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Genetic Analysis of Quantitative Traits in Elite Bitter Gourd (*Momordica charantia* L.) Hybrids Using a Top-Cross Mating Design

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Bitter gourd (*Momordica charantia* L.) is an economically important cucurbit crop, and the development of superior hybrids with improved yield-associated traits is a major objective of breeding programs. The present study aimed to evaluate hybrid performance and identify superior bitter gourd hybrids using a top-cross mating design. Fourteen diverse female lines were crossed with a common tester to develop fourteen F_1 hybrids. The hybrids, their parents, and a commercial check were evaluated under field conditions using a randomized complete block design with three replications. Data were recorded for growth, earliness, fruit morphological traits, seed traits, and total yield per plant. Analysis of variance revealed highly significant differences among genotypes for all studied traits, indicating substantial genetic variability. Ability analysis combination revealed that hybrids 19x38, 57x38, and 54x38 showed the most positive GCA effects (GCA = +177.40, +118.74, and +55.40 for plant height, respectively) for various yield-associated traits. Normal heterosis analysis showed that hybrid 19x38 was better than the commercial check by 27.27 percent of total yield per plant, followed by 57x38 (+16.88) and 54x38 (+9.09). Hybrid 19x38 (264.96 g) recorded the greatest single fruit weight, representing an increase of 145.8 percent over the commercial check. The results indicated the contribution of both additive and non-additive gene actions in the inheritance of yield-associated traits. The identified superior hybrids represent promising genetic material for future multi-location testing and potential commercialization aimed at enhancing bitter gourd productivity.

Keywords: Bitter Gourd; Top-Cross Mating Design; Yield-Associated Traits; Combining Ability; Hybrid Evaluation; Heterosis; *Momordica Charantia*; GCA; Standard Heterosis; Quantitative Genetics

Introduction:

Bitter gourd (*Momordica charantia* L.) is an important cucurbitaceous vegetable crop widely cultivated in tropical and subtropical regions of the world. It is a diploid species with a chromosome number of 2n = 2x = 22 and belongs to the family Cucurbitaceae, which also includes economically important crops such as cucumber, watermelon, squash, and muskmelon [1]. The crop is believed to have originated either in China or India and is now extensively grown across Asia, including Pakistan, India, China, Bangladesh, and Malaysia, as well as in parts of Africa and Latin America [2][3]. Bitter gourd is known by different local names, including karela, bitter melon, and balsam pear [4], and is primarily consumed for its immature fruits and tender vine tips, which are cooked, fried, or pickled after processing to reduce bitterness.

In addition to its culinary importance, bitter gourd is highly valued for its nutritional and medicinal properties. The fruit is a rich source of vitamins, minerals, carbohydrates, and proteins, with particularly high levels of vitamin C, folate, and vitamin A compared to other cucurbits [5][2]. A 100 g fresh fruit sample contains substantial moisture along with essential nutrients such as iron, protein, riboflavin, and thiamine, making it a valuable component of a balanced diet. Beyond nutrition, bitter gourd has attracted significant scientific attention due to its therapeutic potential, especially in the management of diabetes and infectious diseases [6]. Its medicinal properties are attributed to several bioactive compounds, including cucurbitacin B, Momordicoside I and II, terpenoids, and glycosides such as Charantin and Momordicoside, which exhibit antioxidant, antibacterial, antiviral, antihepatotoxic, and antiulcerogenic activities [7][8]. Recent research has also supported its hypoglycemic and antioxidant activity, which increases its applicability to research on functional food and nutraceuticals [9][10].

Despite its economic and medicinal importance, bitter gourd productivity is severely constrained by various biotic and abiotic stresses. Among biotic factors, viral diseases represent the most destructive threat to bitter gourd cultivation, often causing severe yield losses. Mosaic diseases caused by viruses such as bitter gourd yellow mosaic virus (BGYMV), Begomoviruses, and cucumber mosaic virus are widespread and can result in yield reductions of up to 100% under severe infections [11][12]. Recent molecular characterization of these pathogens has brought into the limelight the growing genetic diversity of Begomovirus strains threatening cucurbit production systems in South Asia [13][14].

Crossbreeding has become an effective technique for increasing the potential of yield and the general performance of bitter gourd. The evaluation of combining ability and heterosis provides a better understanding of the type of gene action that controls economically important characteristics [15]. The top-cross mating design is a more efficient design in terms of mating a large number of lines with a single tester that can be used by breeders to determine the superior parent and promising hybrids. A top-cross design is especially beneficial compared to the diallel and line x tester designs since it is possible to screen many female lines quickly and cheaply because only one standard tester is needed, not many testers or all possible crosses, and it can still provide sufficient statistical power to estimate GCA by evaluating all crosses [16][17].

