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RESEARCH ARTICLE

Combining ability, heterosis and variability studies of bread wheat (*Triticum aestivum* L.) for grain yield and its attributes under moisture stress conditions

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Abstract

The study was conducted to identify promising parents and hybrids of bread wheat (*Triticum aestivum* L.) for grain yield and its related traits under moisture stress conditions, given the increasing need for wheat to tolerate limited water due to climate change patterns. In *rabi* 2023–24, six genotypes (RAJ3765, WH730, NIAW3170, MP1378, MACS6768 and UAS375) were crossed in a 6 × 6 half-diallel mating design, resulting in 15 F_1 hybrids. During *rabi* 2024–25, these hybrids, along with two check varieties (UAS347 and HD3090), were evaluated using a Randomised Complete Block Design (RCBD) with three replications under restricted irrigation given at 25 and 45 DAS. Analysis of variance (ANOVA) indicated significant differences among genotypes ($p < 0.05$), demonstrating the presence of considerable genetic variability. WH730, RAJ3765 and NIAW3170 were identified as good general combiners, while NIAW3170 × MACS6768 and RAJ3765 × MP1378 showed high specific combining ability, superior heterosis and better yield performance. High heritability and genetic advance for test weight and grain yield, suggesting scope for direct selection. Grain yield showed positive correlations with spike length, NDVI, SPAD, grains per spike and test weight, while a negative correlation with flowering time suggests early flowering is beneficial under moisture stress. Path analysis highlighted test weight, NDVI and SPAD as the key direct contributors. PCA identified NIAW3170 × MACS6768 as the most promising hybrid. Hybrids like NIAW3170 × MACS6768 and RAJ3765 × MP1378 show strong potential for developing high-yielding and moisture-stress-tolerant wheat varieties.

Keywords: bread wheat; combining ability; half diallel; heterosis; moisture stress; variability

Introduction

Wheat (*Triticum aestivum* L.) is one of the most vital staple food crops worldwide, standing as a cornerstone of global food security. It provides a major source of energy along with essential nutrients, including copper, iron, phosphorus, magnesium, zinc, carbohydrates, protein, fibre and vitamins (1). As the global population is expected to reach nearly 10 billion by 2050, the demand for wheat is estimated to rise by more than 130 million metric tons every year (2). This makes it essential to boost wheat production in the coming decades. However, climate change is bringing more frequent and severe environmental challenges. Environmental stresses such as moisture stress, heat and salinity are the biggest threats among various stresses, causing yield losses of over 50 % in major crops worldwide (3). Within these stresses, moisture stress is the greatest constraint for the wheat crop. Limited water supply leads to problems like reduced photosynthesis, spikelet sterility and ultimately poor grain yield (4). So, crops cannot reach their full potential under such conditions; improving stress tolerance is one of the simplest and most practical ways to secure productivity (5).

Although advanced tools like genome editing, marker-assisted selection, genomic selection, mutation breeding and genetic engineering are available, conventional hybridisation and evaluation of crosses remain the backbone of crop improvement (6). A strong set of initial breeding material is always the first requirement for any breeding programme. In the present study, six bread wheat genotypes (RAJ3765, WH730, NIAW3170, MP1378, MACS6768 and UAS375) were utilised in a half-diallel mating design and the resulting F_1 hybrids were evaluated for genetic variability, combining ability and heterotic performance, with two check varieties (UAS347 and HD3090) included for comparison. The primary objective was to identify superior parental combinations and develop promising lines capable of withstanding moisture stress, thereby contributing to the development of improved wheat varieties for future cultivation.

Materials and Methods

The field experiment was carried out during the rabi seasons of 2023–24 and 2024–25 at the All India Coordinated Research Project on wheat, the main agricultural research Station, University of Agricultural Sciences, Dharwad. This location lies in the Northern Transitional zone (Zone 8) of Karnataka and it is characterised by black soils (Vertisols).

For the study, six bread wheat genotypes, viz., RAJ3765 (P1), WH730 (P2), NIAW3170 (P3), MP1378 (P4), MACS6768 (P5) and UAS375 (P6) were crossed in a half-diallel mating design during rabi 2023–24. A total of 15 F_1 hybrids were generated and subsequently evaluated along with two check varieties (UAS347 and HD3090) in the rabi 2024–25 season in a randomized complete block design (RCBD) with three replications under restricted irrigation which was applied at 25 and 45 days after sowing with a volume of 1.5 L/plot/irrigation, corresponding to crown root initiation and heading stages, respectively.

Each entry was sown with a row spacing of 20 cm and the plot consisted of six rows, every 3 meters in length. All genotypes were maintained at the necessary agronomic levels. Observations were noted on a range of traits including days to 50 % flowering (DFF), plant height (PH in cm), days to maturity (DM), spike length (SL in cm), number of productive tillers per plant, NDVI at grain filling stage (NDVI), chlorophyll content estimation by SPAD meter, test weight (TW in g), number of grains per spike (NGPS) and grain yield per hectare (GYPH in kg/ha).

Six bread wheat genotypes with distinct characteristics were utilised as parents in the hybridisation program. RAJ3765 is a high-yielding cultivar tolerant to temperature stress and rust, well-suited for late sowing conditions. WH730 performs efficiently under moisture stress and exhibits heat tolerance during the grain filling stage. NIAW3170 is a high-yielding type suitable for restricted irrigation, with resistance to both brown and black rust. MP1378 is rich in zinc and iron content (>40 ppm) and adapts well under limited irrigation. MACS6768 has superior protein, zinc and iron (>40 ppm) content and is recommended for irrigated environments. UAS375 is a high-yielding variety with good protein content, suitable for both rainfed and restricted irrigation conditions. Since heat tolerance and moisture stress tolerance are positively associated, the selected parents provided a broad genetic base for crossing. For comparison, two check varieties were included, such as UAS347, a rainfed cultivar suitable for early sowing and HD3090, a late-sown variety with moderate heat tolerance.

Statistical analysis

Heterosis, general combining ability (GCA) and specific combining ability (SCA) were estimated using Griffing's Method II, Model I (1956), with analyses performed in TNAUSTAT and MS Excel. Statistical analyses were conducted using R software (version 4.5.1) with the 'metan' and 'variability' packages for correlation, genetic variability and path analyses; Principal Component Analysis (PCA) was performed using GRAPES software (version 1.1.0).

Results

The ANOVA revealed that differences among genotypes were significant (F = 21, df = 20, p = 0.05, 0.01), supporting the presence of genetic variability (Table 1). Parental lines differed significantly for several traits, while hybrids also showed wide variation. Parents vs. hybrids exhibited significant differences for traits such as DFF, DM, SL, PTP, NDVI, TW and GYPH. Replication effects were largely non-significant.

