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GENETIC ANALYSIS OF PLANT HEIGHT, YIELD COMPONENTS AND SEED YIELD IN RAPESEED (*BRASSICA NAPUS* L.) CULTIVARS

ABSTRACT

Half F_2 diallel crosses of spring rapeseed (*Brassica napus* L.) varieties along with their parents were evaluated for plant height, first siliqua height, siliquae on main axis, siliquae per plant, 1000-seed weight and seed yield. The Analysis of variance based on Cockerham's method revealed significant mean squares of general combining ability (GCA), specific combining ability (SCA) for all the traits indicating that both additive and non-additive genetic effects were involved in controlling these traits. Parent *vs* crosses mean square which is indicating average heterosis was significant for all the traits. In Hayman's method the regression lines estimated for all the traits except plant height cut the Wr axis below the origin in the negative zone, suggesting the overdominance type of gene action for these traits except plant height. Based on the array points on the regression lines, 19H had maximum dominant genes for siliquae on main axis, siliquae per plant 1000-seed weight and seed yield, being closest to the origin, while Option500 possessed recessive genes for first siliqua height, siliquae per plant and 1000-seed weight, as evidenced by its distant position from the origin. Due to low narrow-sense heritability estimates for most of the traits, selection for improving these traits should be carried out in delayed segregating generations.

Key words: additive, diallel, heterosis, heritability, regression lines, seed yield

INTRODUCTION

Due to autumn cultivation of rapeseed (*Brassica napus* L.) in Iran it needs low irrigation, therefore, it has major role in producing self sufficiency in edible oil. Hence, it is necessary to develop the new ideotype varieties based on cross breeding methods with high yield components and seed yield (Rameeh, 2011). Major difficulties involved in yield-

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improvement programs are the accurate selection of parents from which to generate superior breeding combinations, and the selection of high-yielding genotypes from early-segregating generations (Thurling, 1991). The knowledge of genetic control of agronomic traits is very important for plant breeders to formulate proper breeding strategy for improvement new oilseed brassica varieties. Diallel analyses (Hayman, 1954a,b; Jinks, 1954; Griffing, 1956; Cockerham*,* 1963) are commonly used as technique to understand mode of inheritance of a character and also provide information on the nature and amount of genetic parameters. Singh and Chaudhry. (2004) stated that heritability of a trait approaches its maximum in successive generations following hybridization. In addition, the presence of additive gene effects for a trait indicates that selection could be successful for the trait (Kearsey and Pooni, 1996). Estimation of genetic constitution of parents for seed yield and its components can be important for indirect selection for high seed yield in rapeseed (Downeyand Rimer, 1993; Nassimi *et al.*, 2006; Singh *et al.*, 2010; Teklewold and Becker, 2005; Yadav *et al.*, 2005). Although combining ability studies in oilseed *Brassica* spp. are scanty, most of these studies emphasized the preponderance effect of general combining ability (GCA) for yield and its components indicating the importance of additive gene action (Brandle and McVetty, 1989; McGee and Brown, 1995; Woś *et al.*, 1999). On the other hand, Pandey *et al.* (1999) reviewed evidences for the presence of significant specific combining ability (SCA) effects for yield and yield associated traits. Ramsay *et al.*, (1994) reported that variation for both GCA and SCA were responsible for dry matter yield and other quantitative traits in *B. napus*. Significant GCA and SCA effects were reported for siliquae per main raceme, siliquae per plant, siliqua length, number of seeds per siliqua, 1000-seed weight and seed yield in *B. napus* (Leon, 1991; Singh and Murty, 1980; Thakur and Sagwal, 1997), but in other study (Singh *et al.*, 1995) the importance of additive genetic effects for siliquae per plant and 1000-seed weight was emphasized. Singh and Yadav (1980) and Thakur and Sagwal (1997) while examining the genetic control of seed yield in rapeseed found both additive and non-additive gene effects to be involved. Khan and Khan (2005) performed a complete diallel cross analysis in rapeseed according to Hayman (1954b) and Jinks (1954). Vr/Wr graphs indicated that seeds per siliqua, 1000-seed weight and seed yield were controlled by overdominance type of gene action, while siliquae per plant was controlled by additive gene action. Magnitude of narrow-sense heritability was very low for number of seeds per siliqua indicating delaying selection will be more efficient for this trait. Number of siliquae per plant, 1000-seed weight and seed yield per plot had moderate narrow-sense heritability. Shen *et al.* (2005) accomplished genetic analysis for rapeseed genotypes in which F1 hybrids exhibited significant differences in seed and oil yields per plant among F1s and their parents. In this research, although yield heterosis was affected by both additive and non-additive genetic effects, but additive genetic effects were more important for oil content. The KD/KR ratio also revealed excess of dominant alleles in the parents for all the characters, indicating that the parents were diverse and from different populations. Mahmud *et al.* (2009) reported that non-additive genetic effects were more important in the inheritance of yield and its related traits including plant height, number of siliqua per plant, 1000-seed weight and seed yield per plant in rapeseed.

