Annapolis River 2010 Annual Water Quality Monitoring Report

including results from the

Annapolis River Guardians Volunteer Water Quality Monitoring Program

Prepared By:
Chelsae Postma, February 2011

Clean Annapolis River Project
P.O. Box 395,
151 Victoria Street, Annapolis Royal, Nova Scotia
902 532 7533; carp@annapolisriver.ca;
www.annapolisriver.ca

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Acknowledgements

River Guardians is a volunteer-based program that would not have been possible without the dedication of community members throughout the past 19 years. We would like to extend our deepest thanks and appreciation to the volunteers of 2010 who have contributed to the success of the project. The Annapolis River Guardians include:

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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
The Acadia Centre for Estuarine Research, Acadia University
Human Resources and Skills Development Canada

Executive Summary

In 2010, the Annapolis River Guardians completed their 19th year of continuous water quality monitoring on the Annapolis River. Eight volunteers monitored eight sites over the course of the season, which ran from April to November. In 2008, total suspended solids and turbidity were added to the suite of parameters monitored. Others include dissolved oxygen, *E. coli* bacteria, air and water temperature, pH and conductivity, as well as local weather conditions.

E. coli bacteria levels along the Annapolis River during 2010 were similar to those observed in 2009, with 2010 medians being slightly higher at some locations. Both years experienced similar precipitation amounts. Sampling events from May through September often coincided with significant rainfall events, possibly contributing to the elevated overall bacteria counts. As in previous years, *E. coli* counts increased markedly between the sampling stations at Aylesford Road and Victoria Road, indicating the introduction of fecal material between these two locations. In 2009, some additional sampling was performed between these two stations. The results were inconclusive due to the variability of the testing method and can be found in the 2009 River Guardian Report.

Over the 19 years of monitoring, mean dissolved oxygen saturation (DOSAT) levels have remained in the range of 80-94%. In 2010, the mean DOSAT level was 84% compared to 85% in 2009.

The mean summer water temperature for the Annapolis River during 2010 was 19.0ºC, 1.2ºC warmer than for the same period in 2009. As in previous years, water temperatures during the 2010 summer months continued to reach levels stressful to aquatic life ($>$20ºC).

The pH levels at each of the River Guardian sites were consistently within the recommended range for the protection of aquatic life (6.5-9.0). Mean pH values for the eight monitoring locations along the Annapolis River ranged between 6.97 and 7.12.

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 is used as the maximum concentration for preserving aquatic health and 2.9 mg nitrates-nitrogen/L (CCME, 2003) is used as the guideline for reporting nitrates. While elevated total nitrogen results were observed, phosphorus remains a significant concern. During the 2006 to 2010 period, 11% of total nitrogen results exceeded 0.9 mg/L while 41% of total phosphorus results exceeded the suggested guideline level of 0.030 mg/L. 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.

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. This sampling was part of a two-year effort to establish a baseline for turbidity and TSS in the Annapolis River and to develop a numerical relationship between these two parameters. In 2010, 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.

CARP has collected benthic invertebrate samples in the Annapolis River watershed since 2002, using the protocol developed through the Canadian Aquatic Biomonitoring Network (CABIN). A total of three sites are monitored in the Annapolis River, two on the main stem of the river at Paradise and Wilmot, and one on the Southern branch at Millville. The site in Millville is used as a reference site as there are minimal human impacts at this site. There has been no significant change in the Family Biotic Index at the Paradise location over the

period of 2005 to 2009. For the Wilmot location, the Family Biotic Index value increased from 2008 to 2009 indicating a decrease in water quality. The results for 2010 CABIN monitoring had not been processed at the time of writing and were not included in this report.

As part of CARP's Quality Assurance Project Plan, regular quality control samples were collected. The accuracy of River Guardian dissolved oxygen readings were estimated at +/- 0.41 mg/L, compared with +/- 0.38 mg/L recorded in 2009. Travel and field blank samples, collected to check for cross contamination, consistently had *E. coli* counts of 0 cfu/100mL. *E. coli* split samples had a Relative Percent Difference of 25% compared to 29% in 2009.

Introduction

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 is one of the longest running and most extensive volunteer based water quality programs in Eastern Canada. It is also CARP's longest running and only ongoing project. At least 90 volunteers from the Annapolis Valley community have participated in the program over the years, with over 3600 water samples being 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 has been maintained over the past years and eight sites remain actively monitored.

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.
- To develop interest in the Annapolis River and community stewardship to ensure a viable resource for future generations.
- To provide a knowledgeable group of local individuals who can promote the preservation, rehabilitation, and use of these aquatic resources in the future.

Overview of 2010 Monitoring Season

Sample collection for the 2010 season ran from May 2nd to November 1st on a biweekly basis. The parameters monitored were *E. coli* bacteria, dissolved oxygen content, water temperature, air temperature, pH, conductivity, total suspended solids (TSS) and turbidity. TSS and turbidity were introduced in 2008 and continued throughout 2009 and 2010. They were added as 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. The purpose of this was to allow for the calculation of the water quality index (WQI) for the Annapolis River, which would be useful in the annual reporting of the data.

Eight stations were sampled along the Annapolis River. Further information on these sampling locations is contained in Appendix B. The monitoring sites for 2010 were all within the freshwater portion of the Annapolis River (Figure 1). The data collected by the volunteers is stored in a Microsoft Access database at the CARP office.

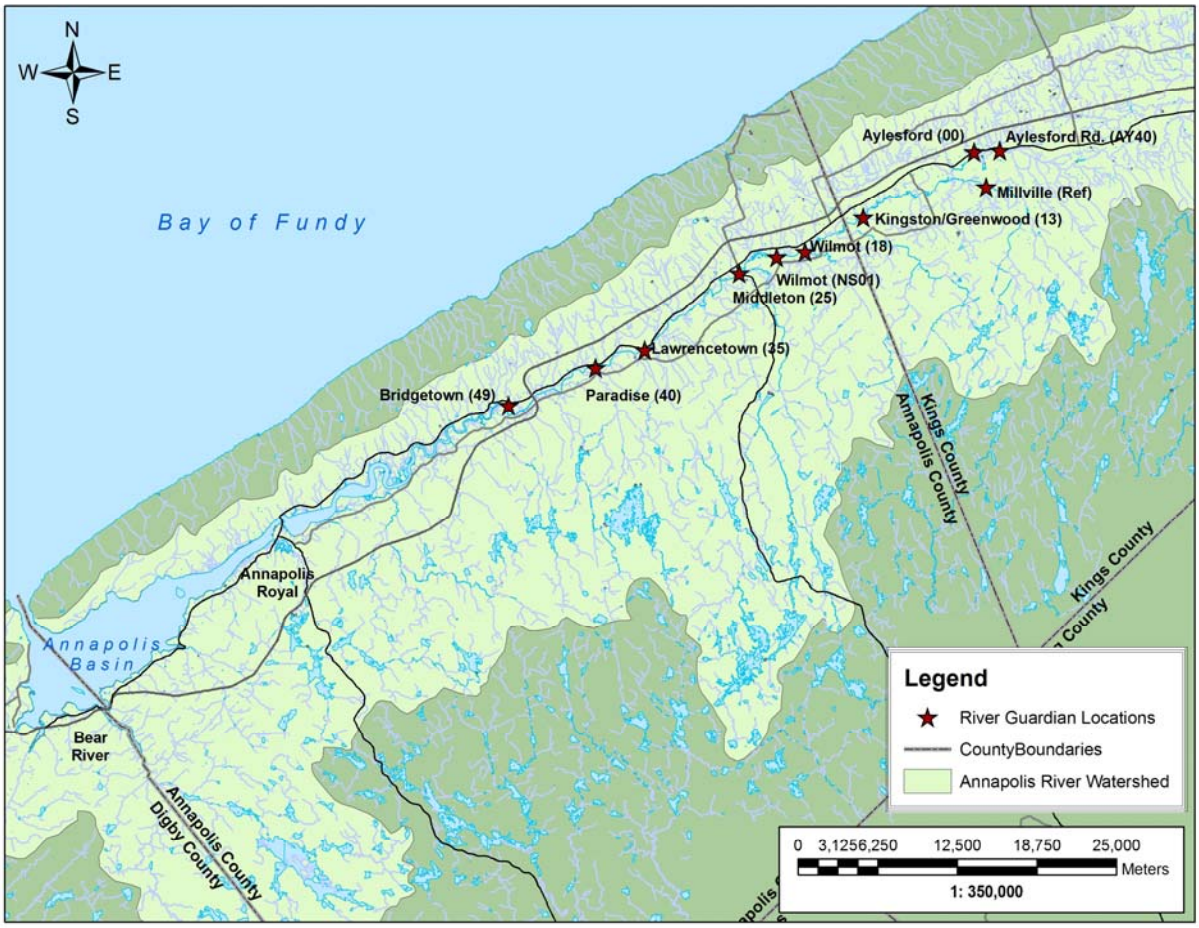

Figure 1. Annapolis River watershed with 2010 River Guardian monitoring sites identified by stars. Sites NS01 and Ref, which were used for nutrient and turbidity/TSS monitoring are also shown on this map.

The 2010 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), each location has a large River Guardians sign (Figure 2) that indicates *E. coli* contamination and overall water quality trends for that location. The signs are updated by the volunteers every two weeks and are on display from May through to November.

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. Site NS01 was also used for TSS/Turbidity sampling.

As part of CARP's Quality Assurance/Quality Control (QA/QC) plan (Sharpe and Sullivan, 2006), additional samples were taken to ensure good data quality. The QA/QC measures taken are detailed in Appendix C.

Figure 2. River Guardian's sign displaying the date, latest bacteria count and overall water quality trend.

Recommendations for CARP:

- Complete the Quality Assurance Project Plan for all of CARP's Water Quality monitoring programs.
- Ensure QA/QC protocols are implemented yearly throughout the entire sampling season, including an information session before the first sampling date.
- Create a manual for the River Guardian facilitator to ensure consistency in analysis and reporting.
- Review and update the Annapolis River Guardian Procedures Manual.

2010 Monitoring Results

E. coli Bacteria

Introduction

Escherichia coli (*E. coli*) are rod-shaped, aerobic, lactose fermenting bacteria. They are Gram-stain negative, thermotolerant and appear as dark blue colonies when cultured in the laboratory. The predominant sources of *E. Coli* bacteria in the 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 of *E. coli* in rivers. These include the type of 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, amount of sunlight, salinity of the water, temperature, as well as composition and abundance of sediment (Davies *et al.,* 1995). There is 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)

Over the period of 1992 to 2009, numerous initiatives have been 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 2009, the Town of Middleton commenced construction of a new sewage treatment plant.

While the core River Guardian monitoring program has been maintained over the period of 1992 to 2010, a number of modifications have been made. For example, in 1996, the collection of *E. coli* samples was standardized to every two weeks. During the period of 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.

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 fecal bacteria contamination into a single table for public awareness purposes (Table 1).

Table 1. Summary of water quality guidelines for fecal coliforms.

cfu*/100ml	Water Use	Explanation/Source
0	Acceptable for drinking	fecal coliforms/100ml. (Health Canada, 2010)
1-50	Acceptable for livestock watering	Interpretation of CCME narrative "high-quality water given to livestock" (total coliforms).
50-100	Acceptable for food crop irrigation	Tentative Maximum Concentration. CCME Guidelines (fecal coliform bacteria/100ml).
100-200	Acceptable for recreational use	Geometric Mean of 5 samples taken during a period not to exceed 30 days, should not exceed 200 cfu/100 ml. (Health Canada, 1992)
>200	Unacceptable for human contact	
*cfu = colony forming units		

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 analysed 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 fecal coliform 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 coliform 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 2009 and 2010 *E. coli* results (Figure 3). The box plot shows the 25th and 75th percentiles as well as the median for each site. The minimum and maximum results are also shown. Note that the y-axis of the graph is plotted using a logarithmic scale (Log *E. coli*) and that the data is artificially capped at 2420 cfu/100mL, as this is the maximum possible value with the IDEXX Colilert testing system. From 1992 to 2010, approximately 2% of the data have exceeded this cap value.

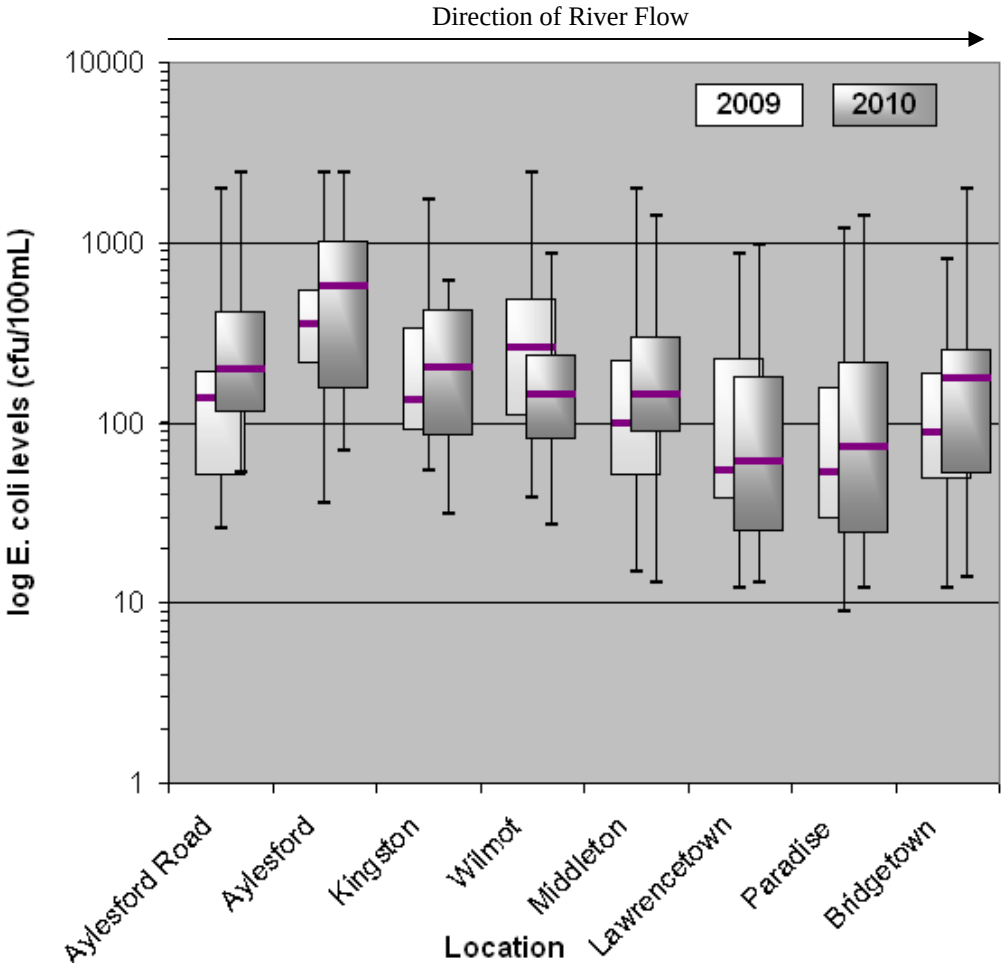

Figure 3. Box and whisker plot of Annapolis River Guardian *E. coli* bacteria results for 2009 and 2010.

In 2010, the median *E. coli* values for seven of the eight sites were higher than in 2009. Wilmot was the only site with a lower median in 2010 than 2009. Aylesford, Kingston, Lawrencetown, Paradise, and Bridgetown all showed greater variability this season, while Wilmot and Middleton showed less. Aylesford Road portrayed a similar range to 2009. 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 fall within each of the ranges specified in Table 1 (Tables 2 through 9). This allows easy visualization of how the *E. coli* readings have fluctuated for each station since CARP began monitoring the Annapolis River. All of the *E. coli* ranges are in units of cfu/100mL.

Table 2. *E. coli* percentages for Aylesford Road.

Year	% in 0 - 50	% 51 - 100	% 101 - 200	% >200
1992				
1993				
1994				
1995				
1996				
1997				
1998				
1999				
2000				
2001				
2002				
2003	20	40	20	20
2004				
2005	33	13	27	27
2006	29	6	6	59
2007	20	20	33	27
2008	8	23	38	31
2009	29	14	36	21
2010	0	23	31	46

Table 3. *E. coli* percentages for Aylesford.

