Reducing Acidification in Endangered Atlantic Salmon Habitat

One Year After: Project Summary

February 2026

Contact: Emily Zimmermann, Biologist

Phone: (207) 446-1003

MAINE DEPARTMENT OF ENVIRONMENTAL PROTECTION
17 State House Station | Augusta, Maine 04333-0017
www.maine.gov/dep

Introduction

Despite restored access to historic habitat in eastern Maine, the population size of Atlantic salmon (*Salmo salar*) has remained low (USASAC 2022). Most Downeast rivers and streams have been identified as acidic (pH <6.5), with headwaters chronically acidic and main stems episodically acidic, due to natural (wetlands, coniferous forests) and anthropogenic (acidified rain) sources (Haines et al. 1990; Whiting and Otto 2008). Loss of fish populations due to acidification of surface waters has been well documented in the North Atlantic region (as reviewed by Clair and Hindar 2005; Dennis and Clair 2012). In addition, numerous studies have demonstrated that episodic exposure to low pH can have detrimental, sub-lethal, or lethal impacts especially when coinciding with key salmon life stages during snow melt and spring runoff (e.g., Kroglund et al. 2008; Lacroix and Knox 2005; as reviewed by McCormick et al. 1998). Adding lime to acidic waters, through application of agricultural lime or lime slurry, has increased salmon populations in Scandinavia and Nova Scotia (as reviewed by Clair and Hindar 2005; Halfyard 2007; Hesthagen et al. 2011), and has been a recommended restoration action for Maine's acidic rivers and streams (NRC 2004). A 2009 Project SHARE pilot study investigating the efficacy of using clam shells to lime small streams suggested a trend towards improved habitat quality (Whiting 2014; for a more detailed project background, see Zimmermann 2018).

To further investigate using shells as a mitigation method, the Downeast Salmon Federation (DSF) started a five-year liming project in the East Machias River watershed in 2019. Clam shells were spread along the stream bottom, as well as along the banks to capture high flow events (i.e., rainfall and snowmelt, when episodic acidity is expected). The project goal was to increase juvenile salmon abundance by application of clam shells to achieve a target pH, and to evaluate changes in the macroinvertebrate community, which provides food sources for salmon. From 2017 through summer 2019, baseline data were collected (see Zimmermann 2019). From 2019-2022, shells were spread along a treatment reach in Richardson Brook over multiple days in August or September. This report investigates any impacts to water quality during the year after the last addition of shells and summarizes the results from all years of the study.

Methods

Study Location

Four tributary streams to the East Machias River were monitored: control sites in Richardson Brook, Creamer Brook, Barney Brook, and Beaverdam Stream, and a treatment site in Richardson Brook (Figs. 1 and 2; for physical and geological characteristics see Appendix I Tables 1 and 2 in Zimmermann 2020). In addition, macroinvertebrates were collected in another tributary, Northern Stream. These are within the homeland of the Passamaquoddy Tribe of Wabanaki. The East Machias River watershed is typical of coastal eastern Maine, with extensive wetlands resulting in colored waters high in organic materials and low in pH, and with high summer temperatures (Project SHARE-USFWS 2009). The existing salmon population in the East Machias River system is low (median large parr density 13.1 per habitat unit, $100m^2$ in 2019; Maine Department of Marine Resources, MDMR). In 2023, 20 redds were observed in the watershed (MDMR). All study streams are stocked by DSF and MDMR, except Barney Brook due to its small size, and the average large parr density observed during fall electrofishing is 12 parr/$100m^2$ at Richardson Brook, 16 parr/$100m^2$ at Creamer Brook, and 13 parr/$100m^2$ at Beaverdam Stream (Fig. 3, MDMR data). These three study streams had on average 5 fish species each year, however the lowest species richness was observed in 2023 (MDMR data).

Water Quality

All water quality monitoring activities followed the EPA-approved Salmon Habitat Monitoring Program Quality Assurance Project Plan (MDEP 2021). Continuous monitoring devices were deployed at all sample sites (except Northern Stream; Fig. 1) to collect water quality data April through November, every hour during baseline (2017-2018) and every half hour 2019-2023 (see Zimmermann 2018 for detailed methods). Continuous data parameters included pH, temperature, dissolved oxygen, and specific conductance. In addition, Onset Hobo MX2501 pH loggers were used to collect pH and temperature data every half hour from November through April at the downstream Richardson Brook site (2019-2023), the upstream Richardson Brook site (2020-2023) and at Creamer Brook (2022-2023). Continuous data were corrected as needed based

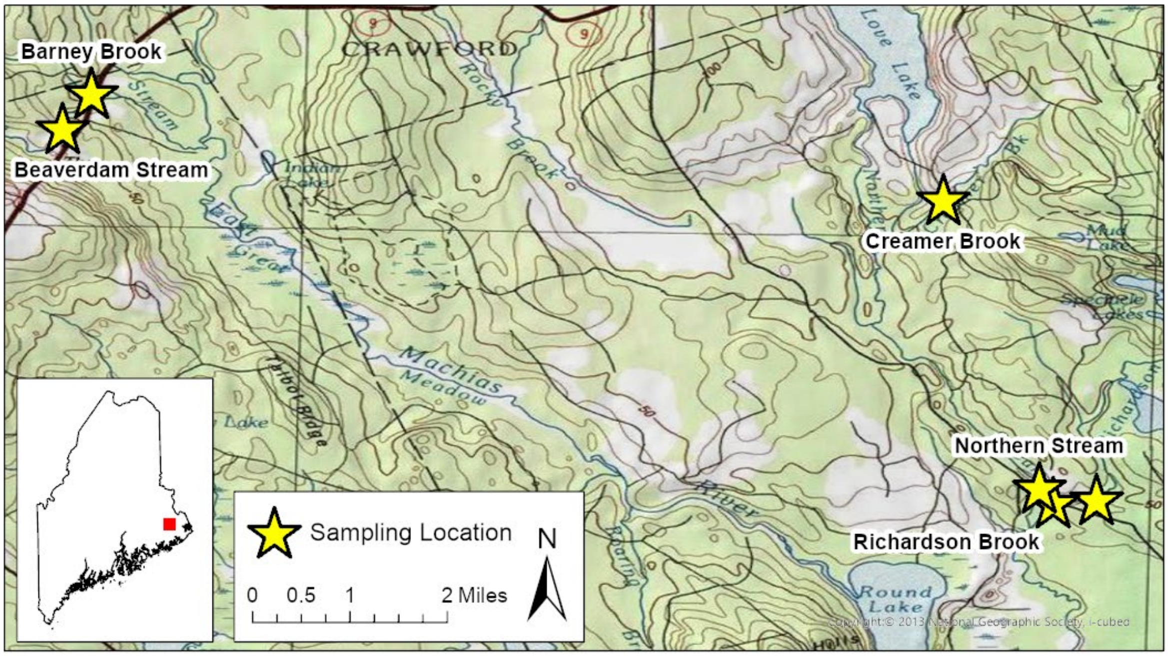


Figure 1. *Map of the study sites on tributaries to the East Machias River. On Richardson Brook, samples were collected at two sites: within the shell treatment reach (Rich-B) and above the shell treatment reach (Rich09; see Fig. 2). Northern Stream was only sampled for macroinvertebrates.*

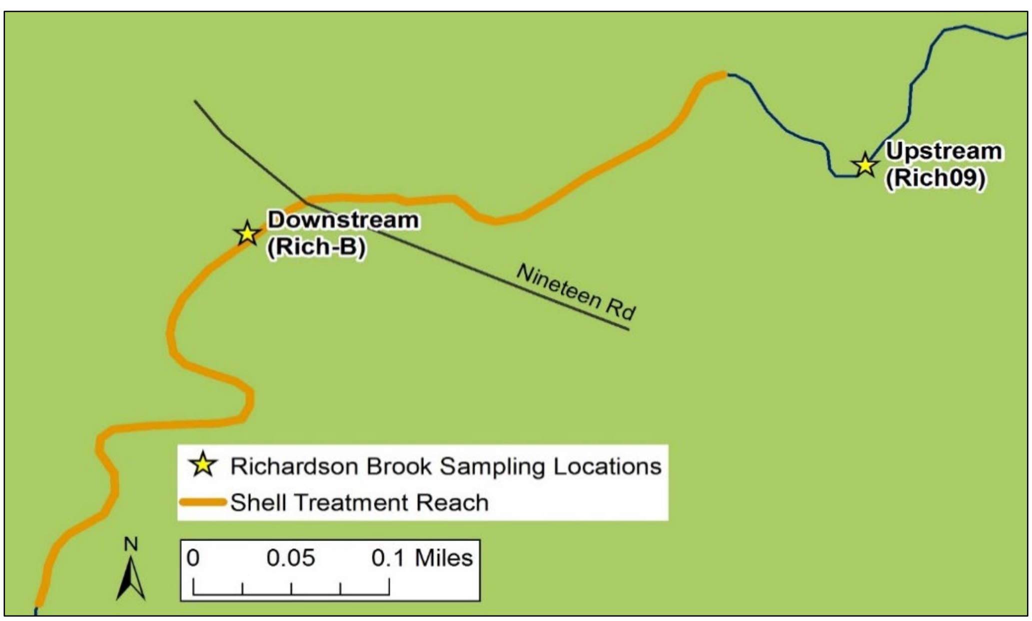


Figure 2. *Map of the study sites on Richardson Brook in relation to the shell treatment reach.*

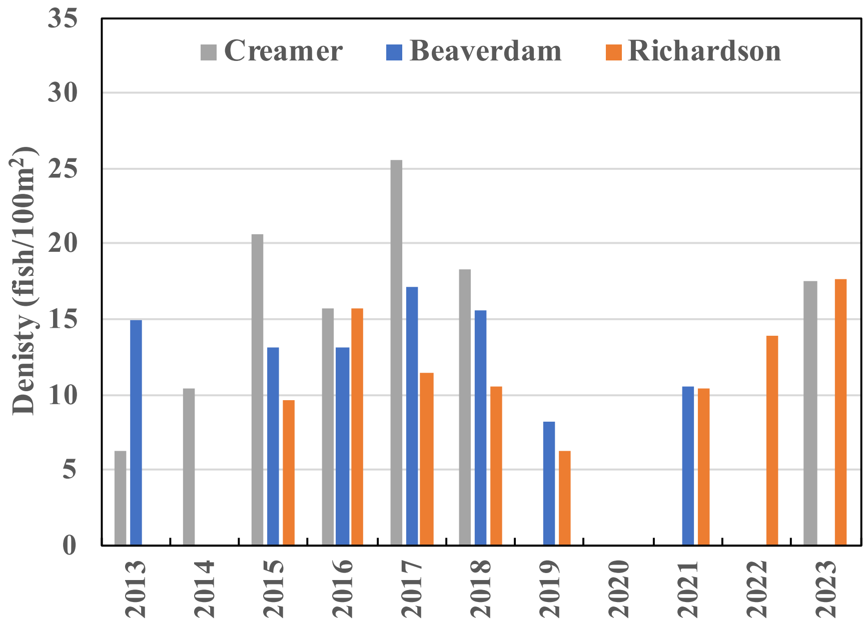


Figure 3. *Salmon density in three of the study streams from 2013-2023. High flows prevented data collection in Creamer Brook in 2019 and 2021. Extremely low flows prevented data collection in 2020. Only Richardson Brook was surveyed in 2022 due to resource limitations. No data for Beaverdam Stream were collected in 2023 due to broodstock collection. Data from MDMR electrofishing surveys.*

on quality control procedures as described in MDEP (2016) and using a sonde as a reference field meter (Eureka Manta2 Sub2).

