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Genetic variability in quantitative traits of field pea (*Pisum sativum* L.) genotypes

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Abstract: The field pea is very important when it comes to the nutrition of domestic animals, due to a high content of proteins in its grains and aboveground biomass. The aim of this study was to examine genetic variability in quantitative traits of newly created field pea hybrids. The researchers studied two cultivars, one line and their F_3 and F_4 hybrids. The following quantitative traits were analysed: number of pods per plant, number of grains per pod, 1000 seed weight and grain yield. The largest number of pods per plant was recorded for the Baccara × L-CC line hybrid, with a statistically significantly larger ($P < 0.01$) number of pods per plant than all the other investigated genotypes, lines and hybrids. L-CC line × Baccara, and Saša × L-CC line had a significantly larger ($P < 0.05$) number of grains per pod. The smallest number of grains per pod was recorded for the Saša and Baccara × Saša hybrid. The genotype vs. year interaction had a significant ($P < 0.05$) influence on 1000 seed weight. Significantly higher ($P < 0.05$) 1000 seed weight was recorded for the Baccara genotype, when compared with all other investigated genotypes, lines and hybrids. The lowest 1000 seed weight was recorded for the L-CC line. The genotype × year interaction had a significant influence on the seed yield of field pea. During this research, high seed yields were achieved by the Baccara genotype and Baccara × L-CC line and Baccara × Saša hybrids.

Keywords: hybrid peas; number of grains per pod; 1000 seed weight; seed yields

The field pea encompasses a large number of agronomic types that generally differ in the colour of flowers, grain shape and colour, and part of the plant used. The pea is very important in nutrition of domestic animals due to its high content of protein, calcium and stimulating substances. It is a good preceding crop for all field crops, except for annual and perennial legumes. Due to its symbiosis with root nodule bacteria, it leaves significant amounts of nitrogen in the soil for the subsequent crop. After the pea is harvested, the soil retains a favourable structure. After cutting or harvesting the pea, there is enough time left for farmers to perform the basic seedbed preparation for the subsequent crop. The field pea has its winter and spring forms. It is suitable to be used as green fodder, hay, grains, silage and flour. Furthermore, it can also be used as green manure.

One of the basic directions of pea breeding is the creation of genotypes with a high genetic potential for fodder yield or grain yield. Genotypes can be targeted for a specific agro-ecological area. The best exploitation of the genetic potential is achieved in the environment for which certain genotypes have been created. In order to obtain new genotypes with a high genetic potential for yield, breeders select and test the most promising hybrids within the pea cultivation program.

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Genetic variations of phenological and morphological properties, such as flowering, plant height and grain mass, may be affected by one or more genes. An increase in pea yields is significantly affected by the selection of genotypes for plant height, number of pods per plant, length of pods and number of days to pea maturity (Kosev 2014). Kumar *et al.* (2013) indicated that grain yields of pea may be increased by choosing genotypes with a larger number of pods per plant and number of grains per pod. In order to make a successful field pea breeding program, breeders must first identify the germplasm of desired traits in the starting material. Afterwards, the breeders have to select good parental lines or genotypes for hybridization and then choose an appropriate selection method. Gixhari *et al.* (2014) stated they used the method of molecular markers for investigating the genetic diversity of 28 local pea populations.

The aim of this study was to examine the genetic variability in quantitative traits of newly created field pea hybrids.

MATERIAL AND METHODS

Parental genotypes and their hybrids were sown in March 2014 in the experimental field of the Agricultural Institute of Republika Srpska in Delibašino Selo. The experiments were conducted during 2014 and 2015. The size of the basic plot was 2.5 m^2. The sowing distance was 12.5 × 8 cm and sowing depth was 4–5 cm. The experiment was done in 4 replications. The basic fertilization in seedbed preparation was 350 kg/ha NPK 8 : 24 : 24. The standard sowing rate was 80 germinated seeds per square meter. Side dressing was done with 100 kg/ha KAN 27% N. A herbicide with the active substance pendimethalin was used for weed control after sowing and before sprouting. When the pea plants grew to 10–12 cm, a corrective herbicide benthon (1680 g/ha) was done. The crops were treated against pests with insecticide at the beginning of flowering and at the stage of blossom fall, using systemic deltamethrin (12.5 g/ha) in 2014 and dimethoate (160 g/ha) in 2015. The grain yield was determined after combining and grain drying.