Year	% in 0 - 50	% 51 - 100	% 101 - 200	% >200
1992	0	0	50	50
1993	9	9	27	55
1994	17	17	17	50
1995	67	0	17	17
1996	62	0	0	38
1997	14	14	29	43
1998	15	8	23	54
1999	9	18	27	45
2000	40	0	20	40
2001	25	19	31	25
2002	6	11	33	50
2003	16	16	58	11
2004	6	0	24	71
2005	29	7	7	57
2006	8	23	8	62
2007	6	6	12	76
2008	0	23	8	69
2009	7	14	0	79
2010	0	21	7	71

Table 4. *E. coli* percentages for Kingston.

Year	% in 0 - 50	% 51 - 100	% 101 - 200	% >200
1992	67	33	0	0
1993	21	21	36	21
1994	33	17	0	50
1995	86	0	0	14
1996	50	19	6	25
1997	19	38	31	13
1998	27	27	27	18
1999	35	18	18	29
2000	40	20	33	7
2001	24	29	18	29
2002	39	28	17	17
2003	13	13	40	33
2004	7	14	43	36
2005	33	7	33	27
2006	7	29	14	50
2007	14	29	14	43
2008	15	0	46	38
2009	0	29	43	29
2010	7	21	21	50

Table 5. *E. coli* percentages for Wilmot.

Year	% in 0 - 50	% 51 - 100	% 101 - 200	% >200
1992	0	33	0	67
1993	19	13	19	50
1994	13	0	31	56
1995				
1996				
1997	28	11	44	17
1998	60	30	10	0
1999	31	25	19	25
2000	50	17	17	17
2001	25	31	25	19
2002	29	35	12	24
2003	20	47	13	20
2004	0	21	57	21
2005	27	7	60	7
2006	21	36	14	29
2007	27	27	27	20
2008	23	8	54	15
2009	15	8	23	54
2010	21	7	36	36

Table 6. *E. coli* percentages for Middleton.

Year	% in 0 - 50	% 51 - 100	% 101 - 200	% >200
1992	0	33	0	67
1993	14	14	43	29
1994	9	9	27	55
1995				
1996	40	10	20	30
1997	13	25	50	13
1998	50	0	25	25
1999	50	8	25	17
2000	60	20	7	13
2001	41	18	24	18
2002	65	29	6	0
2003	36	29	14	21
2004	15	23	38	23
2005	53	20	13	13
2006	43	21	7	29
2007	20	27	27	27
2008	14	36	21	29
2009	29	21	21	29
2010	21	14	36	29

Table 7. *E. coli* percentages for Lawrencetown.

Year	% in 0 - 50	% 51 - 100	% 101 - 200	% >200
1992	0	33	33	33
1993	7	14	21	57
1994	24	6	41	29
1995	43	0	29	29
1996	13	13	33	40
1997	29	36	29	7
1998	42	25	25	8
1999	40	30	30	0
2000	53	20	7	20
2001	56	25	13	6
2002	50	11	17	22
2003	53	20	7	20
2004	21	29	21	29
2005	47	33	20	0
2006	40	7	13	40
2007	57	14	7	21
2008	54	23	8	15
2009	50	14	7	29
2010	50	7	14	29

Table 8. *E. coli* percentages for Paradise.

Year	% in 0 - 50	% 51 - 100	% 101 - 200	% >200
1992	0	0	67	33
1993	14	14	36	36
1994	14	29	0	57
1995	63	0	13	25
1996	29	18	12	41
1997	50	36	7	7
1998	22	44	22	11
1999	42	25	25	8
2000	33	17	8	42
2001	35	18	29	18
2002	59	6	18	18
2003	40	20	27	13
2004	14	21	21	43
2005	36	36	21	7
2006	33	7	13	47
2007	53	27	7	13
2008	54	23	15	8
2009	43	21	14	21
2010	36	29	7	29

Table 9. *E. coli* percentages for Bridgetown.

Year	% in 0 - 50	% 51 - 100	% 101 - 200	% >200
1992				
1993				
1994	14	21	21	43
1995	44	11	22	22
1996	29	18	18	35
1997	35	12	47	6
1998	44	38	13	6
1999	33	22	28	17
2000	60	27	13	0
2001	71	18	0	12
2002	41	35	12	12
2003	33	27	13	27
2004	14	7	50	29
2005	40	47	7	7
2006	27	20	20	33
2007	53	13	0	33
2008	50	29	7	14
2009	29	29	14	29
2010	29	14	14	43

There does not appear to be an indicative trend for *E. Coli* as the values at all sites are quite variable (Tables 2 – 9). The percentage of samples that fell into the range 0-50 cfu/100 mL increased for Kingston and Wilmot, remained the same for Lawrencetown and Bridgetown, and decreased for Aylesford Road, Aylesford, Middleton and Paradise. For the range 51-100 cfu/100 mL, percentages increased in Aylesford Road, Aylesford, and Paradise, and decreased in Kingston, Wilmot, Middleton, Lawrencetown, and Bridgetown. For the range 101-200 cfu/100 mL, Aylesford, Wilmot, Middleton, and Lawrencetown had more values fall into this range than in 2009, Bridgetown had the same, and Aylesford Road, Kingston, and Paradise had less. The percentage of values with >200 cfu/100 mL of *E. Coli* increased for Aylesford Road, Kingston, Paradise, and Bridgetown, remained the same for Middleton and Lawrencetown, and decreased for Aylesford and Wilmot.

The percentage of samples falling into the >200 cfu/100mL and 101-200 cfu/100mL category increased in 2010 when compared to 2009, while the other two categories showed fewer results. This is not entirely unexpected as the two years are quite similar; having approximately the same precipitation amounts. The percentage of data falling into each of these categories for all locations was compiled (Figure 4) as well as the number of samples taken in each year (Table 10).

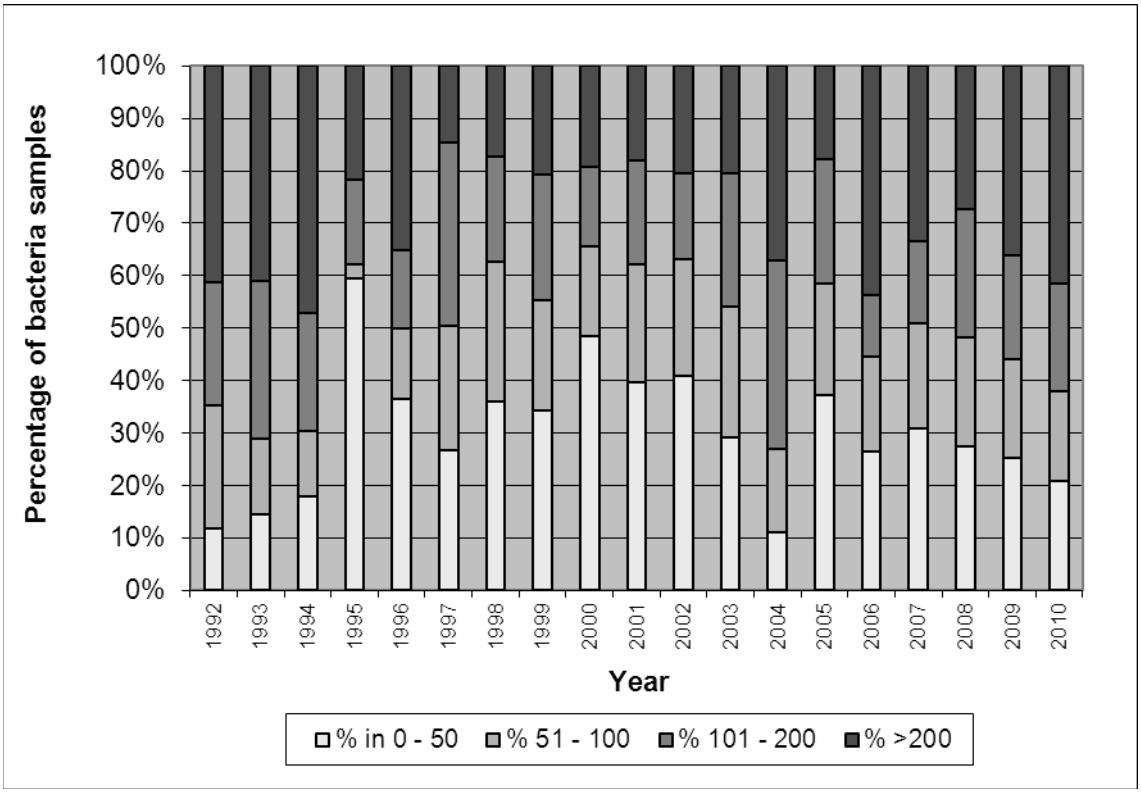

Figure 4. Percentages of fecal bacteria samples (cfu/100 mL) that fall in each water quality category by year.

Table 10. The number of *E. coli* or fecal coliform samples taken each year.

Year	1992	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005	2006	2007	2008	2009	2010
Sample Count	17	83	89	37	88	109	75	96	99	116	122	113	100	118	117	120	106	111	111

It is important to note that in 1992 and 1995, a relatively small number of samples were collected, meaning results for these years may not be as representative as for other years. 1992 showed an extreme low for sample proportions falling into the 0 – 50 cfu/100mL range,

while 1995 showed an extreme high. However, due to the fact that there were so few samples taken in those years, the results may not reflect actual water quality for those years.

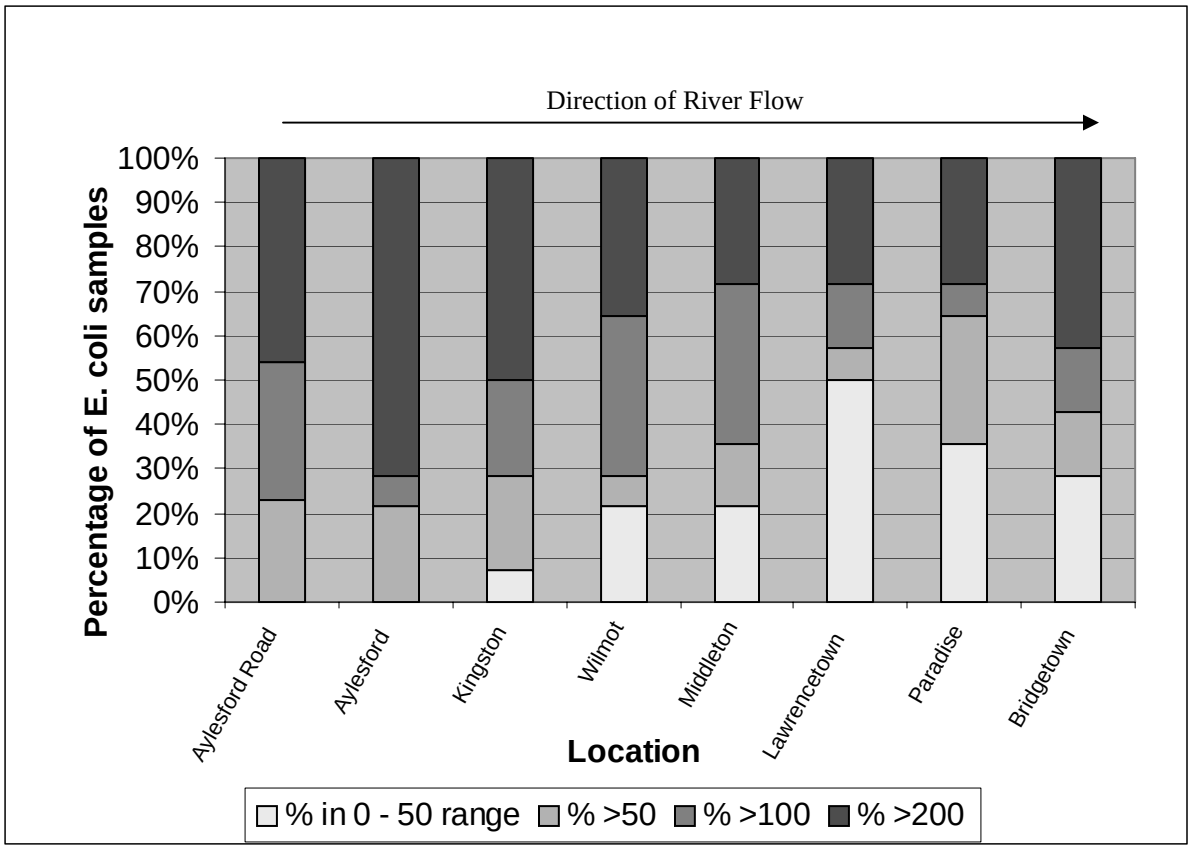

Figure 5. The percentages of 2010 samples falling into the different cfu/100mL ranges, organized by location.

Following a similar pattern as previous years, the highest bacteria counts occurred in Aylesford on Victoria road, while the lowest occurred at the Bridgetown, Paradise and Lawrencetown locations (Figure 5). 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. In 2009, an attempt was made to identify possible sources of this bacterial contamination. These results were inconclusive based on the testing method and are detailed in the 2009 River Guardian Report.

E. Coli Monitoring Recommendations

- Continue regular River Guardian *E. coli* monitoring at the eight main river sample locations.
- Contact livestock owners addressing the issue of restricting animals from the Annapolis River.
- Determine the source of contamination between Aylesford Rd and Victoria Rd
- Investigate correlation between precipitation amounts and *E.coli* levels in the river.

Dissolved Oxygen

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 is able to 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).

Monitoring Results

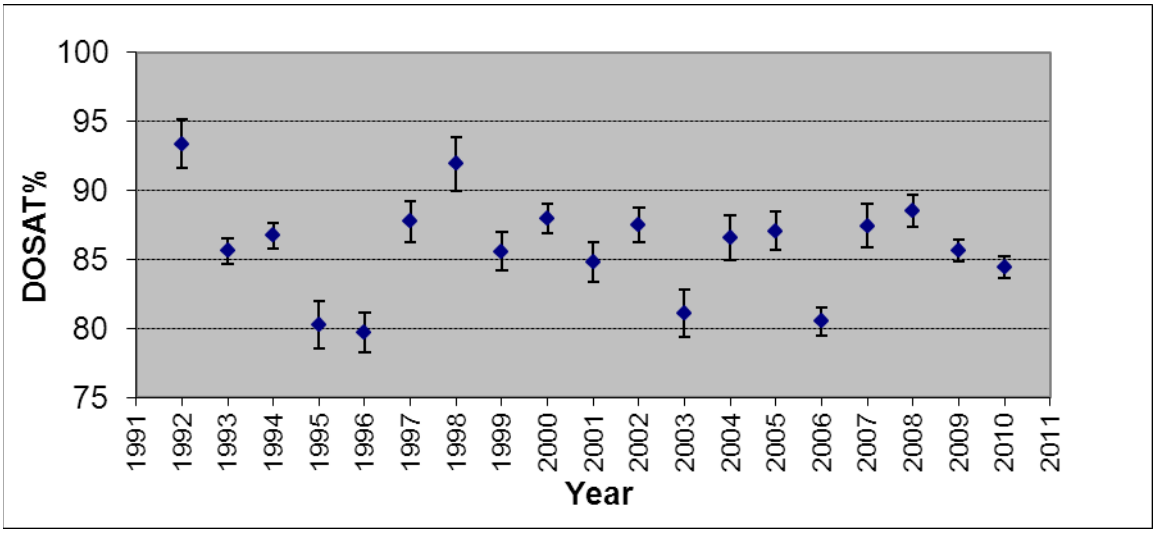

Figure 6. Mean dissolved oxygen saturation (DO SAT) by year, 1992 to 2010 (showing standard error of the mean).

During the period of 1992 to 2010, annual mean dissolved oxygen (percent saturation) levels have varied from a high of 94% in 1992, to a low of 80% in 1996 (Figure 6). The dissolved oxygen varied little in the past two years. In 2010, the mean dissolved oxygen saturation was 84%, compared with 85% in 2009. This value is within the normal range of variability observed for the Annapolis River. The standard error of the mean is shown with error bars, which indicate that there was approximately the same variability in 2010 as in 2009.

The 17-year mean dissolved oxygen (percent saturation) values for each of the main river monitoring sites were calculated (Figure 7). The standard error of this mean is shown with error bars. This is overlaid with the mean values for the 2010 monitoring season. Aylesford road, Kingston and Lawrencetown fell outside the normal DO range, as shown by the bars indicating standard error of the mean. The 2010 averages at Kingston and Lawrencetown were slightly lower than the 16-year mean while Aylesford Road had a higher oxygen saturation

average than the historical mean. Note that the average for Aylesford Road is only for 6 years, and that the Middleton and Wilmot averages are missing some data from 1995 and 1996.