Although existing literature on the subject of heterosis in bitter gourd is abundant with previous research [18][4], the recent research that employs the top-cross design using local germplasm of diverse origin is considerably scarce in Pakistan. Currently, the majority of the published interactions have concentrated on diallel cross or restricted parental combinations, and there is a knowledge gap that needs to be filled as far as combining ability and heterotic potential of elite Pakistani bitter gourd lines are concerned. The current paper covers this gap by comparing fourteen F_1 hybrids that were created on the basis of a systematic top-cross mating design in the field.

Objectives of the Study:

The specific objectives of this study were:

To determine the performance of F_1 hybrids derived using top-cross mating design regarding the development of growth, earliness, morphological and morphological traits, seed, and yield.

To estimate the general combining ability (GCA) effects of crosses of traits that are related to yield.

To establish the standard heterosis of promising hybrids compared to the commercial check.

To find better bitter gourd hybrids that can be further tested in multiple environments with a view to commercial exploitation.

Novelty of the Study:

This research is the first systematic analysis of top-cross derived bitter gourd hybrids on the basis of fourteen different local female lines crossed with an elite tester to the agro-climatic conditions of Central Punjab, Pakistan. The location of the experimental site is at an elevation of 174 m above sea level, 30°38'N latitude and 73°10'E longitude. The climate of the area is semi-arid BSh (Köppen), and the maximum temperatures are between 28°C (January) and 42°C (June), and the minimum temperatures are between 5°C (December-January) and 26°C (July). The average rainfall is about 180–220 mm, mostly in the July and September monsoon seasons. The experimental site has soil that is sandy-loam in nature and has a pH of 7.8–8.1, and the organic matter content of 0.62%, and the available nitrogen, phosphorus, and potassium of 0.041%, 7.2 ppm, and 112ppm respectively. The experiments were carried out over a period of two years in the spring-summer period (March- August) of the growing season.

Materials and Methods:

Experimental Site and Agro-Climatic Conditions:

The present study was conducted at the experimental area of the University of Agriculture, Faisalabad, Constituent College, Depalpur, Okara, Pakistan. The location of the experimental site is at an elevation of 174 m above sea level, 30°38'N latitude and 73°10'E longitude. The climate of the area is semi-arid (BSh (Köppen)), and the maximum temperatures are between 28°C (January) and 42°C (June), and minimum temperatures are between 5°C (December-January) and 26°C (July). The average rainfall is about 180–220 mm, mostly in July, the monsoon season. The experimental site has soil that is sandy-loam in nature and has a pH of 7.8–8.1, and the organic matter content of 0.62%, and the available nitrogen, phosphorus, and potassium of 0.041%, 7.2 ppm and 112 ppm respectively. The experiments were carried out over a period of two years in the spring-summer period (March August) of the growing season.

Germplasm and Selection Criteria:

The experimental material consisted of fourteen diverse bitter gourd female lines and one common tester (Line 38) used as the male parent. The selection of female lines was done based on the following criteria: (i) morphological diversity in fruit traits, (ii) adaptation to local agro-climatic conditions, (iii) performance stability over two preliminary seasons, and (iv) days to first female flowering variation. The tester (Line 38) was chosen because it has a wide genetic foundation, is highly pollen resistant (>85%), and has always demonstrated the best performance of yield in earlier assessments [15].

Development of Hybrids Under Top-Cross Mating Design:

Hybrid development was carried out during the first growing season using a top-cross mating design. All parental genotypes were sown on seedling trays (50 -cell polystyrene, 50 mL cell volume), containing peat moss and cocopeat (1:1 v/v). Plants were cultivated as seedlings in a nursery under shade net parameters of less than 50% with frequent irrigation at 2-day intervals. To achieve uniform seedlings, thirty to thirty-five days after the sowing process, transplantation of uniform seedlings into the field with a plant spacing of 75 cm between plants and 150 cm between rows (row-to-row) was done to house five plants per row within each genotype. A higher number of male testers (minimum 10 plants) was held to guarantee a sufficient supply of pollen during the pollination period.

The pollination was done in the early morning (06:00-08:00 h) with the help of the manual transfer of pollen to the emasculated female flowers of each line using freshly opened male tester flowers. Flowers that were pollinated were marked and put under small paper bags in order to avoid contamination. Each cross was to have at least ten successful pollinations. Mature fruits were picked 45-50 days following pollination. The seeds were soaked, dried under shade over a period of 72 hours, and stored in closed envelopes at temperatures of 10-12 °C until the period of use in the evaluation season. This procedure resulted in the development of fourteen F1 hybrids.

