



Article

Growth and Yield of Strawberry Cultivars under Low Nitrogen Supply in Italy

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Abstract: Nitrogen plays a vital role in plants' biochemical and physiological functions, and it contributes significantly to increasing plant yield and fruit quality. Plants that efficiently absorb and utilize nitrogen enhance the efficiency of fertilizers, reducing their input costs and preventing ecosystem damage. Thus, an adequate nitrogen supply can significantly improve plant growth, fruit quality, and nutritional value. This research focused on evaluating the plant vegetative and productive performance and fruit quality of three short-day strawberry genotypes ("Cristina", "Romina", and "Sibilla") that were fertilized with different amounts of nitrogen, in a crop that was protected under a plastic tunnel. The trial was conducted during two cultivation cycles. The nitrogen rates were 113, 90, and 68 kg/ha for the first year, and 118, 97, and 76 kg/ha for the second. Reduced nitrogen inputs did not significantly affect plant height, indicating that decreased nutritional intake does not harm plant development. The fruit sugar content value remained stable across all nitrogen supplies, as did the fruit titratable acidity. The cultivars maintained a medium fruit firmness at a 60% nitrogen supply, and the Chroma index was not affected. This study found that reducing nitrogen inputs did not have a significant negative impact on the three tested cultivars, making them suitable for cultivation with reduced nitrogen inputs to reduce the environmental impact and save growers' inputs.

Keywords: single cropping; fertilizer; fruit quality; plant performance



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1. Introduction

The vegetative, productive, and qualitative parameters of strawberry plants are influenced by their nutritional status [1,2]. Adequate levels of nitrogen (N), phosphorus (P), and potassium (K) are essential for proper plant growth and development [3,4]. The amount of fertilizer applied for strawberry cultivation in Italy is mainly based on farmers' experience and sensitivity, and there is often an abuse of some nutrients. Plants that are efficient in absorbing and utilizing nutrients greatly enhance the efficiency of applied fertilizers, reducing input costs and preventing losses of nutrients to the ecosystems [5,6]. Therefore, studies on the vegetative, productive, and qualitative plant responses to different nutrient inputs are necessary to achieve the correct supply of nutrients. It is important to define the amount of nutrients to obtain the maximum performance of the plant, along with a low environmental impact. Nitrogen (N) is an essential nutrient that plays a vital role in the biochemical and physiological functions of plants, increasing plant yield and fruit quality [7–10]. The plant genotype, phenological stages, harvesting season, and administered doses determine the amount of nitrogen required by the plant during its cultivation cycle [8,11]. Agehara and

Nunes showed that nitrogen fertilization increased the total production and earliness in strawberries grown under tropical conditions, thus avoiding the application of pre-planting nitrogen [12]. Furthermore, Cvelbar Weber et al. pointed out that different amounts of nitrogen swayed strawberry plant growth, yield, and fruit quality [13]. These studies confirmed the role of appropriate rates of nitrogen on strawberry plant yield and quality, depending on the cultivation conditions. Previous studies [14–17] explained that nitrogen fertilization influences floral initiation and the number of shoots and inflorescences of strawberry plants. According to [18], excessive nitrogen doses inhibit floral induction. On the contrary, runner production is stimulated by nitrogen availability [19]. At the same time, fruit development is accelerated, delaying ripeness, lowering yields, and increasing both fruit acidity and water content [20,21]. Strawberry plants can manifest typical symptoms of nitrogen deficiency such as undersized fruits and yellowish-green foliage with stunted growth and runner numbers [19]. Overall, strawberry cultivation systems need well-defined management of nitrogen and water application to avoid nitrogen loss in the soil and reduce water loss.

Based on these considerations, this study aimed to investigate the effect of different nitrogen regimes on the vegetative, productive, and qualitative responses of three strawberry cultivars grown with a standard early spring cultivation cycle in an open field that was protected under a plastic tunnel.

2. Materials and Methods

2.1. Field Trial

The trial for the identification of strawberry cultivars with reduced nitrogen uptake was conducted for two cycles of cultivation (2016/2017–2017/2018) on short-day cultivars. The experimental trials took place at the ASSAM (Agenzia Servizi al Settore Agroalimentare delle Marche) experimental farm in Petritoli, Marche region, (Italy) (43°04′01.56″ N; 13°41′19.22″ E). The soil was composed of 24% clay, 34% sand, and 42% silt at a pH value of 8.14.

2.2. Plant Material

The single-cropping cultivars studied during the two cultivation cycles (2016/2017 and 2017/2018) were “Romina”, “Sibilla”, and “Cristina”. The plant material comprised “cold stored plant”, category A+ [22]. In the first year, planting took place on 28 July 2016 in an open field; it was covered with a plastic tunnel on 24 February 2017, and fruits were harvested in spring 2017. The same experiment was run for another cycle (2017/2018) with similar planting (26 July 2017) and coverage (21 February 2018) dates. The plant material was provided by Coviro Soc. Cons. a.r.l. (Cervia, Italy).

Plants were placed in 3 different rows, each 54 m in length, each row corresponding to a different nitrogen supply (N100, N80, N60). The experimental design was realized according to the split-plot model, with 3 different levels of nitrogen supply (main plots) and 3 cultivars (sub-plots). Two lysimeters were installed at each of the three rows with different nitrogen treatments. Lysimeters were positioned in the soil between two plants, at different depths (15 cm and 35 cm). These depths corresponded to the area of roots exploration and below. Lysimeters were used to sample soil circulating solutions; then, these samples were analyzed through ion chromatography to detect anionic and cationic species.

