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Original scientific paper

CORRELATION AND PATH COEFFICIENT ANALYSIS FOR PROTEIN YIELD IN CONFECTIONARY SUNFLOWER (*Helianthus annuus* L.)

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The most important criteria for introducing new confectionary hybrids into the production is high protein yield. Path coefficient analysis was used to obtain information on direct and indirect effects of studied traits (seed oil content, kernel oil content, seed yield, kernel protein content, mass of 1000 seeds, kernel ratio and hull ratio) on protein yield. The research was conducted during three vegetation seasons, on 22 experimental confectionary sunflower hybrids created in the breeding program at the Institute of Field and Vegetable Crops. Strong and very strong correlations were found among the largest number of examined traits. A weak negative interdependence was determined between kernel oil content, kernel protein content, mass of 1000 seeds, hull ratio, and protein yield using the analysis of simple correlation coefficients. Positive but weak correlation was determined between protein yield and seed oil content, and kernel ratio. Very strong positive correlation was determined between protein yield and seed yield (0.468^{**}). The seed oil content had a very strong direct negative effect on protein yield ($DE=-0.734^{**}$). The mass of 1000 seeds had a weak negative direct effect on protein yield. Kernel protein content and kernel oil content demonstrated a weak direct positive effect on protein yield. Path coefficient analysis of protein yield showed a very strong positive direct effect of kernel ratio ($DE=1.340^{**}$), seed yield ($DE=0.657^{**}$) and hull ratio ($DE=0.992^{*}$). These findings confirm the effect of seed yield, kernel ratio, and hull ratio on protein yield, and their importance as the selection criteria in confectionary sunflower breeding.

Key words: confectionary sunflower, correlations, path coefficient analysis, protein yield, yield components.

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INTRODUCTION

The major sunflower breeding objectives for all sunflower types should be high yields and quality of oil, proteins and other products for non-food industries and approaches to management of resistance genes, stability of sunflower resistance to certain pathogens (ŠKORIĆ *et al*., 2012). The creation of productive sunflower hybrids demands a formation of a model of hybrids for certain agro-ecological conditions while determining the priority when breeding for most important traits (HLADNI, 2010). Knowing the interdependence of morphophysiological traits with yield is of utmost importance in order for their breeding programs to be successful (ŠKORIĆ *et al*., 2012). For confectionary sunflower the main direction in breeding is directed towards increased mass of 1000 seeds and protein content and quality while lowering the seed oil content and hull ratio (HLADNI *et al*., 2011c). Although the favored seed color of confectionary hybrid in Turkey is white with grey stripes, consumers from Balkan countries such as Serbia, Bulgaria, Moldova and Romania prefer black seeds (SINCIK and GOKSOY, 2014).

A successful breeding program for confectionary sunflower, aimed at determining the associations between the traits, requires knowledge of a well-defined association between agronomical and technological characteristics.

Seed protein content is one of the indicators of sunflower seed quality. According to HLADNI *et al*. (2009a), protein content varies and, depending on the genotype, agroecological conditions and the interaction of the genotype and external environment conditions, it in the range between 16-28%. With kernel increase, the amount of protein in the seed also increases so breeding for increased seed protein amount should be followed by the selection of genotypes with larger kernels (HLADNI *et al*., 2009b). Proteins of sunflower seeds have high digestibility and high biological value and hence the increase in their use as a component of functional foods and a nutritionally balanced diet, especially in this day and age when consumers wish to protect themselves from genetically modified soy protein products (DIMIĆ *et al*., 2006). A strong negative correlation between hull ratio, seed yield and mass of 1000 seeds was determined by (ERGEN and SAGLAM, 2005).

KHOLGHI *et al*. (2011) found that the mass of 1000 seeds had positive direct effects on seed yields of confectionary sunflowers. The results of the path analysis revealed that the seed yield and the kernel protein content had very strong direct positive effects on the protein yield in both experimental years and in the combined data SINCIK and GOKSOY (2014).

Presence or absence of correlations can contribute to the right choice of examined traits so as to enhance the efficiency of some selection criteria. The focus should be placed on traits that have a very strong positive correlation on seed yield (HLADNI *et al*., 2011b).

The aim of this paper was to determine the correlation coefficient, direct and indirect effects by path analyses between sunflower protein yield and seed yield, mass of 1000 seeds, seed oil content, kernel oil content, kernel protein content, kernel ratio, and hull ratio.