The following genotypes of the spring field pea were used for crossing:

(1) Saša – a genotype created at the Agricultural Institute of Republika Srpska, B&H. It is intended for a combined use of green mass and grain.
(2) Baccara – a French field pea genotype intended for grain production.
(3) L-CC field pea line originated from North America. It is suitable for green mass production and has a purple colour of the flower.

Four different combinations were used for analyses: Baccara × L-CC line, Saša × L-CC line, Line L-CC × Baccara and Baccara × Saša.

From the F_2 generation, 60 plants were selected for each hybrid combination. The plants were sown at a distance of 50 × 50 cm. F_3 and F_4 generations were obtained using the pedigree method by selecting 10 plants of the F_2 and F_3 generation, respectively, according to the phenotype. For each hybrid combination in F_3 and F_4 generations, 10 plants in 10 replications were evaluated.

The following quantitative traits were analysed in the paper: number of pods per plant, number of grains per pod, 1000 seed weight (g) and grain yield (kg/ha).

The original experimental data were processed, analysed and evaluated using the following mathematical and statistical methods: the influence of factors on plant traits, analysis of variance (ANOVA) and Fisher's LSD test were used for any significant differences at the $P < 0.05$ levels between the means. All the analyses were conducted using the Statistics 12.0 software package (StatSoft Inc., USA). Testing of significance of the differences between the calculated average values of the investigated factors (year and genotype) was done by using a model of two-factor analysis of variance. The relative dependence was defined by the correlation analysis method (Pearson's correlation coefficients). Obtained coefficients were tested by the t-test for significance levels $P < 0.05$ and $P < 0.01$.

Soil conditions. In the experimental field in Delibašino Selo, valley brown soil dominates on alluvial substrate of the Vrbas River. By mechanical composition, it belongs to a group of clay-loam soil.

The chemical properties of the arable soil layer on which the experiments were laid out are shown in Table 1.

The soil had an alkaline reaction. When it comes to humus content, it was a low-humus soil (1.9%). The availability of easily accessible phosphorus was good (20.3 mg/100 g of soil), while the presence of

Table 1. Results of the chemical analysis of the arable soil layer

Depth (cm)	Humus (%)	pH		P_2O_5	K_2O
		in H_2O	in KCl	(mg/100g of soil)	
0–30	1.9	7.9	6.9	20.3	16.1

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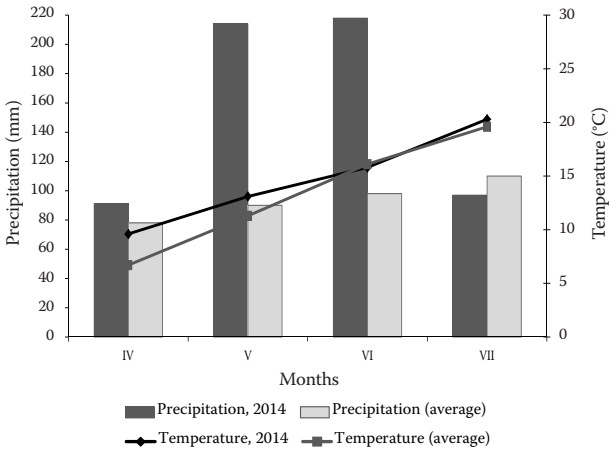

Figure 1. Total precipitation and temperatures in 2014, Banja Luka, Bosnia and Herzegovina

potassium in the soil was moderate and amounted to 16.1 mg/100 g of soil.

Based on the results of the chemical analysis of the arable soil layer, it can be said that the soil provided for the experiment is suitable for the growing of field pea.

Meteorological conditions. Data obtained from Banja Luka hydrometeorological station (Figure 1 and 2) were used for the analysis of weather conditions. Banja Luka is located 4.5 km from Delibašino Selo.

