agronomy

Article

Sustainable Management of Fruit Growing in Rural Areas of Montenegro: The Impact of Location on the Phenological and Nutritional Properties on Raspberry (*Rubus idaeus* L.)

Dejan Zejak [1], Ivan Glisic [2], Velibor Spalevic [3,4], Pavle Maskovic [2] and Branislav Dudic [5,6,*]

1 Faculty of Agriculture, University of Belgrade, Nemanjina 6, 11080 Zemun, Serbia; zejakd@gmail.com
2 Faculty of Agronomy in Cacak, University of Kragujevac, Cara Dusana 34, 32000 Cacak, Serbia; glishoo@yahoo.com (I.G.); pavlem@kg.ac.rs (P.M.)
3 Biotechnical Faculty, University of Montenegro, Podgorica 81000, Montenegro; velibor.spalevic@ucg.ac.me
4 Geography Department, Faculty of Philosophy, University of Montenegro, Niksic 81400, Montenegro
5 Faculty of Management, Comenius University, 82005 Bratislava, Slovakia
6 Faculty of Economics and Engineering Management, University Business Academy, 21102 Novi Sad, Serbia
* Correspondence: branislav.dudic@fm.uniba.sk

Citation: Zejak, D.; Glisic, I.; Spalevic, V.; Maskovic, P.; Dudic, B. Sustainable Management of Fruit Growing in Rural Areas of Montenegro: The Impact of Location on the Phenological and Nutritional Properties on Raspberry (*Rubus idaeus* L.). *Agronomy* **2021**, *11*, 1663. https://doi.org/10.3390/agronomy11081663

Academic Editor: Sophie Parks

Received: 15 July 2021
Accepted: 19 August 2021
Published: 20 August 2021

Abstract: The physical-geographical features of the mountainous area of Montenegro cause difficulties in farmers' life and work. The organization of the agricultural production faces a number of problems that limit the overall development of rural areas. Some agricultural crops, such as raspberry, have found optimal growth conditions and produce appropriate yields associated with good fruit quality in such mountainous area. The Willamette variety dominates the production and has broadly expanded, as some new varieties, most notably, Tulameen, Fertödi Zamatos, and Glen Ample. The aim of this paper was to examine the biological and production characteristics of two raspberry varieties—one floricane (Tulameen) and one primocane (Polka)—grown in two localities. We confirmed the general rule that at lower altitudes, these varieties (570 m a.s.l, Bijelo Polje) are characterized by earlier vegetation, flowering, and fruit ripening in comparison to plants at higher altitude (1040 m a.s.l, Mojkovac). The Tulameen variety started flowering on 15 May in Bijelo Polje (the flowering phenophase lasted for 29 days, until 12 June) and on 25 May in Mojkovac (the flowering phenophase lasted for 27 days, until 20 June). That is a delay of 11 days in relation to different locations. The Polka variety started flowering on 25 June in Bijelo Polje (57 days, until 20 August) and on 1 July in Mojkovac (flowering for 67 days, until 5 September). That is a delay of 7 days. The Tulameen variety started maturation on 14 June in Bijelo Polje (maturation lasted for 27 days, until 10 July) and on 22 June in Mojkovac (26 days, until 17 July), with a delay of 9 days. The Polka variety started maturation on 23 July in Bijelo Polje (the maturation phenophase lasted for 55 days, until 15 September) and on 5 August in Mojkovac (52 days, until 25 September), with a delay of 14 days. The results showed that the Polka variety had significantly higher total phenol content than the Tulameen variety (4.43 and 4.03 mg, respectively). In terms of locality, the Mojkovac raspberries had higher total phenol content than the Bijelo Polje raspberries. Polka raspberries also had a higher content of total flavonoids than Tulameen, whereas differences between localities in regard to the content of total flavonoids in the fruit were not significant. The content of condensed tannins and gallotannins in the raspberry fruit was similar in relation to both varieties and localities. Finally, the total antioxidant capacity was significantly higher in the Polka compared to the Tulameen variety, whereas the differences between localities were not statistically significant.

Keywords: fruit growing; raspberry; *Rubus idaeus* L.; phenological properties; Montenegro

1. Introduction

Montenegro is situated in Southeast Europe along the Adriatic Sea and within the Dinaric Alps, a Western Balkan mountain range comprising mainly NW–SE-oriented ridges [1]. The narrow coastal zone with steep limestone slopes rises to average heights of 800 m, a ria coast is centered on Boka and a large debris cone near Albania [2]. Northeast of this zone lies the high karst zone, which extends about 60 km in NW–SE direction. Parallel to this zone, the elevated Durmitor Flysch region presents a relatively soft lithology dominated by sandstones, siltstones, marls, and conglomerates. The north-western highlands contain limestone and derive from a combination of glacial and karst processes. In the north, the Tara canyon marks the border with the Northern crystalline hills and mainly comprises flysch and sandstone sediments. The Polimlje Region with Prokletije, where the study area of this research is located, contains a varied geology (schists, sand- and limestone, dolomites, volcanic outcrops) with glacial geomorphic features [2–5].

The organization of any agricultural production in such hilly and mountainous landscapes faces a number of problems that limit the overall development of the agricultural area, so that in most cases, the potential of this remote rural area remains large and permanently underutilized.

However, some agricultural crops have found optimal growth conditions and produce appropriate yields accompanied by good fruit quality in the hilly and mountainous area. One of such crops is raspberry, with perennial raspberry plantations, record-breaking yields, excellent fruit quality, and the production of this fruit species in this region, which is one of the world's largest producers at high altitudes [6,7].

Growing raspberries in the north Montenegro has been increasing and is greatly contributing to the development of the region. The Willamette variety dominates the production and, over the last decade, the Meeker variety, as well as some new varieties, most notably, Tulameen, Fertődi Zamatos, and Glen Ample have broadly expanded. To extend the season of supply of fresh raspberries, remontant (primocane) varieties are increasingly represented, even in the plains. Taking into account the growing demand for fresh raspberry fruit, with the aim to make it available on the market for a longer period of time, great attention should be paid to two-crop (remontant) red raspberry varieties (*Rubus idaeus* L.) characterized by excellent fruit quality, firmness, and robustness and suitable for table use. Thanks to the extended maturation period, which, in our agro-ecological conditions takes place from mid-July to September and even until the end of October, the fruits of these varieties can be present on the market over a longer period of time and be sold at significantly higher prices than floricane varieties intended primarily for industrial processing. According to the data of Petrovic and Leposavic [8], Polka fruits are small-to-medium-sized, cone-shaped, light red, attractive, and of better quality than most of the two-crop raspberry varieties [9]. This variety can be grown at higher altitudes.

Tulameen is a Canadian variety. It has been in production since the beginning of the 1990s. The parents of this variety are Nootka and Glen Prosen. The fruits are large (about 2 g heavier than Vilamet), long, conical in shape, bright red in color, aromatic, and sweet-sour in taste. They keep well and stay fresh for a few days after harvest. They are primarily intended for fresh consumption and are also suitable for freezing and other types of processing.

