

Investigation of the Effect of Treated Wastewater on River Temperature and pH

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Abstract

The Squamish Wastewater Treatment Plant (WWTP) utilizes bacteria to biodegrade organic matter and sulphur dioxide for the process of dechlorination, which increase the temperature and reduce the pH of the treated water, respectively. This process can modify the temperature and pH levels of effluent water flowing back into rivers, resulting in detrimental consequences for keystone species in B.C., particularly salmon. This study aims to compare temperature and pH both upstream and downstream of Squamish's WWTP to identify the impact of the treatment process on river properties. We hypothesized that the treated wastewater released from the outflow site would (1) lower river pH and (2) increase river temperature. Using a pH probe and a thermometer, pH levels and temperature were measured 100 feet upstream ($n=5$) and 100 feet downstream of the WWTP outflow site ($n=5$). Collecting data from two separate trials, a two-sample t-test indicated that pH levels upstream and downstream were significantly different in both trial 1 ($p = 0.0363$) and trial 2 ($p = 0.0000343$). Furthermore, the two-sample t-test conducted on the temperature data indicated that the mean temperature difference between the upstream and downstream sites was statistically insignificant in both trial 1 ($p = 0.2826$) and trial 2 ($p = 0.2844$). In conclusion, our findings supported our pH hypothesis and failed to support our temperature hypothesis. This observed alteration of river pH can have adverse implications on salmon survival at all stages of life.

Introduction

Maintaining river pH and temperature levels are vital for British Columbia's aquatic ecosystems and salmon industry. As mentioned by Clark and Bonham (2), British Columbia's rivers are particularly sensitive to acid-base inputs, possessing a low pH buffering capacity, while also being highly susceptible to small changes in temperature. Consequently, this experiment aims to investigate differences in mean river temperature and pH both upstream and downstream of the Squamish Wastewater Treatment Plant (WWTP).

Generally, water downstream of WWTP outflow sites is more basic relative to its upstream counterpart because of the use of nitrogen-fixing bacteria that convert toxic

nitrogenous compounds, such as ammonia (NH_3), into innocuous nitrate (NO_3^-) (Cho et al. 699). As a result, alkaline compounds are added to achieve the optimal pH (7-8) for bacterial activity, resulting in higher river pH (Cho et al. 699; Morrison et al. 479). Similarly, wastewater temperature is often raised to 25-35°C to optimize nitrogen-fixing bacteria in WWTP treatment processes, resulting in higher river temperature (Tchobanoglous et al. 55). Despite the known effects of wastewater treatment on river pH and temperature from previous studies, the Squamish WWTP specifically utilizes a dechlorination process that reduces pH instead of using alkaline additives to increase pH (District of Squamish, 34). These wastewater induced changes are concerning considering the ecological impact on keystone species, such as salmon.

British Columbia's most predominant fish, salmon, play an essential role in transporting nutrients from the ocean to river, fertilizing forests, and serving as a food source for nearly 137 species (McPhail & McPhail 121; Howk). Alterations in river pH and temperature, however, have historically shown adverse effects on salmon proliferation (Lee 3240). As indicated by Daye and Garside (1717), salmon embryo mortality progressively increases as surrounding water pH begins to decline. On the other hand, alkaline water causes gill and fin damage, impairing respiratory function and swimming ability in salmon. The aforementioned damages severely limit their ability to escape predators, lowering species survival (Lease et al. 497). Furthermore, increases in water temperature are coupled with higher metabolic rates, resulting in faster depletion of environmental oxygen. Consequently, this forms local oxygen dead zones, resulting in mass casualties of aquatic organisms (Lee 3240).

We hypothesized that if the Squamish WWTP raises the temperature for greater microbial activity to enhance organic matter decomposition, then downstream river water will be higher in

temperature than upstream. The null hypothesis for our statistical analysis states the difference in mean water temperature between the upstream and downstream sites is statistically insignificant. In addition, we hypothesized that if the Squamish WWTP utilizes sulphur dioxide in their process of dechlorination, then the downstream river water will have a lower pH because the reaction between sulphur dioxide and water produces sulphurous acid. Similarly, the null hypothesis for pH states the difference in mean water pH between the upstream and downstream sites is insignificant.

