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MORPHOLOGICAL CHARACTERISTICS OF FRUITS, DRUPES AND SEEDS IN GENOTYPES OF *ZIZIPHUS JUJUBA* MILL.

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ABSTRACT

The aim of this work is to discriminate 28 *Ziziphus jujuba* Mill. genotypes (ZJ-01 – ZJ-28) based on weight and morphological characteristics of fruit, stones and seeds. These genotypes maintained in a clone repository (Nova Kachovka, Ukraine) were chosen for investigation as potential species for cultivation and use in Slovakia. The fruits were collected at the period of their full maturity (October); their morphometric parameters were following: weight from 2.90 g (ZJ-13) to 28.99 g (ZJ-22), length from 20.73 mm (ZJ-08) to 44.84 mm (ZJ-04), and width from 16.64 mm (ZJ-13) to 38.87 mm (ZJ-22). Isolated stones were also analysed, and their weight was from 0.173 g (ZJ-07) to 0.731 g (ZJ-20), length from 12.84 mm (ZJ-08) to 28.67 mm (ZJ-04), and width from 5.06 mm (ZJ-07) to 9.74 mm (ZJ-01). The weight of the seeds was ranging from 20.00 mg (ZJ-25) to 69.11 mg (ZJ-12), length from 7.24 mm (ZJ-15) to 11.85 mm (ZJ-04), and width was from 3.86 mm (ZJ-05) to 6.84 mm (ZJ-12). Significant differences in the size, shape and color of fruits, stones and seeds were also identified. The pulp, seedless stones and seeds constitute 92.81% (ZJ-26) - 98.32% (ZJ-04), 1.36% (ZJ-04) - 6.27% (ZJ-20), and 0.25% (QA-24, QA-25) - 1.21% (ZJ-16) of the total fruits weight, respectively The shape index of the fruits was found in the range of 1.04 (ZJ-08) to 2.00 (ZJ-12 and ZJ-25). The shape indexes of stones and seeds were found ranging from 1.74 (ZJ-08) to 4.53 (ZJ-05) and from 1.32 (ZJ-23) to 2.74 (ZJ-04), respectively. The relative weight of pulp was determined in the range from 92.76% (ZJ-13) to 98.32% (ZJ-04) and the relative weight of seed was determined in the range of 0.25% (ZJ-24 and ZJ-25) to 1.16% (ZJ-26). The Chinese jujube genotypes of this study were grouped based on hierarchical clustering of the fruit, stone and seed characteristics.

Keywords: Chinese jujube variability; morphological characteristics; fruit; stones; seed

INTRODUCTION

The genus *Ziziphus* has approximately 40 species **(Li et al., 2007)** that has been cultivated for about 4,000 years in China and grown in this country for over 150 years **(Bonner and Rudolf, 1974; Lyrene, 1979; Mowry et al., 1953)**. Among these species, the most known one named *Ziziphus jujuba* Mill. is a thorny rhamnaceous plant **(Li et al., 2007)** and is mainly cultivated in China and grown to some extent in Russia, northern Africa, southern Europe, the Middle East, and the south-western United States **(Shanmugavasan et al., 2011)**. Its common name is Chinese date or Chinese jujube. In older European texts it may be also called the 'common jujube'. It tends to be a more upright tree with bright green, glabrous leaves. The tree is deciduous, losing its leaves in winter; it flowers in the spring, and the fruits mature in autumn. It is resistant to winter cold **(Guijun and Ferguson, 2010)**. It is a hardy tree of arid region, which can be grown successfully in saline soil under hot, arid environment **(Meena et al., 2003)**. Its fruit is an edible oval drupe with single hard stone **(Nowak a Shulzová, 2002; Azam-Ali et al., 2006)**.

