Published June 14, 2016

RESEARCH

Combining Ability Estimation for Grain Yield of Maize Exotic Germplasm Using Testers from Three Heterotic Groups

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ABSTRACT

Combining ability estimates of lines to be used in breeding are useful for maize (*Zea mays* L.) breeders. The objectives of this study were to (i) evaluate combining ability of 25 improved exotic germplasm using different numbers of testers (1 to 3) from Suwan1, Reid, and non-Reid heterotic groups; (ii) study differences in combining ability estimates obtained with different number of testers from the same vs. different heterotic groups; and (iii) determine the appropriate segregating ('S') generation in which line selection should be done to obtain stable general combining ability (GCA) estimates of lines. The results showed that three testers (one from each of the three heterotic groups) were economically best for estimating GCA effects of lines, and if a tester from one of the three heterotic groups was missed, GCA estimates for some lines were biased when compared with GCA estimates with testers from all three heterotic groups. Second, the specific combining ability (SCA) effects of test-crosses were quite similar regardless of whether one or two testers were used from each of the three heterotic groups. Thus, to obtain reliable SCA estimates, at least one tester would need to be used from each of the heterotic groups. Third, the SCA effects were different among crosses between different lines and testers, and to obtain stable estimates of GCA effects for a line, S4 or S5 generation should be the earliest generation in which to begin selection. However, to select lines with diverse genetic backgrounds, S3 should be the key generation for selection.

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Abbreviations: CV, coefficient of variance; GCA, general combining ability; GY, grain yield per plant in gram; H, heterosis; SCA, specific combining ability; SSR, simple sequence repeat.

Genetic theory and breeding practices have established that the introduction of exotic germplasms and their utilization in local breeding programs is highly effective in broadening the maize genetic base (Crossa et al., 1987; Hallauer and Miranda, 1988; Goodman, 2005; Reif et al., 2010; Fan et al., 2015). Two of the key steps in utilizing exotic germplasm are: (i) understanding which heterotic group the introduced lines belong to, and (ii) obtaining combining ability information on the introduced germplasm (Hallauer and Miranda, 1988; Fan et al., 2008a, 2009, 2015).

Proper heterotic group classification greatly improves breeding efficiency (Hallauer and Miranda, 1988; Fan et al., 2014). Because of different heterotic group classification methods used, researchers' classification of maize germplasm into heterotic groups could differ. For example, some researchers classified germplasm widely used in China into four heterotic groups, i.e., Ludahonggu, Sipingtou, Reid, and Lancaster (Fan et al., 2003; Chen et al., 2005; Wu et al., 2007), whereas others clustered germplasm into five or six groups (Yuan et al., 2000; Li et al., 2002, 2003). Li et al. (2003) used simple sequence repeat (SSR) markers to classify 70 maize inbred lines into six heterotic groups. Yuan et al. (2000) used four

Published in Crop Sci. 56:2527–2535 (2016).
doi: 10.2135/cropsci2016.01.0032

types of molecular markers and classified 15 maize inbred lines into five heterotic groups. Li et al. (2002) used the line × tester design to classify 18 maize inbred lines into five heterotic groups.

Currently, the major exotic tropical and subtropical germplasms include Suwan1, Suwan2, ETO, Tuxpeño, and Cuban Flint among others (Fan et al., 2002; Li et al., 2011). Suwan1 population was selected at Kasetsart University (Thailand) from a mixed population developed from 36 elite tropical, subtropical, and temperate maize germplasm (Sriwatanapongse et al., 1993). Following its release, Suwan1 rapidly became the core germplasm in South Asia and East Asia. Cuban Flint is also called Cateto in Brazil, which combines well with Tuxpeño, but it is susceptible to viral diseases (Wang et al., 1998). Germplasms from CIMMYT include more than 200 populations, among them Pop33 which is a medium-maturity subtropical population improved by backcrossing from Pool 33 (Wang et al., 1998) and Pop45 which is a subtropical population that contains germplasm of BSSS (Iowa Stiff Stalk Synthetic), Tuxpeño, Cuban Flint, and Puerto Rico (Yong et al., 2013). Since 2000, many germplasms from CIMMYT have been introduced into China and intensively studied, which has helped broaden the Chinese maize germplasm base and improve grain yield (Chen et al., 2000a, 2000b; Fan et al., 2005, 2008a, 2008b, 2009).

Fan et al. (2002) studied combining ability between five exotic tropical and/or subtropical maize heterotic groups and four commonly used temperate maize heterotic groups in China and found that Suwan1 and POP32 (ETO) were close to Ludahonggu; Antigua and Tuxpeño were close to Reid; and POP28 was close to Sipingtou. Based on combining ability information, Fan et al. (2008a) designated Suwan1 as a new heterotic group, different from both Reid and Lancaster. Fan et al. (2008b) studied GCA and SCA using five local maize lines from Yunnan and five exotic tropical and subtropical lines from CIMMYT, and found that CIMMYT lines had much better GCA effects for ear diameter and ear row number than local elite lines did.