Grab samples for acid neutralization capacity (ANC), calcium, aluminum species, dissolved organic carbon (DOC), closed-cell pH, and base cations were collected in spring (April, except for 2017 when the earliest sample was collected in June), summer (July or August), early fall after shell additions (September or October) and late fall (November; Appendix I Table I-1; Appendix I Table I-2 for shell dates; see Zimmermann 2018 for detailed methods). Exchangeable aluminum (Alx), defined as all dissolved species of aluminum that do not pass through the ion exchange column (including all –(OH), -(SO4), -(F) and –(Cl) complexes and Al^{+3}), was not measured directly but estimated as the difference between dissolved aluminum and organically complexed aluminum. The presence of suspended aluminum could cause an overestimation of the exchangeable aluminum in the samples (Dennis and Clair 2012). The detection of chemical extremes is dependent on sampling frequency, resulting in all grab sample data presented in this report being an estimate rather than an absolute measure of episodicity.

In 2017, macroinvertebrate samples were collected at Creamer Brook and the two Richardson Brook sites using rock bags following the Maine Department of Environmental Protection (MDEP) Biological Monitoring Program sampling methods (MDEP 2014). With the exception of Barney Brook, macroinvertebrates were sampled at all sites every two years 2018-2022, with additional samples collected at Beaverdam Stream and Creamer Brook in 2019 for quality control. DSF staff collected additional macroinvertebrate data in October 2017-2022 at three locations using rock bags, following the Izaac Walton League of America's stream-side identification methods (IWLA 2021; Table 8 in Zimmermann 2023).

Data Visualization and Analysis

Water quality data in this report were analyzed using the Water Resources Database (WRDB) version 7.1.0.35 (Wilson Engineering 2023) and R 4.3.1 (R Core Team 2023). Figure 4 was created using WRDB and figures 5-7 were created using the *ggplot2* package (Wickham 2009). All data are presented as mean ± standard deviation (SD). Due to the small sample sizes, non-parametric Kruskal-Wallis tests were used to compare water grab sample results between sites, seasons, and years, with pairwise Wilcoxon rank sum post-hoc tests with Holm adjustment. Duration of stressful events, based on thresholds for the protection of salmon and other aquatic life, were calculated based on how many consecutive data points exceeded the water quality threshold before recovery.

Quality control issues in 2023, such as being out of calibration, caused rejection of 16% of pH data (on average 32 days per site) and 3% of specific conductance data (28 cumulative days). Equipment malfunction, primarily battery failure, resulted in loss of 5% of continuous data (49 cumulative days) for all parameters. Flagged data (based on data corrections as per MDEP 2021 or best professional judgement) represented 7% of pH data (66 cumulative days).

Results and Discussion

Weather

During the seven years of this study, the area experienced warm, dry springs for three years (2017, 2021 and 2022), a cold, dry spring in 2018, and cold, wet springs in 2019 and 2020 (Fig. 4; NOAA 2023). Summers were either warm and dry, with moderate drought conditions (2017, 2020, 2022) or punctuated with heavy summer rains (NOAA 2023, U.S. Drought Monitor 2023). Severe drought conditions occurred in summer 2020. Rain in the study area is acidic, with an average annual pH around 5.3, having increased approximately 0.8 pH units since the Clean Air Act was amended in 1990 to address the threat of acid rain, and the pH is continuing to trend upward (NADP 2023). Natural rainwater unimpacted by anthropogenic pollution is expected to have a pH of 5.6 due to the dissociation of carbon dioxide into carbonic acid, however naturally occurring acids in the air (such as sulfuric acid, as part of the natural sulfur cycle) could further acidify rainwater regardless of anthropogenic pollution (Charlson and Rodhe 1982). Seasonal variability in the pH of the rain in the study area ranged from the most acidic during winter months (November – April average pH of 5.27) to the least acidic in May (average pH 5.7; NADP 2023). In general, most rain events were small (<5 mm), however wet summers had larger rain events (>20 mm), as did every fall (Weather Underground 2023).

pH

Salmon prefer pH values that are circumneutral (6.5-7.5), rather than acidic (<6.5). The impacts of acidity depend on 1) duration, magnitude, and frequency of the episode, 2) the ability of the fish to avoid adverse water quality conditions, 3) the concentration of exchangeable aluminum (Alx), 4) the buffering capacity of the water (i.e., ANC and calcium) and 5) life stage (see Zimmermann 2018 for overview). pH thresholds used in this analysis are estimates of anticipated impacts to salmon populations and do not include a detailed analysis of the impact of other factors.

Stream pH is generally lowest during the fall and winter, due to factors including increased rainfall, leaf drop (and therefore decreased uptake of nitrogen from soils), and saturation of wetland soils (Driscoll et al. 2003). Winter pH (Nov-April) at the two

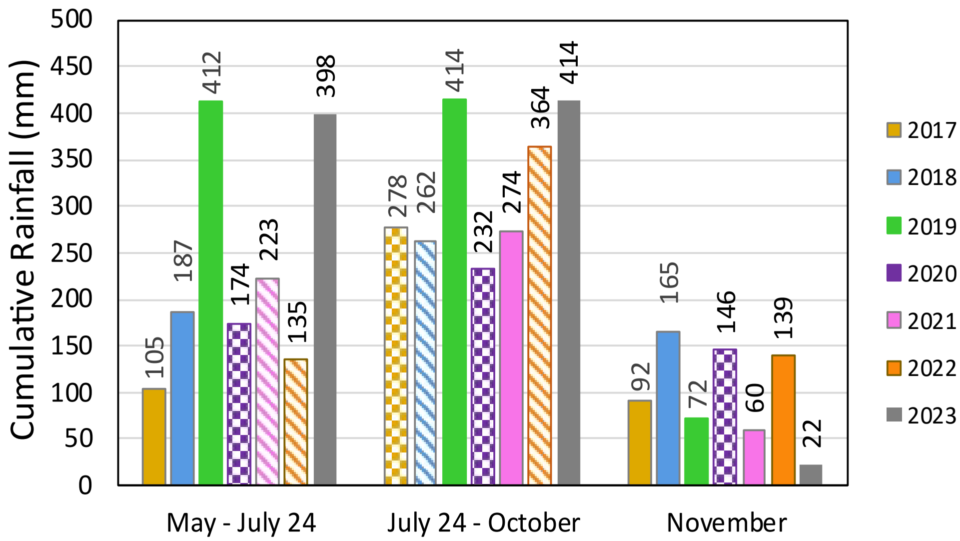


Figure 4. *Cumulative annual rainfall. The three time periods represent spring pre-treatment, summer-fall shell treatment, and November post-treatment, based on shell applications in 2019-2022. Drought conditions are indicated by diagonal hatching (abnormally dry) and checkerboard fill (moderate to severe drought). Data from Weather Underground stations and US Drought Monitor.*

Richardson Brook sites was similar prior to the addition of shells (grand mean 5.1 ± 0.4), with slightly higher pH at the upstream control site (Rich09; mean difference of 0.3 based on paired data points). Following the addition of the first batch of shells, mean winter pH at the treated downstream site (Rich-B) was slightly higher while the upstream control site remained unchanged (5.4 ± 0.3 downstream vs. 5.2 ± 0.2 upstream, with the same mean difference of 0.3). A similar trend was observed between Rich-B and an untreated control stream, Creamer Brook. Measurement accuracy (± 0.1) plus the standard deviations makes it difficult to confirm if mean pH at the treatment site was impacted by the addition of shells.

Compared to the critical stress threshold of 5.5, below which adverse impacts to salmon populations are expected, winter pH (Nov-April) was frequently stressful. pH remained lower than the stress threshold for 62% of the winter on average at the treated downstream Richardson Brook site (2020-2023), compared with 93% of the winter on average at the upstream control site (Haines et al. 1990; Stanley and Trial 1995).

Complete continuous data were only collected 2020-2023, characterizing treatment conditions. To be able to compare across all baseline and treatment years, data from October and November were used as proxy (of all months monitored continuously every year, these had the lowest average monthly minima). These proxy data indicate that prior to the first shell additions, both Richardson Brook sites were below the stress threshold of 5.5 around 80% of the winter on average (± 5% of each other). Following shell additions, the treated site was below the threshold for 40% of the winter on average, compared to 83% at the upstream control. Similar trends were observed at untreated Creamer Brook, with pH remaining below the threshold for 70% of the winter on average. Although rainfall-driven episodic acidity events continue to occur in the winter (Nov-April) after four years of shell additions (Fig. 5), winter pH is less stressful at the treated site than at the control sites.

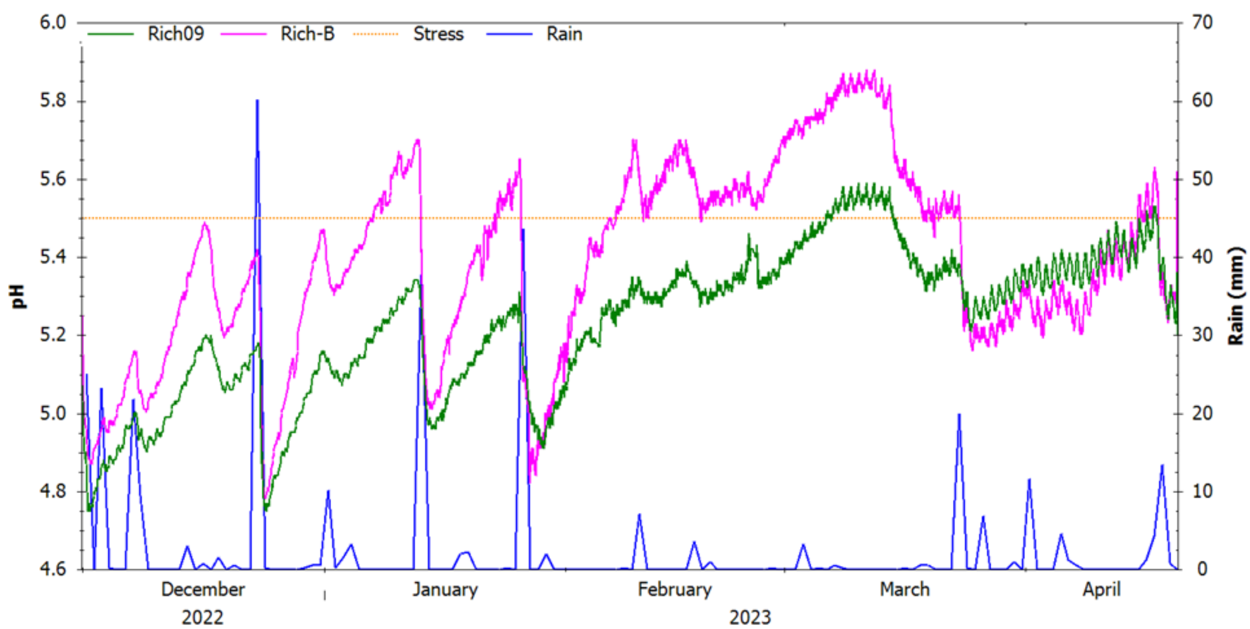


Figure 5. *Winter pH at Richardson Brook (upstream control – Rich09 and downstream treatment – Rich-B), following the final addition of shells in Sept. 2022. Stress threshold from Stanley and Trial 1995 and Haines et al. 1990.*

During the multi-year study, from spring through fall each year, pH remained mostly above the stress threshold of 5.5 at all sites, with episodic depressions following rain events (Fig. 6; Haines et al. 1990; Stanley and Trial 1995). Rain raises the water table from the subsoil into the upper soil horizons where acid neutralization is less effective, resulting in more acidic surface waters, regardless of the pH of the precipitation (Driscoll et al. 2001). Acidic depressions could also be enhanced by ion exchange in soil solution due to the salt effect, in which maritime-origin sodium displaces the hydrogen ions in soil, decreasing soil acidity and increasing surface water acidity (Heath et al. 1992). Barney Brook experienced the highest pH, remaining above 6.5, an optimal minimum pH for the protection of the most sensitive salmon life stages (alevins and smolts), for on average 37 ± 18% every year (e.g., see Fig. 6; Appendix II Tables II-1 and II-5; Kroglund and Staurnes 1999; Kroglund et al. 2008). In contrast, Creamer Brook and the upstream control site at Richardson Brook never exceeded the optimum minimum pH of 6.5, and the treated Richardson Brook site only exceeded this threshold 0.1% of the time prior to shell additions, and 15% of the time after shell additions.

