POLLEN VIABILITY AND SEED SET PERCENTAGE OF NS HYBRIDS AND THEIR PARENTAL LINES

Jovanka D. Atlagic, Nenad Z. Dušanic, Jovan P. Joksimovic, and **Vladimir A. Miklic,** Institute of Field and Vegetable Crops, M. Gorkog 30, 21000 Novi Sad, Serbia and Montenegro
E-mail: atlagic@ifvcns.ns.ac.yu
E-mail: dusanicn@ifvcns.ns.ac.yu
E-mail: jovanj@ifvcns.ns.ac.yu
E-mail: miklicv@ifvcns.ns.ac.yu

Abstract

This research determined the pollen viability, percentage of seed set and seed yield of four commercial sunflower hybrids (HS-H-45, HS-H-111, NS-H-702 and VELJA) and their parental components (four male-fertile lines and three restorers). The results showed that there were significant differences in the mean values for the examined characteristics of most genotypes. The analysis of variance and the calculated component of phenotypical variance showed that the variability of pollen viability was greatly affected by genotype and genotype x years interaction, whereas % seed set and seed yield were greatly affected by all sources of variation. The greatest proportion in phenotypical variance for % seed set was genotype (56.68%), then years (22.72%) and their interactions (14.20%), while the greatest proportion in the phenotypical variance of the seed yield per head was genotype (71.00%) and the years (19.56%). The correlational analysis showed highly significant correlations between percentage of seed set and seed yield, whereas pollen viability had negative but nonsignificant influence on percentage of seed set, that is, positive but nonsignificant influence on seed yield per head.

Introduction

Sunflower (*Helianthus annuus* L.) is an open-pollinated species which tolerates self-pollination. Thanks to the fact that there is a certain percentage of autofertility, it is possible to create inbred lines; that is, hybrids. Many authors have explored the sunflower autocompatibility mechanism. Pollen self–compatibility is the main biological reaction of sunflower plants against self–fertilization, this species being allogamous and entomophilous. Foreign pollen germinates on stigma much faster than its own pollen. Seed set is the culmination of the vegetative phase and is affected by gametogenesis, pollination, fertilization, and embryogenesis. All of these processes are influenced by genetic and environmental factors and flower morphology and structure. Seed set under self–pollination conditions is strongly correlated with a degree of self–compatibility. This phenomenon is common in sunflower, and in some populations is nearly complete (Fick, 1978). While creating inbred lines it is necessary to choose those with a high degree of autocompatibility. Furthermore, it is important to be aware that seed yield largely depends on the degree of autocompatibility as well as on the presence of pollinators and the weather conditions at the time of flowering. It is essential to take care of the given conditions when producing parental lines and hybrids under the conditions of spacial isolation. In light of interesting results from the previous research on attractiveness factors for pollinators under different growing conditions (the amount of fertilizers and microclimates) (Miklič et al., 2003; Joksimović et al., 2003; Atlagić et al., 2003) conducted on commercially important hybrids and their parental components, the goal of this work was to examine pollen viability, seed set (%) and seed yield per head in open pollination conditions on the same material and in equal experiments.

Materials and Methods

The study materials consisted of four widely used commercial sunflower hybrids (HS-H-45, HS-H-111, NS-H-702 and VELJA) and their parental components (four male-fertile inbreds and three restorers) which were grown in a stationary trial established at Rimski Šančevi in 1966, which studies the effect of different fertilization treatments in maize, wheat, sunflower and sugar beet. Over a two-year period during 2000 and 2001, the sunflower hybrids and their parental lines were analyzed under one fertilization treatment (50:50:50) for pollen viability, % seed set and seed yield. Pollen viability was determined by the method of Alexander (Alexander, 1969). This method is based on differential staining of viable and nonviable pollen grains. Pollen from non-dehiscent anthers was taken from three plant heads and from three segments per head per genotype and was suspended in a drop of the stain. Viable and nonviable pollen grains were counted on three slides, at ten locations per slides. Pollen viability was expressed in percentage of total pollen grains.

