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## Combined and Genetic Analysis for Multiple-disease Resistance to Chocolate Spot and Rust on Faba Bean Yield

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### ABSTRACT

This study showed that fungal foliar diseases i.e., chocolate spot (*Botrytis fabae* Sard.) and rust (*Uromyces viciae fabae* (Pers.) Schroet.) caused considerable yield losses in Egypt ranged from 6.54-26.45%. Therefore, the genetic parameters for seed yield and resistance to two diseases (multiple-disease resistance) were assessed in half diallel crosses between five parents of faba bean and their offspring F<sub>1</sub>'s and F<sub>2</sub>'s, to improve the productivity. Highly significant differences among the studied genotypes were detected, pointing to a wide genetic variability for seed yield and two foliar diseases resistant. Nubaria 1 was recorded as good combiner. The general to specific combining ability (GCA/SCA) ratio revealed great additive effects for the two foliar diseases resistant, on the contrary seed yield. Some crosses had useful or true heterosis above mid-parent and/or better parent for resistance to chocolate spot disease and/or seed yield, indicating to high role of non-additive gene action. Then, Giza 40×Giza 429 and Sakha 1×Giza 429 crosses had best level of resistance to chocolate spot disease with potential yield. The results of genetic variance components revealed that additive component of genetic variability D estimates were greater than dominance effect H<sub>1</sub>, suggesting that additive genetic variance is important than non-additive one. Meanwhile, F and K<sub>D</sub>/K<sub>R</sub> confirm that dominant alleles were more recessive ones but (H<sub>1</sub>/D)<sup>1/2</sup> indicated that dominance for resistance was almost partial. As well as the correlation between parental performance (Yr) and parental order of dominance (Wr+Vr) indicated that the faba bean parents possessed mostly negative genes in dominance form for resistance but possessed positive genes for seed yield. Simple correlation coefficients between two diseases in both generations (-0.95 and -0.94) revealed that selection for either chocolate spot resistance would be accompanied by resistance to rust disease (multiple-disease resistance) and would be effective for the improvement of seed yield per plant in faba bean.

**Key words:** *Vicia faba*, combined, diallel crosses, multiple-disease resistance, chocolate spot and rust, *Botrytis fabae* Sard., *Uromyces viciae fabae* (Pers.) Schroet., heterosis, gene action, correlation

### INTRODUCTION

Faba bean (*Vicia faba* L.) is an important temperate legume crop used as a source of protein in human diets, as forage crop for animals and for available nitrogen in the biosphere (Rubiales, 2010). The yield instability is attributed to various biotic and environmental stresses. Faba bean

plants are attacked by foliar diseases, especially chocolate spot by the fungi (*Botrytis fabae* Sard.) and rust (*Uromyces viciae fabae* (Pers.) Schroet.) that are affecting greatly faba bean production and cause considerable yield losses in this crop (Mohamed, 1982; Khalil *et al.*, 1993; Nagwa, 1996). Increasing seed yield and stability are the main objectives of faba bean breeding programs. This could be achieved by reducing cultivar susceptibility to various limiting factors.

Disease management is based mainly on partial protection. However, development of resistant cultivars is the best practical, most efficient and economical approach (Emeran *et al.*, 2011; Fernandez-Aparicio *et al.*, 2011). Although, a number of sources of resistance to both diseases have been identified (Sillero *et al.*, 2010). Given that crops are attacked by different pathogens in a given season, grown varieties should preferably possess "Multiple-disease resistance" which has been defined as host-plant resistance to two or more diseases. Although, there are examples of multiple disease evaluations in some crops (Hussain *et al.*, 2008; Gurung *et al.*, 2009), the common situation is that screenings are usually performed focusing just on one disease. This "One-at-a-time" approach leads to an inefficient employment of resources and time in the production of commercial varieties with multiple-disease resistance. Not only is it needed to multiply the number of screenings but also, once the appropriate genotypes have been identified, genes from different sources must be combined into the same background. Reducing the number of sources would greatly help in simplifying the whole process. In the case of rust and chocolate spot of faba bean there is hardly any information about accessions possessing resistance to both diseases though their response to simultaneous infection were few studied (Omer, 1984; Sillero *et al.*, 2010).

An additional problem when developing a breeding program for disease resistance is that once a resistant variety is obtained, its response may not be durable (Parlevliet, 2002). Hence, resistances used in breeding should be as stable as possible which implies that in different environments and seasons before choosing them for breeding.

Combining ability effects helps the breeder to identify the best combiners to be crossed either to exploit heterosis or to build up the favorable genes. Several authors mentioned that the significance of both general and specific combining ability effects for important agronomic traits, yield and yield components (Abdel-Mohsen, 2004).