The shoots are lush, green in color, with irregularly distributed purple spots in the ground part of the shoot. The native shoots are strong, fairly erect, gray-yellow, with longitudinal cracking of the bark in the base part. The fruits are well distributed and have a relatively long stalk.

Polka is a very popular Polish variety, created in 2001 at the Institute of Fruit Growing and Floriculture in Skernjevice, Poland. The varieties 'Autumn Bliss' and 'Lloyd George' and the species *Rubus crataegifolius* were used for the creation of this variety. Due to the good quality of the fruits, ripening time, yield, and pronounced resistance, Polka has spread very quickly in production plantations in Poland, Serbia, Bosnia and Herzegovina, Ukraine, and other well-known raspberries growers in the world.

The fruits of this variety are small-to-medium-sized, conical, light red in color, very attractive in appearance, and of better quality than most two-species varieties of raspberries. Their primary purpose is for fresh consumption, but due to their good technological characteristics, Polka fruits can also be used for various types of processing. When frozen and stored, the fruits turn to a darker color, which means that they lose their quality.

The aim of this paper was to examine the biological and production characteristics of two raspberry varieties—one floricane (Tulameen) and one primocane (Polka)—grown in two localities. Particular emphasis was laid on the maturation of the fruits of the two varieties in different localities to assess the possibility of expanding the supply of fresh raspberries on the market, especially during the summer tourist season. The second part of the research addresses the fruits' nutritional properties, highlighting that raspberries represent a diverse source of healthy antioxidants and, as such, can be a useful component of our daily diet. The studied raspberry fruits are a rich source of total phenolic compounds and total flavonoids and, thus, could be used as a food additive [10–12].

2. Materials and Methods

2.1. Location of the Study Area

The research work presented in this paper was carried out in two localities in the northern part of Montenegro. This area is mainly mountainous, with the presence of deep valleys incised into limestone ranges. The north Montenegro is rather hilly and underlain by Palaeozoic rocks. In this region, the highest peaks of Montenegro are found, including Komovi (2487 m a.s.l.) and Zla Kolata (2535 m a.s.l.) in the Prokletije Mountains. The rivers in this region drain to the Black Sea, and some of them form deep canyons crossing limestone formations. Further downstream, the River Lim and Tara (close to Mojkovac) form broad valleys flowing through softer Palaeozoic material [1–5].

The research localities are located in the village of Ribarevina (established in 2014), 5 km east of Bijelo Polje (hereinafter referred to as Bijelo Polje Locality) and in the village of Podbisce (established in 2013)—3 km east of Mojkovac (hereinafter referred to as Mojkovac Locality). The Bijelo Polje Locality is located at 570 m a.s.l.; the plot is flat and with western exposure. The Mojkovac Locality is located at 870 m a.s.l; the plantation is set up on a plot with a great inclination and a southwest exposure. The study area is presented in Figures 1–3.

2.2. Climatic Characteristics

The study area is characterized by a moderate continental climate, and in some locations, by a typical mountain climate, as in the neighboring Serbia and other Balkan countries [6,7].

Climate has a high impact on this type of agriculture production. According to Jovovic et al. [13], "Global warming is driving a dynamic hydrological cycle with higher total precipitation and more frequent high intensity rainfall events. Rainfall amounts and intensities expanded around the world and, according to various models, it is expected to increase furthermore during the 21st century. In this Region, we are experiencing increasing temperatures and evapotranspiration, most notably in the northern mountainous region of the studied area. The first two decades of the 21st century were recorded as the warmest since records started, with the most prominent changes within the northern hilly region, corresponding to +1.4 °C and with a decrease in the number of frost days and exceptionally cold days and evenings. A changing precipitation pattern is also forecasted in the near future (less precipitation in summer, more in winter), with an increase of land degradation and erosion processes, water stress (summer), and flood risk (winter). Regarding rainfall, there has been no critical decrease in the normal annual rainfall: precipitation has increased in autumn, whereas it has decreased from springtime to wintertime. There has been a significant increase in the number of extreme weather events".