Variability, heritability and genetic advance

The small difference observed between GCV and PCV for most traits (Table 2) suggests minimal environmental impact. For SL, PTP and NGPS, the relatively higher gap indicates a noticeable environmental influence. TW and GYPH recorded high heritability (>60) and high genetic advance (>20). SL and NGPS showed moderate heritability (30–60) and moderate genetic advance (10–20). DFF, PTP and NDVI showed high heritability but moderate genetic advance. DM and SPAD exhibited high heritability with low genetic advance (<10).

General combining ability and specific combining ability

Analysis of variance (Table 3) showed significant *GCA* and *SCA* effects for all traits. Parent WH730 showed the highest significant GCA effects for DFF, DM, PH, NDVI, SPAD, NGPS, TW and GYPH (Table 4). Among hybrids, NIAW3170 × MACS6768 displayed the highest significant sca effects for DFF, DM, SL, NDVI, SPAD, NGPS, TW and GYPH (Table 5). The SCA/GCA ratio was less than one for most traits except plant height (Table 6).

Per se performance

WH730 showed good overall per se performance among the parents (Table 7). Hybrids RAJ3765 × MP1378, WH730 × NIAW3170, WH730 × MP1378, WH730 × UAS375, NIAW3170 × MACS6768 and MP1378 × MACS6768 recorded higher per se performance than the best check (UAS347) for GYPH. Almost all parents and hybrids showed early flowering.

Table 1. Analysis of variance for morphological, physiological, yield and yield-related traits in the 6 × 6 half diallel population

Characters	Treatments	Replications	Parents	Hybrids	Parent vs. Hybrids	Error	Total
df	**20**	**2**	**5**	**14**	**1**	**40**	**62**
DFF	0.59***	0.59	13.40***	55.31***	52.1***	0.29	21.95
DM	22.37***	0.09	14.48***	25.06***	24.28**	0.54	11.18
PH	26.53*	0.49	31.30	24.56	30.25	11.95	18.78
SL	2.38**	0.02	0.95	3.06***	0.04**	0.64	1.47
PTP	0.58**	0.12	0.50*	0.64**	0.16*	0.14	0.35
NDVI	0.003***	0.0095	0.002***	0.004***	0.0012*	0.0002	0.002
SPAD	5.5025**	7.0876	2.0853	7.0229**	1.5873	1.9135	1.764
NGPS	34.844**	1.7609	25.366*	39.743***	13.644	8.3204	21.09
TW	57.004***	0.6931	9.815**	77.807***	1.7089	4.5906	30.06
GYPH	269334.9***	69540.02	39322.7**	370652.4***	949.50*	21036.8	143340.8

*,**,***significant at 5, 1 and 0.1 % respectively. **DFF** – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length, **PTP** – Productive tillers per plant, **NDVI** – Normalized difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight, **GYPH** – Grain yield per hectare

Table 2. Genetic variability parameters for morphological, physiological, yield and yield related traits in the 6 × 6 half diallel population

Trait	Max	Min	Mean	GCV	PCV	h_{bs}(%)	GA	GAM (%)
DFF	69.00	52.00	61.00	7.87	7.92	98.84	9.86	16.12
DM	102.00	90.00	96.00	3.47	3.56	95.07	6.69	6.97
PH	99.67	82.00	90.97	2.63	4.73	30.89	2.74	3.01
SL	12.50	7.00	9.53	9.43	12.45	57.42	1.40	14.72
PTP	6.00	4.00	5.00	10.00	12.79	61.04	0.74	16.09
NDVI	0.66	0.49	0.59	7.21	7.60	90.00	0.08	14.09
SPAD	48.76	41.22	45.11	4.82	4.98	93.87	4.34	9.63
NGPS	49.00	34.00	42.00	8.19	10.58	59.97	5.59	13.07
TW	54.13	32.31	40.27	12.21	13.28	84.43	9.30	23.11
GYPH	3975.00	2306.25	3087.78	10.98	11.89	85.30	644.04	20.89

DFF – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length, **PTP** – Productive tillers per plant, **NDVI** – Normalized difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight, **GYPH** – Grain yield per hectare

Table 3. Analysis of variance for combining ability of morphological, physiological, yield and yield-related traits in the 6 × 6 half diallel population

Characters	GCA effects	SCA effects	Error
df	**5**	**15**	**40**
DFF	24.8083***	215179***	0.1475
DM	23.2000***	7.1857***	0.2728
PH	33.2019**	6.6241	5.9759
SL	0.3855	1.4631**	0.3207
PTP	0.4458*	0.2958**	0.0976
NDVI	0.0035***	0.0015***	0.0001
SPAD	4.2058*	2.2664*	0.9567
NGPS	30.1576*	13.1776**	4.1597
TW	28.8913***	28.3654***	2.2947
GYPH	119496.7891***	139735.8906***	10521.1816

DFF – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length / Shoot length (depending on crop; most often Spike Length), **PTP** – Productive tillers per plant, **NDVI** – Normalized difference vegetation index, **SPAD** – Soil plant analysis development (Chlorophyll meter reading), **NGPS** – Number of grains per spike, **TW** – Test weight (1000-grain weight or 100-seed weight, depending on crop), **GYPH** – Grain yield per hectare

Table 4. General combining ability effects of parents in the 6 × 6 half diallel population

	DFF	DM	PH	SL	PTP	NDVI	SPAD	NGPS	TW	GYPH
RAJ3765	1.85**	0.56	3.82**	-0.03	0.29**	0.03**	-0.03	0.23	-1.2*	-34.17
WH730	-2.40**	-2.69*	-2.02**	-0.13	0.17	-0.04**	1.22**	2.91**	2.78**	162.9**
NIAW3170	-1.27**	-0.88	0.16	0.4*	0.1	0.01	-0.91**	-0.3	0.75	12.58
MP1378	-0.71**	0.5	0.08	0.09	-0.33**	0.001	0.11	-1.32	0.73	23.15
MACS6768	0.54**	0.06	-1.03	-0.13	-0.02	0.00	0.84**	1.15	0.27	45.77
UAS375	1.98**	2.44	-1.01	-0.2	-0.21	0.01	-1.23**	-2.67**	-2.78**	-210.2**
C. D at 95 %	0.3187	0.4331	2.0280	0.4698	0.2238	0.0084	0.8115	1.6922	1.2569	85.0884
C. D at 99 %	0.5000	0.6794	3.1812	0.7369	0.3510	0.0133	1.2729	2.6543	1.9716	133.467

DFF – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length / Shoot length (depending on crop; most often Spike Length), **PTP** – Productive tillers per plant, **NDVI** – Normalized difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight, **GYPH** – Grain yield per hectare

