkster (LTSAG 2001a). The undeveloped areas are concentrated more in rural areas at the east and southeast part of the Lower 1 and east part of the Middle 1 Branch such as Salem, Superior, and Van Buren (Figure 4, LOSAG 2001b).

The Rouge River suffered significantly from pollution and degradation in the past. Because of that, the Rouge River is a designated Area of Concern (AOC) and governed by management plans to improve water and habitat quality, such as the Rouge River Remedial Action Plan and seven subwatershed management plans (LTSAG 2001a).

The focus of this study is the Lower Branch of the Rouge River (Figure 4 and Figure 5). The Lower Branch is the most southerly branch of the Rouge River with a 246 km² catchment area, separated into Lower 1 and 2 subwatersheds, and shared by Washtenaw and Wayne counties (Figure 4, LOSAG 2001b; LTSAG 2001b).

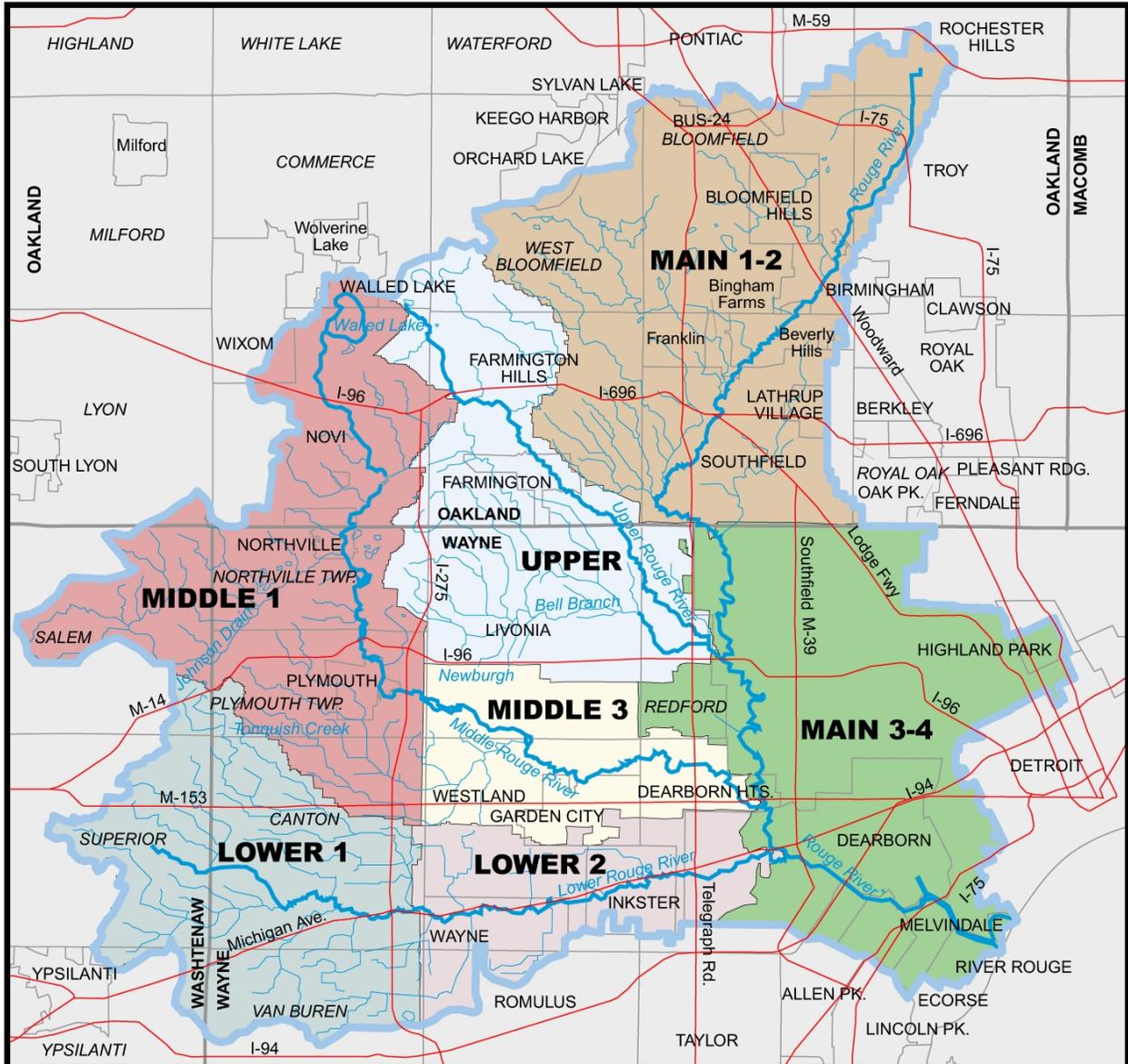


Figure 4. The Rouge River watershed with its four major branches the Main, Upper, Middle and Lower. The four major branches are also divided into the seven subwatersheds Main 1-2, main 3-4, Upper, Middle 1, Middle 3, Lower 1, and Lower 2 (LOSAG 2001a).

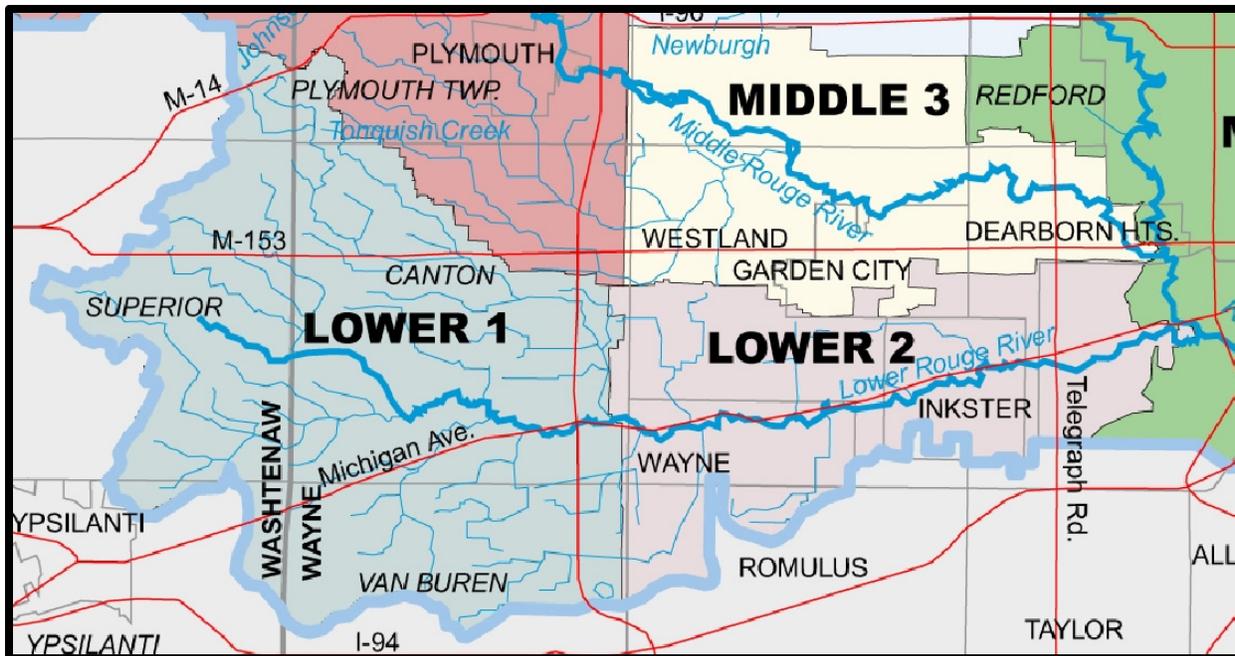


Figure 5. The Lower Rouge River and its two subwatershed Lower 1 and 2 (LOSAG 2001a).

1.4.1. Problem statement

The Lower Rouge River watershed suffered significantly from low baseflow conditions before 1996, especially during summer months (LOSAG 2001b; LTSAG 2001b; Wiley, Seelbach, and Bowler 1998). This resulted in lowest daily average flow ranging from 0.03 to 0.06 m³ (LTSAG 2001b; LOSAG 2001b; Wiley, Seelbach, and Bowler 1998). The low stream discharge rate led to nearly zero dissolved oxygen concentrations and dramatically decreased habitat quality (LOSAG 2001b; LTSAG 2001b). In 1996, the Ypsilanti Community Utilities Authority (YCUA) wastewater treatment plant started to discharge wastewater effluent into the Lower Rouge at Beck Rd in Canton, MI (LTSAG 2001b; LOSAG 2001b). The stream discharge increased to 0.71 m³ for the lowest daily average and increased dissolved oxygen concentration to up to 7 mg/l (LTSAG 2001b). The effluent discharge improved the flow regime and dissolved oxygen concentrations on the Lower Rouge River, but it could also serve as a source of pollution (LTSAG 2001b) by altering the thermal regime of the river.

1.4.2. Pollution in the Rouge River

Chemical and bacterial pollution in the Rouge River is highly prevalent due to the heavy industry adjacent to the watershed and the combined sewer overflows. Heavy metal contamination of the river bed is common in the Rouge River due to the industrial activities of over 100 years along the river (Murray et al. 1999) which are more prevalent at the soil surface at industrial sites close to the east side of the watershed (Murray, Rogers, and Kaufman 2004). Additionally, Murray et al. (1999) found out that the concentration of heavy metal in sediment particles increases with smaller particle size. Kannan et al. (2001) identified several organic pollutants in the sediments of the Lower Rouge such as polychlorinated naphthalene, biphenyl, dibenzo-*p*-dioxins, dibenzofurans and polycyclic aromatic hydrocarbons which were most likely caused by combined sewer outflow and industrial facilities along the river. Additionally, the impact of organic pollution on heterotrophic microbial communities is reflected in higher metabolic diversity (Tiquia 2011) and enzyme activities (Tiquia 2010). Another potential source of pollution into the Lower Rouge River are bacterial pollution coming from domesticated animal and wildlife runoff (Murray et al. 2001). Furthermore, Tiquia et al. (2007) found a shift towards halophilic and halotolerant bacteria communities resulting from the runoff of deicing salts into the Rouge River.

Previous studies on the Rouge River concentrated mainly on chemical and bacterial pollution, whereas Wayne County Michigan conducted research on the water temperature in the Rouge River. Wayne County recorded continuous temperatures from 1994 to 2005 at 8 locations in the Rouge River (Price 2014). From 2007 to 2012, continuous temperature measurements were taken at a minimum of one location within the Rouge River watershed (Price 2014). Temperatures at the Lower Rouge River were continuously measured from 1994 to 2005 at Hannan Rd (US9) in Wayne, MI and Military Ave. (L05D) in Dearborn, MI (Price 2014). Other continuous water

temperature measuring stations are the USGS Gauge 04168400¹ (Dearborn, MI), USGS gauge 04166500² (Detroit, MI), and USGS gauge 04167150³ (Dearborn Heights, MI).

There are a few sites in the Rouge River that are potential source of thermal pollution. The Main Branch receives thermal discharge from AK Steel in Dearborn before it drains into the Detroit River⁴. AK Steel is required to conduct a thermal plume study over four seasons for spring (March through June), summer (July through August), fall (September through November), and winter (December through February) for the recent National Pollutant Discharge Elimination System (NPDES) permit. The thermal plume study started in 2015 and concluded in August 2016. The study included four discharge locations and determined the temporal, spatial, thermal and volumetric characteristics of the thermal plume, and provided the volume, velocity, time of passage and time-temperature information in the intake facilities, in the discharge facilities, and in the centerline of the thermal plume.

1.5.Wastewater treatment plant in Ypsilanti

The wastewater treatment plant (WWTP) is located in southeast Ypsilanti, Michigan. The plant is owned and operated by the YCUA. YCUA provides wastewater treatment for the City of Ypsilanti, Ypsilanti Township, Augusta Township, Pittsfield Township, Sumpter Township, Superior Township, York Township, Canton Township, Northville, and Plymouth (YCUA 2012b). The last three communities are members of Western Township Utilities Authority (WTUA) in Wayne County which agreed with YCUA to expand the YCUA facility to be able to receive

¹ http://waterdata.usgs.gov/mi/nwis/uv/?site_no=04168400&PARAMeter_cd=00065,00060

² http://waterdata.usgs.gov/mi/nwis/uv/?site_no=04166500&PARAMeter_cd=00065,00060

³ http://waterdata.usgs.gov/mi/nwis/uv/?site_no=04167150&PARAMeter_cd=00065,00060

⁴ Information from Sally Petrella (FOTR).

sewage from these communities (YCUA 2012b). In addition, YCUA collects wastewater from the City of Ypsilanti and Ypsilanti Township (YCUA 2012b).

YCUA began operating in 1982, serving all the communities listed above except the WTUA (YCUA 2016). The WWTP was designed for an average daily discharge of 109.4 million liters per day (ML/d) (28.9 million gallons per day (mgd)) (YCUA 2012a). Initially, all of this treated wastewater was discharged to Willow Creek, adjacent to the WWTP and part of the Huron River Watershed. In 1988, the wastewater from the WTUA was added to YCUA. Because of this, a new discharge permit from the Michigan Department of Natural Resources (MDNR) was attained for an average flow of 219.6 ML/d (58 mgd). This resulted in the creation of an additional outfall into the Lower Rouge River. The permit was approved to allow discharge to the Lower Rouge River in order to prevent more nutrient loading into the Bellville Lake (YCUA 2012a; YCUA 2012b). The Willow Creek outfall is still in usage at high flow periods, which occurs just a couple of days of the year. Another reason to move the outfall was to provide higher water quality to the Lower Rouge River and to stabilize the base flow. A two-year nutrient assessment was done on the Lower Rouge River to analyze the impact of the effluent discharge on water quality (YCUA 2012b). The two-year assessment did not show any impairments to water quality. An additional two-year assessment will be conducted once the daily average discharge surpasses 109.4 ML/d (28.9 mgd).

At the time of writing (September 2016), the YCUA average flow capacity is 193.8 ML/d (51.2 mgd) and the average daily use is 41.6 ML/d (11 mgd) (YCUA 2012b; YCUA 2012a; YCUA 2016). The plant does not receive any wastewater from combined sewer overflows and receives only sewage input from separated sewer systems. Because of the lack of stormwater, the concentrated sewage could cause higher water temperatures due to the high microbial activity in

decomposing sewage. Also the water is pumped with a pressure sewer pipe into the Lower Rouge River, which can be another factor in increasing the water temperature.

The WWTP influent goes through primary, secondary, and tertiary treatment. before it is discharged into the Lower Rouge River (YCUA 2014). Tertiary treatment is used to remove fine particles to maximize the disinfection with ultraviolet light. After the treatment with ultraviolet light, the water is pumped to the Lower Rouge River and discharged at the Outfall at Beck Rd. Prior to ultraviolet light disinfection, three million gallons of the treated water is used by YCUA for cooling water, scrubber water, and cleaning water purposes. This water is treated with sodium hypochlorite and returned to the plant influent where it goes through primary, secondary, and tertiary treatment again. Due to the limited usage of sodium hypochlorite, the chlorine in the discharge is reported to be likely non-detectable using conventional measurements methods⁵.

The effluent requires a discharge permit and must meet the regulations for water quality standards set by the Michigan Department of Environmental Quality (MDEQ) (YCUA 2012b). The final effluent regulations set limits and monitoring requirements for carbonaceous biochemical oxygen demand (CBOD₅), total suspended solids, ammonia nitrogen (N), fecal coliform bacteria, total arsenic, total phosphorus, total mercury, pH, and dissolved oxygen (Table 1; YCUA 2012b). The MDEQ has no permit requirement for temperature and chlorine concentration of the discharged effluent.

⁵ Personal communication with Luther Blackburn, Director of Wastewater Operations & Compliance of YCUA.

Table 1. Effluent regulation by MDEQ (YCUA).

PARAMETER	MAX. LIMIT CONCENTRATION			MONITORING FREQUENCY
	monthly	7-day	daily	
CBOD₅	mg/L	mg/L	mg/L	
April-October	4.0	-	10	Daily
November	7.0	-	11	Daily
December-March	10	-	15	Daily
TOTAL SUSPENDED SOLIDS 8500	mg/L	mg/L	mg/L	
	20	30	-	Daily
NITROGEN	mg/L	mg/L	mg/L	
April	-	-	6.5	Daily
May-October	0.5	-	2.0	Daily
November	9.0	-	9.9	Daily
December-March	12	-	17	Daily
FECAL COLIFORM BACTERIA	cts/100 ml			
	200	400	-	Daily
TOTAL ARSENIC	ug/L	ug/L	ug/L	
	10	-	-	Weekly
TOTAL PHOSPHORUS	mg/L	mg/L	mg/L	
May-August	0.7	-	-	Daily
September-April	0.8	-	-	Daily
	12-Month Rolling Average			
TOTAL MERCURY	ng/l			
	5.0			Quarterly
	Min. Daily	Max. Daily		
pH	6.5	9.0		Daily
DISSOLVED OXYGEN	mg/L	mg/L		
April	7.0	-		Daily
May-March	6.0	-		Daily

The YCUA has reported that the discharge does not cause water quality impairments in the Lower Rouge River (YCUA 2012b). The increased base flow is believed to help fish in the Lower Rouge River, although peak flow during storm events may also impair fish communities (LOSAG 2001b). The Lower One Subwatershed Advisory Group (2001b) reports water temperature to support fish species. Data collected by Friends of the Rouge (FOTR) on November 14, 2014 showed average water temperatures of 1.8 °C upstream of the discharge, 16.1 °C at the effluent discharge and elevated water temperature approximately 24 km downstream of the discharge (Figure 6; Table 2). The effluent created a thermal discharge of up to 14.3 °C higher water

temperature in comparison to upstream water temperatures (Table 2). The effects of the elevated water temperature on aquatic ecosystems has not been researched and its impacts are currently unknown for the Lower Rouge River. Based on the data from other rivers, the discharge may have an impact on the thermal regime of the Lower Rouge River. In addition, the possible changes on the thermal regime may affect benthic macroinvertebrate community including the types of functional feeding groups.

Table 2. Water temperature at the Lower Rouge River on November 14th, 2014 (Data from FOTR).

SITE	LOCATION	TRIBUTARY	WATER TEMPERATURE [°C]
FOWL2	Before outflow at Beck Rd.	Fowler Creek	1.8
LR-2	Outflow at Beck Rd.	Lower Rouge	16.1
LR-12	S Morton Taylor Rd.	Lower Rouge	11.7
LR-6	Wayne Rd.	Lower Rouge	7.5
LR-10	Inkster Rd. – Lower Rouge Parkway Dr.	Lower Rouge	5.9

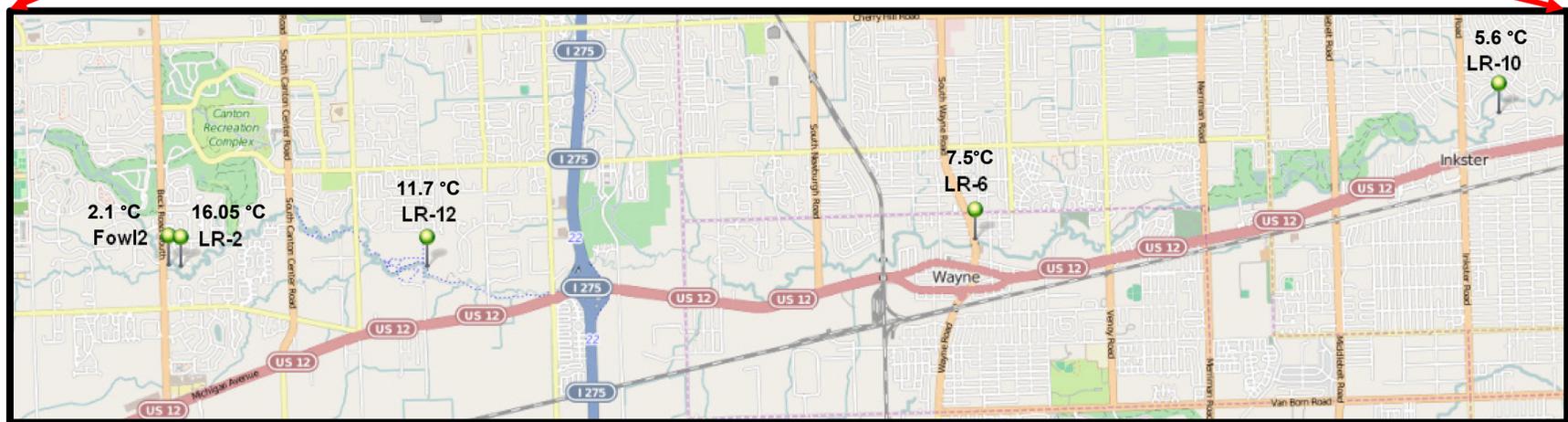
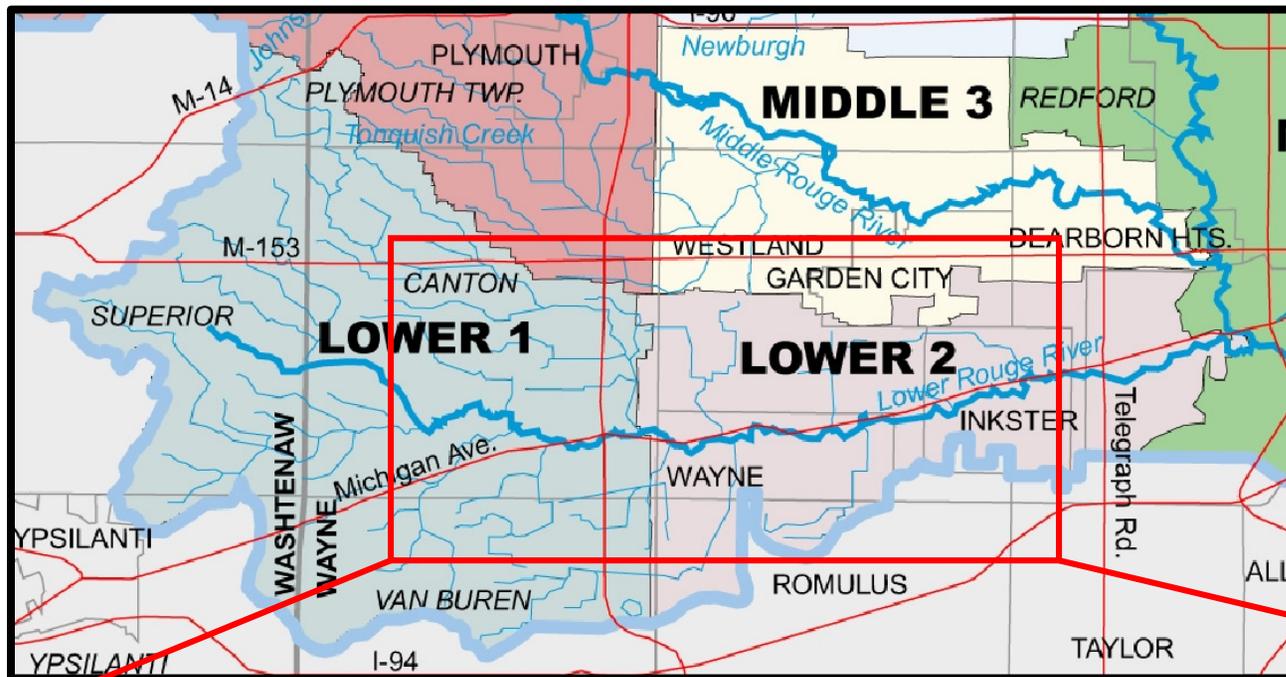


Figure 6. Water temperature in the Lower Rouge River on November 14th, 2014. The discharge is located at LR-2 with 16.06 °C and the lower temperature 2.1 are recorded upstream of the discharge at Fowl2. LR-12, LR-6, and LR-10 showed higher temperature than Fowl2 which are downstream of LR-2 (Courtesy from FOTR and Robert Muller).

1.6. Research focus

The Lower Rouge River has received effluent from the YCUA since 1996. The YCUA states that the discharge does not have any water quality impairments in the Lower Rouge River (YCUA 2012b), yet, there is no research regarding the effect of the YCUA discharge on the thermal regime and macroinvertebrate fauna. The YCUA was not required to do a thermal plume study for the NPDES permit⁶ as AK Steel in Dearborn and I did not find any published data regarding the temperature measurements at the YCUA Outfall. I also did not find any published research regarding temperature recording at the YCUA Outfall and comparing it with upstream temperature. Finally, I did not find any published studies comparing the impact of the discharges from the YCUA Outfall on the water temperature of the Lower Rouge River and the effects of the possibly changed water temperatures on the macroinvertebrate ecosystem. Therefore, the effects of the YCUA discharge on the thermal regime is unknown. It is important to research the effects of the YCUA discharge on the Lower Rouge River water temperature because even small changes in water temperature could have adverse effects on the macroinvertebrate fauna.

Most thermal discharge studies do not provide detailed stream temperature measurements (section 1.2.6). The thermal regime of rivers changes daily and seasonally and therefore a more detailed stream temperature recording would allow for the comparison of temperature data with the macroinvertebrate data. The focus in this study will be on the discharge introduced by the YCUA into the Lower Rouge River and their effects on the thermal regime and how the possibly changed thermal regime affects benthic macroinvertebrate fauna.

The effects of thermal discharges are usually not only related to increased water temperature, but also to contaminants from the discharge, changes in chemical properties,

⁶ Information from Luke Blackburn (YCUA employee).

turbulence or unusual water currents, and scour and siltation (Langford 1990). Thermal discharge is mostly chlorinated, which is most likely the cause of lower abundance of macroinvertebrates rather than elevated water temperatures (Langford 1990; Osborne and Davies 1987; Worthington et al. 2015). It is hard to separate the different influences on the ecosystem, but temperature in my opinion has many direct and indirect influences because it magnifies the properties of chemicals and their toxicity, and of biochemical and physiological properties. My research site uses ultraviolet light for disinfection and a limited amount of sodium hypochlorite for cleaning and cooling at the YCUA. Due to the limited usage of sodium hypochlorite, the chlorine will most likely be non-detectable by conventional methods⁷. Therefore, I did not address the effect of chlorination on benthic macroinvertebrates. For the purpose of this thesis, I concentrated on the impact of temperature. I compared temperature data from a control site to determine the possible effects of the YCUA discharge on water temperature and the possible effects of changed water temperatures on the macroinvertebrate community structure and functional feeding groups.

The purpose of this study was to measure the water temperature hourly at the YCUA Outfall and downstream areas to see if there is any difference in water temperature for spring, summer, and fall in comparison to the control site. Additionally, I sampled benthic macroinvertebrates in spring, summer, and fall to analyze for differences in family richness, diversity and functional feeding groups in relation to water temperature below the Outfall in comparison to the control site.

My motivation for this research was to determine if the discharge from the WWTP has an effect on the stream temperature of the Lower Rouge River and the effects of possibly water

⁷ Personal communication with Luther Blackburn, Director of Wastewater Operations & Compliance of YCUA.

temperatures change on its benthic macroinvertebrates. I assumed that the discharge could lead to higher temperature during summer below the Outfall which would have a greater impact on the benthic macroinvertebrates because stream temperature of the Lower Rouge River during summer is higher in comparison to the other seasons (Figure 7). To test this assumption, I sampled and analyzed the data collected in the spring, summer, and fall and determined the possible correlation with the seasonal temperature changes and the thermal impact of the WWTP discharge.

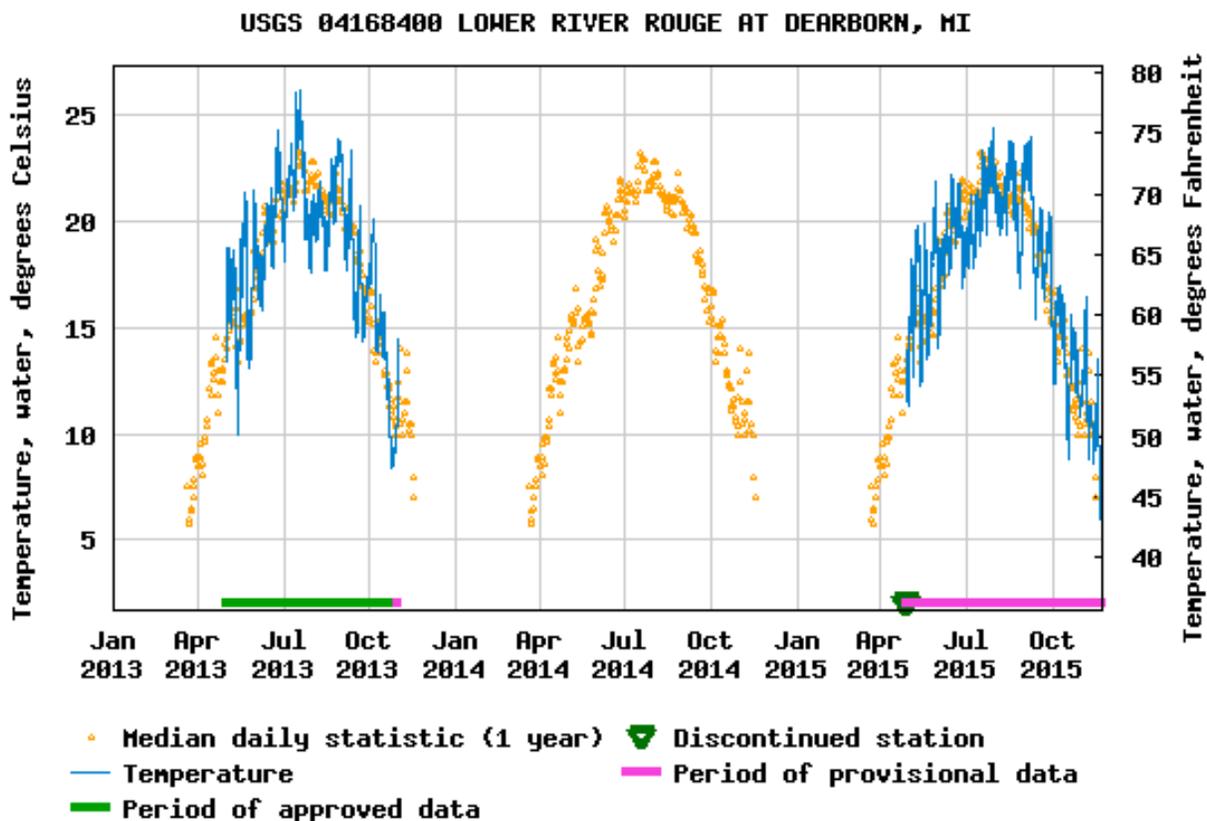


Figure 7. Median daily water temperatures at the Lower Rouge River between 2013 and 2015 measured by the USGS gauge 04168400 at Dearborn, MI. Daily mean water temperature for the year 2014 was statistically determined. (http://nwis.waterdata.usgs.gov/nwis/uv?cb_00010=on&format=gif_stats&site_no=04168400&period=&begin_date=2013-01-01&end_date=2015-11-23). Data from November 23th.

The specific questions I addressed through this research are as follow:

1. How does the effluent from the Ypsilanti Wastewater Treatment Plant affect the stream temperature downstream from the discharge at the Lower Rouge River in spring (April-June 15),

summer (June 16-August), and fall (September-October) 2015?