RESULTS AND DISCUSSION

Meteorological conditions were variable (Popović *et al*. 2015, 2016). In both years, the winter period was warmer compared to the multi-year average. The average annual temperature was 11.1°C, and during the vegetation period it was 13.4°C. The total amount of precipitation in the vegetation period (III–VI) in 1961–2014 was 376.0 mm.

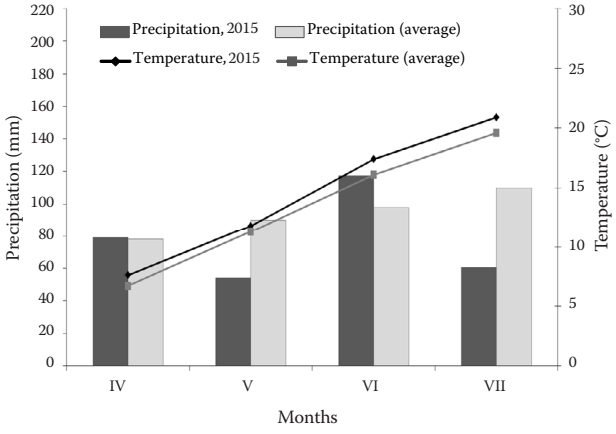

Figure 2. Total precipitation and temperatures in 2015, Banja Luka, Bosnia and Herzegovina

The summer of 2015 was warmer with much less precipitation compared to the multi-year average (Figure 1). In April and July, there was twice as much precipitation as Banja Luka's multi-year average. The average temperatures in both years during the vegetation period were higher than the multi-year average. In the first year of the study, the amount of precipitation during the vegetation period was 244.0 mm higher than the multi-year average. During the vegetation period in 2015, there was 311.2 mm of precipitation, which is 64.8 mm less than the multi-year average (Figure 2).

The genotype, the year and the genotype × year interaction had a significant ($P < 0.05$) influence on the number of pods per plant. For all the investigated

Table 2. Number of pods per plant, number of grains per pod, 1000 seed weight and grain yield of the field pea genotypes

Genotypes	Year	No. of pods per plant	No. of grains per pod	1000-seed weight (g)	Grain yield (kg/ha)
Baccara	2014	5.90	5.70	222.85	5186.20
	2015	4.90	6.20	235.18	4772.50
	average	5.40	5.95	229.02	4979.35
Saša	2014	6.80	5.60	164.47	4186.90
	2015	5.80	5.70	167.35	4418.60
	average	6.30	5.65	165.91	4302.75
L-CC line	2014	6.20	5.80	143.75	3188.90
	2015	6.60	6.60	153.08	3903.10
	average	6.40	6.20	148.41	3546.00
L-CC line × Baccara	2014	5.00	8.10	172.98	4699.10
	2015	5.20	6.90	169.23	4463.20
	average	5.10	7.50	171.11	4581.15
Baccara × L-CC line	2014	6.60	6.20	185.34	4721.50
	2015	6.80	5.90	187.42	4745.40
	average	6.70	6.05	186.38	4733.45
Saša × L-CC line	2014	5.80	7.00	154.82	4264.40
	2015	5.60	7.20	153.04	3962.60
	average	5.70	7.10	153.93	4113.50
Baccara × Saša	2014	6.60	5.60	191.47	4596.80
	2015	6.20	5.60	196.84	4723.40
	average	6.40	5.60	194.16	4660.10
Average	2014	6.12	6.28	176.53	4406.26
	2015	5.57	6.30	180.31	4426.97
	average	6.00	6.29	178.42	4416.61