The second-year experiment was established on a different plot within the same farm (rotation).

2.3. Nitrogen Fertilization Amounts

The total nitrogen application during the two years of trials (from August to June in both years) was maintained to be as homogeneous as possible (Table 1). Nutrient equilibrium was obtained with 10-52-10 (N-P-K) (Peters Professional Plant Starter, ICL Italia Treviso srl, Treviso, Italy) from August until March, and 20-20-20 (N-P-K) (Peters Professional Allrounder, ICL Italia Treviso srl, Treviso, Italy) from April to June. The total

amounts of N fertilization for the N100 trial followed the recommendation for the Marche Region (Delibera 786 of 10/07/2017), while N80 and N60 correspond to reductions of 20% and 40%, respectively. The irrigation system consisted of a dripping hose Aqua-Traxx® FlowControl™ (Toro Ag, Fiano Romano, Italy), with 20 cm spacings between drippers, 16 mm in diameter, and an individual emitter flow rate of 1.01 l/h at 0.7 bar (flow rate per meter of 5.05 l/h/m at 0.7 bar).

Table 1. Nitrogen dose was applied to each treatment (N100, N80, N60) in both strawberry growing seasons (2016–2017 and 2017–2018).

YEAR	N100 (kg/ha)	N80 (kg/ha)	N60 (kg/ha)
1°	113	90	68
2°	118	97	76

2.4. Analyzed Parameters

2.4.1. Water Analysis

The soil circulating solution available to the plants was sampled and then analyzed in terms of electrical conductivity and ion concentration.

Solution sampling. The solution samples were picked up at two different soil depths (15 cm and 35 cm), through lysimeters previously placed. The EC (electrical conductivity) of the circulating solution samples was measured using a WTW 340 conductivity meter (Xylem Water Solutions Italia S.r.l., Lainate, Italy). The conductivity is used to measure the ionic concentration and activity of the solution.

The last analysis was conducted through ion chromatography or IC, an analytical technique for selective ion separation and determination. It adopted ion exchange chromatography, with a reversible exchange between the single ions in the stationary phase and the ions with the same charge in the mobile phase [23]. The analysis of chloride, bromides, nitrate, and sulfate was realized using an ion chromatographer Dionex ICS1000 (Thermo Fisher, Waltham, MA, USA) connected to a laptop.

2.4.2. Vegetative Parameters

The vegetative parameters recorded for evaluation of the effects of the applied N treatments were n° branch crowns/plants, n° inflorescences/plant, n° of leaves/plants, and plant height. The plant height was measured with a ruler and expressed in cm. Measurements were made for 8 plants in each subplot included in the three main plots (treatment). Each subplot was replicated 3 times for each cultivar. The values obtained in two years for the single-cropping cultivars (2017–2018) were averaged.

2.4.3. Productive Parameters

The main productive parameters were evaluated for the different nitrogen regimes: commercial production and average fruit weight (AFW). The commercial production of each cultivar was expressed as average plant production (the plot production for each harvest was divided by the number of plants present in the plot). Then, these values were summarized for all harvests until the end of the season. Starting from the third harvest, twenty uniform fruits in terms of size and ripeness degree were collected from each plot for three consecutive harvests for the qualitative analyses. The methods used were described by Capocasa et al. [24].

2.4.4. Qualitative Parameters

The main qualitative parameters were evaluated for the different nitrogen regimes: sugar content, titratable acidity (TA), fruit color: L* (luminescence), a* (red tone), and b* (yellow tone), Chroma index, and firmness, in accordance with Marcellini et al. [25]. For each thesis (genotype/treatment) and at each harvest, we assessed the Chroma index and firmness of 10 selected fruits. We utilized a Minolta romameter CR 400 (Konica Minolta,

Tokyo, Japan) to evaluate the external color of the fresh fruits, measuring two points on opposite sides of each fruit and recording CIELAB values (L^* , a^* , b^*). The Chroma index was then determined based on the a and b values.

Subsequently, we used a penetrometer (Penetrometer 327, Effegì, Ravenna, Italy) with a 6 mm star probe to measure the firmness of the same fruits. Then, the samples were frozen at -18 °C until evaluations of the total soluble solids (TSS) and titratable acidity (TA) were conducted. The TSS measurements were conducted using a digital refractometer (PR-101, ATAGO, Tokyo, Japan), while the TA was determined through acid–base titration. To calculate the TA, we measured the milliequivalents (mEQ) of 0.1 N NaOH solution per 100 g of fresh weight (FW), using bromothymol blue as a pH indicator.

2.5. Statistical Analysis

The results for the strawberry fruit vegetative, productive, and qualitative parameters are presented as mean \pm standard deviation (SD) for each cultivar/nitrogen treatment. A two-way analysis of variance was used for the short-day cultivars to test for differences among the cultivation years, cultivars, fertilization amounts, and corresponding interactions. Statistically significant means differences were determined with Fisher's (least significant difference, LSD) test ($p \leq 0.05$). The statistical processing was carried out using STATISTICA software (Stasoft, Tulsa, OK, USA).

