

Original Research Article

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Selection of Aphid Tolerant Parents and Crosses Using Combining Ability Analysis for Development of High Yielding Varieties and Hybrids in Safflower (*Carthamus tinctorious* L.)

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ABSTRACT

Combining ability analysis, using Line x Tester mating design was conducted in safflower to select best material for further breeding programme and generation advancement on the basis of general and specific combining ability of the divergent parents and their crosses, respectively. Two CMS lines as female parents, 20 male parents and 40 crosses along with three checks (PKV PINK, AKS 207 and PBNS 12) were evaluated in Randomized Complete Block Design for seed yield under aphid infestation, Aphid Infestation Index (AII), total phenol and chlorophyll content. Study revealed the predominance of non-additive gene action for all the traits under study. Out of 20 males, AKS 325 and AKS 322 were identified as good general combiners for aphid infestation index and phenol content, further, CCC-B2 for aphid infestation index; AKS/S-33 and GMU 6881 for chlorophyll content index; whereas, GMU 3924 and GMU 3876 for seed yield under aphid infestation. Hence, these parents may be exploited in further breeding programme to improve the concerned traits. On the basis of SCA effects, four crosses *viz.*, AKS CMS 2A X GMU 3876, AKS CMS 3A X GMU 3876, AKS CMS 2A X GMU 3924 and AKS CMS 3A X GMU 3924 were found promising for seed yield per plant under aphid infestation and other traits related with aphid tolerance. These crosses may be used for exploitation of heterosis at commercial level. The crosses *viz.*, AKS CMS 2A X GMU 3965, AKS CMS 2A X GMU 3325 and AKS CMS 2A X AKS 325 having highly significant GCA effects of one of the parents and non-significant SCA effects for almost all the traits. Hence, these crosses can be exploited for development of varieties through generation advancement by crossing 'B' lines of female parent (CMS line) again with same male parents and advancing the generations to identify good transgressive segregants in later generations. Further, biparental mating or diallele mating is suggested for varietal development which may be more fruitful instead of regular breeding methods like pedigree, bulk methods.

Keywords

Safflower,
Combining ability,
Aphid tolerance,
A.I.I., Total
phenols,
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yield

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Introduction

Safflower is an important oilseed crop. Its increased popularity in recent years across the county in recent past is the consequence of its

adaptability under drought conditions (Sarode *et al.*, 2008). Safflower is known for its cultivation since time immemorial for orange red dye extracted from its florets and for its much valued oil. Safflower seeds contain 27.5

per cent oil, 15 per cent protein, 41 per cent crude fiber and 2.3 per cent ash. Safflower oil, which on average contains 75 per cent linoleic acid, also contains tocopherols, known to have antioxidant effect and high vitamin E content (Latha and Prakash, 1984). For this reason, safflower oil is used in the diets of patients with cardiovascular disease, and bears great importance for its anti-cholesterol effect (Jhahhariya *et al.*, 2013).

Most of the released varieties like CO-1 are highly susceptible to the attack by aphids [*Uroleucon compositeae* (Theobald)] which recorded 86 aphids per 5 cm twig causing 40-50 per cent seed yield losses; reduction in oil content of damaged seed up to 32 per cent and seed weight by 50.6 per cent (Mane *et al.*, 2012). In addition to the huge direct economic losses as stated above, there are deleterious effects of pesticides which use to remain in the environment. Therefore, development of a cost effective and an environment friendly approach is necessary through improvement of cultivars resistant to aphids. Though, a large number of high yielding varieties and few hybrids have been released, in the country, however, to meet the future requirements, it is utmost necessary to breed resistant / tolerant varieties and hybrids having agronomically superior traits suiting to the need of farmers and its end users as well. Pest resistant cultivar or hybrid is the most suitable non-cash input for marginal farmers and development of such cultivars or hybrids requires identification of the pest resistant/tolerant sources.