The line × tester design has been widely used for estimating GCA and SCA of grain yield (GY), and other genetic parameters (Hallauer and Miranda, 1988; Menkir et al., 2004; Barata and Carena, 2006; Wu et al., 2007; Fan et al., 2008a). Menkir et al. (2004) used the line × tester design and, based on SCA and GY, classified 23 of the 38 inbred lines into two heterotic groups. Wu et al. (2007) used North Carolina design II (NCII) to compute SCA for GY (SCA-GY) of a series of maize inbred lines and classified 27 maize lines into four heterotic groups that are commonly accepted in China (Fan et al., 2003).

Breeders are routinely faced with questions such as the following: How many testers should be used to obtain reliable combining ability estimates of tested lines? Should breeders select testers from the same or different heterotic groups for combining ability estimates? Little or no information is available on these aspects. Therefore, this study was undertaken with the following objectives: (i) evaluate combining ability of 25 improved exotic germplasms in crosses with different numbers of testers chosen from either one, or two, or all three heterotic groups (Suwan1, Reid, and non-Reid); (ii) to investigate if combining ability is affected by use of different numbers of testers from different heterotic groups; and (iii) to determine which segregating ('S') generation is best to start selection in for obtaining stable general combining ability estimates.

MATERIALS AND METHODS

Plant Materials and Experimental Design

Twenty-five improved and adapted tropical maize inbred lines of different populations, obtained from CIMMYT, were crossed to six testers in Yunnan, using two testers from each of the three heterotic groups, i.e., Suwan1, Reid, and non-Reid. The pedigree and ecological type (tropical or temperate) of the maize lines are listed in Table 1. Among the 25 lines, sister lines L3, L4, L5, L6, L7, and L8 were selected in S3, sister lines

Table 1. Pedigree, ecological type, and heterotic group of the 31 inbred lines evaluated.

Line	Pedigree	Ecological type	Heterotic group
L1	P147/P33	Tropical	N/A†
L2	CML329/CML20	Tropical	N/A
L3	CML226/(CATETO DC1276/7619)	Tropical	N/A
L4	CML226/(CATETO DC1276/7619)	Tropical	N/A
L5	CML226/(CATETO DC1276/7619)	Tropical	N/A
L6	CML226/(CATETO DC1276/7619)	Tropical	N/A
L7	CML226/(CATETO DC1276/7619)	Tropical	N/A
L8	CML226/(CATETO DC1276/7619)	Tropical	N/A
L9	CML226/CATETO//CML323/CATETO	Tropical	N/A
L10	P147-F2-152/P45	Tropical	N/A
L11	GLSIY01HGA	Tropical	N/A
L12	FS8BT/MD37	Tropical	N/A
L13	CLA155 = SA3-C4-FS(16/25)	Tropical	N/A
L14	CLA161 = SA4-C2-FS(21/26)	Tropical	N/A
L15	CL-RCY023 = (CL-02439/CML-286)	Tropical	N/A
L16	GLSIY01HGB	Tropical	N/A
L17	CML323/(CATETO DC1276/7619)	Tropical	N/A
L18	CML226/CATETO	Tropical	N/A
L19	P45-C6	Tropical	N/A
L20	P147-F2-114/P45	Tropical	N/A
L21	P147-F2-136/P45	Tropical	N/A
L22	CLA44 = SA3-C4-FS(16/25)	Tropical	N/A
L23	P147-F2-136/P45	Tropical	N/A
L24	P45-C8	Tropical	N/A
L25	DTPY-C9	Tropical	N/A
T1	YML107	Temperate	Reid
T2	YML582	Temperate	Reid
T3	TR2	Temperate	Non-Reid
T4	TRL60	Temperate	Non-Reid
T5	YML146	Tropical	Suwan1
T6	YML132	Tropical	Suwan1

† N/A = not available.

