Bošković-Rakočević L *et al.* (2025)
Notulae Botanicae Horti Agrobotanici Cluj-Napoca
Volume 53, Issue 3, Article number 14598
DOI:10.15835/nbha53314598
Research Article

Impact of nitrogen fertilization on raspberry (*Rubus idaeus* L.) fruit quality and soil fertility

Ljiljana BOŠKOVIĆ-RAKOČEVIĆ[1], Ivan GLIŠIĆ[1], Ivana RADOVANOVIĆ[1], Gorica PAUNOVIĆ[1], Jelena MLADENOVIĆ[1], Mira MILINKOVIĆ[2], Radmila ILIĆ[1]*

[1]*University of Kragujevac, Faculty of Agronomy, Cara Dušana 34, 32000, Čačak, Republic of Serbia; ljiljabr@kg.ac.rs; glishoo@yahoo.com; ivanar.al032@gmail.com; gorica@kg.ac.rs; jelenamala@kg.ac.rs; radmila.nikolic@kg.ac.rs (*corresponding author)*
[2]*Institute of Soil Science, Teodora Drajzera 7, 11000, Belgrade, Republic of Serbia; miramilinkovic@yahoo.com*

Abstract

This study investigates the effects of nitrogen fertilization on raspberry fruit quality and soil fertility over two years. The experiment involved the application of complex NPK fertilizers and varying doses of nitrogen fertilizer (CAN) to raspberry plants of 'Willamette' and 'Tulameen' cultivars in the Ivanjica raspberry region. Results indicate a positive correlation between nitrogen doses and soil fertility, with 'Tulameen' soil showing lower nutrient levels compared to 'Willamette'. Increasing nitrogen doses led to higher levels of primary and secondary chemical compounds in raspberry fruit, particularly in the 'Willamette' cultivar. Physical fruit properties were influenced by both variety and nitrogen doses, with significant effects observed in the first year of the study. Proper fertilizer application is crucial for achieving high and stable raspberry yields and fruit quality, while excessive nutrient application may lead to environmental pollution. Based on the comparison of soil fertility parameters and raspberry fruit quality, the recommended fertilizer dosage for raspberry cultivation in the Ivanjica raspberry region is 500 kg ha^{-1} of NPK fertilizer (16:16:16) and 300 kg ha^{-1} of CAN.

Keywords: interactions; nitrogen fertilization; optimization; raspberry cultivars

Introduction

Due to its biological and pomological characteristics, as well as its nutritional value, the raspberry (*Rubus idaeus* L.) belongs to the group of fruit species that are highly represented in agricultural production. Thanks to modern production technology, the establishment of new raspberry orchards, and the development of modern processing capacities, raspberries are increasingly gaining importance in both global and domestic markets.

Received: 22 May 2025. Received in revised form: 07 Sep 2025. Accepted: 17 Sep 2025. Published online: 23 Sep 2025.
From Volume 49, Issue 1, 2021, Notulae Botanicae Horti Agrobotanici Cluj-Napoca journal uses article numbers in place of the traditional method of continuous pagination through the volume. The journal will continue to appear quarterly, as before, with four annual numbers.

Serbia is among the leading global producers and exporters of frozen raspberries, ranking third worldwide with a production of 940,979.29 tons (FAOSTAT, 2025). The 'Willamette' cultivar dominates production areas, while the 'Tulameen' cultivar is increasingly planted for fresh consumption.

Raspberries grow best on slightly acidic, humus-rich soils, while calcareous and alkaline soils are less suitable (Ubavić *et al*., 2016). Proper fertilization is essential for achieving high yields and quality fruit. Nitrogen fertilization, in particular, plays a crucial role in raspberry production, but excessive use of fertilizers can negatively affect both crop quality and the environment.

Agricultural producers often use larger quantities of fertilizers than necessary, negatively affecting the growth and development of raspberries, and consequently, increasing environmental pollution. According to Ubavić *et al*. (2016), due to its biological specificity, it is necessary to know the quantities of biogenic elements added in different stages of raspberry development, as well as their mutual relationship. For this reason, soil agrochemical analysis and chemical analysis of plant parts are necessary to determine the required quantities of fertilizers and thereby achieve high yield and good fruit quality.

Although the general importance of fertilization in raspberry cultivation is well known, there is limited information on how different nitrogen doses affect both soil fertility and fruit quality in key raspberry cultivars under local growing conditions. Therefore, the aim of this study is to determine the effect of applying nitrogen fertilizer CAN on changes in soil and fruit quality of the raspberry cultivars 'Willamette' and 'Tulameen'.

Materials and Methods

Study site and plant material

The experimental part of the research was conducted during two raspberry growing seasons, 2020 and 2021, in two raspberry orchards of the 'Willamette' and 'Tulameen' cultivars in Serbia, the municipality of Ivanjica, in the village of Đopići (43° 25′ 15″ N; 20° 22′ 27″ E; 929 m above sea level).

The distance between the plots were about 1 km. The area planted with the 'Willamette' cultivar was 6 ares, and with the 'Tulameen' cultivar - 1.5 ares. Both orchards were raised in a system of trellises, i.e. hedges. The 'Willamette' orchard was established in 2017, while the 'Tulameen' orchard was established in 2016.

Classic agro-technical and pomotechnical measures were applied in the orchards, including soil cultivation, fertilization, and pruning, as well as disease and pest control. The orchards were not equipped with irrigation systems and relied solely on rainfall.

In both years, the research was conducted on two raspberry cultivars, 'Willamette' and 'Tulameen'.

