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Combining Ability, Gene Action and Yielding Performance in Maize

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Abstract: Recombinant lines developed from combining Half-sib/S1 evaluation on widely-spaced plants in the direction of high yielding per se, were used in combining ability tests in order to determine gene action under three-way crossing and possible commercial exploitation of such crosses. Combining ability tests consisted of crosses between: a) recombinant lines of common pedigree, b) recombinant lines and freely available inbred lines and c) recombinant lines and commercial F1 maize hybrids (three-way crosses). Heterosis was found to be acceptable, since the best three-way crosses reached the performance of corresponding single-cross hybrids and yielding performance of these three-way crosses was stable across years and experimental fields. Rapid line development from combining Half-sib/S1 evaluation may ensure high and stable crossing performance, based on additive gene action. Stability and uniformity of performance of three-way crosses was due to proper breeding incorporated in single-cross hybrid and to high and stable inbred line performance.

Key words: Three-way crosses, recombinant lines, heterobeltiosis, combining ability

INTRODUCTION

The starting genetic material of a breeding program is very important, since it determines the potential improvement for traits under selection^[1]. Source germplasm used by maize breeders for inbred development includes primarily F2 (elite X elite inbred crosses), backcross and synthetic populations^[2]. Modern maize breeding is based primarily on genetically narrow-based populations^[2,3] including elite-line synthetics with a restricted genetic base, F2 populations of single crosses and backcross populations. For a successful program of recycling, the choice of the germplasm is the first priority^[4]. A successful breeding program in developing new hybrids, depends not only on the germplasm but also on the procedure for developing inbred lines and consequently new promising crosses^[5]. Maize breeders lead their breeding programs to an excessive exploitation of heterosis, which is rendered on the function of alleles showing dominance effects^[6,7]. According to Kearsey and Pooni^[8], heterosis is caused by dispersed genes showing mainly directional dominance and not by heterozygote superiority or complementary epistasis. Additive gene action is of great importance, because this kind of action insures heritable and stable performance^[5,9,10] and may insure high yields for crosses developed under genetically narrow-based crossing programs^[5,11]. Considering crossing between recombinant lines of common pedigree, general combining ability is of small importance, since heterosis was found low and heterobeltiosis was even lower^[5]. These crosses depend

their yielding performance on favourable additive gene effects, common in the two parents and consequently the second-cycle hybrids between them must be in a lower level of heterozygosity compared to the original hybrid^[5,9,11-14]. Genter and Alexander^[15] stated that, if the performance of S1 lines depends mainly on additive effects, then the yield of their crosses would be proportional to their yielding performance per se and thus, it is possible, that selection practiced for improving line performance per se, leads to the accumulation of favourable additive genes. Sotiriou *et al.*^[16] concluded that in such cases the genetic background consist mainly of additive or partially dominant alleles. When crossing unrelated inbred lines, middle-parent heterosis can be more easily realized in comparison to crossing between recombinant lines of common pedigree^[5]. A maize breeder has to balance between the three main goals of breeding (productivity, uniformity, stability) and the cost of hybrid seed production because of the low productivity of inbred parents as well as the need of separate plantings under isolation^[13]. The practical difficulties associated with the low productivity of inbred lines can be overcome by three different approaches:

- a) The use of 3-way or 4-way crosses^[7,13,17].
- b) The use of the backcross method for step-by-step improvement^[7,13].
- c) The improvement of productivity per se of modern inbred lines that serve as parents in crosses^[4,5]. The most common practice involves family selection schemes^[7,13].

The purpose of this study was to explore combining ability in recombinant S-lines in order to determine gene action under three-way crossing and possible commercial exploitation of such crosses, since the S-lines developed from combining Half-sib/S1 evaluation may ensure high and stable per se and crossing performance^[5].

