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Pollen-pistil Interaction in Wide Crosses Involving *Gossypium hirsutum* L. and *Gossypium arboreum* L.

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Abstract: The competitiveness of pollen tube growth in wide crosses involves *Gossypium hirsutum* and *Gossypium arboreum* was studied in six backcross entries. During the process of pollination and fertilization, physical and physiological constraints led to reduction of pollen tube growth under different levels of pistils. The differences in the number of germinating pollen grains on the stigma among the different crosses are maintained when the pollen tubes penetrate the transmitting tissue and grow along the first half of the style. The differences among entries for pollen tube growth at different levels of pistils was more significant at initial level than at later stages. A reduction in the number of pollen tubes growing down the style was recorded in all the treatment over different levels of pistils. The differences in reduction of 50.34% of pollen tube was noticed in the cross (Sahana×PA 255) ×Sahana while (MCU 12×PA 255) ×MCU12 exhibited higher level of pollen tube proliferation. The crosses (MCU 12×PA 255) ×MCU 12 exhibited significantly lower value of pollen tube proliferation at different levels of pistils. Stylar attrition was determined from the pollen tube not reaching the ovary. Stylar attrition was maximum in the cross (MCU 12×PA 255) ×MCU 12 and is minimum in the cross, (Sahana×PA 255) ×Sahana. Though a very low pollen germination over stigmatic surface noticed in (MCU 12×PA 255) ×MCU 12, lower stylar attrition of 33.53% was reported. A reduction in the number of pollen tubes as they grow along the style was observed. The genetic system of pollen pistil interaction seems to be more conspicuous in the upper part of the style than in the lower half and hence fertilization does not depend uniquely on passive physical or physiological constraints by the pistil but also on the genetic interactions among the male gametophytes and the female tissues.

Key words: Pollen tube, palynology, *Gossypium hirsutum*, *Gossypium arboreum*, pollen tube attrition

INTRODUCTION

Post pollination mating success in plants is based primarily on inter and intrasexual mechanism. During the gametophytic phase, large numbers of both male and female gametophytes are produced in plants. However, only a small proportion of these achieve fertilization. The number of ovules exceeds that of seeds produced due to pollen attrition mechanism[1]. Also in most plant species, the number of pollen grains deposited in the stigma greatly exceeds the number of ovules available for fertilization[2]. Anyhow, the final outcome of the fertilization process is the result of a series of steps that determines which male gametophyte among all the possible candidates is going to fertilize the embryosac. The pollen competition in *Gossypium* is that the degree of competition can affect the number of seeds per fruit and this could interfere with the results obtained when studying the vigour of the resultant sporophyte[3]. The pollen tubes have been observed to follow a fixed pattern of reduction since the initial number of pollen tubes is progressively reduced along the style[4]. The pollen tube and style interaction may result in reduced growth of male gametophyes[5]. In the present investigation, the behaviour of pollen under wide crosses was examined through the extent of pollen attrition. The pollen attrition gives a vivid picture upon the nature of compatibility of wide pollen and pistil.

MATERIALS AND METHODS

The experiment was conducted over two years involving different sets of interspecific crosses at Department of cotton, Tamil Nadu Agricultural University, Coimbatore during 2002-2004. The diploid parent (*Gossypium arboreum*) was used as male parent to dust

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over female parent (*Gossypium hirsutum*) of tetraploid in nature. Flowers were collected at time of anthesis and rubbed over the stigma of female parent that had been emasculated at previous day by removing the anthers and petals to make them unattractive to insects[6].

The treatment consisted in the application of different loads of pollen from different sets of diploid male parent to stigma of tetraploid parent. Five pistils per cross from five different crosses were fixed in formalin:acetic acid: ethanol in the ratio of 1:1:18. The pistils were fixed when pollen tube growth had already reached the base of the style, 3 days after pollination. Pollen tube growth along the style was monitored on squash preparation of pistils washed in distilled water, boiled in 5% sodium sulphate to soften tissues and stained with 0.1% aniline blue in 0.1N K_3PO_4[7,8].