Experimental design and fertilization treatments

The trial variants consist of several combinations. For both cultivars, there's a control variant with no treatment, along with two treated variants: one with 45 g of calcium ammonium nitrate (CAN) per trial plot, which translates to 300 kg ha^{-1}, and another with 75 g of calcium ammonium nitrate (CAN) per trial plot, equivalent to 500 kg ha^{-1}.

Before setting up the trials in both growing seasons, soil agrochemical analysis (non-fertilized variant) was performed, after which the plots were fertilized with mineral NPK fertilizer 16:16:16 (500 kg ha^{-1}). Application of mineral fertilizer CAN (27% N) was carried out in both growing seasons at the beginning of the flowering phase, namely: June 1, 2020, and May 30, 2021, for the 'Willamette' cultivar, and June 10, 2020, and June 8, 2021, for the 'Tulameen' cultivar, due to differences in the onset of the flowering phase for these two cultivars in both growing seasons. After raspberry harvesting, soil samples were taken from each fertilized variant, and soil agrochemical analysis was performed.

Soil sampling and analysis

Soil sampling procedures were conducted both before initiating the trials and upon their completion, following a diagonal pattern at a depth of 30 cm, utilizing shovels. Six individual samples were collected in each orchard and combined to form an average soil sample, which underwent various analyses. These analyses included measuring soil pH (pH/H2O) potentiometrically in a soil-water solution (1:2.5 ratio), soil acidity (pH/KCl) potentiometrically in a soil-KCl solution (1:2.5 ratio), humus content determined by the Kotzman method, and phosphorus and potassium content analyzed via the Egner-Riehm method (P_2O_5 measured colorimetrically, K_2O assessed flame photometrically). Additionally, the levels of ammonium (NH_4-N) and nitrate (NO_3-N) nitrogen content were determined using the Bremner method. These soil agrochemical analyses were carried out at the laboratories of both the Faculty of Agronomy in Čačak and the Soil Research Institute in Belgrade.

Fruit sampling and analysis

During the technological ripeness phase of raspberries, 30 fruits (three replicates per ten fruits) were sampled from each trial variant per cultivar. These samples underwent assessment for various fruit quality parameters, including dry matter content, water content, organic acid content measured volumetrically, vitamin C content determined by the Tillmans method, total phenols measured by the Folin-Ciocalteu method, total flavonoids assessed through a method based on their ability to complex metal cations, and total antioxidant capacity measured using the phosphomolybdenum method.

At the end of both growing seasons, fruit mass (g) and fruit dimensions - width and length (cm) were measured. Fruit mass measurement was performed using a digital scale with a measurement range of 5,000 g. Fruit dimensions were measured using a digital caliper Starrett 727 (Athol,NE, USA).

Statistical analysis

The statistical differences between the experimental factors were verified using two-way ANOVA. The source of variation was: two cultivars (A) and three different fertilizer treatments (B). When the F test was significant, all means, including control values were compared with the LSD test at $P \leq 0.05$. Statistical data processing was performed using Microsoft Office Excel Software (Microsoft Corporation, Redmond, WA, USA).

Results and Discussion

Soil agrochemical analysis

Soil pH has a significant influence on the availability of nutrients to plants. The nutrients that are essential for plant growth, such as nitrogen, phosphorus, and potassium, are most available to plants at a soil pH of between 6.0 and 7.0. When the soil pH is too low or too high, the availability of these nutrients decreases and plants may suffer from nutrient deficiencies (Zhang *et al*., 2016).

Soil samples for agrochemical analysis were taken on two occasions, before the experiment setup and after the raspberry harvest, for both examined cultivars, in both growing seasons. The examined parameters of soil agrochemical analysis indicate variations among the soils in the raspberry orchards (Table 1). Before fertilization, the soil's exchangeable acidity on the plot with the 'Tulameen' cultivar in both growing seasons was acidic (pH 5.15 and pH 5.16). With the application of mineral fertilizer CAN at a dosage of 45 g, this value increased in both growing seasons to 5.48 in the first year of research and 5.94 in the second year. Values with 75 g of CAN were approximately in both years of examination (pH 6.00 and pH 5.91).

The control variants for the 'Willamette' cultivar indicate slightly acidic soil reaction (pH 5.57 and pH 6.03). In the experimental variant with 45 g of CAN, pH values slightly decreased in both years of research (pH

5.45 and pH 5.65), which may be associated with greater soil leaching. With 75 g of CAN, pH values increased in the first year compared to the control and the variant with a lower fertilizer dose.

Table 1. Basic agrochemical characteristics of the soil in two experimental orchards

Year	2020					2021				
Variant	pH		Humus	P_2O_5	K_2O	pH		Humus	P_2O_5	K_2O
	H_2O	KCl	(%)	mg 100 g^{-1}		H_2O	KCl	(%)	mg 100 g^{-1}	
'Tulameen'	6.23	5.15	2.95	0.48	13.30	6.09	5.16	3.70	1.70	20.0
'Tulameen' (45 g of CAN)	6.42	5.48	3.71	4.53	37.70	6.57	5.94	4.79	26.30	55.10
'Tulameen' (75 g of CAN)	6.76	6.00	4.90	25.70	75.10	6.58	5.91	4.82	23.10	45.60
'Willamette'	6.37	5.57	4.89	27.40	71.10	6.58	6.03	5.34	70.0	80.10
'Willamette' (45 g of CAN)	6.02	5.45	5.55	93.60	63.80	6.15	5.65	5.76	77.3	79.80
'Willamette' (75 g of CAN)	6.41	5.75	6.04	107.00	70.70	5.66	5.14	6.17	90.5	81.60

Our results showed that higher nitrogen doses significantly increased soil fertility and nutrient availability. Similar findings were reported by Zhang *et al*. (2025), who demonstrated across multiple studies that balanced nitrogen fertilization enhances soil nutrient status while maintaining long-term soil health.

