

## DEVELOPING A SYNTHETIC POPULATION THROUGH SELECTION IN EGYPTIAN CLOVER GENOTYPES (*Trifolium alexandrinum* L.).

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### ABSTRACT

Selection and random cross pollination for fresh and dry forage yields and protein content within and among seven local Egyptian clover genotypes (*Trifolium alexandrinum* L.) was applied for two generations to develop a synthetic population. Syn1 and Syn2 were tested against the base populations and two imported Italian varieties (Nilodi and Sacramento). Enormous improvement was achieved where Syn2 had higher values in all studied traits than the best parent (Gemmeza-1), Syn1 and the imported varieties. The realized gains from selection ranged from 13.2- 34.4% for fresh forage yield, 11.4 to 37.7% for dry forage yield and from 2.8 to 8.9% for protein percentage. Heritability in broad sense were high for seasonal fresh and dry yields (88.7 and 88.2%) and 65.0% for protein percentage. The environmental variations were 11.3, 11.7 and 35.4% for fresh and dry yields and protein percentage. The expected genetic advance for fresh, dry yields and protein percentage were 8.1, 2.3% and 0.8% respectively.

**Keywords:** Synthetic, selection, Egyptian clover, heritability, genotypic, phenotypic and environmental variation, genotypic and phenotypic coefficient of variability.

### INTRODUCTION

Egyptian clover is the main winter leguminous forage crop in Egypt. It is cultivated extensively as an essential source of fodder for animals and improving soil fertility when cultivated after grasses. High attention should be directed to improve the recent varieties and develop new ones to increase the productivity of unit area of land in order to maximize the total production of green fodder which would help in filling the gap between demand and supply of forages.

Developing high yielding cultivars depends mainly upon existing genetic variations among the germplasm under selection. Although plant breeders have made significant improvements in yields of many crops, much work hasn't been done to improve yield potential of Egyptian clover. This may be due to small floral parts which make artificial hybridization difficult. Besides, high degree of self sterility limits the extent to which these plants may be inbred. Therefore, selection procedures may be used to improve forage yield. Johnson and Goforth (1953) reported that four cycles of selection led to increasing yield by 11% in sweet clover. Selection in cross pollinated crops increases the frequency of desirable alleles and leads to produce new genotypes. The word "Synthetic" implies a population of plants artificially produced by the breeder. Synthetic variety is an advanced generation of a seed mixture of strains, clones, inbreds or hybrids among them, maintained for a limited number of generations by open pollination. The

term Synthetic is sometimes incorrectly applied to populations originated from seed mixtures advanced by open pollination without periodic reconstitution. A common use of the synthetic breeding procedure is with forage crops. The theory of the synthetic is to exploit the hybrid vigor from combining superior clones, while avoiding loss vigor from close inbreeding by restricting the number of generations of seed increase (Tysdal and Crandall 1948). Synthetic varieties of forage crops may be developed by combining either strains or individual plants into a composite strain. This is a commonly used procedure in forage crops breeding which originated at the University College of Wales.. However, Radwan *et al* (1983) produced a synthetic variety by selecting 13 superior lots with good combining ability, crossing between the original seed lots and their open pollinated progenies was allowed and the resultant composite seed was considered as the breeder seed of synthetic-79 and the highest actual gain from selection was 13.1% in one cycle and 19.2% in the second cycle of selection.. Younis *et al* (1988) evaluated different selected varieties of Egyptian clover and the results showed that synthetic-79 was slightly higher than the average overall the evaluated populations for total green yield in several locations in two seasons.

Two cycles of selection and cross pollination directed to effective gain in forage yield by 20.58% and 5.11% as reported by Omara and Hussein (1982). Youins *et al* (1986) and Mikhiel (1987) reported that mass selection was effective for improvement forage yield in multi-cut Egyptian clover varieties. The synthetic F2 variety increased dry forage yield by 21.3% in Egyptian clover (Bakheit 1989).

