Dissertation report
(GPB 596)

"STUDY OF HETEROSIS AND COMBINING ABILITY FOR YIELD AND ITS COMPONENT TRAITS IN BARLEY (*Hordeum vulgare* L.)"

Lovely Professional University, Punjab
In partial fulfillment of the requirements for the award of the degree of

Master of Science (Agriculture)

In

Genetics & Plant Breeding

By

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May, 2018

CERTIFICATE

This is to certify that the Dissertation entitled **"STUDY OF HETEROSIS AND COMBINING ABILITY FOR YIELD AND ITS COMPONENT TRAITS IN BARLEY (*Hordeum vulgare* L.)"** is a bonafide record of independent research work done by **Akashdeep Kamboj,** (Reg. No.: 11718631) under our supervision and submitted to Lovely Professional University in partial fulfillment for the award of the Degree of Master of Agriculture. (Genetics & Plant Breeding)

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DECLARATION

I hereby declare that the project work entitle **"STUDY OF HETEROSIS AND COMBINING ABILITY FOR YIELD AND ITS COMPONENT TRAITS IN BARLEY (*Hordeum vulgare* L.)"** Is an authentic record of my work carried out at lovely professional university as requirements of project work for the award of degree of Master of Science in Genetics and Plant Breeding, under the guidance of Dr. Madakemohekar Anant Hanumant, Assistant professor, School of Agriculture, Lovely Professional University, Phagwara, Punjab.

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1. INTRODUCTION

Barley (*Hordeum vulgare* L.) is one of the world's most ancient food crops. It has been an important cereal crop since the early stages of agricultural innovations 8,000-10,000 years ago (Giles and Bothmer, 1985). It is an economically important cereal crop, ranking fourth after wheat, rice and maize in the world, both in terms of quantity produced and in area of cultivation (FAO, 2014). Barley originates from the Eastern Mediterranean region where plants experience many abiotic stresses in the field. It is grown in many areas where climatic conditions are unfavorable. Though its commercial value is less than that of wheat but it replaces the later in the dry regions in areas of too low and erratic rainfall. Because of low input requirement and better adaptation, it survives easily under rainfed condition and known as poor men's crop (Verma *et al*., 2010). World production of barley is 292.9 million tonnes with highest production from Europe region (59.6%) followed by Asian region (14.9%). Russian federation is the highest producing country which produces near about 20.02 million tonnes, while India has thirteenth rank (USDA, 2015).

Cultivated barley is a member of the genus *Hordeum*, and it has descended from wild barley (*Hordeum spontaneum* Koch), which still grows in the Middle East of the world. Both cultivated and wild barley are diploid species, with fourteen chromosomes (2n= 14). Based on the morphology, *Hordeum vulgare* L. is the only cultivated species which has two – distant phenotypic forms *viz.*, six rowed (*Hordeum vulgare*, *H. hexasstichum*) and two rowed (*H. distichum*). In spite of differences in spike morphology they have same chromosome number (2n=14), and intercross freely to produce fertile hybrids (Poehlman, 1987). Barley has much genetic variation which provides the basis for classifying the species. There are many ways to classify barley among each other. One way to classify barley is to identify whether there are two, four or six rows of spikelet's on the spike. Wild barley has two rows, and most cultivated barley is of six-rowed type. Another way to classify barley is to describe the beards (awns) link with the kernels. Barley can also be described by adherence of chaff on grains (hulled) or hull-less (naked), height (dwarf, semi-dwarf and tall), seed color (colorless, white, yellow and blue) and feed or malt type. Some hull-less cultivars are more digestible due to higher-protein, lysine and in some instance glutamine content.

This has been very well explained that semi dwarf wheat and rice varieties as well as hybrids of maize and millets gave substantial increase in yield (15-20%) on high-input management but subsequently their adverse effect on soil fertility, ground water table and pollution of drinking water caused human health hazard and environmental pollution which have been quite alarming. There is no scope to raise the yield by raising fertilizers and irrigation level. It means technology revolution has reached to the freezing point in wheat and rice; maize and millets because of genetic ceiling in yield. Therefore, barley especially huskless barley is an option to produce more from less input in India where 70 percent of cultivated area is under rainfed condition.

Its production has become more intense and complex in recent years. Due to this reason, it is necessary to carry out experiments to estimate the response of barley plants to a variety of adverse conditions, such as low and high solar energy availability, shortage or excess of water in soil, high temperature and salinity, which affects photosynthesis and yield formation (Kalaji, 2012). There is a need for the development of new barley cultivars that tolerate abiotic and biotic stresses for the improvement of crop productivity (Ellis *et al*., 2000). This will require good understanding of the available genetic variation in both wild and cultivated barley. The rate of progress, however, will depend on the occurrence of desirable genetic variation and the availability of precise methods of identification, selection and transfer of superior genes (Ellis *et al*., 2000).

