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Different harvest times affect market quality of *Lycium barbarum* L. berries

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Abstract: Goji berry is a lesser-known fruit species and its berries have been used in traditional medicine for centuries as remedies to treat eye, liver, and kidney ailments, as well as for the prevention of cancer symptoms due to its high phytochemical content. This study aimed to investigate changes in the pomological, biochemical, and phytochemical properties of the berries of 4 goji berry genotypes harvested during 4 months (June, July, August, and September) of the same year. The results indicated that the highest berry dimensions (height and width) and soluble solid content were obtained from Genotype 2, harvested in June. The vitamin C, total phenolic, and total anthocyanin contents and 2,2-diphenyl-1-picrylhydrazyl-hydrate values were between 69.06 and 169.96 mg/100 mL, 219.07 and 558.74 mg GAE/L, 2.73 and 107.94 mg/L, and 75.03 and 500.87 µL/mL, respectively, according to the genotypes and harvest months based on a 3-year (2015–2017) average, indicating great variations in both the genotypes and, in particular, the sampling months. Therefore, consumers must take into consideration the harvest times and berries of the genotypes in order to obtain better contents in the berries for human health.

Key words: Goji berry, harvest time, genotype, bioactive content

1. Introduction

Goji berry is the common name of the species Lycium barbarum, Lycium chinense, and Lycium ruthenicum, belonging to the genus Lycium of the family Solanaceae, and it is known to occur more often in tropical and subtropical regions of the world (Levin and Miller, 2005). The plant is common in Mongolia, the Himalayas, western China, and Tibet (Bucheli et al., 2011), where it occurs in arid, semiarid, and slightly semisaline regions (Fukuda et al., 2001).

Worldwide, goji berry is also known as wolfberry. While *Lycium ruthenicum* has small black berries, *Lycium barbarum* has various colored berries, ranging from orange to dark red. These 2 species are cultivated commercially, with studies having been conducted more often on *Lycium barbarum*. Goji berry is a bush-type perennial plant, which is highly tolerant to climatic and environmental conditions, that is able to uptake necessary nutrients and water from the soil due to its advanced root system. Although *Lycium barbarum* and *Lycium ruthenicum* have similar morphological properties, specific morphological properties are obviously characteristic in distinguishing these 2 species (Wang et al., 2015).

In *Lycium barbarum*, the leaves are long and elliptical, pedicel is 1–2 cm long, calyx usually has 2 lobes that are 2- or 3-ribbed at the end, corolla tube is 8–10 mm long and longer than the lobes, and the berries are red or orange, yellow, rectangular, and oval. There are usually between 4 and 20 brown or yellow seeds per berry. *Lycium ruthenicum* is bushy like *Lycium barbarum* and it is rather branched. Leaves are fleshy, linear, or slightly cylindrical. Calyx is dispersed with 2–4 lobes, pedicel is 5–10 mm long, berries are spherical purple and black, sometimes its head or edge is notched, and the seeds are especially brown (Mi et al., 2015).

Goji berry plants are highly tolerant to adverse environmental conditions and grow at different altitudes ranging from 700 to 2700 m. In China, the plant is very popular and it has been consumed as a food for centuries due to its many benefits for health and its effect on longevity (Gündüz et al., 2015).

The berries, harvested from the beginning of summer to the end of autumn, are consumed either fresh or dried (Amagase and Farnsworth, 2011; Gundogdu et al., 2018). More recently, studies on the goji berries of the species *Lycium* have focused on potential health-benefitting