Due to different estimations of the genetic components for the traits based on application of different and divers genetic materials, the objective of the present study was to estimate genetic parameters for plant height, seed yield and its components in six rapeseed breeding lines to be used in open pollinated and hybrid varieties development programs.

MATERIALS AND METHODS

The material under study consisted of six cultivars including RGS-003, Option500, RW008911, RAS-3/99, 19H and PF7045/91 which were selected based on their different agronomic characters (Table 1). These genotypes were crossed in half diallel method during 2004-05. In order to produce F_2 progenies, fifteen F_1s were selfed at Biekol Agriculture Research Station, located in Neka, Iran (53° 13$'$ E longitude and 36° 34$'$ N latitude, 15 m above sea level) during winter 2005-06. F_2 progenies along with 6 parents were grown in a randomized complete block design with four replications during 2006-07. The plots consisted of four rows 5 m long and 40 cm apart. The distance between plants on each row was 5 cm resulting in approximately 400 plants per plot, which were sufficient for F_2 genetic analysis. Crop management factors like land preparation, crop rotation, fertilizer, and weed control were followed as recommended for local area. All the plant protection measures were adopted to make the crop free from insects. Plant height, first siliqua height, siliquae per main axis and siliquae per plant were recorded based on 20 randomly plants of each plot. Seed yield (adjusted to $kg \times ha^{-1}$) was recorded based on two middle rows of each plot. The combining ability analysis was performed using mean values their F_2 generation along with parents by using Cockerham (1963) method. To determine the adequacy of the additive dominance model, and to assess the validity of some of the assumptions underlying the model, preliminary analysis of the data were done following Hayman (1954a,b) and Jinks (1954). From the data set of P parents with means of parents (m_{lo}) and means of P^2 offsprings (m_{lI}), variance of components of each array (V_r),the mean of variance of components of each array (V'_r), variance components of array means ($V_{r'}$), the covariance of the parents with their offspring in each array (Wr), the mean of covariance of the parents with their offspring in each array (W'_r) and the variance of parental means

(V0L0 = Vp) were calculated. Some of equations related to additive-dominance model with environmental variance (E) are as follow:

$$D = V_p - E$$

$$H_1 = 4 \times V'_r + V_p - 4 \times W'_r - \left(\frac{3 \times P - 2}{P}\right) \times E$$

$$H_2 = 4 \times V'_r - 4 \times V_r - \left[\frac{2 \times (P - 2)}{P^2}\right] \times E$$

$$F = 2 \times V_p - 4 \times W'_r - \left[\frac{2 \times (P - 2)}{P}\right] \times E$$

$$h^2 = 4 \times (m_{1l} - m_{1o})^2 - [4 \times (P - 1) \times P^2] \times E$$

$$F_r = 2 \times \left(V_p - W'_r + V'_r - W_r - V_r\right) - \left[\frac{2 \times (P - 2)}{P}\right] \times E$$

