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Fruit and seed traits of *Berberis croatica* Horvat and *Berberis vulgaris* L.

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Abstract – The three year variability of fruits and seeds was investigated in *Berberis croatica* (Vošac, Mt Biokovo, Rakov Potok near Zagreb) and in *B. vulgaris* (Fran Kušan Pharmaceutical Botanical Garden in Zagreb). *Berberis croatica* had the following dimensions of fruits (seeds): length 7.28–7.88 (4.57–5.03) mm; width 3.85–3.99 (width 1: 2.06–2.20; width 2: 1.44–1.63) mm; weight 0.065–0.078 (0.0116–0.0134) g. Dimensions of *B. vulgaris* fruits (seeds) were: length 10.20–11.29 (5.71–6.24) mm; width 5.29–5.83 (width 1: 2.40–2.71; width 2: 1.60–1.98) mm; weight 0.1602–0.2199 (0.0146–0.0235) g. The fruit shape of both species was similar and the length/width ratio was 1.91–2.04 in *B. croatica* and 1.77–2.07 in *B. vulgaris*. The number of seeds per fruit was 1.23–1.58 in *B. croatica* and 1.36–1.54 in *B. vulgaris*. Generally, fruits and seeds of *B. vulgaris* were significantly longer, wider and heavier than fruits and seeds of *B. croatica*. ANOVA showed significant statistical differences between populations for all analyzed fruit and seed traits while the species significantly differed in all traits, except in the fruit shape and number of seeds in fruit.

Keywords: *Berberis croatica*, *Berberis vulgaris*, fruit, seed

Introduction

Inter- and intraspecific variability in fruit and/or seed traits has been in the focus of interest of many authors (JORDANO 1984, HENDRIX and SUN 1989, WILLSON et al. 1990, HERRERA 1992, OBESO and HERRERA 1994, ERIKSSON 1999) but only few of them address the genus *Berberis*. At the same time, when dealing with the fruit and seed properties of

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barberry species in relation with interannual variation (HERRERA 1998, ALLEN and WILSON 1992), papers are mainly concentrated on the seed and/or fruit production. Even though data regarding basic fruit and seed related traits for some of the European species of the genus *Berberis*, namely *B. vulgaris* (RUDOLF 1974, AKBULUT et al. 2009) and *B. hispanica* (OBESO and HERRERA 1994) could be found, those concerning *B. croatica* are lacking. *B. croatica* is a species with a still unresolved taxonomic status and is still not recognized as a distinctive species by the leading authorities on European flora. Several attempts have been made to crystallize the morphological (KARLOVIĆ et al. 2009) and chemical difference (ZOVKO KONČIĆ et al. 2010) between *B. croatica* and *B. vulgaris*. However, fruit and seed characters have not been included in the analysis to date yet they could help in the characterization of the distinctive morphological traits of these species.

Material and methods

Fruits of *Berberis croatica* Horvat and *B. vulgaris* L. were randomly collected during three years (from 2008 to 2010). *B. croatica* fruits were collected in one natural habitat (Vošac, Mt Biokovo, in text as abbreviation Vo; 43°18' N; 17°02' E; 1422 m a.s.l.), while *B. vulgaris* fruits were collected in one natural (Rakov Potok near Zagreb, in text as abbreviation RP; 45°47' N; 15°42' E; 160 m a.s.l.) and one cultivated habitat (Fran Kušan Pharmaceutical Botanical Garden, Zagreb, in text as abbreviation PBG; 45°50' N; 15°59' E; 195 m a.s.l.). Twenty plants were harvested in the locality Vošac, ten in the locality Rakov Potok and six in the cultivated habitat. According to our permit from the Ministry of Culture of the Republic of Croatia, we were able to analyse 100 fruits for the locality Vošac and so the same number of *B. vulgaris* fruits (100) was collected for each population. Length, width and weight of fruits, as well as seeds, were measured. Since the seeds of the researched *Berberis* species have irregular shapes, the width of the seed was measured on two axes (width 1 and width 2). The ratio between length and width of fruits and length and width (width 1, width 2) of seeds was obtained through mathematical calculation, as well as their mutual (fruit-seed) ratio, which included length, width and weight. A greater ratio indicates narrower fruits (or seeds), while a smaller ratio indicates more orbicular fruits (or seeds).

The differences in fruit and seed traits between species and populations were analyzed by the General Linear Models (GLM) procedure using repeated measures ANOVA and Tuckey's post-hoc test at the $p \leq 0.05$ level. The statistical analysis was performed using Statistica 7 software (StatSoft Inc., Tulsa, OK, USA).

