EFFECT OF MORPHOLOGICAL AND PHYSIOLOGICAL TRAITS ON SEED YIELD AND OIL CONTENT IN SUNFLOWER

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SUMMARY

Path coefficient analysis was used to separate direct and indirect effects of studied traits on seed oil content and seed yield, and to identify traits that could be used as selection criteria in sunflower breeding. Significant and highly significant correlations were found between most of the examined traits. Highly significant negative correlations have been established between stem diameter, total leaf area per plant, head diameter and 1,000-seed weight on one side and seed oil content on the other. Highly significant positive correlations have been established between seed yield on one side and stem diameter, total leaf area per plant, head diameter, total number of seeds per head, and the 1,000-seed weight on the other. The weight of 1,000 seeds had a highly significant direct negative effect on seed oil content and a highly significant direct positive effect on seed yield. The total number of seeds per head has demonstrated a highly significant direct positive effect on seed yield and oil content. The total leaf number per plant has demonstrated a significant direct positive effect on seed yield. A path coefficient analysis of seed yield at phenotypic level indicated that maximum direct effects were shownby the weight of 1,000 seeds, total number of seeds per head and total leaf number per plant. The total number of seeds per head was the most important trait that determined seed yield and oil content. The obtained results can be used in sunflower breeding programs aimed at the development of sunflower hybrids that combine high genetic potentials for seed yield and seed oil content.

Key words: _Helianthus annuus_, **hybrid, morphological and physiological traits, correlation, seed yield, oil content**

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INTRODUCTION

Sunflower is globally one of the most important oil crops and its oil is counted among the highest quality vegetable oils on the market (Škorić *et al.,* 2007). In Serbia, sunflower is the main oil crop. The sunflower acreage varies from 160 to 210 thousand ha, with the seed yield ranging from 1.7 to 2.3 t/ha. Considering the sunflower acreage, Serbia is in the seventh place in Europe and by yield level right behind France and Hungary (Miklič *et al.,* 2007).

The increasing demand from growers on the local and the international market for higher quality and higher yielding sunflower hybrids calls for an immediate replacement of the currently grown hybrids with better, more productive and more stable ones (Miklič *et al.,* 2010).

The main objectives of sunflower breeding at Institute of Field and Vegetable Crops in Novi Sad is the development of hybrids with a high genetic seed yield potential (above 5 t/ha) and high seed oil content (>50%), which would provide the oil yield per hectare of more than 2.5 t (Miklič *et al.,* 2008).

Seed yield and oil content are the two most important economic traits in sunflower. However, they are under a strong influence of environmental factors, they have a complex inheritance and low heritability. That is why the breeding for these complex traits demands knowledge of relationships among plant traits that affect their formation. When starting a breeding program, choice of parents is important for achieving both short and long term objectives.

Increasing seed yield and oil content is the top priority of most sunflower breeding programs. Breeding for high seed yield and seed yield components and development of new sunflower ideotypes demands an increased genetic variability which is accomplished by interspecific hybridization (Škorić *et al.,* 2007). In sunflower breeding for high yield peformance, it is necessary to find morphological and physiological traits, which are easily improved and which show correlation with seed yield and oil content, so that they can be used as selection criteria (Hladni *et al.,* 2008).

Relationships between seed yield and morphological and physiological traits are effectively studied by the simple correlation coefficient analysis (Škorić, 1974; Marinković, 1992; Hladni *et al.,* 2006). As the simple correlation analysis cannot fully explain the relationships between traits, the path coefficient analysis is introduced for a more successful breeding work. This type of analysis enables to partition correlation coefficients to their components which in turn allows to distinguish a direct effect of one variable from indirect effects of other variables, thus giving a clear picture of the individual contribution of each variable to seed yield and oil content.

Total leaf area and plant height (Hladni *et al.,* 2004), total seed per head (Gonzales *et al.,* 2000) and 1,000-seed weight (Marinković, 1992; Gonzales *et al.,* 2000) were found to extend positive direct effects on seed yield.

The cultivated sunflower shows a significant variability in oil content, while the oil content in wild species is generally low (Seiler, 1992). Origin of plant material and weather conditions in the year of study have a high impact on seed oil content (Hladni *et al.*, 2006).

