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## Optimization the selection of lint percentage and fiber length in cotton based on the combining effects of parental forms

Neli Valkova, Valentina Dimitrova, Minka Koleva\*

Field Crops Institute – Chirpan, 2 G. Dimitrov Blvd., 6200 Chirpan, Agricultural Academy - Sofia, Bulgaria

\*E-mail: [m\\_koleva2006@abv.bg](mailto:m_koleva2006@abv.bg)

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**Abstract:** Forty  $F_1$  crosses obtained from  $5 \times 8$  line by tester mating design methodology were studied. The varieties Chirpan-539, Sirius, Darmi (Bulgarian selection), Nazili 954 (Turkish) and FR-H-1002 (Spanish) were used as female parents, selection lines 449, 489, 639, 535, 550, 572, 578 and 678 were used as male parents. The aim of the study was to identify the best  $F_1$  crosses for improving lint percentage and fiber length in new cotton varieties, by applying a new approach to select parents with the highest possible general combining ability (GCA) for both traits. Crosses and their parents were sown in one row, randomized in three replications. The highest values of the GCA effects of the two parental forms for each trait were taken as “ideal” and the Euclidean distances between them and the GCA effects of the two parents for each  $F_1$  cross were calculated. Combining ability analysis of variance showed that females and males differed in general and specific combining abilities. The cultivars Chirpan-539 and Nazili-954 from the females, and lines 550 (Tiara variety) and 578, from the males, appeared to be very good general combiners for lint percentage. The cultivar FR-H-1002 and Darmi variety, from the females, and lines 449, 572, and 578, from the males, appeared to be very good general combiners for fiber length. Some crosses showed high positive SCA effects and heterosis for both traits. Cross combinations FR-H-1002  $\times$  578, Darmi  $\times$  578 and Nazili 954  $\times$  578 had the highest possible GCA for both studied traits and are most valuable for selection programs. A total of seven crosses with short Euclidean distances to the “ideal” population in which desirable segregates are expected to occur were selected.

**Keywords:** cotton; line $\times$ tester crosses; combining ability; “ideal” population; euclidean distances

## INTRODUCTION

A constant task of the cotton breeding in our country is to increase the yield and improve the fiber quality. As a result of intensive selection work, a number of new cotton varieties have been created in recent years and with them the seed cotton yield and quality of the fiber have been significantly improved (Dimitrova et al., 2022). The most recent achievements in cotton breeding are the varieties Tsvetelina (Koleva & Valkova, 2019), Pirin, Perun (Koleva & Valkova, 2023), Aida, Anabel, Tiara, Melani (Dimitrova, 2022a; 2022b; Dimitrova, 2023) and Selena (Dimitrova & Nedyalkova, 2024). Pirin, Perun and Anabel

varieties were obtained by intraspecific crossing and Aida, Tiara, Melani and Selena were created by interspecific hybridization and backcrossing. Tiara, Melani and Selena varieties have improved fiber quality, remaining ones are with improved productivity combined with other valuable qualities. In lint percentage, some of the new varieties are equal to the standard cultivar Chirpan-539 and those with higher fiber quality are significantly inferior to it. Increasing lint percentage is of great importance to increase lint yield  $ha^{-1}$ . Lint percentage and fiber length are in negative correlation and an increase in one of the two traits leads to reduction of the other.

In the selection of cotton in our country, the main selection methods are intraspecific and interspecific hybridization and experimental mutagenesis. Studying the combining ability of parental forms is essential to the success of hybridization programs. Zhang et al. (2016) concluded that the combination of yield and fiber length in  $F_1$  interspecific crosses of *G. hirsutum* L.  $\times$  *G. barbadense* L. depended on the genetic potential and combining ability of parents. Sivia et al. (2017), Nimbale et al. (2019) confirmed that the combining ability analysis is of great importance for the selection of potential parents to produce more desirable segregants. According to Sajjad et al. (2016) the general combining ability (GCA) of genotypes is of greater importance for synthetic selection (for hybridization and selection programs), while the specific combining ability (SCA) is of greater importance for heterosis selection (for hybrid crop exploitation).

Line  $\times$  Tester Mating Design is one of the most used methods in choosing potential parents and  $F_1$  cross combinations to give desirable segregates in the next segregating generations. According to Dabholcar (1992) line  $\times$  tester analysis can provide very useful information regarding females and males as parents for hybridization. Sivia et al. (2017) characterized this method as one of the simplest and most effective methods for evaluating the combining ability of parental forms.