Methods

We conducted our experiment at the Squamish River that neighbours the Wastewater Treatment Plant in Squamish, B.C (Figure 1). To begin, we located the outflow site that was marked with a sign along the water bank. The water outflow pipe was not visible as it was situated underneath the water.

Using distilled water and standard buffer solutions of 4.00 pH, 6.86 pH, and 9.18 pH, in the form of calibration powder, we calibrated our pH probe as preparation for data collection.



Figure 1. Photograph of the Squamish River at our downstream testing site, located 100 feet away from the WWTP outflow site. This was photographed during trial 1.

After locating the WWTP outflow site, we marked our sampling sites 100 feet upstream and 100 feet downstream of this location with masking tape. At the upstream site, we collected $\frac{1}{2}$ cup of water for our sample approximately 2 meters into the river, perpendicular to the shore.



Figure 2. Photographs of Simran Shergill collecting pH data from a sample downstream using a pH probe (on the left) and temperature data using a thermometer (on the right). These are located at the Squamish River, 100ft downstream of the WWTP outflow site.

For this sample, we measured the pH using a pH probe, and roughly confirmed the pH readout by testing the sample with litmus paper (Figure 2). We then measured the temperature of the water sample with a digital thermometer (Figure 2). Once data was recorded, we discarded the sample and cleaned the pH probe. We did this using distilled water, followed by drying with a tissue paper, to prevent cross-contamination between adjacent samples. We collected and tested an additional four samples at the upstream site following the same procedure. We then repeated this data collection procedure at the downstream site, where we collected and tested a total of five water samples.

Two days after the first trial, we conducted a second trial of this experiment at the previously marked upstream and downstream sites. Before data collection for the second trial, we ensured that the pH probe was once again calibrated to account for potential reading errors.

We used Microsoft Excel to perform all statistical analyses for this experiment. Before beginning data analyses, we ensured data was normal for accurate statistical testing, which it was found to be. We determined the normality of the data using the descriptive statistics tool in Microsoft Excel. For trial 1, we performed a two-sample t-test comparing the data for the temperature of the upstream samples with the downstream samples. We then performed a two-sample t-test comparing the data for the pH of the upstream samples and the downstream samples. We then repeated these statistical analyses for trial 2