Positive and highly significant relationships exist between seed oil content on one side and head diameter, number of seeds per head and 1,000-seed weight on the other (Marinković, 1988; Marinković *et al.,* 1994).

In this paper, we studied mutual relationships between several morphological and physiological traits (stem diameter, total leaf number per plant, total leaf area per plant, plant height, and head diameter, total seed number per head, 1,000-seed weight) and seed yield and oil content in a group of new experimental sunflower hybrids.

MATERIALS AND METHODS

In this study we used 21 experimental hybrids developed from new divergent (A) *cms* inbred lines. The female inbred lines, developed by interspecific hybridization from various populations [NS-GS-1, NS-GS-2 (RES-834-1), NS-GS-3 (DEB-SIL-3672), NS-GS-4, NS-GS-5 (PRA-RUN-1321-1), NS-GS-6, NS-GS-7 (DES-1474-2)] and restorer lines possessing good combining characteristics (RHA-R-PL-2/1, RHA-N-49, RUS-RF-OL-168) had been developed at Institute of Field and Vegetable Crops, Novi Sad.

The experiment was established at Rimski Šančevi experiment field of Institute of Field and Vegetable Crops, in a randomized complete block system with three replications, during two growing seasons. The soil in the experiment plots had 2.8% humus content, moderate contents of phosphorus and potassium and pH 6.92 (Vasin *et al.,* 2002).

The examined traits were analyzed in samples that consisted of thirty plants (ten plants per replication) sampled from inside rows of each block.

At the stage of physiological maturity, plant height (PH) and head diameter (HD) were measured (cm) in field. Plants at the flower stage were taken to the laboratory for measurements of the total leaf number per plant (TLN), stem diameter (STD; cm), and total leaf area per plant (TLA; cm^2/plant). The last trait was measured with a leaf area meter (LI-300-LiCor, USA). After harvest, seed yield per individual plants (SY), produced in open pollination, was weighed in laboratory. The number of filled seeds per head (total seed number-TSN) was counted. The weight of 1,000 seeds (M1000S; g) was determined in a random sample of cleaned and air dried seed. Seed oil content (SOC) was analyzed non-destructively by the method of nuclear magnetic resonance (NMR). Mean values and correlation coefficients (r) were determined according to Hadživuković (1991). Direct and indirect effects of the examined traits on seed yield were analyzed by the path coefficient analysis

(Wright, 1921; Dewey, 1952; Ivanović, 1985). Statistical analysis was performed using MSTAT-C (1991) and SAS System Software (2003) programs.

RESULTS AND DISCUSSION

The main objective of sunflower breeding is to develop new hybrids with a high genetic potential for seed yield and oil content. Additional information are still needed to clarify relationships between seed oil content, seed yield and their components. Correlations between different traits are an aspect that should be kept in mind for better planning of breeding programs in sunflower.

Table 1: Simple correlation coefficients of seven yield components and their effects on seed yield and oil content in sunflower.

Trait		TLN	TLA	PH	HD	TSN	M1000S	SOC	SY
		X2	X3	X4	X5	X6	X7	Y1	Y2
STD	X1	-0.514^{**}	0.776^{**}	-0.456^{**}	0.642^{**}	0.038^{ns}	0.379^{*}	-0.419^{**}	0.411^{**}
TLN	X2		-0.202^{ns}	0.566^{**}	-0.452^{**}	-0.075^{ns}	0.010^{ns}	0.168^{ns}	0.087^{ns}
TLA	X3			-0.161^{ns}	0.602^{**}	0.253^{*}	0.461^{**}	-0.457^{**}	0.623^{**}
PH	X4				-0.544^{**}	0.040^{ns}	0.220^{ns}	-0.009^{ns}	0.200^{ns}
HD	X5					0.297^{*}	0.291^{*}	-0.588^{**}	0.446^{**}
TSN	X6						-0.164^{ns}	0.091^{ns}	0.369^{**}
M1000S	X7							-0.786^{**}	0.790^{**}

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

X1	Stem diameter (STD)	X6	Seed number per head (TSN)
X2	Total leaf number per plant (TLN)	X7	Weight of 1000 seeds (M1000S)
X3	Total leaf area per plant (TLA)	Y1	Seed oil content (SOC)
X4	Plant height (PH)	Y2	Seed yield per plant (SY)
X5	Head diameter (HD)		