Preparations were examined with under Nikon microphot-Fx microscope with fluorescence attachment, illuminated with 200 w high pressure UV lamp. The observations were taken with B (380-490 nm) and or BG (650 nm) excitation filters in combination with BA 520 or BA 530 barrier filter. Colour photographs were taken with Kodak Gold 400 ASA film either with barrier filter (greenish yellow) or without barrier filter (blue background with bright white fluorescing pollen tube)[9,10]. The data were analyzed in completely randomized design and treatment means were compared with Duncans Multiple Range Test.

RESULTS AND DISCUSSION

Pollen of backcross progenies recorded poor pollen germination though pollen has better pollen fertility values (Table 1). The nature of pollen deposition varied among stigmata for different backcrosses. The backcross, (MCU 12×PA 255) ×MCU 12 and (Sahana×DLSA 8) ×Sahana recorded higher pollen germination of 36 and 30%, respectively. The nature of pollen attrition was assessed by the behaviour of pollen over the stigma and style of the female sex organ. The differences in the number of germinating pollen grains on the stigma among the different crosses are maintained when the pollen tubes penetrate the transmitting tissue and grow along the first half of the style. The differences among entries for pollen tube growth at different levels of pistils were more significant at initial level than at later stages. A reduction in the number of pollen tubes growing down the style was recorded in all the treatment over different levels of pistils. The pattern of reduction of tubes at different levels of pistils was very similar for all the crosses. The differences in reduction of 50.34% of pollen tube was noticed in the cross (Sahana×PA 255) ×Sahana while (MCU 12×PA 255) ×MCU 12 exhibited higher level of pollen tube proliferation. The crosses (MCU 12×PA 255) ×MCU 12 exhibited significantly lower value of pollen tube proliferation at different levels of pistils. The number of pollen tube was found to be in decreasing trend under in all the wide crosses studied. Only a least number of pollen tube able to reach ovary that suggest the extent of compatibility. Of the successful reach of pollen tube to ovary, only a insignificant seed set was observed in all the crosses. While (Sahana×PA 255) ×Sahana had maximum number of pollen tube of 148, (MCU 12×PA 255) ×MCU 12 had the least of 58 pollen tubes (Table 2). Stylar attrition was determined from the pollen tube not reaching the ovary. Stylar attrition was minimum in the cross (MCU 12×PA 255) ×MCU 12 and is maximum in the cross, (Sahana×PA 255) Sahana. Though a very low pollen germination over stigmatic surface noticed in (MCU 12×PA 255) ×MCU 12, lower stylar attrition of 33.53% was reported (Table 3).

A dramatic reduction in the number of pollen tubes as they grow along the style was observed. The observation of results that different numbers of pollen tubes grow along the style depending on the initial number of pollen grains and that the number of pollen tubes is reduced by the same proportion under different treatment suggest that genetic interactions may play a vital role in the regulation of pollen tube attrition. The reduced distribution of pollen tube in the mid style and ovary attribute to greater chances of pollen attrition. The same results were also reported by Cruzan[11] in *Nicotiana glauca* and Scribailo and Barrett[12] in *Pontederia saginata*. Cruzan[13] observed a higher rate of pollen tube attrition in the lower part of the pistil. Pollen competition designate the extent of gene flow and effect of foreign pollen among wide species[14].

