



Article Morphological and Biochemical Properties in Fruits of Naturally Grown Cornelian Cherry (*Cornus mas* L.) Genotypes in Northwest Bosnia and Herzegovina

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Abstract: The cornelian cherry (Cornus mas L.) is considered to be one of those medicinal plants with important nutritional and therapeutic properties. The plant shows resistance against abiotic and biotic stressors in natural growing conditions and could be suitable to use in sustainable fruit production. This study was carried out on 22 local cornelian cherry (Cornus mas L.) genotypes, which were grown northwest of Bosnia and Herzegovina. Fruits of these 22 genotypes were harvested and analyzed during the ripening period in 2018/2019. Fruit weight, length, and width ranged from 1.38 to 3.01 g, 13.84 to 19.43 mm, and 10.92 to 14.79 mm, respectively. Dry matter content was determined to be the lowest at 11.67% and the highest at 21.89%. The genotypes had vitamin C content between 25.85 and 58.75 mg/100 g. Total phenolic and anthocyanin content were found to be quite variable among genotypes and ranged from 1240 to 6958 mg gallic acid equivalents (GAE) per 100 g fresh weight (FW) and 55.57 to 205.6 mg cyaniding-3-glucoside equivalents (CGE) per 100 g FW, respectively. The content of phosphorus and iron were between 155.52 to 263.06 mg per 100 g and 0.25 to 0.93 mg per 100 g, respectively. Principal Component Analysis (PCA) showed that the first and second components accounted for 44.05% and 60.50% of the total variance, and the major proportion of the first and second components were the morphometric properties and chemical traits of the cornelian cherry fruits. The results revealed that the characterized genotypes could be important for cornelian cherry breeders as ready crossing materials to obtain new cornelian cherry varieties and shows the potential of certain genotypes as a valuable source of natural antioxidants. The results may have served as a guide towards the development of sustainable production programs for cornelian cherries as well.

Keywords: cornelian cherry; morphology; chemical content; diversity; cluster analysis

1. Introduction

Underutilized and forgotten fruits such as mulberry, cornelian cherry, wild strawberry, blackberry, etc., have been growing under different agro-ecological conditions, and they



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Copyright: © 2022 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). supply sustainable fruit production in Bosnia and Herzegovina. These fruits recently received more attention among consumers due to their health benefits. They show a great biological diversity [1–3].

A lot of high-quality berries are grown in Bosnia and Herzegovina. There are large natural populations of wild berry species, including particular black mulberries and cornelian cherries, and all of these fruits are a valuable source of healthy phytonutrients and have been the subject of many studies [4–7].

Cornelian cherry (*Cornus mas* L.) is primarily grown in the temperate zone of Eurasia, up to 1400 m.a.s.l. and is tolerant to diverse abiotic and biotic conditions [8]. It is a tall shrub or small tree from 2 to 8 m high. Cornelian cherry trees begin to flower in early spring but bear the fruit in the late period. The fruits are light or dark red-colored, oval-shaped, and 10–30 mm long with a weight of 2–5 g.

Cornelian cherry fruit is important in human health and nutrition. The colored fruits are rich in natural antioxidants, such as flavonols, vitamin C, anthocyanin, tannins, and phenolics. The fruits of cornelian cherry are used for different purposes: fresh consumption, dried, jams, juices, syrups, or wines. Farmers from Bosnia and Herzegovina produce a special alcoholic beverage—"rakija". There are many old customs related to the Cornelian cherry, especially as the symbol of health. There is a famous saying in the Bosnian tradition: "zdrav kao drijen" (in English: healthy as a cornel).

Its fruit is not only important for nutrients but also highly tolerant to most biotic and abiotic environmental conditions. In addition, the plant is in general grown as wild or semi-wild in natural growing conditions without pesticides treatments. Thus, it is accepted as one of the most important fruit species that could be used in organic fruit production. However, it is not a frequently searched fruit species in Bosnia and Herzegovina yet and needs to study on it [9].

Plant genetic resources are important in plant breeding to evaluate useful traits of genetic resources and combine those traits in one cultivar. In order to achieve this, the genetic resources must be subjected to morphological characterization for describing and classifying the germplasm. This morphological characterization is also important for crossbreeding studies [10].

Many areas of Bosnia and Herzegovina, due to their natural potentials, natural biodiversity, unpolluted areas, and traditionally determined population for agricultural production, have better predispositions in the production of safe food. Cornelian cherry is spread in the natural population in Bosnia and Herzegovina and has good potential for exploitation [11].

The aim of this study was to provide information on the diversity of genetic resources of *Cornus mas* L. genotypes in the area of northwest Bosnia and Herzegovina in terms of fruit characteristics and selection of promising genotypes that would be recommended for the cultivation in modern organic fruit orchards.