The analysis of simple correlation coefficients showed that very high correlations existed between the 1000-seed weight (-0.786^{**}), head diameter (-0.588^{**}), total leaf area per plant (-0.457^{**}), stem diameter (-0.419^{**}) on one side and seed oil content on the other. Very high positive correlations were found between these traits (0.790^{**}; 0.446^{**}; 0.623^{**}; 0.411^{**}, respectively) and seed yield per plant. These results are in agreement with those of Punnia and Gill (1994), who found a significant negative correlation between the 1,000-seed weight and seed oil content. However, our results disagree with those of Marinković *et al.* (1994), who found a highly significant positive correlation between seed oil content on one side and head diameter, total seed number per head and 1,000-seed weight on the other. Highly significant correlations were found between seed yield per plant and the 1,000-seed weight (Yalcin *et al.,* 2007; Behradfar *et al.,* 2009), total leaf area per plant (Merrien *et al.,* 1982; Hladni *et al.,* 2001), and head diameter (Hladni *et al.,* 2003; Mijić *et al.,* 2006; Thitiporn, 2008). A positive relationship between seed yield and stem

diameter was reported by Punnia and Gill (1994), Ashok *et al.* (2000) and Ozer *et al.* (2003).

Simple correlation coefficients showed that the total leaf number per plant and total seed number per head exhibited a positive but low correlation with seed oil content (0.623[**]) and a very high positive correlation with seed yield per plant (0.369[**]). A low positive correlation was found between seed oil content and total seed number per head. This result is in agreement with those of Khan *et al.* (2005) who found a significant negative correlation between seed oil content and total seed number per head. The highly significant correlation between seed yield per plant and total seed number is in accordance with the resuls of Dagustu (2002) and Dušanić *et al.* (2004).

Plant height has exhibited a low negative correlation with seed oil content and a low positive correlation with seed yield per plant (Table 1). A low negative correlation was found between seed oil content and plant height. This result is in agreement with those of Arshad *et al.* (2007). Significant positive correlation was not established between plant height and seed yield per plant although it had been reported by other researchers (Marinković, 1992; Hladni *et al.,* 2003; Mijić *et al.,* 2006; Habib *et al.,* 2007).

The analysis of path coefficients gave a much clearer picture of the effect of certain independent variables on seed yield and oil content in sunflower plants. This analysis allows to separate a direct effect of one variable from indirect effects of other variables. In this way we can see more clearly the actual contribution of individual variables to the forming of the final traits.

The weight of 1,000 seeds exhibited the highest direct negative effect on seed oil content and the highest positive direct effect on seed yield per plant (DE=-0.565[**] and DE=0.789[**], respectively) which explains the very high correlations between these traits (CC=-0.786[**] and CC=0.790[**], respectively; Figures 1 and 2). The direct effect of the weight of 1000 seeds on seed oil content was decreased by the negative indirect effect of the former trait on head diameter (IE=-0.202). A very high positive direct effect of the weight of 1,000 seeds on seed yield per plant brought about a high direct effect of the weight of 1,000 seeds on seed yield per plant, while indirect effects of the weight of 1,000 seeds on seed yield per plant via the other traits were not significant. Many researchers such as Ozer *et al.* (2003) and Khan *et al.* (2005) also reported of having found a positive direct effect of the weight of 1,000 seeds on seed yield.

The highest positive direct effect on seed yield and oil content per plant was exhibited by the total seed number per head (DE=0.485[**] and DE=0.204[**], respectively Figures 1 and 2). The simple correlation coefficient between total seed number per head and seed oil content was slightly lower than the direct effect. Their relationship was masked by the negative indirect effect of total seed number per head via head diameter (IE=-0.206) and by its positive indirect effect via the weight of 1,000 seeds (IE=0.093). The effect of the simple correlation coefficient