3. Results and Discussion

3.1. Water Analysis

The suitability of water for a specific purpose depends on the types and amounts of dissolved salts. Some of the dissolved salts or other constituents may be useful for crops, such as NO_3 [26]. The most important characteristics that determine the quality of irrigation water are the pH; the total concentration of soluble salts, assessed through the EC; the sodium adsorption ratio (SAR), described as the relative proportion of Na to other cations such as Ca and Mg; the concentration of B and other elements that may be toxic to plants; the residual sodium carbonate (RSC), described as the difference between the sum of the carbonates and bicarbonates concentrations and the sum of the Ca and Mg concentrations; and the content of anions such as chloride, sulfate, and nitrate [27].

The trends of the ionic concentrations, specifically cations (Ca, Mg, Na, K, Na + K) and anions (SO_4 , NO_3 , $\text{HCO}_3 + \text{CO}_3$) in the soil circulating solution for the treatment N100, at a depth of 15 cm, are shown in Figure 1A. From the second week of March toward April and May, it is possible to find a light increase in the ionic concentrations due to fertigation. The more evident trend is for calcium and nitrate: the calcium ranges from 5.1 meq/L to 13.2 meq/L, with a monthly average of 7.4 meq/L in March, 11.5 meq/L in April, and 12.16 meq/L in May. The nitrates range from 1.94 meq/L to 12.04 meq/L, with a monthly average of 1.94 meq/L in March, 9.88 meq/L in April, and 9.46 meq/L in May. These trends may be related to the phases of plant development: a stronger uptake of these elements from the plants occurs in March, while this is stable in the following months. The sharp peaks appearing in the graphic are due to a long lag time between treatments. This is clear in the nitrate levels on the 10th and 20th of April of 11.96 and 7.10 meq/L, respectively. The same trend was registered between the 26th of April and the 5th of May, with 13.29 and 6.88 meq/L of nitrates, respectively. The concentrations of the other elements appear to be quite linear during the trial. The trends in ionic concentrations of the soil-circulating solution for the same trial, but at a depth of 35 cm, showed results like those obtained at 15 cm, but with lower amplitudes (results not shown).

