Annapolis River 2021 Annual Water Quality Monitoring Report

Clean Annapolis River Project

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Annapolis River 2021 Annual Water Quality Monitoring Report

Prepared By:

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expanding on the 2014 report by Rajwinder Singh Toor and Lindsey Freeman

Clean Annapolis River Project

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List of Acronyms

ACER	Acadia Centre for Estuarine Research
CARP	Clean Annapolis River Project
CCME	Canadian Council of Ministers for the Environment
CFU	Colony-Forming Units
DO	Dissolved Oxygen
DOSAT	Saturated Dissolved Oxygen
EC	Environment Canada
FNU	Formazin Nephelometric Unit
NO3-N	Nitrate-Nitrogen
NTU	Nephelometric Turbidity Units
OMEE	Ontario Ministry of Environment and Energy
P	Phosphorus
pH	Power of Hydrogen
QA/QC	Quality Assurance/Quality Control
TSS	Total Suspended Solids

Acknowledgements

The River Guardians water quality monitoring program would not have been possible without the dedication of community members throughout the past 30 years.

The success of the River Guardians program is in part due to its approach of bringing together a variety of stakeholders who have an interest in the health of the Annapolis River. We would like to thank the following partners who have worked with us to deliver the Annapolis River Guardians program:

Environment Canada – Atlantic Ecosystems Initiative

Nova Scotia Environment

Saint Mary's University Coop Program

Atlantic Water Network

The Acadia Centre for Estuarine Research

Executive Summary

In 2021, the Annapolis River Guardians completed their 30th year of continuous water quality monitoring on the Annapolis River. CARP staff monitored eight sites over the course of the season, which ran from May to November. Parameters monitored in the 2021 season include dissolved oxygen, *E. coli* bacteria, air and water temperature, pH, conductivity, and turbidity, as well as local weather conditions. In 2008, total suspended solids (TSS) and turbidity were added to the suite of parameters monitored; however TSS sampling ended in 2011, and instead was derived from collected turbidity values.

E. coli bacteria levels along the Annapolis River during 2021 were, on average, higher than those observed in 2020, with 2021 medians being higher at all sites downstream of Aylesford, and only slightly lower or comparable at both the Aylesford Road and Aylesford sites. The amount of precipitation received in 2021 was greater than in 2020, which may explain the observed results. Similar to 2020, there is a large increase in median *E. coli* levels between the Aylesford Road and Aylesford sites, which is consistent with historical data and continues to suggest the presence of a contamination source somewhere between those two sampling locations. In 2021, seven of the eight sample sites recorded a greater proportion of *E. coli* values above 100 cfu/100mL, compared to 2020, and since 2019 the proportion of observations that exceed the detectable threshold has been steadily increasing.

Over the 30 years of monitoring, mean dissolved oxygen saturation (DOSAT) levels have mostly remained in a healthy range. Due to the influence of temperature on dissolved oxygen, the 2021 report includes an analysis of DO that is limited to the summer months (July 1st- September 30th). 2009 and 2014 are the only years since 2003 to drop below an average summer value of 80%, which is still well above the 60% threshold at which aquatic life becomes stressed. In 2021, the mean summer DOSAT level was 87.29%, which is very close to the historical summer average of 87.32%, and the previous year's summer average of 91.27%. In 2021, mean summer dissolved oxygen (DO) concentrations (mg/L) for all sites remained in the range of 7.47 to 9.49 mg/L. The overall mean DO concentration was 8.27 mg/L, similar to 8.23 mg/L in 2020; both these values are lower than their respective full-season averages. The maximum recorded DO and DOSAT levels in 2021 were 16.13 mg/L and 164%, at Lawrencetown and Middleton, respectively.

The mean summer water temperature for the Annapolis River during 2021 was 17.43°C, which is 1.3°C and 0.06°C colder than the same periods in each 2020 and 2019. All eight sample sites recorded a summer average below the 20°C threshold at which temperature aquatic life becomes stressed. However, as in previous years, individual water temperature observations during the 2021 summer months continued to reach and exceed this threshold. The maximum value recorded in

2021 was 23.3°C in Bridgetown, and a total of 16 observations were above 20°C. However this is likely a conservative estimate; contrary to previous years, the 2021 sampling took place in the morning (often before 10am), likely missing the highest water temperatures which are often observed mid-day.

The pH levels at each of the River Guardian sites fell mostly within the recommended range for the protection of aquatic life (6.5-9.0). Ten values fell below the lower 6.5 pH threshold in 2021, with a low of 5.82 recorded at Bridgetown. However only one of these ten values was during the summer period (July 1st to September 30th). The maximum observed pH value was 8.32, recorded at Paradise on August 3rd. Mean summer pH values for the eight monitoring locations along the Annapolis River were all comparable or lower than the averages reported in 2020.

Nitrogen and phosphorus levels were initially measured at Lawrencetown and Wilmot beginning in 2006, and Millville was added as a reference site in 2008. Lawrencetown sampling ceased in 2009. There is much controversy over the level at which nitrogen becomes harmful to aquatic life. For reporting needs, 0.9 mg/L of total nitrogen (Dodds and Welch, 2000) is used as the maximum concentration for preserving aquatic health (CCME, 2003). While elevated total nitrogen results were observed, phosphorus remains a significant concern. During the 2006 to 2020 period, 17% of total nitrogen results exceeded 0.9 mg/L while 76% of total phosphorus results exceeded the suggested guideline level of 0.030 mg/L (OMEE, 1994). These elevated phosphorus concentrations are believed to have a role in excessive periphyton growth along the main stem of the river and depression of dissolved oxygen levels in the tidal portion of the river. It is difficult however to draw any short-term trends because of the small sample size in 2020 (3 observations at Wilmot, and 2 at the Millville reference site), all of which were collected during November, January, and February.

Working in conjunction with Environment Canada, turbidity and total suspended solids (TSS) samples were collected in 2008 and 2009 as part of the regular bi-weekly sample collection and during high flow precipitation events. In 2010 and 2011, samples were only collected after precipitation events of 15 mm or greater in order to assess peak sediment levels in the water column at Bayard Road in Wilmot, Middleton and Paradise. TSS sampling was not continued after 2011, but TSS values were estimated from regular turbidity sample collection, based on a relationship developed from past TSS and turbidity sampling efforts. The maximum observed turbidity value in 2021 was 33.4 FNU, recorded at Kingston. All eight sample sites monitored in 2021 appear have higher turbidity and TSS values than 2020. Turbidity event sampling is no longer part of the standard data collection protocol, and thus peak turbidity levels may not be reflected in the collected data.

1.0 Introduction

1.1 History

The Annapolis River Guardian volunteers began collecting water quality data in the Annapolis River watershed in 1992. The Clean Annapolis River Project (CARP) initiated the program as a public awareness project, and has had numerous volunteers collecting samples over the years. It was one of the longest running and most extensive volunteer-based water quality programs in Eastern Canada. It is also CARP's longest running project. At least 100 volunteers from the Annapolis Valley community have participated in the program over the years, and over 4,700 water samples have been collected and analyzed.

The program was initiated in the early 1990's by Dr. Graham Daborn and Dr. Mike Brylinsky of the Acadia Centre for Estuarine Research (ACER). Many groups were involved in the planning process for the program, including staff from the Nova Scotia Department of Health, the Nova Scotia Department of Environment, Nova Scotia Community College, and CARP. Some modifications have been made over the years, but the core has remained the same.

Originally, the design called for 11 sites to be monitored by 17 volunteers. However, the program was so well received by the community that it was significantly expanded between 1992 and 1994. In 1994, 38 sites were monitored by 43 River Guardians from 36 households (Pittman et al. 2001). This intensity of monitoring placed considerable strain on the capacity of CARP. While some of the initial enthusiasm surrounding the program has subsided, a core group of 8 to 15 dedicated volunteers was maintained until 2014. Due to funding limitations and the costs of running a volunteer-based program, the River Guardians water quality data has been collected by CARP staff since 2015, at eight consistent sample sites along the freshwater portion of the Annapolis River.

1.2 Program Objectives

The Annapolis River Guardians program has four objectives:

- To establish and support a regular observation system that provides an early warning of environmental problems.
- To provide a long-term record of the river's health, made available to public and community audiences.
- To develop interest in the Annapolis River and ensure a viable resource for future generations.

1.3 Overview of 2014 Monitoring Season

Sample collection for the 2021 season ran from May 11th to November 3rd, on a biweekly basis. The parameters monitored were *E. coli* bacteria, dissolved oxygen content, water temperature, air temperature, pH, conductivity, and turbidity. Total suspended solids (TSS) and turbidity event sampling was initiated in 2008, but has not been performed since the 2011 monitoring season. Sampling of these parameters was part of a joint project between CARP and Environment Canada, in order to determine baseline levels in the Annapolis River and to establish a mathematical relationship between the two variables. TSS was estimated from the regular turbidity samples this year, using the line of best fit equation developed in 2014. Bacteria count, DO and temperature data have been collected since the inception of the River Guardians program in 1992, pH has been collected since 2003 and nutrients have been monitored by Environment Canada since 2006. CARP has ceased sampling of benthic invertebrates since the last formal River Guardians report was produced in 2014.

Eight stations were sampled along the Annapolis River. Further information on these sampling locations is contained in Appendix B. The monitoring sites for 2021 were all within the freshwater portion of the Annapolis River (Figure 1). The data collected is stored in a Microsoft Access database at the CARP office.

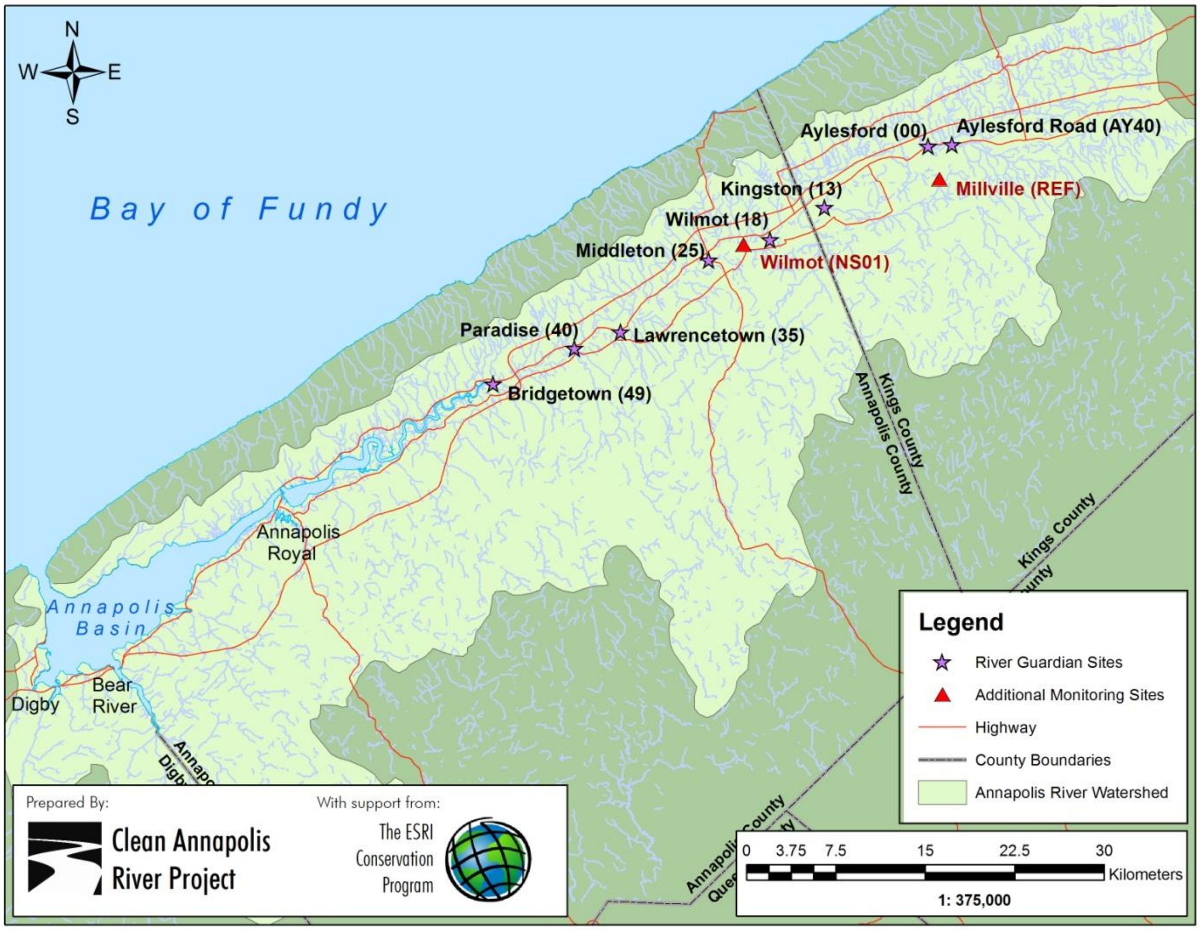

Figure 1. Annapolis River watershed with 2021 River Guardian monitoring sites identified by stars.

The 2021 River Guardian sampling locations (with their identification numbers) were:

49 – Bridgetown	40 – Paradise	35 –Lawrencetown	25 – Middleton
18 – Wilmot	13 – Kingston	00 – Victoria Road, Aylesford	AY40 – Aylesford Road, Aylesford

All sample sites are located on the main stem of the Annapolis River. With the exception of Aylesford Road (Site AY40, hereafter identified as site 60). In the past, each location had a large River Guardians sign (Figure 2), that indicates *E. coli* contamination and overall water quality trends for that location. Historically, the signs have been displayed from May through to November, and were updated by CARP staff every two weeks.

In addition to the regular River Guardians sites, site NS01 (Bayard Road in Wilmot) and REF (South Annapolis River at Millville) are shown in Figure 1. The River Guardians did not monitor these sites, but they were used for the monitoring of nutrients by Environment Canada, as well as for past TSS/Turbidity sampling by CARP.

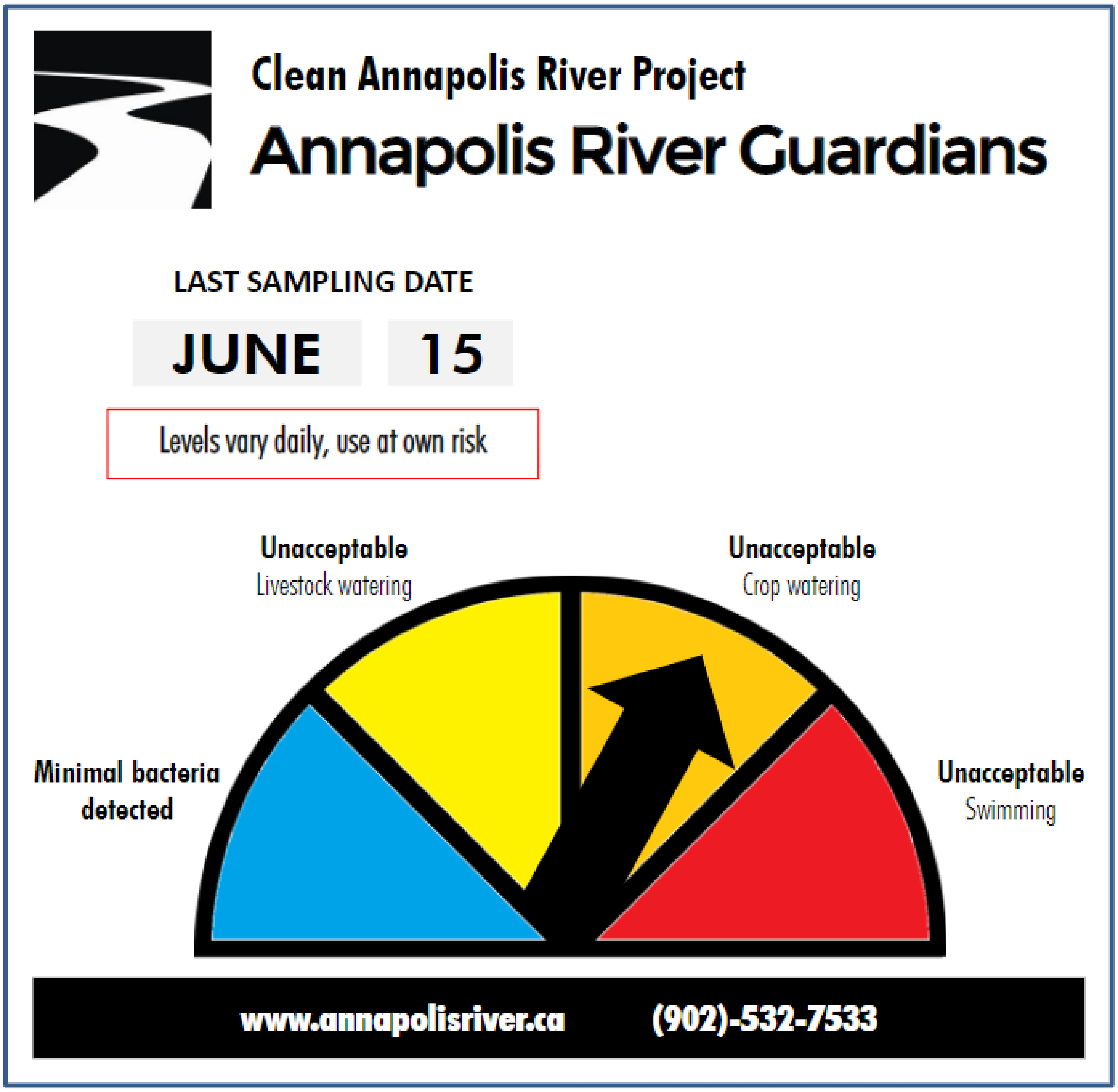

Figure 2. New River Guardian sign to be displayed in 2022. These signs will be stationed at each of the eight sampling locations, and will show the date and E. coli bacterial levels.

2.0 2014 Monitoring Results

2.1 *E. coli* Bacteria

2.1.1 Introduction

Escherichia coli (*E. coli*) are rod-shaped, aerobic, lactose fermenting bacteria that are present in the wastes of humans, animals, and even some fish (Valiela et al., 1991). The predominant sources of *E. coli* bacteria in a watershed include poorly maintained on-site septic systems, malfunctioning central sewage treatment plants, aquatic wildlife, domestic animals, and livestock. Because they occupy the same ecological niche as many human pathogens, such as Cryptosporidium, *E. coli* are used as indicators for the possible presence of other potentially dangerous pathogens. *E. coli* levels have been identified in the past as a major cause of concern in the Annapolis River watershed (Pittman et al., 2001).