Elliott and Whittington (1979) detected that a high degree of additive genetic control the resistance to chocolate spot disease. Mohamed (1982), Khalil *et al.* (1993) and Nagwa (1996) tested the resistance level of several segregating generation, introductions and cultivars under field conditions at Sakha and Nubaria Research Stations. Some of the tested entries showed high level of resistance. El-Hady *et al.* (1998) indicated that the presence of dominant genes for resistance to chocolate spot in faba bean crosses and the additive gene effect were stable over rang of years and narrow sense heritability estimates ranged from 69-95% which indicated the presence of genetic variability among all studied crosses.

Radwan *et al.* (2010) studied the mode inheritance of resistance to chocolate spot disease and reported some crosses had significant constant negative heterosis according to both mid and better parent for resistance to chocolate spot disease. The genotype ILB938 seemed to be the best combiner for chocolate spot disease resistance.

The major aim for any crop breeding program is the development of good genotypes with an adequate resistance/tolerance to yield-reducing stresses. Fungal foliar diseases i.e., chocolate spot and rust are the most destructive diseases of faba bean and cause considerable dramatic damage losses on faba bean yield in Egypt. The present investigation was carried out to select *V. faba* genotypes possessing multiple-disease resistance to both chocolate spot and rust diseases. To this effect, *V. faba* genotypes was evaluated for study the type of gene action and the mode of

inheritance of resistance to both diseases in five faba bean parents and their F<sub>1</sub> and F<sub>2</sub> generations to improve the productivity.

## MATERIALS AND METHODS

Five faba bean genotypes, i.e., Giza 40 (P<sub>1</sub>), Sakha 1 (P<sub>2</sub>), Nubaria 1 (P<sub>3</sub>), Giza 429 (P<sub>4</sub>) and Triple white (P<sub>5</sub>) were selected on the basis of the presence of wide differences among them with respect to certain economically important traits and their reaction with the foliar diseases. The second and the third genotypes possess variable degrees of resistance to foliar diseases (chocolate spot and rust) while another genotypes are susceptible one as shown in Table 1.

In 2010/2011 season under the house cage, all possible cross combinations excluding reciprocals (half diallel) were made between the five parents (were sown in two planting dates to avoid differences in flowering times and to secure enough hybrids seeds during this season). In the 2011/2012 season, hybrids seeds were sown to obtain the F<sub>2</sub> seeds (self-pollinated) and parents were recrossed to obtained adequate hybrid seeds. Parents and derived 10 F<sub>1</sub>'s were grown in 2012/2013 season under free insect cages at Sakha Research Station. The F<sub>2</sub> seeds from the F<sub>1</sub> plants were raised under cages. All genotypes (parents, F<sub>1</sub> and F<sub>2</sub>) were planted in rows 3 m long, 30 cm apart with one seed spaced at 20 cm. Randomized Complete Block Design with three replications was used. The parents, F<sub>1</sub> and F<sub>2</sub> were planted in 2, 3 and 14 rows, respectively. Seed yield per plant, relative of chocolate spot and relative of rust were recorded for each genotype on 10 guarded plants for parents and F<sub>1</sub> plants and on 25 plants for F<sub>2</sub> from each replicate under greenhouse.

**Greenhouse experiments:** The pathogenic isolate of *B. fabae* used in the present investigation from legume and forage crop Diseases Department, Plant Pathology Research Institute, A.R.C. This isolate proved to be aggressive in previous studies. A spore suspension of *B. fabae* was prepared fresh immediately before application at 2.5×10<sup>5</sup> spores/mL. Plants sprayed with tap water only served as the control. All plants were covered with poly ethylene bags for 24 h to maintain high relative humidity and then kept under greenhouse conditions. Meanwhile, *Uromyces viciae-fabae* (Pers.) Schrot., isolate was collected from Kafr El-Sheikh governorate. Twenty days after sowing leaves of the growing seedlings were inoculated using a mixture of 10 mg of *U. viciae-fabae* freshly collected urediospores and 2 g of talc powder were added (1:20 V/V). Mixture was prepared immediately before inoculation with 10 mL water, to make slurry then mixed again. The mixture was agitated while drops were applied to leaves with brush. To achieve a uniform inoculation, one drop of the mixture was placed on the abaxial surface and then brushed all over the leaflet (Stoddard and Herath, 2001).