2. How does the water temperature affect the benthic macroinvertebrates community structure (taxon richness and diversity), and functional feeding groups structure downstream from the discharge into the Lower Rouge River in spring (April-June 15), summer (June 16-August), and fall (September-October) 2015?

A change to the thermal regime of the Lower Rouge River could change the benthic macroinvertebrates' community structure and possibly show a change and an impairment of the functional feeding groups.

I proposed seven general hypotheses to answer the above questions:

- 1) If the Outfall is affecting the water temperature, water temperatures will be lower for the control location Fowler 2 upstream of the Outfall.
- 2) If the Outfall is affecting the water temperature, the water temperature will be higher at the YCUA Outfall and decrease at downstream locations.
- 3) If the Outfall is affecting the water temperature, the difference in water temperature between the Outfall and Fowler 2 will be greater during spring and fall compared to summer.
- 4) If the Outfall is affecting the water temperature, family richness and diversity will be higher at the upstream location Fowler 2 compared to the YCUA Outfall.
- 5) If the Outfall is affecting the water temperature, family richness and diversity will increase in downstream direction from the Outfall.
- 6) If the Outfall is affecting the water temperature, family richness and diversity will be higher for spring and fall compared to summer.

7) If the Outfall is affecting the water temperature, the functional feeding groups diversity of macroinvertebrates will increase with decreasing water temperature.

CHAPTER II

2. MATERIALS AND METHODS

2.1 Sampling locations

I sampled at five locations along the Lower Rouge River (Table 3). The locations and site names were established from Friends of the Rouge (FOTR) sampling sites. The WWTP discharge from YCUA is at LR-2 (Figure 8). LR-12, LR-6, and LR-10 are downstream from the discharge site. Fowl2 is upstream of the discharge site and was used as a control site. Three downstream locations (LR-12, LR-6, and LR-10) were chosen to observe the influence of the Outfall (LR-2) longitudinally.

Table 3. Water temperature sensor and sample locations on the Lower Rouge River. Fowl2 is the control site.

LABELING	TRIBUTARY	LOCATION	GPS COORDINATES	DISTANCE FROM LR-2
LR-2	Lower Rouge	Outflow at Beck Rd.	42°16.91' N 83°30.208' W	0 km
LR-12	Lower Rouge	S Morton Taylor Rd.	42°16.927' N 83°27.966' W	Downstream 4.6 km
LR-6	Lower Rouge	Wayne Rd.	42°17.078' N 83°23.071' W	Downstream 11.2 km
LR-10	Lower Rouge	Inkster Rd. – Lower Rouge Parkway Dr.	42°17.931' N 83°18.420' W	Downstream 19.0 km
FOWL2	Fowler Creek	Before outflow at Beck Rd.	42°16.92' N 83°30.22' W	Upstream 60 m

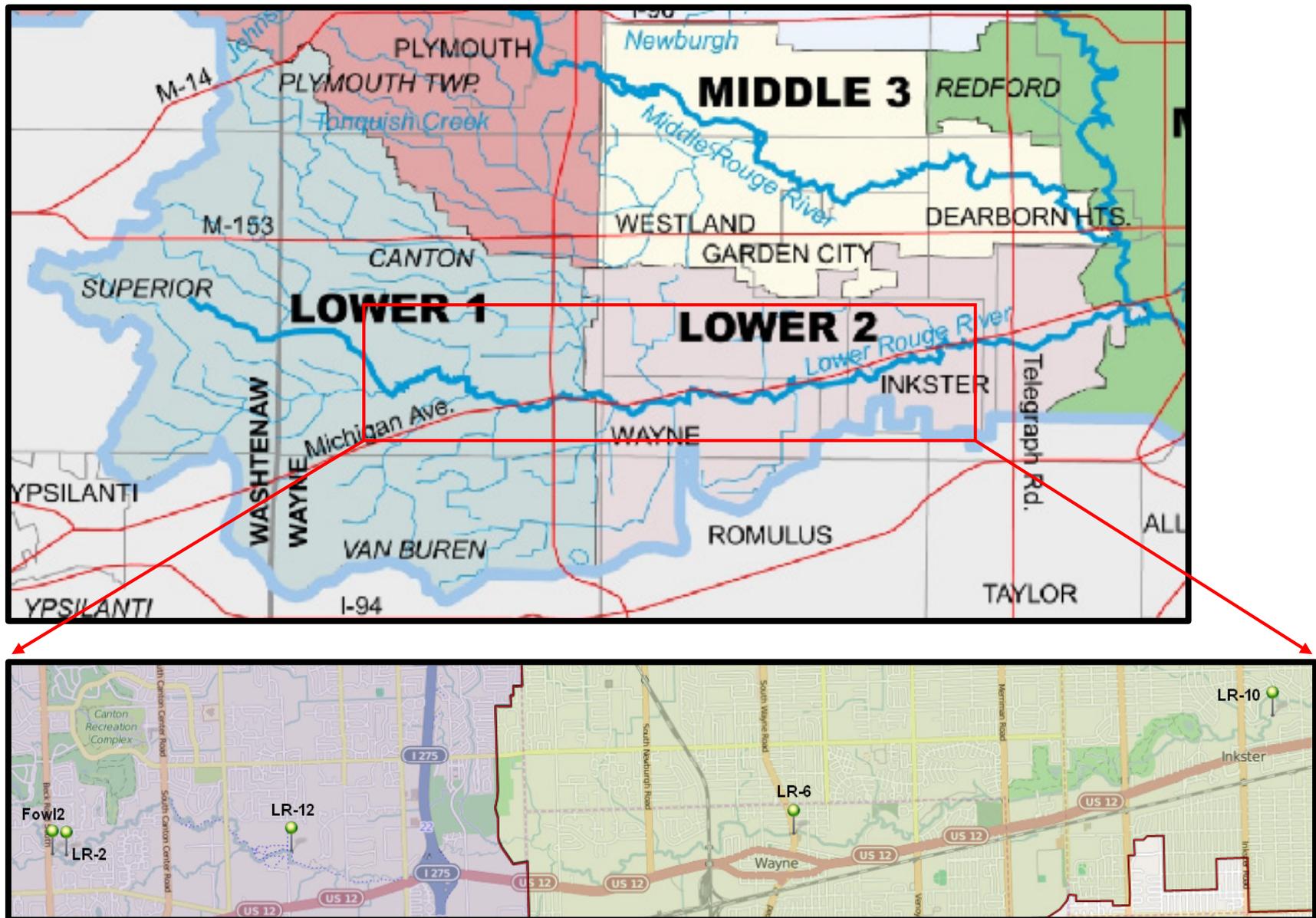


Figure 8. Sampling and temperature sensor locations on the Lower Rouge River. Temperature sensors were installed at Fowl2, LR-2, LR-12, LR-6, and LR-10.

2.2 Air and water temperature determination

2.1.1. Air temperature

Air temperatures were obtained from the National Oceanic and Atmospheric Administration (NOAA) from station GHCND:USC00202015 in Dearborn, MI for the time frame from April to October 2015 (Menne, Durre, Korzeniewski, et al. 2012c; Menne, Durre, Vose, et al. 2012). The station measures air temperature three times in 24 hours, recording the daily maximum and minimum air temperatures as well as the temperature at the time of observation. All available data from 13 April to 26 October 2015 were used except air temperature data from 24 July and 7 September 2015, which were not reported by NOAA.

2.1.2. Water temperature

For the water temperature measurements, I used TidbiT® v2 Water Temperature Data Loggers from Onset Computer Corporation and installed them at the sites LR-2, LR-12, LR-6, LR-10, and Fowl2 starting on 13 April, 2015 (Figure 8). The temperature readings were taken through 26 October, 2015 and were recorded hourly at each site. The TidbiT® v2 Water Temperature Data Loggers were company-calibrated following ISO standards⁸ with an accuracy of $\pm 0.21^{\circ}\text{C}$ for temperatures between 0 to 50 °C. The temperature sensors were placed in PVC casings with 1 cm hole on to top, bottom, and sides and anchored to the land with a steel cable (Figure 9). The PVC casings were additionally connected with 30 cm of steel cable to an automobile brake rotor to stabilize to the location (Figure 10). The temperature sensors were submerged in the water column approximately 30 cm above the riverbed to prevent sedimentation of the temperature sensor over time. The sensors were camouflaged between tree and bush roots to prevent public destruction. Towards the end of the recording period, the steel cables needed to be replaced by chains because

⁸ Obtained Information from Onset Computer Corporation's technical support.

of corrosion. I recorded daily maximum and minimum temperatures and calculated daily mean temperature and standard deviation for 24 hours to analyze the daily water temperature variation at each location. The data were uploaded with the HOBOWare® Pro graphing and analysis software version 3 at each location using the HOBO Optic USB Base Station connected via USB cable to the Computer. The HOBO Optic USB Base Station was attached to an ONSET coupler for TidbiT® v2 Temp data logger to read the water temperatures from the temperature data loggers.

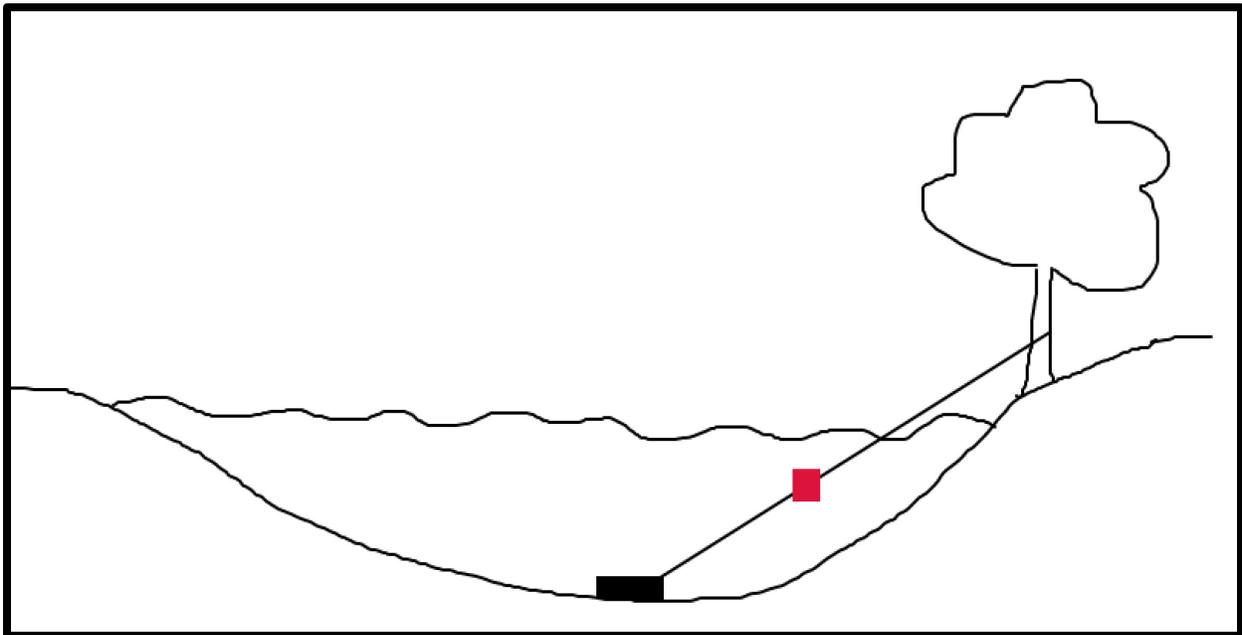


Figure 9. A drawing to visualize how the temperature sensor was placed in the river. Red is the PVS casing with the temperature sensor inside and black is the break rotor to stretch the chain in order to have the temperature sensor in the water column.



Figure 10. Robert Muller holding the PVC casing with the temperature sensor inside and the break rotor.

2.3. Discharge rate from waste water treatment plant and the Inkster, MI gauge

To compare the discharge from the WWTP to the total discharge of Lower Rouge River, I analyzed the daily discharge rate of the WWTP and the Lower Rouge River at Inkster, MI. The Ypsilanti Community Utility Authorities provided the discharge of its wastewater into the Lower Rouge River in million gallons per day (mgd) from April to October 2015. I converted the daily discharge from mgd to cubic meter per second (m^3/s). To compare the discharge rate from the wastewater treatment plant into the Lower Rouge River to the natural discharge rate of the Lower Rouge River, I used the daily discharge rates from April to October given by the United States Geological Survey (USGS) (USGS 2016) 04168000 gauge in Inkster, MI. The Inkster gauge is

located approximately 18.5 river km downstream from the Outfall at Jeffrey Lane in Inkster, MI (Figure 11). I plotted both discharge rates seasonally for spring, summer, and fall to compare the water volume of the wastewater treatment plant discharge to the gauge in Inkster, MI. I also calculated the average discharge rate and standard deviation for spring, summer, and fall for both locations to analyze the variability in water volume.

Precipitation data from the NOAA stations GHCND:US1MIWY0031 (Menne, Durre, Korzeniewski, et al. 2012a; Menne, Durre, Vose, et al. 2012) and GHCND:US1MIWY0068 (Menne, Durre, Korzeniewski, et al. 2012b; Menne, Durre, Vose, et al. 2012) in Canton Michigan were obtained for high peak and mid to low range peak events for the dates May 6, June, 15, 16, August 3, September 4 and 19, 2015. The precipitation data were averaged between the two Canton, MI locations. Both stations were approximately 4.5 km NE of the Outfall.

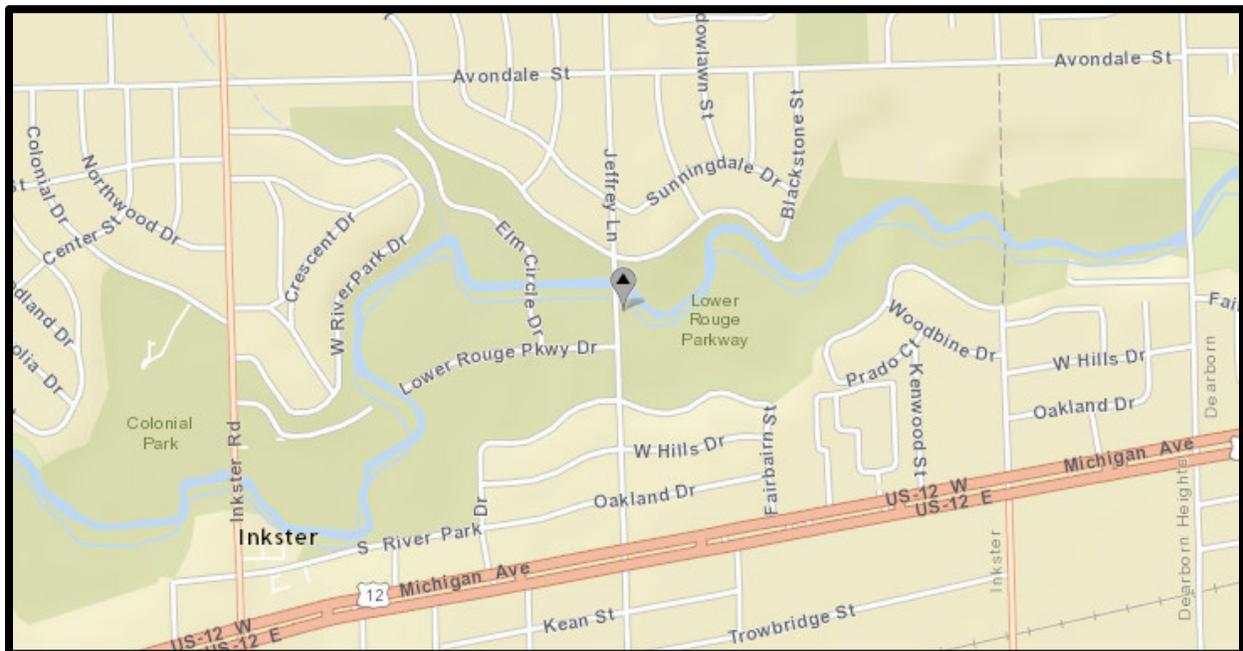


Figure 11. US Geological Survey (USGS 2015) 04168000 gauge at Jeffrey Lane in Inkster, MI.

2.4. Benthic macroinvertebrate sampling and analysis

Since 1998, FOTR and the Wayne County, Department of Public Services - Water Quality Management Division have conducted biomonitoring for benthic macroinvertebrates every spring and fall at the Rouge River. The FOTR collect their data in spring and fall because they provide the highest macroinvertebrate abundance in the water. The thermal regime of temperate stream and rivers varies seasonally for temperate regions because solar radiation changes over the seasons (Caissie 2006; Dallas 2008). Solar radiation contributes the most to the thermal regime of rivers and streams and its input decreases from fall to winter and increases from spring to summer. Because temperature is an important factor affecting the life history of benthic macroinvertebrates (Wallace and Anderson 1996), seasonal sampling of macroinvertebrates is a common practice in temperate regions for both in Europe (Friberg et al. 2006) and some parts of the United States. Seasonal sampling by researchers and both governmental and non-profit entities is routinely carried out to characterize the seasonal variation of taxonomic composition and abundance of benthic macroinvertebrates and to monitor water quality and habitat quality (Bêche, McElravy, and Resh 2006).

I sampled macroinvertebrates at the Lower Rouge River in spring, summer, and fall to observe any effects on the benthic macroinvertebrates caused by a possible thermal regime change and analyzed the community diversity, family richness, and the functional feeding groups of benthic macroinvertebrates at each site. Winter sampling was not conducted for several reasons. First, the winter in Michigan is characterized by high snowfalls and freezing conditions. The Lower Rouge River is often frozen making some of the typical sampling locations hard to access. In addition, the frozen river is a potential safety hazards and makes it difficult to collect adequate and accurate samples at each site which makes it difficult to generate a representative assessment

of the benthic macroinvertebrate community.

I sampled during summer because I hypothesized that the discharge will possibly cause high water temperature with deleterious effects on the benthic macroinvertebrates during summer. This approach is supported by most studies that have found that thermal discharge has the most deleterious effect during summer months with water temperature ranging between 40 and 42 °C - causing decreases in the abundance of macroinvertebrates (Coutant 1962; Durrett and Pearson 1975; Wellborn and Robinson 1996). Spring and fall were sampled because they have the highest abundance of benthic macroinvertebrates and the FOTR is biomonitoring during these seasons.

2.4.1. Macroinvertebrate sampling time periods

Benthic macroinvertebrates were sampled once at each site during spring, summer, and fall 2015. Spring and fall samplings for the site Fowl2 took place during the FOTR's Bug Hunt on 18 April and 17 October, 2015. The remaining sites were sampled by the Wayne County, Department of Public Services – Water Quality Management Division. During spring, the site LR-2, LR-12, and LR-6 were sampled on 17 April and the site LR-10 on 20 April 2015. During fall, the site LR-2 and LR-12 were sampled on 13 October and LR-6 and LR-10 on 20 October, 2015. Additional summer sampling was completed with staff and volunteers from FOTR on 6 July, 2015 for the locations Fowl2, LR-2, and LR-12 and 7 July, 2015 for LR-6, and LR-10. Because the FOTR Bug Hunt took place around the same time each year, I was able to use their data for comparison. Sampling times were also selected according to weather conditions and with consideration for personal safety.

The Lower Rouge River is a flashy river, which reaches high water levels quickly; especially after the winter season when snowmelt results in high discharge rates in March (Figure 12). The dates for summer sampling were on 6 July and 7 July, 2015 because this period was the

earliest time to sample in summer after high discharge rates in June. Fall sampling was conducted on the same dates that FOTR conducted the benthic macroinvertebrate monitoring.

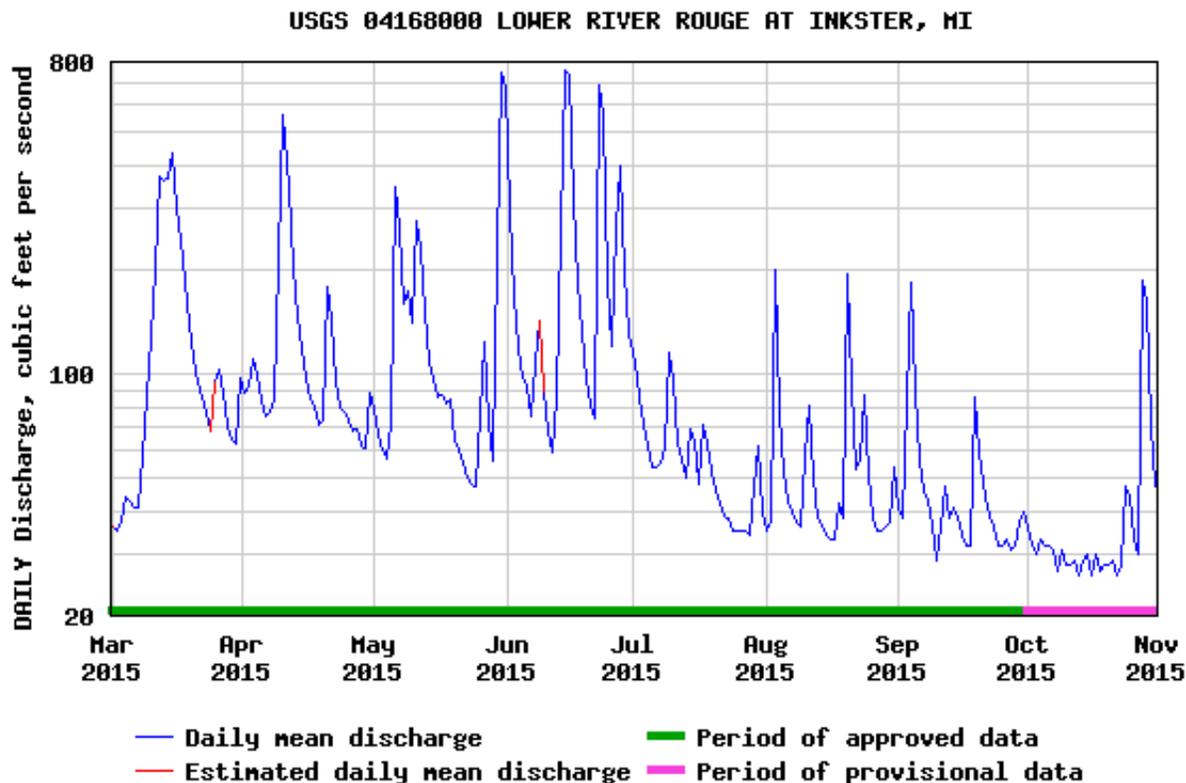


Figure 12. Discharge rate in cubic feet per second at USGS gauge 04168000 Inskter, MI from March to October 2015 (USGS 2016).

2.4.2. Macroinvertebrate sampling method

Benthic macroinvertebrates were sampled at Fowl2, LR-2, LR-12, LR-6, and LR-10 (Table 3). Collected macroinvertebrates were identified to the order level and counted in the field. Voucher specimens were collected and preserved in 95 % ethanol for identification to the family level in the lab.

In order to sample for benthic macroinvertebrates, I used a D-Frame net with a 1.6 mm mesh size. This was used because the Lower Rouge River is mostly a soft-bed river and a multi-habitat approach using a D-Frame net was recommended (Barbour et al. 1999; Stark et al. 2001).

At each location, 30 m of the stream area were sampled for 30 minutes, working from downstream to upstream. Techniques for sampling included placing the D-Frame net downstream and kicking the streambed 3 to 5 times to dislodge the benthic macroinvertebrates from river sediments. Each sampling location mostly consisted of runs and pools. In addition to soft bottom sampling in runs and pools, I scraped rocks and log jams, undercut banks and overhanging vegetation or roots. I placed the samples in trays and picked out the organisms with forceps and plastic spoons and sorted to order level. At each location, we filled 10 to 11 trays with samples.

2.4.3. Macroinvertebrate analysis

Taxonomic classification of macroinvertebrate samples to the nearest family was carried out using a dissecting microscope, keys and description by Bouchard (2004). The spring and fall macroinvertebrates were identified by Bruce McCulloch (volunteer Biologist, FOTR) and Sue Thompson (Department of Public Services Water Quality Management Division, Wayne County). The summer samples were identified by me. Some families were also identified to the genus level using Merritt et al. (2008). Identification to the family and some to the genus level led to the characterization of functional feeding groups (Barbour et al. 1999; Bode et al. 2002). The benthic macroinvertebrate families Heptageniidae, Haliplidae, Hydropsychidae, Veliidae, Hydrophilidae, and Belostomatidae were identified to the genus level.

2.4.4. Historical comparison

The number of families (taxa/family richness) from spring and fall of 2012-2014 (data by FOTR) from each site were analyzed and compared to my 2015 data to determine if my data were unusual to the previous three years in terms of macroinvertebrate family richness and composition. However, FOTR did not sample Fowl2 during spring 2012, Outfall during spring 2013 and fall 2013, and LR-6 during fall 2014. For spring 2012 for Fowl2, I used the last available data which

was from spring 2010. The past three years did not provide individual numbers in each family and they were reported as R for rare with individuals between 1 to 10 and C as common for individuals over 10. Unfortunately, historical water temperatures from the sampling locations are not available. The USGS Inkster gauge has historical water temperature for data for the years 2013 and 2015. However, the USGS Inkster gauge is too far downstream from the Outfall to be used to evaluate the potential impact of the discharge on water temperature.

2.5. Statistical analyses

2.5.1. Temperature

I used descriptive statistics to determine the daily mean, daily maximum, daily minimum water temperature and standard deviation (stdev) for the daily mean water temperatures for every location in order to compare the different daily mean water temperature of each location and their deviation from the Outfall (LR-2) temperature. I evaluated the water temperature in connection with the air temperature data from the National Oceanic and Atmospheric Administration from 2015 (Menne, Durre, Korzeniewski, et al. 2012c). However, air temperature data from 24 July and 7 September, 2015 were not reported by NOAA. The air temperatures were reported in daily maximum and minimum temperature. I calculated the mean air temperature by using the following formula:

$$T_A = \frac{T_{min} \times T_{max}}{2}$$

T_A is the daily average air temperature, T_{min} is the daily minimum air temperature, and T_{max} is the daily maximum temperature. I plotted the mean daily water and air temperature of all locations monthly and seasonally for spring (13 April – 15 June), summer (16 June– 31 August), and fall (1 September – 26 October). However, I used the seasonal data for the statistical comparison of the different locations. The end of spring season was determined by observing mean air temperatures

in June and designated them to either spring and summer. Before 15 June, 2015, the air temperatures were closer to spring air temperatures.

The daily mean water temperature data were tested seasonally for normal distribution with a z-test to evaluate skewness, kurtosis, and standard error (Kim 2013). I used the independent Student's *t*-test with equal variances in Excel 2016 to compare the water temperature between the Outfall (LR-2) and the control location Fowl2. I used the independent Student's *t*-test because Fowl2 and the Outfall receive water from different sources and were normally distributed. Additionally, the variances were unknown. The *t*-test was performed between Fowl2 and LR-2 for spring, summer, and fall. Additionally, I used the independent Student's *t*-test with equal variances to compare Fowl2 and the Outfall with the downstream locations LR-12, LR-6, and LR-10. I chose the independent Student's *t*-test because the downstream locations are 4.5 to 19 river km downstream from the Outfall and therefore were treated independently in regards to geographic location.

2.5.2. Macroinvertebrate statistical analyses

I determined the total number of families, and calculated the Shannon diversity index and, Shannon evenness for macroinvertebrate families and functional feeding groups for spring, summer, and fall. The numbers of each functional feeding group for each location were also determined in spring, summer, and fall. Also the previous three years of macroinvertebrate sampling data from spring and fall given by the FOTR were used to determine variation over the three years and compare them with recent conditions. Also, I used the Bray-Curtis Index to determine the percentage similarity in macroinvertebrate families between the Outfall and Fowl2, between Fowl2 and the downstream, and between Outfall and the downstream sites. In addition, the Bray-Curtis Index was used to analyze the percentage similarity of the functional feeding

groups individuals of Fowl2 to the Outfall, LR-12, LR-6, LR-10 and Outfall to LR-12, LR-6, and LR-10. Covariance and correlation were used to compare the mean daily temperature of the Outfall and the downstream location to Fowl2 and the Outfall. Additionally, all locations daily mean water temperatures were compared to the daily mean air temperatures with covariance and correlation.

The following formula was used for the Shannon Diversity Index (Heip, Herman, and Soetaert 1998):

$$SDI = - \sum_{i=1}^S p_i \times \ln p_i$$

where SDI is the Shannon Diversity Index, S is the total number of macroinvertebrates in the sample, and p_i is the number of individual families or genera (for functional feeding groups diversity it is the individual numbers of macroinvertebrates in each functional feeding group) in proportion to the total macroinvertebrate number in the sample.

The formula below was used for the Shannon Evenness (Heip, Herman, and Soetaert 1998):

$$SE = \frac{SDI}{\ln S}$$

where SE is the Shannon Evenness and S is the number of families (number of functional feeding groups) at each location.

The formula below is the Bray-Curtis dissimilarity (Greenacre and Primicerio 2013):

$$b_{ii'} = \frac{\sum_{j=1}^J |n_{ij} - n_{i'j}|}{n_{i+} + n_{i'+}}$$

where $b_{ii'}$ is Bray-Curtis dissimilarity, n_{ij} are the individual numbers of a family member (functional feeding groups member) at location i, $n_{i'j}$ is the individual numbers of a family member (functional feeding groups member) at location i', n_{i+} is the total number of individuals at location i, and $n_{i'+}$ is the total number of individuals at location i'.

To calculate the percentage of similarity also called Bray-Curtis Index ($B_{ii'}$), I used the formula:

$$B_{ii'} = 100 - 100 \times b_{ii'}$$

CHAPTER III

3. RESULTS

3.1. Water temperature

To compare the data with seasonal benthic macroinvertebrate sampling data, I separated the temperature data into spring, summer, and fall categories. The seasons covered spring from 13 April to 15 June (Figure 13), summer from 16 June to 31 August (Figure 14), and fall from 1 September to 26 October (Figure 15). As stated in the Materials and Methods, Fowl2 is referred to as control, LR-2 as the Outfall, and LR-12, LR-6, and LR-10 as the downstream locations. The statistical analyses are described in sections 3.1.2 and 3.1.3. The mean daily water temperatures are presented by month in Appendix 1.

3.1.1. Mean daily water temperature by seasons

3.1.1.1. Spring

The control site Fowl2 had a mean water temperature of $15.47\text{ }^{\circ}\text{C} \pm 3.90$ for spring (Table 4), which increased towards the end of spring (Figure 13A). The curve shape was characterized by alternating low and high peak mean daily temperatures over a weekly basis. However, the mean daily water temperature at the Outfall site (LR-2) ($15.68\text{ }^{\circ}\text{C} \pm 1.62$) showed less day-to-day variation as the temperature gradually increased throughout the spring (Figure 13B). Although the Outfall and Fowl2 had different mean daily water temperature variations, they had a similar pattern of increase throughout the season (Figure 13). The Outfall had a higher seasonal water temperature

of 0.22 °C on average for spring (Appendix 2).

In comparison to the downstream locations, the Outfall mean water temperature increased steadily during the spring period and varied little by week. The downstream locations showed a parallel and almost overlapping mean daily water temperature trend (Figure 13). In comparison to the Outfall, they showed higher variation with a 1.5-times larger standard deviation (Table 4). The downstream locations LR-12 with 0.12 °C and LR-6 with 0.03 °C on average showed higher seasonal water temperatures than the Outfall for spring (Appendix 3). LR-10 did not show any mean water temperature difference compared with the Outfall during spring.

