



Determining the Heterotic Group of the Nineteen Open Pollinated Maize Varieties from Their Topcrosses with Inbreed Line Testers in Uganda

Netsanet Abera^{1,2*}, Thomas Odong¹ and Lwanga Charles Kasozi³

¹*Makerere University College of Agricultural and Environmental Sciences, Kampala, P.O.Box 7062, Uganda.*

²*Ethiopian Institute of Agricultural Research, Pawe Agricultural Research Center, P.O.Box 25, Ethiopia.*

³*National Crops Resources Research Institute (NaCRRI), Namulonge, Kampala, P.O.Box 7084, Uganda.*

Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JSRR/2018/38177

Editor(s):

(1) Mahmoud Nasr, Sanitary Engineering Department, Faculty of Engineering, Alexandria University, Egypt.

Reviewers:

(1) Habu Saleh Hamisu, National Horticultural Research Institute, Nigeria.

(2) Ahmed Karmaoui, Morocco.

(3) Desimir Knezevic, University of Pristina, Serbia.

Complete Peer review History: <http://www.sciedomain.org/review-history/23237>

Original Research Article

Received 10th October 2017
Accepted 6th December 2017
Published 17th February 2018

ABSTRACT

Knowledge gap on heterotic patterns for yield limits maize breeding program from exploiting genetic potential of open pollinated maize varieties (OPVs) in Uganda. So this study was conducted to determine the heterotic group of the 19 OPVs using the specific combining ability effects of their topcrosses. The topcrosses were evaluated in 4 Agro-ecologies of Uganda using 5x13 α -lattice design replicated twice per location to test the study hypotheses which was stated as all the 19 OPVs fall under divers heterotic groups. The results of this experiment indicated the presence of low variability for grain yield and thus the possibility of selection among the topcross hybrids that are adapted to the different agro ecologies of Uganda is low. An OPV parent was assigned to group A when its cross with inbred line tester A showed a large negative specific combining ability (SCA) value otherwise it was assigned to opposite heterotic group B. Other

*Corresponding author: E-mail: nabera2004@gmail.com;

parents with lower magnitude of SCA were assigned as heterotic group AB. Based on this criterion, 3 parent OPVs (Longe 4, Longe 5RS and SUWAN) were assigned to the heterotic group A, 2 (SITUKA MI and Ambsyn 5) heterotic group B, and other remaining 14 OPVs were grouped under heterotic group AB. The expressed heterosis between female OPV Ambsyn 5 and SITUKA MI when testcrosses with in bred line tester A (CML536) and Longe 4, Longe 5RS and SUWAN with another tester B (CML202) could be exploited to produce topcrosses and OPV KC2014 as general combiner for yield.

Keywords: Heterotic group; heterosis; open pollinated varieties; general and specific combining ability; topcross.

1. INTRODUCTION

Maize is considered as a strategic food and cash crop that provides a significant amount of protein and energy for both humans and livestock in Uganda, [1]. In spite of increasing popularity of hybrids in the country, open pollinated varieties (OPVs) are still an important category of maize cultivars. They are random mating populations developed recombining 8-10 inbred lines with high general combining ability [2] whereas topcrosses are those non-conventional hybrids produced by test crossing open pollinated varieties with one or more inbred line testers. Therefore, the 19 OPVs may fall under different heterotic groups. The concept of heterotic groups and patterns was suggested by [3], they defined a heterotic group "as a group of related or unrelated genotypes from the same or different populations, which display similar combining ability and heterotic response when crossed with genotypes from other genetically distinct germplasm groups". So heterotic grouping means identifying germplasm groups that are genetically divers each other and produce superior hybrids when crossed. Heterotic pools need to be kept separately to ensure they remain unrelated by parentage. Crossing representatives of different heterotic pools will maximize hybrid vigor and finally grain yield of the new hybrids to be developed. Basically, the term heterotic pattern refers to a definite pair of two heterotic groups, which express high heterosis and as a result high hybrid performance in their cross. Heterotic patterns have high importance in crop improvement due to the existence of large magnitude of genetic diversity in germplasm to be used in a hybrid breeding program over decades [4]. The parents having contrasting but complementary heterotic groups are essential in hybrid maize breeding [5]. Generally, there are three heterotic groups which can arbitrary be designated as A, B and AB, to which the parental lines are assigned in maize breeding. This assigning of maize genotypes into heterotic groups is very important to exploit

heterosis or hybrid vigor [6], mostly for grain yield and other yield related traits [7]. One way of achieving high heterosis in the crosses is by combining diverse parents with known heterotic groups.

