SELECTION OF BLACK ELDERBERRY (*Sambucus nigra* L.) AND EVALUATION OF ITS FRUITS USABILITY AS BIOLOGICALLY VALUABLE FOOD

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To carry out domestication, five genotypes (B1, B2, R, VG and RK) were isolated from natural populations by selection of black elderberry (*Sambucus nigra* L.). The most prominent biological-technological characteristics were studied during fruit flowering and ripening. Flowering proceeded in the period from mid-April to the 1st decade of June, while ripening lasted from the 1st decade of August to the 1st decade of October. The largest inflorescences were found in genotype R (14.32 cm in diameter). The highest number of berries per inflorescence (on average, 280), the largest berry (on average, 0.21 g), and the best fruit quality (17.19% dry matter, 11.50% sugars, 35 mg vitamin C, and 2.58 g/l total colored matter) were all demonstrated by genotype VG. In all isolated genotypes fruit demonstrated biologically high quality, which makes it a good raw material desirable for diverse forms of processing. Its high yield level, pronounced resistance to disease...
agents and pests, as well as adaptability to adverse conditions are a recommendation for organic-based production.

Key words: black elderberry, berry, flowering period, inflorescence, fruit quality selection, ripening

INTRODUCTION

Quality human diet is impossible to imagine without fruits and fruit products.

However, in fruit production, in intensifying it, the problem of cultivar susceptibility to disease agents and pests is becoming increasingly important. This is the reason why fruit growing presupposes the application of high amounts of chemical substances (pesticides, fungicides, etc.), which diminishes fruit biological value.

Hence, domestication or introduction into culture of wild fruit crops can be considered one of the methods for improving fruits as food.

Black elderberry (*Sambucus nigra* L.) belongs to the species of the genus *Sambucus*, which grows in Serbia and Montenegro. It is a fruit crop of high quality fruit whose diverse cultivars are already grown on plantations in highly developed countries (USA, Switzerland, Denmark, Austria, etc.) (MÜLLER, 1976; STRAUSS & NOVAK, 1971; BROOKS, 1998). In Denmark its yield levels can be as high as 50 t/ha (McKAY, 2001).

Since black elderberry is encountered only in natural populations of Serbia and Montenegro, the aim of the present work was to select this fruit species, so as to grow and utilize its fruits as biologically valuable food. The isolated selections could be used (together with others) as bio-ameliorative species for bare soil forestation.

MATERIAL AND METHOD

Selection of black elderberry from spontaneous population was conducted in the period 1991-1993 at three locations: Belgrade – in Batajnica, Grocka – in Vinča, and sub-Kopaonik – in the villages of Velika Grabovica and Kriva Reka. The locations chosen differ in ecological and orographic characteristics.

Belgrade location of “Batajnica Woods” is situated at 75 m alt. and is a typical flat land of calcareous chernozem soil type, characterized by moderate continental climate. Grocka location – the village of Vinča-Radmilovac is at 125 m alt. on the slopes of the Danube River, with vertisol soil type in podzolization stage, characterized by moderate continental climate. Sub-Kopaonik location has typical mountain climate, where villages are situated on the slopes of Kopaonik Mt.: Velika Grabovica at 490 m alt. and Kriva Reka at 1400 m alt. with brown-acid soil type.
In the present paper the selection criteria used were as follows: uniformity of flowering, simultaneous ripening of berries in inflorescence, inflorescence and berry size, high yield level and resistance to disease agents and pests. According to determined criteria, individual positive selection was conducted by isolating and marking the genotypes.

Five genotypes were isolated and thereafter analyzed using standard methods:

- Physiological characteristics (period of fruit flowering and ripening) by registering the date of flowering initiation and termination, i.e. mean date of fruit ripeness.
- Physical characteristics of inflorescence and berry – by measuring length and weight in average sample comprising 30 inflorescences and 100 berries.
- Fruit chemical properties were determined on an average sample at the stage of full ripeness:
  1. dry matter - by refractometer;
  2. sugars – Bertrand method;
  3. total acids – by titrimetric method, calculations being done for malic acid content;
  4. juice pH value – by pH-meter
  5. total mineral matter – by roasting in a roasting furnace at 550°C;
  6. vitamin C – by iodine method;
  7. total colored matter – by measuring absorption on Beckman spectrophotometer.