2. Materials and Methods

2.1. Plant Material and Selection

The cornelian cherry trees are abundant in Una–Sana Canton in northwest part of Bosnia and Herzegovina and found as natural populations. The region where cornelian cherries were sampled had similar climatic and soil conditions. The area is characterized by moderate continental climate with cold and humid winters and long warm summers with average altitude of 400 m above sea level. The mean annual temperature and precipitation are 11 °C and 880 to 1880 L/m². A large number of cornelian cherry trees are found alone or as scattered populations in the regions. According to high yield, attractive fruit, and health tree characteristics, a total of 22 promising cornelian cherry genotypes were pre-selected. Fruits of 22 cornelian cherry genotypes were picked up during commercial harvest period of 2018 and 2019 years. The genotypes are found in spontaneous flora in the following areas: Cazin (C1, C2, C3, C4, C5, C6, C7, C8, C9, C10), Bosanska Krupa (BK1, BK2, BK3, BK4, BK5), Bužim (BU1, BU2, BU3, BU4, BU5) and Bihać (BI1, BI2). Thus, genotype codes and numbers represented the sampling area. Fruits were picked at full maturity stage for each genotype (consumption ripeness for morphological and biochemical analysis) and transported to the laboratory. Each sample consisted of 1 kg of fruits per genotype.

2.2. Morphological Analysis

Laboratory analysis of fruits was carried out in the chemical laboratory of the Biotechnical Faculty in Bihać. Biometrical measurements were carried out on fruits, such as fruit weight, fruit length, and fruit width. The weight of the fruit was determined by digital balance. With all 22 genotypes, a sample of 50 fruits (n = 50) was taken, and the individual weights of the fruit were measured in order to calculate the average value. The length and width of the fruit were measured using a digital fruit caliper (HOEGERT, Berlin, Germany).

2.3. Biochemical and Bioactive Content Analysis

After measuring the fruit weight, length, and width, the water, dry matter, ash, acidity, total sugars, anthocyanin, phenols, iron, phosphorus, C vitamins, and pH were determined. All these laboratory analyses were performed in four replicates.

The water content was analyzed by drying with the addition of quartz sand. By this method, water is determined indirectly by measuring the residue that is lagging behind after drying. The addition of sand increases the surface of the sample and accelerates evaporation. It dries at 105 $^{\circ}$ C to constant mass and measure.

Determination of the dry matter content was performed using a refractometer (Carl Zeiss refractometer, Model II). The ash content was determined by incineration at 600 $^{\circ}$ C until white powder was obtained. Five grams of dried fruit were measured, lighted directly, and burned at 600 $^{\circ}$ C until white ash was obtained.

Total sugar was determined using the standard procedures according to Luff-Schoorl described by Vračar 2001 [12].

For titratable acid measurement, potentiometrically method was used, described by Vračar 2001 [12], and results expressed as % citric acid equivalent.

For vitamin C measurement, iodometric titration method was used described by Helmenstine 2007 [13].

For bioactive content analysis (polyphenols and anthocyanins), a simple extraction procedure was performed, and 20 g frozen fruits were added to 20 mL of methanol/HCl 2% (95:5 v/v). Approximately 60 min. later, the berries were homogenized and centrifuged for 15 min at 3000 rpm. The supernatant solution was filtered, and solution was diluted to volume with methanol/HCl 2%. The extraction procedure was repeated twice for each sample.

The pH differential method was used for total anthocyanin determination described by Giusti and Wrolstad (2001) [14]. The results were expressed as mg of cyanidin-3-glucoside per 100 g of fresh weight (FW). Quantification data were the mean of 4 repetition.

Total phenolic compounds were determined by Folin–Ciocalteau's methods described by Singleton and Rossi (1965) [15]. Results were expressed as mg of gallic acid per 100 g of fresh fruits. Quantification data were the mean of 4 repetition.

The phosphorus content was determined by spectrophotometric use of the ammoniumvanadate molybdate method by Gericke and Kurmies (1952) [16].

Measurement of the pH value was carried out with the pH meter by immersing the combined electrode in the sample and reading the values on the device.

The determination of the iron content was performed using the atomic absorption spectrophotometry on the atomic absorption spectrophotometer "Perkin Elmer" AAnalyst-800 with Zeeman correction (flame technique-FAAS). The combination of gases we used for analysis on AAS is acetylene-compressed air, which has reached a temperature of 2400–2700 °C.

2.4. Statistical Analysis

Principal Component Analysis (PCA) analysis, including 14 characteristics of 22 cornelian cherry genotypes, was performed to determine relationship among genotypes [17]. The above-mentioned analyzes of the obtained data set are processed in statistical programs PAST, XLSTAT 13, and SPSS 21 [18]. Post hoc analysis of variance (ANOVA) in cases of statistically significant differences (p < 0.05) was carried out by Tukey's test (p < 0.05). Hierarchical cluster analysis HCPC (Hierarchical Clustering on Principal Components) was used to determine similarities between cornelian cherry genotypes. This type of analysis has been performed using two R packages: FactoMineR for computing HCPC and factoextra to visualize the obtained results.

