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Analysis of Heterosis In Spring Maize (*Zea mays* L.) Germplasm to Heat Tolerance

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Abstract: This study evaluates 21 maize crosses through a series of 7x7 half diallel crosses initiated during the Rabi season in 2022 and planted again in the spring season of 2023 in Prayagraj, Uttar Pradesh. The objective was to assess mid-parent heterosis, heterobeltiosis, and economic heterosis based on the 20 characters analysed in maize. The mean sum of squares for all the characters suggested that parents were quite variable and a considerable amount of variability existed among the hybrids. Crosses P1xP7, P5xP6, P2xP6, P3xP5 and P1xP5 exhibited significant positive mid-parent heterosis values, whereas crosses P2xP6, P5xP6, P3xP5, P4xP5 and P1xP7 showed high heterobeltiosis values. Crosses P1xP7 and P2xP6 demonstrated significant positive economic heterosis. The hybrids P1xP7 and P2xP6 demonstrated superior performance in both yield and heat tolerance. The research provides valuable insights into developing maize cultivars resilient to heat stress crucial for enhancing agricultural sustainability in the face of challenges such as diminished yields, stunted growth and reduced productivity.

Keywords: Maize, Heterosis, Heat, Yield

Maize (*Zea mays* L.) commonly referred to as corn, is believed to have originated from central Mexico approximately 7000 years ago (Ranum et al 2014). Maize cultivation is preferred because of its easy-growing nature, ability to yield significant harvests, convenient storage options and high starch content which can be easily converted into usable energy. Spring maize is sown under low temperature conditions in the months of January and February, and the vegetative growth phase thus takes place at a range of low to medium temperatures in February and March. However, the reproductive stage of spring-sown maize occurs at high temperatures in May, and the crop is harvested under high temperature conditions in June and July. (Yousaf et al 2020). The reproductive stage of maize plants is extremely vulnerable to both suboptimal and excessively high temperatures. Departing from the optimal temperature range can trigger significant heat stress, leading to a notable decrease in growth rate and grain yield. This decline primarily stems from a reduction in the success rate of seed formation and disturbances in various physiological processes.

It is anticipated that by 2050, approximately 45% of the maize production regions worldwide are expected to experience, on average, five days each year during the reproductive stage with maximum temperatures exceeding 35ºC (Gourdji et al 2013). This is of significant concern because even a mere 1ºC increase in the average seasonal temperature has the potential to reduce the economic yield of maize by a substantial margin, ranging from 3% to 13% (Izzaurralde et al 2011). Elevated temperatures during critical stages of development can also negatively affect the quality of maize grains (Siebers et al 2017). Heat stress affects the integrity of the plasma membrane functioning of mitochondria and chloroplast, which further results in the over-accumulation of reactive oxygen species. The activation of a signal cascade subsequently induces the transcription of heat shock proteins. The denaturation and accumulation of misfolded or unfolded proteins generate cell toxicity, leading to death. Therefore, developing maize cultivars with significant heat tolerance is urgently required. (El-Sappah et al 2022). Tassel blast in maize is vital for successful pollination and grain yield, while monitoring leaf firing serves as an indicator of plant stress. The occurrence of tassel blast and leaf firing directly influences grain yield in maize, reflecting the intricate relationship between plant health and productivity.

The purpose of this study was to assess genetic parameters within a 7x7 half diallel cross and evaluate various forms of heterosis, including mid-parent heterosis, heterobeltiosis and economic heterosis, concerning grain yield in spring maize. Additionally, the research aimed to identify the superior heat tolerant germplasm among the maize genotypes under investigation.

MATERIAL AND METHODS

Experimental location: Prayagraj, SE Uttar Pradesh, India, features a subtropical climate with scorching summers (up to 48°C), chilly winters, 983 mm yearly rain (July-October), sandy loam soil, low organic carbon, nitrogen, phosphorus, and potash. The location coordinate is 25.409459° N latitude and 81.851543° E longitude.

Parental materials: The experiment involved seven parent maize lines, namely MILCT-2092 (P1), MILC-43 (P2), MILC-2093 (P3), MILC-2091/A (P4), MILV-1098 (P5), MILC-2050 (P6), and MILCT-145F (P7) which were sourced from the Directorate of Research, Sam Higginbottom University of Agriculture Technology and Sciences, Prayagraj. The research was conducted at Naini Agricultural Institute, Sam Higginbottom University of Agriculture, Technology and Sciences, Prayagraj (Uttar Pradesh) during the spring of 2023.