In selection, the importance of parental forms with high general combining ability in one, two or more traits is usually emphasized. Barut et al. (2000) applied a general approach to select parents having general combining ability as high as possible for all traits. The GCA effects of both parents, for each trait and for each combination, are pooled and ranked, and the highest value for each trait is taken as "ideal". The "ideal" values for each trait are considered to characterize the "ideal" population and the Euclidean distances between the GCA effects of both parents and the "ideal" population values for each  $F_1$  cross are calculated. In our country, in the selection of cotton, this method to select parents with the highest possible general combining ability for all studied traits was applied by Stoilova (2004).

The aim of this study was to improve lint percentage and fiber length of new cotton varieties by applying the general approach to identify and select the parental forms having general combining ability as high as possible for both traits and  $F_1$  crosses with the highest breeding value.

## MATERIAL AND METHODS

The hybrid populations derived from  $5 \times 8$  line by tester crosses mating design methodology, by applying the experimental method I (without reciprocals) of Savchenko (1984), were studied. Three Bulgarian varieties Chirpan-539, Sirius and Darmi and two foreign ones Nazili-954 (Turkish) and FR-H-1002 (Spanish), used as female parents, were crossed with eight advanced lines 449, 489, 535 (Anabel variety), 550 (Tiara variety), 572, 578, 639 and 678, used as male parents, to produce 40 hybrid combinations. The trial was carried out in 2021 at the Field Crops Institute in Chirpan. Parents and hybrids were sown in one row 2.4 m long with a distance of 0.60 m between rows and 0.20 m within row, randomized in three replications. Traditional practices for cotton growing in our country were applied during the growing season. The traits under study were lint percentage and mean fiber length. For each genotype, ten plants from each replication were observed.

Methodology of Savchenko (1984) was applied to evaluate the general combining ability (GCA) and specific combining ability (SCA). The hybrid sum of squares was subdivided into variation due to females, males and the female  $\times$  male interaction. The main effects of females ( $f_i$ ) and males ( $m_j$ ) are equivalent to general combining ability (GCA), and the female  $\times$  male interaction represents specific combining ability (SCA) (Hallauer & Miranda, 1981).

Given that there is a strong negative correlation between the two studied traits some parents may have high GCA for fiber length, some others for lint percentage. To rank the parents for GCA for both traits ( $f_i + m_j$ ) totals were calculated for each trait. The highest ( $f_i + m_j$ ) values were taken

as “ideal” (Barut et al., 2000) and the Squared Euclidean distances (Manly, 1995) were calculated between those “ideals” and the total GCA effects of any two parents for each F<sub>1</sub> cross combination. The shorter the Euclidean distances to the “ideal” population, the more desirable the crosses are. Euclidean distances were determined by the formula:

$$\text{Squared Euclidean distance} = \sum(x_i - x_j)^2$$

Where:  $x_i$  – “ideal” value;  
 $x_j$  – total GCA effects of any two parents

Length and lint percentage were measured in different units and data were pre-standardized.

The year of study 2021 in terms of temperature sum in May-September (the growing season of cotton) was moderately warm to medium (P=41.90%) and in terms of rainfall in May-August was moderately dry (P=74.19%). P% is the coverage factor (coefficient of security) for the temperature sum in May-September and for the rainfall in May-August.

## RESULTS AND DISCUSSION

Results of the variance analysis of combining ability for lint percentage and fiber length are

presented in Table 1. It can be seen from the table that the effects of crosses, the GCA effects of females and males, and the SCA effects for both traits are significant. Therefore, parental forms females and males differed in combining ability for lint percentage and fiber length, and individual analysis can be performed.

The assessments of GCA effects are presented in Table. 2. The lint percentage varied from 37.0% to 39.2% in the females and from 34.7% to 37.5% in the males. Some of the females and males had the same lint percentage, but showed different combining ability for this trait. Of the females, the cultivar Chirpan-539 (Bulgarian selection) had the highest lint percentage and the highest GCA for this trait. The Turkish cultivar Nazili 954, which had lower lint percentage, had also significant positive GCA. Sirius and Darmi varieties, which in lint percentage were equal to the Turkish cultivar Nazili 954, had insignificant negative GCA. The Spanish cultivar FR-H-1002, which in lint percentage was close to the cultivar Chirpan-539, had also insignificant GCA. Of the males, line 550 (Tiara variety) had the highest and significant GCA for lint percentage. Line 578 with the lowest value for this trait had also positive and significant GCA. Line 449 which in lint percentage was equal to line 550 had significant negative GCA. Of the others, 3 lines showed positive and 2 lines were with negative insignificant GCA.