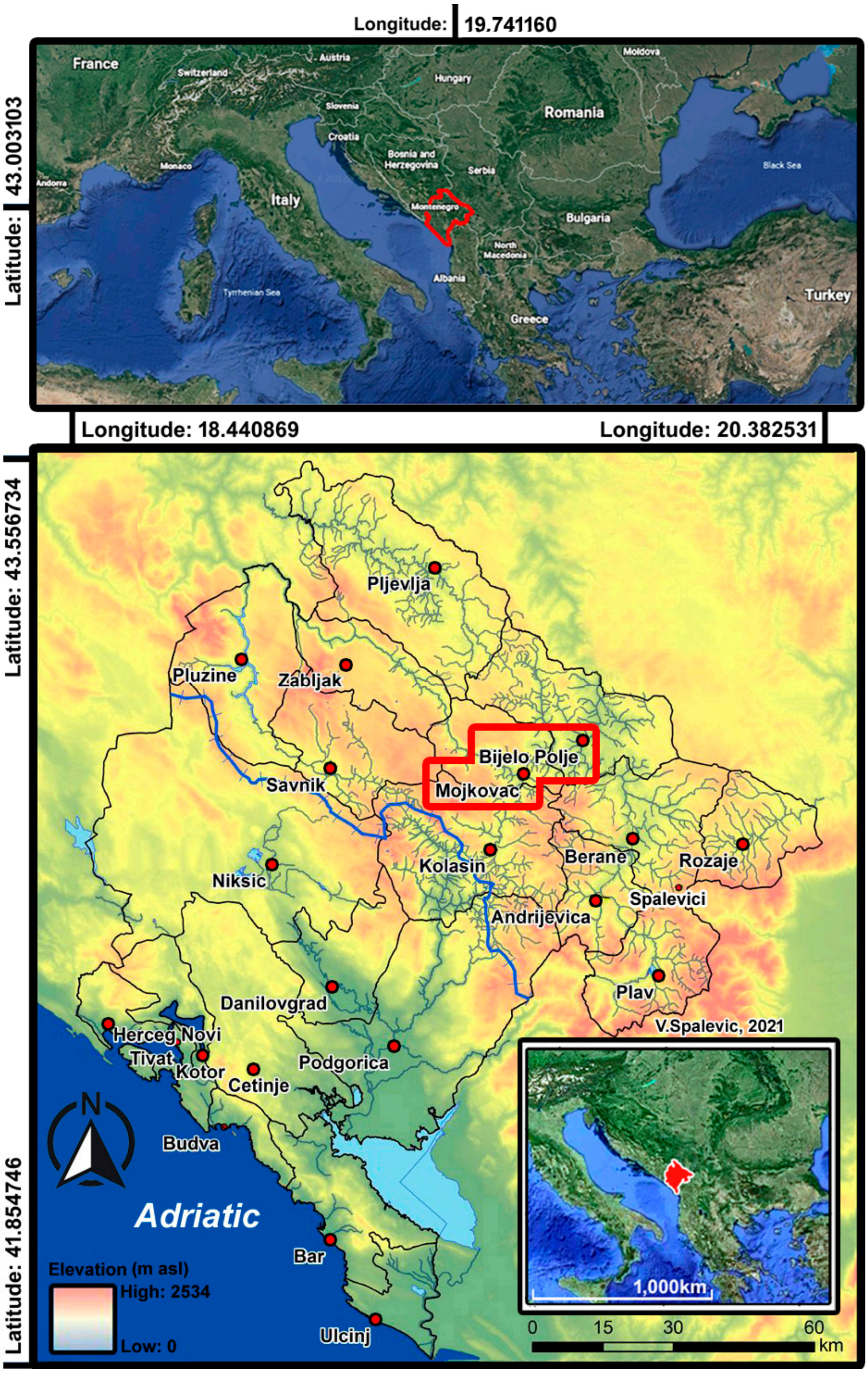

Figure 1. Position of the study areas: Bijelo Polje and Mojkovac, Montenegro.

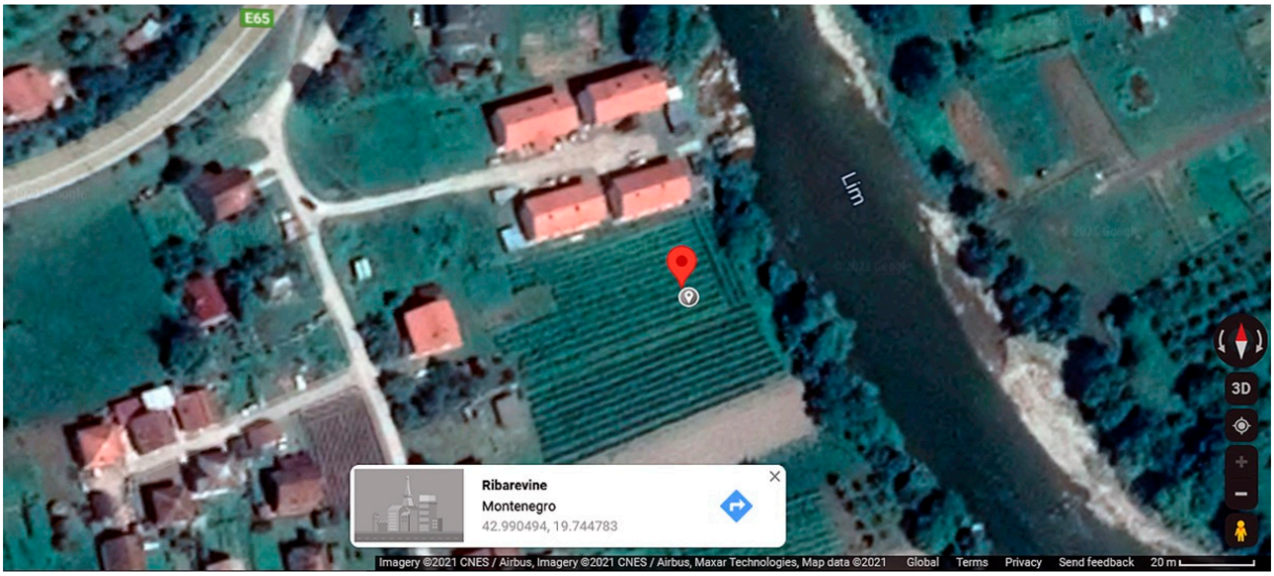

Figure 2. Position of the study area in Ribarevina, Bijelo Polje, Montenegro (42°59′25.8″ N 19°44′41.2″ E; 42.990494, 19.744783), source Google maps.

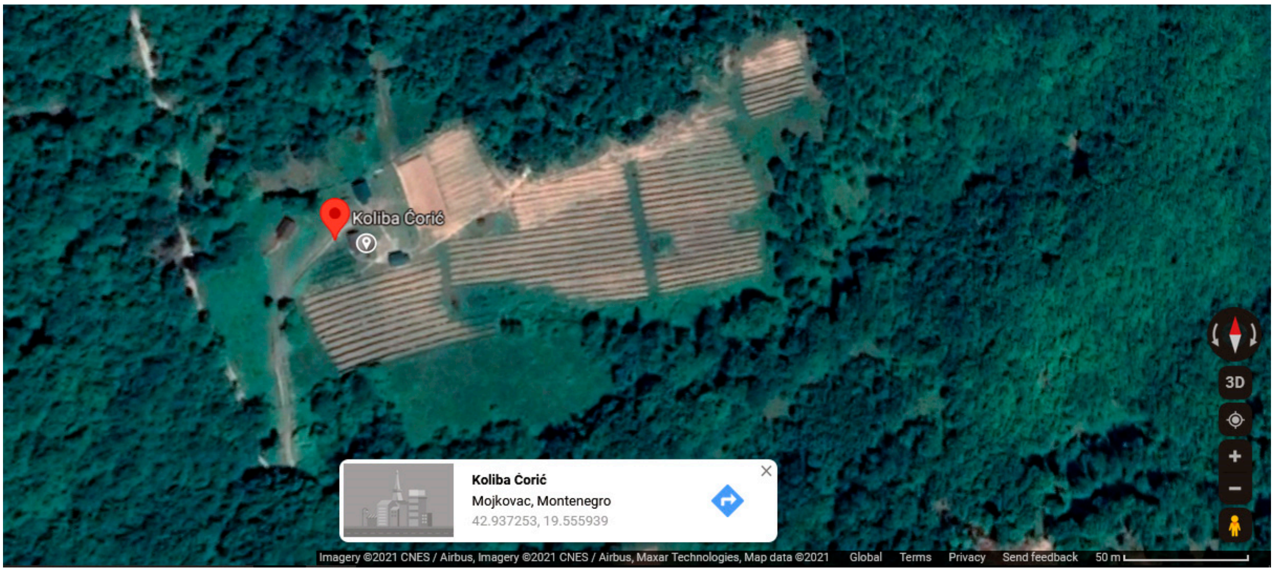

Figure 3. Position of the study area in Podbisce, Mojkovac, Montenegro (42°56′14.5″ N 19°33′22.4″ E; 42.937360, 19.556216), source Google maps.

"Furthermore, storms have become more frequent and more intense as of the begging of the 21st century, resulting in huge amounts of precipitation and high flooding. Flooding, droughts, and heatwaves are progressively impacting natural assets. Flash floods and heavy snowfalls are becoming more common. For this research, we used data provided by the Institute of Hydrometeorology of Montenegro, which considers the studied region as having continental climate, with rainy autumns and springs and cold winters" [4,14,15].