Combining ability studies furnish useful information about selection of suitable parents for effective hybridization programme (Sprague and Tatum, 1942). It is a powerful tool to discriminate good as well as poor combiners to choose appropriate parental material in breeding programmes. It also provides information about the nature of gene action involved in the inheritance of various

characters. The biometrical technique Line x Tester (L x T) analysis appeared to be most useful tool for screening lines rapidly and with reasonable confidence. It also provides information about the general combining ability (GCA) and specific combining ability (SCA) variances and effects.

It is essential to evaluate newly developed parents in cross combinations to test their combining ability for various traits associated with aphid tolerance and seed yield. Therefore, the present investigation was undertaken to study the combining ability for identification of good combiners and promising crosses for future improvement in aphid tolerance and seed yield and in safflower.

Materials and Methods

Plant material

Genetically diverse parents on the basis of their distinguishing characters including tolerance and susceptibility to aphid infestation i.e. two CMS lines *viz.*, AKS CMS 2A and AKS CMS 3A (Cytoplasmic male sterile lines developed by Oilseed Research Unit, Dr. P.D.K.V., Akola) as females and 20 males *viz.*, S-518, CW-99, CCC-B2, GILA, AKS/NS-1, AKS-322, AKS-325, AKS/S-33, GMU-3923, GMU-3965, GMU 3924, GMU 3876, GMU 3863, GMU 3963, GMU 3325, GMU 6877, GMU 6881, GMU 6835, AKS 08R and AKS 10R were deliberately selected.

The crosses were affected in L x T scheme to obtain F₁ seeds of 40 crosses at the experimental field of Oilseeds Research Unit, Dr, P.D.K.V., Akola. Utilising CMS based system, only hand pollination using pollens from protected flowers from male parents was performed with the protected flowers of CMS based females in the morning hours. The parental seeds were multiplied by selfing.

Field trial

A field trial of 65 genotypes, consisting 20 parents, 40 F₁s and three checks viz., PKV PINK, AKS 207 and PBNS 12 was raised in Randomized Block Design with two replications during *rabi* 2017-18 at the field of Oilseeds Research Unit, Dr. PDKV, Akola. Each genotype was planted in a single row of 2 m length with 45 cm spacing between and 20 cm within rows. All the cultivation practices were followed as per recommendations for safflower cultivation. Further, infester rows of susceptible variety viz., "CO-1" were sown one month before sowing of screening block.

Sowing of screening block was executed in such a way that crop should attain age of 35-45 days at peak incidence of aphids. Infester plants were uprooted and speeded across the screening block uniformly when the main crop had attained age of 35-40 days. Necessary plant protection measures were undertaken to avoid wilt and *Alternaria blight*.

Observations

The observations were recorded on randomly selected five competitive plants per plot per replication in parents, F₁s and checks for following traits.

Aphid Infestation Index (A.I.I.) – The observations of foliage drying grade due to aphids were recorded (1-5 scale) when 'CO-1' was completely killed (Anonymous, 2016).

The A.I.I. was estimated by using following formula

$$\text{A.I.I.} = \frac{(1 \times a) + (2 \times b) + (3 \times c) + (4 \times d) + (5 \times e)}{a + b + c + d + e}$$

Where,

1 to 5 = Different foliage drying grades.

a to e = Actual number of plants falling in each of the 5 corresponding foliage drying grades.

Total phenols - Total phenol content in safflower leaves was estimated as per the procedure described by Bray and Thorpe (1954).

Chlorophyll content index - It was determined by using chlorophyll meter, model SPAD-250 plus. For recording the observations; three leaves were randomly collected from observational plants and used for estimation of chlorophyll. Then the mean values of chlorophyll index of three leaves were calculated.

Seed yield per plant (under aphid infestation) in gram was recorded

Statistical analysis

The analysis of variance for all the studied traits was performed as per the procedure given by Panse and Sukthame (1967) from the data obtained in field trial and laboratory. Further, the line x tester analysis was performed as per the standard procedure given by Kempthorne (1957).