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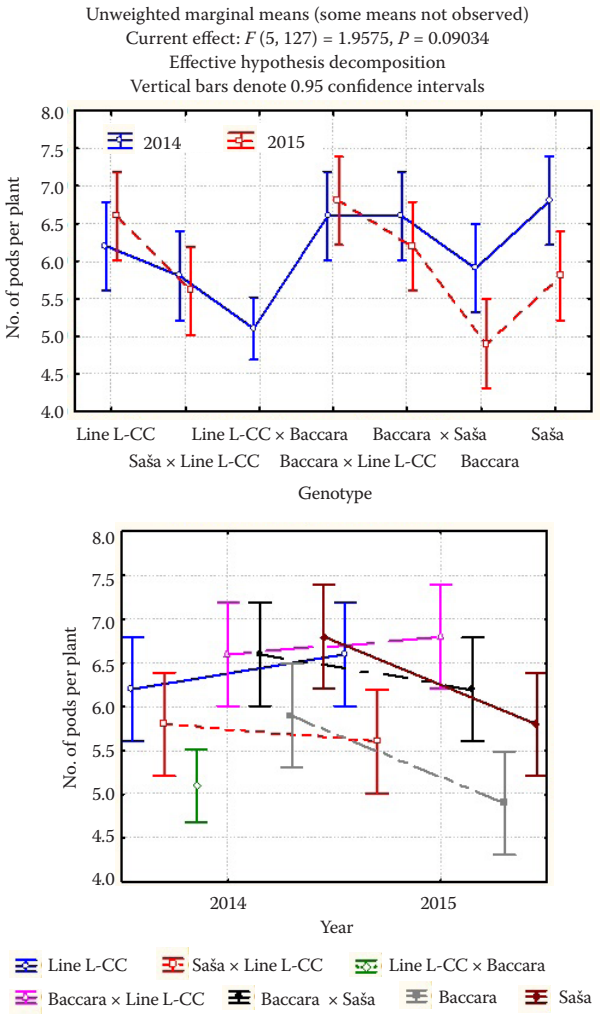

Figure 3. Genotype × year interaction for the number of pods per plant

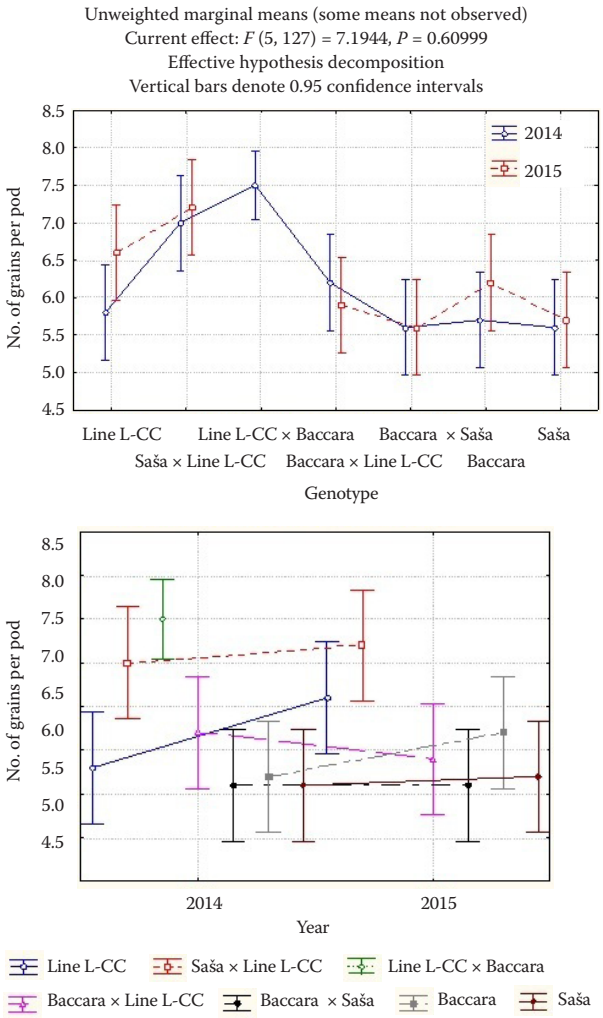

Figure 4. Genotype × year interaction for the number of grains per pod

genotypes, lines and hybrids, the average number of pods was 6.0. There was a significantly larger ($P < 0.05$) number of pods in 2014 than in 2015 (Table 2, Figure 3).

When compared with all investigated genotypes, lines and hybrids, a significantly smaller number of pods per plant was recorded for L-CC line × Baccara hybrid (5.10) and Baccara genotype (5.40). The largest number of pods per plant was recorded for Baccara × L-CC line (6.70). The aforementioned hybrid had a significantly larger ($P < 0.01$) number of pods compared to all investigated genotypes, lines and hybrids (Table 2, Figure 3).