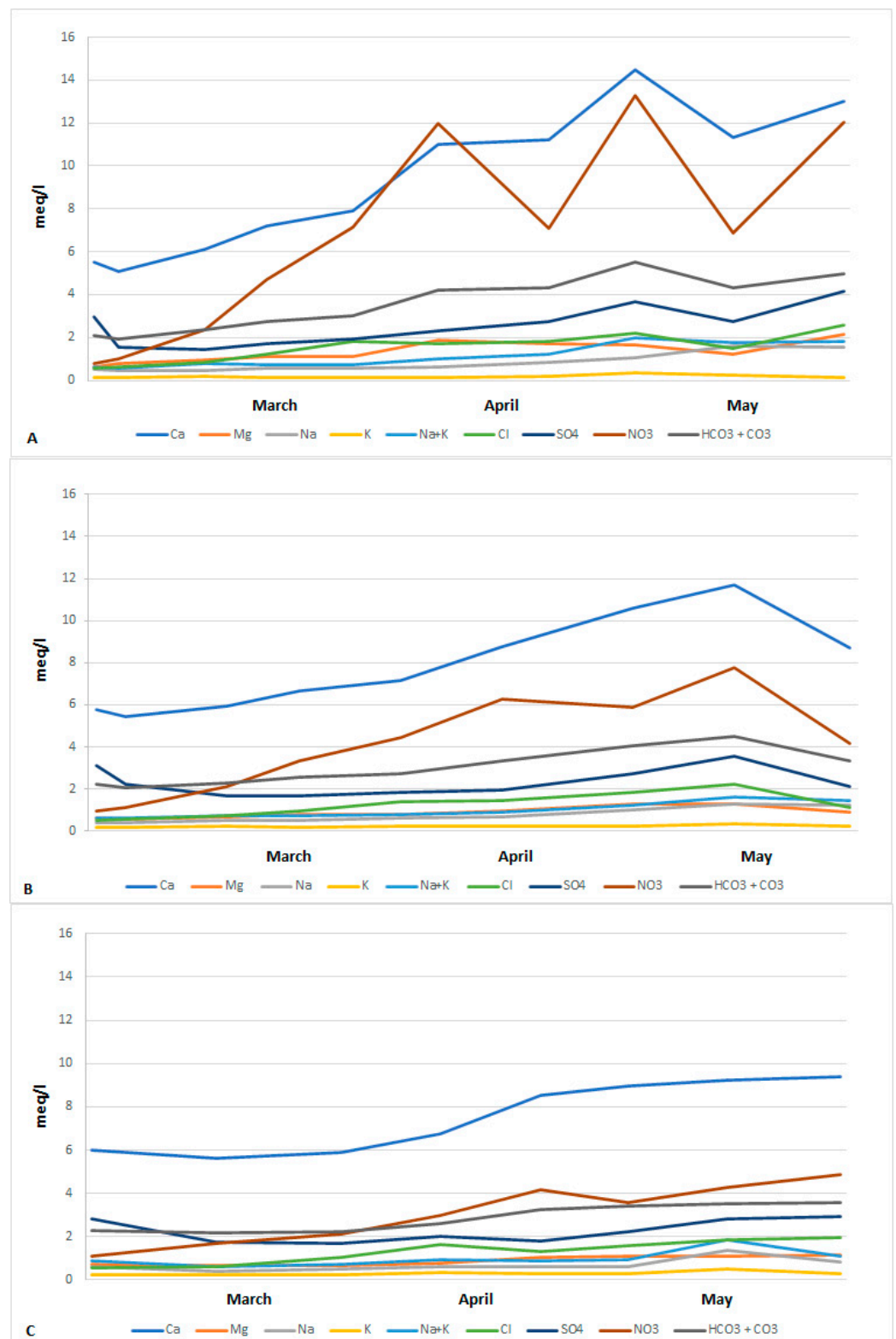


Figure 1. Ionic concentrations in the soil-circulating solution for the N100 (A), N80 (B), and N60 (C) treatments at a depth of 15 cm.

Considering the ion concentration in the soil-circulating solution for the N80 treatment at a depth of 15 cm (Figure 1B), a decrease in overall concentration values compared to N100 is evident. In particular, the lower amounts start from March. The values of the calcium and nitrate concentrations remain the most relevant. Ca ranges from 5.41 to 11.71 meq/L,

with a monthly average of 5.94 meq/L in March, 9.54 meq/L in April, and 8.68 meq/L in May. Nitrates range from 0.95 to 7.74 meq/L, with a monthly average of 1.87 meq/L in March, 6.08 meq/L in April, and 4.16 meq/L in May. There are also similar sharp peaks, but of a smaller entity, with those obtained at N100, corresponding to the 10th and 20th of April (6.28 and 5.86 meq/L) and the 26th of April and 5th of May (7.74 and 4.16 meq/L). The concentrations of the other elements appear to be quite linear during the trial, with fewer variations in comparison to trial N100.

The analyzed soil-circulating solution for the N60 (at 15 cm of depth) thesis shows slightly lower ionic concentrations and a more linear trend in comparison to N80 (Figure 1C). Calcium ranges from 5.62 to 9.38 meq/L, with a monthly average of 5.82 meq/L in March, 7.53 meq/L in April, and 9.30 meq/L in May. Nitrates range from 1.09 to 4.85 meq/L, with a monthly average of 1.49 meq/L in March, 4.54 meq/L in April, and 6.83 meq/L in May. The range in concentrations of the remaining elements is almost linear.

The NO_3 concentrations available in trials N100, N80, and N60 at different soil depths (15 cm/35 cm) are reported in Figure 2. Figure 2A shows that the NO_3 concentration in the soil solution analyzed at 15 cm of depth is higher than that at 35 cm in the N100 trial; there is a generally positive trend for the NO_3 concentrations at both depths during the months of the trial. The inflection points related to the 20th of April and the 5th of May should be due to a wide timing interval that separates two successive fertigation events.

At the same depth (15 cm), the N80 and N100 theses show a halving of the nitrogen concentration (Figure 1B); the reason can be attributed to the lower nitrogen supply at N80, according to the trial; moreover, the maximum reduction in NO_3 availability can be detected in N60 (Figure 1C). Comparing the three theses, a reduction can be noticed in the NO_3 availability from thesis N100 to N60 for both depths. Furthermore, a drop of -31% can be observed for the nitrate value (mg/L NO_3) from N100 to N80 (15 cm), and a decrease of 44% from N100 to N60 (15 cm) (Table 2). The same trend is observed between N100 and N60, but to a smaller extent, in the 35 cm experiment.

Table 2. Mean values (mg/L NO_3) in N100, N80, and N60 (15–35 cm).

Treatment	mg/L NO_3	
Depth	15 cm	35 cm
N100	384.33	189.59
N80	266.13	207.35
N60	167.69	147.81

Strawberries negatively respond to salt stress in terms of growth and yield, so it is a salinity-sensitive species [28]. The hydric stress, due to the electrical conductivity (ECs) of the saturated soil extract, is a factor that contributes to reducing the number of leaves, the leaf area, the shoot dry weight, the number of crowns, the yield, and the fresh weight of the fruit [29,30]. In the study of Barroso and Alvarez [31], the leaves of strawberry cultivars did not develop symptoms of toxicity for EC values lower than $2000 \mu\text{S/cm}$. In the study of HA-Joon et al. [32], the optimal EC value was detected at $1000 \mu\text{S/cm}$ instead of $2000 \mu\text{S/cm}$; the compared parameters were fruit length, diameter, weight, and plant yield. Moreover, the dry branch crowns and dry roots in the $1000 \mu\text{S/cm}$ experiment were heavier than those in the $2000 \mu\text{S/cm}$ experiment.

In our study, the electrical conductivity of the water during April and May 2017 at 15 cm of depth showed higher values for the three nitrogen trials. These results agree with the nutrient concentration explained above. In N100 and N80, the maximum registered value is $1400 \mu\text{S/cm}$ (Figure 3A,B), while in N60 this value is lower, slightly exceeding $1000 \mu\text{S/cm}$ (Figure 3C). The trends are similar, but to a lesser extent, for the water sampled at 35 cm of soil depth.



Figure 2. Evolution of NO₃ amount in the soil solution analyzed at 15 and 35 cm depths for the N100 (A), N80 (B), and N60 (C) treatments, March–April–May 2017.