Results

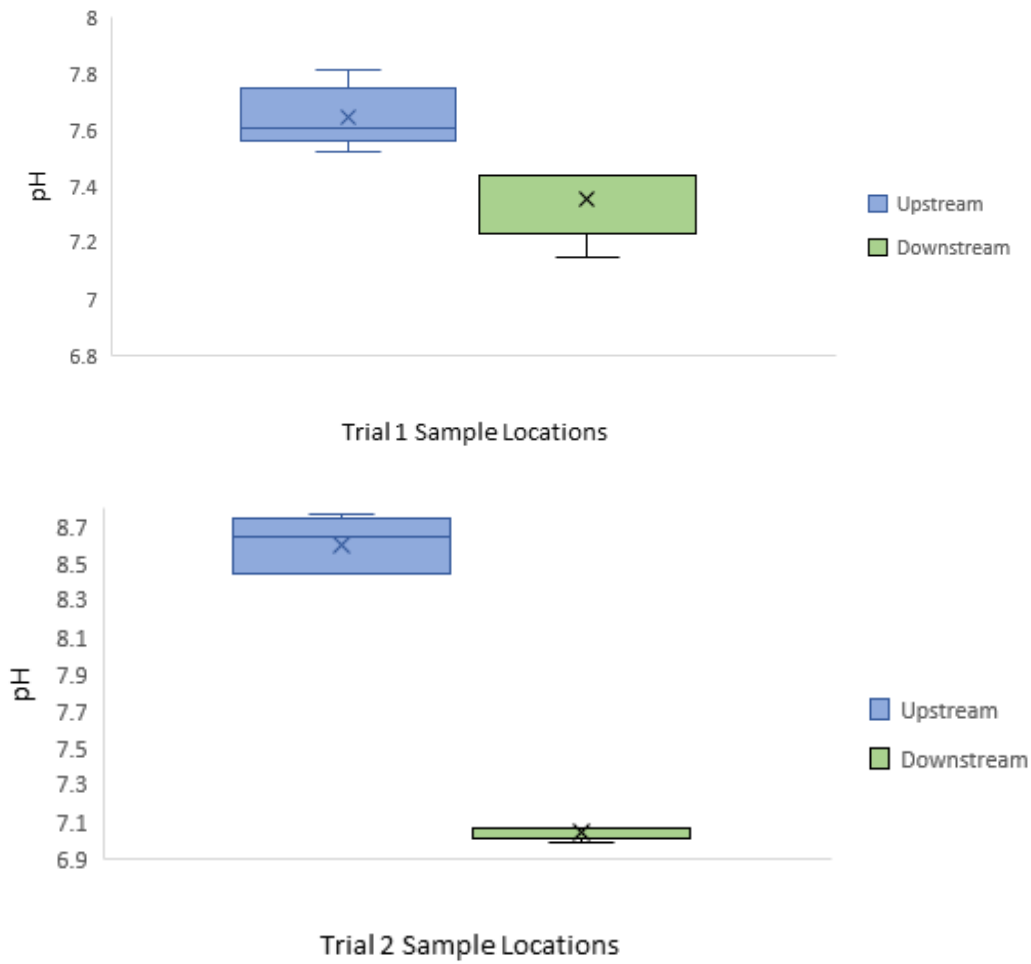


Figure 3. The pH of the Squamish river was sampled at two distinct sites, 100 feet upstream and downstream of the Squamish WWTP. A pH probe was used to measure the pH of the samples in each location (n=5). These boxplots show the median, mean, interquartile ranges, and maximum and minimum values for each sample location. The upper edge and lower edge of the boxes represent the 75th and 25th percentiles, respectively. The horizontal line within the boxes indicates the median, whereas the X corresponds to the mean. The upper and lower whiskers (error bars) extend to the minimum and maximum values found within the data set. In trial 1, it was found that the difference in mean pH between the upstream and downstream location was

statistically significant ($p = 0.0363$). Similar results were found in trial 2 with a p-value of 0.0000343.