Many factors in a particular ecosystem affect the abundance and persistence of *E. coli* in rivers. These include the type of contributing source, the transport mechanism with which the *E. coli* is deposited, and precipitation. The result is that *E. coli* densities in surface waters can be highly variable. Their survival in surface waters is not well understood, and is dependent on many factors. These include predation by other organisms, the amount and intensity of sunlight reaching the water surface, pH, salinity of the water, temperature, as well as composition and abundance of sediment (Wcisto and Chróst, 2000; Davies et al., 1995). The persistence of *E. coli* in river systems is also largely dependent upon the composition and type of media in which they are found. For example, there are a range of estimates for the survival times of the commonly monitored *E. coli* in various media:

Cow pats: 49 days at 37°C, 70 days at 5°C (also dependent on moisture content) (Chalmers et al., 2000)
Drinking water: Between 28 and 84 days (Edberg et al., 2000)
Soil cores with grass roots: 130 days (Chalmers et al., 2000)
Freshwater sediment: 57 days (Davies et al., 1995)

While the core River Guardian monitoring program has been maintained over the period of 1992 to 2021, a number of modifications have been made. For example, in 1996, the collection of *E. coli* samples was standardized to every two weeks. In the period from 1997 to 2002, fecal coliform numbers were determined using the IDEXX Colilert procedure, which specifically identifies *E. coli*. With the change to a new laboratory, the 2003 and 2004 samples were analyzed using the Membrane Filtration procedure, which enumerates fecal coliforms (see Appendix A). In 2005, the Science Advisory Committee for the Annapolis River Guardians advised that bacteria monitoring be switched from fecal coliforms to *E. coli*, to bring the program more in line with current guidance at a national level. To ensure the continuity of the historic dataset, it was decided to collect split samples for the first two months of the season, to allow parallel testing for fecal coliform and *E. coli*. This process confirmed that the two methods do not give statistically different results. Further information on the parallel testing and statistical analysis can be found in the 2005 Annual Report for the Annapolis River Guardians (Beveridge et al., 2006).

The sampling procedure for *E. coli* collection can be found in Appendix A.

2.1.2 Canadian Water Quality Guidelines

Various government agencies have developed water quality guidelines to protect the safety of the general public. Health Canada is responsible for the guidelines for drinking and recreational waters. The Canadian Council of Ministers of the Environment (CCME) has incorporated these guidelines in the comprehensive Canadian Water Quality Guidelines (CCME, 2002). There have been several different guidelines developed for different possible water uses, such as protection of aquatic life, agricultural uses, drinking or recreation. CARP has summarized some of these guidelines for *E. coli* bacteria contamination into a single table for public awareness purposes (Table 1).

Table 1. Summary of water quality guidelines and categories for E. coli.

cfu*/100ml	Water Use	Explanation/Source
0	Acceptable for drinking	*E. coli*/100ml. (Health Canada, 2010)
1-50	Acceptable for livestock watering	Interpretation of CCME narrative "high-quality water given to livestock" (cfu/100mL).
50-100	Acceptable for food crop irrigation	Tentative Maximum Concentration. CCME Guidelines (cfu/100mL).
100-200	Acceptable for recreational use	Interim category.
>200	Unacceptable for human recreational contact**	Geometric Mean of 5 samples taken during a period not to exceed 30 days, should not exceed 200 cfu/100 mL (Health Canada, 1992).
>400	Unacceptable for human recreational contact	Single sample maximum concentration taken in a given period should not exceed 400 cfu/100 mL (Health Canada, 2012).
*cfu = colony forming units		

**These guidelines refer to primary body contact recreational activities, such as swimming, etc. For more information about the Health Canada guidelines for human recreational contact, please refer to http://www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/guide_water-2012-guide_eau/index-eng.php#p2

2.1.3 Monitoring Results

The high variability of fecal bacteria measurements presents a number of challenges with respect to data analysis. Samples collected from a single site, on separate occasions, can vary by two and sometimes three orders of magnitude (e.g. 3 cfu/100 ml to 3000 cfu/100 ml). The use of standard data analysis methods, such as calculating and comparing mean values, inadequately describes the distribution of fecal bacteria results. The following analysis is therefore based on the proportion of samples analyzed that exceed particular water quality thresholds. This approach was chosen as it best presents to decision-makers and resource managers whether the water at a site is unsuitable for particular uses.

While this approach eliminates the bias of calculating means with highly variable data, it presents another type of bias. If the majority of samples one year fall slightly below a guideline threshold (e.g. 200 cfu/100 ml), a small increase in *E. coli* concentration the next year may cause the proportion of samples above 200 cfu/100 ml to increase significantly. This would give the appearance that the water quality had worsened considerably, when in fact the mean concentration may have only increased slightly. In order to ensure the differences observed in the following analysis are real, a box-whisker plot was prepared to compare the distribution of the 2020 and 2021 *E. coli* results (Figure 3). The box plot shows the 25th and 75th percentiles as well as the median for each site. Note that the y-axis of the graph is plotted using a logarithmic scale and that the data is artificially capped at 2419 cfu/100mL, as this is the maximum possible value with the IDEXX Colilert testing system. From 1992 to 2021, approximately 0.9% of the data have exceeded this cap value. This has increased to 1.4% in the last 10yrs, and 3.8% in the 2021 season (4 out of 104 samples).

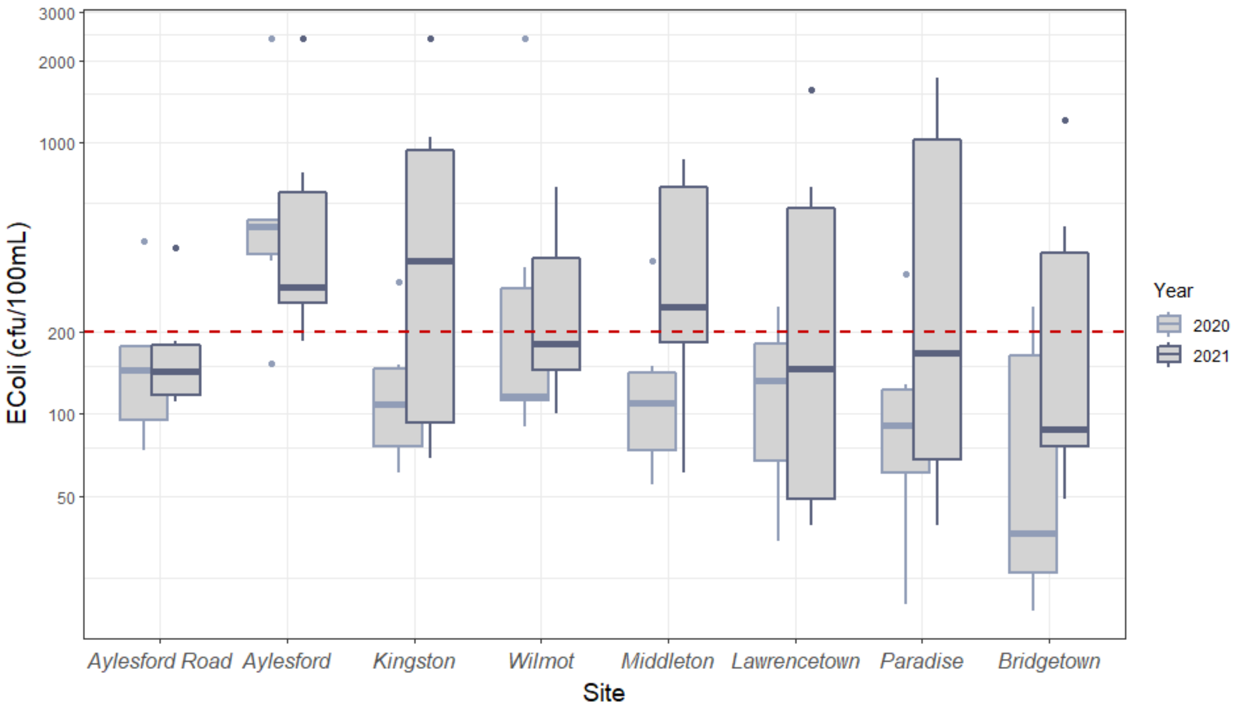

Figure 3. Box and whisker plots of Annapolis River Guardian E. coli bacteria results for 2020 and 2021. To account for differences in sampling period (The 2021 sampling season extended earlier into the Spring and later into the fall), results are compared between the same period from July1st-September 30th (considered the "summer" sampling period for our temperature analysis). The red dashed line indicates the CCME threshold of 200cfu/100mL deemed unacceptable for human recreational contact.

In 2021, the median *E. coli* values for the monitoring sites were higher than in 2020 for all sites, except Aylesford and Aylesford Road, which are the two most upstream locations (Figure 3). Aylesford, Kingston, Middleton, Lawrencetown and Paradise showed greater variability this season, while Aylesford Road, Wilmot, and Bridgetown showed comparable or lower variability to 2020. Contamination continues to be greatest in upstream river sites.

The *E. coli* data for each River Guardian location was calculated as the percentage of samples that fell within each of the ranges specified in Table 1. This allows easy visualization of how the *E. coli* readings have fluctuated over time. All of the *E. coli* ranges are in units of cfu/100mL.

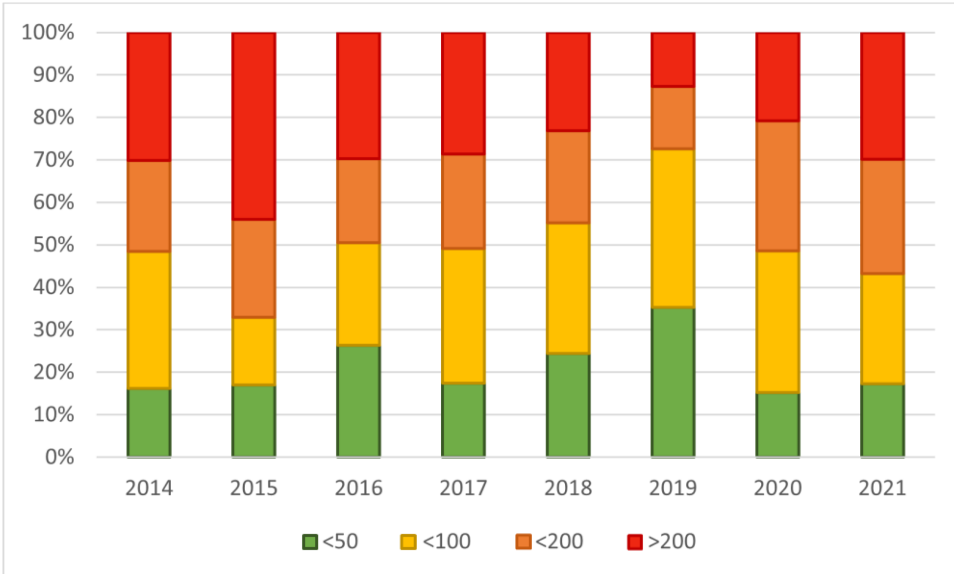

Figure 4. Percentages of E. coli samples (cfu/100mL) that fall into each water quality category by year, since 2014 (the last year that a formal River Guardians report was produced.

The proportion of samples that fell into the range >200 cfu/100 mL decreased between 2015 and 2019; since 2019, this proportion seems to be increasing. Similarly, 2019 also presented the greatest proportion of observations in the <50 cfu/100mL range, which has since declined (Figure 4).

The proportion of samples falling into the >200 cfu/100mL, and the <50 cfu/100mL categories increased in 2021 when compared to 2020, while the 51-100 cfu/100mL, and 101-200 cfu/100mL categories decreased. The distribution for each site in 2020 and 2021 is shown in Figure 5. The bar charts of proportions encompass the entire sampling season, which extended further into the fall during 2021 than it did in 2020. The 2020 sampling season went from June 1st to October 19th, and the 2021 season went from May 11th to November 3rd. This extended sampling season in 2021 compared to 2020 could explain some of the differences in *E. coli* values, as almost 30% of the 2021 observations below 50cfu were observed in the month of May. Contrarily, only 1 of the 31 observations above 200cfu was in the time after October 19th, and is thus unlikely to be a determinant of high *E. coli* values. It is possible though that differences in rainfall could explain some of the observed differences in *E. coli* levels between years. There was significantly more precipitation during the sampling period in 2021 compared to 2020 (~650mm vs. ~465mm) (Data come from CFB Greenwood weather Station). Rainfall can increase the rate of residential septic system failures, and increase the load on municipal water treatment facilities, and so we would expect years with greater rainfall to show a similar rise in *E. coli* levels.

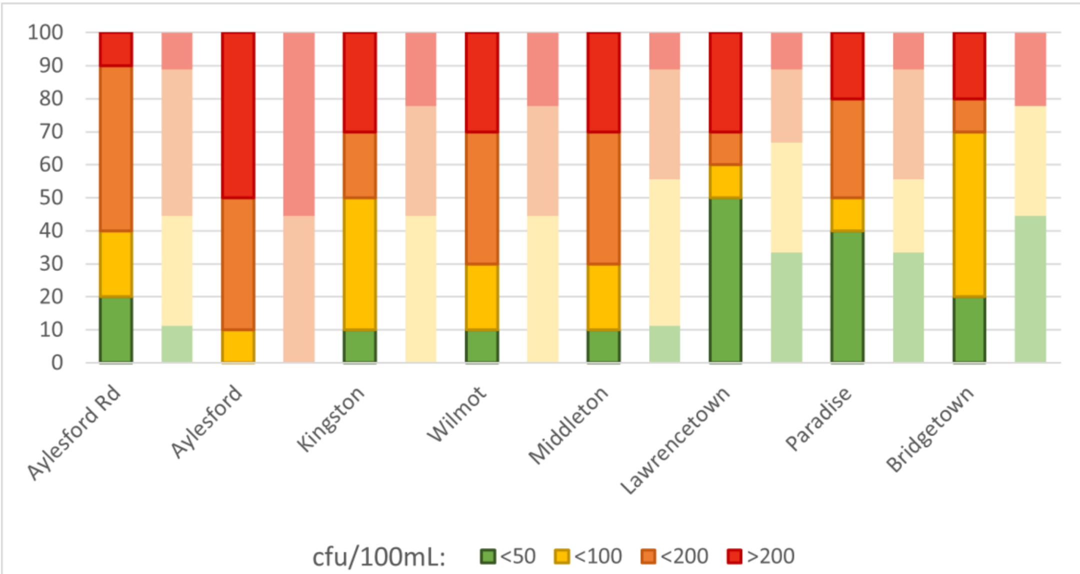

Figure 5. The percentages of 2021 (dark columns) and 2020 (light columns) E. coli samples that fall into the different cfu/100mL ranges. Data are sorted by site and organized by direction of water flow (left to right). 2021 data comes from 104 samples collected between May and October, compared to the 2020 data which constitutes only 72 samples collected between June and October.

There does not appear to be an indicative trend for *E. coli,* as the values at all sites are quite variable between years. However the greatest proportion of bacteria counts >200 cfu/100mL occur quite consistently in Aylesford on Victoria Road. In 2021, the lowest proportion of >200 cfu/100mL samples occurred at the Aylesford Road, Bridgetown, and Paradise locations (Figure 5), but this differs slightly from 2020. The lowest *E. coli* count in 2021 was 16 cfu/100mL at Paradise on June 8th, while the highest was >2419 cfu/100mL (artificially capped), recorded at several sites. There seems to be a source of contamination between the Aylesford Road and Aylesford sites, which may be coming in from one of several tributaries that join the main river between these two sites.

From 1992 to 2011, numerous initiatives were undertaken which have contributed to the improvement of water quality in the Annapolis River. For example, in the winter of 1994, 14 Wing Greenwood discontinued the discharge of untreated aircraft wash-water into a tributary of the Annapolis River. In August 1998, the base discontinued the operation of its own sewage treatment plant, redirecting its waste to the Greenwood municipal facility. In October of 2011, the Town of Middleton completed the construction of a new sewage treatment plant. Since 2013, CARP has worked with farms in the watershed to reduce the amounts of agricultural runoff entering the river.

2.1.4 *E. coli* Monitoring Recommendations

- Continue regular River Guardian *E. coli* monitoring at the eight main river sample locations.
- Collaborate with livestock owners to address the issue of restricting animals from the Annapolis River.
- Continue to investigate the potential source(s) of contamination in the watershed.
- Investigate the correlation between precipitation amounts and *E. coli* levels in the river.

2.2 Dissolved Oxygen

2.2.1 Introduction

Dissolved oxygen (DO) is a widely used and important general indicator of the health of a river system (Addy et al., 1997). Aquatic organisms require oxygen in solution for internal respiration. Oxygen in the atmosphere, which is readily available to terrestrial organisms, must be dissolved into the water and is present at much lower concentrations. Wind, wave action, rainfall, and photosynthesis help aerate waterways and increase dissolved oxygen levels. Sewage, lower rates of photosynthesis, eutrophication, and limited diffusion from the atmosphere due to ice cover can all lead to decreased oxygen levels.

As the temperature of water decreases, a greater concentration of oxygen can dissolve in the water. DO levels are also dependent to a lesser degree on atmospheric pressure and water salinity. The amount of oxygen in water can be reported in two ways, either as a concentration measurement (mg/L) or as percent saturation. Percent saturation represents the actual amount of dissolved oxygen in an amount of water compared to the maximum amount that can be dissolved. This value is given as a percentage. Water reaches its saturation point when it can no longer dissolve any additional oxygen for a given temperature. High levels of photosynthesis or turbulent conditions can "supersaturate" the water, resulting in saturation levels greater than 100%. Dissolved oxygen levels below 60% saturation are known to cause stress to aquatic life, particularly cold-water fish species (Mackie, 2004). Comparatively, CCME guidelines for concentrations of dissolved oxygen (mg/L) for the protection of freshwater warm-water species is 5.5 mg/L, while that for cold water species is 6.5 mg/L (CCME, 2002).

2.2.2 Monitoring Results

To better understand the status of dissolved oxygen levels in the Annapolis River, values for both percent saturation (DOSAT) and concentration (mg/L) were compared. Since the introduction of the Quanta sonde data collection method in 2003, there appears to be discrepancies between the volunteer and sonde probe datafiles, clearly identified by the differences in the reported annual mean values between datasets, which in theory should be the same. The 2021 analyses have been conducted using the sonde probe dataset (including data collected using both Quanta and YSI probes over the years), as that is the sampling method that is still currently in use by the River Guardians program.

In addition, due to differences in sampling period between years, and the influence of temperature on dissolved oxygen levels, DO results summarized here are based on observations recorded during the summer sampling period (July 1st - September 30th). Differences between summer averages and full-season averages can be seen in Figure 6 and Table 2, and illustrate the often higher annual DO levels when including early and late season data collected at colder water temperatures. In particular, 2014 showed a distinct discrepancy between full season and summer averages, however the sampling season that year spanned from late April to late October, which is a wider sampling period than many other years.

Since 2003, summer mean DOSAT levels have varied from a high of 103.7% in 2018, to a low of 70.6% in 2014 (Figure 6). Variation between DOSAT levels has varied minimally over the past three years, however the 2021 mean was slightly lower than the previous 2 years. In 2021, the mean summer DOSAT was 87.3%, compared with 91.3% in 2020 and 90.9% in 2019. This value is within the normal range of variability observed for the Annapolis River. Figure 7 shows the mean DOSAT and standard error of the mean, which indicate that there was similar variability in DOSAT levels in 2021 as in 2020. The maximum DOSAT value recorded in 2021 was 164% on September 20th, at Middleton, while the lowest value was 58.2% on August 3rd at Aylesford. As with the percent saturation, dissolved oxygen concentrations (mg/L) have remained similar over the past several years. The mean summer DO (mg/L) level in 2021 was 8.27 mg/L, which is similar to the recorded mean of 8.23 mg/L in 2020.

Table 2. Annual mean dissolved oxygen levels, including both mg/L and percent saturation values, collected by CARP staff using a Quanta or YSI water quality probe. Averages from the summer sampling period are compared to those calculated over the entire sampling period, which is variable between years.