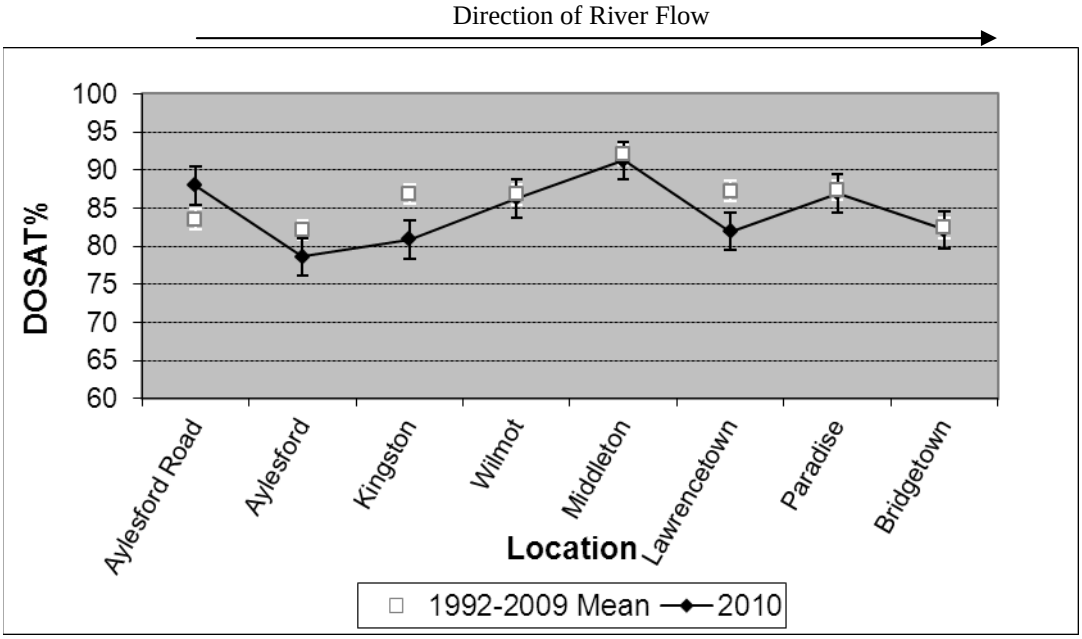

Figure 7. DOSAT results for 2010 as well as mean dissolved oxygen saturation (DOSAT) from 1992 to 2009, organized by sample site. The error bars show standard error of the mean.

The Canadian dissolved oxygen water quality guideline for the protection of freshwater aquatic life is 5.5 mg/L (CCME, 2002). There were no recordings below this level for the 2010 season. Also, there were no samples collected that had dissolved oxygen saturation of 60% or less (Table 11). Out of 111 readings, 100 readings had DO saturation greater than 75%. The high levels of dissolved oxygen observed consistently 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 between the Wilmot and Middleton sites.

Table 11. Dissolved oxygen percent saturation (DOSAT) thresholds for Annapolis River.

Site	Samples less than 60%	Samples within 61-74%	Samples greater than 75%	Total Samples 2010
Aylesford Road	0	0	13	13
Aylesford	0	3	11	14
Kingston	0	3	11	14
Wilmot	0	1	13	14
Middleton	0	0	14	14
Lawrencetown	0	1	13	14
Paradise	0	1	13	14
Bridgetown	0	2	12	14
Totals	0	11	100	111

Dissolved Oxygen Monitoring Recommendations

- Continue regular River Guardian DO monitoring program at the eight main river sample locations.
- Undertake periodic 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.

Water Temperature

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 20ºC, with lethality occurring after prolonged exposures to temperatures over 24ºC (MacMillan *et al.*, 2005).

Monitoring Results

The mean summer water temperature for the Annapolis River in 2010 was 18.9 ºC, which is 1.1 ºC warmer than the same period in 2009. As in previous years, water temperatures during 2010 continued to reach levels stressful to aquatic life during the summer months. The 2010 season average was in the mid to upper range of variability when compared to the previous years (Figure 9). The mean summer water temperature (July, August, September) by year for the eight main River Guardian monitoring sites were compared to the 1992 to 2010 mean summer water temperature (18.5 ºC). The average for 2010 is 0.4 ºC above this average.

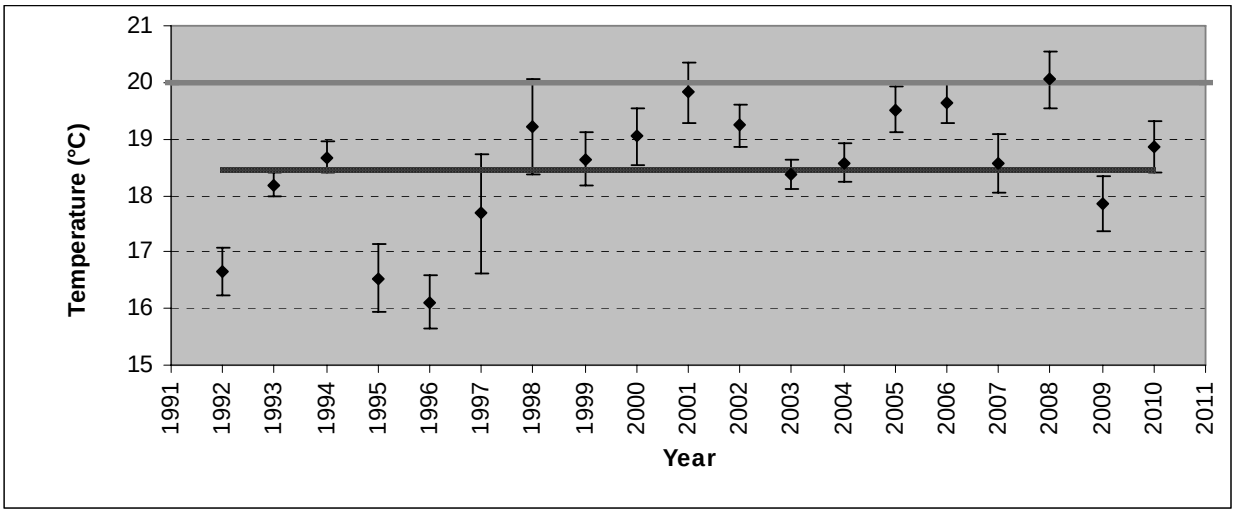

Figure 8. Mean summer water temperature by year (showing standard error of the mean) with 1992-2010 mean shown as a thick line. The 20ºC threshold when fish become stressed is shown as a thick grey line.

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 main Annapolis River in 2010 were compared to the historical averages for those sites (Figure 9). At all sites, the 2010 average was higher than the average from 1992 to 2009. Kingston had the greatest deviation with an average 3ºC warmer than the historical value.

Of the 48 temperature measurements recorded during the months of July, August and September in 2010, 38% exceeded 20ºC. The amount that exceeded 20ºC in 2009 was 23%. The maximum temperature observed was 25.0ºC, recorded at Bridgetown on July 11th, 2010.

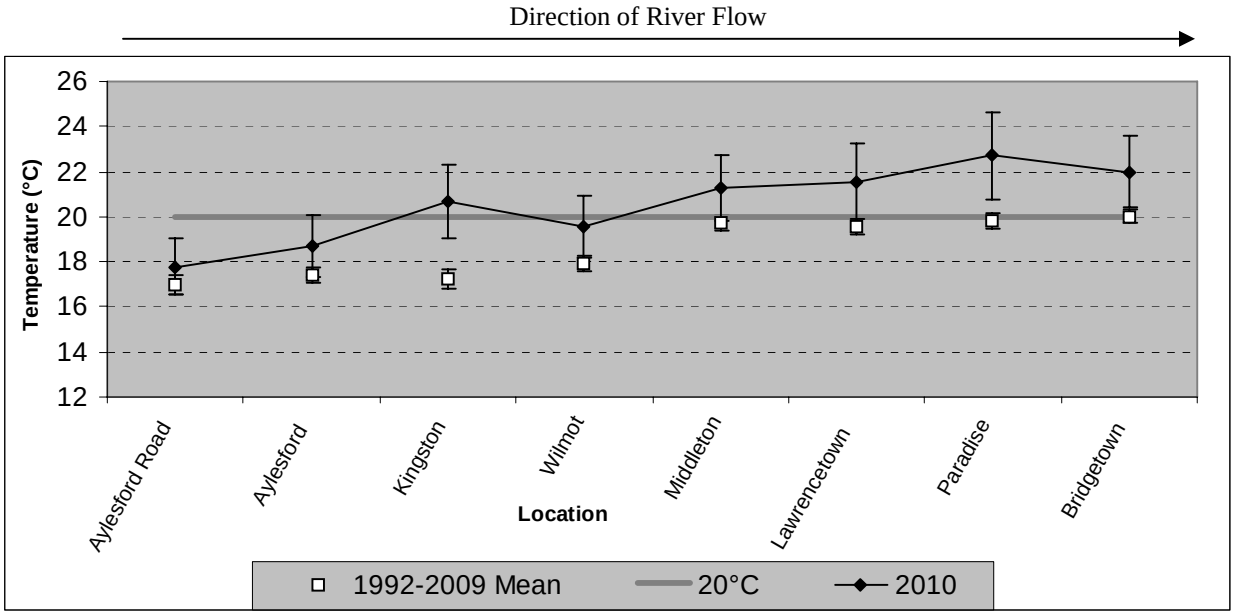

Figure 9. Mean 2010 summer water temperature and historical average temperature (1992 – 2009) by site, with standard error of the mean. The 20°C threshold when fish become stressed is shown as a thick grey line.

Water Temperature Monitoring Recommendations

- Continue regular River Guardian temperature monitoring program at the eight main river locations.
- 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.
- Add a graph displaying the correlation between water and air temperature.

pH

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 vary beyond 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 the underlying soil type and its buffering capacity; however it can also be influenced by anthropogenic means, such as acid precipitation.

pH values are measured on the day following River Guardian collection by CARP staff using the portable HydroLab Quanta water meter (see Appendix A for more details on sampling procedure and meter calibration).

Monitoring Results

pH values all along the Annapolis River are generally good, being only slightly acidic (Figure 10). 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 110 samples of 2010, the lowest value was 6.6 2 in Bridgetown on November 1st while the highest was 7.71 collected in Kingston on May 31st. On average, pH was most acidic in Lawrencetown closely followed by Aylesford Road. Although these two areas had the lowest pH of the sampling sites, their averages were 6.97 and 6.99 respectively including them in the range 6.5 – 9.0 deemed safe for aquatic species by the CCME. There were no values recorded that were out of this range for the 2010 season.

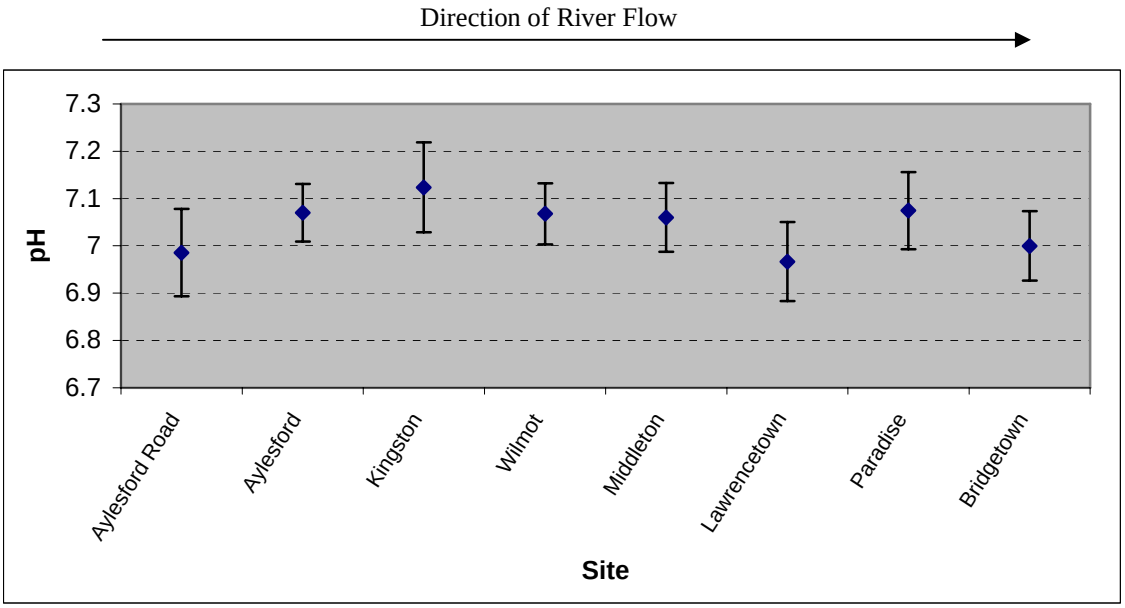

Figure 10. Average pH in 2010 for sampling locations along the Annapolis River (showing standard error of the mean).

Through the past eight years, pH has been in the optimal range, except for 2005 when it fell on the lower end of the scale (Figure 11). The cause of this deviation may possibly have been acid rain and analysis can be reviewed in the 2005 River Guardians report. The pH has increased slightly, becoming less acidic, from 2009 to 2010. During the early years of the Annapolis River Guardians program, pH was

regularly measured at many of the main river sample locations. The average of this previous data was 6.9 and was based on 634 measurements. This historic pH is similar to that observed during the 2003 to 2010 period.

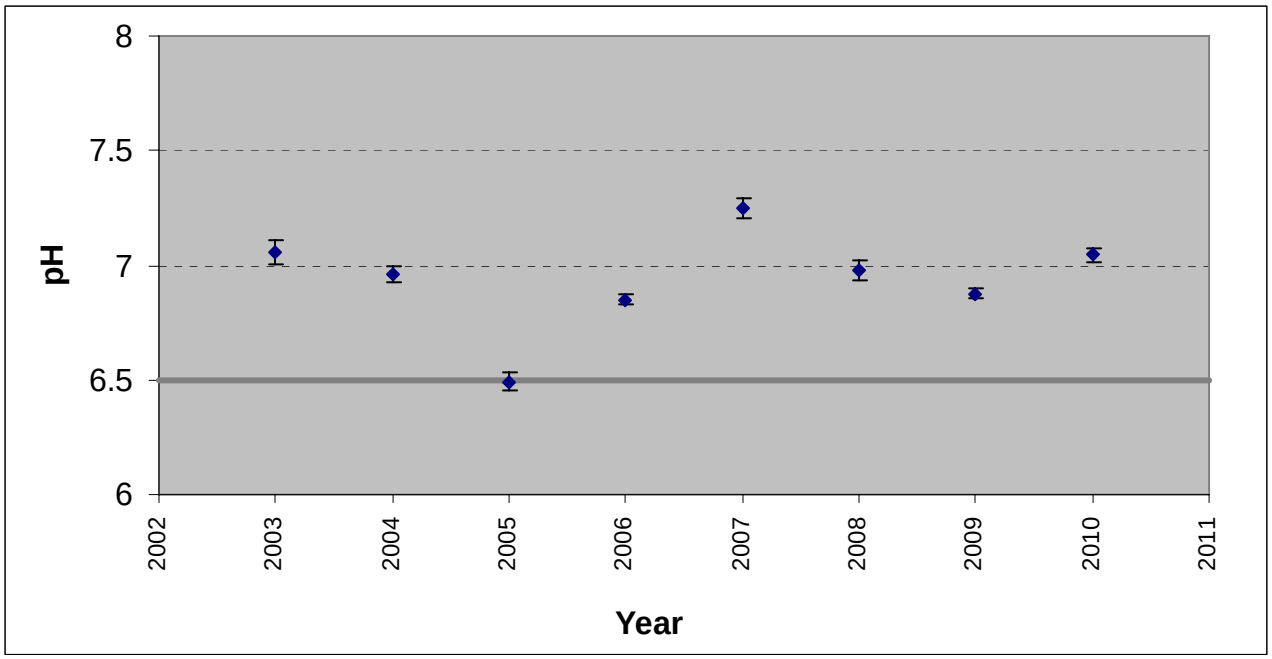

Figure 11. Average pH measured yearly along the Annapolis River (showing standard error of the mean) from 2003-2010. Shown by a thick grey line is the lower threshold of 6.5 for fish species.

pH Monitoring Recommendations

- Regular pH monitoring should be continued at the eight Annapolis River Guardian monitoring locations.
- Investigate acidity in the Nictaux River as a possible reason for the pH drop in Lawrencetown.