Some basic data on meteorological conditions in the course of the experiments are presented in the Table 1.

Table 1. Precipitation and temperature in the period 1948–2020 in the studied area.

	Jan	**Feb**	**Mar**	**Apr**	**May**	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dec**
Max. daily precipitation in mm												
max	68.6	92.8	73	93.3	42.6	58.5	97.8	55.8	95.6	157.6	101.6	79.4
Aver.	23.4	22.8	21.5	24	21.9	20.6	21.7	21.1	25.3	29	29	23.5
St.D.	15.6	18.6	13	15	9.9	12.4	15.2	11.9	17.2	24.6	16.2	14.7
Mean monthly temperatures in °C												
max	2.9	5.8	7.8	12.6	15.8	18.2	20.8	20.9	17.7	12.6	8.6	4.4
min	−5.6	−5.2	−0.7	6.1	9.8	14.1	16.2	14.3	11.3	6.2	−1.6	−4.7
Aver.	−1.6	0.8	4.6	8.9	13.3	16.3	18.1	17.7	14.3	9.4	4.5	0.1
St.D.	2.2	2.7	2.1	1.3	1.3	1	1.1	1.4	1.5	1.4	2.1	2.2
Max. daily temperatures in °C												
max	15.4	20.9	25.6	28.1	32.4	35.5	36.8	39.2	36	29.5	23	19.2
Aver.	11.7	14.5	20.1	23.6	27.6	30.4	32.8	32.8	29.4	24.8	18.6	13.6
St.D.	2.8	3	3.1	2.3	2.2	2.5	2	2.5	2.6	2.6	2.8	3.3
Min. daily temperatures in °C												
min	−27.6	−24.5	- 16.5	−7.5	−4	0	1.2	2.6	−4	−7.2	−15.4	−21.7
Aver.	−15.1	−13	-8.4	−2.8	0.9	4.8	6.5	6.1	2.3	−2.5	−7.3	−12.6
St.D.	5.3	4.7	4.1	1.8	2	1.8	2.1	1.5	2.5	2.3	3.7	4.6

Source: Data from the Hydrometeorological Institute of Montenegro [13,15].

The absolute maximum air temperature ever recorded is 39.2 °C. Winters are severe with negative temperatures as low as −27.6 °C. The average annual air temperature, t0, is 8.9 °C. The average annual precipitation, Hyear, is 873 mm [15].

2.3. The Geological Structure and Soils of the Area

Montenegro is part of the Dinaric Alps, which are included in the complex thrust-and-fold system of the Mediterranean area. This area represents a proxy of long-lasting interactions between Eurasia and Gondwana, resulting in a system of fold-and-thrust belts and associated foreland and back-arc basins. The system cannot be interpreted as the end product of one single Alpine orogeny, as the major suture zones result from various tectonic events which closed different oceanic basins of the former Tethyan Ocean. The Dinarides–Albanides–Hellenides orogenic belt was caused by a Tertiary collision between the Adriatic promontory and the Serbo–Macedonian–Rhodope blocks. The belt is bordered to the west by a foreland basin in the Eastern Adriatic basin filled with Eocene-Quaternary deep marine sediments [14,15].

The study area at large consists of various types of sediment, magmatic, and metamorphic rocks generated in the long, Palaeozoic to Quaternary, interval. Most of the terrain is underlain by Mesozoic formations of carbonate composition, while magmatic and silico-clastic rocks are substantially less present. Palaeozoic geological formations consist of sedimentary and metamorphic, silico-clastic rocks found mostly in the north-eastern parts of Montenegro, while Cainozoic rocks of carbonate and clastic composition occur almost in all regions of Montenegro [14–17].

The main rocks outcropping in the area are clastic and subordinate carbonate rocks from the Palaeozoic, Triassic clastites, volcanites, tuffs, limestone and dolomites, Jurassic clastic rocks with diabasic effusions, metamorphic rocks, and Quaternary, mainly alluvial and colluvial deposits.

According to the results of the field visits and supplementary laboratory analysis, but also using the previous research data of the project Soils of Montenegro (1964–1988) carried out by Fustic and Djuretic [18] and Spalevic [5,19], the most common soil types in the study basin are Dystric Cambisols and Fluvisols and Colluvial Fluvisols in the lower alluvial plain.

2.4. Phenological Characteristics and Chemical Analysis

The varieties examined were Tulameen and Polka. These two varieties are present in a small area of Montenegro's raspberry growing region compared to Willamette. However, as these varieties mature during June and July (Tulameen) and during August and September (Polka), they are very interesting for expansion of the variety structure and extension of the supply, especially, the supply of fresh raspberries on the market. The plantations were set up with a plant spacing of 2.8 $\times$ 0.3 m or 11,900 seedlings per hectare. Standard agrotechnical and protection measures were applied in the plantation.

The analyses were carried out in 2018 and 2019.

The fruit samples were collected from the field in plastic clamshells containing 500 g of fruit (two plastic clamshells for each) in both studied years 2018 and 2019: (1) Polka/Bijelo Polje; (2) Tulameen/Bijelo Polje; (3) Polka/Mojkovac; (4) Tulameen/Mojkovac. The samples were transported the same day after collection from the filed in a small portable 12 Volt mini outdoor mobile freezer to the Laboratory for production monitoring and quality control, Faculty of Agronomy in Cacak, University of Kragujevac, Serbia.

The analyses covered:

– The phenological characteristics—beginning, flow, and end of the flowering and fruit maturation phenophases. The flowering phenophase was determined by registering the start date (when 10% of the flowers were opened) and the end date (when petals fell off of 90% of flowers) of the blooming. The maturation phase was determined by registering the start date (the day of the first harvest) and the end of the harvest (the day of the last harvest). The duration of the flowering and maturation phenophases was expressed in days;
– The primary characteristics of the fruit—weight and content of soluble dry matter. The fruit weight (g) was determined using the analytical-scale FCB 6K (Kern and Sohn GmbH, Belingen, Germany) with accuracy of $\pm$0.1 g, while the soluble dry matter content (°Brix) was determined using the manual refractometer Milwaukee MR 200 (ATC, Rocky Mount, NC, USA) with accuracy of $\pm$0.2%;
– The content of the so-called secondary chemical compounds—total phenols, total flavonoids, condensed tannins and gallotannins, on the basis of which the total antioxidant capacity was calculated. The total phenol content (mg GA/g extract) was determined by the Folin–Ciocalteu reagent method. The total quantity of flavonoids (mg RU/g extract) was determined by spectrophotometry using $AlCl_3$ according to a methodology described by Brighente et al. [20]. The method for the determination of condensed tannins is based on proanthocyanidins deposition using formaldehyde [21]. Gallotannins can be quantitatively determined by the potassium iodate assay. The assay is based on the reaction of potassium iodate (KIO_3) with galloyl esters [21], which leads to the formation of a red intermediate and, eventually, a yellow compound. All determinations were performed in triplicate, and the results are presented as the mean value of three measurements ($\pm$ standard deviation). The abovementioned methods were performed on a HACH DR2800 mobile photometer at the Faculty of Agronomy in Cacak, in the laboratory for testing the quality of food and agricultural products. The total antioxidant activity of the obtained extracts was determined by the phosphomolybdenum method [22]. The method is based on a reduction of Mo (VI) to Mo (V) using antioxidants, resulting in phosphate/Mo (V) complex in an acidic environment. The antioxidant activity of the fruit extracts analyzed was determined by the ABTS+ radical cation method described by Re et al. [23]. Antioxidant activity at the DPPH radical level was determined by the method described by Kumarasamy et al. [24]. The abovementioned methods were performed on a CARY 300 spectrophotometer at the Faculty of Agronomy in Cacak, in the laboratory for testing the quality of food and agricultural products.

