

Article

Optimizing Nitrogen Nutrient Management for the Sustainable Enhancement of Secondary Metabolites and Yield in Onion Cultivation

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Abstract: This study investigates the impact of nitrogen (N), sulfur (S), and iron (Fe) fertilization on secondary metabolites, particularly quercetin and its forms, in onion bulbs (*Allium cepa* L.). Field experiments over two years examined four onion varieties with red, yellow, and white colors of bulbs: Kamal, Robin, Pueblo, and Mundo. The parameters investigated included the yield, dry matter content, and average onion weight. The phenolic and flavonoid contents were also analyzed. The free quercetin, bound quercetin, and total quercetin contents were determined using the HPLC method. The results demonstrated notable increases in yield following the application of nitrogen (NH_4^+) and sulfur (SO_4^{2-}) fertilizers. Incorporating iron (Fe^{2+}) alongside these fertilizers did not yield a significant impact compared to N+S variant. The phenolic and flavonoid content varied with fertilization, while the quercetin content did not yield statistically significant results. Overall, the study highlights the complex relationship between fertilization practices and secondary metabolite production in onions, emphasizing the need for sustainable intensification in modern agriculture.

Keywords: quercetin; phenolics; sustainable agriculture; *Allium cepa* L.



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1. Introduction

In the dynamic landscape of modern agriculture, understanding the intricate interplay between fertilization practices, particularly nitrogen (NH_4^+), iron (Fe^{2+}), and sulfur (SO_4^{2-}) applications, and their impact on the nutritional profile of crops is imperative [1–3]. It is recognized that the selection or recommendation of an optimal fertilization system is crucial for each plant, as it contributes to achieving a high yield and ensuring the quality of plant products for food consumption [4,5]. For example, effective quality improvement technology for garlic (*Allium sativum* L.) involves defining the metabolic transformation of sulfur-containing compounds under exogenous regulation by mineral elements, specifically increasing alliin and organic sulfides in garlic bulbs while maintaining productivity. Applying 125 kg S ha^{-1} of fertilizers and treating with lipoic acid doubles the food and pharmacological value of garlic bulbs [6]. However, nitrogen (NH_4^+) fertilization is also important for members of the *Allium* genus. For instance, nitrogen in onion bulbs accounts for 65% of the nitrogen in the aboveground biomass, and nitrogen use efficiency can be improved with slow- or controlled-release fertilizers [7]. Another study demonstrated that treatments with 25% usable humidity and nitrogen fertilization of 150 and 250 kg ha^{-1} favored the physical, chemical, and bioactive quality of onion bulbs [8]. Additionally, the interaction effect of nitrogen and sulfur significantly increased the main growth parameters and yield of onion bulbs under the application of 200 kg N ha^{-1} and 45 kg S ha^{-1} [9]. Plant nutrition responsibility adheres to a food system and circular economy perspective [10].

Onions (*Allium cepa* L.) have emerged as a noteworthy focus due to their rich content of bioactive compounds, including the health-promoting quercetin and other secondary metabolites [11,12]. Inter- and intra-landrace variability in various onion (*Allium cepa* L.)

varieties was observed in terms of volatile profiles and phenolic compounds [13], which may be influenced by environmental factors. The multifaceted influence of nitrogen and sulfur fertilization on the synthesis and accumulation of these phytochemicals in onion bulbs is a critical aspect of contemporary agricultural research.

Nitrogen and sulfur, two essential nutrients for plant growth and development, play pivotal roles in shaping the metabolic pathways responsible for secondary metabolite production [14,15]. The intricate relationship between these fertilizers and the biochemical processes within the onion plants underscores the need for targeted investigations to optimize their application for enhanced nutritional content [16,17]. The application of boron, copper, and zinc ions has been shown to favor a greater accumulation of micronutrients in onion bulbs [18]. However, studies incorporating Fe as an additional fertilization element, a crucial micronutrient for the photosynthesis process [19], are limited or lacking. Moreover, nowadays, studies align with the principles of sustainable agriculture, aiming not only to maximize yield but also to minimize the environmental impact and promote long-term soil health.