between total seed number per head and seed yield per plant was very high (0.369[**]). The lower simple correlation coefficient than the direct effect resulted from the negative indirect effect of total seed number per head on seed yield per plant via the weight of 1,000 seeds (IE=-0.129). Škorić (1974), Marinković (1987), Patil *et al.* (1996), Lal *et al.* (1997), El-Hosary *et al.* (1999), Taklewold *et al.* (2000) and Dušanić *et al.* (2004) also reported positive direct effects of total seed number per head on seed yield. However, Merrien *et al.* (1982) reported a negative direct effect of total seed number per head on sunflower seed yield.
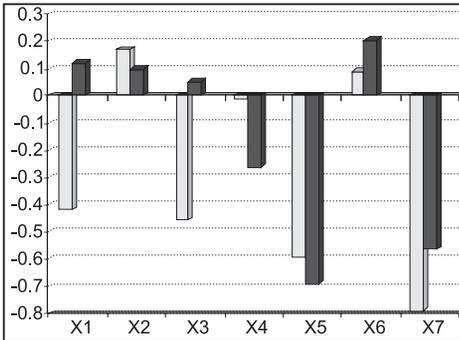

*Figure 1:* Simple correlation coefficients and direct effects of the seven yield components of seed oil content.
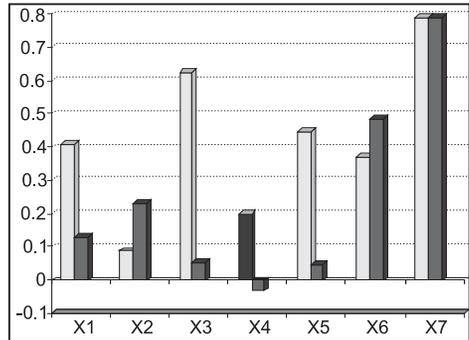
Figure 2: Simple correlation coefficients and direct effects of the seven yield components of seed yield.

X1	Stem diameter (STD)	X5	Head diameter (HD)
X2	Total leaf number per plant (TLN)	X6	Seed number per head (TSN)
X3	Total leaf area per plant (TLA)	X7	Weight of 1000 seed (M1000S)
X4	Plant height (PH)		

Head diameter had a very high negative direct effect on seed oil content (DE=-0.692[**]), with a very high negative simple correlation (CC=-0.588[**]), (Figures 1, 2). The simple correlation coefficient between head diameter and seed oil content was lowered by the negative indirect effect of head diameter via the weight of 1000 seeds (IE=-0.164) and a positive indirect effect of head diameter via plant height (IE=0.142). Although head diameter exhibited a very low positive direct effect on seed yield per plant (DE=0.047), the analysis of the morphological and physiological components of seed yield showed that the direct effect of head diameter on seed yield per plant was masked by the positive indirect effect of head diameter via the weight of 1000 seeds (IE=0.230), total seed number per head (IE=0.144) and the negative indirect effect of head diameter via total leaf number per plant (IE=-0.104). The resulting simple correlation coefficient between head diameter and seed yield per plant was very high (CC=0.446[**]). These results are in agreement with those of Green (1980), Sarno *et al.* (1992), Nirmala *et al.* (2000) and Fick *et al.* (1974) who found a negative direct effect of head diameter on seed yield per plant.