Intensive agriculture can speed up soil acidification through many processes - increasing leaching, addition of fertilizers, removal of produce, and build-up of soil organic matter. Of all the major fertilizer nutrients, nitrogen is the main nutrient affecting soil pH (Purbasha *et al*., 2020), and soils can become more acidic or more alkaline depending on the type of nitrogen fertilizer used. This author also mentioned that soil acidification due to the use of phosphorus fertilizers is small compared to that attributed to nitrogen, due to the lower amounts of this nutrient used and the lower acidification per kg phosphorus and potassium fertilizers have little or no effect on soil pH.

According to the results for humus content (Table 1) in both years of examination, the plot planted with the 'Willamette' cultivar was better supplied with humus compared to the plot under the 'Tulameen' cultivar. The application of increasing doses of fertilizer resulted in an increase in humus content in both years of examination. Comparing the results with data provided by Milošević (1997), Petrović and Milošević (2002), Ubavić *et al*. (2016), and Petrović *et al*. (2020) both plots are fertile and meet raspberry requirements for humus, yet the soil where the 'Willamette' cultivar is planted is more fertile.

In both growing seasons, the content of available phosphorus and potassium significantly differed between the examined plots. On the control variants with the 'Tulameen' cultivar, phosphorus content was below optimal values i.e., below 8-10 mg 100 g^{-1} of soil (Ubavić *et al*., 2016).

Potassium content in the first year of research was also below optimal values (18-20 mg 100 g^{-1} of soil, Ubavić *et al*., 2016), while in the second year of research, it was at an optimal level.

For the 'Willamette' cultivar, both available phosphorus and potassium in both years of examination were above the literature values (Ubavić *et al*., 2016). The application of mineral NPK fertilizers increased the content of available phosphorus and potassium.

Earlier, Sistrunk (1963) reported that the quality of berries is influenced by climatic conditions and soil fertility. Hence, only fertile soil with a regulated pH value and an optimal nutrient and water supply ensures desirable internal quality parameters (Buskiene and Uselis, 2008). For example, the high water availability in the soil increases photosynthesis and carbohydrate accumulation in the leaves of 'Meeker' (Morales *et al*., 2013).

The analysis of nitrogen content (NH_4-N, NO_3-N, and N min) in soil samples (Table 2) revealed significant differences among cultivars and applied fertilizer levels in both years of the study. In 2020, the mean

NH_4-N values were higher in the 'Willamette' than in the 'Tulameen' cultivar. Mean NO_3-N values were higher in the 'Willamette' cultivar compared to the 'Tulameen' cultivar. Regarding Sum N min, the 'Willamette' cultivar exhibited higher values compared to the 'Tulameen' cultivar. In 2021, similar trends were observed, with mean NH_4-N and NO_3-N values being higher in the 'Willamette' cultivar compared to Tulameen. Additionally, the application of different fertilizer levels also significantly influenced nitrogen content in the soil. For instance, in 2020, the application of 75 g CAN resulted in the highest mean NH4-N and NO_3-N values. The control variant pointed the lowest values.

Table 2. Mineral nitrogen content in the soil

Year	Source of variation		Nitrogen content		
			NH_4-N (mg kg^{-1})	NO_3-N (mg kg^{-1})	Sum N min
2020	Cultivar (A)	'Willamette'	26.87 ± 2.15 a	29.20 ± 1.98 a	56.07 ± 5.44 a
		'Tulameen'	12.17 ± 3.05 b	12.25 ± 1.70 b	24.42 ± 3.12 b
	Fertilizer (B)	Control	17.33 ± 2.14 b	16.33 ± 2.01 b	33.67 ± 3.13 b
		45 g CAN	16.33 ± 1.89 b	15.43 ± 1.88 b	31.76 ± 3.44 b
		75 g CAN	21.27 ± 3.13 a	26.30 ± 2.67 a	47.57 ± 4.18 a
2021	Cultivar (A)	'Willamette'	25.20 ± 3.56 a	31.40 ± 2.75 a	56.60 ± 5.14 a
		'Tulameen'	17.17 ± 1.89 b	12.25 ± 2.54 b	29.42 ± 3.11 b
	Fertilizer (B)	Control	15.33 ± 1.99 c	12.25 ± 1.66 c	27.58 ± 3.02 c
		45 g CAN	19.87 ± 2.55 b	23.40 ± 2.08 b	43.27 ± 3.78 b
		75 g CAN	21.27 ± 2.69 a	26.30 ± 3.01 a	47.57 ± 3.66 a
ANOVA 0.05		(A) Cultivar	*	*	*
		(B) Fertilizer	*	*	*
		A x B	*	*	*

Columns means for cultivar, fertilization, and cultivar/fertilization interaction marked with different lowercase letters are significantly different at the P ≤0.05 level based on the LSD test. *- F test significant at P ≤0.05 level; ns - F test not significant

The percentage share of N-NH_4 and N-NO_3 in mineral nitrogen is important in the ecological sense (Zhang *et al*., 2012). Mazur and Mazur (2015) mentioned that besides the type of applied fertilizer, the agronomical properties of soil are also important factors that influenced the content of these two types of nitrogen.

Physical Properties of the Fruit

Among the pomological traits of fruit species, fruit weight is of the greatest importance as an indicator of quality. It is defined as a quantitative hereditary trait that determines the appearance and yield of the fruit, as well as consumer acceptability (Crisosto *et al*., 2004). According to Mratinić (2015) and Eyduran and Agaoglu (2006), this trait depends on cultivar, growing conditions, and the cultivation practices applied in the orchard.