L13 and L22 were selected in S4, and sister lines L21 and L23 were selected in S5 generation, respectively, from the following introgressed segregating populations: Cuban Flint (Cateto), SA3, and P45, after crossing these populations with local elite maize lines. All 25 lines were subjected to selection for plant type, disease resistance, height, fewer growing days from planting to maturity, and other important agronomic traits. Using the line × tester mating design, 150 test-crosses were made in the summer of 2013. The seeds from the 150 test-crosses and a commercial check, Yunrui 999, were planted on 20 April 2015 at three locations: Kunming (25°02′ N, 120°45′ E; 1960 m above sea level), Wenshan (23°19′ N, 104°45′ E; 1540 m above sea level), and Dehong (24°26′ N, 98°35′ E; 913.8 m above sea level). A randomized complete-block design with three replications was used at each location. Each experimental unit (plot) consisted of a single, 4-meter-long row, with a plant-to-plant spacing of 0.25 m within a row. Row-to-row spacing was 0.7 m. Plant density was 55,300 plants ha^{-1}. Number of kernel rows per ear, number of kernels per row, and 1000-kernel weight and single-plant yield (GY) were recorded after drying the kernels to constant moisture of 130 g kg^{-1}.

Statistical Model and Data Analysis

An ANOVA was conducted using the general linear model:

$$Y_{ijkl} = \mu + a_l + b(a)_{kl} + v_{ij} + (av)_{ijl} + e_{ijkl}$$

$$v_{ij} = l_i + t_j + lt_{ij}$$

where Y_{ijkl} = observed value from each experimental unit; μ = population mean; a_l = location effect; $b(a)_{kl}$ = replication effect within each location; v_{ij} = F_1 hybrid effect = $l_i + t_j + lt_{ij}$ (where l_i = ith line effect; t_j = jth tester effect; lt_{ij} = interaction effect between ith line and jth tester); $(av)_{ijl}$ = interaction effect between ijth F_1 hybrid and lth location; and e_{ijkl} = residual effect.

Standard heterosis (H) was computed using the equation:

$$H = 100\% \times (F_1 - CK)/CK$$

where F_1 is GY of a test-cross and CK is GY of a check.

The three locations used in this experiment were treated as a random sample of all possible locations within Southwestern China, as each location represented a unique environment. Therefore, significances for test-crosses, lines, testers, and line × testers were tested against mean squares of the interaction between corresponding source entity (i.e., lines, testers, lines × testers) and locations (Table 2). Significance of locations was tested against replications within locations. Significances of replications within locations and lines × testers × locations was tested against the overall experimental term (Table 2). Combining ability analysis was conducted according to the model and method used by Fan et al. (2009). Data analyses were conducted by use of the SAS 9.1.3 software package (SAS Institute, 2005).

RESULTS AND DISCUSSION

Analyses of Variance for Grain Yield

Results of ANOVA for GY are given in Table 2. All main effects (locations, test-crosses, lines, testers) were highly significant ($P < 0.01$), as were all possible two-way interactions between the main effects. The three-way interaction, lines × testers × locations, was not significant.

Table 2. Analysis of variance for grain yield (g per plant) of test-crosses generated from 25 lines and six testers.

Source	df	Mean squares
Location	2	18882.95**
Rep(Location)	6	3707.09**
Test-crosses	149	3043.10**
Lines	24	4689.39**
Testers	5	34730.98**
Lines × Testers	120	1393.51**
Test-crosses × Location	298	1145.47**
Lines × Location	48	3180.69**
Testers × Location	10	1686.38**
Lines × Testers × Location	240	715.88
Error	894	592.84

** Significant at the 0.01 probability level.

The significant lines × locations and testers × locations interactions strongly suggested that GY of lines and testers depended on the location in which they were grown. Because test-crosses × locations interaction (298 df) is composed of three components, i.e., lines × locations (48 df), testers × locations (10 df), and lines × testers × locations (240 df), and because lines × testers × locations interaction was not significant, we could conclude that test-crosses × locations interaction was mainly significant because of significant lines × locations and testers × locations interactions. The nonsignificant lines × testers × locations interaction implied that lines × testers interaction pattern was consistent across locations and that identification of stable performing hybrids across locations was possible.

Genotype × environment interaction for GY, a complex phenomenon, has been widely studied in maize. In a multi-environment trial, Kang and Gorman (1989) evaluated 17 hybrids, which were affected more by differential fertility and/or cultural practices (environmental index) than by weather factors, and identified hybrids with stable or consistent performance across 12 environments. Using 12 hybrids and 7 inbreds, Tollenaar et al. (2004) studied physiological basis of heterosis of 19 quantitative traits, including GY. They found that genotype × environment interaction was significant for GY and 12 other traits. Fan et al. (2007) studied stability of GY of 13 maize hybrids at two locations in 2 yr and found that hybrids × environments interaction was significant for GY. In contrast, Adebayo and Menkir (2014) studied GY and some other traits under well-watered and drought stress conditions across 2 yr and their results showed that genotypes × years interaction was not significant under both well-watered and drought conditions. These studies suggested that genotype × environment interaction might or might not be significant for GY under different situations. The current study showed that the test-crosses × locations interaction for GY was statistically significant and this result was consistent with the results reported by Tollenaar et al. (2004)

and Fan et al. (2007). One interesting result obtained in this study was that the lines × testers × locations interaction was not significant, though the test-crosses × locations interation was significant. We know that the major component of variance from lines and testers is GCA variance and that from the lines × testers interaction is SCA variance (Fan et al., 2009). Our results showed that both GCA variances (i.e., lines and testers) and SCA variance (i.e., lines × testers) were significant ($P < 0.01$) (Table 2). The GCA × locations interaction (i.e., lines × locations and testers × locations) was significant. However, SCA × locations (i.e., lines × testers × locations) interaction was not significant. These results suggested that the GCA effects were location specific and that a single-location testing would be inadequate.