The pH at the treated Richardson Brook site was 0.5 units higher following shell additions than during baseline years (5.99 ± 0.53 during 2020-2023 vs. 5.48 ± 0.49 during 2017-2019), with a minimum pH 0.8 units higher in 2023 than during baseline years (Fig. 7). In contrast, average pH at the control sites only changed on average 0.08 units between the baseline and treatment periods and mimina varied by 0.3 units. pH at all sites was lower in 2023 compared with prior years, and the difference between the two Richardson Brook sites was less pronounced (Fig. 6). Throughout the study, Beaverdam Stream usually had the second highest pH (6.15 ± 0.46), similar to Barney Brook. However, in 2023 pH at Beaverdam Stream was lower (5.87 ± 0.26) and more similar to the other study streams, including the treated Richardson Brook site (Fig. 6). Reduced pH in 2023 was likely due to the high amount of rain which decreased the buffering capacity at all the study sites.

Stressful conditions continue to occur during the warmer months at the treated Richardson Brook site, despite increases in pH following four years of shell treatments. During baseline years, from spring through fall, both Richardson Brook sites were below the critical stress threshold of 5.5 for around 40% of the time (Baker et al. 1996; Henriksen et al. 1984; Lacroix and Knox 2005; Magee et al. 2003). Following shell additions, the treatment site only fell below 5.5 for around 20% of the time, while no

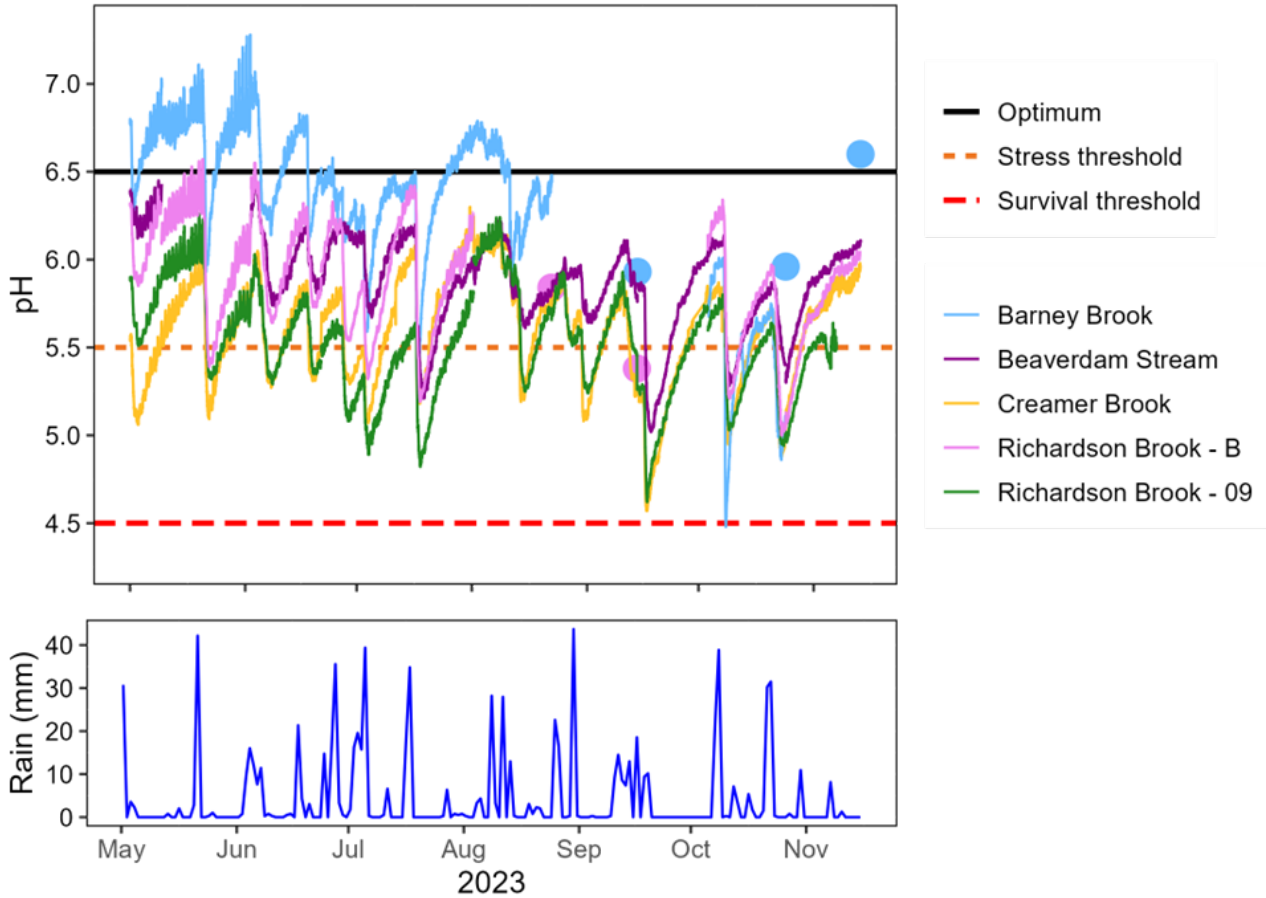


Figure 6. *pH and rainfall at the five study sites in 2023. Gaps in the continuous record are due to equipment malfunctions and/or quality control issues. Dots represent discrete measurements from field meter as proxy for continuous data. Optimum pH from Kroglund and Staurnes 1999 and Kroglund et al. 2008. Stress threshold from Stanley and Trial 1995 and Haines et al. 1990. Survival threshold from Potter 1982. Rainfall data from Weather Underground.*

change was observed at the upstream control site (Fig. 7). During the shell treatment years, the other control sites were below the critical stress threshold on average 4% less of the time than during baseline years, indicating that the increased pH at the treatment site is site specific and therefore due to the addition of shells. The duration of stressful acidic events (<5.5) at the treatment site was reduced by 50-60% following shell additions (average of 2.9 days with a maximum of 14.9 days) compared to baseline years (average of 5.4 days with a maximum of 1.2 months). The duration of

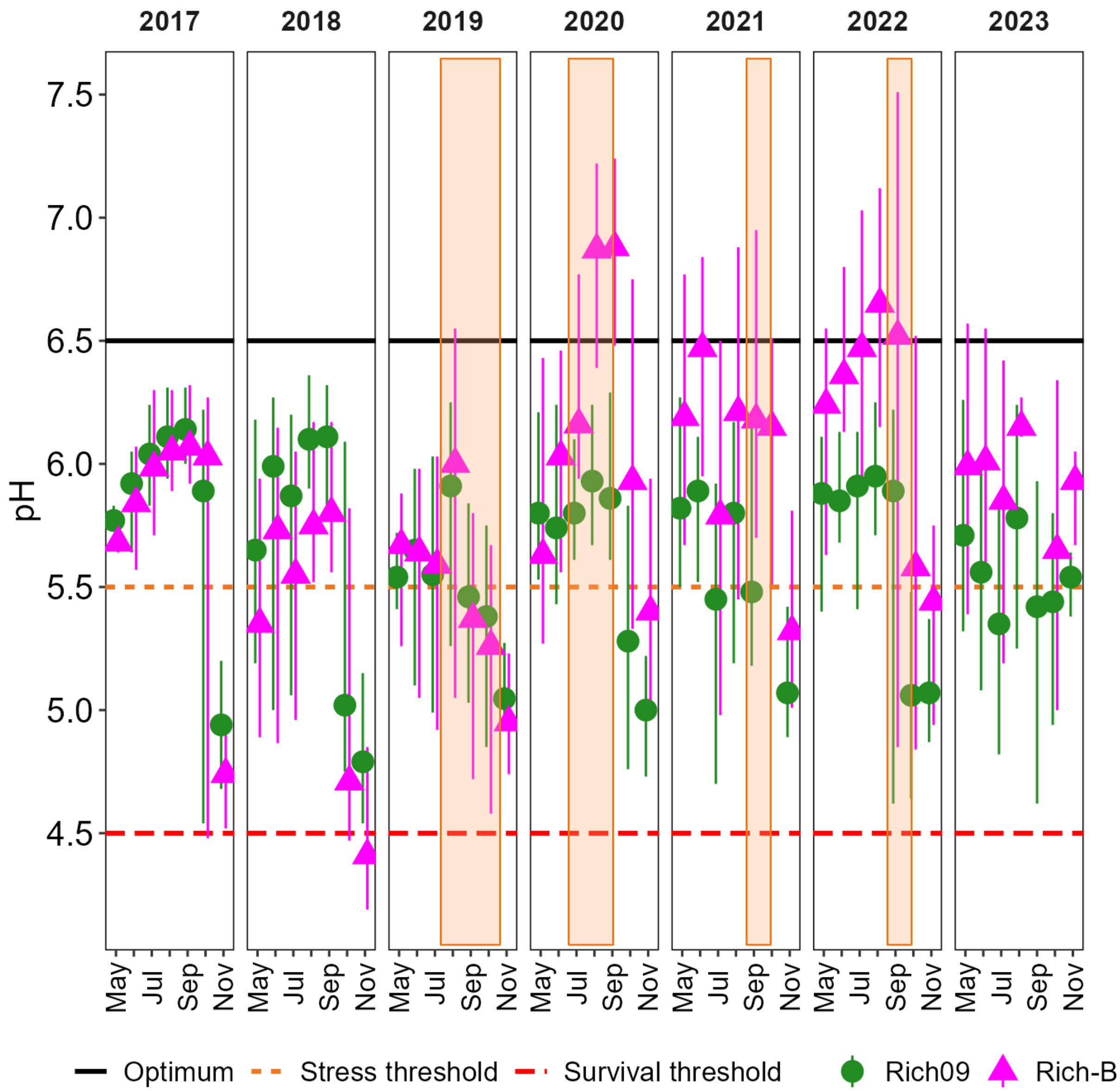


Figure 7. *Monthly pH at the upstream (Rich09, in green) and downstream (Rich-B, in pink) Richardson Brook sites. Each point represents the monthly median, with the whiskers extending to the minimum and maximum values observed. Optimum pH from Kroglund and Staurnes 1999 and Kroglund et al. 2008. Stress threshold from Stanley and Trial 1995 and Haines et al. 1990. Survival threshold from Potter 1982. Orange boxes represent shell additions 2019-2022.*

stressful events was also reduced by 50% at the upstream control site during the shell application years, however the maximum duration was only reduced by 9 days (16%) in comparison with the 22 day reduction (60%) observed at the treatment site. Acidic episodes are still occurring despite the increase in pH at the treatment site following shell additions. However, the decreased maximum duration of these stressful episodes indicates that the addition of shells has increased the buffering capacity at the treatment site, allowing recovery to less stressful pH levels occur more quickly.