Variability components for inflorescence characteristics were determined by monofactorial variance analysis (Jovanović et al., 1992). Coefficients of genetic and phenotypic variation ($CV_g$ and $CV_p$), as relative indicators of those characteristics variability, were found using method of Singh and Choudhary (1976). Coefficient of heritability in a broader sense ($h^2$) was calculated according to Borojević (1992) method as the ratio of genetic to phenotypic variance. Values of variability components and coefficient of heritability are expressed in percent.

RESULTS AND DISCUSSION

Black elderberry flowers after leafing. Of the isolated genotypes, the earliest flowering was recorded in genotypes from Batajnica – B1 and B2 and the latest in the genotype from Kriva Reka – RK (Tab. 1).

The analysis of the table indicates that there is a positive interdependence between flowering period and location altitude as well as a very short flowering period of all isolated genotypes, which complies with the results of Müller (1976) and Atkinson & Atkinson (2002).
Table 1. Average flowering and ripening period of isolated black elderberry (Sambucus nigra L.) genotypes

<table>
<thead>
<tr>
<th>No.</th>
<th>Genotype</th>
<th>Flowering period (date)</th>
<th>Ripening period (date)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>B1</td>
<td>15th – 20th April</td>
<td>around 10th August</td>
</tr>
<tr>
<td>2</td>
<td>B2</td>
<td>20th – 25th April</td>
<td>around 25th August</td>
</tr>
<tr>
<td>3</td>
<td>R</td>
<td>25th – 30th April</td>
<td>around 25th September</td>
</tr>
<tr>
<td>4</td>
<td>VG</td>
<td>25th – 30th May</td>
<td>around 27th September</td>
</tr>
<tr>
<td>5</td>
<td>KR</td>
<td>05th – 10th June</td>
<td>around 5th October</td>
</tr>
</tbody>
</table>

Positive interdependence is also noticeable between ripening and flowering period. The earliest ripening was observed in genotypes B1 and B2 (Aug. 10 and 25) and the latest in genotypes from Kriva Reka (around Oct. 5).

Tab. 2 shows physical characteristics of inflorescences and berries. The data given are average values for the sample of 30 inflorescences, measurements being done during fruit ripening period.

The analysis of the table evidences that the largest inflorescences were exhibited by genotype R (14.32 cm average width and 15.88 cm height). The lowest inflorescence width was demonstrated by genotype KR (10.67 cm), while genotype B2 had the lowest inflorescence height (7.28 cm). The data are in agreement with those reported by GRLIĆ (1989) for the region of Croatia, but they approximate the lower limit of inflorescence size variation, as reported by HEGI (1961).

In studied genotypes the length of inflorescence stem ranged from 5.14 cm (genotype B1) to 7.71 cm (genotype VR), and the number of primary stalks from 4.20 cm (genotype B1) to 5.28 cm (genotypes VG and KR).

The number of berries per inflorescence is noticed to vary a lot per studied genotype - from 131 in genotype KR to 280 in genotype VG. In wild black elderberry, studied in Poland, the number of berries per inflorescence ranged from 100-220, whereas cultivars had between 143 and 380 berries per inflorescence (WAŹBIŃSKA & PUCZEL, 2002).