Percentage of seed set was determined by counting full and empty achenes per head in five plants by repetition for every genotype and was calculated as a ratio between total number of disk flowers formed and full achenes. Seed yield was determined by measuring full seed mass per head by repetiton (three repetitions) for every genotype. Percentage of seed set and seed yield was determined for the conditions of open pollination (heads were not under isolators). Results obtained were statistically processed using the MSTAT program, and analysis of variance, LSD and Duncan's test were performed.

Results and Discussion

Mean values for pollen viability ranged from 67.96% in inbred lines HA-98B to 99.00% in hybrids NS-H-702. Most examined genotypes differed significantly in this characteristic. On the basis of Duncan's test, genotypes were distributed into four groups with from one to four genotypes in a group for significance degree 0.05 (Table 1).

Hybrid NS-H-45 had the highest mean value for seed set (88.32%), and the lowest was in restorer lines RHA-113N and RHA-PL-2/1 (52.88%). At the level of significance there were significant differences between examined genotypes (8 groups with 1, 2 or 4 genotypes in a group). The mean values for seed yield ranged from 23.14g/head (RHA-113N) to 75.49g/head in hybrid VELJA. All examined genotypes differed in this characteristic (Table 1).

Table 1. Mean values and variability for pollen viability, percentage of seed set and seed yield of F1 hybrids and their parental lines.

Genotype	Pollen viability (%)		Seed set (%)		Seed yield (g/plant)	
	X±sx	Signif.	X±sx	Signif.	X±sx	Signif.
NS-H-111	97.01±1.44	AB	68.67±1.72	D	61.86±3.71	C
NS-H-45	98.84±0.15	A	88.32±1.51	A	69.15±5.29	B
VELJA	98.49±0.11	A	77.28±1.82	B	75.49±6.39	A
NS-H-702	99.00±0.07	A	69.39±1.51	D	71.62±4.18	AB
CMS-3-8 B	95.04±1.32	BC	53.07±0.71	E	29.79±1.54	G
HA-26 B	95.21±0.54	BC	70.33±5.84	CD	41.76±4.92	DE
HA-74 B	93.81±1.80	C	74.32±0.94	BC	36.44±3.46	EF
HA-98 B	67.96±2.32	D	72.81±1.62	CD	45.85±2.70	D
RHA-583	98.06±0.28	A	54.88±5.98	E	35.90±3.97	EFG
RHA-113 N	94.93±1.90	BC	52.88±4.35	E	23.14±3.01	H
RHA-PL-2/1	97.18±0.41	AB	52.88±1.86	E	31.90±2.80	FG
LSD 0.05	**2.791**		**4.207**		**6.114**	
LSD 0.01	**3.731**		**5.624**		**8.174**	

The results obtained results show that pollen viability was high in most examined genotypes. The line HA-98B stands out with its lower viability percentage. Most authors have determined a high percentage of pollen viability in cultivated sunflower (Novi Sad line and hybrids) (Atlagić, 1990). The reduced pollen viability was determined in interspecific hybrids obtained by crossing wild species and cultivated sunflower (Georgieva-Todorova, 1990; Atlagić, 1999; Jan, 1997).

Given the data from literature that percentage of cross-pollination in some genotypes had the range 0-100%, the stated values in this research paper for the seed set percentage were at the satisfactory level (>50%). Fick (1997) states that breeders have made hybrids and inbred lines with percentage of self-fertilization of 80-100. Correlation between hybrids and their parental lines for seed set cannot be established.

In the case of seed yield per head, apart from determined variablility for examined genotypes (they all differed significantly in this characteristic), it can be observed that it was higher in hybrids (61.86-75.49%) when compared to parental lines (23.14-45.85%). Interestingly enough, the line HA-98B has the lowest percentage of pollen viability, but very high percentage of seed set; that is, the highest seed yield of all examined lines. Virupakshappa et al. (1992) determined higher autofertility in hybrids than in lines.

The analysis of variance and phenotypical variance component indicated that genotype and genotype x year interaction greatly affected the characteristic of pollen viability. The greatest proportion of phenotypical variance was in genotype (85.95%) (Table 2).

Table 2. ANOVA and components of phenotypic variance for pollen viability.