After ensuring the significance of general combining ability (GCA) and specific combining ability (SCA) effects and their variances, the GCA and SCA variances were calculated for A.I.I., total phenols, chlorophyll content and seed yield.

Results and Discussion

Analysis of variance for combining ability (Table 1) revealed from that the estimates of mean squares due to crosses were significant for all the studied traits and mean squares due

to parents i.e. males; exhibited highly significant differences amongst themselves for all the studied traits indicating presence of substantial genetic variability among the crosses and males for all the traits. Whereas, the mean squares for females and male v/s female crosses were significant for chlorophyll content indicating the presence of substantial genetic variability for this trait. Similar results were also reported by Nai *et al.*, (2014) in safflower.

The SCA effects were found higher than GCA effects indicating predominant role of non-additive gene action for controlling all the traits under study thereby signifying the preponderance of non-additive variance in the expression of these characters.

Jadhav and Gawande (2015) also reported role of non-additive gene effects for seed yield per plant, larval count at vegetative and pod formation stages and percentage of pod damage in chickpea. Hence, there is good scope for development of hybrids for commercial cultivation in safflower from the parental material studied.

None of the parents has recorded significant GCA effects in desirable direction simultaneously for all the characters related to aphid tolerance studied (Table 2). For aphid infestation index and phenol content, the parents AKS 325 and AKS 322 were identified as good general combiners in addition to CCC-B2 for aphid infestation index on the basis of significant GCA effects of the parents for these traits.

The parents AKS/S-33 and GMU 6881 also exhibited significant GCA effects for the trait chlorophyll content index which proved that these parents are the good general combiners for chlorophyll content index. Hence, these identified parents can be exploited as donor parents for the respective trait associated with

aphid tolerance in further breeding programme to improve aphid tolerance in safflower. Further, the parents, GMU 3924 and GMU 3876 were found to be good general combiners for seed yield under aphid infestation, however, none of these two parents showed good GCA effect for any one of the traits associated with aphid tolerance. It may be due to sustaining ability of these genotypes to the attack and infestation by the aphids.

As far as the seed yield under aphid infestation is concerned, these two parents, GMU 3924 and GMU 3876 were recognized as the good parental material among the genotypes studied and same can be exploited as a parent in further breeding programme. Among the female parents, AKS CMS 3B was found to be superior for seed yield per plant along with A.I.I., chlorophyll content index. Whereas, AKS CMS 2B was found superior in desirable direction for phenol content over best check for particular traits.

Since GCA effects are the manifestation of additive properties of genes, parents selected based on GCA effects will be useful for developing breeding lines with high grain yield (Narayanamma *et al.*, 2013) and desirable levels of the trait of interest. While studying genetics of traits associated with pod borer resistance in chickpea; Jadhav and Gawande (2015) noticed that the parent ICC-506 was good general combiner for most of the traits associated with pod borer resistance and ICCV-2 for percent malic acid content.

They considered these two parents as good material for utilizing in further breeding programs as donors for the concerned traits. Similar results were also been reported by Sreelatha *et al.*, (2008) and Narayanamma *et al.*, (2013) in chickpea for pod borer damage and Singh and Singh (2009) in pigeonpea for pod fly resistance.

Table.1 Analysis of variance for combining ability for traits associated with aphid tolerance in safflower

| Sources of variation | Mean sum of squares | | | | |
|-----------------------|---------------------|---------|----------------|---------------------|----------------------|
| | d. f. | A.I.I. | Phenol content | Chlorophyll content | Seed yield per plant |
| Replications | 1 | 0.288 | 0.014 | 9.322 | 1.161 |
| Crosses | 39 | 0.677** | 0.107** | 14.469** | 5.142* |
| Female effects | 1 | 0.008 | 0.072 | 14.860* | 0.677 |
| Male effects | 19 | 1.343** | 0.195** | 23.084** | 9.673** |
| Female x Male effects | 19 | 0.047 | 0.020 | 5.833* | 0.846 |
| Error | 39 | 0.051 | 0.019 | 3.196 | 3.481 |
| GCA variance | | 0.000 | 0.001 | 0.186 | 0.008 |
| SCA variance | | 0.226 | 0.097 | 27.709 | 4.019 |

(* , ** Significant at 0.05 and 0.01 probability level, respectively).