Jujube cultivars vary in size and morphology. After 30 years of growth in an average site, trees of most cultivars will be 40-50 feet tall with a crown diameter of 15-20 feet **(Lyrene and Crocker, 1994)**. Trees bear fruit as early as 1 to 4 years after planting **(Lyrene, 1979)**. Often producing some fruit the second year from seed or grafting **(Lyrene and Crocker, 1994)**. Currently, there are 700 to 800 jujube cultivars in China, including fresh eating, drying, multipurpose (good for both drying and fresh eating), candied, and ornamental. Cultivars for drying, including multipurpose cultivars, formerly dominated and accounted for 90% of the jujube production in China **(Yao, 2012)**. Annual fruit production in China is 450 000 long tons produced on 290 000 hectares **(Hache a Xu, 1995)**. *Zizyphus jujuba* Mill. 'Huizao' is the most commonly planted cultivar in China, with the largest orchard area and the highest annual production **(Shanmugavasan et al., 2011)**. Fruits, leaves, seeds and other parts of *Z. jujuba* plant contain many important biologically active substances. Therefore, their physiotherapeutic effects are used to treat many diseases such as liver disease, fever, sore throat, pharyngitis, bronchitis, diabetes, skin infections, loss of appetite, diarrhea. It is also used for digestive disorders, weakness, urinary problems, and insomnia **(Khare, 1995, Huang et al., 2007, Huang et al., 2008, Adzu et al., 2001, Han and Park, 1986, Kirtikar and Basu, 1984, InAs et al., 2008)**. Alleviates brain nerve disorders **(Heo et al., 2003)**, regulates immune function **(Zhao et al., 2008)** and it's used to reduce blood triglycerides **(Kim, 2002)**. It has anti-cancer **(Lee et al., 2003)**, antifungal, antibacterial, anti-inflammatory **(Al - Reza, et al., 2010, Huang et al., 1990)**, hypotonic, antioxidant **(Zhang et al., 2000, Chang et al., 2010)** and immunostimulatory properties **(Al - Reza**

et al., 2010). It is also used in wound healing **(Ansari et al., 2006)**.

The aim of this work is to determine the differences in basic morphological characteristics of fruit, drupes and seeds between Chinese jujube genotypes, as potential species for cultivation and use in Slovakia.

MATERIAL AND METHODOLOGY

In our experiments, we used the stone fruits (drupes) of 28 Chinese jujube genotypes maintained in a clone repository (Nova Kachovka, Ukraine). The fruits were collected at full maturity (October). At average of 15 fruits, stones and seeds of each genotype were chosen for morphological analysis. The weight (g/mg), height (mm) and width (mm) were determined for all of themusing sliding scale and Axio Vision Rel. 4.8 software **(Carl Zeiss MicroImaging GmbH, Germany)**. Variability of all these parameters was evaluated using descriptive statistics Differences between the genotypes in morphological characteristics were evaluated by analysis of variance and Tukey test. The Pearson correlation coefficients between the selected parameters were calculated. Similarities and/or dissimilarities between genotypes were evaluated by Cluster analysis based separately on fruit, stone and seed data respectively. Statistica 10 software **(StatSoft Inc., USA)** was used to perform all mentioned statistical analyses.

RESULTS AND DISCUSSION

General fruit characteristics

The weight of the whole fruit is one of significant production characteristics of plant species. Further important features of the fruit are its shape, size and color. These parameters of the jujube fruit varied significantly. The fruits were round, oval, apple-, egg- or pear-shaped, etc. Fully mature fruits have brownish-yellow, golden-yellow or reddish to dark brown color. There are also brown spots on the fruit. The images of jujube fruits of various genotypes are shown in Figure 1. High variability of the size, shape and color of these fruits are evident. The fruit pulp also demonstrated varying consistency and different colors (Figure 2).

Fruit weight

As reported by many authors **(Kundi et al., 1989; Gao et al., 2003; Liu et al., 2003; Prasad, 2005; Jiang et al., 2006)**, the average fruit weight of Chinese jujube is usually between 10 and 30 g. **Gao et al., (2012)** determined the fruit weight in the range of 6.7 to 26.7 g, **Karnatovska et al., (2007)** identified in the evaluation of 23 varieties of Chinese jujube in extreme agro ecological conditions in Nova Kachovka (Ukraine), the average weight of the fruit in the range of 1.0 to 9.5 g. **Brindza et al., (2011)** determined the average weight of the fruit in the range of 0.66 to 4.68 g. **Sivakov et al.,** (1988) identified the fruit weight from 5.72 to 10.45 g, **Ecevit et al., (2007)** determined the average fruit weight of 4.52 to 6.12 g. On the other hand, **Guijun and Ferguson, (2010)** determined

Figure 1 Fruits of selected Chinese datle (*Ziziphus jujuba* Mill.) genotypes.