Test-Crosses of Different Testers from Three Heterotic Groups

One of the major purposes of conducting a line × tester experiment is to identify high-yielding test-crosses. Usually, the standard heterosis (H, expressed as % check) can be used to rank hybrids based on GY. The GY means for all test-crosses between 25 lines and 6 testers are given in Table 3. The results showed that GY means of these test-crosses varied significantly. We identified 28 test-crosses with H >10% (in boldface font). The distribution of the 28 test-crosses across the six testers revealed that the largest number (12) of high-heterosis test-crosses were related to TRL60 tester, followed by YML146 (eight test-crosses), and TR2 (seven test-crosses). The rest of the testers yielded one high-heterosis hybrid or test-cross (from YML107 tester) or no high-heterosis hybrids (from YML582 and YML132 testers). These results indicated that the test-crosses from the lines and testers from different heterotic groups produced different proportions of superior hybrids. Apparently, testers from non-Reid heterotic group and Suwan1 heterotic group yielded more test-crosses with high GY, i.e., H >10%, than testers from Reid heterotic group. Thus, to identify test-crosses that could potentially be used as commercial hybrids, use of testers from different heterotic groups would be necessary and the greater the number of testers used, the greater the chances of obtaining superior hybrids.

There were eight test-crosses from YML146 tester (a line from Suwan1 heterotic group) with 10% higher GY than that of the check. Line YML146 was identified by Fan et al. (2008a) from a study of 100 crosses between 25 temperate maize germplasms and four germplasms from CIMMYT. Several studies (Fan et al., 2009, 2014, 2015) have shown YML146 to be a good line that can be used directly in hybrid development. The current study further confirmed YML146 to be a very good germplasm, which should be exploited further in maize hybrid development, as pointed out by Fan et al. (2015).

Table 3. Grain yield (g plant^{-1}) of crosses (25 lines × 6 testers) and top 28 crosses (in boldface) with standard heterosis >10%.

Rank	Line	YML107	Line	YML582	Line	TR2	Line	TRL60	Line	YML146	Line	YML132
1	15	**179.30**	7	168.47	24	**204.08**	6	**193.91**	15	**183.73**	22	162.33
2	24	170.94	23	163.46	3	**181.63**	21	**188.94**	11	**183.30**	17	161.10
3	7	166.70	6	160.93	9	**176.48**	15	**188.41**	21	**181.79**	10	160.69
4	3	163.78	4	159.78	18	**176.21**	1	**187.07**	24	**178.98**	24	160.10
5	1	161.61	1	152.41	17	**175.29**	24	**182.34**	8	**177.81**	6	152.02
6	6	154.40	5	151.44	15	**174.34**	23	**182.30**	23	**175.64**	9	151.66
7	10	152.44	10	149.78	11	**173.65**	20	**180.23**	2	**174.52**	2	151.03
8	20	150.97	15	143.74	2	171.26	2	**177.71**	9	**174.27**	19	150.96
9	17	150.68	11	143.57	12	169.38	9	**176.22**	20	170.41	3	150.14
10	5	150.20	3	142.91	1	167.26	3	**175.82**	7	169.24	16	149.23
11	9	149.72	21	142.51	10	165.35	18	**174.87**	3	163.47	1	147.53
12	8	149.51	2	142.12	21	164.47	8	**174.29**	16	160.84	11	146.59
13	18	145.71	9	141.79	13	162.96	4	172.26	25	159.66	21	146.35
14	2	140.81	19	141.67	22	160.42	5	170.90	13	159.47	20	144.33
15	4	139.74	8	138.39	7	158.18	14	164.20	19	157.56	12	143.97
16	13	136.21	20	134.75	4	157.56	25	163.73	18	154.99	8	141.24
17	11	134.94	17	132.35	8	157.02	17	161.84	10	154.29	4	140.33
18	21	133.23	13	132.12	6	156.66	10	160.27	6	154.13	23	139.09
19	14	133.05	24	127.15	23	154.66	7	158.58	14	153.15	15	135.45
20	23	132.68	22	126.34	20	153.75	22	157.19	1	150.50	25	135.33
21	12	132.39	18	125.93	14	149.73	12	157.18	4	146.70	14	135.18
22	25	125.48	14	122.83	5	147.57	11	152.68	17	145.95	7	131.49
23	19	120.69	12	119.27	19	140.78	13	150.08	5	145.78	5	131.39
24	16	119.64	25	108.02	25	136.06	19	148.25	22	145.46	13	130.51
25	22	116.89	16	106.43	16	121.33	16	135.08	12	145.12	18	127.69
Mean		144.47		139.13		162.24		169.37		162.67		145.03