Progress in the breeding programs depends on the magnitude of genetic variability in the population and the extent of heritability of the desirable characters. Variability analysis has been found useful for getting information about the characters that are expected to respect to selection, (Bakheit 1986).

Heritability in broad sense was estimated by Radwan (1970) in Fahl variety, the obtained results indicated that heritability values were 0.46 and 0.54 for fresh and dry yields, respectively. Ali (1971) reported low to medium heritability estimates in Egyptian clover. On the other hand Bakheit (1986) reported high heritability estimates in Egyptian clover for seasonal fresh and dry yields (89.0 and 91.0%) and protein percentage (89.6 %) indicating that these traits were less influenced by environments. These results were in contrast with those found by Radwan *et al* (1983) who reported that heritability of fresh yield ranged from low to medium values.

This investigation aimed to develop a highly productive synthetic of Egyptian clover through two cycles of direct selection among and within highly productive seven local Egyptian varieties. This Synthetic may have higher productive characters than the base populations, otherwise, the synthetic with accumulated favorable alleles from different varieties would be obtained and consider as breeder seed and or a base population for new synthetic variety.

## MATERIALS AND METHODS

This investigation was carried out at Giza Res. Station during the period from 2002 to 2006 to obtain a highly productive synthetic of Egyptian clover out of seven local varieties. These varieties were previously top yielding and high combining ability in several variety trials over different locations and years as reported by Abdel-Galil *et al.* (1998) and Abdel-Gawad (2003). Also, these varieties mostly represent different regions for berseem production in Egypt; Hellaly, Sakha-4 and Gemmeza-1 represent the Delta region.. Giza-6, Giza-15 and SidsSyn. represent the Middle Egypt and Assiut Pop. represents the Upper Egypt region.

In 2002/2003, the varieties were cultivated in seven plots (non replicated trail) in isolation of other clover varieties. Each plot consisted of 25 rows of 3.5m long, 30 cm apart and 20 cm between hills. Visually selection was carried out for the most vigorous seedling per hill and the other seedlings were thinned. Four cuts were taken and random matting was allowed for the selected plants prior to flowering. Seeds were bulked separately for each variety. All the cultural practices were maintained at the optimum levels for maximum productivity.

In 2003/2004, equal amount of seeds from each variety were bulked and cultivated to form the generation of Syn<sub>1</sub>. The procedures and intensity of selection were applied as in the first season and random matting was allowed among the selected plants. Seeds were bulked to form the sub sequent generation. In 2004/2005 season, the same procedures were followed and seeds were bulked to form the second combination generation (Syn<sub>2</sub>).

In 2005/2006 and 2006/ 2007, a Randomized Complete Block Design traits in three replicates were carried out to evaluate the response to selection and cross pollination for two cycles (Syn<sub>1</sub> and Syn<sub>2</sub>) comparing to the base varieties and two varieties from Italy (Nioldi and Sacramente) which were imported prior to the seasons of this investigation.

Plot size was 4.5m<sup>2</sup> ( 1.5x 3.0 m) and seeds were hand drilled in rows, 20 cm apart at seeding rate of 20 kg fed<sup>-1</sup> and all the cultural practices were maintained as recommended for maximum production of yield.

Fresh and dry yields (FY and DY) were recorded (t fed<sup>-1</sup>) and protein percentage was determined according to A.O.A.C. (1980). Dry matter percentage was determined from plot samples of 300 gm of fresh forage. Data of each season were analyzed and as the data were homogeneous according to Bartlett test, the combined analysis over two seasons was carried out according to Gomez and Gomez (1984), genotypic and phenotypic variance ( $\sigma^2_g$  and  $\sigma^2_p$ ) were estimated according to Al-Jibouri *et al* (1958), genotypic and phenotypic coefficient of variability (G.C.V. and P.C.V.) according to Burton (1952) and heritability in broad sense ( $H^2_b$ ) as outlined by Johanson *et al* (1955).

## RESULTS AND DISCUSSION

The analysis of variance for the studied traits (fresh and dry forage yields and protein percentage) over two years, expressed significant differences among the base populations and the two synthetics (Syn<sub>1</sub> and Syn<sub>2</sub>), Table (1). The interaction of Var. x Years was not significant.