Figure 2 Fruit cross sections of selected Chinese jujube (*Ziziphus jujuba* Mill.) genotypes.

the fruit weight of a certain genotypes to be more than 50 g.

The weight of jujube fruits of present study was in the range of 2.90 to 28.99 g (Table 1). These values are in agreement with those in many reports. The variation coefficient CV characterizes average degree of variability within the tested collection; genotypes reaching extreme values are listed in Table 2.

Fruit length

The jujube fruit reaches the length from 4 to 6 cm. The fruits of the original (wild) forms reach the size of only about 2.5 cm. The pulp is irregularly grooved **(Nowak and Shulzová, 2002, Keys, 1976 Polivka, 2010)**. The given shapes were also determined in genotypes in our collection (Figure 1). The shape of the fruit is conditioned by its height and diameter. The fruit length in our analyses was determined in the range of 20.73 mm (ZJ-08) to 44.84 mm (ZJ-04) (Table 1). The value of the coefficient of variation was 6.11%, which documented low degree of variability of the character within the collection. **Brindza et al., (2011)**

Table 1 The variability of weight and linear parameters of fruits, stones and seeds for the whole collection of Chinese jujube genotypes from the clone repository in Nova Kachovka.

Characteristic	Unit	*min*	*max*	*mean*	*SD*	*CV*
Fruit weight	**G**	2.90	28.99	10.83	0.45	17.93
Stone weight	**G**	0.17	0.73	0.38	0.02	20.34
Seed weight	**mg**	20.00	69.11	48.53	2.69	15.12
Fruit length	**mm**	20.73	44.84	33.68	0.42	6.11
Stone length	**mm**	12.84	28.67	20.62	0.53	5.56
Seed length	**mm**	7.24	11.85	9.74	0.12	3.85
Fruit width	**mm**	16.64	38.87	24.27	0.36	7.07
Stone width	**mm**	5.06	9.47	7.18	0.19	5.82
Seed width	**mm**	3.86	6.84	5.30	0.09	3.95

Legend: *min*, *max* – minimal and maximal values measured, *mean* - arithmetic mean, *SD* - standard error of mean, *CV* - coefficient of variation (%)

Table 2 The fruit/stone/seed weight variability for specific Chinese jujube genotypes from the collection of clone repository in Nova Kachovka.

Genotypes	*mean*	*SD*	*CV*	Genotypes	*mean*	*SD*	*CV*
Fruit weight (g)							
Lowest values				**Highest values**			
ZJ-13	**2.90**	0.08	17.27	**ZJ-22**	28.99	1.14	14.21
ZJ-16	**3.31**	0.08	14.13	**ZJ-17**	26.28	1.95	31.48
ZJ-14	**4.30**	0.10	13.77	**ZJ-04**	18.43	0.72	20.68
ZJ-26	**4.31**	0.07	11.81	**ZJ-02**	18.25	1.22	20.01
ZJ-08	**4.46**	0.11	14.02	**ZJ-10**	15.67	0.60	20.46
Stone weight (g)							
Lowest values				**Highest values**			
ZJ-05	**0.17**	0.01	15.21	**ZJ-20**	0.73	0.03	17.21
ZJ-07	**0.17**	0.01	17.92	**ZJ-11**	0.64	0.03	29.15
ZJ-19	**0.19**	0.01	14.55	**ZJ-10**	0.61	0.04	29.17
ZJ-25	**0.20**	0.00	10.37	**ZJ-03**	0.59	0.03	25.61
ZJ-27	**0.20**	0.01	20.24	**ZJ-12**	0.58	0.03	23.57
Seed weight (mg)							
Lowest values				**Highest values**			
ZJ-25	**20.00**	1.08	16.20	**ZJ-12**	69.11	3.49	15.17
ZJ-05	**25.25**	3.30	26.15	**ZJ-28**	61.50	2.35	12.10
ZJ-07	**26.00**	2.38	25.84	**ZJ-04**	61.00	6.00	13.91
ZJ-13	**34.22**	2.44	21.36	**ZJ-21**	61.00	2.00	4.64
ZJ-24	**42.50**	4.50	21.18	**ZJ-18**	59.57	5.38	23.89