Plant height exhibited a high negative direct effect on seed oil content and a low negative direct effect on seed yield per plant (DE=-0.260[*] and DE=-0.031, respec-
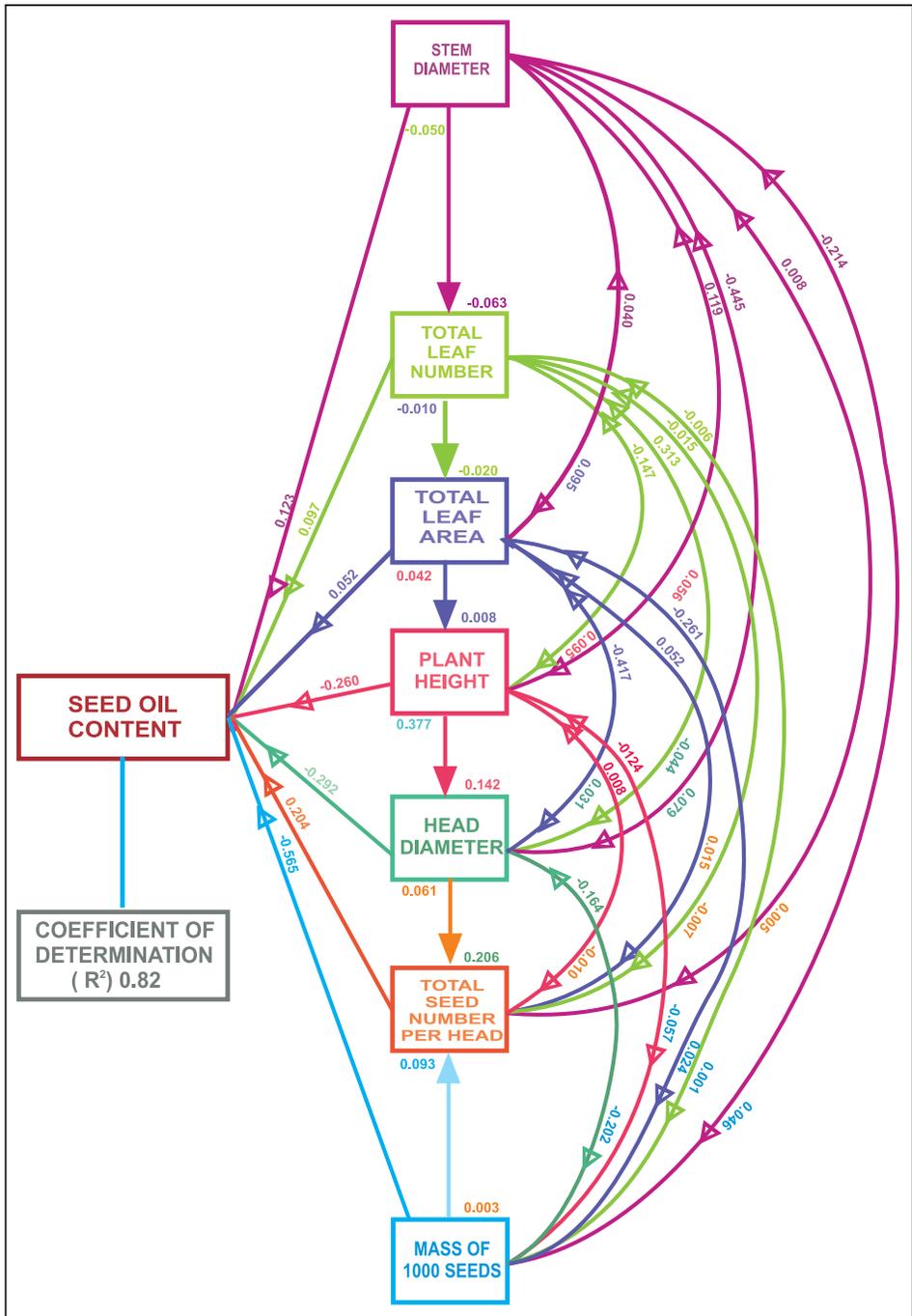
Figure 3: Graphic presentation of the path diagram for seed oil content in sunflower (F₁)

tively). Although the direct effect of plant height on seed oil content was high and negative, their simple correlation coefficient was very low and negative (CC=-0.009). The value of the simple correlation coefficient between plant height and seed oil content would have been higher were it not lowered by the positive indirect effect of plant height via head diameter (IE=0.377[**]) and by the negative indirect effect of plant height via the mass of 1,000 seeds (IE=-0.164). The simple correlation coefficient between plant height and seed yield per plant was low but positive (CC=0.200). The analysis of partial correlations coefficients showed that the direct effect of plant height on seed yield per plant was low and negative, *i.e.*, the effect of plant height on seed yield per plant was indirect, via the weight of 1,000 seeds (IE=0.174) and total leaf number per plant (IE=0.131).

The path analysis showed that total leaf number per plant exhibited a very low positive direct effect on seed oil content (DE=0.097) and a very high positive direct effect on seed yield per plant (DE=0.231[**]). The correlation between total leaf number per plant and seed oil content was low and positive (CC=0.168), same as the direct effect. Total leaf number per plant exhibited a positive indirect effect on seed oil content via head diameter (IE=0.313) and negative indirect effects via plant height (IE=-0.147) and stem diameter (IE=-0.063). The very high positive direct effect of total leaf number per plant on seed yield per plant was masked by its negative indirect effects via most of the studied traits, which explains the very low correlation between total leaf number per plant and seed yield per plant (CC=0.087).