The fruit characteristics of 'Willamette' and 'Tulameen' raspberry cultivars were examined over the years 2020 and 2021, focusing on fruit mass and dimensions (Table 3). In general, 'Tulameen' raspberries exhibited larger fruit dimensions compared to 'Willamette'.

Table 3. The fruit mass and fruit dimensions of 'Willamette' and 'Tulameen' cultivars in 2020 and 2021

Year	Source of variation		Fruit characteristics		
			Fruit mass (g)	Fruit length (cm)	Fruit width (cm)
2020	Cultivar (A)	'Willamette'	3.83 ± 2.11 b	2.72 ± 1.10 a	2.12 ± 0.90 a
		'Tulameen'	5.08 ± 2.99 a	2.62 ± 1.88 b	2.11 ± 1.01 a
	Fertilizer (B)	Control	4.17 ± 2.15 b	2.37 ± 1.15 b	2.10 ±0.98 a
		45 g CAN	4.22 ± 3.01 b	2.46 ± 1.13 a	2.13 ± 0.57 a
		75 g CAN	4.98 ± 2.87 a	2.51 ± 1.55 a	2.12 ± 0.65 a
2021	Cultivar (A)	'Willamette'	4.09 ± 2.15 b	1.89 ± 0.98 b	1.64 ± 0.51 b
		'Tulameen'	4.63 ± 2.18 a	2.10 ± 0.58 a	1.73 ± 0.52 a
	Fertilizer (B)	Control	4.23 ± 2.65 a	1.98 ± 0.68 a	1.68 ± 0.50 a
		45 g CAN	4.38 ± 2.26 a	1.99 ± 1.11 a	1.71 ± 0.65 a
		75 g CAN	4.47 ± 2.38 a	2.01 ± 1.02 a	1.66 ± 0.41 a
ANOVA 0.05		Cultivar (A)	*	*	*
		Fertilizer (B)	*	*	ns
		A x B	*	*	ns

Column means for cultivar, fertilization, and cultivar/fertilization interaction marked with different lowercase letters are significantly different at the P ≤0.05 level based on the LSD test. *- F test significant at P ≤0.05 level; ns - F test not significant

Across different fertilization treatments, there were variations observed. For example, in 2020, fruit mass was highest in the 75 g CAN treatment for both 'Willamette' and 'Tulameen' cultivars, while in 'Tulameen' raspberries showed the highest fruit mass in all fertilization treatments. Regarding fruit length and width, similar trends were observed, with slight variations depending on the cultivar and fertilization treatment. Statistical analysis revealed significant differences between cultivars for all fruit characteristics in both years, while fertilization treatments showed significant differences for some fruit characteristics, especially in fruit mass. Additionally, there were significant interactions between cultivar and fertilization treatment for certain fruit characteristics, indicating that the effects of fertilization treatments varied depending on the cultivar. Overall, these findings suggest that both cultivar and fertilization play significant roles in determining fruit characteristics in raspberry cultivation, highlighting the importance of careful selection and management practices for optimal fruit production.

Our findings align with those of Sawicka *et al*. (2023), who reported that the application of 135 kg N ha^{-1} significantly increased raspberry yield and fruit weight while maintaining adequate fruit firmness and resistance to mechanical damage. Importantly, their results highlighted that balanced nitrogen fertilization improves fruit quality and postharvest characteristics without leading to excessive vegetative growth or reduced fruit storability. Similar to our study, they observed cultivar-specific responses, with some cultivars benefiting more from higher nitrogen doses in terms of yield, while others maintained better dry matter content and earlier ripening.

Table 4 presents the mechanical characteristics of raspberry fruits, including arithmetic diameter (Da), geometric diameter (Dg), sphericity (Φ), elongation (Ra), and surface area (S), as influenced by cultivar and fertilization treatments across two growing seasons. While no significant differences were found between 'Willamette' and 'Tulameen' for Da, Dg, and Φ, significant differences in elongation (Ra) were observed, particularly in 2020, with 'Tulameen' exhibiting higher values than 'Willamette' and vice versa in 2021. This suggests that 'Tulameen' fruits tend to be less elongated in 2020 than in 2021.

Table 4. Mechanical characteristics of raspberry fruits

Year	Source of variation		Fruit characteristics				
			Da	**Dg**	**Φ**	**Ra**	**S**
2020	Cultivar (A)	'Willamette'	2.42 ± 1.65 a	2.30 ± 0.03 a	0.84 ±1.64 a	77.94 ± 0.16 b	16.66 ± 0.15 a
		'Tulameen'	2.36 ± 1.98 a	2.26 ± 0.05 a	0.86 ± 1.15 a	80.53 ± 0.14 a	16.15 ± 0.09 a
	Fertilizer (A)	Control	2.23 ± 0.01 a	2.18± 0.03 a	0.92 ± 0.01 a	88.60 ± 0.81 a	15.01 ± 0.01 a
		45 g CAN	2.29 ± 0.02 a	2.23± 0.01 a	0.90 ± 0.02 a	86.58 ± 0.99 ab	15.68 ± 0.01 a
		75 g CAN	2.31 ± 0.01 a	2.24± 0.01 a	0.89 ± 0.01 a	84.46 ± 0.62 b	15.79 ± 0.01 a
2021	Cultivar (A)	'Willamette'	1.76 + 1.11 a	1.71 ± 0.11 a	0.90 ± 0.05 a	86.77 ± 0.22 a	9.28 ± 0.22 a
		'Tulameen'	1.91 ± 1.15 a	1.84 ± 0.08 a	0.87 ± 0.07 a	82.30 ± 0.99 b	10.69 ± 0.30 a
	Fertilizer (B)	Control	1.83 ± 0.02 a	1.77 ± 0.05 a	0.89 ± 0.06 a	84.84 ± 0.71 a	9.88 ± 0.15 a
		45 g CAN	1.85 ± 0.04 a	1.79 ± 0.06 a	0.90 ± 0.04 a	85.92 ± 0.72 a	10.15 ± 0.11 a
		75 g CAN	1.83 ± 0.03 a	1.76 ± 0.06 a	0.88 ± 0.04 a	82.58 ± 0.61 b	9.82 ± 0.21 a
ANOVA 0.05		Cultivar (A)	ns	ns	ns	*	ns
		Fertilizer (B)	ns	ns	ns	*	ns
		A x B	ns	ns	ns	ns	ns