Source of variation	Degrees of freedom	Mean square		Components of variance	
				σ2	%
Genotype	10	471.233 **	M1	74.374	85.95
Year	1	0.768	M2	0.000	0.00
Genotype x Year	10	24.988 **	M3	6.417	7.42
Error	42	5.736	M4	5.736	6.63
Total	65			86.527	100.00

In the seed set percentage all sources of variation had significant value: genotype, years and their interaction. The greatest proportion in the phenotypical variance was genotype (56.68%), then interaction of genotype x year (22.72%) and then years (14.20%) (Table 3).

Table 3. ANOVA and components of phenotypic variance for percentage of seed set.

Source of variation	Degrees of freedom	Mean square		Components of variance	
				σ2	%
Genotype	10	844.107 **	M1	115.391	56.68
Year	1	1105.527 **	M2	28.902	14.20
Genotype x Year	10	151.759 **	M3	46.241	22.72
Error	42	3.036	M4	13.036	6.40
Total	65			203.570	100.00

The same influence was shown in the expression of seed yield per head (all sources of variation had highly significant values), and in the phenotypical variance the greatest proportion had genotype (71.00%), then years (19.56%), and only a negligible proportion had their interaction (3.61%) (Table 4).

Table 4. ANOVA and components of phenotypic variance for seed yield.

Source of variation	Degrees of freedom	Mean square		Components of variance	
				σ2	%
Genotype	10	2090.163 **	M1	335.255	71.00
Year	1	3126.410 **	M2	92.357	19.56
Genotype x Year	10	78.633 **	M3	17.032	3.61
Error	42	27.538	M4	27.538	5.83
Total	65			472.182	100.00

In defining the pollen viability characteristic, many authors considered this potential for fertilization. However, since sunflower is an open-pollinated plant, the autofertility characteristic is crucial in determnining percentage of seed set and seed yield.

Apart from this, most authors have determined besides genetic determination the important influence of enviromental factors on these examined characteristics, which was also shown by our research.

We have, as many authors before us, confirmed the significant influence of genotype on the characteristic variability. Phenotypical variability was most affected by genotype for all three characteristics, but the influence of year and interaction genotype/year on % seed set and seed yield was also significant.

Vranceanu et al. (1978) claimed that the greatest negative influence on autocompatibility and pollen quality was caused by high temperatures (35/24C) and short day length (period of dark-18h). Roath and Miller (1981) grew three sunflower hybrids and their parental components in five locations for two years and concluded that one of the hybrids and its parental line (maintainer-B analogue) constantly had lower peercnetage of seed set than the other examined genotypes. On the other hand, self-compatibility was not constant under different enviromental conditions.

The calculated correlations showed highly significant positive correlation between percentage of seed set and seed yield, while pollen viability had negative, but nonsignificant influence on percnetage of seed set, that is, positive but nonsignificant influence on seed yield per head (Table 5).

Table 5. Simple correlation coefficient of pollen viability, seed set and seed yield.

Characteristic		Seed set (%) X	Seed yield (g/plant) Y
Pollen viability (%)	X1	-0.0987	0.1781
Seed set (%)	X2		0.7558 **

Significance level (n=11-2) at 0.05 = 0.602; at 0.01 = 0.735

The relation between the examined characteristics, on the basis of calculated simple correlation coefficient, mostly confirms other results from the literature**.** Significant positive correlation between percentage of seed set and seed yield expressed in this way confirms the results of Fick (1983). Negative correlation between percentage of seed set and seed yield was not significant but it was in disagreement with the results of Channa et al. (1975) who claim that by impaired pollen viability (pollen growing old) percantage of seed set is reduced. Viable and fresh pollen yield 94.4% of seed set, while the pollen 84h old yields 2.22% of seed set. The staining method for pollen viability determination used in this paper provides relative evaluation which represents fertilization potential. Unarguably, the use of fluorescent miscoscopy (monitoring growth of pollen tubes) would provide more precise evaluation of pollen viability and at the same time a more objective evaluation of pollen viability's influence on percentage of seed set and seed yield. This was confirmed by Xanthopoulas (1991) who showed the existence of highly significant positive correlation between percemtage of seed set and percentage of penetrated styles.

The results obtained in this research show that NS hybrids and the their parental lines can have good potential (genetic basis) for biologically important characteristics and that given favourable enviromental conditions can yield rich seed yield and oil, which is an ultimate goal in sunflower breeding.

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