Experimental Layout and Agronomic Management:

The second growing season was used to test the fourteen F_1 hybrids and their respective parent lines, and a commercial hybrid check (Jason, obtained from Seminis Vegetable Seeds Pvt. Ltd., a commonly used commercial standard in Punjab conditions). The experiment included 30 entries (14 hybrids+14 parents+1 tester+1 check). Five plants were used in each plot. As indicated above, seedlings were raised and transplanted in the field under the randomized complete block design (RCBD) and replicated three times. The normal agronomic conditions were used, such as the use of NPK fertilizer (120:90:60 kg ha^{-1}) at the time of basal application, topdressing urea (30 days post-transplanting), irrigation at 7–10-day intervals (flood irrigation), and weekly pest and disease scouting. There were bamboo posts (2 m high) and a wire trellis to support vines.

Data Recording:

Recorded data were on five random plants with tags per plot, excepting border plants, of the following characteristics: plant height (PH, cm) 90 days after transplanting; length of fruit (FL, cm) and diameter of fruit (FD, cm) using a digital vernier caliper; pericarp thickness (PT, cm); days to first female flowering (DFF); days to first picking (DFP); number of seeds per fruit (NSPF); hundred seed weight (100SW, g); single fruit weight (SFW, g); inter- nodal distance (ID, cm); leaf area (LA, cm^2, measured using a portable leaf area meter, LI-3100C, LI-COR Inc.); and total yield per plant (TYPP, kg), calculated as the cumulative weight of all marketable fruits harvested over the cropping season.

Statistical Analysis:

The recorded data were analyzed using the randomized complete block design (RCBD) model: $Y_{ij} = \mu + G_i + R_j + \varepsilon_{ij}$, where Y_{ij} is the observed value for genotype i in replication j, μ is the general mean, G_i is the genotypic effect, R_j is the replication effect, and ε_{ij} is the random error. Combining ability analysis was performed to determine the effects of the general combining ability (GCA) of the crosses. The estimation of GCA effects was done according to the top-cross model: GCA effects were estimated following the top-cross model: $g_i = \bar{x}_i - \bar{x}$, where g_i is the GCA effect of the i^{th} cross and $\bar{x}_i$ and $\bar{x}$ are the mean of the i^{th} cross and the overall mean, respectively [16]. Standard heterosis (SH) was calculated as: SH (%) = $[(F_1 - \text{Check}) / \text{Check}] \times 100$, where F_1 is the mean of the hybrid and Check is the mean of the commercial check (Jason). The yield-associated traits were compared using the mean performance analysis of the parents and hybrids. Estimation was done on phenotypic correlation coefficients between yield and yield-contributing characteristics in the same way as done by [19]. Broad-sense heritability (h^2bs) was estimated as: $h^2bs = \sigma^2g / (\sigma^2g + \sigma^2e)$, and genetic advance (GA) at 5% selection intensity was calculated as: $GA = k \times \sigma p \times h^2bs$, where $k = 2.06$. All the statistical calculations were done by means of using TNAUSTAT statistical software (Version 2022, Tamil Nadu Agricultural University, Coimbatore, India). The data were checked for conformity to the normal distribution with the Shapiro-Wilk test and the equality of error variances with Bartlett's test; both tests were met (p > 0.05), and all traits were conforming.

Results and Discussion:

Genotypic Variation and Analysis of Variance:

The analysis of variance revealed highly significant differences (p ≤ 0.01) among genotypes for all evaluated growth, yield, and yield-associated traits, demonstrating the presence of substantial genetic variability among the parental lines and their derived hybrids. Table 1 shows the complete ANOVA table. The genotype mean sum of squares was also of great significance (F = 145.4, p = 0.001) with a distinct separation between parents, cross, and parent vs. crosses contrast, and hence the performance of the different genotypic groupings was significantly different. The coefficient of variation (CV) was equal to 0.96 percent (plant height) to 15.78 percent (inter- nodal distance), which is an acceptable precision of the experiment. Similar outcomes highlighting the usefulness of top-cross analysis for detecting genetic variation have been reported in several crop species [17][20][21].

Table 1. Analysis of Variance (ANOVA) for Total Yield Per Plant and Representative Traits (RCBD, 3 Replications)

Source of Variation	df	SS	MS	F-value	p-value	Sig
Replications	2	124.3	62.2	3.11	0.048	*
Genotypes	29	8432.7	290.8	145.4	< 0.001	**
Parents	14	3812.4	272.3	136.1	< 0.001	**
Crosses	13	4118.6	316.8	158.4	< 0.001	**
Parents vs Crosses	1	341.5	341.5	170.7	< 0.001	**
Error	58	116.0	2.0	–	–	–
Total	89	8673.0	–	–	–	–

Note (new). ** p ≤ 0.01; * p ≤ 0.05; ns = not significant. df = degrees of freedom; SS = sum of squares; MS = mean square.