2.5. Statistical Analysis

Statistical analysis and testing of the significance of the differences obtained were carried out by variance analysis and LSD test for the significance level of $p \leq 0.05$, using the ANOVA statistical software (SPS Statistica, Software 5.0).

3. Results

The results concerning the blooming of the tested raspberry varieties at different localities in the north of Montenegro are shown in Figure 4.

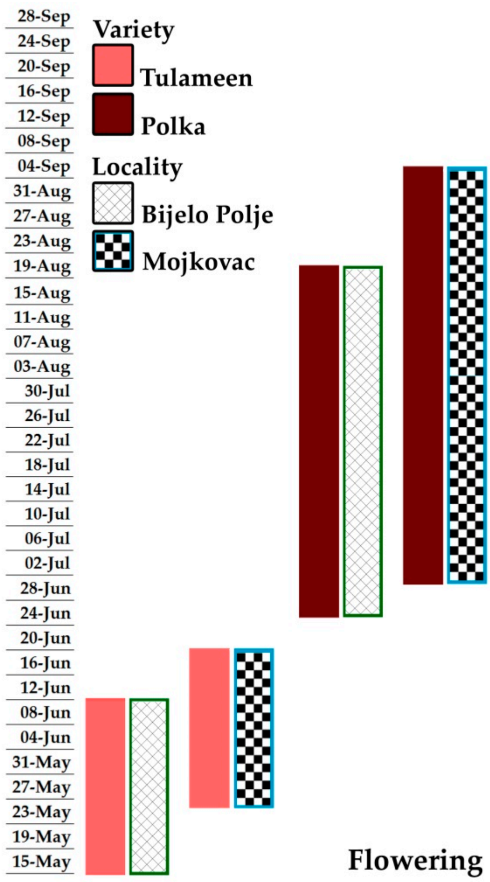

Figure 4. Flowering phenophase of the Polka and Tulameen varieties in the localities Bijelo Polje and Mojkovac—Montenegro (average, years 2018/2019).

The results showed that the flowering phenophase flow varied significantly depending on the variety and on the locality.

At the Bijelo Polje Locality, the Tulameen variety flowered from 15 May to 12 June (the flowering phenophase lasted for 28 days). The same variety at the Mojkovac Locality began flowering 10 days later (25 May), and flowering lasted until 20 June (the flowering phenophase lasted for 26 days).

The Polka variety in both localities flowered later than the Tulameen variety. In the Bijelo Polje Locality, flowering lasted from 25 June to 20 August, while in the Mojkovac Locality, Polka flowered from 1 July to 5 September. In both localities under research, the first flowering phase (flowering in the upper part of the primocanes) lasted about 20–25 days, followed by a shorter recess in flowering. The second flowering phase (on the same primocanes, only in their central and lower parts) was shorter by 15 to 20 days. A similar pattern was also observed in relation to the fruit maturation phenophase (Figure 5).

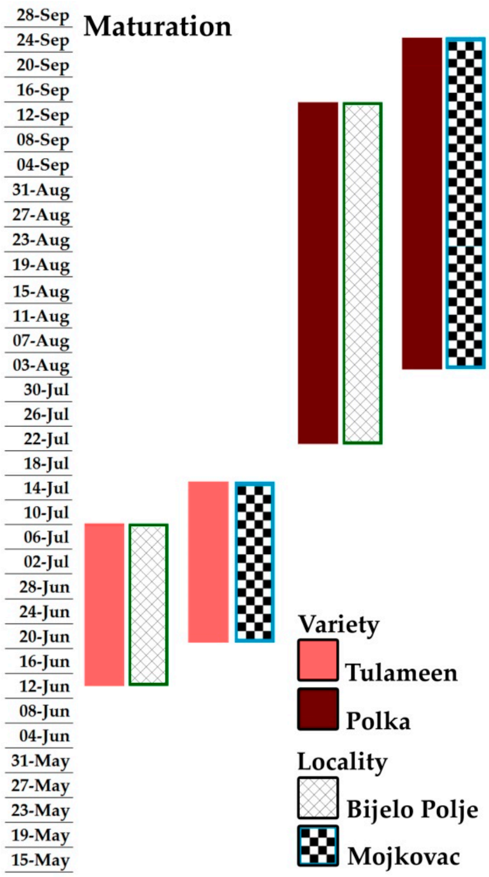

Figure 5. Maturation phenophase of the Polka and Tulameen varieties in the localities Bijelo Polje and Mojkovac—Montenegro (average, years 2018/2019).

The Tulameen variety is a floricane variety that maturated before the Polka variety. At the Bijelo Polje Locality, the Tulameen maturation period was from 14 June to 10 July (the harvesting period lasted 26 days). In the same Locality, Polka fruit maturation began on 23 July. The first harvest wave lasted until 15 August, followed by a brief recess in maturation. In the lower cane parts, fruit maturation started around 22 August and lasted until 15 September.

Fruit maturation in the Mojkovac Locality began later than in the Bijelo Polje Locality. In the Mojkovac Locality, the Tulameen variety matured from 22 June to 17 July, which is, on average, 8 days later than in the Bijelo Polje Locality. The Polka variety matured in the Mojkovac Locality from 5 August to 25 September (10–15 days later than in the Bijelo Polje Locality). In the Mojkovac Locality, fruit maturation of the Polka variety was continuous, i.e., there it did not stop as in Bijelo Polje. The main reason for this phenomenon is that Mojkovac is at a higher altitude; temperatures in summer are moderate and pleasant, exceptionally favorable for raspberry maturation [6]. High temperatures that can occur during the summer, especially on southern slopes and at lower elevation, can cause significant problems in fruit bearing, leading to smaller fruits, the appearance of sunburns, and even a halt in flowering or fruiting [7].

Based on the results shown, it can be stated that the conditions of the locality significantly affected the dynamics and duration of the flowering and fruiting phases.

Climate parameters significantly influence the beginning and flow of phenophases in raspberry development [25,26].