Table 5. Specific combining ability effects of crosses in the 6 × 6 half diallel population

	DFF	DM	PH	SL	PTP	NDVI	SPAD	NGPS	TW	GYPH
RAJ3765× WH730	4.80**	0.91*	1.61	-0.13	0.88**	0.08**	-0.40	-4.24*	-3.15*	-167.42*
RAJ3765× NIAW3170	-4.32**	-0.40	-1.32	1.30**	0.44	-0.03**	-2.38**	0.92	3.56**	32.89
RAJ3765× MP1378	0.12	0.72	0.10	0.50	-0.63*	0.01	2.43**	5.85**	6.30**	606.33**
RAJ3765× MACS6768	-1.13**	-0.84*	4.71*	0.12	-0.44	-0.01	-1.61**	-5.47**	-5.53**	-412.79**
RAJ3765× UAS375	3.43**	3.29**	2.85	-1.25**	-0.75**	-0.01	-0.83**	-1.85	-3.59**	-391.29**
WH730 × NIAW3170	-0.07	-1.15**	-0.49	0.19	0.06	0.00	1.58**	0.89	2.85*	129.83
WH730× MP1378	-5.63**	-1.03*	-2.90	-1.35**	0.00	-0.05**	0.72*	-0.98	5.00**	243.77**
WH730× MACS6768	-4.88**	-3.09**	0.54	-2.28**	-0.31	-0.05**	-0.09	-0.50	-6.29**	-334.86**
WH730× UAS375	-0.32	0.04	1.19	1.64**	-0.63*	-0.01	2.47**	4.27**	5.10**	420.64**
NIAW3170 ×MP1378	-1.76**	-5.34**	0.20	-1.28**	0.06	-0.00	-2.16**	-3.72*	-8.11**	-464.92**
NIAW3170× MACS6768	-8.01**	-1.90**	-1.72	1.59**	-0.25	-0.05**	3.12**	5.11**	9.69**	649.46**
NIAW3170× UAS375	6.55**	0.72	1.34	-1.13*	0.44	0.04**	-0.67*	-0.42	-1.84	-19.54
MP1378× MACS6768	4.93**	3.72**	1.67	0.75	0.69**	0.03**	2.20**	2.14	3.63**	192.39*
MP1378× UAS375	-3.01**	-0.65	-1.75	0.12	-0.13	-0.01	-1.59**	-4.89**	-4.22**	-414.61**
MACS6768× UAS375	-1.26**	-2.21**	2.03	0.89*	-0.44	0.04**	-3.20**	-2.51	-1.49	-114.73
CD at 95 %	0.7259	0.9864	4.6184	1.0698	0.5096	0.0193	1.8480	3.8535	2.8624	193.7680
CD at 99 %	1.0035	1.3637	6.3850	1.4790	0.7045	0.0267	2.5549	5.3275	3.9572	267.8829

*,**,***significant at 5, 1 and 0.1 % respectively. **DFF** – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length, **PTP** – Productive tillers per plant, **NDVI** – Normalized difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight (1000-grain weight or 100-seed weight, depending on crop), **GYPH** – Grain yield per hectare

Table 6. Variance of genetic components of morphological, physiological, yield and yield-related traits in the 6 × 6 half diallel population

Genetic components	σ_{gca}	σ_{sca}	GCA/SCA ratio
DFF	3.0826	21.3704	0.144246
DM	2.8659	6.9129	0.414573
PH	3.4033	0.6482	5.250386
SL	0.0081	1.1425	0.00709
PTP	0.0435	0.1982	0.219475
NDVI	0.0004	0.0012	0.3222
SPAD	0.4061	1.3096	0.31010
NGPS	3.2497	9.0178	0.360365
TW	3.3246	26.0707	0.127522
GYPH	13621.95	129214.7	0.105421

DFF – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length; most often Spike Length), **PTP** – Productive tillers per plant, **NDVI** – Normalized difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight, **GYPH** – Grain yield per hectare, σ^2GCA – Genetic variance due to general combining ability, σ^2SCA – Genetic variance due to specific combining ability

Table 7. *Per se* performance of the parents and their crosses of 6 × 6 half diallel population

	DFF	DM	PH	SL	PTP	NDVI	SPAD	NGPS	TW	GYPH
RAJ3765 (P1)	63	95	94.66	9.2	5	0.62	46.55	46	39.26	3193
WH730 (P2)	59	93	87.00	10.25	5	0.525	45.60	49	44.25	3275
NIAW3170 (P3)	62	98	92.33	10	4	0.615	44.00	41	38.87	2956.5
MP1378 (P4)	62	98	92.49	10.35	4	0.6	44.9	41	40.62	3060
MACS6768 (P5)	67	98	85.33	8.75	5	0.605	46.65	46	39.92	2956.5
UAS375 (P6)	62	100	86.16	9	5	0.57	44.15	40	37.92	2934.25
UAS347 (check 1)	69	100	89.83	9.9	5	0.595	46.10	44	39.05	3281
HD3090 (check 2)	62	99	91.40	8.95	4	0.64	42.90	40	37.39	2868.75
RAJ3765 × WH730	65	95	94.42	9.25	6	0.655	45.85	42	38.90	3056.25
RAJ3765 × NIAW3170	57	95	93.66	11.2	6	0.585	42.50	44	43.56	3106.25
RAJ3765 × MP1378	62	98	95.00	10.1	4	0.625	47.25	48	46.28	3690.5
RAJ3765 × MACS6768	62	96	98.5	9.5	4	0.6	43.85	39	33.46	2694
RAJ3765 × UAS375	68	102	96.67	8.05	4	0.605	42.85	39	32.89	2459.5
WH730 × NIAW3170	57	91	88.66	10	5	0.555	47.05	47	46.84	3400
WH730 × MP1378	52	93	86.16	8.15	5	0.5	47.00	44	48.97	3525.12
WH730 × MACS6768	54	90	88.5	7	4	0.495	46.45	46	36.68	2968.75
WH730 × UAS375	60	96	89.16	10.85	4	0.545	47.80	47	45.56	3468.5
NIAW3170 × MP1378	57	90	91.45	8.75	4	0.59	41.70	38	33.83	2665.62
NIAW3170 × MACS6768	52	93	88.42	11.4	4	0.54	48.35	49	50.63	3803
NIAW3170 × UAS375	68	98	91.505	8.6	5	0.64	42.30	40	36.58	2878
MP1378 × MACS6768	66	100	91.72	10.25	5	0.615	48.30	47	44.54	3356.25
MP1378 × UAS375	59	98	88.33	9.55	4	0.585	42.30	34	34.18	2493.75
MACS6768 × UAS375	62	96	90.99	10.1	4	0.63	41.35	39	35.92	2816

DFF – Days to 50 % flowering, **DM –** Days to maturity, **PH –** Plant height, **SL –** Spike length, **PTP –** Productive tillers per plant, **NDVI –** Normalized difference vegetation index, **SPAD –** Soil plant analysis development, **NGPS –** Number of grains per spike, **TW –** Test weight, **GYPH –** Grain yield per hecta

Heterosis

The heterosis analysis presented in Tables 8-10 revealed considerable hybrid vigour across agronomic and physiological traits. Hybrids NIAW3170 × MACS6768, RAJ3765 × MP1378 and WH730 × MP1378 showed significant positive heterosis over the better parent for grain yield. These hybrids also outperformed standard checks. For flowering and maturity, hybrids NIAW3170 × MACS6768, WH730 × MP1378 and WH730 × MACS6768 displayed significant negative heterosis over checks. Yield-related traits such as NGPS, TW and PTP exhibited significant heterosis in WH730 × MP1378 and NIAW3170 × MACS6768.