	Summer (July, August, September)		**Whole season**	
Site	**DO(mg/L)**	**DO (%)**	**DO(mg/L)**	**DO (%)**
2003	7.98	86.59	9.07	87.24
2004	8.66	89.28	9.48	87.98
2005	7.81	83.85	8.55	83.85
2006	7.84	83.90	8.37	83.02

2007	8.80	92.93	9.09	89.73
2008	8.17	85.31	8.81	83.66
2009	7.73	79.69	8.66	84.70
2010	8.00	84.02	8.99	88.98
2011	8.14	85.32	9.11	88.63
2012	7.70	84.93	8.32	85.45
2013	8.40	88.74	8.82	87.74
2014	6.72	70.55	8.11	79.01
2015	7.82	86.29	8.75	89.05
2016	8.56	94.89	8.62	90.97
2017	8.62	86.49	8.69	88.69
2018	9.83	103.65	10.23	103.00
2019	8.72	90.95	9.80	95.80
2020	8.23	91.27	8.36	90.93
2021	8.27	87.29	8.84	87.74
2003-2021 Mean	**8.22**	**87.32**	**8.87**	**87.97**

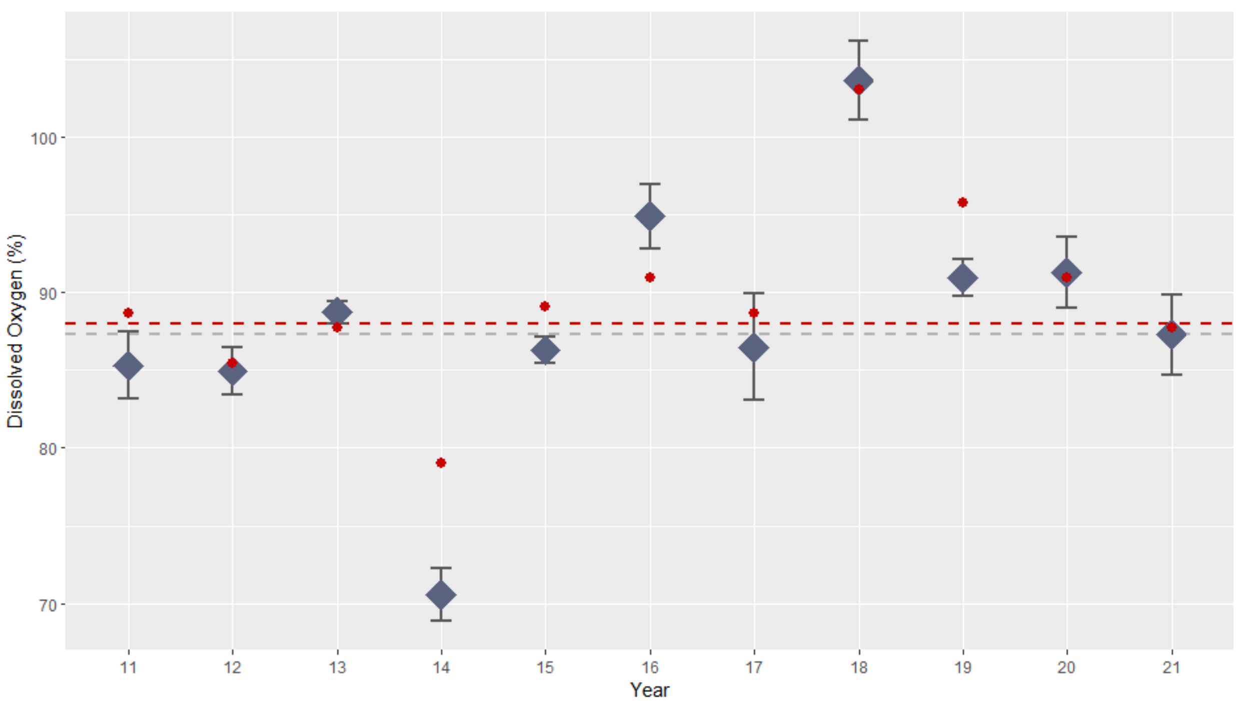

Figure 6. Annual mean DOSAT (%) by year, from 2011 to 2021. Diamond points show the annual summer average (July-September), and the red dots represent the average of the full sampling period. The grey line indicates the cumulative historic summer mean (2003-2021) of 87.32%, and the red line indicates the cumulative full-season mean of 87.97%.

Mean DOSAT (%) levels were comparable to the historic mean for Kingston, Wilmot, and Paradise, while mean DOSAT values fell marginally below historical averages at Aylesford Road, Aylesford, Lawrencetown, and Bridgetown. DOSAT average was the highest in Middleton, and also showed the highest variability at that site (Figure 7). Out of 48 summer observations, 38 readings had DO saturations greater than 75%, and throughout the entire sampling period, 87 of the 104 observations were above 75% (Table 4). Three of the samples collected in 2021 had a DOSAT of 60% or less, all observed on August 3.

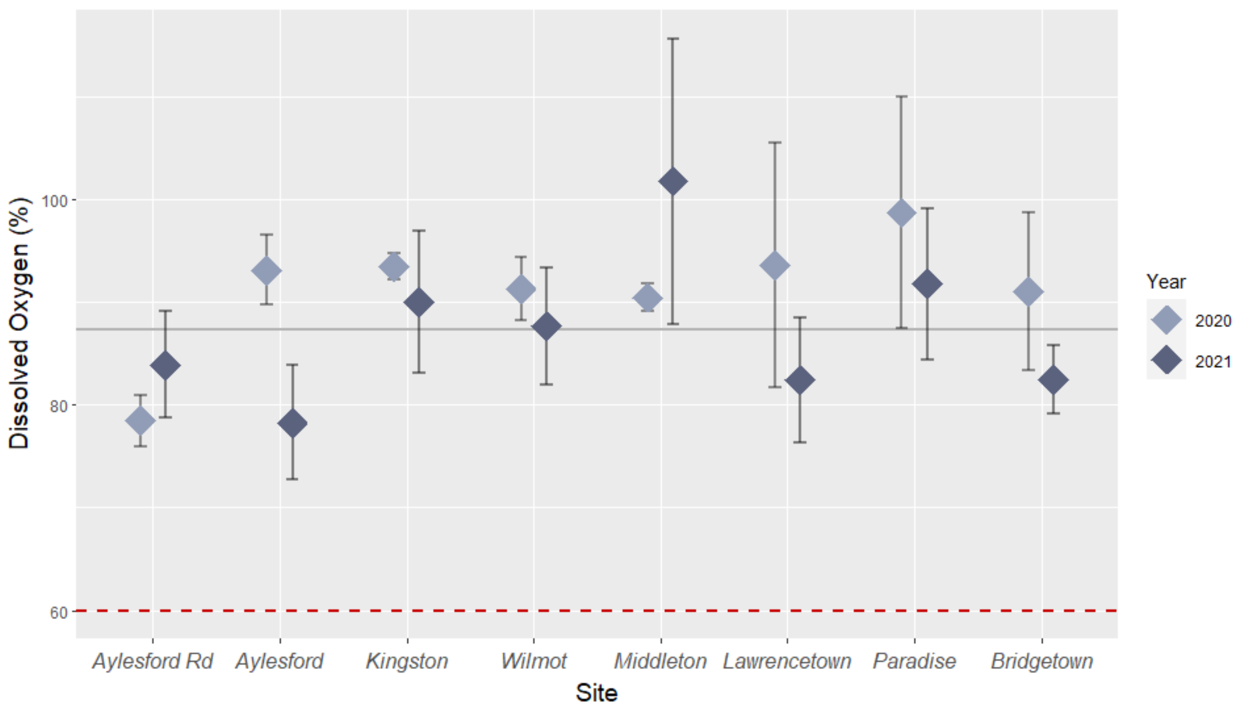

Figure 7. Average summer DOSAT results for 2021, compared to 2020, grouped by sample site. The error bars show standard error of the mean. The solid gray line indicates the historical summer average since 2003 (87.32%). The dashed red line shows the CCME guideline of 60%.

Table 3. Summer (July 1st – September 30th) average DO measurements at each of the eight River Guardians sampling sites. Data from 2021 are compared to the historical average encompassing 1992-2021 (including both the Quanta and volunteer data).

	DO(mg/L)		DO(%)	
Site	*1992-2021*	***2021***	*1992-2021*	***2021***
Aylesford Road	8.57	**8.27**	88.19	**83.90**
Aylesford	8.31	**7.77**	84.61	**78.28**
Kingston	8.79	**8.75**	90.07	**90.03**

Wilmot	8.67	**8.61**	89.31	**87.65**
Middleton	9.05	**9.49**	96.63	**101.77**
Lawrencetown	8.54	**7.60**	91.15	**82.40**
Paradise	8.74	**8.26**	93.80	**91.78**
Bridgetown	7.58	**7.47**	83.18	**82.48**

With the exception of Middleton, all sites had lower dissolved oxygen values in 2021 than the historic mean previously observed at each site (Table 3). This was true for both DO (mg/L) and DOSAT readings.

The mean dissolved oxygen values, calculated in mg/L were calculated for each of the main river monitoring sites and compared to data from 2020 (Figures 7 and 8). The standard error of this mean is shown with error bars. The mean summer values for the 2021 monitoring season show lower variability and lower mean values in the three most downstream sites when compared to 2020 values. Variability was much greater in 2021 at the Aylesford Rd, Kingston and Middleton sites, and the average DO value was greater as well. Aylesford, Lawrencetown and Bridgetown fall below the historical summer average (2003-2021) in both DO(mg/L) and DOSAT(%). The lowest DO (mg/L) value that was recorded in 2021 was 5.47 mg/L in Lawrencetown on August 3rd, and the highest was 16.13 mg/L on October 4th, also in Lawrencetown.

Table 4. Dissolved oxygen percent saturation (DOSAT) thresholds for the Annapolis River. Full season counts, as well as counts for the summer subset of July-September (shown in brackets).

Site	Samples less than 60%	Samples within 61-74%	Samples greater than 75%	Total Samples 2021
Aylesford Road	1 (1)	0 (0)	12 (5)	13 (6)
Aylesford	1 (1)	4 (1)	8 (4)	13 (6)
Kingston	0 (0)	1 (1)	12 (5)	13 (6)
Wilmot	0 (0)	2 (1)	11 (5)	13 (6)
Middleton	0 (0)	1 (1)	12 (5)	13 (6)
Lawrencetown	1 (1)	2 (1)	10 (4)	13 (6)
Paradise	0 (0)	1 (1)	12 (5)	13 (6)
Bridgetown	0 (0)	3 (1)	10 (5)	13 (6)
Totals	**3 (3)**	**14 (7)**	**87 (38)**	**104 (48)**

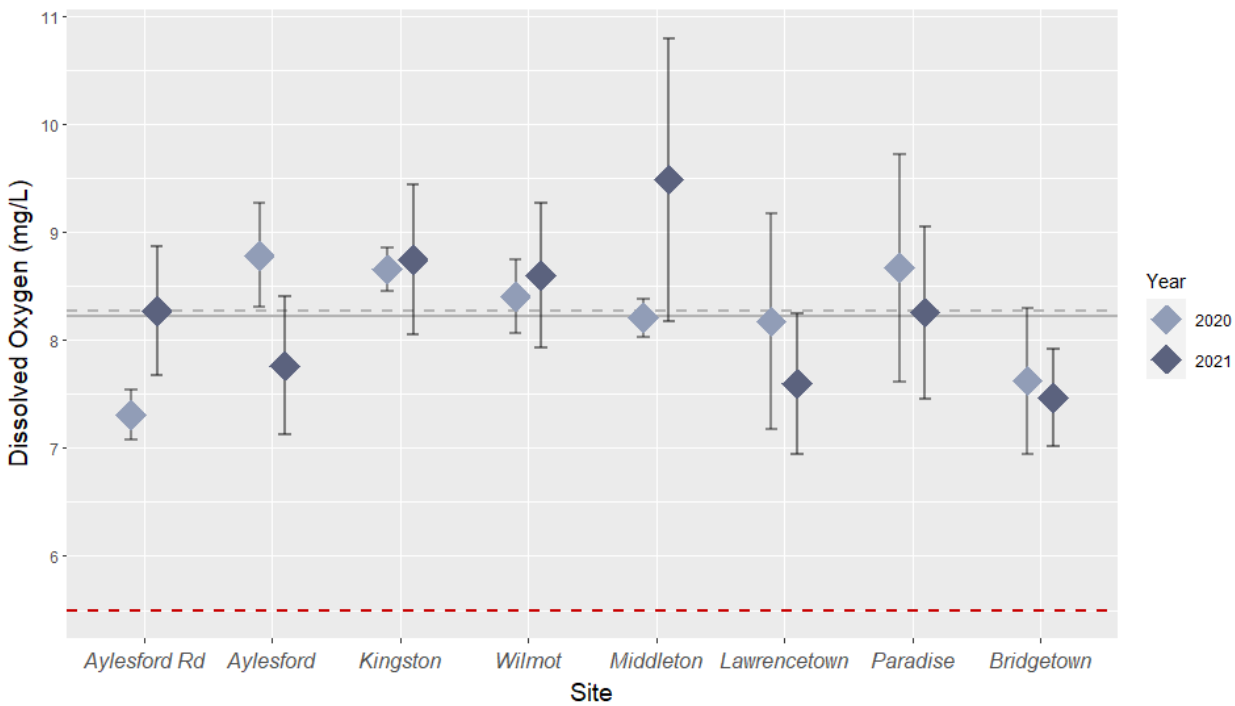

Figure 8. Average summer DO (mg/L) results for 2021, compared to the 2020 results (mg/L), organized by sample site. The error bars show standard error of the mean. The grey dashed line shows the summer average from 2021 (8.27mg/L), and the solid gray line shows the historical summer average since 2003 (8.22mg/L). The red line indicates the CCME guideline threshold.

All sites measured still had mean values that were well above CCME guidelines for cold water species (i.e. 5.5 mg/L). The site with the highest mean DO (mg/L) in 2021 was Middleton. Only one observation fell below the threshold of 5.5mg/L, and a large majority of samples were above 6.5 mg/L (Table 5).

Over the entire sampling season, only one observation fell below the 5.5 mg/L value in 2021 (5.47 mg/L; Lawrencetown August 3), and an additional 14 recordings fell below 6.5 mg/L (Table 5), 7 of which occurred on August 3rd. The high levels of dissolved oxygen historically observed at Middleton are likely due to input from the Nictaux River tributary, which is fast flowing and well oxygenated. The Nictaux River joins with the Annapolis River approximately 400 m upstream from the Middleton site.

Table 5. Dissolved oxygen (mg/L) thresholds for the Annapolis River. Full season counts, as well as counts for the summer subset of July-September (shown in brackets).

Site	<5.5 mg/L	5.5 to 6.5 mg/L	>6.5 mg/L	Total Samples 2021
Aylesford Road	0 (0)	1 (1)	12 (5)	13 (6)
Aylesford	0 (0)	4 (2)	9 (4)	13 (6)

Kingston	0 (0)	1 (1)	12 (5)	13 (6)
Wilmot	0 (0)	1 (1)	12 (5)	13 (6)
Middleton	0 (0)	1 (1)	12 (5)	13 (6)
Lawrencetown	1 (1)	2 (1)	10 (4)	13 (6)
Paradise	0 (0)	1 (1)	12 (5)	13 (6)
Bridgetown	0 (0)	3 (2)	10 (4)	13 (6)
Totals	**1 (1)**	**14 (10)**	**89 (37)**	**104 (48)**

2.2.3 Dissolved Oxygen Monitoring Recommendations

- Continue the regular River Guardian DO monitoring program at the eight main river sample locations.
- Undertake DO monitoring of the Annapolis River estuary in the late summer and early autumn. These times are most likely to display depressed levels of DO. Depth profiling should be included as part of this monitoring.
- Investigate atmospheric pressure readings to determine whether or not they vary enough to affect dissolved oxygen readings.
- Compile the historic volunteer data and Quanta probe data into a single dataset that includes all DO observations for easier and more comprehensive analyses.

2.3 Temperature

2.3.1 Introduction

Water temperature, like dissolved oxygen, serves as a broad indicator of water quality. The temperature of water has a direct bearing on the aquatic species present and their abundance. For example, trout and salmon species experience stress at water temperatures in excess of 20°C, with lethality occurring after prolonged exposures to temperatures over 24°C (MacMillan et al., 2005).

2.3.2 Monitoring Results

The mean summer water temperature for the Annapolis River in 2021 was 17.43°C, which is 1.3°C and 0.06°C colder than the same periods in 2020 and 2019, respectively. As in previous years, water temperatures continued to reach and exceed levels stressful to aquatic life during the summer months (Figure 9). The 2012 season had the highest recorded mean summer water since the inception of the River Guardians program. This was followed by a decline in summer water temperatures in both 2013 and 2014. 2021 temperature data had a narrower range of variability as compared to data recorded in previous years (Figure 9).

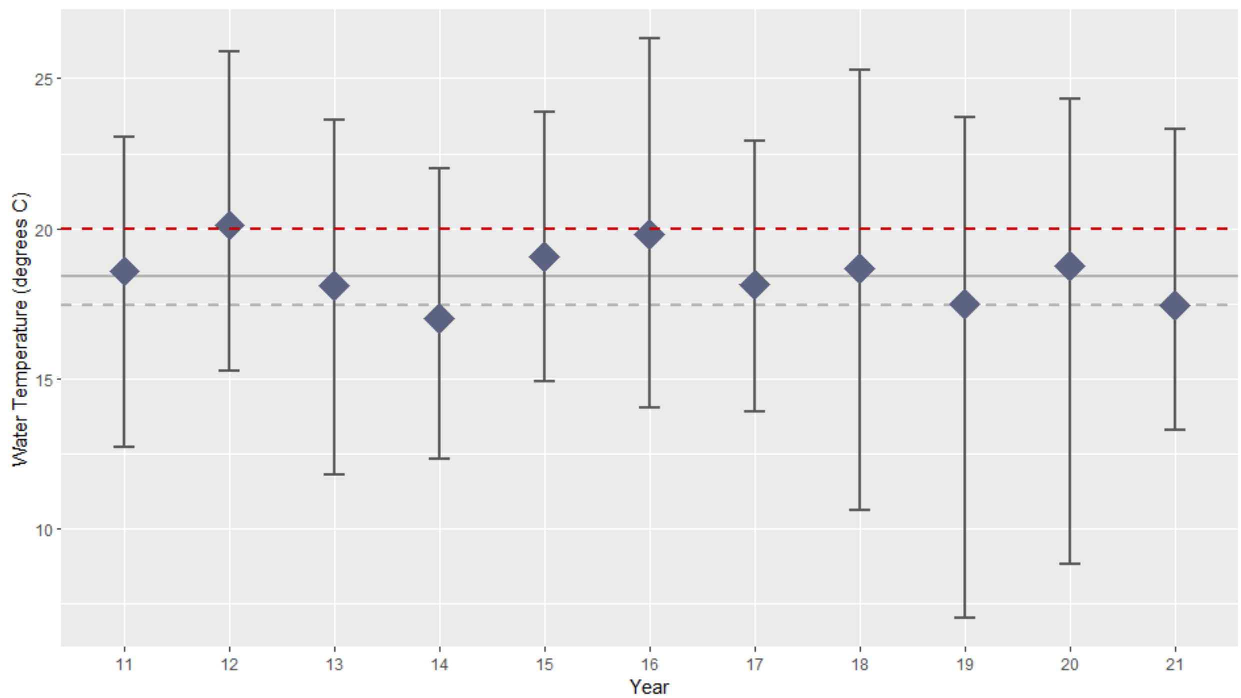

Figure 9. Mean summer water temperatures by year (showing full range of temperature values) with the 2003-2021 mean shown as a solid gray line. The 20°C threshold where fish become stressed is shown as a dashed red line, and the summer average for the 2021 season is shown by the grey dashed line.

