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Effects of Long-Term Nitrogen Fertilization and Application Methods on Fruit Yield, Plant Nutrition, and Soil Chemical Properties in Highbush Blueberries

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Abstract: Nitrogen (N) fertilizer is routinely applied in highbush blueberry (*Vaccinium corymbosum* L.) production. The recommended N fertilizer rate increases as the plants mature, and is usually determined based on regional growing conditions. However, the effects of N fertilizer rates and application methods over the long term remain poorly understood. In this study, ammonium sulfate was applied as an N source at the recommended rate (100%), which corresponds to a maximum of 155 kg N ha⁻¹ for plants older than eight years, along with higher rates at 150% and 200% of the recommended level, as well as a control treatment of no N. Treatments were applied to the blueberry cultivar 'Duke' as either broadcast (BROAD) or fertigation (FERT), and impacts were analyzed after 12 and 13 years of treatment. In the 14th year, the 100% N rate was uniformly applied as BROAD across all plants to separate the effects of different N rates from those caused by long-term soil condition changes. The BROAD treatment at the 100% N rate achieved the highest yield, and the FERT treatment at 200% resulted in the lowest yield in the 12th year, suggesting that excessive N rates can reduce fruit yield. However, no significant yield differences were observed in the 13th year. Higher N rates were associated with reduced titratable acidity in fruits and fewer flower buds. The soil pH declined across all N treatments, with the FERT at 200% showing the most significant reduction. All N treatments generally increased soil electrical conductivity (EC). High N rates also decreased plant accumulation of magnesium, calcium, and copper, with the latter reaching deficiency levels. These findings emphasize the importance of adhering to recommended N application rates and adjusting soil pH and EC to mitigate the adverse effects of prolonged N treatments.

Keywords: fertigation; electrical conductivity; soil pH; leaf nutrient levels; ammonium N (NH_4^+); titratable acidity; flower buds



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1. Introduction

The northern highbush blueberry (*Vaccinium corymbosum* L.), a member of the Ericaceae family, was originally cultivated as a fruit crop in North America. Today, it is widely grown in many regions worldwide [1]. These plants are adapted to acidic soils with a pH range of 4.2 to 5.5 [2]. Given the limited availability of nutrients in low-pH soils, blueberries have evolved to grow with low nutrient requirements. However, in commercial cultivation, a proper fertilization regime primarily, if not exclusively, composed of nitrogen (N) fertilizer is necessary for optimal fruit production [2,3]. Additionally, blueberries have a shallow root system, with most roots concentrated within the top 30 cm of soil [4]. As the root zone can quickly become depleted of water during periods of high demand, making the plants more susceptible to drought stress, commercial productions also require frequent irrigation [5].

In Canada, highbush blueberry production is primarily concentrated in the province of British Columbia [6]. In this region, as in many other parts of the world, blueberries are planted at high densities in raised beds covered with sawdust mulch. Irrigation is

commonly provided through drip systems [1,7]. The N fertilizer application follows age-based recommendations, usually developed for the growing region as appropriate for the soil type and fertility levels [2,8]. According to the British Columbia Blueberry Production Guide, 15 kg N ha $^{-1}$ is recommended for one-year-old plants, assuming 2470 plants per hectare. As the plants mature, the N rate gradually increases, reaching 115 kg N ha $^{-1}$ at eight years, with recommendations of up to 155 kg N ha $^{-1}$ for older plants [9].

Blueberry plants take up inorganic N as nitrate (NO_3^-) and ammonium (NH_4^+) forms. However, in the acidic soils where they naturally grow, applying the NH_4^+ form of N leads to better growth and higher N accumulation, showing a preference for the NH_4^+ form [10,11]. Consequently, the most commonly used N fertilizers for blueberries include urea and ammonium sulfate, traditionally applied as granular (broadcast) applications. However, applying fertilizers through the irrigation system (fertigation) is becoming increasingly popular [2].

Several studies have investigated the effects of varying N rates and application methods on blueberry production. These studies show that applying N fertilizer via fertigation (FERT) may enhance plant growth and yield compared to broadcast (BROAD) applications, particularly in young, establishing plants [8,12–16]. However, because the NH₄+ form of N is less mobile and drip systems may not distribute fertilizer uniformly around the base of the plants, FERT might require a higher amount of N fertilizer than BROAD applications [8,17]. Although applying adequate fertilizer is important, excessive rates can lead to adverse effects, including reduced yield and growth [12,18,19] and, in some cases, even plant injury or death [8,20].

Despite the above research on N application methods and rates for blueberries, current knowledge is mostly limited to short-term evaluations of about 2 to 4 years, primarily conducted on establishing fields. However, northern highbush blueberries are perennials grown in the field for many years. The plants take about eight years to mature and can remain in fruit production for approximately 20 to 30 years after planting [21]. Therefore, long-term treatments are necessary to understand how sustained fertilizer rates and application methods affect fruit production and soil properties in blueberry orchards.

The objective of this study was to evaluate the effects of prolonged N fertilizer treatments, in relation to application rates and application methods, on flower bud development, fruit yield and quality. We also assessed the evolution of key soil properties, including pH, EC, and soil mineral N. Results of soil properties pertaining to this long-term site were previously reported [22].

2. Materials and Methods

2.1. Experimental Site and Study Design

The study was conducted in a northern highbush blueberry planting (*Vaccinium corymbosum*) of cv. 'Duke' at the Agassiz Research and Developmental Center of the Agriculture and Agri-Food Canada, located in Agassiz, British Columbia (49°14′ N, 121°45′ W). The details of the field setup, climate conditions and soil properties have been previously described [13,22,23]. The weather data for the evaluation period and the normal weather data are provided in Supplementary Table S1. The planting was carried out in 2008 after applying elemental sulfur at 1120 kg ha⁻¹ to lower the initial pH from 5.6 to 5.0. At the start of the experiment, the total C was 3.24%, and the total N was 0.26%. The soil mineral N content within 30 cm depth was 29 kg N ha⁻¹ [13]. Planting was carried out in 1 m wide, 20 cm high raised beds, with 90 cm spacing between plants. The beds were mulched with an approximately 8 to 10 cm thick layer of Western hemlock and Douglas fir sawdust, which was replenished roughly every other year.

The experiment was designed as a randomized complete block design with six replicates. Each plot consisted of six plants and a guard plant or row spacing separating the adjacent plots (Supplementary Figure S1). Each planting row was irrigated with two drip lines suspended along the edges of the row, approximately 19 cm from the center and at

a height of 60 cm. The plants were treated with approximately $11.5 \text{ kg P}_2\text{O}_5 \text{ ha}^{-1}$ (triple super phosphate) and $15.8 \text{ kg K}_2\text{O ha}^{-1}$ (K-Mag 0-0-21.5) twice a year, based on foliar analysis until plant maturity. Since 2015, no fertilizer treatments have been applied other than the N treatments described below. Irrigation was applied based on weather conditions, and plants were exposed to open pollination by natural pollinators. Plants were pruned annually during dormancy according to industry standards, with approximately 30% of the wood removed from each plant, focusing on less vigorous and twiggy branches [9,24].