Correlation and path analysis

Grain yield showed positive associations with SL, SPAD, NGPS and TW (Fig. 1). DFF and DM were negatively correlated with yield. Lower NDVI during grain filling was associated with higher yield. Thousand-grain weight exerted the strongest direct positive effect on yield (β =0.783), followed by SPAD (β =0.186) and NDVI at grain filling (β =0.148 Indirect effects of NGPS through TW (β = 0.548) and SPAD through TW (β = 0.513) were highest. The residual value was 0.0516.

Principal component analysis

Principal component analysis (PCA) identified ten components, with the first three explaining 76.54 % of the total variation (Table 11 & 12). PC1 contributed the most (45.44 %), followed by PC2 (20.23 %) and PC3 (10.86 %). Key traits influencing PC1 were TW, GYPH and NGPS, while DFF, NDVI and DM were dominant in PC2. PC3 was mainly explained by SL and PTP (Table 13) and these results are aligned with other studies (31, 32). The genotype–trait (G-T) biplot (Fig. 2) effectively distinguished both traits and genotypes. The hybrid NIAW3170 × MACS6768 (P3 × P5) aligned strongly with yield-contributing traits such as NGPS, TW, SPAD and GYPH. Conversely, RAJ3765 × UAS375 (P1 × P6) appeared on the opposite side of the major trait vectors. Hybrids like P1 × P3, P4 × P5 and most of the parents clustered near the origin.

Table 8. Mid-parent heterosis for the hybrids of a 6 × 6 half-diallel population

	DFF	DM	PH	SL	PTP	NDVI	SPAD	NGPS	TW	GYPH
RAJ3765× WH730	3.17**	-0.53	-0.26	-9.76	9.09	5.65*	-1.16	-14.69*	-12.14*	-6.67
RAJ3765× NIAW3170	-9.52**	-3.06**	-1.05	12	0.00	-5.65*	-10.03**	-4.37	10.97	-2.71
RAJ3765× MP1378	-1.59	-0.51	0.35	-2.42	-27.27**	0.81	-2.53*	4.15	13.92*	15.58**
RAJ3765× MACS6768	-7.46**	-2.55**	4.05	3.26	-18.18*	-3.23	-4.88**	-15.28*	-16.18**	-15.73**
RAJ3765× UAS375	7.94**	2.0*	2.12	-12.5	-27.27**	-2.42	-3.37**	-15.63*	-16.23**	-22.97**
WH730 × NIAW3170	-8.06**	-7.14**	-3.98	-2.44	0.00	-9.76**	3.52**	-5.31	5.83	3.83
WH730× MP1378	-16.13**	-5.61**	-6.85	-21.26*	-10.00	-16.67**	3.87**	-11.22	10.65*	7.63
WH730× MACS6768	-19.4**	-8.16**	1.72	-31.71**	-10.00	-18.18**	1.07	-5.2	-17.11**	-9.34*
WH730× UAS375	-3.23**	-4.5**	2.49	5.85	-20.00*	-4.39	4.77**	-3.27	2.94	5.91
NIAW3170 ×MP1378	-8.06**	-8.16**	-1.14	-15.46	0.00	-4.07	-5.35**	-8.53	-16.74**	12.88**
NIAW3170× MACS6768	-22.39**	-5.1**	-4.24	14	-10.00	-12.2**	3.37**	6.66	26.83**	18.96**
NIAW3170× UAS375	9.68**	-2.00*	-0.9	-14	0.00	4.07	-5.08**	-3.42	-5.88	-2.66
MP1378× MACS6768	-2.24*	2.04*	-0.84	-0.97	0.00	1.65	3.58**	-2.07	9.65	4.99
MP1378× UAS375	-4.84**	-2.00*	-4.51	-7.73	-20*	-2.5	-4.86**	-17.17*	-15.85**	-18.51**
MACS6768× UAS375	-7.46**	-4.00**	5.61	12.22	-20.00*	4.13	-10.87**	-15.17*	-10.01	-11.92*
S.E	0.543	0.738	3.457	0.800	0.441	0.014	0.5440	2.884	2.142	145.059
CD at 95 %	1.541	2.091	9.782	2.27	1.25	0.041	5.650	5.653	4.199	284.317
CD at 99 %	2.160	3.701	13.661	3.18	1.76	0.057	7.941	7.438	5.519	373.693

DFF – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length, **PTP** – Productive tillers per plant, **NDVI** – Normalized difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight, **GYPH** – Grain yield per hectare