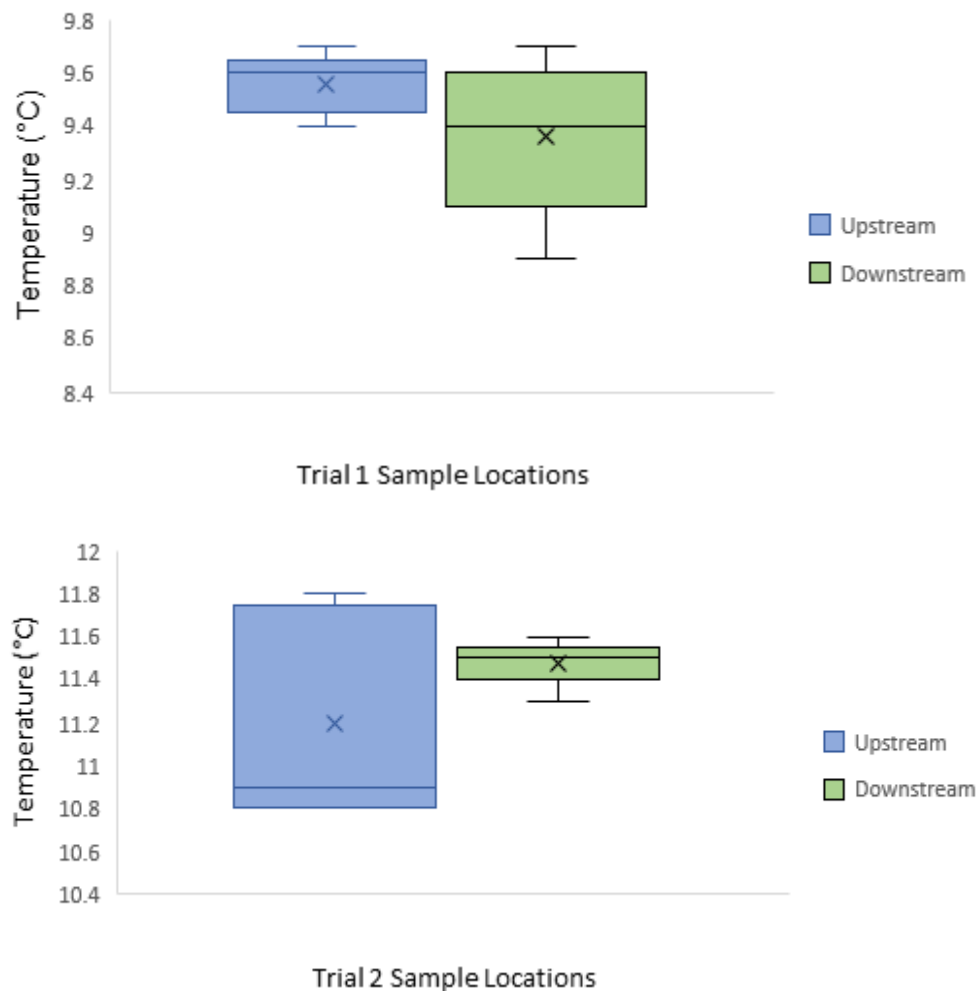


Figure 4. The temperature of the Squamish river was sampled at two distinct sites, 100 feet upstream and downstream of the Squamish WWTP. A digital thermometer was used to measure the temperature of the samples in each location ($n=5$). These boxplots show the median, mean, interquartile ranges, and maximum and minimum values for each sample location. The upper edge and lower edge of the boxes represent the 75th and 25th percentiles, respectively. The horizontal line within the boxes indicates the median, whereas the X corresponds to the mean. The upper and lower whiskers (error bars) extend to the minimum and maximum values found within the data set. In trial 1, it was found that the difference in mean temperature between the

upstream and downstream location was not statistically significant ($p = 0.2826$). Similar results were found in trial 2 with a p-value of 0.2844.

pH:

In trial 1, the mean pH value for the upstream site was evaluated to be 7.648, whereas the downstream site had a mean pH of 7.358. In comparison, trial 2 showed a much larger discrepancy between the upstream and downstream pH means. The upstream site in trial 2 had a mean pH of 8.604, whereas the downstream site had a mean pH of 7.046. Further statistical analysis was conducted on the Microsoft Excel platform. The descriptive statistics tool on Microsoft Excel showed the mean, median, and mode were approximately the same which indicates that the collected data was normal. Then a two-sample t-test was performed to determine whether the difference in the mean water pH of the downstream and upstream site was significant. Both trials yielded a p-value less than 0.05 (α), with trial 1 producing a p-value of 0.0363 and trial 2 producing a p-value of 0.0000343.

Temperature:

In trial 1, the mean temperature of the upstream site was calculated to be 9.56°C, whereas the mean temperature of the downstream site was calculated to be 9.36°C. These values did not show much discrepancy. For trial 2, the mean temperature for both sites also displayed similar consistency. The mean temperature of the upstream site for trial 2 was found to be 11.20°C, and the mean temperature of the downstream site was found to be 11.48°C. The same statistical analyses were performed on the temperature data set as the pH data set. Both trials yielded a p-value greater than 0.05 (α), with trial 1 generating a p-value of 0.2826 and trial 2 generating a p-value of 0.2844.