Column means for cultivar, fertilization, and cultivar/fertilization interaction marked with different lowercase letters are significantly different at the P ≤0.05 level based on the LSD test. *- F test significant at P ≤0.05 level; ns - F test not significant. Da: Mean arithmetic fruit diameter; Dg: Mean geometric fruit diameter; ϕ: Sphericity; Ra: Fruit weight/length ratio; S: Fruit surface area

Fertilization also did not have a significant influence on all mechanical traits except Ra. Although the differences between fertilizer treatments were generally small, the control variant consistently showed the highest values for elongation (Ra), especially in 2020, while 75 g CAN treatment often resulted in reduced Ra values. This trend may indicate a slight compacting effect of higher nitrogen application on fruit shape.

Interaction effects between cultivar and fertilizer (A × B) were not statistically significant for any of the traits, suggesting that the response to fertilization was similar across both cultivars.

The shape of a fruit is usually expressed in terms of its sphericity. It is an important trait used in fluid flow and heat and mass transfer calculations. Fruits are developed in accordance with biological and genetic principles (dynamics of cell division and elongation) (Jennings, 1971) that can be channelled by weather conditions, especially in the last days of maturation. Namely, berry development at this time depends on adequate supplies of carbohydrates and water; any limitation will adversely affect fruit size and shape (Harris *et al.*, 1968). In our study, berry sphericity is lower than 1 (100%) and could be assumed as oval (Habibeh *et al.*, 2011).

Chemical fruit characteristics

Table 5 shows the content of primary chemical compounds in raspberry fruits-total soluble solids (TSS), titratable acidity (TA), ripening index (TSS/TA), and vitamin C content-as influenced by cultivar and nitrogen fertilization over two growing seasons.

Significant differences were observed between the two cultivars in all examined parameters in both years. 'Tulameen' consistently exhibited higher TSS and ripening index values compared to 'Willamette', indicating sweeter fruits with a more favorable sugar-acid balance. In contrast, 'Willamette' had significantly higher titratable acidity and vitamin C content. However, the vitamin C content for both cultivars, in both years of the study, increased with higher doses of nitrogen fertilizer, which is consistent with the findings reported by Ali (2012). For both growing seasons, the vitamin C content was within or slightly above the values reported by Stojanov (2019) in his research.

Table 5. Content of primary chemical compounds in raspberry fruit

Year	Source of variation		TSS (%)	TA (%)	Ripening index	Vitamin C (mg 100 g^{-1})
2020	Cultivar (A)	'Willamette'	11.00 ± 0.17 b	1.30 ± 0.02 a	10.61 ± 1.22 b	24.55 ± 2.12 a
		'Tulameen'	14.55 ± 0.16 a	0.79 ± 0.02 b	18.43 ± 1.18 a	21.65 ± 1.98 b
	Fertilization (B)	Control	10.93 ± 0.15 c	0.64 ± 0.01 c	16.99 ± 1.21 a	20.78 ± 2.01 c
		45 g CAN	12.63 ± 0.16 b	0.93 ± 0.01 b	13.82 ± 1.24 b	23.13 ± 1.98 b
		75 g CAN	14.78 ± 0.18 a	1.32 ± 0.02 a	12.75 ± 1.00 c	25.40 ± 2.00 a
2021	Cultivar (A)	'Willamette'	12.32 ± 1.12 b	1.50 ± 0.07 a	8.46 ± 1.04 b	27.67 ± 2.43 a
		'Tulameen'	15.28 ± 1. 23 a	0.77 ± 0.03 b	17.96 ± 1.19 a	23.30 ± 2.38 b
	Fertilization (B)	Control	11.53 ± 1.16 c	0.90 ± 0.05 c	11.11 ± 1.01 b	21.50 ± 2.12 c
		45 g CAN	13.80 ± 1.14 b	1.13 ± 0.07 b	14.32 ± 1.07 a	26.00 ± 2.43 b
		75 g CAN	16.08 ± 1.15 a	1.38 ± 0.08 a	14.21 ±1.08 a	28.95 ± 2.51 a
ANOVA 0.05		Cultivar (A)	*	*	*	*
		Fertilizer (B)	*	*	*	*
		(A) x (B)	ns	ns	ns	ns

Column means for cultivar, fertilization, and cultivar/fertilization interaction marked with different lowercase letters are significantly different at the P ≤0.05 level based on the LSD test. *- F test significant at P ≤0.05 level; ns - F test not significant. TSS: Soluble solids content; TA: Titratable acidity

The obtained results for TSS are within the range reported by Petrović and Milošević (2002) and Veličković (2007), and are somewhat higher than the values obtained in the study by Daubeny and Anderson (1991). It seems that geographic region i.e. site with specific pedo-climatic conditions also play important role in the biosynthesis of above compounds.

Fertilization significantly affected all chemical parameters. The highest TSS and vitamin C content were recorded with the 75 g CAN treatment in both years, suggesting that higher nitrogen rates can enhance TSS accumulation and ascorbic acid synthesis. However, this treatment also led to increased acidity, particularly in 2020, which reduced the ripening index compared to lower nitrogen rates. Interestingly, the control treatment showed the lowest TSS and vitamin C values, highlighting the role of nitrogen in improving fruit quality traits.

