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Exploration of Foliar Epidermal Pattern in *Cucumis pubescens* Wild.

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ABSTRACT

The Indian wild taxa *Cucumis pubescens*Willd.is a weed belonging to family Cucurbitaceae, a family that grows wild in the arid and shadow regions of India. In the present investigation the plant were subjected to detailed and critical analyses of the foliar epidermal characteristics. For this work parameters like number of stomata and epidermal cell per mm^2, length and breadth of stomata, pore area, guard cell index, pore area index and total pore area were analysed. To the best knowledge, the foliar epidermal work has been done for the first time. The leaves were amphistomatic and differed in the morphology of the walls of the normal epidermal cells. In plant, the numbers of stomata were more on the abaxial surface than adaxial surface. The mean values of number of stomata and epidermal cells (per mm^2), length and breadth of stomata (µm), pore area, Guard cell index, pore area index and total pore area on abaxial and adaxial epidermal surfaces are 501, 251, 5.2, 3.7, 26, 49, 145, 0.7 and 254, 89, 5.7, 4.3, 23, 40, 104, 0.2 respectively.

Keywords: *Cucumis pubescens*, foliar epidermal pattern analyses, abaxial surface, adaxial surface.

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INTRODUCTION

Cucumis pubescens Willd. is a weed belonging to family Cucurbitaceae, a family that grows wild in the arid and rain shadow regions of India. In India this plant is mainly found in Rajasthan, Madhya Pradesh, Chhattisgarh, Bihar, Maharashtra, etc. Although due to its ability to adapt quickly, it can be found in many more regions as well. Globally it is common in Malaysia, Australia, Africa, and few south Asian countries. The various vernacular names of *Cucumis pubescens*in india are ‘chitrakala’ (Sanskrit), ‘Selni’ (Marathi), ‘kachri’ (Hindi), ‘bangaumukh’ (Bangali), ‘kachari’ (Rajasthani and Gujrati), ‘pehta’ (Bihari), ‘demu’ (Chhattisgarhi), ‘sane’ (Madhya Pradesh) and ‘chibbad’ (Punjabi) [1].

It is a small brown or yellow melon. It has a bitter sweet flavour. But after getting ripe, it tastes sweet and sour and can be consumed directly. As all cucumber crops, it has hairy stem, yellow flowers and triangular leaves (fig.1). It is used as chutney, kachri powder and sabzi. The

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fruits resemble a tiny melon (fig.2). Around 15-20 years back, it was a well-known crop, but these days it is losing its identity. It is even used to make vegetables in many households.

The main problem with the plant is that it was not cultivated purposely. Due to the wild growth in nature, it grows on a huge area surrounding the crops. People and farmers use it as refreshment and have never seen it as a cash crop. But in Rajasthan in later stages, people started cultivating it on a very small scale. Therefore it holds a significant part in Rajasthani Cuisine.

Along with its uses in vegetables, fruits and pickles the plant has a number of medicinal properties also. The fruit also helps in kidney stones and other urinary related issues. It has the property to remove constipation and aid better bowel movement. It holds anti-inflammatory and analgesic value, thus, it provides relief in stomach upset. It is beneficial in treating prickly heat, bedsores, ear ache etc. The fruit juice is also used as a tonic [4].

Despite of all these benefits the plant is losing its identity. It should have been included in the list of super foods, but on the contrary, it is fighting for its recognition. It is considered as a food for the poor and is not even cultivated purposely in other parts of the nation outside Rajasthan. If given proper information it can add an excellent commercial value in other hot and arid areas also. These are not even costly if compared with other vegetables and masala ingredients. This very fact can serve to be its USP (Unique Selling Proposition) in the market. All their varieties- raw, ripped, dried and powdered can be used as a good source of income on a large scale.

As far as literature is concerned very scanty material about plant morphology is available. Most of the information is about the ethnobotanical aspects of plant. Though extensive embryological work has been reported [3]. The purpose of the present work was to explore the foliar epidermal pattern of the plant which is to the best of our knowledge this work has been done for the first time.