2.2. Nitrogen Treatments and Experiment Period

Ammonium sulfate (21-0-0) was used as the N fertilizer, applied either as a BROAD treatment or dissolved in irrigation water for the FERT treatment. From 2008 to 2012, N was applied at 50%, 100%, and 150% of the recommended rates, calculated on a per-plant basis, according to the provincial blueberry production guide [9]. As the plants matured, a supplementary amount of N equivalent to 50% of the recommended rate was added to each treatment. Accordingly, from 2013 onward, the plants received 100%, 150%, and 200% of the recommended N rate [22,23]. For plants aged eight years or older, the recommended rate (100%) is 115 kg N ha $^{-1}$ to 155 kg N ha $^{-1}$ [9]. A 0% control was also included, which received no N fertilizer throughout the study. All fertilizer treatments were applied as split applications. BROAD treatments involved three equal applications once a month starting from late April to mid-May. FERT treatments were applied as 10 to 12 weekly applications during the same period.

All treatments were continued annually as described above for 13 years. In the 14th year, 100% N was applied as a BROAD treatment to all plants, including controls that had not received any N since field establishment. The data presented in this work show the impacts of these sustained treatments after 12 and 13 years, corresponding to the calendar years 2020 and 2021. Additionally, plant responses to the uniform N treatment in the 14th year are presented in 2022 data.

2.3. Bud Development Assessments, Berry Yield, and Fruit Quality

Flower and leaf bud developments were assessed in 2021. Briefly, four canes per plant (25 cm long, without branching) were cut from four plants per plot, and the total number of leaf and flower buds was counted. When two buds occurred in a particular position, they were counted separately. Since this analysis showed a differential response to FERT treatment only, we further assessed the number of flowers per inflorescence and fruit set percentage in FERT treatments. For this, two branches per plant were randomly selected, and the second and third flower bud positions from the tip were assessed in mid-June 2021. The number of pedicels to which flowers and developing fruits attached was counted to determine the number of flowers produced. The fruit set was determined through the visual assessment of actively developing fruits.

For yield assessments, harvesting was carried out by hand in 2020. Since hand harvesting of these mature blueberry plants is labor-intensive, only the first four blocks were selected, and only three plants from the center of each plot were harvested. Two rounds of harvesting were carried out as the fruit ripened. Unripe fruits were also picked during the second round to avoid a third round of picking. In 2021 and 2022, all six blocks were machine-harvested, including all plants except the guard plants separating adjacent plots. The guard plants were deblossomed to prevent accidental mixing of fruits during harvesting. The harvest was completed in a single round when the fruits were fully ripe, though there were occasionally some unripe fruits. Fruits were stored at $-20\,^{\circ}\text{C}$ after harvesting until analysis. The average yield per hectare was calculated based on an estimated density of 2470 plants per hectare [9].

For titratable acidity (TA) and total soluble solids (TSS), fruit samples were processed approximately two months after harvest. Briefly, berries were thawed at room temperature for 30 to 60 min. About 100 to 150 g of berries were weighed and ground with a handheld blender. For TA, 5 g of ground fruit samples were homogenized with 100 mL of water and

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titrated with 0.1 N NaOH using a potentiometric titrator (TitroLine Easy, Schott Instruments, Mainz, Germany) to an endpoint of pH 8.1. The TA was expressed as a percentage of citric acid equivalent (g of citric acid per 100 g) [25,26]. The percentage of TSS (°Brix) was measured using a digital refractometer (PR-32α, Atago Co., Ltd., Tokyo, Japan).

2.4. Leaf Nutrient Analysis

Leaf samples were collected from early to mid-August in 2021 and 2022. Briefly, fully opened young leaves were collected from lateral branches on five plants per plot [27]. The samples were dried at $60\,^{\circ}\text{C}$ for approximately 72 h, until the weight stabilized, and ground using a Wiley mill equipped with a 0.5 mm screen. A quantity of 5 g of the sample was microwave-digested in concentrated HNO₃, and total element content was analyzed using an iCap 7400 ICP-OES analyzer (Thermo Fisher Scientific, Waltham, MA, USA). N analysis was carried out in a LECO CN828 C and N analyzer using 0.1 g of ground leaf sample (LECO Corp., St. Joseph, MI, USA).

2.5. Soil Analysis

Soil and sawdust samples were collected in September 2020, November 2021, and November 2022. The sawdust mulch samples were collected by hand and included the approximately 10 cm thick mulch layer. Soil cores were taken at depths of 0–15 cm, 15–30 cm, and 30–60 cm after removing the sawdust, using a 2 cm diameter auger. Samples were collected from the first four blocks in 2020, all six blocks in 2021, and the first five in 2022, based on resource availability. Four to six samples were collected from each plot, representing the center of the bed between plants, the region along the drip line, and the lower half of the bed to represent different regions of the raised bed. All samples from the same plot were mixed before analysis.

 ${
m NH_4^+}$ and ${
m NO_3^-}$ N analysis in soil was conducted as described in [28]. Briefly, 5 g of moist soil (2020 and 2021) or air-dried soil (2022) was mixed with 50 mL of 2M KCl by shaking for 1 h at 84 rpm in a platform shaker. The mixture was then allowed to settle for approximately 1 h and filtered with a Q2 grade filter paper (Fisher Scientific, Ottawa, ON, Canada). The extracts were analyzed using a Lachat QuikChem 8500 Series 2 flow injection analysis system (Hach, Loveland, CO, USA), and all data were presented on a dry-weight basis. For pH and EC analysis, 10 g of fresh soil (2020 and 2021) or air-dried soil (2022) was mixed with 20 mL of deionized water, except for a few samples where further dilutions were made when EC values exceeded the 1413 μ S cm⁻¹ range of the standard solution. The samples were analyzed using a Multilab IDS 4010-3 (YSI, Yellow Springs, OH, USA) or a Seven Excellence meter (Metler Toledo, Mississauga, ON, Canada) [29,30]. The EC values are presented on a 1:2 soil dry weight-to-water volume (EC_{1:2}) basis.

2.6. Statistical Analysis

The data were analyzed using SigmaPlot software, version 15.0 (Grafiti LLC, Palo Alto, CA, USA). Replicates were treated as random effects in all assessments. When comparing the treatment effects with the control that received no N fertilizer, the combined effects of fertilizer rates and application methods were analyzed using one-way analysis of variance (ANOVA), followed by the Holm–Sidak post hoc test. To compare the impacts of N fertilizer rates and application methods, a two-way ANOVA was used, followed by the Holm–Sidak post hoc test, with data from control treatments omitted since the application method was not applicable. The number of replications for each assessment ranged from 4 to 6, as data were not collected from all six blocks each year (the specific number of samples is provided within the data). Statistical significance was declared at $p \le 0.05$.

3. Results

3.1. Berry Yield, Fruit Quality, and Flower Buds

In 2020, berry yield was influenced by the N application rate but not the application method (Supplementary Table S2). The BROAD-100% treatment produced the highest average yield of 22,076 kg ha $^{-1}$, while the FERT-200% treatment resulted in the lowest average yield of 13,873 kg ha $^{-1}$. Although not statistically significant, the average yield of the FERT-200% treatment tended to be lower than the 17,486 kg ha $^{-1}$ average yield obtained in the controls that received no N (Figure 1A). In contrast, yields in 2021 were lower across all treatments compared to 2020, with average yields of 12,366 kg ha $^{-1}$, 12,847 kg ha $^{-1}$, and 11,524 kg ha $^{-1}$ for the control, BROAD-100%, and FERT-100% treatments, respectively, showing no significant impact in response to different N rates or application methods (Figure 1B, Supplementary Table S2).