Table 9. Standard heterosis for the hybrids of 6 × 6 half diallel population

	DFF		DM		PH		SL		PTP	
	UAS 347	HD 3090	UAS 347	HD 3090	UAS 347	HD 3090	UAS 347	HD 3090	UAS 347	HD 3090
RAJ3765× WH730	-5.80**	4.84**	-5.50**	-4.55**	5.10	3.30	-6.57	3.35	20.00*	50.00**
RAJ3765× NIAW3170	-17.39**	-8.06**	-5.00**	-4.04**	4.27	2.48	13.13	25.14*	10.00	37.50**
RAJ3765× MP1378	-10.14**	0.00	-2.50*	-1.52	5.76	3.94	2.02	12.85	-20.00*	0.00
RAJ3765× MACS6768	-10.14**	0.00	-4.50**	-3.54**	9.65*	7.77	-4.04	6.15	-10.00	12.50
RAJ3765× UAS375	-1.45	9.68**	2.00*	3.03**	7.61	5.77	-18.69*	-10.06	-20.00*	0.00
WH730 × NIAW3170	-17.39**	-8.06**	-9.00**	-8.08**	-1.30	-3.00	1.01	11.73	0.00	25.00*
WH730× MP1378	-24.64**	-16.13**	-7.50**	-6.57**	-4.08	-5.73	-17.68	-8.94	-10.00	12.50
WH730× MACS6768	-21.74**	-12.90**	-10.00**	-9.09**	-1.48	-3.17	-29.29**	-21.79*	-10.00	12.50
WH730× UAS375	-13.04**	-3.23**	-4.50**	-3.54**	-0.73	-2.44	9.60	21.23*	-20.00*	0.00
NIAW3170 ×MP1378	-17.39**	-8.0**	-10.00**	-9.09**	1.80	0.05	-11.62	-2.23	-10.00	12.50
NIAW3170× MACS6768	-24.64**	-16.13**	-7.00**	-6.06**	-1.57	-3.26	15.15	27.37**	-10.00	12.50
NIAW3170× UAS375	-1.45	9.68**	-2.00*	-1.01	1.86	0.11	-13.13	-3.91	0.00	25.00*
MP1378× MACS6768	-5.07**	5.65**	0.00	1.01	2.10	0.35	3.54	14.53	0.00	25.00*
MP1378× UAS375	-14.49**	-4.84**	-2.00*	-1.01	-1.67	-3.36	-3.54	6.70	-20.00*	0.00
MACS6768× UAS375	-10.14**	0.00	-4.00**	-3.03**	1.30	-0.44	2.02	12.85	-20.00*	0.00
S.E	0.363	0.363	0.618	0.618	2.471	2.471	0.589	0.589	0.306	0.306
CD at 95 %	1.090	1.090	1.853	1.853	7.411	7.411	1.767	1.767	0.918	0.918
CD at 99 %	1.502	1.502	2.553	2.553	10.207	10.207	2.434	2.434	1.264	1.264

DFF – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length, **PTP** – Productive tillers per plant, **NDVI** – Normalised difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight, **GYPH** – Grain yield per hectare.

Table 10. Standard heterosis for the other hybrids of 6 × 6 half diallel population

	NDVI		SPAD		NGPS		TW		GYPH	
	UAS 347	**HD 3090**	**UAS 347**	**HD 3090**	**UAS 347**	**HD 3090**	**UAS 347**	**HD 3090**	**UAS 347**	**HD 3090**
RAJ3765× WH730	10.08^{**}	2.34	0.66	6.67^{**}	-4.89	5.69	-0.44	4.00	-3.06	6.54
RAJ3765× NIAW3170	-1.68	-8.59^{**}	-0.51	5.43^{**}	-0.46	10.62	11.55	16.52^{*}	-1.47	8.28
RAJ3765× MP1378	5.04	-2.34	-9.43^{**}	-4.03^{**}	8.42	20.48^{*}	18.51^{**}	23.79^{**}	17.05^{**}	28.63**
RAJ3765× MACS6768	0.84	-6.25^{*}	3.21^{*}	9.36^{**}	-11.72	-1.90	-14.33^{*}	-10.51	-14.56^{*}	-6.10
RAJ3765× UAS375	1.68	-5.47^{*}	-3.96^{**}	1.77	-12.17	-2.40	-15.79^{*}	-12.04	-21.99^{**}	-14.27^{*}
WH730 × NIAW3170	-6.72^{*}	-13.28^{**}	-6.76^{**}	-1.19	5.57	17.32	19.93^{**}	25.27^{**}	7.85	18.53^{**}
WH730× MP1378	-15.97^{**}	-21.88^{**}	-1.60	4.27^{**}	-1.02	9.99	25.40^{**}	30.98^{**}	11.80^{*}	22.87^{**}
WH730× MACS6768	-16.81^{**}	-22.66^{**}	1.86	7.94^{**}	5.69	17.45	-6.07	-1.89	-5.84	3.49
WH730× UAS375	-8.40^{**}	-14.84^{**}	2.21	8.31^{**}	7.85	19.85^{*}	16.66^{*}	21.85^{**}	10.01	20.90^{**}
NIAW3170 ×MP1378	-0.84	-7.81^{**}	2.05	8.14^{**}	-14.56	-5.06	-13.38^{*}	-9.52	-15.45^{**}	-7.08
NIAW3170× MACS6768	-9.24^{**}	-15.63^{**}	3.09^{*}	9.24^{**}	11.15	23.51^{*}	29.64^{**}	35.41^{**}	20.62^{**}	32.55^{**}
NIAW3170× UAS375	7.56^{**}	0.00	-5.62^{**}	0.01	-10.13	-0.13	-6.31	-2.14	-8.72	0.31
MP1378× MACS6768	3.36	-3.91	-8.66^{**}	-3.21^{*}	2.05	13.40	14.07^{*}	19.15^{**}	6.45	16.99^{**}
MP1378× UAS375	-1.68	-8.59^{**}	-4.36^{**}	10.59^{**}	-22.64^{**}	-14.03	-12.46	-8.56	-20.92^{**}	-13.09*
MACS6768× UAS375	5.88^{*}	-1.56	-8.34^{**}	-2.87^{*}	-11.60	-1.77	-8.01	-3.92	-10.69^{*}	-1.85
S.E	0.010	0.010	0.3723	0.3723	2.320	2.320	1.634	1.634	111.466	111.466
CD at 95 %	0.030	0.030	1.1162	1.1162	6.958	6.958	4.899	4.899	334.191	334.191
CD at 99 %	0.041	0.041	1.5375	1.5375	9.584	9.584	6.749	6.749	460.301	460.301

NDVI – Normalised difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight, **GYPH** – Grain yield per hectare

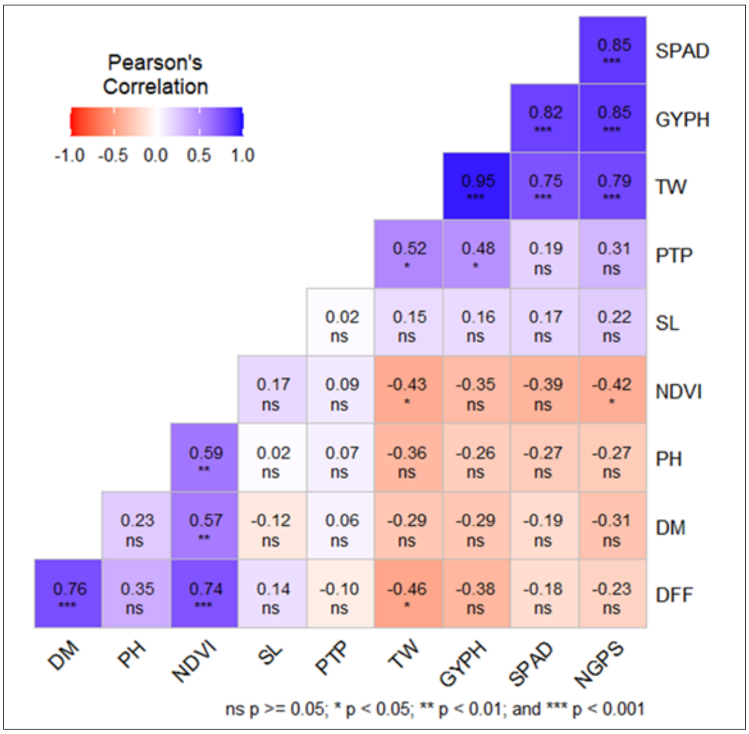

Fig. 1. Heat map depicting the correlation of grain yield and its attributing traits in a 6 × 6 half diallel population.

