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Cytological and Histological Studies of Jasmine (*Jasminum sambac* Ait.) Flower Petals

X. Alex Isac*, K. R. Rajadurai[1] and M. Jawaharlal[2]

ABSTRACT: *An investigation on gland morphology, anatomy and the chemical composition of the floral fragrance in the jasmine (Jasminum sambac cv. Gundumalli) flower petals was carried out at Department of Floriculture and Landscaping, Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore during 2014-15. In this study, the epidermal cell types and their distribution on abaxial and adaxial surface, floral trichome and stomatal availability on flower petals of Jasminum sambac cv. Gundumalli were identified using Light Microscopy (LM) and Scanning Electron Microscopy (SEM), and the systematic significance of these characters was evaluated. The major epidermal type like Papillose Conical cells with Striations (PCS) on the adaxial surface and Tabular Rugose cells with seriations (TRS) cells on the abaxial surface were recognized on the petal surface. Floral stomata were present on the abaxial surface and floral trichomes are present more on abaxial tip surface. This is apparently one of very few studies in which both odour and osmophore are characterised in jasmine flower petals. Structures responsible for floral scent production were localised with neutral red staining and histochemical assays for lipids and starch. Their morphology and anatomy were studied with scanning electron microscopy and light microscopy thin sections, respectively. Anatomical evidence suggests that unicellular trichomes on the abaxial surface of the petals are osmophores. These are rich in stored lipids, while the parenchyma surrounding the vascular bundles contains starch. Only freshly opened flowers produced odours, while the floral trichomes are in developing condition in its bud stage of the flowers. Gland anatomy and position of these scent glands on the flower petals plays a key role in release pattern of the fragrant volatile compounds.*

INTRODUCTION

Jasminum sambac commonly called as Arabian Jasmine is one of the important commercial loose flower crops from time immemorial. Jasmines have been cultivated since antiquity for a range of different purposes *viz.,* making garlands, adorning hairs, concrete extraction and in home gardens. More than 200 species of Jasmines occur in tropical and sub tropical regions, however around 40 are found in Indian sub-continent. Among them, *Jasminum sambac* Ait. (Gundumalli), *J.auriculatum* Vahl. (Mullai), *J.grandiflorum* Linn. (Pitchi) and *J.multiflorum* (Kakkada) are four commercially important species. Among the four species, *J.sambac* cv. Gundumalli occupies the leading position as a loose flower due to its intoxicating perfume emanating from its flowers.

The petal epidermal surface is important in pollination, as it influences the way in which pollinators perceive and interact with the flower. There is evidence that petal epidermal type and its surface affect colour depth (Kay *et al.,* 1981), scent production (Kolossova *et al.,* 2001), temperature (Dyer *et al.,* 2006) and provides tactile cues (Whitney *et al.,* 2009). Previous reports by several authors have classified and analyzed the distribution of the epidermal surface of petals within the angiosperms (Kay *et al.,* 1981) and extensive and detailed analyses have been provided for a few groups, such as Asteraceae (Hansen, 1991) and Leguminosae (Stirton, 1981). In this between 80% of the angiosperm species analysed have at least one petal having this epidermal type on the adaxial surface (or upper side, towards the floral axis) of the petals (Kay *et al.,* 1981; Christensen and Hansen, 1998).

The major and the minor volatile constituents of *J. sambac* were previously studied by Amr *et al.* (2008). However, there are no reports on the morphology and anatomy of petal and the features of secretory structures of *J. sambac.* Therefore, aims of the present study were to (a) characterize the epidermal cell types

*[1] Department of Floriculture and Landscaping, Tamil Nadu Agricultural University, Coimbatore, **E-mail: senthamil.alex@gmail.com*

[2] Horticultural College & Research Institute for Women, Navalur kutapattu, Trichy

in dorsal (adaxial), lateral or ventral (abaxial) surface (b) presence of trichomes (c) distribution of stomata and relate these patterns to our current understanding of the scent release pattern of this species

MATERIALS AND METHODS

Jasminum sambac cv. Gundumalli flower at fully matured unopened bud stage and fully opened stage were collected from the field of Tamil Nadu Agricultural University (TNAU), Coimbatore, Tamil Nadu, India to study the surface cell arrangement and cross section. The jasmine flower petals were sampled and used immediately for experiments. Petals of unopened bud stage and fully opened stage were made to thin section and the surface was observed under "QUANTA 250" Scanning Electron Microscope (SEM) with out Energy Dispersive X-Ray Spectroscopy (EDAX). Different parts of the flower petals *viz.*, tip portion, middle portion and basal portion of the petals in both the adaxial and abaxial surfaces were studied. After the specimens had been compared under Light Microscope (LM). For LM study, the sample preparation was made according to the procedure given by Johanson (1940).

