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Developmental anatomy and cytochemistry of *Consolea* style

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ABSTRACT

To identify and locate the site of incompatibility, the structural anatomy of the style and the cytochemistry of its secretions during the stages of development in Consolea have been investigated using light microscope. The pollen-pistil interaction studies show strong incompatibility response at the style. The style in Consolea is half closed. It is open with a stylar canal towards the stigma and closed with a core of stylar transmitting tissue towards the ovary. The stylar canal is lined with a morphologically distinct oblong to square stylar canal cells. This canal becomes progressively narrow and closed towards the ovary. The transmitting tissue is compact and oval shaped and forms large intercellular spaces filled with secretions rich in proteins, lipids and enzymes. The secretions reaches its peak value, when the stigma is at receptive and at post pollination stages. The anatomy and cytochemical differences between the metabolites at different regions of the style during the development are discussed from the aspect of pollen-pistil interaction.

Key words: *Consolea*, stylar transmitting tissue, cytochemistry, secretions, pollen-pistil interaction, incompatibility.

INTRODUCTION

In angiosperm pistils, the pollen tube grow through the style and this activity is restricted to a group of cells called stylar transmitting tissue. It is present in a compact core of loosely arranged cells in the solid styles (closed style) and a glandular layer lining the stylar canal cells in the hollow styles (open style). The cells of the transmitting tissue produce extra cellular secretions containing metabolites and enzymes which nourish the pollen grains during germination [17, 7]. The ultra structure of the style has been studied in number of species [8, 14, 15,16,5, 9, 10, 18, 6]. However studies on the anatomy and cytochemistry of the styles have been limited to few taxa. The present paper form a part of a major research investigation into the incompatibility and pollen-pistil interaction in *Consolea* with a view to unrevealing the reasons for non-seed setting[13]. We have investigated the anatomy and the cytochemical differences between metabolites and their role in pollen-pistil interaction.

MATERIALS AND METHODS

Styles of six following developmental stages were collected on the basis of their size and age (time before and after anthesis) of the flower buds / flowers.
Stage – I – Early floral bud
Stage – II – Medium floral bud
Stage – III – Late floral bud
Stage – IV- Early anthesis
Stage – V – At the time of anthesis
Stage – VI – Late anthesis

These developmental stages were determined based upon the size and age (time before and after anthesis) of the flower buds/flowers.

For the cytochemical localization of enzymes and metabolites, fresh semi-thin sections were and stained with the methods listed in Table-I. The stain intensity was observed and the response of the tissue was given score to indicate the magnitude of the particular enzyme / metabolite studied.

TABLE-I: Methods followed for cytochemical localization

Sr.No	Metabolites / Enzymes	Method	Reference
1	Proteins	Coomassie brilliant blue	Heslop-Harrison,1979
2	Insoluble polysaccharides	Periodic acid-Schiff's reagent	Hotchkiss, 1948
3	Lipids	Sudan black-B	Bayliss-High,1982
4	Peroxidase	P-phenylene diamine	Raa ,1973
5	Esterase	α - naphthyl acetate-fast blue	Nachlar & Seligman,1949

RESULTS AND DISCUSSION

The style of *Consolea* is half closed (Fig. 1-4). It is with stylar canal at the upper half towards the stigma (Figs.1, 2) and with compact transmitting tissue at the lower half towards the ovary (Figs. 4, 6). It reveals that the style consists of a single layered epidermis, a parenchymatous cortex and a central core of transmitting tissue at the basal region (Fig.6). It further reveals the presence of a single layer of morphologically distinct, oblong to square cells lining the inner surface of the style (stylar canal cells) at the upper half (Fig.2) with a canal which becomes progressively narrow and closed towards the ovary (Figs.4, 6). The transmitting tissue appears oval shaped in cross section (Fig.5) from the young to the mature pollinated stage. Presence of half closed style has also been reported in *Vigna unquiculata* by [24]. In *V.unquiculata*, the style is solid (closed) towards the stigma and it is hollow (open) towards the ovary. In contrast to this, in the present investigation in *Consolea*, the style is hollow towards the stigma and solid towards the ovary (Figs. 2-6).

Table.2. Behavior of stylar tissue and stylar secretion from fresh stigmas to different cytochemical tests.