Fowl2 and the downstream locations LR-12, LR-6, and LR-10 showed a similar mean daily water temperature trend (Figure 13, A, C, D, E respectively). The curves almost overlap and the mean daily water temperature varied little after the first third of spring. Fowl2 and the downstream locations showed a greater similarity in daily mean water temperature patterns compared to the Outfall. The downstream locations showed higher temperatures than Fowl2 by 0.33 °C for LR-12, 0.25 °C for LR-6, and 0.21 °C for LR-10 (Appendix 2).

In comparison to mean daily water temperatures, mean daily air temperatures showed both higher and lower temperature peaks due to the difference in thermal capacity between air and water (Figure 13). In addition, the air temperature peaks occurred generally before the water temperature peaks (Figure 13).

Table 4. Mean temperature (mean T) in °C and standard deviation (STDEV) with maximum and minimum average water temperature for spring from 13 April to 15 June 2015. Temperatures mean was calculated from mean daily water temperatures from spring. LR-2 is the Outfall.

	FOWL2	LR-2	LR-12	LR-6	LR-10	AIR
MEAN T	15.47	15.68	15.80	15.71	15.68	15.28
STDEV	3.90	1.62	2.58	3.12	3.23	5.82
MAX	21.78	18.36	21.47	21.59	21.77	21.78
MIN	6.11	11.75	9.92	8.14	7.79	6.11

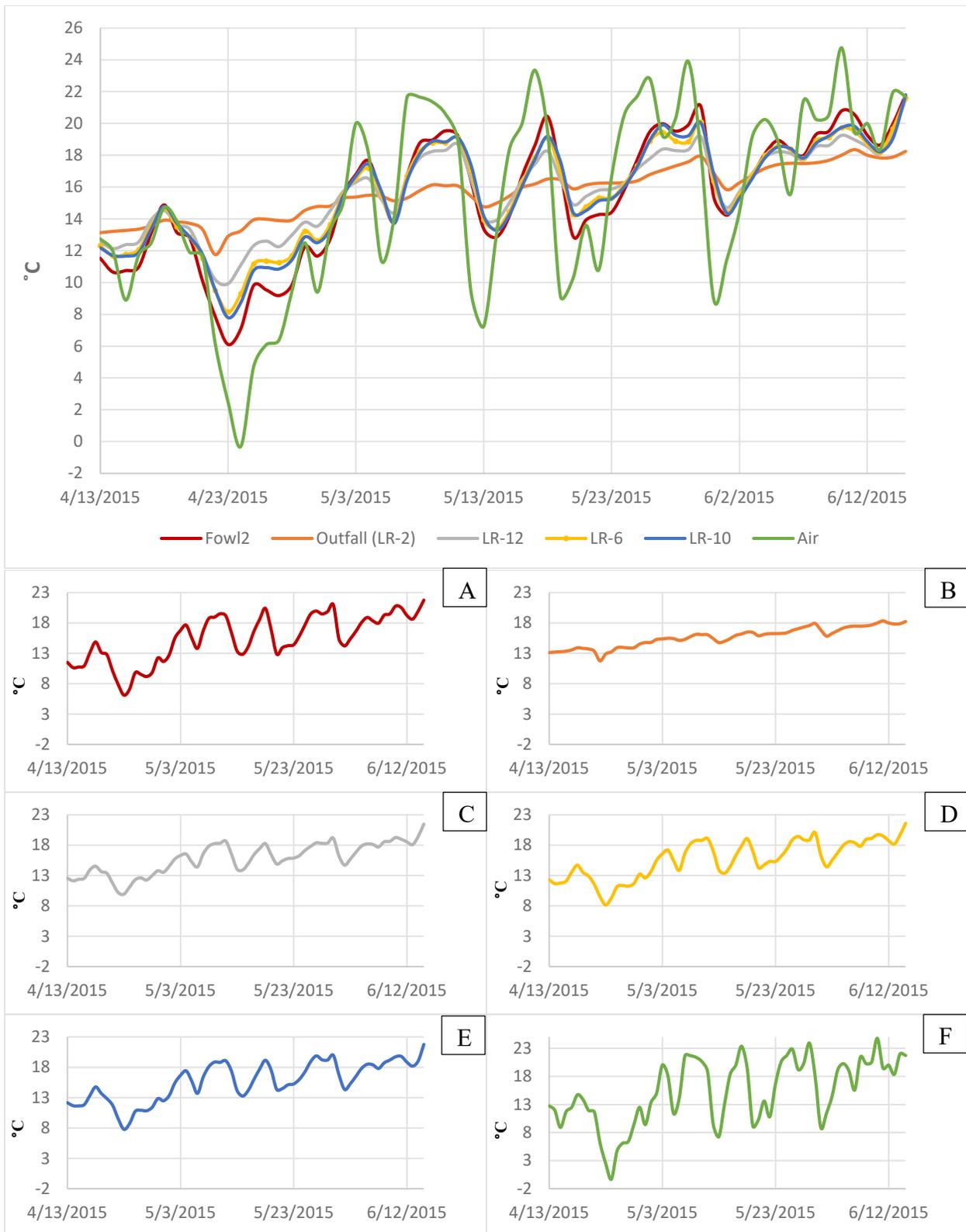


Figure 13. Mean daily water and air temperatures for spring measured in °C from 13 April to 15 June 2015. The water temperature was measured hourly and averaged for a 24-hour period. The mean daily air temperature was calculated from daily maximum and minimum air temperatures. The top graph shows all the following temperatures. A) Fowl2, B) LR-2, C) LR-12, D) LR-6, E) LR-10 and F) air.

3.1.1.2. Summer

During summer months, the water temperature of Fowl2 varied from day-to-day (Figure 14). The mean daily water temperature of $20.59\text{ }^{\circ}\text{C} \pm 1.57$ with its highest mean daily water temperature on 19 July 2015 of $23.65\text{ }^{\circ}\text{C} \pm 0.86$ and its lowest mean daily water temperature on 28 August 2015 of $16.67\text{ }^{\circ}\text{C} \pm 1.18$ (Figure 14A). Fowl2 ($20.59\text{ }^{\circ}\text{C} \pm 1.57$) had a higher mean water temperature for summer compared to the Outfall ($19.79\text{ }^{\circ}\text{C} \pm 1.01$) (Table 5). The mean daily water temperature at the Outfall was almost constant with an increase of $2.86\text{ }^{\circ}\text{C}$ by the end of summer. Towards the end of summer, the Outfall had a higher mean daily water temperature than Fowl2 of $21.57\text{ }^{\circ}\text{C} \pm 0.17$ on 31 August 2015. Fowl2 had water temperatures that were on average $0.81\text{ }^{\circ}\text{C}$ higher than those recorded at the Outfall (Appendix 2).

The downstream locations had parallel and almost overlapping water temperatures (Figure 14). The mean water temperature for the Outfall was lower compared to the downstream locations (Figure 14). Over the course of the summer, the Outfall showed a constant rise in mean water temperature. In contrast, the three downstream locations had greater day-to-day variations in water temperature over the summer period. Towards the end of summer, the Outfall had a higher mean daily water temperature than the downstream locations, which reached their summer low water temperature on 26 August 2015 for LR-12 and LR-6 and on 27 August 2015 for LR-10. In general, the Outfall showed low similarity to the downstream locations, even though modest parallel day-to-day changes were observed. The downstream locations had higher average temperatures of $0.47\text{ }^{\circ}\text{C}$ for LR-12, $0.70\text{ }^{\circ}\text{C}$ for LR-6, and $0.83\text{ }^{\circ}\text{C}$ for LR-10 during the summer (Appendix 3).

Fowl2 and the three downstream locations had similar mean water temperatures and similar trends in daily mean water temperature patterns (Figure 14). In general, the average water temperatures of the three downstream locations were $20.26\text{ }^{\circ}\text{C} \pm 0.97$ for LR-12, $20.49\text{ }^{\circ}\text{C} \pm 1.25$

for LR-6, and $20.62\text{ }^{\circ}\text{C} \pm 1.41$ for LR-10. These temperatures were similar to the average water temperature at Fowl2 ($20.59\text{ }^{\circ}\text{C}$) (Table 5). The mean water temperature and the standard deviation of Fowl2 and LR-10 were the most similar (Table 5). Compared to Fowl2, the downstream locations LR-12 and LR-6 had lower water temperature of $0.34\text{ }^{\circ}\text{C}$ and $0.10\text{ }^{\circ}\text{C}$, respectively (Appendix 2).

During summer, the average air temperature had a higher variation than water temperature (Figure 14). The curve is characterized by high and low alternating temperatures. The pattern of variation in air temperature is similar to variations in water temperature at Fowl2 and downstream locations; however, the high and low peaks in air temperature slightly preceded the peaks in water temperatures. The Outfall showed the lowest similarity to the patterns of variation of air temperature, even though modest parallel movements were observed. The downstream locations had on average higher water temperatures than the Outfall during summer by $0.47\text{ }^{\circ}\text{C}$ for LR-12, $0.70\text{ }^{\circ}\text{C}$ for LR-6, and $0.83\text{ }^{\circ}\text{C}$ for LR-10 (Appendix 3).

Table 5. Mean temperature (mean T) in $^{\circ}\text{C}$ and standard deviation (STDEV) with maximum and minimum average water temperature for summer from 16 June to 31 August 2015. Temperature mean was calculated from mean daily water temperatures from summer. LR-2 is the Outfall.

	FOWL2	LR-2	LR-12	LR-6	LR-10	AIR
MEAN T	20.59	19.79	20.26	20.49	20.62	20.94
STDEV	1.57	1.01	0.97	1.25	1.41	2.82
MAX	23.65	21.57	22.14	22.96	23.20	23.65
MIN	16.67	17.83	17.62	17.45	17.42	16.67

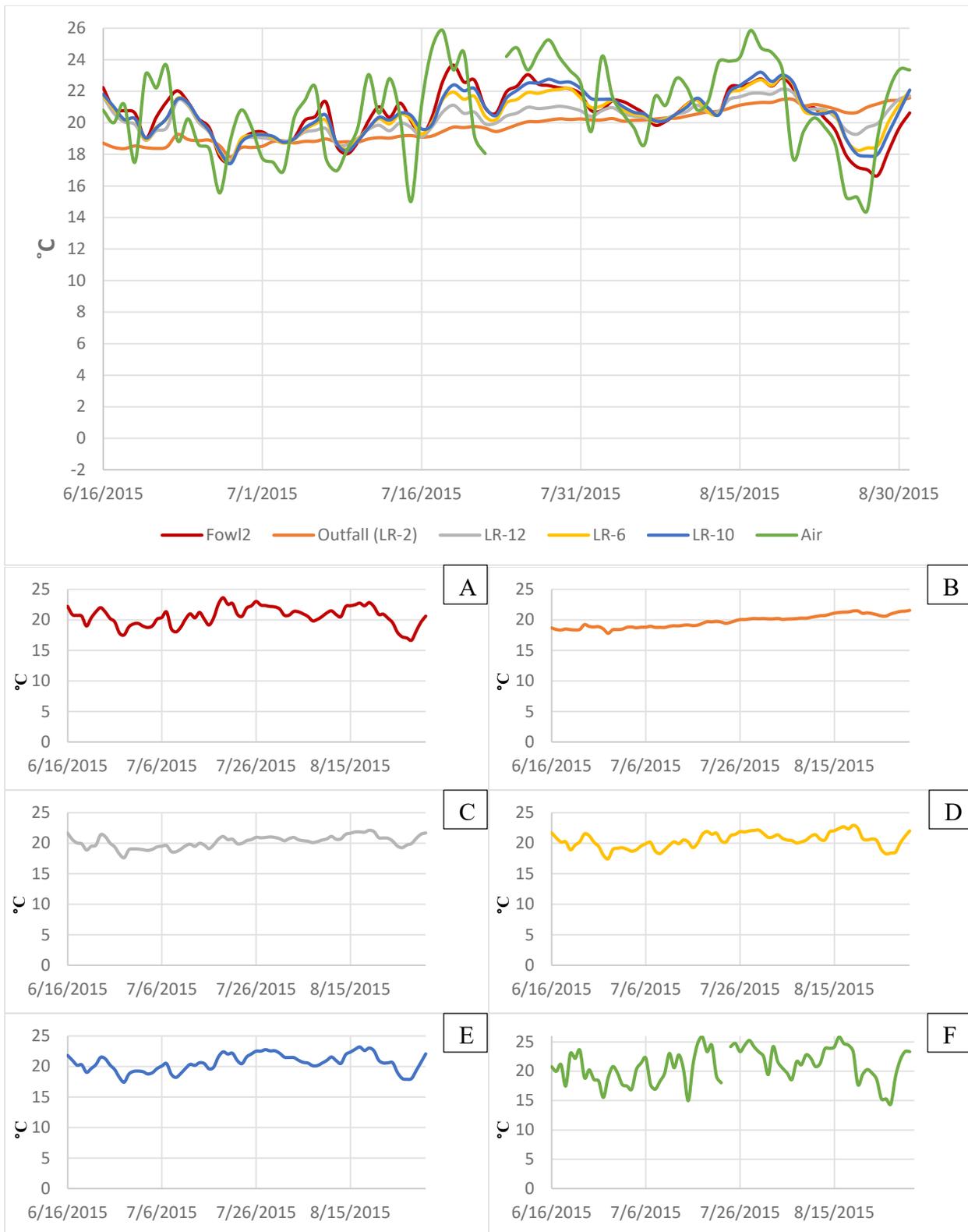


Figure 14. Mean daily water and air temperatures for spring measured in °C from 16 June to 31 August 2015. The water temperature was measured hourly and averaged for a 24-hour period. The mean daily air temperature was calculated from daily maximum and minimum air temperatures. The top graph shows all the following temperatures. A) Fowl2, B) LR-2, C) LR-12, D) LR-6, E) LR-10 and F) air.

3.1.1.3.Fall

The mean water temperature and standard deviation did not differ appreciably (0.20 °C) for Fowl2 and the Outfall during fall 2015 (Table 6). The mean daily water temperature for both locations followed a similar trend with the exception of the period between 3 - 14 October (Figure 15). The average daily water temperatures for the Outfall and Fowl2 decreased rapidly in early September and continued to decrease towards the end of October. The mean water temperature for Fowl2 was 0.20 °C higher than that at the Outfall during the fall time period (Appendix 2).

The Outfall and the downstream locations showed similar mean daily water temperature trends except for the period between 3 - 14 October (Figure 15). The mean water temperature decreased from LR-12 downstream to LR-10. The mean daily water temperature at the Outfall and the downstream locations decreased rapidly in early September and continued to decrease towards the end of October. The downstream locations had higher mean temperatures than the Outfall with 3.20 °C for LR-12, 1.81 °C for LR-6, and 1.11 °C for LR-10 during fall (Appendix 3).

The mean daily water temperature trends for Fowl2 were parallel to the daily mean water temperature trends in downstream locations (Figure 15). Fowl2 had the lowest mean fall water temperature whereas LR-12 had the highest mean fall water temperature. For all locations, the mean daily water temperature decreased rapidly in early September and continued to decrease towards the end of October.

During early to mid-October, the mean daily air temperature followed an almost parallel trend for all locations with the exception of the Outfall (Figure 15). The air temperature decreased towards the end of October similarly to the other locations. The downstream locations showed

higher mean water temperatures than Fowl2 by 3.00 °C for LR-12, 1.61 °C for LR-6, and 0.91 °C for LR-10 during fall (Appendix 2).

Table 6. Mean temperature (mean T) in °C and standard deviation (STDEV) with maximum and minimum average water temperature for fall from 1 September to 26 October 2015. Temperature mean was calculated from mean daily water temperatures from fall. LR-2 is the Outfall.

	FOWL2	LR-2	LR-12	LR-6	LR-10	AIR
MEAN T	16.57	16.37	19.57	18.18	17.48	15.69
STDEV	3.40	3.63	1.87	2.88	3.41	5.31
MAX	23.78	22.33	22.65	23.22	23.53	23.78
MIN	10.55	10.54	15.77	12.05	10.18	10.55

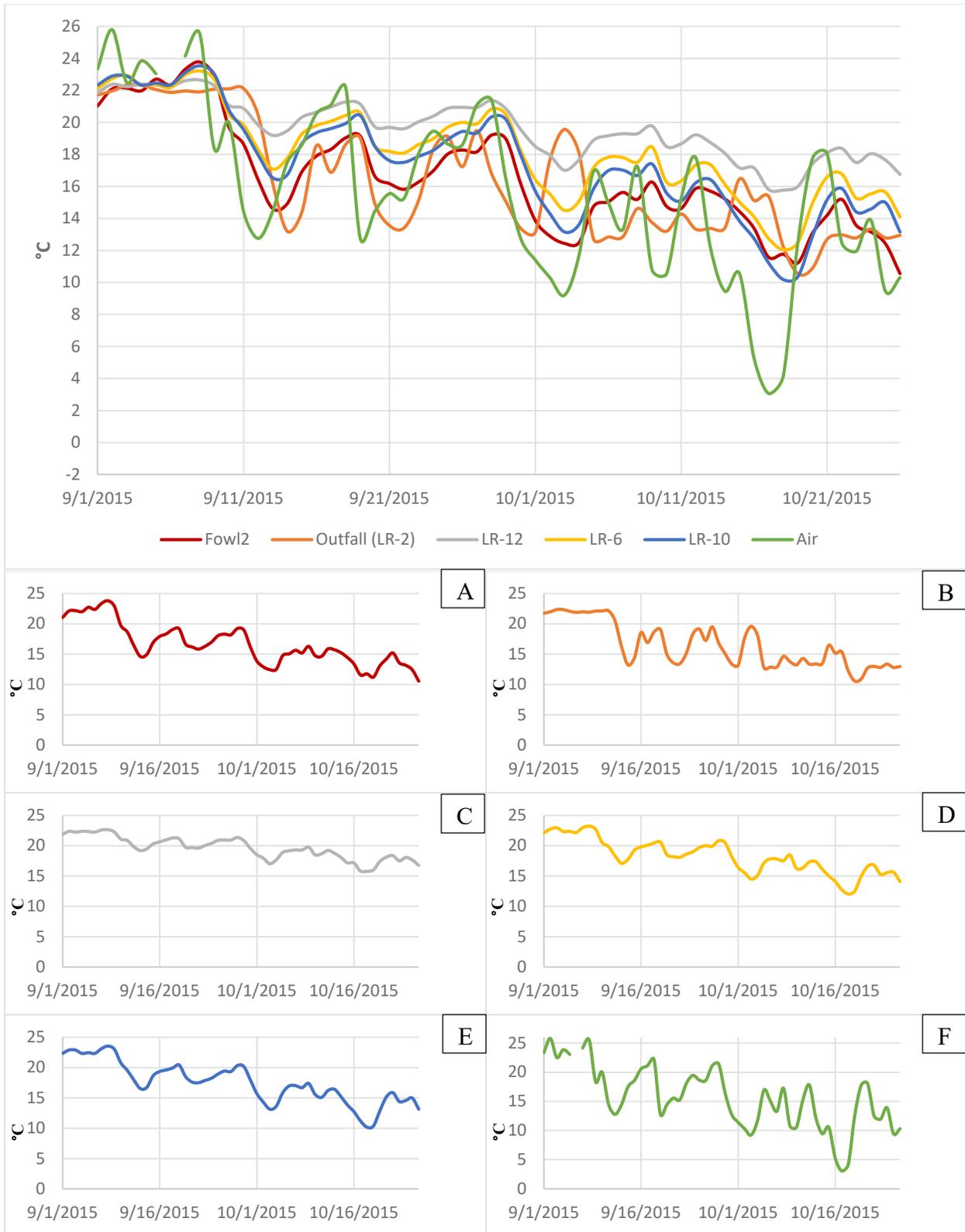


Figure 15. Mean daily water and air temperatures for spring measured in °C from 1 September to 26 October 2015. The water temperature was measured hourly and averaged for a 24-hour period. The mean daily air temperature was calculated from daily maximum and minimum air temperatures. The top graph shows all the following temperatures. A) Fowl2, B) LR-2, C) LR-12, D) LR-6, E) LR-10 and F) air.

3.1.2. Comparison of water temperatures by sites

The seasonal daily mean water temperature data were tested for a normal distribution in order to assess skewness and kurtosis (Kim 2013). The daily mean water temperature showed a normal distribution for each season (Table 7) with an acceptable absolute z-value for skewness and kurtosis below 3.29 according to Kim (2013).

For the seasonal macroinvertebrate data, the *t*-test was calculated seasonally by designating 13 April to 15 June as spring, 16 June to 31 August as summer, and 1 September to 26 October as fall. The independent Student *t*-test with equal variances was chosen because Fowl2 and the Outfall receive water from different sources. For this *t*-test, I used the mean daily temperature. In addition, I applied the independent Student's *t*-test with equal variances to compare the daily mean water temperature for Fowl2 and the Outfall with the downstream locations LR-12, LR-6, and LR-10 for each season. The independent Student's *t*-test with equal variances was chosen because the downstream locations range from 4.5 to 19 river km downstream from the Outfall and therefore were treated independently based on geographic location.

Table 7. The daily mean water temperature of each location was tested on normal distribution by using skewness and kurtosis for the seasons spring (13 April to 15 June), summer (16 June to 31 August), and fall (1 September to 26 October) in 2015. Reported are the z-values for skewness and kurtosis. The normal distribution applies for absolute z-values below 3.29 for sample size between 50 and 300. Sample size for spring was 64, for summer was 77, and for fall was 56.

	FOWL2	LR-2	LR-12	LR-6	LR-10
SPRING (SKEWNESS)	-1.47	-0.95	-1.04	-1.29	-1.36
SPRING (KURTOSIS)	-1.21	-1.37	-1.12	-1.24	-1.18
SUMMER (SKEWNESS)	-1.72	0.46	-0.90	-0.77	-0.67
SUMMER (KURTOSIS)	-0.52	-2.28	-0.71	-1.17	-1.32
FALL (SKEWNESS)	1.46	1.34	-0.45	-0.16	-0.18
FALL (KURTOSIS)	-0.88	-1.95	-1.28	-1.05	-0.95

From April to October, the mean water temperature for Fowl2 and LR-2 did not differ significantly (Table 8). However, the standard deviation for Fowl2 was almost 1/3 higher than that for the Outfall. One reason for this difference may be that the Outfall showed less daily variation

in temperature changes than Fowl2 (see section 3.1.1.1 and 3.1.1.2). LR-12 had the highest mean water temperature whereas LR-2 showed the lowest mean water temperature for the whole seven months. The mean water temperature decreased gradually downstream from LR-12 to LR-10. Fowl2 had a lower mean water temperature than the downstream locations.

Table 8. Mean water temperature (T) in °C and standard deviation (STDEV) for all locations from April through October.

	FOWL2	LR-2	LR-12	LR-6	LR-10
MEAN T	17.784	17.483	18.615	18.282	18.121
STDEV	3.792	2.905	2.722	3.185	3.451

The Student's *t*-test indicated a significant difference in mean daily water temperatures for the Outfall and Fowl2 for summer ($p < 0.05$, Table 9). During spring and fall, mean water temperature for Fowl2 and the Outfall did not differ significantly.

Table 9. Independent Student's *t*-test with equal variance for the locations Fowl2 and Outfall for each season. The *t*-tests were calculated from the mean daily temperatures of Fowl2 and the Outfall. P-values below 0.05 are considered significantly different. * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$

SEASONS	MEAN ± SEM FOWL2	MEAN ± SEM LR-2	P-VALUE	SIGNIFICANCE
SPRING	15.466 ± 0.487	15.682 ± 0.203	0.683	Not significant
SUMMER	20.595 ± 0.179	19.788 ± 0.116	2.206 × 10⁻⁴	***
FALL	16.570 ± 0.455	16.371 ± 0.485	0.766	Not significant

The independent Student's *t*-test showed a significant difference in daily mean water temperatures for the Outfall and the three downstream locations during summer ($p < 0.05$, Table 11). During fall, the Outfall and Fowl2 had a significant difference in daily mean water temperature for LR-12 and LR-6 (Table 10, Table 11). During spring, the three downstream locations had similar daily mean water temperatures as Fowl2 and the Outfall.

Table 10. P-values for the independent Student’s t-test with equal variances comparing Fowl2 with the downstream locations LR-12, LR-6, and LR-10 for spring, summer, and fall 2015. P-values below 0.05 are considered significantly different.

	FOWL2	LR-12		LR-6		LR-10	
SEASONS	Mean ± SEM	Mean ± SEM	P-Value	Mean ± SEM	P-Value	Mean ± SEM	P-Value
SPRING	15.466 ± 0.487	15.798 ± 0.323	0.571	15.711 ± 0.391	0.695	15.678 ± 0.404	0.738
SUMMER	20.595 ± 0.179	20.259 ± 0.111	0.112	20.493 ± 0.143	0.656	20.618 ± 0.161	0.925
FALL	16.570 ± 0.455	19.573 ± 0.250	6.955×10⁻⁸	18.180 ± 0.384	0.008	17.481 ± 0.455	0.160

Table 11. P-values for the independent Student’s t-test with equal variances comparing Fowl2 and the Outfall with the downstream locations LR-12, LR-6, and LR-10 for spring, summer, and fall 2015. P-values below 0.05 are considered significantly different.

	OUTFALL	LR-12		LR-6		LR-10	
SEASONS	Mean ± SEM	Mean ± SEM	P-Value	Mean ± SEM	P-Value	Mean ± SEM	P-Value
SPRING	15.682 ± 0.203	15.798 ± 0.323	0.761	15.711 ± 0.391	0.946	15.678 ± 0.404	0.993
SUMMER	19.788 ± 0.116	20.259 ± 0.111	0.004	20.493 ± 0.143	1.828×10⁻⁴	20.618 ± 0.161	4.680×10⁻⁵
FALL	16.371 ± 0.485	19.573 ± 0.250	4.779×10⁻⁸	18.180 ± 0.384	0.004	17.481 ± 0.455	0.098

3.1.3. Covariance and Correlation

To assess the relationship between temperature at Fowl2, the Outfall, and the downstream locations, I calculated the covariance and correlation coefficient for spring (13 April to 15 June), summer (16 June to 31 August), and fall (1 September to 26 October).

3.1.3.1.Spring

The covariance was positive for all locations and data points clustered closely along the trend line indicating that all locations shared a similar water temperature pattern (Figure 16). Fowl2 shared a higher similarity in its water temperature pattern with the downstream sites, which is reflected in the high correlation coefficient (Table 12) and in the close clustering along the trend line (Figure 16B, C, and D). However, as stated in section 3.2.1.1, the Outfall was less similar in

its water temperature pattern compared to Fowl2 and the downstream sites because they showed the lowest correlation and the data points were not as closely clustered as for the correlation of Fowl2 with the downstream sites (Figure 16, Table 12). All locations showed a high correlation with the daily mean air temperature.

Table 12. Covariance and correlation coefficient of mean daily water and air temperatures in °C for the locations Fowl2/Outfall, Fowl2/LR-12, Fowl2/LR-6, Fowl2/LR-10, Outfall/LR-12, Outfall/LR-6, Outfall/LR-10, Air/Fowl2, Air/Outfall, Air/LR-12, Air/LR-6, and Air/LR-10 for spring (13 April – 15 June 2015).

	COVARIANCE	CORRELATION COEFFICIENT
FOWL2/OUTFALL	5.559	0.894
FOWL2/LR-12	9.736	0.982
FOWL2/LR-6	11.924	0.994
FOWL2/LR-10	12.328	0.993
OUTFALL/LR-12	3.855	0.935
OUTFALL/LR-6	4.549	0.913
OUTFALL/LR-10	4.670	0.905
AIR/FOWL2	12.126	0.994
AIR/OUTFALL	4.609	0.910
AIR/LR-12	7.999	0.990
AIR/LR-6	9.766	0.999
AIR/LR-10	10.111	0.999

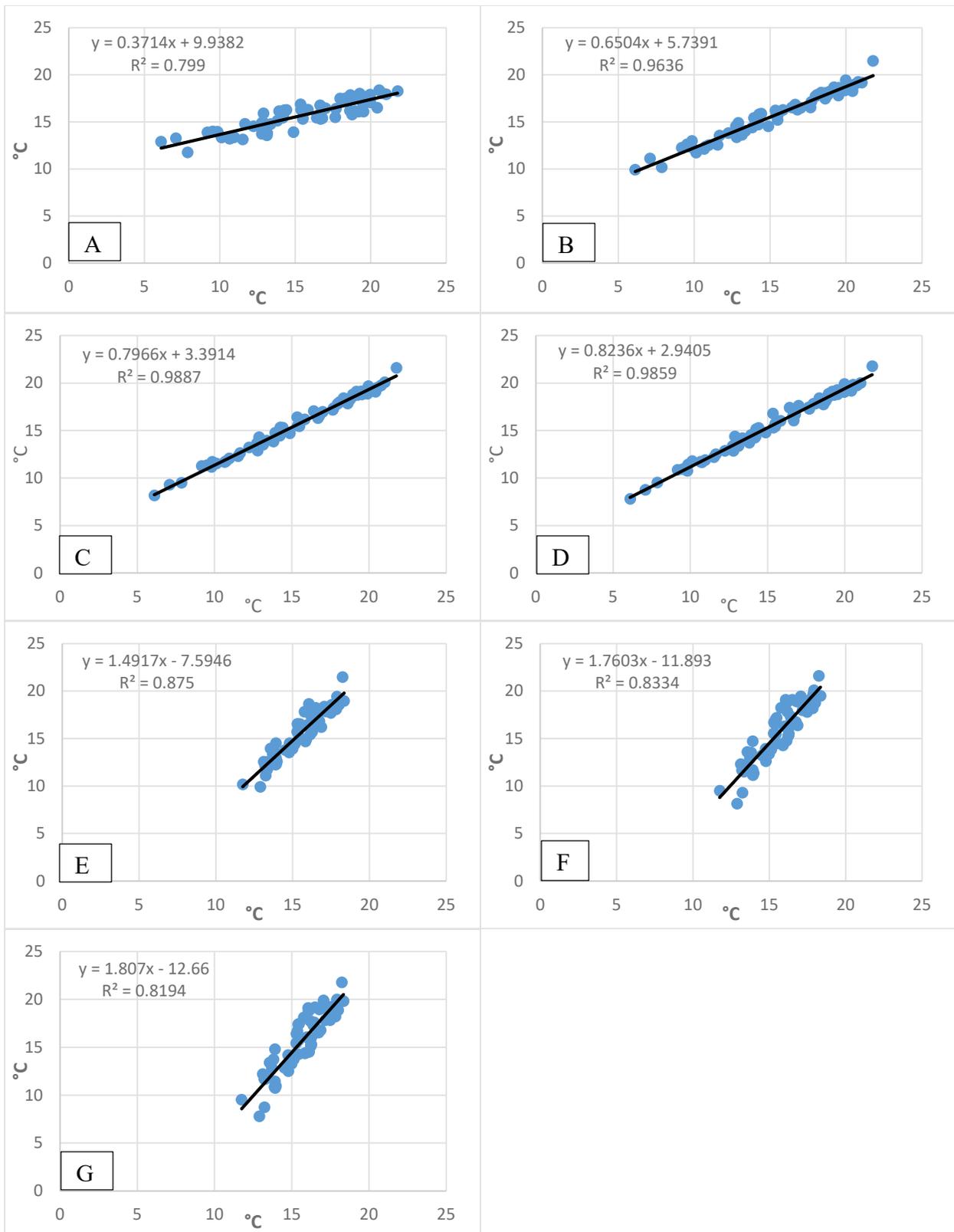


Figure 16. Correlation of mean daily water temperatures (°C) of the following locations: A) Fowl2/Outfall (LR-2), B) Fowl2/LR-12, C) Fowl2/LR-6 D) Fowl2/LR-10, E) Outfall (LR-2)/LR-12, F) Outfall (LR-2)/ LR-6, and G) Outfall/LR-10 for spring (April – 15 June 2015).