The number of pods per plant is a variable property. Our results are in agreement with the results of Kosev (2015). Erić *et al.* (2004) reported that during their research on the impact of sowing time on yield components they obtained 5.45 to 7.15 pods per plant, and the average of 6.3 pods per plant, which is also in accordance with our results. Yücel Özveren (2013) recorded 9.3–17.4 pods per plant during his research on field pea yield components.

Table 3. The analysis of variance for the number of pods per plant, number of grains per pod, 1000 seed weight and grain yield

Parameter	LSD	Genotype	Year	G × Y
No. of pods per plant	0.05	0.584	0.315	0.826
	0.01	0.298	0.413	1.082
No. of grains per pod	0.05	0.617	0.329	0.873
	0.01	0.807	0.432	1.143
1000 seed weight	0.05	14.444	7.720	20.427
	0.01	18.910	10.107	26.743
Grain yield	0.05	418.099	223.812	592.194
	0.01	547.368	293.012	775.291

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The results shown in Tables 2–4 indicate that the genotype had a great influence on the number of seeds per pod. The genotype and the genotype × year interaction had a significant ($P < 0.05$) influence on the number of seeds per pod (Figure 5).

A significantly larger ($P < 0.05$) number of grains per pod than in all other genotypes, lines and hybrids was recorded for the following hybrids: L-CC line × Baccara (7.50) and Saša × L-CC line (7.10). The smallest number of seeds per pod was recorded for Saša genotype and Baccara × Saša hybrid (Table 2 and 4, Figure 4).

The number of seeds per pod varies and it can be small (3–4), medium (5–6) and large (7–12). During these experiments, the hybrids L-CC line × Baccara and Saša × L-CC line had, on average, more than 7 seeds per pod. It can be said for these hybrids that they have a large number of seeds per pod. The hybrid Baccara × L-CC line and L-CC line had, on average, more than 6 seeds per pod. Similar results were achieved by Ali *et al.* (2012) and Rashwan and El-Shaieny (2016). Sibhatu *et al.* (2016) found that the sowing spacing did not affect significantly the number of grains per pod. The aforementioned authors reported that the number of grains per pod ranged from 3.33 to 4.33.

The genotype and the genotype × year interaction had a significant ($P < 0.05$) influence on 1000 seed weight (Tables 2 and 4). Significantly higher ($P < 0.05$) 1000 seed weight than in all other investigated genotypes, lines and hybrids was recorded for the Baccara genotype (229 g). Baccara × L-CC line and Baccara × Saša had greater 1000 seed weight than all other genotypes, lines and hybrids, except for the Baccara genotype. The lowest 1000 seed weight was recorded for the L-CC line.

The 1000 seed weight is an important trait, especially when determining the amount of seeds for sowing. By the 1000 seed weight we distinguish small-seeded genotypes (up to 150 g), medium- (150–250 g) and large-seeded genotypes (more than 250 g). According

Table 4. Fisher's LSD test for the number of grains per pod, 1000 seed weight and grain yield