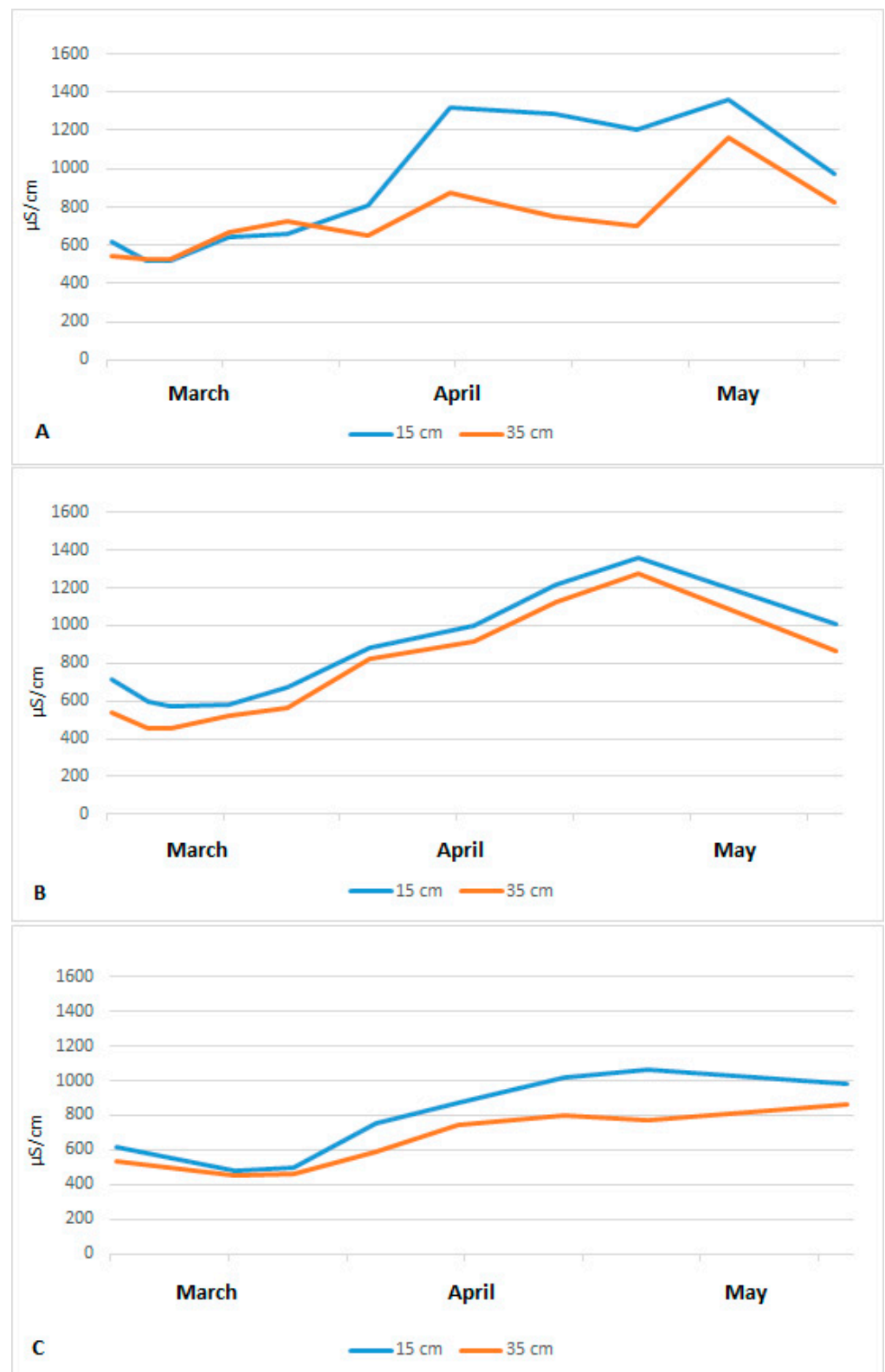


Figure 3. Water electrical conductivity was analyzed at 15 and 35 cm for the N100 (A), N80 (B), and N60 (C) treatments, March–April–May 2017.

3.2. Vegetative Parameters

The vegetative crops' growth is dependent on the organic matter, which enhances both the soil's microbiological and biochemical activities [33]. Nevertheless, the soil organic matter content hides the effect of mineral nitrogen intake, the principal macro-element

for the growth of plants [34]. This study took place in soil that was poor in both nitrogen concentration (0.90 g/kg) and organic matter (11.9 g/kg) so that the fertilization practices were not masked by the soil properties. Given an overview of the studied vegetative parameters (Table 3), it is evident that that year of cultivation (a) resulted as an impacting factor influencing the branch crowns, plant height, and leaf number. Cultivar (b) was decisive for the branch crowns and plant height. Treatment (c) seemed to have a single key role in the branch crown number. Furthermore, it is remarkable that the interaction (a) × (b) impacted all of the vegetative features. The interaction (a) × (c) showed a significant incidence for the branch crowns. The interaction (a) × (b) × (c) seemed to have an insignificant impact on the studied parameters, as well as interaction (b) × (c). Considering the majority of the studied parameters, each cultivar responded similarly to every treatment.

Table 3. Two-way analysis of variance (ANOVA) for the vegetative parameters.