3. Results and Discussion

3.1. Morphological Properties

Results of morphological characteristics of cornelian cherry fruit are shown in Table 1. There were statistically significant differences among genotypes in terms of fruit weight, length, and width at the 0.05 level. The fruit weight ranged from 1.38 g (C3) to 3.01 g (BK3). These results correspond to the results of [9,19,20]. Obtained results of fruit weight are greater than the reported results by [11,21] but lower than the results of [8,22–25]. The fruit length (13.84–19.43 mm) and width (10.92–14.79 mm) were measured similarly to previously evaluated genotypes from different regions [10,20,21,25]. Variability of cornelian cherry fruit weight and dimensions was evident in the studied populations. Genotypes from Bosanska Krupa were a larger fruit size.

Table 1. Morphological properties of cornelian cherry fruits.

Genotypes	Fruit Weight (g)	Fruit Length (mm)	Fruit Width (mm)
CA1	1.54 ± 0.21 c,d,e	16.47 ± 0.71 a,b	12.17 ± 0.81 c,d
CA2	1.91 ± 0.41 c,b,d	$14.97\pm1.21~\mathrm{a}$	$13.79\pm1.11~\mathrm{b}$
CA3	1.38 ± 0.15 c,d,e	$15.90\pm0.83~\mathrm{a}$	$11.84\pm0.75~\mathrm{c}$
CA4	1.66 ± 0.26 d,e	$15.54\pm1.07~\mathrm{a}$	12.63 ± 0.78 c,d
CA5	1.64 ± 0.18 d,e	15.85 ± 0.74 a,b	$12.42\pm0.68~\text{c,d}$
CA6	1.57 ± 0.25 c,d,e	$14.86\pm0.87~\mathrm{a}$	$11.24 \pm 0.78 \text{ d}$
CA7	1.68 ± 0.26 d,e	15.02 ± 0.84 a,b	$11.11\pm0.84~\mathrm{d}$
CA8	1.49 ± 0.24 c,d,e	13.84 ± 0.86 a	$10.97\pm0.72~\mathrm{c}$
CA9	$2.08\pm0.38~\mathrm{b}$	$15.53\pm1.63~\mathrm{a}$	$12.03\pm1.04~\mathrm{c}$
CA10	$1.53\pm0.21~\mathrm{e}$	13.88 ± 0.88 a	$10.84\pm0.72~\mathrm{d}$
BU1	1.73 ± 0.31 d,e	16.62 ± 0.96 a	12.45 ± 1.07 c,d
BU2	1.64 ± 0.26 d,e	16.55 ± 0.97 a	12.16 ± 0.72 c,d
BU3	$1.59\pm0.26~\mathrm{e}$	16.45 ± 0.84 a,b	12.13 ± 1.37 c,d
BU4	1.95 ± 0.57 b,d	17.52 ± 1.36 a	$12.81\pm1.45~\mathrm{c}$
BU5	1.66 ± 0.24 d,e	16.52 ± 0.83 a,b	12.11 ± 1.12 c,d
BK1	2.22 ± 0.39 b,c	16.96 ± 1.12 a	13.23 ± 1.22 b
BK2	2.02 ± 0.27 b,d	17.75 ± 1.14 a	13.01 ± 0.73 b
BK3	$3.01\pm0.52~\mathrm{a}$	18.36 ± 1.01 a	$14.79\pm1.20~\mathrm{a}$
BK4	2.29 ± 0.34 b	19.43 ± 0.96 a	14.71 ± 0.96 a,b
BK5	$2.29\pm0.34~\mathrm{b}$	$18.27\pm1.55~\mathrm{a}$	$13.65\pm0.73~\mathrm{b}$
BI1	1.89 ± 0.32 c,d,e	$15.65\pm1.08~\mathrm{a}$	$11.77\pm0.83~\mathrm{d}$
BI2	1.77 ± 0.49 b,c,d,e	$15.24\pm2.03~\mathrm{a}$	$10.92\pm1.05~d$

Different letters within the same column indicate significant differences among genotypes (p < 0.05).

Previous studies conducted on cornelian cherry genotypes showed that yield, fruit weight, and fruit size are important parameters for cornelian cherry selection. Fruit weight is also the most distinct character which affects consumer behavior as well positively correlated with the percentage of mesocarp in the total fruit weight [8,22,24,26]. Considering this fact, the fruit of genotypes BK1, BK3, BK4, and BK5 could be interesting for the selection of promising cornelian cherries for use in future breeding programs in Bosnia and Herzegovina.

3.2. Biochemical and Bioactive Content

Results of biochemical and bioactive properties of cornelian cherry fruit are shown in Tables 2 and 3. There were statistically significant differences among genotypes in terms of all biochemical and bioactive content at the 0.05 level.