The most important climate parameters are temperature, sunlight quality, and daylight duration [27]. Our results related to flowering duration (26–28 days for Tulameen and 55–65 days for Polka), as well as to the duration of fruit bearing, are similar to the results of several other authors [8,28–31].

Orographic factors, primarily altitude, significantly modify climate parameters. The Mojkovac Locality is located 470 m higher than the Bijelo Polje Locality, which caused significant differences in the time of beginning and in the flow of the flowering and fruit-bearing phenophases.

For the same variety, due to the influence of latitude, altitude, climate conditions, terrain exposure, etc., the difference in time between the phenophases can be several weeks [30], as shown by the results of our paper.

The results related to the fruit characteristics of the raspberry varieties under research in different localities are summarized in Tables 2 and 3.

Table 2. Raspberry fruit weight (g) and soluble dry matter content.

	Fruit Weight (g)	Soluble Dry Matter (°Brix)
Bijelo Polje locality		
Tulameen	4.76 ± 0.17 a	12.24 ± 0.40 a
Polka	3.47 ± 0.23 b	13.63 ± 0.59 b
Mojkovac locality		
Tulameen	4.14 ± 0.20	12.87 ± 0.48
Polka	4.09 ± 0.18	12.99 ± 0.50
ANOVA		
A	*	*
B	ns	ns
A × B	*	*

The table contains the average values of the studied parameters for two years ± a standard error; * indicates significant differences, and ns not significant differences at $p \leq 0.05$ according to the F test; the various lowercase letters in the respective columns indicate significant differences at $p \leq 0.05$ according to the LSD test.

Table 3. Secondary chemical compounds content in raspberry fruits.

	Total Phenols (mg GAE/g)	Total Flavonoids (mg RUE/g)	Condensed Tannins (mg GAE/g)	Gallotannins (mg GAE/g)	Total Antioxidant Capacity (µg AA/g)
Variety (A)					
Tulameen	4.03 ± 0.11 b	1.98 ± 0.09 b	0.53 ± 0.02	0.47 ± 0.01	1.14 ± 0.06 b
Polka	4.43 ± 0.09 a	2.16± 0.10 a	0.54± 0.02	0.47± 0.01	1.27± 0.07 a
Locality (B)					
Bijelo Polje	4.18 ± 0.10 b	2.07 ± 0.08	0.52 ± 0.02	0.48 ± 0.01	1.20 ± 0.05
Mojkovac	4.29 ± 0.09 a	2.08 ± 0.09	0.55 ± 0.03	0.46 ± 0.01	1.21 ± 0.07
ANOVA					
A	*	*	ns	ns	*
B	*	ns	ns	ns	ns
A × B	ns	*	ns	ns	ns

The table contains the average values of the studied parameters for two years ± a standard error; * indicates significant differences, and ns not significant differences at $p \leq 0.05$ according to the F test; the various lowercase letters in the respective columns indicate significant differences at $p \leq 0.05$ according to the LSD test.

The results related to the fruit characteristics of the raspberry varieties under research in different localities are presented in Tables 2 and 3.

The differences in fruit weight as well as in soluble dry matter content between the localities were not statistically significant. However, the differences between varieties were significant. The Tulameen variety fruits were significantly larger than those of Polka (4.76 g and 3.47 g, respectively), which is consistent with their variety characteristics and the results of various authors, i.e., Daubeny and Anderson [32], Peral Eyduran and Sabit Ağaoğlu [33], Milivojevic et al. [34], Leposavic et al. [35].

As regards the soluble dry matter content, the results showed that the values of Polka were higher (13.63 °Brix) than those of Tulameen (12.24 °Brix). In the study of Markuszewski et al. (2019), Polka showed a larger soluble dry matter content than Tulameen. Peral Eyduran and Sabit Ağaoğlu (2006) presented a significantly higher value of soluble dry matter content in the Tulameen variety compared to our research. On the other hand, Cortellino et al. [36] obtained values lower than ours, which only confirms that soluble dry matter content is a complex characteristic that depends on a large number of factors, such as tending, locality, and harvest season, i.e., sampling time [37,38].

Fruit and vegetables contain a wide range of antioxidant compounds that can help protect cellular systems against oxidative damage and reduce the risk of chronic diseases [39]. Such benefits for cellular systems can be attributed to the antioxidant capacity and specific interrelationships of compounds present in the raspberry fruit, such as total phenols, total flavonoids, condensed tannins and gallotannins, often referred to as secondary chemical units [40].

The results showed that the Polka variety had significantly higher total phenol content than the Tulameen variety (4.43 and 4.03, respectively). When considering localities, the Mojkovac Site raspberries had higher total phenol content than the Bijelo Polje raspberries. Polka also had a higher content of total flavonoids in the fruit than Tulameen, whereas differences between the localities for the content of total flavonoids in the fruit were not significant. The content of condensed tannins and gallotannins in the raspberry fruit was similar both between the varieties and between the localities. Finally, the total antioxidant capacity was significantly higher in Polka compared to Tulameen, whereas the differences between localities in this regard were not statistically significant.

Phenolic compounds in raspberry fruits are known to have antioxidant, anti-cancerous, anti-inflammatory, and anti-neurodegenerative biological properties [41].

The results obtained by Milivojevic et al. [42] demonstrated that the average content of total phenols for eight raspberry varieties varied from 2.34 to 4.66 mg GAE g^{-1} FW. Moore et al. [43], examining the content of total phenols in 10 raspberry varieties, found an average value of 3.04 mg GAE g^{-1} FW.

Some results suggest that the genotype may have a very strong influence on the content of bioactive compounds in fruits [44]. The research of Milivojevic et al. [45] established a negative correlation between fruit size and most nutritive quality parameters. In particular, raspberry varieties with a smaller fruit size (such as Polka, in our case) showed a higher nutritional value, expressed in high values of soluble dry matter and phenols. Similar results were obtained by Remberg [46], with a negative correlation between fruit weight and total antioxidant capacity in raspberry fruits. Our results are consistent with those of previous studies. As a variety characterized by larger fruits, Tulameen had a slightly lower antioxidant capacity value and lower soluble dry matter content values than Polka.

With our research, we confirmed the findings of Augšpole et al. [47], Liu [39], Krauze-Baranowska M. and Majdan [40] that raspberry represents a diverse source of potentially healthy antioxidants and, as such, can be a useful component of our daily diet. Raspberry cultivar fruits are a rich source of total phenols, total flavonoids, titrable acidity, and total soluble sugars, which demonstrates their potential use as a food additive.

4. Conclusions

Raspberry flowering in the Bijelo Polje Locality was, on average, 7–11 days earlier than flowering in the Mojkovac Locality. Irrespective of the locality, Tulameen flowered 35–40 days earlier than Polka.