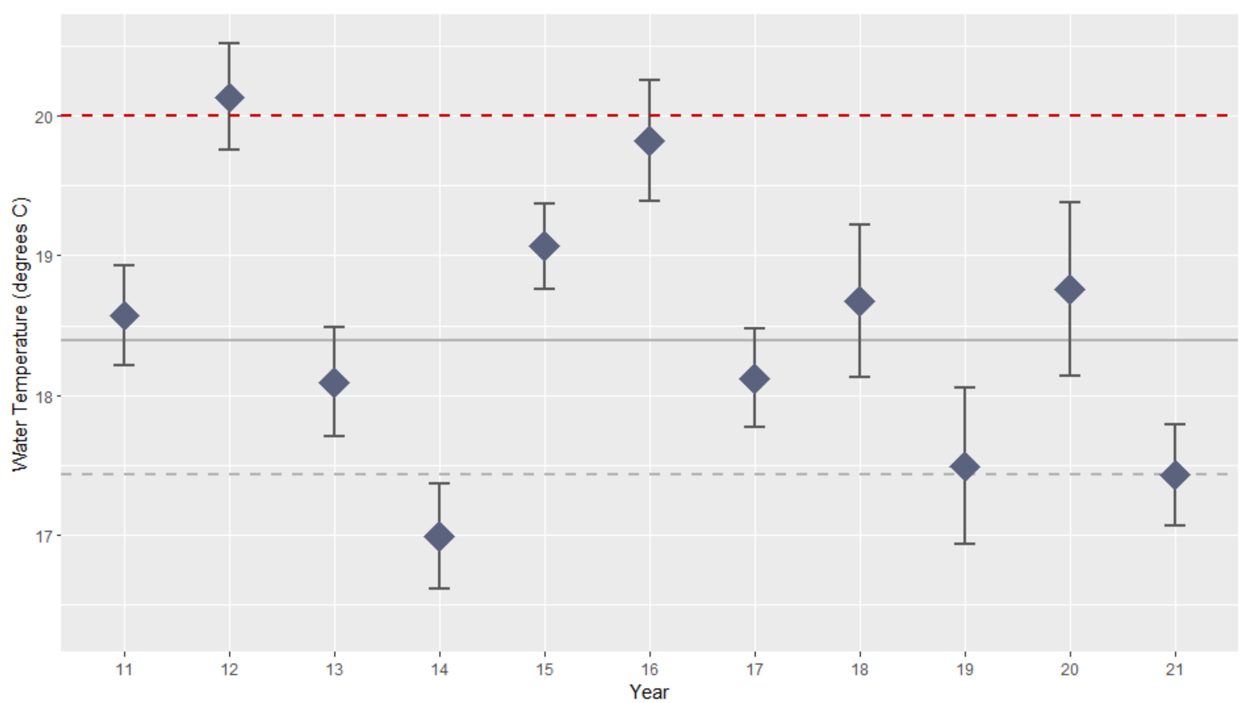

Figure 6. Mean summer water temperatures by year (showing standard error of the mean) with the 2003-2021 mean shown as a solid gray line. The 20°C threshold where fish become stressed is shown as a dashed red line, and the summer average for the 2021 season is shown by the grey dashed line.

The mean summer water temperature (July 1st – September 30th) by year was compared to the 2003 to 2021 historical summer average water temperature (Figure 10). The average water temperature for 2021 is 0.96 °C below the historical average. The data from previous River Guardians annual reports suggested a gradual increase in temperature in the lower river sites, particularly in the summer data. The mean summer water temperature values along the Annapolis River in 2021 were compared to the 2020 averages for those sites (Figure 11), and all sites show a decline in average water temperature compared to last year's data. In 2021, Lawrencetown, Paradise, and Bridgetown are the only sites that had averages above the historical average. All other sites were below the historical average of 18.39°C. Aylesford Road had the greatest deviation with an average temperature 2.6°C colder than the historical mean.

Of the 48 discrete water temperature measurements recorded during the months of July, August and September in 2021, only 15% exceeded 20°C, compared to 44% in 2020. The maximum water temperature observed was 23.3°C, recorded at Bridgetown on August 17th, 2014.

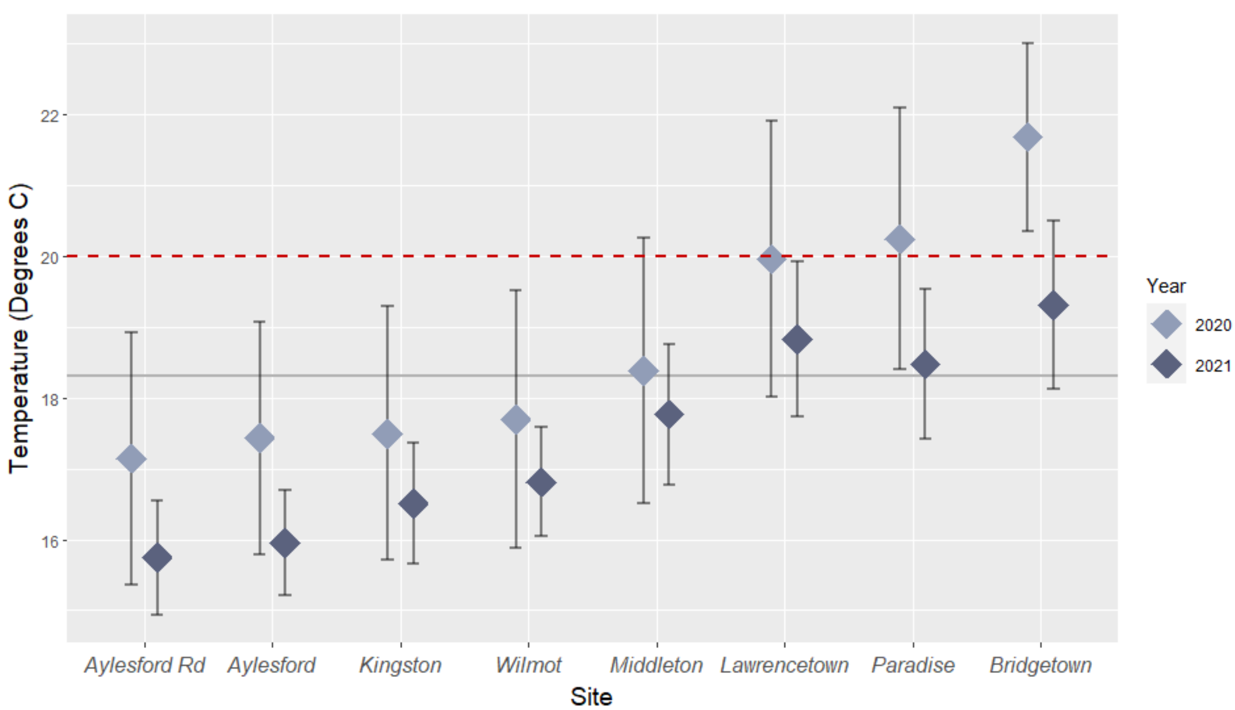

Figure 7. Mean summer water temperatures in 2021 and 2020, grouped by site, showing standard error of the mean. The 20°C threshold where fish become stressed is shown as

a dashed red line and the solid gray line denotes the historic summer average (2003-2021) of 18.39°C.

In previous years, dataloggers have been installed at the Aylesford Road and Kingston monitoring sites over the summer to passively monitor water temperature. While these methods were not repeated in 2021, the insights from the 2014 season are still of interest: dataloggers deployed in 2014 indicated that the most stressful temperatures exist around 3pm. In 2021, each River Guardians sampling day was completed by a single field crew, moving from Bridgetown in the upstream direction, and ending with Aylesford Road. Due to this limitation, sampling occurred over a period of up to 3 hours, and took place as early as 7:15 and only as late as10:50am: well before even the 12pm sampling time that was followed during the volunteer-run program. This sampling method could have implications for the analysis, as temperatures are likely to reach higher values than what is reported. This also makes it more difficult to draw comparisons to data from previous years, as sampling methods are not consistent.

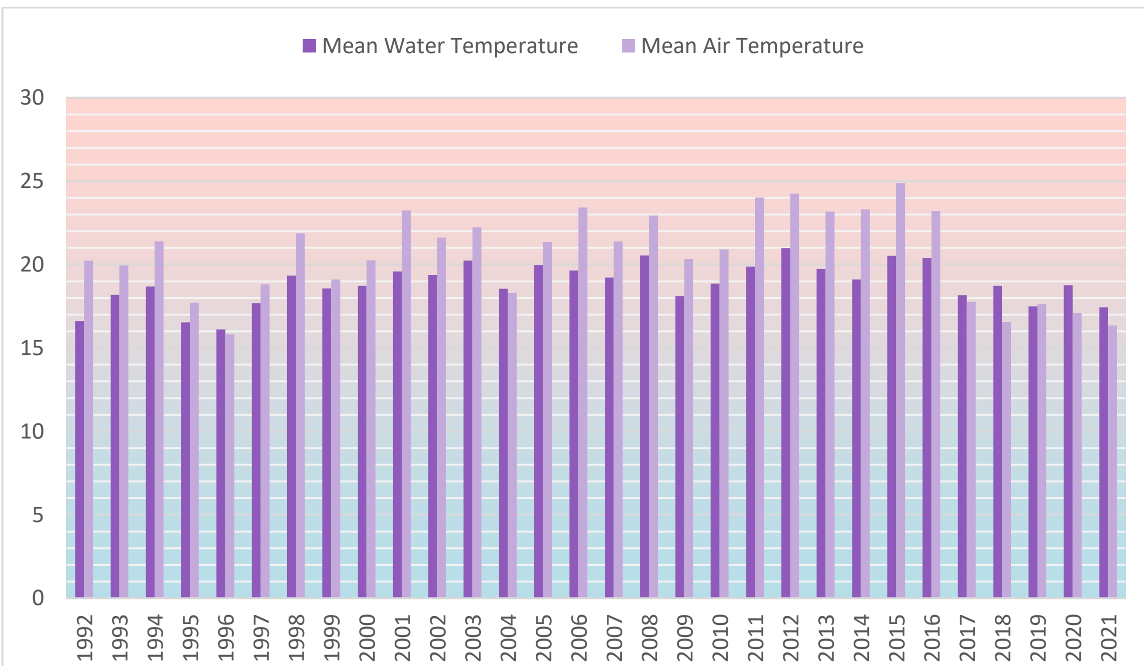

Figure 12. Mean summer air and water temperatures (1992-2021) by year. Since 2018, mean water temperatures have been greater than mean air temperature.

The mean summer water and air temperatures are shown by year in Figure 12, for each year from 1992 to 2021. Water and air temperature data in this graph both come from the Volunteer data archives, as it is the only place that reports air temperature readings. Historically, higher air temperatures have coincided with higher water temperatures, except in 1996 and 2004, where mean air temperatures were slightly below mean water temperatures. It is possible that River Guardian sampling dates in these years fell on colder days of the summer, which may explain the slightly lower air temperature values. However 4 of the past 5 years have documented a lower mean summer air temperature than the mean summer water temperature. The mean summer air temperature for 2021 was 16.3˚C, which was 0.75˚C and 1.29˚C lower than the mean annual air temperature in 2020 and 2019, respectively. The mean air temperature in 2021 was 1.08˚C lower than the average water temperature, compared to 1.66˚C lower in 2020. The earlier and extended daily sampling windows that are observed after volunteer involvement was ceased could explain this trend. Air temperatures are cooler in the early hours of the day, and while water temperatures fluctuate similarly, the magnitude of fluctuation is often greater for air temperature.

The coefficient of determination (R^2) in Figure 13 denotes a value of 0.7292. This means that 72.9% of the variance in water temperature values can be explained by changes in air temperatures, a decrease from the value of 74.9% reported in 2014. A perfect correlation (data all on the trendline) would have an R^2 of 1, while data with little to no correlation would have values closer to 0. Therefore, there is a strong positive correlation between the air and water temperatures (Figure 13).

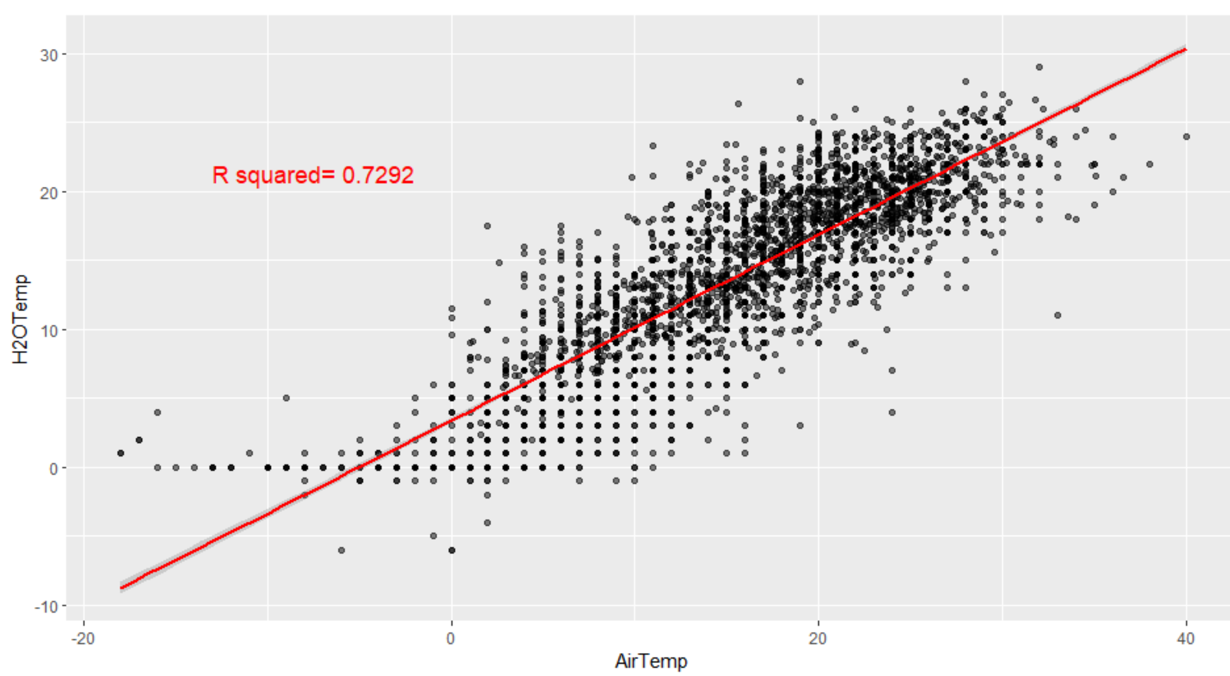

Figure 13. Correlation between air and water temperature values from 1992 to 2021.

2.3.3 Water Temperature Monitoring Recommendations

- Continue regular River Guardian temperature monitoring program at the eight main river locations.
- Continue temperature logger installations at regular monitoring sites along the Annapolis River.
- Install temperature loggers in candidate streams to assess for fish habitat improvements.
- Temperature data loggers should be calibrated immediately prior to deployment and at least once in situ. These procedures should be added to the QA/QC Project Plan.
- Investigate the temperature increase on the Annapolis River between Aylesford and Lawrencetown. This may include collection of thermal status data on tributaries to the Annapolis River.

- Conduct sampling later in the day to conform with historical data collection methods, and to increase the likelihood of capturing maximum daily temperatures, or consider deploying passive data loggers at those sites for the entire sampling period.

2.4 pH

2.4.1 Introduction

pH is a measure of the acidic/basic nature of water and is determined by measuring the concentration of the hydrogen ion (H^+). It is expressed on a logarithmic scale from 0 to 14, with zero being the most acidic and 14 the most basic. As pH is an inverse logarithmic scale, every unit decrease in the pH scale represents a tenfold increase in acidity. To ensure the health of freshwater aquatic life, pH levels should not fall outside the range of 6.5-9.0 (CCME, 2002). Levels below 5.0 are known to adversely affect many species of fish, including salmon and trout. pH varies naturally depending on a river system's underlying bedrock and soil composition, as well as by the amount of aquatic plants and organic material present, but can also be influenced by anthropogenic means such as acid precipitation and increased atmospheric CO_2 concentrations (Dodds and Whiles, 2010).

2.4.2 Monitoring Results

Unlike a vast majority of river systems in Nova Scotia, pH values all along the Annapolis River are generally good, being only slightly acidic (Figure 14). The probable cause is the Torbrook Geological Formation, which is carved by many of the rivers tributaries, and contains limestone that helps buffer the watershed from acidification. Out of the 48 summer samples of 2021, the lowest value recorded within the summer sampling period (July 1st – September 30th) was 6.45

at Aylesford Road on September 7th; this was the only summer observation to fall outside the 6.5-9.0 range, however when considering the entire sampling season, an additional 9 observations are outside this range. The highest pH was 8.32 collected in Paradise on August 3rd. On average, pH was lowest at Aylesford Road and Wilmot, whose average pH values were 7.03 and 7.11, respectively (Figure 14).

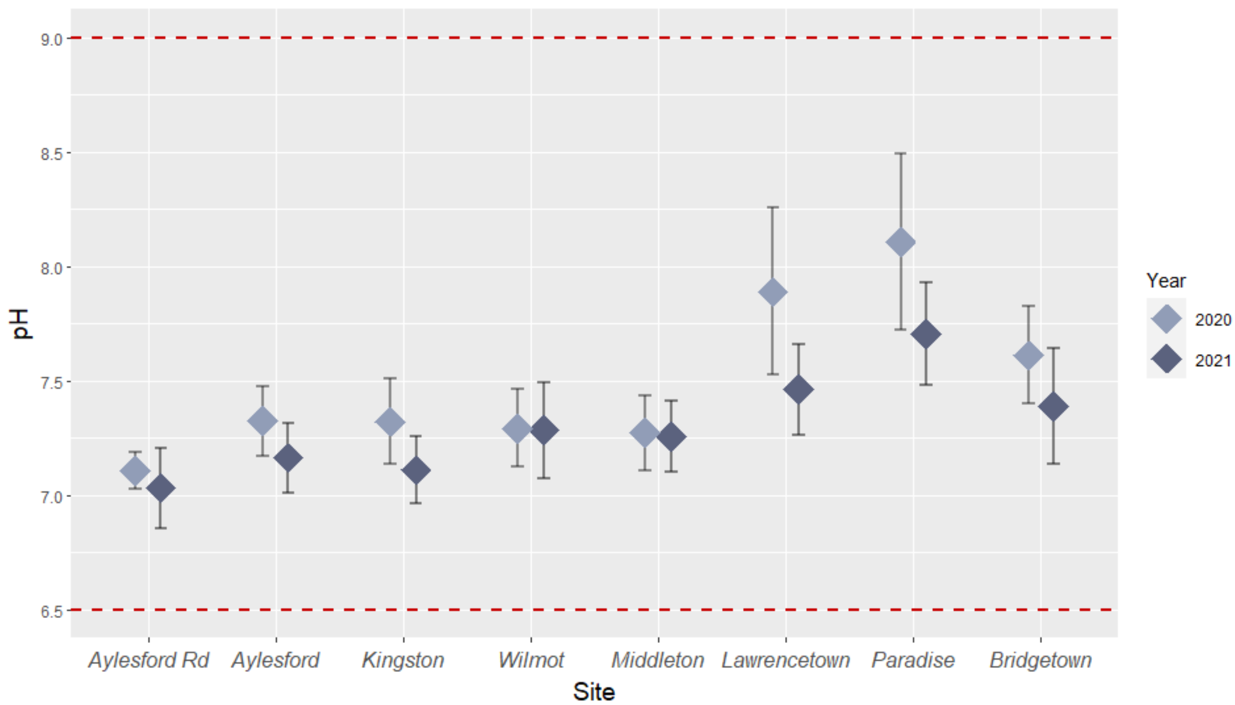

Figure 14. Average pH in summer 2021 for sampling locations along the Annapolis River (showing standard error of the mean), compared to values from 2020.