Genotypes	Dry Matter (%)	Water (%)	Ash (%)	рН	Total Sugar (%)	Total Acidity (%)
C1	$19.19\pm0.96~\mathrm{f}$	$80.82 \pm 4.04 \mathrm{d_{c}c}$	$0.82\pm0.04~\mathrm{a}$	3.23 ± 0.16 a	$11.3\pm0.57\mathrm{b}$	$1.27\pm0.09~\mathrm{d}$
C2	$17.66\pm0.88~{\rm g}$	82.33 ± 4.11 b,c	0.42 ± 0.02 c,d	3.09 ± 0.15 b,c	$11.88\pm0.59\mathrm{b}$	1.33 ± 0.09 d,e
C3	16.10 ± 0.81 g	83.83 ± 4.19 b,c	0.62 ± 0.03 b,c,d	3.21 ± 0.16 a	8.73 ± 0.44 d,c	$0.95\pm0.07~{\rm f}$
C4	$11.67\pm0.58\mathrm{\check{h}}$	88.32 ± 4.41 a	0.25 ± 0.01 c,d,e	$3.05\pm0.15~\mathrm{c}$	7.72 ± 0.39 d,e	$0.91\pm0.06~{\rm f}$
C5	$14.77\pm0.74~{ m g}$	85.22 ± 4.26 b,c	0.52 ± 0.02 c,d,e	3.17 ± 0.16 b,c	$8.36\pm0.59~\mathrm{d}$	$1.06\pm0.07~{\rm f}$
C6	15.12 ± 1.51 g	84.88 ± 8.48 b,c	0.44 ± 0.04 c,d	3.12 ± 0.31 b,c	8.67 ± 0.61 c,d	$0.97\pm0.09~{\rm f}$
C7	15.65 ± 1.57 g	84.33 ± 8.43 b,c	0.54 ± 0.05 c,d,e	3.14 ± 0.31 b,c	8.28 ± 0.58 c,d	1.09 ± 0.10 c,d
C8	17.75 ± 1.78 g	82.25 ± 8.22 c,d	0.42 ± 0.04 b,c,d	$3.07\pm0.31~{\rm c}$	$10.63\pm0.74\mathrm{b}$	1.06 ± 0.09 d,c
C9	17.68 ± 1.77 g	82.31 ± 8.23 c,d	0.44 ± 0.04 c,d	$2.85\pm0.29~d$	$12.18\pm0.85~\mathrm{a}$	$1.88\pm0.17~\mathrm{c}$
C10	21.33 ± 2.13 d,e	78.66 ± 5.50 c,d	$0.76\pm0.05\mathrm{b}$	$3.02\pm0.21~\mathrm{c}$	$12.54\pm0.88~\mathrm{a}$	$1.81\pm0.09~{\rm c}$
BU1	$11.92\pm1.19\mathrm{h}$	83.98 ± 5.87 b,c	0.31 ± 0.02 c,d,e	$3.03\pm0.21~\mathrm{c}$	9.42 ± 0.66 c,d	1.45 ± 0.07 d,e
BU2	$16.25\pm1.14~\mathrm{g}$	83.74 ± 5.86 b,c	0.57 ± 0.03 b,c	$3.02\pm0.21~\mathrm{c}$	9.64 ± 0.67 c,d	$1.62\pm0.08~\mathrm{e}$
BU3	$20.47\pm1.43~\mathrm{f}$	79.52 ± 5.56 b,c,d	0.48 ± 0.03 c,d	$2.94\pm0.21~\mathrm{d}$	$11.65\pm0.58\mathrm{b}$	2.92 ± 0.15 a
BU4	$20.09\pm1.41~\mathrm{f}$	79.91 ± 3.99 c,d	0.62 ± 0.03 b,c	$2.99\pm0.15~\mathrm{d}$	$9.53\pm0.48~\mathrm{c}$	$2.35\pm0.12b$
BU5	$20.69\pm1.45~\mathrm{e}$	79.31 ± 3.96 c,d	0.63 ± 0.03 b,c	$3.01\pm0.15~\mathrm{c}$	9.77 ± 0.98 c,d	$2.35\pm0.12b$
BK1	$20.36\pm1.62~\mathrm{f}$	79.75 ± 3.98 c,d	0.64 ± 0.03 b,c	2.97 ± 0.15 c,d	9.31 ± 0.93 c,d	$2.11\pm0.11~\mathrm{b}$
BK2	$21.89\pm1.75~\mathrm{a}$	78.11 ± 3.90 c,d	0.54 ± 0.02 b,c	$2.97\pm0.15~\text{c,d}$	$9.95\pm091~{\rm c}$	$2.17\pm0.10\mathrm{b}$
BK3	$21.37\pm1.71\mathrm{b,c}$	78.62 ± 3.93 c,d	0.61 ± 0.03 b,c	2.97 ± 0.15 c,d	$11.78\pm0.71~\mathrm{a}$	$2.17\pm0.08~b$
BK4	$19.26\pm1.54~\mathrm{f}$	80.73 ± 4.03 c,d	0.78 ± 0.03 b,c	2.98 ± 0.15 c,d	$9.81\pm0.59~\mathrm{c}$	$2.11\pm0.11~\mathrm{b}$
BK5	$21.84\pm2.18b$	78.16 ± 3.90 c,d	$0.68\pm0.03\mathrm{b}$	$3.02\pm0.15~\mathrm{c}$	$11.64\pm0.72\mathrm{b}$	$2.13\pm0.11~\mathrm{b}$
BI1	$14.31\pm1.43~\mathrm{g}$	$85.69\pm4.28\mathrm{b}$	0.49 ± 0.02 b,c,d	3.11 ± 0.16 b,c	6.82 ± 0.41 d,e	$1.67\pm0.08~\mathrm{e}$
BI2	18.36 ± 1.83 f,g	81.75 ± 4.08 c,d	0.84 ± 0.04 a	3.17 ± 0.16 b,c	8.55 ± 0.51 c,d	1.98 ± 0.13 c,d