Nutrients: Nitrogen and Phosphorus

Introduction

Nutrients are essential for the growth of both plant and animal 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. Currently, nutrient monitoring is only carried out in Wilmot and Millville. Although nitrogen and phosphorus are naturally occurring, there are many anthropogenic sources. Any kind of wastewater discharges (domestic, municipal, industrial), agricultural chemicals such as fertilizers and atmospheric deposition can all contribute to elevated nutrient levels in a river system.

Nitrogen and phosphorus occur naturally in very small amounts and thus are often the limiting factor for plant growth. When nutrient levels rise, they can cause excessive periphyton and macrophyton growth in freshwater systems. Upon death and decomposition, they deplete oxygen to levels, which can threaten aquatic life.

There is much disparity between literature sources identifying unacceptable levels of these two nutrients. Dodds and Welch (2000) compiled many different criteria from literature sources for unacceptable levels of both nitrogen and phosphorus. For total nitrogen, depending on the water quality target, the upper limit ranged from 0.25 mg/L to 3.0 mg/L. For dissolved nitrate, the limits are defined to be anywhere from 0.02 mg/L to 1.0 mg/L. The CCME (2003) has established a guideline for nitrates at 2.9 mg/L nitrates (NO_3) as nitrogen (N) for the protection of aquatic life.

There appears to be greater consensus for guidelines for phosphorus. The Ontario Ministry of Environment and Energy (OMEE) set a guideline of 0.030 mg/L total P, above which excessive plant growth occurs. Mackie (2004) suggested that total phosphorus levels in excess of 0.030 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.

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 2010 was performed in Wilmot, near the bridge and gauging station on Bayard Road and in Millville, near the bridge on Victoria Road.

The results for monitoring of total nitrogen, nitrates and total phosphorus can be seen in Figures 12, 13 and 14 respectively.

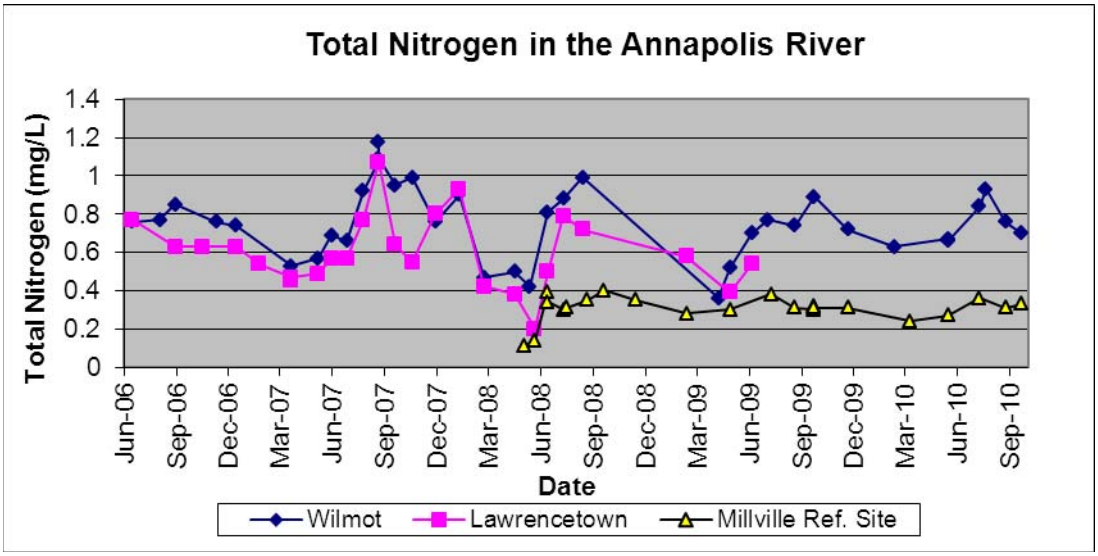

Figure 12. Total nitrogen results from 2006-2010 for Wilmot, 2006-2008 for Lawrencetown, and 2008-2010 for Millville Reference Site.

Total nitrogen has a fairly large range of values that appear to follow a slight yearly trend. Wilmot and Lawrencetown values range from 1.18 mg/L on August 21st, 2007 in Wilmot to 0.2 mg/L on May 21st, 2008 in Lawrencetown. At the Millville Reference Site, the initial reading is the minimum recorded, of 0.11mg/L on May 1st, 2008. It peaks on September 19th, 2008 at 0.4 mg/L. Total nitrogen in Wilmot and Lawrencetown follow a varying seasonal trend while the reference site in Millville is fairly consistent (Figure 12). Values in all three locations peak in the summer season and drop in winter season. Total nitrogen starts to decrease around August and continues to decline until near April when values again climb to climax in early summer. This variation may be the result of agriculture fertilizers and other anthropogenic factors affecting land surrounding the river in Wilmot and Lawrencetown. 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 causes adverse ecological effects, described by Dodds and Welch (2000).

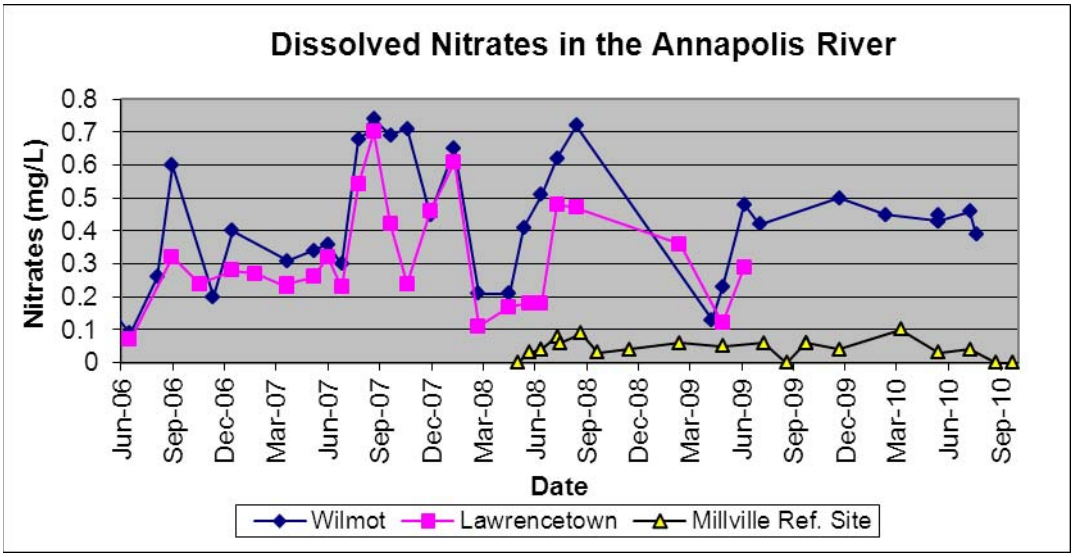

Figure 13. Nitrate results from 2006-2010 for Wilmot, 2006-2008 for Lawrencetown, and 2008-2010 for Millville Reference Site.

Similar to total nitrogen in the Annapolis River (Figure 12), dissolved nitrates peak in the summer season and drop during the winter (Figure 13). The magnitude of variation is less than in Figure 12 as dissolved nitrates only contribute in part to the overall total found in the river. The highest level of nitrate was observed at Wilmot on August 21st, 2007 at 0.74 mg/L while the lowest was 0.07 mg/L in

Lawrencetown on June 16th, 2006. Nitrates at the Millville site were consistently measured between 0.03 mg/L and 0.10 mg/L. All of these levels are far below the CCME guideline.

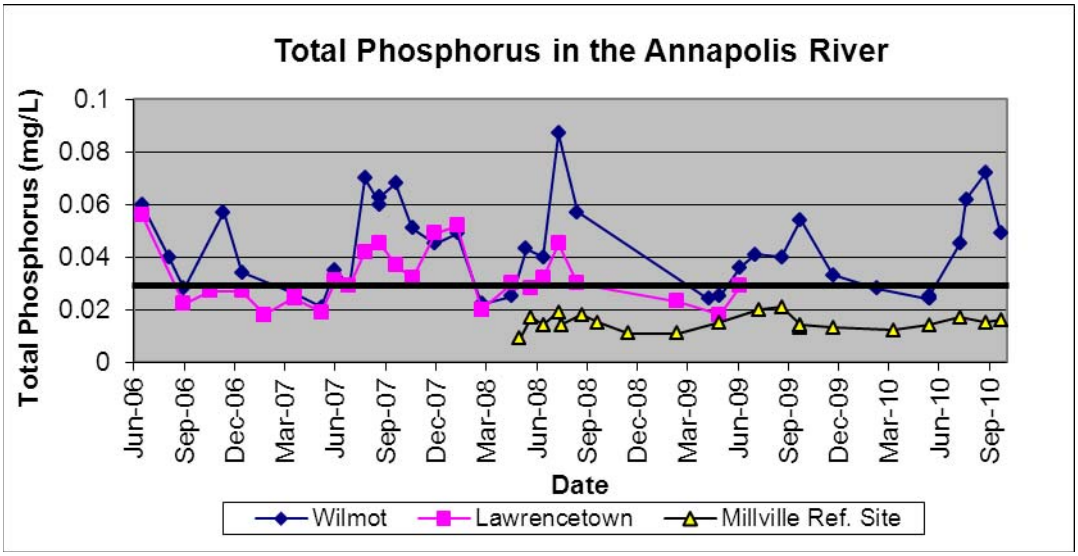

Figure 14. Total phosphorus results from 2006-2010 for Wilmot, 2006-2008 for Lawrencetown, and 2008-2010 for Millville Reference site. The solid line represents the phosphorus guideline of 0.030 mg/L (Mackie, 2004).

The general trend of total phosphorus observed in the Annapolis River increases from spring to summer and decreases from summer to winter (Figure 14). Although Lawrencetown and Wilmot values follow a similar trend, the data is not as closely paralleled as seen in the nitrogen data. The maximum total phosphorus of 0.09 mg/L was observed at Wilmot on July 10th, 2008 and the minimum of 0.018 mg/L was in Lawrencetown on April 28th, 2009. Of all the data collected, 67% from Wilmot and 38% from Lawrencetown were above the recommended upper limit of 0.030 mg/L. Millville values were consistently below this guideline with a minimum value of 0.015mg/L on November 14th, 2008 and a maximum of 0.021 mg/L on September 22, 2009.

In the past, large algal blooms have occurred on the Annapolis River. On July 27th, 2008, the River Guardian volunteer for Bridgetown noted a green colour to the water. On August 1st, 2008, CARP staff observed a dark green colour to the water at this location only. This colour seems to be indicative of an algal bloom and may have been a result of excess levels of nitrogen and/or phosphorus. On the next collection day, August 10th, 2008, the green colour was no longer observable. No instances of an algal bloom were noted in 2009 or 2010 although the river is not regularly monitored for this phenomenon.

Table 12. Average, minimum and maximum for total nitrogen, dissolved nitrates, and total phosphorus at each location. Results are from 2008-2010 for Millville, 2006-2010 for Wilmot, and 2006-2009 for Lawrencetown.

	Total Nitrogen (mg/L)			Dissolved Nitrates (mg/L)			Total Phosphorus (mg/L)		
Location	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
Millville Ref Site	0.31	0.11	0.40	0.05	0.03	0.10	0.015	0.009	0.021
Wilmot	0.76	0.36	1.18	0.44	0.09	0.74	0.043	0.021	0.087
Lawrencetown	0.60	0.20	1.07	0.31	0.07	0.70	0.031	0.018	0.056

Wilmot has a higher nutrient concentration for total nitrogen, dissolved nitrates, and total phosphorus than either Lawrencetown or Millville (Table 12). Millville has the lowest average for all three nutrients when comparing all three sites. Therefore, the locations in order of

increasing nutrients, and thus decreasing river health are Millville, Lawrencetown and Wilmot. Wilmot is located upstream of Lawrencetown on the main stem of the Annapolis River. 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. Wilmot is the next downstream site before Lawrencetown. Also, between Wilmot and Lawrencetown, the Nictaux River, Black River and other tributaries enter the Annapolis River, possibly diluting the nutrient results at Lawrencetown.

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.

Benthic Invertebrates

Introduction

River systems are host to many different forms of life, and many of them can help indicate the river's water quality. Of particular interest are the benthic invertebrates that live on the streambed. These include insects (e.g. mayflies), molluscs (e.g. clams) and other organisms that spend part or all of their life cycle on the bottom of watercourses. Some invertebrates are very sensitive to pollution, while others are pollution tolerant and can thrive in a contaminated environment. The relative abundance and diversity of benthic invertebrates present at a site can provide information on the water quality.

Benthic invertebrate sampling adds another dimension to ecological monitoring. While the measurement of physical and chemical parameters provides a picture of the river's health at a given time, the type of organisms existing in the system can provide a longer-term indication of its health. For example, a rainfall event can cause a river's total suspended solid count to spike for a short period and then quickly return to normal, whereas benthic life will show a greater sensitivity to long-term effects.

The sampling is ideally performed in the latter portion of the summer, late August to mid-September, during relatively low water levels when streams are safer to work in. CARP makes use of the sampling and analysis procedure developed through the Canadian Biomonitoring Network (CABIN).

Benthic Invertebrate Monitoring in the Annapolis River Watershed

The CABIN sampling program undertaken by CARP has pursued three objectives:

- To collect a sufficient number of samples from reference, or pristine, sites in order to allow the development of a reference condition approach model (RCA) for Nova Scotia or Atlantic Canada. The development of a RCA model is a long-term objective, requiring contributions from many partners and the collection of samples from across the region.
- To annually collect benthic invertebrate samples from water quality monitoring sites along the main Annapolis River in order to allow a time series analysis to be performed, highlighting temporal changes. This objective has been undertaken with the view that the CABIN analysis will compliment CARP's traditional chemical and physical water quality monitoring activities.
- To utilize benthic invertebrates as a tool to assess before and after changes in aquatic quality at sites undergoing habitat restoration activities.

CARP has worked with Environment Canada since 2002 to build a network of benthic invertebrate sample stations in the Annapolis watershed. Table 10 describes the location and status of CABIN samples collected in the Annapolis watershed by CARP, with CABIN samples collected by Environment Canada staff shown in Table 11. The locations of these samples are shown in Figure 15.