**Disease assessment:** The faba bean plants grown, free from chemical treatments were applied to the plants during the assay. The disease severity of chocolate spot and rust diseases was recorded at mid-February and mid-March, respectively using the scale of Bernier *et al.* (1993), as follows:

Table 1: Name, origin and special remarkable characters of faba bean parental genotypes

Parent	Origin	Special remarkable
P <sub>1</sub> (Giza 40)	Egypt	Early mature and high susceptibility to foliar diseases
P <sub>2</sub> (Sakha 1)	Egypt	Early mature and resistant to foliar diseases
P <sub>3</sub> (Nubaria 1)*	Egypt	Late mature, large seeded type and high resistant to foliar diseases
P <sub>4</sub> (Giza 429)	Egypt	Tolerant to <i>Orobanche</i> and high susceptibility to foliar diseases
P <sub>5</sub> (Triple white)	Sudan	Early mature, small seeded type and susceptible to foliar diseases

\*Selected from Spanish genotype Rena Blanca

• **Chocolate spot scale:**

- 1 = No disease symptoms or very small specks (highly resistance)
- 3 = Few small disease lesions (resistant)
- 5 = Some coalesced lesions, with some defoliation (moderately resistant)
- 7 = Large coalesced sporulating lesions, 50% defoliation and some dead plants (susceptible)
- 9 = Extensive, heavy sporulation, stem girdling, blackening and death of more than 80% of plants (highly susceptible)

• **Rust scale:**

- 1 = No pustules or very small non-sporulating flecks (highly resistant)
- 3 = Few scattered pustules covering less than 1% of the leaf area and few or no pustules on stem (resistant)
- 5 = Pustules common on leaves covering 1-4% of leaf area, little defoliation and some pustules on stem (moderately resistant)
- 7 = Pustules very common on leaves covering 4-8% of leaf area, some defoliation and many pustules on stem (susceptible)
- 9 = Extensive pustules on leave, petioles and stem covering 8-10% of leaf area, many dead leaves and several defoliation (highly susceptible)

**Statistical analysis:** Analyses of variance (ANOVA) were carried out for the evaluated generation with and without infection. Before running the combined analysis, Bartlett's test was used to satisfy the assumption of homogeneity of variances. The analysis of variance for combining ability and estimated of genetic effects were done following the technique of Griffing (1956) Method II, Model I. In this approach, the combining ability were estimated which, in turn, would be translated into different genetical components, such as additive and non-additive gene action under certain assumptions.

Heterosis was determined as outlined by Foolad and Bassiri (1983). Appropriate t-test was made for the significance of the  $F_1$ 's from the mid and better parent (heterobeltiosis superiority of  $F_1$  hybrids over the best parent) values (Wynne *et al.*, 1970). The components of genetic variance were estimated according to Hayman (1954a, b).

**RESULTS AND DISCUSSION**

**Combined analysis of variance for yield:** The results of combined analysis of variance in both  $F_1$  and  $F_2$  seed yield (Table 2) showed significant differences among the two different infection types

Table 2: Mean squares of combined analysis of variance for both  $F_1$  and  $F_2$  yield traits under different infection types and genotypes

SOV	df	Yield	
		$F_1$	$F_2$
Replication	2	0.64	0.85
Infection	1	1395.55**	874.23**
Error	2	345.10**	200.47**
Genotype	14	445.12**	396.84**
Infection* Genotype	14	10.51**	13.55**
Error	56	2.11	2.66
Total	89		

\*,\*\*Significant at the 0.05 and 0.01 probability levels, respectively

(with/without), therefore, infection severity affect the yield and productivity (Khalil *et al.*, 1993; Nagwa, 1996). In respect to genotypes in both F<sub>1</sub> and F<sub>2</sub>, significant differences were detected for yield trait which indicate the presence of sufficient genetic variability among genotypes since as can be exploited in faba bean breeding program for improving yield and other traits (Farang and Afiah, 2012). In terms of the interaction between infection and genotypes, there were significant differences for yield trait, meaning the different effects range of infection on genotypes yield.

Results in Table 3 revealed comparison of estimates of faba bean seed yield under infection conditions (with/without) for chocolate spot and rust and calculated yield losses percentage in F<sub>1</sub> and F<sub>2</sub> genotypes. Results showed that Sakha 1 (P<sub>2</sub>) parent had the highest seed yield (56.00 and 45.30 g) under infection free or with infection, respectively. While, F<sub>1</sub> crosses P<sub>3</sub>×P<sub>5</sub> and P<sub>3</sub>×P<sub>4</sub> recorded (64.20 and 53.40) and (63.10 and 53.30 g) without infection and with infection, respectively. But F<sub>2</sub> cross P<sub>1</sub>×P<sub>3</sub> had the highest seed yield (62.40 and 60.87 g) under infection free or with infection, respectively.

Therefore, chocolate spot and rust diseases could affect greatly the faba bean production by losing 26.45% of this crop yield, these results are harmony with those obtained by Mohamed (1982), Khalil *et al.* (1993) and Nagwa (1996) who reported that infection recorded considerable yield losses in Egypt. It is worth mentioning that the Nubaria 1(P<sub>3</sub>) parent and P<sub>1</sub>×P<sub>3</sub> cross had the largest seed yield and the lowest loss in F<sub>2</sub> seed yield at all, followed by P<sub>1</sub>×P<sub>4</sub> cross, meaning that this cross was more resistant or tolerant to chocolate spot and rust diseases.