**Table 1.** Analysis of combining ability variance for lint percentage and fiber length in 5 × 8 F<sub>1</sub> line by tester crosses

Sources on variation	Degrees of freedom	Mean squares	
		Lint percentage	Fiber length
Crosses	39	4.534 <i>F<sub>exp.</sub></i> 22.557+++	2.482 <i>F<sub>exp.</sub></i> 11.333+++
Errors	78	0.201	0.219
GCA-females	4	4.311 <i>F<sub>exp.</sub></i> 63.397++	1.888 <i>F<sub>exp.</sub></i> 25.863++
GCA-males	7	0.386 <i>F<sub>exp.</sub></i> 5.676+	0.619 <i>F<sub>exp.</sub></i> 8.479++
SCA-females × males	28	1.392 <i>F<sub>exp.</sub></i> 20.471++	0.728 <i>F<sub>exp.</sub></i> 9.973++
Errors	78	0.068	0.073

**Table 2.** Assessments of general combining ability effects for lint percentage and fiber length of the parents

Traits	Lint percentage %		Fiber length mm	
	×	GCA	×	GCA
<b>Females</b>				
Chirpan-539	39.2	0.756	25.7	-0.612
Sirius	37.3	-1.136	25.9	-0.007
Darmi	37.0	-0.094	27.1	0.372
Nazili 954	37.6	0.531	26.1	-0.324
FR-H-1002	38.9	-0.057	26.1	0.571
Standard error		0.159		0.165
<b>Males</b>				
449	37.3	-0.570	25.5	0.305
489	36.9	0.010	26.8	-0.015
535 (Anabel)	37.4	0.017	25.2	-0.055
550 (Tiara)	37.5	0.357	25.6	-0.555
572	37.2	0.010	24.5	0.245
578	34.7	0.283	26.5	0.485
639	36.7	-0.050	26.6	-0.002
678	37.5	-0.057	26.5	-0.408
Standard error		0.201	0.596	0.209

Fiber length varied from 25.7 mm to 27.1 mm in the females and from 24.5 mm to 26.8 mm in the males. Some of the female and male parents with the same fiber length similar to lint percentage exhibited different combining abilities. Of the females, the Spanish cultivar FR-H-1001 had the highest and significant GCA for fiber length. Darmi variety, with the longest fiber, had also positive and significant GCA. The cultivar Chirpan-539, with the shortest fiber, had the highest negative GCA for this trait. The Turkish cultivar Nazili 954 also had negative and significant GCA. Sirius variety, which in fiber length approached the cultivar Chirpan-539, had negative but non-significant GCA. Of the males, line 578 had the highest GCA followed by line 449. Line 572, with the shortest fiber, was also with positive and significant GCA, while line 489, with the longest fiber, had insignificant negative GCA. Lines 550 (with comparatively short fiber) and 678 (with comparatively longer fiber) had significant negative GCA for fiber length, lines 535 (Anabel

variety) and 639, also differing in fiber length, had insignificant negative GCA.

The results of the assessments of combining ability of the female and male parents showed that the genotypes with the same lint percentage and the same fiber length could differ in combining ability for these two traits. Some authors (Thomson & Luckett, 2011; Zhang *et al.*, 2016) reported that the genotypes with the highest indicators had the highest GCA, which was not fully confirmed in this study. In our climatic conditions, usually drought occurs in the months of July and August, during the period of flowering and fruiting, and foreign cultivar, as well as some Bulgarian varieties with longer fiber, cannot realize their genetic potential. Drought has very adverse effect not only on seed cotton yield, but also causes strong reduction in fiber length.