Stream Temperature

Salmon prefer cold waters. Stream temperatures were similar at all study streams, remaining below the threshold for optimal growth of 20°C for most of the sampling period (85% at all streams combined; Appendix II Tables II-1 and II-5; Jonsson et al. 2001; USEPA 1986). Warmest temperatures occurred in June, July, and August. The stress threshold of 22°C was exceeded 6.1% of the time (Cunjak et al. 2005; Elliott and Elliott 2010; Lund et al. 2002), USEPA's short-term maximum for survival of 23°C was exceeded 3.4% of the time (USEPA 1986), the lethal temperature for adult salmon survival (26-27°C) was exceeded only 0.07% of the time (Shepard 1995 as cited in Frechette et al. 2018), and the lethal temperature for parr (28-29°C) was never exceeded (Elliott 1991 as cited in Stanley and Trial 1995; Garside 1973 as cited in Lund et al. 2002; Grande and Andersen 1991 as cited in Elliott and Elliott 2010).

Barney and Creamer Brooks remained the coldest, especially during hot, dry summers, possibly due to the relative influence of groundwater during low flows (Appendix II Table II-1). Exceedances of the stress threshold of 22°C lasted 10 ± 5 hours at a time. Daily temperature ranges of 2.9 ± 1.6°C may help provide nighttime temperature refugia when daily maximum temperature is a few degrees above the stress threshold. The longest period above 22°C occurred at Beaverdam Stream in July 2023, lasting 10.7 days. When temperatures remain above the stress threshold for long periods of time, such as during the hottest parts of the summer at all sites, sub-lethal stress may be occurring.

Specific Conductance

Specific conductance is a measure of the concentration of ions in the water, or the ability of water to conduct electricity. The streams in the study area have very low specific conductance (29 ± 12 μS/cm at all sites combined; Appendix II Table II-1), which can increase the difficulty of accurate pH measurements and electrofishing (Hovind 2010 as cited in Garmo et al. 2014; Zimmermann 2018). Lab-analyzed closed-cell pH grab samples closely matched sonde pH (within 0.14 ± 0.22), with the closest match at the Richardson Brook sites (within 0.01 ± 0.19), providing confidence in the pH data discussed above. Beaverdam Stream and Barney Brook had the highest specific conductance (38 ± 16 μS/cm and 34 ± 12 μS/cm, respectively). Shell additions caused no persistent impacts to specific conductance. However, concentrations at the treated Richardson Brook site increased to levels similar to the two highest streams for approximately a month following each shell addition, likely due to the increase in dissolved solids such as sodium, calcium, and chloride, originating from the shells. Rain events reduced the elevated concentrations to pre-treatment levels. No negative impacts to aquatic life are expected from the range of specific conductance observed, however increased ion concentrations (such as calcium) may increase buffering capacity.

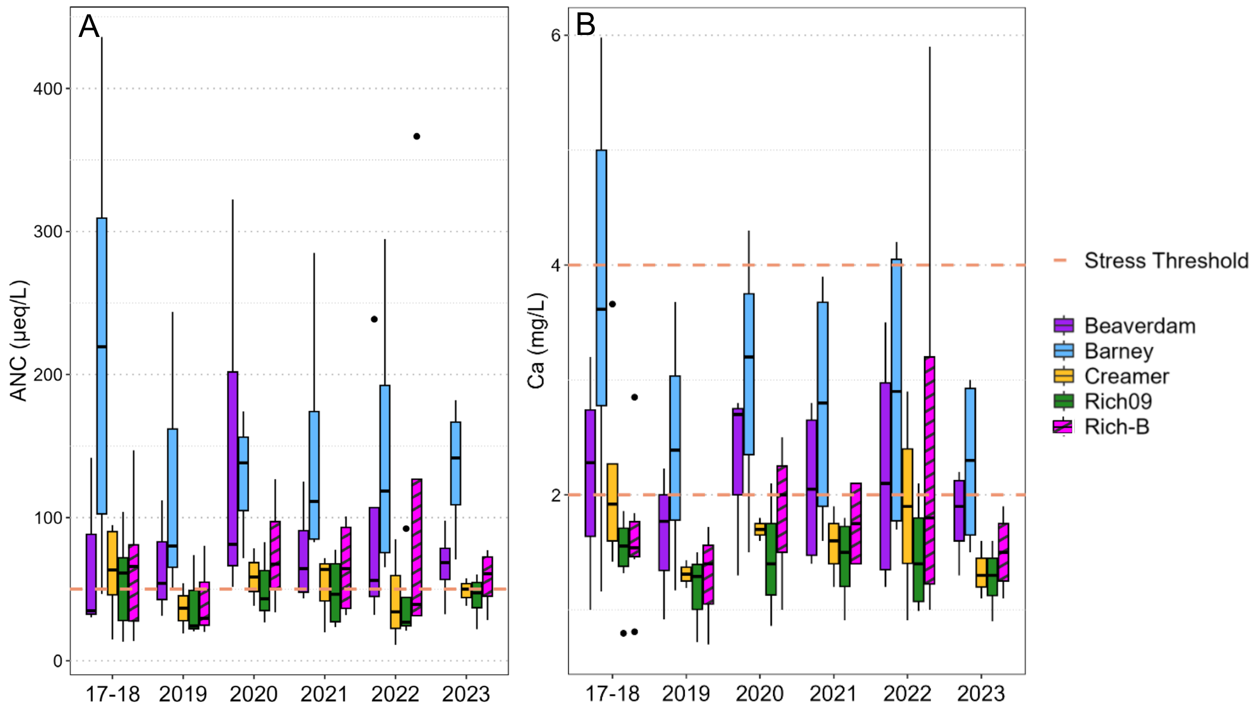
Dissolved Oxygen (DO)

Salmon prefer well oxygenated waters. As in prior years, DO levels were within a healthy range for fish and aquatic life, in addition to the preferred range for salmon of >6-7 mg/L, for most of the study period (96%; Appendix II Tables II-1 and II-5; Stanley and Trial 1995; USEPA 1986). DO concentrations fell below the Maine Water Quality Standard of 7 mg/L at all sites, especially during hot, dry summers, lasting on average 12 ± 7 hours, with a maximum duration of almost 11 days at the downstream Richardson Brook site in October 2017 (38 M.R.S. §§ 465.2.B). During the lowest flow conditions during these hot, dry summers (2017, 2018 and 2020), DO dropped below USEPA's threshold for acute impairment of 5 mg/L, lasting on average 9 ± 3 hours, with a maximum duration of almost 2 days at Barney Brook in 2018 when flows likely ceased completely leaving a stagnant pool (USEPA 1986). Hot, dry summers resulted in DO minima that coincided with the warmest temperatures, increasing stress. In addition, these conditions coincided with the lowest flows, possibly preventing movement of salmon and other aquatic organisms to oxygen and temperature refugia, if any existed nearby.

Acid Neutralization Capacity (ANC)

Streams with higher ANC have a higher capacity to buffer against changes in acidity. At all streams, ANC was highest during summer baseflow. ANC remained above the threshold of acid sensitivity for the protection of the most sensitive aquatic species and life stages of 50 µeq/L for on average 62% of the samples across all sites (Fig. 8.A; Appendix II Table II-2; Driscoll et al. 2001). During spring and fall, when the water table rises to the upper soil horizons which have reduced acid buffering capacity, ANC fell below the Norwegian 20-30 µeq/L critical limit for salmon at Creamer and Richardson Brooks, representing on average 30% of the samples at those sites (Baker et al. 1990; Driscoll et al. 2001; Lien et al. 1996; Kroglund et al. 2002). ANC was likely high enough (>100 µeq/L) for maintenance of the calcium concentration necessary to buffer against the negative impacts of acidity (2 mg/L) during summer baseflows at Barney Brook (58% of samples) and Beaverdam Stream (25% of samples; Fig. 8 A; Brocksen et al. 1992). Richardson Brook also exceeded 100 µeq/L at both sites in 2017, and at the downstream treatment site every summer while shells were applied (2020-2022). Despite higher ANC during summer baseflow, there were no statistically significant differences at the treated Richardson Brook site between years, compared to the other sites, or pre- and post-shell additions (Appendix II Table II-3).

In low DOC waters, ANC is an approximate surrogate for alkalinity, but it can be significantly higher than alkalinity when DOC is higher (Garmo et al. 2014). When calculated from ANC, alkalinity was 7.2 ± 5.2 mg/L across all sites, with highest values at Barney Brook and Beaverdam Stream. Only one sample was above USEPA's recommended ambient water quality criteria of at least 20 mg/L alkalinity (21.8 mg/L at Barney Brook during summer baseflow 2017), however this threshold does not apply where values are naturally lower (USEPA 1986). Relatively low ANC values indicate a deficit of buffering materials in the watershed due to thin soils (Potter 1982), allowing



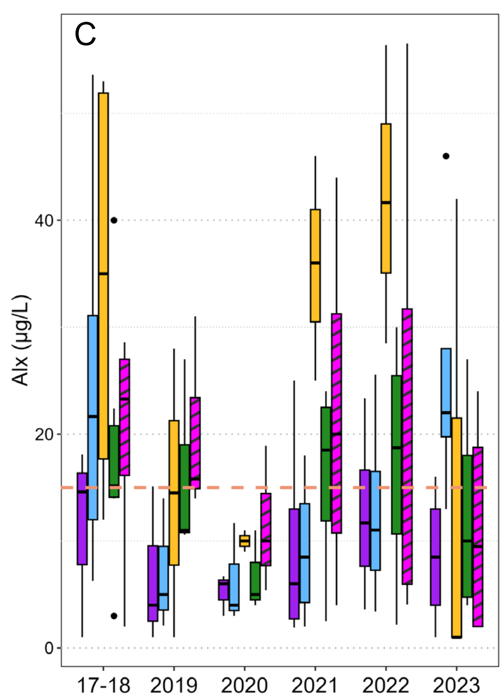


Figure 8. *(A) Acid neutralization capacity (ANC), (B) calcium (Ca) and (C) exchangeable aluminum (Alx). 2017-2018 represents baseline conditions. Shells were added to Rich-B 2019-2022. Each box represents the interquartile range, with the horizontal line representing the median, and whiskers extending to the minimum and maximum values observed, except where values are considered statistical outliers (dots). Most sample sizes n = 4. ANC stress threshold of <50 µeq/L from Driscoll et al. 2001. Calcium stress thresholds of <4 mg/L from Marcus et al. 1986, as cited in Brocksen et al. 1992, and <2 mg/L from Baker et al. 1990 and Baldigo and Murdoch 2007. Alx stress threshold of >15 µg/L from EIFAC as cited in Dennis and Clair 2012.*

swings in pH after rain inputs and increasing the potential for salmon mortality (Fig. 6; MacAvoy and Bulger 1995). Buffering capacity at the treated Richardson Brook site may have been trending higher during summer baseflow with the addition of shells, however the increase was not statistically significant. Summer baseflow coincides with periods of highest pH and therefore lowest stress due to acidity, so increases in ANC may not have been biologically significant. With the end of this project, the stream is expected to revert to the pre-treatment acidified state relatively quickly due to the low buffering capacity of the watershed.

Calcium

Higher calcium concentrations enable faster growth and higher survival in fish. Calcium can increase the efficiency of ion regulation, buffering against the detrimental impacts of toxic aluminum, but only at concentrations above 2 mg/L in acidic streams (Baldigo and Murdoch 2007; McDonald et al. 1980; Wood et al. 1990). Streams with higher pH and ANC also had higher calcium concentrations, remaining above the survival threshold of 2 mg/L for more than half of the sampling events at Beaverdam Stream and Barney Brook (Fig. 8 B; Appendix II Tables II-3 and II-6; Baker et al. 1990; Baldigo and Murdoch 2007). In contrast, streams with lower pH (Creamer and Richardson Brooks) were above the survival threshold for only 16% of the sampling events on average. The difference in summer baseflow calcium concentrations between the treated and control sites on Richardson Brook increased from 0.14 ± 0.07 mg/L during baseline years to 0.40 ± 0.17 mg/L following shell additions. Increases in calcium were also observed at Beaverdam Stream during the shell addition years, with 4% more samples above 2 mg/L, however the biggest increase was observed at the treatment site (+27%, as compared to +15% at the upstream control site).