Non-significant interaction was found between cultivar and fertilizer (A × B) for any of the chemical traits, implying that both cultivars responded similarly to the applied nitrogen treatments.

However, some researchers have found that fertilizers containing N (Ballinger and Kushman, 1969) increased TA content and pH juice (Alleyne and Clark, 1997) in berries, whereas N fertilization decreased SSC in red raspberry fruit (Papp *et al*.,1984). In addition, total acid content in raspberries is related to cultivar, location and agricultural practice. Hence, data from relevant literature, confirm the major effect of genetic, physiological, climatic and production factors on acidity of raspberries (Orhan *et al*., 2006; Morales *et al*., 2013).

The specific nature of the response of different berry species and their cultivars to fertilization management in different sites and soil types was previously reported (Hargreaves *et al*., 2008; Ali, 2012).

Table 6 summarizes the content of secondary chemical compounds in raspberry fruits-total phenols, total flavonoids, and antioxidant capacity (measured by ABTS assay)-as influenced by cultivar and nitrogen fertilization in two consecutive years.

Table 6. Content of secondary chemical compounds in raspberry fruit

Year	Source of variation		Fruit characteristics		
			Total phenols (mg GAE 100 g^{-1})	Total flavonoids (mg QE 100 g^{-1})	ABTS (μg AA g^{-1})
2020	Cultivar (A)	'Willamette'	416.32 ± 24.33 a	204.07 ± 18.91a	121.11 ± 16.22 a
		'Tulameen'	402.22 ± 22.46 b	198.65 ± 17.53 b	113.68 ± 15.78 b
	Fertilizer (B)	Control	405.3 ± 29.12 c	200.3 ± 19.09 c	116.55 ± 12.34 c
		45 g CAN	406.76 ± 19.99 b	201.53 ± 18.78 b	117.6 ± 12.01 b
		75 g CAN	415.76 ± 21.03 a	202.25 ± 18.99 a	118.05 ± 11.98 a
2021	Cultivar (A)	'Willamette'	431.2 ± 25.62 a	184.2 ± 16.43 a	139.33 ± 14.76 a
		'Tulameen'	413.6 ± 24.91 b	133.13 ± 15.82 b	119.96 ± 15.04 b
	Fertilizer (B)	Control	408.3 ± 21.19 c	145.7 ± 14.41 c	125.5 ± 13.09 c
		45 g CAN	421.25 ± 27.32 b	161.35 ± 15.04 b	127.95 ± 13.68 b
		75 g CAN	437.65 ± 30.05 a	168.95 ± 15.87 a	135.5 ± 14.05 a
ANOVA 0.05		Cultivar (A)	*	*	*
		Fertilizer (B)	*	*	*
		A x B	ns	ns	ns

Column means for cultivar, fertilization, and cultivar/fertilization interaction marked with different lowercase letters are significantly different at the P ≤0.05 level based on the LSD test. *- F test significant at P ≤0.05 level; ns - F test not significant

Statistical analysis revealed significant differences between cultivars for all measured parameters. In both years, 'Willamette' had higher total phenol and flavonoid contents, as well as greater antioxidant activity, compared to 'Tulameen'. This indicates a stronger antioxidant profile in 'Willamette', which may be of interest from a nutritional and functional food perspective.

Fertilization had a significant effect on all parameters as well. The highest concentrations of phenols, flavonoids, and antioxidant activity were consistently observed in the 75 g CAN treatment, followed by the 45 g CAN treatment, while the control had the lowest values. These results suggest that increasing nitrogen availability positively influences the biosynthesis of bioactive compounds in raspberry fruits, likely due to improved plant vigour and metabolic activity. However, the values of total phenolic content were consistent with the previous findings of Glišić *et al.* (2018).

Our findings are in line with recent studies reporting that organic fertilizers enhance fruit weight and antioxidant activity, whereas mineral fertilizers may increase phenolic compounds and acidity (Valentinuzzi *et al.*, 2018; Frias-Moreno *et al.*, 2021). Combining organic and mineral sources could therefore optimize both yield and fruit quality while supporting soil sustainability.

There were non-significant interactions between cultivar and fertilization (A × B), meaning that both cultivars responded similarly to nitrogen application in terms of secondary metabolite accumulation.

The results (Table 6) indicate that the total phenolic content differed between the studied cultivars, confirming previous findings by Ali (2012), who established that total phenolic content depends on the cultivar, climatic conditions, and applied agrotechnical practices. The same author found that increasing the amounts of applied nitrogen and potassium led to higher total phenolic content, which was also confirmed in this study, as the total phenolic content increased with higher nitrogen doses. While "Willamette" stands out for its naturally higher levels of secondary metabolites, increasing nitrogen input, particularly to 75 g CAN, further enhances the functional quality of the fruit (Table 6). In contrast, excessive N doses are also reported to decrease some of the antioxidant contents of crops, probably due to rapid plant growth and development, and thus preferential allocation of resources are directed to growth processes rather than secondary metabolism (Mitchell *et al.*, 2007). Obviously, fertilizers containing higher amounts N, have caused the largest content of bioactive compounds in this study.

The values of total flavonoid content in 2020 were within the range reported by Glišić *et al*. (2018), while the values in 2021 were lower, most likely due to weather conditions. An increase in the dose of nitrogen fertilizers led to higher total flavonoid content, which also confirmed the previous findings of Ali (2012). The total antioxidant capacity depends on numerous factors, among which the most significant are the cultivar and applied agrotechnical practices, primarily the use of organic and mineral fertilizers (Moore *et al*., 2008). The influence of cultivar on antioxidant capacity was previously established by Glišić *et al*. (2018), who, by examining the same cultivars, also found lower antioxidant capacity in the 'Tulameen' cultivar compared to 'Willamette'.