Results

Table 1 presents descriptive statistics for the analyzed traits of fruits and seeds of *Berberis croatica* and *B. vulgaris*. The average length of *B. croatica* fruits ranged from 7.28 mm (Vo 2010) to 7.88 mm (Vo 2009) while the width ranged from 3.85 mm (Vo 2010) to 3.99 mm (Vo 2008). Weight of *B. croatica* fruits ranged from 0.065 g (Vo 2008) to 0.078 g (Vo 2010). Fruits of *B. vulgaris* were the largest and their length ranged from 10.20 mm (PBG 2009) to 11.29 mm (RP 2010), while the width ranged from 5.29 mm (RP 2008) to 5.83 mm (PBG 2009). Weight of *B. vulgaris* fruits ranged from 0.1602 g (RP 2009) to 0.2199 g (PBG 2009). The shapes of the two species were similar and the ratio between

length and width of fruits was from 1.91 (Vo 2010) to 2.04 (Vo 2009) in *B. croatica* and from 1.77 (PBG 2009) to 2.07 (RP 2010) in *B. vulgaris*. The most variable trait was fruit weight, ranging from 25.61 % (Vo 2010) to 28.55 % (Vo 2008) and from 22.84 % (RP 2010) to 36.54 % (PBG 2008) in *B. croatica* and *B. vulgaris*, respectively. The least variable trait was fruit length, which ranged from 5.84 % (Vo 2009) to 9.62 % (Vo 2010) and from 6.37 % (PBG 2009) to 9.83 % (RP 2008) in *B. croatica* and *B. vulgaris*, respectively (Tab. 1).

The number of seeds per fruit ranged from 1.23 (Vo 2010) to 1.58 (Vo 2009) in *B. croatica* and from 1.36 (RP 2008, RP 2010) to 1.54 (PBG 2010) in *B. vulgaris*.

Tab. 1. Statistics for analyzed traits of fruits and seeds of *Berberis croatica* – Vošac (Vo) and *B. vulgaris* – Rakov Potok (RP) and Pharmaceutical Botanical Garden »Fran Kušan« (PBG). Min, Max and Std. dev. are in mm (length, width) and g (weight); CV (coefficient of variability) is in %. Minimum and maximum are bolded.

Statistics		<i>B. croatica</i>			<i>B. vulgaris</i>					
		Vo 2008	Vo 2009	Vo 2010	RP 2008	RP 2009	RP 2010	PBG 2008	PBG 2009	PBG 2010
Fruit traits										
Length	N	100	100	100	100	100	100	100	100	100
	Mean	7.83	7.88	7.28	10.89	10.39	11.29	10.42	10.20	10.41
	Min	7.00	6.89	5.26	5.34	8.64	5.84	8.94	9.01	7.33
	Max	9.04	9.07	9.43	12.74	12.10	13.26	12.54	12.47	12.12
	Std. dev.	0.50	0.46	0.70	1.07	0.79	0.91	0.74	0.65	0.67
	CV	6.39	5.84	9.62	9.83	7.60	8.06	7.10	6.37	6.44
Width	N	100	100	100	100	100	100	100	100	100
	Mean	3.99	3.91	3.85	5.29	5.30	5.55	5.42	5.83	5.71
	Min	3.03	2.86	2.23	3.91	4.22	4.16	3.76	4.59	3.50
	Max	5.03	5.06	5.01	6.90	6.90	7.04	7.76	7.58	7.43
	Std. dev.	0.42	0.44	0.51	0.69	0.62	0.54	0.94	0.68	0.73
	CV	10.53	11.25	13.25	13.04	11.70	9.73	17.34	11.66	12.78
Length/ Width	N	100	100	100	100	100	100	100	100	100
	Mean	1.98	2.04	1.91	2.05	1.98	2.07	1.97	1.77	1.85
	Min	1.46	1.52	1.40	1.22	1.55	1.30	1.49	1.47	1.12
	Max	2.88	2.88	3.05	2.77	2.78	2.40	2.60	2.07	2.66
	Std. dev.	0.25	0.25	0.22	0.19	0.22	0.19	0.30	0.16	0.25
	CV	12.63	12.25	11.52	9.27	11.11	9.18	15.23	9.04	13.51
Weight	N	100	100	100	100	100	100	100	100	100
	Mean	0.065	0.071	0.078	0.182	0.160	0.209	0.180	0.220	0.206
	Min	0.034	0.032	0.036	0.066	0.082	0.107	0.062	0.102	0.051
	Max	0.119	0.113	0.132	0.307	0.250	0.328	0.362	0.376	0.348
	Std. dev.	0.018	0.020	0.020	0.050	0.037	0.048	0.066	0.0531	0.062
	CV	27.06	28.55	25.61	27.48	22.85	22.84	36.54	24.15	30.05