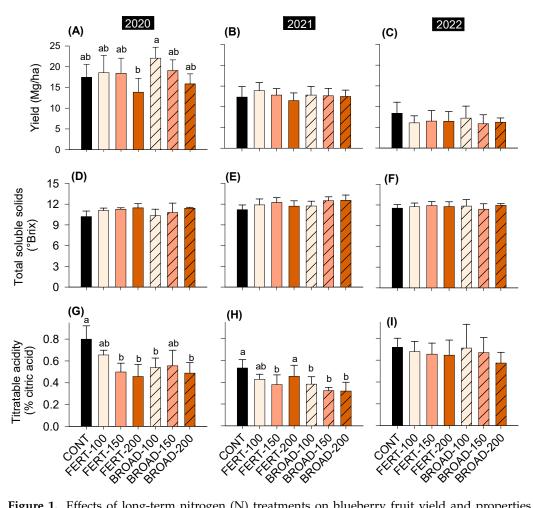


Figure 1. Effects of long-term nitrogen (N) treatments on blueberry fruit yield and properties. (A–C) Average fruit yield, (D–F) total soluble solids (TSS) content, and (G–I) titratable acidity (TA) of fruits in 2020 (A,D,G), 2021 (B,E,H), and 2022 (C,F,I). Plants were treated with N at 100%, 150%, or 200% of the rates recommended by the British Columbia Blueberry Production Guide, applied either as fertigation (FERT) or broadcast (BROAD) until 2021. In 2022, all plants received N at 100% as BROAD treatments, but data are presented according to historical N treatments. Data are means \pm SD (n = 4 for 2020; n = 6 for 2021 and 2022). Different letters indicate significant differences (one-way ANOVA followed by Holm–Sidak post hoc test; $p \le 0.05$).

The TSS content was influenced by the application method only in 2020 (Figure 1D,E; Supplementary Table S2). The TA was affected by the N rate in 2021, with a similar trend observed in 2020 (Supplementary Table S2). The influence of the N rate on TA was clearly

visible when the combined effects of the application method and N rate were analyzed, with most N treatments resulting in lower TA compared to the control in both years (Figure 1G,H). The 200% N rate resulted in the lowest average TA, with reductions of approximately 43% and 39% in the FERT and BROAD treatments, respectively, compared to the controls.

The N rate did not influence the number of leaf and flower buds or the percentage of flower buds; however, all were significantly affected by the application method (Supplementary Table S2). The combined effect of the application method and rate showed that the FERT-200% treatment had more leaf buds and fewer flower buds, both in number and percentage, compared to the BROAD-100% treatment (Figure 2A). Since the N rate alone did not influence the number of flower buds, we further investigated whether the treatment rate might affect the number of flowers produced per inflorescence. This assessment was carried out only within the FERT treatment and the control. When the second and third flower bud positions were examined, each position produced an average of 7–8 flowers per bud across all treatments, with no significant differences observed in response to the treatments (Figure 2B). Additionally, the fruit set percentage assessed based on the visual assessment of actively developing fruits in each inflorescence showed no significant differences (Figure 2C,D).

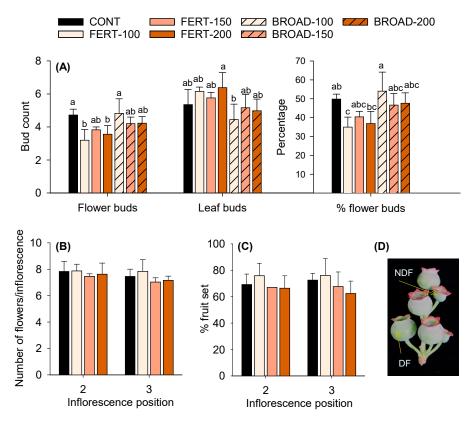


Figure 2. Effects of nitrogen (N) fertilization on reproductive development in blueberry plants. (**A**) Number of flower and leaf buds and the percentage of flower buds relative to the total bud count, measured in 25 cm long canes in 2021. Plants received N at 100%, 150%, or 200% of the rates recommended by the British Columbia Blueberry Production Guide, applied as fertigation (FERT) or broadcast (BROAD). (**B**) Number of flowers per inflorescence in the second and third inflorescence positions. (**C**,**D**) Percentage of fruit set, determined by visually estimating the proportion of developing fruits (DF) compared to non-developing fruits that remained in the cluster (NDF) or dropped. Data are means \pm SD. For bud counts, n = 4–5, with each sample composed of 16 canes. For assessment of flowers per inflorescence and fruit set percentage, n = 4, with each sample composed of eight canes. Different letters denote significant differences. No significant differences were detected in graphs without letters (one-way ANOVA followed by Holm–Sidak post hoc test; $p \le 0.05$).

3.2. Soil N Availability, pH, and Electrical Conductivity

The $\mathrm{NH_4}^+$ and $\mathrm{NO_3}^-$ levels showed considerable variability within treatments at a given soil depth, likely due to the uneven distribution of applied fertilizer. Despite this variation, notable differences emerged in response to N application methods and rates, particularly in the 0–15 cm soil layer when analyzed in 2021 (Supplementary Table S3). The sawdust mulch layer generally contained higher levels of $\mathrm{NH_4}^+$ under the BROAD treatment than the FERT treatment. This was especially pronounced in the BROAD-200% treatment, where the average $\mathrm{NH_4}^+$ level reached 3000 mg kg $^{-1}$, compared to an average of 150 mg kg $^{-1}$ in the FERT-200% treatment and 30 mg kg $^{-1}$ in the control treatment in 2021, with similar trends observed in 2020 (Figures 3 and 4). Variation in $\mathrm{NO_3}^-$ levels was also observed in 2021 but not in 2020. In 2021, the average soil $\mathrm{NO_3}^-$ level was approximately 2 mg kg $^{-1}$ in the control, 40 mg kg $^{-1}$ in the FERT-200%, and 50 mg kg $^{-1}$ in the BROAD-200% treatment at the 0–15 cm soil depth. The $\mathrm{NO_3}^-$ levels showed relatively little to no variation across different soil depths (Figures 3 and 4 and Supplementary Figure S2).

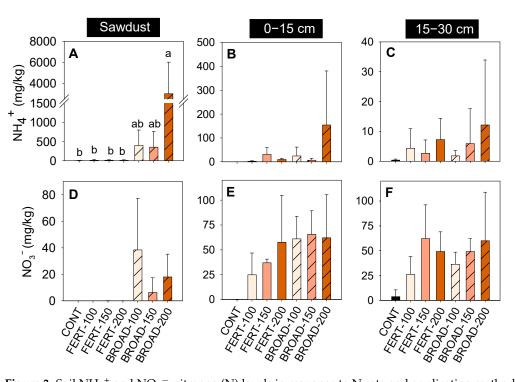


Figure 3. Soil NH₄⁺ and NO₃⁻ nitrogen (N) levels in response to N rate and application method in 2020. NH₄⁺ levels (**A–C**) and NO₃⁻ levels (**D–F**) were measured in the sawdust mulch (**A,D**) and at 0–15 cm (**B,E**), and 15–30 cm (**C,F**) soil depths in September 2020. Blueberry plants were treated with N at 100%, 150%, or 200% of the rates recommended by the British Columbia Blueberry Production Guide, applied as fertigation (FERT) or broadcast (BROAD). Data are presented as means (dry-weight basis) \pm SD (n = 4). Different letters indicate significant differences; graphs without letters found no significant differences (one-way ANOVA followed by Holm–Sidak post hoc test; $p \le 0.05$).