The only site with concentrations regularly above the suggested threshold of 4 mg/L to prevent deformities and other stress was Barney Brook (21% of the sampling events; Marcus et al. 1986, as cited in Brocksen et al. 1992). During summer baseflow 2022, after three years of shell additions, the treated Richardson Brook site attained an anomalously high calcium concentration of 5.9 mg/L, but none of the other study sites had concentrations higher than 4 mg/L, making the single high value inconclusive.

Despite the increase in calcium following shell additions, there were no statistically significant differences at the treated Richardson Brook site (Appendix II Table II-3). Summer baseflow coincides with periods of highest pH and therefore lowest stress due to acidity, so increases in the buffering capacity (through high ANC and calcium) may not have biological significance.

Aluminum

Aluminum concentrations were highly variable throughout the study. Average total aluminum per stream ranged from 118 to 205 µg/L, well below the acute Maine Ambient Water Quality Criteria maximum (Criteria Maximum Concentration, CMC) of 750 µg/L (based on a pH of 6.5-9 and dissolved organic carbon (DOC) <5 mg/L, which are significantly different from values observed in the study streams; Appendix II Tables II-1, II-2 and II-4; MDEP CMR Chapter 584). The chronic Maine Ambient Water Quality Criteria (Criterion Continuous Concentration, CCC) of 87 µg/L was exceeded in 73 ± 11% of samples at each site, however this threshold is exceeded in many high quality waters across the country, according to USEPA (MDEP CMR Chapter 584), such as streams high in tannins.

USEPA's site-specific maximum criterion (CMC; ranging from 5-1,500 µg/L depending on DOC, total hardness, and pH at each sample site; USEPA 2018) was exceeded in 53 ± 31% of samples per year and the chronic criterion (CCC; ranging from 3 to 560 µg/L) was exceeded in 60 ± 24% of samples per year. The study streams were outside the upper range of EPA's model inputs for DOC (>12.0 mg/L) in 43% of the samples and the lower range for pH (<5.0) in 3% of the samples. Low pH increases the solubility, and therefore toxicity, of aluminum, meaning the criteria may be biased high for samples more acidic than the model inputs. In contrast, DOC can help buffer against the toxic impacts of aluminum in acidic waters by binding aluminum into inert organic complexes that are biologically unavailable (Baldigo and Murdoch 2007; Kroglund et al. 2008; Tipping et al. 1991). Therefore, EPA's site-specific criteria may be biased low for samples with DOC higher than the model inputs, if DOC is helping buffer against toxic aluminum, which may counterbalance the high bias from acidic samples. Despite these biases, the majority of samples at Creamer and Richardson Brooks exceeded the acute criteria, indicating conditions stressful for aquatic life. Streams with higher buffering capacity experienced fewer exceedances (33% of Beaverdam Stream samples and 10% of Barney Brook samples).

Exchangeable aluminum (Alx, also known as inorganic monomeric or labile aluminum) can cause respiratory distress when it binds to the gills of fish (e.g., Magee et al. 2003). Although inert organic aluminum was the dominant species every year at every site, representing 70 ± 29% of aluminum, likely due to the relatively high concentrations of dissolved organic carbon (see next section), Alx represented 9.3 ± 6.0% of aluminum species. Across all years, 22 ± 6% of samples at each site exceeded the threshold for the protection of aquatic life of 15 µg/L (Fig. 8 C; Appendix II Tables II-4 and II-5; Howells et al. 1990 as cited in Dennis and Clair 2012; Kroglund and Staurnes 1999; McCormick et al. 2009). Aluminum is more soluble and therefore more toxic in acidic waters. In addition, solar gain can heat brown colored waters, such as those in the study area, breaking down organic complexes and freeing ionic metals such as Alx, iron, and mercury.

There were no statistically significant differences at the treated Richardson Brook site between years, compared to the other sites, or pre- and post- shell additions (Appendix II Table II-3). Compared to other regions that have conducted liming studies, total aluminum concentrations in the study streams were similar to baseline conditions in Wales (150-190 µg/L, compared to 60-100 µg/L post-liming; Bradley and Ormerod 2002) and average Alx concentrations were similar to baseline conditions in Norway (70-160 µg/L, compared to post-liming 7 ± 4 µg/L; Hesthagen et al. 2011). Despite increases in pH and summer baseflow calcium and buffering capacity following shell additions at the treated Richardson Brook site, sub-lethal stress due to toxic Alx may still decrease smolt tolerance to saltwater when they migrate out of their freshwater habitat (Kroglund and Staurnes 1999; McCormick et al. 2009; Monette et al. 2008; Staurnes et al. 1995).

Dissolved Organic Carbon (DOC) and Anions

The toxic impacts of Alx may be buffered by the presence of DOC. Downeast streams, including those studied here, are naturally highly colored, with relatively high organic content (and therefore high DOC) due to wetlands and coniferous forests (Haines et al. 1990). DOC at all sites combined ranged from 6.7 to 27 mg/L, with an average of 12.6 ± 5.0 mg/L (Appendix II Table II-2). DOC was higher during the 'post' shell addition time period (>July 26, 2020) at all sites compared to 'pre' shell additions, but there were no statistically significant differences. The negative correlation between DOC and pH observed during base flows ($r = 5.6$, $R^2 = 0.39$, $p < 0.001$) suggests baseflow pH is driven by natural organic acids (Garmo et al. 2014). During the spring and fall the correlation was less strong ($r = 0.88$, $R^2 = 0.01$, $p = 0.38$) and trended positive more months than negative (64% of the time), indicating that low pH in spring and fall correlates with low DOC and suggesting that seasonal pH depressions are not driven by organic acids.

Anion composition indicates that inorganic processes drive seasonal pH depressions, but not necessarily due to anthropogenically acidified rain. Inorganic anions (chloride, sulfate, and nitrate) dominated over organic anions (DOC) in all samples. However, sulfate (an indicator of sulfur dioxide emissions) represented on average only 32% of the anions, exceeding 50% of the total anion composition in just 9% of samples (mostly at Barney and Creamer Brooks; Kahl et al. 1992). The dominance of inorganic anions in relation to DOC suggests that pH depressions are driven by inorganic processes such as rainfall-induced elevation of the water table into soil layers with reduced acid neutralization capacity, resulting in acidification of surface waters (Driscoll et al. 2001). However, based on the overall high DOC concentrations in the study streams, it is expected that some buffering of Alx is occurring despite low pH values.

Base Cation Surplus

Base cation surplus (BCS) is an effective indicator of anthropogenic acidification (as distinguished from natural acidity) because it reduces the influence of natural acidity from DOC, which can be highly variable (Lawrence et al. 2007; Baldigo et al. 2009). BCS is the difference between the sum of cations (calcium, potassium, magnesium, and sodium) and anions (chloride, nitrate, sulfate, and strong organic anions as defined as 0.071*DOC-2.1; Lawrence et al. 2007). The threshold for aluminum mobilization occurs at a BCS around 0, regardless of DOC values. During the study period, BCS ranged from 35 µEq/L at Creamer Brook to 369 µEq/L at the treated site on Richardson Brook (Appendix II Table II-6). In general, lowest values were observed in the fall and the spring, when the water table rises to the upper soil horizons which have reduced acid buffering capacity, and corresponding with the lowest pH values. Low values during the summer of 2023 were likely driven by the wet weather. As expected based on calcium, ANC, and pH (Figs. 5 and 7), Barney Brook had the highest average BCS, and thus the highest capacity to buffer against the mobilization of toxic aluminum. All of the study streams may have the potential to buffer against toxic aluminum, particularly during summer baseflow.

Nutrients

Nutrient levels were similar across all study streams, although Barney Brook had approximately twice as much biologically available nitrogen (nitrate + nitrite, N+N) as the other streams (Appendix II Table II-7). N+N at all sites combined ranged from 0.007 to 0.11 mg/L, with an average of 0.041 ± 0.029 mg/L. Total Kjeldahl nitrogen (TKN) ranged from 0.29 to 1.35 mg/L, with an average of 0.62 ± 0.21 mg/L. Average TKN was higher than the 0.5 mg/L maximum usually seen in natural, undisturbed streams in Maine (based on MDEP's Biological Monitoring dataset), however this is not surprising due to the highly colored, tannin-rich streams in the study area. Total phosphorus ranged from 9 to 42 µg/L, with an average of 18 ± 7 µg/L. Phosphorus in the study area was on the high end of the spectrum for class A streams based on MDEP's Biological Monitoring dataset. At all sites, nutrient levels were typical of natural, undisturbed streams in Maine.

Algae

All study streams attained Maine's highest aquatic life water quality classification (see Appendix III for 2022 results; 38 M.R.S.§§ 465). Across all sites, algae that are intolerant of pollution (including nutrients and sedimentation) represented 35 ± 4% of species present, while pollution tolerant algae only represent 1-4% of species present. Sewage fungus was not present at any of the study streams. At Creamer Brook, nuisance algae (filamentous algae >1 cm long and periphyton mats >1 mm thick) represented 25 ± 19% of the percent cover across all transect stations (ranging from 0 to 60%). This could suggest an excess of nutrients, however total phosphorus was similar to the other study sites, and within the range expected for natural, undisturbed streams, as was nutrient-tolerant diatom richness. The algae assemblage in the study streams were representative of natural, undisturbed streams.

Macroinvertebrates

All study streams attained Maine's highest aquatic life water quality classification (see Appendix III for 2022 results; 38 M.R.S.§§ 465; Davies et al. 2016; but see Zimmermann 2018 for an explanation of the one anomalous result at Beaverdam Stream). Across all years sampled, the dominant taxa were genera of mayflies and caddisflies that most often occur in areas of little current (Appendix II, Table II-7). A predatory caddisfly (*Oecetis*) joined the dominant taxa at two sites in 2020 (Beaverdam Stream and the upstream Richardson Brook site), likely due to its tolerance of high temperatures and low flows. In 2023, two non-biting midges, *Thienemannimyia* and *Psectrocladius*, were among the dominant taxa at the upstream control site on Richardson Brook. Mayflies represented around one third of the generic richness (ranging from 10-77%, depending on the site and year), suggesting a healthy macroinvertebrate assemblage requiring good water quality.