Table 2. Chemical properties of cornelian cherry fruits.

Different letters within the same column indicate significant differences among genotypes (p < 0.05).

Dry matter content in cornelian cherry fruit in the literature ranged from 9.8 to 32.37% [8,9,19,21–28], and our results are in accordance with these results (11.67 to 21.89%). Several factors are responsible for the amount of dry matter in cornelian cherry fruits, including genetic background, growing conditions, plant age, climatic and soil properties, etc. [24].

Among vitamins, vitamin C is of special importance because the human body cannot synthase it. It is a vital vitamin for the strength of the immune system and plays an essential role in protecting against cardiovascular diseases [29]. When compared to the other vitamin-C-rich fruits, cornelian cherry fruits contain more vitamin C than fruits of kiwifruits, lemons, oranges, etc. [30]. In this study, Vitamin C content ranged between 25.58 and 58.75 mg/100 g FW among genotypes in previous studies [26] found vitamin C values ranging from 14.95 to 22.23 mg/100 g. Previous research conducted by [28] at genotypes in Montenegro showed that the average vitamin C in cornelian cherry fruits varied between 48–108 mg/100 g Ref. [8] found 31–70 mg/100 g; Ref. [31] found 24.0–36.0 mg/100 g; Ref. [32] found 25.36–44.06 mg/100 g; Ref. [33] found 19.9–43.3 mg/100 g in [25] found 29.0–103.3 mg/100 g in Turkey.

Cornelian cherry fruits are either used for fresh consumption or processed into several products without the application of heat treatment or low heat treatment. Thus the thermos labile substances, including vitamin C, remain preserved in the product [34].

Humans intake an important amount of minerals, biologically valuable molecules, with a diet mostly of plant origin [24]. In this study, the total amount of ash varied from 0.25% (C4) to 0.80% (C1). A previous study also indicated variation in ash content ranging from 0.51 to 1.13% in cornelian cherry fruits grown in Turkey [8]. The cornelian cherry genotypes sampled from Montenegro gave ash amounts of 0.89–1.16% [24].