Soil pH was significantly affected by the N application rate, particularly in the 0–15 cm layer, with similar trends also observed in deeper soils (Supplementary Table S3). All N treatments resulted in lower soil pH than the controls at the 0–15 cm depth. The controls had an average pH of 4.9 to 5.3 at the 0–15 cm depth. The 100% N rates maintained soil pH at or near 4.2, the lower limit recommended for blueberries (Figure 5A,C). However, higher N rates, particularly the 200% rate, led to a pH below 4.2. The impact of the application method on soil pH was most noticeable at the 15–30 cm and 30–60 cm depths (Supplementary Table S3). Only the FERT treatments had lower pH than the control at 15–30 cm soil depth, with similar trends also at the 30–60 cm depth, indicating a faster pH decline with FERT than BROAD treatments. However, pH at these depths generally remained within

the recommended range, except in the FERT-200% treatment at the 15–30 cm depth, where the pH dropped to 4.2 or below (Figure 5A,C).

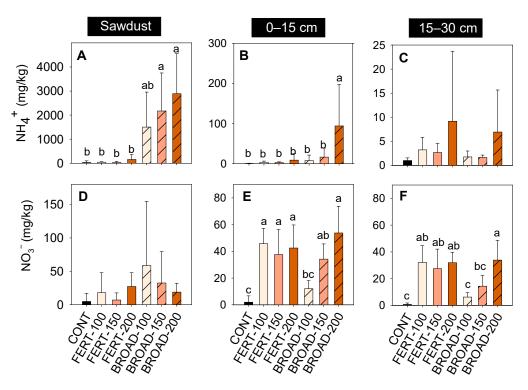


Figure 4. Soil NH₄⁺ and NO₃⁻ nitrogen (N) levels in response to N rate and application method in 2021. NH₄⁺ levels (**A–C**) and NO₃⁻ levels (**D–F**) were measured in the sawdust mulch (**A,D**) and at the 0–15 cm (**B,E**), and 15–30 cm (**C,F**) soil depths in November 2021. Blueberry plants were treated with N at 100%, 150%, or 200% of the rates recommended by the British Columbia Blueberry Production Guide, applied as fertigation (FERT) or broadcast (BROAD). Data are presented as means (dry-weight basis) \pm SD (n = 6). Different letters indicate significant differences; graphs without letters found no significant differences (one-way ANOVA followed by Holm–Sidak post hoc test; $p \le 0.05$).

The soil EC values did not show a consistent response to the N application rate or method (Supplementary Table S3). However, compared to the control, multiple treatments resulted in higher EC values (Figure 5B,D). For example, the BROAD-200% treatment had a significant impact in both years, with EC values at least four times higher than those in the control in the 0–15 cm layer. The effect was evident even at the 30–60 cm depth, where the FERT-150% and 200% treatments and the BROAD-200% treatment showed increased EC compared to the control in both years.

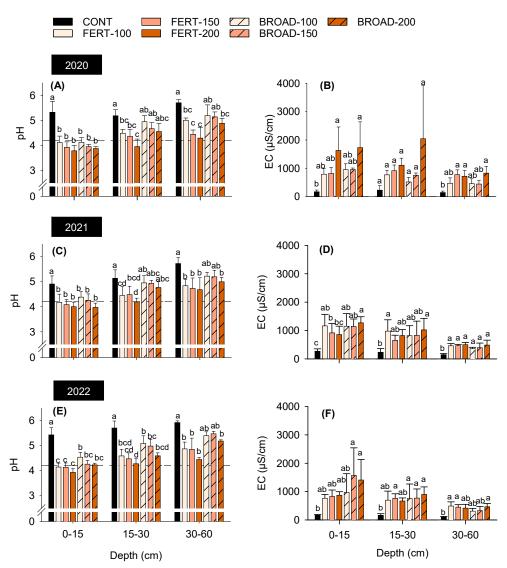


Figure 5. Effects of nitrogen (N) fertilization on soil pH and electrical conductivity (EC). Soil pH (**A**,**C**,**E**) and EC (**B**,**D**,**F**) were assessed at three different soil depths in 2020 (**A**,**B**), 2021 (**C**,**D**) and 2022 (**E**,**F**). Plants were treated with N at 100%, 150%, or 200% of the rates recommended by the British Columbia Blueberry Production Guide, applied either as fertigation (FERT) or broadcast (BROAD) until 2021. In 2022, all plants received N at 100% as BROAD treatments; however, data are presented according to historical N treatments. Data are means \pm SD (n = 4, 6, and 5 in 2020, 2021, and 2022, respectively). Different letters indicate significant differences (one-way ANOVA followed by Holm–Sidak post hoc test; $p \le 0.05$).

3.3. Plant Nutrient Accumulation

To assess the influence of the treatments and the resulting changes in soil pH and EC on plant nutrient uptake, macro- and microelements in blueberry leaves were analyzed in 2021. Leaf N levels were influenced by both the N rate and application method (Supplementary Table S4). All N treatments resulted in higher leaf N levels than the control (Table 1), which had N levels below the recommended range of 1.4% to 2.2% [9]. Among the N-treated plants, the BROAD-200% treatment resulted in higher leaf N levels than the 100% and 150% FERT treatments (Table 1).

Table 1. Leaf nutrient levels in blueberries in response to long-term nitrogen treatments analyzed in 2021. Plants were treated with N at 100%, 150%, or 200% of the rates recommended by the British Columbia Blueberry Production Guide, applied either as fertigation (FERT) or broadcast (BROAD) until 2021.

Nutrient ‡	CONTROL	FERT-100	FERT-150	FERT-200	BROAD-100	BROAD-150	BROAD-200
N	1.23 \pm 0.04 c†	$1.46\pm0.06^{\text{ b}}$	$1.48\pm0.05^{\ b}$	$1.53\pm0.08~^{ab}$	$1.53\pm0.07~^{ab}$	$1.53\pm0.07^{~ab}$	1.60 ± 0.05 a
P	770 ± 54	873 ± 85	803 ± 71	839 ± 65	852 ± 66	813 ± 97	880 ± 101
K	3061 ± 140	3262 ± 465	$\textbf{3338} \pm \textbf{250}$	$\textbf{3288} \pm \textbf{168}$	3296 ± 167	$\textbf{3018} \pm \textbf{280}$	3176 ± 192
Mg	$2278\pm319~^{a}$	$2077\pm350~\text{ab}$	$1779\pm140~^{\mathrm{bc}}$	$1803\pm276^{\ bc}$	$1733\pm165^{\ bc}$	$1562\pm113~^{\rm c}$	$1598\pm104^{\text{ c}}$
Ca	$10,085 \pm 1708$ a	$8670\pm768~\mathrm{ab}$	$7146\pm813~\mathrm{bc}$	$6842\pm1203~^{\mathrm{bc}}$	$7738\pm2260~^{abc}$	6411 ± 968 bc	$6067 \pm 912^{\text{ c}}$
S	1331 ± 205	1596 ± 353	1426 ± 303	1449 ± 355	1541 ± 216	1407 ± 101	1477 ± 200
Al	137 ± 24	157 ± 51	155 ± 37	160 ± 51	134 ± 19	140 ± 17	135 ± 7
Fe	110 ± 18	132 ± 44	157 ± 68	142 ± 47	121 ± 18	130 ± 19	125 ± 15
Mn	180 ± 49	212 ± 73	220 ± 54	236 ± 119	181 ± 49	161 ± 22	160 ± 26
Cu	3.4 ± 0.3 a	3.2 ± 0.5 ab	3.0 ± 0.4 $^{ m abc}$	2.5 ± 0.3 c	$3.0\pm0.4~\mathrm{abc}$	3.0 ± 0.4 $^{ m abc}$	2.7 ± 0.3 bc
Zn	11.6 ± 2.1	10.7 ± 1.6	9.4 ± 0.9	9.3 ± 1.7	10.9 ± 1.4	11.8 ± 1.5	10.7 ± 0.4

