

FERTILITY STUDIES IN EGYPTIAN CLOVER WITH PARTICULAR REFERENCE TO POLLEN STAINABILITY¹

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ABSTRACT

Fertility studies were conducted in Egyptian clover (*Trifolium alexandrinum* L.) during winter of 1979-80. Pollen stainability, pollen production and pollen size were recorded. The range of pollen size was grouped into three different classes. An optimum of stainable pollen production and stainability percentage indicated B₂-1, UPB-101 and JB-1 as the promising strains. The results for correlation coefficient and test of independence indicated no correlation between pollen stainability and pollen size and size of sterile pollen to be independent of the size of fertile pollen. The fertile pollen of the same genotype, however, were observed to be bigger than their sterile counterparts.

INTRODUCTION

Egyptian clover is an important protein rich forage crop. In view of developing superior varieties systematic breeding programmes including hybridization are indispensable. A prior information on fertility status of the parents may substantially provide an insight towards their crossability in hybridization. The present study, therefore, was made to index out the pollen fertility level of some of the promising strains of Egyptian clover (*Trifolium alexandrinum* L.)

MATERIALS AND METHODS

Nine promising genotypes of Egyptian clover were grown during winter of 1979-80. The pollen production, stainability and size studies were made by collecting pollen from fresh flowers around 9.00 a.m. on the glass slides containing Muntzing's mixture of glycerol

and 1 per cent acetocarmine solution. In each slide, twentyfive randomly selected microscopic fields were considered for evaluating the number of pollen grains and stainability after 6 hr staining. The stainability was used to refer as fertility and unstainability as an index of morphological sterility. The shape and size of the pollen grains were recorded from a random sample in each microscopic field in each genotype.

The pollen size was categorised in three groups viz., small ($\leq 15 \mu\text{m}$), medium ($>15 \mu\text{m}$ and $<30 \mu\text{m}$) and large ($\geq 30 \mu\text{m}$), analogous to a classification made by Singh *et al.* (1968). The correlation coefficient between stainability percentage and stainable pollen size, test of independence of size of unstainable pollen on the size of stainable pollen through X^2 , difference in means in the size of stainable pollen and unstainable pollen using Fisher's 't' test were work-

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ed out (Steel and Torrie, 1960; Chandel, 1965).

RESULTS AND DISCUSSION

The shape of almost all the pollen grains from each genotype was found to be spherical with very few oval. The observations (Table I) showed that all the strains except C₃-94 had large sized stainable pollen grains in a big proportion. The size of stainable pollen ranged from 21 to 36 μ m and unstainable ones from 12 to 33 μ m (Table II). The data for variability in the stainable pollen size showed that the five genotypes namely, B₂-1, UPB-101, UPB-102, C₃-94 and Syn-

1 had an equal range of variation of 12 μ m and the remaining ones had a range of 9 μ m. Regarding unstainable pollen size maximum range of variation was recorded in S 99-1. The large variations in size and the substantial presence of large sized pollen have often been considered as being the result of polyploidy (Rangaswamy, 1936; Mukherjee, 1951; Sampat and Ramanathan, 1951 and Nair, 1960 a, b). This corroborated a known origin of a strain S 99-1, to be a tetraploid culture. The pollen grains within the same strain falling in more than one categories of sizes could possibly be due to natural course of evolu-