Table 1: Proportion mean±SE of pollen grain deposition over stigma

Treatments	Pollen grains in stigma	Germinated pollen grain	Pollen germination (%)	Pollen fertility (%)
(MCU 5×PA 255) ×MCU 5	475±13.16a	138±2.10	27cd	46.88
(MCU 5×DLSA 8) ×MCU 5	383±6.26bc	112±1.83	29c	64.29
(Sahana×PA 255) ×Sahana	293±4.78d	78±1.35	27cd	23.08
(Sahana×DLSA 8) ×Sahana	450±7.37ab	135±2.34	30bc	59.38
(MCU 12×PA 255) ×MCU 12	335±6.47c	128±2.22	38a	40.00
(MCU 12×DLSA8) ×MCU 12	475±7.78a	171±2.97	36ab	35.42

Means followed by a common letter(s) are not significantly different at 5% by DMRT

Table 2: Mean±SE of pollen tubes at different levels of pistil

	Pistil level					
Treatments	Stigma	Transmitting tissue	¼ style	½ style	¾ style	Ovary
(MCU 5×PA 255) ×MCU 5	326±5.65a	288±5.00ab	210±3.64b	175±3.04cd	150±2.60c	35±2.34ab
(MCU 5×DLSA 8) ×MCU 5	296±5.13b	201±3.49c	185±3.21d	170±2.95d	148±2.57cd	125±2.14c
(Sahana×PA 255) ×Sahana	294±5.09bc	252±4.37b	204±3.54bc	188±3.21c	175±3.04a	148±2.57a
(Sahana×DLSA 8) ×Sahana	321±5.57ab	298±5.17a	268±4.65a	200±3.47b	172±2.99ab	140±2.42ab
(MCU 12×PA 255) ×MCU 12	173±3.10c	152±2.63d	135±2.34e	105±1.82e	85±1.47e	58±1.07e
(MCU 12×DLSA 8)×MCU 12	310±5.37ab	287±4.97ab	265±4.57e	225±3.90a	176±3.06a	120±2.09d

Means followed by a common letter(s) are not significantly different at 5% by DMRT

Table 3: Pollen attrition per cent for different wide backcrosses

Treatments	Pistil distribution over stigmatic surface	Pistil distribution inside ovary	Pollen attrition
(MCU 5×PA 255) ×MCU 5	326a	135b	41.41c
(MCU 5×DLSA 8) ×MCU 5	296b	125c	42.23cd
(Sahana×PA 255) ×Sahana	294b	148a	50.34e
(Sahana×DLSA 8) ×Sahana	321a	140ab	43.61d
(MCU 12×PA 255) ×MCU 12	173c	58e	33.53a
(MCU 12×DLSA 8) ×MCU 12	310ab	120cd	38.71ab

Means followed by a common letter(s) are not significantly different at 5% by DMRT

The different backcross entries of wide crosses may provide a useful tool to study pollen tube attrition. Hormaza and Herrero[15] pointed the variability among the stigmata in the number of pollen grains for distribution of pollen tube in the style. The restriction in the distribution of pollen tube may be due to various reasons as pointed out by different authors. Herrero[16] reported that physical limitation could be a constraint of the width of the transmitting tissue of the style. Mulcahy and Mulcahy[17] pointed out that restriction in the nutrient supply and requirement of large amount of nutrients to produce cell wall may contribute to restricted pollen tube distribution. Since the rate of pollen tube attrition is more or less constraint along the pistil, it seems that superimposed on the physical or physiological constraints presented by the pistil is a genetic interaction between the pollen tubes or between those tubes and the style that is contributing in a substantial way to the determination of the attrition pattern. It is also inferred that in the lower part of the pistil, the number of growing pollen tube behave independent of the initial number of germinating pollen grains on the stigma. The genetic system of pollen pistil interaction seems to be more conspicuous in the upper part of the style than in the lower half and hence fertilization does not depend uniquely on passive physical or physiological constraints by the pistil but also on the genetic interactions among the male gametophytes and the female tissues. Cruzan[13], Hormaza and Herrero[15] suggested that differences among pollen donors after pollinations which lead to stylar inhibition of pollen tube growth rather than to differences in pollen vigour.

The progamic phase in wide hybrids of *Gossypium* was generally influenced by genetic interaction and physical constraint involving a reduction in diameter of transmitting tissue and in availability along the style.

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