Dissolved Oxygen and Temperature on the Viability of Spawning Salmon

Arrthy Thayaparan, Fatima Syed, Lisa Liang, & Nicole Dodge

Abstract

To observe how dissolved oxygen (DO) levels and air and water temperature affect the viability of spawning salmon at Baker Creek (BC) in Quesnel, British Columbia and at Salish Creek (SC) in Vancouver, British Columbia. BC supports spawning *Oncorhynchus gorbuscha* (pink salmon) and SC used to support spawning Oncorhynchus kisutch (coho salmon). At each location, samples were taken to determine DO levels, air temperatures, water temperatures and a qualitative study of plant life at an upper and lower site at each creek was observed. The three null hypotheses are: DO levels will be the same at both BC and SC, water temperatures will be the same at both BC and SC, and air temperatures will be the same at both BC and SC. The alternative hypothesis is that DO levels and temperatures will be different at BC and SC. The data for DO levels, air temperatures and water temperatures was analyzed using a Welch's t-test in R Software. After this analysis, the three null hypotheses were rejected and the alternative hypothesis was accepted. DO levels were significantly higher at SC compared to BC and water and air temperatures at SC were significantly higher compared to BC. The relationship between temperature and DO level is illustrated by Henry's Law which indicates that colder temperatures equate to higher DO levels in water. Thus, our results illustrate an unexpected difference between expected DO levels and temperatures where pink salmon spawn in BC. At SC, DO levels were within a range to sustain coho salmon life, but the water temperature was outside of this range and may explain why spawning does not occur in SC.

Introduction

The purpose of our field study was to determine how dissolved oxygen level, air temperature, and water temperature affect the viability of spawning salmon. Dissolved oxygen (DO) is the amount of oxygen that gets diffused from the atmosphere into the water, and is extremely important for respiration and breaking down organic matter (Manahan, 2005). Salmon are a keystone species and are critical within their ecosystem (Manahan, 2005; Hyatt & Godbout, 1999). Humans depend on salmon as a source of food and a huge source of revenue for the Canadain economy (Fisheries and Ocean Canada, 2019). In 2017, commercial fishing contributed around \$100 million to the economy and British Columbia was the main contributor (Fisheries and Ocean Canada, 2019). Salmon are also culturally significant to Indigenous communities (Hutchings et al., 2012). Changes in temperatures and DO levels caused by increasing infrastructure, fisheries and climate change resulted in decreased wild salmon populations (Smith et al., 2011; Ohlberger et al., 2018).

Oxygen enters water predominantly via the atmosphere, but also via photosynthesis from surrounding plant life (Manhan, 2005). DO is used for respiration by organisms and DO levels are indicative of water productivity which is the water's ability to sustain life (Manhan, 2005). Appropriate DO levels can increase biodiversity within a body of water (Manhan, 2005). If DO levels become too high, eutrophication occurs, which results in DO being used up too quickly, thus creating critically low levels (Manahan, 2005). Different factors can affect DO levels, such as aeration, the introduction of air into the water by physical movement such as wind or turbulence, and photosynthesis (Manahan, 2005). Plants near streams produce oxygen which gets diffused into the stream (Manahan, 2005). Additionally, DO levels can decrease due to organic matter such as leaves, biological waste or organisms decomposing in the water (Manhan, 2005). Henry's Law states that the colder the temperature, the more soluble a gas (oxygen) is, causing the DO levels in water to increase with colder temperatures (Manhan, 2005). Furthermore, plants provide shade, which results in lower water temperature and higher DO levels (Manahan, 2005).

Salmon need specific DO levels and temperature ranges to survive and reproduce (Brett & Blackburn, 1981). After hatching, in order to survive their spawning migration from a freshwater environment to the ocean, salmon require large amounts of oxygen (National Park

Service, 2019; Eliason & Farrell, 2016). Spawning *Oncorhynchus kisutch* (coho salmon) prefer DO levels above 6.3 mg/L and water temperatures between 4.4-9.4 °C (McMahon, 1983). Spawning *Oncorhynchus gorbuscha* (pink salmon) prefer DO levels of 8 mg/L and water temperatures between 9.2-13.7 °C (Raleigh & Patrick, 1985). However, healthy salmon embryos require a higher level of DO (Brett & Blackburn, 1981). Coho embryos need DO levels above 8mg/L and a water temperature range between 4.4-13.3 °C (McMahon, 1983) . Pink salmon embryos require DO levels of at least 8 mg/L and a temperature range between 8-10 °C (Raleigh & Patrick, 1985). Anything below these levels causes salmon to eventually die from hypoxia (Carter, 2005; Remen et al., 2012). Hypoxia occurs when not enough oxygen reaches an organism's tissues (Carter, 2005; Remen et al., 2012).