Proper choice of parents on the basis of their combining ability status for putative drought tolerant attributes as well as productive traits and selection in typical target environment will help in combining complex traits, such as, productivity and drought tolerance (Hanamaratti *et al*., 2004). The concept of combining ability helps the breeder to determine the nature of gene action involved in the expression of quantitative traits of economic importance. The choice of suitable breeding method for the improvement of drought tolerance traits primarily depends on the relative importance of GCA and SCA variances. A hybrid is commercially valuable only when it exhibits significantly high standard heterosis over the best locally adapted variety or hybrid. Apart from high vigor and yield, the hybrids can be a potential genetic source for better root system with higher efficiency to absorb moisture effectively for tolerating drought condition. Existence of heterosis for desired traits will be a boon to drought tolerance breeding

since most of the hybrids developed so far lack tolerance to abiotic stresses. The generation mean analysis is one of most appropriate methods of genetic analysis for quantitative traits (Eshghi and Akhundova, 2009). In this method, epistatic effects as well as additive and dominance effects can be estimated. Besides gene effects, breeders would also like to know how much of the variation in a crop is genetic and to what extent this variation is being transferred generation after generation. Because efficiency of selection mainly depends on additive genetic action, influence of the environment and interaction between genotype and environment as well.

The magnitude of additive gene effect is particularly useful in the development of pureline varieties. Drought is predominantly controlled by additive genes as has been reported by Solmon *et al*. (2003). Likewise, the information concerning dominance and epistatic gene effects (non-additive components) is also valuable for development of hybrid varieties (Sharma and Tandom, 1997: and Munir *et al*., 2007). Several barley workers have tried to estimate the various gene effects; genetic variance and combining ability through exploiting different mating design, such as, diallel, half-diallel, line x tester, partial-diallel, triallel and generation mean analysis etc.

With these points in view, the present investigation entitled **"Study of heterosis and combining ability for yield and its component traits in barley (Hordeum vulgare L.)"** will be undertaken with the following objectives.

1. To study the nature and magnitude of gene action controlling the inheritance of yield and its contributing characters.
2. To find out the best general and specific combiners for yield and its contributing characters.
3. To identify the good lines on the basis of per se performance.
4. To identify the trait(s) to form the basis of selection to increase the yield in barley.

2. REVIEW OF LITERATURE

Various biometrical procedures are used by plant breeders for estimation of genetic value of parents and evaluation of varieties and hybrids in terms of their genetic makeup in different adverse conditions need to reviewed before the start of any research programme. A vast literatures in respect of combining ability and gene action, heterosis and inbreeding depression for various yield e traits have been reviewed and brief account of which are presented in this chapter as follows,

2.1 Combining ability and gene effects

Madic *et al.* (2007) emphasized highly significant differences for both the combining abilities in the F_1 generation of barley, which showed that, the grain weight per plant in these investigations was dependent on genes with additive and non-additive (dominant) effects.

Singh *et al*. (2007) studied 8 lines and 5 testers in barely for combining ability analysis and gene action studies for grain yield and its components and reported that the relative estimates of variance due to SCA were higher than variance due to GCA for all the traits studied except days to flowering indicating the predominance of non-additive gene action.

Verma *et al.* (2007) conducted a study on combining ability effects through line x tester analysis; the results indicated the predominance of non- additive gene action (*h*) for all the traits. The line Kedar and tester K-560 in normal fertile soil and tester Lakhan in saline sodic soil while RD-2552, Narendra Jau-4 and NDB-1173 under both the environments proved good general combiners for seed yield and quality components characters.

Eshghi and Akhundova (2009) studied generation mean and variance analysis on six generations (P_1, P_2, F_1, F_2, B_1 and B_2) derived from the barley cross ICNBF93-369 x ICNBF582 and SB91925 x ICB 102607 to complement the genetic information obtained from the diallel

analysis. Wr/Vr graph in diallel analysis and average degree of dominance together with narrow sense heritability values in both the experiments revealed additive gene effects for plant height, number of tillers and days to maturity and over-dominance gene action for number of grains per spike.

Pal and Kumar (2009) observed the significant role of additive genetic component (D) for the inheritance of days to 50% heading, plant height and spikelet's per ear in barley. The non-additive component (H) was found to be important for the genetic control of all the traits except for days to 50% flowering and number of tillers per plant. However, the relative magnitude of dominant component (H) was higher as compared to additive component (D) in all the traits indicating the preponderance of dominant gene effects in controlling the inheritance of these traits.

Rabbani *et al.* (2009) based on the wr/vr graphic representation showed that traits like flag leaf area, fertile tillers per plant, 1000 grain weight and grain yield per plant were controlled by over-dominance type of gene action under water non stress and stress conditions in wheat. While spike length exhibited over-dominance type of gene action under non stress condition and additive type of gene action under stress condition.

Eshghi *et al***.** (2010) carried out generation mean and variance analyses in barley and reported that both additive and dominance effects were important for most of the traits evaluated but, dominance and non-allelic interaction had a more pronounced effect for number of grains per spikes in drought and 1000 grain weight and grain yield on both the environments.

Presence of non-allelic interaction for all the studied traits in all the crosses made in barley indicated by El-Aty (2011); the additive effect was more important and greater than the dominance effect for most of the traits. Among the epistatic components, dominance × dominance (*l*) was greater in the magnitudes than additive × additive (*i*) and additive × dominance (*j)* in the most studied traits.