Legend: *mean* - arithmetic mean; *SD* - standard error of *the mean*; *CV* - coefficient of variation (%)

Table 3 Correlation between the fruit weight (g) and other morphological characteristics of evaluated fruits, stones and seeds shown with Pearson coefficients of correlation

Characteristic	Unit	*r*	*sr*	Confidence interval		r^2		*p*
				min	*max*			
Fruit length	**mm**	0.77	4.99	0.56	0.89	0.59		0.00
Fruit width	**mm**	0.97	1.32	0.94	0.99	0.94		0.00
Stone weight	**mm**	0.58	0.14	0.26	0.78	0.33		0.00
Stone length	**mm**	0.69	3.44	0.43	0.85	0.48		0.00
Stone width	**mm**	0.34	1.18	-0.04	0.63	0.12		0.07
Stone thickness	**mm**	0.25	1.35	-0.13	0.57	0.06		0.19
Seed weight	**g**	0.41	0.01	0.01	0.69	0.17		0.04
Seed length	**mm**	0.84	0.92	0.67	0.93	0.71		0.00
Seed width	**mm**	0.51	0.82	0.15	0.75	0.26		0.00

Legend: *r* - Pearson correlation coefficient, *min/max* - 95% confidence interval for *r*, r^2 - coefficient of determination, *p* - significance level ofr

determined the average length of the fruit from 16.77 to 21.67 mm. **Karnatovska et al., (2007)** identified in the evaluation of 23 varieties of Chinese jujube an average length of fruit in the range of 13.0 to 32.2 mm. **Zhang et al., (2011)** determined the average length of fruit at 26.1 mm. **Klymenko and Grygorieva, (2008)** in the range of 14.98 to 29.74 mm. The results of our experiments correspond with the results obtained by the given authors.

Fruit width

In our experiments the fruit width was determined in the range of 16.64 mm (ZJ-13) to 38.87 (ZJ-22) (Table 1). The variation coefficient (7.07%) confirmed moderate degree of variability within the collection. **Brindza et al., 2011** determined fruit width in the range of 12.67 mm to 16.97 mm. **Karnatovska et al., (2007)** identified in the evaluation of 23 varieties of Chinese jujube average fruit width in the range of 11.06 to 23.8 mm. **Zhang et al., (2011)** determined an average width of the fruit at 25.80 mm. **Klymenko and Grygorieva, (2008)** identified in the evaluation of 23 varieties of Chinese jujube fruit width in the range of 11.45 to 18.71 mm. The results of our experiments correspond with the results obtained by the given authors.

Shape index of fruits

The ratio between the length and width of the fruit is known as the shape index. In the assessment of genotypes we determined the shape index ranging from 1.04 (ZJ-08, ZJ-26 – roundish shape) to 2 (ZJ-07, ZJ-25) (Figure 5). We also identified significant differences among the genotypes in the color and shape of the fruit, as illustrated in figure 1.

Stone characteristics

Stone weight

The fruits of Chinese jujube (drupes) contain endocarps (stones), which are highly variable in their shape and size (Figure 2 and Figure 3). Each drupe contains only one stone placed in the middle of the fruit. The stone is hard,

spindle-shaped, elliptic, oblong, pointy at both ends, pointed, deeply wrinkled, red or dark brown to dark gray, 2.5 to 5 cm long **(Dinavand, Zarinkamar, 2006 Bonner and Rudolf, 1974 Husak et al., 1996)**. In evaluated genotypes we determined the weight of stones in the range of 0.173 (ZJ-07) to 0.731 g (ZJ-20) (Table 1). The value of

Figure 3 Stones of selected Chinese jujube (*Ziziphus jujuba* Mill.) genotypes.