In the face of a growing global population, the significance of sustainable agricultural practices cannot be overstated in ensuring food security while preserving the nutritional quality of crops [20,21]. Sustainable agriculture plays a crucial role in mitigating the environmental impact of fertilization practices and enhancing the resilience of agroecosystems. This study aims to provide actionable insights for farmers, policymakers, and researchers by exploring the intricate relationships between fertilization, crop varieties, and the phytochemical composition. Specifically, the research focuses on unraveling the distinct effects of nitrogen, iron, and sulfur fertilization on the quercetin and secondary metabolite content in onions, with an emphasis on promoting the sustainable intensification of agriculture.

2. Materials and Methods

2.1. Characteristics of Onion Varieties

Kamal: Kamal is a red onion variety known for its premium quality and delightful taste. Its bulbs are uniform with thin necks, exhibiting good resistance to mold and ensuring excellent storage capability. Sowing typically occurs outdoors between March and April, with a maturation period of approximately 24 weeks.

Robin: Robin, a dark red variety introduced in 2005, features semi-erect to horizontal leaves of a moderate length. The bulbs are medium to large and broadly ovate with a narrow neck. They tend to sprout moderately late to late during storage.

Pueblo: Pueblo is a very early white onion variety producing round bulbs suitable for both fresh market consumption and drying purposes, yielding outstanding results. Sowing usually takes place in April, with harvesting in August and storage until October. The growing period spans approximately 115 days from sowing, with a seeding rate of 1 million seeds per hectare.

Mundo: Mundo, a late-maturing variety, is ideal for spring sowing. It features moderately long, upright leaves with a slight droop and a light green hue. The bulbs are large and round with a moderate neck thickness, boasting firm white flesh suitable for year-round storage. Mundo has been a registered variety since 1998 (Figure 1).



Figure 1. Cont.



Figure 1. Experimental onion varieties: (A) Kamal, (B) Robin, (C) Pueblo, (D) Mundo.

2.2. Field Small-Plot Nutrition Experiment Description

A two-year field small-plot nutrition experiment with four varieties of kitchen onion (*Allium cepa* L.) was established in the cadastral area of PD Madunice. The region of Dolné Považie, where the village of Madunice is located, belongs to a warm, moderately dry climatic zone with mild winters. The average annual air temperature (30-year average) reaches 9.2 °C. The coldest month is January, with an average temperature of −2 °C, and the warmest is July, with an average temperature of 18.9 °C. The experimental site in the Madunice locality is formed by moderately heavy fluvial soils. The onion varieties Kamal, Robin, Pueblo, and Mundo were used in the field experiment.

The nutrition experiment with onion varieties was established in the form of a small-plot trial on a site with a fluvial soil type, the agrochemical characteristics of which are provided in Table 1. The soil was a medium–heavy fluvisol containing fluvial silicate and carbonate sediments with textural diversity and varying mineral richness. It was characterized as fertile to very fertile according to the productivity potential. Before the onion seeds were sown directly in spring, germinating plants (weeds) on the soil surface were removed, and the surface layer (to a depth of 30 mm) was cultivated.

Table 1. The results of the soil agrochemical analysis before sowing.

Year	pH	Humus (%)	Nutrient Content (mg kg ^{−1} of Soil)						
			N _{an}	S	Fe	P	K	Ca	Mg
1 year	5.96	2.82	7.2	18.0	19.68	35	260	4200	830
2 year	6.90	4.16	13.5	27.5	14.13	17.5	242.5	5600	935.0

The experimental onion varieties were sown at the optimal agronomic term using a precision seeder for precise seeding into four double rows spaced 0.3 m apart and at a depth of 20 mm. The field bed width was 1.5 m. As part of the cropping sequence, maize was planted as the preceding crop in the first year, followed by annual sunflower in the second growing year.