In genotype VG average berry weight is also very pronounced (0.21 g), while in all other studied genotypes it falls within the limits average for non-selected elderberry (JANDA, 1980). JOHNSON et al. (1985) reported that average fruit weight in wild elderberry was 0.0784 g. WAŹBIŃSKA & PUCZEL (2002) found that berry weight in wild elderberry ranged from 0.132 – 0.190 g, and in grown cultivars from 0.176 – 0.33 g. ATKINSON & ATKINSON (2002) reported that berry weight of black elderberry picked from 20 locations in Great Britain amounted to 0.118 g.
Table 2. Inflorescence and berry characteristics of isolated black elderberry (Sambucus nigra L.) genotypes

<table>
<thead>
<tr>
<th>No.</th>
<th>Property</th>
<th>Inflores width (cm)</th>
<th>Inflores height (cm)</th>
<th>Length of infloresc stem (cm)</th>
<th>Number of primary stalks</th>
<th>Number of berries per inflorescence</th>
<th>Berry weight (g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>B1</td>
<td>12.53</td>
<td>8.01</td>
<td>5.14</td>
<td>4.20</td>
<td>242</td>
<td>0.15</td>
</tr>
<tr>
<td>2</td>
<td>B2</td>
<td>11.44</td>
<td>7.28</td>
<td>5.33</td>
<td>4.88</td>
<td>210</td>
<td>0.17</td>
</tr>
<tr>
<td>3</td>
<td>R</td>
<td>14.32</td>
<td>15.88</td>
<td>6.46</td>
<td>5.04</td>
<td>146</td>
<td>0.13</td>
</tr>
<tr>
<td>4</td>
<td>VG</td>
<td>12.50</td>
<td>15.26</td>
<td>7.71</td>
<td>5.28</td>
<td>280</td>
<td>0.21</td>
</tr>
<tr>
<td>5</td>
<td>KR</td>
<td>10.67</td>
<td>14.47</td>
<td>5.56</td>
<td>5.28</td>
<td>131</td>
<td>0.15</td>
</tr>
</tbody>
</table>

LSD 0.05 0.933 0.481 0.236 0.251 33.50
LSD 0.01 1.368 0.704 0.346 0.367 49.10

Analysis of variance and individual LSD-tests evidenced very significant differences in inflorescence characteristics between studied genotypes (Tab. 2). Since phenotypic value of a certain characteristic is a sum of genetic effects and those of the environment, it is important to know which part of phenotypic values is conditioned by hereditary basis and which one by environmental conditions. Comparisons of all studied inflorescence characteristics (Tab. 3) showed that inflorescence height had the highest coefficient of both genetic and phenotypic variation (CV_g=34.27%; CV_f=34.32%), however, the lowest number of primary stalks in inflorescence (CV_g=1.75%; CV_f=1.96%).

Table 3. Coefficients of genetic (CV_g), phenotypic variation (CV_f) and coefficients of heritability in a broader sense (h^2) for inflorescence characteristics in isolated black elderberry (Sambucus nigra L.) genotypes

<table>
<thead>
<tr>
<th>Property</th>
<th>CV_g (%)</th>
<th>CV_f (%)</th>
<th>h^2 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Length of infloresc. stem (cm)</td>
<td>17.53</td>
<td>17.88</td>
<td>95.98</td>
</tr>
<tr>
<td>Infloresc. height (cm)</td>
<td>34.27</td>
<td>34.32</td>
<td>98.73</td>
</tr>
<tr>
<td>Infloresc. width (cm)</td>
<td>2.15</td>
<td>2.18</td>
<td>97.36</td>
</tr>
<tr>
<td>Number of primary stalks</td>
<td>1.75</td>
<td>1.96</td>
<td>79.99</td>
</tr>
<tr>
<td>Number of berries per infloresc.</td>
<td>6.12</td>
<td>6.12</td>
<td>98.99</td>
</tr>
</tbody>
</table>
In studied inflorescence characteristics the values for coefficient of heritability are markedly high. The highest value was found for the number of berries in inflorescence (98.99%) and the lowest for the number of primary stalks in inflorescence (79.99%). Such values of coefficients of heritability for inflorescence characteristics evidence small effects of the environment on manifesting those characteristics. In addition, they indicate that it is possible to improve those characteristics by selection and employ studied genotypes in future breeding activities.

Of chemical properties, studies were conducted on the most important parameters which determine fruit quality – contents of dry matter, sugars, total acids, pH value, total mineral matter content, vitamin C and total colored matter (Tab. 4).