Wide genetic diversity exists between open pollinated maize varieties (OPVs) and the study parental lines hence high level of heterosis can be expressed in their topcrosses. In a rational exploitation of the genetic potential of OPVs, determining the genetic divergence between them and the inbred line testers is paramount, if heterosis is to be maximized in their topcrosses. Thus information on relationships between breeding materials is an important requirement for selection of parents in plant breeding programmes. There was therefore a need to assess the genetic potential of topcrosses generated from crosses between OPVs and inbred line testers. The objective of this study was to determine the heterotic groups of the local and introduced OPVs based on their specific combining abilities with the two inbred line testers, as exhibited in ensuing topcrosses evaluated in 4 different agro-ecologies of Uganda.

2. MATERIALS AND METHODS

The research was conducted at four sites: i) The National Crops Resources Research Institute (NaCRRRI) Namulonge (Central Uganda) ii), National Semi-Arid Resources Research Institute (NaSARRI) Serere (Eastern Uganda) iii) Bulindi Zonal Agricultural and Development Research Institute (Western Uganda) and iv) Ngetta Zonal Agricultural and Development Research Institute, Ngetta (Northern Uganda). The detail description of the sites is provided in the Table 1.

2.1 Genetic Materials

Nineteen open pollinated maize varieties and two inbred line testers sourced from East African

Countries (Uganda, Tanzania, Kenya and Ethiopia) and CIMMYT, respectively, were used in this study with their identities shown in Table 2. The heterotic groups of the 19 OPVs were not known while the two inbred lines namely CML536 and CML202 were from the heterotic groups A and B, respectively. OPVs are random mating populations and hence heterosis may be expressed when testcrossed with the inbred lines. When OPVs are testcrossed with inbred line testers, the resulting hybrid is called a topcross.

2.2 Nursery for Generating Topcrosses

The 19 OPVs were crossed to the two inbred line testers A (CML536) and B (CML202), using the Line x Tester mating design. The OPVs were used as female and the two testers as male parent; the study was carried out during the first cropping season of 2015 (season A) at NaCRRRI. One row plot of length 6.25 m was used. A spacing of 0.75 m and 0.25 m was used between rows and within plants, respectively. The female plants were shoot bagged before they started

silking (female flowering) to protect them from outcrossing with unwanted pollen. On the other hand, at pollen shedding stage, the male flowers (tassels) of the two inbred line testers were covered with water-proof paper bags (pollen bags). The tassels were covered for at least 24 hours before pollination, to ensure death of unwanted pollen from other plants, such that whatever pollen being shed in the pollen bag belongs to the target plant (to avoid contamination). Hand pollination was achieved by shaking the pollen bags covering the tassels in order to aid pollen shed, and introducing the pollen (in its bag) to the receptive silks of the target OPV. After pollination the silks were left covered with the respective pollen bags until harvesting still to avoid contamination with unwanted pollen. Hand pollinations were usually done in the morning from around 10:00 am when pollen begins to shed. Each cross was labelled by writing the name of the female and male parents, the name of the person who made crosses and the date of crossing as indicated in Fig. 1. After these series of steps, 38 topcrosses were produced (Table 2).

Table 1. Description of the study site used for testing sixty-five maize genotypes in Uganda, 2015B

Site	Latitude (° North)	Longitude (° East)	Altitude (m.a.s.l.)	Annual rainfall (mm)	Average temperature °C
Namulonge	0° 32'	32° 37'	1200	1242	22.0
Serere	1° 31'	33° 28'	1140	1250	31.3
Bulindi	1° 29'	31° 26'	1218	1400	23.9
Ngetta	2° 14'	32° 54'	1180	1300	23.6

