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GENOTYPIC VARIATIONS IN SOME EGYPTIAN CLOVER (*TRIFOLIUM ALEXANDRINUM L.*) VARIETIES FOR FORAGE YIELD

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ABSTRACT

The recent study's objective was to detect variability in Egyptian multi-cut barseem genotypes via studying ten different commercial varieties regarding forage yield production during two successive winter seasons (2018/19 and 2019/20). These varieties, namely; Hartuor, Narmar, Sinai 2, Giza 6, Helaly, Gemiza, Serw1, Sakha 4, Khadrawy and Meskawi. Four cuttings were taken in each season. The analysis of variance in each season revealed that the mean squares due to berseem genotypes were significant for all studied traits. A wide range of variation was observed for plant height traits). Berseem varieties had fresh seasonal yield ranging from 114.70 to 137.72 and averaging 125.20 ton acre-1 in the first season. In the second season, the genotypes had a mean seasonal new yield of 146.89 with a range of 120.26 to 162.07 ton acre-1. Seasonal forage dry yield of the varieties ranged from 20.72 to 26.53 with a grand mean of 24.31 ton acre-1 in the first season, and it went from 25.10 to 29.84 with a great standard of 28.65 ton acre-1 in the 2nd season. Based on *per se* performance, the variety Khadrawy was identified for total fresh and dry forage yields. The values of PCV for different characters were higher than corresponding GCV values. Estimates of heritability were high for plant height at 3rd cut in the two seasons, plant height at the 1st and 2nd cuts, and seasonal dry yield in the second season. High heritability values coupled with high genetic advance were recorded for plant height at 2nd and 3rd cut in the two seasons, at 4th cut in the first season, and 1st cut in the second season. Genetic advance as percent of the mean (GAM) was found to be the highest for plant height at 1st cut in the second season (21.78 %) followed by seasonal dry yield in 2nd season (16.07 %).

Keywords: barseem clover, forage yield, PCV, GCV, heritability, genetic gain

INTRODUCTION

The Egyptian clover or berseem (*Trifolium alexandrinum* L) is the main winter legume forage crop fed as a common practice in Egypt. Berseem is grown through the winter season from (early October to May) for forage production, and at the end of the season (late April and early May), seed crops have sometimes taken (El- Nahrawy *et al.,* 1997). In Egypt, About 30% of the winter cropped area is devoted to barseem. Berseem is an imperative forage crop and highly nutritional eminence for animal feed (Graves *et al.,* 1996*).* It is a legume with its ability to improve soil nitrogen in Egypt and is of better quality than grasses and alfalfa in protein and mineral contents (Laghari *et al.,* 2000). Besides, berseem is fulfilling more than 60% of animal requirements in green fodder and hay in Egypt and is considered the cheapest feed source (El-Aidy 2003).

Forage and seed yield improvement depends essentially on the prevailing genetic variability. The knowledge of genetic variability present in a given crop species for the character under progress is of paramount importance for any plant breeding program (Allard, 1960). Knowledge of the genetic variability of the local Egyptian clover genotypes was limited. Therefore, there is a need to generate information on genetic variability, genotypic coefficient of variation, heritability, and genetic advance of the Egyptian clover to estimate the progress of their breeding program in the future. Martiniello *et al.,* (1992) evaluated the phenotypic variability and adaptability for bio-agronomic traits in thirty-two berseem genotypes (*Trifolium alexandrinum* L) under field conditions. They found that the range of variation among the bio-agronomic traits was higher when the cut was applied at early flowering. The broader range of variation was with plant height and dry matter traits. Thirty-two accessions of multi-cut Egyptian clover were evaluated under regular irrigation and drought conditions for forage yield by Bakheit and El-Hinnawy (1993). They found that a considerable variation among accessions for both fresh and dry forage yield. El-Nahrawy *et al.,* (2006) estimated the phenotypic variability of new forage yield in some Egyptian clover cultivars under two locations. They reported that the phenotypic coefficient of variation (P.C.V) was low in all fresh forage yield cuts.

