AN ASSESSMENT OF AUTO-AND CROSS-FERTILIZATION AND THEIR IMPACT ON BREEDING SUDANESE FABA BEAN (VICAI FABA L.) CULTIVARS

***Seif, M. Gasim[1], Awadalla, A. Abdelmula[1] and Jamal, E. Khalifa[2]**
[1]Deparment of Agronomy, Faculty of Agriculture, University of Khartoum, 13314, Shambat, Sudan
[2]Hudeiba Research station, Agricultural Research Corporation, Wad Medani, Sudan
*Corresponding author's e-mail: seifgasim@hotmail.com

Abstract

Faba bean (Vicia faba L.) is a partially allogamous grain legume grown for its high seed protein content. Seed yield of the crop is greatly dependant on the pollinating insects which carry out both self- and cross-fertilization. The degree of cross-fertilization and autofertility is a crucial factor in any breeding program. The degree of auto- and cross-fertlization of four faba bean cultivars was assessed at three locations in Sudan using hilum colour as a morphological marker. The variation between cultivars and between locations for the degree of cross-fertilization was extremely small which ranged from 10.6% to 12.7%. The degree of auto-fertility was high in the four cultivars, reaching up to 88%. The Sudanese faba bean cultivars were characterized by a relatively high autofertility which could be improved by availability of pollinating insects or manipulation of the variation in floral biology towards a clear genetic switch of the mode of reproduction to either full autogamy and handling the population for improvement and maintenance by conventional breeding methods or ~100% cross-fertilization and manipulate the variation towards cross-fertilization and handle the improvement of population through heterosis breeding.

Key words: *Vicia faba* L.; breeding, cross-fertilization; auto-fertility; line cultivar; self-pollinated cultivar

Introduction

Faba bean (*Vicia faba* L.) is an insect -pollinated species with a genuine mixed mating system. The crop is a main source of edible protein for food and feed. In addition, it contributes to soil fertility through nitrogen fixation and minimizing the impact on the environment (Palmer *et. al*, 2009).

In Sudan, the productivity of the crop is low (2860 kg/ha) compared to, the productivity in Egypt (3267 kg/ha) (AOAD, 2005), where it is also grown by irrigation. One of the major constraints that limit the realization of full yield potential of faba bean and cause this instability in yield is thought to be the partial dependence of the crop on insects for pollination, since it is a partially cross-fertilizing species. Pollinators help increase seed set and self-pollination, but more importantly enhance cross-fertilization (Richards, 2001).

The average degree of cross-fertilization was reported to range from 12% to 18% at Hudeiba (Salih 1987) and from 14.2% to 17.4% at Shambat (Salih *et al*. 1994). At present, seed stock of the conventional cultivars suffered a great deal of mechanical and genetic mixing with each other which resulted in obvious losses of their agronomic and genetic features.

In order to improve both yield and yield stability of the present Sudanese faba bean cultivars, information about their reproductive mode is essential. The objective of this study was to estimate the degree of auto-fertility and cross-pollination and their effect on improving the present cultivars.

Materials and methods

A field survey was conducted and seeds were collected from faba bean fields at the northern Sudan (Dongla, Hudeiba) and Shambat (north Khartoum) during the harvest time in season 2006/2007. Seeds were collected on the basis of hilum color. Five seeds of each of about 500 white hilum plants and 200 black hilum plants from each of the known cultivars grown in these areas, namely Hudeiba/93, Bassbier, Ed-Damar and Selaim, were harvested individually. Two experiments (Exp. 1 and Exp. 2) were conducted:

Experiment 1, which was carried out in order to estimate the degree of cross-fertilization each population based on hilum color. Hilum colour is a monogenic trait with black is fully dominant and determined by the genotype of the mother plant on which a given seed grew**,** hence, seeds harvested from a plant are uniform as to their hilum color. The white and black hilum seeds of each cultivar were bulked in a ratio of 100 white to 300 black and sown in 2007/2008 season at Dongla, Hudeiba, and Shambat. At maturity period, only white hilum seed were harvested at the three locations and sown in 2008/2009 at Shamabt to estimate the degree of cross-fertilization that occurred at the three locations.

The degree of cross-fertilization was estimated according to Metz *et al*. (1994) as the frequency of heterozygotes in the progeny of homozygous

mother plant since all heterozygous descendents should have originated from cross-fertilization, because homozygous recessive mother plants are homozygous for the recessive allele

Experiment 2: the degree of autofertility was assessed by taking the rest of the white and black hilum seeds of each of the four cultivars from the three locations and sown at Shambat in bee-proof isolation cages, covering an area of 60 m^2 each. Three generations, S_0, S_1 and S_2, were derived and evaluated in 2007/2008, 2008/2008 and 2009/2010 seasons (Table 2) for their degree of autofertility following the method used by Link (1990). In this experiment, 8 plants were grown per plot in two replications. These genotypes produced 3-5 flowers per inflorescence. At the first eight flowering nodes, the number of flowers was reduced to two, resulting in a total of 16 flowers per plant. Other inflorescences and tillers were removed. All flowers were left untripped. Rate of fertilization was calculated as (number of seed containing pods/16) ×100, in this way, the relative proportion of flowers that self-fertilized without being tripped were scored, thus measuring the degree of autofertility (Drayner, 1959).