[‡] All nutrient levels are presented as mg kg⁻¹, except N levels, which are presented as a percentage. N = nitrogen, P = phosphorus, K = potassium, Mg = magnesium, Ca = calcium, S = sulfur, Al = aluminum, Fe = iron, Mn = manganese, Cu = copper, Zn = zinc. [‡] Data are means \pm SD (n = 6). Different letters denote significant differences (one-way ANOVA followed by Holm–Sidak post hoc test; $p \le 0.05$). Letters in bold represent values below the lower limits of sufficient levels (N: 1.4%, P: 800 mg/kg, K: 4000 mg/kg, Mg: 1000 mg/kg, Ca: 4000 mg/kg, S: 1000 mg/kg, Al: not given, Fe: 45 mg/kg, Mn: not given, Cu: 3 mg/kg, Zn: 8 mg/kg) as per the British Columbia Blueberry Production Guide [9]. The underlined values are above the recommended levels (>8000 mg/kg for Ca).

Potassium (K) and copper (Cu) were the only other nutrients that displayed deficiencies (Table 1). The K levels were up to 25% lower than the recommended lower limits across all treatments. Cu deficiencies were observed in response to high N fertilization, with a 10% and 17% decline from the recommended levels in the FERT and BROAD treatments, respectively, at the 200% rate. Although still within the recommended range, the fertilization also impacted several other nutrient levels. Manganese (Mn) and zinc (Zn) levels were influenced by the N application method, calcium (Ca) by the N rate, and magnesium (Mg) by both the N rate and application method. Compared with the control, the high N treatments resulted in lower Mg, Ca, and Cu levels, irrespective of the application method. None of the treatments showed any significant effect on phosphorus (P), K, iron (Fe), or sulfur (S) levels (Supplementary Table S4).

3.4. Influence of Historical Treatments on Plant, Fruit, and Soil

To differentiate the impacts of long-term fertilization on soil conditions from those caused by N rates in the treatment year, we applied 100% N to all treatments, including the controls, as a BROAD application in 2022, assuming that differences in soil conditions resulting from long-term treatments would persist. As expected, when assessed based on the historical treatments, soil pH remained lower in soils that had been treated with N fertilizer compared to the control (Figure 5E). The average soil pH in the 0–15 cm soil layer was 5.4 in the historical control, 4.2 in the BROAD-200%, and 3.9 in the FERT-200% treatments. In addition, the average pH values in all the historical FERT treatments were slightly below the recommended low pH of 4.2, while they were at 4.2 or higher in the BROAD treatments. Although the influence of historical N treatments on pH values was evident even below the 0–15 cm depth, they were within the acceptable range, except in the controls, where the average pH was 5.7 and 5.9 at the 15–30 cm and 30–60 cm depths, respectively.

The soil EC generally remained high for the historical treatments that received high N rates, particularly those applied as BROAD (Figure 5F). The EC values in the historical BROAD-200% treatments were significantly higher than the controls across all soil depths

assessed, with approximately 9-fold and 5.5-fold increases in EC at the 0–15 cm and 15–30 cm soil depths, respectively.

The $\mathrm{NH_4^+}$ levels also showed persistent effects, with all historical BROAD treatments showing high $\mathrm{NH_4^+}$ levels compared to the historical control. In contrast, $\mathrm{NH_4^+}$ levels in the historical FERT treatments did not differ from those of the control (Figure 6). The prominently high $\mathrm{NH_4^+}$ levels observed in the sawdust mulch layer in response to the BROAD-200% treatments during earlier years declined to an average of about 740 mg kg $^{-1}$, representing nearly a fourfold reduction from the 2021 levels. Despite this decline, $\mathrm{NH_4^+}$ levels in the BROAD-200% treatments remained higher compared to both the historical control and the FERT treatments. No clear trends related to historical treatments were observed in the $\mathrm{NO_3^-}$ levels.

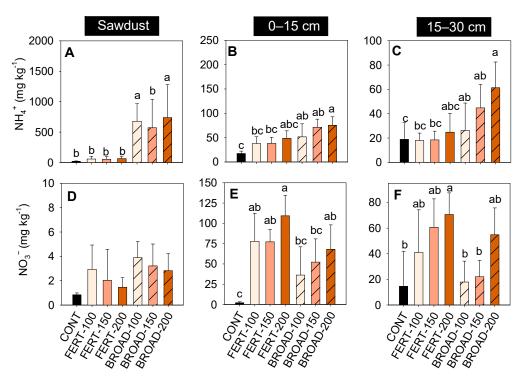


Figure 6. Soil NH₄⁺ and NO₃⁻ nitrogen (N) levels in response to historical N application rates and methods. NH₄⁺ levels (**A–C**) and NO₃⁻ levels (**D–F**) were measured in the sawdust mulch (**A,D**) and at the 0–15 cm (**B,E**) and 15–30 cm (**C,F**) soil depths. Treatments included N fertilizer applied at 100%, 150%, or 200% of the rates recommended by the British Columbia Blueberry Production Guide, using either fertigation (FERT) or broadcast (BROAD) methods until 2021. In 2022, all plants received N at 100% using BROAD applications, but data are presented based on historical N treatments. Values are expressed as means (dry-weight basis) \pm SD (n = 5). Different letters indicate significant differences; graphs without letters found no significant differences (one-way ANOVA followed by Holm–Sidak post hoc test; $p \leq 0.05$).

The average fruit yield was relatively similar across all treatments, showing no significant impact of historical treatments (Figure 1C). The fruit TSS level, which was not affected by different N treatments, again showed no influence due to historical treatments (Figure 1F). On the other hand, the differences in fruit TA caused by N treatments in the earlier years disappeared in response to equal N fertilization (Figure 1I).

The application of N fertilizer in 2022 was sufficient to eliminate the N deficiency previously observed in control plants, but levels remained lower than in plants that regularly received N (Table 2). Conversely, the reduced Cu levels in high N-treated plants in 2021 continued into 2022. All historical N treatments resulted in lower Cu levels than the control, with the 200% BROAD and FERT treatments continuing to show deficiency levels. As in 2021, Mg and Ca levels were also lower in plants that historically received N compared to

the control (Table 2). Additionally, leaf aluminum (Al) and S levels were influenced by the historical N rate and application method, respectively (Supplementary Table S4). S levels were generally higher in all BROAD treatments than in controls and the FERT treatments. Leaf P levels were elevated in plants that historically received high N rates compared to the controls (Table 2). However, these effects were not consistent with the previous year observations.

Table 2. Leaf nutrient levels in blueberries in response to long-term nitrogen treatments analyzed in 2022. Plants were treated with N at 100%, 150%, or 200% of the rates recommended by the British Columbia Blueberry Production Guide, applied either as fertigation (FERT) or broadcast (BROAD) until 2021. In 2022, all plants received N at 100% as BROAD treatments, but data are presented according to historical N treatments.