Table.2 Estimates of general combining ability effects of parents for traits associated with aphid tolerance

| S.N. | Genotypes | Aphid infestation index (A.I.I.) | Phenol Content | Chlorophyll content index | Seed yield per plant |
|------|-------------------|----------------------------------|----------------|---------------------------|----------------------|
| | Lines (Males) | | | | |
| 1 | S-518 | -0.378** | 0.310** | -1.391 | 0.625 |
| 2 | CW-99 | -0.503** | 0.125 | 1.834 | 1.032 |
| 3 | CCC-B2 | -0.553** | 0.036 | 1.559 | 1.155 |
| 4 | GILA | 1.498** | -0.427** | -5.891** | -3.768** |
| 5 | AKS/NS-1 | 0.248* | -0.187* | -1.691 | -0.903 |
| 6 | AKS-322 | -0.603** | 0.256** | 2.659* | 1.005 |
| 7 | AKS-325 | -0.653** | 0.272** | 1.509 | 1.462 |
| 8 | AKS/S-33 | -0.528** | 0.175* | 4.509** | 0.377 |
| 9 | GMU-3923 | 0.273* | -0.154 | -1.344 | -1.300 |
| 10 | GMU-3965 | 0.298* | -0.131 | -0.916 | -1.758 |
| 11 | GMU-3924 | -0.603** | 0.222** | 1.659 | 2.302* |
| 12 | GMU-3876 | -0.353** | 0.189* | 1.184 | 2.537** |
| 13 | GMU-3863 | 0.448** | -0.178* | -1.366 | -0.430 |
| 14 | GMU-3963 | -0.428** | 0.258** | 1.009 | 1.085 |
| 15 | GMU-3325 | 0.323** | -0.198* | -0.316 | -1.253 |
| 16 | GMU-6877 | 0.448** | -0.241** | 0.534 | -0.373 |
| 17 | GMU-6881 | -0.428** | 0.162* | 3.109** | 1.327 |
| 18 | GMU-6835 | 0.448** | -0.165* | -2.491* | -1.355 |
| 19 | AKS-08R | 0.673** | -0.167* | -1.841 | -1.118 |
| 20 | AKS-10R | 0.373** | -0.157* | -2.314* | -0.650 |
| | SE(gi) | 0.035 | 0.0243 | 0.325 | 0.293 |
| | CD (0.05) | 0.071 | 0.492 | 0.657 | 0.593 |
| | CD (0.01) | 0.095 | 0.0658 | 0.880 | 0.794 |
| | Testers (Females) | | | | |
| 1 | AKS CMS 2B | -0.010 | 0.030 | 0.431 | -0.092 |
| 2 | AKS CMS 3B | 0.010 | -0.030 | -0.431 | 0.092 |
| | SE(gi) | 0.111 | 0.0769 | 1.027 | 0.928 |
| | CD (0.05) | 0.224 | 0.155 | 2.079 | 1.877 |
| | CD (0.01) | 0.300 | 0.208 | 2.783 | 2.513 |

(* , ** Significant at 0.05 and 0.01 probability level, respectively)

