

## TRICHOMES DIVERSITY IN FAMILY SOLANACEAE FROM MUZAFFARABAD DIVISION, AZAD JAMMU AND KASHMIR, PAKISTAN

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### ABSTRACT

Trichomes of twenty two taxa of the genus *Solanum*, *Withania*, *Petunia*, *Capsicum*, *Atropa*, *Lycopersicon*, *Cestrum*, *Hyoscyamus*, *Datura*, *Physalis* and *Nicotiana* were examined by using light microscope and photomicroscope. The indumentums shows considerable variations among different species, and therefore, useable characters in de-limitation of species. The study revealed the characters of anatomic interest of leaf and stem epidermal features that have not been previously reported before and has been conducted first time. Size, shape and types of trichomes were measured with considerable variability and the presence of glandular and non-glandular trichomes were observed. Distribution of foliar trichomes emerged as a supportive taxonomic and anatomic tool, which are present in the species mostly are unicellular, bicellular and non-glandular, glandular with bulbous base.

**Keywords:** Epidermis, anatomy, Trichomes; glandular and non –glandular

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### INTRODUCTION

Solanaceae is an important family for possessing countless ornamental, medicinal and nutritious species (Heywood, 1993). The family is represented by about 94 genera and 3000 species of nearly cosmopolitan distribution (Mabberley, 1987) mostly found in tropical and temperate regions with centers of diversity in the southern hemisphere, particularly in South America. Center of speciation occurs in Australia, Africa, Europe and Asia D'Arcy (1976). It is most closely related to the Scrophulariaceae, The family was divided by Wettstein (1985) into the tribes: Nicandreae, Solanaeae, Datureae, Cestreae and Salpiglossideae. The taxonomic value of the indumentum as well as its importance in systematic are well known in Lamiaceae and its related families Acanthaceae, Bignoniaceae, Scrophulariaceae, Verbenaceae, Fagaceae, Malvaceae, Convolvulaceae, Asteraceae, Moraceae and Cucurbitaceae (Ahmad, 1964a; Wagner et al., 2003; Porto *et al.*, 2008 (Xiang et al., 2010).

In Pakistan the family Solanaceae is represented by 14 genera and 52 species (Nasir, 1985), 27 species are native, 6 exotic and the rest of cultivated. Some chief genera of Solanaceae found in Pakistan are *Solanum* (Potato, nightshade), *Datura* (Jimsonweed), *Nicotiana* (tobacco), *Lycium* (Kaffir thron), *Atropa* (belladonna), *Hyoscyamus* (henbane), *Capsicum* (pepper chili), *Lycopersicon* (tomato) and *Petunia*. Efforts have been made to trace the taxonomical and anatomical characteristics of trichomes of leaf and stem of present day Solanaceae.

Trichomes were among the first anatomical features of plants to be recognized by early microscopists and they have played a key role in plant taxonomy (Behnke, 1984). Systematic and phylogenetic relationship and taxonomy is well known in Lamiaceae, Solanaceae and such related families as Verbinaceae and Scrophulariaceae on the leaf surface (Metcalf and Chalk 1950; Cantino, 1990) (Wagner, 1991; Khokhar, 2009). The botanical literature contains more than 300 descriptions of trichomes which categorize their great variations (Kohkhar, 2009). Anatomical studies have shown that foliar features are strictly comparable over wide taxonomy range. It plays very significant role in identification of incomplete plants (Stace, 1965). Angiospermic families viz; Restionaceae, Malpighiaceae, Malvaceae, Fagaceae and Centrolepidaceae individual species can be identified on the form and type of their trichomes alone (Cutler, 1985). Anatomical and morpho-taxonomical have been both for the classification and identification purposes by scientists (Xiang *et al.*, 2010).

Trichomes may serve to protect buds of some plants until defense phytochemical produced (Johanson, 1975). Simple and non-glandular trichomes serves the plant and humans in many ways. Leaf trichomes have been shown to reduce insect herbivory in a number of plant species. (Marquis, 1992; Elle *et al.*, 1999; Romeis *et al.*, 1999; Hare and Smith, 2005). Glandular trichomes can affect host diseases and pest resistance based on phytochemicals. Functionally trichomes protect plant from insect, pathogens, herbivores, heat and sunlight (Croteau, 1977; Werker, 1993; Duke, 1994). Glandular trichomes produce various substances, which are stored at the plant surface (Wagner, 1991; Kolb and Muller, 2004).