Genotypes	C Vitamin (mg/100 g FW)	Anthocyanin (mg CGE/100 g FW)	Total Phenolic Content (mg GAE/100 g FW)	Phosphorus (mg/100 g FW)	Iron (mg/100 g FW)
C1	$29.89\pm1.49~\mathrm{e}$	197.16 ± 9.86 a	$4264 \pm 213.20 \text{ c}$	$228.87 \pm 11.44 \text{ c}$	$0.39\pm0.02~\mathrm{e}$
C2	$32.84 \pm 1.64~\mathrm{e}$	$186.27\pm9.31\mathrm{b,c}$	$2165\pm108.25~\mathrm{f}$	$157.36\pm7.87~\mathrm{f}$	$0.31\pm0.02~\mathrm{f}$
C3	$31.67\pm1.58~\mathrm{e}$	$88.4\pm4.42~\mathrm{f}$	1971 ± 98.55 f,g	$245.87 \pm 12.29 \ b$	$0.38\pm0.02~\mathrm{e}$
C4	$46.91\pm2.35~\mathrm{c}$	$79.5\pm3.98~\mathrm{f}$	$1621 \pm 81.05 \text{ g}$	$198.48\pm9.92~\mathrm{e}$	$0.35\pm0.02~\mathrm{e}$
C5	$29.17\pm1.75~\mathrm{e}$	55.57 ± 3.33 g	1285 ± 77.10 g,h	$175.15 \pm 10.51 \text{ g}$	$0.25\pm0.02~\mathrm{f}$
C6	$28.12\pm1.69~\mathrm{e}$	60.82 ± 3.65 g	1555 ± 93.33 g	$250.86 \pm 15.05 {\rm b}$	$0.47\pm0.03~\mathrm{d}$
C7	$34.38\pm2.06~\mathrm{e}$	57.9 ± 3.47 g	1717 ± 103.02 g	263.06 ± 15.78 a	$0.46\pm0.03~\mathrm{d}$
C8	$39.89 \pm 2.39 \text{ d}$	65.93 ± 3.96 g	1240 ± 74.49 h	$189.76 \pm 11.39 \mathrm{d_{e}}$	$0.28\pm0.02~\mathrm{f}$
C9	$50.94\pm3.06\mathrm{b}$	123.89 ± 7.43 e	1441 ± 86.46 g,h	$214.04 \pm 12.84 \text{ e}$	$0.30\pm0.02~{\rm f}$
C10	$31.53\pm2.21~\mathrm{e}$	$101.05\pm7.07~\mathrm{h}$	2051 ± 143.57 f,g	$225.20 \pm 15.76 \text{ c}$	$0.29\pm0.02~\mathrm{f}$
BU1	$32.74\pm2.29~\mathrm{e}$	$79.45\pm5.56~\mathrm{f}$	$1801 \pm 126.07 \text{ g}$	$188.93 \pm 13.22 \text{ d}$	$0.44\pm0.03~\mathrm{d}$
BU2	$40.34\pm2.82~d$	$119.7\pm8.38~\mathrm{e}$	3853 ± 269.71 d	$155.52\pm10.89~\mathrm{f}$	$0.55\pm0.04~\mathrm{c}$
BU3	$58.75\pm4.11~\mathrm{a}$	178.06 ± 12.46 b,c	$2241\pm156.87~\mathrm{f}$	$209.31 \pm 14.65 \text{ e}$	$0.57\pm0.04~\mathrm{c}$
BU4	$37.07 \pm 2.97 \text{ d}$	$157.69 \pm 12.62 \text{ d}$	$3681 \pm 294.48 \ d$	194.28 ± 15.54 d,e	$0.45\pm0.04~\mathrm{d}$
BU5	$30.14\pm2.41~\mathrm{e}$	$205.55\pm16.44~\mathrm{a}$	$2509\pm200.72~\mathrm{f}$	$177.36 \pm 14.19 \text{ g}$	$0.59\pm0.05\mathrm{b}$
BK1	$28.51\pm2.28~\mathrm{e}$	$128.88 \pm 10.31 \text{ e}$	$3416 \pm 273.28 \ { m e}$	199.13 ± 15.93 e	$0.55\pm0.04~\mathrm{c}$
BK2	$34.54\pm2.76~\mathrm{e}$	$194.88 \pm 15.59 \text{ b}$	$4056 \pm 324.48 \text{ c}$	$220.14 \pm 17.61 \text{ c}$	$0.52\pm0.04~{ m c}$
BK3	$39.32 \pm 3.15 \text{ d}$	$192.55 \pm 15.40 \text{ b}$	6958 ± 556.64 a	$187.35 \pm 14.99 \text{ d}$	$0.53\pm0.04~\mathrm{c}$
BK4	$34.75\pm3.48~\mathrm{e}$	$88.54\pm8.85~\mathrm{f}$	$2116 \pm 211.63 \text{ g}$	$202.39 \pm 20.24 \text{ e}$	$0.93\pm0.09~\mathrm{a}$
BK5	$31.3\pm3.13~\mathrm{e}$	176.46 ± 17.65 b,c	$6475 \pm 647.50 {\rm b}$	$197.48 \pm 19.75 \ \mathrm{e}$	$0.53\pm0.05~\mathrm{c}$
BI1	$25.85\pm2.59~\mathrm{e}$	$66.56 \pm 6.65 \text{ g}$	$1954\pm195.42~{\rm g}$	$205.85 \pm 20.59 \text{ e}$	$0.62\pm0.06~\mathrm{b}$
BI2	$15.73\pm1.57~\mathrm{f}$	$79.39\pm7.94~\mathrm{f}$	3318 ± 331.81 e	$246.60\pm24.66~b$	$0.53\pm0.05~\mathrm{c}$

Table 3. Vitamin C, anthocyanin, total phenolic, phosphorus and iron content of cornelian cherry fruits.

Different letters within the same column indicate significant differences among genotypes (p < 0.05).

Cornelian cherry fruits are rich in organic acids, and among organic acids, malic acid is predominant [9]. In the present study, the total acidity ranged from 0.91% in the C4 genotype to 2.91% in the Bu3 genotype. Our total acidity results were in agreement with the previous studies conducted in mostly Balkan and eastern Europe countries, including Serbia, Montenegro, Turkey, Romania, and the Czech Republic [21,23,25–28], which varied from 0.22% to 4.10%.

Cornelian cherries include plenty of sugars in their fruit and have a significant energy value. Consumers are directly affected by the sugar content of many fruits, and also sugar content strongly affects the fermentation processes during various types of processing [24]. When comparing the studies previously conducted in Bosnia and Herzegovina [9,32], our total sugar content results (6.82–12.54%) found similar to previous results (6.2–12.04%). However, Ref. [26] reported higher total sugar content (13.49–20.68%) in cornelian cherry fruits from Serbia and 10.94–14.47% in Montenegro [24].