**Table 1: The combined analysis of variance for fresh and dry forage yields (t fed<sup>-1</sup>) and protein percentage (%) over two years.**

S.O.V.	df	Mean squares		
		Fresh yield	Dry yield	Protein
Years	1	20.41	1.90	0.875
Rep. / Year	4	5.68	0.457	0.088
Varieties	10	145.96	10.44	2.134
Var. x Y.	10	1.01	0.313	0.263
Error	40	3.08	0.248	0.168

The evaluation of Syn<sub>1</sub> and Syn<sub>2</sub> populations and the imported varieties against the original parents for fresh and dry forage yields and protein percentage over two years is presented in Table (2). Significant differences among the tested entries were found and enormous improvement was achieved after two cycles of selection and cross pollination. Syn<sub>2</sub> located first significantly higher than the best parent (Gemmeza-1), Syn<sub>1</sub> and Nilodi variety by 22.0, 13.2, and 18.7% for fresh forage yield. The realized gain from selection ranged from 13.2 to 34.4 % for fresh yield. Regarding the dry yield, the same performance was detected, where Syn<sub>2</sub> was significantly higher than the best parent (Gemmeza-1) and Noldi variety by 23.9 and 20.0%, respectively.

**Table 2: Means of total fresh and dry forage yields (t fed<sup>-1</sup>), protein (%) and realized gain (RG %) of Syn<sub>2</sub> over two years.**

Var./Char.	FY (t fed <sup>-1</sup> )	RG (%)	DY (t fed <sup>-1</sup> )	RG (%)	Protein (%)	RG (%)
Hellaly	51.3	28.7	7.9	30.7	19.5	8.5
Sakha-4	52.4	26.0	8.2	28.1	19.4	8.9
Gemmeza-1	54.1	22.0	9.2	23.9	20.0	6.1
Giza-6	50.9	29.7	7.1	37.7	19.7	7.5
Giza-15	49.1	34.4	7.3	36.0	19.5	8.5
Sids-Syn.	49.7	32.8	7.8	31.6	19.5	8.5
Assiut Pop.	50.1	31.7	7.8	31.6	19.8	7.0
Sacromente	52.5	25.7	8.6	24.6	20.1	5.6
Nilodi	55.6	18.7	9.5	20.0	20.2	5.4
Syn <sub>1</sub>	58.3	13.2	10.1	11.4	20.7	2.8
Syn <sub>2</sub>	66.0		11.4		21.3	
Mean	53.6		8.6		20.0	
L.S.D.at 0.05	2.896		0.822		0.676	

It is worth to mention that the two imported varieties (Nolodi and Sacromente) had a competitive performance against the local varieties. Nilodi variety recorded 55.6 t fed<sup>-1</sup> of fresh forage yield which was significantly higher than the local varieties except Gemmeza-1 variety. Significant differences were found between Syn<sub>1</sub> and Syn<sub>2</sub> and the best varieties by 5.4, 5.6 and 6.1% for protein percentage. The realized gains from selection indicate that significant responses to selection were achieved for fresh and dry forage yields and protein content. The obtained results are in agreement with the findings of Omara and Hussein (1982), where significant response to selection for forage yield was obtained after the first cycle of selection and the two cycles of selection directed to gain effects in forage yield by 20.6 and 5.1%. Furthermore, the present results are in the line with those obtained by Bakheit *et al* (2007), where they found significant response for two cycles of selection for fresh and dry forage yields and protein content of Fahl clover. Also, Bakheit (1989) reported that the synthetic F<sub>2</sub> variety has increase in the dry forage yield by 21.3%.