General Combining Ability Effects

Another purpose of a line × tester experiment is to identify superior lines that can be effectively used in hybrid and/or inbred development programs. Usually, GCA effects of individual lines are considered to be controlled by genes with additive effects and these effects can be passed on to the next generation (Hallauer and Miranda, 1988; Kang 1994). Thus, GCA effects are a major criterion for evaluating lines for their potential application in hybrid- and inbred line-development programs (Fan et al., 2008b, 2014; Yao et al., 2013). To investigate whether GCA effects were the same or different when different numbers of testers from the same or different maize heterotic groups were used, the six testers were grouped as follows (Table 4): Group1 included all six testers; Group2, three testers, one from each of the 3 heterotic groups; Group3, two testers from non-Reid heterotic group; and Group4, two testers from Reid heterotic group; and Group5, two testers from Suwan1 heterotic group. The GCA effects of the 25 lines with the five groups of testers are shown in Figure 1. The GCA effects of the 25 lines tested against all six testers of Group1 were used as standard GCA for each line. Upon comparing the GCA effects from other four groups with the standard GCA effects, we found that the absolute magnitudes of GCA effects differed. However, the relative magnitudes of the GCA effects for the 25 lines tested in all groups were quite similar, with only a few exceptions. To further confirm this finding, a correlation analysis was conducted. Correlation coefficients for GCA effects were as follows: Group1 vs. Group2, $r = 0.92$ ($P < 0.01$); Group1 vs. Group3, $r = 0.90$ ($P < 0.01$); Group1 vs. Group4, $r = 0.84$ ($P < 0.01$); and Group1 vs. Group5, $r = 0.56$ ($P < 0.01$). The results revealed that three testers (one from each of the three different heterotic groups) gave almost consistent GCA effects with all testers used and would be economically most effective for estimating GCA effects of lines. In addition, though significant correlation coefficients were detected between Group1 and Group3 ($r = 0.90$, $P < 0.01$), Group4 ($r = 0.84$, $P < 0.01$), and Group5 ($r = 0.56$, $P < 0.01$), the peaks and troughs of GCA effects for some lines were quite different (Fig. 1). This suggested that when testers did not represent the three heterotic groups, the estimates of GCA effects from some lines would not be as reliable as when all heterotic groups were represented. Thus, we could conclude that (i) in a maize hybrid- or inbred-line development-program, three testers, i.e., one tester from each maize heterotic group, should provide the best GCA estimates; (ii) for GCA estimates to be reliable and/or correct, all three heterotic groups must be represented.

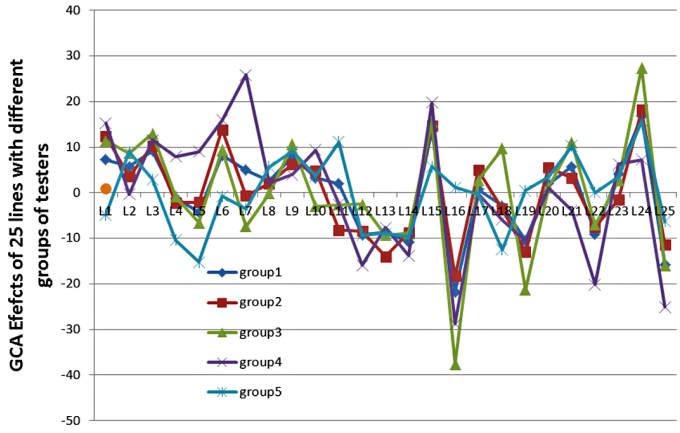

Fig. 1. General combining ability (GCA) effects of 25 exotic lines with five groups of testers.

Specific Combining Ability Effects

The SCA effects of individual test-crosses between 25 exotic lines and the 6 testers included in different groups are shown in Figure 2.

First, the SCA effects were almost identical in the test-crosses between the lines and YML107, TRL60, and YML132 testers used in both Group1 and Group2 (Fig. 2a, d and f). As Group1 contains all six testers (two testers per heterotic group) and Group2 includes three testers, with one tester from each of the three heterotic groups, these results suggested that SCA effects were quite similar regardless of whether one or two testers were used from each of the three maize heterotic groups. Thus, to obtain reliable estimates of SCA effects for test-crosses, at least one tester from each of the available heterotic groups should be included.