MATERIALS AND METHODS

The research was conducted during three vegetation seasons on 22 experimental confectionary sunflower hybrids, produced within the breeding program at the Institute of Field and Vegetable Crops. Twenty-two new high protein two-line confectionary hybrids: NS-H-1, NS-H-2, NS-H-3, NS-H-4, NS-H-5, NS-H-6, NS-H-7, NS-H-8, NS-H-9, NS-H-10, NS-H-11, NS-H-12, NS-H-13, NS-H-14, NS-H-15, NS-H-16, NS-H-17, NS-H-18, NS-H-19, NS-H-20, NS-H-21, NS-H-22, created by crossing cytoplasmic male sterile female line and male line with a fertility restorer gene, were examined during three vegetation seasons 2008, 2009, 2010 at three locations Rimski Šančevi, Erdevik in the Vojvodina region, and Kula in Central Serbia. The following traits were examined: mass of 1000 seeds, seed oil content, kernel oil content, kernel protein content, hull ratio, kernel ratio, seed yield and protein yield. The plot where the experiment was conducted was 28 m^2 and (70cm x 28cm plant density) seeds were planted by hand in 4 rows (in April) and all plants from two mean rows were harvested (in September) except first plants at each plot. The experiment was done in a randomized complete block design with 3 replications. Seed yield was measured on a scale and calculated to 11% seed humidity content. Seed oil content and kernel oil content (%) from absolutely clean and air-dried seed was determined with the NMR-analyzer. Kernel protein content was determined using the Kjeldahl method. Mass of 1000 seeds (g) was measured on a random sample of an absolutely clean and air-dried seed. Protein yield was determined from seed yield and seed protein content (tha^{-1}). The determination of the hull/kernel ratio in seed was performed by dehulling the seed sample and separation into kernel and hull.

Mutual relations between relations between relationships of the examined characteristics, and direct and indirect effects on seed yield, were analyzed by the path coefficient analysis (WRIGHT, 1921; DEWEY and LU, 1959; IVANOVIĆ and ROSIĆ, 1985). Statistical analysis was performed using MSTAT-C (1991) and SAS System Software (2003) programs.

RESULTS AND DISCUSSION

When creating new high-protein hybrids for confectionary use it is important to find traits that are easily determined and at the same time show their interdependence with protein yield, based on which that those traits can become selection criteria (HLADNI *et al*. 2011a).

The analysis of simple correlation coefficients revealed a negative weak correlation between kernel protein content, mass of 1000 seeds and hull ratio, and protein yield, a positive weak correlation between seed oil content and kernel ratio, and protein yield, and a positive very strong correlation between seed yield (0.468^{**}) and protein yield (Table 1).

Table 1. Simple correlation coefficients between different observing characters

Trait		KOC	SY	KPC	MTS	KR	HR	PY
		X2	X3	X4	X5	X6	X7	Y
SOC	X1	0.552^{**}	0.542^{**}	-0.398^{*}	-0.716^{**}	0.850^{**}	-0.811^{**}	0.006
KOC	X2		0.032	-0.340^{*}	-0.255^{*}	0.441^{**}	-0.402^{*}	-0.104
SY	X3			-0.327^{*}	-0.494^{**}	0.605^{**}	-0.592^{**}	0.468^{**}
KPC	X4				0.278^{*}	-0.510^{**}	0.495^{**}	-0.041
MTS	X5					-0.601^{**}	0.589^{**}	-0.099
KR	X6						-0.973^{**}	0.165
HR	X7							-0.118

** *F test significance at level P<0.01* * *F test significance at level P<0.05* ns- *not significantly different*

X1	Seed oil content (SOC)	X5	Mass of 1000 seeds (MTS)
X2	Kernel oil content (KOC)	X6	Kernel ratio (KR)
X3	Seed yield (SY)	X7	Hull ratio (HR)
X4	Kernel protein content (KPC)	Y	Protein yield (PY)

The correlation analyses of the combined two years of data revealed that protein yields were significant and positively correlated with the mass of 1000 seeds (r=0.699) and seed yield (r=0.852) (SINCIK and GOKSOY 2014).

The analysis of simple correlation coefficient showed a very strong positive correlation between seed oil content and seed yield (0.542^{**}); this is in agreement with the research performed by KAYA *et al*. (2007), but in contradiction to the research performed by HLADNI *et al*. (2008) who determined a strong negative correlation between seed yield and seed oil content (-0.649^{**}). A negative weak correlation between seed oil content and seed yield was determined by ARSHAD *et al*. (2010). Simple correlation coefficient showed a very strong negative correlation between the mass of 1000 seeds and seed yield; these results are in disagreement with the results of DUŠANIĆ *et al*. (2004), KAYA *et al*. (2008), BEHRADFAR *et al*. (2009), MIJIĆ *et al*. (2009), HLADNI *et al*. (2010) and SINCIK and GOKSOY (2014).