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contents such as polysaccharides, fatty acids, carotenoids, antioxidants, and minerals (Amagase and Farnsworth, 2011; Jin et al., 2013). The results have indicated that *L. barbarum* berries possess significantly more antioxidant values than many vegetables and fruits (USDA, 2016). Antioxidants protect cells and cell components against oxidative damages (Prior et al., 1999). *L. barbarum* berries contain major components like essential polysaccharides, carotenoids, and flavonoids. Dried goji berries contain 5% to 8% polysaccharides from water-soluble glycoconjugates, known as goji berry polysaccharides. This polysaccharide contains 6 different monosaccharaides, consisting of xylose, glucose, and lower concentrations of arabinose, mannose, rhamnose, and galactose (Amagase and Farnsworth, 2011; Tekin and Yılmaz, 2018). The dry berries also contain secondary major components like 0.03% to 0.5% carotenoids, where 56% of this carotenoid is composed of zeaxanthin, beta-cryptoxanthin palmitate, zeaxanthin monopalmitate, and beta-carotene (Inbaraj et al., 2008; Altındal and Altındal, 2018). The berries contain essential fatty acids like hexadecanoic, linolenic, and myristic acids. The third major component is phenolic acid, consisting of gallic, caffeic, protocatechuic, *p*-coumaric, and chlorogenic acids (Qian et al., 2004; Okatan, 2018; Usanmaz et al., 2018).

Oil can be extracted and tea can be brewed from the fruits and leaves of the plant, which can also be used to flavor and color chocolate, cakes, and salads. The daily dry fruit recommendation for adults is usually 20–30 g. In addition, because of its positive impacts on sun stains, scars, acne, and skin spots as well as antiaging, skin elasticity, and vitality, in addition to its antioxidants and vitamins, which also have an accelerating effect on cell renewal in the body, it has begun to be used as an auxiliary component in cosmetic products in recent years (Luo et al., 2004).

Goji berry was recently introduced to Turkey and its cultivation in Turkey has mostly been carried out with *Lycium barbarum* and partly with *Lycium chinense*, as well as the NQ series and hybrids of these species. Moreover, it has also been cultivated with seed-grown plants. Due to the different genetic backgrounds of seed-grown plants, remarkable differences have occurred both in the morphology of the plants and pomology of the berries. These differences cannot provide a harvest with the desired standards, which reduces the market value of the berries. More recently, commercial goji berry plantations have been established in Turkey and producers do not have enough information about the effects of different harvesting times on the content of the berries. Thus, the current study seems to be important in terms of informing producers about the pomological, biochemical, and phytochemical contents of the berries at different harvest times. No research exists focusing on the effect of different harvest times on goji berry quality, which makes the current study original.

2. Materials and methods

2.1. Materials

This study was carried out between 2015 and 2017 in the district of Çivril in the province of Denizli (38°11′10.87″N, 29°39′24.63″E). A commercial orchard was established with a drip irrigation system and 3-year-old goji berry plants (4 genotypes) with spacing distances within and between rows of 1.2 × 3.0 m. Berries were harvested from the plants in June, July, August, and September. Genotypes belonging to *Lycium barbarum* were used as the seed propagation material. They had different berry shapes and were named Genotypes 1–4. Climatic data for the district of Çivril in the province of Denizli are presented in Table 1. The study area, Çivril, has a warm and mild climate. There is much more precipitation in winter than in summer. According to the Köppen-Geiger system, the climate is Csb. The annual average temperature and precipitation for Çivril is 10.2 and 398 mm, respectively. A soil sample of 0–30 cm in depth was taken from the experiment area in 2015 and the analysis of the collected samples was performed by the Uşak Provincial Food and Agriculture Livestock Directorate Laboratory. Accordingly, the obtained analysis results are presented in Table 2.

2.2. Methods

Approximately 1 kg of berries per genotype was harvested during the 4 months and some pomological, biochemical, and phytochemical features were analyzed. Samples were taken by harvesting from the same plants in the first weeks of June, July, August, and September. Samples were taken in 3 replicates with 10 plants per replicate.

2.2.1. Determining pomological properties

From 2015 to 2017, the berry weights (g), widths (mm), and lengths (mm) were measured from 20 randomly selected fruits per genotype.

2.2.2. Determining biochemical properties

Biochemical properties (soluble solid content, pH, titratable acidity) of the goji berry samples were determined between 2015 and 2017.

2.2.2.1. Amount of soluble solids content (SSC)

The SSC of the berries was determined by distilling a sufficient amount of juice obtained by filtering through cheesecloth on the prism of digital refractometer.