Second, the SCA effects for most of the crosses between the lines and a single tester were different when testers used were not from all available heterotic

Table 4. Six testers of three heterotic groups (Reid, Non-Reid, and Suwan1) were included in five groups. Group1 included all six testers; Group2 included three testers, with one from each of the three heterotic groups; Group3 included two testers, both from the non-Reid heterotic group; Group4 included two testers, both from the Reid heterotic group; and Group5 included two testers, both from the Suwan1 heterotic group.†

	Reid		Non-Reid		Suwan1	
Tester group	YML107	YML582	TR2	TRL60	YML146	YML132
Group1	+	+	+	+	+	+
Group2	+	–	–	+	–	+
Group3	–	–	+	+	–	–
Group4	+	+	–	–	–	–
Group5	–	–	–	–	+	+

† (+) indicates that the line is included in the group; (–) indicates that the line is not included in the group.

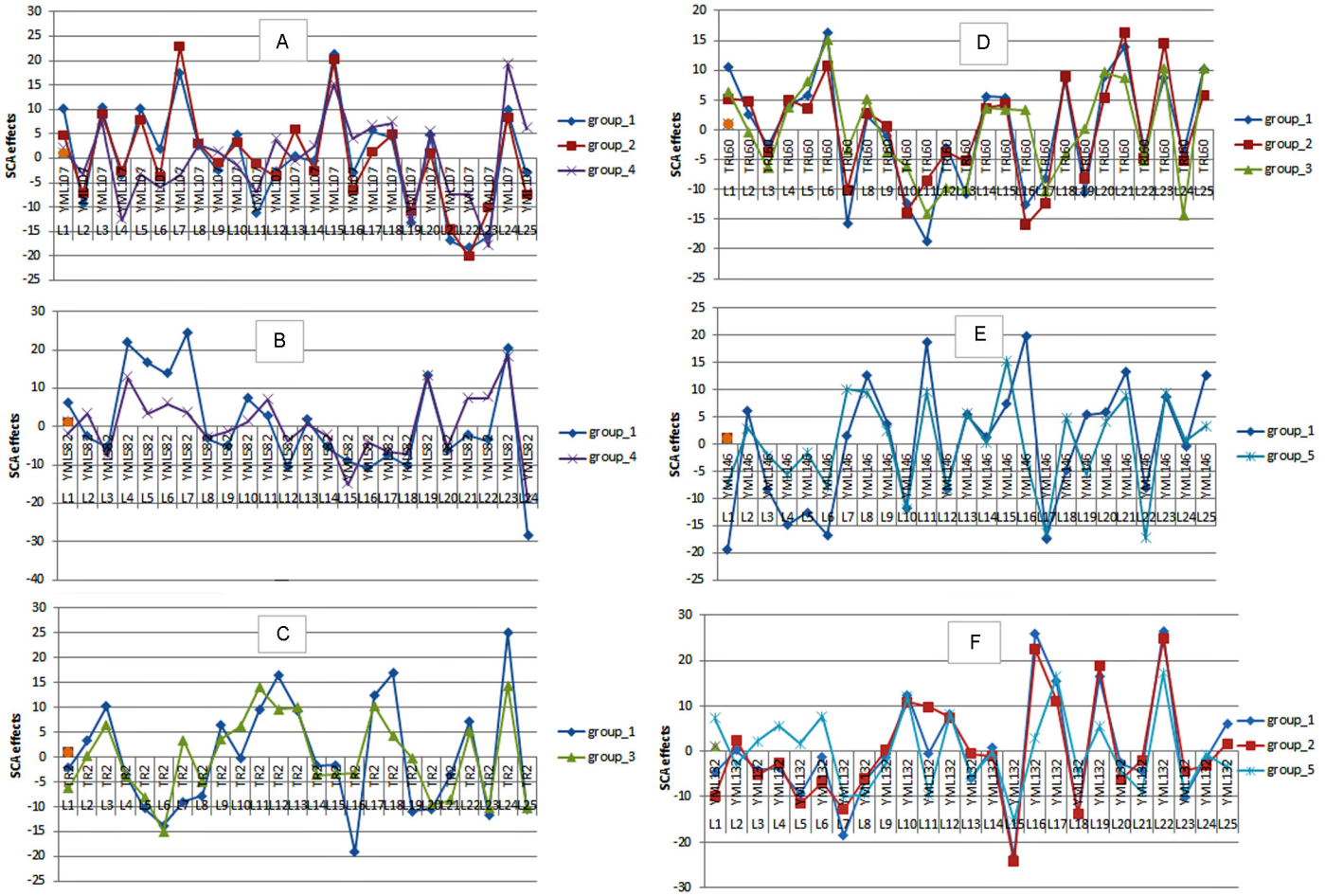

Fig. 2. Specific combining ability (SCA) effects of crosses between 25 exotic lines and six testers that are included in different groups. (A) YML107 used in Group1, Group2, and Group4. (B) YML582 used in Group1 and Group4. (C) TR2 used in Group1 and Group3. (D) TRL60 used in Group1, Group2, and Group3. (E) YML146 used in Group1 and Group5. (F) YML132 used in Group1, Group2, and Group5.