The components under the simple additive-dominance model are: *D*, the component of variation due to additive effects of the genes; H_1 and H_2 the component of variation due to dominance effects of the genes; h is the direction of dominance effects whether it is toward positive or negative; *F*, provides an estimate of the relative frequency of dominant to recessive alleles in the parents, and variation in dominance over the loci. These estimates were obtained by removing the environmental component (Mather & Jinks, 1982). For F_2 data, these parameters resemble to F_1 data with small modifications due to reduced heterozygosity (Jinks, 1956). Non significant of *t* value for *Wr-Vr* indicates its uniformity and therefore validity of genetic hypotheses. The second test for the adequacy of the additive-dominance model is regression coefficient analysis. Failure of this test indicating that: (1) non-allelic interaction (epistasis) is present; (2) genes are not independent in their action, or (3) there is non-random association among parents. All the analyses were performed using Dial98, MS-Excel and SAS version9 softwares (Zhang and Kang1997).

Table 1

The origin and some of characteristics of the rapeseed studied genotypes

Cultivars	Origin	Growth type	Quality of Seed
1-RAS-3/99	Germany	Spring type	Double zero[1]
2-RW008911	Germany	Spring type	Double zero
3-19H	Pakistan	Spring type	Double zero
4-RGS 003	Germany	Spring type	Double zero
5-Option 500	Canada	Spring type	Double zero
6-PF7045/91	Germany	Spring type	Double zero

[1]: Double zero: Fatty acid of oil is less than of 2% and also glucosinolate of meal is less than 30 μM × g^{-1}

RESULTS AND DISCUSSION

Diallel analysis based on Cockerham's method (1963) for the traits including plant height, first siliqua height, siliquae on main axis, siliquae per plant, 1000-seed weight and seed yield is presented in Table 2. Significant mean square of the genotypes for all the traits indicating genetic differences among the genotypes and it be confirmed for the parents and their F_2 crosses regard to significant mean squares of the parents and crosses, respectively. Therefore, the Hayman's model could be used for genetic analysis of these traits. Significant mean squares of general and specific combining ability estimates (GCA and SCA, respectively) were detected for all the traits indicating the importance of additive and non-additive genetic effects for controlling these traits. Similarly, Ramsay *et al.*, (1994) reported that variation for both GCA and SCA were responsible for yield and other quantitative traits in *B. napus*. Significant GCA and SCA effects were also reported for siliquae per main raceme, siliquae per plant, siliqua length, number of seeds per siliqua, 1000-seed weight and seed yield in *B. napus* (Leon, 1991; Singh and Murty, 1980; Thakur and Sagwal, 1997), but in other study (Singh *et al.*, 1995) the importance of additive genetic effects for siliquae per plant and 1000-seed weight was emphasized. Parent *vs* crosses mean square which is indicating average heterosis was significant for all the traits. Shen *et al.* (2005) were reported the importance of additive and non-additive genetic effects yield and yield associated heterosis in rapeseed.

Table 2

Diallel analysis (Cockerham, 1963) of plant height ,yield components and seed yield.

S.O.V	df	M. S					
		Plant heigh [cm]t	First siliqua height [cm]	Siliquae on main axis	Siliquae per Plant	1000-Seed Weight [g]	Seed Yield
Replication	3	195.16**	81.92**	81.92**	1057.04**	0.23	276606.58
Genotypes	20	319.96**	323.56**	323.56**	1478.50**	0.64**	456133.05**
Parents	5	913.03**	679.02**	229.64**	1310.75**	0.52**	467001.77*
Parents vs crosses	1	177.14*	88.92**	1909.72**	5255.60**	0.60**	2505191.17**
Crosses	14	118.35**	216.29**	218.79**	1266.32**	0.68**	307151.27*
GCA	5	111.39*	195.21**	305.49**	1142.03**	1.27*	162317.28
SCA	9	112.21*	228.08**	170.63**	1335.36**	0.36*	387614.60*
Error	60	43.86	11.45	11.46	234.57	0.08	146217.31

* and **: Significant at the 5% and 1% levels of probability, respectively

The remaining three assumptions i.e. no multiple allelism, absence of non allelic interaction and independent distribution of genes among the parents were observed through two tests i.e. uniformity of *Wr-Vr* and joint regression analysis. Both the tests validated additive-dominance model for all the traits except seeds per siliqua therefore the model was considered completely adequate for these traits except seeds per siliqua for which one test only validated indicating the model was partially adequate (Table 3).