Tab. 1. – continued

Statistics		<i>B. croatica</i>			<i>B. vulgaris</i>					
		Vo 2008	Vo 2009	Vo 2010	RP 2008	RP 2009	RP 2010	PBG 2008	PBG 2009	PBG 2010
Seed traits										
Length	N	145	158	123	136	138	136	142	149	154
	Mean	5.03	4.92	4.57	5.88	5.71	5.99	6.14	5.94	6.24
	Min	4.13	3.97	3.58	4.30	4.23	4.29	4.81	5.14	4.51
	Max	5.72	5.84	5.74	6.94	6.88	6.99	7.29	6.64	7.75
	Std. dev.	0.31	0.30	0.38	0.47	0.52	0.47	0.45	0.36	0.49
	CV	6.16	6.10	8.32	7.99	9.11	7.85	7.33	6.06	7.85
Width 1	N	145	158	123	136	138	136	142	149	154
	Mean	2.10	2.06	2.20	2.57	2.55	2.71	2.46	2.40	2.50
	Min	1.28	1.22	1.30	1.43	1.54	2.03	1.78	1.87	1.68
	Max	2.90	2.51	2.60	3.09	3.16	3.14	3.28	3.14	3.32
	Std. dev.	0.26	0.23	0.18	0.27	0.35	0.23	0.33	0.32	0.31
	CV	12.38	11.17	8.18	10.51	13.73	8.49	13.41	13.33	12.40
Width 2	N	145	158	123	136	138	136	142	149	154
	Mean	1.49	1.44	1.63	1.94	1.82	1.98	1.61	1.60	1.66
	Min	0.82	0.92	0.85	0.87	1.09	0.96	0.50	0.79	0.83
	Max	2.27	1.97	1.96	2.48	2.75	2.57	2.36	2.24	2.66
	Std. dev.	0.26	0.21	0.22	0.30	0.25	0.30	0.34	0.34	0.36
	CV	17.45	14.58	13.50	15.46	13.74	15.15	21.12	21.25	21.69
Length/ Width 1	N	145	158	123	136	138	136	142	149	154
	Mean	2.43	2.41	2.09	2.30	2.26	2.22	2.53	2.50	2.52
	Min	1.87	1.93	1.66	1.88	1.87	1.71	1.88	2.01	1.90
	Max	3.62	3.53	2.83	3.30	2.96	2.67	3.40	3.07	3.96
	Std. dev.	0.30	0.26	0.20	0.23	0.22	0.21	0.31	0.24	0.30
	CV	12.35	10.79	9.57	10.00	9.73	9.46	12.25	9.60	11.90
Length/ Width 2	N	145	158	123	136	138	136	142	149	154
	Mean	3.48	3.48	2.86	3.12	3.18	3.10	3.95	3.88	3.89
	Min	2.04	2.45	2.17	2.22	2.15	2.15	2.62	2.58	2.44
	Max	5.09	4.95	5.01	5.65	4.71	5.46	6.11	6.67	6.72
	Std. dev.	0.57	0.51	0.45	0.59	0.42	0.59	0.80	0.84	0.83
	CV	16.38	14.66	15.73	18.91	13.21	19.03	20.25	21.65	21.34
Width 1/ Width 2	N	145	158	123	136	138	136	142	149	154
	Mean	1.45	1.46	1.37	1.35	1.42	1.40	1.58	1.56	1.55
	Min	0.56	0.92	1.01	1.07	0.76	0.98	1.02	1.03	0.93
	Max	2.12	1.91	1.89	2.40	2.20	2.29	2.51	2.96	2.81
	Std. dev.	0.29	0.22	0.16	0.19	0.20	0.21	0.32	0.34	0.30
	CV	20.00	15.07	11.68	14.07	14.08	15.00	20.25	21.79	19.35