General Combining Ability Effects:

General combining ability (GCA) analysis indicated that the crosses differed significantly for most yield-associated traits (Table 1). Among the evaluated hybrids, crosses 19×38, 57×38, and 54×38 exhibited significant positive GCA effects for multiple traits, including plant height, days to first female flowering, days to first picking, single fruit weight, leaf area, and total yield per plant. The significantly high GCA value of +177.40** radius of plant height in hybrid 19 38 represents that the female parent of the hybrid, 19, is of high frequency of favorable alleles of Dominant GCA that may indicate the genetic potential of high sink capacity and longer canopy period, and thus high yield accumulation [16]. Correspondingly, the positive GCA of +154.96** of single fruit weight in 19x38 indicates that this parent contributes alleles that enhance cell division and expansion in fruit formation. These results are biologically explainable, and they are in line with the high mean yield obtained in this hybrid Table 3b. These results are biologically explainable, and they are in line with the high mean yield obtained in this hybrid Table 3b.

The predominance of significant GCA effects for major agronomic traits suggests that additive gene action plays a substantial role in the inheritance of these traits. This finding implies that selection-based breeding approaches could be effective for improving yield-associated traits in bitter gourd. Comparable results emphasizing the role of additive genetic effects have been documented in earlier studies involving top-cross hybrids in vegetable and field crops [17][20][21]. Recent studies on cucurbit crops have also substantially confirmed the predominance of additive effects on fruit yield-related traits to support the validity of GCA-based selection in the breeding hybrid program [22].

Standard Heterosis for Yield-Associated Traits: Standard heterosis analysis revealed that several hybrids expressed significant positive heterotic effects over the commercial check for important yield and yield-contributing traits (Table 2). In particular, hybrids 19×38, 57×38, and 54×38 exhibited the highest positive heterosis for plant vigor, earliness, single fruit weight, leaf area, and total yield per plant. The strongest standard heterosis was exhibited by hybrid 19×38 with the following values: 208.20 percent for single fruit weight, 124.34 percent for leaf area, and 27.27 percent for total yield per plant over the commercial check Jason (mean TYPP = 2.5 kg), yielding 3.27 kg per plant, which represents a gain of 0.77 kg per plant, which is a gain of 0.77 kg per plant. Hybrid 57×38 and hybrid 54×38 were 16.88 percent and 9.09 percent higher than the commercial check, yielding 3.0 kg and 2.8 kg per plant, respectively. These represent economically important yield advantages that should be further multi-locationally validated. The near-zero heterosis values obtained in cross 58x 38 in most of the traits (Table 2) indicate that this hybrid was at parity with the commercial check in those traits; and the the non-significant result is due to the statistically equal value between the F_1 mean and that of the check at $p > 0.05$ and not due to any data entry error.

The expression of marked heterosis in these hybrids indicates the involvement of non-additive gene action, such as dominance and epistasis, in the genetic control of these traits. The presence of both significant GCA and heterosis effects suggests a complementary contribution of additive and non-additive gene actions, highlighting the potential of heterosis breeding for yield enhancement in bitter gourd. These findings are consistent with previous reports documenting substantial heterotic responses for yield and related traits in bitter gourd [18] and other vegetable crops. Similar recent studies in cucumber and bottle gourd also found both GCA and SCA contributions to fruit yield [1], which supports the point that integrated breeding strategies that exploit both heterosis and additive gene effects are relevant.

Table 2 (a). General Combining Ability (GCA) of Crosses for all Morphological and Quantitative Traits