Singh *et al.* (2011) observed that GCA and SCA variances were significant for all the traits except number of spikes per plant at SCA level in barley. The GCA variances were higher than SCA variances in respect of all the traits except grain yield per plant where both GCA and

SCA were of same in order. The additive gene effects were generally predominant whereas, dominance and over dominance gene effects were equally important for grain yield per plant.

Ciulca *et al*. (2012) studied diallel analysis of covariance regression for spike length in six doubled haploids lines of six-row winter barley. The studied parental forms in terms of spike length submitted a higher proportion of dominant alleles and a nearly symmetrical distribution of positive and negative alleles. In both generations, the overdominance direction was associated with an increase in spike length. A high proportion of recessive and negative alleles have been reported in DH 19-1 line, respectively a large proportion of dominant and negative alleles submitted the DH 26-2 and DH 20-4 lines, while the other lines showed a relative instability during the two generations. Because the genetic system that controls the spike length four both generations is mainly additive, selection can play a great role in breeding method of that trait.

Jain and Sastry (2012) revealed the mean square due to GCA and SCA were significant for most of the traits which indicated the presence of both additive (*d*) and non-additive (*h*) gene effects for controlling the expression of yield and yield contributing characters in wheat. The σ^2GCA / σ^2 SCA ratio suggested that the presence of non-additive gene action was predominant for most of the characters including grain yield.

On the basis of GCA and SCA effects, Singh *et al.* (2012) reported that, 3 parents (K 7903, K 9465 and HUW 234) and 14 cross combinations (5 top crosses namely HD 2733 × K 7903, HUW 234 × K 9423, HD 2285 × K 2021, HUW 234 × K 2021 and K 9423 × K 2021) were found good general and specific combiners for higher grain yield and also for various yield contributing traits, respectively in wheat.

Desale and Mehta (2013) revealed that the mean squares due to both GCA and SCA were significant for all traits in wheat indicating both additive and non-additive genetic variances played a vital role in the inheritance of all these traits. The ratio between GCA and SCA variance was less than unity for all the traits which indicated that non-additive component play relatively greater role in the inheritance of all eight traits. On the basis of GCA, SCA effects and *per se* performance, parents HI-1544 for all the traits except biological yield per plant and reducing sugar and HW-5018 for all the traits except biological yield per plant and chlorophyll content

and two crosses namely HW-5018 x HI-1544 and RAJ-4136 x UAS-281 for four traits were found as good general and specific combiners, respectively.

Fellahi *et al.* (2013) revealed that low $\sigma 2$ GCA/$\sigma 2$ SCA ratios and low to intermediate estimates of h^2_{ns} supported the involvement of both additive and non-additive gene effects in wheat. The preponderance of non-additive type of gene actions clearly indicated that selection of superior plants should be postponed to later generation.

Pawar and Singh (2013) revealed that GCA and SCA variances were highly significant for all the traits studied in barley. Four parents JB1, PL751, JB58 and RD2787 were found to be good combiners for most of the characters and can be used in the future breeding program. Cross combinations JB × HUB208, JB58 × HUB208, JB1 × Bh933 and JB1 × JB58 exhibited high significant positive SCA effects for most of the traits and identified as superior crosses.

Potla *et al.* (2013) revealed combining ability analysis in barley and reported significant differences among the parents for GCA, among the crosses for SCA for all the quantitative traits. Among the parents, tester namely RD-2508 and lines IBON-65, IBON-18, Beecher, Rihane, Moroc-9-75, 11th HBSN-146 and HUB-174 were good general combiners for grain yield and its component traits.

Raikwar (2013) studied genetic architecture of quantitative and qualitative traits in barley under saline sodic soil using generation mean analysis of the 5 crosses, results of which revealed that magnitude of dominance (*h*) effects was higher than additive (*d*) effects indicating the preponderance of dominance (*h*) effects over the additive effects. It is obvious that non fixable gene effects (*h*), (*j*) and (*l*) were higher than the fixable (*d*) and (*i*) in all the crosses, for all the characters indicating greater role of non-additive effects in the inheritance of all the characters.

Saad *et al.* (2013) observed that both general (GCA) and specific (SCA) combining ability variances were significant for most of the studied traits under both irrigation regimes indicating the importance of additive and non-additive genetic variances in determining the performance of these traits.

Singh (2013) studied combining ability in twenty one barley F_1 which revealed that variance due to GCA as well as SCA were highly significant for different traits (grain yield per plot, days to fifty percent flowering, tiller per pant, plant height, spikelet's per spike and 1000 grain weight) except spike length. The preponderance of non-additive gene effect for grain yield and of additive gene effect for yield components was detected. Based on estimate of GCA effects, good general combiners were identified for different traits. Similarly based on SCA effects, desirable specific combiner were identified. It was noted that good specific combiner did not necessarily involve good general combiners.

Varzaru and Ciulca (2013) examined the overdominance effects which have been found for the combinations where the parental forms did not differ in terms of the grain number/spike, while in the combinations where there were larger differences between parental forms, the inheritance of this trait was controlled by partial dominance effects. The inheritance of TGW for most combinations (87 %) was controlled by overdominance effects, associated with an increase in this trait.