Regarding the genetic variances in Table (3), heritability estimates were high for seasonal fresh yield (88.7%); seasonal dry yield (88.2%) and it was medium for protein percentage (65.0%), Table 3). These results indicate that these traits were less influenced by environment and the dry forage yield should be considered in the estimation of the average yield as the heritability estimate of dry yield was slightly larger than in fresh yield. These results are in agreement with the findings of Bakheit (1986), where he found high heritability of fresh forage yield (89.0%) and dry forage yield (91.0%) in fifty six accessions of multi-cut Egyptian clover. On the other hand, the results are slightly contrasted with those found by Radwan *et al* (1983) who stated that heritability of fresh yield ranged from low to medium values in Egyptian clover.

**Table 3: Estimates of genetic parameters of fresh yield (FY), dry yield (DY) and protein percentage.**

Char.	X̄	σ <sup>2</sup> <sub>g</sub>	σ <sup>2</sup> <sub>p</sub>	E.V.	G.C.V.	P.C.V.	H <sup>2</sup> <sub>b</sub>	Ga units	Ga (%)
FY	53.6	24.16	27.24	11.3	9.2	9.7	88.7	4.3	8.1
DY	8.6	1.88	2.13	11.7	15.9	17.0	88.2	0.2	2.3
Protein%	20.0	0.31	0.48	35.4	2.8	3.5	65	0.2	0.8

Genotypic variance (σ<sup>2</sup><sub>g</sub>), phenotypic variance (σ<sup>2</sup><sub>p</sub>), environmental variation (E.V.), genotypic coefficient of variation (G.C.V), phenotypic coefficient of variation (P.C.V), heritability in broad sense (H<sup>2</sup><sub>b</sub>), genetic advance (Ga) and genetic advance as percentage of mean (Ga%).

The environmental variation ((σ<sup>2</sup><sub>p</sub>-σ<sup>2</sup><sub>g</sub>)/σ<sup>2</sup><sub>p</sub>\*100) reflects the influence of environment on the studied traits which were 11.3, 11.7 and 35.4% for fresh and dry forage yields and protein percentage, Table (3). The estimates of expected gains from selection showed that selection within the best entries will lead to an increase of 4.3 t fed<sup>-1</sup> (8.1%) in fresh forage yield, 0.2 t fed<sup>-1</sup> (2.3%) in dry forage yield and 0.2 t fed<sup>-1</sup> (0.8%) in protein yield over the average of all entries. These results are in agreement with those found by Bakheit (1986) and Bakheit (1989).

In conclusion, selection and cross pollination procedures in isolation, seems to be a helpful technique to develop a high productive synthetic population out of selected genotypes. Syn<sub>2</sub> population should be tested over many locations and years in Egypt to evaluate its performance over different environments.

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**إنتاج عشيره تركيبه عن طريق الإختخاب فى بعض عشائر البرسيم المصرى  
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تم الإختخاب للمحصول الأخضر ، المحصول الجاف ونسبة البروتين في ٧ أصناف محلية من البرسيم المصري لدورتين إنتخابيتين للمحصول علي عشيره تركيبه وتم الوصول إلي تحسين كبير في إنتاجية المادة العلفية من خلال استخدام الإختخاب والتلقيح المفتوح فى دورتين إنتخابيتين .

حققت العشيرة التركيبه  $Syn_2$  أعلى قيم للصفات المدروسة متفوقة معنويًا عن أحسن الأبياء (جيزة - ١) والعشيرة  $Syn_1$  والأصناف المستورده. تراوحت نسبة التحسين بين ١٣,٢ - ٣٤,٤% للمحصول الأخضر ، ١١,٤ - ٣٧,٧% للمحصول الجاف و ٢,٨ - ٨,٩% في نسبة البروتين.

وأظهرت النتائج ارتفاع درجة التوريث في المدى الواسع للمحصول الأخضر والجاف (٨٨,٧ ، ٨٨,٢%) و ٦٥,٠% لنسبة البروتين بما يشير إلي انخفاض التأثير البيئي وزيادة تأثير العوامل المضيفة .

كما أوضحت النتائج أن تأثير التباين البيئي على الصفات المدروسة كان بنسبة ١١,٣ ، ١١,٧ ، ٣٥,٤% للمحصول الأخضر والجاف ونسبة البروتين على التوالي.