Significant value of additive component (D) which is indicating the importance of additive genetic effect was detected for all the traits except siliquae per plant and seed yield (Table 4). Dominance components (H_1 and H_2) were significant for all the traits except plant height for which only H_1 was significant. This result was also supported by the result of Cockerham's method (1963) with significant GCA and SCA mean squares for all the traits. The positive value of H_1-H_2 for all the traits indicating un quality of frequency of the alleles with significant positive and negative effects among the parents for controlling these traits. The value of $(H_1/D)^{0.5}$ was exhibited more than unity for all the traits which indicating the control of over dominance gene actions for these traits. $H_2/4H_1$ was less than 0.25 for all the traits except seed yield denoted asymmetry at loci showing dominance which evidenced the asymmetrical distribution of genes as assumed in Hayman's analysis of variance for the traits except seed yield. Non deviation of $H_2/4H_1$ for seed yield indicating symmetrical distribution of genes with positive and negative effects among the parents for this trait. The overall dominance effect was determined by h^2 estimates, which gave the total sum over all the loci in a heterozygous state was significant for first siliqua height, siliquae per plant and seed yield. The proportion *KD/KR* of all the traits were greater than unity indicated the excess of dominant genes than recessive ones among the parents. The h^2/H_2 ratio denotes an approximate number of genes or groups of genes controlling the traits, i.e., exhibiting dominance, and was more than one for first siliqua height and siliquae on main axis. Similar nature of gene action for yield associated traits had been reported in rapeseed by Satija *et al.* (2001), and Singh *et al.* (1995). Straight forward selection from the segregating population of the characters does not seem to be possible, the genetic variation existed in these traits could be improved successfully following reciprocal recurrent selection. Furthermore these traits in which over dominance was involved may advantageously to be utilized by the breeders to develop hybrid, as suggested by Khan *et al.* (2005). However, before deriving some conclusive inferences, this information must be substantiated.

Broad-sense heritability estimate ranged from 0.95 to 0.71 related to first siliqua height and seed yield, respectively. This implied that a high estimate of genetic variance and a low estimate of environmental variance for these traits in rapeseed genotypes. First siliqua height, 1000-seed weight and plant height with 0.95, 0.89 and 0.82 broad-sense heritability, respectively were less affected by environment than the other traits. Narrow-sense heritability estimate was varied from 0.33 to 0.05 related to first siliqua height and 1000-seed weight, respectively.

Table 3

Uniformity of Vr-Wr and regression coefficient test for different traits of diallel crosses of rapeseed

Traits	.t- test for uniformity WR-WR	Regression analysis (.t value of b)		Conclusion
		H0: b=0	H1: b=1	
Plant height	1.04	0.59	0.71	Model was adequate shown by three tests
First siliqua hight	0.62	3.72**	1.17	Model was adequate shown by three tests
Siliquae on main axis	0.09	1.79*	1.15	Model was adequate shown by three tests
Siliquae per Plant	0.45	1.22	0.71	Model was adequate shown by three tests
Seeds per Siliqua	2.54	3.15**	-1.18	Model was adequate shown by three tests
1000-Seed Weight	0.44	1.07	079	Model was adequate shown by three tests
Seed Yield	1.25	1.80*	-0.08	Model was adequate shown by three tests

* and **: Significant at the 5% and 1% levels of probability, respectively

Table 4

Statistical indices and genetic parameters for plant height, yield components and seed yield