Anthocyanins are colorful bioactive substances widely found in colored fruits and have an antioxidant effect on human health, preventing cardiovascular disease and cancer [35]. We found total anthocyanin content between 55.57 and 205.6 mg CGE/100 g FW, depending on the peel epidermis color, which varies from pink to dark red. Previously in Bosnia and Herzegovina [9], it was reported to be between 38.98 and 103.37 mg CGE/100 g FW, and in the Czech Republic [33] between 61 and 253 mg CGE/100 g FW). The other studies conducted in Turkey, Serbia, and Montenegro have reported lower values. For example, Ref. [23] reported 65 mg CGE/100 g FW. Ref. [26] reported 47.60–116.38 mg CGE/100 g FW. Ref. [28] reported 11.85–195.43 mg CGE/100 g FW, and Ref. [24] reported 32.54–157.06 mg CGE/100 g FW), respectively.

The other important bioactive substances found in fruits are phenolic compounds. It has been revealed that regular consumption of phenolic compounds reduces the risk of many human diseases [36]. Among the fruit species, cornelian cherry fruits are rich in phenolic substances, which contribute to the antioxidant value of cornelian cherry fruits [24]. In our study, the content of total phenolics differs from one genotype to another. and the highest content of total phenolics was determined in the Bk3 genotype (6958 mg GAE/100 g FW). The lowest total

polyphenols were found in the fruits of the C8 genotype (1240 CGE/100 g FW). In other studies, total phenols values are in range from 1191 to 2306 mg CGE/100 g FW [9] 1823 to 6143 mg CGE/100 g FW [33], 6878 mg CGE/100 g FW [23], 2663 mg CGE/100 g FW [37], 1580 to 5910 mg CGE/100 g FW [28]. In addition to the genetic constitution, the number of phenolic compounds in fruits is influenced by climatic and geographical factors and applied agro technics and fruit ripeness [24].

In our study, phosphorus content varied between 155.52 and 263.06 mg/100 g FW. which is lower than those reported by [27] in Czech (259.9–435.1 mg CGE/100 g FW and reported by [38] from Coruh valley in Turkey (280–350 mg CGE/100 g FW). After calcium, phosphorus is the most abundant mineral in the body. These two important nutrients work closely together to build strong bones and teeth [38,39].

When we observe the iron (Fe) content, twenty-two cornelian cherry genotypes have different results. The results have a variation between 0.25 and 0.93 mg/100 g. Fe is one of the necessary elements and plays an important role in oxygen and electron transfer in the human body. It is also vital for the normal functioning of the central nervous system [40].

3.3. Multivariate Data Analysis

Table 4 presents the results of the five main components of the PCA analysis, and Table 5 indicates a contribution of each of the 14 analyzed traits in the total variability. The variables with the highest values of eigenvectors in the first five main components are presented:

PCA1-dry matter, water, and anthocyanins;

PCA2—ash content, pH, and vitamin C;

PCA3—fruit length, fruit width, total sugars, and Fe;

PCA4—total phenols;

PCA5—fruit weight, total acidity, and phosphorus.

Table 4. Eigenvalues, the proportion of variance and cumulative variance associated with the first five main components (PCA) estimated from a 14-variable correlation matrix for 22 cornelian cherry genotypes.

Variables	PCA1	PCA2	PCA3	PCA4	PCA5
Eigenvalue	6.166	2.304	2.080	1.176	0.648
Proportion of variance (%)	44.046	16.459	14.852	8.399	4.6262
Cumulative variance (%)	44.046	60.505	75.357	83.756	88.383

Table 5. Eigenvalues are the proportion of variance and cumulative variance associated with the first five main components (PCA) estimated from a 14-variable correlation matrix for 22 cornelian cherry genotypes.

	PCA1	PCA2	PCA3	PCA4	PCA5
Fruit weight-FW	8.788	0.197	11.351	0.030	15.027
Fruit length-FL	9.656	0.042	13.813	0.019	0.331
Fruit width-FWW	8.118	2.530	12.000	8.821	3.829
Dry matter-DM	11.145	2.959	8.667	0.418	0.097
Water-W	11.897	2.036	7.877	0.314	0.351
Total ash-TA	3.670	26.414	0.824	0.144	0.095
pH	5.548	15.772	0.121	15.429	0.063
Total Sugars-TS	5.431	2.361	19.735	2.061	7.020
Vitamin C-VC	0.459	28.499	1.953	6.144	5.745
Total acidity-TAA	10.967	0.046	0.437	13.227	13.455
Anthocyanins-AA	9.690	0.341	4.676	6.769	3.011
Total phenols-TP	9.023	2.495	0.112	11.506	3.968
Phosphorus-P	1.337	13.732	1.322	19.048	42.280
Iron-Fe	4.270	2.576	17.111	16.071	4.727

The analysis of the obtained results shows that they are through the dominant eigenvectors in the first main component (PCA), which amounted to 44.05% of the total variance of the research. The most significant properties of the first component relate to the morphometric and chemical properties of cornelian cherry fruit. The highest values of eigenvectors were water and dry matter content (11.897 and 11.145). Anthocyanin content had lower values of eigenvectors (9.690). As part of the second main component, which amounts to (60.505%) of the total variability of the experiment, most of the properties with a high value for the eigenvector are related to the chemical composition of cornelian cherry fruits. The properties with the highest value of all of the eigenvectors from above are vitamin C content and total ash (28.499 and 26.414) (Table 4).