2.2.2.2. pH

A total of 20 randomly selected fruits were collected from each genotype at harvest time and mashed in a cup to obtain fruit juice, and then the pH of the fruit juice was measured using a pH meter.

2.2.2.3. Amount of titratable acidity

In order to identify the fruit acidity, 20 randomly selected fruit samples were taken into consideration. A homogeneous fruit juice mixture was extracted from the fruits. Next, 10 mL of fruit juice and 20 mL of distilled

Table 1. Average temperature values for the district of Çivril.

Temperatures	Average °C	Minimum °C	Maximum °C
January	−0.7	−4.6	3.3
February	1.0	−3.3	5.4
March	4.6	−0.9	10.1
April	9.4	3.2	15.7
May	13.7	7.0	20.4
June	17.3	10.0	24.6
July	20.3	12.5	28.1
August	20.1	12.2	28.1
September	16.6	8.7	24.5
October	11.4	4.6	18.3
November	6.4	0.9	2.1
December	1.9	11.9	5.9

Source: www.climate-data.org.

Table 2. Soil properties of the experimental area.

Properties	Values	Status
pH	8.55	Strong alkaline
Salt (µm/cm)	379	Salt filter
Lime (%)	6.7	Medium lime
Organic matter (%)	0.20	Slightly
Structure	Tinned	Tinned
N (%)	0.010	Poor
P (ppm)	0.06	Very poor
K (ppm)	486	Too much
Ca (ppm)	7283	Too much
Mg (ppm)	740	Too much
Na (ppm)	154	Sufficient
Fe (ppm)	1.74	Inadequate
Cu (ppm)	7.76	Sufficient
Zn (ppm)	1.79	Sufficient
Mn (ppm)	10.96	Sufficient

water, at room temperature, were put into a beaker. The electrode of a digital manual pH meter was submerged into this mixture and it was mixed until the pH meter reached 8.1 (acid-base transformation point in the fruit juice), and then 0.1 N NaOH was added (Navarro et al., 2011). Next, all of the values were substituted in the following formula and the titratable acidity was expressed as a percentage of citric acid (Karacalı, 2002):

Percent of acidity: (used NaOH amount × 0.1 × 0.061/10 (10 mL of fruit juice)) × 10 Normality of the base: 0.1 mEq value of the citric acid: 0.061

2.2.3. Phytochemical properties

The vitamin C, total phenolic, and total anthocyanin contents and free radical scavenging activity (1,1-diphenyl-2-picrylhydrazyl, DPPH) were analyzed.

2.2.3.1. Vitamin C

The spectrophotometrical method was used in order to determine the amount of vitamin C. For the reduction of the coloring material, 2,6-dichlorophenolindophenol (2,6-D), ascorbic acid was utilized. For this purpose, a standard curve was obtained with the solutions prepared using oxalic acid, ascorbic acid, and 2,6-D as coloring material. First, 10 mL of fruit juice was extracted and diluted with oxalic acid 10 times; then it was drawn into 2 tubes, 1 mL for each, where one was mixed with 9 mL of distilled water while the other was mixed with 9 mL of the 2,6-D coloring material. The obtained solutions were read by a spectrophotometer at 518 nm. Absorbance values were substituted in the standard curve and the corresponding ascorbic acid amounts were found. Because the fruit juice was 10-fold diluted, the resulting value was multiplied by 10 and given as × mg/100 mL (Hışıl, 1993).

2.2.3.2. Determining total phenolic matter

The total phenolic amount was determined using Folin–Ciocalteu reagent as defined by Singleton and Rossi (1965). The fruit extract was mixed with Folin–Ciocalteu reagent and distilled water at a ratio of 1:1:18, left to rest for 8 min, and then added to 7% sodium carbonate. After 2 h of incubation in a dark room, the absorbance of the bluish solution was measured using a spectrophotometer at 750 nm. The results were calculated as mg GAE/L fresh weight basis.