Discussion

We hypothesized that treated wastewater released from Squamish's WWTP outflow site would alter the pH and temperature of the downstream river because of specific treatment processes. Generally, wastewater treatment involves raising water temperature for enhanced biodegradation and utilizing pH-reducing compounds to remove chlorine (District of Squamish 34; Cho et al. 687). Results from the two-sample t-tests performed on the pH data for trial 1 ($p = 0.0363$) and trial 2 ($p = 0.0000343$) rejected our null hypothesis of the difference in mean water pH of the downstream and upstream locations being insignificant, suggesting differences in acidity between both locations. In contrast, for trials 1 ($p = 0.2826$) and 2 ($p = 0.2844$), similar statistical analyses for temperature data supported our null hypothesis that differences in mean water temperature of the downstream and upstream sites were insignificant. These results were determined using the conventional 95% confidence level that most experimental analyses adhere to in the biological field.

Most studies suggest that WWTP's employ alkaline-dependent bacteria for biodegradation (Cho et al. 687). This effectively removes organic matter but requires the addition of alkaline additives (pH 7-8) to maximize bacterial activity (Cho et al. 699; Morrison et al, 479). Our findings contradict these studies and support our predictions as we found that downstream river pH was lower, which may be attributed to specific treatments used by the Squamish WWTP. In the drinking-water system, residual chlorine is required, but if left untreated, may cause harm to aquatic life in the receiving water (Brungs 2180). The Squamish WWTP dechlorinates wastewater using sulphur dioxide, which significantly reduces the pH of the treated effluent, and thus the receiving river water (District of Squamish 34). Research also shows that

WWTPs release warmer water at the outflow site because biodegrading bacteria have optimal functional temperature ranges of 25-35°C (District of Squamish 34). Contradicting this, we found the difference in mean temperature between both sites to be insignificant.

As a result, these specific changes in river pH can have severe implications on local salmon populations. River acidification can decrease salmon survival at the larval, adult, and reproductive stages. Firstly, egg hatching is severely impaired at low pH, where eggs can fail to hatch or have delayed hatching times (Peterson 773). Hatching complications result in higher rates of unsuccessful births and underdeveloped salmon that are more susceptible to early predation (Peterson 773). Secondly, feeding behavior is severely impaired at low pH, resulting in death through starvation or lowered metabolism (Moore 497). Finally, salmon pheromone detectors are progressively hindered with lowering pH. Chemical cues, such as testosterone or urine from an ovulating female, elicit reproductive responses which are severely reduced with impaired pheromone detection (Leduc et al. 8). Consequently, if the Squamish WWTP fails to neutralize treated water, this could lead to significant reductions in local salmon populations and prevent propagation into future generations.

In our experiment, it is important to consider the influence of variation and uncertainty on our findings. One source of variation in river pH between trial 1 and 2 could be attributed to the Squamish WWTP having specific outflow intervals for maximal dispersion of treated wastewater before subsequent release. Visible outflow solely in trial 2 is indicative of this outflow interval. Furthermore, pH may be varied due to wind-induced mixing. High wind intensity in trial 1 (14-16 km/h) could have effectively mixed the treated wastewater with normal inflowing water, thus keeping the pH relatively stable. However, in trial 2, where wind intensity was much lower

(6 km/h), poor mixing could have resulted in areas of varying pH. Moreover, unusually high pH readings in the upstream site in trial 2 could be attributed to pH probe calibration errors.

However, since the pH readout would have been consistent within trials, and the statistical analysis was performed within trials, this variation was not of major concern to our results.

Furthermore, the slight variation in temperature measurements could be the result of inconsistent recording methods. In some trials, the sample cup was placed on a cool, rocky surface, whereas in others, it was held by a student who could have transferred body heat to the sample. While there was variation in the individual readings, the high heat capacity of river water could have prevented large temperature fluctuations (Park et al. 6).

To minimize variation and uncertainty in future experiments, this study could be conducted with a larger sample size, more locations, strict procedural methods, and more accurate measuring tools. To account for changes in abiotic factors, the weather forecast can be consulted to plan data collection on days with similar temperature forecasts, wind speeds, light intensity, and cloud cover.