groups (Fig. 2). The SCA effect differences among crosses between the 25 lines and the three testers, with one from each of the three heterotic groups, i.e., Reid, non-Reid, and Suwan1, are shown in Figure 3. Even when testers were from the same heterotic group, the SCA effects for test-crosses between some of the lines and the testers were quite different (Fig. 4). Therefore, to obtain estimates of SCA effects of crosses between the lines and different testers, all possible crosses should be made.

Combining Ability Variation in Sister Lines

It is known that GCA mainly relates to additive genetic effects (Kang, 1994) and genes with such effects accumulate as cyclic selection progresses (Hallauer and Miranda, 1988). However, since the GCA accumulation can be positive or negative, little or no knowledge exists about which segregating generation should be used to begin selection of lines to obtain stable GCA effects. To determine whether or not GCA effects were stable, the coefficient of variation (CV) for GCA effects was calculated for three groups of sister lines selected at different breeding stages from different populations. Line_Group1 included L3, L4, L5, L6, L7, and L8; Line_Group2 included L13 and L22; and Line_Group3 included L21 and L23 (Table 1). The GCA effects for L3, L4, L5, L6, L7, and L8 (Line_Group1) were 9.14, −1.09, −4.27, 8.19, 4.95, and 2.56, respectively (CV = 161.7%); the GCA effects for L13 and L22 (Line_Group2) were −8.59 and −9.05, respectively (CV = 22.0%); and the GCA effects for L21 and L23 (Line_Group3) were 5.73 and 4.15, respectively (CV = 3.6%). As indicated earlier, the lines from Line_Group1 were selected in S3 and the lines from Line_Group2 and Line_Group3 were selected in S4 and S5. The results relative to differences in CV revealed that sister lines selected in S3 generation were less stable than those selected in later generations (S4 or S5) as far as GCA variation is concerned. The relatively large CV for Line_Group1 was indicative of the fact that the lines had variable or unstable performance.

Furthermore, on examining the GCA effects of Line_Group1, Line_Group2, and Line_Group3, we learned that all the GCA effects were negative in Line_Group2 (i.e., GCA of L13 = −8.59, GCA of L22 = −9.05). All the GCA effects were positive in Line_Group3 (i.e., GCA of L21= 5.73, GCA of L23 = 4.15), and the GCA effects were either positive or negative in Line_Group1. Actually, in Line_Group1, L4 (GCA = −1.09) and L5 (GCA= −4.27)

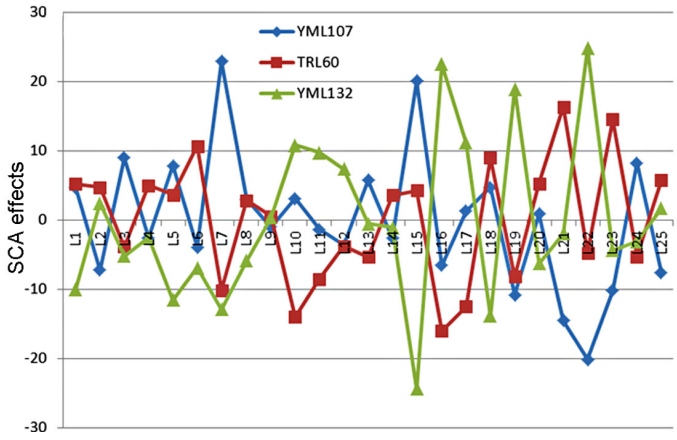

Fig. 3. Specific combining ability (SCA) effects for crosses between 25 lines and three testers, one each from Reid, non-Reid, and Suwan1 heterotic groups.

were selected in S5 from the same S3 line, and L6 (GCA = 8.19), L7 (GCA =4.95), and L8 (GCA = 2.56) were selected in S5 from the same S3 family. These results strongly imply the direction (positive vs. negative) of GCA effects was determined at S3 level and the S3 was a critical stage for diversifying genetic base of the selected lines.