2.2.3.3. Determining total anthocyanin

The total anthocyanin content in the fruit was determined using the absorbance values taken spectrophotometrically at different pH ranges according to Giusti et al. (1999). For the measurement of the diluted extracts, pH 1.0 (hydrochloric acid-potassium chloride) and pH 4.5 buffer solutions were prepared and measured at 531 and 700 nm. The total anthocyanin content (molar extinction coefficient of 28,000, cyanidin-3-glucoside) and absorbance [(A531 – A700) pH 1.0 – (A531 – A700) pH 4.5] were calculated for milligrams per 100 g fresh weight.

2.2.3.4. Free radical scavenging activity percentage (DPPH)

The percentage of free radical scavenging activity was conducted according to the method of Boskou et al. (2006) by determining the scavenging rate of the methanolic extracts in the samples of the DPPH radical. In this method, 100 µL of methanolic extract diluted at a 1/10 ratio was mixed with 3.9 mL of 6×10^5 M DPPH solution and left to rest for 30 min, and then the results were measured using a double-beam UV-Vis spectrophotometer at 515 nm, in 3 parallel runs.

2.3. Statistical analysis

The data obtained from the experiment, as the means of 3 replications, were analyzed using MINITAB (Minitab Inc., State College, PA, USA) and the averages were found by applying the Tukey test.

3. Results and discussion

3.1. Pomological values

The average berry lengths (mm), widths (mm), and weights (g) were determined from 2015 to 2017 and are presented as 3-year averages in Table 3. In general, the greatest berry lengths (mm), widths (mm), and weights (g) were obtained from Genotype 2 in June, followed by Genotype 1 in July. The biggest berries were obtained from Genotype 1 in August and Genotype 2 in September. The greatest berry lengths were obtained from Genotype 2 in June, Genotype 2 in July, Genotype 1 from August, and Genotype 1 in September. The greatest berry weights were obtained from Genotype 2 in June, Genotype 2 in July, Genotype 1 in August, and Genotype 2 in September.

Berry size is highly important in goji berry cultivation and fresh consumption. A few studies exist on the pomological properties of *Lycıum barbarum* in the literature. For example, Donno et al. (2015) reported a berry weight of 0.67 g, width of 8.15 mm, and length of 11.50 mm. In their study, Kafkaletou et al. (2018) reported average berry lengths among goji berry genotypes of between 0.42 and 0.86 g.

3.2. Biochemical properties

Biochemical properties such as the SSC, pH, and titratable acidity, analyzed based on the averages from 2015 to 2017,

Table 3. Fruit sizes according to the harvest times of the goji berry genotypes from 2015 to 2017.

Harvest time	Genotypes	Fruit length (mm)	Fruit width (mm)	Fruit weight (g)
June	Genotype 1	19.20 ab*	10.59 ab	1.04 b
	Genotype 2	21.50 a	11.17 a	1.32 a
	Genotype 3	13.56 cd	9.30 bc	0.62 d
	Genotype 4	13.23 cd	9.50 bc	0.46 d
July	Genotype 1	17.78 a	10.09 ab	1.00 c
	Genotype 2	19.91 ab	10.64 ab	1.27 a
	Genotype 3	12.55 c	8.86 bc	0.59 d
	Genotype 4	12.25 c	9.04 bc	0.45 d
August	Genotype 1	19.69 a	10.74 a	1.06 b
	Genotype 2	15.19 b	10.63 a	0.91 c
	Genotype 3	13.59 bc	9.16 bc	0.89 c
	Genotype 4	11.79 d	8.88 bc	0.58 d
September	Genotype 1	14.11 bc	8.40 cd	0.54 d
	Genotype 2	13.36 bcd	8.62 cd	0.61 d
	Genotype 3	12.16 cd	8.00 cd	0.39 d
	Genotype 4	12.37 cd	7.53 d	0.42 d

* Statistically significant differences ($P < 0.05$) among genotypes with different letters in the same column.

are presented in Table 4. The highest SSC was obtained in Genotype 1 (22.63%) in June, while the lowest was in Genotype 4 (13.90%) in September. The highest pH value was obtained in Genotype 4 (5.18%) in July, while the lowest was in Genotype 4 (3.95%) in June. The highest titratable acidity was determined in Genotype 2 (0.84%) in September, while the lowest was in Genotype 1 (0.36%) in June.