Table 13. CABIN samples collected by CARP

Site Code	Date Sampled	River	Number of	Reference	Comments
	dd/mm/year		Samples	or Test	
ANN0102	9/5/2002	Fales River	1	Reference	
ANN0202	9/24/2002	East Round Hill River	1	Reference	
ANN0302	9/24/2002	West Round Hill River	1	Reference	
ANN0402	9/25/2002	Black River	1	Reference	
ANN0502	10/11/2002	South Annapolis River	1	Reference	
ANN0703	10/8/2003	Skinner Brook	1	Test	
ANN0803	10/8/2003	Leonard Brook	1	Test	
ANN0903	10/8/2003	Leonard Brook	1	Test	
ANN1003	10/9/2003	Slokum Brook	1	Reference	
ANN0103	10/9/2003	Fales River	1 + 2	Reference	Repeat of 2002 Reference Site; QA/QC samples collected
ANN1104	10/18/2004	Annapolis River at Aylesford	1	Test	Long-term monitoring site
ANN1204	10/19/2004	Acacia Brook	1	Reference	
ANN1304	10/19/2004	West Branch Bear River	1	Reference	
ANN1404	10/20/2004	Annapolis River at Kingston	1	Test	Long-term monitoring site
ANN1504	10/20/2004	East Round Hill River	1	Reference	Repeat of 2002 Reference Site
ANN1604	10/21/2004	West Branch Moose River	1	Reference	
ANN1704	10/21/2004	West Branch Moose River	1	Reference	
ANN1804	10/21/2004	East Branch Moose River	1	Reference	
ANN1105	9/13/2005	Annapolis River at Aylesford	1	Test	Long-term monitoring site
ANN1405	9/13/2005	Annapolis River at Kingston	1	Test	Long-term monitoring site
ANN1905	9/13/2005	Annapolis River at Middleton	1	Test	Long-term monitoring site
ANN2005	9/14/2005	Annapolis River at Paradise	1 +2	Test	Long-term monitoring site; QA/QC samples collected
ANN2006	11/10/2006	Annapolis River at Paradise	1	Test	Long-term monitoring site
ANN2106	11/10/2006	E. Branch of S. Annapolis @ Morristown	1	Reference	
ANN2206	11/10/2006	Annapolis River at Wilmot	1	Test	Co-located with EC gauging & Hydrolab placement
ANN2007	11/9/2007	Annapolis River at Paradise	1	Test	Long-term monitoring site
ANN2107	11/9/2007	Annapolis River at Wilmot	1	Test	Co-located with EC gauging & Hydrolab placement
ANN2307	11/9/2007	Fash Brook	1	Test	
ANN2008	17/9/2008	Annapolis River at Paradise	1	Test	Long-term monitoring site
ANN2208	17/9/2008	Annapolis River at Wilmot	1	Test	Co-located with EC gauging & Hydrolab placement
ANN2308	9/9/2008	S. Annapolis River at Millville	1 + 2	Reference	Co-located with EC tubidity & TSS station
ANN2408	8/9/2008	Thornes Brook at Karsdale	1	Reference	
ANN2508	8/9/2008	Fash Brook-West Branch	1	Reference	
ANN2608	9/9/2008	Shearer Brook	1	Reference	
ANN2009	13/9/2009	Annapolis River at Paradise	1	Test	Long-term monitoring site
ANN2209	13/9/2009	Annapolis River at Wilmot	1	Test	Co-located with EC gauging & Hydrolab placement
ANN2309	13/9/2009	S. Annapolis River at Millville	1	Reference	Co-located with EC tubidity & TSS station

Table 14. CABIN samples collected by Environment Canada

Site Code	Date Sampled	River	Number of	Reference
	dd/mm/year		Samples	or Test
EELBRK01	3/10/2006	Eel Weir Brook (inflows to Annapolis River near Lawrencetown)	1	Reference
ROXBRK01	3/10/2006	Roxbury Brook (inflow to Annapolis River near Paradise)	1	Reference
OAKBRK01	4/10/2006	Oakes Brook (inflow to Nictaux River near Albany	1	Reference
BLKRIV01	5/10/2006	Black River (inflow to Annapolis River)	1	Reference
SNSHBK01	6/10/2006	Snowshoe Brook south of Lawrencetown on the South Mountain	1	Reference
SANN01	7/10/2006	South Annapolis River	1	Reference

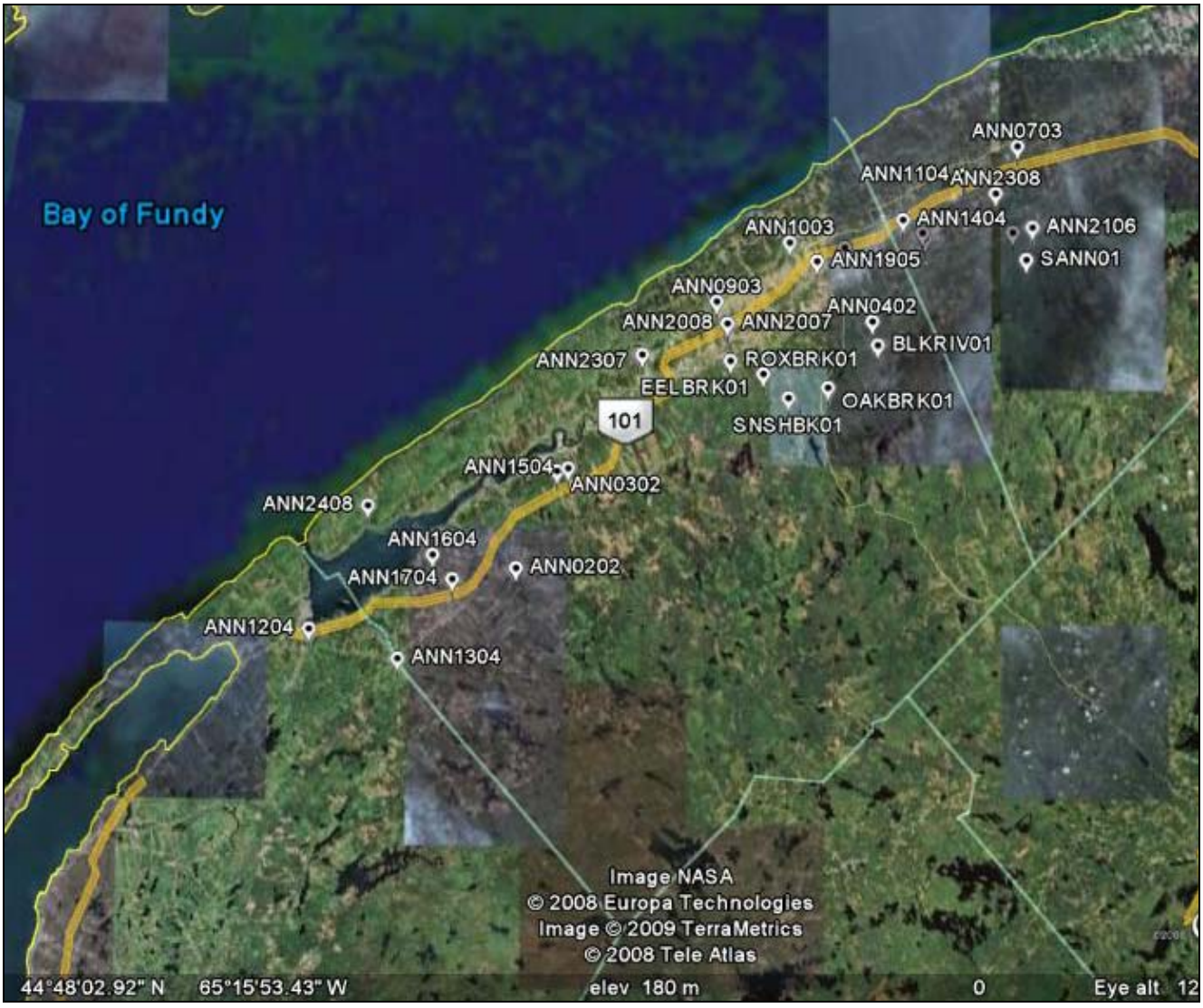

Figure 15. CABIN sample locations in the Annapolis River watershed (collections by CARP and Environment Canada)

Monitoring Results

Benthic invertebrate samples have been collected from the Annapolis River at Paradise since 2005 and at Wilmot since 2006. To present these results, the Family Biotic Index has been used, as indicated by the CABIN analysis procedure. The index produces a value from 0 to 10, 0 being excellent water quality and 10 being poor water quality. The CABIN procedures outline categories for evaluation of water quality using the Family Biotic Index (Reynoldson *et al.,* 2002). These categories are presented below, in Table 15.

Table 15. Evaluation of water quality using the Family Biotic Index.

Family Biotic Index Score	Stream Condition
0.00 – 3.75	Excellent
3.76 – 4.25	Very good
4.26 – 5.00	Good
5.01 – 5.75	Fair
5.76 – 6.50	Fairly poor
6.51 – 7.25	Poor
7.26 – 10.00	Very poor

The tolerance values for the Family Biotic Index calculation were taken from Applied Aquatic Ecosystem Concepts (Mackie, 2004). If they were not listed there, the values were taken from either the CABIN procedures (Reynoldson *et al.,* 2004) or from the Quality Assurance Work Plan for Biological Stream Monitoring in New York State (Bode *et al,* 1991).

Figure 16 presents the results for the Family Biotic Index calculations for the Paradise site from 2005-2009. The result for 2009 was 4.39. The result for each year falls between 4.26 and 5.00, which is a 'good' score, according to the table above.

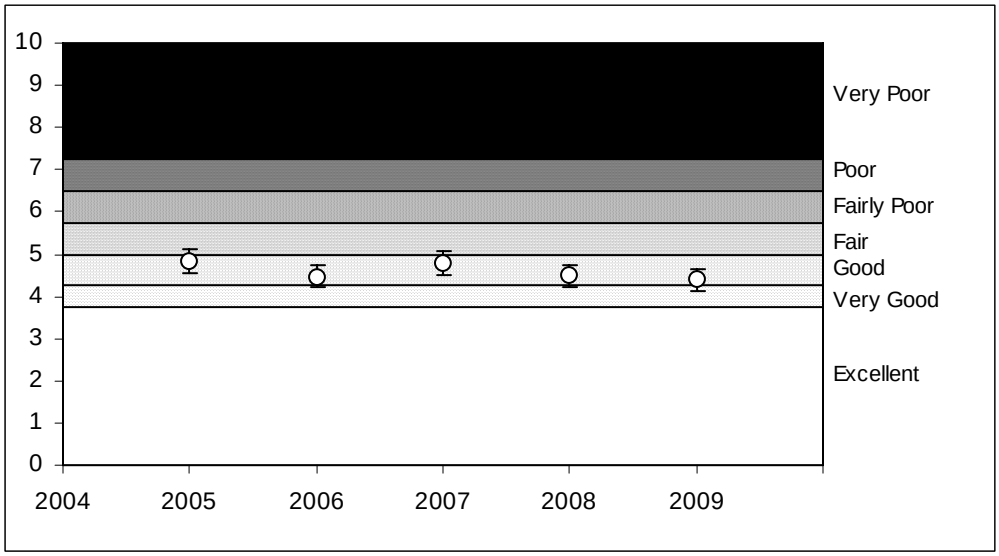

Figure 16. Family Biotic indices for 2005 – 2009 for the Paradise location. The error bars are displaying a 12% error, which was calculated using the QA/QC replicate data.

The 2006-2009 index results for Wilmot are presented below in Figure 17. The 2006 and 2009 indices fall into the 'fair' category while the results for 2007 and 2008 fall into the 'good' category. The Family Biotic Index for Wilmot in 2009 was 5.17.

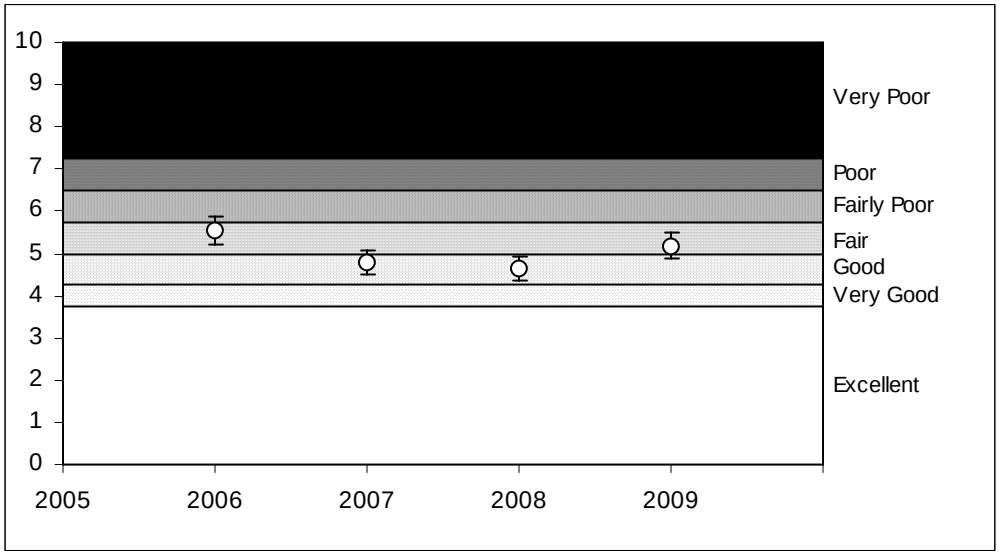

Figure 17. Family Biotic indices for 2006 – 2009 for the Wilmot location. The error bars are displaying a 12% error, which was calculated using the QA/QC replicate data.

The 2009 results for both sites were calculated using the unverified identifications performed by CARP, as the verified identifications were not yet available. The verified results were used to calculate the indices for 2005 to 2008.

In addition to the Family Biotic Index, several other measures were used to characterize the benthic invertebrate samples for Paradise and Wilmot. The tables displaying these results are below

Table16. Benthic invertebrate results for Paradise.

	2005	2005 (QA1)	2005(QA2)	2006	2007	2008	2009
Family Biotic Index	4.83	5.02	4.71	4.45	4.76	4.32	4.39
Taxonomic Richness	31	29	21	24	20	33	25
Total EPT	293	503	197	95	211	291	241
Percentage EPT in sample	50.52%	43.63%	55.18%	32.65%	44.80%	56.50%	53.79%
Diversity	3.21	2.69	3.00	2.99	3.12	3.34	3.44
Hmax	3.43	3.25	3.04	3.05	3.00	3.28	3.22
Evenness	0.93	0.83	0.99	0.98	1.04	1.02	1.07
Intolerant Organism Count	86	149	67	58	87	94	135
Tolerant Organism Count	5	14	13	12	11	13	5
Tolerant - Intolerant Ratio	0.06	0.09	0.19	0.21	0.13	0.14	0.04

Table 17. Benthic invertebrate results for Wilmot

	2006	2007	2008	2009
Family Biotic Index	5.56	4.68	4.68	5.17
Taxonomic Richness	21	21	34	29
Total EPT	41	73	177	61
Percentage EPT in sample	10.79%	24.33%	31.44%	17.63%
Diversity	1.53	2.62	3.47	2.75
Evenness	0.50	0.86	1.05	0.82
Intolerant Organism Count	21	63	108	45
Tolerant Organism Count	32	0	46	11
Tolerant - Intolerant Ratio	1.52	0.00	0.43	0.24

The different measurements are described below.

- Taxonomic Richness refers to the number of different types of families in the sample.
- Total EPT refers to the number of organisms in the sample that come from the orders of Ephemeroptera, Plecoptera or Trichoptera. These organisms tend to have low pollution tolerance, so larger relative numbers of them tend to indicate less contaminated waters.
- The Diversity Index measures the relative abundance of each family. Mackie (2004) describes guidelines for using the species diversity index in assessing water quality. Since the samples taken by CARP were not identified to species, the index was modified to be used at the family level. A diversity index of <1 indicates polluted water, an index result of 1-3 indicates sub-polluted water and an index of >3 indicates clean water. However, Mackie does emphasize that these results treat all organisms as identical and does not take into account the pollution sensitivity of each different taxonomic grouping. The test is also optimized for analysis at the genus level of taxonomy and loses reliability at higher levels, such as family.
- Evenness also measures how the organisms are distributed between families. The closer the sample is to an even distribution, the closer this value will be to 1. Stresses to the aquatic environment tend to cause some taxa to shrink in number or disappear while causing others to increase in population resulting in populations skewed toward a small number of taxa. Thus, evenness results close to 1 tend to indicate a relatively uncontaminated environment.
- Intolerant organism counts measure the amount of organisms that come from families with a Hilsenhoff tolerance value of 3 or less; tolerant organism counts measure the amount of organisms that come from families with a Hilsenhoff tolerance value of 7 or greater.

Benthic Invertebrates Monitoring Recommendations

- Continue to collect annual benthic invertebrate samples from the Paradise and Wilmot locations.

Total Suspended Solids and Turbidity

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 in turn 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, which can then 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.

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, samples were taken after 15 mm of precipitation had fallen.

Monitoring results

Turbidity and TSS data collected May through to December in 2008-2010 along the Annapolis River is compiled in Figures 18 and 19. There are several spikes in the data throughout each year, which corresponds to major precipitation events. The most notable occurred in September 2008, March 2009, July 2009, February 2010 and December 2010 (Figures 18 and 19). All of these dates correspond to rainfall amounts of greater than 50 mm with the exception of March 30, 2009 when only 17mm of rainfall was recorded. It is possible that there was significant snowmelt in occurrence with the rainfall, which led to high and turbid river levels.