Heritability and genetic advance are important selection parameters. Heritability estimate of a character is essential for plant breeders because it provides information on the extent to which a particular name can be transmitted from the parent to the progeny (Allard, 1960; and Poehlman and Sleper, 1995). Similarly, the genetic advance is also considered necessary because genetic advance shows the degree of the gain obtained in character from one selection cycle. Heritability estimates and expected genetic gain are more useful than the heritability value alone in predicting the resultant effect for selecting the best genotypes (Johnson *et al.,* 1955). High genetic advance coupled with high heritability estimates offers the most suitable condition to decide selection criteria (Allard, 1960; Poehlman and Sleper, 1995; Syukur *et al.,* 2012). Radwan and Abo El-Zahab (1972) estimated heritability from a combined analysis of variance of three progeny tests for multi-cut Egyptian clover. They obtained estimates of 26.0, 31.0, 30.0, and 39.0 % for green forage yield of the three successive cutting and seasonal crops, respectively. Ahmed *et al.,* (2015) found that broad sense-heritability percentages were 98.23, 98.7 for fresh forage yield and dry forage yield at the first cutting of berseem clover. Thirteen populations of Egyptian clover were evaluated for forage yield by Badawy *et al.,* (2018). They found a wide range of variability for fresh forage yield and dry forage yield traits. The phenotypic coefficient of variance (P.C.V. %) varied by 0.23 % for plant height to 2.2% for fresh forage yield, and the genotypic coefficient of variation (G.C.V %) 0.22 % for plant height to 2.1% for new forage yield. Bahaa *et al.,* (2018). The genotypic variance relative to the environmental conflict (σ2p / σ2g) was high for the studies traits. The percentage of environmental variation from the phenotypic variance ranged from 8.2% for fresh forage yield to 19.2% for 1000 seed weight. The phenotypic and genotypic coefficients of variation descended from fresh forage yield to 1000-seed weight to the number of florets/ inflorescence to the number of seeds/inflorescence. The broad-sense heritability estimates were 91.8, 86.0, 86.4, and 80.8% for fresh forage yield, the number of florets/ inflorescence, seed/inflorescence, and 1000 seed weight, respectively. Because of these, the present study was done with the objectives to assess genetic variability, heritability, and genetic advance of ten Egyptian clover genotypes for forage yield.

MATERIALS AND METHODS

The present study was carried out at the Experimental Station, Faculty of Agriculture, Cairo University, Giza, Egypt, during two successive winter seasons (2018/19 and 2019/20). In both seasons, ten varieties of Egyptian clover (*Trifolium alexandrinum* L.) were used. It's namely, Hartuor, Narmar (these varieties were developed by agronomy Dept., Fac. of Agric., Cairo Univ.), Sinai 2, Giza 6, Helaly, Gemiza, Serw1, Sakha 4, Khadrawy and Meskawi (varieties developed by the Forage Res. Dept., Field Crop Res., Inst. ARC, Egypt). The experimental design was Randomized Complete Block Design (RCBD), with four replications. The plot size was 16m^2 (4x4). The experiments were sown on November 5th and November 28th for the first and second seasons, respectively. The Seed was sown broadcast with a seeding rate of 20 kg/acre. The recommended cultural practices of growing Egyptian clover were followed to raise average plant growth.

Four cuttings were taken in each season. The first cut was taken 60 days after sowing, and other cuts were taken at 40-day intervals in both seasons. At each cut, plant height (cm) was recorded on ten plants randomly taken from each plot. Whole plots were cut for fresh yield determination and converted to ton acre^{-1}. Samples of 500gm were taken and dried to constant weight in an oven-controlled at 105°C, and percent of dry matter was calculated, and dry forage yield was determined. Seasonal fresh and dry products (ton acre^{-1}) were calculated by summation the result of four cuts at each season.