Throughout the past 10 years, average pH has been in the optimal range, although the 2019 average fell toward the lower end of the scale, reaching an average of 6.62 (Figure 15). On average, the pH has increased since 2011, although all locations had similar or lower pH levels in 2021, compared to 2020 (Figure 14).

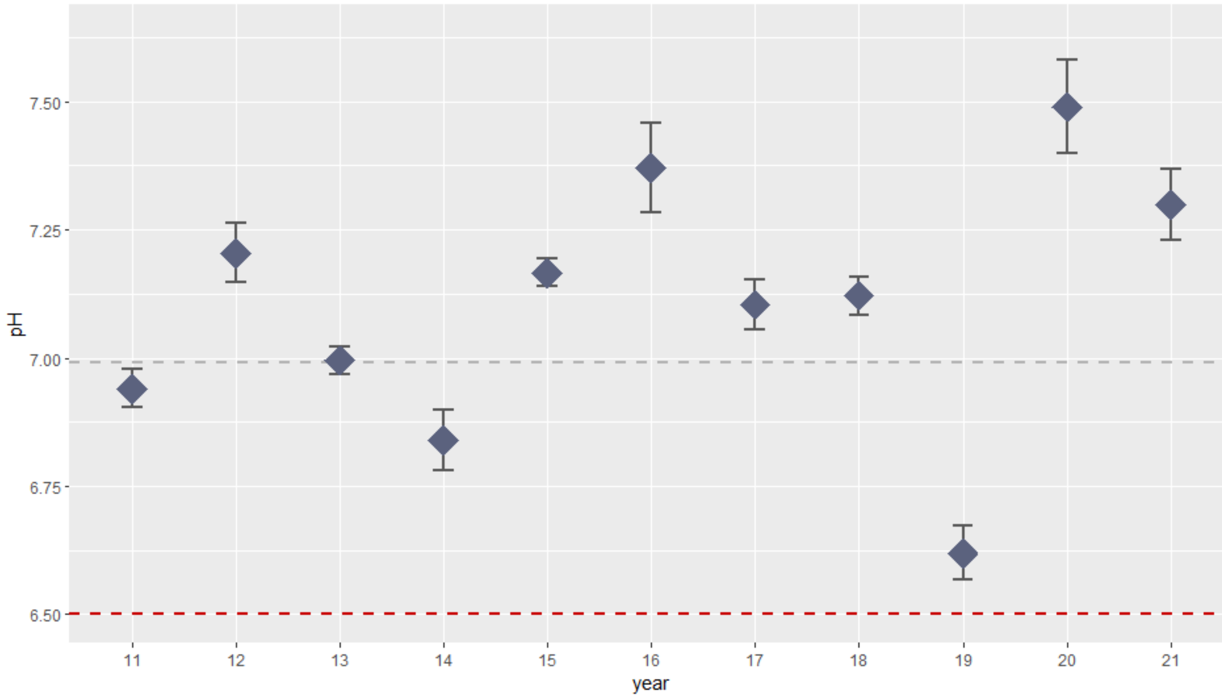

Figure 15. Average summer pH measured yearly along the Annapolis River (showing standard error of the mean) from 2011-2021. Shown by a thick red line is the lower threshold of 6.5 for fish species. The grey dashed line shows the historic average pH of 6.99 (encompassing all sites and dates since 2003).

2.4.3 pH Monitoring Recommendations

- Regular pH monitoring should be continued at the eight Annapolis River Guardian monitoring locations.

2.5 Nutrients: Nitrogen and Phosphorus

2.5.1 Introduction

Nutrients are essential for the growth of both plant and animal life. They can occur naturally, or as a result of anthropogenic activities. Two nutrients commonly monitored in freshwater systems are nitrogen and phosphorus, which are often found to be the limiting factors of plant growth in aquatic systems. When the levels of these nutrients rise, either from natural inputs or from anthropogenic sources such as wastewater or agricultural runoff, excessive periphyton and macrophyton growth can result. Upon the death and decomposition of these plants, oxygen levels can become depleted to such an extent as to threaten aquatic life.

In 2006 and 2007, Environment Canada monitored two locations along the Annapolis River for a large range of water quality parameters including nitrogen and phosphorus. In 2008, a reference site on the South Annapolis River in Millville was added and in 2009, the Lawrencetown sample site was dropped. Nutrient monitoring is currently only carried out at Wilmot and Millville.

Dodds et al. (1998) compiled information from hundreds of streams in the US and from the EPA eutrophication survey in order to compare criteria for measuring nutrients in streams. As nitrogen can be present in various soluble and insoluble forms in freshwater systems, differing criteria for nitrogen have been outlined for both total nitrogen and dissolved nitrates. Dodds and Welch (2000) determined that acceptable total nitrogen criteria ranged from 0.25 mg/L to 3.0 mg/L. This guideline, however, does not account for the effects of eutrophication, and was therefore determined to be too high for a threshold value in the Annapolis river watershed. An interim guideline of 0.9 mg/L total nitrogen was set as a criterion for the watershed, based on information obtained from Dodds and Welch (2000). It is believed that this value is more representative of that in which impairment through eutrophication is likely to occur. Total nitrogen was used as a threshold rather than dissolved nitrate as it measures all the nitrogen in a system rather than a portion of it.

In the case of phosphorus, there seems to be less variability between recommended criteria. The Ontario Ministry of Environment and Energy (OMEE) set a guideline of 0.03 mg/L total P, above which excessive plant growth occurs. Mackie (2004) suggested that total phosphorus levels in excess of 0.03 mg/L indicate that the surface waters are eutrophic. Dodds and Welch (2000) list upper limits ranging from 0.02 mg/L to 0.07 mg/L. For evaluation of phosphorus in the Annapolis River watershed, a criterion of 0.03 mg/L was used to indicate potential impairment through eutrophication.

2.5.2 Monitoring results

The nutrient results shown in this section were collected and analyzed by Environment Canada. Environment Canada collects regular water quality samples at one location on the Annapolis River and one location on the South Annapolis River. Grab sampling for 2020 was performed in Wilmot, near the bridge and gauging station on Bayard Road and in Millville, near the bridge on Victoria Road. Results for the 2021 season were not included in this report, as they were not yet available at the time the report was created.

Total nitrogen exhibits a wide range of values that appear to follow a slight annual trend. Wilmot values range from 1.22 mg/L on February 11th, 2009 to 0.36 mg/L on April 8th, 2009 (Figure 16). At the Millville Reference site, the initial reading is the minimum recorded, at 0.11 mg/L on May 1st, 2008, with a peak reading on December 13th, 2016 at 0.65 mg/L. In the past three years, only 1 of the 24 samples collected at Wilmot has exceeded the 0.9 mg/L guideline, and that was on October 12th, 2018 with a reading of 1.22 mg/L. In 2020, all three samples collected were below this guideline.

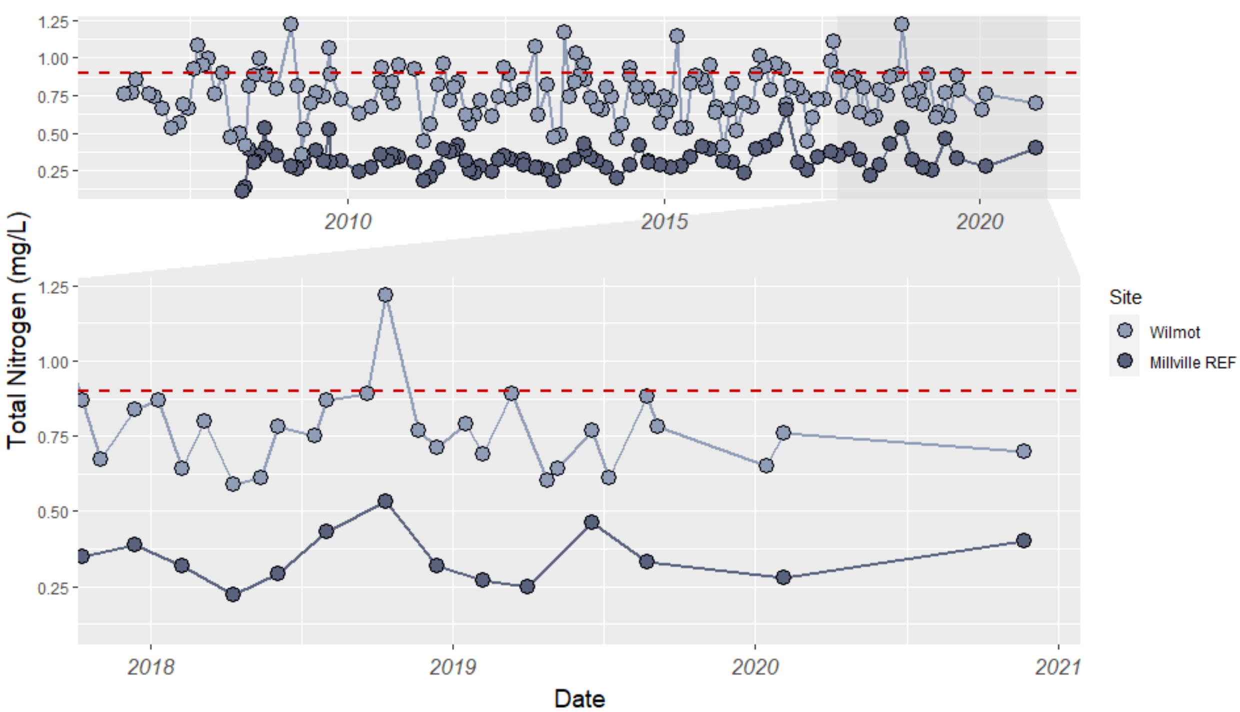

Figure 16. Total nitrogen results from 2006-2020 for Wilmot, and 2008-2020 for the Millville Reference site. The dashed red line represents the threshold value of 0.9 mg/L, above which conditions are deemed to be unacceptable. The bottom panel shows a subset of the sampling period from 2018 to 2021.

Total nitrogen at all three sites exhibit seasonal fluctuations, with greater variability observed in the values recorded at the Wilmot locations, compared to the Millville reference site (Figure 16). Values at both locations peak in the summer season and drop in the winter. In general, total nitrogen starts to decrease in the late summer and continues to decline until near April when values begin to climb again, although several spikes are observed periodically at the Wilmot location. This variation may be the result of agricultural fertilizers and other anthropogenic factors affecting land surrounding the river. Also, groundwater in the Wilmot area has been shown in the past to have elevated nitrate levels (Nova Scotia Environment, 2009). Most results fall above the upper limit of 0.25 mg/L to 3.0 mg/L that can cause adverse ecological effects, described by Dodds and Welch (2000), but most fall below the 0.9 mg/L guideline for the Annapolis watershed. In the 2020 field season, total nitrogen levels were recorded at lower levels than in past records (Figure 17), however the sample size in 2020 is small by comparison, with no observations from the summer season.

The general trend of total phosphorus observed in the Annapolis River increases from spring to summer and decreases from summer to winter (Figure 17). The maximum total phosphorus of 0.171 mg/L was observed at Wilmot on October 12th, 2018 and the minimum of 0.006 mg/L was recorded at the Millville Reference site on April 9th, 2013. Of all the data collected, 76% of samples from Wilmot were above the recommended upper limit of 0.030 mg/L. Millville values were generally below this guideline, however, on September 15th, 2009 the total phosphorus value reached a maximum of 0.08 mg/L.

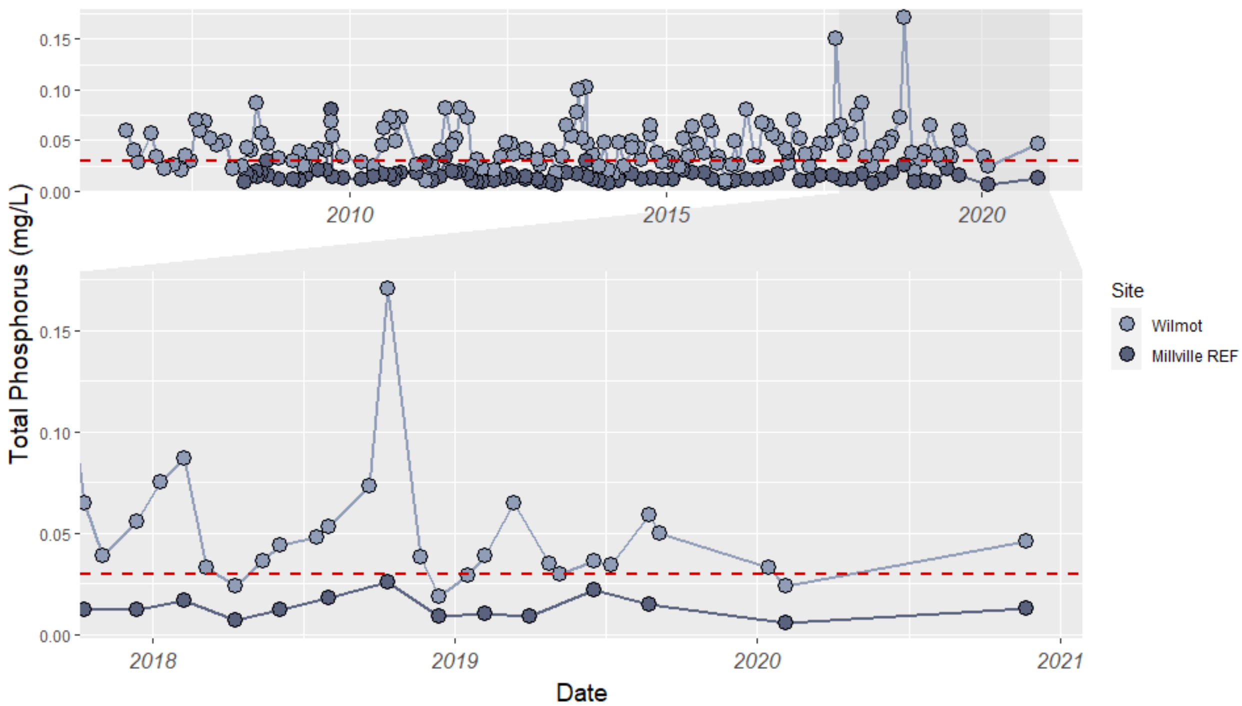

Figure 17. Total phosphorus results from 2006-2020 for Wilmot, and 2008-2020 for the Millville Reference site. The dashed red line represents the phosphorus guideline of 0.03 mg/L (Mackie, 2004). The bottom panel shows a subset of the sampling period from 2018 to 2021.

Table 6. Mean, minimum, and maximum values for total nitrogen and total phosphorus at each location. Results are from 2008-2020 for Millville, 2006-2020 for Wilmot, and 2006-2009 for Lawrencetown.

Location	Total Nitrogen (mg/L)			Total Phosphorus (mg/L)		
	Mean	Min	Max	Mean	Min	Max
Millville Ref Site	0.40	0.11	0.65	0.032	0.006	0.08
Wilmot	0.69	0.36	1.22	0.040	0.010	0.171
Lawrencetown	0.60	0.20	1.07	0.031	0.018	0.056

Table 6 summarizes the average, as well as maximum and minimum nutrient values recorded at each of the nutrient monitoring sites, including the Lawrencetown site in which sampling ceased in 2009. Overall, Wilmot exhibited a higher nutrient concentration for total nitrogen and total phosphorus than either Lawrencetown or Millville (Table 13). Millville has the lowest average for total nitrogen, and is only slightly above the Lawrencetown total phosphorus average, although this could be attributed to the difference in the number of sampling years. Therefore, the locations in order of increasing nutrients, and thus decreasing river health are Millville, Lawrencetown and Wilmot. High *E. coli* values observed at Aylesford and Kingston may help explain the high nutrient values at Wilmot, as they both can be an indicator of a contamination source. Also, between Wilmot and Lawrencetown, the Nictaux River, Black River and other tributaries enter the Annapolis River, possibly diluting the nutrients resulting in lower concentrations at Lawrencetown.

2.5.3 Nutrient Monitoring Recommendations

- Work in collaboration with Environment Canada to ensure the continued collection of nitrogen and phosphorus samples at Millville and Wilmot.
- Examine flow rates in the Annapolis River near the nutrient sample collection points, as flow has a great influence on nutrient concentrations.
- Conduct analyses for traceable compounds found in fertilizers and wastewater treatment discharges to determine sources of nutrient inputs.
- Take more nutrient samples at various sites along the river. Add nutrient monitoring to the regular monitoring regime.

2.6 Total Suspended Solids and Turbidity

2.6.1 Introduction

Total suspended solids (TSS) and turbidity are both terms that describe the amount of suspended particulate matter in water, although they are measured in different ways. TSS describes the physical mass of the particulate matter, while turbidity refers to the extent that light will penetrate the sample. Highly turbid waters have poor light penetration, which can hinder the growth of aquatic plants and can affect the health of aquatic animals.

Throughout 2008 and 2009, CARP and Environment Canada worked together in order to establish baseline levels of TSS and turbidity for the Annapolis River, to be used in determining a water quality objective for these parameters. This water quality objective could then be used in the calculation of a water quality index for the Annapolis River, which would be useful for annual reporting. The monitoring was also conducted to help determine the relationship between TSS and Turbidity. The two measurements are related, but this relationship is unique for every waterway and must be determined. In order to develop this relationship, TSS and turbidity samples were taken simultaneously for each station along the Annapolis River for the duration of the 2008 and 2009 sampling season. In 2010, samples were only taken at Bayard Road, Wilmot, Middleton, and Paradise after 15 mm of precipitation had fallen to assess peak sediment levels in the river. In 2011, event samples were taken from Lawrencetown and Millville in addition to the other sites.