The analysis of path coefficients indicated that stem diameter had a low positive direct effect on seed oil content (DE=0.123) and seed yield per plant (DE=0.129) (Figures 3 and 4). The very high simple correlation coefficient between stem diameter and seed oil content (CC=0.419**) resulted from high negative indirect effects of stem diameter on seed oil content via head diameter (IE=-0.445) and the weight of 1,000 seeds (IE=-0.214).

The simple correlation coefficient between stem diameter and seed yield per plant, which was very high and positive (CC=0.411[**]), resulted from a low positive direct effect of stem diameter on seed yield per plant which was masked by a positive indirect effect of stem diameter on seed yield per plant via the weight of 1,000 seeds (IE=0.299) and its negative indirect effect via total leaf number per plant (IE=-0.119) (Table 2).

Total leaf area per plant had very low direct effects on seed oil content and seed yield per plant (DE=0.052 and DE=0.050, respectively) (Figures 3 and 4). There was a very high negative correlation between total leaf area per plant and seed oil content (CC=-0.457[**]), which resulted from negative indirect effects of total leaf area per plant on seed oil content via head diameter (IE=0.417), weight of 1,000 seeds (IE=-0.161) and total leaf number per plant (IE=-0.020). There was a very high positive correlation between total leaf area per plant and seed yield per plant (CC=0.623**), which resulted from a positive indirect effects of total leaf area per plant on seed yield per plant via the weight of 1000 seeds (IE=0.364), total seed
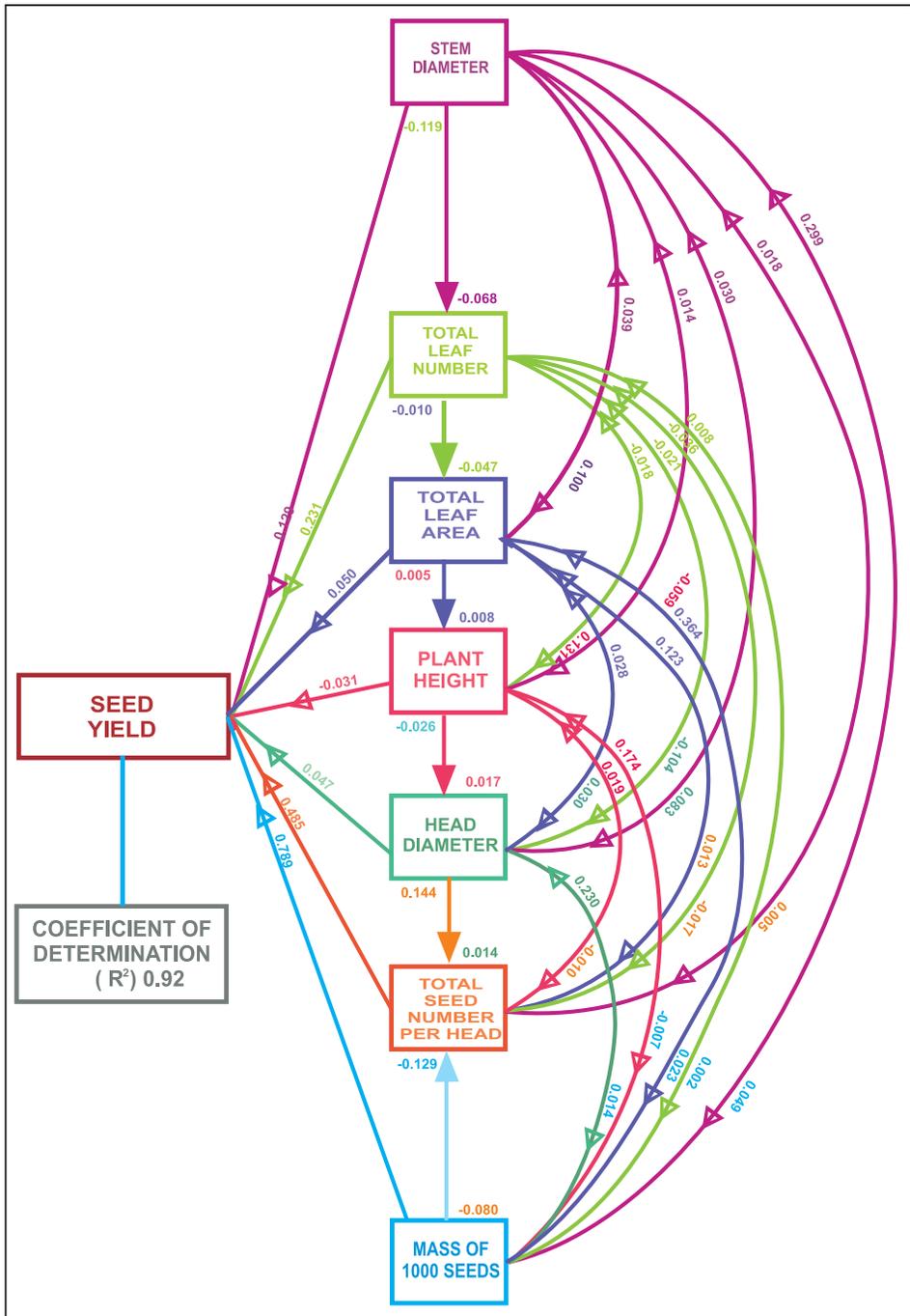
Figure 4: Graphic presentation of the path diagram for seed yield in sunflower (F₁)