Statistical indices and genetic parameters	Plant height [cm]	First siliqua height [cm]	Siliquae on main axis	Siliquae per Plant	1000-Seed Weight [g]	Seed yield [kg × ha^{-1}]
D	215.5**±41.7	71.2**±16.8	0.11**±0.01	2.74±1.61	4.64**±1.74	9187±9014
F	255.2**±101.8	53.43±41.21	0.10±0.09	3.16±3.92	6.95**±4.27	3450±22022
H_1	230.9**±105.8	245.89**±42.81	0.33**±0.10	18.19**±4.08	20.06**±4.44	71594**±22884
H_2	147.1±94.6	208.80**±38.25	0.28**±0.01	15.52**±3.64	17.22**±3.96	70422**±20443
h^2	21.6±63.6	307.39**±25.74	7.42±6.16	14.51**±2.45	0.26±2.67	67829**±13759
H_1-H_2	83.80	37.07	0.05	2.67	2.84	1172
$H_2/4H_1$	0.16	0.21	0.21	0.21	0.22	0.25
KD/KR#	3.67	1.50	1.71	1.56	2.12	1.14
.h^2/H_2	0.15	1.47	26.50	0.93	0.02	0.96
$HB \times S$	0.82	0.95	0.79	0.73	0.89	0.71
$HN \times S$	0.31	0.33	0.20	0.16	0.05	0.12
$(H1/D)^{0.5}$	1.03	1.86	1.74	2.57	2.07	2.79

#: $[(4DH1)^{0.5} + F]/[(4DH1)^{0.5} - F]$
**: Significant at the 1% level of probability

The scatter of array points along the regression line for different characteristics suggested the existence of genetic diversity in the parental material meaning thereby that sufficient variation is present in the plant material which could be explored by following an suitable breeding program. The distribution of parents along the regression line of Wr on Vr for all the traits are presented in Fig. 1 to 6. A zero, positive or negative intercept indicates complete, partial or over-dominance gene action, respectively. Parents closer to the origin possess more dominant alleles and those farther from the origin contain more recessive alleles for the respective traits.

For plant height, the graphical presentation (Fig.1) exhibited that regression line passed above the point of origin depicting partial dominance type of gene action. The distribution of array points on regression line revealed the concentration of dominant genes in the parent PF7045/91 and while the parent RGS003 possessed maximum recessive genes. Due to high amount of plant height was related to RGS003 (Table 5), therefore recessive genes had more important role in controlling plant height for these genotypes.

The regression line for first siliqua height cut the *Wr* axis below the origin in the negative zone, suggesting the overdominance type of gene action (Fig. 2); this is also supported by the greater than unity $(H1/D)^{0.5}$ ratio (Table 4). The distribution of array points along the regression line indicated that RW008911 and RGS003, being closest to the origin, contain maximum dominant genes, while Option500, being farthest from the origin, had maximum recessive genes.

For siliquae on main axis, the regression line passed the *Wr* axis below the origin, indicating over- dominance gene action (Fig. 3), which is evidenced by the greater than unity $(H1/D)^{0.5}$ ratio (Table 4). Based on the array points on the regression line, 19H had maximum dominant genes, being closest to the origin, while RW008911 possessed recessive genes, as evidenced by its distant position from the origin. The means performance of 19H and RW008911 for this trait were 46.33 and 33.10, respectively, therefore it seems that high amount of siliquae on main axis was more affected by recessive genes.

Over dominance was noted for siliquae per plant as regression line intercepted Wr axis below the point of origin (Fig. 4), which is evidenced by the greater than unity $(H1/D)^{0.5}$ ratio (Table 4). The parents 19H and RGS003 being closer to the origin contained maximum dominant genes and the parents PF7045/91 and Option500 possessed maximum recessive genes as they were farther from the origin.