Data were subjected to regular analysis of variance (ANOVA) according to the method outlined by Gomez and Gomez (1984) using the MSTATC computer program (MSTATC, Michigan State Univ., 1992). L.S.D judged differences between means. Test at 5% probability levels. The variability present in the population was estimated by measure mean, range, phenotypic and genotypic variance. To estimate the phenotypic and genotypic variance, genotypic and phenotypic coefficients of variation were estimated based on the formula Syukur *et al.,* (2012) as follow:

$\sigma^2_e = MS_e / r$

$\sigma^2_g = [(MS_g) - (MS_e)] / r$

$\sigma^2_p = [\sigma^2_g + (\sigma^2_e)]$,

Where: σ^2_e = environmental variance; σ^2_g = Genotypic variance; σ^2_p = Phenotypic variance; MS_e = mean square of error from the analysis of variance; MS_g = mean square of genotypes; MS_e = error mean square;

r = number of replications.

Genotypic coefficient of variation (GCV) = $[(\sigma^2_g)^{1/2} / \bar{X}] \times 100$;

Phenotypic coefficient of variation (PCV) = $[(\sigma^2_p)^{1/2} / \bar{X}] \times 100$,

Where: σ^2 = Genotypic variance; σ^2_p = Phenotypic variance; $\bar{X}$ is grand mean of a character.

Broad sense heritability (h^2_b) of the all traits was calculated according to the formula as described by Allard (1960) as follow:

$h^2_b = [(\sigma^2_g) / (\sigma^2_p)] \times 100$,

Where: h^2_b = heritability in broad sense; σ^2_g = Genotypic variance; σ^2_p = Phenotypic variance.

Genetic advance (GA) was determined as described by Johnson *et al.,* (1955):

$GA = K (\sigma_p) h^2_b$,

Where: K = the selection differential (K = 2.06 at 5% selection intensity); σ_p = the phenotypic standard deviation of the character; h^2_b = broad sense heritability.

The genetic advance as percentage of the mean (GAM) was calculated as described by Johnson *et al.,* (1955) as follow: GAM (%) = $(GA/\bar{X}) \times 100$,

Table 1: Mean squares of plant height for four cuts and seasonal fresh and dry yield at 1st and 2nd seasons.

Source of variation	Degrees freedom	First season						Second season					
		Plant height				Seasonal forage yield (ton acre-1)		Plant height				Seasonal forage yield (ton acre-1)	
		Cuts						Cuts					
		1	2	3	4	Fresh	Dry	1	2	3	4	Fresh	Dry
Reps	3	21.50	52.89	4.09	29.68	50.25	3.110	20.35	10.38	13.38	31.62	84.25	5.68
Genotypes	9	13.21*	26.09*	54.83*	67.45*	9.82*	7.32 *	50.55*	31.77*	21.34*	16.71*	7.70 *	5.35 *
Error	27	36.38	25.53	32.91	59.90	14.81	0.623	26.24	11.96	15.20	41.79	12.35	0.72

* = significant difference at $p < 0.05$.

Table 2: Ranges, means, and L.S.D0.05 of plant height for four cuts and seasonal fresh and dry yield at 1st and 2nd seasons

Trait	Cuts	First Season				Second Season			
		Ranges		Means	L.S.D.0.05	Ranges		Means	L.S.D.0.05
		Max	Min			Max	Min		
Plant height (cm)	1	63.95	70.33	67.5	5.58	53.75	63.00	58.5	5..95
	2	77.91	85.76	80.74	6.08	67.40	75.35	71.10	4.16
	3	78.90	91.18	83.85	7.18	68.88	77.00	73.39	4.683
	4	73.93	96.63	82.99	7.13	92.23	98.25	97.76	5.96
Seasonal forage yield (ton acre-1)	Fresh	114.70	137.72	125.20	4.63	120.26	162.07	146.89	4.23
	Dry	20.72	26.52	24.31	0.95	25.10	29.84	28.65	1.02

Table (3): Estimates of genotypic variation ($\sigma^2 g$), phenotypic variation ($\sigma^2 p$), genotypic coefficient of variation (GCV), phenotypic coefficient of variation (PCV), heritability in broad sense (h2b), genetic advance (GA), genetic advance as percentage of the mean (GAM) for studied traits of berseem at first and second seasons.