Despite improvements to pH and other chemical properties following shell additions, no statistically significant differences in the macroinvertebrate assemblage were observed at the downstream Richardson Brook site between years, compared to the other sites, or pre- and post-shell additions (Fig. 9). Mayflies are the most sensitive group of aquatic insects to acidity (Wiederholm 1984) and their relative abundance increased by 12 percent at the treatment site following shell additions, in contrast to the upstream control (-5%) and the other study streams (-7.5% at Creamer Brook and +3% at Beaverdam Brook). Liming projects in other locations had modest effects on macroinvertebrates, with increased abundances of acid sensitive taxa, although only within a few years following treatment in some cases (Bradley and Ormerod 2002, Fjellheim et al. 2001, Tipping et al. 2002 from Kowalik and Ormerod 2006). Natural inter-annual variation may limit the ability to detect any changes due to the addition of

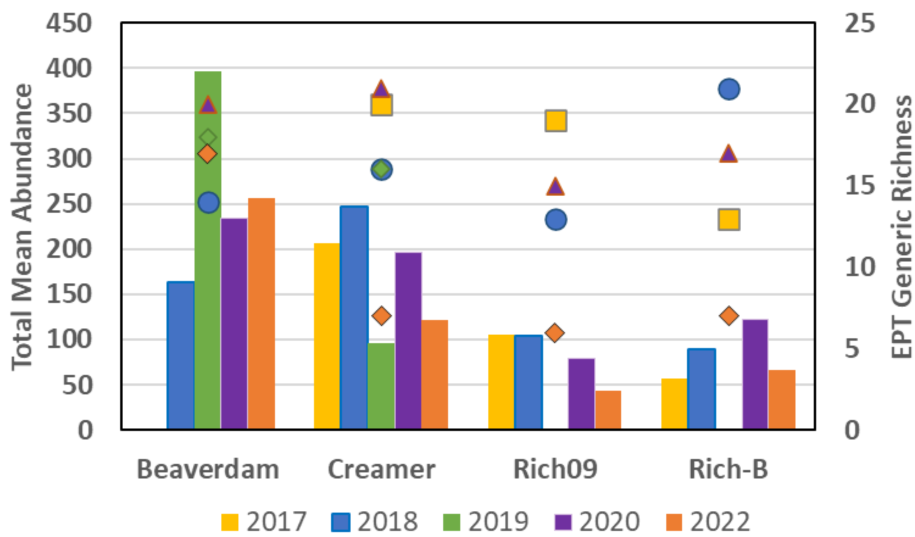


Figure 9. *Total mean abundance (bars) and generic richness of EPT taxa (Ephemeroptera, Plecoptera, and Tricoptera; points). Sample size n = 4, except for Creamer Brook where n =5.*

shells. Episodic acidity (<5) may have a detrimental impact on any acid-sensitive macroinvertebrates present in the study streams, however, as episodic acidity events have been occurring for decades, the macroinvertebrate assemblage in Downeast streams may be tolerant of low pH pulses. Salmon are opportunistic feeders, targeting the most abundant and optimally sized aquatic invertebrates, but also seasonally changing their diet from drifting to benthic prey (Grader and Letcher 2006; Ojala 2008). Changes in macroinvertebrate abundance may therefore have a stronger impact on salmon populations than changes in macroinvertebrate composition (Scott and Crossman 1973 as cited in Stanley and Trial 1995).

Conclusion

Following four years of clam shell additions, despite a rainy final year of the study, pH in the treated section of Richardson Brook was higher than in baseline years, as well as higher than all untreated control sites. Sub-lethal stress due to low pH and aluminum toxicity is likely still occurring during episodic, precipitation-driven acidity events. Eastern Maine is prone to rain-driven acidic episodes due to the naturally acidic pH of rain (~5.6, regardless of anthropogenic inputs) and inorganic processes within the acidic upper soil horizons, including the salt effect of maritime-origin sodium deposition and the naturally low weathering rate of the granitic bedrock. However, these stressful acidic events lasted for shorter durations at the treatment site due to increased buffering from the dissolution of the clam shells. The most sensitive salmon life stages (alevins and smolts) are present in the streams from March through June, however low pH events occurring during the autumn rainy season may also impact hardier life stages (parr and adults).

Any life stages present in the study area during summer baseflow conditions may experience additional sublethal stress from warm temperatures and low dissolved oxygen, particularly during hot, dry summers. Although not statistically significant, increases in acid neutralization capacity (ANC), calcium, and base cation surplus (BCS) were observed at the treated Richardson Brook site during summer baseflow, suggesting increased buffering capacity. However, summer baseflow coincides with periods of highest pH and therefore lowest stress due to acidity, so increases in chemical buffering capacity may not be biologically significant for reducing sub-lethal stress. Smolts are likely still experiencing sub-lethal stress due to toxic exchangeable aluminum, which had no measurable change following the addition of shells, however all streams in the study may provide some buffering capacity through high dissolved organic carbon (DOC) and BCS concentrations. Changes to stream chemistry resulted in no measurable biological changes in either the macroinvertebrate assemblage, which may be adapted to episodic acidity, nor salmon density, which may be due to the relatively short study period compared with the life cycle of Atlantic salmon (5+ years). With no further shell additions, Richardson Brook is expected to revert to the pre-treatment acidified state relatively quickly due to the low buffering capacity of the watershed.

Works Cited

Baker, J.P., Bernard, D.P., Christensen, S.W., Sale, M.J., Freda, J., Heltcher, K., Marmorek, D., Rowe, L., Scanlone, P., Suter, G., Warren-Hicks, W., and Welbourn, P. 1990. Biological effects of changes in surface water acid-base chemistry. NAPAP Report 13. In: National Acid Precipitation Assessment Program, Acidic Deposition: State of Science and Technology. Vol. II.

Baker, J.P., Van Sickle, J., Gagen, C.J., DeWalle, D.R., Sharpe, W.E., Carline, R.F., Baldigo, B.P., Murdoch, P.S., Bath, D.W., Kretser, W.A., Simonin, H.A., Wigington, P.J., Jr. 1996. Episodic acidification of small streams in the northeastern United States: effects on fish populations. Ecological Applications. 422-437.

Baldigo, B.P., and Murdoch, P.S. 2007. Effect of stream acidification and inorganic aluminum on mortality of brook trout (*Salvelinus fontinalis*) in the Catskill Mountains, New York. Canadian Journal of Fisheries and Aquatic Science. 54: 603-615.

Baldigo, B.P., Lawrence, G.B., Bode, R.W., Simonin, H.A., Roy, K.M., and Smith, A.J. 2009. Impacts of acidification on macroinvertebrate communities in streams of the western Adirondack Mountains, New York, USA. Ecological Indicators. 9: 226-239.

Bradley, D.C., and Ormerod, S.J. 2002. Long-term effects of catchment liming on invertebrates in upland streams. Freshwater Biology. 47: 161-171.

Brocksen, R.W., Marcus, M.D., and Olem, H. 1992. Practical guide to managing acidic surface waters and their fisheries. Lewis Publishers, Inc. Chelsea, Michigan. 190 p.

Charlson, R.J. and Rodhe, H. 1982. Factors controlling the acidity of natural rainwater. Nature. 295: 683-685.

Clair, T.A., and Hindar, A. 2005. Liming for the mitigation of acid rain effects in freshwaters: a review of recent results. Environmental Reviews. 13: 91-128.

Cunjak, R.A., Roussel, J.-M., Gray, M.A., Dietrich, J.P., Cartwright, D.F., Munkittrick, K.R., and Jardine, T.D. 2005. Using stable isotope analysis with telemetry or mark-recapture data to identify fish movement and foraging. Oecologia. 144: 1-11.

Davies. S.P., Drummond, F., Courtemanch, D.L., Tsomides, L., and Danielson, T.J. 2016. Biological water quality standards to achieve biological condition goals in Maine rivers and streams: Science and policy. Maine Agricultural and Forest Experiment Station. Technical Bulletin 208.

Dennis, I.F. and Clair, T.A. 2012. The distribution of dissolved aluminum in Atlantic salmon (*Salmo salar*) rivers in Atlantic Canada and its potential effect on aquatic populations. Canadian Journal of Fisheries and Aquatic Science. 69: 1174-1183.

Driscoll, C.T., Lawrence, G.B., Bulger, A.J. Butler, T.J., Cronan, C.S., Eagar, C., Lambert, K.F., Likens, G.E., Stoddard, J.L., and Weathers, K.C. 2001. Acidic deposition in the Northeastern United States: sources and inputs, ecosystem effects, and management strategies. BioScience. 51.3: 180-198.

Driscoll, C.T., Driscoll, K.M., Mitchell, M.J., and Raynal, D.J. 2003. Effects of acidic deposition on forest and aquatic ecosystems in New York State. Environmental Pollution. 123: 327-336.

Elliott, J.M., and Elliott, J.A. 2010. Temperature requirements of Atlantic salmon *Salmo salar,* brown trout *Salmo trutta* and Arctic charr *Salvelinus alpinus*: predicting the effects of climate change. Journal of Fish Biology. 77: 1793-1817.

Fjellheim, A., Tysse, A., and Bjerknes, V. 2001. Reappearance of highly acid-sensitive invertebrates after liming of an alpine lake ecosystem. Water, Air and Soil Pollution. 130: 1391-1396.

Frechette, D.M., Dugdale, S.J., Dodson, J.J., and Bergeron, N.E. 2018. Understanding summertime thermal refuge use by adult Atlantic salmon using remote sensing, river temperature monitoring, and acoustic telemetry. Canadian Journal of Fisheries and Aquatic Sciences. 75: 1999-2010.

Garmo, Ø.A., Skjelkvåle, B.L., de Wit, H.A., Colombo L., Curtis, C., Fölster, J., Hoffmann, A., Hruška, J., Høgåsen, T., Jeffries, D.S., Keller, W.B., Krám, P., Majer, V., Monteith, D.T., Paterson, A.M., Rogora, M., Rzychon, D., Steingruber, S., Stoddard, J.L., Vuorenmaa, J., and Worsztynowicz, A. 2014. Trends in surface water chemistry in acidified areas in Europe and North America from 1990 to 2008. Water, Air, and Soil Pollution. 225: 1880.

Grader, M., and Letcher, B.H. 2006. Diel and seasonal variation in food habits of Atlantic salmon parr in a small stream. Journal of Freshwater Ecology. 21(3): 503-517.

Haines, T.A., Norton, S.A., Kahl, J.S., Fay, C.W., Pauwels, S.J., and Jagoe, C.H. 1990. Intensive studies of stream fish populations in Maine. EPA/600/3-90/043.

Halfyard, E. 2007. Initial results of an Atlantic salmon river acid mitigation program. MSc Thesis, Acadia University, 164 p.

Heath, R.H., Kahl, J.S., and Norton, S.A. 1992. Episodic stream acidification caused by atmospheric deposition of sea salts at Acadia National Park, Maine, United States. Water Resources Research. 28(4): 1081-1088.

Henriksen, A., Skogheim, O.K., and Rosseland, B.O. 1984. Episodic changes in pH and aluminum-speciation kill fish in a Norwegian salmon river. Vatten. 40: 255-260.

Hesthagen, T., Larsen, B.M., and Fiske, P. 2011. Liming restores Atlantic salmon (*Salmo salar*) populations in acidified Norwegian rivers. Canadian Journal of Fisheries and Aquatic Sciences. 68: 224-231.

Izaac Walton League of America (IWLA). 2021. Biological monitoring instructions for stream monitors. URL https://www.iwla.org/water/resources-for-monitors.

Jonsson, B., Forseth, T., Jensen, A.J., and Næsje, T.F. 2001. Thermal performance of juvenile Atlantic Salmon, *Salmo salar*. Functional Ecology. 15: 701-711.

Kahl, J.S., Norton, S.A., Haines, T.A., Rochette, E.Z., Heath, R.H., and Nodvin, S.C. 1992. Mechanisms of episodic acidification in low-order streams in Maine, USA. Environmental Pollution. 78: 37-44.

Kowalik, R.A., and Ormerod, S.J. 2006. Intensive sampling and transplantation experiments reveal continued effects of episodic acidification on sensitive stream invertebrates. Freshwater Biology. 51: 180-191.

Kroglund, F., and Staurnes, M. 1999. Water quality requirements of smolting Atlantic salmon (*Salmo salar*) in limed acid rivers. Canadian Journal of Fisheries and Aquatic Sciences. 56: 2078-2086.

Kroglund, F., Wright, R.F., and Burchart, C. 2002. Acidification and Atlantic salmon: critical limits for Norwegian rivers. Norwegian Institute for Water Research, Oslo. Report nr 111.