As known, quality factors more or less genetically controlled, as the level of the bioactive compounds vary according to cultivar (Dale and Daubeny, 1985). Besides cultivar, many factors can affect the total antioxidant capacity of fruit such as cultivation site and technique (i.e. inorganic fertilizers or organic manure), rainfall, mean growing temperature (Hargreaves *et al*., 2008; Castilho Maro *et al*., 2013) and light levels (Atkinson *et al*., 2006).

Conclusions

This two-year study demonstrates that nitrogen fertilization strongly influences both soil fertility and raspberry fruit quality. Higher nitrogen doses, particularly 75 g CAN per plot (equivalent to 500 kg ha^{-1}), improved soil nutrient availability and enhanced fruit quality traits, especially in the 'Willamette' cultivar. Both cultivars benefited from fertilization in terms of fruit size, with 'Tulameen' producing larger fruits overall. However, these findings also highlight the risk of over-fertilization, which can lead to environmental degradation and nutrient imbalances. For sustainable raspberry production, growers should carefully balance nitrogen inputs to optimize yield and quality while safeguarding soil health and environmental integrity.

Authors' Contributions

Conceptualization: LBR, JM, MM; Data curation: LBR, IR; Formal analysis: LBR, IG, IR, MM, RI; Funding acquisition: LBR, JM, MM, IG, IR, RI, GP; Investigation: LBR, IG, IR, GP, JM, RI, MM; Methodology: LBR, IG, GP, RI; Software: LBR, IR, RI; Supervision: JM, MM; Validation: LBR, IR, RI; Roles/Writing - original draft: LBR, IG, GP, RI; and Writing - review & editing: LBR, RI.

All authors read and approved the final manuscript.

Acknowledgements

This work is supported by the Ministry of Science, Technological Development and Innovation of Republic of Serbia (grants 451-03-136/2025-03/200088 and 451-03-136/2025-03/200011). We hereby express our sincerest gratitude to them for their financial support.

Conflict of Interests

The authors declare that there are no conflicts of interest related to this article.

The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript, or in the decision to publish the results.

References

Ali L (2012). Pre-harvest factors affecting quality and shelf-life in raspberries and blackberries (*Rubus* spp. L.). PhDThesis. Swedish University of Agricultural Sciences, Alnarp.

Alleyne V, Clark JR (1997). Fruit composition of 'Arapaho' blackberry following nitrogen fertilization. HortScience 32(2):282-283. *https://doi.org/10.21273/hortsci.32.2.282*

Atkinson CJ, Dodds PAA, Ford YY, Le Miére J, Taylor JM, Blake PS, Paul N (2006). Effects of cultivar, fruit number and reflected photosynthetically active radiation on *Fragaria* × *ananassa* productivity and fruit ellagic acid and ascorbic acid concentration. Annals of Botany 97(3):429-441. *https://doi.org/10.1093/aob/mcj046*

Ballinger WE, Kushman LJ (1969). Relationship of nutrition and fruit quality of Wolcott blueberries grown in sand culture. Journal of the American Society for Horticultural Science 94:329-335. *https://doi.org/10.21273/jashs.94.3.329*

Buskiene L, Uselis N (2008). The influence of nitrogen and potassium fertilizers on the growth and yield of raspberries cv. Polana. Agricultural Research 6(1):27-35.

Castilho Maro LA, Pio R, Santos Guedes MN, Patto de Abreu CM, Nogueira Curi P (2013). Bioactive compounds, antioxidant activity and mineral composition of fruits of raspberry cultivars grown in subtropical areas in Brazil. Fruits 68(3):209-217. *https://doi.org/10.1051/fruits/2013068*

Crisosto CH, Garner D, Andris HL, Day KR (2004). Controlled delayed cooling extends peach market life. HortTechnology 14:99-104. *https://doi.org/10.21273/HORTTECH.14.1.0099*

Dale A, Daubeny HA (1985). Genotype-environmental interaction involving British and Pacific Northwest red raspberry cultivars. HortScience 20(1):68-69. *https://doi.org/10.21273/hortsci.20.1.68*

Daubeny HA (1996). Brambles. In: Janick J, Moore JN (Eds). Fruit Breeding, Volume II, Vine and Small Fruits. John Wiley & Sons, Inc, New York, NY pp. 109-190.

Daubeny HA, Anderson A (1991). 'Tulameen' red raspberry. HortScience 26(10):1336-1338. *https://doi.org/10.21273/HORTSCI.26.10.1336*

Eyduran SP, Agaoglu YS (2006). A preliminary examination of ten raspberry cultivars. Research Journal of Agriculture and Biological Science 2(6):375-379.

FAOSTAT (2025). Food and Agriculture Organization of the United Nations. Retrieved 2025 February 15 from *https://www.fao.org/faostat/en/#data*

Frias-Moreno MN, Parra-Quezada RA, González-Aguilar G, Ruíz-Canizales J, Molina-Corral FJ, Sepulveda DR, Salas-Salazar N, Olivas GI (2021). Quality, bioactive compounds, antioxidant capacity, and enzymes of raspberries at different maturity stages, effects of organic vs. conventional fertilization. Foods 10(953):1-15. *https://doi.org/10.3390/foods10050953*

Glišić I, Milošević T, Paunović G, Vukosavljević V, Ilić R, Mladenović J, Mašković P, Nikolić M (2018). Kvalitet i zdravstvena ispravnost voća i grožđa [Quality and health safety of fruits and grapes]. Agronomski fakultet, Čačak pp 25-27.