Development of hybrids: In the previous *rabi* season of 2022, a set of experimental maize hybrids were developed using a 7x7 half diallel mating design. This resulted in a total of 21 maize hybrids, denoted as P1×P2, P1×P3, P1×P4, P1×P5, P1×P6, P1×P7, P2×P3, P2×P4, P2×P5, P2×P6, P2×P7, P3×P4, P3×P5, P3×P6, P3×P7, P4×P5, P4×P6, P4×P7, P5×P6, P5×P7 and P6×P7. These hybrids were generated through diallel mating involving seven carefully chosen parental genotypes during the Rabi season of 2022. The seeds of the 21 F1 hybrids, along with the initial seven parent lines and SHIATS Makka-3 (check), were gathered and stored in the Department of Genetics and Plant Breeding at Naini Agricultural Institute, Sam Higginbottom University of Agriculture Technology and Sciences, situated in Prayagraj, Uttar Pradesh. These seeds were subsequently stored and then cultivated once more during the scheduled spring season, which was from February to June in the year 2023.

Evaluation of experimental materials: During spring 2023, study was conducted in F1 maize populations, their seven parent plants and the check in an experimental field, with three replications. Plant characteristics were assessed, encompassing the timing of tassel and silk emergence, the duration between these events, tassel blast percentage, plant height, plant girth, leaf length and width, leaf firing extent, and days to maturity. Moreover, the chlorophyll content, indicative of plant health, was also examined. Following the harvest, measurements were collected for cob length, cob girth, cob weight, number of grain rows per cob, number of grains in each row, total grain count per cob, shank weight, seed index, and grain yield per plant. To gather this data, observations were made on five healthy maize plants within each replication, except for specific traits such as tassel and silk emergence, tassel blast, leaf firing and days to maturity, which were recorded for each plot.

Statistical analysis: The collected data were analysed using the statistical software TNAUSTAT (Nadarajan et al 2016), to estimate various genetic parameters. The software calculated three types of heterosis: mid-parent heterosis (MPH), heterobeltiosis (HB), and economic heterosis (EH). For the computation of standard heterosis, the high-yielding variety SHIATS Makka-3 was chosen as the standard check. Heterosis was assessed using three formulas: mid-parent heterosis (MPH), heterobeltiosis (HB), and economic heterosis (EH), which respectively measure the difference between F1 hybrid and parental means, the better parental mean, and a high-yielding standard check.

RESULTS AND DISCUSSION

Analysis of characters in maize: The mean sum of squares for all characteristics in the hybrid group showed significance (Table 1). Among the parent group, the mean sum of squares for all characteristics was significant, except for leaf width and days to maturity. When comparing the mean sum of squares between parents and hybrids, most characteristics exhibited statistical significance, except for chlorophyll content, number of grain rows per cob and seed index. These findings suggest that the parents displayed significant variability, and there was a substantial amount of diversity among the hybrids. Furthermore, the presence of heterosis was observed in most of the characteristics studied in the hybrids. Similar results were reported by Patil et al (2016) and Jebaraj et al (2023). The outcome indicated variations in the performances of the crosses, which were also compared to their respective parent plants. The level of diversity within the population is sufficient to facilitate the process of selection and the development of heterotic combinations by employing a diverse range of parent plants.

Mid-parent heterosis values of hybrid varieties: Among the twenty-one hybrids that were analysed for grain yield, the top five performing hybrids were P1xP7, P5xP6, P2xP6, P3xP5 and P1xP5 in decreasing order (Table 2). The best performing hybrids ranged in value from 66.78% in P7xP1 to 32.67% in P1xP5.

Heterobeltiosis values of hybrid varieties: The hybrids P2xP6, P5xP6, P3xP5, P4xP5, and P1xP7 displayed significant levels of heterobeltiosis for grain yield in decreasing order (Table 3). Particularly noteworthy, heterobeltiosis values were highly significant for genotypes P2xP6 and P5xP6. Values ranged from 44.98% in P2xP6 to 19.23% in P1xP7.

Economic heterosis values of hybrid varieties: Two hybrids, P1xP7 with 19.79% and P2xP6 with 18.5% outperformed the control variety SHIATS Makka-3 in terms of grain yield (Table 4). For traits such as days to 50% tasselling, days to 50% silking, anthesis silking interval, days to maturity, plant height, leaf firing, and tassel blast, a significant number of the crosses that showed pronounced negative heterosis, held a dominant influence. On the contrary, for other traits like plant girth, leaf length, leaf width, cob length, cob girth, chlorophyll content, cob weight, number of grain rows per cob, number of grains per row, number of grains per cob, shank weight, seed index and grain yield per plant, the majority of the crosses displayed significant positive heterosis. This indicates that, in the case of these traits, genes with positive effects held dominance.