Kroglund, F., Rosseland, B.O., Teien, H.-C., Salbu, B., Kristensen, T., and Finstad, B. 2008. Water quality limits for Atlantic salmon (*Salmo salar*) exposed to short term reductions in pH and increased aluminum simulating episodes. Hydrology and Earth Systems Sciences. 12: 491-507.

Lacroix, G.L., and Knox, D. 2005. Acidification status of rivers in several regions of Nova Scotia and potential impacts on Atlantic salmon, Canadian Technical Report of Fisheries and Aquatic Sciences, 2573.

Lawrence, G.B., Sutherland, J.W., Boylen, C.W., Nierzwicki-Bauer, S.W., Momen, B., Baldigo, B.P., and Simonin, H.A. 2007. Acid rain effects on aluminum mobilization clarified by inclusion of strong organic acids. Environmental Science and Technology. 41 (1): 93-98.

Lien, L., Raddum, G.G., Fjellheim, A., Henriksen, A. 1996. A critical limit for acid neutralizing capacity in Norwegian surface waters, based on new analyses of fish and invertebrate responses. The Science of the Total Environment. 177: 173-193.

Lund, S.G., Caissie, D., Cunjak, R.A., Vijayan, M.M., and Tufts, B.L. 2002. The effects of environmental heat stress on heat-shock mRNA and protein expression in Miramichi Atlantic salmon (*Salmo salar*) parr. Canadian Journal of Fisheries and Aquatic Sciences. 59: 1553-1562.

MacAvoy, S.E., and Bulger, A.J. 1995. Survival of brook trout (*Salvelinus fontinalis*) embryos and fry in streams of different acid sensitivity in Shenandoah National Park, USA. Water, Air, and Soil Pollution. 85: 445-450.

Magee, J.A., Obedzinski, M., McCormick, S.D., and Kocik, J.F. 2003. Effects of episodic acidification on Atlantic salmon (*Salmo salar*) smolts. Canadian Journal of Fisheries and Aquaculture Science. 60: 214-221.

Maine Department of Environmental Protection Code of Maine Rules (MDEP CMR). Chapter 584: Surface Water Quality Criteria for Toxic Pollutants.

Maine Department of Environmental Protection. 2014. QAPP for Biological Monitoring of Maine's Rivers, Streams, and Freshwater Wetlands. Appendix Di: Methods for Biological Sampling and Analysis of Maine's Rivers and Streams. DEP-LW-0387-C2014, revised date 4/1/2014.

Maine Department of Environmental Protection. 2016. Continuous Monitoring of Water Quality SOP, revision No. 1, effective date 6/7/2016.

Maine Department of Environmental Protection (MDEP). 2021. Quality Assurance Project Plan for the Salmon Habitat Monitoring Program. Effective date 2/22/2021.

Maine Revised Statutes (M.R.S.). Title 38: Waters and navigation. Chapter 3: Protection and improvement of waters. Article 4-A: Water Classification Program. Section 465. URL http://www.mainelegislature.org/ legis/statutes/38/title38sec465.html.

McCormick, S.D., Hansen, L.P., Quinn, T.P, and Saunders, R.L. 1998. Movement, migration, and smolting of Atlantic salmon (*Salmo salar*). Canadian Journal of Fisheries and Aquatic Science. 55 (Suppl. 1): 77-92.

McCormick, S.D., Keyes, A., Nislow, K.H., and Monette, M.Y. 2009. Impacts of episodic acidification on in-stream survival and physiological impairment of Atlantic salmon (*Salmo salar*) smolts.

Canadian Journal of Fisheries and Aquatic Science. 66: 394-403.

McDonald, D.G., Hobe, H., and Wood, C.M. 1980. The influence of calcium on the physiological responses of the rainbow trout, *Salmo gairdneri*, to low environmental pH. Journal of Experimental Biology. 88: 109-131.

Monette, M.Y., Björnsson, B.T., and McCormick, S.D. 2008. Effects of short-term acid and aluminum exposure on the parr-smolt transformation in Atlantic salmon (*Salmo salar*): disruption of seawater tolerance and endocrine status. General and Comparative Endocrinology. 158: 122-130.

National Atmospheric Deposition Program (NADP). 2023. Station ME94, Indian Township and station ME98, Acadia National Park – McFarland Hill. NADP Program Office, Wisconsin State Laboratory of Hygiene, Madison, WI. URL https://nadp.slh.wisc.edu/maps-data/ntn-interactive-map/.

National Oceanic and Atmospheric Administration (NOAA). 2023. Gulf of Maine region quarterly climate impacts and outlook. June, September and December 2023. URL https://gulfofmaine.org/public/climate-network/climate-outlook/.

National Research Council (NRC). 2004. Atlantic Salmon in Maine. Washington, DC: The National Academies Press. URL https://doi.org/10.17226/10892.

Ojala, J.V. 2008. Invertebrate phenology and prey selection of three sympatric species of salmonids; implications for individual fish growth. University of Massachusetts Amherst masters thesis 1911 – February 2014. pp 97.

Potter, W. 1982. The effects of air pollution and acid rain on fish, wildlife, and their habitats – rivers and streams.

U.S. Fish and Wildlife Service, Biological Services Program, Eastern Energy and Land Use Team, FWS/OBS-80/40.5. 52 pp.

Project Share and U.S. Fish and Wildlife Service (USFWS). 2009. Restoring salmonid aquatic/riparian habitat: a strategic plan for the Downeast Maine DPS rivers.

R Core Team. 2023. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org/.

Stanley, J.G., and Trial, J.G. 1995. Habitat suitability index models: nonmigratory freshwater life stages of Atlantic salmon. U.S. Department of the Interior. Biological Science Report 3.

Staurnes, M. Kroglund, F., and Rosseland, B.O. 1995. Water quality requirement of Atlantic salmon (*Salmo salar*) in water undergoing acidification or liming in Norway. Water, Air, and Soil Pollution. 85: 347-352.

Tipping, E., Woof, C., and Hurley, M.A. 1991. Humic substances in acid surface waters; modelling aluminum binding, contribution to ionic charge-balance, and control of pH. Water Resources. 25(4): 425–435.

United States Atlantic Salmon Assessment Committee (USASAC). 2022. Annual Report, no. 34 – 2021 activities.

United State Drought Monitor. 2023. National Drought Mitigation Center, Lincoln, NE. URL https://droughtmonitor.unl.edu/Maps/MapArchive.aspx

United States Environmental Protection Agency (USEPA). 1986. Quality Criteria for Water. EPA 440/5-86-001. URL https://www.epa.gov/sites/default/files/2018-10/documents/quality-criteria-water-1986.pdf.

United States Environmental Protection Agency. 2018. Final Aquatic Life Ambient Water Quality Criteria for Aluminum. EPA- 822-R-18-001.

Weather Underground. 2023. Tom's Backyard Personal Weather Station KMEALEXA2, Alexander Elementary School KMEBAILE9, HeatherWood Gardens KMEBARIN2, Eastridge KMECOOPE3, Accurite KMECOOPE5, Heather Wood Farm KMEPRINC2. URL ttps://www.wunderground.com/.

Whiting, M.C. 2014. Final report for Project SHARE's Clam Shell Pilot Project. Maine Department of Environmental Protection: Bangor, Maine.

Whiting, M.C. and Otto, W. 2008. Spatial and temporal patterns in the water chemistry of the Narraguagus River: a summary of the available data from the Maine DEP Salmon Rivers Program. Maine Department of Environmental Protection: Bangor, Maine.

Wickham, H. 2009. ggplot2: Elegant Graphics for Data Analysis. Springer-Verlag New York.

Wiederholm, T. 1984. Responses of aquatic insects to environmental pollution. In V.H. Resh and D.M. Rosenberg (Eds.), *The Ecology of Aquatic Insects*. (pp. 530-535). Praeger Publishers.

Wilson Engineering, LLC. 2023. Water Resources Database (WRDB). St. Louis, Missouri. URL wrdb.com.

Wood, C.M., McDonald, D.G., Ingersoll, C.G., Mount, D.R., Johannsson, O.E., Landsberger, S., and Bergman, H.L. 1990. Effects of water acidity, calcium, and aluminum on whole body ions of brook trout (*Salvelinus fontinalis*) continuously exposed from fertilization to swim-up: a study by instrumental neutron activation analysis. Canadian Journal of Fisheries and Aquatic Sciences. 47: 1593-1603.

Zimmermann, E. 2018. Reducing acidification in endangered Atlantic salmon habitat: baseline data. Maine Department of Environmental Protection: Augusta, ME. URL https://www.maine.gov/dep/water/monitoring/rivers_and_streams/salmon/2017-downeast-baseline-report.pdf.

Zimmermann, E. 2019. Reducing acidification in endangered Atlantic salmon habitat: baseline data summary. Maine Department of Environmental Protection: Augusta, ME. URL https://www.maine.gov/dep/water/ monitoring/rivers_and_streams/salmon/2018-downeast-baseline-summary-report.pdf.

Zimmermann, E. 2020. Reducing acidification in endangered Atlantic salmon habitat: first year of clam shells. Maine Department of Environmental Protection: Augusta, ME. URL https://www.maine.gov/dep/water/monitoring/rivers_and_streams/salmon/first-year-of-clam-shells.pdf.

Zimmermann, E. 2023. Reducing acidification in endangered Atlantic salmon habitat: fourth year of clam shells. Maine Department of Environmental Protection: Augusta, ME. URL https://www.maine.gov/dep/water/monitoring/rivers_and_streams/salmon/fourth-year-of-clam-shells.pdf.

Appendix I – Methodology

Table I-1. Analytical parameters, methods, laboratories and certification.

Analyte	Method	Analysis Lab	DEP Certified?
ANC	E600/4-87/26 5.5.1	UMO Sawyer Water Research Lab	No*
Aluminum (speciation)	SW6010B	UMO Sawyer Water Research Lab	No*
pH (closed-cell)	E600/4-87/26 19.0	UMO Sawyer Water Research Lab	No*
Calcium and other cations	E200.7	Maine Environmental	Yes
DOC	SM5310B	Maine Environmental	Yes
Anions (chloride and sulfate)	E300.0	Maine Environmental	Yes
Nitrate	E353.2	Eastern Analytic, Inc.	Yes

**Research method with no state certification, approved by the DEP biologist, and used for continuity of data to compare with prior liming projects.*

Table I-2. Clam shell application dates and quantities.

Year	Application Date	Quantity (m^3)	Clam Shell Type
2019	July 24 – Oct. 8	23	*Mya arenaria* (soft-shell clam)
2020	July 26 and Aug. 13	26	*Mya arenaria* (soft-shell clam)
2021	Sept. 7 – 10	23	*Mya arenaria* (soft-shell clam) and *Merceneria mercenaria* (quahog)
2022	Sept. 8 – 9	18	*Merceneria mercenaria* (quahog)

Appendix II – Data Summary Tables

Table II-1. Continuous data summary statistics. Mean ± standard deviation (SD), minimum and maximum of measurements from Manta+ 20 sondes, May to Nov. 2023 (n ~ 8,000).