In the experiment conducted using a block method with the arrangement of experimental variants in strips and with the regular distribution of replications within individual blocks, the influence of different nutrition and fertilization variants on the yield and content substances of the four onion varieties was investigated.

In each block, four fertilization variants were monitored for each variety:

- 1 Variant—unfertilized control (further referred to as “0” in the text).
- 2 Variant—fertilization at a rate of 140 kg N ha^{−1} (NH₄⁺).
- 3 Variant—fertilization at a rate of 140 kg N ha^{−1} + fertilization at a rate of 19 kg of available sulfur (SO₄²⁺) per hectare (N+S).
- 4 Variant—fertilization at a rate of 140 kg N ha^{−1} + fertilization at a rate of 19 kg of available sulfur (SO₄²⁺) per hectare + 5 kg of iron (Fe²⁺) per hectare (N+S+Fe).

Each fertilization variant was replicated four times, with the size of one replication (experimental plot) being 5 m², meaning the area of one variant amounted to 20 m².

2.3. Fertilization Application Description

The nitrogen application aimed at reaching a soil level of 140 kg ha⁻¹. It was calculated based on soil N content analysis. Split into two parts, the calculated doses (120 kg N ha⁻¹ in the first year and 80 kg N ha⁻¹ in the second year) were applied. The first part (80 kg N ha⁻¹ in the first year and 53 kg N ha⁻¹ in the second year) was applied one week post-sowing, while the second part (40 kg N ha⁻¹ in the first year and 27 kg N ha⁻¹ in the second year) was applied at the fifth true leaf stage of the onion.

To meet the specified sulfur level (19 kg ha⁻¹), 33.88 kg S ha⁻¹ was applied to the third and fourth fertilization variants in the first year, and 11.65 kg S ha⁻¹ was applied in the second year using ammonium sulfate. Iron was applied with nitrogen and sulfur for the fourth variant: 2.5 kg Fe ha⁻¹ as a soil spray post-sowing and at the fifth true leaf stage, foliarly as FeSO₄·7H₂O.

Phosphorus fertilization occurred in autumn via simple superphosphate application (8% P) based on soil analysis. Potassium fertilization was unnecessary due to the soil's high potassium content.

Granulated urea, ammonium sulfate, and urea were used for the first partial nitrogen application across variants, while the second application utilized DAM-390 for all fertilized variants.

2.4. Determining Yield and Other Fundamental Parameters

In the field experiment with kitchen onion, the following key parameters were determined: the yield, dry matter content, dry matter yield, and average weight of one onion.

The quantity of fresh onions harvested per hectare (t ha⁻¹) was measured to assess the productivity of the crop. The percentage of dry matter in the harvested onions was analyzed, providing insight into the proportion of solid material present in the crop. Based on the dry matter content and the total yield, the yield of dry matter per hectare (t ha⁻¹) was calculated. The average weight of a single onion (in grams) was determined, helping to understand the size distribution of the harvested onions.

2.5. Determining the Content of Free Quercetin in Kitchen Onion

After harvest maturity, the content of free quercetin in the onion was determined in an extract of 80% ethanol using an HPLC/MS/MS system (AGILENT 1260, Santa Clara, CA, USA) with a DAD detector, Triple Quadrupole 6410 MS/MS detector, following the method described by Patil et al. (1995), expressed in mg kg⁻¹ of fresh weight [22].

2.6. Sample Preparation for HPLC Analysis

Sample preparation involved obtaining 20 g of tissue from the middle section around the equator of the bulb, excluding dry skin. This tissue was blended with 80 mL of 80% ethanol for 1 min and then filtered through filter paper. The filtrate was stored in screw-capped vials at −20 °C until analysis. A 5 mL aliquot was evaporated to dryness under vacuum at 50 °C using a Buchler evaporator (Evapomix, Washington, DC, USA) and then re-suspended in 1 mL of ethanol (80%). The extracts were filtered through 0.45 µm nylon membrane filters (Sigma-Aldrich, Merck, Germany), and a 10 µL volume of this solution was injected into the HPLC system for analysis.