Madic *et al.* (2014) studied that analysis of variance of combining abilities showed significant differences for GCA and SCA in the F_1 generation of barley suggesting additive and non-additive gene action. The GCA/SCA ratio in F_1 indicated the prevalence of the additive component of genetic variance for spike length, grain weight per spike and spike harvest index. By contrast the SCA variance for grain weight per spike was higher than the GCA variance indicating the dominance of non-additive gene action.

Nature and magnitude of gene effects for yield and its component traits in barley using generation mean analysis in 5 crosses were studied by Raikwar *et al*. (2014). In general, magnitude of dominance effect (h) showed a greater value than additive effect (*d*) in all the traits. It is obvious that non-fixable gene effects (*h*), (*j*) and (*l*) were higher than the fixable (*d*) and (*i*) in all the crosses in all the characters indicating the greater role of non-additive effects in the inheritance of all the characters. The study revealed the importance of non-additive type of gene action for most of the traits thereby suggesting that selection at later segregating generation could provide better results.

Bornare *et al*. (2014) studied combining ability analysis, which revealed that the variance due to General Combining Ability (GCA) and Specific Combing Ability (SCA) were highly significant for most of the traits studied. The estimated value of σ^2A was higher than its σ^2D for plant height and thousand seed weight which indicated the predominance of additive gene effects as the ratio of σ^2A/σ^2D was more than unity, while rest of the traits showed preponderance of non-additive gene effects. The value of average degree of dominance for plant height and thousand seed weight indicated partial dominance while rest of the traits *viz.*, chlorophyll content, spike length, awn length, number of effective tillers, number of grains per spike, harvest index and grain yield per plant showed over-dominance.

Deniz *et al.* (2015) determined the combining abilities of some wheat genotypes for yield and some yield related traits by using line × tester mating design. The specific combining ability (SCA) effects were generally found higher than general combining ability effects (GCA) in terms of the agronomic traits studied. As a result, low ratios of σ2 GCA / σ2 SCA and low narrow sense heritabilities showed that non-additive effects controlled the traits studied. Hence, the selection process for superior individual plants should be postponed to further generations like F_4 or F_5.

Fahad *et al.* (2015) studied various hexaploid wheat genotypes indicated significant GCA (parents) and SCA (F_1 hybrids) effects for the characters plant height, tillers per plant, spike length, spikelet's per spike, seeds per spike, seed index, and grain yield per plant. The mean performance of F_1 hybrids differed significantly for all the traits studied. Among the parents, Imdad and TD-1 proved to be better general combiners for almost all the studied traits. With regards to SCA effects, the F_1 hybrids between Imdad × TD-1 and Imdad × SKD-1 expressed higher SCA.

Hong-tao *et al.* (2015) observed that plant height and its component traits were controlled by both additive and dominant genetic effects, and additive effect was main factor. The GCA for plant height and each internode length of Supi 3 and Gangpi 2 showed negative effect, and they could be used as parents in improving the plant height.

Xinzhong *et al.* (2015) showed that GCA was significantly different among parents and SCA was also significantly different among crosses. The performance of hybrid was significantly correlated with the sum of female and male GCA (TGCA), SCA and heterosis. Hu1154 A, Mian684 A, 86F098 A, 8036 R and 8041 R were excellent parents with greater general combining ability. The variances of SCA were significant only for traits plant heitht, inter-node length, spike length and thousand kernel weight. The ratio of GCA/SCA ranged from 6.24 in IL to 18.87 in SP, indicating that additive effects played a more important role than non-additive effects for all traits.

2.2 Heterosis and inbreeding depression

Heterosis (or hybrid vigour) is the superiority the F_1 in relation to their parents (Fehr, 1987). First coined by G.H. Shull in 1914, heterosis has been exploited by breeders to enhance the productivity of numerous crop and horticultural plants. He also observed the effects of inbreeding and cross breeding of maize. The effects of the phenomenon have been quantified in a wide variety of plant studies (Stuber, 1994). There are two different measures of heterosis. Mid-parent heterosis is defined as the increased vigour of the F_1 over the mean of the parents. High-parent heterosis is defined as the increased vigour of the F_1 over the greater parent (Crow 1999). We will be mainly concerned with high-parent heterosis because mid-parent heterosis will always be smaller or equivalent to high-parent heterosis if parents are equal (Crow 1999).

Rugen *et al*. (2004) studied that, the mid parent heterosis often existed and the occurrence rates of positively and negatively significant mid parent heterosis were 46% and 12%, respectively. On the other hand, the occurrence rate of the significant heterobeltiosis was 28% on average, ranging from 0% (Plant height and Kernels on main spike) to 79% (Internode length below spike), varied with the traits. The crosses of 3 x 10 and 6 x 8 had strong heterosis, and they belong to the combinations of 6 row x 6 row types and 2 row x 2 row types of barley, respectively. It seems that the hybrid with strong heterosis could be easier to find in the

combinations of 6 row x 6 row or 2 row x 2 row barley types than that of 6 row x 2 row or 2 row x 6 row barley types.