Dry matter content varied between 14.80% (in genotype B2) and 17.19% (in genotype VG). Comparisons of those data with the ones in literature indicate that dry matter content in genotypes B1, B2 and KR falls within the limits average for non-selected elderberry (MRATINIĆ & KOJIĆ, 1998), however, dry matter content in genotypes R and VG is slightly higher than mentioned.

Table 4. Fruit chemical properties of isolated black elderberry (Sambucus nigra L.) genotypes

<table>
<thead>
<tr>
<th>No.</th>
<th>Property</th>
<th>Dry matter (%)</th>
<th>Total sugar content (%)</th>
<th>Invert sugar content (%)</th>
<th>Organic acids content (%)</th>
<th>pH value</th>
<th>Mineral matter content (%)</th>
<th>Vitamin C content (mg%)</th>
<th>Total colored matter content (g/l)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>B1</td>
<td>15.20</td>
<td>8.90</td>
<td>6.20</td>
<td>1.15</td>
<td>4.00</td>
<td>0.90</td>
<td>20.00</td>
<td>1.39</td>
</tr>
<tr>
<td>2</td>
<td>B2</td>
<td>14.80</td>
<td>9.20</td>
<td>5.70</td>
<td>1.00</td>
<td>4.10</td>
<td>0.98</td>
<td>23.00</td>
<td>1.67</td>
</tr>
<tr>
<td>3</td>
<td>R</td>
<td>16.75</td>
<td>11.20</td>
<td>5.60</td>
<td>1.03</td>
<td>4.10</td>
<td>0.91</td>
<td>27.00</td>
<td>1.41</td>
</tr>
<tr>
<td>4</td>
<td>VG</td>
<td>17.19</td>
<td>11.50</td>
<td>6.78</td>
<td>1.04</td>
<td>4.10</td>
<td>0.99</td>
<td>35.00</td>
<td>2.58</td>
</tr>
<tr>
<td>5</td>
<td>KR</td>
<td>15.89</td>
<td>9.85</td>
<td>5.39</td>
<td>1.09</td>
<td>4.00</td>
<td>0.90</td>
<td>30.00</td>
<td>2.31</td>
</tr>
</tbody>
</table>

On the other hand, total sugars content ranging from 8.90% (genotype B1) to 11.50% (genotype GV) is higher only in genotypes R and VG than average for non-selected elderberry.

Organic acids content is fairly uniform and ranges from 1.03% (genotype R) to 1.15% (genotype B1). Those results are in agreement with the ones reported by MÜLLER (1976) and are at the average level for non-selected elderberry, as pointed out by MRATINIĆ & KOJIĆ (1998).

Like acids content, pH value is very uniform and varies from 4.00 (in genotypes B1 and KR) to 4.10 (in genotypes B2, R and VG). The data obtained for this parameter are slightly higher than those reported by MÜLLER (1976) for black elderberry cultivar Don, where pH value is 3.80.

Black elderberry fruit is very rich in total mineral matter, which is confirmed by the results obtained for the studied genotypes. Mineral matter content varies slightly depending on the genotype – from 0.90% (in genotypes B1
and KR) to 0.99% (in genotype VG) and complies with the reports of a majority of authors (GRLIĆ, 1989; MÜLLER, 1976; MRATINIĆ & KOJIĆ, 1998 and others).

Vitamin C content, a very important biological value parameter of elderberry fruit quality, ranges from 13.00 mg% (genotype B2) to 35.00 mg% (genotype VG). Comparisons between data indicate positive interdependence of this parameter and altitude of location where genotypes come from. Our data coincide with reports of KAACK & AUSTED (1998) who found that average vitamin C content in 13 studied cultivars was from 6-25 mg%.

Comparisons of our results with those from literature allow us to conclude that in all isolated elderberry cultivars this parameter is either higher (BUKVIĆ, 1995) or at the level average for non-selected elderberry (MRATINIĆ & KOJIĆ, 1998).

