

## IN VIVO POLLEN GERMINATION STUDIES OF DISTYLOUS *PENTAS LANCEOLATA* VAR. WHITE

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Fresh flower buds of *Pentas* var. white pin morph were pollinated with both pin (self) and thrum pollen grains under controlled conditions. The stigmas were collected after 8h and 24 h and studied using aniline blue fluorescent method. The self-compatibility of pin morph flowers atypical of heterostylous plants is discussed.

**Keywords :** Distyly; *Pentas lanceolata*; Pin; Thrum.

### Introduction

Distyly is a simple inherited polymorphism, in which floral morphs are easily identified under field conditions, long styled pin and short styled thrum. According to Ford<sup>1</sup> population studies using ecological genetic approaches offer attractive opportunities for investigation of natural selection maintenance and break down of heterostyly.

Some of the earlier investigated tropical heterostylus species are *Oldenlandia umbellata*<sup>2</sup>, *Pentas lanceolata*<sup>3</sup>, *Morinda tomentosa*<sup>4</sup> and *Guettarda speciosa*<sup>5</sup> all belonging to Rubiaceae. Heim<sup>6</sup> was the first to notice heretostyly in three species of *Pentas*, while studying the peculiarities and distributions of Rubiaceae in tropical Africa. Extensive study of heterostyly in this genus was carried by Verdcourt<sup>7</sup>, which has revealed that fifteen species exhibit dimorphic heterostyly. *Pentas lanceolata* was one among them, which showed distyly. In the present investigation an attempt was made to access the nature of compatibility / incompatibility exhibited by the pin morphs of *Pentas lanceolata* var. white.

Experimental studies on the pollination biology of heterostylus plants have enabled analysis of the function and adaptive significance of the polymorphism<sup>8</sup>. Research over the last few decades has brought to limelight a significant number of cases where plants with style length polymorphism exhibit various combinations of heterostylus and non heterostylus characters<sup>9a,9b</sup>. The latter include strong self-compatibility, multi allelic incompatibility, monomorphic stamen heights and an absence of ancillary polymorphism.

### Material and Methods

*Pentas lanceolata* var. white - *Pentas lanceolata* are woody erect shrubs growing to a height of four to six meters. The var. white is distylus, producing both pin (long styled) and thrum (short styled) flower bearing plants.

Details of pollen germination and pollen tube growth in pollinated pistils were studied using aniline blue fluorescent methods<sup>10</sup>. The pistils, after twenty hrs of self and cross pollination with pin pollen and thrum pollen, respectively, were fixed in acetic - alcohol (absolute ethanol - glacial acetic acid 3:1) for twenty hrs and then cleared in NaOH at ambient temperature. The pistils, made soft after clearing, were carefully rinsed twice with tap water and mounted on 0.005% aniline blue, prepared in 0.05 molar Na<sub>2</sub>HPO<sub>4</sub> (pH > 8.2). A drop of 50% glycerin was added to the stain to prevent drying of pistils. The pistils were given a longitudinal slit with a scalpel before lowering the coverglass. The tissue was spread by applying gentle pressure on the cover glass. The preparations were observed under fluorescent microscope.

**Table 1.** *P. lanceolata* var. white self and cross-pollinations.

Combinations	% of pollen germination	Tube length(mm) after pollination	
		8h	24h
*W <sub>p</sub> × W <sub>p</sub>	56.6	11.4 ± 0.61	30.3 ± 1.06
W <sub>p</sub> × *W <sub>T</sub>	34.1	56.2 ± 1.23	16.1 ± 0.65