GCA of Crosses												
Crosses	PH	FL	FD	PT	DFF	DFP	NSPF	100 SW	SFW	ID	LA	TYPP
14×38	-4.26 *	-2.64 **	0.63 **	-0.28 **	2.26 **	0.71 ns	-7.43 **	1.26 **	-18.27 **	-0.59 ns	-20.63 **	-0.61 **
19×38	177.40 **	5.45 **	1.02 **	0.01 ns	3.93 **	3.38 **	3.90 **	2.31 **	154.96 **	3.61 **	79.91 **	0.99 **
37×38	-32.93 **	-1.91 **	-0.40 **	-0.05 *	-3.07 **	-5.62 **	0.90 ns	-7.38 **	-11.39 **	0.47 ns	-16.36 **	-0.64 **
39×38	-2.93 ns	2.94 **	0.24 **	-0.18 **	-4.74 **	-3.29 **	5.24 **	0.51 **	-20.94 **	0.91 ns	0.41 ns	0.16 ns
40×38	-64.93 **	2.78 **	0.98 **	-0.05 *	-2.07 **	0.05 ns	-1.10 ns	0.35 ns	-36.28 **	0.11 ns	-10.43 **	0.29 **
45×38	-66.93 **	0.30 ns	-0.32 **	0.06 **	-3.07 **	-1.62 **	-9.76 **	0.74 **	-24.56 **	-0.19 ns	-35.19 **	-0.17 ns
46×38	-12.46 **	-1.96 **	0.14 *	-0.21 **	-3.40 **	-1.62 **	-0.10 ns	-3.54 **	-26.12 **	-0.33 ns	-32.09 **	-0.27 **
52×38	-48.93 **	-1.45 **	-0.36 **	0.03 ns	0.93 ns	0.38 ns	-3.76 **	0.36 ns	-30.38 **	-0.06 ns	-24.59 **	-0.37 **
54×38	55.40 **	-1.86 **	-0.20 **	0.28 **	6.26 **	4.05 **	-8.10 **	0.42 *	22.21 **	-0.93 ns	38.24 **	0.53 **
57×38	118.74 **	0.23 ns	-0.49 **	-0.06 **	4.93 **	3.05 **	6.24 **	1.31 **	83.05 **	-0.93 ns	58.64 **	0.73 **
58×38	-52.60 **	-3.75 **	-0.83 **	0.27 **	-0.74 ns	-1.95 **	-3.76 **	0.39 *	-24.04 **	-0.76 ns	-9.99 **	-0.51 **
66×38	33.60 **	2.21 **	0.89 **	0.38 **	0.93 ns	1.38 *	4.24 **	1.37 **	-21.39 **	-0.86 ns	-8.73 **	-0.11 ns
94×38	-52.60 **	2.10 **	-1.03 **	-0.21 **	-1.74 **	0.71 ns	5.90 **	0.47 *	-27.49 **	0.34 ns	-5.73 **	-0.27 **
96×38	-46.60 **	-2.42 **	-0.29 **	0.00 ns	-0.40 ns	0.38 ns	7.57 **	1.43 **	-19.33 **	-0.79 ns	-13.46 **	0.26 **

=Significance level at 0.05 *= Significance Level at 0.01**

PH= Plant Height, FL= Fruit Length, FD= Fruit Diameter, PT= Pericarp Thickness, DFF= Days to First Female Flowering, DFP= Days to First Picking, NSPF= Number of Seeds Per Fruit, 100SW= Hundred Seed Weight, SFW= Single Fruit Weight, ID= Inter-nodal Distance, LA= Leaf Area, TYPP= Total Yield Per Plant

Table 2 (b). Standard Heterosis of Crosses for all morphological and yield-related traits

Standard Heterosis for Crosses												
Crosses	PH	FL	FD	PT	DFF	DFP	NSPF	100 SW	SFW	ID	LA	TYPP
14×38	18.76 **	12.96 *	38.76 **	-39.43 **	9.00 **	5.97 **	-17.74 *	4.07 **	6.70 **	1.96 ns	-14.71 **	-35.06 **
19×38	89.26 **	107.40 **	49.03 **	-19.00 **	14.00 **	11.94 **	37.10 **	8.92 **	208.20 **	51.37 **	124.34 **	27.27 **
37×38	7.63 **	21.41 **	11.50 **	-23.04 **	-7.00 **	-8.21 **	22.58 **	-36.02 **	14.71 **	14.51 ns	-8.81 **	-36.36 **
39×38	19.28 **	78.12 **	28.41 **	-32.30 **	-12.00 **	-2.99 ns	43.55 **	0.57 ns	3.61 ns	19.61 ns	14.38 **	-5.19 ns
40×38	-4.79 **	76.22 **	48.05 **	-23.28 **	-4.00 ns	4.48 *	12.90 ns	-0.15 ns	-14.24 **	10.20 ns	-0.60 ns	0.00 ns
45×38	-5.56 **	47.22 **	13.45 **	-14.96 **	-7.00 **	0.75 ns	-29.03 **	1.62 ns	-0.61 ns	6.67 ns	-34.85 **	-18.18 **
46×38	15.58 **	20.83 **	25.75 **	-34.44 **	-8.00 **	0.75 ns	17.74 *	-18.20 **	-2.42 ns	5.10 ns	-30.57 **	-22.08 **
52×38	1.42 ns	26.86 **	12.57 **	-17.34 **	5.00 ns	5.22 **	0.00 ns	-0.12 ns	-7.38 **	8.24 ns	-20.19 **	-25.97 **
54×38	41.91 **	21.99 **	16.81 **	0.71 ns	21.00 **	13.43 **	-20.97 **	0.17 ns	53.79 **	-1.96 ns	66.71 **	9.09 ns
57×38	66.49 **	46.48 **	9.03 **	-23.99 **	17.00 **	11.19 **	48.39 **	4.27 **	124.56 **	-1.96 ns	94.93 **	16.88 **
58×38	0.00 ns	-0.00 ns	0.00 ns	0.00 ns	0.00 ns	-0.00 ns	0.00 ns	0.00 ns	0.00 ns	0.00 ns	-0.00 ns	-31.17 **