Table 2. Analysis of direct and indirect effects of observed characters on protein yield

Components	Direct effect via: DE(P)	Indirect effect via: IE(Pxr)							CC(r)
		SOC	KOC	SY	KPC	MTS	KR	HR	
SOC	-0.734^{**}		0.066	0.356	-0.051	0.034	1.139	-0.805	0.006
KOC	0.120	-0.405		0.021	-0.044	0.012	0.591	-0.399	-0.104
SY	0.657^{**}	-0.398	0.004		-0.042	0.024	0.811	-0.587	0.468
KPC	0.128	0.292	-0.041	-0.215		-0.013	-0.683	0.491	-0.041
MTS	-0.048	0.526	-0.030	-0.325	0.036		-0.805	0.584	-0.099
KR	1.340^{**}	-0.624	0.053	0.397	-0.065	0.029		-0.965	0.165
HR	0.992^{*}	0.595	-0.048	-0.389	0.063	-0.028	-1.304		-0.118

Coefficient of R^2 determination R= 0.391

Seed yield exerted a strong negative interdependence with kernel protein content, very strong negative correlation with mass of 1000 seeds and hull ratio, and very strong positive correlation with kernel ratio and seed oil content. These results are in disagreement with GOKSOY and TURAN (2007), AHMAD *et al*. (2013), and RADIĆ *et al*. (2013), who determined the strong positive correlation between seed yield and mass of 1000 seeds.

Strong negative correlations were found between kernel protein content and seed oil content, a very strong negative correlation between hull ratio, mass of 1000 seeds and seed oil content, and very strong positive correlation between seed kernel content and kernel ratio and seed oil content. These results are in agreement with research by OZER *et al*. (2003), MIJIĆ *et al.* (2009), ZHANG *et al*. (2010), XIANG *et al*. (2010) and HLADNI *et al*. (2010).

ZHANG *et al*. (2010) determined a strong positive correlation between seed oil content and kernel oil content, and a strong negative correlation between seed oil content and hull ratio. XIANG *et al*. (2010) and OZER *et al*. (2003) determined a very strong positive correlation between seed oil content and kernel ratio.

MIJIĆ *et al.* (2009) and HLADNI *et al*. (2010) determined strong negative interdependence between mass of 1000 seeds and seed oil content.

These results are in disagreement with research by OZER *et al*. (2003) who determined strong positive correlation of mass of 1000 seed and seed oil content, and RADIĆ *et al*. (2013) who determined positive and weak correlations between mass of 1000 seeds and seed oil content.

Kernel protein content demonstrated a very strong negative interdependence with kernel ratio, a strong interdependence with a positive direction with mass of 1000 seeds, and a very strong positive correlation with hull ratio. The analysis of simple correlation coefficient shows a very strong negative correlation between mass of 1000 seeds and kernel ratio, and very strong positive correlation with hull ratio, which is consistent with the research of LI *et al*. (2010). A very strong correlation with a negative direction was found between kernel ratio and hull ratio; these results are in agreement with the research of ZHANG *et al*. (2010), who determined a significantly negative correlation between kernel ratio and hull ratio in seed.

Higher protein yield is an ultimate objective of confectionary sunflower researchers.

The focus should be placed on the traits with very strong direct effects on seed protein yield.

Presence or absence of correlations can contribute to the right choice of examined traits so as to enhance the efficiency of some selection criteria. The focus should be placed on traits that have a very strong positive direct effect on protein yield. Plant breeders commonly prefer yield components that indirectly increase yield (KAYA *et al*., 2007).

Path coefficient analysis provides a much clearer picture of the effect of certain independent variables on protein yield. Kernel ratio demonstrated very strong direct effect of a positive direction on protein yield (DE=1.340**), while the simple correlation coefficient demonstrated a weak positive correlation. The effect of the simple correlation coefficient was masked by the negative indirect effect of the kernel ratio through seed oil content (IE=-0.624), and the positive indirect effect of kernel ratio through seed yield (IE=0.397). Seed yield demonstrated a very strong highly significant positive direct effect on protein yield (DE=0.657**) which is in accordance with the simple correlation coefficient (Table 2). These results are in agreement with SINCIK and GOKSOY (2014). Hull ratio demonstrated a strong positive direct effect (DE=0.992*) on protein yield while the simple correlation coefficient is weak and with a negative direction.