MATERIALS AND METHODS

Recombinant lines developed from combining Half-sib/S1 evaluation on widely-spaced plants in the direction of high yielding per se, were used in combining ability tests in order to determine gene action under three-way crossing. The recombinant lines were developed from the F2 generation of F1 commercial single-cross maize hybrid Lorena (PR3183), according to the procedure described by Ipsilandis and Koutsika-Sotiriou^[5].

Combining ability tests consisted mainly of crosses between: a) Recombinant Lines (RL) of common pedigree, b) recombinant lines and freely available (FR) inbred lines and c) recombinant lines and commercial F1 maize hybrids (three-way crosses). No balanced design was used.

Evaluation of crosses was conducted in Technological Education Institute farm of Larissa, Greece, in two different fields, in five years (from 1998 to 2002). In 1998, the materials evaluated were:

- Seven F1 single-cross commercial hybrids (Lorena, Dracma, Costanza, Rio Grande, Marista, Prisma and Aris).
- Two F2 from single-cross hybrids (Costanza F2, Prisma F2).
- Thirteen three-way crosses (nine 3-ways with RL lines, four with FR lines).
- One double cross (Prisma X Costanza).
- Five single crosses (one FR X FR, four RL X FR).

In 1999, the materials evaluated were:

- Eight F1 single-cross commercial hybrids (Aligreen, Costanza, CS1251, Prisma, Dracma, LG2360, Golden West 623 and 621).
- Fourteen three-way crosses (seven 3-ways with RL lines, seven with FR lines).
- Three single crosses (RL X FR).

In 2000, the materials evaluated were:

- Six F1 single-cross commercial hybrids (Aligreen, Costanza, CS1251, Prisma, Rio Grande and Dias).
- One double hybrid (Aligreen X Rio Grande).
- Fourteen three-way crosses (seven 3-ways with RL lines, seven with FR lines).
- Five single crosses (one RL X RL, four RL X FR).

In 2001, the materials evaluated were:

- Three F1 single-cross commercial hybrids (Aligreen, Costanza and Volusia).
- One double hybrid (Costanza X Aligreen).
- Ten three-way crosses (six 3-ways with RL lines, seven with FR lines).
- Eight single crosses (five RL X RL, two RL X FR, one FR X FR experimental hybrid).

In 2002, the materials evaluated were:

- One F1 single-cross commercial hybrid (Costanza).
- Four three-way crosses (with RL lines).
- Seven single crosses (three RL X RL, three RL X FR, one FR X FR).

The experimental design, was the Randomized Complete Block (RCB) with 4 replications for all field trials. In all yield tests the experimental plot consisted of two 5 m long rows, spaced 80 cm apart. All plots contained 50 plants, i.e. 25 plants/row giving a density of 6.25 plants/m². Plots were overplanted and thinned to desired stand at the seedling stage. Nitrogen and P fertiliser were applied at regular quantities. Trials were regularly irrigated (to avoid drought stress). Grain moisture was adjusted at 15.5%.

The eleven inbred lines used in experiments (single and 3-way crosses) were previously selected in the basis of productivity per se, after comparison to free-release inbred line B73 (in RCB design, similar to the one described above).

Middle-parent Heterosis (MPH%), Heterobeliosis to second parent in cross (SPHB%) and Heterobeliosis to best parent in cross (BPHB%) for all types of crosses were computed by formulas proposed by Koutsika-Sotiriou and Bos^[18] and applied by Ipsilandis and Koutsika-Sotiriou^[5].

RESULTS

Table 1 clearly demonstrated that the yielding performance per se of all inbred lines used in crosses, was very high (up to 8000 kg ha⁻¹). Yields of best performing line A1 reached 211% of free-release inbred line B73, 56% of F1 hybrid Lorena and 58% of average yield of F1 hybrid Costanza (Table 2-6). The average yield of Costanza is equal to the yielding performance of year 1998, the first year of experimentation (base yield), facilitating comparisons of lines and crosses across years. Nine inbred lines (recombinant and free-release) differed significantly when compared to B73, while two inbred