3.1.3.2. Summer

All locations showed a positive covariance during summer indicating that water temperature had a similar pattern at the different locations (Figure 17). The correlation of Fowl2/Outfall was the lowest among the locations indicating a lower similarity in water temperature pattern than Fowl2 with the downstream sites (Table 13). The correlation coefficients comparing Fowl2 with LR-6 and LR-10 was the highest among the sites indicating the highest similarity in water temperature pattern (Table 13). However the correlation coefficient of the Outfall with LR-6 and LR-10 showed a lower correlation than with Fowl2, which had closely clustered data points indicating a low similarity in water temperature pattern (Figure 17). All location except for the Outfall correlated high with the daily mean air temperature.

Table 13. Covariance and correlation coefficient of mean daily water temperatures in °C for the locations Fowl2/Outfall, Fowl2/LR-12, Fowl2/LR-6, Fowl2/LR-10, Outfall/LR-12, Outfall/LR-6, Outfall/LR-10, Air/Fowl2, Air/Outfall, Air/LR-12, Air/LR-6, and Air/LR-10 for summer (16 June– 31 August 2015).

	COVARIANCE	CORRELATION COEFFICIENT
FOWL2/OUTFALL	0.377	0.240
FOWL2/LR-12	1.126	0.749
FOWL2/LR-6	1.777	0.916
FOWL2/LR-10	2.053	0.940
OUTFALL/LR-12	0.704	0.725
OUTFALL/LR-6	0.671	0.535
OUTFALL/LR-10	0.668	0.474
AIR/FOWL2	1.831	0.887
AIR/OUTFALL	0.633	0.475
AIR/LR-12	1.057	0.829
AIR/LR-6	1.511	0.919
AIR/LR-10	1.744	0.942

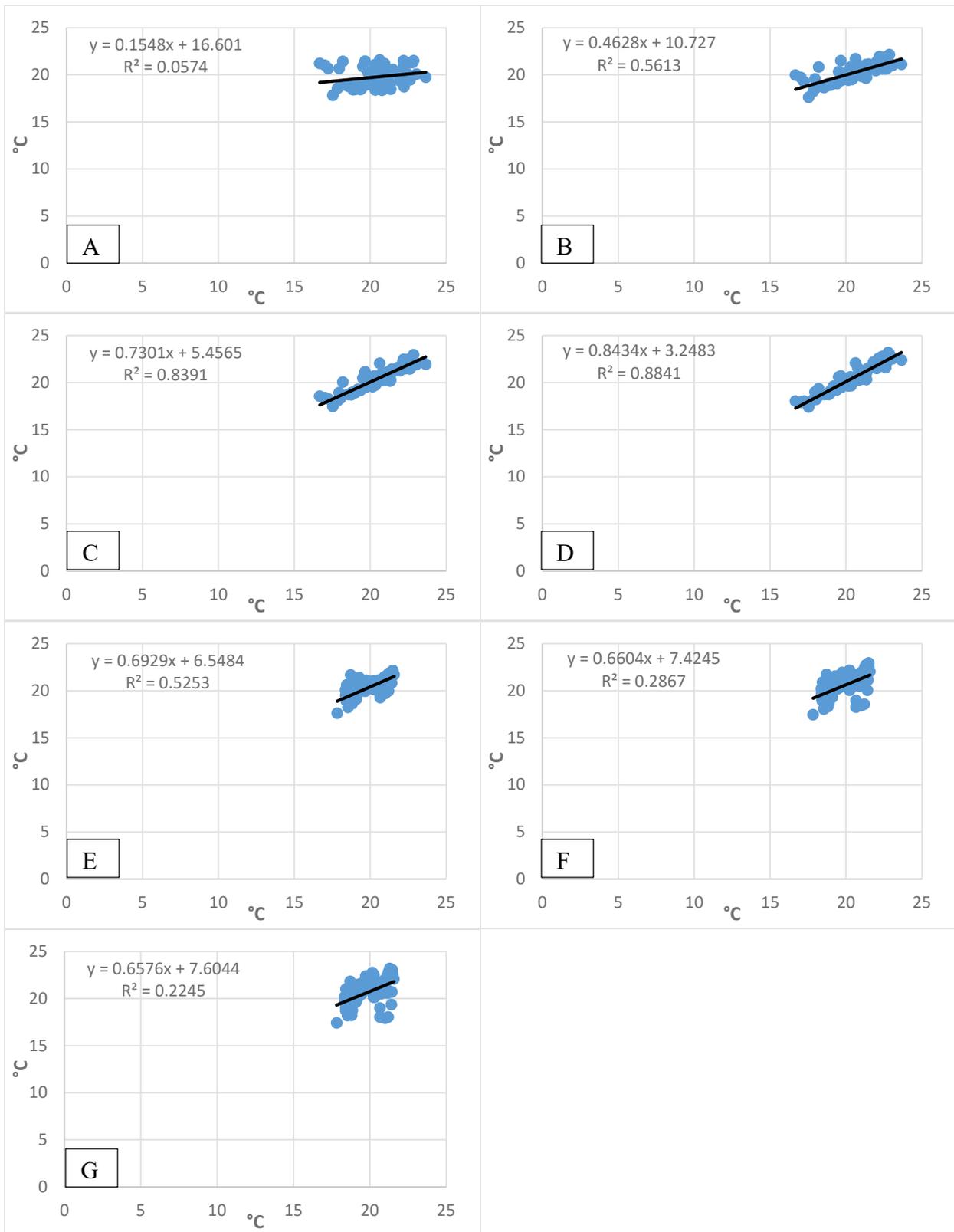


Figure 17. Correlation of mean daily water temperatures in °C of the following locations: A) Fowl2/Outfall (LR-2), B) Fowl2/LR-12, C) Fowl2/LR-6 D) Fowl2/LR-10, E) Outfall (LR-2)/LR-12, F) Outfall (LR-2)/ LR-6, and G) Outfall/LR-10 for summer (16 June – 31 August 2015).

3.1.3.3.Fall

All locations showed a positive covariance during fall indicating a similar water temperature pattern (Figure 18). The correlation coefficient of Fowl2/Outfall (Figure 18A), Outfall/LR-12, Outfall/LR-6, Outfall/LR-10 (Figure 18E-F) were less similar in their water temperature pattern than Fowl2 with the downstream locations, which is reflected in the scattered data points in Figure 18A, E, F, and G (Figure 18B-D, Table 14). Fowl2/LR-12 had the closest correlation, indicating the highest similarity in their water temperature pattern. All locations correlated high with the daily mean air temperature. Outfall correlated the lowest with the daily mean air temperature.

Table 14. Covariance and correlation coefficient of mean daily water temperatures in °C for the locations Fowl2/Outfall, Fowl2/LR-12, Fowl2/LR-6, Fowl2/LR-10, Outfall/LR-12, Outfall/LR-6, Outfall/LR-10 Air/Fowl2, Air/Outfall, Air/LR-12, Air/LR-6, and Air/LR-10 for fall (1 September– 26 October 2015).

	COVARIANCE	CORELLATION COEFFICIENT
FOWL2/OUTFALL	9.552	0.787
FOWL2/LR-12	6.026	0.963
FOWL2/LR-6	9.343	0.920
FOWL2/LR-10	11.041	0.800
OUTFALL/LR-12	4.946	0.741
OUTFALL/LR-6	7.598	0.741
OUTFALL/LR-10	9.107	0.749
AIR/FOWL2	8.396	0.813
AIR/OUTFALL	8.630	0.780
AIR/LR-12	4.688	0.820
AIR/LR-6	7.114	0.808
AIR/LR-10	8.721	0.837

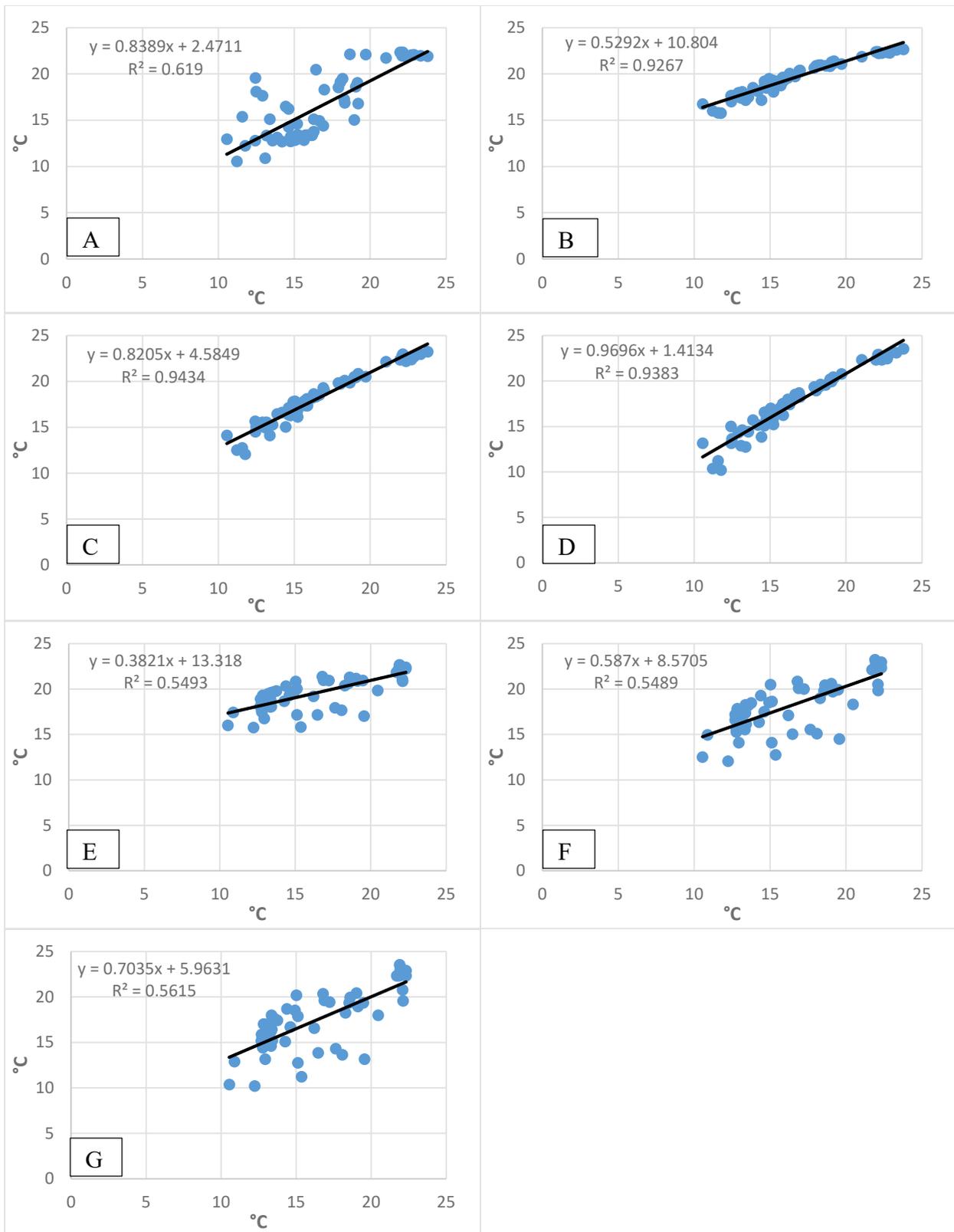


Figure 18. Correlation of mean daily water temperatures in °C of the following locations: A) Fowl2/Outfall (LR-2), B) Fowl2/LR-12, C) Fowl2/LR-6 D) Fowl2/LR-10 E) Outfall (LR-2)/LR-12 F) Outfall (LR-2)/ LR-6 G) Outfall/LR-10 for fall (1 September – 26 October 2015).

3.1.4. Wastewater treatment plant discharge and the Inkster USGS gauge discharge

The daily discharge from the WWTP at the Outfall into the Lower Rouge River was reported by YCUA from April to October 2015 (Appendix 4 and 5) in millions of gallons per day (mgd). I converted the discharge rate to cubic meter per sec (m^3/sec). The USGS gauge at Inkster, MI measures the discharge rates in cft/sec (cubic feet per sec) every 15 minutes. I used the mean daily data given by the United States Geological Service (USGS 2016) and converted it to m^3/sec . Additionally, precipitation data from the NOAA from the stations GHCND:US1MIWY0031 (Menne, Durre, Korzeniewski, et al. 2012b; Menne, Durre, Vose, et al. 2012) and GHCND:US1MIWY0068 (Menne, Durre, Korzeniewski, et al. 2012a; Menne, Durre, Vose, et al. 2012) in Canton Michigan were obtained for high peak and mid to low range peak events for the dates May 6, June, 15, 16, August 3, September 4 and 19, 2015. The precipitation data were averaged between the two Canton, MI locations for a 24-hour period. The stations were approximately 4.5 km NE of the Outfall.

3.1.4.1.Spring

The mean discharge rate for the WWTP during spring was $1.1 \text{ m}^3/\text{sec}$ (26 mgd) with a standard deviation of $0.2 \text{ m}^3/\text{sec}$ (5.4 mgd) (Figure 19). The daily discharge rate from the WWTP remained almost constant in spring, which is also reflected in the low variation of the standard deviation.

The Inkster gauge had a mean discharge rate of $4.1 \text{ m}^3/\text{sec}$ (93 mgd) in spring with a standard deviation of $4.1 \text{ m}^3/\text{sec}$ (94 mgd) (Figure 19). It showed high variations in the daily discharge rate ranging from $1 \text{ m}^3/\text{sec}$ (23 mgd) to $21 \text{ m}^3/\text{sec}$ (479 mgd). The highest discharge rates at the Inkster gauge occurred toward the end of spring. The highest discharge peak of $21.2 \text{ m}^3/\text{sec}$ (484 mgd) on 15 June 2015 was associated with a total precipitation of 33.8 mm in 24 hours.

Before 15 June 2015, the total precipitation was 7.2 mm in 24 hours. A mid-range peak on 6 May 2015 with 9.8 m³/sec (224 mgd) was associated with a total precipitation amount of 24.2 mm in 24 hours.

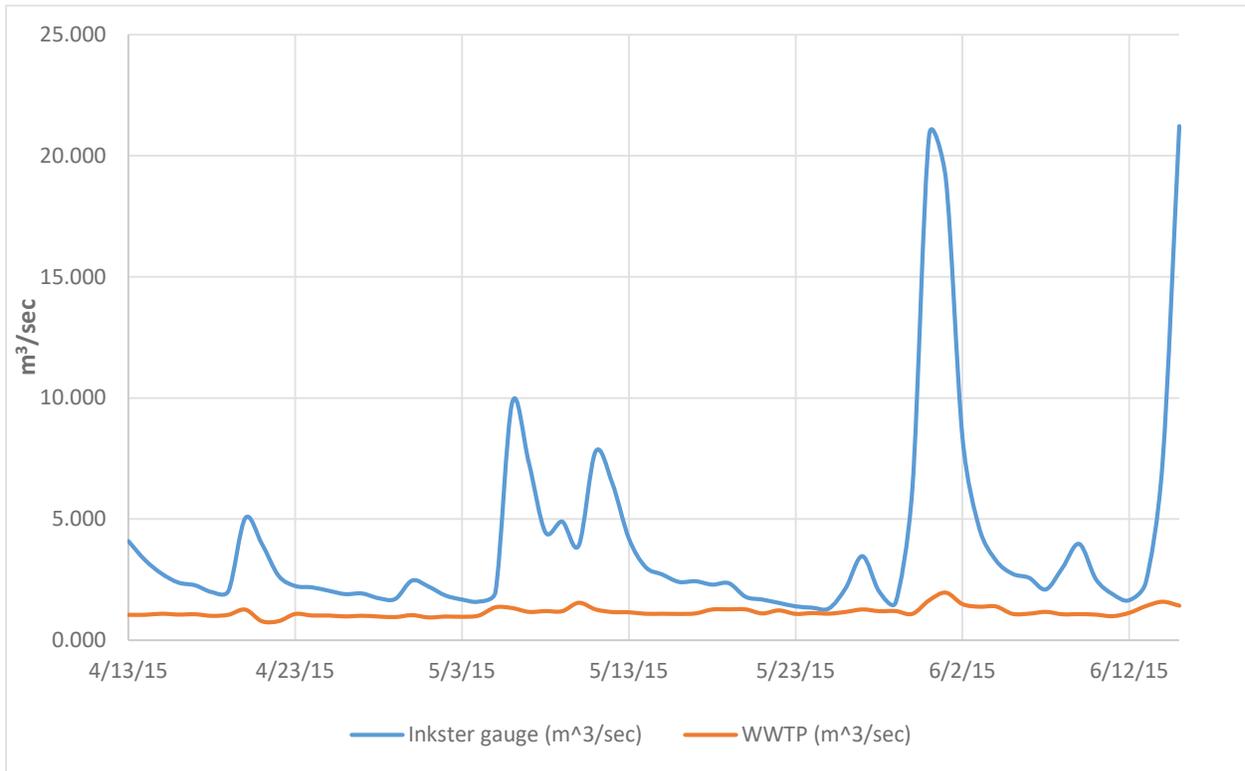


Figure 19. Daily discharge rate from the wastewater treatment plant (WWTP) and Inkster, MI USGS gauge during spring (13 April -15 June 2015). Discharge rates for the WWTP were converted from million of gallons per day into cubic meter per second (m³/sec). The daily discharge rate at Inkster, MI was calculated by USGS based on measurements made each 15 minutes (USGS 2016) in cft/sec and was converted to m³/sec.

3.1.4.2. Summer

The mean discharge rate for the WWTP during summer was 1 m³/sec (23 mgd) with a standard deviation of 0.1 m³/sec (3.1 mgd) and showed a reduced discharge rate compared to spring (Figure 20). The daily discharge rate from the WWTP remained almost constant in summer.

The Inkster gauge had a mean discharge rate of 2.8 m³/sec (64 mgd) with a standard deviation of 3.7 m³/sec (85 mgd), which was reduced compared to spring (Figure 20). The highest discharge peak was on 20.6 m³/sec (469 mgd) on 16 June 2015 with a total precipitation of 15.9 mm in 24 hours. A lower peak on August 3, 2015 with 5.6 m³/sec (127 mgd) was associated with

a total precipitation amount of 25.2 mm in 24 hours. The difference in precipitation and discharge peaks between the two days could be attributed that the location of the weather station, which does not reflect the precipitation of the headwaters of the Lower Rouge River.

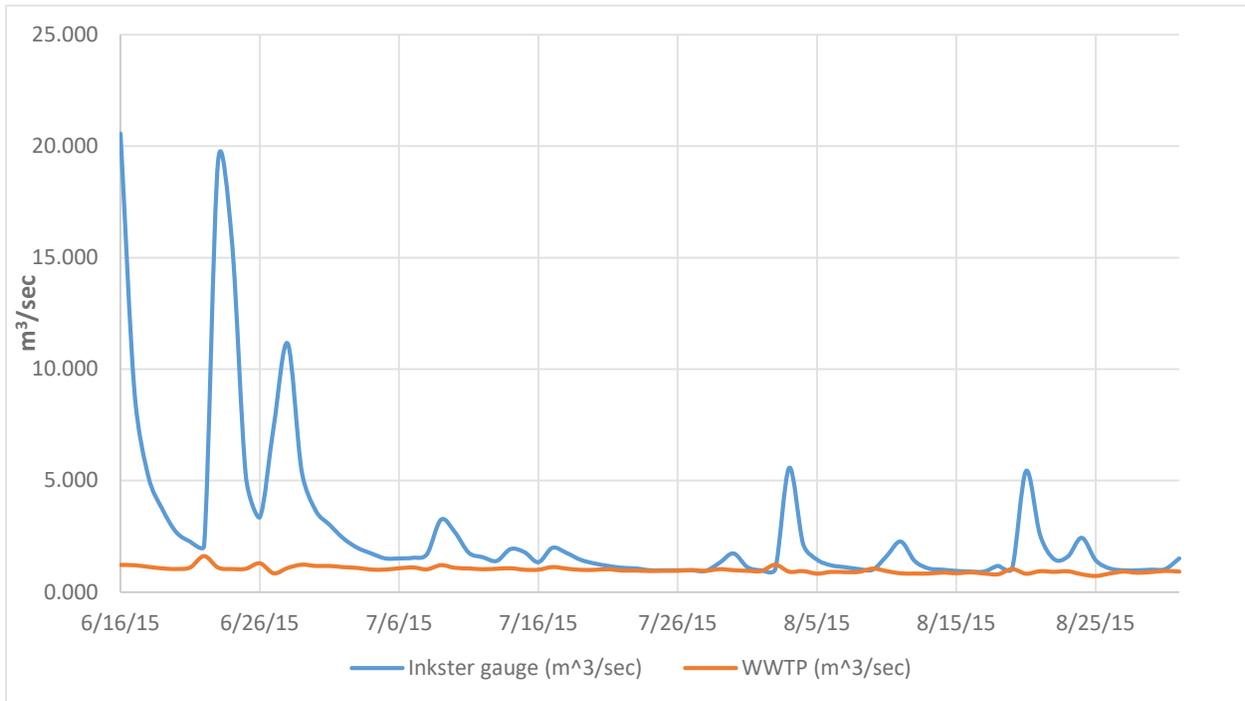


Figure 20. Daily discharge rate from the wastewater treatment plant (WWTP) and Inkster, MI USGS gauge during summer (16 June – 13 August 2015). Discharge rates for the WWTP were converted from million of gallons per day into cubic meter per second (m^3/sec). The daily discharge rate at Inkster, MI was calculated by USGS based on measurements made each 15 minutes (USGS 2016) in cft/sec and was converted to m^3/sec .

3.1.4.3. Fall

The mean discharge rate for the WWTP during fall was $0.9 m^3/sec$ (20 mgd) with a standard deviation of $0.2 m^3/sec$ (4.2 mgd) and was the lowest discharge rate of the three seasons (Figure 21). The WWTP daily discharge rate was slightly lower than in summer and remained almost constant in fall

The Inkster gauge had a daily discharge rate of $1.1 m^3/sec$ (26 mgd) with a standard deviation of $0.7 m^3/sec$ (15 mgd) and had the lowest discharge rate of the three seasons (Figure 21). However, for fall 2015, the Inkster gauge had a reduced discharge rate compared to summer. It showed high discharge rates at the beginning of fall with $5.2 m^3/sec$ (118 mgd) on September 4,

2015 with a total precipitation of 36.1 mm 24 hours. A lower peak on September 19, 2015 with 2.4 m³/sec (55 mgd) received a total precipitation amount of 14.2 mm over a 24-hour period.

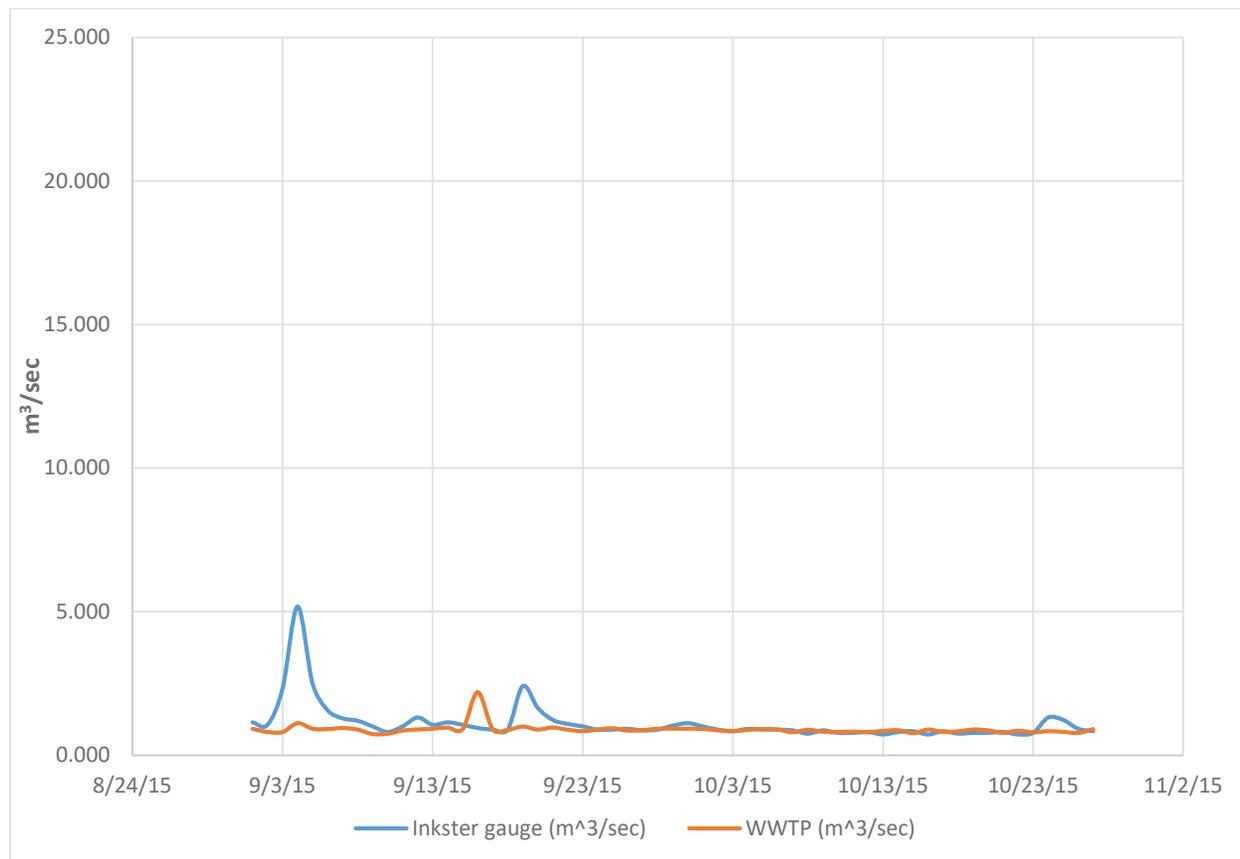


Figure 21. Daily discharge rate from the wastewater treatment plant (WWTP) and the Inkster, MI USGS gauge during fall (1 September – 26 October 2015). Discharge rates for the WWTP were converted from million of gallons per day into cubic meter per second (m³/sec). The daily discharge rate at Inkster, MI was calculated by USGS based on measurements made each 15 minutes (USGS 2016) in cft/sec and was converted to m³/sec.

3.2. Macroinvertebrates

3.2.1. Macroinvertebrate species diversity and habitat quality

Macroinvertebrates were sampled in spring, summer, and fall at the locations Fowl2, LR-2, LR-12, LR-6, and LR-10. The macroinvertebrates were identified to the family level; and some were also identified to the genus level to categorize them into functional feeding groups. Additionally, based on the taxonomic level of order, the macroinvertebrates were divided into “sensitive”, “somewhat sensitive”, and “tolerant” using the Stream Macroinvertebrate Datasheet by the Michigan Clean Water Corps (MCWC 2005). To compare the diversity among locations

and the distribution of taxa at individual locations, the Shannon Diversity Index and Shannon's Evenness (Heip, Herman, and Soetaert 1998) were calculated by using the numbers of individuals in each family. Family richness was determined by calculating the number of families at each location. Taxa percentages higher than 10% were considered as high abundance. In addition, the similarity of family taxa with Fowl2 and the Outfall with the downstream locations was compared using the Bray-Curtis similarity (Greenacre and Primicerio 2013). Trends of the macroinvertebrate order taxa were analyzed from 2012 to 2014. However, no data were available for Fowl2 for spring 2012, Outfall for spring 2013 and 2013, and LR-6 for fall 2014. For spring 2012 for Fowl2, I used the last available data which as in spring 2010. The past three years did not provide individual numbers in each family and they were reported as R for rare with individuals between 1 to 10 and C as common for individuals over 10 (Appendix 6).

3.2.1.1.Spring

3.2.1.1.1. Comparison of Fowl2 and Outfall (LR-2)

In a comparison of Fowl2 and the Outfall, Fowl2 had 10 taxa families present with a total of 115 individuals whereas the Outfall had 14 family taxa with 262 individuals (Table 15). The Outfall had more than double the number of individuals compared to Fowl2. Fowl2 had almost 7-times more individual sensitive taxa (Heptageniidae 27) than the Outfall (Limnephilidae 1, Leptophlebiidae and Heptageniidae 3). Fowl2 had one sensitive taxa (Heptageniidae (Ephemeroptera)), while the Outfall had three sensitive taxa (Limnephilidae (Trichoptera), Leptophlebiidae (Ephemeroptera) and Heptageniidae (Ephemeroptera)). The Outfall showed the highest abundance of macroinvertebrates in the somewhat sensitive group with Simuliidae (Diptera) (100) followed by Hydropsychidae (Trichoptera) (50) and Amphipoda (30). Chironomidae (Diptera) (50) had the highest number of individuals in the Outfall's tolerant group.

In contrast, Fowl2 had the highest abundance of macroinvertebrates in the tolerant group with Chironomidae (40). At Fowl2, Simuliidae (Diptera) (26) had the highest abundance in the somewhat sensitive group. The highest abundance of macroinvertebrates at the Outfall were the somewhat sensitive group with 193 (74%) individuals followed by the tolerant taxa groups with 65 (25%) individuals and the sensitive group with 4 (1.5%) individuals. In contrast, Fowl2 showed the highest abundance in the tolerant group with 46 (40%) individuals followed by the somewhat sensitive group with 42 (36.5%) individuals and the sensitive group with 27 (23.5%) individuals. The number of the somewhat sensitive group and the tolerant group were close to each other.

The Shannon Diversity Index was higher for Fowl2 (1.70) than the Outfall (1.63). The Shannon evenness however showed a more evenly distributed macroinvertebrate taxa at Fowl2 (0.77) than at the Outfall (0.64). Both locations' samples showed a similarity of 40.9% (Table 17).

Analysis of the family richness in Fowl2 for the years 2010, 2013, 2014 and 2015 varied between 8 to 16 with the current value of 10 within this range (Table 32). The taxa Heptageniidae (Ephemeroptera), Amphipoda, Isopoda, Oligochaeta (Annelida), and Chironomidae (Diptera) reappeared at Fowl2 in spring 2010, 2013, 2014, and 2015. The taxa Haliplidae (Coleoptera) and Simuliidae (Diptera) reappeared in 2010, 2013, and 2015 and Decapoda reappeared in 2013 and 2015. The comparison of spring data with the Outfall from previous years is limited because FOTR has data only for 2014. The taxa Simuliidae (Diptera), Coenagrionidae (Zygoptera), Hydropsychidae (Trichoptera), Amphipoda, Isopoda, Oligochaeta (Annelida), and Chironomidae (Diptera) were present in 2014 and 2015 spring at the Outfall (Table 33). Family richness varied from 14 for 2015 to 9 for 2014.