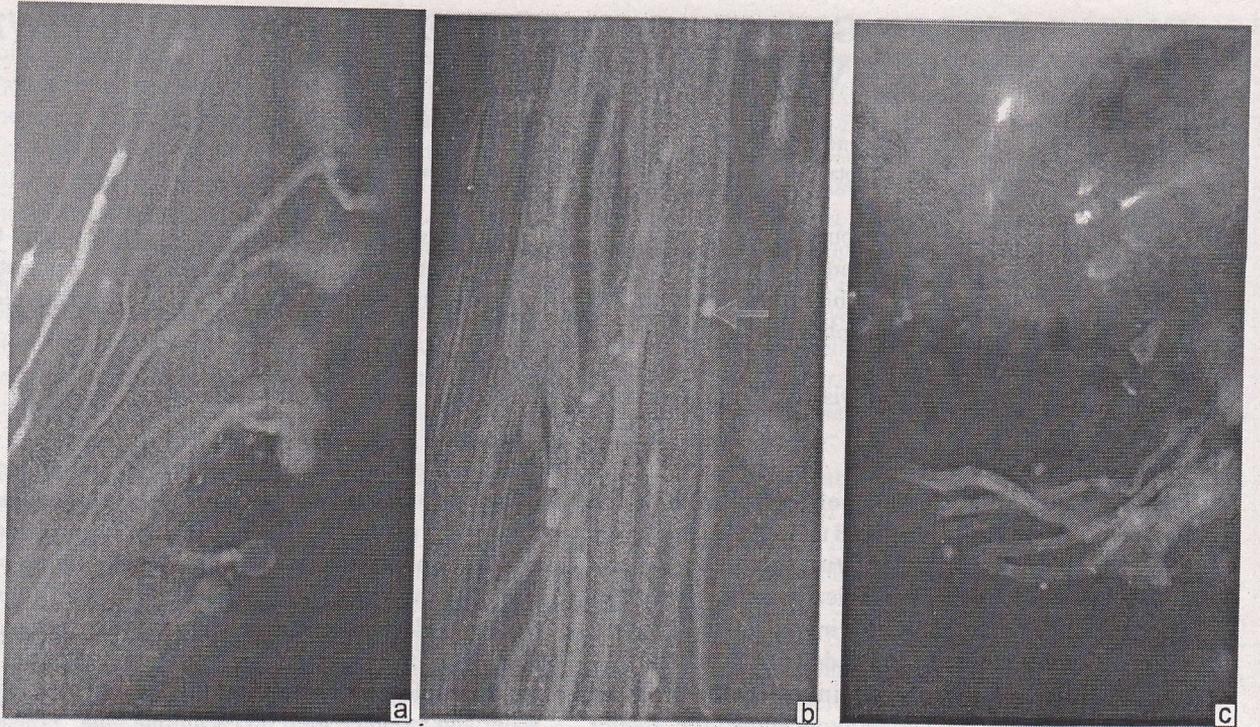
All values represent a mean of six replicates