Habibeh N, Sadegh S, Jafar H, Mohammad A (2011). Some post-harvest properties of Iranian genotype of raspberry (*Rubus ideaus* L.). Australian Journal of Agricultural Engineering 2(6):155-159.

Hargreaves J, Sina Adl M, Warman PR, Vasantha Rupasinghe HP (2008). The effects of organic amendments on mineral element uptake and fruit quality of raspberries. Plant Soil 308:13-226. *https://doi.org/10.1007/s11104-008-9621-5*

Harris JM, Kriedemann PE, Possingham JV (1968). Anatomical aspects of grape berry development. Vitis 7(2):106-109.

Jennings DL (1971). Some genetic factors affecting fruit development in raspberries. New Phytologist 70:361-370. *https://doi.org/10.1111/j.1469-8137.1971.tb02534.x*

Mazur Z, Mazur T (2015). Effects of long-term organic and mineral fertilizer applications on soil nitrogen content. Polish Journal of Environmental Studies 24(5):2073-2078. *https://doi.org/10.15244/pjoes/42297*

Milošević T (1997). Specijalno voćarstvo [Special fruit growing]. Agronomski fakultet i Zajednica za voće i povrće, Čačak-Beograd.

Mitchell AF, Hong Y, Koh E, Barrett DM, Bryant DE, Denison RF, Kaffka S (2007). Ten-year comparison of the influence of organic and conventional crop management practices on the content of flavonoids in tomatoes. Journal of Agricultural and Food Chemistry 55(15):6154-6159. *https://doi.org/10.1021/jf070344+*

Moore PP, Perkins-Veazie P, Weber CA, Howard L (2008). Environmental effect on antioxidant content of ten raspberry cultivars. Acta Horticulturae 777:499-501. *https://doi.org/10.17660/actahortic.2008.777.76*

Morales CG, Pino MT, Del Pozo A (2013). Phenological and physiological responses to drought stress and subsequent rehydration cycles in two raspberry cultivars. Scientia Horticulturae 162:234-241. *https://doi.org/10.1016/j.scienta.2013.07.025*

Mratinić E (2015). Kupina [Blackberry]. Partenon, Beograd p 108.

Orhan E, Esitken A, Ercisli S, Turan M, Sahin F (2006). Effects of plant growth promoting rhizobacteria (PGPR) on yield, growth and nutrient contents in organically growing raspberry. Scientia Horticulturae 111:38-43. *https://doi.org/10.1016/j.scienta.2006.09.002*

Papp J, Kobzos-Pápai I, Nagy J (1984). Effect of nitrogen application on yield, leafnutrient status and fruit chemical composition of raspberry and red currant varieties. Acta Agronomica Academiae Scientiarum Hungaricae 33:337-343.

Petrović S, Milošević T (2002). Malina-tehnologija i organizacija proizvodnje. Agronomski fakultet, Čačak.

Petrović S, Milošević T, Jevremović D, Glišić I, Milošević N (2020). Jagodasto voće - tehnologija gajenja, zaštite i prerade [Berry fruits - growing, protection, and processing technology]. Agronomski fakultet, Čačak.

Purbasha P, Kumar C, Mrutyunjay D, Tusarkanta B, Ambika M (2020). Fertilizer use and soil acidity. Biomolecule Reports- An International eNewsletter BR/02/19/17

Sawicka B, Barbaś P, Skiba D, Krochmal-Marczak B, Pszczółkowski P (2023). Evaluation of the quality of raspberries (*Rubus idaeus* L.) grown in balanced fertilization conditions. commodities 2(3):220-245. *https://doi.org/10.3390/commodities2030014*

Sistrunk WA (1963). Field condition and processing practices relating to frozen strawberry quality. Proceedings of the American Society for Horticultural Science 83:440-446.

Stojanov D (2019). Uticaj organskih, organo-mineralnih i mineralnih hraniva na vegetativni rast, rodni potencijal i fizičko-hemijske osobine ploda maline (*Rubus idaeus* L.) [Impact of organic, organo-mineral and mineral fertilizers on vegetative growth, yield potential and physico-chemical properties of raspberry (*Rubus idaeus* L.)]. PhD Tesis. Univerzitet u Kragujevcu, Agronomski fakultet, Čačak.

Ubavić M, Bošković-Rakočević LJ, Paunović G (2016). Ishrana voćaka [Fruit tree nutrition]. Univerzitet u Kragujevcu, Agronomski fakultet, Čačak, 1-242.

Valentinuzzi F, Pii Y, Mimmo T, Savini G, Curzel S, Cesco S (2018). Fertilization strategies as a tool to modify the organoleptic properties of raspberry (*Rubus idaeus* L.) fruits. Scientia Horticulturae 240:205-212. *https://doi.org/10.1016/j.scienta.2018.06.024*

Veličković N (2007). Malina-praktični priručnik [Raspberry – practical manual]. PEGAZ, Bijelo Polje p 65.

Zhang JB, Zhu TB, Cai ZC, Qin SW, Müller C (2012). Effects of long-term repeated mineral and organic fertilizer applications on soil nitrogen transformations. European Journal of Soil Sciences 63(1):75. *https://doi.org/10.1111/j.1365-2389.2011.01410.x*

Zhang Q, Li Y, Yang X (2016). Soil pH and nutrient availability regulate soil microbial communities in a long-term fertilization experiment. Soil Biology and Biochemistry 98:37-49. *https://doi.org/10.1016/j.soilbio.2016.04.016*

Zhang X, Li S, An X, Song Z, Zhu Y, Tan Y, Guo X, Wang D (2025). Effects of nitrogen, phosphorus and potassium formula fertilization on the yield and berry quality of blueberry. PLoS ONE 20(1):e0318032. *https://doi.org/10.1371/journal.pone.0318032*