Negative correlation between the two traits was also manifested in the combining ability of genotypes. The cultivars Chirpan-539 and Nazili 954 with positive GCA for lint percentage had negative GCA for fiber length. Darmi variety and

the cultivar FR-H-1002 with positive GCA for fiber length had negative, but insignificant GCA for lint percentage, which indicates that the negative correlation between the two traits was less pronounced. The same applies to male parents. Exceptions were line 578, with significant positive GCA for both traits, and line 572 with significant positive GCA for fiber length and insignificant positive GCA for lint percentage. These genotypes may prove very valuable for breeding programs to improve both traits simultaneously. Line 505 with significant positive GCA for lint percentage had significant negative GCA for fiber length and line 449 with significant positive GCA

for fiber length had significant negative GCA for lint percentage.

The assessments of SCA effects are presented in Table 3. Seventeen crosses (42.5%) manifested positive SCA for lint percentage. Five crosses showed high lint percentage (39.3-40.3%) and high SCA effects. Other three also had high SCA effects, but they were with lower lint percentage. The crosses with high SCA effects showed mid patent heterosis (to the average of the two parents) from 103.7% to 108.0%, compared to the better parent (heterobeltiosis) from 101.0% to 107.2%. Karademir & Gencer (2010) reported heterosis of 5.52% in only one cross among studied dial-

**Table 3.** Mean values, effects of specific combining ability (below line) and variances ( $\sigma^2S_i$ ;  $\sigma^2S_j$ )

Females	Males								$\sigma^2S_i$
	449	489	535	550	572	578	639	678	
	Lint percentage, %								
Chirpan-539	37.5	38.1	38.5	39.8	39.6	37.8	38.7	38.5	
	-0.476	-0.489	-0.096	0.897	1.010	-1.029	0.204	-0.023	0.478
Sirius	35.3	37.7	35.8	36.2	37.2	37.4	36.0	37.6	
	-0.784	1.069	-0.904	-0.811	0.536	0.462	-0.571	1.002	0.716
Darmi	38.0	35.5	37.1	38.5	38.3	37.8	38.3	38.1	
	0.907	-2.239	-0.646	0.414	0.627	0.179	0.654	0.461	1.062
Nazili 954	39.3	40.3	40.0	38.1	35.8	39.1	37.5	36.3	
	1.582	2.002	1.696	-0.577	-2.531	0.529	-0.771	-1.931	2.953
FR-H1002	35.9	37.4	37.7	38.2	38.1	38.2	38.2	38.2	
	-1.230	-0.343	-0.050	0.077	0.357	0.217	0.483	0.490	0.319
$\sigma^2S_j$	1.413	2.623	1.022	0.486	2.050	0.400	0.398	1.289	
SE(SCA)	0.369								
	Fiber length, mm								
Chirpan-539	26.9	25.4	25.9	25.6	25.5	26.9	25.5	24.1	
	0.878	-0.268	0.238	0.438	-0.495	0.664	-0.215	-1.241	0.473
Sirius	26.1	26.7	27.1	24.9	27.0	26.0	26.4	26.4	
	-0.559	0.427	0.834	-0.832	0.401	-0.772	0.047	0.454	0.399
Darmi	26.0	26.8	25.8	26.9	27.1	26.8	26.7	27.5	
	-1.005	0.114	-0.845	0.722	0.154	-0.352	0.002	1.208	0.544
Nazili 954	27.4	25.0	25.9	26.2	25.0	27.6	25.4	25.5	
	1.124	-0.989	-4.917	0.717	-1.249	1.144	-0.569	-0.129	0.844
FR-H1002	26.8	27.6	26.7	25.3	28.3	26.7	27.6	26.2	
	-0.438	0.715	-0.178	-1.045	1.188	-0.685	0.735	-0.292	0.610
$\sigma^2S_j$	0.879	0.430	0.367	0.745	0.842	0.727	0.220	0.819	
SE(SCA)	0.384								

lel crosses. The cultivar Chirpan-539 from the females and lines 550 and 578 from the males with high GCA for lint percentage had low variances of SCA. The low variances of SCA indicate that their high GCA was due to mainly of additive gene effects and these genotypes are very suitable for synthetic (pedigree) selection. The cultivar Nazili 954 from the females, with high GCA for this trait, had high variances of SCA. The high variances of SCA show that its high GCA was due to additive and non-additive (dominance and epistasis) gene actions, which makes it more suitable for heterosis selection (to exploit heterosis).