TSS was measured by the River Guardian program from 1992 to 2002. Although it was recognized that TSS is an important parameter for the Annapolis River, sampling was discontinued in 2003. It was felt that the procedure was time-consuming, failed to record the inherent variability of the parameter and was producing unreliable results (Dill, 2003). The revised protocol used in 2008 and 2009 required biweekly sample collection in addition to samples gathered after events of significant rainfall or snowmelt. These event readings were taken by either CARP staff or volunteers. At first, event samples were gathered after rainfall amounts of at least 5 – 10 mm, but it was found that this amount of rainfall had very little effect on the TSS and turbidity readings. The collection protocol was subsequently revised, with samples only being collected for rainfall amounts of at least 20 – 30 mm. In 2010 and 2011, samples were taken after at least 15 mm of precipitation had fallen.

The relationship curve developed for the Annapolis River (Figure 18), was used to estimate TSS loadings to the river in the 2021 analysis. Turbidity readings were not collected in 2018 or 2019, and the turbidity data collected in 2020-2021 are measured in FNU, as opposed to the previously used NTU measurements. The two units are measured differently, but are essentially equal values (1NTU = 1FNU).

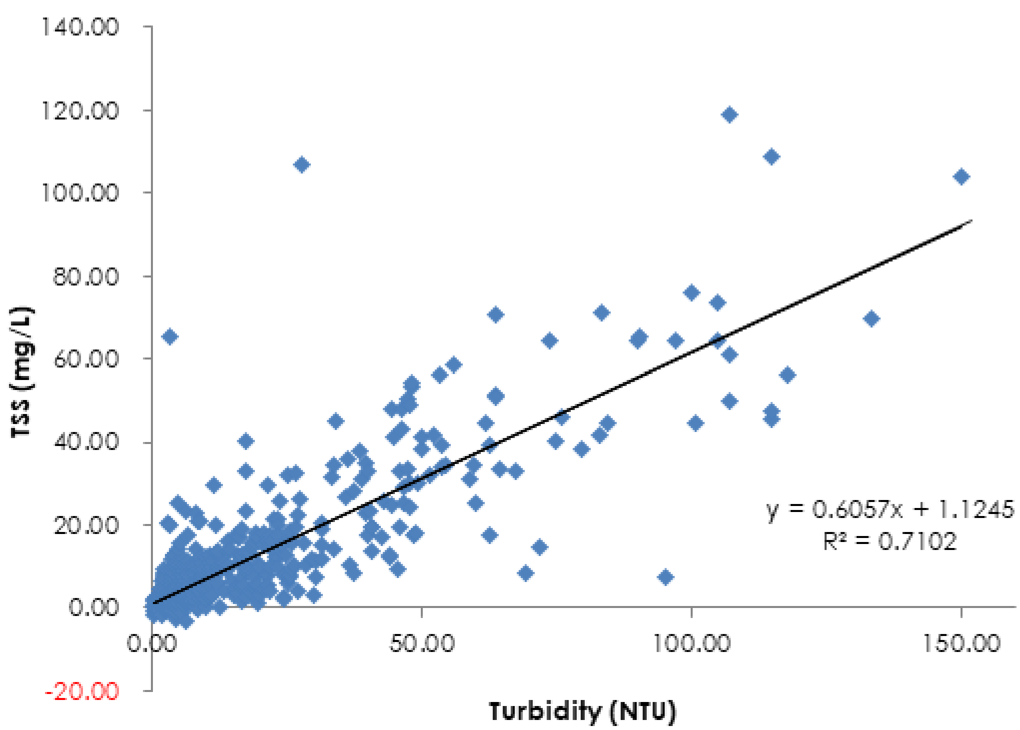

Figure 18. TSS in mg/L vs. turbidity in NTU for all sampled locations along the Annapolis River with the best-fit line and equation

2.6.2 Monitoring results

Turbidity data has been gathered as part of the regular biweekly monitoring regime of the River Guardian program since 2009. Turbidity data collected from April to October can be found in Figure 19. TSS values in Figure 20 were estimated based upon the preliminary relationship developed for the river with a best fit line equation. The best-fit line and equation was generated in 2014, using historical data, and is illustrated in Figure 18. The R^2 value derived from the regression analysis was determined to be 0.71, meaning that 71% of the variance in TSS readings can be explained by changes in turbidity. Recommendations in 2014 suggested that more data be collected to improve the accuracy of this relationship and to test the validity of the best fit equation. This has not yet been done, and should be prioritized during the 2022 season.

Turbidity levels in the river to date have ranged from lows of 0 NTU on several occasions, to a high of 398 NTU on October 18th, 2010. The maximum turbidity value observed in 2021 was 33.4 FNU, recorded at Kingston on October 18th, but since event sampling is no longer a regular part of the data collection methods, it is possible that turbidity levels exceeded this. The lowest turbidity value observed in 2021 was 0.06 FNU, recorded on May 25th at Kingston.

Figure 19 shows turbidity fluctuations between individual sites, which can be compared to TSS values derived from the regression analysis in Figure 20. Mean turbidity levels in the Annapolis River were below the determined safe level (interim guideline of 10 NTU) for all sites. Aylesford and Kingston had the highest mean turbidity values in 2021, which was a pattern observed in both turbidity and inferred TSS values. Aylesford Road had the lowest mean turbidity value in 2021. Many riverine turbidity analyses compare rainfall event turbidity levels to a standard background level for the particular waterway, and denote an increase of 8NTU to be of concern for short-term exposure. An increase as little as 2NTU can have negative impacts when sustained for long periods (CCME 2002). Similarly, a baseline level of 3NTU sustained for a period of over 10.5 months can have an impact on aquatic life, notable primary productivity (Birtwell et al. 2008). Average turbidity values observed in 2021 all exceed 3FNU, and so demonstrate the importance of continued turbidity monitoring in this system.

2.6.3 TSS/Turbidity Monitoring Recommendations

- Continue assessment to establish an accurate relationship between TSS and turbidity, which can be used to calculate TSS from the biweekly turbidity readings in the River Guardian Program
- Investigate possible correlations between TSS/Turbidity data, *E. coli* readings and rainfall amounts.

- Possibly extend the sampling season to exceed 10 months for monitoring whether long-term exposure to low turbidity levels may be impacting the system.

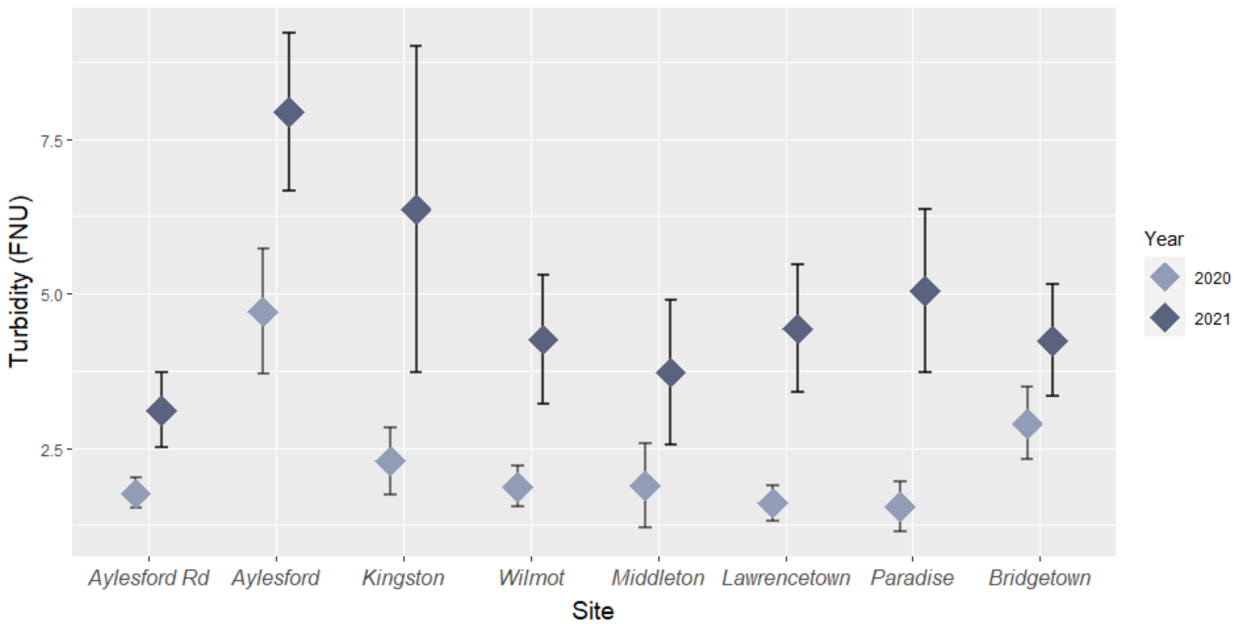

Figure 19. Turbidity (FNU) averages for each site, comparing data from 2021 and 2020 seasons.

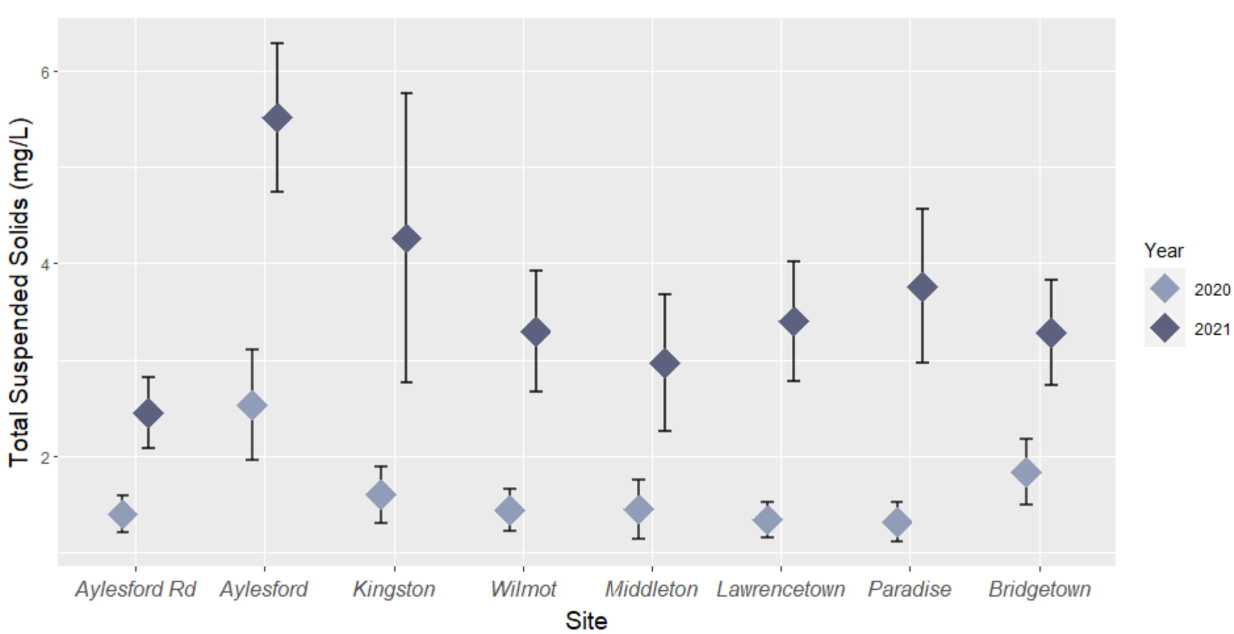

Figure 20. Total suspended solids (mg/L) averages for each site, inferred from turbidity (FNU) values using the best-fit equation from historical values (see Figure 18). Data from 2021 are compared to data from 2020.

3.0 Trend Analysis

3.1 Purpose

Beginning in 2006, Trend analyses have been completed for several of the water quality monitoring parameters. The results have been included in the annual River Guardians Report Cards. Trends have been calculated since 2008 using a Shapiro-Wilks parametric analysis test and a Mann-Kendall or seasonal Kendall non-parametric test. If trends were found, they were reported as either increasing or decreasing, otherwise the parameter was reported as having no discernible trend.

3.2 Background Information

There are several different ways of reporting trends in a series of data, depending on the nature of the data set. Many of the statistical methods fall under two broad categories, parametric and non-parametric. Parametric methods are used for normally distributed data, while non-parametric methods are suited for non-normally distributed data. Methods of each type were attempted for the trend analysis of the water quality data.

The parameters assessed using these two methods were bacteria counts, DOSAT, temperature and pH. DOSAT was selected over DO because DO values are dependent on temperature, therefore, temperature trends might cause DO trends to be masked or indicated when they are not appropriate. Nutrient trends were also analyzed for Wilmot using parametric methods.

The procedure used for the non-parametric analysis performed in 2014 was based on a procedure provided by D. Parent of Environment Canada and used by Glozier, Crosley, Mottle and Donald (2004). This procedure involved:

- separation of the data by station for each parameter
- a visual assessment of the data time series, which includes dividing the data into season according to the box-plot
- checking outliers for errors in measurement
- the Kruskal-Wallis test for seasonality
- either the Seasonal Kendall test or the Mann-Kendall test depending on whether the data displayed seasonality.

R code and methodology instructions for the boxplots, outlier checks, and Kruskal-Wallis tests were not available for the 2021 analysis. Time and resource limitations prevented us from filling these gaps during the 2021 analysis, and so it is recommended that this be prioritized during the 2022 analysis. The non-parametric analyses that were conducted this year were not checked for outliers beyond a visual assessment of max and min values in the Access database and Excel spreadsheets exports. Any outliers identified this way were cross-referencing with the original raw datasheets in the CARP office. This assessment did identify several errors that have since been corrected. In addition, the 2021 data were not tested for seasonality due to missing code for the Kruskal-Wallis tests. Parameters were instead analyzed using either the Seasonal Kendall or Mann-Kendall test in accordance with what was done in 2014 (*E. coli* was the only parameter for which the Seasonal Kendall test was used). We cannot ignore the possibility that seasonality is present in other parameters, and results should thus be interpreted with caution.

The Kruskal-Wallis test would otherwise be performed using R Studio and the Kendall tests were performed using a free DOS-based computer program for the Kendall family of trend tests developed by the United States Geological Survey. The program is available at http://pubs.usgs.gov/sir/2005/5275/downloads/ (Helsel, Mueller and Slack, 2006)

The parametric procedures as performed in 2014, were suggested by Drs. Y. Zhang and M. Brylinsky of Acadia University (pers. comm, December 2008). This procedure involved:

- separation of the data by station for each parameter
- a visual assessment for correlations between locations using scatterplot matrices
- a check for autocorrelation for each parameter and location
- an assessment for normality using the Shapiro-Wilks test
- transformations of the data if the parameter was found to be non-normal
- a linear regression of the data to determine whether a trend was present.

Similarly, the scatterpot matrices and tests for autocorrelation, which would otherwise be conducted in R studio, was missing R code, and could not be performed. Due to this, we cannot be certain that autocorrelation is not present in any of the parameters. The linear regressions and trend analyses were conducted despite this, but should be interpreted with caution.

The analysis procedures for parametric, non-parametric, and autocorrelation tests can be found in Appendix A, and should be referenced when re-writing the corresponding R code for future analyses. All trend analyses used the volunteer dataset and encompass a timeframe from 1992-2021. As noted earlier, there are some discrepancies between the volunteer and the Quanta data files, and so it might be on interest to collate these data into a single file moving forward. It may also be worthwhile to conduct an additional trend analyses in future years that illustrates a more recent window of time, e.g the last 10 years.

3.3 Results

The results for the non-parametric tests (Table 7) and the results for the parametric tests (Table 8) were compiled, and the trends are reported in Tables 9-11. Autocorrelation tests were not performed in 2021 due to missing R code. In 2014, autocorrelation tests were performed on all of the parameters to test for significant serial dependence, and none of the individual sites were found to exhibit significant serial dependence, while all sites together exhibited dependence. Based on this, trend analysis for the 2021 data was performed on individual sites only.

Table 7. Statistically significant trends* and rates of change using non-parametric procedures.

	Bacteria Count	DOSAT (%)	DO (mg/L)	pH	Water Temperature	Air Temperature
Aylesford Road	Yes (- 12.0cfu/ 100mL/year)	No	No	No	Yes (- 0.99˚C/year	No
Aylesford	No	Yes (+ 0.51 %/year)	No	No	Yes (+ 0.14˚C/year)	Yes (+ 0.16°C/year)
Kingston	Yes (+ 2.64cfu/ 100mL/year)	Yes (+0.48 %/year)	No	Yes (+ 0.01/year)	Yes (+ 0.22˚C/year)	Yes (+ 0.20°C/year)
Wilmot	No	Yes (+0.35 %/year)	No	No	Yes (+ 0.22˚C/year)	Yes (+ 0.17°C/year)
Middleton	No	Yes (+0.25 %/year)	No	Yes (+ 0.01/year)	Yes (+ 0.17˚C/year)	Yes (+ 0.10°C/year)
Lawrencetown	No	No	No	Yes (+ 0.02/year)	Yes (+ 0.27˚C/year)	Yes (+ 0.17°C/year)
Paradise	No	No	Yes (+ 1.0 mg/L/year)	Yes (+ 0.02/year)	Yes (+ 0.22˚C/year)	Yes (+ 0.24°C/year)
Bridgetown	No	Yes (- 0.22 %/year)	Yes (- 0.05 mg/L/year)	No	Yes (+ 0.17˚C/year)	Yes (+ 0.26°C/year)

*Statistically significant trends ($p<0.05$) using Seasonal Kendall and Mann-Kendall tests.

Table 8. Statistically significant trends* and rates of change using parametric procedures.

	Bacteria Count	DOSAT (%)	DO (mg/L)	pH	Water Temp	Air Temp	Turb	Total P (mg/L)	Total N (mg/L)
Aylesford Road	Yes (- 23.57cfu/ 100mL/year)	No	No	No	Yes (- 0.1°C/year)	No	No		
Aylesford	No	Yes (+ 0.26 %/year)	No	No	Yes (+ 0.16°C/year)	Yes (+ 0.18°C/year)	Yes (- 0.99FNU/year)		

Kingston	Yes (+9.46cfu/100mL/year)	No	Yes (- 0.08 mg/L/year)	No	Yes (+ 0.25°C/year)	Yes (+ 0.22°C/year)	No		
Wilmot	No	Yes (+ 0.2 %/year)	Yes (- 0.04 mg/L/year)	No	Yes (+ 0.23°C/year)	Yes (+ 0.19°C/year)	No	No	No
Middleton	No	Yes (+0.13 %/year)	Yes (- 0.05 mg/L/year)	Yes (+ 0.01 /year)	Yes (+ 0.45°C/year)	Yes (+ 0.13°C/year)	Yes (- 0.66FNU/year)		
Lawrence town	No	Yes (+ 0.15 %/year)	Yes (- 0.06 mg/L/year)	Yes (+ 0.02 /year)	Yes (+ 0.29°C/year)	Yes (+ 0.17°C/year)	No		
Paradise	No	Yes (+ 0.32 %/year)	Yes (- 0.03 mg/L/year)	Yes (+ 0.02 /year)	Yes (+ 0.24°C/year)	Yes (+ 0.25°C/year)	No		
Bridgetown	No	No	Yes (- 0.04 mg/L/year)	Yes (+ 0.03 /year)	Yes (+ 0.24°C/year)	Yes (+ 0.1°C/year)	Yes (- 0.43FNU/year)		

*Statistically significant trends ($p<0.05$, residual plot randomly distributed, initial confidence interval range does not overlap with final confidence interval range) using linear regression fit.