## MATERIALS AND METHODS

Twenty two species of family Solanaceae were collected during the years 2013 and 2015. Eight field trips were conducted during the month of March-September at the different localities of Muzaffarabad Division, Azad Jammu and Kashmir (Fig 1). Dried plants were mounted on herbarium sheets deposited mainly in the herbarium of Botany department, Azad Jammu and Kashmir University Muzaffarabad.

The plants were identified with the help of flora of Pakistan (Ali and Qaiser, 2009). Paradermal sections of the upper and lower epidermis of many fresh leaves were made with the sharp razor blade and transferred into Petridish. Trichomes were obtained from leaves and stems studied with a light microscope. For light microscopy, the leaves and stems have been sectioned at 20–30  $\mu\text{m}$  by using hand microtome. Safranin (2%) and light green (1%) combinations were used (Gerlach, 1977) with some modifications. Permanent slides were prepared by dehydrating the epidermal sections that have been washed free of excess stain for 2 minutes in 30, 50, 70 and 90 % alcohol, clearing then in few drops of pure xylene for 2 minutes and mounted in Canada balsam for microscopic examination (Chaudhary and Imran 1997; Johanson 1975, Cotton 1974). Length and width of trichome of plant species were taken as an average (three readings) Excel software 2016. Micrographs of the trichomes were taken by Nikon (FX 35) camera equipped with photomicroscope (IRMECO 910 Germany). Basic terminology was used according to Pyne (1978) and Harris and Harris (2001).

## RESULTS AND DISCUSSION

Micro anatomical characteristics of leaf epidermis and stem and distribution of trichome was examined in the present study. A total of 22 dicot plant species were carried out which were separated in 11 genera of family Solanaceae for trichome detail (Table 1). The genus *Solanum* has 9 species which are most prevailing while *Capsicum*, *Datura* and *Petunia* have 2 species and remaining 7 genera were represented by single species (Table 1). The epidermis and stem possesses a number of taxonomically important characters which offer variable clues for the identification, like the indumentation characteristics, composition, distribution and structural peculiarities of epidermal cell wall (Dickison, 2000). The taxonomic studies of a number of families and genera have been made on the basis of leaf epidermis Adedeji and Jewoola (2008). In the most of the taxon studied the leaf surfaces are glabrous varying from pilose to ovate, hairy, dentate and entire, densely covered with glandular and non-glandular, and unicellular to multicellular trichomes.