Parameter	Branch Crowns	Inflorescences Number	Plant Height	Leaves Number
Year (a)	**	NS	**	*
Cv (b)	**	NS	**	NS
Treatment (c)	*	NS	NS	NS
Year × Cv (a) × (b)	**	**	**	**
Year × Treatment (a) × (c)	**	NS	NS	NS
Cv × Treatment (b) × (c)	NS	NS	NS	NS
Year × Cv × Treatment (a) × (b) × (c)	NS	NS	NS	NS

** = significant interaction with $p < 0.01$; * = significant interaction for $p < 0.05$; NS = no significant interaction.

The plant height, branch crown number, and inflorescence number did not exhibit statistically significant differences among the treatments (Tables 4 and 5). A possible reason could be that all of the plants followed a standard fertilization plan from September until March and reduced nitrogen treatments during spring when the plants had already a developed vegetative structure. It is conceivable that a reduction in the fertilizer administered in September may have resulted in different vegetative features between cultivars managed under different treatments. A previous study [17] demonstrated the crucial importance of fertilization timing for short-day (SD) strawberry plants. It is demonstrated that, under controlled photoperiod conditions conducted in a phytotron, feeding SD strawberry plants one week from the beginning of the SD period could double the number of flowers compared to plants treated two weeks before the start of SD conditions [17].

Among the vegetative parameters, the only significant result seemed to be the “Sibilla”’s leaf number (Table 4). The “Sibilla” cultivar treated with 60% nitrogen administration showed a significant foliage decrease (26.2 ± 5.6 leaves at N100 and 23.6 ± 4.3 leaves at N60) in contrast with the other two tested plant cultivars, which did not show any significant differences among the treatments. Medeiros et al. [35] observed the same feature of the “Sibilla” cultivar in the “Oso Grande” cultivar, namely a great increase in terms of the leaves number with increasing nitrogen intake.

Table 4. Effects of nitrogen on plant height and leaf number in different strawberry cultivars.

Cultivar	Plant Height (cm)			Leaves Number			
	Treatment	N100	N80	N60	N100	N80	N60
“Cristina”		33.3 ± 3.2 c	34.2 ± 3.5 c	33.3 ± 4.0 c	25.4 ± 5 ab	24.6 ± 4.0 ab	24 ± 6.1 ab
“Romina”		38.9 ± 2.9 ab	40.2 ± 3.0 a	40.2 ± 2.6 a	24.1 ± 7.1 ab	25.8 ± 6.2 ab	24.4 ± 6.2 ab
“Sibilla”		38.5 ± 7.6 ab	37.9 ± 5.9 b	38.3 ± 5.6 b	26.2 ± 5.6 a	25.1 ± 6.7 ab	23.6 ± 4.3 b
AVERAGE		36.9 ± 5.6 NS	37.4 ± 5 NS	37.3 ± 5.1 NS	9.7 ± 1.1 NS	9.6 ± 1 NS	9.7 ± 1.3 NS

Values with the same lower-case letter among treatments for the same parameter were not statistically different for Fisher’s LSD test ($p < 0.05$). Average values for each parameter with the same uppercase letter were not statistically different for Fisher’s LSD test ($p < 0.05$). NS = not significant. Values are expressed as means of two years (2017–2018) ± standard deviation (SD).

Table 5. Effects nitrogen on branch crowns number and inflorescences number in different strawberry cultivars.

Cultivar	Branch Crowns Number			Inflorescences Number			
	Treatment	N100	N80	N60	N100	N80	N60
“Cristina”		4.1 ± 1.4 bc	4.2 ± 1.4 abc	3.6 ± 1.2 c	11.9 ± 3.5 ab	12.1 ± 3.2 a	11.3 ± 3.3 ab
“Romina”		4.8 ± 2 ab	4.9 ± 2.3 a	4.8 ± 2 ab	11.1 ± 2.8 ab	11.4 ± 2.9 ab	10.6 ± 3.4 b
“Sibilla”		4.8 ± 2.1 ab	4.6 ± 2.1 ab	4.4 ± 1.7 ab	11.3 ± 2.7 ab	10.9 ± 3 ab	11.8 ± 3.4 ab
AVERAGE		4.5 ± 1.9 NS	4.6 ± 2 NS	4.3 ± 1.8 NS	11.4 ± 3 NS	11.5 ± 3.1 NS	11.3 ± 3.4 NS

Values with the same lower-case letter among treatments for the same parameter were not statistically different for Fisher’s LSD test ($p < 0.05$). Average values for each parameter with the same uppercase letter were not statistically different for Fisher’s LSD test ($p < 0.05$). NS = not significant. Values are expressed as means of two years (2017–2018) ± standard deviation (SD).

3.3. Productive Parameters

In considering the productive parameters (Table 6) year (a), cultivar (b), treatment (c), and interaction (a) × (b) generally exhibited a great impact on the average fruit weight (AFW) and commercial production. The interaction (a) × (c) seemed to influence the AFW, as well as the interaction (a) × (b) × (c). Interaction (b) × (c) did not manifest any correlation with any of the productive parameters. The evaluation of the productive parameters did not highlight any great change among the different treatments.

Table 6. Two-way analysis of variance (ANOVA) for the productive parameters.