Figure 4 Seeds of selected Chinese jujube (*Ziziphus jujuba* Mill.) genotypes.

the coefficient of variation was 20.34%, which documents a high degree of variability of the characteristic within the collection. Genotypes reaching extreme values of a given characteristic are shown in Table 2. The coefficients of variation also documented a significant difference in weight of stones within genotypes. **Brindza et al., 2011** determine the average weight of drupes in the range of 0.153 to 0.845 g. **Karnatovska et al., (2007)**, in the evaluation of 23 varieties of Chinese jujube in extreme agro ecological conditions in Nova Kachovka (Ukraine), identified the average weight of stones in the range of 1.0 to 2.1 g. **Ecevit et al., (2007)**, in the study of 52 genetic resources, determined the average weight of stones in the range from 0.34 to 0.41 g, **Sivakov et al., (1988)** identified the stone weight in the range of 0.28 to 0.65 g and **Ghosh and Mathew, (2002)**, in the study of nine genotypes, in the range of 0.6 to 1.9 g.

Stone length and width

In the assessment of individual fruits in genotypes we determined the stone length and width in the ranges of 12.84 (ZJ-08) - 28.67 mm (ZJ-04) and 5.06 mm (ZJ-07) - 9.74 mm (ZJ-01), respectively (Table 1). Variation coefficients (5.56–5.82%) confirmed low degree of variation in both these parameters. **Brindza et al., (2011)** in the evaluation of 19 genotypes of Chinese jujube determined the average stone length in the range of 10.58–14.35 mm.

Seed characteristics

Seed weight

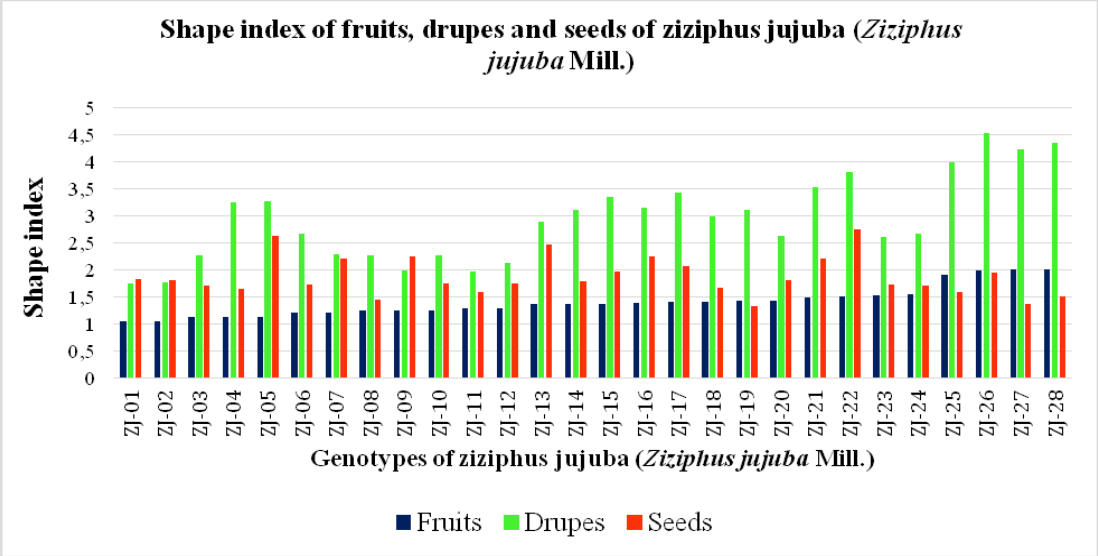

Figure 5 Comparison of the tested Chinese jujube genotypes (*Ziziphus jujuba* Mill.) in the shape index of fruit, drupes and seeds.