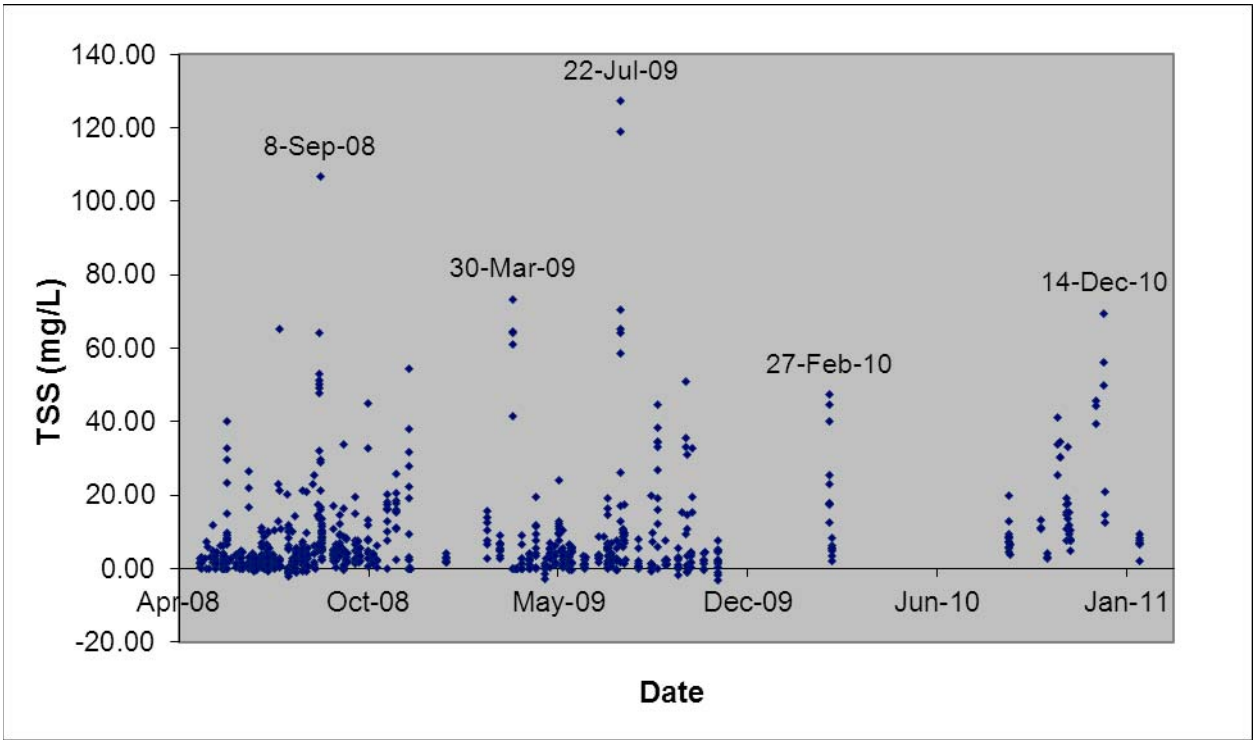

Figure 18. 2008-2010 Total Suspended Solids (TSS) results in mg/L by date.

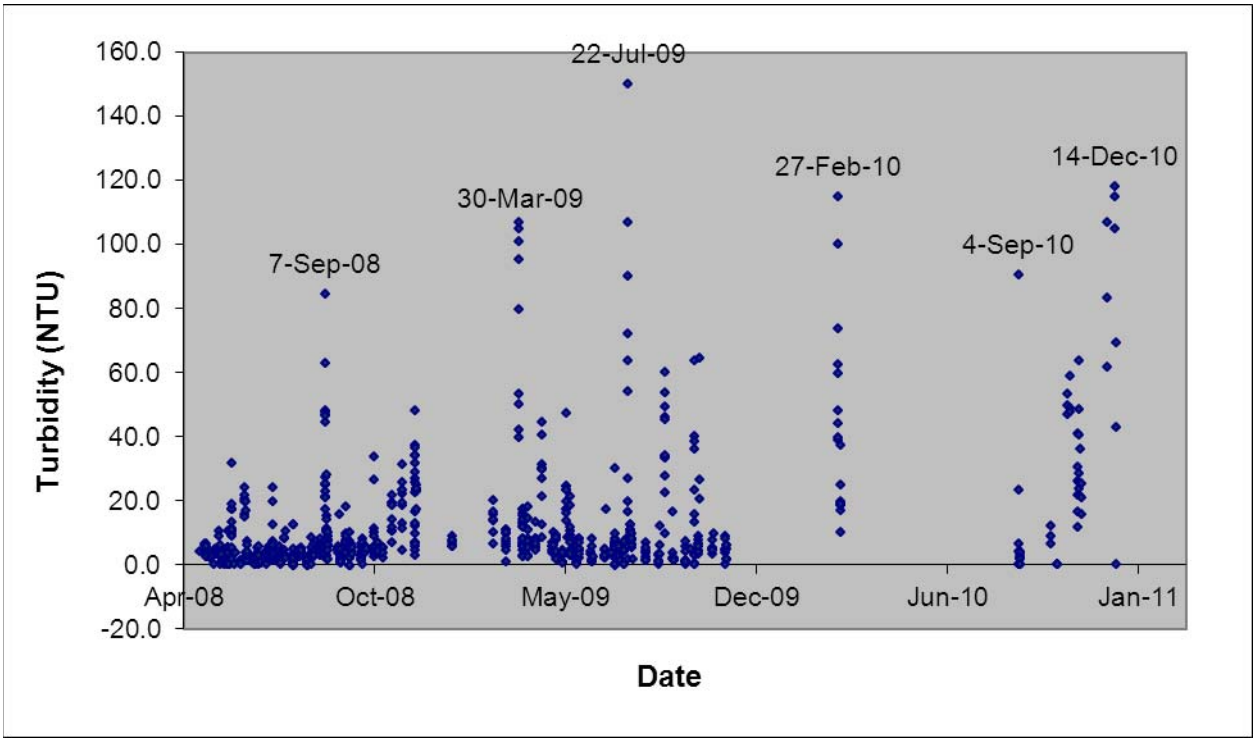

Figure 19. 2008-2010 turbidity results in Nephelometric Turbidity Units (NTU) by date.

Data for the turbidity and TSS sample grabs for 2008-2010 were compiled in box and whisker plots to show the variability of the parameters between stations (Figures 20 and 21). Event grabs were overlaid onto the routine grabs to demonstrate how peak sediment in the river compares to baseline concentrations. The results have a large range and are shown in a logarithmic scale.

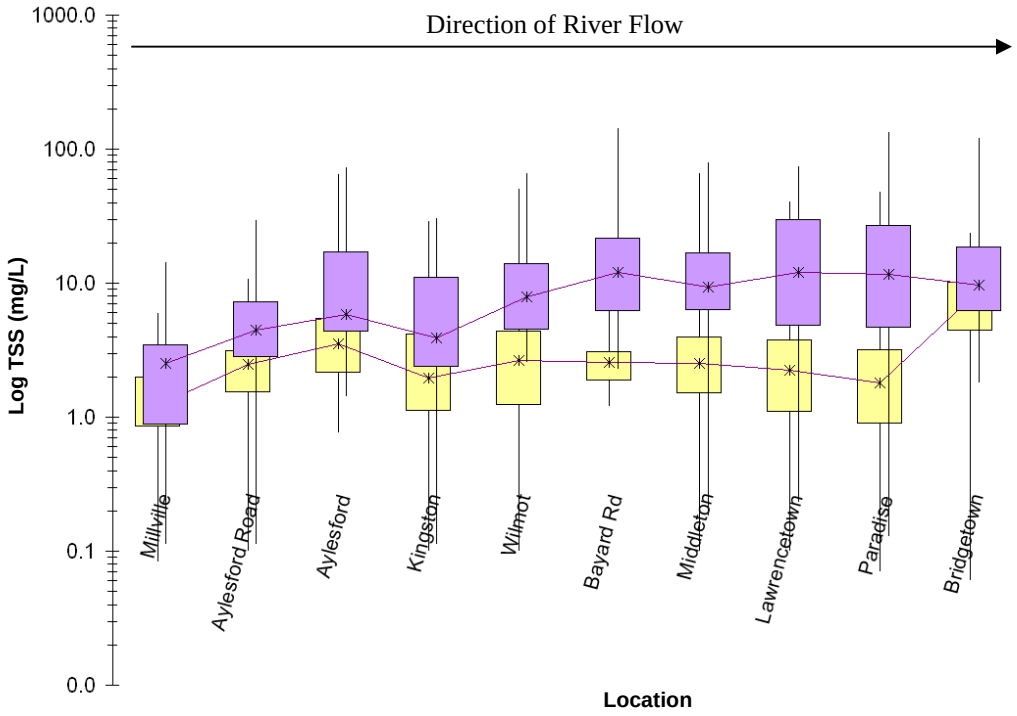

Figure 20. Routine (yellow) and Event (purple) TSS (mg/L) samples gathered at all locations from 2008-2010.

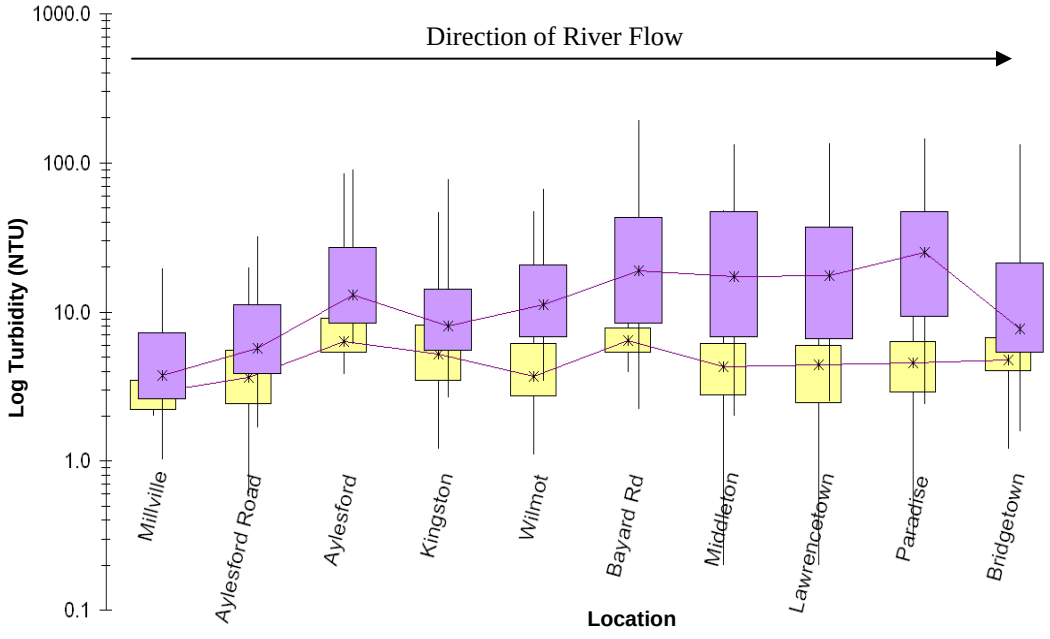

Figure 21. Routine (yellow) and Event (purple) Turbidity (NTU) samples gathered at all locations from 2008-2010.

Event grabs showed an overall higher TSS (mg/L) average than routine grabs, as is expected. Maximum levels reached over 100 mg/L with the highest value of 127.32 mg/L from Bayard Road, Wilmot on July 22, 2009. The Millville reference site has the lowest amounts of suspended solids in the water column for both routine and event samples. The samples for turbidity follow the same general trend. The highest turbidity measured was 150 NTU and was taken from the same sample that produced the highest TSS value. Note that the Bridgetown location shows high TSS readings when compared with the other sites, which is not reflected in the turbidity results. The Bridgetown location is the only monitored location that periodically has salt water due to tidal influence, which may be a possible explanation for this discrepancy.

One of the purposes of measuring these two parameters was to establish a relationship between TSS and turbidity (Figure 22). Upon visual examination, it seems as though these two variables are directly correlated. Continued collections and analysis is required to accurately establish this relationship as the data only encompasses a three-year period under the direction of Environment Canada.

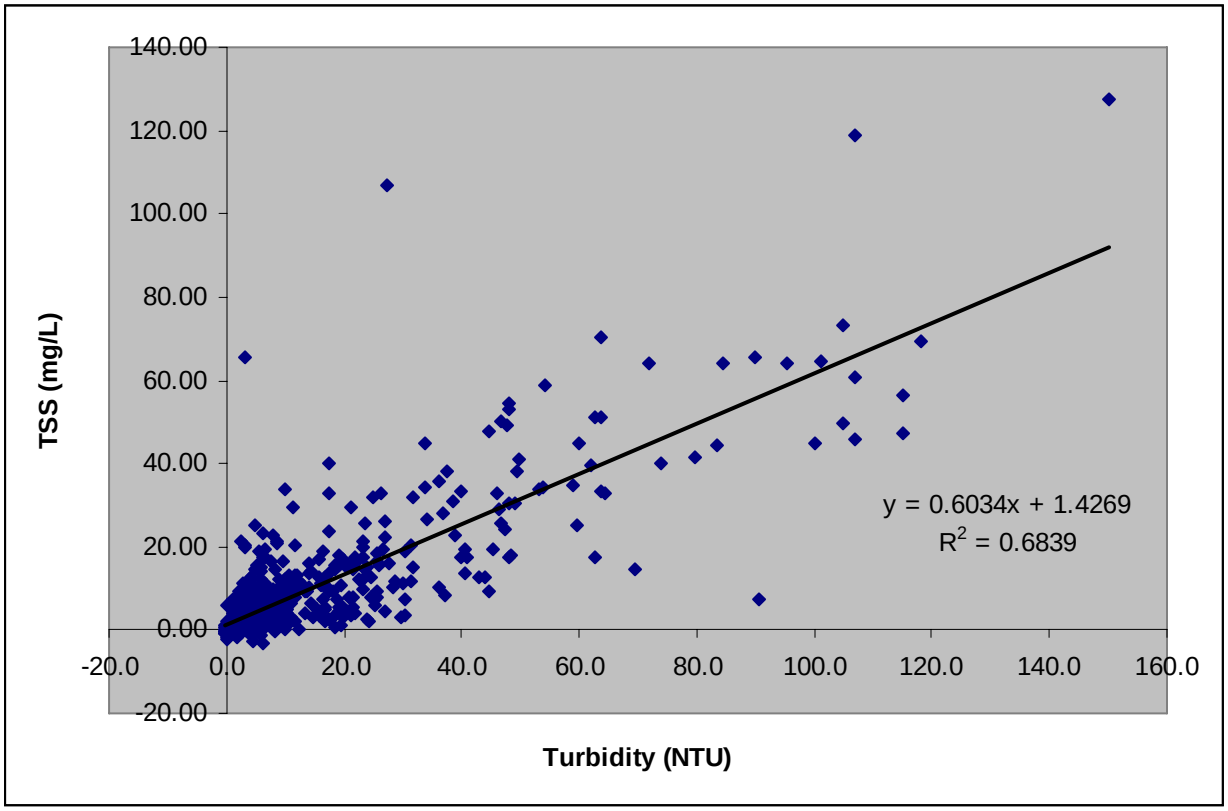

Figure 22. TSS in mg/L vs. turbidity in NTU for all sampled locations along the Annapolis River with the best-fit straight line and equation.

Although the best-fit straight line and equation are included in the chart above, this is only a preliminary estimate of the correlation between TSS and turbidity for the Annapolis River. This relationship will be modified and adjusted as CARP continues to collect these samples. Once sufficient data is collected, a more accurate relationship will be developed to enable Total Suspended Solids to be calculated from turbidity readings.

TSS data from 1992 to 2002 was compared to data from 2008 and 2009 gathered during routine biweekly collections (Figure 23). The medians of the two data sets are similar, but the spread of the original data is larger and has a greater number of small values (between -15 and 1 mg/L). This might be due to the size of the original data set, which contains 9 years of data, whereas the 2008/2009 data set is only for 2 years. Note that the scale of the y-axis is logarithmic, therefore, negative values cannot be shown. However the minimum values for both sets of data are negative: -78.0 mg/L for the historical data and –3.21 mg/L for the 2008/2009 data.

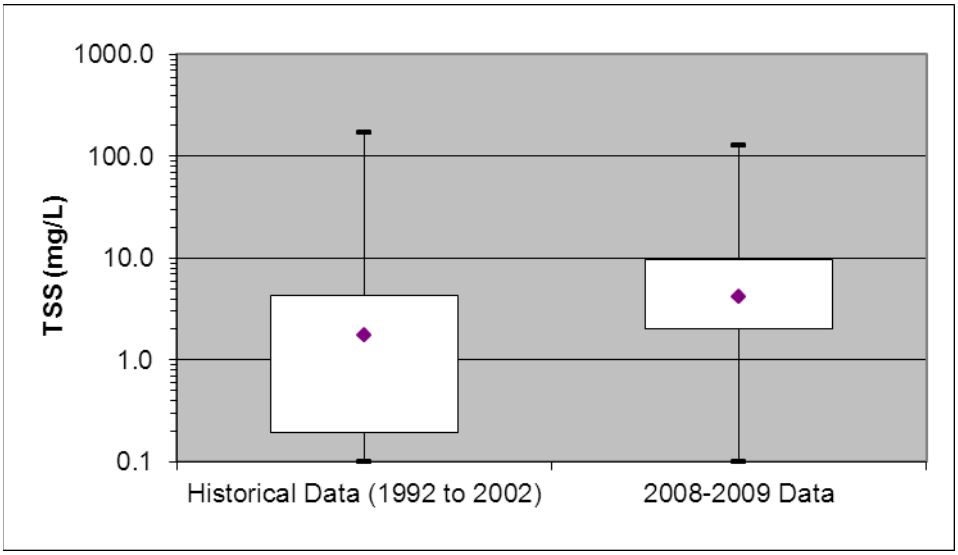

Figure 23. Comparison of the historical River Guardians TSS data (1992 – 2002) and the 2008/2009 TSS data collected as part of the TSS/Turbidity project.