2.7. Determination of the Bound Quercetin Content in Onion

To determine the bound quercetin content in kitchen onion, a method of extraction in 62.5% ethanol and 6 M HCl was employed, followed by quantification using an HPLC instrument according to the method described by Hertog et al. (1992) [23]. The results were calculated in mg kg⁻¹ of fresh weight of the onion.

2.8. Determination of the Total Phenolic Content in the Onion

The phenolic content in the kitchen onion was determined according to Ragazzi and Veronese (1973) in an extract of 2% trichloroacetic acid (TCA) using the Folin-Ciocalteu method, expressed in mg of gallic acid (GA) per 1000 g of fresh weight of the onion [24].

2.9. Determination of the Total Flavonoid Content in the Experimental Onion Varieties

The flavonoid content was determined according to Bergamini et al. (2004) in an extract of 2% trichloroacetic acid (TCA), expressed in mg of catechin per 1 kg of fresh weight of the onion [25]. Note: In subsequent text, the flavonoid content will be reported in units of mg kg^{-1} , indicating mg of catechin per kilogram of fresh weight of the onion.

3. Results

All onion varieties exhibited the highest yields of fresh weight in the variant fertilized with nitrogen and sulfur (Variant 3 (N+S)). There was a significant increase in the yield of 15% for the Mundo cultivar, 29% for the Robin cultivar, 15% for the Kamal cultivar, and 19% for the Pueblo cultivar.

The highest yield of dry matter, on average over 2 years, was observed for the white variety Pueblo in the variant with N+S+Fe fertilization, reaching 5.11 t ha^{-1} . Following this, the red varieties Kamal (16%) and Robin (17%) were observed, and the lowest dry matter content was determined for the yellow variety Mundo (13%) (Table 2).

The phenol content in onion varieties varied due to nitrogen fertilization in all its experimental variants. On average, over 2 years, nitrogen fertilization had the most significant impact on the phenol content in onions, especially in the Kamal variety (0.929 mg g^{-1}) and Robin variety (0.865 mg g^{-1}). For the white variety Pueblo and the yellow variety Mundo, N+S+Fe fertilization seemed to be the most effective (Table 3).

Table 2. The yield parameters of the experimental onion cultivars (2-year experiment).

Variant	Yield (t ha^{-1})	Dry Matter Content (%)	Dry Matter Yield (t ha^{-1})	Average Weight of One Onion (g)
Cultivar Mundo				
1 (0)	26.12 _a	12.01 _a	3.13 _a	59.66 _a
2 (N)	29.04 _b	11.93 _a	3.45 _b	68.99 _b
3 (N+S)	30.57 _b	12.32 _a	3.69 _c	65.55 _{ab}
4 (N+S+Fe)	29.92 _b	12.41 _a	3.79 _{bc}	77.05 _c
LSD _{0.05}	2.35	0.74	0.30	7.82
Cultivar Robin				
1 (0)	23.72 _a	13.28 _b	3.06 _a	61.64 _a
2 (N)	31.57 _b	12.71 _a	3.99 _b	84.83 _c
3 (N+S)	33.50 _b	13.24 _b	4.40 _b	81.72 _c
4 (N+S+Fe)	32.76 _b	13.05 _b	4.22 _b	73.64 _b
LSD _{0.05}	3.35	0.33	0.44	7.67
Cultivar Kamal				
1 (0)	23.68 _a	14.53 _{ab}	3.37 _a	39.96 _a
2 (N)	26.62 _b	14.14 _a	3.67 _{ab}	45.67 _{bc}
3 (N+S)	28.93 _c	14.61 _b	4.23 _c	48.61 _c
4 (N+S+Fe)	27.82 _{bc}	14.54 _b	3.94 _{bc}	42.94 _{ab}
LSD _{0.05}	2.03	0.38	0.35	5.44