A recent study found a discrepancy between the temperature and expected DO levels when spawning salmon were present (Fellman et al., 2018). Henry's Law can calculate the expected amount of DO in water by using the water's temperature (Manahan, 2005). Fellman et al. (2018) were expecting to determine a certain DO level given the temperature of the water, however, where salmon were spawning they found the DO level was not at the expected level (Fellman et al., 2018). As this was not the main focus of the study by Fellman et al. (2018), more research needs to be done in this area in order to test this discrepancy. The effects of changing temperatures on species due to climate change has resulted in more research in this area. Although research has been done on how warming temperatures affect pink salmon, very little is known about how DO levels affect pink salmon. Pink salmon seem resistant to warming temperatures and scientists predict that this is due to their cardiorespiratory system (Clark et al., 2011). Therefore, we wanted to build on the limited research done on how DO levels affect spawning pink salmon.

Our research question is, how does dissolved oxygen level and temperature affect the viability of spawning salmon? To test this we went to two different creeks, Baker Creek (BC) and Salish Creek (SC). At BC, there are currently pink salmon spawning. Historically, coho salmon spawned at SC, but they no longer do so (Vibert, 2019; Natural Resource Management Specialist, 2019). Testing DO levels and air and water temperatures, as well as qualitatively observing plant life at these locations, might give insight into salmon spawning viability at SC and illustrate if there are other factors controlling salmon spawning. Our three null hypotheses are: DO levels will be the same at both BC and SC, water temperature will be the same at both BC and SC. Our alternative hypothesis is DO levels and temperature will be different at BC and SC. We predict that BC will have higher DO levels since we expect temperatures to be colder because BC is further north compared to SC.

Methods

Dissolved oxygen levels and temperature were measured at two different creeks to evaluate their effect on the viability of salmon spawning. BC is located in Quesnel, British Columbia and the creek has active spawning of pink salmon. SC, in Vancouver, British Columbia, used to be one of the few streams that supported spawning coho salmon. However, due to urban development, Salish Creek is no longer used by spawning salmon. Due to the heavy influence of plant life on both dissolved oxygen levels and temperature, qualitative data of plant life between the two creeks was observed. Figure 1 is a map of British Columbia showing the locations of Quesnel and Vancouver.



Figure 1. Map of British Columbia with red dots indicating the approximate location of Baker Creek in Quesnel and Salish Creek in Vancouver.

Prior to the field experiment, random.org was used to generate randomized transect numbers. Five randomized transect numbers were assigned independently for the upper creek and lower creek of both BC and SC. The transect numbers generated for the upper creek of BC were 1 m, 2 m, 8 m, 10 m, and 12 m while the transect numbers generated for the upper creek of SC were 2 m, 3 m, 15 m, 16 m, and 20 m. The transect numbers generated for the lower creek of BC were 1 m, 2 m, 4 m, 8 m, and 16 m and the transect numbers generated for the lower creek of SC were 2 m, 3 m, 15 m, 16 m, and 20 m. The transect numbers generated for the lower creek of BC were 1 m, 2 m, 4 m, 8 m, and 16 m and the transect numbers generated for the lower creek of sC were 2 m, 3 m, 8 m, 15 m, and 20 m. Measurements of air temperature, water temperature and dissolved oxygen levels were taken at the same five sets of transect numbers corresponding to its location, which totaled to 10 replicates. As salmon spawning activity were located in the upper creek for both locations, we chose to compare the data of abiotic factors collected from the upper part of each creek.