Our results were similar to those of Ban et al. (2015), who obtained SSC values of between 17.32% and 21.68% in goji berries. Jatoi et al. (2018) reported the average SSC as 18.5% in goji berries grown in Croatia. In the same study, the titratable acidity was between 0.90% and 0.93%. Donno et al. (2015) obtained a pH of 3.41, titratable acidity of 267.67 mEq/L, and SSC of 11.63%. In Greece, the majority of goji berry cultivars had SSC values ranging between 7.5% and 16.3% and titratable acidity between 2.2% and 8.3% (Drogoudi et al., 2017). In another study conducted in Greece, titratable acidity ranged between 0.3% and 0.4%, SSC varied from 21% to 25%, and the average pH was 5.35 in goji berries (Kafkaletou et al., 2017). In China, the SSC of mature goji berries ranged between 14.7% and 19.3%. The titratable acidity ranged between 0.8% and 2.7% (Zhang et al., 2015). Similar observations were found in other studies (Cantin et al., 2010; Kwon et al., 2015; Drogoudi et al., 2016). Although our results are in accordance with other studies, some variations were thought to have been due to the harvest time and differences among the genotypes.

3.3. Phytochemical properties

The vitamin C, total phenolic, and total anthocyanin contents and DPPH value averages from 2015 to 2017 are presented in Table 5. The highest vitamin C content in the analyzed genotypes was determined in Genotype 2 (169.96 mg/100 mL) in September, while the lowest was in Genotype 3 (69.06 mg/100 mL) in September. The highest total phenolic content was determined in Genotype 1 (558.74 mg GAE/L) in September, while the lowest was in Genotype 3 (219.07 mg GAE/L) in August. The highest total anthocyanin content was determined in Genotype 1 (107.94 mg/L) in August, while the lowest was in Genotype 3 (2.73 mg/L) in September. The highest DPPH value was obtained in Genotype 2 (500.87 µL/mL) in June, while the lowest was in Genotype 4 (75.03 µL/mL) in August .

More recently, some fruits, particularly berries including blueberries, raspberries, rosehip, blackberries, mulberries, and goji berries, have been accepted as super fruits or super foods. Super fruits have achieved common popularity in the past decade because of their high nutritive and bioactive properties (Jamin, 2009; Karp, 2012). In a study conducted in Italy on goji berries, the total polyphenolic compound

Table 4. SSC, pH, and titratable acidity averages from 2015 to 2017.

Harvest time	Genotypes	SSC%	pH	TA%
June	Genotype 1	22.63 a*	4.92 f	0.36 h
	Genotype 2	23.10 a	4.68 g	0.50 c
	Genotype 3	18.87 e	4.48 h	0.49 c
	Genotype 4	19.40 e	3.95 i	0.41 ef
July	Genotype 1	19.40 e	5.00 cd	0.42 e
	Genotype 2	17.94 f	5.11 b	0.42 e
	Genotype 3	20.83 cd	4.92 f	0.41 ef
	Genotype 4	20.27 d	5.18 a	0.40 fg
August	Genotype 1	20.77 d	5.05 c	0.39 g
	Genotype 2	21.70 b	4.97 def	0.39 g
	Genotype 3	21.80 b	4.98 de	0.37 h
	Genotype 4	21.50 bc	5.05 c	0.45 d
September	Genotype 1	17.13 g	4.92 f	0.44 d
	Genotype 2	17.83 fg	4.92 f	0.84 a
	Genotype 3	14.07 h	5.02 cd	0.49 c
	Genotype 4	13.90 h	4.93 ef	0.55 b

* Statistically significant differences ($P < 0.05$) among genotypes with different letters in the same column.

Table 5. Vitamin C, total phenolic (mg GAE/L), and total anthocyanin (mg/L) contents and DPPH (µL/mL) value averages.