Table 2. *Cont.*

Variant	Yield (t ha ^{−1})	Dry Matter Content (%)	Dry Matter Yield (t ha ^{−1})	Average Weight of One Onion (g)
Cultivar Pueblo				
1 (O)	22.86 _a	19.05 _b	4.17 _a	62.25 _a
2 (N)	25.78 _b	18.61 _b	4.77 _b	73.51 _c
3 (N+S)	28.60 _c	17.93 _a	5.00 _b	69.42 _{bc}
4 (N+S+Fe)	28.37 _c	18.59 _b	5.11 _b	65.50 _{ab}
LSD _{0.05}	2.35	0.53	0.37	5.47

Note: The same letters in the averages of the observed characteristics indicate a statistical difference; LSD = least significant difference, at $p = 95\%$ ($\alpha = 0.05$).

Table 3. The level of phenolics and flavonoids in the experimental onion cultivars (2-year experiment).

Variant	Cultivar			
	Kamal	Robin	Pueblo	Mundo
Phenolics (mg gallic acid equivalents per g FW)				
1 (O)	0.808 _a	0.776 _a	0.395 _a	0.752 _a
2 (N)	0.929 _b	0.865 _a	0.458 _b	0.858 _b
3 (N+S)	0.861 _{ab}	0.842 _a	0.453 _b	0.858 _b
4 (N+S+Fe)	0.856 _{ab}	0.849 _a	0.500 _c	0.921 _b
LSD _{0.05}	0.089	0.098	0.036	0.093
Flavonoids (mg g ^{−1} FW)				
1 (O)	0.273 _b	0.278 _a	0.104 _a	0.160 _a
2 (N)	0.282 _b	0.353 _b	0.105 _a	0.158 _a
3 (N+S)	0.286 _b	0.279 _a	0.105 _a	0.154 _a
4 (N+S+Fe)	0.238 _a	0.299 _a	0.116 _a	0.158 _a
LSD _{0.05}	0.033	0.030	0.018	0.013

Note: The same letters in the averages of the observed characteristics indicate a statistical difference; LSD = least significant difference, at $p = 95\%$ ($\alpha = 0.05$).

Fertilization had a differential impact on the flavonoid content during the 2-year experiment across various onion varieties. The highest flavonoid content in the N+S-fertilized variant was observed in the red variety Kamal (0.286 mg g^{−1} FW). In the N+S+Fe-fertilized variant, the highest flavonoid content was found in the red variety Robin (0.299 mg g^{−1} FW). However, the highest overall flavonoid content was achieved in the N-fertilized variant, with the red variety Robin reaching a level of 0.353 mg g^{−1} FW. The yellow variety Mundo did not show significant changes in flavonoid levels under the effect of variant fertilization (0.160 mg g^{−1} FW in the control variant (O)). The lowest contents of flavonoids were recorded for the white variety Pueblo in all experimental variants with fertilization (ranging from 0.104 to 0.116 mg g^{−1} FW). At the same time, in the N+S-fertilized variant, the white variety Pueblo showed an increased level of flavonoids by 10% compared to the control variant (without fertilization (O)).

Over the span of 2 years, we observed statistically significant variations in the determination of free, bound, and total quercetin contents among different years and onion varieties (Table 4). However, the impact of fertilization on the quercetin content and its fractions did not yield statistically significant results. The hypothesis suggesting that the foliar application of fertilizer containing iron would elevate the quercetin concentration in onions was not substantiated. Conversely, the crucial metabolic roles of nitrogen and sulfur in quercetin formation were confirmed, evident in the increased concentration of

bound quercetin with the combined application of nitrate and sulfate nutrition in red onion varieties. In a 2-year experiment involving the red Kamal variety, the highest quercetin production (17.46 kg ha^{-1}) was observed in the N+S fertilization variant.