Table 1: The inbred lines used in crosses their type and origin, yield per se in kg ha⁻¹, relative yield as percentage: of B73 (%B73), of F1 hybrid Lorena (%Lorena), of F1 hybrid Costanza in year 1998, 1999, 2000, 2001, 2002 and average of five years, respectively; %Cost98, %Cost99, %Cost00, %Cost01, %Cost02 and %CostAvg

Inbred line	Type	Origin	Yield	%B73	%Lorena	%Cost98	%Cost99	%Cost00	%Cost01	%Cost02	%Cost Avg
A1	RL	Ipsilandis and Koutsika-Sotiriou ^[2]	8000	211	56	58	55	61	52	68	58
A2	RL	Ipsilandis and Koutsika-Sotiriou ^[2]	6000	158	42	44	41	46	39	51	44
A3	RL	Ipsilandis and Koutsika-Sotiriou ^[2]	5100	134	36	37	35	39	33	43	37
P6GR	FR	Greece	5000	132	35	36	34	38	33	43	37
P1GR	FR	Greece	4800	126	34	35	33	37	31	41	35
D3GR	FR	Greece	4700	124	33	34	32	36	31	40	34
D4GR	FR	Greece	4600	121	32	34	32	35	30	39	34
B84	FR	USA	4550	120	32	33	31	35	30	39	33
Va26	FR	USA	4500	118	31	33	31	34	29	38	33
A4	RL	Ipsilandis and Koutsika-Sotiriou ^[2]	4100	108	29	30	28	31	27	35	30
A5	RL	Ipsilandis and Koutsika-Sotiriou ^[2]	4000	105	28	29	28	31	26	34	29
B73	FR	USA	3800	100	27	28	26	29	25	32	28
LSD ₀₅ = 650,			Average = 4929								

Table 2: Yield in kg ha⁻¹, relative yield as percentage of F1 hybrid Costanza (%Cost), Middle-parent Heterosis (MPH%), Heterobeltiosis to second parent in cross (SPHB%) and Heterobeltiosis to best parent in cross (BPHB%) for all types of crosses in year 1998

Cross	Yield	%Cost	MPH%	SPHB%	BPHB%
Lorena	14300	104			
Dracma	14040	102			
Costanza	13715	100			
Rio Grande	13130	96			
Aris	11895	87			
Prisma	11505	84			
Marista	11310	82			
Average F1	12842	94			
Costanza F2	7573	55			
Prisma F2	6793	50			
Average F2	7183	52			
Marista X A3 RL	12935	94	58	154	14
Costanza X A1 RL	12643	92	16	58	-8
Prisma X A3 RL	12383	90	49	143	8
RioGrande X A1 RL	11050	81	5	38	-16
Costanza X A4 RL	11018	80	24	169	-20
Prisma X A4 RL	10985	80	41	168	-5
Prisma X A1 RL	10628	77	9	33	-8
Rio Grande X A4 RL	10563	77	23	158	-20
Rio Grande X A2 RL	10530	77	10	76	-20
Average F1 X RL	11415	83			
Costanza X Va26 FR	11635	85	28	159	-15
Prisma X Va26 FR	10075	73	26	124	-12
Marista X Va26 FR	9100	66	15	102	-20
Rio Grande X P1GR FR	8808	64	-2	83	-33
Average F1 X FR	9904	72			
Average 3-way	10950	80			
A4 RL X P6GR FR	12350	90	171	201	147
A2 RL X Va26 FR	12025	88	129	167	100
A3 RL X B84 FR	10303	75	114	126	102
A1 RL X Va26 FR	10303	75	65	129	29
Average RL X FR	11245	82			
D3GR FR X D4GR FR	8808	64	89	91	87
Average single-cross	10758	78			
Prisma X Costanza	10985	80			
Average double-cross	10985	80			
LSD ₀₅ = 1170, Average = 11121			81		

lines although with better performance were at the same level of productivity (no significant difference) when compared to B73.