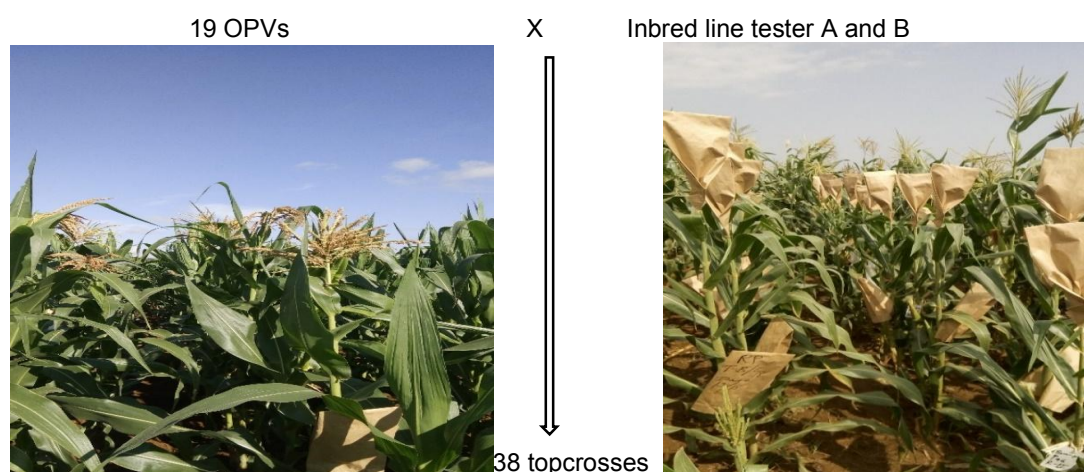


Fig. 1. Line x Tester mating design of 19 OPVs by two inbred line testers A and B carried out to generate 38 topcrosses at NaCRRRI in season 2015A

Table 2. Names and pedigrees of topcrosses generated in season 2015A and used in determination of heterotic groups of the 19 OPVs in Uganda in season 2015B

Entry	Name	Pedigree
1	Topcross1	MM3/CML536
2	Topcross2	MM3/CML202
3	Topcross3	Longe4/CML536
4	Topcross4	Longe4/CML202
5	Topcross5	Longe5/CML536
6	Topcross6	Longe5/CML202
7	Topcross7	Longe5D/CML536
8	Topcross8	Longe5D/CML202
9	Topcross9	Longe5RS/CML536
10	Topcross10	Longe5RS/CML202
11	Topcross11	SITUKA/CML536
12	Topcross12	SITUKA/CML202
13	Topcross13	STAHA/CML536
14	Topcross14	STAHA/CML202
15	Topcross15	TMV1/CML536
16	Topcross16	TMV1/CML202
17	Topcross17	ECAVL1/CML536
18	Topcross18	ECAVL1/CML202
19	Topcross19	ECAVL2/CML536
20	Topcross20	ECAVL2/CML202
21	Topcross21	ECAVL17/CML536
22	Topcross22	ECAVL17/CML202
23	Topcross23	ECAVL18/CML536
24	Topcross24	ECAVL18/CML202
25	Topcross25	KakSyn-II/CML536
26	Topcross26	KakSyn-II/CML202
27	Topcross27	AmbSyn2/CML536
28	Topcross28	AmbSyn2/CML202
29	Topcross29	AmbSyn5/CML536
30	Topcross30	AmbSyn5/CML202
31	Topcross31	KC2014/CML536
32	Topcross32	KC2014/CML202
33	Topcross33	SUWAN/CML536
34	Topcross34	SUWAN/CML202
35	Topcross35	VP MAX/CML536
36	Topcross36	VP MAX/CML202
37	Topcross37	OUI-1/CML536
38	Topcross38	OUI-1/CML202
Total		38

2.3 Experimental Design Used for Evaluating the 38 Topcross Hybrids

The 38 topcross hybrids, their 19 OPV parents and 8 checks (including single, double, 3-way and varietal cross hybrids) were evaluated in four different agro-ecologies of Uganda. The experimental design used was 5 x 13 α -Lattice design with two replications. Two row plots of 5 m long were used with an inter-row spacing of 0.75 m and intra-row spacing of 0.25 m. In order

to ensure a standard population density of 53,333 plants per hectare, two seeds were planted per hill and later thinned to one plant per hill 2 – 3 weeks after germination. Harvesting was done for each of 4 locations (Namulonge, Bulindi, Serere, and Ngetta) on February 5th, March 2nd, 17th, and 19th, respectively. However, trial at Ngetta was affected by prolonged drought.