Tab. 1. – continued

Statistics		<i>B. croatica</i>			<i>B. vulgaris</i>					
		Vo 2008	Vo 2009	Vo 2010	RP 2008	RP 2009	RP 2010	PBG 2008	PBG 2009	PBG 2010
Weight	N	145	158	123	136	138	136	142	149	154
	Mean	0.012	0.012	0.013	0.022	0.022	0.024	0.018	0.017	0.015
	Min	0.001	0.001	0.005	0.004	0.007	0.010	0.003	0.003	0.002
	Max	0.018	0.021	0.019	0.035	0.034	0.035	0.038	0.046	0.033
	Std. dev.	0.003	0.003	0.003	0.007	0.013	0.005	0.008	0.007	0.006
	CV	24.14	23.97	20.15	31.78	57.72	22.13	44.32	43.79	42.47
Seed and fruit traits										
Seeds in Fruit	N	100	100	100	100	100	100	100	100	100
	Mean	1.45	1.58	1.23	1.36	1.38	1.36	1.42	1.49	1.54
	Min	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
	Max	2.00	3.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
	Std. dev.	0.50	0.52	0.42	0.48	0.49	0.48	0.50	0.50	0.50
	CV	34.49	32.91	34.15	35.29	35.51	35.29	35.21	33.56	32.47
Seed/Fruit Length	0.64	0.62	0.63	0.54	0.55	0.53	0.59	0.60	0.60	
Seed width 1/Fruit Width	0.61	0.62	0.54	0.43	0.43	0.40	0.47	0.43	0.44	
Seed width 2/Fruit Width	0.87	0.89	0.74	0.59	0.60	0.56	0.73	0.67	0.68	
Seed/Fruit Weight	0.177	0.170	0.172	0.119	0.137	0.113	0.098	0.077	0.071	

Seeds of *B. vulgaris* were also larger than seeds of *B. croatica*. The length of *B. croatica* seeds ranged from 4.57 mm (Vo 2010) to 5.03 mm (Vo 2008), width 1 from 2.06 mm (Vo 2009) to 2.20 mm (Vo 2010), width 2 from 1.44 mm (Vo 2009) to 1.63 mm (Vo 2010) and the weight from 0.0116 g (Vo 2008) to 0.0134 g (Vo 2010). In *B. vulgaris* seeds the length ranged from 5.71 mm (RP 2009) to 6.24 mm (PBG 2010), width 1 from 2.40 mm (PBG 2009) to 2.71 mm (RP 2010), width 2 from 1.60 mm (PBG 2010) to 1.98 mm (RP 2010) and the weight from 0.0146 g (PBG 2010) to 0.0235 g (RP 2010). The most variable trait was seed weight, ranging from 20.15 % (Vo 2010) to 24.14 % (Vo 2008) and from 22.13 % (RP 2010) to 57.72 % (RP 2009) in *B. croatica* and *B. vulgaris*, respectively. The least variable trait was seed length and it ranged from 6.10 % (Vo 2009) to 8.32 % (Vo 2010) and from 6.06 % (PBG 2009) to 9.11 % (RP 2009) in *B. croatica* and *B. vulgaris*, respectively (Tab. 1).

ANOVA showed significant statistical differences between species for most analyzed fruit and seed traits (Tab. 2). The only exception was the ratio between fruit length and width as well as the number of seeds in the fruit. The analysis of fruit traits between species showed that fruits of *B. vulgaris* were significantly longer, wider and heavier than fruits of *B. croatica* (Tab. 3). Nevertheless, fruit shape (based on ratio between length and wide) was similar in the two species.

Tab. 2. Results of ANOVA for analysed traits of fruit and seed. Asterisks indicate significant p values at $p \leq 0.05$.

Effect	Sum of Squares	Degr. of freedom	Mean Square	F	p-level
Fruit Length					
Population	1765.34	2	882.67	1608.8	<0.0001*
Species	1725.57	1	1725.57	2536.49	<0.0001*
Fruit Width					
Population	523.84	2	261.92	643.30	<0.0001*
Species	513.08	1	513.08	1161.04	<0.0001*
Fruit Length/Width					
Population	4.504	2	2.252	40.52	<0.0001*
Species	0.202	1	0.202	2.89	0.0904
Fruit Weight					
Population	2.99660	2	1.49830	832.78	<0.0001*
Species	2.94489	1	2.94489	1497.422	<0.0001*
Seed Length					
Population	347.93	2	173.97	977.2	<0.0001*
Species	335.54	1	335.54	1608.2	<0.0001*
Seed Width 1					
Population	49.319	2	24.659	300.74	<0.0001*
Species	43.625	1	43.625	454.14	<0.0001*
Seed Width 2					
Population	31.795	2	15.898	189.02	<0.0001*
Species	15.041	1	15.041	119.48	<0.0001*
Seed Length/Width 1					
Population	16.161	2	8.081	126.2	<0.0001*
Species	1.598	1	1.598	15.92	0.0001*
Seed Length/Width 2					
Population	142.38	2	71.19	165.92	<0.0001*
Species	18.93	1	18.93	25.67	<0.0001*
Seed Width 1/Width 2					
Population	6.560	2	3.280	48.44	<0.0001*
Species	0.826	1	0.826	10.08	0.0016*
Seed Weight					
Population	0.023150	2	0.011575	44.116	<0.0001*
Species	0.016446	1	0.016446	59.0474	<0.0001*
Seeds in fruit					
Population	2.047	2	1.023	4.241	0.0153*
Species	0.005	1	0.005	0.020	0.8870

BERBERIS CROATICA AND *B. VULGARIS* FRUIT AND SEED TRAITS

Tab. 3. *p* values from Tukey post-hoc test for variability of fruits of *Berberis croatica* in Vošac and *B. vulgaris* in Rakov Potok and Pharmaceutical botanical garden »Fran Kušan« (PBG) populations. Asterisks indicate significant *p* values at $p \leq 0.05$.