Sr. No	Metabolite/ enzymes	Stage	Stylar tissue			Stylar canal exudates
			Stylar cortical tissue	Stylar transmitting tissue	Stylar canal cells	
1	Proteins	I	+	+	++	_
		II	+	++	++	_
		III	+	++	+++	_
		IV	++	+++	+++	+
		V	++	+++	+++	+
		VI	++	+++	+++	+
2	Insoluble polysaccha-rides	I	++	+++	+++	_
		II	++	+++	+++	+
		III	+++	+++	+++	+
		IV	++	++	++	+
		V	+	++	++	_
		VI	+	++	++	_
3	Lipids	I	+	+	+	_
		II	+	+	+	_
		III	+	+	+	_
		IV	+	++	+	+
		V	+	++	+	+
		VI	_	++	+	+
4	Peroxidases	I	+	+	+	+
		II	+	++	+	+
		III	+	++	+	+
		IV	+	++	++	+
		V	++	++	++	+
		VI	++	++	++	+
5	Esterases	I	_	_	_	_
		II	_	+	_	_
		III	+	+	+	_
		IV	+	+	+	+
		V	+	+	+	+
		VI	_	+	_	_

-- *Undetectable* + *Meagre* ++ *Moderate* +++ *Intense*

Figure-1. Stigmatic head with papillae (X 285)

Figure-2. Style L.S. Open stylar canal towards stigma (X300)

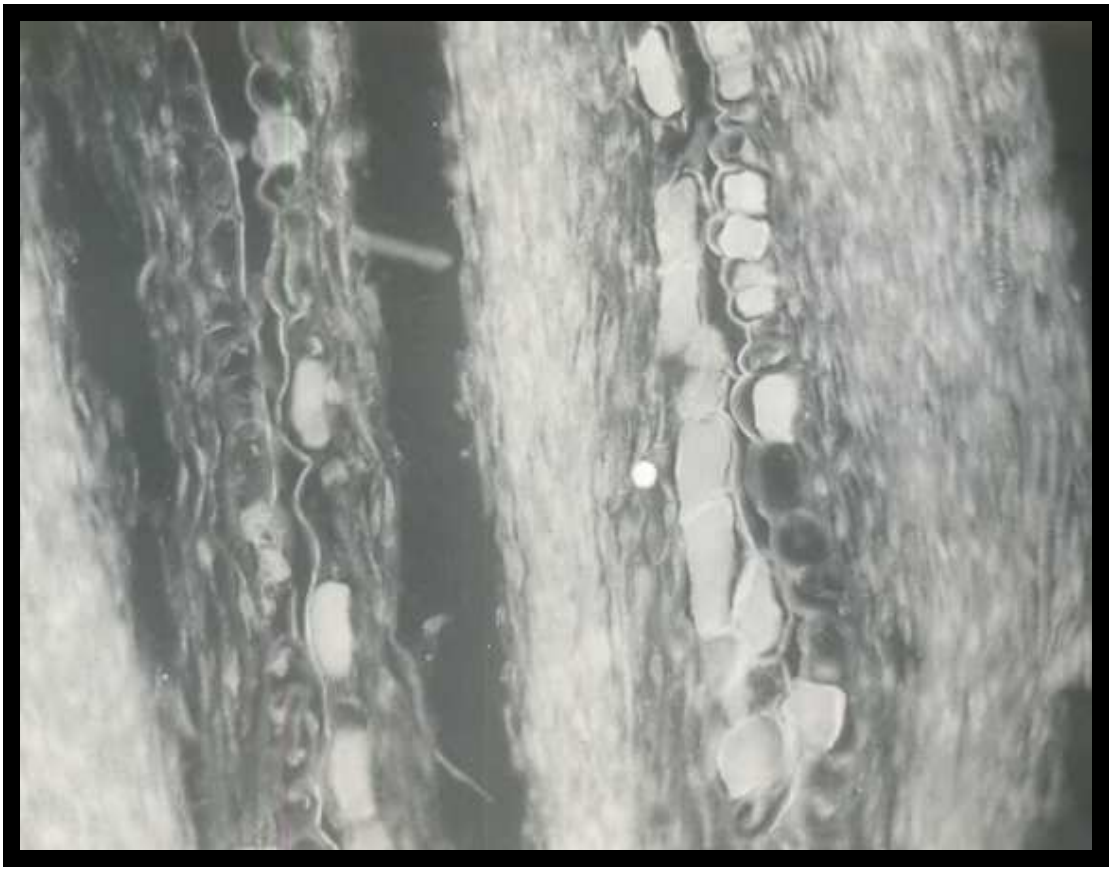


Figure-3. Style L.S. Stylar canal getting closed towards ovary (X300)