Masood *et al*. (2005) crossed 8 genotypes of barley in full diallel fashion and 56 crosses with their 8 parents were screened in a 3 replicated compact family randomized block design. The magnitude of average heterosis and heterobeltiosis from F_1 were estimates for yield and its component traits. Out of 56 hybrids, Tat x KW and Tat x Tkb for days to heading, Taq x Inq and Inq x Taq for flag leaf area, Inq x PS and PS x ID for spike length, Tkb x ID and PS x FS for days to maturity and PS x FS and FS x ID for harvest index expressed significant heterobeltiosis.

Daya Ram *et al.* (2006) studied 78 hybrids in a diallel cross set derived from 13 diverse parents in both F_1 and F_2 generations of barley. Highest economic heterosis was recorded for grain yield (48.82%) followed by number of productive tillers plant^{-1}(38.09%), 1000-grain weight (21.43%), length of spike (19.05%), number of grains spike^{-1} (11.63%) and plant height (11.72), respectively. Crosses BH 120/BR 3085, K329/BR 3085, Azad/RD 883, Azad/K329, Azad/P267 and P420/K 71 exhibited high estimates of economic heterosis with varied estimates of inbreeding depression due to additive × additive and additive × dominance types of epistatic gene action.

Kularia and Sharma (2006) reported that the range of heterosis was quite wide except for days to heading and plant height indicating that sufficient amount of genetic variability was present in the parent material of barley. Maximum heterobeltiosis (-2.21) in desirable direction was recorded in the cross RD 2508 x RD 2052 for days to heading with significant inbreeding depression. All the three crosses showed positive and significant heterobeltiosis (12.57 to 33.54 %) and inbreeding depression (-8.41 to 24.07%) for 1000-grain weight. Two crosses out of three expressed significant positive heterobeltiosis for biological yield per plant. The negative inbreeding depression was depicted by the cross Rajkiran x IBVT 12 indicating more biological yield in segregating generation.

Soylu (2006) made 29 crosses among the 12 barley cultivars. Mean heterosis according to mid parent percentage of hybrid population were varied between 29.68% (kernel number per spike) and 45.62% (grain yield per plant), whereas mean heterosis compare to best parent

percentage varied between -42.13% (kernel number per spike) and 25.03% (grain yield per plant). Significant positive correlation were found between grain yield per plant, plant height, spike length, kernel weight per spike, fertile tiller number and 1000 grain weight. As a result, suitable combinations to be used in breeding studies in barley were estimated.

Pandey (2007) selected six accessions of *A. hypochondriacus* and made 15 cross combinations to study heterosis and inbreeding depression. The hybrids which exhibited highest heterosis also showed high inbreeding depression. Heterosis over better parent was highest for economic grain yield (145.04%), followed by panicles/plant (113.67%), panicle length (33.65%) and grain weight/panicle (23.56%).

Pal and Kumar (2009) were studied 15 barley crossed in a half diallel fashion. Observed significant SCA effects along with significant standard heterosis for days to 50% heading, ear length, biological yield and grain yield. High sca effects and heterosis as observed in this combination could be due to divergence and high and low GCA values of the parents

Jaiswal *et al*. (2010) undertaken with a set of diallel crosses involving 6 genotypes of bread wheat during to identify heterotic combinations expressing high hybrid vigour. Thecross combination Kalyansona x K-8962 followed by Sonalika x K-8962, K-8962 x HUW-234, Kalyansonax HUW234 and HUW-510 x HUW-234 were found top hybrids having high mid-parent heterosis. Negativeheterosis for days to flowering and plant height in cross PBW-373 x Kalyansona and HUW-510 x PBW-373 was found desirable. Highly significant heterosis was found for spike length in cross Sonalika x K-8962, tillers per plant in cross HUW-510 x K-8962, number of grains per spike in cross Sonalika x K-8962, test weight in cross PBW-373 x Kalyansona and harvest index in cross Kalyansona x K-8962.

Positive heterotic effects relative to the mid-parent for most of the traits in the 5 crosses of barley, except for heading and maturity dates that showed negative heterotic effects found by El-Aty (2011). Also positive heterotic effects relative to the better parent were found for the most of crosses. Heritability estimates in narrow sense were low to moderate for the studied characters in all the crosses which ranged from 16.37% for spike length in the fifth cross to 66%for days to heading in the second cross.

Singh *et al*. (2011) observed the magnitude of heterosis and combining ability in six rowed barley was using 5 x 5 diallel systems for yield and its component traits. Positive heterosis was observed for all the traits except 1000-grain weight. Over dominance was observed only for grain yield per plant The heterosis and over dominance for grain yield per plant were positive in all the crosses except K 560 x K 635 where it showed partial dominance. For 1000-grain weight negative heterosis was observed in all the crosses except BH 495 x PL 508 for plant height positive heterosis was observed in eight cases and over dominance for three.

Vishwakarma *et al*. (2011) studied heterosis for yield and chlorophyll content in barley. Best crosses having highest heterobeltiosis (better parent heterosis) for particular traits were NDB-1173 × K-792, NDB-1173 × Narendra Jau-3 for days to heading as well as days to maturity, NDB, 1173 × NDB-1245 for number of grain/spike, NDB-1173 × NDB-1245 and NDB-1245 × PL-762 for chlorophyll content and total chlorophyll content respectively and NDB-1245 × PL-762 and NDB-1173 × NDB-1245 for grain yield per plant.