Nutrient ‡	CONTROL	FERT-100	FERT-150	FERT-200	BROAD-100	BROAD-150	BROAD-200
N	1.47 ± 0.06 ^{b†}	1.72 ± 0.07 a	1.74 ± 0.06 a	1.77 ± 0.04 a	1.76 ± 0.05 a	1.75 ± 0.06 a	1.81 ± 0.05 a
P	$896\pm52^{ m \ b}$	991 \pm 77 $^{ m ab}$	$1005\pm46~\mathrm{ab}$	$1018\pm62~^{\rm a}$	$983 \pm 45~\mathrm{ab}$	$1038\pm63~^{a}$	$1023\pm71~^{\mathrm{a}}$
K	3322 ± 346	3449 ± 286	3483 ± 146	3469 ± 495	3378 ± 355	3556 ± 503	3403 ± 290
Mg	1797 \pm 178 $^{\rm a}$	$1553\pm142~^{ab}$	$1458\pm189^{\text{ b}}$	$1494\pm146^{\text{ b}}$	1535 \pm 72 $^{\mathrm{b}}$	$1495\pm131^{\ \mathrm{b}}$	$1447\pm89^{\text{ b}}$
Ca	$5972 \pm 571~^{\rm a}$	4340 ± 425 ^b	$4202\pm707^{\:b}$	$4028\pm445^{\text{ b}}$	4583 \pm 341 $^{\mathrm{b}}$	$4288\pm316^{\;b}$	$4023\pm458^{\:b}$
S	$1047\pm65~^{\rm c}$	1157 ± 34 bc	$1127\pm62~^{\rm c}$	1167 ± 51 bc	1302 \pm 74 $^{\rm a}$	$1259\pm116~^{\rm ab}$	1324 \pm 77 $^{\rm a}$
Al	99 ± 19	103 ± 6	110 ± 20	119 ± 7	110 ± 11	107 ± 12	117 ± 10
Fe	99 ± 15	104 ± 9	112 ± 20	114 ± 15	112 ± 20	113 ± 21	112 ± 11
Mn	77 ± 18	95 ± 23	87 ± 31	94 ± 16	90 ± 14	83 ± 15	85 ± 15
Cu	4.2 ± 0.5 a	3.4 ± 0.3 b	3.0 ± 0.4 bc	$2.7\pm0.3~^{\rm c}$	3.2 ± 0.2 bc	3.2 ± 0.4 bc	2.8 ± 0.4 bc
Zn	11.0 ± 2.5	11.1 ± 2.9	9.8 ± 0.7	11.8 ± 4.2	10.4 ± 0.9	11.7 ± 1.4	10.7 ± 0.6

[‡] All nutrient levels are presented as mg kg⁻¹, except N levels, which are presented as a percentage. N = nitrogen, P = phosphorus, K = potassium, Mg = magnesium, Ca = calcium, S = sulfur, Al = aluminum, Fe = iron, Mn = manganese, Cu = copper, Zn = zinc. [†] Data are means \pm SD (n = 6). Different letters denote significant differences (one-way ANOVA followed by Holm–Sidak post hoc test; $p \le 0.05$). Letters in bold represent values below the lower limits of sufficient levels (N: 1.4%, P: 800 mg/kg, K: 4000 mg/kg, Mg: 1000 mg/kg, Ca: 4000 mg/kg, S: 1000 mg/kg, Al: not given, Fe: 45 mg/kg, Mn: not given, Cu: 3 mg/kg, Zn: 8 mg/kg) as per the British Columbia Blueberry Production Guide [9].

4. Discussion

4.1. High Nitrogen Rates over the Long Term Do Not Improve Fruit Yield

Berry yield data from 2020 and 2021 indicate that applying N fertilizer at rates higher than recommended does not enhance fruit production and may, in fact, negatively impact yield. This negative trend is evident from the lower berry yield observed in the FERT-200% treatment compared to the BROAD-100% treatment in 2020. Historical yield data from the same plots further support the adverse effects of high N rates on fruit yield. For instance, in 2016, the FERT-200% treatment did not result in a significant change in fruit yield compared to the control, while lower N rates led to increased yields. Average yields in 2017 and 2018 also showed negative trends in response to increasing N rates [23]. Despite mostly being short-term evaluations, no or negative influence of increasing N rates on blueberry yield has also been reported in previous studies [16,19,20,31-33]. The lack of beneficial response to increasing N levels may be attributed to N no longer being a limiting factor for reproductive development, with other factors potentially becoming limiting. For example, limitations in photosynthetic capacity can restrict fruit production in blueberries, as fruit growth primarily relies on immediately assimilated carbon rather than on carbohydrate reserves [34]. Conversely, plant death or injuries caused by higher N rates have been suggested as contributing factors to the reduced yield observed in some studies [31]. Excess N availability late in the growing season can trigger late growth flushes, increasing plant susceptibility to cold injuries [27,31]. Additionally, the differences observed in flower bud development (Figure 2) suggest

that variations in resource allocation between reproductive and vegetative growth in response to N availability may also contribute to these effects.

Although high N rates are not generally beneficial, there may be opportunities to improve current N recommendations for British Columbia to achieve maximum yields. For example, in 'Duke', applying higher N rates (150% of the recommended rates for British Columbia) as FERT treatments generally resulted in better yields than recommended rates and BROAD applications in establishing fields [13,22]. This increased N requirement in FERT treatments is likely due to the release of fertilizer in areas where roots have not yet been fully established, unlike in BROAD treatments, where fertilizer is applied around the base of the plants [8,17]. Therefore, slightly increasing the recommended rates when using FERT treatments could benefit growers during the early years of field establishment. In contrast, the lack of significant yield differences between controls that received no N and those that received 100% of the recommended N rate in mature plants in the present study (Figure 1A,B), along with the negative trends observed in response to high N rates, suggests that there may be potential to further reduce N rates without negatively impacting crop yield in mature plants, irrespective of the application method. Consistent with this possibility, a three-year study in a mature 'Duke' planting found no yield differences when N fertilizer was applied at rates of 60 and 120 kg N ha^{-1} [35].

In addition to assessing lower annual N fertilizer rates over the long term, evaluating alternate-year N treatments or alternating between relatively lower and higher rates would be valuable. While young plantings typically show an increase in yield during their establishment period [13,22], mature blueberries do not necessarily produce stable yearly yields. Instead, yields can fluctuate between relatively high and low production years. For example, a 10-year study on 'Bluecrop' and 'Duke' found that yields in 'Bluecrop' fluctuated without a specific trend, whereas in 'Duke', a peak year was followed by medium- and low-yield years [36]. Although our historical data showed no specific pattern [22,23]; the three-year period presented here shows a similar trend following a peak production in 2020. However, we cannot rule out that the lower yield in 2021 could be due to the extreme heat event that affected this growing region, leading to a significant fruit loss [6,37]. Apart from yield fluctuations, an earlier study reported a trend where leaf N levels were lower in high-producing years compared to low-producing years [32]. These fluctuations in N levels and variations in berry yield suggest that maintaining the same annual N rates for established plants may not be necessary. Instead, there may be opportunities to adjust N supply annually based on the previous year yield.