Traits / Estimates	First season						Second season					
	Plant height (cm) cut				Seasonal forage yield (ton/acre)		Plant height (cm) cut				Seasonal forage yield (ton/acre)	
	1	2	3	4	Fresh	Dry	1	2	3	4	Fresh	Dry
$\sigma^2 G$	4.11	19.71	46.62	52.48	6.13	0.58	43.98	28.79	17.55	6.27	3.8	5.17
$\sigma^2 P$	13.21	25.55	54.85	67.43	15.96	1.3	50.54	31.78	21.35	16.71	7.68	5.35
GCV (%)	3.00	5.49	8.15	8.73	1.98	3.13	11.34	7.56	5.71	2.56	1.33	7.94
PCV (%)	5.38	6.26	8.52	9.89	3.19	4.69	12.15	7.93	6.29	4.39	1.86	8.06
h2b (%)	31.11	77.14	84.99	77.83	38.41	44.62	87.02	90.59	82.20	37.52	49.48	96.64
GA	2.33	8.03	12.97	13.16	3.16	1.05	12.74	10.57	7.80	3.16	2.79	4.60
GAM %	3.45	9.95	15.46	15.86	2.52	4.31	21.78	14.79	10.66	3.23	1.92	16.07

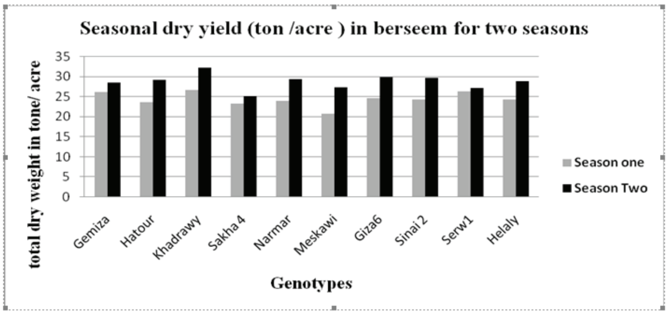

Figure 1: Seasonal dry yields of the ten berseem varieties in 1st and 2nd seasons.

Where: GAM = genetic advance as percentage of the mean, GA = genetic advance, and $\bar{X}$ = grand mean of a character

RESULTS AND DISCUSSION

Variation of mean performance among berseem genotypes: The characterization of the genetic variation in the available germplasm is important for further improving crop yield and imparting resistance to biotic and abiotic stresses (Kour and Singh, 2004). The variance analysis in each season revealed that the mean squares due to berseem genotypes were significant for all studied traits, indicating the presence of sufficient variability in the material studied, and selection could be effective for improving those characters (Table 1).Similar results have also been reported by Bakheit and El-Hinnawy (1993), Badawy *et.al*., (2018) and Salama, *et.al*.,(2020).

The average, minimum, and maximum values of the studied traits are shown in Table 2. A wide range of variation was observed for plant height traits. In the 1st season, the plant height had an overall mean and range of 67.5 and 6.95 to 70.33cm, respectively at first cut. It ranged, at 2nd cut, from 77.91 to 85.76 cm with a grand mean of 80.74cm. At the 3rd cut, plant height ranged from 78.92 to 91.18cm with a grand mean of 83.85cm. The range of plant height trait at the 4th cut was 73.93 to 96.63cm with a grand mean of 82.99cm. In the 2nd season, plant height ranged from 53.75 to 63.00 cm with a grand mean of 58.50cm at first cut, while it ranged from 67.40 to 75.35 cm with a grand mean of 71.10cm at 2nd cut. The range of plant height trait at the 3rd cut was from 68.88 to 77.00 with grand mean of 73.39cm, while at the 4th cut, it ranged from 92.23 to 98.25cm with a grand mean of 97.76cm.

The extent of variability concerning seasonal forage yield in different diverse genotypes of berseem measured in terms of general mean and range for seasonal fresh and dry yields (Table 2). Berseem varieties had fresh seasonal yield ranging from 114.70 to 137.72 and averaging 125.20 ton acre^{-1} in the first season. In the second season, the genotypes had a mean seasonal fresh yield of 146.89 with a range of 120.26 to 162.07 ton acre^{-1}. Seasonal forage dry yield of the varieties ranged from 20.72 to 26.53 with a grand mean of 24.31 ton acre^{-1} in the first season, and it ranged from 25.10 to 29.84 with a grand mean of 28.65 ton acre^{-1} in the 2nd season. A wider range of variation between thirty-two genotypes of berseem for plant height and dry matter was reported by Martiniello *et al.,* (1992).