Table 11. Phenotypic path analysis for morphological, physiological, yield and its attributing traits in the 6 × 6 half diallel population

	DFF	DM	PH	SL	PTP	NDVI_2	SPAD	NGPS	TW
DFF	**0.06653**	-0.05595	-0.00487	-0.00619	-0.00125	0.09731	-0.02899	-0.02759	-0.3652
DM	0.04902	**-0.07594**	-0.00239	-0.00145	0.00139	0.07089	-0.03089	-0.03404	-0.24119
PH	0.0181	-0.01012	**-0.01791**	-0.00027	-0.00031	0.05795	-0.03279	-0.02718	-0.20639
SL	-0.01178	0.00315	0.00014	**0.03495**	0.00003	0	0.02278	0.03586	0.40808
PTP	0.0082	0.0104	-0.00054	-0.0001	**-0.01017**	0.02184	0.02576	0.00444	0.08387
NDVI_2	0.0459	-0.03816	-0.00736	0	-0.00157	**0.14849**	-0.06664	-0.0448	-0.33345
SPAD	-0.01035	0.01258	0.00315	0.00427	-0.0014	-0.05043	**0.18641**	0.08265	0.51312
NGPS	-0.01505	0.0212	0.00399	0.01027	-0.00037	-0.05182	0.12631	**0.12197**	0.54839
TW	-0.031	0.02337	0.00472	0.01819	-0.00109	-0.06001	0.12203	0.08533	**0.78386**

Residual effect – 0.0516, **DFF** – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length, **PTP** – Productive tillers per plant, **NDVI** –Normalised difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test

Table 12. Eigen values for 20 traits for 6 × 6 half diallel population

Principal components	eigenvalue	Percentage of variance	cumulative percentage of variance
PC1	4.544	45.443	45.443
PC2	2.024	20.238	65.681
PC3	1.086	10.862	76.543
PC4	0.979	9.794	86.337
PC5	0.671	6.713	93.05
PC6	0.247	2.474	95.523
PC7	0.237	2.37	97.893
PC8	0.125	1.248	99.141
PC9	0.058	0.58	99.721
PC10	0.028	0.279	100

Table 13. % contribution of various traits towards principal components

Variables	PC1	PC2	PC3
DFF	8.466	19.804	3.188
DM	6.413	15.714	3.175
PH	5.189	7.587	1.233
PTP	2.812	12.743	29.507
SL	0.391	6.032	56.583
NDVI	9.95	18.952	0.015
SPAD	14.211	5.237	2.483
NGPS	16.149	4.842	1.785
TW	18.48	3.304	1.49
GYPH	17.941	5.785	0.542

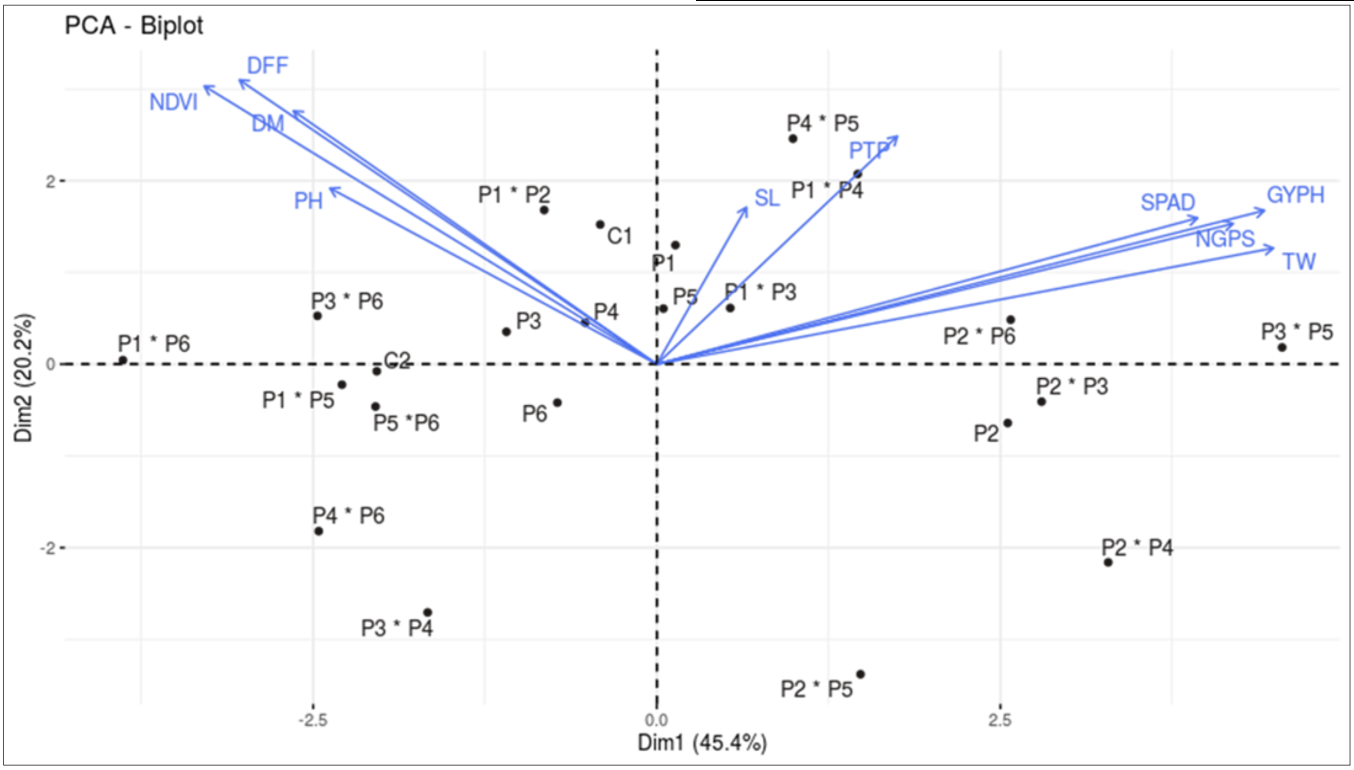

Fig. 2. PCA Biplot and distribution of genotypes under moisture stress conditions in a 6 × 6 half diallel population.