There were no differences between the localities in terms of fruit weight and soluble dry matter content. The Tulameen variety was significantly larger but had a lower soluble dry matter content.

The Polka variety had higher total phenols and total flavonoids, as well as a higher total antioxidant capacity compared to Tulameen. The locality had no significant effect on these properties, except for total phenol contents, which was higher in the raspberry fruit from the Mojkovac Locality.

In general, the good raspberry growing results in both localities for both varieties indicate that production of the raspberry varieties Tulameen and Polka can be implemented in the northern part of Montenegro, which will provide a continuous and long-term supply of fresh raspberry on the market, as well as fruits of good quality.

Author Contributions: Conceptualization, D.Z., I.G., V.S., P.M. and B.D.; methodology, D.Z., I.G., V.S., P.M. and B.D.; formal analysis, D.Z., I.G., V.S., P.M. and B.D.; investigation, D.Z., I.G., V.S., P.M. and B.D.; data curation, D.Z. and V.S.; writing—original draft preparation, D.Z., I.G., V.S., P.M. and B.D.; writing—review and editing, D.Z., I.G., V.S., P.M. and B.D.; supervision, I.G. and V.S. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Kerckhof, A.; Spalevic, V.; Eetvelde, V.; Nyssen, J. Factors of land abandonment in mountainous Mediterranean areas: The case of Montenegrin settlements. *SpringerPlus* **2016**, *5*, 485. [CrossRef] [PubMed]
2. Nyssen, J.; Van den Branden, J.; Spalevic, V.; Frankl, A.; Van de Velde, L.; Curovic, M.; Billi, P. Twentieth century land resilience in Montenegro and consequent hydrological response. *Land Degrad. Dev.* **2014**, *25*, 336–349. [CrossRef]
3. Frankl, A.; Lenaerts, T.; Radusinovic, S.; Spalevic, V.; Nyssen, J. The regional geomorphology of Montenegro mapped using land surface parameters. *Z. Geomorphol.* **2016**, *60*, 1–14. [CrossRef]
4. Nikolic, G.; Spalevic, V.; Curovic, M.; Khaledi Darvishan, A.; Skataric, G.; Pajic, M.; Kavian, A.; Tanaskovik, V. Variability of Soil Erosion Intensity Due to Vegetation Cover Changes: Case Study of Orahovacka Rijeka, Montenegro. *Not. Bot. Horti Agrobo.* **2018**, *47*, 237–248. [CrossRef]
5. Spalevic, V. Impact of Land Use on Runoff and Soil Erosion in Polimlje. Ph.D. Thesis, Faculty of Agriculture, University of Belgrade, Belgrade, Serbia, 2011; pp. 1–260.
6. Leposavic, A.; Milenkovic, S.; Cerovic, R. Raspberry production in the hilly mountains region of Serbia. *J. Mt. Agric. Balk.* **2004**, *7*, 317–332.
7. Glisic, I.; Milosevic, T. *Potentials of Hilly and Mountainous Areas for Raspberry Production: Current Situation and Perspectives. Proceedings of the Symposium: Improvement of Agriculture, Forestry and Water Management in Karst, Hilly and Mountainous Areas—Rational Use and Protection*; Academy of Sciences and Arts of Bosnia and Herzegovina: Sarajevo, Bosnia and Herzegovina, 2017; pp. 75–84, Special edition.
8. Petrovic, S.; Leposavic, A. *Raspberry. New Technologies of Cultivation, Protection and Processing*; Scientific Fruit Growing Society of Serbia: Cacak, Serbia, 2016; pp. 1–265.
9. Leposavic, A.; Jankovic, M.; Sretenovic, D.; Stevanovic, S.; Jevremovic, D. Biological and pomological properties of some red raspberries cultivars. *J. Mt. Agric. Balk. Troyan* **2006**, *9*, 803–815.
10. Grajkowski, J.; Ochmian, I. Influence of three biostimulants on yielding and fruit quality of three primo cane raspberry cultivars. *Acta Sci. Pol. Hortorum Cultus* **2007**, *6*, 29–36.
11. Wang, A.Y.; Chen, C.-T.; Wang, C.Y. The influence of light and maturity on fruit quality and flavonoid content of red raspberries. *Food Chem.* **2009**, *112*, 676–684. [CrossRef]
12. An, B.; Wei, H.; Li, L.; Guo, P. Nutrient Uptake and Utilization and Antioxidants of Fruits in Red Raspberry (*Rubus idaeus* L.) Cultivar 'Autumn Bliss' in response to Fertilization under Extended Photoperiod. *Not. Bot. Horti Agrobot.* **2018**, *46*, 440–448. [CrossRef]
13. Jovovic, Z.; Dolijanovic, Z.; Spalevic, V.; Dudic, B.; Przulj, N.; Velimirovic, A.; Popovic, V. Effects of Liming and Nutrient Management on Yield and Other Parameters of Potato Productivity on Acid Soils in Montenegro. *Agronomy* **2021**, *11*, 980. [CrossRef]
14. Spalevic, V.; Lakicevic, M.; Radanovic, D.; Billi, P.; Barovic, G.; Vujacic, D.; Sestras, P.; Khaledi Darvishan, A. Ecological-Economic (Eco-Eco) Modelling in the River Basins of Mountainous Regions: Impact of Land Cover Changes on Sediment Yield in the Velicka Rijeka, Montenegro. *Not. Bot. Horti Agrobot.* **2017**, *45*, 602–610. [CrossRef]
15. Spalevic, V.; Barovic, G.; Vujacic, D.; Curovic, M.; Behzadfar, M.; Djurovic, N.; Dudic, B.; Billi, P. The Impact of Land Use Changes on Soil Erosion in the River Basin of Miocki Potok, Montenegro. *Water* **2020**, *12*, 2973. [CrossRef]
16. Zejak, D.; Radovic, A.; Spalevic, V.; Glisic, I. Production of planting material of raspberry variety "Glen Ample" in the North Montenegro. *Agric. For.* **2021**, *67*, 245–259.
17. Spalevic, V.; Zejak, D.; Curovic, M.; Glisic, I.; Radovic, A. Analysis of the impact of fruit growing development on the intensity of soil erosion and runoff: Case study of Krusevo, Bijelo Polje, Montenegro. *Agric. For.* **2021**, *67*, 37–51.
18. Fustic, B.; Djuretic, G. *Soils of Montenegro [Zemljista Crne Gore]*; Spalevic, V., Ed.; University of Montenegro, Biotechnical Institute: Podgorica, Montenegro, 2000; pp. 1–626.
19. Spalevic, V. Application of Computer-Graphic Methods in the Studies of Draining Out and Intensities of Ground Erosion in the Berane Valley. Master's Thesis, Faculty of Agriculture of the University of Belgrade, Belgrade, Serbia, 1999; 135p.
20. Brighente, I.M.C.; Dias, M.; Verdi, L.G.; Pizzolatti, M.G. Antioxidant activity and total phenolic content of some Brazilian species. *Pharm. Biol.* **2007**, *45*, 156–161. [CrossRef]