number per head (IE=0.123) and stem diameter (IE=0.100). High positive direct effects of total leaf area per plant on seed yield per plant were observed by Razi *et al.* (1999) and Nirmala *et al.* (2000).

Table 2: Path coefficient analysis of seed yield and oil content in sunflower.

Component		SOC			SY		
		DE (P)	IE (Pxr)	CC (r)	DE (P)	IE (Pxr)	CC (r)
Stem diameter (STD)	X1	**0.123**			**0.129**		
Indirect effect TLN			-0.050			-0.119	
Indirect effect TLA			0.040			0.039	
Indirect effect PH			0.119			0.014	
Indirect effect HD			-0.445			0.030	
Indirect effect TSN			0.008			0.018	
Indirect effect M1000S			-0.214			0.299	
Total				-0.419**			0.411**
Total leaf number (TLN)	X2	**0.097**			**0.231****		
Stem diameter STD			-0.063			-0.066	
Indirect effect TLA			-0.010			-0.010	
Indirect effect PH			-0.147			-0.018	
Indirect effect HD			0.313			-0.021	
Indirect effect TSN			-0.015			-0.036	
Indirect effect M1000S			-0.006			0.008	
Total				0.168			0.087
Total leaf area (TLA)	X3	**0.052**			**0.050**		
Indirect effect STD			0.095			0.100	
Indirect effect TLN			-0.020			-0.047	
Indirect effect PH			0.042			0.005	
Indirect effect HD			-0.417			0.028	
Indirect effect TSN			0.052			0.123	
Indirect effect M1000S			-0.261			0.364	
Total				-0.457**			0.623**
Plant height (PH)	X4	**-0.260***			**-0.031**		
Indirect effect STD			0.056			-0.059	
Indirect effect TLN			0.055			0.131	
Indirect effect TLA			-0.008			-0.008	
Indirect effect HD			0.377			-0.026	
Indirect effect TSN			0.008			0.019	
Indirect effect M1000S			-0.124			0.174	
Total				-0.009			0.200
Head diameter (HD)	X5	**-0.692****			**0.047**		
Indirect effect STD			0.079			0.083	
Indirect effect TLN			-0.044			-0.104	
Indirect effect TLA			0.031			0.030	
Indirect effect PH			0.142			0.017	

Table 2: Path coefficient analysis of seed yield and oil content in sunflower.