Parameter	Average Fruit Weight	Commercial Production
Year (a)	**	**
Cv (b)	**	**
Treatment (c)	**	**
Year × Cv (a) × (b)	**	**
Year × Treatment (a) × (c)	**	NS
Cv × Treatment (b) × (c)	NS	NS
Year × Cv × Treatment (a) × (b) × (c)	**	NS

** = significant interaction with $p < 0.01$; NS = no significant interaction.

More specifically, the average fruit weight values did not show significant differences among the three nitrogen treatments (Table 7). Similar results but at lower nitrogen doses (60, 40, 20 kg N/ha) were obtained in the trial conducted by Cvelbar Weber et al. [13]. Regardless of the nitrogen supplied to the plants, “Cristina” exhibited a higher average fruit weight than “Romina” (of about 13–15 g) and “Sibilla” (about 10–13 g).

Table 7. Effects of nitrogen on commercial production and average fruit weight in different strawberry cultivars.

Cultivar	Commercial Production (g/Plant)			Average Fruit Weight (g)			
	Treatment	N100	N80	N60	N100	N80	N60
“Cristina”		783.1 ± 114.9 a	744.9 ± 124.4 ab	710.5 ± 170.1 abc	32.4 ± 3.3 a	31.2 ± 3.9 a	33.4 ± 2.3 a
“Romina”		529.9 ± 95.1 de	533.7 ± 70.6 de	496.4 ± 60.1 e	18.8 ± 2.6 b	18.3 ± 2.9 b	18.3 ± 2.2 b
“Sibilla”		635.8 ± 51.2 bcd	609.6 ± 34.6 cd	600 ± 39.3 de	21.3 ± 2.3 b	21.4 ± 3.2 b	20.7 ± 2.1 b
AVERAGE		649.6 ± 136.9 NS	629.4 ± 120.2 NS	602.3 ± 134.6 NS	24.1 ± 6.6NS	23.7 ± 6.5 NS	24.1 ± 7.1 NS

Values with the same lowercase letter among treatments for the same parameter were not statistically different for Fisher’s LSD test ($p < 0.05$). Average values for each parameter with the same uppercase letter were not statistically different for Fisher’s LSD test ($p < 0.05$). NS = not significant. Values are expressed as means of two years (2017–2018) ± standard deviation (SD).

The statistical analysis confirmed an insignificant difference in the commercial production among the different nitrogen supplies (Table 7). Similar findings were already observed in the studies from Darnell and Stutte, Cantliffe et al., and D’Anna et al. [36–38], which indicated that plant growth and yield may not be reduced by the volume of the administrated NO_3^- , but by the capacity to reduce and assimilate the NO_3^- . This capacity could be inhibited by the lack of reductants, like NADH and NADPH, or the deficiency of skeleton carbohydrates needed for the assimilation. In our study, the commercial production originating from plants with the lowest dose of nitrogen showed a slight downward trend of -7% compared to N100. In the N100 trial, “Cristina” appeared to be commercially more productive, followed by “Sibilla”, and then “Romina”. This tendency was maintained for all of the nitrogen trials, although the reduction in this element provided no significant decrease in production for all three cultivars. In other studies, the effect of higher rates of N fertilization on marketable yield was clear, with a significant increase with respect to the lower doses [12,39,40].

In general, from the productive point of view, the results of our study showed that there was not a negative effect of nitrogen reduction on plant yield, and this outcome could be read as an indication of the need to better test the nitrogen requirements of the different cultivars in different conditions, in order to reduce the environmental impact that can be derived from the excessive use of nitrogen.

3.4. Qualitative Parameters

In evaluating all of the studied qualitative parameters, year (a) seemed to affect the sugar content, firmness, brightness, and redness of the fruit of the three tested cultivars. Among the factors, cultivar (b) showed the greatest influence in terms of the fruit sugar content, titratable acidity, firmness, brightness *L, redness *a, yellowness *b, and the Chroma index. The combination of (a) × (b) greatly influenced the fruit sugar content, firmness, brightness *L, redness *a, yellowness *b, and the Chroma index. The impacts of treatment (c) and the interactions (a) × (c), (b) × (c), and (a) × (b) × (c) were not relevant to the studied parameters (Table S1).

According to our results, genotype was the principal factor affecting the sugar content and titratable acidity of strawberry fruit (Table 8), and this outcome is in line with those other previous studies [41–44]. In particular, the fruits of “Romina” and “Sibilla” revealed the highest sugar content in all of the nitrogen trials (Table 8). The fruit sugar content and titratable acidity did not show significant differences among treatments for these cultivars, as registered by Sedri and Farami [45]. However, these results contrast with other studies [38–40,46], which pointed out the correlation between lower doses of nitrogen supply and higher fruit soluble solids content.

Table 8. Effects of nitrogen on sugar content and titratable acidity in different strawberry cultivars.