Best parents and hybrids

WH730, RAJ3765 and NIAW3170 emerged as the best parents based on per se performance and GCA (Table 14). Hybrids NIAW3170 × MACS6768 and RAJ3765 × MP1378 showed superior performance under moisture stress based on heterosis and sca effects (Table 15).

Discussion

The ANOVA revealed that differences among genotypes were significant (F = 21, df = 20, *p* = 0.05, 0.01), which was aligned with previous results (8, 9), supporting the presence of genetic variability (Table 1). Parental lines differed significantly for several traits, confirming the presence of diversity among parents, while hybrids also showed wide variation, reflecting the influence of parental combinations. Parents vs. hybrids exhibited significant differences, suggesting that hybrids, on average, performed distinctly from their parental lines, reflecting heterosis for key traits. Replication effects were largely non-significant, which was aligned with other findings validating uniform experimental conditions under RCBD (10, 11).

Variability, heritability and genetic advance

The small difference observed between GCV and PCV for most traits (Table 2) aligned with other research indicates that the environment had minimal impact on their expression (12). However, for SL, PTP and NGPS, the relatively higher gap indicates a noticeable influence of environment on these traits aligned with other studies (13). TW and GYPH with high heritability (>60) and high genetic advance (>20) were aligned with other studies (14, 15 and 16) are controlled by

Table 14. Evaluation of wheat parental lines for *per se* performance and GCA effects

Parents	*Per se* performance	*gca* effects	*Per se,* performance and *GCA* effects
RAJ3765	DM, PTP, NDVI, SPAD, NGPS, GYPH	PTP, NDVI	PTP, NDVI
WH730	DFF, DM, PH, SL, NDVI, SPAD, NGPS, TW, GYPH	DFF, DM, PH, NDVI, SPAD, NGPS, TW, GYPH	DFF, DM, PH, NDVI, SPAD NGPS, TW, GYPH
NIAW3170	DFF, NDVI	DFF, SL	DFF
MP1378	DFF, SL, NDVI, NGPS, TW	DFF	DFF
MACS6768	PH, PTP, NDVI, SPAD	-	-
UAS375	DFF, PH, PTP	-	-

Table 15. Top two performing hybrids based on morphological, physiological and yield traits in a 6 × 6 half diallel cross

Sl. No	**Trait**	**Cross**	**F_1 *per se* mean**	***sca* effect**	**H_{bp} (%)**	**Best SH (%)**	
						UAS 347	**HD 3090**
1	DFF	NIAW3170 × MACS6768	52.00	-8.01**	-22.39**	-24.64**	-16.13**
		WH730 × MP1378	52.00	-5.63**	-16.13**	-24.64**	-16.13**
2	DM	NIAW3170 × MP1378	90.00	-5.34**	-8.16**	-10.00**	-9.09**
		WH730 × MP1378	93.00	-3.09**	-5.61**	-7.50**	-6.57**
3	PH	WH730 × MP1378	86.165	-2.90	-6.85	-4.08	-5.73
		MP1378 × UAS375	88.33	-1.75	-4.51	-1.67	-3.36
4	SL	WH730 × UAS375	10.85	1.64**	5.85	9.60	21.23*
		NIAW3170 × MACS6768	11.40	1.59**	14.00	15.15	27.37**
5	PTP	RAJ3765 × WH730	6.00	0.88**	9.09	20.00*	50.00**
		MP1378 × MACS6768	5.00	0.69**	0.00	0.00	25.00*
6	NDVI	RAJ3765 × WH730	0.655	0.08**	5.65*	10.08**	2.34
		NIAW3170 × UAS375	0.64	0.04**	4.07	7.56**	0.00
7	SPAD	NIAW3170* MACS6768	48.35	3.12**	3.37**	3.09*	9.24**
		WH730 × UAS375	47.80	2.47**	4.77**	2.21	8.31**
8	NGPS	RAJ3765 × MP1378	48.00	5.85**	4.15	8.42	20.48*
		NIAW3170 × MACS6768	49.00	5.11**	6.66	11.15	23.51*
9	TW	NIAW3170 × MACS6768	50.63	9.69**	26.83**	29.64**	35.41**
		RAJ3765 × MP1378	46.289	6.30**	13.92*	18.51**	23.79**
10	GYPH	NIAW3170 × MACS6768	3803.00	649.46**	18.96**	20.62**	32.55**
		RAJ3765 × MP1378	3690.50	606.33**	15.58**	17.05**	28.63**

DFF – Days to 50 % flowering, **DM** – Days to maturity, **PH** – Plant height, **SL** – Spike length, **PTP** – Productive tillers per plant, **NDVI** – Normalized difference vegetation index, **SPAD** – Soil plant analysis development, **NGPS** – Number of grains per spike, **TW** – Test weight (1000-grain weight or 100-seed weight, depending on crop), **GYPH** – Grain yield per hectare.

additive gene action and can be improved through direct selection (Table 2). Traits like SL and NGPS, showing moderate heritability (30–60) and genetic advance (10–20), are influenced by both additive and non-additive effects, leading to gradual improvement. DFF, PTP and NDVI with high heritability but moderate genetic advance suggest partial additive control, while DM and SPAD with high heritability and low genetic advance (<10) are mainly affected by non-additive genes and environment, limiting selection efficiency.

General combining ability (GCA) and specific combining ability (SCA)

The analysis of variance in Table 3 showed significant GCA and *S*CA effects, confirming the involvement of both additive and non-additive gene actions in governing the traits. To identify the best parents and the best crosses, it is required to know the GCA and SCA effects, respectively (17). In Table 4, parent WH730 showed the highest significant GCA effects for key traits, identifying it as the best general combiner, aligned with another study (18). In Table 5, among the hybrids, NIAW3170 × MACS6768 displayed the highest significant *sca* effects for DFF, DM, SL, NDVI, SPAD, NGPS, TW and GYPH, making it the most promising cross combination with high yield, which can thrive under moisture stress conditions, aligned with (19, 20).

The SCA/GCA ratio was less than one for most of the characters, except for plant height, indicating that non-additive effects contributed more strongly to trait expression (Table 6). This outcome is consistent with earlier findings, where the majority of traits were also governed by non-additive variance, apart from spike length (21). In contrast, PH showed a higher contribution of additive variance. Although wheat is a self-pollinated crop where additive effects are generally more valuable for varietal development, the presence of higher non-additive variance suggests scope for exploiting heterosis and can also produce large F_2 and F_3 populations from the crosses to develop transgressive segregants which can be stabilised through subsequent selfing (22).