4.2. Flower Bud Set and Fruit Qualities Are Influenced by the Nitrogen Fertilizer Rate

The lower flower bud numbers and the higher leaf bud numbers in the FERT-200% treatment, compared to the BROAD-100% treatment, indicate that the yield reductions observed with high N rates are at least partly due to the increased development of leaf buds causing fewer flower buds (Figure 2). Studies in both annual and perennial plants show N availability or C to N ratio as a contributing factor in regulating flowering initiation [38,39]. Nitrogen levels, in particular, influence the activity of transcription factors that control flowering regulatory genes [40]. While the exact mechanisms are not clear, a lower carbohydrate-to-N ratio is also believed to restrict flower bud development in blueberries [24], likely leading to observed differences in flower bud numbers. Nonetheless, the lack of difference in the number of flowers produced per inflorescence and the uniformity of fruit set percentage across different N rates within the FERT treatment suggest that the bud differentiation and fruit set may not significantly influence yield.

The differences observed in flower and leaf bud numbers in 2021, however, did not translate into yield differences. Moreover, if the number of flower buds was the primary factor driving yield differences in response to N rates, we would expect to see yield differences in 2022 as well, despite the equal N rates, because blueberry flower buds develop at the end of the previous growing season [41]. The absence of clear yield differences in 2021 and 2022 suggests that other factors, such as the weight of individual

fruits, are also influencing yield. A higher crop load in blueberries tends to result in smaller berries [42]. This inverse relationship likely compensates for the lower percentage of flower buds and may account for the absence of significant yield differences in those years.

Compared to controls, N treatments reduced fruit TA in 2020 and 2021, with higher N rates generally having the greatest effect. This reduction in TA in response to high N levels aligns with previous studies on the highbush blueberry cv. 'Wolcott' [43,44], as well as findings in other fruit crops, such as apple [45] and citrus [46]. However, short-term N treatments in establishing plants did not influence TA in the cv. 'Duke' [13]. Nonetheless, when equal fertilizer rates were applied in 2022, no difference in TSS and TA was observed, suggesting that the reduction in TA is likely a direct response to N availability rather than to changes in soil properties caused by long-term treatments. The N-dependent regulation of fruit TA has not been extensively studied. However, possible explanations include variations in the allocation of photoassimilates to fruit versus vegetative growth based on N availability. Additionally, fruit temperature and transpiration could vary due to shading from vegetative growth. These differences could, in turn, influence organic acid metabolism and thereby affect fruit TA [47]. The lack of N-dependent TA difference reported in the establishing plants [13] could be due to a less pronounced change in TA in young plants.

4.3. Fertilizer Distribution and Plant Availability Are Affected by the Application Method

The N fertilizer applied in the $\mathrm{NH_4}^+$ form is converted to $\mathrm{NO_3}^-$ through nitrification by soil microbes. Although the inhibited activity of nitrifying microbes in the acidic soils where blueberries grow slows nitrification, once $\mathrm{NH_4}^+$ is converted to $\mathrm{NO_3}^-$, it becomes less preferable for blueberries and more susceptible to leaching due to its high mobility [2,23]. Unlike in the FERT treatment, where $\mathrm{NH_4}^+$ tends to move more effectively through the sawdust mulch, a significant portion of N fertilizer applied as BROAD treatments likely remains in the sawdust mulch. This observation aligns with previous findings that that more $\mathrm{NH_4}^+$ is retained in the mulch layer under BROAD treatments, whereas more $\mathrm{NO_3}^-$ is leached when fertilizer is applied as a FERT treatment [23].

BROAD-applied fertilizer directly under the drip lines is more likely to dissolve in the irrigation water and be transported to the root zones, while at least a portion of the rest remains in the sawdust mulch (Figure 7). During the rainy season, which usually lasts from October to April in this region (Supplementary Table S1), the fertilizer in the mulch may gradually dissolve and become available to the plants over time. The high NH₄⁺ levels observed in the BROAD-200% treatment in the 0–15 cm layer in November 2021 (Figure 4B), the increased N accumulation in the leaves under the BROAD-200% treatment (Table 1), and the precipitation-dependent accumulation of N in the leachate in response to BROAD treatments observed earlier [23] support this possibility. While plants are unlikely to take up N during dormancy, they may take up a portion of this leftover N before and after winter dormancy. However, our observations align with previous studies indicating that high NH₄⁺ levels left in the mulch layer do not enhance fruit yield once the N supply exceeds the recommended levels [22,35]. Furthermore, when N levels in the leachate were assessed, NO₃⁻ and NH₄⁺ concentrations were comparatively lower in the BROAD treatment than in the FERT treatment [23]. This observation, together with the four-fold decline in NH_4^+ levels in the sawdust mulch layer from 2021 to 2022, suggests that not all residual N in the sawdust mulch may become available in the root zone or be leached away. Instead, a significant portion of N applied through the BROAD method may be lost due to volatilization.

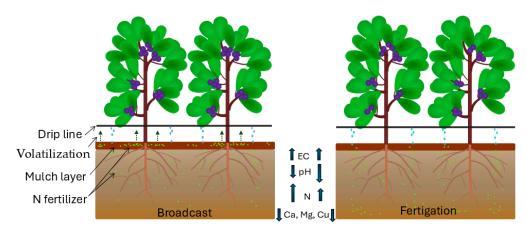


Figure 7. Impact of long-term nitrogen (N) fertilization and application methods. N fertilizer applied as broadcast (BROAD) treatments initially remains in the sawdust after application, gradually becoming available in the root zone as it dissolves with irrigation water. Fertilizer not directly under drip irrigation tends to dissolve more slowly, with some loss due to volatilization. In contrast, fertigation (FERT)-applied N passes through the mulch layer more readily and becomes immediately available to plants, though a higher portion may be lost through leaching compared to BROAD treatments. All N treatments increase electrical conductivity (EC) and reduce pH, but the pH decline occurs more rapidly with FERT treatments (illustrated by arrows, with arrow heights indicating the extent of change). These soil changes also reduce the availability of calcium (Ca), magnesium (Mg), and copper (Cu) to plants, while N availability appears slightly higher in BROAD treatments due to differences in the rate and pattern of fertilizer distribution.

4.4. Long-Term Fertilization May Result in Soil pH and Electrical Conductivity Levels Undesirable for Blueberries

The soil EC, which generally indicates soil salinity but also serves as an indicator of soluble nutrients, is a major determinant of the physical, chemical, and biological properties of soil. The desired soil EC is usually crop-dependent, and exceeding threshold levels often leads to reduced yield potential [48]. An earlier study on highbush blueberries recommended maintaining soil EC values below 2000 μS cm $^{-1}$ for optimal plant growth [49]. Although none of the treatments resulted in EC values exceeding this limit, our previous work has shown negative correlations between blueberry yield and even lower EC values, which stayed below 1000 μS cm $^{-1}$ at the 0–30 and 30–60 cm soil depths when analyzed at 5, 6, and 10 years after the treatment initiation [22,23]. Therefore, while the different N rates applied as either BROAD or FERT treatments over thirteen years do not appear to have had a detrimental effect on soil EC leading to an obvious impact on plant growth performance, increasing EC values could be a contributing factor to the lower yield potentials observed with high N rates.

Soil pH is another crucial parameter that influences the availability and toxicity of various elements to both plants and soil microorganisms. Most plant nutrients are optimally accessible to plants when the soil pH is between 6.0 and 7.5 [48]. While blueberries have evolved to grow in the 4.2 to 5.5 pH range with low nutrient requirements, excessively low pH can reduce plant growth. In such conditions, toxic ions such as Al^{3+} become available to plants, which can have deleterious effects on plant development and yield [2,50]. Therefore, it is important to pay attention to soil pH when N fertilizer is applied over the long term. Ammonium-based fertilizers such as ammonium sulfate used in this study are associated with a decline in soil pH due to the release of H^+ ions when plants take up NH_4^+ , and when NH_4^+ is nitrified to NO_3^- [2].