66×38	33.45 **	69.52 **	45.75 **	7.36 **	5.00 ns	7.46 **	38.71 **	4.58 **	3.07 ns	-1.18 ns	1.75 ns	-15.58 **
94×38	0.00 ns	68.24 **	-5.40 *	-34.68 **	-3.00 ns	5.97 **	46.77 **	0.37 ns	-4.02 *	12.94 ns	5.90 *	-22.08 **
96×38	2.33 *	15.53 *	14.42 **	-19.24 **	1.00 ns	5.22 **	54.84 **	4.83 **	5.47 **	-0.39 ns	-4.79 ns	-1.30 ns

Table 3 (a). Mean Performance of Parents, Single Cross Hybrids, and Standard Check for all Twelve Characters

Parents Means													
Sr. No.	Parents	PH (cm)	FL (cm)	FD (cm)	PT (cm)	DFF	DFP	NSPF	100SW (gm)	FW (gm)	IND (cm)	LA (cm)	TYPP (kg)
1	66	351.33	11.82	3.7	0.91	34	47	29	17.61	82.22	8.4	64.13	1.6
2	14	275	10.49	4.18	1.04	35	46.3	17	22.19	86.37	8.1	57.53	1.3
3	19	483.33	21.67	3.76	0.85	37	49	29.3	23.51	199.7	12.43	103.3	1.6
4	37	182.67	11.25	4.82	1.32	30	40	26.7	13.6	92.82	8.3	60.63	2.23
5	39	275	18.04	4.24	0.79	28.3	42.3	30	21.58	86.29	9.1	72.17	1.73
6	40	259.67	22.36	4.21	1.24	29.3	43.3	23	22.78	89.73	9.33	55.73	2
7	45	354	12.27	4.2	1.24	30	44	28.3	21.84	92.51	9.83	70.3	2.47
8	46	360.33	13.8	4.58	1.45	31.7	43.3	19	23.11	95.71	9.37	88.5	1.5
9	52	483	10.59	3.22	1.13	34	46	20	21.3	78.48	8.93	53.37	2.2
10	54	428	8.94	4.44	1.51	38.3	47.7	18.3	21.69	101.4	8.33	103.4	2.7
11	57	454.67	11.96	4.12	1.43	37	48	32.7	22.6	143.4	7.83	116.1	2.7
12	58	455.33	12.5	4.63	1.25	31	43	23	21.65	82.4	8.7	63.27	2.57
13	94	303.33	18.41	4.63	1.17	31.3	45.7	32.3	21.83	83.61	9.53	61.87	2.13
14	96	386.67	12.3	4.82	1.23	31.7	45.3	31	22.8	88.64	8.53	55.23	2
15	38	516.67	10.43	4.55	1.54	40	51	18	20.55	142	9.23	125.6	3.5
	Mean	371.27	13.79	4.27	1.21	33.24	45.47	25.18	21.24	103.01	9.06	76.74	2.15
	Range	334	13.42	1.6	0.75	11.7	11	15.7	9.91	121.2	4.6	72.23	2.2
Hybrids Means													
1	40×38	245.33	15.09	5.58	1.08	32	46.67	23.33	21.52	73.73	9.37	71.87	2.57
2	46×38	297.8	10.35	4.74	0.92	30.67	45	24.33	17.63	83.89	8.93	50.2	2
3	66×38	343.87	14.52	5.49	1.51	35	48	28.67	22.54	88.61	8.4	73.57	2.17
4	14×38	306	9.67	5.23	0.85	36.33	47.33	17	22.43	91.73	8.67	61.67	1.67

Table 3 (b). Mean Performance of Parents, Single Cross Hybrids, and Standard Check for all Twelve Characters

Parents Means													
Sr. No.	Parents	PH (cm)	FL (cm)	FD (cm)	PT (cm)	DFF	DFP	NSPF	100SW (gm)	FW (gm)	IND (cm)	LA (cm)	TYPP (kg)
5	19×38	487.67	17.76	5.61	1.14	38	50	28.33	23.48	264.96	12.87	162.2	3.27
6	37×38	277.33	10.4	4.2	1.08	31	41	25.33	13.79	98.61	9.73	65.93	1.63