Due to the fact that samples were being collected by different people at BC and SC, three of the following measures were used to standardize our measurements and avoid inconsistency. Firstly, the same thermometer was used to measure both air and water temperature at each transect at each of the creeks. Air temperature was measured before water temperature. Secondly, to take measurements of dissolved oxygen levels, the level recorded was the value on the Texas Instruments TI-84 calculator after it had stabilized and stopped changing. The last standardization was to take pictures of the surroundings of each creek and, as a group, qualitatively compare the difference between the plant density.

The experiment was conducted simultaneously at the two creeks at around 1pm Pacific Standard time on November 8th, 2019. Upon arriving at SC, we were not able to lay out a 20 meter long transect line at both the lower and upper creek without disrupting the surrounding life. As a result, we decided to restrict the transect length to 15 meters. Previously assigned transect numbers above 15 were randomly re-assigned through random.org. The new transect numbers at the upper creek of SC were changed to 1 m, 2 m, 3 m, 4 m, and 15 m. The new transect numbers at the lower creek of SC were changed to 2 m, 3 m, 8 m, 14 m, and 15 m. The transect numbers assigned for BC remained unchanged as the full 20 meters was fully accessible without disrupting the area.

Air temperature was taken by holding a Fisher thermometer directly above the assigned transect number for 30 seconds. Water temperature was taken by dipping approximately half of

the length of the thermometer into the stream at the assigned transect number and holding it in the stream for 30 seconds. In between each transect number, a kinwipe was used to dry off the thermometer before taking the readings of the next sample.

To measure dissolved oxygen levels, water samples from each transect were collected in five clear plastic cups. To ensure consistency, each sample was collected by fully submerging each plastic cup just beneath the water surface of each transect. After collecting the water samples, a Vernier dissolved oxygen probe was inserted into a Texas Instruments TI-84 calculator. Then the probe was dipped into the water sample in the plastic cup and swirled around for about 30 seconds, without touching the sides of the plastic cup. We recorded the oxygen level once the numerical value on the Texas Instruments TI-84 calculator had stabilized. The same process was repeated for each sample of water collected. To avoid cross contamination, we ensured that the probe and clear plastic cups were rinsed thoroughly with distilled water before reusing again for the next sample. Figure 2 is images of air temperature, water temperature, and DO levels being measured.



Figure 2. Images of air temperature, water temperature, and dissolved oxygen levels being measured at Baker Creek in Quesnel, British Columbia.

Plant life observation was done by taking pictures of the surroundings of each creek and comparing the plant life density. To identify possible differences in vegetation at the two creeks, tree and shrub dichotomous keys were used.

At each of the two creeks, there were 10 replicates for each of the abiotic factors. The means, standard deviations, t-values, degrees of freedom, and P-values were calculated using Welch's t-test in R software. These results were then inputted into Microsoft Excel to generate bar graphs.

Results

The average DO levels were calculated from a sample size of ten from the lower and upper stream of BC and from a sample size of ten from the lower and upper stream of SC. The mean DO levels and standard deviation were calculated using R Software. The mean DO level at BC was 8.82 mg/L with a standard deviation of 0.30. The mean DO level at SC was 9.14 mg/L with a standard deviation of 0.26. The t-value, degrees of freedom, and P-value for the average DO levels were calculated using Welch's t-test in R Software. The t-value is 2.56, with 17 degrees of freedom and a P-value of 0.0196. Since the P-value is less than 0.05, we reject the null hypothesis that oxygen levels are the same at BC and SC. Figure 1 compares the mean DO levels at BC and SC.



Figure 3. Mean oxygen levels for Baker Creek, Quesnel, British Columbia and Salish Creek, Vancouver, British Columbia with 95% confidence interval bars. The sample size for Baker Creek was 10 and the sample size for Salish Creek was 10. The t-value is 2.56, with 17 degrees of freedom and a P-value of 0.0196. Asterisks indicates significance difference between Baker Creek and Salish Creek, *p <0.05.

The average air temperature was calculated from a sample size of ten from the lower and upper stream of BC and from a sample size of ten from the lower and upper stream of SC. The average air temperature at BC was 6.50 °C with a standard deviation of 0 °C. The average air temperature at SC was 11.95 °C with a standard deviation of 0.16 °C. The t-value, degrees of freedom, and P-value for the average air temperature was calculated using Welch's t-test in R software. The t-value is 109.00, with 9 degrees of freedom, and a P-value of 2.33e-15. Since the

P-value is less than 0.05, we reject the null hypothesis that the average air temperature is the same at BC and SC. Figure 2 compares the mean air temperature at BC and SC.