Table.3 Estimates of specific combining ability effects of crosses for traits associated with aphid tolerance

| Sr. No. | Genotypes | Aphid infestation index (A.I.I.) | Phenol Content | Chlorophyll content | Seed yield per plant |
|---------|------------------------|----------------------------------|----------------|---------------------|----------------------|
| 1 | AKS CMS 2A X S-518 | 0.110 | 0.027 | -3.506* | -0.588 |
| 2 | AKS CMS 2A X CW-99 | 0.135 | -0.105 | -1.231 | -0.506 |
| 4 | AKS CMS 2A X CCC-B2 | 0.085 | 0.211 | -1.406 | -0.588 |
| 4 | AKS CMS 2A X GILA | 0.135* | -0.034* | -0.006 | -0.106* |
| 5 | AKS CMS 2A X AKS/NS1 | 0.085 | -0.019 | -0.256 | -0.356 |
| 6 | AKS CMS 2A X AKS-322 | -0.115 | -0.060 | 1.794 | -0.033 |
| 7 | AKS CMS 2A X AKS-325 | -0.015 | 0.002 | 0.544 | 0.009 |
| 8 | AKS CMS 2A X AKS/NS-33 | -0.190 | -0.029 | 0.244 | -0.726 |
| 9 | AKS CMS 2A X GGMU-3923 | -0.140 | -0.039 | 0.042 | 0.032 |
| 10 | AKS CMS 2A X GGMU-3965 | 0.235 | 0.015 | 0.869 | 0.139 |
| 11 | AKS CMS 2A X GGMU-3924 | -0.015 | 0.046 | -0.356 | -0.125* |
| 12 | AKS CMS 2A X GGMU-3876 | -0.115** | 0.080** | -0.081 | 1.085** |
| 13 | AKS CMS 2A X GGMU-3863 | 0.085 | -0.052 | 1.469 | 0.567 |
| 14 | AKS CMS 2A X GGMU-3963 | -0.090 | 0.089 | -0.856 | 0.082 |
| 15 | AKS CMS 2A X GGMU-3325 | -0.040 | -0.043 | 0.419 | 0.410 |
| 16 | AKS CMS 2A X GGMU-6877 | -0.065 | 0.030 | -0.031 | -0.001 |
| 17 | AKS CMS 2A X GGMU-6881 | -0.040 | 0.004 | 0.894 | 0.310 |
| 18 | AKS CMS 2A X GGMU-6835 | -0.015 | 0.015 | -0.356 | -0.223 |
| 19 | AKS CMS 2A X AKS-08R | 0.010 | -0.046 | 1.844 | 0.735 |
| 20 | AKS CMS 2A X AKS-10R | -0.040 | -0.094 | -0.033 | -0.118 |
| 21 | AKS CMS 3A X S-518 | -0.110 | -0.027 | 3.506* | 0.588 |
| 22 | AKS CMS 3A X CW-99 | -0.135 | 0.105 | 1.231 | 0.506 |
| 23 | AKS CMS 3A X CCC-B2 | -0.085 | -0.211* | 1.406 | 0.588 |
| 24 | AKS CMS 3A X GILA | -0.135* | 0.034 | 0.006 | 0.106* |
| 25 | AKS CMS 3A X AKS/NS1 | -0.085 | 0.019 | 0.256 | 0.356 |
| 26 | AKS CMS 3A X AKS-322 | 0.115 | 0.060 | -1.794 | 0.033 |
| 27 | AKS CMS 3A X AKS-325 | 0.015 | -0.002 | -0.544 | -0.009 |
| 28 | AKS CMS 3A X AKS/S33 | 0.190 | 0.029 | -0.244 | 0.726 |
| 29 | AKS CMS 3A X GGMU-3923 | 0.140 | 0.039 | -0.042 | -0.032 |
| 30 | AKS CMS 3A X GGMU-3965 | -0.235 | -0.015 | -0.869 | -0.139 |
| 31 | AKS CMS 3A X GGMU-3924 | 0.015 | -0.046 | 0.356 | 0.125* |
| 32 | AKS CMS 3A X GGMU-3876 | 0.115** | -0.080* | 0.081 | -1.085** |
| 33 | AKS CMS 3A X GGMU-3863 | -0.085 | 0.052 | -1.469 | -0.567 |
| 34 | AKS CMS 3A X GGMU-3963 | 0.090 | -0.089 | 0.856 | -0.082 |
| 35 | AKS CMS 3A X GGMU-3325 | 0.040 | 0.043 | -0.419 | -0.410 |
| 36 | AKS CMS 3A X GGMU-6877 | 0.065 | -0.030 | 0.031 | 0.001 |
| 37 | AKS CMS 3A X GGMU-6881 | 0.040 | -0.004 | -0.894 | -0.310 |
| 38 | AKS CMS 3A X GGMU-6835 | 0.015 | -0.015 | 0.356 | 0.223 |
| 39 | AKS CMS 3A X AKS-8R | -0.010 | 0.046 | -1.844 | -0.735 |
| 40 | AKS CMS 3A X AKS-10R | 0.040 | 0.094 | 0.033 | 0.118 |
| | SE (Sij) ± | 0.157 | 0.108 | 1.453 | 1.312 |
| | CD (0.05) | 0.317 | 0.219 | 2.940 | 2.655 |
| | CD (0.01) | 0.425 | 0.294 | 3.936 | 3.554 |