The results of graphic analysis for 1000-seed weight (Fig. 5) showed that the intercept of regression line on *Wr* axis was negative suggesting the presence of over dominance type of gene action. The parents RAS3/99 and 19H being closer to the origin possessed maximum dominant genes and the parent Option500 being farther from the origin possessed maximum recessive genes for the trait. The parents RAS3/99 and 19H with means of 4 and 4.25 g had high amount of this trait, therefore high amount of 1000-seed weight was controlled by dominance genes in rapeseed.

For seed yield, the regression line passed the *Wr* axis below the origin, indicating overdominance gene action (Fig. 6), which is evidenced by the greater than unity $(H1/D)^{0.5}$ ratio (Table 4). Based on the array points on the regression line, 19H and RGS003 had maximum dominant genes, being closest to the origin, while RW008911 possessed recessive genes, as evidenced by its distant position from the origin. The means performance of 19H and RW008911 for this trait were 2447.92 and 2505.83 kg × ha^{-1}, respectively. Khan *et al.* (2005) and Amiri-Oghana *et al.* (2009) have also reported a similar type of gene action for seed yield.

Table 5

The means of plant height, yield components and seed yield of six parents of *B.napus*

Parents	Plant height [cm]	First siliqua height [cm]	Siliquae on main axis	Siliquae per Plant	1000-Seed Weight [g]	Seed yield [kg × ha^{-1}]
1-RAS-3/99	165.50	107.39	43.94	130.50	4.00	2635.52
2-RW008911	150.42	94.33	46.73	123.00	3.95	2505.83
3-19H	140.43	88.70	33.10	114.67	4.25	2447.92
4-RGS 003	178.40	122.97	46.20	149.55	3.75	2971.84
5-Option 500	139.26	89.60	24.73	121.67	3.43	2218.58
6-PF7045/91	153.10	100.05	38.65	160.08	3.30	3131.92
LSD(α=0.05)	9.37	4.79	4.79	21.66	0.40	540.77
LSD(α=0.01)	12.18	6.22	6.22	28.16	0.52	703.00