Figure 4. Mean air temperature at Baker Creek, Quesnel, British Columbia and Salish Creek, Vancouver, British Columbia with 95% confidence interval bars. The sample size for Baker Creek was 10 and the sample size for Salish Creek was 10. The t-value is 109.00, with 9 degrees of freedom and a P-value of 2.33e-15. Asterisks indicates significance difference between Baker Creek and Salish Creek, *p <0.05.

The average water temperature was calculated from a sample size of ten from the lower and upper stream of BC and from a sample size of ten from the lower and upper stream of SC. The average water temperature at BC was 3.96 °C with a standard deviation of 0.08 °C. The average water temperature at SC was 10.25 °C with a standard deviation of 0.34 °C. The t-value, degrees of freedom, and P-value for the average water temperature was calculated using Welch's t-test in R software. The t-value is 54.96, with 9 degrees of freedom and a P-value of 9.08e-14. Since the P-value is less than 0.05, we reject the null hypothesis that the average water temperature is the same at BC and SC. Figure 3 compares the mean water temperature at BC and SC.



Figure 5. Mean water temperature at Baker Creek, Quesnel, British Columbia and Salish Creek, Vancouver, British Columbia with 95% confidence interval bars. The sample size for Baker Creek was 10 and the sample size for Salish Creek was 10. The t-value is 54.96 with 9 degrees of freedom and a P-value of 9.08e-14. Asterisks indicates significance difference between Baker Creek and Salish Creek, *p <0.05.

On the day of the field experiment, we observed that BC had less plant biodiversity than SC. The upper stream of BC had black cottonwood, white sweet clover, and russet sedge. The lower stream of BC had black cottonwood. Figure 6 is images of different plant species at BC.



Figure 6. a) Image of black cottonwood at the upper stream of Baker Creek, Quesnel, British Columbia. b) Image of russet sedge at the upper stream of Baker Creek, Quesnel, British Columbia. c) Image of white sweet clover at the upper stream of Baker Creek, Quesnel, British Columbia.

The upper stream of SC had holly trees, red alder, and sword fern. The lower stream of

SC had snowberry, western red cedar, and sword fern. Figure 7 is images of different plant

species at SC.



Figure 7. a) Image of sword fern at the lower stream of Salish Creek, Vancouver, British Columbia. b) Image of holly tree at the upper stream of Salish Creek, Vancouver, British Columbia. c) Image of red alder at the upper stream of Salish Creek, Vancouver, British Columbia.

Discussion

The aim of this study was to determine whether there were significant differences found in DO levels and temperature levels at BC and SC. These findings could then be used to establish why salmon spawn at BC and not at SC. Due to its location in northern British Columbia, we anticipated that BC would have lower air and water temperatures and therefore have higher DO levels.

Our alternative hypothesis is that the DO levels and temperature measurements will be different at BC and SC. Whereas, our null hypotheses is that there would be no difference in DO levels and temperatures between the two creeks. According to the data collected, we can conclude that the results found between BC and SC are significantly different from each other. Based on the results of Welch's t-test we can confidently reject both null hypotheses. Therefore, we accept the alternative hypothesis that there is significant differences in DO levels and temperatures between BC and SC. Our prediction that BC would have lower air and water temperatures was correct, but our prediction that BC would have higher DO levels than SC was incorrect.

The results differed from our prediction as BC had a DO level of 8.86 mg/L which was lower than the DO level of 9.14 mg/L at SC. We correctly predicted that BC would have a lower water temperature. The water temperature at BC was 3.96 °C compared to the water temperature at SC which was 10.25 °C. The difference in observed DO levels and the expected DO levels due to temperature is similar to the discrepancy found in the 2018 study by Fellman et al. The following discussion will further analyze the significance of our results to determine whether the results provide evidence to explain why salmon are viable in BC and not in SC.

During the data collection period, salmon were not spawning in either creek. Pink salmon spawn in BC during the fall season up until the end of October (Vibert, 2019). Currently, SC has no coho salmon returning to spawn. From our results, we hope to determine if differences in DO levels and temperatures at BC and SC contribute to pink salmon spawning in BC and no coho salmon spawning in SC.