Table 9. Non-parametric trend interpretations of water quality. Green up arrows correspond to increasing water quality conditions, and red down arrows correspond to decreasing water quality conditions. White horizontal arrows suggest no discernable trend.

	Bacteria Count	DOSAT (%)	DO (mg/L)	pH	Water Temp	Air Temp
Aylesford Road	↑	↔	↔	↔	↑	↔
Aylesford	↔	↑	↔	↔	↓	↓
Kingston	↓	↑	↔	↓	↓	↓
Wilmot	↔	↑	↔	↔	↓	↓
Middleton	↔	↑	↔	↓	↓	↓
Lawrencetown	↔	↔	↔	↓	↓	↓
Paradise	↔	↔	↑	↓	↓	↓
Bridgetown	↔	↓	↓	↔	↓	↓

Table10. Parametric trend interpretations of water quality.

	Bacteria Count	DOSAT (%)	DO (mg/L)	pH	Water Temp	Air Temp	Total P (mg/L)	Total N (mg/L)
Aylesford Road	↑	↔	↔	↔	↑	↔		
Aylesford	↔	↑	↔	↔	↓	↓		

Kingston	↓	↔	↓	↔	↓	↓		
Wilmot	↔	↑	↓	↔	↓	↓	↔	↔
Middleton	↔	↑	↓	↓	↓	↓		
Lawrencetown	↔	↑	↓	↓	↓	↓		
Paradise	↔	↑	↓	↓	↓	↓		
Bridgetown	↔	↔	↓	↓	↓	↓		

Table 11. Overall water quality ranking for each monitored parameter, and the number of sites that show increasing, decreasing, or no trends since 2003.

Parameter	RANK	comments	Trends (2003-2021)
E. coli	FAIR	31 of 104 above 200cfu/100mL (threshold for human rec contact)	1 inc 1 dec. 6 none.
DOSAT	FAIR	101 of 104 above 60% DOSAT	5 inc. 0 dec 3 none
pH	GOOD	10 of 104 between 6.5-9ph	0 inc. 4 dec. 4 none.
Water Temp	GOOD	7 of 48 summer (Jul-sept) samples above 20 degrees	1 inc. 7 dec. 0 none
Turbidity	GOOD	12 of 104 below 10FNU	3 inc. 0 dec. 5 none (2014 stated "not enough information to evaluate trends")

4.0 Recommendations

4.1 Summary of Recommendations for the River Guardians Program

- Continue regular River Guardian *E. coli*, DO, temperature, pH, and turbidity monitoring at the eight main river sample locations.
- Address the issue of restricting livestock from the Annapolis River.
- Investigate the correlation between precipitation amounts and *E. coli* levels in the river.
- Continue monitoring efforts in the Annapolis River estuary, and develop regular estuary monitoring sites.

- Install temperature loggers in candidate streams to assess for fish habitat improvements.
- Temperature data loggers should be calibrated immediately prior to deployment and at least once in situ. These procedures should be added to the QA/QC Project Plan.
- Investigate the temperature increase on the Annapolis River between Middleton and Lawrencetown. This may include collection of thermal status data on tributaries to the Annapolis River.
- Work in collaboration with Environment Canada to ensure the continued collection of nitrogen and phosphorus samples at Millville and Wilmot, and consider adding nutrient data collection to the suite of parameters monitored.
- Examine flow rates in the Annapolis River near the nutrient sample collection points, as flow has a great influence on nutrient concentrations.
- Conduct analyses for traceable compounds found in fertilizers and wastewater treatment discharges to determine sources of nutrient inputs.
- Continue analysis of TSS/Turbidity to establish an accurate relationship, which can be used to calculate TSS from the biweekly turbidity readings in the River Guardian Program
- Investigate possible correlations between TSS/Turbidity data, *E. coli* readings and rainfall amounts.
- Review current and historic air photos of the Aylesford area and other amasses data, to identify land use changes and possible sources of contamination.
- Continue to identify possible sources of contamination along the Annapolis River in Aylesford.
- Research and implement a more definitive test for autocorrelation.
- Regularly perform volunteer and employee training and overview before each season to ensure proper technique and sampling consistency

4.2 Recommendations for CARP

- Continue to implement the Quality Assurance Project Plan for all of CARP's Water Quality monitoring programs.
- Calibrate the YSI every two or three weeks for pH, conductivity, dissolved oxygen and turbidity.
- Continue to update the manual for the River Guardian facilitator to ensure consistency in analysis and reporting.
- Continue to update the Annapolis River Guardian Procedures Manual on a continual basis.

- Continue to ensure QA/QC protocols are implemented yearly throughout the entire sampling season, including an information session before the first sampling date.
- Fill in the gaps in the data analysis instructions and corresponding R code.
- Consider collating the volunteer data with the Quanta data, to create a single, all-encompassing datafile for future analyses.

5.0 References

Addy, K. and L. Green. 1997. Dissolved Oxygen and Temperature. Natural Resources Fact Sheet No. 96-3. University of Rhode Island.

Australian and New Zealand Environment and Conservation Council. 2000. Australian Guidelines for Water Quality Monitoring and Reporting. Agriculture and Resource Management Council of Australia and New Zealand.

Beveridge, M., A. Sharpe, D. Sullivan. March 2006. Annapolis River 2005 Annual Water Quality Monitoring Report, Clean Annapolis River Project.

Birtwell, I.K., Farrell, M., and Jonsson A. 2008. The validity of including turbidity criteria for aquatic resource protection in Land Development Guidelines. (Pacific and Yukon Region). Canadian Manuscript Report of Fisheries and Aquatic Sciences. 2852: xiii + 72 p.

Bouchard Jr., R.W., 2004. Guide to Aquatic Invertebrates of the Upper Midwest. 4th Edition, Chironomidae Research Group, University of Minnesota, Saint Paul, Minnesota.

Canadian Council of Ministers of the Environment. 2002. Including Summary of Existing Canadian Environmental Quality Guidelines (December 2003).

Canadian Council of Ministers of the Environment. 2002. Canadian water quality guidelines for the protection of aquatic life: Total particulate matter. In: Canadian environmental quality guidelines, 1999, Canadian Council of Ministers of the Environment, Winnipeg.

Canadian Council of Ministers of the Environment. 2003. Water Quality for the Protection of Aquatic Life: Nitrate. Retrieved June 15, 2011, from http://st-ts.ccme.ca/?lang=en&factsheet=140.

Chalmers, R.M., H. Aird and F.J. Bolton. 2000. Waterborne Escherichia coli 0157. Journal of Applied Microbiology Supplement. 88: 124-132.

Chambers P.A., M. Guy, E.S. Roberts, M.N. Charlton, R. Kent, C. Gagnon, G. Grove, and N. Foster. 2001. Nutrients and their impact on the Canadian environment. Agriculture and Agri-Food Canada, Environment Canada, Fisheries and Oceans Canada, Health Canada and Natural Resources Canada. 241p.

Cooke, S. 2006. Water Quality in the Grand River: A Summary of Current Conditions and Long Term Trends. Grand River Conservation Authority.

Daborn, G.R., A.M. Redden, and R.S. Gregory, Ecological Studies of the Annapolis Estuary, 1981-82, The Acadia University Institute, Number 29, Wolfville, 1982.

Dalziel, J.A., P.A. Yeats and B.P. Amirault. 1998. Inorganic Chemical Analysis of Major Rivers Flowing Into The Bay Of Fundy, Scotian Shelf and Bras D'Or Lakes, Canadian Technical Report of Fisheries and Aquatic Sciences 2226. Science Branch, Department of Fisheries and Oceans, Dartmouth.

Davies, C.M., J.A.H. Long, M. Donald, and N.J. Ashbolt. 1995. Survival of Fecal Microorganisms in Marine and Freshwater Sediments. Applied and Environmental Microbiology. 61: 1888-1896.

Dill, M. 2003. Annapolis River Guardians Volunteer Water Quality Monitoring Program. 2002 – 2003 Annual Report. Clean Annapolis River Project.

Dodds, W.K, and E.B. Welch. 2000. Establishing Nutrient Criteria in Streams. Journal of the North American Benthological Society. 19(1): 186-196.

Dodds, W.K., Jones, J.R., and E.B. Welch, 1998. Suggested Criteria for Stream Trophic State: Distributions of Temperate Stream Types by Chlorophyll, Total Nitrogen and Phosphorus. Water Research. 31: 1738-1750.

Dodds, W. and Whiles, M., 2010. Freshwater Ecology. 2nd Edition, Elsevier publishing/Academic Press, San Diego, California.

Edberg, S.C., E.W. Rice, R.J. Karlin and M.J. Allen. 2000. Escherichia coli: the best biological drinking water indicator for public health protection. The Society for Applied Microbiology. 88: 106-116.

Environment Canada. 2010. Canadian Aquatic Biomonitoring Network Field Manual. (T. Paull, Ed.) National Water Research Institute, Environment Canada.

Freeman, L. 2012. Annapolis River 2011 Annual Water Quality Monitoring Report. Clean Annapolis River Project.

Freeman, L. 2013. Annapolis River 2012 Annual Water Quality Monitoring Report. Clean Annapolis River Project

Glenen, J., A. Sharpe. January 2010. Annapolis River 2009 Annual Water Quality Monitoring Report. Clean Annapolis River Project.

Glozier, N. E., R. W. Crosley, L. A. Mottle, D. B. Donald. 2004. Water Quality Characteristics and Trends for Banff and Hasper National Parks: 1973-2002. Environmental Conservation Branch, Ecological Sciences Division, Prairie and Northern Region.

Health Canada. 1992. Guidelines for Canadian Recreational Water Quality. Retrieved June 15, 2011, from http://www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/guide_water-1992-guide_eau/section3-eng.php#sec3_1_1.

Health Canada. 2010. Guidelines for Canadian Drinking Water Quality – Summary Table. Retrieved June 15, 2011, from http://www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/2010-sum_guide-res_recom/index-eng.php#a8.

Health Canada. 2012. Guidelines for Canadian Recreational Water Quality, Third Edition. Retrieved May 7, 2013, from http://www.hc-sc.gc.ca/ewh-semt/pubs/water-eau/guide_water-2012-guide_eau/index-eng.php.

Helsel, D. R., R. M. Hirsch. 2002. U.S. Geological Survey, Techniques of Water-Resources Investigations Book 4. Chapter A3: Statistical Methods in Water Resources. U.S. Department of the interior, United States Geological Survey. (http://water.usgs.gov/pubs/twri/twri4a3/)

Helsel, D. R., D. K. Mueller, J. R. Slack. 2006. Computer Program for the Kendall Family of Trend Tests. U.S. Department of the interior, United States Geological Survey. (http://pubs.usgs.gov/sir/2005/5275/pdf/sir2005-5275.pdf)

Hirsch, R. M., R. B. Alexander, R. A. Smith. 1991. Selection of Methods for the Detection and Estimation of Trends in Water Quality. Technical Memorandum. (http://water.usgs.gov/admin/memo/BSA/BSA91.01.pdf)

IDEXX Quanti-Tray®/2000 MPN Table (per 100mL) with 95% Confidence Limits (No date). Taken from the IDEXX website, accessed January 14, 2009. (http://www.idexx.com/water/refs/qt2k95.pdf)

Ironside, G., 2001. Nutrients In The Canadian Environment: Reporting on the State of Canada's Environment. Indicators and Assessment Office, Environment Canada.

Janssen, M. 2010. Chapter 4: Primer on Temporal Autocorrelation. In J.D. Ackerman (Ed). Spatial and Temporal Analysis in Ecology: A Primer. University of Guelph, Guelph, ON.

Jessop, B.M., Physical and biological survey of the Annapolis River, 1975, Freshwater and Anadromous Division Resource Branch, Fisheries and Marine Service, Department of Environment, Data Record Series No. Mar/D-76-8, 1976.

Mackie, G., 2004, Applied Aquatic Ecosystem Concepts. 2nd Edition, Kendall/Hunt Publishing Company, Dubuque, Iowa.

MacMillan, JL., D. Cassie, J.E. LeBlanc, T.J. Crandlemere. 2005. Characterization of water temperature for 312 selected sites in Nova Scotia. Canadian Technical Report of Fisheries and Aquatic Sciences 2582.

Meko, M. 2011. GEOS 585A, Applied Time Series Analysis. University of Arizona, Tucson, AZ. Accessed January 2012. (http://www.ltrr.arizona.edu/~dmeko/geos585a.html)

Nova Scotia Environment. 2009. Nova Scotia Groundwater Observation Well Network. Accessed February 2010. (http://www.gov.ns.ca/nse/groundwater/docs/GroundwaterObservationWellNetwork2009Report.pdf)

OMEE – Ontario Ministry of Environment and Energy, 1994, as cited in P. Chambers 2001, p. 145.

Pittman S. and R. Jones. 2001. Annapolis River Guardians Volunteer Monitoring Program. Unpublished.

Postma, C. February 2011. Annapolis River 2010 Annual Water Quality Monitoring Report. Clean Annapolis River Project.

Reynoldson, T.B., C. Logan, T. Pascoe, S.P. Thompson. 2002. CABIN (Canadian Aquatic Biomonitoring Network) Invertebrate Biomonitoring Field and Laboratory Manual. National Water Research Institute, Environment Canada.

Sharpe, A. March 2007. Report on the Investigation of Low Dissolved Oxygen Levels in the Annapolis River Estuary. Clean Annapolis River Project.

Sharpe, A. March 2008. Annapolis River 2007 Annual Water Quality Monitoring Report. Clean Annapolis River Project.

Sharpe A. and D. Sullivan. March 2004. Aylesford East Baseline Research Project: Summary Report of Findings. Clean Annapolis River Project.

Sharpe A. and D. Sullivan. 2006. CARP Quality Assurance/Quality Control Project Plan.

Valiela, I., Alber, M. and M. LaMontagne. 1991. Fecal Coliform Loadings and Stocks in Buttermilk Bay, Massachusetts, USA, and Management Implications. Enviromental Management. 15: 659-674.

Wcisło, R. and R.J. Chróst. 2000. Survival of Escherichia coli in Freshwater. Polish Journal of Environmental Studies. 9: 215-222.

6.0 Appendices

A.0 Appendix A

A.1 Parameters Tested and Methodologies

Table A1. Current and previous parameters measured throughout the program.

Parameters Analyzed in 2021	Additional Parameters Analyzed in Previous Years of the Program
E. coli bacteria densities	Salinity
Dissolved Oxygen	Chlorophyll a
Temperature (Water and Air)	Nitrate-N, Nitrite, Chloride, Sulphate, Total Phosphate
Weather conditions	Colour
pH, Conductivity, Total Dissolved Solids	Transparency
Total Nitrogen, Total Phosphorus	
Turbidity	

A.1.1 Water Collection for *E. coli* Bacteria Analysis

Following the contamination of some sampling equipment in 2003, a new collection procedure for fecal coliform samples was developed in 2004 and has been in place since. The sampling units (Figure A1) allow for representative sampling from mid-span of bridges at the sampling sites.

Figure A1. Collection unit used for *E. coli* samples in 2012.

The open sample bottle is secured in the clamp, and lowered from the mid-span of the bridge into the river, to a depth of 1 meter. Samples are collected on the upstream side of bridges, where a safe pedestrian walkway exists. After collection, water samples are refrigerated until delivery to the lab, typically within 24 hours of collection.

A.1.2 Enumeration of *E. coli* Bacteria

Prior to the 2005 season, bacterial samples collected by Clean Annapolis River Project's Annapolis River Guardians program were tested for Fecal Coliforms (FC) using the membrane filtration method. During the winter of 2005, the program's Science Advisory Committee suggested that the program switch to testing for *E. coli* (EC) using the Most Probable Number method (used in the Valley Regional Hospital), to bring testing more in line with national guidelines. In order to ensure the continuity of the dataset, a period of duplicate analysis with the two methods was conducted. Duplicate samples were analysed using both methods over a two-month period (four biweekly sample events at eight locations along the river). Analysis of the paired results indicated no significant difference between the two testing methods.

All *E. coli* bacteria samples are submitted to the Valley Regional Hospital Microbiology Laboratory in Kentville, Nova Scotia. The Valley Regional lab is accredited by Nova Scotia Environment to perform bacterial water quality analysis. From 1997 to 2003 and again since 2005, fecal bacteria densities were determined using the IDEXX Colilert procedure, to give a Most Probable Number of *E. coli* bacteria present. For the 2021 sample season, analysis was performed using the IDEXX Colilert procedure.

A.1.3 Water Chemistry

All 2021 water chemistry parameters were collected by CARP staff using a YSI professional plus water quality probe. Measurements are taken at a depth of 1m. This has been the standard methodology since the volunteer program was ceased in 2014. Detailed field methodology from the volunteer program can be found in past River Guardians reports.

A.1.4 Trend Analysis

Before any trend analyses were performed, outlier tests were conducted. The mean and standard deviation of a particular data set were calculated and each value was compared to the mean. If any value differed from the mean by more than twice the standard deviation, it was considered an outlier and was checked against the original data sheets. If there was reason to suspect the data point of being invalid, the data was not included in the trend analysis. If no notes or calculation errors were made on the original data sheet, the outlier value was retained in the data set. The analysis for the temperature data was performed only on data from the summer months (July, August and September), as elevated water temperatures that occur in the summer months are the principal concern.

The outlier analysis was not performed on the bacteria data, as the nature of the data is not conducive to outlier analysis. The data is highly variable with a wide range of 0 to 2419 cfu/100 mL and is capped at 2419 cfu/100 mL. The cap of 2419 cfu/100 mL is due to method limitations; the IDEXX Colilert testing method will not produce a reading greater than this number. Some of the earlier data was analyzed using a different method that was not capped, so any data point above the 2419 cfu/100 mL threshold was artificially capped at 2419 cfu/100 mL for consistency purposes.

A.1.4.1 Non-Parametric Analysis

In the past, a box and whisker plot was made for each parameter, with the data grouped by month, and the box plots were then visually assessed for similarities across months. Adjacent months with similar medians and ranges were grouped together as a season (Figure A2). This was not conducted in 2021because the corresponding R code for producing the boxplots and conducting seasonal Kruskal-Wallis analyses were not available.