**Diallel analysis and its genetic components for diseases:** In diallel hybrids, such information about general and specific combining ability for parents and their hybrids may be helpful breeders

Table 3: Comparison of estimates of faba bean seed yield under infection conditions (with/without) for chocolate spot and rust and yield losses% in F<sub>1</sub> and F<sub>2</sub> genotypes

Genotype	Seed yield without infection		Seed yield with infection		Mean yield losses (%)	
	F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>
P <sub>1</sub> (Giza 40)	36.67		26.97		26.45	
P <sub>2</sub> (Sakha 1)	56.00		45.30		19.11	
P <sub>3</sub> (Nubaria 1)	40.20		37.57		6.54	
P <sub>4</sub> (Giza 429)	46.50		38.03		18.22	
P <sub>5</sub> (Triple white)	36.20		27.57		23.84	
P <sub>1</sub> ×P <sub>2</sub>	41.70	36.27	34.70	30.40	16.73	16.18
P <sub>1</sub> ×P <sub>3</sub>	42.10	62.40	39.87	60.87	5.23	2.45
P <sub>1</sub> ×P <sub>4</sub>	55.40	51.30	45.87	49.00	17.20	4.48
P <sub>1</sub> ×P <sub>5</sub>	51.20	38.37	44.47	33.00	13.14	14.00
P <sub>2</sub> ×P <sub>3</sub>	36.60	39.47	30.50	32.80	16.60	16.90
P <sub>2</sub> ×P <sub>4</sub>	52.80	49.00	45.07	41.17	14.59	15.98
P <sub>2</sub> ×P <sub>5</sub>	45.30	40.00	34.97	34.67	22.75	13.33
P <sub>3</sub> ×P <sub>4</sub>	63.10	46.60	53.30	36.17	15.49	22.38
P <sub>3</sub> ×P <sub>5</sub>	64.20	40.50	53.40	35.67	16.82	11.93
P <sub>4</sub> ×P <sub>5</sub>	47.70	42.00	39.77	38.80	16.62	7.62
Mean	47.70	44.10	39.82	37.87	16.62	14.63
LSD	2.24	2.41	2.59	3.01		

Table 4: Mean squares obtained from preliminary analysis and combining ability in F<sub>1</sub> and F<sub>2</sub> generations 5×5 diallel in faba bean for yield and diseases resistance

SOV	df	Chocolate spot		Rust		Yield	
		F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>
Genotypes	14	5.48**	5.38**	3.52**	3.12**	201.88**	228.87**
GCA	4	5.50**	3.45**	4.03**	3.02**	41.49**	45.66**
SCA	10	0.36	1.13**	0.03	0.25	77.61**	88.54**
Error	28	0.17	0.25	0.30	0.18	0.80	1.08
GCA/SCA		15.28	3.05	134.33	12.08	0.53	0.52

\*\*Significant at 0.01 probability levels

to identify the best combiners which may be hybridized to build up favorable fixable genes. Therefore, faba bean plants are challenged by fungal pathogens, such as chocolate spot disease and rust diseases which strongly affect crop yield in Egypt. Genetic resistance is considered the most desirable control method since it is more cost effective and environment friendly than the use of chemicals. Thus, genetic parameters offer a great opportunity to improve the knowledge in resistance inheritance mechanisms against faba bean pathogens and identify effective resistance genes against them.

The mean squares and their significance for yield and disease scores of chocolate spot and rust calculated in the F<sub>1</sub> and F<sub>2</sub> generations and parents are presented in Table 4. The preliminary statistical analysis of all genotypes (parents and their possible hybrids) was highly significant in both F<sub>1</sub> and F<sub>2</sub> generations, pointing to a wide genetic variability for yield, chocolate spot and rust resistance which may facilitate genetic improvement using such genetic pools of faba bean. These results were in harmony with those reported by Awaad *et al.* (2005) and Noorka and El-Bramawy (2011).

ANOVA of the diallel data set with respect to faba bean seed yield and its components attributes as well as disease severity, revealed a highly significant general and specific combining ability (GCA and SCA) effects (Table 4). The GCA variance contains additive epistasis effect, while SCA variance contains dominance epistasis as outlined by Griffing (1956). Hence, the significant estimates of both GCA and SCA variances for disease scores of chocolate spot and rust in F<sub>2</sub> generation only and seed yield in both generations suggested that each of additive and non-additive in the nature of gene actions were involved in controlling these characters through all faba bean genotypes. These results confirmed those findings by Ibrahim (2010) and El-Bramawy and Osman (2012) who reported the significant genetic variation among faba bean genotypes (parents and their hybrids) in respect to yield and its components attributes. Moreover, the plant resistance character against the disease severity of chocolate spot and rust possessed a little bit significant difference compared to the above-mentioned characters via yield components. Moreover the variances due to GCA were larger than those for SCA and the ratio of  $\sigma^2_{GCA}/\sigma^2_{SCA}$  exceeded the unity for two diseases resistance in both generations, revealing that the largest part of the total genetic variance associated with resistance being the result of additives types on gene action. This indicated that direct selection could useful for improving resistance traits. In contrast, yield trait was mainly controlled by non-additive (dominance) gene action.