Table 4. Quercetin fractions in onions and their share of the total quercetin content (mg g^{-1} FW), averaged over 2 years.

Variant	Free Quercetin	% of Total Quercetin	Bound Quercetin	% of Total Quercetin	Total Quercetin	Rel. %
Cultivar Mundo						
1 (0)	0.065 _{ab}	20	0.265 _{ab}	80	0.329 _a	100
2 (N)	0.063 _{ab}	19	0.263 _{ab}	81	0.327 _a	100
3 (N+S)	0.072 _b	22	0.262 _a	78	0.334 _a	100
4 (N+S+Fe)	0.053 _a	15	0.308 _b	85	0.360 _a	100
LSD _{0.05}	0.013	-	0.044	-	0.048	-
Cultivar Robin						
1 (0)	0.052 _c	17	0.255 _a	83	0.307 _a	100
2 (N)	0.040 _{ab}	12	0.286 _{ab}	88	0.326 _{ab}	100
3 (N+S)	0.048 _{bc}	14	0.306 _b	86	0.354 _b	100
4 (N+S+Fe)	0.038 _a	12	0.265 _a	88	0.303 _a	100
LSD _{0.05}	0.007	-	0.003	-	0.028	-
Cultivar Kamal						
1 (0)	0.030 _a	6	0.434 _a	94	0.464 _a	100
2 (N)	0.043 _b	8	0.477 _b	92	0.520 _{bc}	100
3 (N+S)	0.034 _{ab}	6	0.514 _c	94	0.547 _c	100
4 (N+S+Fe)	0.032 _a	6	0.471 _b	94	0.503 _b	100
LSD _{0.05}	0.010	-	0.031	-	0.033	-
Cultivar Pueblo						
1 (0)	0.008 _b	15	0.005 _b	85	0.006	100
2 (N)	0.005 _a	14	0.003 _a	86	0.004	100
3 (N+S)	0.004 _a	14	0.002 _a	86	0.003	100
4 (N+S+Fe)	0.004 _a	15	0.002 _{ab}	85	0.003	100
LSD _{0.05}	0.0002	-	0.0001	-	0.0001	-

Note: The same letters in the averages of the observed characteristics indicate a statistical difference; LSD = least significant difference, at $p = 95\%$ ($\alpha = 0.05$).

In our experiment, a dosage of sulfur of 19 kg ha^{-1} was not shown to have an impact on the quercetin content. At the same time, an enhanced level of bound quercetin with the combined application of nitrate and sulfate nutrition in red onion varieties was observed.

4. Discussion

Integrated nutrient management practices, such as precision fertilization, nutrient cycling, and agroecological approaches, play a crucial role in promoting sustainable agriculture. Overall, the optimization of N, S, and Fe fertilization in the context of sustainable agriculture represents a critical frontier in agricultural science, with implications for food security, environmental sustainability, and human well-being [26]. The results of the presented research work demonstrated notable increases in the onion yield following the application of nitrogen (N) and sulfur (S) fertilizers. Incorporating iron (Fe) alongside these fertilizers did not yield a significant impact compared to N+S. Al-Fraihat (2009) [9]

demonstrated that the application of $200 \text{ kg N ha}^{-1} + 100 \text{ kg S ha}^{-1}$ resulted in increased total and marketable yields of onion bulbs, specifically of the Giza 20 cultivar. Another study conducted in India revealed that the application of 150 kg N ha^{-1} and 90 kg S ha^{-1} led to significant improvements in the growth, yield attributes, and overall yield of Rabi onion under the conditions of Rajasthan, India [27]. These findings suggest that even higher dosages of nitrogen and sulfur may have a considerable impact on onion yield.

Sulfur is essential for protein synthesis, including the formation of crucial sulfur-containing amino acids like methionine and cysteine, as well as for producing vitamins and chlorophyll [28]. Once considered deficient only in specific soils during the early 1950s, sulfur deficiency has become universal. Additionally, sulfur plays a vital role in activating specific enzymes and serves as a fundamental component of ferredoxin, a molecule critical in photosynthesis. Insufficient sulfur availability compromises plants' ability to achieve maximum yield potential and efficiently utilize applied nitrogen [29].