Chen et al. (1994) studied how sister lines could improve GY of a hybrid. They found that if two sister lines (Lines A and B) were selected in S2 (or S3) and when Line A was used to substitute for Line B that was originally used in a hybrid, the hybrid could have 60 to 130% greater GY. When two sister lines were selected at S4 (or S5), the hybrid only had about 5% increase in GY following sister-line substitution. Their results indirectly suggested that sister lines developed at earlier generations (i.e., S2 or S3) would have wider genetic differences than when sister lines were developed in later generations (i.e., S4 or S5). This would be expected because the S2 or S3 lines would be more heterozygous and/or heterogeneous than S4 or S5 lines. Li et al. (2006) studied combining ability of 38 sister lines selected in five different generations and found that GCA and SCA effects segregated among sister lines. A large amount of segregation was found among the sister lines selected during second to fourth generation. Results from this study clearly showed that for GCA effects, a larger amount of variation was detected among the sister lines developed in S3 generation than those among sister lines developed at the S4 or S5 level. Thus, we could conclude that to obtain stable GCA for maize lines, selection must start in S4 or later generations. However, if a sister line is to be substituted for another sister line in a hybrid to improve GY of the hybrid, sister lines developed at S2 and S3 stage should be used to obtain greater GY because of the existence of a relatively large amount of genetic variation at earlier stages.

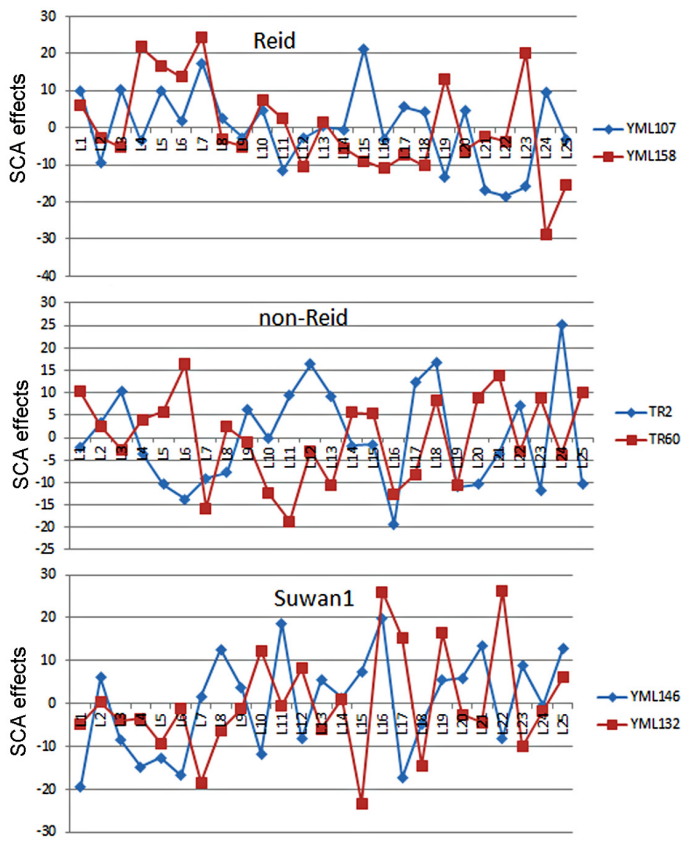

Fig. 4. Specific combining ability (SCA) effects for crosses between 25 lines and six testers from three maize heterotic groups.

CONCLUSIONS

We summarize our results as follows:

1. Though two testers from each of the different heterotic groups of maize might produce similar order of magnitude of GCA effects in most of the tested lines, using one tester from each of the available heterotic groups would provide best estimates of GCA effect. Therefore, for determining which lines to be selected with best GCA effects for a maize hybrid- or inbred line-development program with three heterotic groups and for obtaining reliable GCA estimates, at least one tester from the three heterotic groups should be used.

2. SCA effects for crosses between tested lines and a specific tester would be quite similar when at least one line from each of the available heterotic groups is used as tester. The SCA effects of crosses between tested lines and different testers would be different; thus, SCA effects of these lines with other germplasm not used as tester cannot be predicted and use of testers from different heterotic groups would be necessary for detecting possible crosses with high GY.

3. The S3 seems to be the key generation for selecting maize lines with diverse genetic bases, and S4 or S5 might be the earliest generations for a breeder to start selection from offspring in a cross because by S4 or S5,

most of the additive-effect genes are expected to be fixed for a line to have stable GCA effect.

Acknowledgments

We sincerely acknowledge the financial support from Yunnan Leading Talent Project (2014HA002) and the Crucial New Production Project of Yunnan (2012BB012). We acknowledge the support given by CIMMYT in supplying the lines included in the current study. Help from Yunnan Tian Rui Seed Company and personnel from experimental locations in Wenshan and Dehong is also greatly appreciated.

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