**Degree of disease infection performance:** Results of the present investigation are presented, interpreted and discussed in order to cover the most of the genetical aspects regarding faba bean

Table 5: Mean performance of parents, F<sub>1</sub>'s, F<sub>2</sub>'s for chocolate spot and rust diseases

Genotypes	Chocolate spot		Rust	
	F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>
P <sub>1</sub> (Giza 40)		7.0		6.7
P <sub>2</sub> (Sakha 1)		3.2		3.7
P <sub>3</sub> (Nubaria 1)		2.7		3.3
P <sub>4</sub> (Giza 429)		6.8		6.3
P <sub>5</sub> (Triple white)		6.0		6.0
P <sub>1</sub> ×P <sub>2</sub>	3.9	3.7	5.2	4.9
P <sub>1</sub> ×P <sub>3</sub>	4.0	3.7	4.8	4.3
P <sub>1</sub> ×P <sub>4</sub>	5.3	3.8	6.7	5.3
P <sub>1</sub> ×P <sub>5</sub>	5.7	4.1	6.2	5.1
P <sub>2</sub> ×P <sub>3</sub>	2.9	2.7	3.7	3.5
P <sub>2</sub> ×P <sub>4</sub>	3.7	3.7	5.3	5.0
P <sub>2</sub> ×P <sub>5</sub>	4.7	4.7	4.7	5.0
P <sub>3</sub> ×P <sub>4</sub>	4.3	4.0	5.0	4.0
P <sub>3</sub> ×P <sub>5</sub>	4.7	4.7	4.7	4.0
P <sub>4</sub> ×P <sub>5</sub>	5.7	4.6	5.8	5.3
LSD 5%	1.19	1.44	1.58	1.21

resistance to chocolate spot and rust disease. Chocolate spot and rust disease scale of parents, F<sub>1</sub>'s and F<sub>2</sub>'s is shown in Table 5. Results revealed that the relative ranking scores of tested parental genotypes in descending order for chocolate spot and rust diseases resistance were Nubaria 1 (P<sub>3</sub>) and Sakha 1 (P<sub>2</sub>) as resistant genotypes with (2.7 and 3.3) and (3.2 and 3.7), respectively while Giza 40 (P<sub>1</sub>) and Giza 429 (P<sub>4</sub>) were susceptible checks (7.0 and 6.7) and (6.8 and 6.3), respectively. The absence of complete resistance and susceptibility suggests the involvement of polygenic system (Abo-El-Zahab *et al.*, 1994).

Crosses involving the highly resistant parents exhibited the highest levels of resistance Sakha 1 (P<sub>2</sub>) ×Nubaria 1 (P<sub>3</sub>) with (2.9 and 3.7) and (2.7 and 3.5) in F<sub>1</sub>'s and F<sub>2</sub>'s, respectively. However, crosses involving the susceptible one parent as Giza 40 (P<sub>1</sub>) and Giza 429 (P<sub>4</sub>) showed the least level of resistance (7.0 and 6.8); in F<sub>1</sub>'s Giza 40 (P<sub>1</sub>) ×Triple white (P<sub>5</sub>) and Giza 429 (P<sub>4</sub>) ×Triple white (P<sub>5</sub>) but Giza 40 (P<sub>1</sub>) ×Giza 429 (P<sub>4</sub>) and Giza 40 (P<sub>1</sub>) ×Triple white (P<sub>5</sub>) were susceptible for chocolate spot and rust, respectively. While, in F<sub>2</sub>'s, Sakha 1 (P<sub>2</sub>) ×Triple white (P<sub>5</sub>) and Nubaria 1 (P<sub>3</sub>) ×Triple white (P<sub>5</sub>) but Giza 40 (P<sub>1</sub>) ×Giza 429 (P<sub>4</sub>) and Giza 429 (P<sub>4</sub>) ×Triple white (P<sub>5</sub>) were susceptible for chocolate spot and rust, respectively.

From these findings may suggest that the above mentioned parents (Sakha 1 and Nubaria 1) and F<sub>1</sub> and F<sub>2</sub> crosses (Sakha 1 (P<sub>2</sub>) ×Nubaria 1(P<sub>3</sub>)) may be of value for improving seed yield of faba bean through improvement of resistance to chocolate spot and rust disease. Similar results were obtained by Rehab *et al.* (2012), to chocolate spot resistance.