MATERIALS AND METHODS

The study material mature leaves of *Cucumis pubescens* were subjected to analysis of the foliar epidermal characteristics (fig.3). For this, a series of parameters listed below were analysed for both abaxial as well as adaxial epidermis. To the best of our knowledge, this is the first information available on the epidermal pattern of *C. pubescens.*

For epidermal study standard techniques were used [5, 6, and 7], the mature leaves were fixed in FAA (10% Formaldehyde:50% Alcohol: 5% Acetic Acid + 35% Water). The middle portion of the leaves of about $1cm^2$ was taken for the study. These blocks of mature leaves were placed in 1:1 solution of glacial acetic acid: hydrogen peroxide in petriplates and kept in oven at 60^0C. The adaxial and abaxial epidermis was separated out carefully. The epidermis was stained in 1% safranin, washed with water and mounted in 4% glycerine. The number of stomata and epidermal cells were calculated in X45 and size of stomata and pore area were measured in X100.

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For pore area Camera Lucida drawing were used. The photomicrographs were taken from temporary preparations.

For the present study the stomata were defined as the structure comprising slits guarded by two guard cells each. Following epidermal parameters were explored.

(a) Number of epidermal cells per mm^2,
(b) Number of stomata per mm^2,
(c) Guard cell index (GCI),
(d) Size (length and breadth) of stomata (µm),
(e) Area of stomatal pore (μm^2),
(f) Pore area index (PAI),
(g) Total pore area per mm^2 (TPA) and
(h) Any abnormality

The TPA is defined as the percent of the total area of the pores per mm^2. The GCI, PAI and TPA were calculated with the help of following formulae:

$$GCI = \frac{\text{Number of guard cells in stomata per mm}^2}{\text{(Number of guard cells + subsidiary cells + Epidermal cells) per mm}^2} \times 100$$

$$PAI = \frac{\text{Mean pore area } (\mu m^{2})}{\text{Mean length} \times \text{breadth of stomata } (\mu m)} \times 100$$

$$TPA = \frac{\text{Mean pore area } (\mu m^2) \times \text{mean number of stomata per mm}^2}{10^6\,(1000 \times 1000\ \mu m)} \times 100$$

OBSERVATIONS

The mature leaves of *Cucumispubescens* were subjected to analysis of the foliar epidermal characteristics (fig.3). It was observed that the leaves were amphistomatic and differed in the morphology of the walls of the normal epidermal cells (fig. 4, 5). The walls of the epidermal cells of abaxial surface were chiefly wavy while those of adaxial epidermis were relatively straight.

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The data related to number and length and breadth of stomata was taken in 10×40 magnification. The mean number of epidermal cells and stomata per mm^2 on abaxial surface was found to be 501 and 251(fig.4), and on adaxial surface 254and 89 respectively (fig.5). The range of stomata on abaxial and adaxial surfaces varied from 76 to 516 and 34 to 135 respectively. The data related to mean number of epidermal and stomata are show in table 1.

The mean length and breadth of stomata on abaxial and adaxial surface was found to be 5.2 and 3.7; 5.7 and 4.3 respectively. The mean pore area on abaxial and adaxial surface was found to be 26 and 23 μm^2 respectively. Parameters like GCI, PAI and TPA were also calculated by using formulae in materials and methods. The mean value of GCI, PAI and TPA on abaxial and adaxial surface were found to be 49, 145, 0.7 and 40, 104, 0.2 respectively.

Some abnormalities were also observed as absence of one guard cell and both guard cells. Both these abnormalities were found on the abaxial surface though only first was found on adaxial surface. Out of these two abnormalities the former was in high frequency on the abaxial surface (fig.6 and fig.7).

Fig. 1. Flowering plant of *C. pubescens*

Fig. 2. Fruiting in *C. pubescens*

Fig. 3. Mature leaf of *C. pubescens*

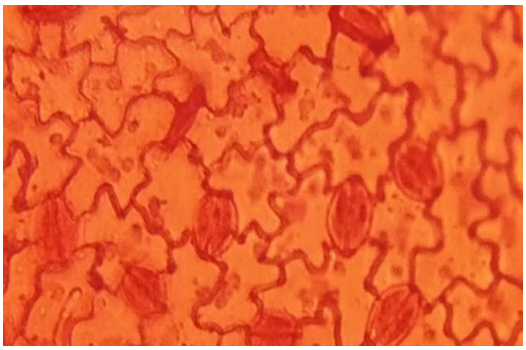

Fig. 4. Abaxial surface of leaf of *C. pubescens* (40X)