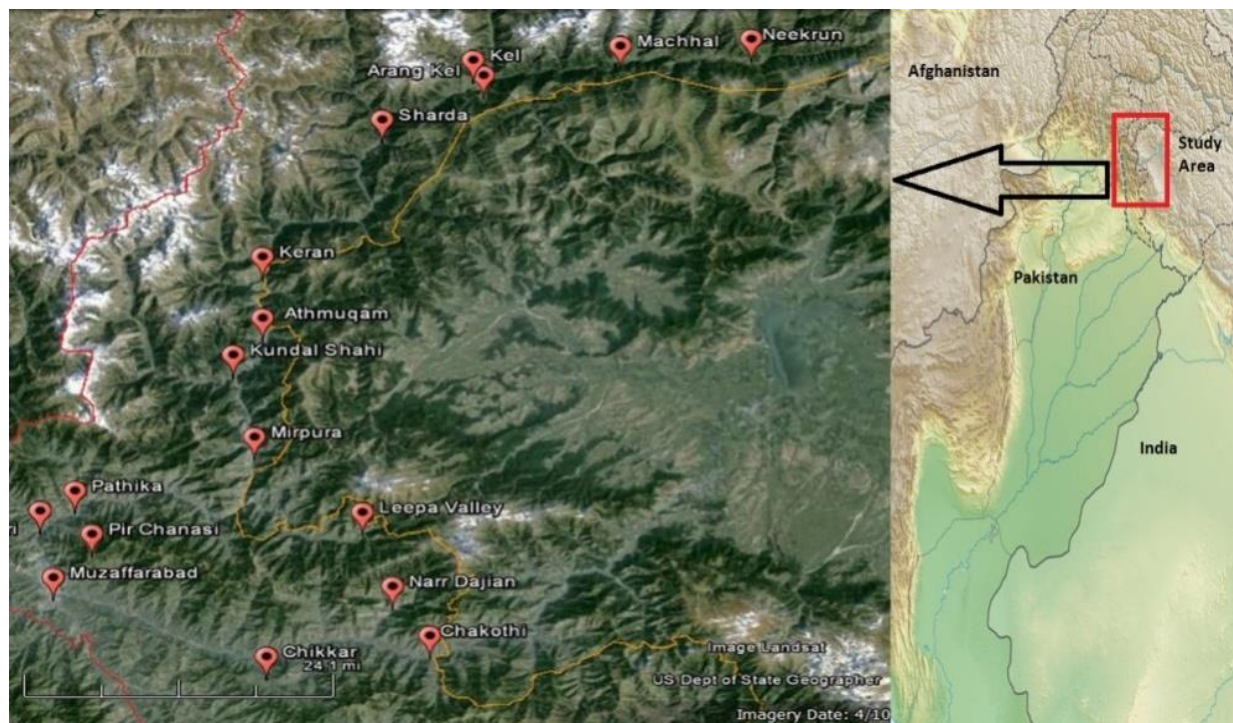
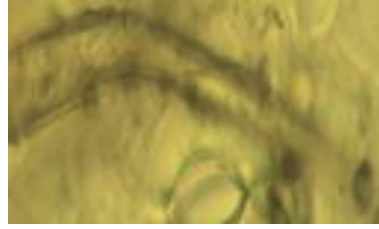
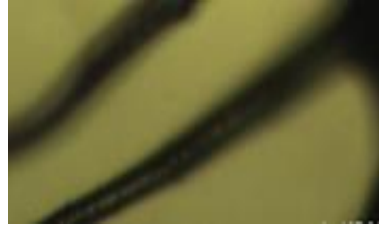
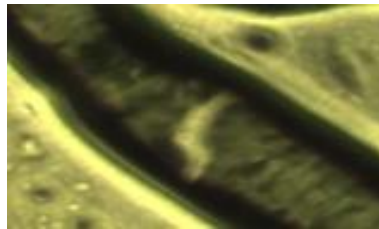
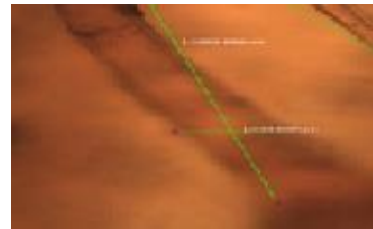
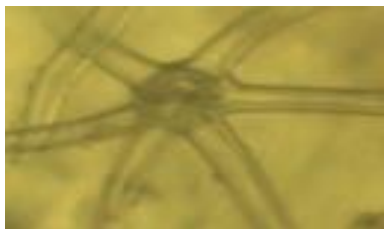
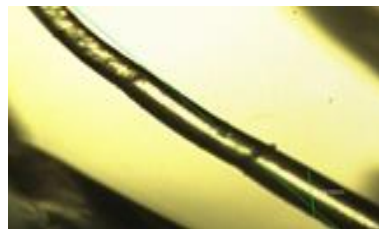
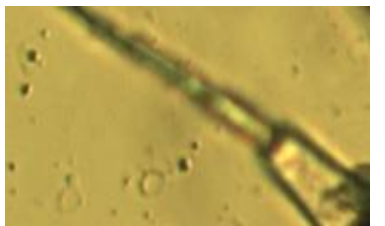


Fig 1 Map of the area

Table 1. Size and type of trichome (adaxial and abaxial) surface of 22 species of family Solanaceae.