DO levels around 8 mg/L are ideal for spawning salmon (Brett & Blackburn, 1980). BC and SC were both at this required level. However, while both creeks are at the ideal DO level, salmon only spawn at BC. When salmon are spawning, DO levels usually drop significantly due to the large oxygen intake from the salmon (Fellman et al., 2018). DO levels that fall below 4 mg/L will not be able to support salmon populations which results in spawning salmon not returning to those specific bodies of water (Brett & Blackburn, 1980). This may be the case of what happened at SC.

As the spawning season was over during the time of our data collection, we cannot determine if DO levels at BC were higher during spawning season. In accordance with Henry's Law, due to the lower temperature at BC, DO levels were expected to be much higher than what was found. The DO levels in BC were above 4 mg/L which is sustainable for pink salmon. Assuming that the DO levels were much higher before and during the spawning season, we can assume that the lower DO levels in BC, compared to SC, are likely due to oxygen usage by spawning salmon. However, further study is required to be confident in this reasoning.

If salmon were to spawn in SC, the immense usage of oxygen could potentially have DO levels fall below 4 mg/L, which would lead to negative effects in salmon behaviour and embryo development (Brett & Blackburn, 1980). However, further study is required to ensure that this is a possible reason behind the difference in salmon viability. Until more research can be done, we can only assume that coho salmon do not return to SC because of the effects of other abiotic factors, as the DO level was within researched ranges that would sustain salmon populations.

Water temperature levels between 5-17 °C are required for all salmon to survive (Lee et al., 2003). Coho salmon prefer a temperature range between 4.4-9.4 °C, while pink salmon prefer a temperature range between 9.2-13.7 °C (McMahon, 1983; Raleigh & Patrick, 1985). Based on our results, we can conclude that at the time of data collection, both creek sites fall out of the temperature range preferred by pink or coho salmon.

At BC, we are aware that the pink salmon spawning season comes to an end at the end of October (Vibert, 2019). The average air temperature in Quesnel in October 2019 was 9.22 °C

("Quesnel, British Columbia, Canada Monthly Weather", 2019). Therefore, though the water temperature at BC was not at the required temperature range for pink salmon embryo to survive, we cannot conclude that in October the water temperature was outside of the temperature range required for the survival of pink salmon embryos. Based on a study by Clark et al. (2011), pink salmon have shown to thrive regardless of drastic environmental changes. Although, BC's water temperature was not in the range that is required for pink salmon, it is not necessarily an unhealthy environment for spawning salmon as they are observed to return to BC yearly.

In comparison, SC is in the temperature range required for all salmon and is slightly above the preferred temperature range for coho salmon. While studies have shown that pink salmon are able to thrive regardless of temperature changes, coho salmon have not been studied to determine if they have that same aptitude. Therefore, we can hypothesize that the warmer temperatures at SC are a factor in the lack of spawning coho salmon at SC, but further study would be required to confirm this hypothesis.

Air temperature can affect water temperature and DO levels directly. When there is lower air temperature, based on Henry's Law, the water temperature will be colder and the DO levels will be higher due to oxygen being more soluble at lower temperatures (Manahan, 2005). However, in regards to our results this relationship was not clearly seen. Though BC had a much lower mean air temperature, the DO levels were lower in SC where the temperature was higher.

Lower DO levels in BC could be due to the effects of spawning salmon earlier in the season. Compared to SC, BC was observed to have less plant biodiversity near the stream. Therefore, DO levels at SC could be higher due to oxygen output from the thick foliage near the stream. Specifically, BC had bare deciduous trees in the distance and two small plants close to

the water, whereas SC had both coniferous and deciduous trees nearby and many shrubs and ferns close to the water. The drastically different composition of plant life in the two areas further substantiates a possibility in the difference between the DO levels. Though we would need further study to measure how DO levels are impacted by plant biodiversity and plant density.

We are able to address the discrepancy of DO levels between BC and SC by proposing that DO levels were not at expected levels given the temperature and the oxygen intake by spawning salmon and other organisms in Baker Creek. While we are unable to confidently prove that this is the reason behind the anomaly, further study and data collection before and during spawning season may provide further evidence.