In Table 2, the best yielding F1 hybrid found to be Lorena (14300 kg ha⁻¹). F1 hybrid Costanza reached

Table 3: Yield in kg ha⁻¹, relative yield as percentage of F1 hybrid Costanza (%Cost), Middle-parent Heterosis (MPH%), Heterobeltiosis to second parent in cross (SPHB%) and Heterobeltiosis to best parent in cross (BPHB%) for all types of crosses in year 1999

Cross	Yield	%Cost	MPH%	SPHB%	BPHB%
Prisma	14850	102			
Aligreen	14776	102			
Costanza	14534	100			
CS1251	14068	97			
Dracma	13500	93			
Golden West 623	11850	82			
Golden West 621	11730	81			
LG2360	9344	64			
Average F1	13082	90			
Costanza X B84 FR	14244	98	47	195	-2
Prisma X Va26 FR	13020	90	33	173	-12
Costanza X Va26 FR	12810	88	33	169	-12
Dracma X Va26 FR	11940	82	31	150	-12
LG2360 X B84 FR	11746	81	66	144	26
LG2360 X Va26 FR	10682	73	51	124	14
Dracma X B84 FR	10426	72	14	116	-23
Average F1 X FR	12124	83			
Costanza X A1 RL	14112	97	23	66	-3
Dracma X A1 RL	13752	95	25	62	2
Prisma X A1 RL	12856	88	10	52	-13
Costanza X A4 RL	12600	87	33	190	-13
Dracma X A3 RL	12060	83	28	123	-11
LG2360 X A1 RL	11610	80	30	37	24
Dracma X A5 RL	10544	73	19	149	-22
Average F1 X RL	12505	86			
Average 3-way	12314	85			
A3 RL X B84 FR	14240	98	178	195	163
A4 RL X P6GR FR	13750	95	185	216	159
A1 RL X Va26 FR	12690	87	92	166	50
Average single-cross	13560	93			
Prisma X Dracma	10200	70			
Average double-cross	10200	70			
LSD ₀₅ = 1020, Average = 12613			87		

13715 kg ha⁻¹. The F2 generation of Costanza reached 55% of original hybrid, while the average F2 performance of Costanza and Prisma reached 52% of Costanza F1 and 57% of average performance between Costanza F1 and Prisma F1 (12610 kg ha⁻¹). The average yield of F1 found to be 12842 kg ha⁻¹.

The average of 3-way crosses found to be 10950 kg ha⁻¹. Three-way crosses between F1 and RL

Table 4: Yield in kg ha⁻¹, relative yield as percentage of F1 hybrid Costanza (%Cost), Middle-parent Heterosis (MPH%), Heterobeltiosis to second parent in cross (SPHB%) and Heterobeltiosis to best parent in cross (BPHB%) for all types of crosses in year 2000

Cross	Yield	%Cost	MPH%	SPHB%	BPHB%
Aligreen	13854	106			
Costanza	13112	100			
Prisma	12906	98			
CS1251	12796	98			
Rio Grande	12174	93			
Dias	11146	85			
Average F1	12665	97			
Costanza X B84 FR	13644	104	56	214	4
Aligreen X Va26 FR	13408	102	48	212	-3
Aligreen X B84 FR	13290	101	46	206	-4
Costanza X P6GR FR	12640	96	41	164	-4
CS1251 X Va26 FR	12080	92	41	181	-6
Costanza X Va26 FR	11926	91	37	177	-9
Rio Grande X Va26 FR	9372	71	14	118	-23
Average F1 X FR	12337	94			
Rio Grande X A1 RL	13094	100	32	71	8
Costanza X A1 RL	12774	97	23	67	-3
Aligreen X A1 RL	12652	96	18	65	-9
Rio Grande X A2 RL	12024	92	34	110	-1
CS1251 X A4 RL	11706	89	40	199	-9
Costanza X A4 RL	10886	83	28	178	-17
Aligreen X A2 RL	10464	80	7	82	-24
Average F1 X RL	11943	91			
Average 3-way	12140	93			
A1 RL X Va26 FR	12894	98	116	200	69
A2 RL X Va26 FR	12836	98	156	198	124
A1 RL X B84 FR	12616	96	110	190	65
A4 RL X P6GR FR	11008	84	153	181	130
Average RL X FR	12338	94			
A1 RL X A2 RL	11994	91	79	109	57
Average single-cross	12270	94			
Aligreen X Rio Grande	8376	64			
Average double-cross	8376	64			
LSD ₀₅ = 1200, Average = 12141	93				