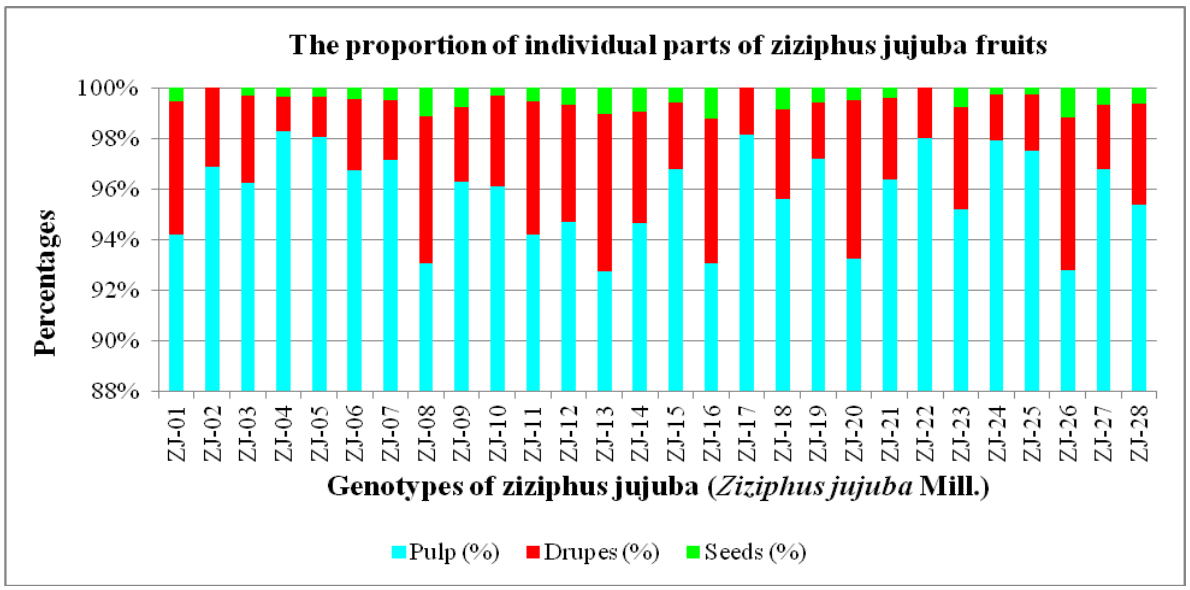

Figure 6 The weight of the pulp, drupes and seeds (%) from the total weight of the fruit (100%) of Chinese jujube (*Ziziphus jujuba* Mill.).

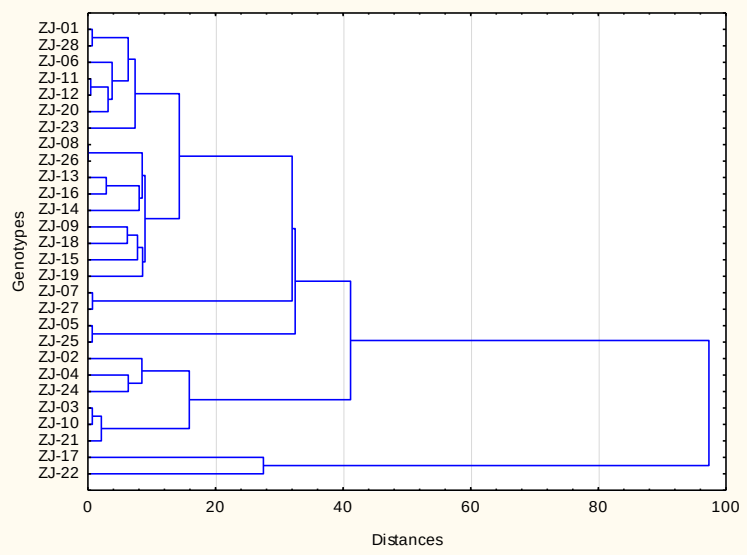

Figure 7 Dendrogram of 28 genotypes of Chinese jujube based on morphometric characteristics of fruits.