Heat tolerance of maize hybrids: The hybrid P2xP6 exhibited substantial negative mid-parent, heterobeltiosis and economic heterosis values for leaf firing, while the hybrid P1xP7 demonstrated similarly significant negative mid-parent and economic heterosis values for tassel blast. These findings suggest that these two genotypes P1x P7 and P2xP6 possess heat tolerance and high yielding potential. Upon comparing all three types of heterosis for both grain yield per plant and heat tolerance, genotypes P1xP7 and P2xP6 performed better than the other genotypes. Similar findings were reported by Dash et al (2020) and Talekar et al (2021). The significant positive value of heterosis suggests a wide genetic divergence between the parents. The presence of both significantly positive and significantly negative values of heterosis in specific hybrid combinations highlights variations in gene action attributed to the genetic makeup of the parents. This is of paramount importance for identifying superior crossbreeding combinations. The results reveal a connection between heterosis, grain yield, and heat tolerance in the studied hybrids. The hybrids displaying substantial heterosis demonstrate enhanced grain yield, particularly under heat stress conditions. This correlation highlights the potential synergy between heterosis and heat tolerance, suggesting avenues for developing resilient crop varieties to mitigate the impacts of climate change on agricultural productivity.

Table 1. Analysis of variance of characters in maize

Characters	Mean sum of squares					
	Replicates	Treatments	Parents	Hybrids	Parents Vs. Hybrids	Error
DF	[2]	[27]	[6]	[20]	[1]	[54]
D50T	0.3	13.17 **	28 **	7.88 **	30.04 **	2.32
D50S	0.58	13.13 **	29.21 **	7.43 **	30.73 **	2.36
ASI	0.32	0.53 **	0.49 **	0.50 **	1.29 **	0.25
TB	0.1	35.78 **	24.41 **	39.96 **	20.51 **	12.3
PH	64.99	1019.39 **	356.94 **	855.43 **	8273.20 **	527.42
PG	0.28	0.45 **	0.24 **	0.44 **	1.97 **	0.24
LL	17.7	142.64 **	119.54 **	114.31 **	847.73 **	71.68
LW	0.33	1.11 **	0.53	1.17 **	3.50 **	0.52
CC	0.77	5.69 **	8.88 **	4.99 **	0.61	2.7
LF	1.8	106.68 **	118.79 **	103.69 **	93.87 **	41.08
DM	0.11	6.44 **	3.22	3.95 **	75.57 **	3.1
CL	0.5	11.81 **	15.48 **	6.66 **	92.77 **	5.06
CG	0.27	1.89 **	1.58 *	1.94 **	2.65 *	0.96
CW	175.05	902.39 **	844.33 **	494.01 **	9418.45 **	352.8
NGRC	0.32	4.51 **	4.05 **	4.81 **	1.3	2.27
NGR	1.41	54.49 **	90.64 **	38.57 **	156.04 **	22.62
NGC	64.12	10369.31 **	14997.03 **	6985.14 **	50286.41 **	4114.73
SW	2.07	21.65 **	15.98 **	22.56 **	37.49 **	9.06
SI	4.26	6.05 **	10.53 **	5.00 **	0.18	3.41
GY	0.48	642.88 **	679.69 **	590.59 **	1467.89 **	268.79

* Significant at 5% and ** Significant at 1% respectively

D50T- Days to 50% Tasselling, D50S- Days to 50% silking, ASI-Anthesis silking interval, TB- Tassel blast, PH- Plant height, PG- Plant girth, LL- Leaf length, LW- Leaf width, CC- Chlorophyll content, LF- Leaf firing, DM- Days to maturity, CL- Cob length, CG- Cob girth, CW- Cob weight, NGRC- Number of grain rows per cob, NGR- Number of grains per row, NGC- Number of grains per cob, SW- Shank weight, SI- Seed index and GY- Grain yield per plant

Table 2. Mid-parent heterosis values of hybrid varieties

Genotype	P1xP7	P5xP6	P2xP6	P3xP5	P1xP5
D50T	-2.56	-2.59	-1.42	0.5	-2.13
D50S	-2.7	-2.73	-1.83	0.49	-2.52
ASI	-33.33**	-14.29	-17.65*	-20*	-20*
DM	-3.17**	-0.89	-2.65**	1.26	-1.42
LF	35.26**	15.48*	-26.33**	-17.19*	8.65
TB	-54.17**	33.93**	49.14**	2.82	2.82
PH	20.46**	20.11**	18.04*	8.26	14.71
PG	4.97	3.18	10.06*	14.75**	-0.76
LL	16.83**	11.53	12.68*	-0.05	1.57
LW	8.19	12.86*	32.6**	11.53*	13.13*
CL	44.64**	16.48*	40.72**	11.46	39.95**
CG	24.16**	13.07**	9.8*	0.79	13.48**
CC	-8.31**	-2.92	4.85*	-0.88	3.04
CW	72.7**	35.92**	62.61**	19.48*	51.38**
NGRC	13.67*	15.23**	3.49	-2.16	7.85
NGR	34.77**	33.56**	38.4**	14.78	43.46**
NGC	38.68**	26.81**	27.89**	18.32*	53.98**
SW	21.57*	13.13	37.11**	-11.76	50.79**
SI	17.54**	6.11	10.32	13.06*	8.42
GY	66.78**	52.64**	42.28**	36.49**	32.67*