lines were better performing when compared to 3-way between F1 and FR lines. Single crosses (in average) yielded close to the average performance of the three-way crosses and close to the performance of the double cross Prisma X Costanza.

Similar data are presented in Table 3-6, where the average performance of F1 was always better than the average of all other type of crosses. In general, three-way crosses between F1 and RL lines were better performing when compared to 3-way between F1 and FR lines (Table 2-6). A few experimental single crosses found to be better than F1 hybrids (the average in year 1999, Table 3). Various three-way crosses yielded near or better than the corresponding F1 hybrid, exhibiting high Heterobeltiosis to second parent (around 200%) and satisfactory Middle-parent Heterosis (over 50%) and in some cases Heterobeltiosis to F1 parent.

The Coefficient of Variation (%) is presented in Table 7. This includes crosses with average CV across years <10%. Costanza X Va26 and Marista X Va26 exhibited the lower Cvs, that is only 3% in average

Table 5: Yield in kg ha⁻¹, relative yield as percentage of F1 hybrid Costanza (%Cost), Middle-parent Heterosis (MPH%), Heterobeltiosis to second parent in cross (SPHB%) and Heterobeltiosis to best parent in cross (BPHB%) for all types of crosses in year 2001

Cross	Yield	%Cost	MPH%	SPHB%	BPHB%
Costanza	15287	100			
Aligreen	15254	100			
Volusia	14232	93			
Average F1	14924	98			
Volusia X B84 FR	13758	90	43	171	-3
Costanza X P6GR FR	10285	67	-1	85	-33
Aligreen X P6GR FR	9688	63	-7	74	-36
Average F1 X FR	11243	74			
Volusia X A1 RL	14608	96	26	64	3
Costanza X A1 RL	13459	88	11	51	-12
Costanza X A5 RL	13107	86	33	194	-14
Costanza X A2 RL	13081	86	19	96	-14
Prisma X A2 RL	12433	81		86	
(P6GR X A4 RL)					
X A2 RL	9534	62		43	
Aligreen X A3 RL	8110	53	-23	43	-47
Average F1 X RL	12047	79			
Average 3-way	11806	77			
B84 FR X P6GR FR	14515	95	173	186	160
Average FR X FR	14515	95			
A1 RL X P6GR FR	13357	87	84	140	50
A4 RL X Va26 FR	13279	87	177	191	165
Average RL X FR	13318	87			
A5 RL X A1 RL	12075	79	81	171	35
A4 RL X A1 RL	11937	78	77	161	34
A3 RL X A1 RL	11259	74	54	98	26
A5 RL X A4 RL	10519	69	133	136	130
A5 RL X A2 RL	9089	59	63	104	36
Average RL X RL	10976	72			
Average single-cross	12004	79			
Costanza X Aligreen	11982	78			
Average double-cross	11982	78			
LSD ₀₅ = 990, Average = 12311	81				

Table 6: Yield in kg ha⁻¹, relative yield as percentage of F1 hybrid Costanza (%Cost), Middle-parent Heterosis (MPH%), Heterobeltiosis to second parent in cross (SPHB%) and Heterobeltiosis to best parent in cross (BPHB%) for all types of crosses in year 2002