Positive and significant SCA for fiber length was found for 15 (37.5%) crosses. In these crosses fiber length varied from 26.2 mm to 28.3 mm. Heterosis to the average of the two parents (mid parent heterosis) was from 101.1% to 111.9%, compared to the better parent it was from 100.4% to 108.4%. Çoban et al. (2015), Çoban & Ünay (2017) in *G. hirsutum* × *G. barbadense* crosses reported mid parent heterosis for fiber length in all cross combinations between 2.05% to 16.99%. Positive heterosis for fiber length in this hybridization was reported by Roy et al. (2018). According to Ekinçi & Basbag (2018) among the studied *G. hirsutum* × *G. barbadense* crosses the determined heterosis (Ht) values for fiber length varied from -1.77 to 3.81%, the estimated heterobeltiosis (Hb) values for this trait varied from -7.87% to -0.03%. Heterobeltiosis for this trait in this hybridization was also reported by Ünay et al. (2018). Chapara & Madugula (2021) reported heterosis for Upper Half Mean Length (UHML) and mean length in intraspecific line × tester crosses and the highest values were 21.34% and 25.37%, heterobeltiosis for UHML ranged between 7.53 to 20.73%.

Crosses Nazili 954 × 489 and Nazili 954 × 535 (Anabel variety) exhibited the highest heterosis for lint percentage, crosses FR-H-1002 × 572 and FR-H-1002 × 639 manifested the highest heterosis for fiber length (data for heterosis are not given). Only one cross Nazili 954 × 449 demonstrated high and positive SCA for both traits, with heterobeltiosis for lint percentage 104.5% and for fiber length 105.0%. This cross combination may

prove very valuable for breeding programs to combine high lint percentage and longer fiber in one genotype. Darmi variety and line 578 of the females, with high GCA for fiber length, had low variances of SCA and are suitable for breeding programs, the cultivar FR-H-1002 and lines 449 and 572, with high GCA for this trait and high SCA variances are more suitable for heterosis selection.

Totals GCA effects ( $f_i + m_j$ ) of the two parents for each trait (fiber length and lint percentage) and ranked crosses based on the Euclidean Distance to the “ideal” population are presented in Table 4. It can be seen from the table that, based on the calculated Euclidean distances, the closest to the “ideal” population and therefore the most desirable of the crosses were FR-H-1002 × 578, Darmi × 578 and Nazili 954 × 578. The parental forms of these crosses demonstrated high and positive totals GCA effects for both traits.

The cultivar Nazili 954 had significant positive GCA for lint percentage, while the cultivar FR-H-1002 had significant positive GCA for fiber length. These two cultivars had high variances of SCA, which means that their high GCA was determined by additive and non-additive gene actions and selection for the relevant traits, in the hybrid combinations with them, should be conducted in the later hybrid generations. Darmi variety had high GCA for fiber length and low variances of SCA. Line 578, used as male parent, demonstrated very good combining ability with all three cultivars used as females. This line had high GCA for both traits and low variances of SCA. Therefore, in the cross combination Darmi × 578, efficient selection for both traits can be conducted in the early hybrid generations.

Another four crosses with relatively short Euclidean distances were also reliable. In these ones, the parental forms had high positive combining effects for fiber length and very weak negative combining effects for lint percentage. The crosses Chirpan-539 × 550, Chirpan-539 × 578, Chirpan-539 × 535, Chirpan-539 × 572, Chirpan-539 × 639 and Chirpan-539 × 489 manifested high positive totals combining effects of parental forms only for lint percentage, while for

**Table 4.** Totals general combining ability effects ( $f_i + m_j$ ) of the two parents for each trait and Squared Euclidean distances to the “ideal” values