3.2.1.1.2. Comparison of downstream locations and the Outfall

The downstream locations LR-12 (128) and LR-6 (45) had lower numbers of individuals

than the Outfall (262) (Table 15) However, the downstream location LR-10 (309) had higher numbers of individuals than the Outfall. The downstream location LR-10 had the same number of individuals of Simuliidae (Diptera) (100) and Hydropsychidae (Trichoptera) (50) as the Outfall, but the number of Chironomidae (Diptera) at LR-10 was 2-times higher with 100 individuals. Additionally, LR-10 had a higher abundance of Calopterygidae (Zygoptera) (25) and Annelida (Oligochaeta) (25). LR-12 showed high numbers of individuals of Simuliidae (Diptera) (25), Hydropsychidae (Trichoptera) (41), and Chironomidae (Diptera) (35) as the Outfall but the number of individuals at LR-12 were lower. LR-10 also showed high numbers of individuals of Simuliidae (Diptera) (100), Hydropsychidae (Trichoptera) (50), and Chironomidae (Diptera) (100) as the Outfall but the number of individuals at LR-12 were lower. LR-6 had lower number of individuals.

The most abundant taxa at LR-6 were similar to the downstream locations and the Outfall (Hydropsychidae (Trichoptera) (13) and Chironomidae (Diptera) (12)). However, LR-6 did not have any Simuliidae. The highest number of families occurred at the locations LR-6 (15) followed by LR-12 (14) and LR-2 (14). LR-10 had the lowest family richness (11) for spring. The highest abundance of macroinvertebrates was mostly in the somewhat sensitive taxa group. LR-12 (Heptageniidae 1, Baetidae 1) and LR-6 (Baetidae 1) were the only downstream locations with a sensitive taxon with low numbers.

The Shannon Diversity Index was the lowest for LR-10 (1.58) and the highest for LR-6 (2.13). LR-6 (0.78) showed the highest evenness and the locations LR-12 (0.70), LR-10 (0.66), and the Outfall (0.64) did not vary much from each other. The Outfall and LR-10 sites had the highest Bray-Curtis Index similarity with 73.6% compared to the other downstream locations LR-12 and LR-6 (Table 17).

In the past three years, the downstream locations had a reappearance of the taxa Decapoda,

Hydropsychidae (Trichoptera), Oligochaeta (Annelida), and Chironomidae (Diptera) at each location (Table 34, Table 35, Table 36). Additionally, in the past three years, Elmidae (Coleoptera) and Simuliidae (Diptera) reappeared at LR-12, Coenagrionidae (Zygoptera) reappeared at LR-6, and Calopterygidae (Zygoptera) at LR-10. The family richness varied between 13 to 16 for LR-12, 9 to 15 for LR-6, and 11 to 12 for LR-10.

3.2.1.1.3. Comparison of downstream locations and Fowl2

Fowl2 and the downstream locations differed in terms of the distribution of the high abundance taxa, except for Simuliidae (Diptera) and Chironomidae (Diptera) (Table 15). Fowl2 had high abundance of taxa in all three tolerant groups (Heptageniidae 27, Simuliidae 26, Chironomidae 40), whereas the downstream locations had the highest abundance of taxa in the somewhat sensitive group (Simuliidae, Hydropsychidae) followed by the tolerant group (Chironomidae). In comparison, the Outfall and the downstream locations shared a higher similarity of high abundance taxa than Fowl2 and the downstream locations (see 3.2.1.1.2). Additionally, the downstream locations LR-12 (14), LR-6 (15), and LR-10 (11) showed a higher number of families than Fowl2 (10).

The Shannon Diversity Index for LR-10 (1.58) was the lowest, followed by Fowl2 (1.70). The site LR-6 (2.13) had the highest Shannon Diversity Index, however the Shannon Evenness (0.78) did not differ much from Fowl2 (0.77). Fowl2 and downstream samples showed a medium to low Bray-Curtis Index similarity between 56.8% to 27.5% respectively (Table 17).

A comparison of the downstream data from FOTR of the past three years with the 2015 downstream data showed a reappearance of the taxa Decapoda, Hydropsychidae (Trichoptera), Oligochaeta (Annelida), and Chironomidae (Diptera) at each location (Table 34, Table 35, Table 36).

Table 15. Percentage (%) and number of individuals (#) in each family of macroinvertebrates in spring calculated by the raw data. Macroinvertebrates are categorized to sensitive, somewhat sensitive, and tolerant groups (Michigan Clean Water Corps). The number (N) of family taxa is the number of families at each location. LR-2 is the Outfall.

TOLERANCE	CLASS	ORDER	FAMILIES	FOWL2		LR-2		LR-12		LR-6		LR-10	
				%	#	%	#	%	#	%	#	%	#
Sensitive		Trichoptera	Limnephilidae	0	0	0.4	1	0	0	0	0	0	0
		Ephemeroptera	Heptageniidae	23.5	27	0	0	0.8	1	0	0	0	0
		Ephemeroptera	Leptophlebiidae, Heptageniidae ⁹	0	0	1.1	3	0	0	0	0	0	0
		Ephemeroptera	Baetidae	0	0	0	0	0.8	1	2.2	1	0	0
Somewhat sensitive		Coleoptera	Elmidae	0	0	0.4	1	4.7	6	0	0	0	0
		Coleoptera	Haliplidae	1.7	2	0	0	0	0	2.2	1	0	0
		Pelecypoda	Sphaeriidae	0	0	0	0	1.6	2	2.2	1	0	0
		Diptera	Simuliidae	22.6	26	38.2	100	19.5	25	0	0	32.4	100
		Diptera	Tipulidae	0	0	0.4	1	1.6	2	2.2	1	0.3	1
		Decapoda		3.5	4	0	0	2.3	3	8.9	4	1.3	4
		Zygoptera	Coenagrionidae	0	0	1.1	3	0	0	2.2	1	0	0
		Zygoptera	Calopterygidae	0	0	0.4	1	0	0	2.2	1	8.1	25
		Odonata	Aeshnidae	0	0	0	0	1.6	2	0	0	0.3	1
		Trichoptera	Hydropsychidae	0	0	19.1	50	32.0	41	28.9	13	16.2	50
		Amphipoda		2.6	3	11.5	30	2.3	3	0	0	0.3	1
		Isopoda		6.1	7	2.7	7	0.8	1	8.9	4	0.3	1
Tolerant	Oligochaeta	Annelida		0.9	1	1.9	5	3.1	4	4.4	2	8.1	25
	Hirudinea	Annelida		0	0	3.8	10	0	0	2.2	1	0.3	1
		Diptera	Chironomidae	34.8	40	19.1	50	27.3	35	26.7	12	32.4	100
	Gastropoda	Hygrophila	Physidae	0	0	0	0	0	0	2.2	1	0	0
	Gastropoda	Basommatophora	Ancylidae	0	0	0	0	1.6	2	2.2	1	0	0
		Hemiptera	Gerridae	0	0	0	0	0	0	2.2	1	0	0
		Hemiptera	Veliidae	3.5	4	0	0	0	0	0	0	0	0
		Diptera	Ceratopogonidae	0.9	1	0	0	0	0	0	0	0	0
Total				100.0	115	100.0	262	100.0	128	100.0	45	100.0	309
N of families				10		14		14		15		11	

⁹ The location LR-2 voucher specimens did not include all specimens from the field. Therefore, I could not differentiate between Leptophlebiidae, and Heptageniidae.

Table 16. Shannon diversity index and Shannon's Evenness in spring for each location. LR-2 is the Outfall.

LOCATION	DATE	SHANNON DIVERSITY INDEX	SHANNON'S EVENNESS
FOWL2	4/18/2015	1.70	0.77
LR-2	4/17/2015	1.63	0.64
LR-12	4/17/2015	1.84	0.70
LR-6	4/17/2015	2.13	0.78
LR-10	4/20/2015	1.58	0.66

Table 17. Bray-Curtis Index for spring. The Bray-Curtis Index shows the percentage similarity in macroinvertebrate community between the two locations Fowl2/LR-2, Fowl2/LR-12, Fowl2/LR-6, Fowl2/LR-10, LR-2/LR-12, LR-2/LR-6, and LR-2/LR-10. LR-2 is the Outfall.

	FOWL2/ LR-2	FOWL2/ LR-12	FOWL2/ LR-6	FOWL2/ LR-10	LR-2/ LR-12	LR-2/ LR-6	LR-2/ LR-10
BRAY-CURTIS INDEX	40.9 %	56.8 %	27.5 %	34.4 %	56.9 %	22.8 %	73.6 %

3.2.1.2. Summer

3.2.1.2.1. Comparison of Fowl2 with the Outfall (LR-2)

During summer, the family richness of Fowl2 (10) and the Outfall (10) were identical, but showed fewer family taxa number than in spring (Table 18, Table 15). Fowl2 and the Outfall had the same number of individuals, with 94 each (Table 18). The highest number of individuals in a single taxa was the tolerant taxa Chironomidae (Diptera) with 57 at Fowl2 and 55 at the Outfall. The remaining taxa were mostly distributed in the somewhat sensitive group with the highest abundance in Hydropsychidae (Trichoptera) (15) at LR-2 and Decapoda (14) at Fowl2. LR-2 did not have any sensitive taxa present during the summer, while Fowl2 had two sensitive taxa Heptageniidae (2) and Baetidae (Ephemeroptera) (9).

Both locations showed a similar Shannon diversity of 1.34 and taxa were similarly unequally (0.30) distributed (Table 19). Therefore, the taxa diversity is lower and the taxa are less equally distributed than in spring. Fowl2 and the Outfall showed a high similarity with 73.4% (Table 20) in samples than in spring with 40.9% (Table 17).

3.2.1.2.2. Comparison of the Outfall with downstream locations

During summer, the Outfall (10) and the downstream locations (LR-12 14, LR-6 7, and LR-10 9) had different family taxa numbers (Table 18). LR-12 had the highest number of family taxa and LR-6 showed the lowest taxa number, whereas in spring LR-6 (15) had the highest number of family taxa (Table 15). LR-10 showed the highest number of individuals with 232, followed by the Outfall (94), LR-12 (73), and LR-6 (39). The Outfall and the downstream locations had high abundance of the somewhat sensitive taxa Hydropsychidae (Trichoptera) and the tolerant taxa Chironomidae (Diptera) (Table 18). However, they had different distribution of these taxa among the locations. LR-10 had the highest number of Hydropsychidae (Trichoptera) (101) and LR-6 (13) the lowest. The Outfall had the highest number of Chironomidae (Diptera) (55) followed by LR-6 (16). However, LR-10 showed very low abundance of Chironomidae (Diptera) (3) in comparison to the other locations. In addition, LR-12 showed the highest abundance of Elmidae (Coleoptera) (13), LR-10 had Baetidae (Ephemeroptera) (109), and LR-6 also had a high abundance of Decapoda (6).

LR-12 had the highest diversity of macroinvertebrates (2.46) and the highest evenness (0.63) of species distribution, LR-10 showed the lowest diversity (1.11) with the lowest evenness (0.20). The Outfall had a Bray-Curtis Index similarity of 47.9% with LR-12 and 51.1% with LR-6 (Table 20). The downstream location LR-10 showed the lowest similarity between the downstream locations with 15.9% with the Outfall.

3.2.1.2.3. Comparison of Fowl2 with downstream locations

LR-10 had the highest number of individuals with 232 followed by Fowl2 (94), LR-12 (73), and LR-6 (39) (Table 18). Fowl2 (60.6%) and the downstream locations (LR-12 13.7%, LR-6 41.0%) showed both high abundance in Chironomidae (Diptera) except LR-10 (3). Additionally,

Fowl2 (14%) and LR-6 (6%) had a high percentage of Decapoda. Fowl2 also had a high abundance in the sensitive order Ephemeroptera (Total of 11), but LR-10 had 10-times more (109) of the sensitive order Ephemeroptera. The number of family taxa at Fowl2 (10) was in general lower than LR-12 (14). However, the family taxa number was lower for LR-6 (7) and LR-10 (9) compared with Fowl2.

The Shannon Diversity Index of Fowl2 (1.34) and LR-6 (1.40) were close to each other, however Fowl2 (0.30) macroinvertebrates were less evenly distributed than at LR-6 (0.38) (Table 16). LR-10 had the lowest Shannon Diversity Index (1.11) and evenness (0.20) and LR-12 had the highest diversity (2.46) and evenness (0.63). Fowl2 and the downstream location showed a Bray-Curtis Index similarity in their samples with 31.1% for LR-12, 48.1 % for LR-6, and 17.1 % for LR-10 (Table 20).

Table 18. Percentage (%) and number of individuals (#) in each family of macroinvertebrates in summer calculated by the raw data. Macroinvertebrates are categorized to sensitive, somewhat sensitive, and tolerant groups (Michigan Clean Water Corps). The number (N) of family taxa is the number of families at each location. LR-2 is the Outfall.

TOLERANCE	CLASS	ORDER	FAMILIES	FOWL2		LR-2		LR-12		LR-6		LR-10	
				%	#	%	#	%	#	%	#	%	#
Sensitive		Ephemeroptera	Heptageniidae	2.1	2	0	0	1.4	1	0	0	0	0
		Ephemeroptera	Baetidae	9.6	9	0	0	1.4	1	2.6	1	46.8	109
Somewhat Sensitive		Coleoptera	Elmidae	2.1	2	8.5	8	31.5	23	2.6	1	0	0
		Coleoptera	Hydrophilidae	0	0	0	0	1.4	1	0	0	0	0
		Coleoptera	undetermined	0	0	0	0	0	0	0	0	0	0
		Diptera	Simuliidae	1.1	1	2.1	2	9.6	7	2.6	1	0.9	2
		Diptera	Tipulidae	0	0	4.3	4	1.4	1	0	0	0	0
		Decapoda		14.9	14	3.2	3	4.1	3	15.4	6	3.0	7
		Zygoptera	Coenagrionidae	0	0	0	0	1.4	1	0	0	0.4	1
		Zygoptera	Calopterygidae	0	0	1.1	1	0	0	0	0	0.4	1
		Odonata	Aeshnidae	0	0	0	0	2.7	2	0	0	0	0
		Trichoptera	Hydropsychidae	6.4	6	16.0	15	21.9	16	33.3	13	43.3	101
		Amphipoda		1.1	1	2.1	2	1.4	1	0	0	0	0
	Isopoda		1.1	1	2.1	2	0	0	0	0	1.7	4	
Tolerant	Turbellaria			0	0	0	0	2.7	2	0	0	0	0
	Oligochaeta	Annelida		1.1	1	0	0	5.5	4	2.6	1	1.7	4
		Diptera	Chironomidae	60.6	57	58.5	55	13.7	10	41.0	16	1.3	3
		Hemiptera	Veliidae	0	0	2.1	2	0	0	0	0	0	0
Total				100.0	94	100.0	94	100.0	73	100.0	39	100.0	232
N of families				10		10		14		7		9	

Table 19. Shannon diversity index and Shannon's Evenness in summer for each location. LR-2 is the Outfall.

LOCATION	DATE	SHANNON DIVERSITY INDEX	SHANNON'S EVENNESS
FOWL2	7/6/2015	1.34	0.30
LR-2	7/6/2015	1.34	0.30
LR-12	7/6/2015	2.46	0.63
LR-6	7/7/2015	1.40	0.38
LR-10	7/7/2015	1.11	0.20

Table 20. Bray-Curtis Index for summer. The Bray-Curtis Index shows the percentage similarity in macroinvertebrate community between the two locations Fowl2/LR-2, Fowl2/LR-12, Fowl2/LR-6, Fowl2/LR-10, LR-2/LR-12, LR-2/LR-6, and LR-2/LR-10. LR-2 is the Outfall.

	FOWL2/ LR-2	FOWL2/ LR-12	FOWL2/ LR-6	FOWL2/ LR-10	LR-2/ LR-12	LR-2/ LR-6	LR-2/ LR-10
BRAY-CURTIS INDEX	73.4 %	31.1 %	48.1 %	17.1 %	47.9 %	51.1 %	15. %

3.2.1.3.Fall

3.2.1.3.1. Comparison of Fowl2 and Outfall (LR-2)

During the fall, the Outfall (10) had one more taxa than Fowl2 (9), but Fowl2 (419) had over 4-times more individuals than the Outfall (90) (Table 21). Both locations had a high abundance of Chironomidae (Diptera) (Outfall 20, Fowl2 258) with Fowl2 having over 10-times more Chironomidae (Diptera). Most of their taxa were concentrated in the somewhat sensitive and tolerant group. Fowl2 had the second highest abundance in the somewhat sensitive group with Isopoda (126). In comparison, the Outfall showed high abundance in the somewhat sensitive group in Amphipoda (20) and Hydropsychidae (Trichoptera) (20). The abundant families of Amphipoda, Hydropsychidae (Trichoptera), and Chironomidae (Diptera) composed equal percentages of individuals at the Outfall (22.2 %).

The Outfall (2.00) had a Shannon Diversity Index that was two times higher than that for Fowl2 (1.00) as well as a higher species evenness value (LR-2 0.44, Fowl2 0.17) (Table 22). Fowl2 and the Outfall had a low Bray-Curtis Index similarity with 14.2% for the samples collected in fall

(Table 23).

In the past three years, the taxa Amphipoda, Isopoda, and Chironomidae (Diptera) reappeared at Fowl2 and the Outfall (Table 37, Table 38). Additionally, in the past three years, Heptageniidae (Ephemeroptera) and Decapoda reappeared at Fowl2 (Table 37). For the past three years, the family taxa number varied between 8 to 13 for Fowl2 and between 8 to 12 for the Outfall.

3.2.1.3.2. Comparison of Outfall with downstream locations

The comparison of the Outfall and the downstream locations in the fall showed the highest taxa at LR-10 (15) and the lowest at LR-2 (10) and LR-6 (11) (Table 21). LR-12 had the highest number of individuals with 93, followed by the Outfall with 90, LR-10 with 83, and LR-6 with 48. All of the downstream locations and the Outfall had a high abundance of Hydropsychidae (Trichoptera) (LR-2 20, LR-12 20, LR-6 17, LR-10 28). The Outfall did not have any sensitive taxa. LR-10 (14) had more than 3-times the number of sensitive taxa Baetidae (Ephemeroptera) and Heptageniidae (Ephemeroptera) than LR-6 (4). LR-12 (20) showed the highest sensitive taxa percentage with Baetidae (Ephemeroptera) among the locations. In addition, LR-12 had high abundance in Simuliidae (Diptera) (20) and the abundant families (Baetidae, Simuliidae, and Hydropsychidae) were evenly distributed with 21.5 % for each of them. LR-6 had additionally high abundance in the somewhat sensitive Decapoda (5) and Calopterygidae (Zygoptera) (6) and tolerant taxa Oligochaeta (5). LR-10 showed high abundance in the sensitive taxa Baetidae and Heptageniidae (14), and the somewhat sensitive taxa Hydropsychidae (28) and Isopoda (15) and did not have high abundant taxa in the tolerant group.

The Shannon Diversity Indices were very close to each other with LR-12 (2.05) having the highest value followed by LR-2 (2.00), LR-6 (1.91), and LR-10 (1.84). All of them showed almost the same evenness (LR-2 0.44, LR-12 0.45, LR-6 0.49, LR-10 0.42) (Table 22). The Outfall and

the downstream locations showed a mid-range similarity in their samples with 49.2 % for LR-12, 53.6 % for LR-6, and 46.2 % for LR-10 (Table 23).

During the past three years, the downstream locations showed a reappearance of the taxa Decapoda, Hydropsychidae (Trichoptera), and Chironomidae (Diptera) (Table 39, Table 40, Table 41). However, the downstream location LR-6 was not sampled during fall 2014. One common taxa reappeared over the last three years between the Outfall and the downstream location is Chironomidae (Diptera). Additionally, over the past three years, the taxa Baetidae (Ephemeroptera) reappeared at LR-12 and LR-10, Simuliidae (Diptera), Tipulidae (Diptera), and Calopterygidae (Zygoptera) at LR-12, and Isopoda, Oligochaeta (Annelida), and Ancyliidae (Gastropoda) at LR-6. The family taxa number varied for the Outfall from 10 to 12, for LR-12 from 12 to 19, for LR-6 from 8 to 10, and for LR-10 from 6 to 14 (Table 38, Table 39, Table 40, Table 41).

3.2.1.3.3. Comparison of Fowl2 with downstream locations

Fowl2 (9) had the lowest family taxa number compared to the downstream location (LR-12 12, LR-6 11, and LR-10 15) (Table 21). Fowl2 had distinctly more individual macroinvertebrates with 419 followed by LR-12 (93), LR-10 (83), and LR-6 (48). The sites did not have any abundant taxa in common except for LR-10. Fowl2 (126) and LR-10 (15) showed a high abundance for the somewhat sensitive taxa Isopoda.

The downstream locations had Shannon Diversity Indices that were approximately two times higher (LR-12 2.05, LR-6 1.91, and LR-10 1.84) than Fowl2 (1.00) and showed higher Evenness values (LR-12 0.45, LR-6 0.49, LR-10 0.42, than Fowl2 1.67) (Table 22). Fowl2 and the downstream locations showed low Bray-Curtis Indices similarity with 2.3% for LR-12, 5.6% for LR-6, and 11.6% for LR-10 (Table 23).

Over the past three years, the taxa Chironomidae (Diptera) were present at Fowl2 and the downstream locations (Table 37, Table 39, Table 40, Table 41).

Table 21. Percentage (%) and number of individuals (#) in each family of macroinvertebrates in fall calculated by the raw data. Macroinvertebrates are categorized to sensitive, somewhat sensitive, and tolerant groups (Michigan Clean Water Corps). The number (N) of family taxa is the number of families at each location. LR-2 is the Outfall.

TOLERANCE	CLASS	ORDER	FAMILIES	FOWL2		LR-2		LR-12		LR-6		LR-10	
				%	#	%	#	%	#	%	#	%	#
Sensitive		Ephemeroptera	Heptageniidae	3.1	13	0	0	0	0	0	0	0	0
		Ephemeroptera	Baetidae, Heptageniidae ¹⁰	0	0	0	0	0	0	8.3	4	16.9	14
		Ephemeroptera	Baetidae	0	0	0	0	21.5	20	0	0	0	0
Somewhat Sensitive		Coleoptera	Elmidae	0	0	0	0	3.2	3	6.3	3	3.6	3
		Pelecypoda	Sphaeriidae	0	0	0	0	0	0	0	0	1.2	1
		Diptera	Simuliidae	0	0	3.3	3	21.5	20	0	0	0	0
		Diptera	Tipulidae	0	0	3.3	3	5.4	5	0	0	1.2	1
		Decapoda		0.5	2	5.6	5	4.3	4	10.4	5	4.8	4
		Zygoptera	Coenagrionidae	0	0	0	0	0	0	0	0	1.2	1
		Zygoptera	Calopterygidae	0	0	7.8	7	7.5	7	12.5	6	2.4	2
		Zygoptera	Undetermined	0	0	0	0	0	0	0	0	1.2	1
		Odonata	Aeshnidae	0.2	1	0		2.2	2	0	0	0	0
		Trichoptera	Hydropsychidae	0	0	22.2	20	21.5	20	35.4	17	33.7	28
		Amphipoda		1.7	7	22.2	20	0	0	0	0	6.0	5
	Isopoda		30.1	126	4.4	4	0	0	6.3	3	18.1	15	
Tolerant	Turbellaria			0	0	0	0	1.1	1	0	0	0	0
	Oligochaeta	Annelida		2.1	9	3.3	3	0	0	10.4	5	7.2	6
	Hirudinea	Annelida		0	0	5.6	5	7.5	7	0	0	0	0
		Diptera	Chironomidae	61.6	258	22.2	20	3.2	3	6.3	3	1.2	1
	Gastropoda	Hygrophila	Physidae	0.5	2	0	0	0	0	0	0	0	0
	Gastropoda	Basommatophora	Ancylidae	0	0	0	0	1.1	1	2.1	1	0	0
		Hemiptera	Belostomatidae	0	0	0	0	0	0	0	0	1.2	1
		Hemiptera	Veliidae	0	0	0	0	0	0	2.1	1	0	0
	Hemiptera	Corixidae	0.2	1	0	0	0	0	0	0	0	0	
Total				100.0	419	100.0	90	100.0	93	100.0	48	100.0	83
N of families					9		10		12		11		15

¹⁰ The locations LR-6 and LR-10 voucher specimens did not include all specimens from the field. Therefore, I could not differentiate between Baetidae, and Heptageniidae.

Table 22. Shannon diversity index and Shannon’s Evenness in fall for each location. LR-2 is the Outfall.

LOCATION	DATE	SHANNON DIVERSITY INDEX	SHANNON'S EVENNESS
FOWL2	10/17/2015	1.00	0.17
LR-2	10/13/2015	2.00	0.44
LR-12	10/13/2015	2.05	0.45
LR-6	10/20/2016	1.91	0.49
LR-10	10/20/2016	1.84	0.42

Table 23. Bray-Curtis Index for fall. The Bray-Curtis Index shows the percentage similarity in macroinvertebrate community between the two locations Fowl2/LR-2, Fowl2/LR-12, Fowl2/LR-6, Fowl2/LR-10, LR-2/LR-12, LR-2/LR-6, and LR-2/LR-10. LR-2 is the Outfall.

	FOWL2/ LR-2	FOWL2/ LR-12	FOWL2/ LR-6	FOWL2/ LR-10	LR-2/ LR-12	LR-2/ LR-6	LR-2/ LR-10
BRAY-CURTIS INDEX	14.2 %	2.3 %	5.6 %	11.6 %	49.2 %	53.6 %	46.2 %

3.2.2. Functional feeding groups

The functional feeding groups were determined by identifying the macroinvertebrates to family- and genus-level. The Shannon diversity, Shannon Evenness, and numbers of individual are reported for each functional feeding groups. In addition, the Bray-Curtis Index was determined to compare the percentage similarity of functional feeding groups individuals of Fowl2 to the Outfall, LR-12, LR-6, LR-10 and Outfall to LR-12, LR-6, and LR-10 for each season.

3.2.2.1.Spring

In general, all locations showed a dominance of collector-gatherers and collector-filterers (Figure 22). In addition, Fowl2 had a greater number of scrapers.

Fowl2 and the Outfall showed a similarity of 61.9 % in their functional feeding groups (Table 24) and the Shannon diversity and evenness was higher at Fowl2 compared to the Outfall (Table 25). The Outfall had higher abundance in collector-gatherers (94) and collector-filterers (150) than Fowl2 (51, 26) (Figure 22). Fowl2 had higher number of shredders (6) and scrapers

(27), which were lower at the Outfall. Engulfer-predator and piercer-predator were found in low numbers at Fowl2 (1, 2) and the Outfall (4, 0). Piercer-predators were missing at the Outfall.

The downstream locations LR-12, LR-6, and LR-10 had a higher Shannon diversity index and evenness compared to the Outfall during spring 2015 (Table 25). LR-6 was the only downstream location with a higher Shannon's diversity and evenness than Fowl2. LR-12 (67) and LR-10 (150) showed a higher numbers of collector-filterers than collector-gatherers (LR-12 50, LR-10 127, Outfall 94) as the Outfall (150) (Figure 22). LR-6 had higher numbers of collector-gatherers (19) than collector-filterers (14) as Fowl2. However, Fowl2 showed a higher similarity with LR-12 (93.8%) than LR-6 (57.0%) (Table 24). LR-6 is the only downstream location which had all functional feeding groups represented (Figure 22). LR-12 was missing piercer-predators and LR-10 was missing scrapers. The Outfall and LR-10 showed the highest similarity (89.8%) whereas the Outfall and LR-6 showed the lowest similarity (30.3%) (Table 24).

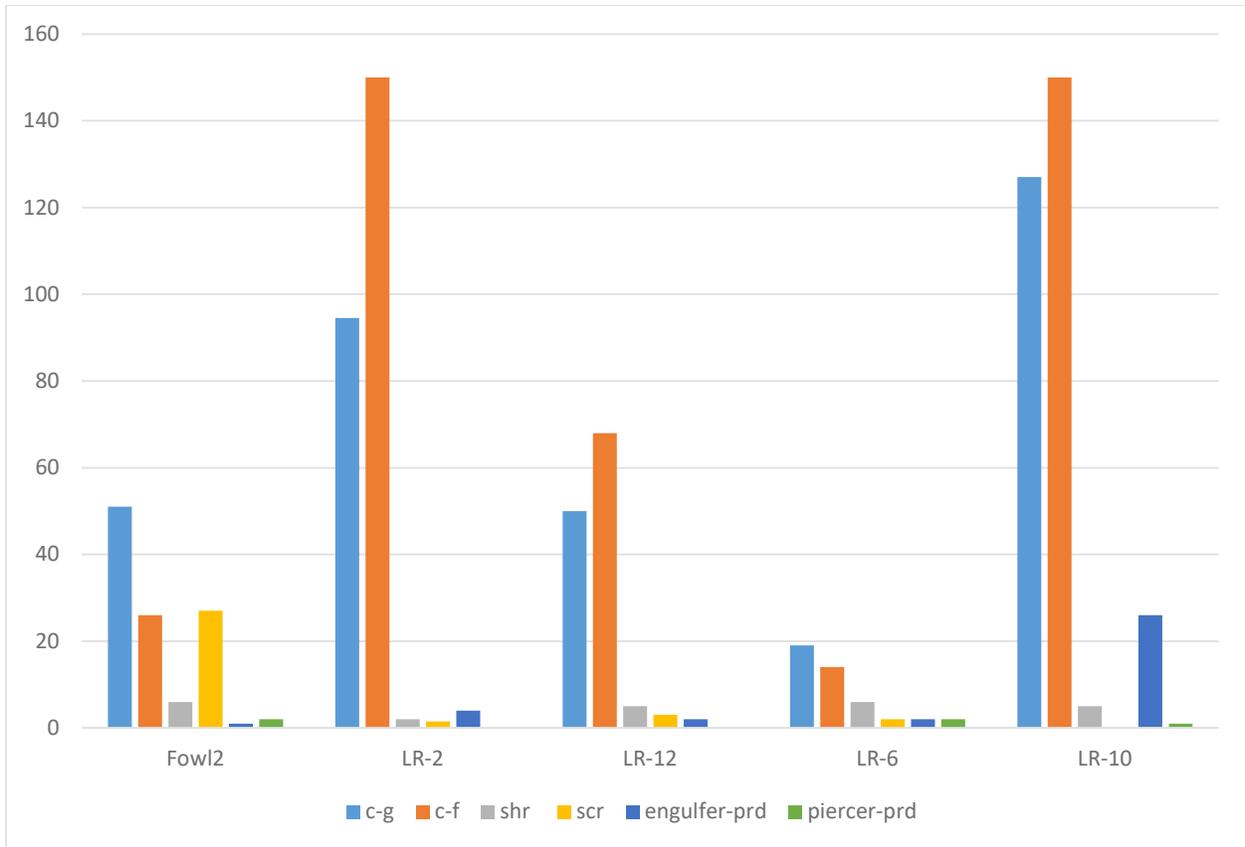


Figure 22. Numbers of individuals of functional feeding groups in spring. Collector-gatherers (c-g), collector-filterers (c-f), shredders (shr), scrapers (scr), engulfer-predators (engulfer-prd), and piercer-predators (piercer-prd). LR-2 is the Outfall.