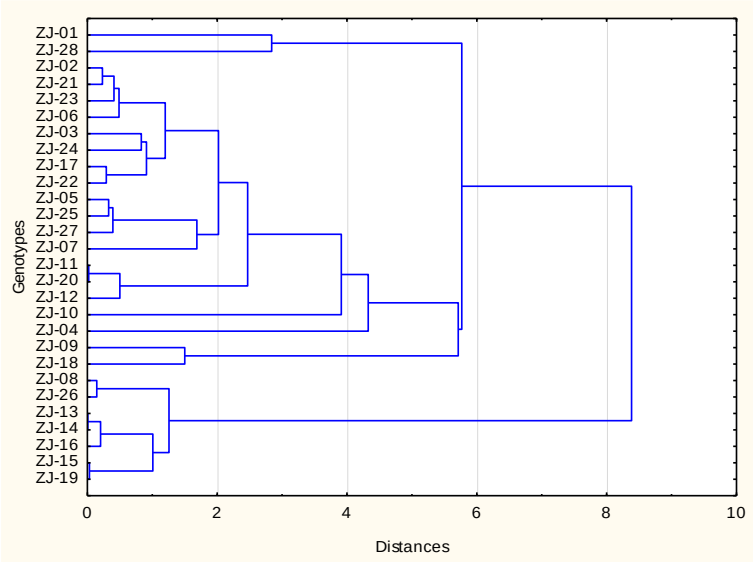

Figure 8 Dendrogram of 28 genotypes of Chinese jujube based on morphometric characteristics of stones.

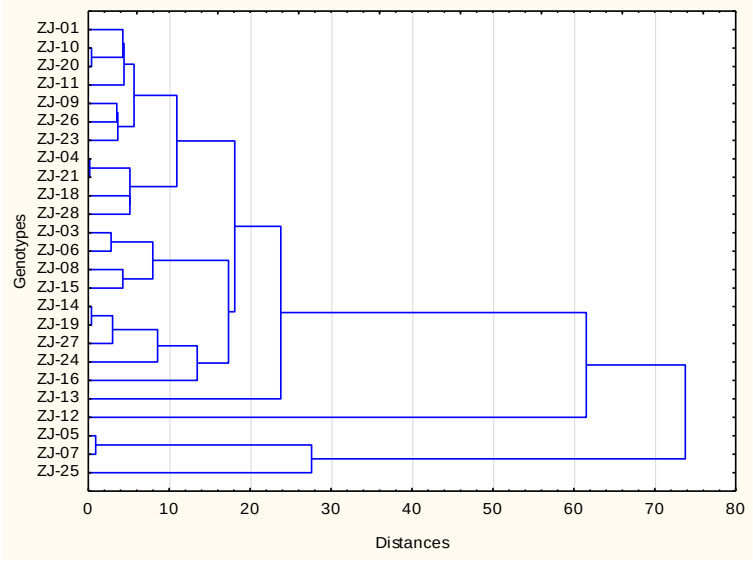

Figure 9 Dendrogram of 28 genotypes of Chinese jujube based on morphometric characteristics of seeds.

Seeds of Chinese datle (up to three) are enclosed in a hard woody inner endocarp – stone. However, some fruits are seedless and formed in a parthenocarpic way **(Pareek, 2001)**. The seeds of Chinese jujube have significant economic value as a raw material for obtaining valuable oil, which has a wide practical use. This is supported by findings of many literary sources. **Al-Reza et al., (2010)** determined 23 components in the oil from Chinese jujube seed. Among the major components included in this oil were eucalyptol, chavicol, eugenol, isoeugenol, ledol, veridiflorol, tumeron and more. Evaluating a collection of genotypes, we determined the seed weight in the range of 20.00 mg (ZJ-25) to 69.11 mg (ZJ-12) (Table 1). The value of variation coefficient (15.22%) documents a medium degree of variability of the characteristic within the collection. Genotypes reaching extreme values of a given characteristic are shown in Table 2. The coefficients of variation also documented a significant difference in the weight of the seeds within genotypes. Some genotypes do not form seeds (ZJ-02, ZJ-17, ZJ-22).

Seed length and width

In the collection of evaluated genotypes we determined the length of seeds in the range of 7.24 (ZJ-15) to 11.85 mm (ZJ-04) (Table 1). Coefficients of variation 3.85% documented low degree of variability of the characteristic within the collection. The width of the seeds was determined in the range of 3.86 mm (ZJ-05) to 6.84 mm (ZJ-12) (Table 1). The value of the coefficient of variation 3.95% documents also a low degree of variability of the characteristic within the collection. Among the genotypes, we also identified significant differences in the color and shape of the seeds, as illustrated in Figure 4. In the evaluated collection of genotypes, we determined the length of the seeds in the range of 3.86 (ZJ-25) to 6.84 mm (ZJ-12) (Table1). Variation coefficient (3.95%) documented low degree of variability of the characteristic within the collection.