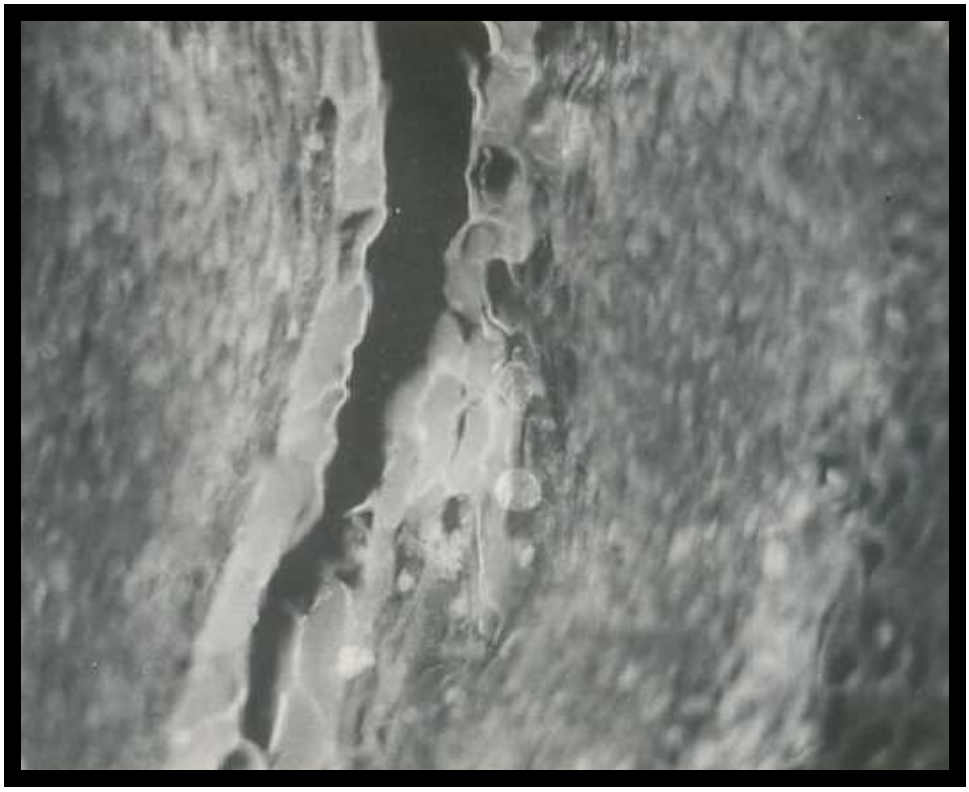


Figure-4. Style L.S. Stylar canal closed towards ovary (X362)