Conclusion

The results of this experiment were important as they provided insight into the implications of Wastewater Treatment Plants on salmon's ability to survive and ultimately contribute to future generations. This provides helpful information to the Squamish WWTP so they can consider tighter regulation of treatment processes to prevent drastic alterations in the river pH at the outflow site. Statistical analyses provided results that supported the hypothesis that wastewater outflow into the Squamish River would alter its pH, but did not provide results supporting our hypothesis that wastewater outflow would alter the river's temperature in the

immediate area. To strengthen the validity of our results, further testing can be done at additional Wastewater Treatment Plant locations to increase our sample size. A larger sample size and varying testing locations would allow us to extrapolate treatment-water induced pH and temperature changes rather than having our findings solely focus on the Squamish Wastewater Treatment Plant.

Acknowledgements

We would like to thank Dr. Celeste Leander and the teaching staff of BIOL 342 for setting up this virtual learning environment and patiently addressing the students' concerns, questions and needs. Additionally, we would like to thank UBC for providing us with the opportunity to take BIOL 342, especially in a virtual setting. Lastly, we would like to acknowledge the traditional, ancestral, and unceded territory of the Sḵwx̱wú7mesh and Stó:lō community on which we are learning, working and conducting our experiments.

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Appendix A: Raw Data

A1 - Table 1. Trial 1 data from the downstream site collected on March 22nd, 2021. The pH and temperature (°C) were recorded for each sample of water (n=5).

Sample #	pH	Temperature
Sample 1	7.15	9.7 C
Sample 2	7.32	9.5 C
Sample 3	7.44	9.4 C
Sample 4	7.44	8.9 C
Sample 5	7.44	9.3 C

A2 - Table 2. Trial 1 data from the upstream site collected on March 22nd, 2021. The pH and temperature (°C) were recorded for each sample of water (n=5).

Sample #	pH	Temperature
Sample 1	7.69	9.4 C
Sample 2	7.81	9.7 C
Sample 3	7.61	9.5 C
Sample 4	7.61	9.6 C
Sample 5	7.52	9.6 C

A3 - Table 3. Trial 2 data from the downstream site collected on March 24th, 2021. The pH and temperature (°C) were recorded for each sample of water (n=5).

Sample #	pH	Temperature
Sample 1	6.99	11.3 C
Sample 2	7.07	11.6 C
Sample 3	7.07	11.5 C
Sample 4	7.03	11.5 C
Sample 5	7.07	11.5 C

A4 - Table 4. Trial 2 data from the upstream site collected on March 24th, 2021. The pH and temperature (°C) were recorded for each sample of water (n=5).

Sample #	pH	Temperature
Sample 1	8.77	10.9 C
Sample 2	8.64	10.8 C
Sample 3	8.73	11.7 C
Sample 4	8.44	11.8 C
Sample 5	8.44	10.8 C

A5 - Table 5. The Computed pH and Temperature Averages for Both the Upstream and Downstream Locations in Trial 1

Location	Average pH	Average Temperature
Upstream	7.648	9.56
Downstream	7.358	9.36

A6 - Table 6. The Computed pH and Temperature Averages for Both the Upstream and Downstream Locations in Trial 2

Location	Average pH	Average Temperature
Upstream	8.604	11.20
Downstream	7.046	11.48

Appendix B: Two-Sample t-Test Results**B1 - Table 1:** pH two-sample t-test result (*p-value*) for both trial 1 and 2.

Trial	<i>p -value (Two-tail)</i>
1	0.0363
2	0.0000343

B2 - Table 2: temperature two-sample t-test result (*p-value*) for both trial 1 and 2.

Trial	<i>p -value (Two-tail)</i>
1	0.2826
2	0.2844

Appendix C: Microsoft Excel Descriptive Statistics Tool

<i>Column1</i>	
Mean	11.48
Standard Error	0.048989795
Median	11.5
Mode	11.5
Standard Deviation	0.109544512
Sample Variance	0.012
Kurtosis	2.916666667
Skewness	-1.293233816
Range	0.3
Minimum	11.3
Maximum	11.6
Sum	57.4
Count	5

C1 - Figure 1. Example output of the descriptive statistics tool from Microsoft Excel. Normality of the data can be determined by comparing the mean, median and mode. Approximately equal mean, median and mode values indicate that the data is normal.