TABLE I
POLLEN FREQUENCY DISTRIBUTION UNDER DIFFERENT CATEGORIES IN EGYPTIAN CLOVER

| Sl. No. | Genotype | Pollen characteristic | Pollen frequency | | | Total |
|---------|--------------------|-----------------------|------------------|--------|-------|-------|
| | | | Small | Medium | Large | |
| 1 | S 99-1 | (+) | — | 2 | 23 | 25 |
| | | (—) | — | 24 | 1 | 25 |
| 2 | Mescavi | (+) | — | 6 | 19 | 25 |
| | | (—) | — | 25 | — | 25 |
| 3 | B ₂ -1 | (+) | — | 3 | 22 | 25 |
| | | (—) | 3 | 21 | — | 24 |
| 4 | UPB-101 | (+) | — | 5 | 20 | 25 |
| | | (—) | 1 | 24 | — | 25 |
| 5 | UPB-102 | (+) | — | 3 | 22 | 25 |
| | | (—) | 1 | 24 | — | 25 |
| 6 | UPB-103 | (+) | — | 4 | 22 | 26 |
| | | (—) | 2 | 24 | — | 26 |
| 7 | C ₃ -94 | (+) | — | 10 | 16 | 26 |
| | | (—) | 5 | 21 | — | 26 |
| 8 | JB-1 | (+) | — | 9 | 16 | 25 |
| | | (—) | 2 | 23 | — | 25 |
| 9 | Syn-1 | (+) | — | 2 | 23 | 25 |
| | | (—) | — | 20 | — | 20 |

N.B. (+) Stainable
(—) Unstainable

TABLE II

POLLEN GRAIN PARAMETERS IN CERTAIN GENOTYPES OF EGYPTIAN CLOVER

| Sl. No. | Genotype | Average stainable pollen no./microscopic field | Stainability (%) (A) | Size (μ m)* | | Comparison between stainable & unstainable size (μ m) |
|---------|--------------------|--|----------------------|---------------------|-----------------------|--|
| | | | | Stainable (B_1) | Unstainable (B_2) | Difference due to mean ¹ ($B_1 - B_2$) |
| 1 | S 99-1 | 6.72 | 75.00 | 33.00 (27-36) | 24.00 (18-33) | 9.00 |
| 2 | Mescavi | 9.16 | 88.75 | 29.64 (24-33) | 20.88 (18-27) | 8.76 |
| 3 | B ₂ -1 | 12.76 | 95.90 | 31.56 (24-36) | 19.80 (15-24) | 11.76 |
| 4 | UPB-101 | 16.32 | 87.74 | 30.00 (21-33) | 20.64 (15-27) | 9.36 |
| 5 | UPB-102 | 8.16 | 70.88 | 30.32 (24-36) | 20.16 (15-27) | 10.16 |
| 6 | UPB-103 | 8.73 | 60.21 | 31.80 (24-33) | 21.16 (15-24) | 10.64 |
| 7 | C ₂ -94 | 5.56 | 77.20 | 30.12 (21-33) | 18.96 (12-24) | 11.16 |
| 8 | JB-1 | 10.24 | 88.58 | 28.08 (24-33) | 20.64 (15-27) | 7.44 |
| 9 | Syn-1 | 6.28 | 88.70 | 31.44 (24-36) | 18.24 (18-27) | 13.20 |

Statistics

- (i) Correlation coefficient : $r (AB_1) = -0.4347$ (Non-significant)
- (ii) Chi-square Test of independence of B_2 on B_1 : $(\chi^2)^1 = 1.7300$ (Non significant)
 $Z = 2.0129$ (Non significant)
- (iii) Calculated 't' values for $(B_1 - B_2)$ at 48 DF for each strain = > 3.4960 (significant at 0.001 level of probability)

N.B. : *Figures under parentheses characterising the genotypes are variation range in size (μ m).

¹Based on a random sample from each of the 25 microscopic fields.

tion accompanying frequent open pollination with the other cultivated varieties and wild types (Rogers, 1961).

There was no correlation between stainability percentage and size of stainable pollen (Table II). This is to the

contrary of the findings of Nair (1960a). For such phenomenon, one of the reasons could be that all the stainable pollen are not necessarily able to germinate while the unstainable pollen are certainly sterile (Sharma and Sharma, 1965). The

correspondence between size of stainable and unstainable pollen could not be established as the X^2 and Z values were non-significant. These findings indicated that pollen size might not be treated as a fertility index. However, within the strain stainable pollen were found to be bigger than the sterile ones (Table II).

Further, the stainable pollen production and stainability percentage were recorded to be maximum in UPB-101 and B₂-1 respectively. In view of the optimum production potential for fertile pollen and stainability percentage, the strains B₂-1, UPB-101 and JB-1 appeared to show healthy signs of fertility ensuring thereby an effective pollination and in turn better seed setting in the hybridization.

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