The data collected within this period may not be usable. With regards to this data, Dill (2003) stated:

> The current [TSS] data in the River Guardians database is flawed by the fact that 15% of the samples have a negative value for [suspended particulate matter], which is not possible. The problem of negative values has occurred as recently as 2001 and is distributed through most of the years.

The data taken from 2008-2010 also contained negative values. Before correction, approximately 15% of the data was negative. However, as part of the project's Quality Assurance/Quality Control (QA/QC) plan (see Appendix C), blank samples were also processed. Many of the blank samples produced negative numbers as well, and using these results as a correction factor, the sample data was adjusted. The result was that only approximately 3% of the corrected data was negative. The absence of similar QA/QC data for the original data set makes it difficult to work with the results. In addition, although some of the 2008-2010 data was negative, the 1992 to 2002 data tended to be negative to a much greater degree (as much as –78.0).

TSS/Turbidity Monitoring Recommendations

- Continue Event sampling after precipitation amounts greater than 20mm
- 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.

Trend Analysis

Purpose

A trend analysis has been done for several of the water quality monitoring parameters since 2006. The results of this analysis were included as part of the annual River Guardians Report Card. These trend calculations were simple three-year rolling average comparisons, and a trend was indicated for a certain parameter if it had changed by a given percentage, which varied according to the parameter. If a trend was found, it was reported as either increasing or decreasing, otherwise the parameter was reported to have no trend indicated.

Background Information

In 2008, new methods of performing trend analyses were researched in an effort to increase the statistical validity of the results. Literature sources consulted included: Australian and New Zealand Environment and Conservation Council (2000), Helsel and Hirsch (2002), Hirsch, Alexander and Smith (1991) and Cooke (2006). 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.

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 used 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 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.

The Kruskal-Wallis test was performed using Systat 8.0 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, Slack, 2006)

The parametric procedures that were performed on the data 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.

Systat 8.0 was used to produce scatterplot matrices and autocorrelation plots; the Analyse-It add-on for Microsoft Excel was used to perform the Shapiro-Wilks test and regression analyses.

Methodology

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.

Non-Parametric Analysis

In 2008, a box and whisker plot was made for each parameter, with the data grouped by month. For the temperature, bacteria and pH data, months of January through March were excluded, as very little data was recorded for those months. 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 24).

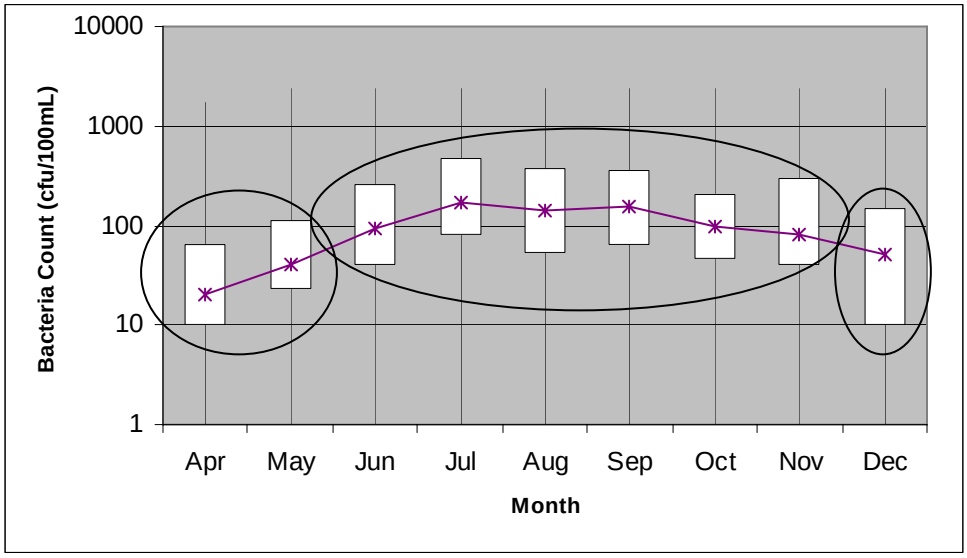

Figure 24. Bacteria count data for all years grouped by month. The circles indicate the seasons that were determined from this plot. There was very little data for the January to March period; these months were not used in the analysis. A 'dummy season' containing no data was used in the analysis to 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.

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 25).

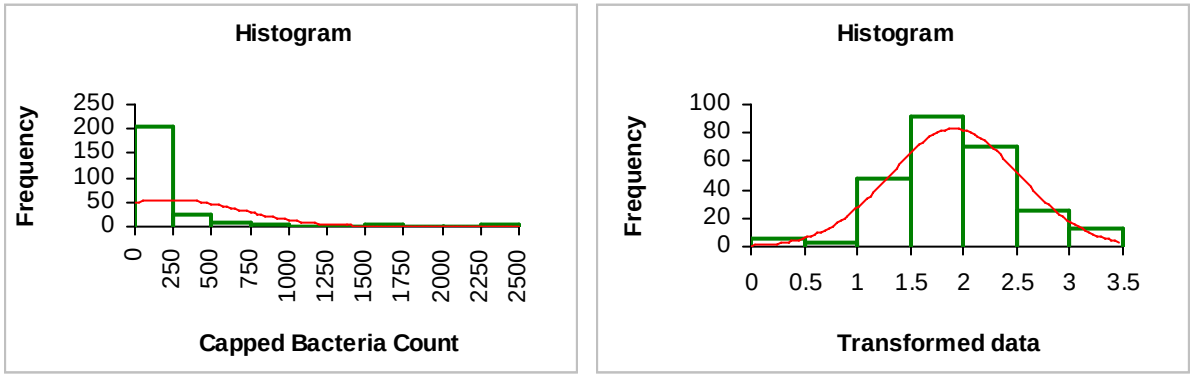

Figure 25. Lawrencetown (#35) bacterial count data distribution before transformation (left) and after transformation (right).

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 the trend analysis procedure below was completed, the bacteria data trend results had to be transformed back using an inverse logarithmic function.

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 26).
- examination of the residual plot and distribution. If the residual scatterplot was randomly distributed and the associated histogram resembled a normal distribution, this test was passed. Non-linear correlation of the data would be indicated if this test was failed (Figure 28).

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 26 and 27 below show that the Kingston data set passed the other two significance tests as well, therefore the trend slope of –0.6 %/year was accepted as significant. This indicates that dissolved oxygen levels are decreasing at the Kingston location.

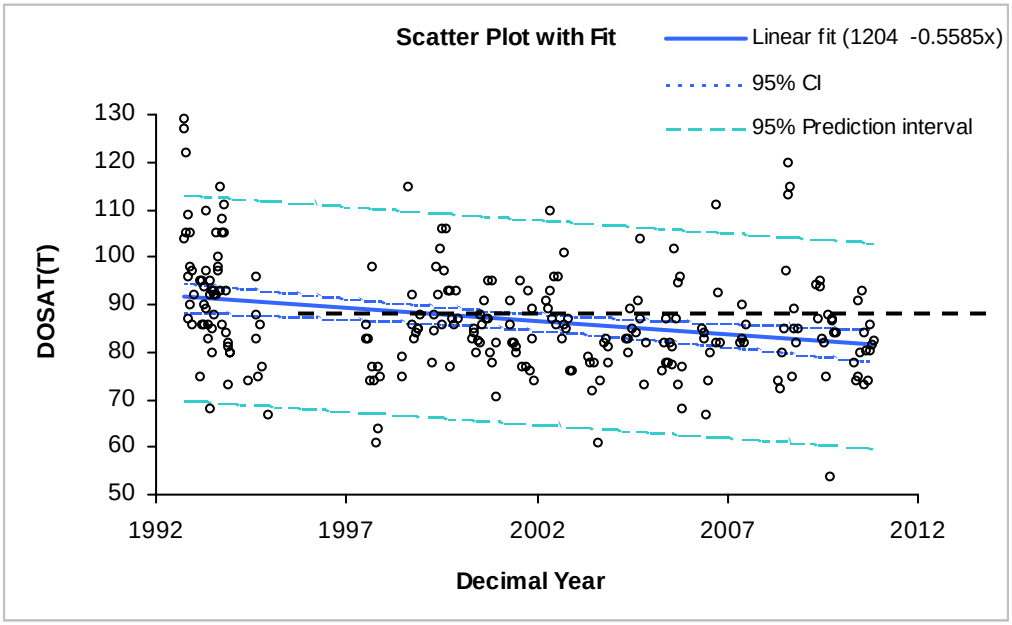

Figure 26. Linear regression for DOSAT data at the Kingston location. The thick dashed line is for the purpose of comparing the confidence interval range at the beginning and end of the data set. If this horizontal line had remained within the confidence interval range for the entire domain of the data set, a trend could not be concluded. This did not occur for the Kingston DO data set; this data set passes this significance test.

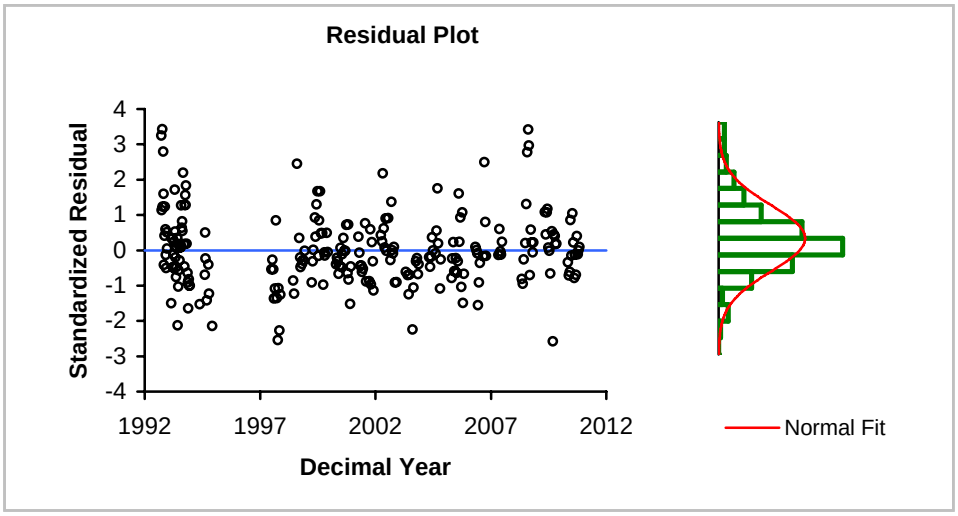

Figure 27. Residuals plot for the DOSAT regression for the Kingston location. The scatterplot does not display significant clustering and appears to be randomly distributed and though the histogram displays a small spike around the centre, it still resembles a normal distribution; therefore this test is passed for the Kingston DO data.

Autocorrelation and Serial Dependence

Autocorrelation is an important consideration for both parametric and non-parametric statistical trend analyses (Helsel and Hirsch, 2002) as its existence invalidates most statistical tests. Autocorrelation refers to serial dependence within a data set, meaning that observation pairs separated by a constant time lag are correlated (Australian and New Zealand Environment and Conservation Council, 2000). One of the assumptions of the linear regression fit is that there must be no correlation between data points (i.e. data points must be independent). 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 28), as well as for the entire data set for each parameter (Figure 29).

Significant serial dependence is indicated when the vertical bars extend beyond the 95% confidence curves. In the Kingston 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 29).

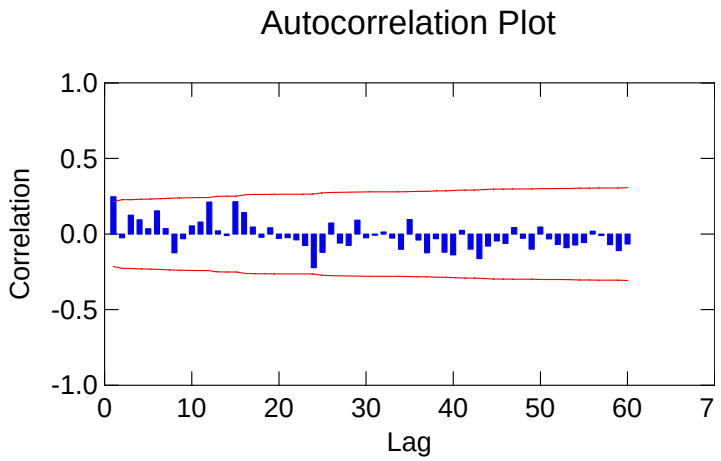

Figure 28. Autocorrelation plot for temperature at the Kingston location.

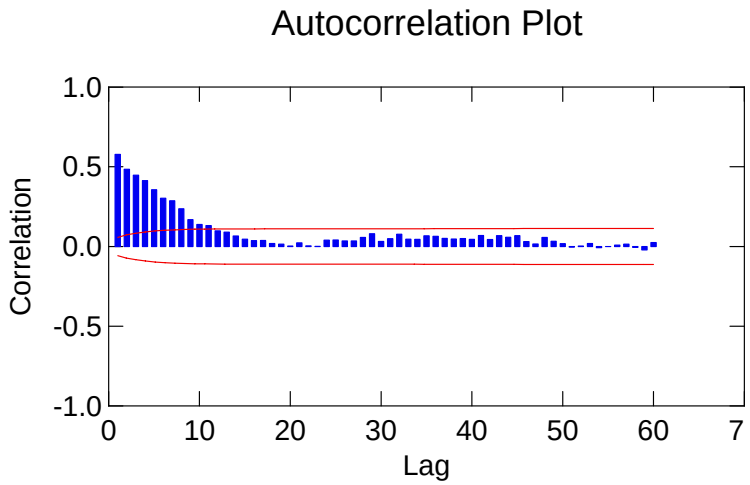

Figure 29. Autocorrelation plot for the entire temperature data set. Several of the bars extend beyond the confidence interval range; therefore significant serial dependence is indicated.

Results

The results for the non-parametric tests (Table 18) and the results for the parametric tests (Table 19) were compiled.

Table 18. Statistically significant trends* and rates of change using non-parametric procedures.

	Bacteria Count	Dissolved Oxygen	pH	Temperature
Aylesford Road	No	No	No	No
Aylesford	Yes(+ 10 cfu/100mL/year)	No	No	No
Kingston	Yes (+4 cfu/100mL/year)	Yes (-0.5 %/year)	Yes (+ 0.03/year)	Yes (+0.13°C/year)
Wilmot	No	Yes (+0.3 %/year)	No	No
Middleton	No	No	Yes (+0.04/year)	No
Lawrencetown	Yes (-4 cfu/100mL/year)	No	Yes (+0.04/year)	Yes (+0.11°C/year)
Paradise	No	No	No	No
Bridgetown	No	Yes (-0.4 %/year)	No	No

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

Table 19. Statistically significant trends* and rates of change using parametric procedures.

	Bacteria Count	Dissolved Oxygen	pH	Temperature	Total Nitrogen	Total Phosphorus
Aylesford Road	No	No	No	No		
Aylesford	Yes (+9 cfu/100mL/year)	No	No	No		
Kingston	Yes (+9 cfu/100mL/year)	Yes (-0.6 %/year)	No	No		
Wilmot	No	No	No	No	No	No
Middleton	No	No	No	No		
Lawrencetown	No	No	No	No		
Paradise	No	No	No	No		
Bridgetown	No	No	No	No		

*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.

Values resulting from these calculations indicate a statistically significant trend as a rate of change, with a positive value as an increasing trend and a negative value as a decreasing trend. The two test types generate slightly different results, but were mostly consistent. Both indicate increasing bacteria trends upriver, at Kingston and Aylesford. The non-parametric tests also produced a decreasing bacteria trend at Lawrencetown. Both methods display a decreasing DO trend upriver, especially at Kingston and the non-parametric tests show a decreasing trend at Lawrencetown. Small increases in pH were found at Kingston, Middleton and Lawrencetown using non-parametric analysis, while parametric results did not show any trend. No temperature trends were found using the parametric methods. The non-parametric tests showed an increasing temperature trend at Kingston and Lawrencetown. No nutrient trends were shown for either nitrogen or phosphorus at Wilmot. The Aylesford Road site has only been monitored since 2003, and was only monitored sporadically until 2006, so there is not a large amount of data for that location. The data produced confidence intervals with a wide range; therefore no trends could be concluded. Similarly, nutrient data has only been collected since 2006, usually with 6 – 8 samples at one location per year, which may be why no trends were concluded for these parameters.