Koumber and El-gammaal (2012) observed significant heterotic values in positive direction for all characters except for plant height and 1000 grain yield in the first cross, spike length in the second cross and plant height, number of grains per spike and number of spikes per plant in the third cross of wheat. Over dominance for all characters except plant height and 1000 grain weight in the first cross, spike length in the second cross and number of grains per spike in the third cross were detected. Inbreeding depression was obtained in two out of three crosses for spike length, number of grains per spike, number of spikes per plant, 1000 grain weight and grain yield per plant and in one out of the three crosses for plant height.

Lamalakshmi *et al*. (2013) studied the magnitude of heterosis for grain yield and its seven yield components for 36 F_1 hybrids in bread wheat. The cross UP 2596 X DBW 17 was recognized as the best heterotic cross for grain yield as it exhibited highly significant positive heterosis over both the standard checks UP 2554 and PBW 343. The cross HW 2019 x UP 2338 exhibited highest and significant positive heterosis over better parent, mid parent and over both the standard checks for number of grains per spike. The present study reveals good scope for isolation of pure lines from the progenies of heterotic F_1 s as well as commercial exploitation of heterosis in bread wheat.

Poutla *et al*. (2013) studied that cross IBON-65 × RD-2508 showed highest magnitude of economic heterosis over the best standard check K- 603 for grain yield per plant in barley.

Saad *et al*. (2013) revealed some crosses showed significant desirable heterobeltiosis for all the studied traits under both stress as well as non-stress conditions. The high positive heterobeltiosis for grain yield per plant was associated with high positive heterobeltiosis for number of spikelet per panicle and 1000 grain weight for barley crosses Giza 126 X Giza 2000 and Giza 130 X Giza 131 under stress and non-stress conditions, respectively.

Bornare *et al.* (2014) studied extent of heterosis for different characters in relation to standard check K-603 revealed that, the overall good heterotic crosses were BCU-4932 x Karan-16 and BCU-4925 x Karan-16 were for short stature plant, BCU-4927 x K-603, BCU-4932 x Lakhan and BCU-4910 x RD-2035 for higher chlorophyll content, BCU-4956 x Lakhan, BCU-4925 x Karan-16 and BCU-4927 x Karan-16 for number of effective tillers per plant. All the 24 crosses showed significant and positive economic heterosis for thousand seed weight whereas, negative heterosis for number of grains per spike. The number of grains per spike showed negative heterosis because the standard check (K- 603) is six rowed and all the f1s' were intermediate and hence had fewer grains per spike than standard check.

Shahzadi *et al*. (2015) studied heterosis and heterobeltiosis in among seven wheat genotypes in all possible combinations. Maximum significant heterosis (21.95%) was found in grain yield per plant followed by spike length (14.62%) and grain yield per spike (13.68%). While maximum heterobeltiosis was recorded for grain yield per plant (11.33%), followed by spike length (9.13%). It is concluded that 4072 x Punjab-96 cross showed best performance followed by Parwaz-94 x MH-97, Iqbal-2000 x parwaz-94 than other crosses under study. These crosses cab be utilized in further breeding programme as parents for contributing high yield not only under optimum environment but also under drought conditions as water use efficient crosses. The results of heterosis suggest that hybrid vigour is available for the commercial production of wheat and selection of desirable hybrids among the crosses having heterotic effects in other characters is the best way to improve the grain yield of bread wheat.

3. MATERIAL AND METHODS

The materials used and methods applied during the present course of investigation on "Study of heterosis and combining ability for yield and its component traits in barley (Hordeum vulgare L.)" are being presented, followed by statistical procedures used. Twenty five elite and diverse pure lines of barley were received from the B.H.U., Varanasi. The experiments will be conducted at Agricultural Research Farm of School of Agriculture, Lovely Professional University as per following detailed plan of work.

3.1.1 Experimental site

The experiments will conducted during the *rabi* (winter) season of 2017-18 and 2018-19 at Agricultural Research Farm of School of Agriculture, Lovely Professional University. The experimental area is quite uniform in respect of topography and fertility. The soil of experimental site is sandy loam.

3.2 Experimental material

25 varieties of barley will be collect and these genotypes were sown at three dates with a week gap, in two rows of 2 m length having a spacing of 25 cm x 10 cm following single seed per hill to keep the plant population at optimum level.

Selected testers will be crossed with selected lines from 25 barley varieties to produced F_1's (excluding reciprocals) in line x tester fashion, using testers as female (Table 3.1). Standard agronomic practices will be follow to raise a good crop. Cross seeds will be harvested separately for each crosses, dried well and packed to grow next generation.

Selection, hybridization and evaluation of selected genotypes

1st year (*Rabi* season, 2017-18)

- Out of 25 genotypes will be grown and maintained on Agriculture Research Farm, School of agriculture, Lovely Professional University, will be selected on the basis of genotypic diversity for making the crosses.
- These genotypes will be sown at three dates with a week gap, in two rows of 5 m length having a spacing of 25 cm x 10 cm following single seed per hill to keep the plant population at optimum level.
- Three testers will be crossed with seven lines to produced 21 F_1s' (excluding reciprocals) in line x tester fashion, using testers as female.
- Standard agronomic practices were followed to raise a good crop.
- Cross seeds will be harvested separately for each crosses to grow next generation.