Per se, performance of hybrids vs parents

GYPH and DFF are the two important traits, as the grain yield is the ultimate goal and earliness helps the crop to escape during moisture stress conditions.WH730 shows the good overall per se performance among the parents (Table 7). Hybrids such as RAJ3765 × MP1378, WH730 × NIAW3170, WH730 × MP1378, WH730 × UAS375, NIAW3170 × MACS6768, MP1378 × MACS6768 recorded greater per se performance than the best check (UAS347) for GYPH, . This suggests that these crosses showed superior performance across multiple traits and may exhibit high *SCA* and heterosis (23). Almost all the parents and hybrids showed early flowering indicating as which helps in crop escape during moisture stress.

Heterosis

The heterosis analysis presented in Table 8-10 across various agronomic and physiological traits revealed considerable hybrid vigour, indicating opportunities for hybrid exploitation and genetic improvement. For grain yield per hectare, hybrids such as NIAW3170 × MACS6768, RAJ3765 × MP1378 and WH730 × MP1378 showed significant positive heterosis over the better parent, which aligns with similar outcomes for several traits (24). These hybrids also outperformed the standard checks, indicating the role of favourable non-additive interactions in enhancing yield, a trend supported by other studies for number of grains per spike and test weight (25). For flowering and maturity, hybrids like NIAW3170 × MACS6768, WH730 × MP1378 and WH730 × MACS6768 displayed significant negative heterosis over checks, suggesting their potential as early maturing lines, a trait particularly valuable in environments where escaping terminal moisture stress is crucial for yield stability. Similarly, yield-contributing traits such as NGPS, TW and PTP exhibited significant heterosis in WH730 × MP1378 and NIAW3170 × MACS6768, which is consistent with the other finding (26). Overall, the repeated and significant heterotic expression of NIAW3170 × MACS6768 and WH730 × MP1378 across several yield and physiological traits emphasises their value both as promising hybrids for direct exploitation and as important parental sources in breeding programs.

Correlation and path analysis among the traits

Grain yield exhibited strong positive associations with SL, SPAD, NGPS and TW, confirming that these traits directly support higher productivity (Fig. 1). In contrast, DFF and DM were negatively correlated with yield, indicating that early flowering and early maturity are beneficial under stress, consistent with earlier reports (27, 28). Lower NDVI values during the grain-filling stage under stress were also advantageous, as they suggest efficient remobilisation of assimilates to the grain, thereby supporting yield, which was aligned with rice studies (29). Thousand-grain weight had the strongest direct positive effect on GYPH (β =0.783), followed by SPAD (β =0.186) and NDVI at the grain-filling stage (β =0.148), as shown in Table 11. In terms of indirect effects, NGPS through TW (β =0.548) and SPAD through TW (β =0.513) were highest, underscoring the central role of test weight in yield improvement. The residual value of 0.0516 indicates that approximately 95 % of the variation in grain yield was accounted for by the traits considered, reflecting the robustness of the model.

Principal component analysis (PCA)

In PCA, key traits influencing PC1 were thousand-grain weight, grain yield and number of grains per spike, while DFF, NDVI and DM were dominant in PC2. PC3 was mainly explained by spike length and productive tillers per plant (Table 12-13) and these results are aligned with other studies (30, 31). The genotype–trait (G-T) biplot (Fig. 2) effectively distinguished both traits and genotypes. Vectors with greater length contributed more to population variability, while genotypes farther from the origin may contribute good transgressive segregants in the next generations. The hybrid NIAW3170 × MACS6768 (P3 × P5) aligned strongly with yield-contributing traits such as NGPS, TW, SPAD and GYPH, highlighting its potential for yield improvement under stress. Conversely, RAJ3765 × UAS375 (P1 × P6) appeared on the opposite side of the main trait vectors, showing weak association with yield traits. Hybrids like P1 × P3, P4 × P5 and most of the parents clustered near the origin, indicating average performance across traits. Overall, hybrids displayed greater variability than parents, reflecting heterosis and wider trait divergence, consistent with earlier findings (32).

Best parents and hybrids

Evaluation of per se performance and General Combining Ability (GCA) among the six wheat parents showed clear variability in genetic potential. Among parents, WH730, RAJ3765 and NIAW3170 emerged as the most promising. WH730 consistently excelled across several traits, while RAJ3765 contributed strongly to PTP and NDVI at grain filling (Table 14). Both carried favourable additive genes, making them suitable for pedigree or recurrent selection. Hybrids such as NIAW3170 × MACS6768 and RAJ3765 × MP1378 demonstrated superior performance under moisture stress conditions based on significant positive heterosis and high *sca* effects (Table 15). Notably, NIAW3170 × MACS6768 exhibited early flowering, aiding stress escape. This indicates that high specific combining ability was not always linked to high parental GCA effects, potentially due to the importance of intra-allelic interaction or additive × additive gene action in high × high combinations (23). It also emphasises that strong *sca* effects can arise even from parents with contrasting GCA, underscoring that this can result in transgressive segregants, likely due to the influence of dominant gene interactions (33, 34). Such crosses offer promising opportunities to improve yield and stress resilience in wheat breeding.

Conclusion

This 6×6 half-diallel study identified a few outstanding hybrids exhibiting strong heterosis and combining ability under moisture stress conditions in wheat. WH730, RAJ3765 and NIAW3170 were effective general combiners, while NIAW3170 × MACS6768 had high specific combining ability and standard heterosis along with RAJ3765 × MP1378. The predominance of non-additive variance indicates strong potential for generating superior recombinants. So, breeders can produce large F_2 and F_3 populations from the crosses like NIAW3170 × MACS6768 and RAJ3765 × MP1378 to develop transgressive segregants.

In the above genotypes, grain yield was positively correlated with key yield-contributing traits and negatively with days to flowering, showing that early-flowering genotypes can better escape terminal drought by completing their life cycle before soil moisture is severely depleted. These genotypes offer strong potential for breeding high-yielding, moisture-stress-tolerant wheat. The high-yielding superior lines can be further utilised in hybridisation and also the development of Multi-parent Advanced Generation Inter-Crossing (MAGIC) populations to combine diverse traits and enhance tolerance to moisture stress during crop growth. There is a scope of molecular breeding, like Marker-assisted selection (MAS), to identify the best genotypes in early segregating generations.

Authors' contributions

KR contributed to the conduct of research experiments and drafting the manuscript, UG assisted in formulating research and analysing results. SN, KDL, VGM and BLS were involved in drafting and reviewing the article. All authors have read and approved the final version of the manuscript.

Compliance with ethical standards

Conflict of interest: The Authors do not have any conflict of interest to declare.

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During the preparation of this manuscript, the authors utilised Scribbr to enhance clarity and readability. All suggestions were applied with care, followed by thorough review and editing by the authors themselves. The final responsibility for the content of the publication rests entirely with the authors.

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