**Combining ability:** The estimates of GCA effects “gi” listed in Table 6, where differed from one individual parent to another and from trait to trait. The parental genotypes P<sub>2</sub> (Sakha 1) and P<sub>3</sub> (Nubaria 1) had significance negative (favorable) general combining ability effects (gi) in F<sub>1</sub>'s and F<sub>2</sub>'s for chocolate spot with (-0.95 and -0.99) and (-0.71 and -0.82), respectively, as well as rust



Table 6: General Combining Ability (GCA) effects of parents for yield and chocolate spot and rust resistance

Genotypes	Chocolate spot		Rust		Yield	
	F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>
P <sub>1</sub> (Giza 40)	0.67**	0.45	0.71*	0.56*	-2.87**	0.00
P <sub>2</sub> (Sakha 1)	-0.95**	-0.71*	-0.72*	-0.46*	-0.44	0.35
P <sub>3</sub> (Nubaria 1)	-0.99**	-0.82**	-0.91**	-0.92**	1.90**	1.92**
P <sub>4</sub> (Giza 429)	0.63**	0.52	0.61*	0.47*	3.02**	2.00**
P <sub>5</sub> (Triple white)	0.63**	0.57*	0.30	0.35	-1.60**	-4.27**
S.E. (g-g <sub>c</sub> )	0.22	0.27	0.29	0.22	0.45	0.56

\*,\*\*Significant and highly significant at 0.05 and 0.01 level of probability, respectively

Table 7: Percentage of heterotic relative to mid and better parent for yield and diseases resistance

Crosses	Chocolate spot		Rust		Yield	
	Mid parent	Better parent	Mid parent	Better parent	Mid parent	Better parent
P <sub>1</sub> ×P <sub>2</sub>	-22.94*	22.81ns	0.00ns	40.87*	-3.97ns	-23.40**
P <sub>1</sub> ×P <sub>3</sub>	-17.27ns	49.81**	-3.40ns	45.05*	23.55**	6.12*
P <sub>1</sub> ×P <sub>4</sub>	-22.92**	-21.96**	2.62ns	50.37ns	41.13**	20.62**
P <sub>1</sub> ×P <sub>5</sub>	-12.77ns	-5.50ns	-2.60ns	20.83ns	63.08**	61.31**
P <sub>2</sub> ×P <sub>3</sub>	-0.17ns	9.74ns	4.86ns	10.21ns	-26.39**	-32.67**
P <sub>2</sub> ×P <sub>4</sub>	-26.82**	14.69ns	6.60ns	45.23*	8.16**	-0.52ns
P <sub>2</sub> ×P <sub>5</sub>	1.52ns	45.94**	-3.41ns	27.25ns	-4.03ns	-22.81**
P <sub>3</sub> ×P <sub>4</sub>	-8.84ns	62.17**	3.52ns	50.15*	41.01**	40.14**
P <sub>3</sub> ×P <sub>5</sub>	7.73ns	74.91**	0.11ns	40.24ns	63.97**	42.15**
P <sub>4</sub> ×P <sub>5</sub>	-11.61ns	-5.56ns	-6.41ns	-30.83ns	21.24**	4.56ns

\*,\*\*Significant and highly significant at 0.05 and 0.01 level of probability, respectively, ns: Non-significant; \*Significant, \*\*Highly significant

disease scale with (-0.72, -0.91) and (-0.46 and -0.92), respectively. This proved that these parents seemed to be the good combiners for chocolate spot and rust resistance. Also, the parent P<sub>3</sub> (Nubaria 1) and P<sub>4</sub> (Giza 429) showed positive and highly significant value for seed yield. Consequently, it could be concluded that previously mentioned parental genotypes would prospect in faba bean breeding and therefore P<sub>3</sub> (Nubaria 1) may be valuable for improving seed yield and its components including resistance to diseases infection. Similar findings were earlier reported by Ibrahim (2010) and El-Bramawy and Osman (2012).

**Heterosis:** Respective phenotypes of the different cross combinations of faba bean reactions for chocolate spot disease were tested for the expression of mid-parent heterosis or heterobeltiosis (Table 7).