RESULTS AND DISCUSSION

Scanning electron microscopic study of adaxial surface

The adaxial petal surface of both the bud and open flower of *J.sambac* cv. Gundumalli had papillose conical cells with striations (PCS) with pentagonal shape. There is no variation in the cell shape through at the basal portion, middle and tip portion of the petals. A glandular duct is present on the tip of each conical cell. (Table 1, Fig 1 & 2). It is proven earlier that these conical cells are controlled by Myb factor named Mixta in *A.majus* (Noda *et al.*, 1994) and this shape plays an important role in guiding pollinating insects. Morphogenesis of conical papillate epidermal cells in the adaxial epidermis of the jasmine flowers appears similar to that in *Arabidopsis,* Rose and wall flower *Erysimum cheiri, Mirabilis jalapa* (Uta effmert *et al.*, 2005) and it is characteristic of some 80% of different plant species of angiosperms which was surveyed by Kay *et al.* (1981). In *C.bewari* using RNA insitu hybridization, some enzymes contributing to scent production are expressed uniformly and exclusively in these cells of the epidermal layer of the petals (Duradeva *et al.*, 1996). Kolosova *et al.* (2001) demonstrated that S-adenosyl-L-methionine: benzoic acid carboxy methyl transferase (BAMT) an enzyme involved in scent biosynthesis was localized in the conical cells of the epidermal layer. On basis of these observations it was hypothesized that papillate cell shape also influences directionally in enhancing volatile emission in *Jasminum sambac*.

Stomata is absent in the adaxial surface of the petals (Fig. 1). The lack of stomata on the adaxial epidermis in which the papillate conical cells are present is consistent with the observation that the MIXTA transcription factor, which is required for conical cell differentiation (Noda *et al.*, 1994), reduces the stomatal density in tobacco leaves when over expressed. Whereas trichomes and conical papillate cells appear to have a common developmental pathway, the developmental pathway of stomata is different and mutually exclusive from that of trichomes and conical papillate cells as suggested by Martin and Glover (1998).

Scanning electron microscopic study of abaxial surface

The abaxial petal surface of jasmine buds had tabular rugose cells (TRS) cells more or less isodiametric with longitudinal and dense striations. Stomata are present in the tip of the flower which is seen in both bud and open flower. Closed stomata at the bud stage (Fig 4) and open stomata in the open flower (Fig. 3b). However, in the beginning of scent release, stomata of the abaxial epidermis was mostly open indicate that they manage the extensive gas exchange due to intensive metabolism caused by volatile production (Skubatz *et al.*, 1995).

Table 1

Surface	*Major epidermal type*	*Abbreviation*	*Trichomes*	*Stomata distribution*	*Figure*
Adaxial					
Open flower	Papillose conical cells	PCS	-(absent)	-	Fig. 1
Fully mature bud stage	Papillose conical cells	PCS	-(absent)	-	Fig. 2
Abaxial					
Open flower	Tabular rugose cells	TRS	Present	+(Only in the tip portion)	Fig. 3a. & 3b.
Fully mature bud stage	Tabular rugose cells	TRS	Developing	+(Only in the tip portion)	Fig. 4