SIMPLE coefficient of the correlation of hull ratio is negative and weak in comparison to the direct effect; that relation was masked by a positive indirect effect of the hull ratio through seed oil content (IE=0.595), and a negative indirect effect through seed yield (IE=-0.389).

Seed oil content has very strong direct effect of a negative direction on protein yield (DE=-0.734**), while the simple correlation coefficient demonstrated a weak positive correlation; that relation was masked by a positive indirect effect of the seed oil content through kernel ratio (IE=1.139) and a negative indirect effect through hull ratio (IE=-0.805).

The direct effect of kernel protein content on protein yield is weak and of positive direction. Simple correlation coefficient demonstrated a weak negative correlation; its effect was masked by the positive indirect effect of the kernel protein content through the hull ratio

(IE=0.491) and seed oil content (IE=0.292), and the negative indirect effect of kernel protein content through seed yield (IE=-0.215) and kernel ratio (IE=-0.683).

Relation between mass of 1000 seeds and protein yield is weak and of negative direction (CC=-0.099), with direct effect. KHOLGHI *et al*. (2011) found that the mass of 1000 seeds weight had positive direct effects on seed yields of confectionary sunflowers.

CONCLUSION

Higher protein yield is an ultimate objective of confectionary sunflower researchers. The focus should be placed on traits with very strong direct effects on seed protein yield. The path analysis indicated that the kernel ratio and seed yield demonstrated a very strong positive direct effect on protein yield (DE=1.340**), (DE=0.657**) and hull ratio (DE=0.992*) showed a strong positive direct effect on protein yield. Seed oil content had a very strong negative direct effect on protein yield (DE=-0.734**). On the basis of these investigations it was observed that the kernel ratio, seed yield, and hull ratio were the most important traits for protein yield research, and can be used for the improvement of protein yield of confectionary sunflowers.

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KORELACIJA I PATH KOEFICIJENT ANALIZA ZA PRINOS PROTEINA KOD KONZUMNOG SUNCOKRETA (*Helianthus annuus* L.)

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Izvod

Najvažniji kriterijum za uvođenje novih konzumnih hibrida u proizvodnju je visok prinos proteina. Path koeficijent analiza je primenjena radi dobijanja informacija o direktnim i indirektnim efektima proučavanih svojstava (sadržaj ulja u semenu, sadržaj ulja u jezgru, prinos semena, sadržaj proteina u jezgru, mase 1000 semena, udeo jezgra i udeo ljuske) na prinos proteina suncokreta. Svojstva koja imaju najveći uticaj na povećanje prinosa proteina suncokreta će se koristiti kao selekcioni kriterijum u oplemenjivanju konzumnog suncokreta. Istraživanje je sprovedeno tokom tri vegetacione sezone sa 22 eksperimentalna konzumna hibrida suncokreta, stvorena u oplemenjivačkom programu Instituta za ratarstvo i povrtarstvo. Između največeg broja ispitivanih svojstava postojale su jake i vrlo jake korelacije. Analiza prostih koeficijenata korelacije je pokazala da postoji slaba negativna međuzavisnost prinosa proteina sa sadržajem ulja u jezgru, sadržajem proteina u jezgru, masom 1000 semena, udelom ljuske. Uočena je pozitivna slaba korelacija prinosa proteina sa sadržajem ulja u semenu i udelom jezgra. Pozitivna veoma jaka korelacija prinosa semena (0.468^{**}) sa prinosom proteina. Sadržaj ulja u semenu ima jako jak direktan negativan uticaj na prinos proteina ($DE=-0.734^{**}$). Masa 1000 semena je imala slab negativan direktan uticaj na prinos proteina. Sadržaj proteina u jezgru i sadržaj ulja u jezgru su pokazali slab pozitivan direktan uticaj na prinos proteina. Path koeficijent analiza je pokazala jako jak pozitivan direktni efekat udela jezgra ($DE=1.340^{**}$), prinosa semena ($DE=0.657^{**}$) i udela ljuske ($DE=0.992^{*}$) na prinos proteina. To pokazuje da udeo jezgra, prinos semena i udeo ljuske imaju velik uticaj na prinos proteina i predstavljaju važne selekcione kriterijume kod oplemenjivanja konzumnog suncokreta.

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