The application of sulfur modifies the physicochemical properties of soil, improving nutrient availability and facilitating plant growth and development. This enhances nutrient translocation to reproductive organs, boosts photosynthesis, and, consequently, significantly enhances yields and yield components [30,31].

However, excessive fertilizer application leads to the emission of harmful greenhouse gases into the atmosphere, contributing to environmental pollution. Unused fertilizers become pollutants in the air, water, and soil [32]. Moreover, excessive nitrogen fertilizer application can harm *Actinobacteria*, posing a threat to microorganisms in agricultural systems [33]. Hence, an optimal fertilization approach entails the application of 140 kg N ha^{-1} combined with 19 kg of available sulfur per hectare (N+S), as supported by the observed outcomes. These results indicate a notable increase in yield, with improvements of 15% for the Mundo cultivar, 29% for the Robin cultivar, 15% for the Kamal cultivar, and 19% for the Pueblo cultivar.

Numerous studies have examined onion skin as an abundant source of fructooligosaccharides, dietary fibers, polyphenols, and antioxidants [34,35]. Two primary subgroups of flavonoids present in onions are anthocyanins and quercetin and its derivatives. These compounds are responsible for imparting yellow to purple hues to onion skins [36]. A comparative study involving fifteen Indian onion cultivars with diverse skin colors revealed that red-colored varieties exhibit the highest levels of flavonoids and natural antioxidants [37]. It is also noteworthy that phenolic compounds in the black (or red/purple) onion variety remained stable during the digestion process, exhibiting a higher bioaccessibility index [38]. In the presented study, the red onion varieties showed an evident increase in the concentration of bound quercetin with the combined application of nitrate and sulfate.

The red onion varieties Kamal and Robin exhibited the highest flavonoid contents in the N+S-fertilized variant, with values of 0.286 mg g^{-1} and 0.299 mg g^{-1} , respectively. Meanwhile, in the N+S+Fe-fertilized variant, the highest flavonoid content recorded was 0.353 mg g^{-1} , matching that of the N-fertilized variant. Simultaneously, within the N+S-fertilized variant, the white variety Pueblo exhibited a 10% elevation in flavonoid levels compared to the control variant (without fertilization). The study from 2024 highlighted the significant potential of optimizing nitrogen fertilizer sources to enhance the nutritional quality of white short-day onions, encompassing phenolics, flavonoids, and free radical scavenging activity—specifically, the synergistic effect of pre-plant CaCN_2 application (80 kg N ha^{-1}) [39]. However, it is conceivable to suggest that the experimental combination of N+S fertilizers may increase the level of flavonoids in onion varieties, with a more pronounced enhancement observed in red onion varieties.

Fe is involved in the synthesis of chlorophyll and in other enzymatic and metabolic processes without which plants cannot carry out the plant lifecycle [40,41]. Therefore, in the one fertilization variant N+S+Fe of our experiment, we used Fe at a dose of 5 kg ha^{-1} . At the same time, a significant increase in the growth and yield parameters compared to those of the variant N+S was not observed. Previous studies found that the shoot growth decreased and the root length increased in soybean seedlings with increasing Fe application

doses [42]. However, an interactive effect of N and Fe fertilization on the growth and yield of soybean was demonstrated in soil with high bicarbonate and pH. There was a positive interaction between N and Fe at N rates up to 80 kg N ha⁻¹ [3]. We suppose that the interaction between N and Fe fertilization may vary depending on the type of plant, soil, and dosage. For example, it was found that plants of the *Poaceae* family are more susceptible to nanoscale zero-valent iron than *Fabaceae* [43]. It is possible that in our experimental variant, the dose of N was high enough to observe some positive effects of Fe on the growth and qualitative parameters of the experimental onion varieties.