Cross	Yield	%Cost	MPH%	SPHB%	BPHB%
Costanza	11746	100			
Average F1	11746	100			
Costanza X A1 RL	11634	99	25	70	-1
Damao X A1 RL	9286	79		36	
Robinia X A1 RL	8984	76		31	
Costanza X A3 RL	7584	65	-6	74	-35
Average 3-way	9372	80			
A4 RL X A2 RL	11364	97	163	224	121
A5 RL X A3 RL	6160	52	58	80	41
A3 RL X A4 RL	6044	51	53	72	38
Average RL X RL	7856	67			
A3 RL X Va26 FR	10174	87	147	164	133
A2 RL X B84 FR	8144	69	80	109	58
A3 RL X B84 FR	6720	57	63	72	54
Average RL X FR	8346	71			
B84 FR X Va26 FR	6510	55	68	69	67
Average single-cross	7874	67			
LSD ₀₅ = 1722, Average = 8696	74				

(1% for Costanza X Va26 in 1998). Costanza F1 had a CV = 9% in average (Table 7).

Combining data from results, three-way crosses between Costanza X A1 were high yielding (94.756% of

Table 7: The Coefficient of Variation % as a parameter of stability for years: 1998 (CV98), 1999 (CV99), 2000 (CV00), 2001 (CV01) and average CV (CV Avg), only for crosses with average CV<10%

Crosses	CV98	CV99	CV00	CV01	CV Avg
Costanza X Va26 FR	1.0	4	4		3.0
Marista X Va26 FR	3.0				3.0
Costanza X A1 RL	5.5	5	7	7	6.1
Lorena	6.0				6.0
Costanza X A4 RL	6.5	9	9		8.2
Rio Gande	6.5	9	9	7	7.9
Prisma X A1 RL	7.5	8			7.8
A1 RL X Va26 FR	8.0	9	9		8.7
A3 RL X B84 FR	8.5	9			8.8
Dracma	9.0	8			8.5
Costanza	10.0	8	10	8	9.0
Aligreen		9	10	9	9.3
A2 RL X Va26 FR	9.0		10		9.5
Prisma	11.0	9	11	9	10.0

Costanza F1 yield) with a low CV (near 6%) and the same was found for Rio Grande X A1 (90.22% of Costanza F1 yield). These three-way crosses to the best performing inbred line A1 were always in the top of ranking and the average yield was at 89.5% of Costanza F1. In general, the average yield for all three-way crosses was at 82.1% of Costanza F1. Three-way crosses to RL lines exhibited better performance in comparison to FR lines (83.15% against 81.05% of Costanza F1). Costanza and Rio Grande were crossed successfully to RL lines (87.25% and 85.186% of Costanza F1 in average, respectively), but only Costanza was crossed successfully to FR lines (89.95% of Costanza F1 in average). Prisma and Dracma crossed satisfactory to RL lines (near 84% of Costanza F1) and Aligreen to FR lines (near 89% of Costanza F1). Costanza X Va26 (average yield 87.97% of Costanza F1) and Costanza X B84 (101% of Costanza F1, Middle-parent Heterosis around 50%) three-way crosses were successful. The average yield of three-way crosses to inbred line B84 was found at 91% of Costanza F1.

DISCUSSION

It is expected, high yielding inbred lines used in crosses to express favorable additive gene action when considering yielding performance per se or in crosses where the level of heterozygosity is low^[5,9,11,19]. Another consequence of this fact is that hybrids formed by crossing high yielding inbred lines may depend a portion of their vigor on favorable additive gene action, since such kind of loci may be alike in the two parents especially when they derived from common pedigree^[5,9,19,20].

The average of the F2 evaluated in 1988 were the expected one by the theory^[10,13], near 55% of the respective F1 yielding performance. Three-way crosses, single crosses and double crosses were found to be at the same level of productivity and in general, three-way

crosses between F1s and RL lines were better performing when compared to 3-way between F1s and FR lines. Various three-way crosses yielded near or better than the corresponding F1 hybrid, exhibiting high heterobeltiosis to second parent and satisfactory Middle-parent heterosis and in some cases Heterobeltiosis to F1 parent. These findings indicate that, new combinations formatting three-way crosses could exploit both favorable additive and dominant gene action because of the lower level of heterozygosity in comparison to F1 and the high level of productivity of the inbred parents^[5,8,9,11-14].