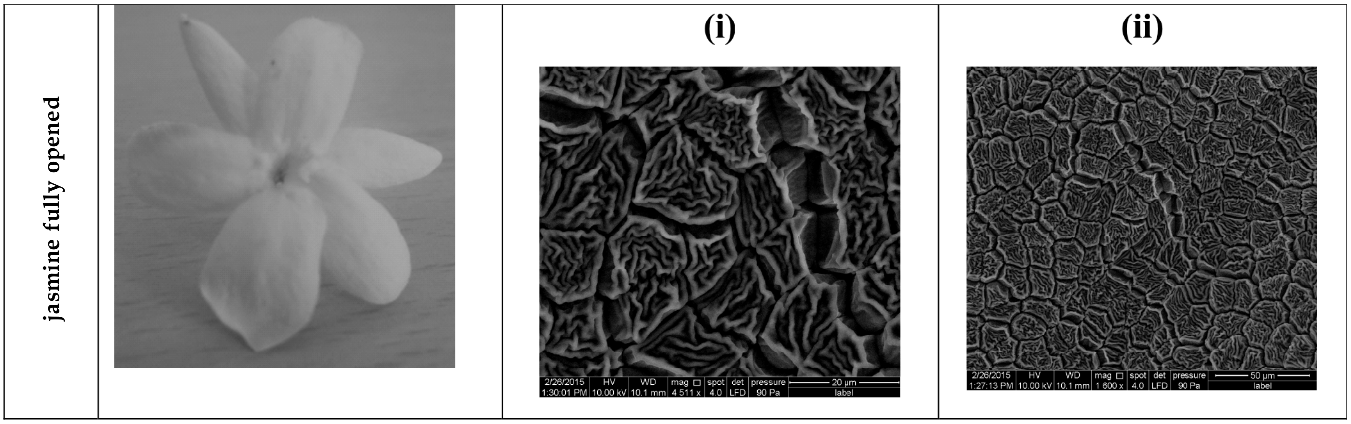


Figure 1: Adaxial surface of open jasmine fully opened flower

(i) 600X magnification, (ii) 2400X magnification

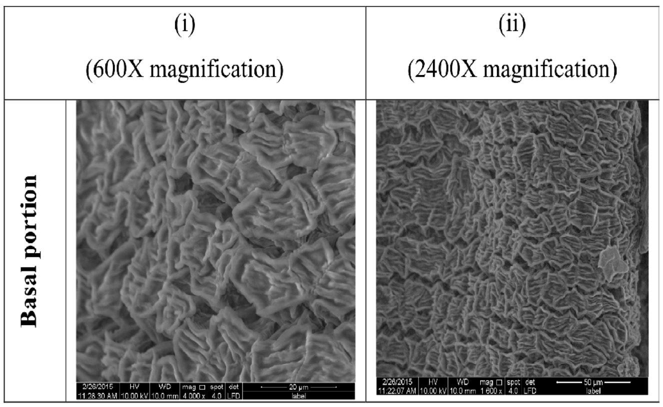


Figure 2: Adaxial surface of jasmine mature bud

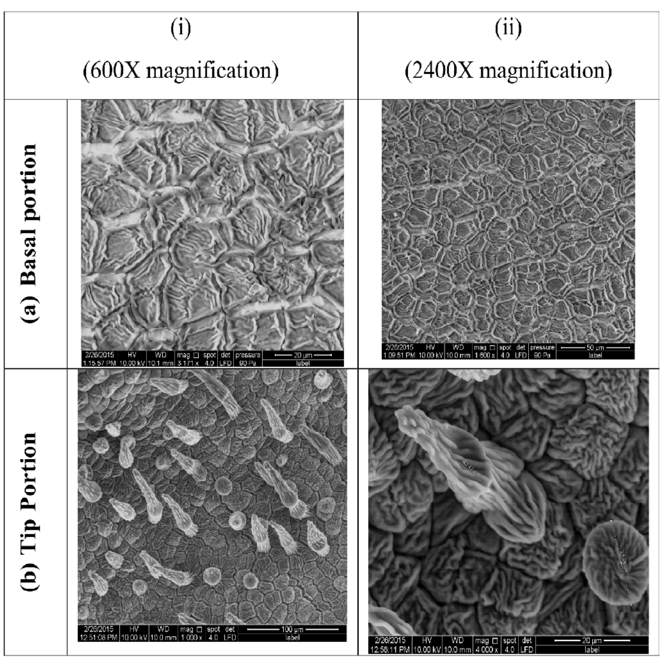


Figure 3a: Abaxial surface of fully opened jasmine flower

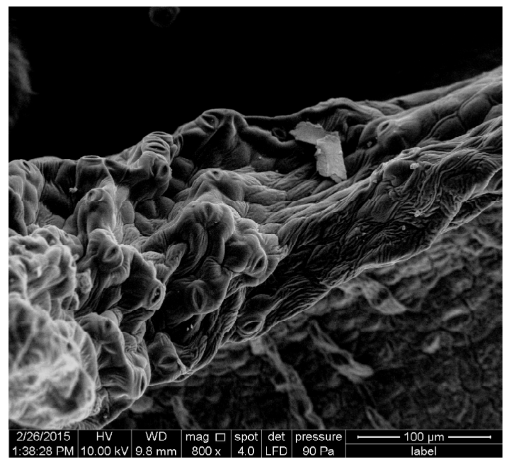


Figure 3b: Stomata on the abaxial epidermal cells of *J.sambac* cv. Gundumalli

Floral tirchomes are absent in the jasmine flower petals because it is also an important component to contribute in fragrance emission (Table 1).