Shape indexes of fruits, stones and seeds

The shape of each object can be characterized by the shape index, i.e. the length to width ratio. Figure 5 represents the shape indexes of fruits, stones and seeds. The shape index of the fruits was found in the range of 1.04 (ZJ-08) to 2.00 (ZJ-12 and ZJ-25), so the genotype collection demonstrate significant variability in the shape of the fruit, as seen in Figure 1. The shape indexes of stones and seeds were found ranging from 1.74 (ZJ-08) to 4.53 (ZJ-05) and from 1.32 (ZJ-23) to 2.74 (ZJ-04), respectively. According to Figure 5, it is evident that the shapes of fruits, stones and seeds did not correlate to each other, but were highly specific for jujube genotypes.

Relative weights f pulp, stone and seed

The relative values of pulp (stone, seed) weight in relation to the total fruit weight (relative weight of the pulp, stone or seed) are very important characteristics for practical use of many fruiting plants. A comparison of the evaluated genotypes in a given characteristic is presented in Figure 6. This Figure shows differences between genotypes. The relative weight of pulp was determined in the range from 92.76% (ZJ-13) to 98.32% (ZJ-04). The relative weight of seed was determined in the range of 0.25% (ZJ-24 and ZJ-25) to 1.16% (ZJ-26). These findings have a practical significance for genotype evaluation.

The relationship between specific characteristics

Because of the complexity of addressing the issues, we have identified a relationship between fruit weight and other assessed characteristics on the fruit, with the application of a simple correlation analysis. The results of the analysis are given in Table 3. The results indicated high correlations between the fruit weight and the fruit width (r = 0.971), stone length (r = 0.843), fruit length (r = 0.771) and stone height (r = 0.694). Slight correlation was found between the fruit weight and the stone thickness (r = 0.254) and width (r = 0.341).

Clustering of Chinese jujube e genotypes based on fruit characteristics

The Chinese jujube genotypes of this study were grouped based on hierarchical clustering of the fruit, stone and seed characteristics (Figure 7, Figure 8 and Figure 9). The three dendrograms, obtained from the cluster analysis of the selected morphometric characteristics (fruits, seeds and stones) gave different results. The fruit characters generated dendrogram with three different clusters where in the most dissimilar cluster can be identified genotypes ZJ-17 and ZJ-22 the both with high proportion of pulp in fruits. Dendrogram created from morphometric parameters of stones separated 3 different groups, where genotypes (ZJ-01 and ZJ-28) in the first group and genotypes (ZJ-08, ZJ-26, ZJ-13, ZJ-14, ZJ-15 and ZJ-19) in the second group were different from the rest of the tested genotypes. Performed cluster analysis based on characteristics of seeds created group of distinct genotypes (ZJ-12, ZJ-05, ZJ-07 and ZJ-25).

CONCLUSION

The results of the experiment presented in this work are consistent with the results reported earlier. In evaluating 28 genotypes of Chinese jujube we determined the weight of the fruits in the range from 2.90 g (ZJ-13) to 28.99 g (ZJ-22), length from 20.73 mm (ZJ-08) to 44.84 mm (ZJ-04), and width from 16.64 mm (ZJ-13) to 38.87 mm (ZJ-22). Isolated stones were also analysed, and their weight was from 0.173 g (ZJ-07) to 0.731 g (ZJ-20), length from 12.84 mm (ZJ-08) to 28.67 mm (ZJ-04), and width from 5.06 mm (ZJ-07) to 9.74 mm (ZJ-01). Presented results also showed that significant differences in the evaluated characteristics were found for the studied jujube genotypes. Obtained results are important for breeding new varieties of Chinese jujube as well as their practical use.

The results obtained by these experiments confirmed differences between the evaluation of Chinese jujube genotypes in morphological characteristics of fruit, drupes and seeds. The results also demonstrate that the fruits of Chinese jujube, with their high proportion of pulp, can be used for direct consumption and for preparation of many food products. This species is very suitable for cultivation, dissemination and utilization in Slovakia. It is temperature resistant to -25 °C. Besides fruits, other parts of this plant can be useful, especially for pharmacological and cosmetic use. Seeds are valuable for their oil content. Growing and

utilization of Chinese jujube plant in Slovakia would be mainly used for socio-economic development of family farms.

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