Table 24. The Bray-Curtis Index was determined to compare the percentage similarity of functional feeding groups individuals of Fowl2 to the Outfall, LR-12, LR-6, LR-10 and Outfall to LR-12, LR-6, and LR-10 for spring 2015.

	FOWL2/ OUTFALL	FOWL2/LR- 12	FOWL2/LR- 6	FOWL2/LR- 10
BRAY-CURTIS INDEX	61.9%	93.8%	57.0%	53.6%
		OUTFALL/ LR-12	OUTFALL/ LR-6	OUTFALL/ LR-10
BRAY-CURTIS INDEX		67.4%	30.3%	89.8%

Table 25. Shannon diversity index and Shannon's evenness for the functional feeding groups at each location during spring 2015.

LOCATION	SHANNON DIVERSITY INDEX	SHANNON'S EVENNESS
FOWL2	1.31	0.73
LR-2	0.81	0.50
LR-12	0.98	0.61
LR-6	1.41	0.79
LR-10	1.01	0.63

3.2.2.2. Summer

During summer, collector-gatherers had a high number at all locations, however, collector-filterers had higher numbers at the downstream locations and the Outfall (Figure 23).

Fowl2 had a lower Shannon diversity than the Outfall (Table 27). Fowl2 (71) and the Outfall (61) had a high number of collector-gatherers and had a high similarity (97.3%) of functional feeding groups (Figure 23, Table 26). Collector-filterers showed the second highest number of functional feeding groups at the Outfall (17) and third highest at Fowl2 (7). Shredders had the second highest percentage at Fowl2 (14). Fowl2 had a low number of scrapers (2) and was missing engulfer-predator and piercer-predator. The Outfall was missing piercer-predator and the remaining functional feeding groups were represented in low numbers.

The downstream sites had a higher Shannon diversity and evenness than Fowl2 and LR-12 and LR-6 had a higher Shannon diversity than the Outfall (Table 27). The downstream locations also had high numbers of collector-gatherers (LR-12 40, LR-6 19, and LR-10 120) similar to Fowl2 and the Outfall (Figure 23, Table 26). LR-12 shared the highest similarity with Fowl2 (88.1%) and the Outfall (90.8%) among the downstream locations. The downstream locations showed a higher number of collector-filterers (LR-12 (23), LR-6 (14), and LR-10 (103)) than at Fowl2 (7). The remaining functional feeding groups were present at low numbers. LR-12 was missing piercer-predators, LR-6 did not have scrapers, engulfer-predator, and piercer-predator, and LR-10 was missing scraper and piercer-predators. LR-6 and LR-10 showed the lowest similarity with Fowl2 (LR-6 58.7%, LR-10 57.7 %) and the Outfall (LR-6 60.9%, LR-10 55.5%) (Table 26).

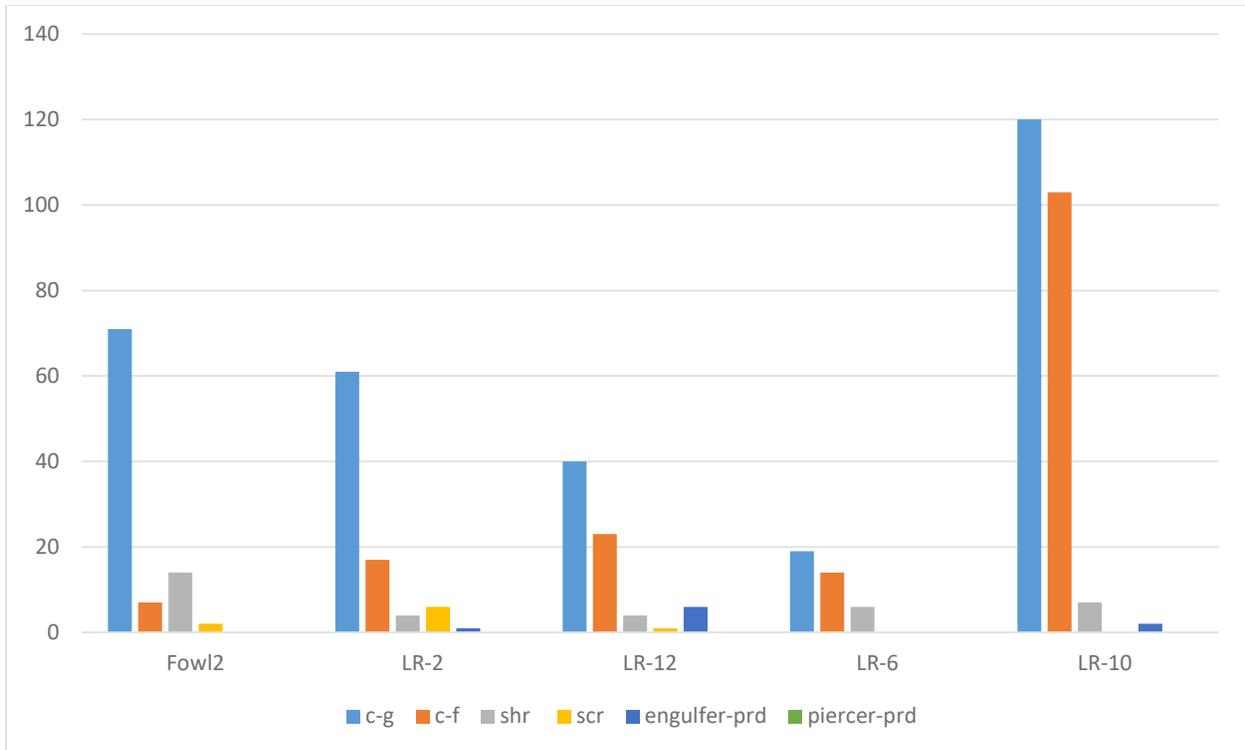


Figure 23. Numbers of individuals of functional feeding groups in summer. Collector-gatherers (c-g), collector-filterers (c-f), shredders (shr), scrapers (scr), engulfer-predators (engulfer-prd), and piercer-predators (piercer-prd).

Table 26. The Bray-Curtis Index was determined to compare the percentage similarity of functional feeding groups individuals of Fowl2 to the Outfall, LR-12, LR-6, LR-10 and Outfall to LR-12, LR-6, and LR-10 for summer 2015.

	FOWL2/ OUTFALL	FOWL2/ LR-12	FOWL2/LR-6	FOWL2/ LR-10
BRAY-CURTIS INDEX	97.3%	88.1%	58.7%	57.7%
		OUTFALL/ LR-12	OUTFALL/ LR-6	OUTFALL /LR-10
BRAY-CURTIS INDEX		90.8%	60.9%	55.5%

Table 27. Shannon diversity index and Shannon's evenness for the functional feeding groups at each location during summer 2015.

LOCATION	SHANNON DIVERSITY INDEX	SHANNON'S EVENNESS
FOWL2	0.77	0.56
LR-2	0.95	0.59
LR-12	1.12	0.69
LR-6	1.01	0.92
LR-10	0.85	0.61

3.2.2.3.Fall

All locations a high number of collector-gatherers and collector filterers with the exception of Fowl2, which did not have any collector-filterers (Figure 24).

Fowl2 had lower Shannon diversity and evenness than the Outfall (Table 29). Fowl2 (400) and the Outfall (47) had the highest number of collector-gatherers however they share a low similarity (35.4%) (Figure 24, Table 28). Fowl2 showed almost 100% collector-gatherers and a low number of shredders (2) scrapers (15), engulfer-predator (1), and piercer-predator (1). The Outfall had almost all functional feeding groups present with the exception of scrapers.

The downstream sites had a higher Shannon diversity and evenness than Fowl2 and the Outfall (Table 29). The downstream sites LR-12 (40) and LR-10 (29) showed a higher number of collector-filterers than Fowl2 (0) and the Outfall (23) (Figure 24). LR-10 did not differ appreciably between collector-gatherers (30) and collector-filterers (29). Most of the downstream location had all functional feeding groups present except piercer-predators were missing at LR-6 and LR-10. Fowl2 shared a low similarity with the downstream regions (LR-12 36.3%, LR-10 30.7%) with the lowest at LR-6 (19.8%) whereas the Outfall had a higher similarity with the downstream locations (LR-12 98.4%, LR-6 67.7%, LR-10 91.6%) (Table 28).

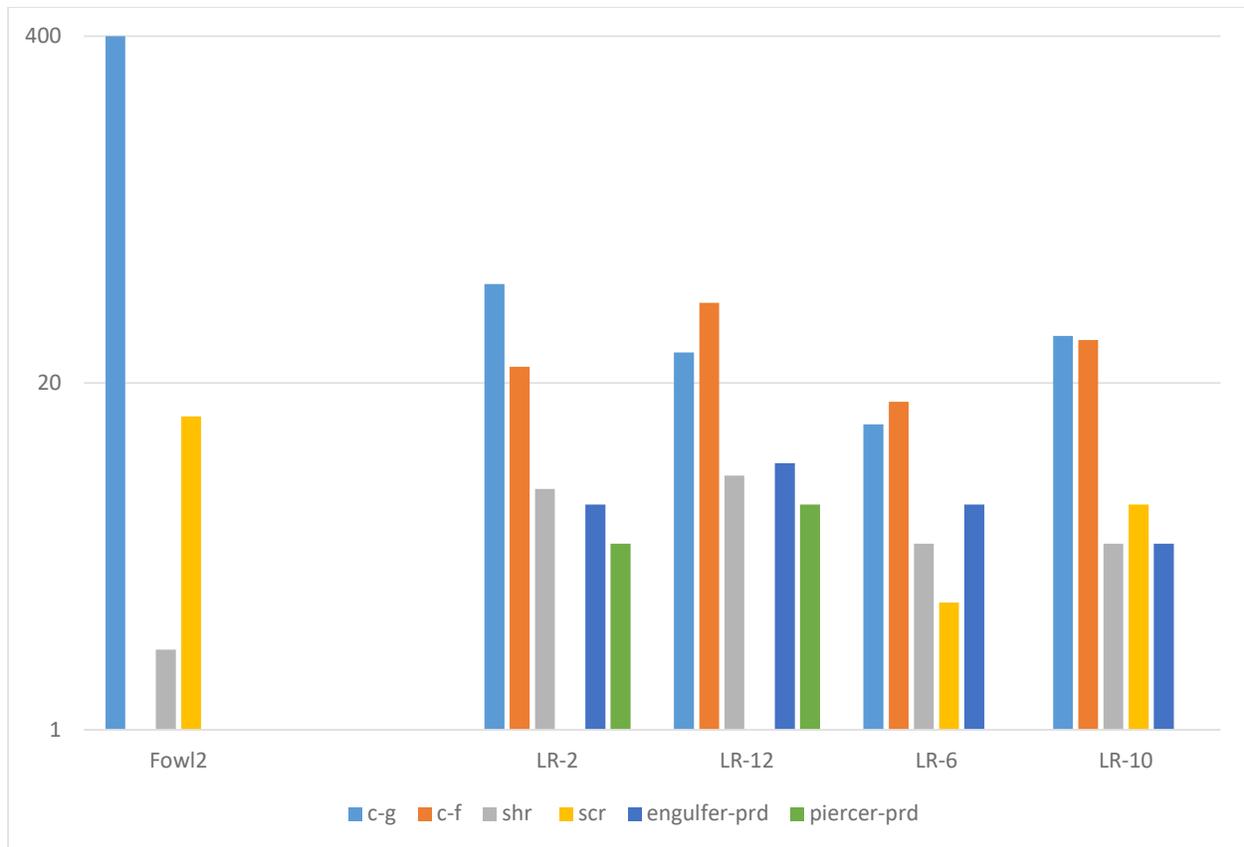


Figure 24. Numbers of individuals of functional feeding groups in fall. Collector-gatherers (c-g), collector-filterers (c-f), shredders (shr), scrapers (scr), engulfer-predators (engulfer-prd), and piercer-predators (piercer-prd). Y-axis was changed to logarithmic scale for a better visualization.

Table 28. The Bray-Curtis Index was determined to compare the percentage similarity of functional feeding groups individuals of Fowl2 to the Outfall, LR-12, LR-6, LR-10 and Outfall to LR-12, LR-6, and LR-10 for fall 2015.

	FOWL2/ OUTFALL	FOWL2/ LR-12	FOWL2/LR-6	FOWL2/ LR-10
BRAY-CURTIS INDEX	35.4%	36.3%	19.8%	30.7%
		OUTFALL/ LR-12	OUTFALL/ LR-6	OUTFALL/ LR-10
BRAY-CURTIS INDEX		98.4%	67.7%	91.6%

Table 29. Shannon diversity index and Shannon's evenness for the functional feeding groups at each location during fall 2015.

LOCATION	SHANNON DIVERSITY INDEX	SHANNON'S EVENNESS
FOWL2	0.22	0.16
LR-2	1.10	0.68
LR-12	1.23	0.77
LR-6	1.44	1.31
LR-10	1.31	0.95

CHAPTER IV

4. DISCUSSION

4.1. Thermal regime

The goal of this study was to evaluate the effects of the YCUA discharge on the thermal regime and macroinvertebrate family richness and diversity at the Lower Rouge River in Southeast Michigan. To do this, water temperatures were measured upstream and downstream of the YCUA discharge into the Lower Rouge River for three seasons from 13 April to 26 October, 2015 to observe daily and seasonal changes in water temperature. Additionally, the family richness, diversity, and functional feeding groups diversity of benthic macroinvertebrates below the discharge was determined to analyze the influence of the water temperature from the Outfall. Macroinvertebrates were sampled in spring, summer, and fall 2015 and past data of three years of macroinvertebrates sampling from FOTR were used for further analysis. Key results are discussed along with the original hypotheses for this thesis.

4.1.1. Temperature

Hypothesis 1: If the Outfall is affecting the water temperature, water temperatures will be lower for the control location Fowler 2 upstream of the Outfall.

This hypothesis was not supported by my data because the daily mean water temperatures at the Outfall were not warmer than Fowl2 during the research period. The daily mean water temperatures differed significantly for Fowl2 and the Outfall only for summer 2015. In contrast,

the daily mean water temperatures for spring and fall 2015 did not show any significant difference. The daily mean water temperatures at the Outfall were not higher than Fowl2, but during spring and summer 2015 both locations showed a difference in their thermal regime pattern. The Outfall showed an almost constant daily mean water temperature, while Fowl2 fluctuated in daily mean water temperatures. During fall 2015, the Outfall also showed a day-to-day variation in daily mean water temperatures that was almost comparable to the fluctuations of Fowl2 with few exceptions. The Outfall water temperature closely followed the air temperature pattern.

My data indicate that the Outfall did not represent a thermal discharge during the study timeframe, which is in contrast to the FOTR water temperature measurements from 14 November, 2014. The discharge even turned into a cool water discharge during summer 2015. Similar results were observed downstream of hypolimnetic impoundments discharges resulting in cooler summer water temperatures (Lehmkuhl 1972; Preece and Jones 2002). The daily mean water temperatures were similar for spring and fall 2015, however the day-to-day variation in thermal regime suggest that the Outfall affects the thermal regime pattern rather than the daily mean water temperature. The effects on downstream areas are discussed in the second hypothesis. The fall thermal regime pattern of the Outfall indicates that the discharged water was more likely influenced by the air temperature during fall 2015 compared to spring and summer 2015.

Thermal discharges can stabilize unstable thermal regime (Langford 1990). Even though, the Outfall was not a thermal discharge during spring and summer 2015, the-day to-day water temperature fluctuations were stabilized to an almost constant water temperature with little variation below the Outfall. The YCUA discharge affected the thermal regime below the Outfall during spring and summer 2015. The longitudinal influence on water temperature and thermal regime of the YCUA discharge in regards to the downstream sites will be discussed in the second

hypothesis.

Other studies have shown that thermal discharges from power plants increased the water temperature below the discharge (Alston et al. 1978; Coutant 1962; Mann 1965; Massengill 1976; Osborne and Davies 1987; Wellborn and Robinson 1996; Worthington et al. 2015). However, the YCUA discharge did not have higher water temperature compared to Fowl2. One of the reasons could be that a power plant discharge differs in their operation from a WWTP and therefore in the discharge temperature as well. The differences between the discharge from power plant and the WWTP is that power plants use the water from an adjacent water body for cooling purposes. During this process, water absorbs the heat and is discharged back into the waterbody, while a WWTP does not involve any human made external heating during the processes before discharging.

Hypothesis 2: If the Outfall is affecting the water temperature, the water temperature will be higher at the YCUA Outfall and decrease at downstream locations.

This hypothesis was rejected by my data because the daily mean water temperature at the Outfall was not higher than the downstream locations LR-12, LR-6, and LR-10. The Outfall and the downstream sites were similar in their daily mean water temperatures for spring 2015. However, Fowl2 was also similar in its daily mean water temperature to the Outfall and the downstream sites. All locations daily mean water temperature correlated highly with the daily mean air temperature. Therefore, all locations were highly influenced by the air temperature rather than the YCUA discharge temperature. During summer 2015, the Outfall differed significantly in mean daily water temperatures from the downstream sites and also during fall 2015 for the downstream sites LR-12 and LR-6. Surprisingly the Outfall did not discharge warmer water as expected from the November 2014 data, but was cooler than the downstream locations.

Interestingly, Fowl2 was similar in daily mean water temperatures to the downstream sites during summer 2015. Fowl2 and the downstream sites were more influenced in their water temperature by the air temperature rather than the Outfall. In addition, The Outfall was less influenced by the air temperature during summer 2015 compared to spring and fall 2015. The day-to-day variation of the mean water temperatures between the Outfall and the downstream locations during spring and summer 2015 showed little correlation. While the downstream sites fluctuated on a day-to-day basis in their mean daily water temperatures, the Outfall remained almost constant. In addition, Fowl2 and the downstream sites showed almost the same day-to-day variation in mean daily water temperature for spring and summer 2015.

The similar daily mean water temperature at the Outfall and the downstream site during spring 2015 could suggest an influence on the thermal regime from the Outfall to the downstream sites. On the other hand, the similarity of the daily mean water temperatures of all locations during spring indicates that the temperature influences from the Outfall are not solely contributing to the thermal regime of the downstream sites. The cooler water temperature could be retained during transportation due to the 10.8 km and 2.1 m deep underground pipe. The similar daily mean water temperatures at Fowl2 and the downstream sites indicates that the downstream locations show no detectable influence in water temperature by the Outfall. Additionally, the day-to-day variation in mean daily water temperatures indicates that the Outfall did not have a detectable influence on the downstream sites during spring and summer 2015. Also the similar day-to-day pattern in mean daily water temperature between Fowl2 and the downstream sites indicates that the downstream regions follow a natural temperature fluctuation. The downstream sites are more influenced by the air temperature during spring and summer 2015 rather than by the water temperature from the Outfall. It seems that the cool water temperature and its influence on the downstream thermal

regime is limited to its immediate downstream areas of the Outfall and returns to its natural thermal regime before LR-12.

Nonetheless, Fowl2 differed significantly from the downstream sites in daily mean water temperatures while being similar with the Outfall during fall 2015 which indicates that Fowl2 and the Outfall had no detectable influence on the water temperatures of the downstream sites. During fall, the riparian vegetation loses canopy cover which could result in stronger influence of solar radiation on the river water temperature (Caissie 2006). Therefore, the air temperature had a high influence on each site. In addition, the river discharge had its lowest discharge volume during fall 2015 which could result in higher influence of air temperature on the water temperature due to the fact less water is present and less energy required to heat the water. As discussed in the previous section, the almost constant mean daily water temperature pattern of the Outfall during spring and summer 2015 was not observed during fall 2015. The day-to-day mean temperature variation of the Outfall seemed to follow roughly the pattern of the other locations and the air temperature. However, a more detailed study during fall is required to understand the thermal regime pattern of the Outfall.

The dimension of the thermal regime alteration by thermal discharges depends on their temperature and the water temperature of the receiving water body (Coulter et al. 2014). Additionally, the ratio of the volume of the thermal discharge and the river could provide insight into the mass flow dynamics of the discharge on the river for assessing thermal alteration. Similar approaches have been done in considering the ratio of maximum storage capacity to median annual runoff of impoundments and their influence on downstream river temperature (Maheu et al. 2016).

Hypothesis 3: If the Outfall is affecting the water temperature, the difference in water temperature between the Outfall and Fowler 2 will be greater during spring and fall compared to summer.

My data did not support this hypothesis because the daily mean water temperature difference during spring and fall 2015 was not higher compared to summer 2015. During spring and fall 2015, the Outfall and Fowl2 were similar in daily mean water temperatures while during summer 2015 the difference was significant. The Outfall retained a cooler temperature during summer 2015 than the upstream location at Fowl2. One possible explanation for this is the underground effluent transport from the YCUA to the Outfall.

The seasonal difference between the Outfall and the downstream sites indicated higher daily mean water temperature variation during summer and fall 2015 than spring 2015. The downstream sites showed unexpectedly warmer daily mean water temperatures than the Outfall during summer and fall 2015, while during spring 2015 they were similar (i.e. insignificant variation). The warmer temperature of the downstream sites during summer could be explained by higher air temperature and the influence of atmospheric conditions on water temperature (Caissie 2006). Riparian vegetation could be a buffer for influencing solar radiation on water temperature, by blocking it from reaching the river. However, during fall, the riparian vegetation loses canopy cover, which allows more solar radiation to reach the water surface and heat it up. These reasons could lead to cooler water temperatures during summer and warmer conditions during fall. The different outcome of Fowl2 and the downstream site in regard to the Outfall for fall 2015, suggests that the close geographic location between the Outfall and Fowl2 resulted in less variation than the downstream sites.

4.2. Macroinvertebrates

Hypothesis 4: If the Outfall is affecting the water temperature, family richness and diversity will be higher at the upstream location, Fowler 2, compared to the YCUA Outfall.

This hypothesis was rejected because the upstream control location Fowl2 did not show a higher family richness than the Outfall from spring to fall 2015 and additionally, Fowl2 did not had higher macroinvertebrate diversity during summer and fall 2015. Even though, the mean water temperatures for spring 2015 were similar for the two sites, the Outfall showed a higher number of individuals and higher family richness than Fowl2. One reason for higher family taxa in spring at the Outfall could be that during the sampling time (between April 17 to 20, 2015), the daily mean water temperatures for the Outfall were in general higher than Fowl2 which could have contributed to higher number of macroinvertebrates. A mild increase in water temperature can support abundance and secondary production of benthic macroinvertebrates because it improves growth and production which results in greater species success (LeRoy Poff and Matthews 1986; Wellborn and Robinson 1996; Worthington et al. 2015).

However, Fowl2 showed higher diversity than the Outfall which could be due to the riffle habitat under the bridge at Fowl2 whereas Outfall was lacking in riffle habitats and mainly constituted of runs and pools. Riffle habitats are shallow and more turbulent which results in more oxygenated water and allow more sensitive taxa to inhabit these areas.

The temperature between both locations differed significantly during summer 2015, but there were no differences in family richness and diversity. These findings indicate that the difference in temperature during summer 2015 was not high enough to cause significant difference in family richness and diversity. Studies showed that thermal discharges that lead to water temperatures between 38 and 42°C resulted in the reduction of macroinvertebrates (Coutant 1962;

Wellborn and Robinson 1996). Durance and Ormerod (2007) observed a decrease in springtime abundance in a small river for a 1°C increase in water temperature. Fowl2 showed warmer water temperature than the Outfall during summer 2015, but this difference did not exceed 1 °C for the mean seasonal water temperature. One would assume that the Outfall would show more family richness and diversity during summer because the Outfall was cooler than Fowl2. Nevertheless, the Outfall decreased in the number of family richness from spring to summer 2015 while Fowl2 remained the same. One reason for the decrease in family richness at the Outfall during summer could be that many macroinvertebrates emerge to adult insects from spring to early fall and leave the water for breeding (Wallace and Anderson 1996).

During fall 2015, the Outfall showed higher family richness and diversity than Fowl2, even though both locations had similar daily mean water temperatures. Water temperatures at these five sites during the research period seemed to not contribute to a change in macroinvertebrate community. These results indicate that something other than the water temperature might be causing the differences in family richness, such as a change in nutrient distribution which is discussed later.

The spring and fall data of Fowl2 and the Outfall (unfortunately had data for the past one year) for the past three years, showed a variation in family richness for each site. The families Heptageniidae, Amphipoda, Isopoda, Oligochaeta, and Chironomidae reappeared at Fowl2 in the past three years, while I cannot report the families for the last three years for the Outfall. The macroinvertebrate community and family richness seemed to vary for Fowl2 each year and each seasons with the exception of the mentioned five families. This variation in family richness and community makes it difficult to relate water temperature with macroinvertebrate community. A long-term analysis is required in order to understand better the relationship of water temperature

and macroinvertebrate community. In addition, the Bray-Curtis similarity showed low similarity between both locations during spring and fall 2015, even though both locations are geographically close to each other. One reason could be the difference in nutrient distribution. However, the Bray-Curtis similarity was higher during summer 2015, which could be due to the fact that many macroinvertebrates are emerging to adult insect and leaving the water, while a low diversity of insect remains in the water, which could have caused a more similar diversity and family richness.

Hypothesis 5: If the Outfall is affecting the water temperature, family richness and diversity will increase in downstream direction from the Outfall.

This hypothesis was not supported by my data because I did not observe an increase in family richness and diversity in the downstream direction except during spring 2015 from LR-2 (Outfall) to LR-6. During spring 2015, family richness and diversity increased from the Outfall to LR-6; however, LR-10 showed a lower diversity and family richness than the Outfall. Additionally, the Bray-Curtis similarity index varied between the Outfall and the downstream sites for each location. Since the mean daily water temperature was not significantly different, water temperature is not the contributor to the difference in the macroinvertebrate community. Something other than water temperature seems to be responsible for different family richness and diversity. One reason for the variation could be a change in nutrient distribution, which is discussed in the section about functional feeding groups.

Summer and fall 2015, on the contrary, showed significant difference in water temperature between the Outfall and the downstream sites; however, both seasons had a different outcome. In both cases, the family richness did not increase in the downstream direction from the Outfall, but rather varied for each location. Also the Bray-Curtis similarity varied for each location between the Outfall and the downstream sites. Interestingly, the Shannon diversity showed a decrease from

LR-12 to LR-10 in the downstream direction. The data from the past three years also showed a variation of macroinvertebrate family richness at each location for each year, during spring and fall. As mentioned before, the variation in macroinvertebrate family richness and diversity is most likely related to other factors than the temperature, which is discussed later.

Hypothesis 6: If the Outfall is affecting the water temperature, family richness and diversity will be higher for spring and fall compared to summer.

This hypothesis was partially supported by my data with the exception of Fowl2 and LR-2. During summer 2015, all locations showed a lower number of individuals and family taxa number than in spring 2015, except for LR-12. One of the reasons for the lower family richness and lower number of individuals could be that many macroinvertebrates leave the water, metamorphosing into adults usually during late spring to early fall for breeding (Wallace and Anderson 1996). The individual numbers of sensitive taxa were low at every location with the exception of LR-10, which had a high number of Baetidae. All locations showed a high number of collector-gatherers in the functional feeding groups and collector-filterers did not dominate the Outfall and downstream sites as in spring, which suggests a lower amount of suspended fine particulate organic matter. These results indicate a shift in nutrient distribution and availability which either is the result of a change in the Outfall discharge composition during summer or is due to the difference in nutrient availability between seasons or both.

Interestingly, Fowl2 showed its highest individual numbers of macroinvertebrates in fall in comparison to spring and summer, while the other sites had low numbers of individuals. In comparison to spring, all locations, with the exception of LR-10, showed a decrease in family richness while LR-10 showed an increase. One reason for this could be that many macroinvertebrates emerged to the adult stage during early spring to summer for breeding. The

freshly-hatched macroinvertebrates during summer and early fall could be too small to be sampled by the D-net I used for this study.

For spring, summer, and fall 2015, I did not observe any impairments to the macroinvertebrate community and population to the downstream sites in comparison to the control, even though Fowl 2 and LR-12 and LR-6 significantly differed in water temperature during fall 2015. One possible explanation for this is that the mean water temperature difference for each season was less than 1 °C between the control site and the Outfall. Other studies (Coutant 1962; Wellborn and Robinson 1996) have reported changes in macroinvertebrates below the thermal discharge in density and biomass, however these studies were experiencing higher water temperature changes of over 38 °C than those observed at the YCUA discharge. Hogg et al. (1995) and Hogg and Williams (1996) observed a reduction of macroinvertebrate densities with an increase of 2 to 3.5 °C in water temperature and Durance and Ormerod (2007) observed a decrease in macroinvertebrates in springtime abundance in a small river for an increase of water temperature of 1 °C. Yet, during my study period, the Outfall did not reach an increase of 1 °C for the mean seasonal water temperature in spring, summer, and fall 2015 compared to the control site. Interestingly, the discharge was colder during summer months than the control site and the downstream locations.

Some macroinvertebrate families might be indicators for thermal pollution. Alston et al. (1978) reported a dominance of Oligochaeta and Chironomidae below thermal discharges and LeRoy Poff and Matthews (1986) saw an increase in Hydropsyche *Cheumatopsyche* further downstream of thermal discharges. Benda and Proffitt (1974) also observed an increase in Chironomidae. In my study, Chironomidae and Hydropsyche were two of the dominant taxa which indicates a possible thermal influence from the discharge. However, another study analyzed

Chironomidae in groundwater reservoir Southern Ontario and their sensitivity to thermal changes (Tixier, Wilson, and Williams 2008). This study found that Chironomidae abundance and species composition are sensitive to temperature changes. Hogg and Williams (1996) also observed lower densities of Chironomidae. These studies show that thermal discharges do not produce uniform effects especially for mild temperature increases. Durance and Ormerod (2007) reported that variation in family abundance of macroinvertebrate is more likely related to water quality rather than to thermal discharge. Additionally, the response to changes in water temperature or other factors, such as velocity and chemistry, depends on each species genetic variation, their variation among sites and the gene flow (Hogg et al. 1995; Hogg and Williams 1996). Therefore, abundance and community structure depend on more factors than just temperature.

4.2.1. Functional feeding groups

Hypothesis 7: If the Outfall is affecting the water temperature, functional feeding groups diversity of macroinvertebrates will increase with decreased water temperature.

My hypothesis was not supported by my data because the diversity of functional feeding groups did not show a relationship to water temperature. Spring 2015 showed the lowest mean seasonal water temperature and the highest diversity of functional feeding groups at Fowl2, while summer and fall 2015 had lower diversity. On the other hand, the Outfall and the downstream sites LR-12, LR-6, and LR-10 had the highest diversity during fall 2015. One reason for this difference was explained in the previous section with the emergence of macroinvertebrates to adult insects and leaving the water from late spring to early fall. Even though the daily mean water temperature is similar for all location during spring 2015, all location had different functional feeding groups diversity.