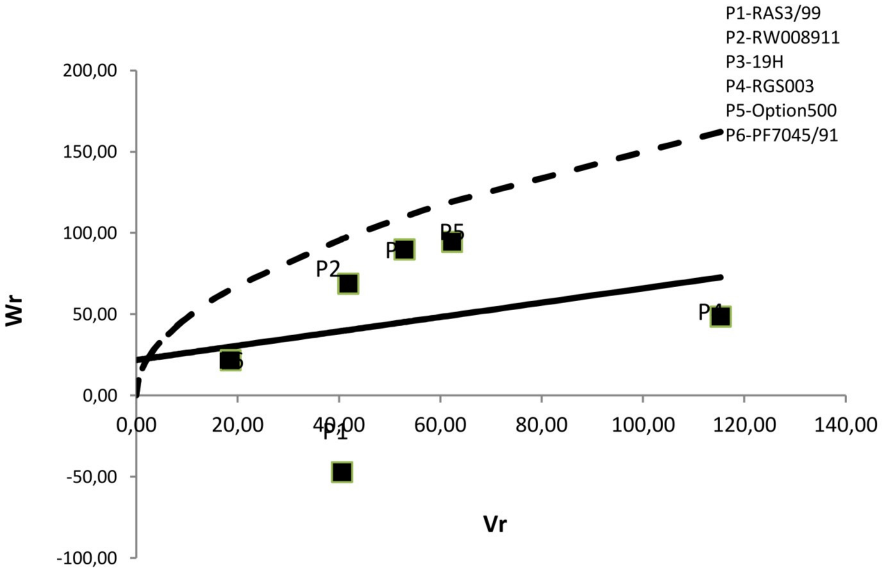

Fig 1. (Vr, Wr) graph for plant height

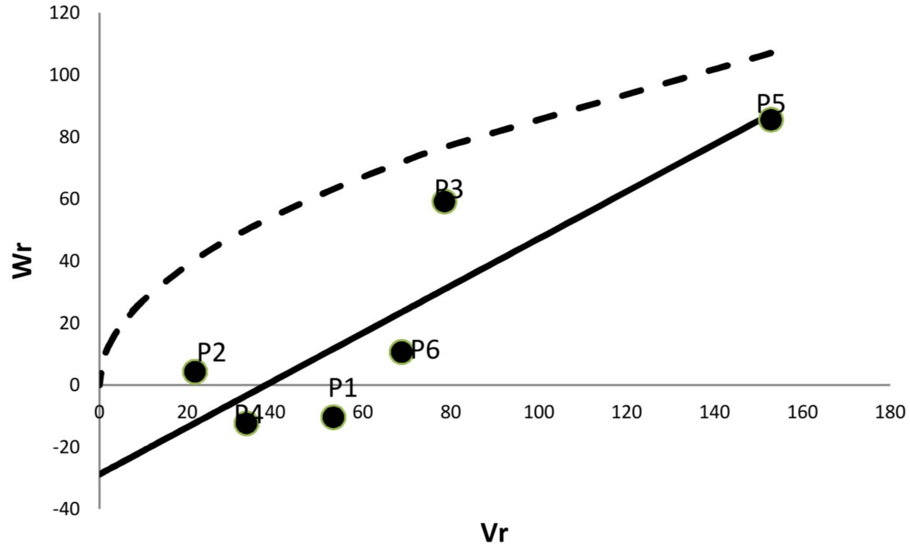

Fig 2. (Vr, Wr) graph for first siliqua height

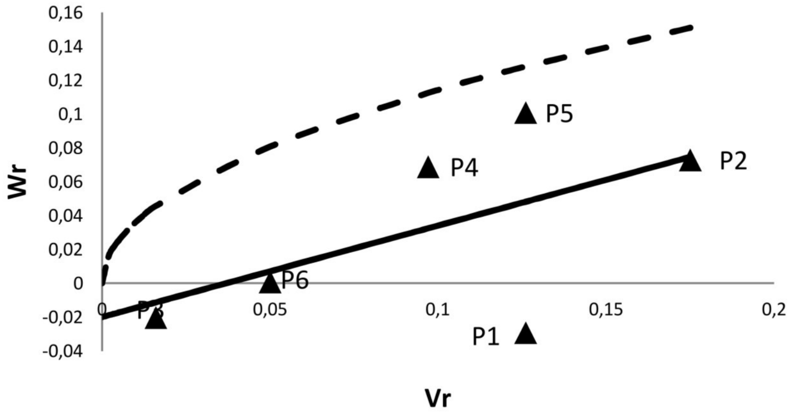

Fig 3. (Vr, Wr) graph for siliquae on main stem

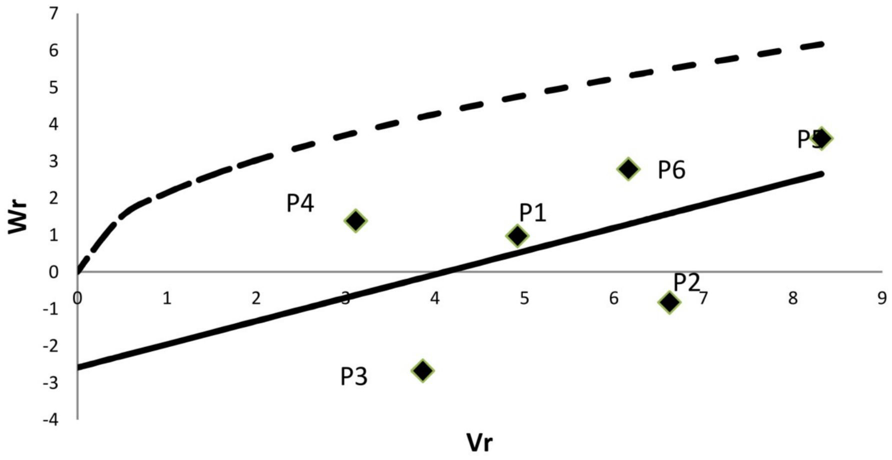

Fig 4. (Vr, Wr) graph for siliquae per plant

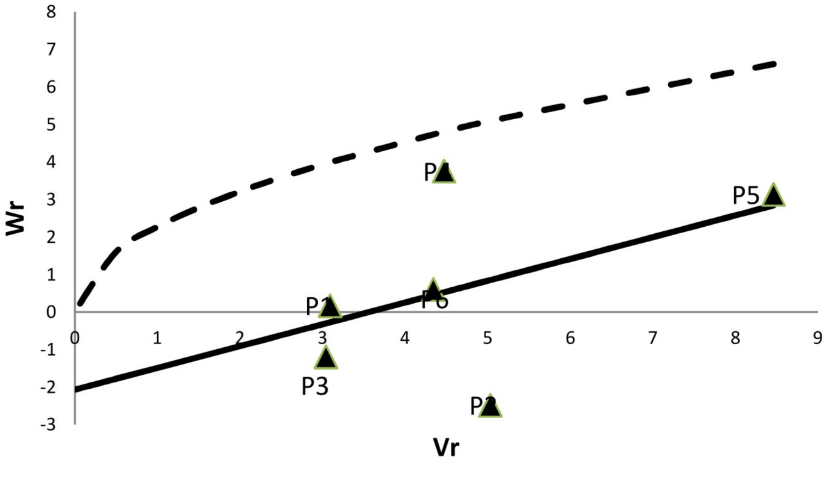

Fig 5. (Vr, Wr) graph for 1000-seed weight

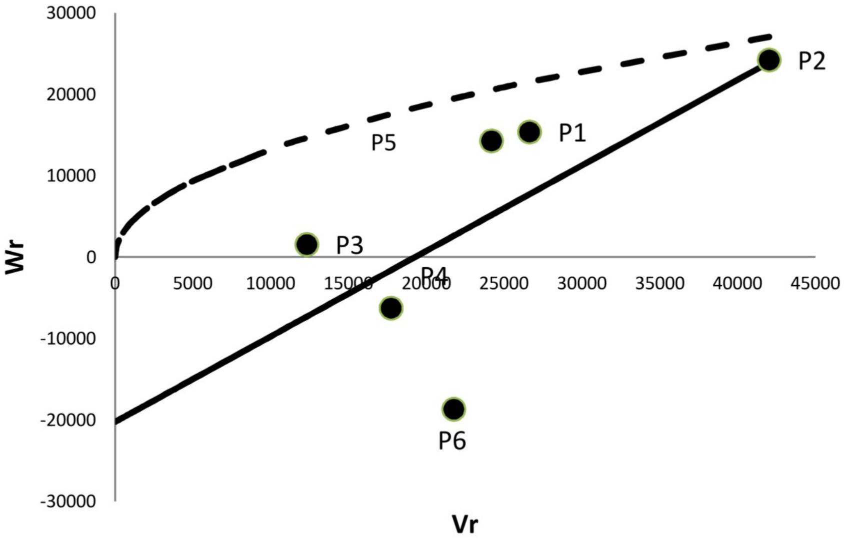

Fig 6. (Vr, Wr) graph for seed yield

CONCLUSION

Significant dominance components (*H1* and *H2*) for all the traits except plant height for which only H_1 indicated the importance of non additive genetic effects for all the traits. This result was also supported by significant GCA and SCA mean squares for all the traits in Cockerham's method. The value of $(H1/D)^{0.5}$ was exhibited more than unity for all the traits which indicating the control of over dominance gene actions for these traits and this type of gene action caused significant average heterosis which was detected by Cockerham's method. Due to low narrow-sense heritability estimates for most of the traits, selection for improving these traits should be carried out in delayed segregating generations. Based on the array points on the regression lines, 19H had maximum dominant genes for siliquae on main axis, siliquae per plant 1000-seed weight and seed yield, being closest to the origin, while Option500 possessed recessive genes for first siliqua height, siliquae per plant and 1000-seed weight, as evidenced by its distant position from the origin.

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