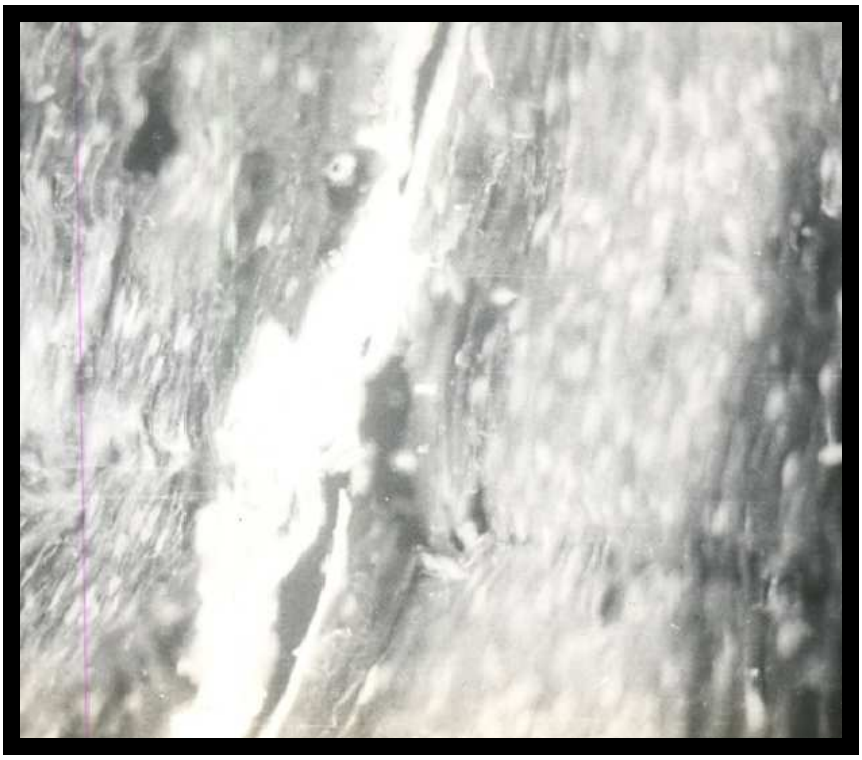


Figure-5. Style C.S. Open stylar canal towards stigma (X300)

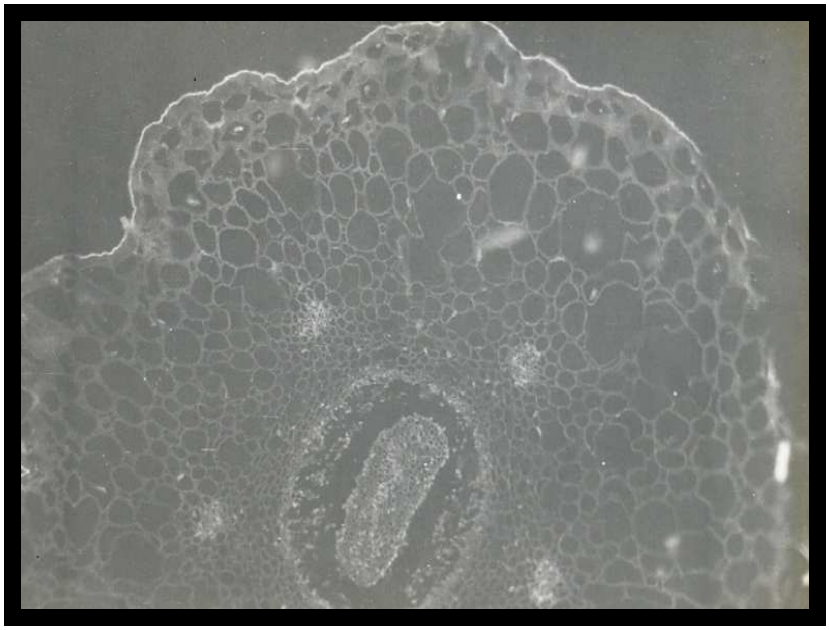
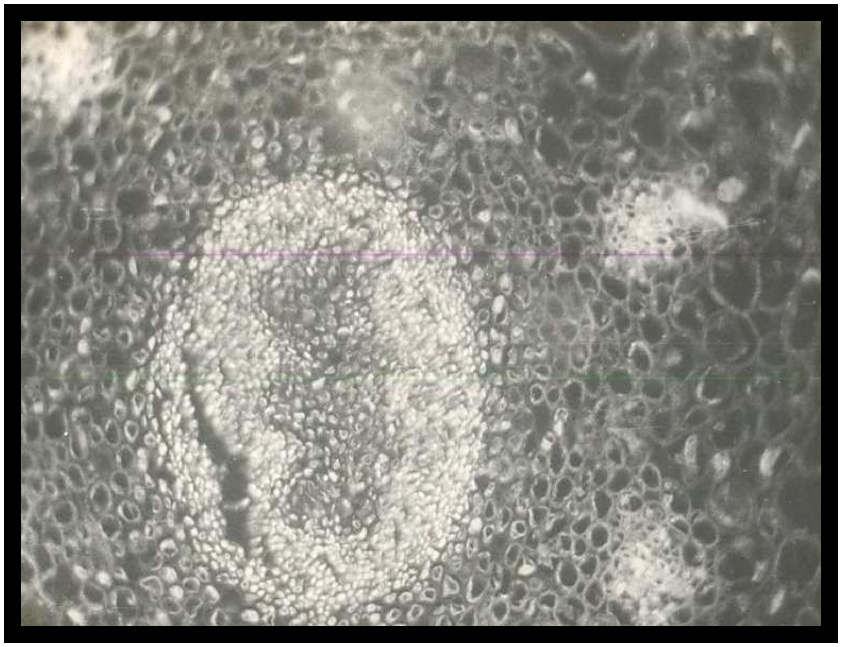