Harvest time	Genotypes	Vitamin C (mg/100 mL)	Total phenol (mg GAE/L)	Anthocyanin (mg/L)	DPPH (µL/mL)
June	Genotype 1	118.95 bcd*	395.66 h	23.89 h	118.60 j
	Genotype 2	103.65 d	475.90 b	12.45 k	500.87 a
	Genotype 3	111.54 cd	325.54 l	10.78 l	255.43 f
	Genotype 4	143.46 abc	465.25 d	25.61 g	279.90 e
July	Genotype 1	128.40 bcd	448.89 e	88.00 b	144.82 i
	Genotype 2	109.53 cd	326.58 k	2.33 o	132.09 ij
	Genotype 3	121.12 bcd	468.89 c	16.35 j	224.10 g
	Genotype 4	55.88 e	272.31 o	45.50 c	80.77 k
August	Genotype 1	110.14 cd	310.22 m	107.94 a	240.57 fg
	Genotype 2	137.03 abcd	367.87 j	20.14 i	81.52 k
	Genotype 3	116.12 cd	219.07 p	41.50 d	451.69 b
	Genotype 4	134.17 bcd	370.21 i	32.29 f	75.03 k
September	Genotype 1	122.98 bcd	558.74 a	3.75 n	323.35 d
	Genotype 2	169.96 a	282.95 n	38.06 e	389.76 c
	Genotype 3	69.06 e	412.80 g	2.73 o	256.93 f
	Genotype 4	150.74 ab	426.30 f	5.42 m	185.77 h

* Statistically significant differences ($P < 0.05$) among genotypes with different letters in the same column.

content was recorded as 268.5 mg GAE/100 g FW, vitamin C content of different species of goji berry was 48.94 mg/100 g FW, and DPPH was 19.36% (Donno et al., 2015). The averages of the DPPH analyses and total phenolic content for goji berry extracts were reported as 40.6 mg Trolox/g dried fruit and 11.5 mg GAE/g dried fruit (Jeszka-Skowron et al., 2017). Endes et al. (2015) indicated that dried goji fruit water extract contained 3.44 mg GAE/100 mL of total phenolic content and its antioxidant activity in DPPH analysis was equal to 20.78%. In China, the total phenolic content ranged from 26.9 to 73.4 mg GAE/g FW and the DPPH free radical-scavenging activities of extracts from berries of the goji genotypes ranged from 36 to 85 µM TE/g FW (Zhang et al., 2015). In Greece, there were significant differences among *L. barbarum* genotypes in terms of antioxidant activity (Skenderidis et al., 2018). There are several reports available mentioning the high antioxidant activity levels in horticultural plants, varying from species to species and their maturity stages (Kähkönen et al., 2001; Ercisli et al., 2008a, 2008b; Serce et al., 2010; Sengul et al., 2011; Zia-Ul-Haq et al., 2014; Bendokas et al., 2018). Wang and Lin (2000) conducted experiments to compare the antioxidant activity in berry species and found the highest in strawberries, followed by black raspberries (*Rubus occidentalis* L.), blackberries (*Rubus* sp.), and red raspberries (*Rubus idaeus* L.). The differences between our study and the others were thought to derive from the different goji genotypes and harvest times. It was also thought that different applications in cultivation, such as fertilization or irrigation, affect the chemical contents of the fruit species.

3.4. Conclusions

Our results indicate that goji berry has rich contents in terms of nutritional elements, vitamins, and other components compared to other fruit and vegetable species, and it is a fruit with highly important increased value, both in terms of its contributions to human nourishment and its use in cosmetics. On the other hand, owing to its wide cultivation altitude range from 700 to 2700 m, its broad harvesting period from June to the end of October, and its tolerance to excessive hot and extreme cold, drought, and other environmental factors, goji berry is an alternative plant with high added value that can be cultivated in all regions of Turkey, except the coastal regions. In agreement with the abovementioned studies, we report that goji berry is a remarkable source of antioxidant compounds when compared to other fruits. Research supports its deep-colored fruits as potent antioxidant sources. Berries and dried fruit compose a relatively small part of the average diet, but they are important antioxidant sources. Highly pigmented berries have the highest antioxidant activity. Such fruits are rich in antioxidant compounds that are known for their enhanced stability and bioaccessibility. Based on our findings and the cited literature, we can suggest that goji berry be listed among red fruits that provide antioxidants.

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