Total colored matter content varied between 1.39 g/l (genotype B1) and 2.58 g/l (genotype VG). It can be stated that in all studied genotypes this content is markedly high, because compared to the most colored fruit of small fruits – blackberry – it is higher several times (ANDROS, 1941). According to NAKAJAMA et al. (2004), there are four types of anthocyanins in elderberry and their concentration amounts to 635-1969 mg% (ATKINSON & ATKINSON, 2002). The USDA report (2003) indicated that in raw elderberry fruit anthocyanidin content ranged from 379.15 to 1067.33 mg%, while flavonol content was between 29 and 60 mg%.

Lastly, considering all analyzed chemical parameters of elderberry fruit quality, it can be concluded that black elderberry fruit falls into the category of biologically high-quality fruits (BULATOVIĆ & NIKETIĆ, 1974), therefore it could be processed into various products, such as preserves, stewed fruit, syrups, juices, jams etc.

This statement and the fact that isolated genotypes are high-yielding, extremely resistant to disease agents and pests, but of modest demands for cultural practices, all this recommend them for organic-based production.

CONCLUSION

The analysis of biological-technological characteristics of 5 selected black elderberry (Sambucus nigra L.) genotypes leads to the conclusions as follows:

1. Flowering proceeded in the period from mid-April (genotype B1) to the 1st decade of June (genotype KR).
2. Fruit ripening lasted from the 1st decade of August (genotype B1) to the 1st decade of October (Genotype KG).
3. Positive interdependence was perceived between fruit flowering and ripening period.
4. Genotype R demonstrated the largest inflorescences whose average breadth was 14.32 cm and average height 15.88 cm.
5. Genotype VG exhibits conspicuously high number of berries, having 280 berries per inflorescence.
6. Analysis of variance and individual LSD-tests indicated very significant differences in inflorescence characteristics between studied genotypes.

7. The highest coefficients of genetic and phenotypic variation (CV\(_g\)=34.27%; CV\(_f\)=34.32%) were found for the characteristic of inflorescence height, while the highest value of coefficient of heritability was for the number of berries in inflorescence (98.99%).

8. All isolated genotypes demonstrated biologically high quality fruit, so they can be used for various forms of processing.

9. Genotype VG demonstrated best quality fruit for all analyzed chemical parameters (17.19% dry matter, 11.50% total sugars, 10.4% organic acids, 0.99% total mineral matter, 35 mg% vitamin C, and 2.58 g/l total colored matter).

10. All isolated genotypes can be recommended for growing in organic-based production, because they are yielding, resistant to disease agents and pests and of very modest cultural practices requirements.

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Selekcija crne zove (Sambucus nigra L.) i ocena upotrebljivosti plođova kao biološki vredne hrane

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IZVOD

Selekcijom crne zove (Sambucus nigra L.) iz prirodnih populacija izdvojeno je 5 genotipova (B₁, B₂, R, VG i RK), u cilju njihove domestifikacije. Kod izdvojenih genotipova proučene su najvažnije biološko-tehnološke osobine i vreme cvetanja zrenja ploda. Cvetanje se odvijalo u periodu polovine aprila-I dekade juna, a zrenje od I dekade avgusta do I dekade oktobra. Najkrupnije cvasti nađene nađene su kod genotipa R (sa prečnikom od 14,32 cm). Najviše bobica po cvasti (prosečno 280), najkrupnija bobica (prosečno 0,21 g) i najbolji kvalitet ploda (sa 17,19% suve materije, 11,5% šećera, 35 mg vitamina C i 2,58 g/l ukupnih bojenih materija) ispoljio je genotip VG. Kod svih izdvojenih genotipova plod je ispoljio biološki visok kvalitet, što ga preporučuje kao dobru sirovinu za različite vidove prerade. Njihova dobra rodnost, izražena otpornost prema prouzrokovanoj bolesti i štetočina, kao i adaptivnost na lošije uslove, preporučuje ih za gajenje po principu organske proizvodnje.