Parents	Totals general combining effects of the parents ( $f_i + m_j$ )		Squared Euclidean distance
	Lint percentage	Fiber length	
FR-H-1002 × 578	0.226	1.056	1.539
Darmi × 578	0.189	0.857	1.800
Nazili 954 × 578	0.814	0.161	2.804
FR-H-1002 × 572	-0.047	0.816	2.821
Darmi × 572	-0.084	0.617	3.435
FR-H-1002 × 489	-0.047	0.556	3.452
FR-H-1002 × 535	-0.040	0.516	3.557
FR-H-1002 × 639	-0.107	0.569	3.689
Darmi × 489	-0.084	0.357	4.406
Darmi × 535	-0.077	0.317	4.562
Chirpan-539 × 578	1.039	-0.127	4.604
Darmi × 639	-0.144	0.370	4.635
FR-H-1002 × 550	0.300	0.016	4.843
Nazili 954 × 572	0.541	-0.079	4.868
FR-H-1002 × 678	-0.114	0.163	5.562
FR-H-1002 × 449	-0.627	0.876	6.027
Nazili 954 × 449	-0.039	-0.019	6.388
Darmi × 550	0.263	-0.183	6.452
Darmi × 449	-0.664	0.677	6.647
Chirpan-539 × 572	0.766	-0.367	6.882
Nazili 954 × 489	0.541	-0.339	7.027
Darmi × 678	-0.151	-0.036	7.039
Nazili 954 × 639	0.481	-0.326	7.050
Nazili 954 × 535	0.548	-0.379	7.383
Chirpan-539 × 449	0.186	-0.307	7.778
Sirius × 578	-0.853	0.478	8.656
Chirpan-539 × 639	0.706	-0.614	9.478
Chirpan-539 × 489	0.766	-0.627	9.532
Chirpan-539 × 535	0.773	-0.667	9.970
Nazili 954 × 678	0.474	-0.732	11.292
Sirius × 572	-1.126	0.238	12.000
Nazili 954 × 550	0.888	-0.879	12.389
Sirius × 489	-1.126	-0.022	13.618
Sirius × 535	-1.119	-0.062	13.845
Sirius × 639	-1.186	-0.009	14.059
Chirpan-539 × 678	0.699	-1.02	14.481
Sirius × 550	-0.779	-0.562	15.593
Chirpan-539 × 550	1.113	-1.167	16.220
Sirius × 449	-1.706	0.298	17.427
Sirius × 678	-1.193	-0.415	17.502

fiber length they were negative. In these crosses, an increase in lint percentage and reduction in fiber length could be expected more pronounced in the first cross with the highest negative combining effects (for fiber length) and weaker in the second cross. The cultivar Chirpan-539 used as female parent and line 578 as male parent had high GCA for lint percentage and low variances of SCA. Line 639 with insignificant negative GCA effects and line 535 (Anabel variety) with weak positive GCA effects, also had low variances of SCA. In these cross combinations selection for lint percentage can be made in the earlier hybrid generations, while line 489 with weakly expressed positive GCA effects had high variances of SCA. In the crosses FR-H-1002  $\times$  449, Darmi  $\times$  449 and FR-H-1002  $\times$  639 showed high positive combining effects of parental forms for fiber length and negative for lint percentage, improvement of fiber length and reduction of lint percentage could be expected. Darmi variety with high GCA for fiber length had low variances of SCA, but the cultivar FR-H-1002 and line 449, with high GCA, had high variances of SCA and selection for this trait should be conducted in later hybrid generations.

## CONCLUSIONS

Genotypes used as female and male parents differed in general and specific combining ability.

The cultivars Chirpan-539 and Nazili-954 from the females and lines 550 and 578 from the males appeared to be very good general combiners for lint percentage. The cultivar Chirpan-539 and both lines had low SCA variances and are very valuable for breeding programs, while the cultivar Nazili-954 had high SCA variances and is more suitable for heterosis selection.

The cultivars FR-H-1002 and Darmi variety from the females and lines 449, 572 and 578 from the males appeared to be very good general combiners for fiber length. Darmi variety and line 578 had low SCA variances and are very suitable for synthetic selection, while the cultivars FR-H-1002 and lines 572 and 578 had high SCA vari-

ances and are more suitable for heterosis selection.

Some crosses showed high positive SCA effects and heterosis for lint percentage and for fiber length. Heterosis, compared to the better parent, for lint percentage ranged from 101.0% to 107.2%, for fiber length ranged from 100.4% to 108.4%.

The cross combination Nazili 954  $\times$  449 exhibited high SCA and heterosis for both traits, heterobeltiosis for lint percentage was 104.5% and for fiber length was 106.2%.

Cross combinations FR-H-1002  $\times$  578, Darmi  $\times$  578 and Nazili 954  $\times$  578 showed the highest possible GCA for both traits. The parental forms of these crosses had positive and high totals general combining effects for both traits.

A total of seven crosses with short Euclidean distances to the “ideal” population in which desirable segregates are expected to occur were selected.

The approach for selection of parental forms having general combining ability as high as possible for all studied traits gives very valuable information about their breeding value and helps to identify the most promising crosses, as well as to optimize the existing negative correlation dependence between them.

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