	Taxon	(length $\mu\text{m}$ )			(width $\mu\text{m}$ )		(length $\mu\text{m}$ )		(width $\mu\text{m}$ )	
		Trichome type	Abaxial Surface		Abaxial Surface		Adaxial Surface		Adaxial Surface	
1	<i>Solanum nigrum</i> L.	Non glandular	122.16	$\pm 0.685$	55.16	$\pm 5.87$	112.47	$\pm 0.327$	19.71	$\pm 2.483$
2	<i>Solanum villosum</i> L.	Non glandular	128.53	$\pm 0.985$	9.56	$\pm 0.865$	80.81	$\pm 2.846$	6.23	$\pm 0.412$
3	<i>Solanum surratense</i> Brunf.	Star shaped	122.76	$\pm 12.198$	9.96	$\pm 1.100$	58.79	$\pm 0.482$	6.53	$\pm 0.408$
4	<i>Solanum cordatum</i> Forssk.	Non glandular	90.00	$\pm 5.197$	8.76	$\pm 1.262$	51.50	$\pm 1.268$	6.36	$\pm 0.245$
5	<i>Solanum pseudocapsicum</i> L.	Glandular	129.33	$\pm 1.425$	10.73	$\pm 1.252$	70.33	$\pm 0.944$	12.47	$\pm 0.799$
6	<i>Solanum melangena</i> L.	Star shaped	120.06	$\pm 8.087$	9.25	$\pm 0.533$	64.03	$\pm 1.447$	7.76	$\pm 0.489$
7	<i>Solanum tuberosum</i> L.	Star shaped	137.33	$\pm 11.213$	8.80	$\pm 0.152$	129.43	$\pm 8.451$	8.66	$\pm 0.789$
8	<i>Solanum erianthum</i> D. Don.	Non glandular	91.36	$\pm 1.633$	8.20	$\pm 0.393$	57.90	$\pm 4.547$	5.99	$\pm 0.421$
9	<i>Solanum dalcumara</i> L.	Glandular	115.08	$\pm 0.579$	8.13	$\pm 0.983$	86.93	$\pm 6.223$	6.56	$\pm 0.518$
10	<i>Hyoscyamus niger</i> L.	Non glandular	57.73	$\pm 1.035$	5.96	$\pm 0.413$	34.53	$\pm 1.878$	7.76	$\pm 0.395$
11	<i>Capsicum annum</i> L.	Glandular	118.66	$\pm 1.624$	10.433	$\pm 0.491$	69.16	$\pm 4.849$	6.26	$\pm 0.422$
12	<i>Capsicum frutescens</i> L.	Glandular	74.73	$\pm 7.624$	10.300	$\pm 0.276$	73.17	$\pm 2.013$	10.76	$\pm 0.986$
13	<i>Petunia alba</i> Lindl.	Non glandular	111.96	$\pm 9.969$	10.500	$\pm 0.260$	103.76	$\pm 3.828$	9.90	$\pm 0.428$
14	<i>Petunia hybrida</i> L.	Glandular	134.60	$\pm 5.967$	11.167	$\pm 0.355$	71.33	$\pm 14.435$	10.10	$\pm 0.374$
15	<i>Withania somnifera</i> (L.) Dunal	Non glandular	83.06	$\pm 1.549$	10.167	$\pm 0.886$	50.36	$\pm 3.891$	7.77	$\pm 0.370$
16	<i>Cestrum nocturnum</i> L.	Glandular	114.96	$\pm 8.233$	6.86	$\pm 0.423$	53.44	$\pm 3.219$	7.03	$\pm 0.540$
17	<i>Datura Stramonium</i> L.	Star shaped	126.20	$\pm 2.391$	11.367	$\pm 0.451$	106.83	$\pm 3.246$	10.43	$\pm 0.476$
18	<i>Datura innoxia</i> Miller.	Non glandular	148.46	$\pm 13.840$	17.033	$\pm 1.218$	160.76	$\pm 7.200$	15.50	$\pm 0.492$
19	<i>Atropa acuminata</i> Royale.	Glandular	108.43	$\pm 4.675$	14.300	$\pm 0.694$	95.36	$\pm 6.029$	10.43	$\pm 0.262$
20	<i>Physalis minima</i> Auctt.	Non glandular	76.30	$\pm 5.867$	10.433	$\pm 0.220$	76.46	$\pm 3.267$	9.33	$\pm 0.231$
21	<i>Nicotiana tabacum</i> L.	Non glandular	75.94	$\pm 3.435$	9.33	$\pm 0.423$	55.47	$\pm 2.191$	8.03	$\pm 0.593$
22	<i>Lycopersicon esculentus</i> Miller.	Non Glandular	128.63	$\pm 2.050$	17.167	$\pm 1.220$	99.78	$\pm 2.717$	13.26	$\pm 0.654$