Another reason than the water temperature for the different diversity during spring 2015 could be the turbulence at the Outfall and nutrient introduction created by the YCUA effluent discharge (LeRoy Poff and Matthews 1986). YCUA effluent discharge needs to meet permit discharge regulations by the MDEQ to protect the stream water quality (YCUA 2012b); nevertheless, even modern WWTPs will introduce additional nutrients into the stream (Gücker, Brauns, and Pusch 2006; Spänhoff et al. 2007). This additional nutrient load could have an impact on the macroinvertebrates food web structure. The most abundant families at Fowl2, Outfall, and the downstream sites LR-12 and LR-10 represent mainly the functional feeding groups collector-gatherers and collector-filterers with Outfall and LR-12 having the highest abundance. Both functional feeding groups consume fine particulate organic matter and microbiota; however, they differ in their feeding preference. Collector-gatherers are deposit feeders while collector-filterers are suspension feeder (Allan and Castillo 2007b). The shift in functional feeding groups towards higher numbers of collector-filterers as opposed to collector-gatherers at the Outfall, LR-12, and LR-10 suggests a change in nutrient distribution and availability. LeRoy Poff and Matthews (1986) reported an increase in collector-filterers and scrapers below thermal discharge locations. They attributed the increase mainly to the reduced processing of organic matter by the macroinvertebrates which resulted in a higher availability of suspended fine particulate organic matter. The Outfall, LR-12, and LR-10 showed an increase in collector-filterers, while the number of scrapers were lower for the Outfall and LR-12. These data suggest an increase of suspended fine particulate organic matter below the Outfall. They suggested the nutrient output from the thermal discharge was the main culprit of a shift in functional feeding groups. However, the study by LeRoy Poff and Matthews (1986) was a short-term study and concentrated during the winter months from December 1982 to February 1983. This short-term study during the winter of 1982

and 1983 limits the applicability to the other seasons spring, summer, and fall. However, my data indicate the shift in nutrient availability was most likely caused by the YCUA effluent and the effect of the turbulence discharge in relocating deposit food into the water column. Nonetheless, non-point pollution is probably another source of nutrient and organic matter input.

4.3. Conclusion

This study was conducted to determine if the YCUA effluent discharge has an effect on the thermal regime of the Lower Rouge River and if the effects on the thermal regime could be observed on the macroinvertebrate family richness, diversity, and functional feeding groups. Water temperatures were measured from 13 April to 26 October, 2015 at two control stations, one at the Outfall, and three downstream of the Outfall to observe the daily water temperature and its longitudinal magnitude. In addition, macroinvertebrates were sampled at five temperature locations during spring, summer, and fall 2015 to determine family richness, diversity, and functional feeding groups.

The Outfall was not a thermal discharge during spring, summer, and fall 2015, contrary to the measurements made by FOTR during the fish survey in November, 2014. In contrast, the Outfall turns into a cooler discharge during summer and fall 2015 months. In addition, the Outfall had an almost constant daily mean water temperatures during spring and summer 2015, while fall 2015 daily mean water temperatures showed daily variations. The influence of the Outfall on the thermal regime was not observed at the downstream regions which showed a more similar thermal regime to Fowl2. The water temperatures at the downstream sites are more influenced by the air temperature rather than by the Outfall. Therefore, the impact of the Outfall on the thermal regime is limited to its immediate downstream areas. Interestingly, family richness and diversity in general were higher at the Outfall in comparison to the control station Fowl2, which suggests a positive

impact of the YCUA effluent on macroinvertebrate community. However, family richness and diversity at the downstream sites varied at each location. These variations were possibly the result of additional nutrients and a change in their distribution caused by the YCUA effluent, which was observed in the shift of the functional feeding groups from collector-gatherer to collector-filterer. My results suggest that the YCUA effluent discharge had no detectable influence on the water temperature and thermal regime downstream of the Outfall during the research period from April 13 to October 26, 2015.

4.4.Future research

For the future, a more concentrated water temperature measurement over a longer period of time between the Outfall and the first downstream location LR-12 is needed to understand the thermal influences of the WWTP on the daily and seasonal water temperature and the thermal regime on the immediate downstream areas. Additionally, a more frequent sampling of the macroinvertebrate community with a focus on temperature sensitive taxa at each water temperature logger would allow observations of the effects of the WWTP on macroinvertebrates over a longer period of time below the discharge.

APPENDIX 1

Monthly mean daily water and air temperature from April to October 2015

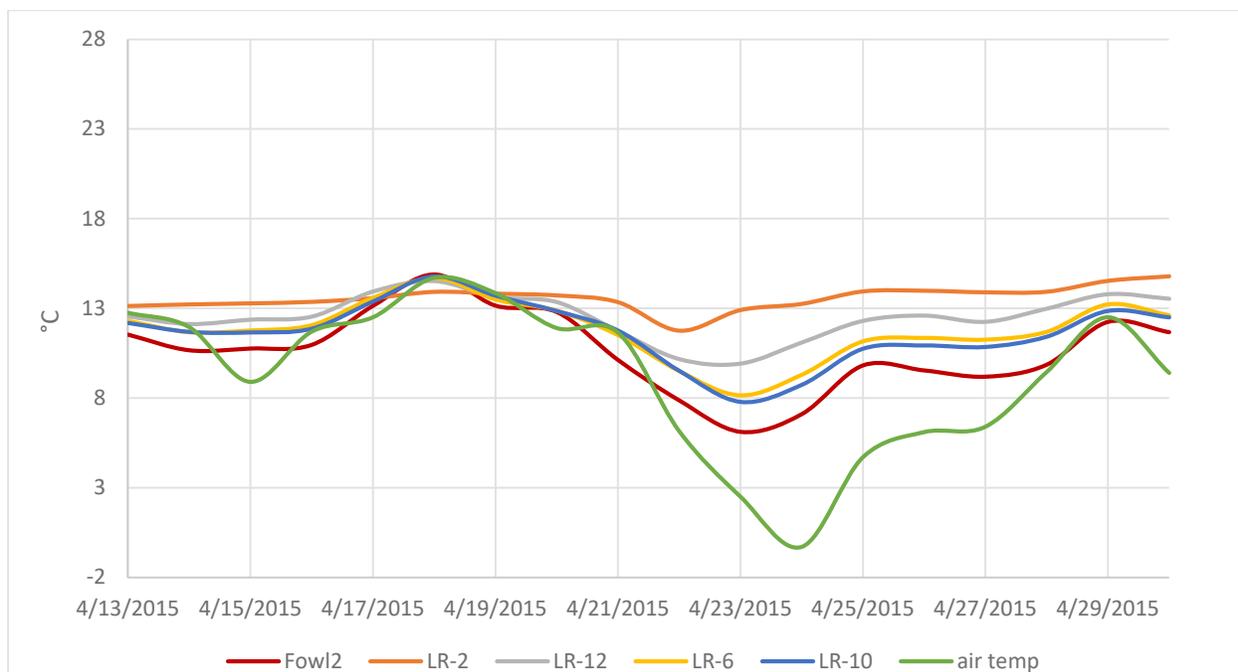


Figure 25. Mean daily water and air temperature for April 2015 calculated from the raw 24 hourly measured water temperature data.

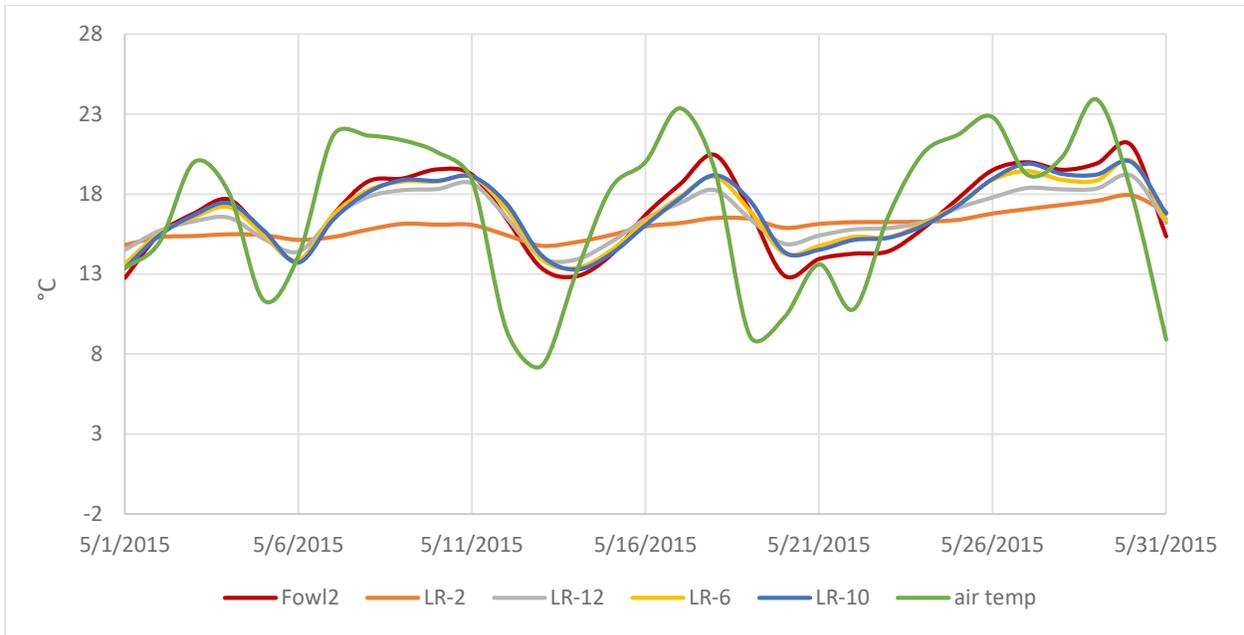


Figure 26. Mean daily and air water temperature for May 2015 calculated from the raw 24 hourly measured water temperature data.

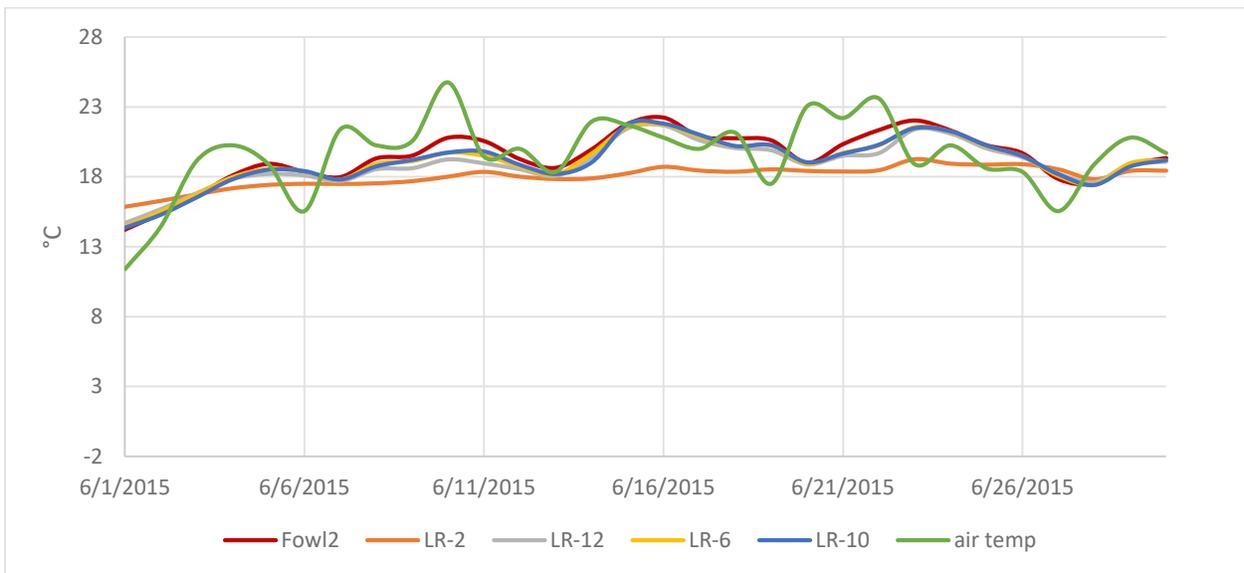


Figure 27. Mean daily and air water temperature for June 2015 calculated from the raw 24 hourly measured water temperature data.

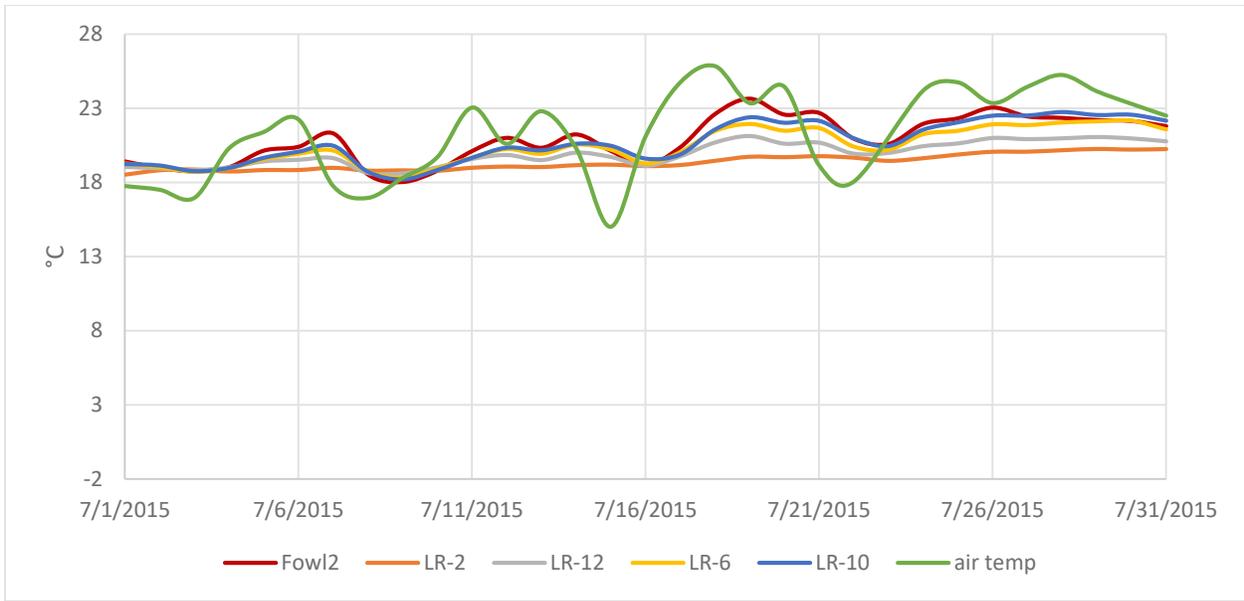


Figure 28. Mean daily water and air temperature for July 2015 calculated from the raw 24 hourly measured water temperature data.

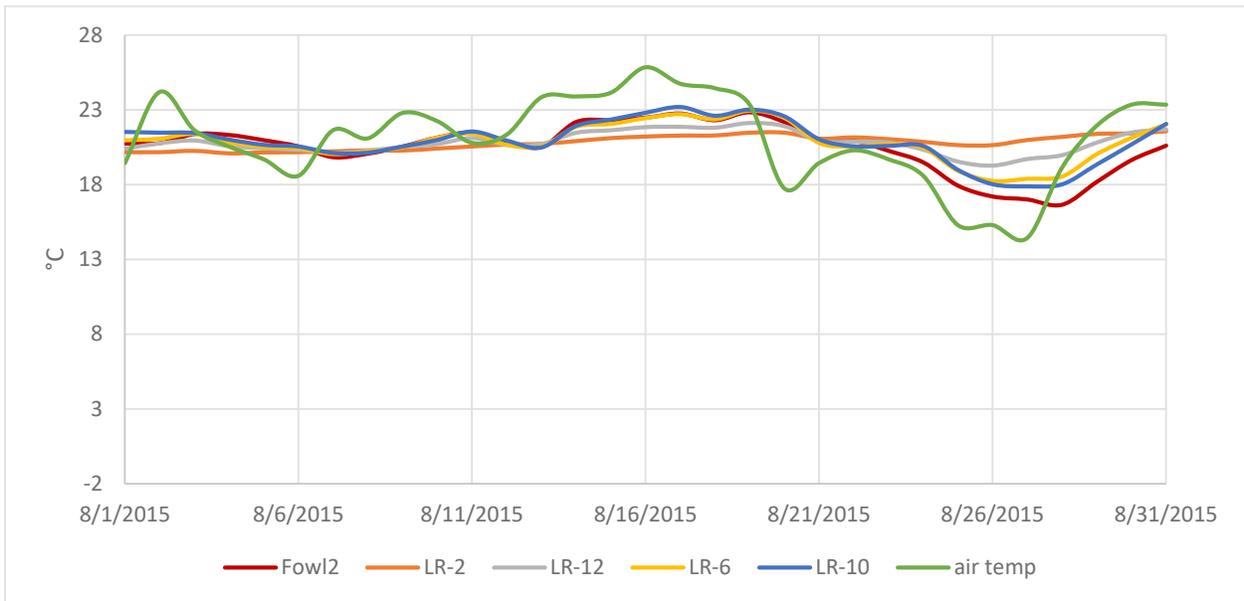


Figure 29. Mean daily water and air temperature for August 2015 calculated from the raw 24 hourly measured water temperature data.

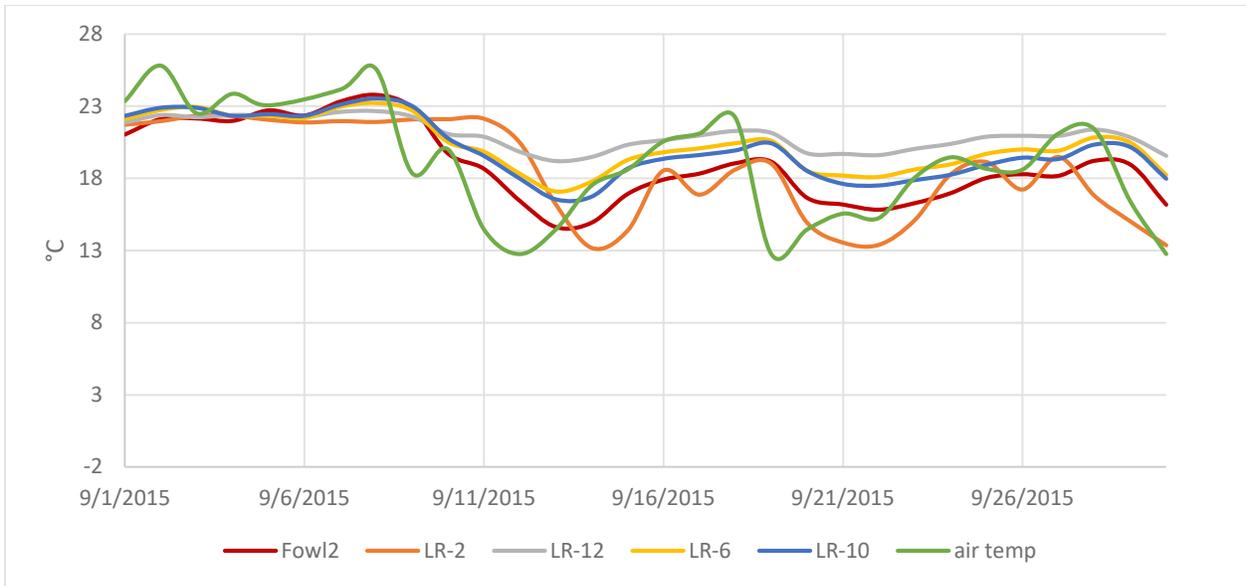


Figure 30. Mean daily water and air temperature for September 2015 calculated from the raw 24 hourly measured water temperature data.

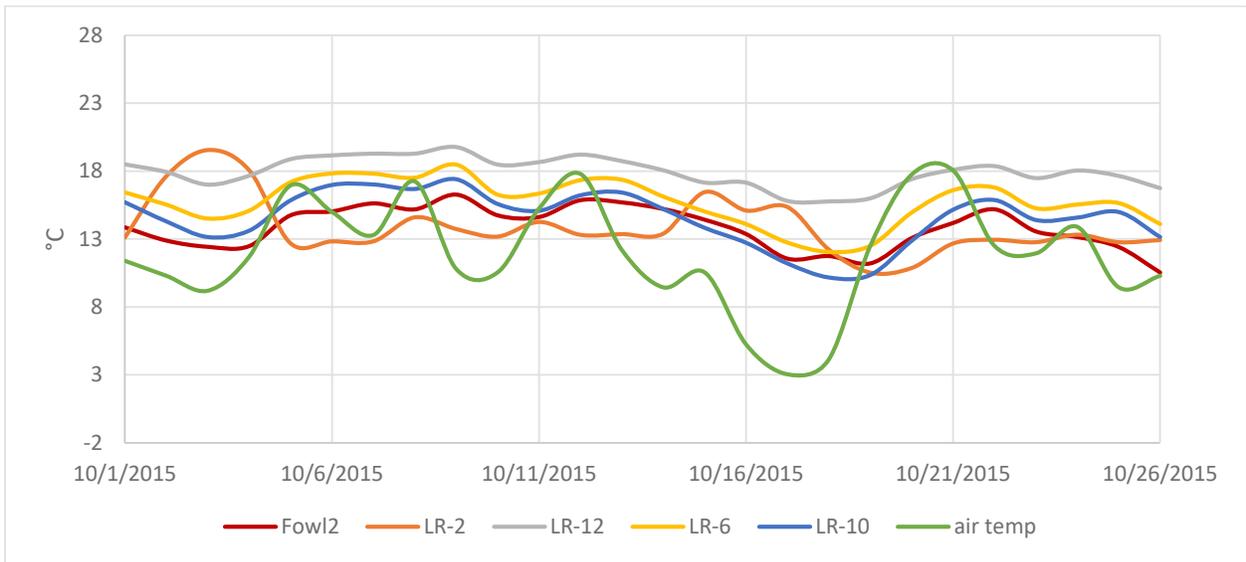


Figure 31. Mean daily water and air temperature for October 2015 calculated from the raw 24 hourly measured water temperature data.

APPENDIX 2

Mean water temperature difference between the control site and LR-2, LR-12, LR-6, and LR-10

Table 30. Mean water temperature difference between the control site Fowl2 and the sites LR-2, LR-12, LR-6, and LR-10 for the three seasons spring, summer, and fall 2015.

SEASONS	T_WATER DIFFERENCE	T_WATER DIFFERENCE	T_WATER DIFFERENCE	T_WATER DIFFERENCE
	LR-2 - FOWL2	LR-12 - FOWL2	LR-6 - FOWL2	LR-10 - FOWL2
SPRING	0.22	0.33	0.25	0.21
SUMMER	-0.81	-0.34	-0.10	0.02
FALL	-0.20	3.00	1.61	0.91

APPENDIX 3

Mean water temperature difference between the Outfall and LR-12, LR-6, and LR-10

Table 31. Mean water temperature difference between the Outfall and the downstream sites LR-12, LR-6, and LR-10 for the three seasons spring, summer, and fall 2015.

SEASONS	T_ WATER DIFFERENCE	T_ WATER DIFFERENCE	T_ WATER DIFFERENCE
	LR-12 - OUTFALL	LR-6 - OUTFALL	LR-10 - OUTFALL
SPRING	0.12	0.03	0.00
SUMMER	0.47	0.70	0.83
FALL	3.20	1.81	1.11

APPENDIX 4

Discharge rate at the Outfall and at the Lower Rouge River in Inkster, MI from April to October

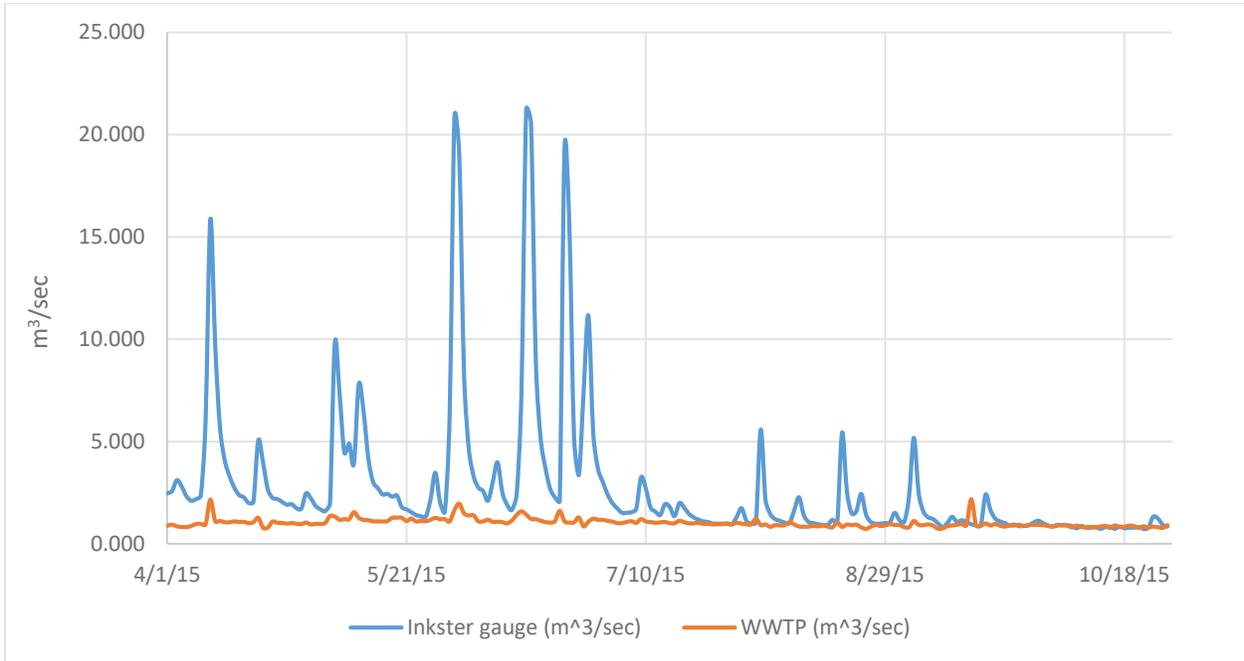


Figure 32. Discharge rate in cubic meter per second (m³/sec) for the Outfall and Inkster USGS gauge from April to October 2015.

APPENDIX 5

Monthly discharge rate at the Outfall and at the Lower Rouge River in Inkster, MI

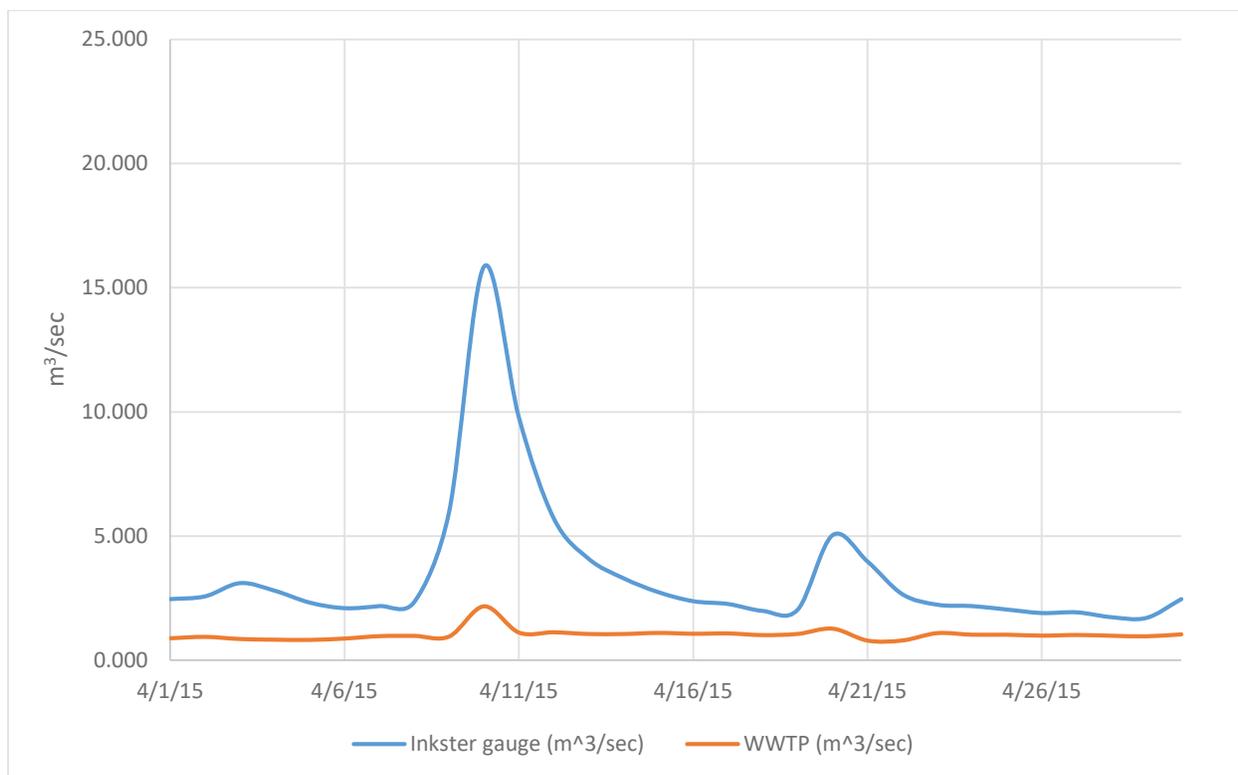


Figure 33. Discharge rate in cubic meter per second (m³/sec) for the Outfall and Inkster USGS gauge in April 2015.

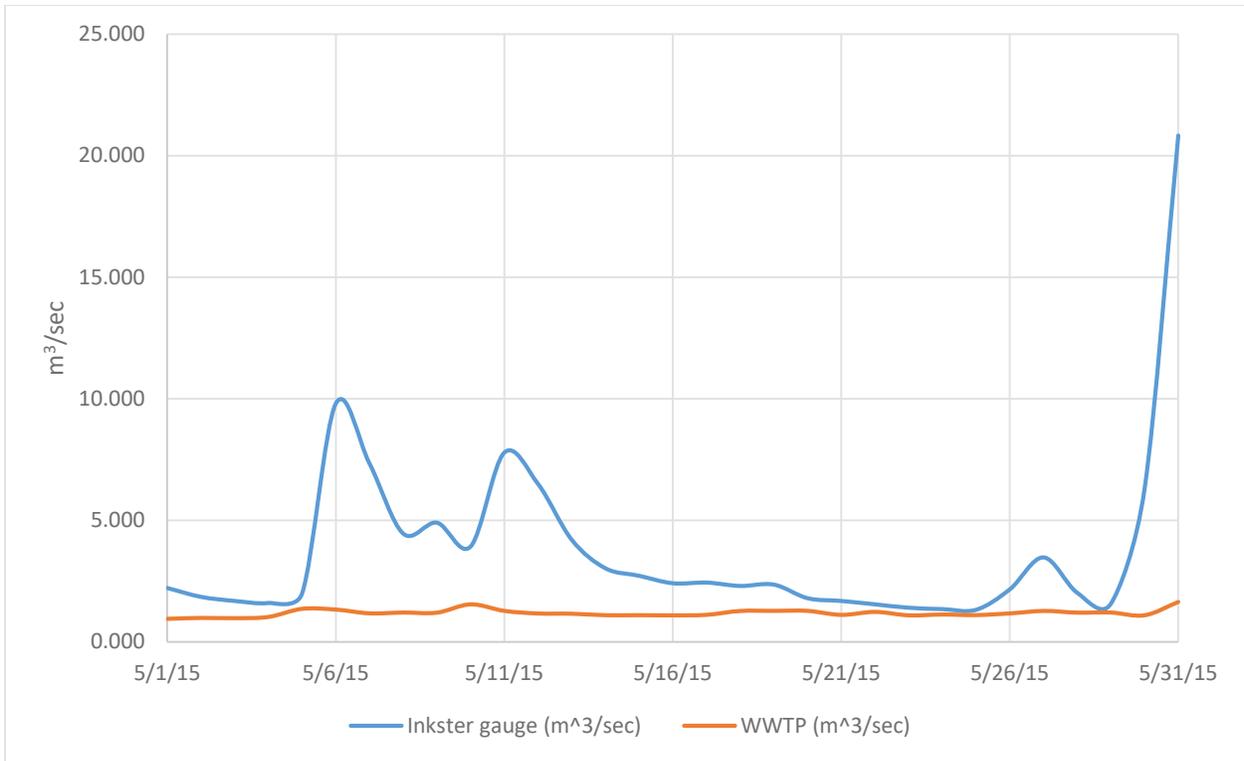


Figure 34. Discharge rate in cubic meter per second (m³/sec) for the Outfall and Inkster USGS gauge in May 2015.