Population	Vošac	Rakov Potok	Population	Vošac	Rakov Potok
	Length			Length/Width	
Vošac			Vošac		
Rakov Potok	<0.0001*		Rakov Potok	0.1867	
PBG	<0.0001*	0.0001*	PBG	0.9058	0.0757
	Width			Weight	
Vošac			Vošac		
Rakov Potok	<0.0001*		Rakov Potok	<0.0001*	
PBG	<0.0001*	0.4388	PBG	<0.0001*	0.9738

ANOVA of seed traits showed similar results (Tab. 4). In general, seeds of *B. vulgaris* were significantly longer, wider and heavier than fruits of *B. croatica*. Interspecies and

Tab. 4. *p* values from Tukey post-hoc test for variability of seeds of *Berberis croatica* in Vošac and *B. vulgaris* in Rakov Potok and Pharmaceutical botanical garden »Fran Kušan« (PBG) populations. Asterisks indicate significant *p* values at $p \leq 0.05$.

Population	Vošac	Rakov Potok	Population	Vošac	Rakov Potok
	Length			Length/Width 2	
Vošac			Vošac		
Rakov Potok	<0.0001*		Rakov Potok	0.0002*	
PBG	<0.0001*	<0.0001*	PBG	<0.0001*	<0.0001*
	Width 1			Width 1/Width 2	
Vošac			Vošac		
Rakov Potok	<0.0001*		Rakov Potok	0.0198*	
PBG	<0.0001*	0.0058*	PBG	0.0004*	<0.0001*
	Width 2			Weight	
Vošac			Vošac		
Rakov Potok	<0.0001*		Rakov Potok	<0.0001*	
PBG	0.0031*	<0.0001*	PBG	<0.0001*	<0.0001*
	Length/Width 1			Seeds in fruit	
Vošac			Vošac		
Rakov Potok	0.0008*		Rakov Potok	0.4001	
PBG	0.0163*	<0.0001*	PBG	0.9030	0.6651

intraspecies variability for seed traits was something higher than for fruit traits. The least inter- and intraspecies variability was recorded for the number of seeds in fruit.

Discussion

The least variable traits in this analysis were fruit and seed length while the most variable traits were fruit and seed weight. That was expected since dimensionality considerations predict that, all else being equal, the coefficients of variability (CVs) of mass-related traits will be larger than those of linear or surface measurements (LANDE 1977), such as length and width of fruits and seeds. The largest variability for fruit and seed weight was also reported by OBESO and HERRERA (1994) which state CVs for these traits ranging generally from 20–30 % for eight investigated species, including *Berberis hispanica*. The same authors also reported fruit length and width variability ranging generally between 5 and 12 % (7.6 % and 7.4 % in *B. hispanica* for fruit length and fruit width, respectively). These results are in keeping with the results reported in this paper.

Regarding fruit traits of *B. vulgaris*, AKBULUT et al. (2009) reported much lower values for the wild population of *B. vulgaris* growing in Turkey. For example, average fruit length, width and mass are reported to be 7.69 mm, 3.32 mm, and 0.07 g, (compared with the average value range for the same traits in this investigation, that is, 10.20–11.29 mm, 5.29–5.83 mm and 0.1602–0.2199 g). This could be explained by environmental factors but also by great intraspecific variation in *Berberis* already reported by LANDRUM (1999).

Generally, fruits and seeds of *B. vulgaris* were significantly longer, wider and heavier than fruits and seeds of *B. croatica*. This is in congruence with the results of KARLOVIĆ et al. (2009) who, when analyzing distinguishing morphological traits between these two species, stated that *B. croatica* is characterized by reduced growth/size traits compared with *B. vulgaris*. Our results support this thesis. Results regarding fruit and seed traits show significant differences between species for most analyzed traits (10 out of 12). Nevertheless, whether the observed differences between *B. croatica* and *B. vulgaris* are merely non-hereditary modifications caused by environmental factors or species-related differences, should be further tested by means of genetic analysis.

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