(* , ** Significant at 0.05 and 0.01 probability level, respectively)

Out of forty, two crosses *viz.*, AKS CMS 2A XGMU-3876 and AKS CMS 3A X GILA found to be superior combinations for aphid infestation index. Further, AKS CMS 2A X GILA, AKS CMS 2A X GMU-3876, AKS CMS 3A X GILA and AKS CMS 3A XGMU-3876 were found to be the good specific combinations for phenol content. For chlorophyll content, only one cross *i.e.* AKS CMS 3A X S-518 found significantly superior amongst all the cross combinations. For seed yield per plant under aphid infestation, the crosses *viz.*, AKS CMS 2A X GMU-3924, AKS CMS 2A X GMU 3876, AKS CMS 3A X GMU 3924 and AKS CMS 3A X GMU 3876 exhibited significant SCA effects. Amongst these four crosses, two male parents were found to be common *viz.*, GMU 3924 and GMU 3876, which were also found to be good general combiners for the concerned trait. Hence, there is appreciable role of GCA effect of the parents in these four crosses to be significantly superior on the basis of their SCA effects for seed yield per plant under aphid infestation. Significant SCA effects for pod borer damage in chickpea was also reported by Narayanamma *et al.*, (2013) and Jadhav and Gawande (2015) (Table 3).

Four crosses *viz.*, AKS CMS 2A X GMU 3876, AKS CMS 3A X GMU 3876, AKS CMS 2A X GMU 3924 and AKS CMS 3A X GMU 3924 were identified as promising crosses on the basis of SCA effects of crosses and good GCA effects of parents for seed yield per plant under aphid infestation and other traits related with aphid tolerance. Further, in accordance with the results obtained, the SCA variances for almost all the traits under studies were found to be greater than GCA variance. Thus, these crosses may be evaluated in preliminary or multilocation hybrids trials for further exploitation at commercial level. The crosses *viz.*, AKS CMS 2A X GMU 3965, AKS CMS 2A X GMU 3325 and AKS CMS 2A X AKS 325

had highly significant GCA effects of one of the parents involved in the cross and low SCA effects (non-significant) for almost all the traits. It is therefore recommended to utilize these crosses further by generation advancement by crossing again by using 'B' lines of female parent (CMS line) with same male parents and advancing the generations to identify and select good transgressive segregants in later generations for development of varieties. The use of conventional breeding methods such as, pedigree, single seed descent and bulk method are associated with the weakness of causing rapid homozygosity and low genetic variability, especially in the presence of linkage blocks and inverse relationships among the desirable traits (Clegg *et al.*, 1972). Therefore, superior transgressive segregation may be obtained from this material either through biparental mating or diallel selective mating (Jensen, 1970). Further, Malhotra *et al.*, (1980) opined that diallel selective mating among the parents based on GCA may result in breaking up some undesirable linkages and in turn releases greater genetic variability.

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