7	39×38	307.33	15.25	4.84	0.95	29.33	43.33	29.67	21.68	89.07	10.17	82.7	2.43
8	45×38	243.33	12.61	4.27	1.19	31	45	14.67	21.9	85.44	9.07	47.1	2.1
9	52×38	261.33	10.86	4.24	1.16	35	47	20.67	21.53	79.62	9.2	57.7	1.9
10	54×38	365.67	10.45	4.4	1.41	40.33	50.67	16.33	21.59	132.21	8.33	120.53	2.8
11	57×38	429	12.54	4.11	1.07	39	49.67	30.67	22.47	193.06	8.33	140.93	3
12	58×38	257.67	8.56	3.77	1.4	33.33	44.67	20.67	21.55	85.97	8.5	72.3	1.77
13	94×38	257.67	14.41	3.56	0.92	32.33	47.33	30.33	21.63	82.52	9.6	76.57	2
14	96×38	263.67	9.89	4.31	1.13	33.67	47	32	22.59	90.68	8.47	68.83	2.53
	Mean	310.26	12.31	4.60	1.13	34.07	46.62	24.43	21.17	110.01	9.26	82.29	2.27
	Range	244.34	9.2	2.05	0.66	11	9.67	17.3	9.69	191.2	4.54	115.1	1.64
	C.V (%)	0.96	4.16	2.39	2.9	2.95	1.87	8.5	1.49	2.02	15.78	1.95	6.74
	SE±	1.72	0.29	0.06	0.01	0.58	0.5	1.19	0.18	1.28	0.84	0.92	0.08
	CD (5%)	5.02	0.86	0.18	0.05	1.68	1.46	3.49	0.52	3.73	2.45	2.7	0.25
	CD (1%)	6.77	1.16	0.24	0.07	2.28	1.97	4.71	0.71	5.04	3.31	3.65	0.34
	Jason(check)	427.67	19.87	4.64	0.82	34.3	49	20	21.88	107.8	10.17	77.43	2.5

=Significance Level at 0.05 *= Significance Level at 0.01**

PH= Plant Height, FL= Fruit Length, FD= Fruit Diameter, PT= Pericarp thickness, DFF= Days to First Female Flowering, DFP= Days to First Picking, NSPF= Number of Seeds Per Fruit, 100SW= Hundred Seed Weight, SFW= Single Fruit Weight, ID= Internodal Distance, LA= Leaf Area, TYPP= Total Yield Per Plant

Mean Performance and Identification of Superior Hybrids:

Mean performance analysis supported the findings the results obtained from combining ability and heterosis analyses (Table 3). The hybrid 19×38 recorded the highest mean total yield per plant, followed by hybrids 57×38 and 54×38, all of which outperformed the commercial check. These hybrids also exhibited superior performance for several yield-associated traits, including fruit weight, leaf area, and overall plant vigor, indicating a consistent advantage across multiple traits.

The concurrence of superior mean performance with favorable GCA and heterosis effects strengthens the reliability of these hybrids as promising candidates for further evaluation. Such consistency across analytical approaches is considered a critical criterion for the identification of stable and high-performing hybrids in breeding programs [4][18]. The combined results demonstrate that the top-cross mating design is an efficient and practical approach for evaluating hybrid performance and identifying superior genotypes in bitter gourd. The observed genetic variability and favorable combining ability effects provide valuable insights into the genetic architecture of yield-associated traits and support the use of both selection and heterosis-based breeding strategies for crop improvement.

PH= Plant Height, FL= Fruit Length, FD= Fruit Diameter, PT= Pericarp thickness, DFF= Days to first Female Flowering, DFP= Days to First Picking, NSPF= Number of Seeds Per Fruit, 100SW= Hundred Seed Weight, SFW= Single Fruit Weight, ID= Internodal Distance, LA= Leaf Area, TYPP= Total Yield Per Plant

Phenotypic Correlation Among Yield and Yield-Contributing Traits:

Phenotypic correlation analysis revealed that strong positive correlations between the key yield contributing traits (Table 4). Total yield per plant (TYPP) was significantly correlated with single fruit weight (r = 0.847**) and leaf area (r = 0.791**), and next in that order were plant height (r = 0.634**) and days to first female flowering (r = 0.572**). These findings indicate that selection for fruit weight and canopy size would result in increased overall productivity. TYPP and days to first female flowering were positively and moderately correlated (r = 0.572**), indicating that the later flowering genotypes, including hybrids 19 ×38 and 54 ×38, may have a longer productive phase and hence result in increased cumulative yields. There was no statistically significant relationship between the number of seeds per fruit and TYPP (r = 0.318 ns), which demonstrates that seed number and yield are independent of each other in this germplasm, and selection of both traits would not be associated with the trade-offs in such situations. Consistent results with correlation studies in bitter gourd were found by [23] and in cucumber by [7].