Figure-6. Style C.S. Closed stylar canal towards ovary (X360)



The cells of the young stylar tip are elongate and are loosely arranged. These cells project in to a common canal above the style tip at stage-I. The tissue placed above and surrounding the stylar canal forms the stigma. Presences of extra cellular secretion consists of enzymes (esterases, phosphatases and peroxidases) and metabolites (Carbohydrates, proteins, lipids, nucleic acids etc.) is one of the important character of stylar tissue during its development as studied[14, 18, 22, 6]. This was not evident during the early stages of style development in *Consolea* (Stage I & II) which is however distinct and evident in the stylar canal and stylar transmitting tissue from stage-III. It was found increased continuously up to anthesis and pollination (Table-2) as revealed by various cytochemical tests (Table-1).

The stylar transmitting tissue shows intense staining for all metabolites from early to later stages. Higher intensity of stain was noticed in the stylar transmitting tissue than the stylar cortical tissue (Table-2). The canal lining cells which are present in the upper half of the style shows moderate to intense stain for all metabolites studied cytochemically (Table-2).

Protein content increased tremendously from the initial (Stage-I) to final stages (Stage-VI) of development. Intense staining was noticed during and after pollination (Table- 2). This may be due to the release of protein from the pollen wall to the pistil after contact. [1, 2] also reported that the protein housed in the pollen as well as pistil play an important role in pollen- pistil interaction. [4] noticed the synthesis of new proteins after pollination. According to [11, 14], these proteins present on the stigma and style are involved in the recognition of pollen during fertilization.
Peroxidase activity was localized throughout the stylar tissue in the form of patches. Very slight increase in its activity was noticed after pollination (Table- 2). Involvement of peroxidases in incompatibility and its increase after cross pollination was studied by [3]. Less peroxidase activity in incompatible pollination as observed by [3, 12] may affect the rate of respiration which reflects a reduction in release of energy necessary for the growing pollen tube. In the present study in *Consolea* the activity of peroxidase does not show any increase after pollination (Table- 2). The pollen tube may fail to reach the ovary due to the lack of available energy. Peroxidase may also involve in nullifying the effect of hydrogen peroxide, which is released during terminal respiration, since it is a terminal oxidizer.

Presence of esterase in the transmitting tissue was noticed using α - naphthyl acetate-fast blue reaction. The intensity of the reaction was found negligible. The stylar cortical tissue does not show any positive reaction after pollination (Table- 2). [21, 19] also studied the activity of non-specific estrases and reported that it indicates the onset of stigma and stylar secretion and receptivity of the stigma.

Lipids are consistently noticed in all the regions of the style [20]. High intensity was localized during pollination and post-pollination (Stages IV-VI) (Table- 2) in the transmitting tissue. Higher lipid content with comparatively low esterase activity could be correlated in *Consolea*, since it is the enzyme which hydrolyses fat and lipid contents. Due to the diffused esterase activity at various regions of the style, the accumulation of lipids may be high and which might also act as a barrier for the pollen tube to grow through the style.

Stylar transmitting tissue showed higher polysaccharides than other regions of the style [23]. Stylar canal does not show positive reaction and the distribution of PAS-positive substances was not uniform at all regions (Table- 2). The intensity was more or less similar in the stylar canal cells and stylar cortical region (Table- 2). [7] reported the presence of abundant starch in the stylar tissue of unpollinated pistil in *Aegle marmelos*, *Fritillaria* and *Lilium*. Following pollination and pollen tube growth, the starch is broken down and it is probably used up for the growth of the pollen tube. Various metabolites and enzymes present in the secretions of style nourish the pollen grain and pollen tube. It was also reported by [17, 7]. Depending upon the nature of pollination (self/ cross) metabolites play a major role in screening the pollen and either accepts and allow the pollen to germinate (compatible) or rejects the pollen (incompatible).