Due to the visualization of the level of statistical significance of the separation of individual genotypes and groups of cornelian cherry genotypes in the first three graphs (Figures 1–3), ellipsoids were set up where the tested cornelian cherry genotypes were classified into appropriate populations according to varietal affiliation with 95% confidence.



Figure 1. Grouping and relationships of 14 variables analyzed on 22 cornelian cherry genotypes, versus the first two main components (PC1 and PC2).



Figure 2. Grouping of 22 cornelian cherry genotypes concerning the first two main components (PCA1 and PCA2). calculated via correlation matrix for 14 characteristics.



Figure 3. Grouping of 22 cornelian cherry genotypes concerning the first two main components (PCA1 and PCA2), calculated via correlation matrix for 14 characteristics.

Figures 2 and 3 present the distribution of 22 cornelian cherry genotypes based on the first two main components by using 14 examined morphological and chemical characteristics. According to the first two components, which contain 60.50% of the total variability, twenty-two cornelian cherry genotypes were grouped mostly around the center of the coordinate system. Figure 3 also shows a partial degree of overlap between the analyzed cornelian cherry genotypes. This could be explained by the long growing period as there was an exchange of genetic material, perhaps also confirming the anthropogenic impact in terms of retention and spread of cornelian cherry genotypes in northwestern Bosnia.

Figure 4 shows the separation of the tested cornelian cherry genotypes. Graph 4 (biplot) also includes an analytical presentation of the original analyzed properties. From this graph, it is noticeable that there is a strong positive correlation between iron content, dry matter, total phenols, and fruit length. Then a positive correlation is seen between pH and phosphorus content. The presence of a negative correlation was noted between the characteristics (total acidity, anthocyanins, total sugars, fruit weight, and vitamin C).



Figure 4. Biplot 22 cornelian cherry genotypes separated based on morphological and chemical properties by analysis of major components. The original properties are shown as vectors where the direction indicates the value for each property, and the length indicates the level of variability.

Furthermore, a hierarchical cluster analysis was conducted (Figure 5). After the hierarchical cluster analysis, the existence of divergence of the examined cornelian cherry



genotypes is visible. The separation of all analyzed samples into three different clusters is visible.

Figure 5. Cluster analysis of the 22 selected genotypes on the basis of 14 quantitative characteristics.

The first-largest cluster (blue color) includes 11 cornelian cherry genotypes. In the second (yellow color) cluster, a total of three genotypes of cornelian cherry are classified, which are from the group Cazin (2) genotypes, and (1) genotype from Bihać. In the third cluster (grey color), 8 samples from a total of 22 cornelian cherry genotypes were classified. The number of cornelian cherry genotypes classified in the third cluster originates from the group Cazin (6), Bužim (1), and Bihać (1). In the first, the largest cluster is (11) genotypes from the group Bosanska Krupa (5) genotypes, (4) genotypes from the group Bužim and (2) genotypes from the group Cazin.

To identify promising cornelian cherry genotypes, i.e., those that are characterized by desirable characteristics, the average values of all monitored quantitative traits were analyzed. The results of the factor analysis are completely in agreement with the results of the obtained hierarchical cluster analysis, which classifies cornelian cherry genotypes into three separate clusters (Figure 6). In the right part of the coordinated system, cornelian cherry genotypes were grouped, which showed the highest average values of the examined characteristics, while in the left part, cornelian cherry genotypes, which were characterized by lower values of the examined characteristics, were grouped.



Figure 6. Arrangement of analyzed cornelian cherry genotypes into separate clusters using hierarchical cluster analysis over factor data analysis.

Previous studies are also indicating great diversity among seed propagated genotypes belonging to different horticultural plants [41–48].

4. Conclusions

Cornelian cherry fruits collected in natural populations are a valuable source of natural bioactive compounds and gene stock for breeding programs. Bosnian genotypes of cornelian cherry showed high content of C vitamin, total phenolic, anthocyanins, and Iron (Fe) content. Therefore, cornelian cherry fruits from natural populations could be considered a good source of natural antioxidants. The best balance of biochemical elements in combination with good yield and fruit size measurements is found in selections BK3 and BK4. The present study shows the potential value of cornelian cherry germplasm, and the results are useful in attempting to select superior genotypes for sustainable cornelian cherry production in Bosnia and Herzegovina.

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