* Significant at 5% and ** Significant at 1% respectively

Table 3. The value of heterobeltiosis of the hybrid varieties

Genotype	P2xP6	P5xP6	P3xP5	P4xP5	P1xP7
D50T	-5.02 **	-5.48 **	-2.91	-0.48	-3.69 *
D50S	-5.29 **	-5.73	-2.82	-0.47	-3.57 *
ASI	-22.22 *	-25.00 *	-33.33 **	-14.29	-33.33 **
DM	-2.82 **	-1.77	1.08	-4.58 **	-3.17 **
LF	-27.01**	8.04	-26.05 **	-47.01 **	30.00 **
TB	39.20 **	29.28 **	-2.45	-32.57 **	-56.22 **
PH	10.92	13.89	4.43	8.38	13.05
PG	8	1.49	11.99 *	10.9	1.73
LL	8.67	4.33	-7.18	23.99**	15.14*
LW	31.87**	6.97	3.48	3.98	4.81
CL	19.23*	15.16*	8.1	4.4	18.12*
CG	9.8	10.95*	-4.19	-5.07	20.87**
CC	4.85*	-3.37	-1.9	6.58**	-12.49**
CW	40.82**	35.61**	9.46	8.51	42.08**
NGRC	-11.10 **	11.67	-2.71	-11.90 **	8.81
NGR	28.46**	28.86**	10.01	9.24	1.83
NGC	25.83 **	16.58 *	11.2	19.16 **	5.83
SW	14.7	-2.51	-21.86 *	18.16	6.5
SI	6.92	0.56	12.94	-16.89 **	4.83
GY	44.98 **	42.63**	27.92*	23*	19.23*

* Significant at 5% and ** Significant at 1% respectively

Table 4. Economic heterosis values exhibited by high-performing hybrids

Genotype	P1xP7	P2xP6
D50T	-2.56*	-4.15*
D50S	-3.57*	-5.29**
ASI	-25*	-12.5
DM	-4.51**	-4.17**
LF	31.86**	-27.46**
TB	-19.87*	67.31**
PH	-2.37	-9.06
PG	-4.54	-2.11
LL	4.44	-8.1
LW	9.55	20.6**
CL	10.1	2.33
CG	4.27	2.39
CC	2.42	10.49**
CW	-5.46	-3.24
NGRC	-3.89	1.83
NGR	10.3	0.52
NGC	6.89	5.8
SW	-21.37**	-13.81*
SI	-2.17	-11.18*
GY	19.79*	18.5*

* Significant at 5% and ** Significant at 1% respectively

D50T- Days to 50% Tasselling, D50S- Days to 50% silking, ASI-Anthesis silking interval, DM- Days to maturity, LF- Leaf firing, TB- Tassel blast, PH- Pant height, PG- Plant girth, LL- Leaf length, LW- Leaf width, CL- Cob length, CG-Cob girth, CC-Chlorophyll content, CW- Cob weight, NGRC- Number of grain rows per cob, NGR- Number of grains per row, NGC- Number of grains per cob, SW- Shank weight, SI-Seed index and GY- Grain yield per plant

CONCLUSION

Based on all three forms of heterosis concerning both grain yield per plant and heat tolerance, genotypes P1xP7 and P2xP6 excelled in comparison to the other genotypes. The connection between heat tolerance and grain yield underscores the importance of breeding and selecting maize varieties that exhibit strong heat tolerance. By doing so, farmers can mitigate the negative effects of heat stress and achieve more consistent and higher grain yields, even in regions prone to heat waves and rising temperatures due to climate change. This interplay between heat tolerance and grain yield in maize is a key consideration for ensuring food security and sustainable agriculture in the face of changing environmental conditions.

AUTHORS CONTRIBUTION

Anu George, as the first author, executed the research. My advisors, Shailesh Marker and Vaidurya Pratap Sahi, provided guidance and supervision. Co-authors Reuben James Melvin and M.L. Sharin helped in data collection and recording. Grateful to my senior, Venkata Krishna Thupakula, for his support in fieldwork and creating the crosses.

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