*S. americanum**S. villosum**S. surratense**S. cardatum**S. Pseudocapsicum**S. Melongena**S. Tuberosum**S. Erianthum**S. Dulcamara**H. niger**C. annum**C. Frutescense**P. alba**P. hybrid**W. somnifera**C. nocturnum**D. stramonium**D. innoxia*

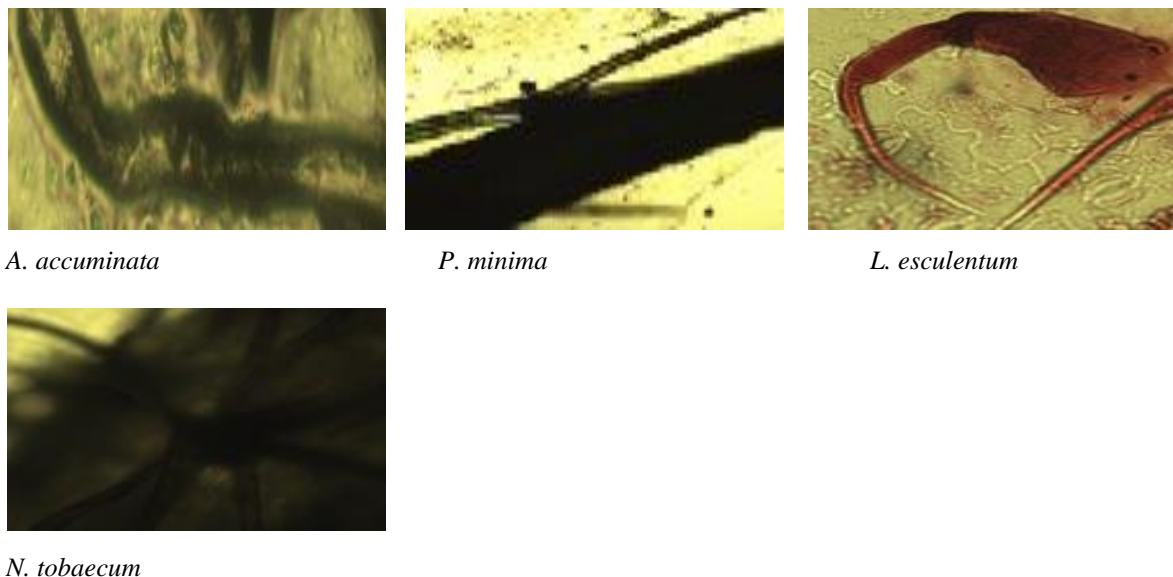


Fig. 2. Micrographs of trichomes of 22 species of family Solanaceae.

The Micro anatomical characteristics of leaf and stem epidermis and distribution of trichomes on the abaxial and adaxial surfaces were contributed in the classification of Asteraceae, Apocyanaceae, Brassicaceae, Boraginaceae, Euphorceae, Fagaceae, Malvaceae, Menispermaceae, Polygonaceae, Rosaceae and Solanaceae Baranova (1972) Bhatia (1984) and Dias *et al.* (2013). Different anatomical characters which indicate the close relationship among the species and genera include size, shape, type and number of trichome on the amphistomatic leaf surfaces. 16 species shows non-glandular type of trichome viz; *Solanum nigrum*, *Solanum surratense*, *Solanum melongena*, *Solanum villosum*, *Solanum erianthum*, *Hyoscyamus niger*, *Capsicum annum*, *Petunia alba*, *Petunia hybrida* and *Solanum tuberosum*, *Cestrum nocturnum*, *Atropa accuminata*, *Physalis minima* *Nicotiana tabaecum*, *Withania somnifera*, *Solanum pseudo capsicum* (Table 1) while 6 species consist of glandular type of trichome. Viz; *Lycopersicon esculentum*, *Solanum cordatum*, *Datura innoxia*, *Datura stramonium*, *Solanum dulcamara* and *Capsicum frutescense*. 8 species shows tricellular, 9 species shows multicellular, 3 species shows bicellular and 2 species shows unicellular trichome on both sides of leaf epidermis. Maximum mean length and width size of trichome is recorded in *Datura innoxia* on the abaxial and adaxial side of leaf measured as  $148.46 \mu\text{m} \pm 13.84\mu$ ,  $17.03\mu\text{m} \pm 1.21\mu$ ,  $160.76\mu\text{m} \pm 7.20 \mu\text{m}$  and  $15.50 \mu\text{m} \pm 0.49 \mu\text{m}$  respectively ( Table- 1). While minimum length and width were noted in *Capsicum frutescense*, is measured as  $74.73 \mu\text{m} \pm 7.62 \mu\text{m}$ ,  $10.30 \mu\text{m} \pm 0.27 \mu\text{m}$ ,  $73.17 \mu\text{m} \pm 2.01 \mu\text{m}$  and  $10.76 \mu\text{m} \pm 0.98 \mu\text{m}$  On the abaxial and adaxial surfaces of leaf , rest of the species shows lowest values (Table 1; Fig. 2) Xiang (2010).