Genotype	Line L-CC	Saša × Line L-CC	Line L-CC × Baccara	Baccara × Line L-0CC	Baccara × Saša	Baccara	Saša
No. of grains per pod							
Line L-CC	–	0.006074	0.000095	0.642660	0.065145	0.439687	0.090574
Saša × Line L-CC	0.006074	–	0.217168	0.001450	0.000008	0.000512	0.000015
Line L-CC × Baccara	0.000095	0.217168	–	0.000015	0.000000	0.000004	0.000000
Baccara × Line L-CC	0.642660	0.001450	0.000005	–	0.165365	0.757021	0.217168
Baccara × Saša	0.065145	0.000008	0.000000	0.757021	–	0.279879	0.877042
Baccara	0.439687	0.000512	0.000004	0.757021	0.279873	–	0.354037
Saša	0.090574	0.000015	0.000000	0.217168	0.877042	0.354037	–
1000 seed weight							
Line L-CC	–	0.453919	0.002461	0.000001	0.000000	0.000000	0.018702
Saša × Line L-CC	0.453919	–	0.020919	0.000021	0.000000	0.000000	0.105408
Line L-CC × Baccara	0.002461	0.020919	–	0.039618	0.002112	0.000000	0.480385
Baccara × Line L-CC	0.000001	0.000021	0.039618	–	0.291456	0.000000	0.006139
Baccara × Saša	0.000000	0.000000	0.002112	0.291456	–	0.000005	0.000189
Baccara	0.000000	0.000000	0.000000	0.000000	0.000005	–	0.000000
Saša	0.018702	0.105408	0.480385	0.006139	0.000189	0.000000	–
Grain yield							
Line L-CC	–	0.008812	0.000004	0.000000	0.000001	0.000000	0.000546
Saša × Line L-CC	0.008812	–	0.030181	0.004317	0.011563	0.000086	0.376658
Line L-CC × Baccara	0.000004	0.030181	–	0.476559	0.711918	0.064247	0.194216
Baccara × Line L-CC	0.000000	0.004317	0.476559	–	0.731523	0.251178	0.045585
Baccara × Saša	0.000001	0.011563	0.711918	0.731523	–	0.136977	0.096352
Baccara	0.000000	0.000086	0.064247	0.251178	0.136977	–	0.001900
Saša	0.000546	0.376658	0.194216	0.045585	0.096352	0.001900	–

Probabilities for post hoc tests; degrees of freedom (df) =127.00; mean squared error = 1.0402 (No. of grains per pod), 539 (1000 seed weight) and 4550 E2 (grain yield)

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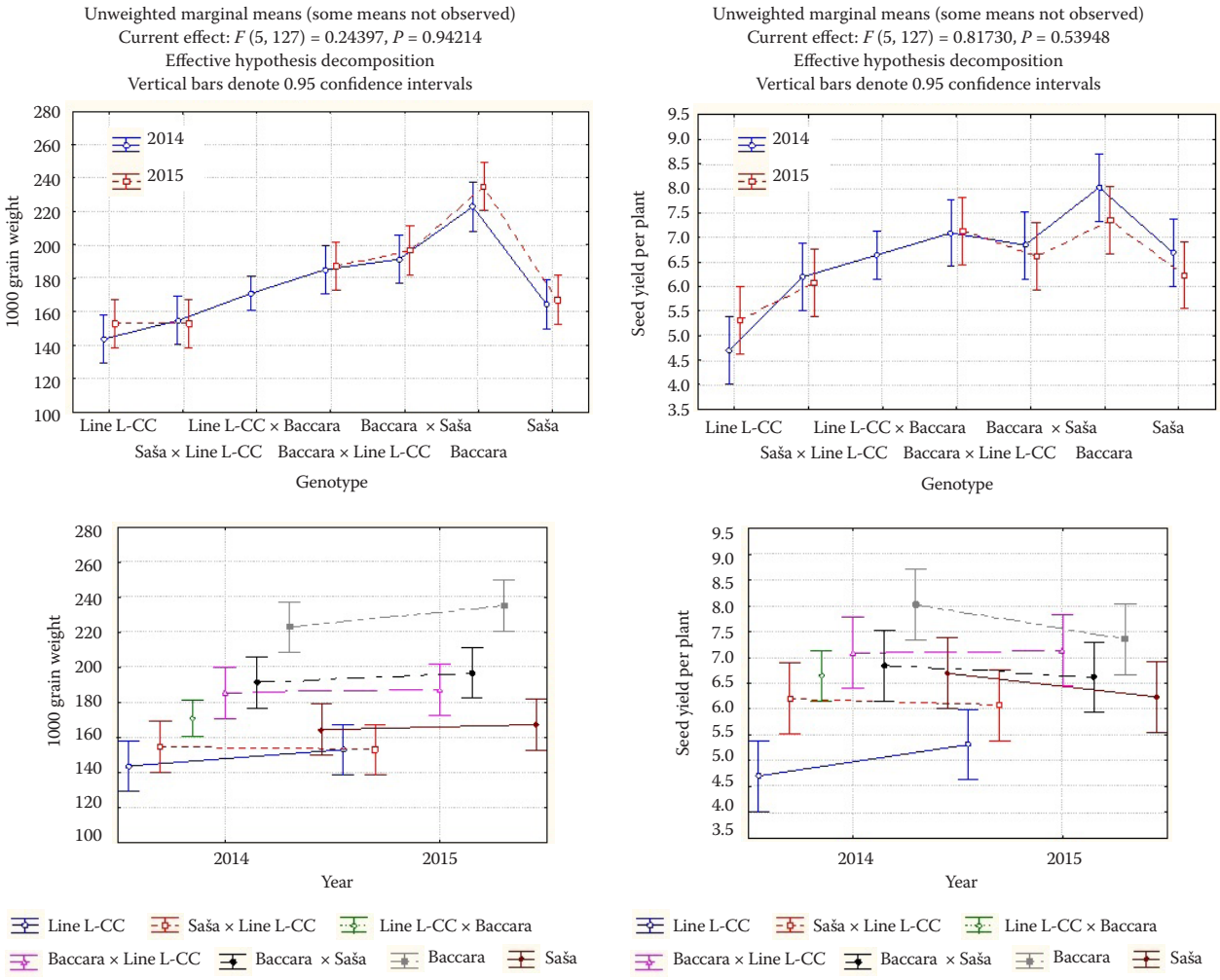