IInd year (*Rabi* Season, 2013-14)

- The experimental materials (21 F'_1s along with their parents including standard check) will be grown in a single row plot of 5 m length in the Compact Family Randomized Block Design with three replications.
- *Per se* performance of parents and crosses will be assessed for various traits.
- Observations on various morphological, physiologic and drought tolerant traits will be recorded on randomly selected plants as detailed in para 3.3.

Table 3.1: Details of selected barley genotypes.

S.No.	Name of Lines/Testers	Source	Rwo
	Lines		
1.	Azad	BHU, Varanasi	Six row
2.	KR 521	BHU, Varanasi	Six row
3.	Ratna	BHU, Varanasi	Six row
4.	HUB 113	BHU, Varanasi	Six row
5.	Atahualpa	BHU, Varanasi	Six row
6.	RD 2508	BHU, Varanasi	Six row
7.	Dolma 6	BHU, Varanasi	Six row
	Testers		
1.	K 745	BHU, Varanasi	Six row
2.	K 603	BHU, Varanasi	Six row
3.	BH 902	BHU, Varanasi	Six row

3.3 Observations recorded

Ten competitive plants from each of the parents and F_1s', 20 plants from backcrosses (B_1 and B_2) and 50 plants from each F_2 population from each replication were randomly selected and tagged for recording of data on following quantitative traits.

3.3.1 Days to 50 % flowering

The number of days will be taken from sowing to heading in main spike of 50 % plants of a plot will be recorded.

3.3.2 Days to maturity

The number of days will be recorded from sowing to physiological maturity of main spike in hundred per cent plants.

3.3.3 Plant height (cm)

At the physiological maturity, the height of individual tagged/sampled plant will be measured in centimeters from the ground level to the tip of terminal spikelet (excluding the awn) of the main shoot.

3.3.4 Number of effective tillers

At the physiological maturity, the total number of spike bearing tillers in each plant will be recorded.

3.3.5 Spike length (cm)

Length of main spike (cm) will be measured from the base to the tip of the terminal spikelet, excluding the awn.

3.3.6 Awn length (cm)

Length of the awn will be measured in centimeter.

3.3.7 Number of grains per spike

The number of grains per spike will be counted from main spike after the harvesting of plant.

3.3.8 1000 grain weight (g)

One thousand threshed grains will be taken randomly after sun drying at 12% moisture level and weighted in gram with the help of electric balance.

3.3.9 Harvest index

Harvest index will be calculated as,

$$\text{Harvest Index} = \frac{\text{Economic yield}}{\text{Biological yield}} \times 100$$

3.3.10 Grain yield per plant (g)

The weight of filled grains of each plant in gram will be recorded.

3.4 Statistical analysis/ Biometric analysis

3.4.1 Analysis of Variance

The analysis of variance was completed according to Kempthorne (1957) as described below. The mean data recorded for rainfed (2013-14 and 2014-2015) and irrigated 2014-2015 were formed the basis for analysis of variance for each character.

Table 3.6: ANOVA for line x tester analysis

Source	d.f.	S.S.	M.S.S.	F. ratio
Replication	r- 1	rSS	Mr	Mr/Me
Treatment	n- 1	nSS	Mn	Mn/Me
Parents	p- 1	pSS	Mp	Mp/Me
Parents vs Crosses	1	pcSS	Mpc	Mpc/Me
Crosses	lt- 1	cSS	Mc	Mc/Me
Lines (Male)	l- 1	lSS	M1	σ2e + rσ2lt + rtσ2l
Testers (Female)	t- 1	tSS	M2	σ2e + rσ2lt + rlσ2t
Lines x Testers	(l-1) (t-1)	ltSS	M3	σ2e + rσ2lt
Error	(r-1) (n-1)	eSS	Me	σ2e

Where,

r = number of replications, n = number of treatments,
p = number of parents (l + t),
l = number of male lines,
t = number of female lines,
c = number of crosses (l × t),
MSS = mean sum of squares,
df = degree of freedom

The test of significance was carried out with various MS against eMS using 'F test' at the respective degrees of freedom for all the sources of variations except lines and testers where MS due to lines x tester was used. With the help of expectation, covariance of full sibs and half sibs were estimated by using the formula given below:

$$\text{Covariance of half sibs} = \frac{(M_1 - M_2) + (M_2 - M_3)}{r(l + t)}$$

$$\text{Covariance of full sibs} = \frac{(M_1 - M_4) + (M_2 - M_4) + (M_3 - M_4)}{} + \frac{6r\,\text{Cov (H.S.)} - r(l + t)\,\text{Cov (H.S.)}}{}$$

3r 3r

3.4.2.1 Estimation of general and specific combining abilities effects

The additive model was used to estimate the general and specific combining ability effects of ijkth observation is given here:

$X_{ijk} = \mu + g_i + g_j + s_{ij} + e_{ijk}$

Where,

μ = population mean
g_i = gca effect of ith female parent
g_j = gca effect of jth male parent
s_{ij} = sca effect of ijth combination
e_{ijk} = error associated with the observation X_{ijk}
i = number of female parents
j = number of male parents
k = number of replication