Results and discussion

Table 1 shows the mean percentage of cross-fertilization of the four faba bean cultivars at each of the three locations (Dongla, Hudeiba and Shambat). The variation between cultivars and between locations for the degree of cross-fertilization was extremely small. The estimates of the degree of cross-fertilization in this study ranged from 5% to 23% for the individual plant of each cultivar at the three locations with an average ranging from 10.6% to 12.7% for both cultivars and locations. Nearly similar degree of cross-fertilization of 12% to 18% was reported at Hudeiba (Salih, 1987) and 14.2% to 17.4% at Shambat (Salih *et al*. 1994) which are in variance with those of Link *et al.* 1994, Suso *et al*. (2001) and Gasim *et al*. (2004) who reported on the effects of genotype and environment on the degree of cross-fertilization. Such low estimates of the degree of cross-fertilization in this material may be due to few and poor activities of pollinators as in case of Shambat where few bees were observed in the field or the cultivars which rather behave predominantly like self-fertilizers. However, this is not a disadvantage since it offers the opportunity to maximize selection intensity and makes breeding easer, associating with reducing inbreeding depression and increasing autofertility (Link 1990), hence ensure seed production in the absence or poor activity of insect pollinators (Drayner, 1959). This is confirmed by the high estimates of degree of autofertility in the four cultivars, which reached up to 88% (Table 2). Therefore, inbred lines could be developed from these cultivars. Still, however, without pollinator visits, most of the faba bean genotypes admit a need of manual tripping to induce successful pollination and fertilization (Link 2006). As a consequence of absence of pollinators, yield of pure self seeds in cages was variable and mostly low. In cages, tripping can be done manually, to substitute for missing pollination thereby allowing true selfing and high seed set, but this is very costly procedure (Link 1990).

Table 1. Mean degree of cross-fertilization of four faba bean cultivars grown at three locations (Dongla, Hudeiba and Shambat) in Sudan

Location/cultivar	No.of tested plants		Degree of cross-fertilization (%)			
	Mother	Progeny	Min	Max	Mean	±sd*
Dongla						
Hudeiba/93	40	1189	5.00	21.43	10.95	4.66
Basabier	52	1663	5.00	22.58	10.64	4.51
Ed-Dammar	40	1221	5.26	21.43	10.96	4.10
Selaim	43	1323	5.00	23.53	11.05	5.12
Mean	**43.75**	**1349**	**5.07**	**22.24**	**10.90**	**4.60**
Hudeiba						
Hudeiba/93	44	1126	6.45	23.33	12.15	5.05
Basabier	42	1158	6.45	20.69	10.60	4.70
Ed-Dammar	42	1103	5.26	20.69	11.40	4.61
Selaim	39	1064	6.45	22.22	11.87	4.79
Mean	**41.75**	**1112.7**	**6.15**	**21.73**	**11.50**	**4.79**
Shambat						
Hudeiba/93	45	1290	5.56	25.00	12.00	4.92
Basabier	37	1167	5.13	20.83	12.71	4.60
Ed-Dammar	35	1078	5.00	25.00	11.11	5.05
Selaim	40	1116	5.00	23.33	11.59	5.14
Mean	**39.25**	**1162**	**5.17**	**23.54**	**11.85**	**4.93**

***Sd= Standard Deviation**

Table 2. Mean percentage of autofertility of open-pollinated (So), once-selfed (S1) and twice-selfed (S2) generations of four faba bean cultivars at Shambat

cultivar	Autofertility/Generation			Mean	LSD (0.05)
	So	S1	S2		
Hudeiba/93	0.92	0.87	0.85	0.88	0.08
Bassbeir	0.88	0.86	0.84	0.86	0.06
Ed-Damar	0.90	0.89	0.85	0.88	0.06
Selaim	0.89	0.86	0.85	0.86	0.07

Therefore, it may be worthwhile to check which insects could be relatively best pollinators for our faba bean in order to multiply and breed them by improving their foraging places and nesting sites. Otherwise, a manipulation of the variation in the floral biology in faba bean towards a clear genetic switch of the mode of reproduction to either ~100% self-fertilization and handling the population for improvement and maintenance by conventional breeding methods (as in case of pea) or to ~100% cross-fertilization and manipulate the variation toward cross-fertilization and handle the improvement of populations through heterosis breeding (as in maize).

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