Figure 5. Genotype × year interaction for 1000 grain weight

Figure 6. Genotype × year interaction for seed yield

to the determined values of 1000 seed weight, the investigated hybrids and genotypes were grouped into the medium-sized grain genotypes. The L-CC line is an exception, which had the average 1000 seed weight of 148.4 grams and according to the above-mentioned classification it belongs to the small-sized grain genotypes. Field pea cultivars are usually medium-sized and are grown for grain production or combined use. The results of 1000-seed test are in accordance with the results of Türk *et al.* (2011), Kosev (2014, 2015). In local pea populations from Eastern Anatolia, Turkey, Tan *et al.* (2013) found a large variation in 1000 seed weight, ranging from 63.3 (Balcesme) to 207.7 g (Incili).

The genotype and the genotype × year interaction had a significant ($P < 0.05$) influence on grain yield (Table 2 and 4). During these investigations, a significantly lower grain yield than in all other genotypes, line and hybrids was recorded for the L-CC line (3546 kg/ha).

During these tests a high grain yield was achieved by Baccara and Baccara × L-CC line and Baccara × Saša hybrids (Table 2 and 4, Figure 6).

For all the investigated genotypes, lines and hybrids the average grain yield was 4416.6 kg/ha. Total average yields between the years were not statistically significantly different. However, Baccara and Saša × L-CC line had a significantly higher ($P < 0.01$) yield in 2014 than in 2015. L-CC line × Baccara had a significantly higher ($P < 0.05$) grain yield in 2014 than in 2015. During 2015, L-CC and Saša achieved higher yields than in the previous year (Table 2 and 4, Figure 6).

The seed yield of field pea depends on a number of factors, such as genotype, growing conditions and genotype × environment interaction. During these experiments, the average grain yields of field pea were high. Our results are in accordance with the results of Erić *et al.* (2004), Spies *et al.* (2010) and Rasaei *et al.* (2012). When Bokan *et al.*(2013) studied several genotypes of spring field pea, the pea

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grown for fodder and grain yielded 1.370–2.540 kg/ha and the pea intended for grain 1.930–2.780 kg/ha.

CONCLUSION

Based on the results of the investigation of variability in quantitative traits of field pea, one can draw the following conclusions:

A statistically significantly larger number of grains per plant, compared to all other genotypes, lines and hybrids, was recorded for the hybrids L-CC line × Baccara and Saša × L-CC line.

The genotype and genotype × year interaction had a significant influence on 1000 seed weight.

Significantly higher 1000 seed weight compared to all the other investigated genotype, lines and hybrids was recorded for the Baccara genotype.

The genotype and genotype × year interaction had a significant influence on the grain yield of field pea.

High grain yields were achieved by the Baccara genotype and its hybrids: Baccara × L-CC line and Baccara × Saša.

Based on the results of this research, it is evident that two tested genotypes, Baccara × L-CC line and Baccara × Saša, due to their average grain yield, 1000 seed weight and number of pods per plant, can be taken for further research.

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