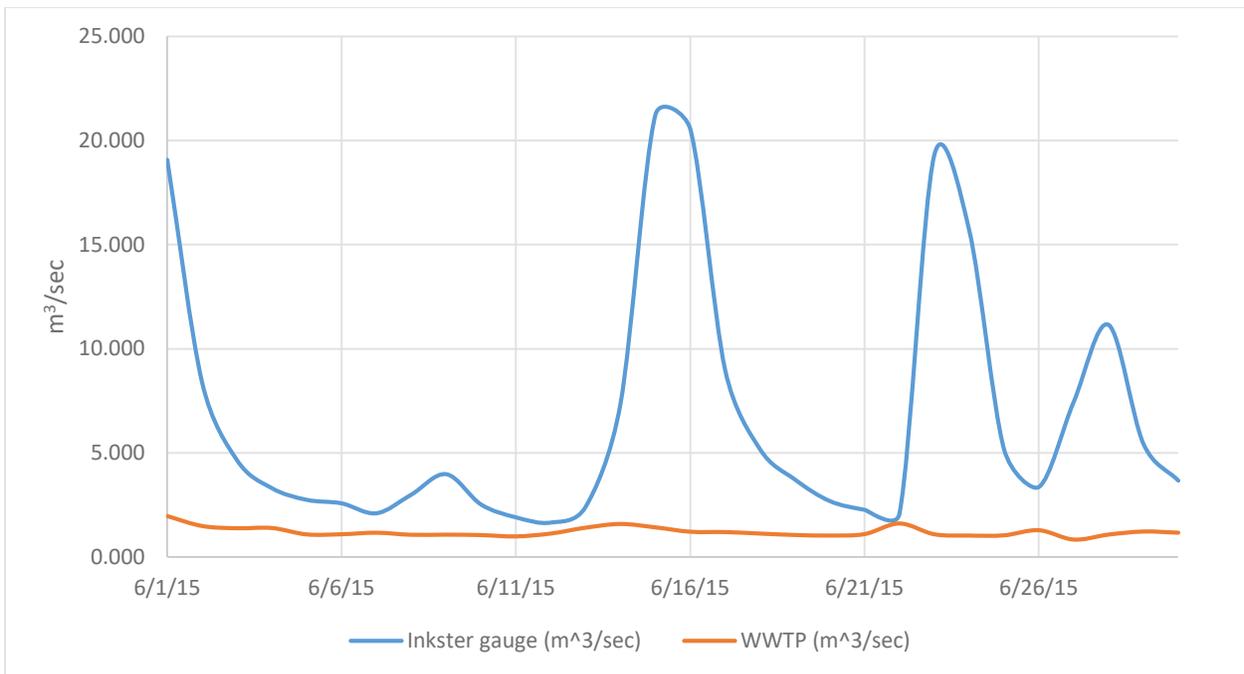


Figure 35. Discharge rate in cubic meter per second (m³/sec) for the Outfall and Inkster USGS gauge in June 2015.

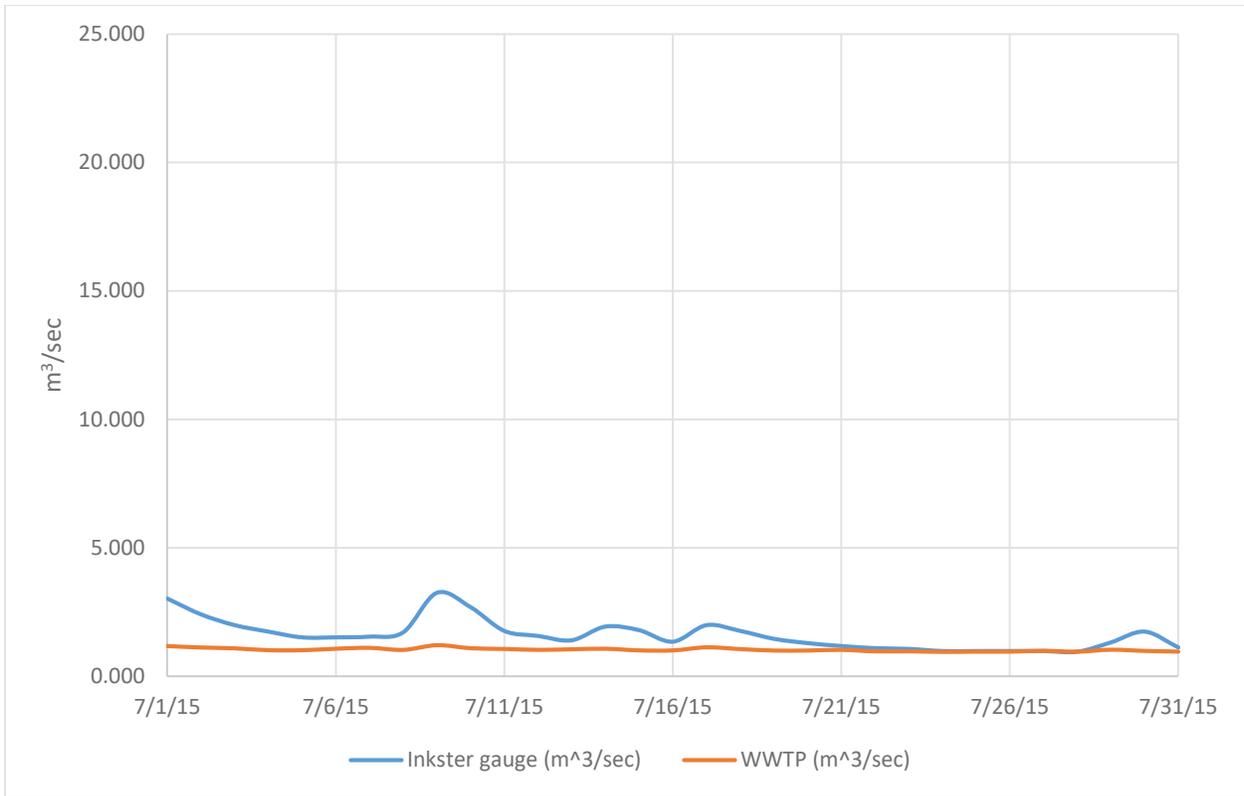


Figure 36. Discharge rate in cubic meter per second (m³/sec) for the Outfall and Inkster USGS gauge in July 2015.

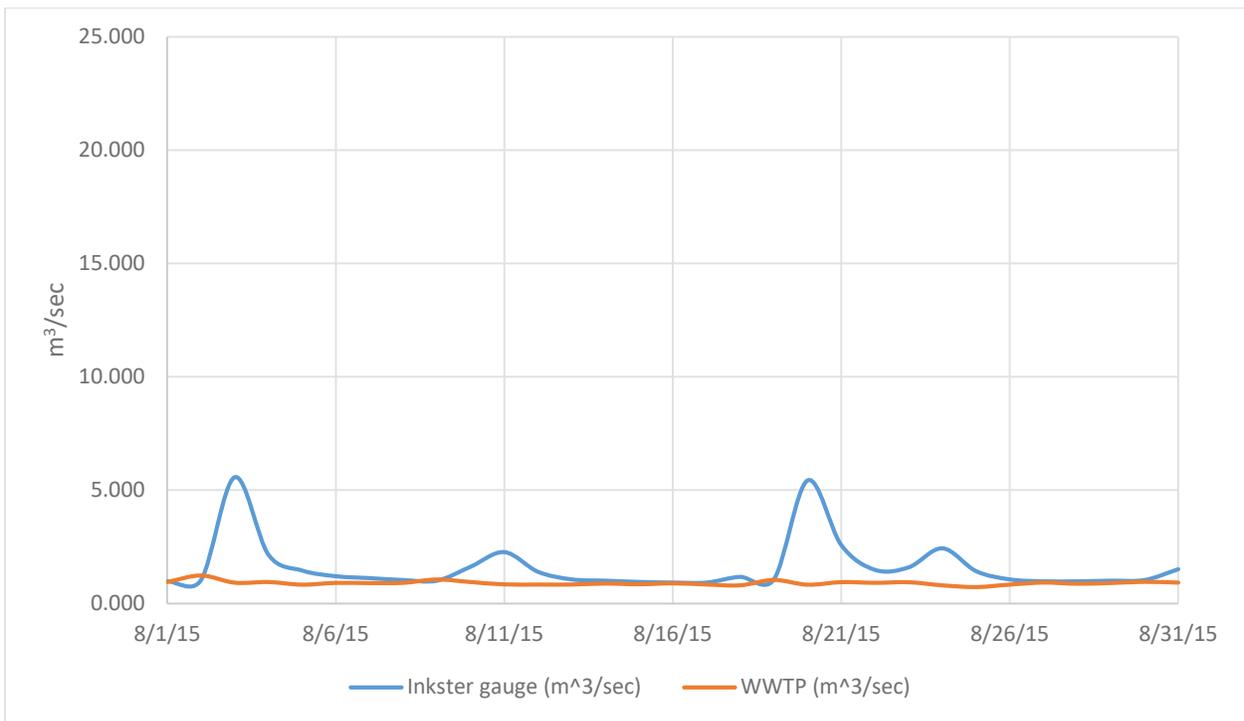


Figure 37. Discharge rate in cubic meter per second (m³/sec) for the Outfall and Inkster USGS gauge in August 2015.

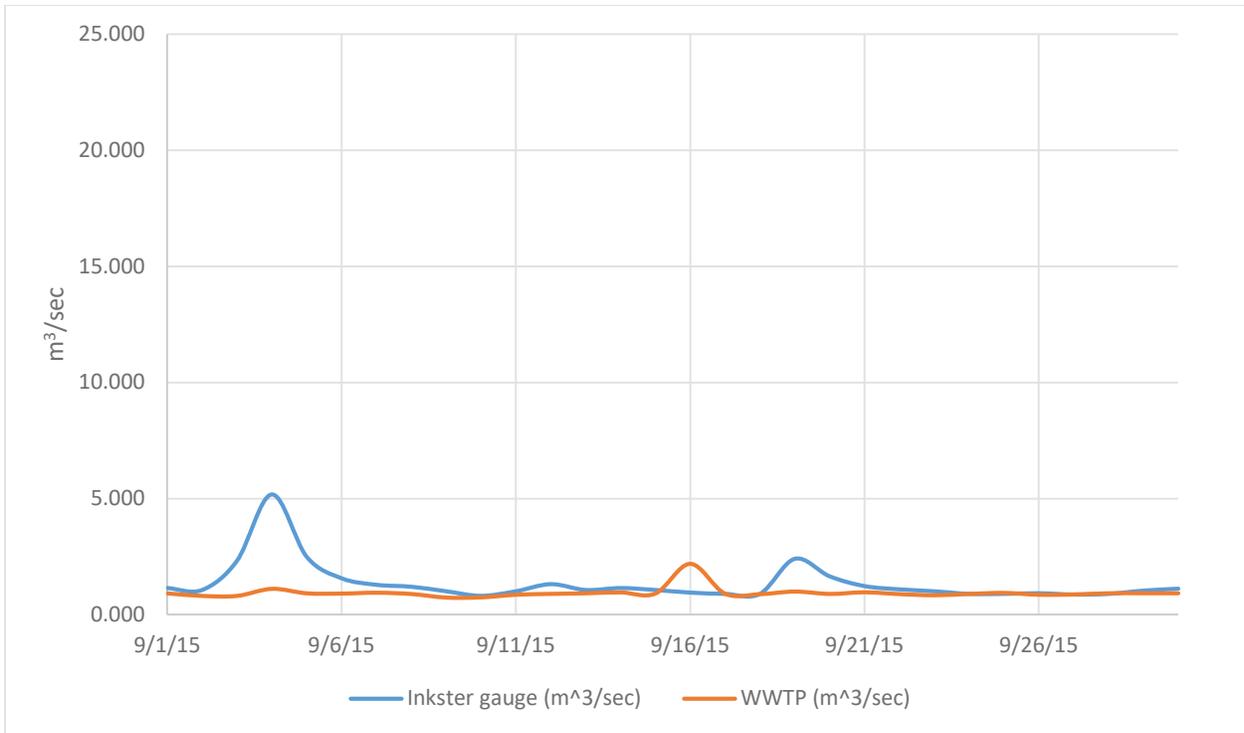


Figure 38. Discharge rate in cubic meter per second (m³/sec) for the Outfall and Inkster USGS gauge in September 2015.

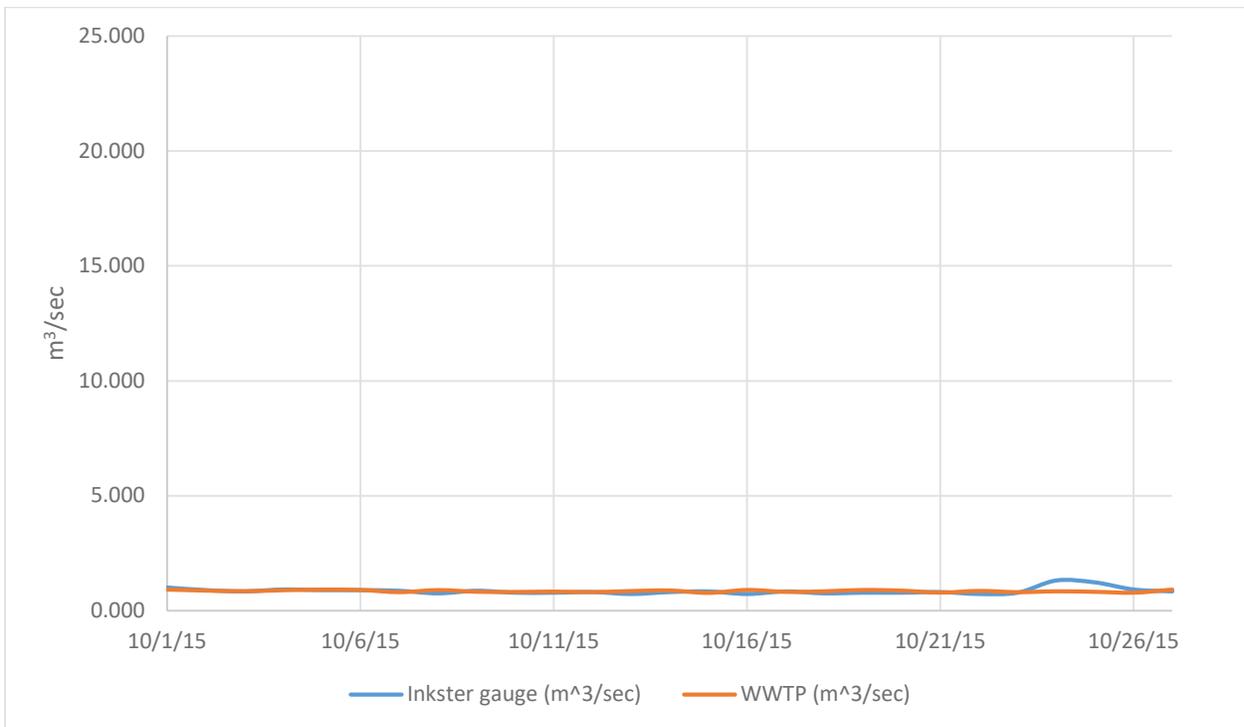


Figure 39. Discharge rate in cubic meter per second (m³/sec) for the Outfall and Inkster USGS gauge in October 2015.

APPENDIX 6

Past data of macroinvertebrate families for each location from 2012 to 2015 for spring and fall

Table 32. Past spring benthic macroinvertebrate data for Fowl2 2012 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES (CLASS)	DATE	DATE	DATE	DATE
FOWL2			4/18/2015	4/12/2014	4/27/2013	4/24/2010
Caddisfly larvae	Trichoptera	Limnephilidae	-	R	-	R
Caddisfly larvae	Trichoptera	Uenoidae	-	-	-	
Stonefly nymphs	Plecoptera	Nemouridae	-	-	-	R
Mayfly nymphs	Ephemeroptera	Heptageniidae (<i>Stenacron</i>)	C	R	R	C
Beetle	Coleoptera	Haliplidae (<i>Peltodytes</i>)	R	-	R	
Beetle larvae	Coleoptera	Dytiscidae	-	-	-	R
Beetle adults	Coleoptera	Elmidae	-	-	-	
Black fly larvae	Diptera	Simuliidae	C	-	C	C
Crayfish	Decapoda		R	-	R	-
Damselfly	Zygoptera	Coenagrionidae	-	C	-	-
Damselfly	Zygoptera	Calopterygidae	-		-	C
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	-	-	R	R
Scuds	Amphipoda	Gammaridae (<i>Gammarus</i>)	R	R	R	C
Sowbugs	Isopoda		R	C	R	R
Aquatic worms	Annelida	<i>Oligochaeta</i>	R	C	R	R
Midge larvae	Diptera	Chironomidae	C	C	C	C
Pouch snails	Gastropoda	Physidae	-	-	-	R
True bugs	Hemiptera	Veliidae (<i>Microvelia</i>)	R	-	-	-
True bugs	Hemiptera	Corixidae	-	-	R	-
True bugs	Hemiptera	Gerridae	-	-		R
Other true flies	Diptera	Culicidae	-	-	R	-
Other true flies	Diptera	Ceratopogonidae	R	-	-	-
FAMILY RICHNESS			10	8	12	16

Table 33. Past spring benthic macroinvertebrate data for the Outfall 2014 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES (CLASS)	DATE	DATE
LR-2			4/17/2015	4/11/2014
Caddisfly larvae	Trichoptera	Limnephilidae	R	-
Mayfly nymphs	Ephemeroptera	Heptageniidae (<i>Stenacron</i>)	R	-
Mayfly nymphs	Ephemeroptera	Leptophlebiidae		-
Beetle adults	Coleoptera	Elmidae	R	-
Black fly larvae	Diptera	Simuliidae	C	C
Clams			-	R
Crane fly larvae	Diptera	Tipulidae	R	-
Crayfish			-	R
Damselfly nymphs	Zygoptera	Coenagrionidae	C	R
Damselfly nymphs	Zygoptera	Calopterygidae	R	-
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	C	C
Scuds	Amphipoda		C	R
Sowbugs	Isopoda		R	C
Aquatic Worms	Annelida	<i>Oligochaeta</i>	R	R
Leeches	Annelida	<i>Hirudinea</i>	R	-
Midge larvae	Diptera	Chironomidae	C	C
FAMILY RICHNESS			14	9

Table 34. Past spring benthic macroinvertebrate data for LR-12 2012 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES (CLASS)	DATE	DATE	DATE	DATE
LR-12			4/17/2015	4/11/2014	5/9/2013	4/25/2012
Caddisfly larvae	Trichoptera	Leptoceridae	-	-	-	R
Mayfly nymphs	Ephemeroptera	Baetidae	R	-	R	R
Mayfly nymphs	Ephemeroptera	Heptageniidae (<i>Stenacron</i>)	R	R	-	
Beetle adults	Coleoptera	Elmidae	R	R	R	R
Black fly larvae	Diptera	Simuliidae	C	C	C	C
Clams	Pelecypoda	Unionidae	-	-	R	-
Clams	Pelecypoda	Sphaeriidae	R	C		R
Crane fly larvae	Diptera	Tipulidae	R	R	-	-
Crayfish	Decapoda		R	R	R	C
Damselfly nymphs	Zygoptera	Coenagrionidae	-	-	R	C
Damselfly nymphs	Zygoptera	Calopterygidae	-	R		
Dragonfly nymphs	Odonata	Aeshnidae	R	-	-	R
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	C	C	C	C
Scuds	Amphipoda		R	-	C	R
Sowbugs	Isopoda		R	C	C	-
Aquatic worms	Annelida	<i>Oligochaeta</i>	R	R	R	R
Leeches	Annelida	<i>Hirudinea</i>	-	R	R	-
Midge larvae	Diptera	Chironomidae	C	C	C	C
Limpets	Gastropoda	Ancylidae	R	-	-	-
Pouch snails	Gastropoda	Physidae	-	-	R	-
	Gastropoda	Planorbidae	-	-		-
True bugs		Belostomatidae	-	R	-	-
FAMILY RICHNESS			14	13	16	14

Table 35. Past spring benthic macroinvertebrate data for LR-6 2012 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES (CLASS)	DATE	DATE	DATE	DATE
LR-6			4/17/2015	4/14/2014	5/1/2013	4/25/2012
Mayfly nymphs	Ephemeroptera	Heptageniidae (<i>Stenacron</i>)	-	R	R	R
Mayfly nymphs	Ephemeroptera	Baetidae	R	-	-	-
Beetle adults	Coleoptera	Elmidae	-	R	-	-
Beetle larvae	Coleoptera	Elmidae	-	-	-	R
Beetle adults	Coleoptera	Haliplidae	R	-	-	-
Clams	Pelecypoda	Sphaeriidae	R	-	R	-
Crane fly larvae	Diptera	Tipulidae	R	-	-	-
Crayfish	Decapoda		R	R	R	R
Damselfly nymphs	Zygoptera	Coenagrionidae	R	C	R	R
Damselfly nymphs	Zygoptera	Calopterygidae	R		-	
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	C	R	C	R
Scuds	Amphipoda		-	-	-	R
Sowbugs	Isopoda		R	R	R	R
Aquatic worms	Annelida	<i>Oligochaeta</i>	R	C	C	C
Leeches	Annelida	<i>Hirudinea</i>	R	-	-	-
Midge larvae	Diptera	Chironomidae	C	C	C	C
Pouch snail	Gastropoda	Physidae	R	-	-	R
Limpets	Gastropoda	Ancylidae	R	-	R	
True bugs	Hemiptera	Gerridae	R	-	-	-
FAMILY RICHNESS			15	9	9	12

Table 36. Past spring benthic macroinvertebrate data for LR-10 2012 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES (CLASS)	DATE	DATE	DATE	DATE
LR-10			4/20/2015	4/22/2014	5/9/2013	4/25/2012
Mayfly nymphs	Ephemeroptera	Heptageniidae (<i>Stenacron</i>)	-	-	R	-
Beetle adults	Coleoptera	Haliplidae	-	-	R	-
Black fly larvae	Diptera	Simuliidae	C	R	-	C
Clams	Pelecypoda	Sphaeriidae	-	R	-	R
Crane fly larvae	Diptera	Tipulidae	R	R	-	R
Crayfish	Decapoda		R	C	R	R
Damselfly nymphs	Zygoptera	Calopterygidae	C	C	R	C
Damselfly nymphs	Zygoptera	Coenagrionidae	-			
Dragonfly nymphs	Odonata	Aeshnidae	R	-	-	R
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	C	C	C	C
Scuds	Amphipoda		R	R	R	R
Sowbugs	Isopoda		R	-	R	-
Aquatic worms	Annelida	<i>Oligochaeta</i>	C	R	R	R
Leeches	Annelida	<i>Hirudinea</i>	R	R	-	-
Midge larvae	Diptera	Chironomidae	C	C	C	C
Limpets	Gastropoda	Ancylidae	-	-	R	R
FAMILY RICHNESS			11	11	11	12

Table 37. Past fall benthic macroinvertebrate data for Fowl2 2012 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES/CLASS	DATE	DATE	DATE	DATE
FOWL2			10/17/2015	10/25/2014	10/19/2013	10/20/2012
Mayfly nymphs	Ephemeroptera	Heptageniidae (<i>Stenacron</i>)	C	C	R	R
Beetle adults	Coleoptera	Elmidae	-	R	-	R
Clams	Pelecypoda	Sphaeriidae	-	-	R	R
Crayfish	Decapoda		R	C	R	R
Damselfly nymphs	Zygoptera	Coenagrionidae	-	-	-	R
Damselfly nymphs	Zygoptera	Calopterygidae	-	R	R	
Dragonfly nymphs	Odonata	Aeshnidae	R	-	-	-
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae	-	R	R	-
Scuds	Amphipoda		R	C	R	R
Sowbugs	Isopoda		C	R	R	C
Aquatic worms	Annelida	<i>Oligochaeta</i>	R	R	-	R
Midge larvae	Diptera	Chironomidae	C	C	C	C
Pouch Snail	Hygrophila	Physidae (<i>Gastropoda</i>)	R	-	-	R
True bugs	Hemiptera	Veliidae	-	-	-	R
True bugs	Hemiptera	Corixidae	R	-	-	
FAMILY RICHNESS			9	9	8	13

Table 38. Past fall benthic macroinvertebrate data for LR-2 2012 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES/CLASS	DATE	DATE	DATE	DATE
LR-2			10/13/2015	10/23/2014	10/11/2013	10/11/2012
Mayfly nymphs	Ephemeroptera	Heptageniidae (<i>Stenacron</i>)	-	-	R	-
Beetle adults	Coleoptera	Elmidae	R	-	-	R
Beetle adults	Coleoptera	Hydrophilidae	-	R	-	-
Black fly larvae	Diptera	Simuliidae	R	R	-	R
Crane fly larvae	Diptera	Tipulidae	R	R	-	R
Crayfish	Decapoda		R	R	-	R
Damselfly nymphs	Zygoptera	Calopterygidae	R	R	R	-
Damselfly nymphs	Zygoptera	Coenagrionidae	-	-		R
Dragonfly nymphs	Odonata	Aeshnidae	-	R	R	-
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	C	C	-	C
Scuds	Amphipoda		C	R	R	R
Sowbugs	Isopoda		R	R	R	R
Aquatic Worms	Annelida	<i>Oligochaeta</i>	R	R	R	-
Leeches	Annelida	<i>Hirudinea</i>	R	-	R	-
Midge larvae	Diptera	Chironomidae	C	R	C	C
Pouch snails	Gastropoda	Physidae	-	-	R	-
Limpets	Gastropoda	Ancylidae	-	R	-	R
True bugs	Hemiptera	Veliidae	-	-	-	R
FAMILY RICHNESS			11	12	10	11

Table 39. Past fall benthic macroinvertebrate data for LR-12 2012 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES/CLASS	DATE	DATE	DATE	DATE
LR-12			10/13/2015	10/23/2014	10/15/2013	10/12/2012
Mayfly nymphs	Ephemeroptera	Heptageniidae (<i>Stenacron</i>)	-	-	C	C
Mayfly nymphs	Ephemeroptera	Baetidae	C	C		
Beetle adults	Coleoptera	Elmidae	R	R	C	R
Beetle adults	Coleoptera	Hydrophilidae	-	-		
Beetle adults	Coleoptera	Dytiscidae	-	-		
Beetle larvae	Coleoptera	Elmidae	-	-	-	C
Black fly larvae	Diptera	Simuliidae	C	C	C	C
Clams	Pelecypoda	Sphaeriidae	-	-	C	R
Crane fly larvae	Diptera	Tipulidae	R	R	R	C
Crayfish	Decapoda		R	R	R	R
Damselfly nymphs	Zygoptera	Calopterygidae	R	R	R	R
Dragonfly nymphs	Odonata	Aeshnidae	R	-	R	-
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	C	C	C	C
Scuds	Amphipoda		-	R	R	-
Sowbugs	Isopoda		-	-	-	R
Aquatic worms	Annelida	<i>Oligochaeta</i>	-	R	R	R
Flatworm		<i>Turbellaria</i>	R		-	
Leeches	Annelida	<i>Hirudinea</i>	R	R	R	-
Midge larvae	Diptera	Chironomidae	R	C	R	C
Pouch snails	Gastropoda	Physidae			R	R
Limpets	Gastropoda	Ancylidae	R	-		
True bugs	Hemiptera	Nepidae	-	-	-	R
True bugs	Hemiptera	Veliidae	-	-	R	
True bugs	Hemiptera	Belostomatidae	-	-	-	-
True flies	Diptera	Ceratopogonidae	-	-	R	-
FAMILY RICHNESS			12	12		18

Table 40. Past fall benthic macroinvertebrate data for LR-6 2012 to 2015 from FOTR. Fall 2014 was not sampled. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES/CLASS	DATE	DATE	DATE	DATE
LR-6			10/20/2015		10/11/2013	10/12/2012
Mayfly nymphs	Ephemeroptera	Baetidae	R		C	-
Mayfly nymphs	Ephemeroptera	Heptageniidae (Stenacron)			-	R
Beetle adults	Coleoptera	Elmidae	R		-	-
Crane fly larvae	Diptera	Tipulidae	-		R	-
Crayfish	Decapoda		R		R	C
Damselfly nymphs	Zygoptera	Coenagrionidae	-		R	C
Damselfly nymphs	Zygoptera	Calopterygidae	R		-	-
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	C		C	C
Sowbugs	Isopoda		R		R	R
Aquatic worms	Annelida	<i>Oligochaeta</i>	R		R	R
Leeches	Annelida	<i>Hirudinea</i>	-		R	-
Midge larvae	Diptera	Chironomidae	R		R	C
Limpets	Gastropoda	Ancylidae	R		C	C
True bugs	Hemiptera	Veliidae (<i>Microvelia</i>)	R		-	-
FAMILY RICHNESS			10		10	8

Table 41. Past fall benthic macroinvertebrate data for LR-10 2012 to 2015 from FOTR. R means rare (1-10) and C means common (>10).

COMMON NAME	ORDER	FAMILIES/CLASS	DATE	DATE	DATE	DATE
LR-10			4/20/2015	10/23/2014	10/11/2013	10/12/2012
Mayfly nymphs	Ephemeroptera	Baetidae	C	R	C	C
Mayfly nymphs	Ephemeroptera	Heptageniidae (Stenacron)		-	-	-
Beetle adults	Coleoptera	Elmidae	R	-	-	-
Clams	Pelecypoda	Sphaeriidae	R	-	R	R
Crane fly larvae	Diptera	Tipulidae	R	-	-	-
Crayfish	Decapoda		R	R	R	C
Damselfly nymphs	Zygoptera	Coenagrionidae	R	-	-	R
Damselfly nymphs	Zygoptera	Calopterygidae	R	R	R	-
Net-spinning caddisfly larvae	Trichoptera	Hydropsychidae (<i>Cheumatopsyche</i>)	C	R	C	C
Scuds	Amphipoda		R	R	-	-
Sowbugs	Isopoda		C	R	-	-
Aquatic worms	Annelida	<i>Oligochaeta</i>	R	R	-	R
Leeches	Annelida	<i>Hirudinea</i>	-	-	-	R
Midge larvae	Diptera	Chironomidae	R	C	R	C
Limpets	Gastropoda	Ancylidae	-	R	-	R
True bugs	Hemiptera	Belostomatidae (<i>Belastoma</i>)	R	-	-	-
FAMILY RICHNESS			14	9	6	9

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