The results indicate that there is no clear explanation for the difference in viability for spawning salmon between BC and SC. Additional research with comparisons to other bodies of water can possibly conclude whether the lack of salmon in SC and the presence of salmon in BC is due to the measured DO levels and temperatures.

While collecting data for our studies, our group had to divide into two groups so that we could take measurements on the same day and at the same time. In doing so, there could be differences in the equipment used and in the collection methods due to unconscious bias. To minimize errors, our group practiced together in the lab before our collection day and asked the lab technicians how to operate the easy pro link probe to ensure that the calibration was the same for the equipment. In future studies, we would recommend having more replicates and vary the times of data collection to before, during, and after spawning season. In doing so, this would help to verify the theories we have discussed as well as further substantiate our results.

Conclusion

Based on our results, we can conclude that there is a difference in the DO levels and air and water temperatures found between BC and SC. Therefore, we reject both of our null hypotheses stating that the DO level and temperature will be the same at BC and SC, and accept our alternative hypothesis that the DO level and temperature will be different at the two sites. In our discussion, we have analyzed possibilities as to why these results are varied in these two different locations. DO levels in BC and SC varied significantly and this may be due to pink salmon spawning in BC and/or the difference in foliage density. Water and air temperature in BC and SC varied significantly due to differences in geographical location. However, the exact impact this had on the creeks and DO levels cannot be verified without further study. Our study had a discrepancy in regards to DO levels not being as high as expected for the temperature measurements at BC. We believe this may be due to spawning salmon consuming large quantities of DO. Our results fail to explain why pink salmon spawn in BC based on the temperatures being outside of the range of 9.2-13.7 °C which is required for pink salmon survival. To investigate this further, DO level and temperature measurements need to be taken during spawning season. A possible reason coho salmon are not spawning in SC is due to a high water temperature. A study of DO levels and temperature needs to be replicated at a time when coho salmon historically spawned in SC. Although we found a significant difference between DO levels and air and water temperatures between SC and BC, we cannot use this difference to conclude why pink salmon spawn in BC and coho salmon do not spawn in SC.

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Appendix

Lower creek time BC: 1:30 pm Upper creek time BC: 2:01 pm

Lower creek time SC: 2:15 pm Upper creek time SC: 1:15 pm

Change in Methods: the transect lines at both the upper and lower stream of SC was not able to be a full 20 m, changed to 15 m (both) new transect numbers from random.org *=change

1	1 m	2 m	8 m	10 m	12 m
BC Upper [O2] mg/L	8.5	8.9	9.5	9.5	9.6
2	1 m*	2 m	3 m	4 m*	15 m
SC Upper [O2] mg/L	8.7	8.1	7.7	7.8	7.9
3	1 m	2 m	4 m	8 m	16 m
BC Lower [O2] mg/L	8.7	9.3	8.2	9.0	7.0
4	2 m	3 m	8 m	14 m*	15 m
SC Lower [O2] mg/L	9.4	9.6	10.4	10.5	11.3

Table 1. Oxygen Concentrations at Upper and Lower stream of BC and SC

Table 2. Wat	er Temperature	at the Upper a	nd Lower Stream	s of BC and SC
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1	1 m	2 m	8 m	10 m	12 m
BC Upper Water Temp. (°C)	4.0	4.0	4.0	3.8	3.8

2	1 m*	2 m	3 m	4 m	15 m
SC Upper Water Temp.(°C)	10	10	10	10	10
3	1 m	2 m	4 m	8 m	16 m
BC Lower Water Temperature (°C)	4.0	4.0	4.0	4.0	4.0
4	2 m	3 m	8 m	14 m*	15 m
SC Lower Water Temperature (°C)	11	10.5	11	10	11

Table 3. Air Temperature at the Upper and Lower Streams of BC and SC

1	1 m	2 m	8 m	10 m	12 m
BC Upper Air Temperature (°C)	6.0	6.0	6.0	6.0	6.0
2	1 m*	2 m	3 m	4 m*	15 m
SC Upper Air Temperature (°C)	14	14	14	14	13.5
3	1 m	2 m	4 m	8 m	16 m
BC Lower Air Temperature (°C)	7.0	7.0	7.0	7.0	7.0

4	2 m	3 m	8 m	14 m*	15 m
SC Lower Air Temperature (°C)	10	10	10	10	10