Stream Name	pH	pH Min	pH Max	Temperature (Temp; °C)	Temp Min	Temp Max	Specific Conductance (SPC; µS/cm)	SPC Min	SPC Max	Dissolved Oxygen (DO; mg/L)	DO Min	DO Max
Barney Brook	6.32 ± 0.47	4.48	7.31	14.50 ± 3.47	6.18	22.12	27 ± 5	19	43	9.74 ± 1.06	7.76	13.47
Beaverdam Stream	5.87 ± 0.26	5.02	6.45	16.43 ± 5.08	2.24	26.49	28 ± 6	4	63	9.03 ± 1.26	7.25	13.25
Creamer Brook	5.60 ± 0.30	4.57	6.30	14.91 ± 4.74	1.59	23.48	20 ± 2	16	25	9.56 ± 1.29	7.66	14.11
Richardson Brook – 09 (upstream control)	5.52 ± 0.30	4.62	6.26	15.90 ± 4.89	1.89	26.00	20 ± 2	5	25	8.87 ± 1.40	6.51	13.63
Richardson Brook – B (treatment)	5.89 ± 0.33	5.00	6.57	15.73 ± 4.98	1.54	25.00	21 ± 2	18	32	9.50 ± 1.30	7.36	14.00

Table II-2. Discrete data summary statistics. Mean ± SD, minimum and maximum from grab samples collected 2017-2023. n = 24, except n = 18 at Creamer Brook (no April samples) and n = 20 at Beaverdam Stream (no 2017 data).

Stream Name	Calcium (Ca; mg/L)	Ca Min	Ca Max	Dissolved Organic Carbon (DOC; mg/L)	DOC Min	DOC Max	ANC (µeq/L)	ANC Min	ANC Max	pH (closed-cell)	pH cc Min	pH cc Max
Barney Brook	2.94 ± 1.33	1.16	5.98	11.6 ± 5.7	3.4	27	158.6 ± 102.0	46.6	435.9	6.45 ± 0.33	5.82	6.96
Beaverdam Stream	2.08 ± 0.73	0.92	3.50	12.4 ± 5.5	6.1	26	88.9 ± 73.9	31.4	322.3	6.09 ± 0.42	5.28	6.88
Creamer Brook	1.74 ± 0.66	0.91	3.66	13.0 ± 4.7	7.6	23	51.7 ± 26.2	11.2	94.9	5.77 ± 0.41	4.96	6.37
Richardson Brook - 09 (upstream control)	1.39 ± 0.40	0.72	2.10	13.0 ± 4.7	5.6	23	48.0 ± 26.4	13.3	104.0	5.64 ± 0.43	4.80	6.25
Richardson Brook – B (treatment)	1.78 ± 1.02	0.70	5.90	12.7 ± 4.3	6.8	22	71.1 ± 72.0	13.9	366.5	5.92 ± 0.48	4.94	6.86

Table II-3. Treatment summary. Mean values (± SD) before any change in pH was observed at the treated Richardson Brook site ('pre': May 30, 2017 – July 26, 2020) and after pH increased ('post': July 26, 2020 – Nov. 13, 2023).

Stream Name	**pH Pre** *n ~ 30,000*	**pH Post** *n ~ 43,000*	**Ca (mg/L) Pre** *n = 11**	**Ca (mg/L) Post** *n = 13**	**Alx (µg/L) Pre** *n = 11**	**Alx (µg/L) Post** *n = 13**	**ANC (µEq/L) Pre** *n = 11**	**ANC (µEq/L) Post** *n = 13**
Barney Brook	6.3 ± 0.3	6.3 ± 0.4	3.2 ± 1.6	2.7 ± 1.0	16.5 ± 15.6	15.0 ± 12.1	177 ± 128	143 ± 75
Beaverdam Stream	6.1 ± 0.5	6.1 ± 0.4	1.9 ± 0.8	2.1 ± 0.7	8.9 ± 6.9	10.7 ± 7.7	107 ± 104	79 ± 55
Creamer Brook	5.7 ± 0.4	5.5 ± 0.4	1.9 ± 0.8	1.6 ± 0.6	29.5 ± 18.0	31.9 ± 17.5	58 ± 30	47 ± 23
Richardson Brook – 09+ (upstream control)	5.6 ± 0.4	5.4 ± 0.4	1.3 ± 0.4	1.4 ± 0.4	15.8 ± 10.6	14.5 ± 10.2	51 ± 31	46 ± 23
Richardson Brook – B (treatment)	5.5 ± 0.5	5.8 ± 0.5	1.5 ± 0.6	2.0 ± 1.3	18.6 ± 9.4	18.0 ± 16.9	62 ± 45	79 ± 90

* Creamer Brook was not sampled in April 2018-2022 (n = 8 pre, 10 post). Beaverdam Stream was not sampled in 2017 (n = 8 pre).
+ Rich09 includes samples collected from Rich-A (a site 360m downstream) in 2017, 2018, and April 2019 (pre).

Table II-4. Aluminum species data summary statistics. Mean ± SD, minimum and maximum from grab samples collected 2017-2023. n = 24, except n = 18 at Creamer Brook (no April samples) and n = 20 at Beaverdam Stream (no 2017 data).

Stream Name	**Total Aluminum (µg/L)**	**Tot. Al. Min**	**Tot. Al. Max**	**Dissolved Aluminum (µg/L)**	**Diss. Al. Min**	**Diss. Al. Max**	**Exchangeable Aluminum (Alx; µg/L)**	**Alx Min**	**Alx Max**
Barney Brook	199 ± 108	40	537	172 ± 94	32	465	16 ± 14	2	54
Beaverdam Stream	153 ± 71	54	386	131 ± 64	32	332	11 ± 7	<1	25
Creamer Brook	256 ± 115	94	557	229 ± 106	92	505	31 ± 17	1	56
Richardson Brook – 09+ (upstream control)	202 ± 79	101	448	184 ± 69	75	388	15 ± 10	2	40
Richardson Brook – B (treatment)	196 ± 81	88	474	181 ± 75	79	432	18 ± 14	2	57

Table II-5. Exceedance summary. Percentage of data observations that exceeded stress threshold values for sonde data (pH, temperature and DO) April-Nov. 2023 (n ~ 8,000). Grab sample data (calcium and exchangeable aluminum) combine all seven years of the study 2017-2023 (n = 24, except n = 18 at Creamer Brook (no April samples) and n = 20 at Beaverdam Stream (no 2017 data).

Stream Name	pH <5.5	pH <6.5	Temperature >20.0 °C	DO <5 mg/L	DO <7 mg/L	Calcium <2.0 mg/L*	Calcium <4.0 mg/L*	Alx >15 µg/L*
Barney Brook	6.8	55.7	3.4	0	0	33.3	75.0	33.3
Beaverdam Stream[a]	9.5	100	26.5	0	0	45.0	100	25.0
Creamer Brook	43.7	100	11.0	0	0	83.3	100	66.7
Richardson Brook – 09[+] (upstream control)	45.4	100	19.3	0	3.8	91.7	100	37.5
Richardson Brook – B (treatment)	22.0	99.2	18.2	0	0	70.8	95.8	54.2

Table II-6. Base cation surplus (BCS) summary statistics. Mean ± standard deviation from grab samples collected April-Nov. 2019-2023. Cations include calcium, potassium, magnesium, and sodium. Anions include chloride, nitrate, sulfate, and strong organic anions (0.071*DOC-2.1, from Lawrence et al. 2007). Data converted from mg/L. n = 17, except n = 13 at Creamer Brook (no April samples).

Stream Name	Cations (µEq/L)	Anions (µEq/L)	BCS (µEq/L)	BCS Min	BCS Max
Barney Brook	412.2 ± 31.5	84.4 ± 28.2	327.8 ± 3.3	325.4	330.1
Beaverdam Stream	383.5 ± 28.9	246.8 ± 97.0	88.6 ± 184.9	88.6	184.9
Creamer Brook	230.6 ± 8.35	137.6 ± 75.3	93.0 ± 66.9	45.7	140.3
Richardson Brook – 09 (upstream control)	210.4 ± 8.4	68.6 ± 26.7	141.8 ± 35.1	117.0	166.7
Richardson Brook – B (treatment)	229.2 ± 11.8	81.0 ± 36.4	148.2 ± 48.2	114.1	182.2

Table II-7. Nutrient data summary statistics. Mean ± standard deviation from grab samples collected during summer baseflow 2018-2023. n = 5, except n = 6 at Creamer Brook for N+N due resampling in 2019.

Stream Name	Nitrate + Nitrite as Nitrogen (N+N; mg/L)	Total Kjeldahl Nitrogen (TKN; mg/L)	Total Phosphorus (µg/L)
Barney Brook	0.068 ± 0.032	0.49 ± 0.14	20 ± 4
Beaverdam Stream	0.023 ± 0.014	0.78 ± 0.36	20 ± 12
Creamer Brook	0.048 ± 0.032	0.49 ± 0.14	17 ± 3
Richardson Brook – 09 (upstream control)	0.030 ± 0.027	0.68 ± 0.10	16 ± 5
Richardson Brook – B (treatment)	0.032 ± 0.020	0.67 ± 0.10	15 ± 5

Table II-8. Macroinvertebrate summary. Samples were collected in August using rock bags following the Biological Monitoring Unit's protocol (MDEP 2014) and analyzed by a certified taxonomist to the lowest possible level (species). EPT taxa include mayflies (Ephemeroptera), stoneflies (Plecoptera), and caddisflies (Trichoptera).

Log #	Stream Name	Station ID	Years Sampled	Total Mean Abundance	Generic Richness	EPT Generic Richness	Relative Ephemeroptera Abundance
2687	Beaverdam Stream	S-1149	2018	164	39	14	14%
2764	Beaverdam Stream	S-1149	2019	396	36	18	10%
2833	Beaverdam Stream	S-1149	2020	234	57	20	16%
2977	Beaverdam Stream	S-1149	2022	256	46	17	25%
2589	Creamer Brook	S-1115	2017	207	40	20	28%
2690	Creamer Brook	S-1115	2018	246	37	16	77%
2763	Creamer Brook	S-1115	2019	96	36	16	36%
2834	Creamer Brook	S-1115	2020	194	53	21	37%
2980	Creamer Brook	S-1115	2022	121	21	7	62%
2591	Richardson Brook - A	S-1117	2017	106	37	19	49%
2689	Richardson Brook - A	S-1117	2018	104	31	13	42%
2836	Richardson Brook - A	S-1117	2020	80	40	15	42%
2981	Richardson Brook - A	S-1117	2022	43	14	6	39%
2590	Richardson Brook - B	S-1116	2017	56	31	13	31%
2688	Richardson Brook - B	S-1116	2018	89	43	21	30%
2835	Richardson Brook - B	S-1116	2020	122	47	17	27%
2982	Richardson Brook - B	S-1116	2022	67	17	7	58%

Appendix III – Biological Monitoring Program Reports

Macroinvertebrates
Beaverdam Stream – Station 1149 URL
https://www.maine.gov/dep/gis/datamaps/lawb_biomonitoring/station_web/S-1149M.html

Creamer Brook – Station 1115 URL
https://www.maine.gov/dep/gis/datamaps/lawb_biomonitoring/station_web/S-1115M.html

Richardson Brook – Station 1117 URL
https://www.maine.gov/dep/gis/datamaps/lawb_biomonitoring/station_web/S-1117M.html

Richardson Brook – Station 1116 URL
https://www.maine.gov/dep/gis/datamaps/lawb_biomonitoring/station_web/S-1116M.html

Algae
Beaverdam Stream – Station 1149 URL
https://www.maine.gov/dep/gis/datamaps/lawb_biomonitoring/station_web/S-1149A.html

Creamer Brook – Station 1115 URL
https://www.maine.gov/dep/gis/datamaps/lawb_biomonitoring/station_web/S-1115A.html

Richardson Brook – Station 1117 URL
https://www.maine.gov/dep/gis/datamaps/lawb_biomonitoring/station_web/S-1117A.html

Richardson Brook – Station 1116 URL
https://www.maine.gov/dep/gis/datamaps/lawb_biomonitoring/station_web/S-1116A.html