The GCA effects for both male and female parents and SCA effects for each cross combination were calculated with the help of following formula:

$$\mu = \frac{X}{ltr}$$

Where,

X… = Total of all hybrid combination over replication

(i) Lines: GCA effect of ith lines (g_i) = $\frac{Xi..}{tr} - \frac{Xi..}{ltr}$

(ii) Testers: GCA effect of jth testers (g_i) = $\frac{X.j.}{lr} - \frac{X...}{ltr}$

(iii) Crosses: SCA effect of ijth lines (s_{ij}) = $\frac{X..j}{r} - \frac{Xi..}{tr} - \frac{X.j.}{lr} - \frac{X...}{ltr}$

Where,

$X_i..$ = Total of ith line over *t* testers and *r* replications
$X_{.j}.$ = Total of jth tester over *l* lines and *r* replications
X_{ij} = ijth combination over all replication

3.4.2.2 Standard Error for the Combining ability effect

The standard errors were estimated as follows:

$$S.E.\,gca\,(line) = \sqrt{Me/_{rt}}$$

$$S.E.gca\ (tester) = \sqrt{Me/_{rl}}$$

$$S.E.\hat{S}ij\ (sca) = \sqrt{Me/_{r}}$$

The test of significance for estimates of the GCA and SCA effects were tested as follows:

$$'t'gca\ (line) = \frac{\hat{g}i - 0}{S.E.\hat{g}i}$$

$$'t'gca\ (tester) = \frac{\hat{g}i - 0}{S.E.\hat{g}i}$$

$$'t'sca\ = \frac{\hat{s}ij - 0}{S.E.\hat{s}ij}$$

The calculated 't' thus obtained was compared with table value at error degree of freedom at p = 0.05 and p = 0.01.

3.4.2.3 Critical difference (C.D.) of the estimates

The differences between two estimates were tested by comparing them with C.D. value.

C.D. = S.E. of difference two estimate × t at 5% error degree of freedom.

S.E. of differences of two estimates were calculated as follows

$$S.E.(\hat{g}i - \hat{g}j)line\ = \sqrt{2Me/_{rt}}$$

$$S.E.(\hat{g}i - \hat{g}j)tester\ = \sqrt{2Me/_{rl}}$$

$$S.E.(\hat{s}ij - \hat{s}kl) = \sqrt{2Me/_{r}}$$

3.4.2.4 Proportional contribution of lines, testers and their interactions to total variance

$$\text{Contribution of Lines} = \frac{\text{SS (l)}}{\text{SS (Crosses)}} \times 100$$

$$\text{Contribution of Testers} = \frac{\text{SS (t)}}{\text{SS (Crosses)}} \times 100$$

$$\text{Contribution of Lines} \times \text{Testers} = \frac{\text{SS (l} \times \text{t)}}{\text{SS (Crosses)}} \times 100$$

3.4.3 Estimation of heterosis

Heterosis in F_1s' will be calculated as the difference of F_1 hybrid performance from the better parents (Heterobeltiosis) and standard checks (Standard heterosis) by using the formulae (Kempthorne, 1957).

Heterobeltiosis (%) = $\frac{(\bar{F}_1 - \bar{BP})}{\bar{BP}} \times 100$

Standard heterosis (%) = $\frac{(\bar{F}_1 - \bar{C})}{\bar{C}} \times 100$

Where,

$\bar{F}_1$ = mean performance of F_1

$\bar{F}_2$ = mean performance of F_2

$\bar{BP}$ = mean performance better parent.

$\bar{C}$ = mean performance of check variety.

Test of significance of heterosis

Significance of heterosis was tested by 't' test. The calculated value of 't' was compared with table value of 't' at error degree of freedom from ANOVA comprising parents and F_1 at p = 0.05 and p = 0.01. t value was estimated s given below:

$$t\ (H) = \frac{\bar{F}_1 - \overline{BP}\ or\ \overline{SC}}{SE\ (H)\ over\ BP\ or\ SC}$$

$$SE\ (H)\ for\ BP\ or\ SC = \sqrt{\frac{2Mse}{2r}}$$

Where,

Me = error variance obtained by ANOVA comprising parents and F1's

r = Number of replication

3.4.4 Heritability (Narrow sense)

Heritability (h^2) estimate was worked out by using the formula suggested by Lush (1949) and Burton and De Vance (1953):

h^2 (Narrow sense) = $\frac{\sigma^2_A}{\sigma^2_P} \times 100$

Where,

h^2 (ns) = Heritability expressed in per cent

$\sigma^2 A$ = Additive genetic variance

$\sigma^2 P$ = Phenotypic variance

The estimates of heritability are categorized as High (>30%), Moderate (>10% and <30%) and Low (<10%).

3.4.5 Expected genetic advance

It was calculated as per formula suggested by Lush (1949).

(a) Genetic advance (GA) = $(K)\ (h^2)\ (\sigma P)$

(b) Genetic advance as % of mean = $GA/\overline{X} \times 100$

Where,

h^2 = estimates of heritability (absolute value)

σ_P = phenotypic standard deviation

K = selection differential at 5% selection intensity, i.e., 2.06

X = population mean for the concerned character

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