2.4 Data Collection

Data on yield for 38 topcrosses was collected. Heterotic grouping was determined by estimating the specific combining ability of the topcrosses. Therefore, all the mean yield data of 38 topcrosses at 4 sites were taken to determine the heterotic grouping of the OPVs.

2.5 Statistical Analysis

The yield data only for each of the 38 topcrosses were taken and analysed in Genstat edition 12th to get the specific combining ability for of each the crosses so as to determine heterotic groups the heterotic groups of parent OPVs. The following North Carolina (NC) II linear model was used:

$$Y_{ij} = \mu + GCA_f + GCA_m + SCA_{ij} + \varepsilon_{ij}$$

Where: Y_{ij} is the observed value of i^{th} crosses in j^{th} replication, μ is over all mean crosses, GCA_f is the general combining ability of female, and GCA_m is the general combining ability of male, SCA_{ij} is specific combining ability of the interaction effect between the i^{th} female and the j^{th} male and ε_{ij} is random error.

3. RESULTS

The mean squares for genotypes and its components across in four sites for grain yield in season 2015B are presented in Table 3. The mean square of location was significant ($P \leq 0.001$) whereas GCA_m (Testers) and GCA_f (OPVs) and SCA_{fxm} was non-significant at ($P \leq 0.05$) and the interaction effects $Loc \times GCA_f$ and $Loc \times SCA_{fxm}$ was not also significant too.

The SCA effects of the 38 topcrosses are shown in Table 4. The results of this experiment revealed low heterosis for grain yield. Five parents namely AmbSyn 5, SITUKA MI, Longe 4, Longe 5RS and SUWAN showed positive SCA and greater magnitude than the standard error when testcrossed with inbred line testers A and B. The highest SCA value was recorded for

Table 3. Mean squares of ANOVA across sites for selected maize open pollinated maize and their topcrosses for grain yield in Uganda, 2015B

SOV	d.f.	TSS	MS	Fcal.	F prb.
Loc	3	792.63	264.21	565.40	< 0.001
GCAf	18	21.54	1.20	1.47	0.14
GCAm	1	19.60	19.60	4.78	0.12
Loc*GCAf	54	44.09	0.82	1.78	0.004
Loc*GCAm	3	12.30	4.10	8.78	< 0.001
SCAfxm	18	10.50	0.58	0.95	0.53
Loc*SCAfxm	54	33.31	0.62	1.32	0.09
Pooled Error	150		0.46		

ANOVA- Analysis of variance, SOV- source of variation, d.f- degree of freedom, TSS- total sums of squares, MS- mean squares, Fcal- F calculated, Fprb- F probability, Loc- Location, GCAf-general combining ability of female, GCAm- general combining ability of male, SCAfxm- specific combining ability of crosses

Table 4. Estimates of SCA effects of L x T crosses evaluated for grain yield in 2015B

Female	Male		Mean	GCA _f	SCA _{f x m}		Het.G
	TA	TB			TA	TB	
MM3	4.75	3.95	4.35	-0.55	0.04	-0.04	AB
Longe 4	4.61	4.81	4.71	-0.19	-0.46	0.46	A
Longe 5	5.42	4.67	5.05	0.15	0.01	-0.01	AB
Longe 5D	4.85	4.15	4.50	-0.40	-0.01	0.01	AB
Longe 5RS	5.32	5.34	5.33	0.43	-0.37	0.37	A
SITUKA MI	5.75	4.1	4.93	0.03	0.47	-0.47	B
STAHA	5.48	4.9	5.19	0.29	-0.07	0.07	AB
TMV1	5.73	4.35	5.04	0.14	0.33	-0.33	AB
ECAVL1	5.58	5.09	5.34	0.44	-0.11	0.11	AB
ECAVL2	5.53	4.89	5.21	0.31	-0.04	0.04	AB
ECAVL17	5.28	4.71	5.00	0.10	-0.07	0.07	AB
ECAVL18	5.62	4.5	5.06	0.16	0.20	-0.20	AB
KakSyn-II	5.01	4.33	4.67	-0.23	-0.02	0.02	AB
AmbSyn 2	4.19	4.05	4.12	-0.78	-0.29	0.29	AB
AmbSyn 5	5.19	3.41	4.30	-0.60	0.53	-0.53	B
KC2014	5.99	5.27	5.63	0.73	0.00	0.00	AB
SUWAN	4.79	4.9	4.85	-0.05	-0.41	0.41	A
VP MAX	5.30	4.36	4.83	-0.07	0.11	-0.11	AB
OUI-1	5.48	4.46	4.97	0.07	0.15	-0.15	AB
Mean	5.26	4.54	4.90	-0.001	-0.0005	0.0005	
GCA _M	0.36	-0.36	0.00				
GM			4.90				
SE	0.08	0.08		0.24	0.34	0.34	