Seasonal dry yields of the ten berseem varieties in the 1st and 2nd seasons were shown in Figure (1). Concerning the total dry yield of the two seasons, variety khadrawy was the best (p>0.05), producing 26.52 and 32.18 ton acre^{-1} in the 1st and 2nd seasons, respectively. Based on *per se* performance, the variety khadrawy was identified for total fresh and dry forage yields. Significant differences among berseem genotypes for different traits have been reported by Martiniello *et al.,* (1992), Bakheit and El-Hinnawy (1993), El-Nahrawy *et al.,* (2006), Zayed *et al.,* (2011), and Abd El-Naby *et al., (2015)*.

The estimates of phenotypic and genotypic variances, genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV), broad-sense heritability, genetic advance under selection and genetic advance as a percentage of the grand mean for studied traits of berseem varieties in two seasons are presented in Table (3).

The GCV and PCV were computed to access the existing variability in the characters. The GCV values were ranged from 1.33 % for fresh seasonal yield in the 2nd season to 11.34% for plant height at the first cut. Similarly, the PCV values ranged from 1.86% for fresh seasonal yield in the 2nd season to 12.15% for plant height at the first cut in the 2nd season (Table 3). The values of PCV for different characters were higher than the corresponding GCV value. However, GCV was near to PCV for the characters like plant height at 2nd and 3rd cuts in the two seasons, seasonally fresh and dry yields in the second season (Table 3), indicating the high contribution of genotypic effect for phenotypic expression of such characters. A similar result was reported by Abo El-Goud *et al.,* (2015). In other traits, there are relatively wider gaps between the estimate of PCV and GCV showed distinct contributions of environmental factors and genotypic effect for the expression of the traits.

Estimation of heritability is essential for plant breeders as it helps calculate the expected gains from any selection scheme (Falconer and Mackay, 1996). According to Singh (2001) that heritability values greater than 80% were very high, values from 60–79% were moderately high, values from 40–59% were medium, and values less than 40% were low. Accordingly, estimates of heritability were ranged from medium to moderately high for fresh seasonal yield in the two seasons, seasonal dry yield in 2nd season, and plant height at 2nd and 4th cuts in the first season. Estimates of heritability were high for plant height at the 3rd cut in the two seasons, plant height at the 1st and 2nd cuts, and seasonal dry yield in the second season. The characters having high heritability indicated a relatively small contribution of the environmental factors to the phenotype, and selection for such characters could be pretty easy due to the high additive effect. High estimates of broad-sense heritability have also been reported by Bakheit (1985) for plant height and seasonal dry yield and by Zayed *et al.,* (2011) for Dry yield of berseem. Genetic advance (GA) under selection referred to the improvement of characters in genotypic value for the new population compared with the base population under one cycle of selection at a given selection intensity (Singh, 2001). Hence, knowledge on heritability coupled with genetic advances is more beneficial. Estimates of GA values for all characters studied are displayed in Table 3. In the present investigation, high heritability values coupled with high genetic advance (Table 3) were recorded for plant height at 2nd and 3rd cut in the two seasons, at 4th cut in first season, and 1st cut in the second season, indicating these characters were controlled by additive gene effects and phenotypic selection for these characters is likely to be effective. Genetic advance as percent of the mean (GAM) was found to be the highest for plant height at 1st cut in the second season (21.78 %), followed by seasonal dry yield in 2nd season (16.07 %) (Table 3). According to Jonhson *et al.,* (1955), high heritability estimates along with the high genetic advance as per mean is usually more helpful in predicting gain under selection than heritability alone

CONCLUSION

Knowledge of the genetic variability of the Egyptian clover genotypes for the character under improvement is paramount for any plant breeding program's success. A wide range variation was observed for plant height and forage yield traits of studied genotypes of berseem. High heritability values coupled with high genetic advances were recorded for plant height. This is usually more helpful in predicting gain under selection. Based on *per se* performance, the variety Khadrawy was identified for total fresh and dry forage yields.

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