Cultivar	Sugar Content (°Brix)			Titratable Acidity (meqNaOH/100 g Fruit Weight)			
	Treatment	N100	N80	N60	N100	N80	N60
“Cristina”		6.6 ± 0.5 c	6.4 ± 0.6 c	6.4 ± 0.5 c	10.9 ± 1abcd	10.4 ± 1 d	10.9 ± 1.7 abcd
“Romina”		7.6 ± 0.6 ab	7.2 ± 0.5 b	7.5 ± 0.7 ab	11.1 ± 0.8 abcd	10.6 ± 0.7 cd	10.6 ± 0.8 bcd
“Sibilla”		7.8 ± 0.8 a	7.9 ± 0.7 a	7.6 ± 1.1 ab	11.5 ± 1.4 a	11.4 ± 1.8 ab	11.3 ± 1.6 abc
AVERAGE		7.3 ± 0.9 NS	7.2 ± 0.8 NS	7.2 ± 0.9 NS	11.2 ± 1.1 NS	10.8 ± 1.3 NS	10.9 ± 1.4 NS

Values with the same lowercase letter for the same parameter were not statistically different for Fisher’s LSD test ($p < 0.05$). Average values for each parameter with the same uppercase letter were not statistically different for Fisher’s LSD test ($p < 0.05$). NS = not significant. Values are expressed as means of two years (2017–2018) ± standard deviation (SD).

The fruits’ firmness did not seem to be influenced by the administration of nitrogen (Table 9). The “Sibilla” fruits showed the hardest texture, followed by “Romina” and “Cristina” in all the nitrogen trials. These results were in contrast with the studies conducted by D’Anna et al., Asghari et al., and Cardenosa et al. [38,40,47], which found a higher fruit consistency when reducing the administration of nitrogen. According to Van der Boon [48], the excess nitrogen could cause an irregular ripening of fruits, resulting in soft consistency and poor taste. An excessive nitrogen presence in the fruits corresponds to a calcium reduction, which decreases the texture and consequently, the shelf life of the product, considering that calcium is an intermolecular binding agent that is responsible for the pectin–protein complexes of the middle lamella [13]. Therefore, a decrease in nitrogen administration should determine a better quality of fruit firmness.

Table 9. Effects of nitrogen on firmness and Chroma index in different strawberry cultivars.

Cultivar	Firmness (g/cm ²)			Chroma Index			
	Treatment	N100	N80	N60	N100	N80	N60
“Cristina”		292.8 ± 58.3 c	297 ± 43.7 c	298.8 ± 59.5 c	44.1 ± 3.6 c	43.8 ± 3.4 c	44.3 ± 3.3 c
“Romina”		356.9 ± 64.9 b	348.9 ± 47.8 b	352.8 ± 51.9 b	48.9 ± 1.5 b	49.4 ± 1.8 b	49.4 ± 1.9b
“Sibilla”		414.2 ± 104.3 a	425 ± 82 a	415.5 ± 102.5 a	52.1 ± 2.4 a	52.1 ± 2.3 a	50.6 ± 6.7 ab
AVERAGE		354.7 ± 91.9 NS	356.9 ± 79.5 NS	355.7 ± 87.6 NS	48.3 ± 4.2 NS	48.4 ± 4.3 NS	48.1 ± 5.2 NS

Values with the same lowercase letter for the same parameter were not statistically different for Fisher’s LSD test ($p < 0.05$). Average values for each parameter with the same uppercase letter were not statistically different for Fisher’s LSD test ($p < 0.05$). NS = not significant. Values are expressed as means of two years (2017–2018) ± standard deviation (SD).

In the present study, changing the nitrogen doses in all of the treatments did not sway the Chroma index (Table 9). The only difference was found in comparing the studied cultivars: “Sibilla” had the darkest fruits, followed by “Romina”, and then “Cristina”. On the contrary, the study conducted by Yoshida et al. [49] exhibited a correlation between anthocyanin synthesis and nitrogen fertilization. Lack of nitrogen during the fertilization treatment seemed to lower the anthocyanins accumulation in fruit. Anthocyanins accumulate in the inner strawberry flesh over the course of the late stage of fruit development, and improper fertilization may reduce the biosynthesis of these molecules.

4. Conclusions

The objective of this study was to verify any changes in the vegetative, productive, and qualitative responses of short-day cultivars under different nitrogen supplies. The purpose of this project was to achieve a positive tradeoff between good plant performance and a low environmental impact. The most interesting results obtained can be disclosed and applied by growers working in open field conditions in a crop protected under a plastic tunnel, in soil conditions with low organic matter and total N levels. Analyzing

these results, it seemed that a decrease in nutritional intake, from about 115 kg/ha (N100) to about 70 kg/ha (N60) of nitrogen, did not negatively affect plant development. Total production is one of the main productive parameters. No significant difference was noticed for this parameter among short-day cultivars with lower nitrogen rates. In general, the average fruit weight was not affected by nitrogen reduction. Regarding the qualitative parameters, the sugar content value remained stable in fruits from the short-day cultivars for all nitrogen supply levels. In each N treatment, all the cultivars maintained high values of fruit titratable and acidity and kept a medium fruit firmness at a 60% nitrogen supply (N60). The Chroma index was not influenced by different nitrogen amounts.

From these data, it is clear that there is not a marked negative impact of nitrogen reduction on the three tested cultivars for any of the analyzed parameters. There are some differences in the response of the tested cultivars to nitrogen reduction for the vegetative, productive, and qualitative parameters but, in general, all of them are suitable for cultivation with a reduced input of nitrogen, reducing the environmental impact and saving inputs from growers.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/horticulturae9111165/s1>, Table S1: Two-way analysis of variance (ANOVA) for the qualitative parameters.

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