Anatomical studies of the flower tissue using light microscopy

Cross section of the petals from *J. sambac* cv. Gundumalli at two different stage *viz.,* bud when the flowers are still closed *i.e.,* at the bud stage and open flowers which shown in Fig. 1 provided the information of the jasmine petal anatomy. The petal consists of epidermis, fundamental tissues and vascular bundles (Fig. 1).

Epidermal cells

At the bud stage, abaxial epidermal cells had more of less rectangular in shape. Epidermal cells are alive and have obvious nucleus and dense cytoplasm. Their cell walls and cuticle are thin and closely arranged.

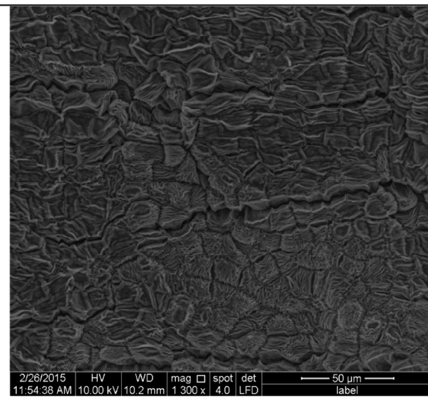
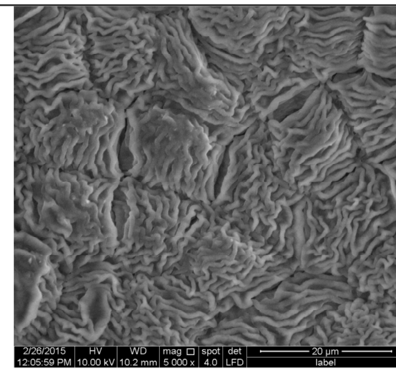
Jasmine Matured Bud	(i) 600X Magnification	(ii) 1200 X Magnification
		

Figure 4: Abaxial surface of jasmine flower bud

In the open flower stage when flower began to senescence the abaxial cells attain conical shaped cells with slight intercellular space. Furthermore, only a few mesophyll cells were observed. They were separated by an extensive intercellular system. The cells of the abaxial epidermis are generally larger than those of the adaxial epidermis and surrounded with a cell wall and thin cuticle. The sections of the petals confirmed the delicate epidermis of both the adaxial and abaxial side of the flower, as observed with the electron microscopy.

Fundamental cells/ Spongy parenchyma and Vascular bundles

Fundamental tissues of petals locating under epidermis at the central part of the petal which was filled with a spongy parenchyma, composed of loosely attached cells with large intercellular spaces. Most of the cells are circular in shape. Vascular bundles which are simple in structure distributed in fundamental tissue. In the open flower, spongy parenchyma was practically devoid of cells.

Role of the adaxial epidermis in emission of fragrance

The role of the adaxial epidermis in emission of fragrance was also supported by morphological studies using SEM and light microscopy. This segment of the flower displayed a distended epidermis surface with PCS type of cells on the adaxial surface, which was strongly suggestive of the conical cells found in the epidermis of rose and *Petunia* petals and in snapdragon (Kolosova *et al.*, 2001). This structure has also been described for lobe margins of *Bougainvillea stipitata* flowers, Nyctaginaceae (Lopez and Galetto, 2002). This typical epidermis would also fulfill a feature found in osmophores, because surface enlargement is supposed to be a precondition for optimal volatile emanation (Vogel, 1962). Floral volatiles are released from the epidermal cells of the petals. Indeed, enzymes catalysing the final steps in volatile phenylpropanoid/benzenoid production have been localized predominantly to the petal epidermis in some species such as rose and snapdragon (Veronique *et al.*, 2007; Long *et al.*, 2009), although, to some extent, localization to sub epidermal or mesophyll cell layers has been reported for some of them as well (Rohrbeck *et al.*, 2006).

However, cross-sections of the petals examined by Light Microscopy confirmed a fragile structure of both epidermises of supporting facilitated emission, but the nearly very few mesophyll cells with more intercellular space indicate that the scent has a diffuse emission.

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