CONCLUSION

Consolea is a succulent xerophytic cactus grows well in all ecological conditions like climatic, biotic and edaphic. The plant flowers well but no fruit setting. Present study is to understand the nature of stigma and style and find out where the incompatibility lies exactly. Changes in the level of macromolecules were studied biochemically and histochemically. The style is half closed. It is open with a stylar canal towards the stigma and closed with a core of stylar transmitting tissue towards the ovary. The stylar canal is lined with a morphologically distinct, oblong to square stylar canal cells. This canal becomes progressively narrow and closed towards the ovary. The transmitting tissue is compact and oval shaped and forms large intercellular spaces filled with secretions rich in proteins, lipids and enzymes. Protein content increased tremendously from the initial to final stages. Diffused esterase activity with high accumulation of lipids was observed in stylar transmitting tissue. Activity of peroxidase is very important for the release of energy necessary for the growing pollen tube, but peroxidase does not show any increase after pollination. This may affect the rate of respiration which reflects a reduction in release of energy. Present pollen-pistil interaction studies in *Consolea* show strong incompatibility response at the style.

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REFERENCES

[1] E. Pacini, G. Frenchi, G. Sarfatti, *Ann.Bot*, **1981**,47, 405-408.
[2] G. Dumas, R.B. Knox, T. Gaude, *Inter.Rev.Cyto*, **1984**, 90, 239-272.

[3] G.M.M. Bremedeijer, *Theor. Appl.Genet*, **1982**, 62, 305-309.
[4] H.F. Linskens and J. Tupy, *Zuchter*, **1966**, 36, 151-158.
[5] H.J. Wilms, *Acta.Bot.Neerl*, **1980**, 29, 33-47.
[6] H.K. Kandasamy and U. Kristen, *Bot. Acta*, **1990**, 103, 384-391.
[7] I.K. Vasil, Fertilization in Higher plants, North Holland Pub., Amsterdam, **1974**, pp.33-47.
[8] J. Bell, and G. Hicks, *Planta*, **1976**, 131,187-200.
[9] J. Heslop – Harrison and Y. Heslop-Harrison, *Ann. Bot*, **1982**, 50, 635-645.
[10] J. Heslop – Harrison, *Ann.Bot*, **1979**, 44, 1-47.
[11] J. Heslop – Harrison, R.B. Knox, Y. Heslop-Harrison, *Theor. Appl. Genet*, **1974**, 44, 113-137.
[12] J. Raa, *Physiol.Plant*, **1973**,28,132
[13] K. Saroja, Ph. D of Gujarat University, **1992**.
[14] M. Cresti, C.J. Keijzer, A. Tiezzi, F. Ciampolini, S. Focardi, *Amer. J. Hot*, **1986**, 73, 1713-1722.
[15] M. Cresti, F. Ciampolini, S. Sansavini, *Science Horticulture*, **1980**, 12, 327-337.
[16] M. Herrero and H.C. Dickinson, *Journal of Cell. Sci*, **1979**, 36, 1-18.
[17] M. Kroh and J.P.F.G. Helsper, Fertilization in higher plants, North Holland publishing Company, Amsterdam, **1974**, Pp.167-175.
[18] M. Sedgley and A.E. Clarke, *Nordic Journal of Botany*, **1986**, 6, 591-598.
[19] M.M. Nachles and A.M. Seligman, *Journal of the National Cancer Institute*, **1949**, 9,415.
[20] O, Bayliss – High, Manual of histological techniques, Churchill, Livingstone, Edinburgh, **1982**.
[21] O. Matteson, R.B. Knox, J. Heslop – Harrison, Y. Heslop – Harrison, *Nature*, **1974**, 247,298-300.
[22] P.K. Kuruvilla and J.J. Shah, *Ann.Bot*, **1988**, 61,269-281.
[23] R.D. Hotchkiss, *Arch. Biochem*, **1948**, 16, 131-141.
[24] S. Ghose and K.R. Shivanna, *Bot.Gaz*, **1982**, 143, 311-319.