It was shown that sulfur fertilization increased the quercetin and total polyphenol contents, along with the antioxidant activity in experimental onions. The red cultivars statistically significantly exhibited the highest polyphenol and quercetin contents at a sulfur dose of 40 kg·ha⁻¹. The yellow cultivars showed the highest polyphenol contents at a sulfur dose of 50 kg·ha⁻¹. The white cultivars had the lowest polyphenol and quercetin contents, with the highest values observed at a sulfur dose of 40 kg·ha⁻¹ [44]. In our experiment with a dosage of sulfur of 19 kg ha⁻¹, no impact of such fertilization on the quercetin content was shown. At the same time, an enhanced level of bound quercetin with the combined application of nitrate and sulfate nutrition in red onion varieties was observed.

The correct balance of nutrients is particularly important for high sulfur-using crops like canola and alfalfa [45]. Balanced and judicious nutrient management practices for a specific plant object are crucial for promoting sustainable agriculture. Excessive applications of sulfur and nitrogen can lead to environmental pollution. Nitrogen can leach into groundwater, causing contamination, while sulfur can contribute to acid rain formation when released into the atmosphere as sulfur dioxide [46–48]. High doses of sulfur and nitrogen can negatively impact soil biodiversity by favoring certain plant species over others. This can lead to a decline in beneficial soil organisms and a loss of biodiversity in agricultural ecosystems.

This study underscores the critical importance of precise nutrient management tailored to specific plant species, particularly onions (*Allium cepa* L.), for sustainable agricultural practices. Our experiments focused on the effects of nitrogen (NH₄⁺), sulfur (SO₄²⁺), and iron (Fe²⁺) fertilization on secondary metabolites, notably quercetin, across four onion varieties. Over two years, we observed increased onion yields with nitrogen–sulfur (N+S) fertilization, while the addition of iron did not significantly affect the yield compared to N+S fertilization. The phenolic and flavonoid contents varied with fertilization but showed no significant changes in the quercetin content. Effective nutrient management practices are essential for promoting sustainable agriculture while ensuring environmental stewardship. While excessive sulfur and nitrogen application can lead to environmental degradation, judicious fertilization, particularly the synergistic effects of nitrogen and sulfur, enhances the onion yield and nutritional quality. Our findings highlight the need for balanced fertilization strategies tailored to specific crop requirements to mitigate environmental pollution and preserve soil biodiversity. Striking a balance between optimal fertilization and environmental sustainability is paramount for advancing sustainable agriculture and securing long-term agricultural viability.

5. Conclusions

Effective nutrient management practices tailored to specific plant species are imperative for fostering sustainable agricultural systems. The overapplication of sulfur, nitrogen, and iron can result in detrimental environmental consequences, highlighting the importance of optimal dosages for onion cultivars. Our experimentation revealed that sulfur application at a dosage of 19 kg ha⁻¹ did not significantly impact the quercetin content in onions. Conversely, a combined nitrogen and sulfur (N+S) fertilizer approach led to increased levels of bound quercetin, particularly evident in red onion varieties. Notably, the application of N+S fertilizers significantly enhanced the flavonoid levels across all onion cultivars, resulting in notable increases in the yield. However, the addition of iron alongside N+S did not yield significant growth or yield improvements compared to N+S

alone. Our findings underscore the beneficial effects of sulfur in promoting onion growth and development. Furthermore, the interaction between nitrogen and iron fertilization suggested that high nitrogen doses might overshadow the potential benefits of iron for onion growth and quality parameters.

Integrated nutrient management practices are crucial for sustainable agriculture, highlighting the importance of precision fertilization and agroecological approaches. Optimal fertilization strategies are essential for food security, environmental sustainability, and human well-being. Balanced fertilization, particularly the synergistic effects of nitrogen and sulfur fertilization, enhances the onion yield and nutritional quality, contributing to long-term sustainability in onion cultivation.

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