The use of three-way commercial hybrids is a low-cost technique for seed companies^[13] but the lack of genetic uniformity may lead in competition effects and unstable yielding performance^[9]. Fasoulas^[9,21], Fasoula and Fasoula^[22-24] proposed selection of parents in absence of competition and evaluation in many places from the first stages of breeding and of course evaluation of the final crosses in various environments to achieve stability of yielding performance. Fasoulas^[9,21], Fasoula and Fasoula^[22-24] consider that Coefficient of Variation (CV) is a meter of uniformity and stability. From this point of view, the low CVs of a number of three-way crosses indicate uniformity and stability. Additionally, many three-way crosses exhibited high and stable yields across years of evaluation. Visual observation revealed satisfactory uniformity within plots.

Since a number of three-way crosses exhibited high yields accompanied by uniformity and stability, depending their performance on the productivity and stability of the two parents, it seems to be a means for exploiting heterosis under low-cost hybrid production programs. F1 hybrid Costanza proved to be a good mother parent for development of three-way crosses with high and stable yielding performance. The RL inbred line A1 was combined easily to F1 hybrids to produce high and stable three-way crosses. It is clear that this line contributed useful additive gene action, which usually leads to stable performance^[5,9,11]. The B84 inbred line exhibited considerable specific combining ability, in some cases and Va26 inbred line contributed in stable performance.

The expected Middle-parent Heterosis (MPH%) in three-way crosses involving recombinant lines (RL), may be computed by the equation; $MPH = 76.166 + 0.411Y$, where, Y is the relative yield of the cross in comparison to Costanza F1 ($r = 0.62$, $p < 0.001$). Heterobeltiosis to the best parent (BPHB%) may be computed by the equation; $BPHB = 90.516 + 0.544Y$, where, Y is the relative yield of the cross ($r = 0.73$, $p < 0.001$). No relation between Heterobeltiosis to inbred line parent (SPHB%) and three-way cross performance was found, indicating different combining performance of RL inbred lines.

The expected Middle-parent Heterosis (MPH%) in three-way crosses involving free-release inbred lines (FR), may be computed by the equation; $MPH = 27.296 + 0.515Y$, where, Y is the relative yield of the cross in comparison to Costanza F1 ($r = 0.77$, $p < 0.001$). Heterobeltiosis to the best parent (BPHB%) may be computed by the equation; $BPHB = 88.654 + 0.504Y$, where, Y is the relative yield of the cross ($r = 0.57$, $p < 0.005$). Heterobeltiosis to the inbred line parent (SPHB%) may be computed by the equation; $SPHB = 37.891 + 0.305Y$, where, Y is the relative yield of the cross ($r = 0.98$, $p < 0.001$). In general, heterotic phenomena were alike in the two kinds of three-way crosses. This is not in agreement with that found for single crosses^[24] and may be attributed in heterotic gene action contributed by the hybrid parent.

As a conclusion, the development of three-way crosses by using modern maize hybrids, may be an easy method to exploit heterosis and achieve high and stable yielding performance. The cost of this method is considerably low since the parents used in this study (F1 hybrids and inbred lines) are highly productive. In such cases, high and stable yields may depend on both useful additive and dominant gene action.