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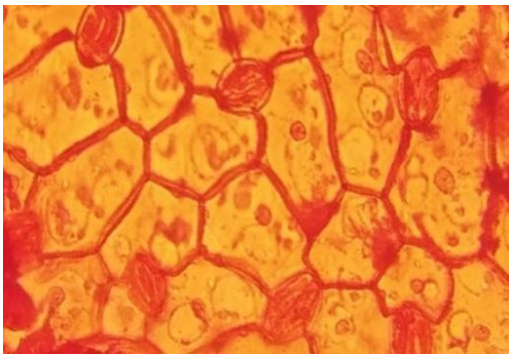

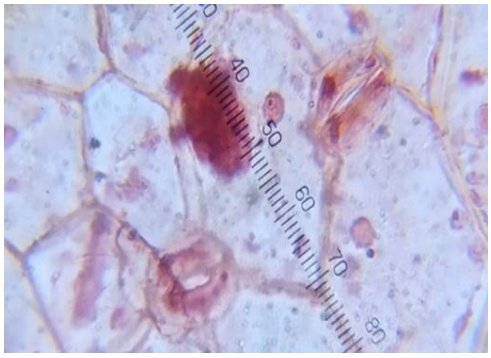

Fig. 5. Adaxial surface of leaf of *C. pubescens* (40X) **Fig. 6. Stomata of *C. pubescens* in 100 X**

RESULTS AND DISCUSSIONS

The stomata were found on both the abaxial and adaxial surfaces (amphistomatic). But the number of stomata was more on the abaxial surface. The mean number of epidermal cells on abaxial and adaxial surface was 501 and255 respectively. The mean number of stomata on abaxial and adaxial surface was found to be 251 and 90 respectively. The mean number of stomata on abaxial surface ranged from 76-516 while on adaxial surface ranged from 33-135.

To the best of our knowledge, this is the first work that has been done on the foliar epidermal pattern of *C. pubescens*Willd. Inter-specific and intra-specific variations in the values of GCIs pointed out the difference in the proportions of guard cells over that of epidermal cells (including guard cells). In fact, guard cells are specially programmed epidermal cells. The difference in their proportions indicated the difference in the genotype dependent pattern of genetic program distribution within the cells of the same tissue. Differences in the values for pore area indices (PAIs) gave information about the proportion of the area of stomatal complex that could differentiate as pore. Accessions dependent significant differences in the mean values of total pore area clearly indicated the genotypic control on the distribution of stomata, in addition to the environmental control.

As the plant material was collected from wild a number of abnormalities like absence of one guard cell and both guard cells were observed. Absence of one guard cell was very common. *C. pubescens* is not only medicinally important but has edible qualities too, this plant requires urgent attention in order to explore it properly and conserve it by latest advanced phytochemical, cytogenetic, biotechnologies and *in- vitro* techniques.

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Table 1. Table showing mean data of epidermal and stomatal cells per mm^2

	ABAXIAL LEAF SURFACE						ADAXIAL LEAF SURFACE					
	Mean	±	SE	Range			Mean	±	SE	Range		
No. of Epidermal cells	501	±	50	144	-	864	254	±	21	101	-	406
No. of stomatal cells	251	±	27	76	-	516	89	±	8	34	-	135

Table 2. Table showing mean length and breadth of stomata in micrometer^2

	ABA XIAL LEAF SURFACE						ADAXIAL LEAF SURFACE					
	Mean	±	SE	Range			Mean	±	SE	Range		
Length	5.2	±	0.6	4.4	-	6.4	5.7	±	0.6	4.8	-	6.8
Breadth	3.7	±	0.4	2.8	-	4.8	4.3	±	0.4	3.2	-	5.2

Table 3. Table showing mean Pore area micrometer^2

	ABAXIAL LEAF SURFACE						ADAXIAL LEAF SURFACE					
	Mean	±	SE	Range			Mean	±	SE	Range		
Pore Area	26.3	±	10.27	15.5	-	49.6	23.0	±	11.28	10.23	-	54.25

Table 4. Table showing mean values of GCI, PAI and TPA

	ABAXIAL LEAF SURFACE						ADAXIAL LEAF SURFACE					
	Mean	±	SE	Range			Mean	±	SE	Range		
GCI	49.4	±	2.4	46.3	-	54.2	40.2	±	3.7	33.3	-	46.4
PAI	144.7	±	65.3	57.5	-	287.0	103.5	±	73.8	44.7	-	326.0
TPA	0.68	±	0.42	0.14	-	1.7	0.21	±	0.12	0.05	-	0.48

GCI - Guard cell index, PAI - Pore area index, TPA - Total pore area per mm^2

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