Chocolate spot values revealed that only three crosses P<sub>1</sub>×P<sub>2</sub>, P<sub>1</sub>×P<sub>4</sub> and P<sub>2</sub>×P<sub>4</sub> (×) had negative significant (favorable) mid-parent heterosis -22.94, -22.92 and -26.82%, respectively. While only one cross P<sub>1</sub>×P<sub>4</sub> were significantly negative heterobeltiosis with -21.96. These crosses displayed highly resistance above mid and better-parent value (Radwan *et al.*, 2010). However, there is no useful or true heterosis above mid or best parent for resistance to rust disease. Meanwhile, mid-parent heterosis and heterobeltiosis percentages for seed yield were significantly positive for four crosses P<sub>1</sub>×P<sub>4</sub> (41.13 and 20.62%), P<sub>1</sub>×P<sub>5</sub> (63.08 and 61.31%), P<sub>3</sub>×P<sub>4</sub> (41.01 and 40.14%) and

Table 8: Estimates of genetic variance components and related statistics in both F<sub>1</sub> and F<sub>2</sub> generations for yield and diseases resistance

Genetic parameter	Chocolate spot		Rust		Yield	
	F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>
E	0.17**	0.25ns	0.31**	0.18**	0.801ns	1.08ns
D	4.07**	3.99**	2.18**	2.29**	59.61ns	59.33ns
F	1.63**	3.71**	-0.11ns	1.02**	105.02ns	102.94ns
H <sub>1</sub>	0.94**	3.61**	-0.68**	0.45**	314.81**	375.82**
H <sub>2</sub>	0.63**	2.59**	-0.52**	0.32**	237.49*	297.80**
h <sup>2</sup>	0.98**	3.49**	-0.20**	0.70**	126.66**	43.75**
Bh <sup>2</sup>	96.90	95.35	91.48	94.23	99.26	98.71
Nh <sup>2</sup>	81.89	77.34	87.95	88.53	74.94	62.58
(H1/D) <sup>1/2</sup>	0.48	0.95	0.56	0.45	2.30	2.52
UV(H <sub>2</sub> /4H <sub>1</sub> )	0.17	0.18	0.19	0.17	0.19	0.20
K <sub>D</sub> /K <sub>R</sub>	2.42	2.91	0.93	2.93	2.26	2.05
Yr and Wr+Vr	0.64	0.39	-0.02	0.56	-0.40	-0.44
r <sup>2</sup>	0.41	0.15	0.00	0.31	0.16	0.19
h <sup>2</sup> /H <sub>2</sub>	1.56	1.35	0.39	2.19	0.54	0.15

\*,\*\*Significant and highly significant at 0.05 and 0.01 level of probability, respectively, ns: Non-significant; \*Significant, \*\*Highly significant

P<sub>3</sub>×P<sub>5</sub> (63.97 and 42.15%), respectively, as well as P<sub>2</sub>×P<sub>4</sub> cross with 8.16% for mid-parent heterosis percentages. Crosses involving the resistant parent P<sub>2</sub> (Sakha 1) showed the highest levels of resistance. Results concluded that P<sub>1</sub>×P<sub>4</sub> and P<sub>2</sub>×P<sub>4</sub> crosses had best level of resistance and yield.

**Genetic components of variance:** The separating of the total genetic variance to its parts via additive and dominance gene effects and other statistics derived from these estimates for yield and chocolate spot and rust resistance are tabulated in Table 8. Estimates of the environmental variance (E) were highly significant for chocolate spot in F<sub>1</sub> and for rust resistance in both generations, indicating that the resistance has been affected by environmental factors. Additive components of genetic variability (D) were highly significant in both generations for the resistance scale of two diseases (Elliott and Whittington, 1979). This suggesting the importance of additive variance for the resistance to chocolate spot and rust and selection for resistance traits in segregating generations would be effective. These results were in agreement with those reported by Ibrahim (2010), Rehab *et al.* (2012) and El-Bramawy and Osman (2012). Dominance components of variation (H<sub>1</sub> and H<sub>2</sub>) were significant in both generations for diseases and seed yield. These values differed from zero and H<sub>1</sub> was greater than H<sub>2</sub> in both generations, indicating that positive and negative alleles at loci for all traits were not in proportional equal for parents. Moreover, D values (4.07 and 3.99) and (2.18 and 2.29) were greater than H<sub>1</sub> (0.94 and 3.61) and (-0.68 and 0.45) for chocolate spot and rust, respectively. The last results meaning that additive genetic variance is important than dominance (non-additive) variance for the resistance to diseases, conflict yield trait. The smaller H<sub>2</sub> than H<sub>1</sub> in F<sub>1</sub> indicates that the positive U and negative V allele's frequencies at the loci for the resistance to chocolate spot and rust in question are not equal in proportion in parents. This was reflected in estimates of covariance of additive and dominance effects (F) which were highly significantly positive in both generations for chocolate spot and in F<sub>2</sub>'s for rust that indicate the excess of dominant alleles compared to recessive ones but yield value was insignificant positive.

The values of UV were slightly below the maximum value of 0.25 (symmetrical) as in both generations which arises when  $U = V = 0.5$  overall loci of diseases resistance and seed yield. This indicates that the distribution seemed to be nearly symmetrical distribution of genes with positive and negative effects among the parents as in  $F_1$  and  $F_2$ . The ratio of total number of dominance to recessive genes  $K_D/K_R$  was more than one in both generations for all traits except for  $F_1$  in rust, pointing that the presence of more dominant genes in the parents than recessive ones. These results confirmed with the finding reported by El-Bramawy and Osman (2012).