		Direct	Indirect	Total	Direct	Indirect	Total
Indirect effect TSN			0.061			0.144	
Indirect effect M1000S			-0.164			0.230	
Total				-0.588**			0.446**
Total seed number per head (TSN)	X6	**0.204****			**0.485****		
Indirect effect STD			0.005			0.005	
Indirect effect TLN			-0.007			-0.017	
Indirect effect TLA			0.013			0.013	
Indirect effect PH			-0.010			-0.001	
Indirect effect HD			-0.206			0.014	
Indirect effect M1000S			0.093			-0.129	
Total				0.091			0.369**
Weight of 1000 seed (M1000S)	X7	**-0.565****			**0.789****		
Indirect effect STD			0.046			0.049	
Indirect effect TLN			0.001			0.002	
Indirect effect TLA			0.024			0.023	
Indirect effect PH			-0.057			-0.007	
Indirect effect HD			-0.202			0.014	
Indirect effect TSN			-0.033			-0.080	
Total				-0.786**			0.790**
Coefficient of determination, R^2		**0.815**			**0.922**		

X1: Stem diameter (STD)   X5: Head diameter (HD)
X2: Total leaf number per plant (TLN)   X6: Seed number per head (TSN)
X3: Total leaf area per plant (TLA)   X7: Weight of 1000 seed (M1000S)
X4: Plant height (PH)

It is important for sunflower breeding for increased productivity to find morphological and physiological traits that are easy to score and at the same time demonstrate causal relationships with seed oil content and seed yield per plant and therefore could be used as selection criteria. The focus should be placed on traits that have a very high positive direct effect on seed yield and oil content.

The path analysis indicated that total seed number per head had high direct effects on seed oil content and seed yield per plant (Figures 1 and 2). Very high positive direct effects on seed yield per plant were exhibited by the weight of 1,000 seeds, total seed number per head and total leaf number per plant. Marinković (1992), Goksoy and Turan (2007) and Behradfar *et al.* (2009) reported positive direct effects of the weight of 1,000 seeds and total seed number per head on seed yield of sunflower. In our study, highest negative direct effect on seed yield per plant were exhibited by head diameter and the weight of 1,000 seeds (Figures 1 and 2).

Having examined the direct and indirect effects of the group of morphological and physiological traits on seed yield and oil content, we concluded that the direct effects of the stem diameter, total leaf number per plant, total leaf area per plant, plant height, total seed number per head and the weight of 1000 seeds on seed oil content were masked by the high indirect effect of head diameter. The direct effects

of the stem diameter, total leaf area per plant, plant height, head diameter and total seed number per head on seed yield per plant was masked by the high indirect effect of the weight of 1000 seeds (Table 2).

The coefficients of determination (R^2) for seed yield and oil content were 0.922 and 0.815, respectively, which indicated that the studied traits affected 92% and 82%, respectively, of the total variability for seed yield and oil content (Figures 3 and 4).

Differences in the results obtained in this and other studies may be due to different plant materials used in these studies.

The research results discussed in this paper indicate that, among the studied traits, total number of seeds per head had the largest impact on seed yield and oil content. It can be used in improvement of seed yield and oil content as well as for assessment of sunflower breeding materials.

CONCLUSIONS

Determination of relationships between seed yield and oil content on the one hand and morphological and physiological traits of the sunflower plant on the other facilitates the utilization of genetic resources in sunflower breeding for a high genetic potential for seed yield.

The analysis of simple correlation coefficients showed that the weight of 1,000 seeds and stem diameter had a high negative correlation with seed oil content and a high positive correlation with seed yield per plant. Further, the total leaf number per plant and total seed number per head had a positive but low correlation with seed oil content and a very high positive correlation with seed yield per plant. Head diameter had a very high negative correlation with seed oil content and a very high positive correlation with seed yield per plant. Plant height had a low negative correlation with seed oil content and a low positive correlation with seed yield per plant.

The applied path coefficient analysis gave a somewhat different picture. It partitioned the direct and indirect effects of the morphological and physiological yield components on seed yield and oil content, which allowed us to detect the components that exhibited the highest direct effects on seed oil content and seed yield expression. Total seed number per head was found to be a good indicator for seed yield and oil content. The data obtained in this investigation, as well as various literature data, indicate that the weight of 1,000 seeds, total seed number per head and total leaf number per plant are the main seed yield components which are applicable as selection criteria in sunflower breeding.

The coefficients of determination (R^2) of 0.922 and 0.815 indicated that the traits involved in this study affected the total variability of seed yield and oil content by 92% and 82%, respectively.

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