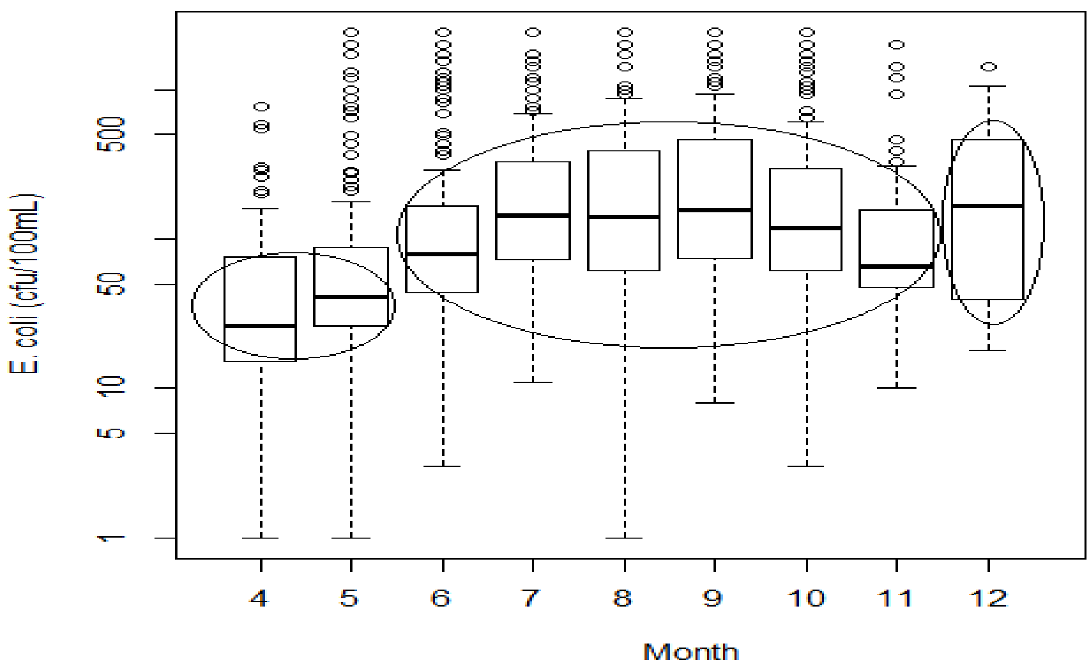

Figure A2. Bacteria count data for all years grouped by month. The circles indicate the seasons that were determined from this plot. There was very little data for the January to March period; these months were not used in the analysis. A 'dummy season' containing no data was used in the analysis to represent the January to March period.

Three seasons were indicated by the bacteria count box plot shown above and a fourth season was included in the analysis to represent the January to March months. The bacteria data was grouped according to these seasons and the Kruskal-Wallis for seasonality test was performed. Bacteria count data was indicated as being seasonal, while the pH, DOSAT and summer temperature data were not. Based on this, the Seasonal Kendall test was performed on the bacteria count data and the Mann-Kendall test was performed on pH, DOSAT and temperature data. These tests produce a linear trend equation and a probability statistic (p-value), which indicates whether or not the trend is statistically significant. A trend was considered significant if the p value was less than 0.05. Non-parametric analyses were not performed on the nutrient data as there was not enough data to assess the seasonality of the data set.

A.1.4.2 Parametric Analysis

The data was grouped by parameter and location, and the Shapiro-Wilks test was performed on each data set. The Shapiro-Wilks test is a test for non-normality and produces a histogram of the data overlaid with a normal distribution curve as well as some significance and probability statistics. For this procedure, the histogram and normal curve are examined to determine whether the data visually resembles a normal distribution. If the data does not resemble a normal distribution (in the case of *E. coli*), the data set can be transformed until it resembles a normal distribution. CARP's *E. coli* data distribution resembled a logarithmic distribution, so the data was transformed by taking the base-10 logarithm of the bacteria results. The logarithmic transformation produced a normally distributed data set (Figure A3).

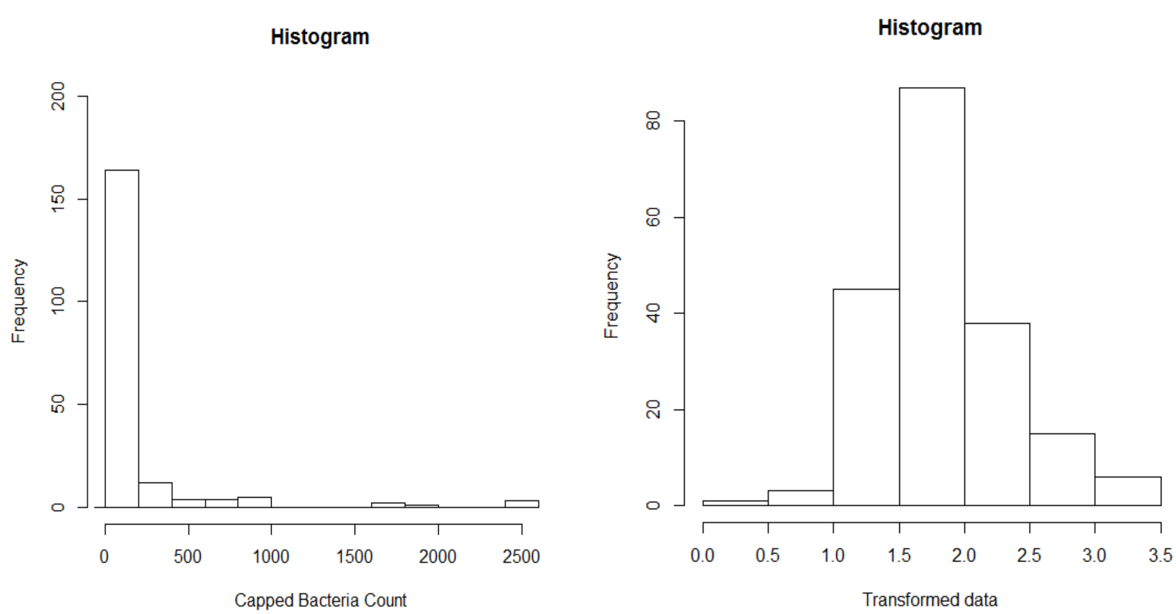

Figure A3. Lawrencetown (#35) bacterial count data distribution before transformation (left) and after transformation (right).

The transformed data much more closely resembles a normal distribution and can be used for the regression analysis. The data for DO, temperature and pH did not require transformation to resemble a normal distribution.

After normality was established for each parameter, a linear regression was performed on its data set. This produced a linear slope of the trend, as well as a confidence interval, prediction interval, probability value and residual histogram. The trend slope provides the rate of change of the variable by year, the confidence interval and probability value allow for the determination of statistical significance of the trend and the residual plot and histogram indicate whether the data set varies in a non-linear fashion, which would indicate that the linear regression calculation is not appropriate for the data set. For the determination of statistical significance, three tests were performed. If any of these tests were failed, the trend was not considered significant. The three tests included:

- verification of the slope's p value. If the value was less than 0.05, this test was passed.
- examination of the confidence intervals of the regression plot. If the confidence interval range at the beginning of the data set overlaps with the range at the end of the data set, this test was passed (Figure A4).
- examination of the Scatterplot and distribution. If the scatterplot appeared to the randomly distributed and the associated histogram resembled a normal distribution, this test was passed.

As an example, the DOSAT data for the Kingston location is displayed below. The p value for the slope produced by the regression analysis was <0.0001. This value is less than the 0.05 threshold, therefore, the data passed this significance test. Figures A4 and A5 below show that the Kingston data set passed the other two significance tests as well, therefore the trend slope of -0.43 %/year was accepted as significant. This indicates that dissolved oxygen levels are decreasing at the Kingston location.

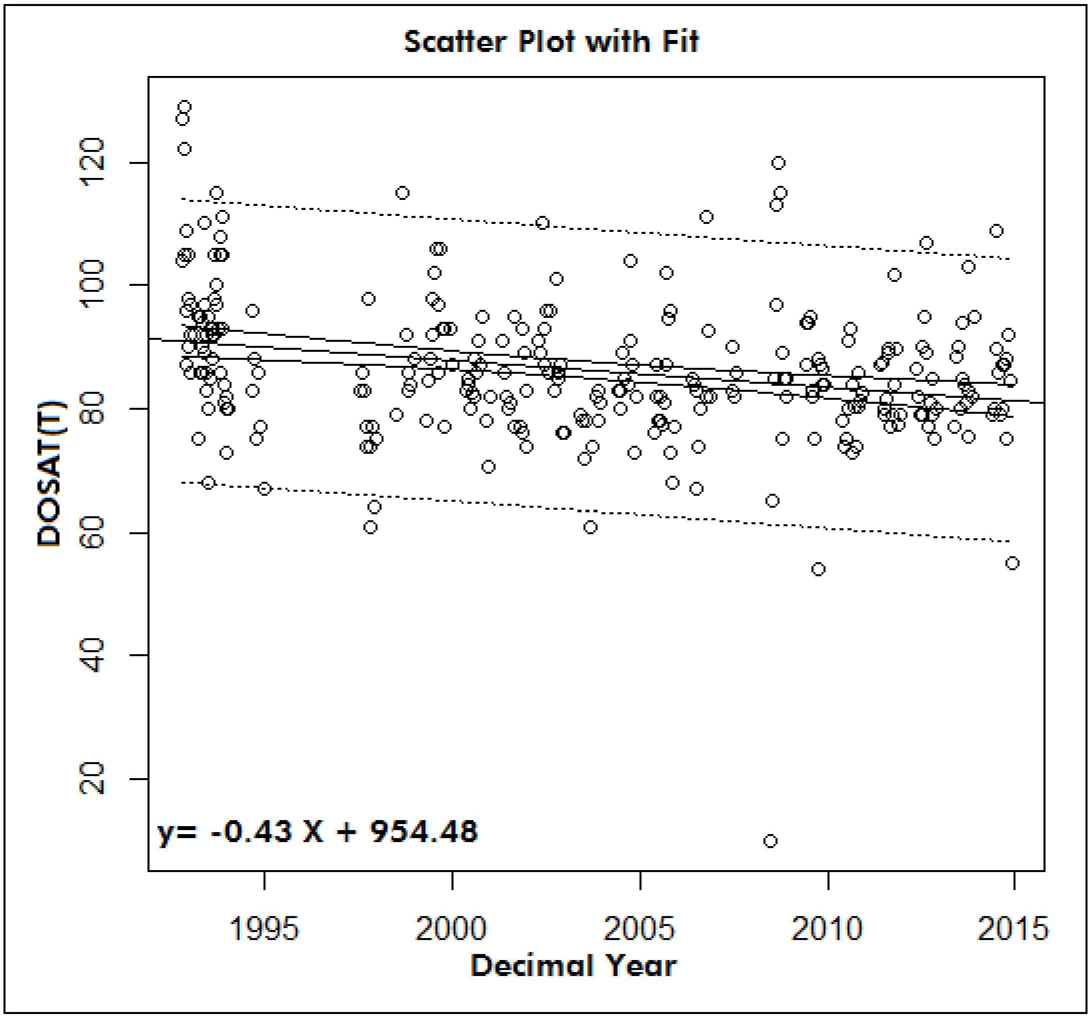

Figure A4. Linear regression for DOSAT data at the Kingston location. The curved line represents the 95% confidence interval range at the beginning and end of the dataset.

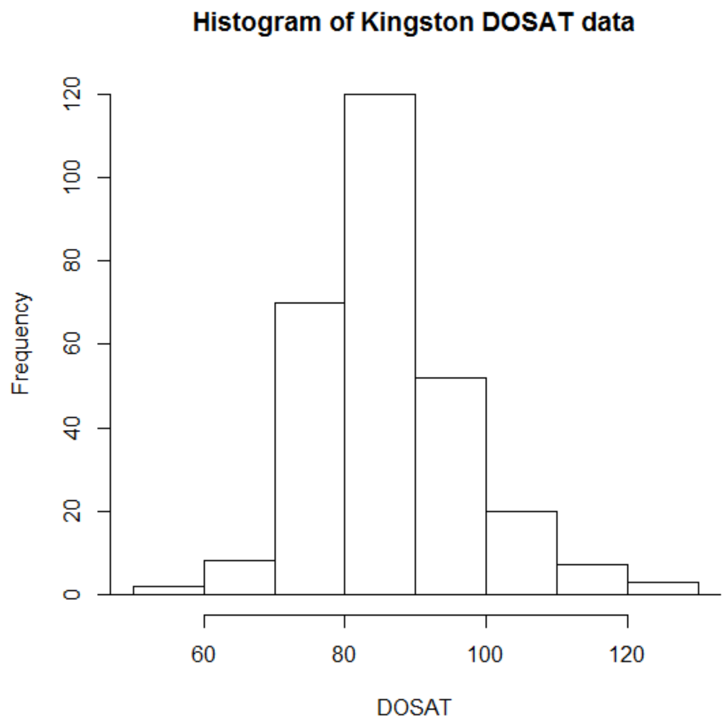

Figure A5. Histogram of Kingston DOSAT data. Data appears to be normally distributed. Since the plot is also randomly distributed, this significant test is passed.

Autocorrelation and Serial Dependence

Autocorrelation is an important consideration for both parametric and non-parametric statistical trend analyses (Helsel and Hirsch, 2002) because its existence invalidates most statistical tests, as they assume data points to be independent and uncorrelated to one another. Autocorrelation refers to the correlation of a set of data points across either space or time. If a set of data displays temporal autocorrelation, (a.k.a. their data points, when separated by a unit of time, known as a lag, demonstrate a correlation) they are said to show serial dependence (Australian and New Zealand Environment and Conservation Council, 2000). For example, if a turbidity sample was taken during a storm event, and then another taken a few hours later, the likelihood of the readings for both samples being affected by this event is high, and so the sample values are not independent from one another. To test for autocorrelation, a data series is plotted against a time lagged version of itself and the correlation value between the data points measured. These values are then plotted on an Autocorrelation plot (Figures A6 and A7) for each lagged unit of time, and compared against a 95% confidence interval to test for serial dependence. (Meko, 2011; Janssen, 2010). Significant serial dependence is indicated when the vertical bars extend beyond the 95% confidence curves.

The linear regression fit assumes that there must be no correlation between data points. In the case of water quality data, the potential existed for data points collected temporally close or along the same stretch of river to be correlated. To assess whether the data was affected by this serial dependence, an autocorrelation plot for each variable at each location was performed (Figure A6), as well as for the entire data set for each parameter (Figure A7). In the Paradise plot, most of the bars do not extend beyond the confidence interval, thus serial dependence is not indicated. When an autocorrelation plot was made for all locations, significant serial dependence was displayed; therefore a trend analysis was not performed on the data for all locations (Figure A7). Autocorrelation analyses were not conducted in 2021 because the corresponding R codes were not available.

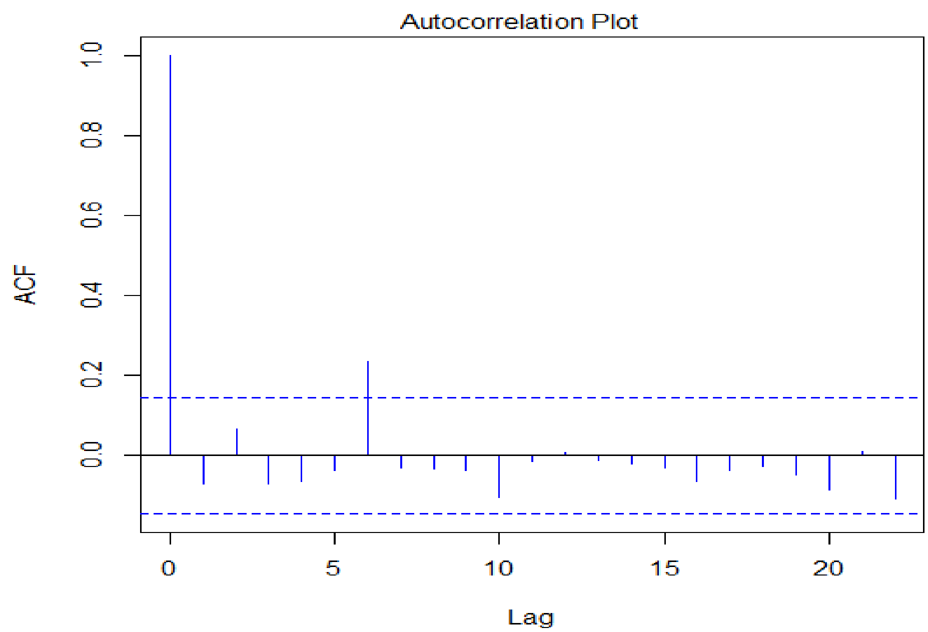

Figure A6. Autocorrelation plot for E. coli at the Paradise site.

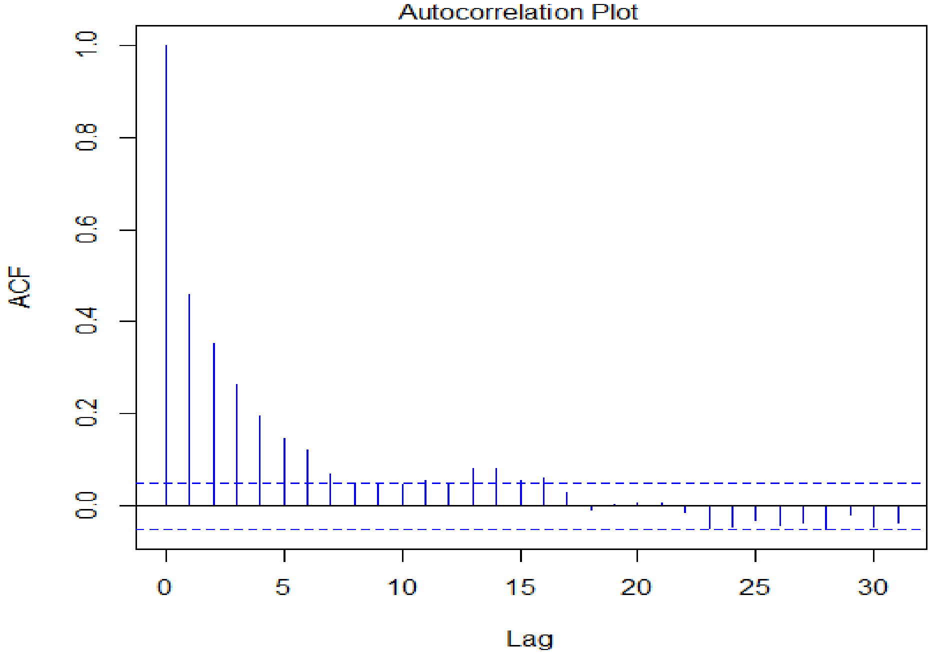

Figure A7. Autocorrelation plot for the entire E. coli dataset.

B.0 Appendix B

B.1 Sites Monitored

Water samples were collected during 2012 by the Annapolis River Guardians program at several different locations (Table B1). Coordinates are reported in latitude and longitude, as recorded on a hand-held GPS unit.

Table B1. Coordinates and descriptions for Annapolis River Guardian and TSS/turbidity sample locations.

Site Code	Latitude	Longitude	Site Name	Site Name (Long with Reference Points)
60	N45 01.699	W64 48.617	Aylesford Road	Bridge at Aylesford Rd, near Hwy 1
Ref	N45 00.122	W64 49.381	Millville	Bridge on Victoria Rd, South Annapolis River
00	N45 01.606	W64 50.148	Aylesford	Bridge on Victoria Rd, near Hwy 1
13	N44 58.713	W64 56.663	Kingston	Bridge on Bridge St. near Stronach Park
18	N44 57.199	W65 00.096	Wilmot	Bridge on Old Mill Road
NS01	N44 56.942	W65 01.769	Wilmot	Bridge on Bayard Road
25	N44 56.213	W65 03.969	Middleton	Bridge on Hwy 10, near Riverside Park
35	N44 52.850	W65 09.476	Lawrencetown	Bridge on Lawrencetown Lane
40	N44 52.045	W65 12.384	Paradise	Bridge on Paradise Lane
49	N44 50.335	W65 17.492	Bridgetown	Bridge on Queen Street

The NS01 and Ref sites were sampled for nutrients by Environment Canada.

End of Report.