The average degree of dominance for all loci  $(H_1/D)^{1/2}$  was less than one for diseases resistance, indicating that dominance was almost partial or incomplete in both generations. Meanwhile,  $(H_1/D)^{1/2}$  was more than unity (one) for yield in both generations, these findings indicated the involvement of an over dominance expression. However, the over-dominance observed in such characters may not be an index of true over-dominance, since the degree of dominance could be biased due to linkage, epistasis or both together (Comstock and Robinson, 1952). Overall dominant effects of heterozygous loci ( $h^2$ ) were highly significant confirming that dominance is unidirectional positive or negative in both generations which confirmed the  $H_1$  and  $H_2$  results. This finding was in harmony with the result detected by Lithourgidis *et al.* (2003). The number of effective factors ( $h^2/H_2$ ) was estimated in  $F_1$  and  $F_2$  generations. It may be noted that this value is underestimated either when the dominant effects of all the genes concerned are not equal in size and direction or when distribution of the genes is correlated or when both above two conditions are fulfilled (Jinks, 1954). Bond *et al.* (1972) suggested that resistance was determined by two kind's genes.

Heritability in broad ( $Bh^2$ ) and narrow ( $Nh^2$ ) sense values (Table 8) estimated in  $F_1$  and  $F_2$ . Chocolate spot and rust diseases resistance and seed yield  $Bh^2$  recorded (96.90 and 95.35%), (91.48 and 94.23%) and (99.26 and 98.71%) in  $F_1$  and  $F_2$ , respectively. Meanwhile,  $Nh^2$  recorded (81.89 and 77.34%), (87.95 and 88.53%) and (74.94 and 62.58%) for the same traits in  $F_1$  and  $F_2$ , respectively (El-Hady *et al.*, 1998). The ( $Nh^2$ ) results indicated that the large proportion of total genetic variation due to the additive genetic effects and confirmed that the environmental effects constitute a major portion of the seed yield phenotypic variation. Then, it can improve the resistance to chocolate spot and rust more facility and available than seed yield through selection. The correlation of parental mean performance ( $Y_r$ ) and their order of dominance ( $W_r+V_r$ ) were positive in resistance cases except rust in  $F_1$ 's, indicated that the faba bean parents possessed mostly negative genes in dominance form for resistance but possessed positive genes for seed yield.

The results of study indicated that dominant alleles were more the recessive ones but dominance for resistance was almost partial. And genetic variation due to the additive effects and recorded narrow sense heritability, therefore, selection would be effective in early generations to improve disease resistance of chocolate spot and rust in faba bean through selection among segregating generations.

**Correlation:** Simple correlation coefficients between pairs of studied characters, in both generations, are presented in Table 9. The results revealed that seed yield per plant was negatively and highly significant correlated with chocolate spot (-0.409 and -0.418) and rust infection (-0.458 and -0.394) in  $F_1$  and  $F_2$  generations, respectively. Meanwhile, the two diseases were positively and significant correlated both with other (0.952 and 0.949) in both generations,

Table 9: Correlation coefficients between all possible pair's combination of yield Chocolate spot and Rust disease severity

Trait	Generation	Seed yield	Chocolate spot	Rust
Seed yield	F <sub>1</sub>	1.00	-0.409**	-0.458**
	F <sub>2</sub>	1.00	-0.418**	-0.394**
Chocolate spot	F <sub>1</sub>		1.00	0.952**
	F <sub>2</sub>		1.00	0.949**
Rust	F <sub>1</sub>			1.00
	F <sub>2</sub>			1.00

\*,\*\*Indicate significant at 0.05 and 0.01 level of probability, respectively

respectively. Then, selection for either chocolate spot resistance would be accompanied by resistance to rust disease (multiple-disease resistance) and would be more effective for the improvement of seed yield per plant in faba bean.

## CONCLUSION

Faba bean genotypes uniform with high seed yield potential and resistances to chocolate spot and rust (multiple-disease resistance) are necessary to be considering as a main target for faba bean production. Therefore, breeding efforts in the current study emphasized on enhancement of seed yield components and resistance to leaf spot diseases. Our genetic analysis confirmed that additive and dominance gene action was very important in controlling the behavior of most agronomic characters, including the resistance to both diseases infection. For achieving faba bean genotypes, Sakha 1 and Nubaria 1 as well as their respective crosses were the best genotypes for high yield potential and adapted/tolerant to diseases infection. Once the appropriate genotypes have been identified, genes from different sources must be combined into the same background. The results confirmed that the environmental effects constitute could be a major portion of the total phenotypic variation in the studied traits, especially seed yield and the selection should be effective for certain of these characters.

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