When compared to the results of the 2009 trend analysis, the results are fairly consistent. The non-parametric method indicated the same trends were present for DO and Temperature in 2010 as in 2009, but with slightly different magnitudes. In 2010, there was an increasing trend for bacteria count in Aylesford, which is expected from a p value just above 0.05 in 2009. pH increased at three sites in 2010, however no trends were determined in 2009. The parametric methods indicated the same trends for bacteria count, DO, pH and temperature in 2010 as found in 2009. Because of the presence of the serial dependence, it was not possible to conduct trend analysis for all of the sites as a single data set.

Recommendations

Summary of Recommendations for the River Guardians Program

- Complete the Quality Assurance Project Plan for all of CARP's Water Quality monitoring programs.
-
- Continue regular River Guardian *E. coli* monitoring at the eight main river sample locations.
- Contact livestock owners addressing the issue of restricting animals from the Annapolis River.
- Investigate correlation between precipitation amounts and *E.coli* levels in the river.
- Continue regular River Guardian DO monitoring program at the eight main river sample locations.
- Undertake periodic 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.
- Continue regular River Guardian temperature monitoring program at the eight main river locations.
- 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.
- Regular pH monitoring should be continued at the eight Annapolis River Guardian monitoring locations.
- Investigate pH drop in Lawrencetown.
- 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.
- Continue Event sampling after high precipitation amounts
- 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.
- Resume sampling locations in Aylesford for *E. coli* next season using an analysis method other than Coliscan Easygel.
- Review current and historic air photos of the Aylesford area to identify land use changes and possible sources of contamination.
- Conduct a foot survey along the Annapolis River between Victoria Road and Aylesford Road as well as along Patterson, Parker and Skinner brooks to identify possible contamination sources.
- Research and implement a more definitive test for autocorrelation.
- Regularly perform volunteer training and overview before each season to ensure proper technique and sampling consistency
- Implement QA/QC techniques throughout the entire sampling season

Recommendations for CARP

- Complete the Quality Assurance Project Plan for all of CARP's Water Quality monitoring programs.
- Calibrate the Quanta Hydrolab every two or three weeks for pH, conductivity, and dissolved oxygen.
- Create a manual for the River Guardian facilitator to ensure consistency in analysis and reporting.
- Ensure QA/QC protocols are implemented yearly throughout the entire sampling season, including an information session before the first sampling date.

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Appendices

Appendix A – Parameters Tested and Methodologies

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

Parameters Analyzed in 2010	Additional Parameters Analyzed in Previous Years of the Program
E. coli bacteria densities	Salinity
Dissolved Oxygen	Chlorophyll a
Temperature (Water and Air)	Nitrate-N, Chloride, Sulphate, Total Phosphate
Weather conditions	Colour
pH, Conductivity, Total Dissolved Solids	Transparency
Nitrate, Nitrite, Phosphate	
Total Suspended Solids (TSS)	
Turbidity	

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 and used during the 2004 through 2010 seasons. The sampling units (Figure A1) allow for representative sampling from mid-span of bridges at the sampling sites.

Figure A1. Collection unit used for fecal coliform samples in 2010.

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.

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. Further information on the comparison of the two testing methodologies is presented in the 2005 Annapolis River Guardian Report, Appendix C, which is available at the CARP office.

All E. Coi bacteria samples aresubmitted 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 2004 sample season, analysis was performed using the membrane filtration method.

Dissolved Oxygen Content

Dissolved oxygen samples are collected from the mid-span of bridges using a horizontal van Dorn sampler, at a depth of 1 meter. Dissolved oxygen in mg/L is determined using the modified Winkler titration using pre-packaged Hach reagents. The Winkler titration procedure is a widely recognized standard for determining dissolved oxygen. The procedure is reported to have an accuracy of at least +/- 1 mg/L. Dissolved oxygen as percent saturation is determined using Rawson's nomogram. Further information on the collection and analysis procedure for dissolved oxygen can be found in the Annapolis River Guardians Procedure Manual, which is available at the CARP office.

Temperature

Van Dorn samplers collect water at 1m depth, and temperature readings are immediately taken directly from the Van Dorn or from a 1L plastic bottle. The Annapolis River Guardians used a combination of glass/alcohol and digital thermometers during 2010. Prior to the start of the season, all thermometers were compared with the temperature reading from CARP's HydroLab Quanta water meter. This unit had recently been serviced and calibrated, with a reported accuracy of +/- 0.10 ^{o}C. From this comparison, a correction factor was determined for each River Guardian thermometer. These correction factors were applied to all River Guardian temperature measurements.

pH and Conductivity

Water chemistry data, including pH and conductivity, was collected using CARP's portable HydroLab Quanta water quality monitoring meter. Data was collected on a biweekly basis by CARP staff, typically the day following the volunteers' sampling day, at a set location on the riverbank at each River Guardian site. The meter was placed in the river approximately 1 to 2 meters away from the bank, and allowed to stabilize, usually for two to three minutes before a reading was taken. Once stabilized, the values were stored in the meter's memory and recorded on the data sheets upon return at the CARP office. The data is stored using an in-house Microsoft Access database. the multi-sensor water meter was calibrated for pH, conductivity and dissolved oxygen approximately every two to three weeks according to the directions in the Operating Manual (Hydrolab Corporation, 2002).

Procedures for TSS/Turbidity collection and processing

Samples were taken using either van Dorn Samplers or by hand from the shore near the bridge. If a van Dorn sampler was used, it was used mid-span of the bridge at a depth of approximately 30 to 60 cm. If taken from the shore, the bottle was dunked in an area where the water's flow was constant and at a depth the length of a forearm, approximately 30 to 40 cm. The collection method was not recorded for particular samples. A collection of approximately 1 litre of water was attempted for each collection. Field Turbidity was assessed using the Quanta Hydrolab at the time of collection. The collection sites for after April 2010 were NS01, 25, and 40. Some collection at other River Guardian sites occurred earlier in the year.

TSS data was processed through filtration. Filters were stored in a desiccator for at least 24 hours and were then weighed in a weighing boat on an analytical balance. The weight of the filter paper and the weighing boat together were recorded on the weighing boat. The filters used were Ahlstrom brand, grade 161, 4.7 cm in diameter, or Whatman brand, grade 934AH, 4.7 cm in diameter. The water sample was passed through one of the pre-weighed filters using a suction filtration procedure. The filter paper was carefully placed back in its weighing boat and dried in an oven at ~90 degrees Celsius before being stored back in the desiccator. After remaining in the desiccator for approximately 24 hours, the filters and boats were removed and reweighed. The original weight was subtracted from the new weight of the filter and boat, and this number was divided by the sample volume to give a g/L TSS reading. The balance used was an A&D Electronic Balance ER-120A.

Appendix B – Sites Monitored

Water samples were collected during 2010 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)
AY40	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, while NS01 was also sampled for TSS and turbidity.

Appendix C – Quality Assurance / Quality Control Data

Introduction

Following a bacterial contamination event in 2003, CARP initiated a number of procedures to ensure the quality of data collected. In addition to instituting a new collection procedure for fecal bacteria, CARP has put in place a program of regular quality control checks on sampling equipment and methods. Further information on the quality assurance/quality control (QA/QC) program can be found in CARP's draft QA/QC Project Plan (Sharpe and Sullivan, 2006). An important initial step in the QA/QC program is the training of volunteers. CARP staff conducted visits with each of the Annapolis River Guardian volunteers on collection days in order to both collect a series of blank and split samples, as well as to ensure the consistency in collection procedures. In total, twenty-two QA/QC samples were collected during the 2010 season. These were, in summary:

- 5 Dissolved oxygen split samples
- 4 *E. coli* travel blanks
- 5 *E. coli* duplicate samples
- 7 *E. coli* field samples
- 3 split turbidity/TSS samples

Background

For the purposes of CARP's water quality monitoring programs, a blank sample is a sample that is known not to contain any of the substance in question. For CARP's monitoring of *E. coli* bacteria, either distilled or un-chlorinated tap water is added to the sample bottle. There are two types of blank samples that are collected for QA/QC analysis:

- Travel blanks are obtained by filling the sample bottle with distilled/tap water before the start of a sampling day, and placing them in the same cooler among other surface water samples. Travel blanks are used to ensure there is no cross-contamination between samples while they are being transported in the same cooler. They should always produce plates with no fecal bacteria growth.
- Field blanks are obtained by performing the entire sampling protocol (i.e.: attaching the bottle to the clamp, removing the cap and lowering the apparatus to the water surface) but NOT submerging the bottle. The bottle is instead lifted up empty and filled with distilled/tap water on the bridge. This type of blank sample is used to test the sampling procedure and should also always produce plates with no fecal bacteria growth. A positive result on a field blank would lead to further investigations to determine the source of contamination (for example: operator, equipment, distilled water, etc).

Split samples are used to measure both precision and accuracy. Precision is expressed as the degree of agreement among repeated measurement of the same parameter and provides information on the reproducibility and consistency of the methods used. Accuracy, on the other hand, consists of how close a measurement is to the "true" value.

A split sample is a single sample volume that is divided in two samples that are analysed separately. Split samples can provide information on the precision of the lab method (i.e.: the precision of Valley Regional Hospital's *E. coli* analysis). Split samples can also provide information on the accuracy of the method used (i.e.: the accuracy of volunteers at the Winkler titration compared to staff).

The degree of variability between two split samples can be evaluated by calculating their relative percent difference (RPD). The RPD is expressed as the absolute difference of the two measurements multiplied by 100 and divided by the average of the two values:

$$RPD = \frac{|X_1 - X_2| \times 100}{(X_1 + X_2)/2}$$

When more than two samples are to be compared, the degree of variability is estimated by calculating their Relative Standard Deviation (RSD). Both the RPD and the RSD are expressions of precision, the smaller the value, the greater the precision.

$$RSD = \frac{s}{X_m} \times 100$$

s = standard deviation
X_m = mean of duplicate samples

Accuracy is estimated by taking the absolute difference between the "true" value and the "test" value. When there are multiple measurements, the true value is subtracted from the average of the test measurements. The result is compared to acceptable accuracy standards for each individual method. The staff value is considered the "true value" for the purpose of comparison.

Accuracy = Test/Average value – True Value

Dissolved Oxygen

Dissolved oxygen split samples were taken in 2010 using a single volume of water from a van Dorn sampler. The accuracy of volunteer DO measurements was assessed through the collection of seven split samples. The Winkler titration (described in Appendix A) is widely recognized has a standard for determining dissolved oxygen and is reported to have an accuracy of at least +/- 1 mg/L. Results from the split samples (Table C1) indicate that the volunteers attained an average accuracy of +/- 0.406 mg/L (RPD = 4.5%). For comparison purposes, the average DO accuracy for 2009 was +/- 0.38 mg/L (RPD = 4.2%).

Table C1. Volunteer's level of accuracy at measuring dissolved oxygen using the Winkler titration.

Site #	Date	Volunteer Result	QA/QC Result	Accuracy	% Difference
49	17-Oct-10	9	9.4	0.4	4.35
40	17-Oct-10	9.52	9.74	0.22	2.28
35	03-Oct-10	7.83	7.58	0.25	3.24
25	03-Oct-10	9.8	8.7	1.1	11.89
18					
13					
00	19-Sep-10	8.2	8.26	0.06	0.73
AY40					
			Mean	0.406	4.50

E. coli Bacteria

Throughout the sampling season, a series of blank samples were submitted blind for analysis to the microbiology laboratory at Valley Regional Hospital. The four travel blanks analyzed all had coliform counts of 0 cfu/100ml, which indicates that no cross-contamination was occurring during transportation of the samples. A field blank was collected at each River Guardian site and all eight samples showed 0 cfu/100m, indicating that the sample collection procedure was not contaminating the samples.

Throughout the 2010 sampling season, a total of five split samples were collected during the sampling visits with the volunteers. These samples were submitted to the Valley Regional Hospital Microbiology Laboratory under fictitious sample identification numbers. The purpose of this was to assess the reproducibility of the *E. coli* MPN analysis method used. The mean RPD for these split samples in 2010 was 25.0% (Table C2). The mean RPD for the 2008 and 2009 seasons was 23.3% and 28.8%, respectively.

The 2009 RPD mean is slightly higher than the 2008 value, with the 2010 value in between the two. This seems to indicate that the testing precision has been consistent the last few years. The test performed is the Colilert Most Probable Number analysis, and it is performed at the Microbiology Laboratory at Valley Regional Hospital.

Table C2. Relative percent difference in duplicate samples analysed for fecal coliforms.

Site #	Date	Volunteer Result	QA/QC Rsult	Accuracy	% Difference
49	17-Oct-10	167	206	39	20.91
40	17-Oct-10	250	166	84	40.38
35	03-Oct-10	299	206	93	36.83
25	03-Oct-10	154	152	2	1.31
18					
13					
00	19-Sep-10	1120	866	254	25.58
AY40					
			Mean	94.4	25.00

All analysis methods have inherent variability; this is particularly the case with IDEXX, as the Most Probable Number result is statistically derived (Table C3). The variability values are taken from the IDEXX Quanti-Tray®/2000 MPN Table (per 100mL) with 95% Confidence Limits (no date). For each volunteer result, the 95% confidence range was found and compared to the confidence range of the QA result. If these ranges overlapped, then the variability between the two results can be explained by the inherent variability of the procedure. None of the volunteer results had a value whose confidence range that did not overlap with that of the QA result, however, the Paradise volunteer results just bordered the 95% confidence interval of the QA/QC result.

Table C3. Confidence interval limits for IDEXX Colilert Most Probable Number procedure.

	95% Confidence	
MPN	Lower Limit	Upper Limit
0	0	3.7
10	5	18
50	36	69
100	81	121
150	124	181
200	166	242
500	405	619
1000	740	1320
1500	1010	2350
2000	1220	3300
>2419	1440	infinite

Turbidity/TSS QA/QC

Split samples were used in order to perform QA measurements for TSS and turbidity in 2008-2010. In 2008-2009, samples were taken from volunteer van Dorn samplers, and in 2010, split samples were analyzed for TSS from the same collection. Results from the 2009 collection season are included. (Tables C4 and C5).

Table C4. Relative percent difference in duplicate samples analysed for total suspended solids.

Site	Date	Volunteer result	QA/QC result	Accuracy	Percent difference
49	18-May-09	7.40	7.01	-0.39	5.41
40	18-May-09	5.29	10.41	5.12	65.22
35	18-May-09	9.87	7.37	-2.5	29.00
18	31-May-09	2.17	2.89	0.72	28.46
00	31-May-09	2.94	3.41	0.47	14.80
13	20-Sep-09	-2.22	-1.37	0.85	47.35
			Mean	0.7117	31.709

Table C5. Relative percent difference in duplicate samples analysed for turbidity.

Site	Date	Volunteer result	QA/QC result	Accuracy	Percent difference
49	18-May-09	32.10	20.40	-11.7	44.57
40	18-May-09	52.10	24.80	-27.3	71.00
35	18-May-09	23.50	17.40	-6.1	29.83
18	31-May-09	3.16	2.86	-0.3	9.97
00	31-May-09	4.36	4.15	-0.21	4.94
13	20-Sep-09	2.98	2.89	-0.09	3.07
			Mean	-7.617	27.228

A. Cook of Environment Canada suggested that negative TSS readings could be related to not pre-washing the filters (pers. comm., February 2010). Filters can contain soluble materials that are washed out during filtration, which would contribute to the loss of mass of the filter. Also, TSS and turbidity sampling both have a high degree of inherent variability, especially at higher numbers.

For the TSS and turbidity samples, the type of weighing boat used had a large effect on the results, which is reflected in the blank sample results (Table C6). No weighing boats were used for the first month of sampling in 2008. After that, plastic boats were used until spring of 2009, after which aluminum boats were used. These procedural blanks produced variable results, which may be related to soluble materials in the filter paper.

Table C6. Average results for blank TSS and turbidity samples, organized by weigh boat type.

Boat type	TSS (mg/L)	Lab turbidity (NTU)
None	-0.80	0.597
Plastic	0.04	
Aluminum	-2.47	