The soil pH in the study field, initially set to 5.0 by the application of elemental sulfur in 2008, decreased to an average of 4.0 at the 0–15 cm depth by 2021 in response to both FERT-200% and BROAD-200% treatments, while the controls remained at 4.9. Even at the 100% rate, the pH fell to 4.2 and 4.4 in the FERT and BROAD treatments, respectively. At

the 15–30 cm depth, the pH was 4.2 and 4.8 for the FERT-200% and BROAD-200% rates, respectively, while it was 5.1, 4.5, and 5.0 for the control, FERT-100%, and BROAD-100% treatments, respectively (Figure 5). These results indicate a general decline in soil pH in response to all N treatments and application methods, with a more pronounced decrease at higher rates applied through fertigation (Figure 7).

These changes in soil pH are consistent with trends observed in earlier years, which showed a faster decline in pH in response to FERT treatments, evident as early as 2013, just five years after the treatments began. At that time, the FERT-200% treatment resulted in a pH of 4.34, while BROAD treatments maintained an average pH of 4.8 across different rates when analyzed at the 0–30 cm soil depth [22]. A soil analysis conducted in 2018, ten years after treatment initiation, continued to show this trend [23]. Although a direct comparison is not possible due to differences in the depth of soil layers analyzed, the data suggest a gradual but consistently faster pH decline in response to FERT treatments throughout the evaluation period. Similarly, another study reported lower soil pH in response to ammonium sulfate applied as a FERT treatment compared to BROAD treatments within five years [16]. Altogether, these observations show that long-term FERT treatments, especially when applied as ammonium sulfate, require close monitoring of soil pH and implementing pH correction methods. Alternatively, urea can help slow soil acidification, as it releases only half the amount of H⁺ compared to ammonium sulfate and is less prone to leaching [16].

4.5. Long-Term Nitrogen Fertilization Influences Plant Nutrient Accumulation

The leaf N level of 1.2% in the controls was below the recommended minimum of 1.4% when assessed in 2021. The application of N at the 100% rate in 2022 was sufficient to correct this deficiency (Table 1). However, the comparatively lower N levels in these historical controls, relative to plants that consistently received N, suggest that the controls likely did not receive adequate N or sufficient time to achieve equivalent N levels within a single growing season. While not applying N led to a N deficiency, none of the long-term high N rates exceeded the recommended maximum leaf N levels of 2.2% [9].

When leaf N levels in 2021 were compared across different N fertilizer rates, the BROAD-200% treatment did not differ significantly from the lower BROAD rates. Similarly, the FERT-200% treatment did not differ from the lower FERT rates. However, the BROAD-200% rate resulted in higher leaf N levels than the FERT-100% and FERT-150% treatments (Table 1). As discussed earlier, this finding supports the observation that plants tend to accumulate more N with BROAD treatments compared to FERT treatments. The extended duration of N availability in the root zone, resulting from the gradual movement of BROAD-applied N from the sawdust mulch layer, may enhance N uptake (Figure 7). While we cannot exclude the possibility that FERT-applied N may be lost when not released near the root zone, the placement of drip lines near the center of the rows and the broad expansion of the root system in mature plants reduces the likelihood of such losses.

Blueberries are efficient at taking up Ca²⁺ and have low Ca requirements. However, they lack mechanisms to regulate Ca²⁺ influx, which can lead to high accumulation at high Ca²⁺ levels [2]. Lower plant Ca levels in response to high N fertilizer have been previously reported [51]. The solubility of certain metal ions increases at lower pH, which can cause plant-available nutrients like Ca, Mg, and P to decline due to leaching or precipitation as metallic salts [52]. Therefore, the high Ca levels observed in the controls, which exceeded the recommended levels in 2021, and the comparatively lower Ca levels in all treatments receiving N above 100% are likely attributed to the pH differences caused by the N fertilizer treatments. However, even the pH levels below the recommended range observed in response to the 200% treatments did not result in Ca levels falling below the recommended levels.

Similarly to the comparatively lower levels of Ca, the leaf Mg levels were also reduced in plants that received N compared to the controls, but did not reach deficiency levels. This decline is also likely due to the pH decline observed in the fertilized soils compared to the

controls, as acidic conditions increase Mg leaching and impair uptake [53]. Additionally, the reduced pH may explain the lower Cu levels observed in response to N treatments, even reaching deficiency levels at the 200% rate in both FERT and BROAD treatments. Although Cu deficiencies are typically a concern when soil pH is too high and are generally not a major issue in blueberry production systems, highly acidic soil can limit plant-available Cu [2], potentially leading to the observed reductions. On the other hand, the lower K levels are likely independent of the long-term treatments, as no differences in K levels were observed between fertilized plants and the controls. Although leaf K levels were below the recommended range, we did not observe symptoms typically associated with K deficiency, such as chlorosis at the leaf margins of older leaves, scorching along the margins, curling, or necrotic spots [2].

5. Conclusions

Our findings suggest that applying N fertilizer at rates exceeding the recommendations in British Columbia offers no additional benefits to growers and may even lead to adverse effects over the long term. The impact of N rates is often more pronounced when considered with the application method. Long-term FERT treatments tend to decrease soil pH faster, while BROAD treatments lead to greater N accumulation, both of which likely contribute to observed differences. Additionally, soil EC increases with prolonged N treatments regardless of the application method, potentially contributing to a reduction in the effectiveness of N treatments over time. These changes in soil conditions can decrease the availability of certain nutrients, leading to deficiencies. While increased N fertilization rates can reduce fruit TA, this is likely independent of changes in soil pH and EC.

Excessive long-term application of N, especially through FERT treatments, can decrease flower bud formation and increase leaf bud formation, and may lead to reduced fruit yield. While BROAD treatment at the currently recommended rate for mature plants is likely to produce the best fruit yield, long-term fertilization at those rates may not consistently benefit crop production, possibly due to the adverse effects of prolonged fertilization on soil conditions. Overall, while the current regional recommendations for N fertilizer for highbush blueberries are adequate for the cv. 'Duke', there are likely opportunities to further reduce N rates for mature plantings without negatively impacting fruit yield.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/horticulturae10111205/s1, Figure S1: Study Design; Figure S2: Soil NH₄⁺ and NO₃⁻ nitrogen (N) levels in response to N rate and application method at the 30–60 cm soil depth; Table S1: Monthly temperature and total precipitation at the experimental location; Table S2: Results of ANOVA showing the impact of nitrogen fertilizer application rates and application methods on fruit yield, titratable acidity (TA), total soluble solids (TSS), and the leaf and flower bud counts; Table S3: Results of ANOVA showing the impact of nitrogen fertilizer application rates and application methods on soil NO3– and NH4+ levels, soil pH, and electrical conductivity (EC) in the sawdust mulch layer and at the 0–15 cm, 15–30 cm, and 30–60 cm soil depths; Table S4: Results of ANOVA showing the impact of nitrogen fertilizer application rates and application methods on leaf nutrient levels.

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Data Availability Statement: The data presented in the study are included in the article/Supplementary Materials. Further inquiries can be directed to the corresponding author.

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