OPV-open pollinated varieties, TA-tester A, TB-tester B, GCA_f and GCA_M-general combining ability of female and male parent, SCA_{f x m}-specific combining ability of crosses, SE- Standard error of crosses, GM-over all mean, Het.G-heterotic group, L x T- Line by Tester mating design

parent AmbSyn 5 when test crossed with inbred line tester A while the least value was observed when test crossed with Tester B.

4. DISCUSSION

The results of this experiment indicated the presence of poor (non-significant SCAfxm) for

grain yield among crosses (see Table 3). This could be an indication that the parents used to produce these crosses are related. Therefore, except few parents, the possibility of selection of most parents which can be involved in making topcross hybrids in Uganda is low. Even though the expressed additive (GCA) and non-additive gene effects (SCA) seem to be small on the

average, they may be very important for specific cross combinations [8]. For example, the GCA of parent KC2014 was significant positive for grain yield indicating the increased concentration of favorable alleles. [9] suggested that the performance of a single-cross progeny could be adequately predicted on the basis of GCA, if SCA is not significant. On the other hand, [10] reported that, on average, hybrids produced by crossing inter-population lines have more positive SCA effects than those produced by crossing intra-population lines which tend to have more negative SCA effects.

Based on these results (in Table 4), 3 parental OPVs (Longe 4, Longe 5RS and SUWAN) were assigned to heterotic group A, 2 OPVs (SITUKA MI and Ambsyn 5) to heterotic group B, and the remaining 14 OPVs which showed similar performance and lower SCA value than the SE when crossed with tester A or tester B were assigned to both heterotic group AB. The low heterosis expressed in terms of grain yield revealed limited complementarities (low hybrid vigor) between the test OPVs and the two inbred line testers, suggesting that the two testers and the 19 OPVs may not be distantly related. Consequently, the possibility of selection among high yielding topcross hybrids that are adapted to different agro-ecologies of Uganda is low. The GCA of parent KC2014 was positive and significant for grain yield indicating the increased concentration of favorable alleles and could thus be good general combiner. [9] suggested that the performance of a single-cross progeny could be adequately predicted on the basis of GCA, if SCA is not significant. The choice of heterotic groups is fundamental because heterotic groups and heterotic patterns are important tools for predicting and exploiting heterosis of the trait of interest [4]. Therefore, the higher the levels of heterosis in a cross indicate wide genetic diversity between the parents and ultimately high potential for generating superior hybrids. Similar study with this concept was reported by [11].

5. CONCLUSION AND RECOMMENDATIONS

The heterosis expressed between OPVs Ambsyn5 and SITUKA MI when test crossed with in bred line tester B (CML536) and Longe 4, Longe 5RS and SUWAN with tester A (CML536) indicates that the two sets of OPVs belong to the complementary heterotic groups. That is OPV Ambsyn 5 and SITUKA MI belong to heterotic group A, OPVs Longe 4, Longe 5RS and

SUWAN belong to heterotic group B, while the remaining 14 OPVs which neither exhibited high heterosis levels with tester A nor tester B belong to AB heterotic group. Therefore, Ambsyn 5 and SITUKA MI can be used as OPV testers A, while Longe 4, Longe 5RS and SUWAN can be used as open pollinated testers B. In all OPVs Ambsyn5, SITUKA MI, Longe 4, Longe 5RS and SUWAN and KC2014 are recommended for use in the breeding program for cultivar development. On the other hand, variety KC2014 which exhibited positive and significant GCA towards grain yield can be utilized in selection of parents for variety improvement. The better heterosis found for grain yield of these OPVs as heterotic group A and B indicated the potential of those OPVs for inbred line and hybrid development. However, molecular techniques would be required to validate the suitability of the listed OPVs.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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