\*W<sub>p</sub> -White Pin

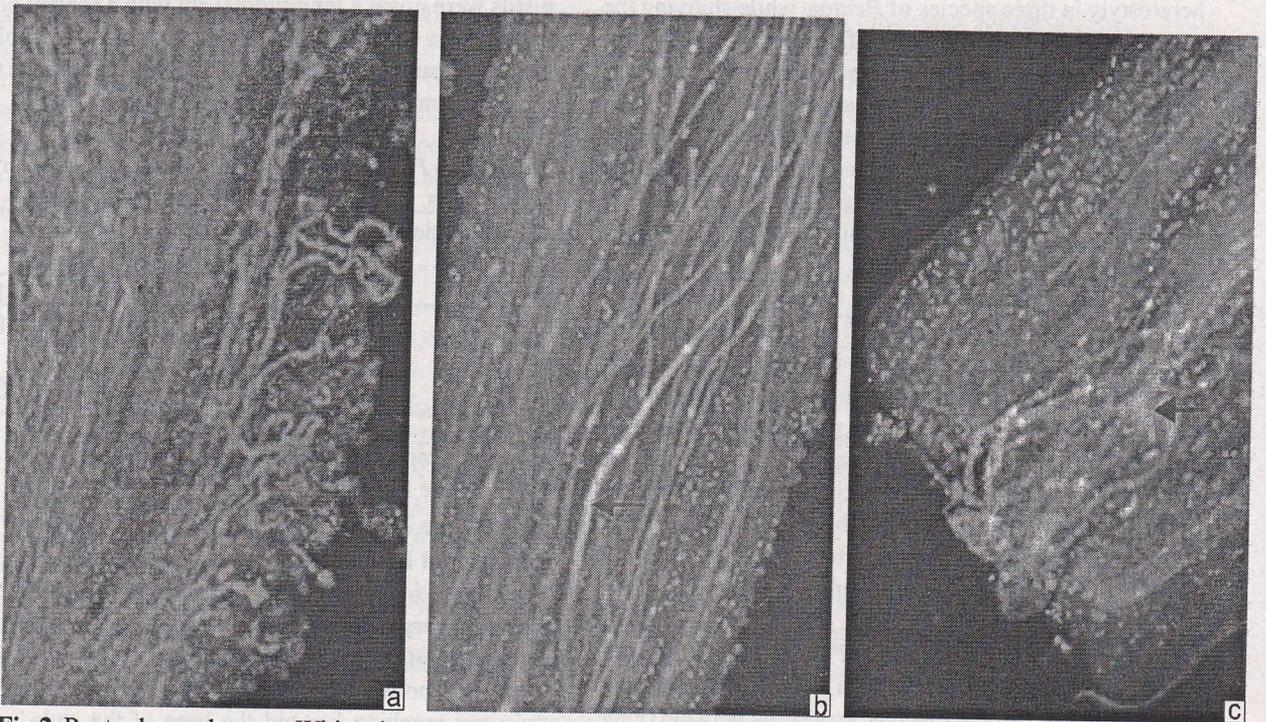
\*W<sub>T</sub> -White Thrum

### Results and Discussion

The stigma of *Pentas lanceolata* var. white, pin morph showed a very good percentage of self-pollen adherence, percentage of pollen germination and mean tube length. Numerous pollen tubes were seen to grow through the style and reaching beyond the base of the style, which showed callose plugs interspersed at regular and large



**Fig.1.** *Pentas lanceolata* var. White pin self pollination ; a. Pin pollen on pin stigma; b. Pollen tube passing through style showing low callose deposition; c. Pollen tube extending out of the base of style after compatible self pollination.



**Fig.2.** *Pentas lanceolata* var. White pin cross pollination ; a. Thrum pollen on pin stigma; b. Pollen tube passing through style showing irregular deposition of callose; c. Pollen tube at the base of style showing more callose depositions.

intervals (Fig.1. a-d) . When the pin stigmas were pollinated with thrum pollen grains, the percentage of pollen adherence, pollen germination and mean tube length were also found to be low. The pollen tubes showed larger number of irregular callose plugs (Fig.2 a-d).

In the present investigation, *Pentas lanceolata* self and cross pollen show a massive germination but self pollen tubes grow faster than cross pollen (thrum) in pin stigma. Both, self and cross pollen could grow through the entire length of the style without any inhibition at any region or abnormal growth pattern. The self pollen, however, show a more regular deposition of callose plugs which is rather irregular in cross pollination. Observation in *Nicotiana glauca*<sup>11</sup>, has shown that compatible pollen tubes typically showed a uniform layer of callose deposition in the walls, in small plugs, spaced at regular intervals within the tubes whereas in incompatible tubes there was an irregularity of callose deposition in both walls and plugs. In *Pentas lanceolata* a similar observation was made. Here self-pollination is, hence, found to be more compatible in pin flowers although both pin and thrum pollen could germinate on pin stigmas<sup>12</sup>. Thus it appears that the timing and site of incompatible pollen arrest varies in different species.

It may be concluded that in *Pentas lanceolata* *in vivo* germination on self-pin stigma, there is no self-incompatibility, which is atypical for a heterostylous plant. Recent studies on *Psychotria ipecacuantha*<sup>13</sup> has also

revealed that pollen tube growth in style, fruit production after spontaneous self-pollination and intra-morph pollination indicated partial intramorph and self compatibility in this heterostylous plant species.

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