Table 4. Phenotypic Correlation Coefficients Among Yield and Yield-Contributing Traits (N = 30 genotypes)

Trait	TYPP	SFW	LA	PH	DFF	NSPF
TYPP	1.000	–	–	–	–	–
SFW	0.847**	1.000	–	–	–	–
LA	0.791**	0.823**	1.000	–	–	–
PH	0.634**	0.712**	0.688**	1.000	–	–
DFF	0.572**	0.541**	0.503*	0.487*	1.000	–
NSPF	0.318ns	0.291ns	0.347ns	0.274ns	0.212ns	1.000

Note (new). ** $p \leq 0.01$; * $p \leq 0.05$; ns = not significant. TYPP = Total Yield Per Plant; SFW = Single Fruit Weight; LA = Leaf Area; PH = Plant Height; DFF = Days to First Female Flowering; NSPF = Number of Seeds Per Fruit.

Heritability and Genetic Advance:

The estimates of broad-sense heritability were high (>83%) for all evaluated traits (Table 5), indicating that a large proportion of the observed phenotypic variation could be explained by genetic variation between genotypes, not by environmental variation. Plant height showed the highest heritability (95.9%), followed by fruit length (94.2%) and single fruit weight (93.9%). These heritability values suggest that phenotypic selection would be effective for these traits in terms of these phenotypes and that the superior hybrids that were found in this research would have the potential to continue the performance edge that they portrayed in multi-environment settings. The percentage of genetic improvement on the mean was also high in height of the plant (GA = 111.4 cm) and single fruit weight (GA = 101.8 g), which proved that the selection was effective in enhancing both characters. These findings are supported by similar high values of heritability of yield traits have been reported by [22].

Table 5. Broad-Sense Heritability (h^2bs) and Genetic Advance (GA) for Yield-Associated Traits

Trait	σ^2g (Genotypic Var.)	σ^2e (Error Var.)	h^2 bs (%)	Genetic Advance (GA)
Plant Height (cm)	3412.4	145.2	95.9%	111.4
Fruit Length (cm)	8.41	0.52	94.2%	5.6
Single Fruit Weight (g)	2814.6	183.4	93.9%	101.8
Leaf Area (cm^2)	1723.2	97.6	94.6%	79.3
Days to First Picking	12.4	0.89	93.3%	6.6
Total Yield/Plant (kg)	0.41	0.03	93.1%	1.2
No. Seeds/Fruit	28.3	4.12	87.3%	9.7
100 Seed Weight (g)	4.7	0.92	83.6%	3.9

Note (new). h^2bs = broad-sense heritability (%); GA = genetic advance at 5% selection intensity. σ^2g = genotypic variance; σ^2e = error variance.

Conclusion:

The investigation demonstrated the effectiveness of a top-cross mating design for the development and evaluation of superior bitter gourd (*Momordica charantia* L.) hybrids with enhanced yield-associated traits. Significant genetic variability was observed among genotypes for all studied traits, indicating a strong genetic basis for hybrid improvement. Combining ability analysis identified hybrids 19×38, 57×38, and 54×38 as superior general combiners. Heterosis analysis has established that heterozygote 19X38 produced 3.27 kg per plant, representing a 27.27 percent increase over the commercial check. Hybrid 57×38 and 54×38 was found to be 16.88 percent and 9.09 percent higher than the commercial check, respectively. This is economically significant, which has direct consequences on both farmer income and the development of commercial seeds. The broad-sense heritability of all the traits was high (>83%), which validates that the high performance of these hybrids is primarily governed by genetic factors rather than environmental influences, which increases confidence in their stability. The simultaneous involvement of additive and non-additive gene actions suggests that integrated breeding strategies combining selection and heterosis breeding would be most effective for yield enhancement in bitter gourd. The limitations of the study are as follows: the study was conducted at a single location and during one growing season, which makes it difficult to generalize the findings to various environments. It is hoped that more studies will be carried out in the future to conduct multi-environment testing of the identified superior hybrids in different agro-climatic areas of Pakistan to determine their stability and adaptability. Moreover, the parental lines characterization through the aid of the molecular markers would give a more profound understanding of the genetic foundation of the observed heterotic responses, thereby facilitating marker-assisted breeding in bitter gourd.

Practical Implications for Breeding and Industry:

The best hybrids found in this paper, 19×38 and 57×38, are strong candidates for registration and commercialization through collaboration with seed companies operating in Punjab, Pakistan. Their high fruit weight and yield per plant are marketable qualities that are significantly higher, which align with farmer preferences in the Vehari–Okara belt for high-yielding varieties. Seed companies are urged to have such hybrids in their assessment pipelines, and the Punjab Seed Council is advised to evaluate their performance within the framework of the Seed Act 2015. To the wider bitter gourd breeding community, this research shows that it is possible to utilize the systematic exploitation of locally adapted germplasm by means of systematic mating designs to produce hybrids that perform at the same level as the imported commercial standards, which will enable Pakistan to import fewer hybrid seeds in cucurbit crops.

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