REFERENCES

1. Fountain, M.O. and A.R. Hallauer, 1996. Genetic variation within maize breeding populations. *Crop Sci.*, 36: 26-32.
2. Bauman, L.F., 1981. Review of methods used by breeders to develop superior corn inbreds. *Proceedings Annual Corn and Sorghum Industrial Res. Conf.*, 36: 199-208.
3. Hallauer, A.R., 1979. Corn breeding opportunities in the 1980's. Des Moines IA: Annual Corn Iowa Seed Dealers Association.
4. Duvick, D.N., 1996. Plant breeding, an evolutionary concept. *Crop Sci.*, 36: 539-548.
5. Ipsilandis, C.G. and M. Koutsika-Sotiriou, 2000. The combining ability of recombinant S-lines developed from an F2 maize population. *J. Agril. Sci., Cambridge*, 134: 191-198.
6. Smith, O.S., 1984. Comparison of effects of reciprocal recurrent selection in the BSSS(R), BSCB1(R) and BS6 population. *Maydica*, 24: 1-8.
7. Falconer, D.S., 1989. *Introduction to Quantitative Genetics*. 3rd Edn. London: Longman.
8. Kearsey, M.J. and H.S. Pooni, 1992. The Potential of Inbred Lines in the Presence of Heterosis. In: *Reproductive Biology and Plant Breeding* (Eds. Dattee, Y., C. Dumas and A. Gallais), London: Springer-Verlag, pp: 371-386.
9. Fasoulas, A.C., 1993. *Principles of Crop Breeding*. A.C. Fasoulas. P.O. Box 1555, Thessaloniki, GR-54006.
10. Hallauer, A.R. and J.B. Miranda Fo., 1981. *Quantitative Genetics in Maize Breeding*. 1st Edn. Iowa State Univ. Press. Ames, Iowa.
11. Ipsilandis, K. and M. Koutsika-Sotiriou, 1997. High Line Performance per Se under Low Stress Conditions. In: *XVIIth Conference on Genetics, Biotechnology and Breeding of Maize and Sorghum*. EUCARPIA (Ed. Tsafaris, A.). Thessaloniki, pp: 191-196.
12. Genter, C.F., 1967. Inbreeding without inbreeding depression. In: *Proceedings, 22nd Hybrid Corn Industry-research Conference*. Washington D.C. American Seed Trade Association, pp: 82-90.
13. Fehr, W.R., 1987. *Principles of Cultivar Development*. Vol. 1. Macmillan Publ. Co.
14. Koutsika-Sotiriou, M., I. Bos and A. Fasoulas, 1990. Hybrid reconstruction in maize. *Euphytica*, 45: 257-266.
15. Genter, C.F. and M.W. Alexander, 1966. Development and selection of productive S1 inbred lines of corn (*Zea mays* L.). *Crop Sci.*, 6: 429-431.
16. Sotiriou, A., M. Koutsika-Sotiriou and E. Gouli-Vavdinoudi, 1996. The effect of honeycomb selection for grain yield on a maize population. *J. Agril. Sci., Cambridge*, 127: 143-149.
17. Hallauer, A.R., 1990. Methods used in developing maize inbreds. *Maydica*, 35: 1-16.
18. Koutsika-Sotiriou, M. and I. Bos, 1996. Heterosis after several numbers of cycles of mass honeycomb selection in maize. *J. Genetics and Plant Breeding*, 50: 7-14.
19. Ipsilandis, K., 1996. The possibility to predict combining ability between maize inbred lines based on best cross performance. Ph.D. Thesis, Aristotle University of Thessaloniki. Greece.
20. Ipsilandis, C.G. and B.N. Vafias, 1998. Selection of elite lines derived from F2 generation of an F1 commercial single maize hybrid (In: Greek with English summary). *Proceedings, 7th Hellenic Congress on Plant Breeding*. Heraklion, Crete, pp: 17-25.
21. Fasoulas, A.C., 1988. *The Honeycomb Methodology of Plant Breeding*. A. Altidjis. Thessaloniki, GR-54006.
22. Fasoula, D.A. and V.A. Fasoula, 1997a. Competitive ability and plant breeding. *Plant Breed. Rev.*, 14: 89-138.
23. Fasoula, D.A. and V.A. Fasoula, 1997b. Gene action and plant breeding. *Plant Breed. Rev.*, 15: 315-374.
24. Fasoula, V.A. and D.A. Fasoula, 2000. Honeycomb breeding: Principles and applications. *Plant Breed. Rev.*, 18: 177-250.