21. Verrmeris, W.; Nicholson, R. *Phenolic Compound Biochemistry*; Springer: Dordrecht, The Netherlands, 2006. [CrossRef]
22. Prieto, P.; Pineda, M.; Aguilar, M. Spectrophotometric quantitation of antioxidant capacity through the formation of a phosphomolybdenum complex: Specific application to the determination of vitamin E. *Anal. Biochem.* **1999**, *269*, 337–341. [CrossRef]
23. Re, R.; Pellegrini, N.; Proteggente, A.; Pannala, A.; Yang, M.; Rice Evans, C. Antioxidant activity applying an improved ABTS adical cation decolorization assay. *Free Radic. Biol. Med.* **1999**, *26*, 1231–1237. [CrossRef]
24. Kumarasamy, Y.; Byres, M.; Cox, P.J.; Jaspars, M.; Nahar, L.; Sarker, S.D. Screening seeds of some Scottish plants for free-radical scavenging activity. *Phytother. Res.* **2007**, *21*, 615–621. [CrossRef] [PubMed]
25. Ivanovic, M.; Pavlovic, A.; Mitic, M.; Pecev Marinkovic, E.; Krstic, J.; Mrmosanin, J. Determination of Total and Individual Anthocyanins in Raspberries Grown in South Serbia. In Proceedings of the XXI Savetovanje o biotehnologiji, Čačak, Serbia, 11–12 March 2016; pp. 263–267.
26. Neocleous, D.; Papadopoulos, I.; Vasilakakis, M. Growing Red Raspberry in Soilless Culture Under Different Chilling Treatments for Early Summer Production. *Small Fruits Rev.* **2005**, *4*, 37–48. [CrossRef]
27. Sønsteby, A.; Heide, O. Effects of photoperiod and temperature on growth and flowering in the annual (primocane) fruiting raspberry (*Rubus idaeus* L.) cultivar 'Polka'. *J. Hortic. Sci. Biotechnol.* **2009**, *84*, 439–446. [CrossRef]
28. Marinkovic, D.; Misic, P.D.; Zec, G.; Colic, S. Pomoloske osobine sorti maline u Pancevackom ritu. *Jugosl. Vocar.* **2004**, *38*, 91–99.
29. Marinkovic, D.; Zec, G.; Colic, S.; Jankovic, Z. Uticaj ekoloskih faktora na fiziologiju cvetanja i zrenja plodova maline. *Vocarstvo* **2005**, *152*, 493–501.
30. Petrovic, S.; Milosevic, T. *Raspberry from Serbia*; Faculty of Agronomy: Cacak, Serbia, 2005.
31. Nikolic, M.; Milivojevic, J. *Jagodaste Vocke. Tehnologija Gajenja*; Univerzitet u Beogradu, Poljoprivredni Fakultet: Beograd, Serbia, 2015.
32. Daubeny, H.A.; Anderson, A. "Tulameen" Red Raspberry. *HortScience* **1991**, *26*, 1336–1338. [CrossRef]
33. Eyduran, S.P.; Agaoglu, Y.S. A Preliminary Examination Regarding Ten Raspberry Cultivars. *Res. J. Agric. Biol. Sci.* **2006**, *2*, 375–379.
34. Milivojevic, J.; Maksimovic, V.; Nikolic, M.; Bogdanovic, J.; Maletic, R.; Milatovic, D. Chemical and antioxidant properties of cultivated and wild Fragaria and Rubus berries. *J. Food Qual.* **2011**, *34*, 1–9. [CrossRef]
35. Leposavic, A.; Jankovic, M.; Djurovic, D.; Veljkovic, B.; Keserovic, Z.; Popovic, B. Fruit quality of red raspberry cultivars and selections grown in Western Serbia. *Hortic. Sci.* **2013**, *40*, 154–161. [CrossRef]
36. Cortellino, G.; De Vecchi, P.; Lo Scalzo, R.; Ughini, V.; Granelli, G.; Bucchieri, M. Ready-to-eat raspberries: Qualitative and nutra-ceutical characteristics during shelf-life. *Adv. Hortic. Sci.* **2018**, *32*, 399–406.
37. Markuszewski, B.; Wysocki, K.; Kopytowski, J. Summer- and autumn-fruiting raspberry quality as influenced by the harvest date. *Acta Agrophys.* **2019**, *26*, 47–56. [CrossRef]
38. Mazur, S.P.; Nes, A.; Wold, A.B.; Remberg, S.F.; Aaby, K. Quality and chemical composition of ten raspberry (*Rubus idaeus* L.) genotypes during three harvest seasons. *Food Chem.* **2014**, *160*, 233–240. [CrossRef]
39. Liu, R.H. Health benefits of fruit and vegetables are from additive and synergistic combinations of phytochemicals. *Am. J. Clin. Nutr.* **2003**, *78*, 517–520. [CrossRef]
40. Krauze-Baranowska, M.; Majdan, M. Raspberries—A source of therapeutically valuable secondary metabolites and vitamins (in Polish). *Panacea* **2009**, *1*, 14–15.
41. Bomser, J.; Madhavi, D.L.; Singletary, K.; Smith, M.A.L. In vitro anticancer activity of fruit extracts from Vaccinium species. *Planta Med.* **1996**, *62*, 212–216. [CrossRef]
42. Milivojevic, M.J.; Nikolic, D.M.; Dragisic-Maksimovic, J.J.; Radivojevic, D.D. Generative and fruit quality characteristics of primocane fruiting red raspberry cultivars. *Turk. J. Agric. For.* **2011**, *35*, 289–296.
43. Moore, P.P.; Perkins Veazie, P.; Weber, C.A.; Howard, L. Environmental effect on antioxidant content of ten raspberry cultivars. *Acta Hortic.* **2008**, *777*, 499–504. [CrossRef]
44. Anttonen, M.J.; Karjalainen, R.O. Environmental and genetic variation of phenolic compounds in red raspberry. *J. Food Compos. Anal.* **2005**, *18*, 759–769. [CrossRef]
45. Milivojevic, J.; Nikolic, M.; Radivojevic, D.; Poledica, M. Yield components and fruit quality of floricane fruiting raspberry cultivars grown in Serbia. *Acta Hortic.* **2012**, *946*, 95–99. [CrossRef]
46. Remberg, S.F. Studies of Antioxidant Activity in Fruits and Berries. Effects of Cultivars and Postharvest Condition. Ph.D. Dissertation, Norwegian University of Life Science, Ås, Norway, 2006.
47. Augšpole, I.; Dimiš, F.; Romanova, I.; Linia, A. Characterizationof red raspberry (*Rubus idaeus* L.) for their physicochemical and morphological properties. *Agron. Res.* **2021**, *19*, 1227–1233.