Unicellular trichomes were observed in *Solanum surratense* and *Physalis minima*. (Fig 16). The average length and width size of trichome was examined in *Solanum surratense* is (  $122.76 \mu\text{m} \pm 12.19 \mu\text{m}$  ,  $9.96 \mu\text{m} \pm 1.10 \mu\text{m}$  ) on the abaxial surface while ( $58.79 \mu\text{m} \pm 0.48 \mu\text{m}$  and  $6.53 \mu\text{m} \pm 0.40 \mu\text{m}$ ). Bicellular trichomes were observed in *Solanum nigrum*, *Solanum cordatum* and *Withania somnifera*. (Fig. 2). The values of the trichomes were given in the (Table 1). Tricellular trichome were studied in the species of *Solanum erianthum* ( $91.36 \mu\text{m} \pm 1.63 \mu\text{m}$  ) *Solanum dulcamara* ( $115.08 \mu\text{m} \pm 0.57 \mu\text{m}$ ), *Solanum tuberosum* ( $137.33 \mu\text{m} \pm 11.21\mu\text{m}$ ), *Cestrum nocturnum* ( $114.96 \mu\text{m} \pm 8.23 \mu\text{m}$ ), *Hyoscyamus niger* (  $57.73 \mu\text{m} \pm 1.03 \mu\text{m}$ ), *Nicotiana tabacum* ( $75.94 \mu\text{m} \pm 3.43 \mu\text{m}$  ) , *Solanum pseudo-capsicum* ( $129.33 \mu\text{m} \pm 1.42 \mu\text{m}$ ) and *Lycopersicon esculentum* ( $128.63 \mu\text{m} \pm 2.05 \mu\text{m}$ ). Whereas, rest of the species viz; *Solanum villosum*, *Solanum melongena*, *Capsicum annum*, *Capsicum frutescense*, *Petunia alba*, *Petunia hybrida*, *Datura innoxia*, *Datura stramonium* and *Atropa accuminata* (Fig. 2) respectively. Star shaped trichome were observed in three species viz; *Petunia alba*, *Petunia hybrida*, and *Datura innoxia* (Fig.2). The average length and width size was calculated as  $111.96 \mu\text{m} \pm 134.60 \mu\text{m} \pm 8.23 \mu\text{m} \pm 134.60 \mu\text{m} \pm 5.96 \mu\text{m} \pm 148.46 \mu\text{m} \pm 13.84 \mu\text{m}$  respectively. Non-glandular trichomes were observed in eighteen species Croteau, and Johansan (1984) (Fig. 2) while glandular trichomes were observed in four species out of twenty-two species of Solanaceae.

Unicellular, Bicellular, Tricellular and Multicellular with sessile and glandular trichomes were previously reported by Metcalf and Chalk (1950); Walas (1959); Inamdar and Choan (1969); Ramaya and Rao (1976); Adedeji and Dloh (2004); Celka *et al.* (2006); Xiang *et al.* (2010) and Osman (2012) in the families of Asteraceae, Boraginaceae, Fagaceae, Lamiaceae, Malvaceae, Solanaceae and Verbenaceae.

*Petunia alba* and *Petunia hybrida* both have stellate trichome which were triradiate to multiradiate, stalked and non-glandular. In *Cestrum nocturnum* and *Physalis minima* and *Hyoscyanum niger* non-glandular stalked type trichome are found on the abaxial and adaxial surface of leaf of epidermis which are multi-cellular. In case of *Atropa accuminata* non-glandular trichome are present on both surfaces of leaf which are multi-cellular type. Ahmad (1964b).

During this study very valuable variation in the configuration of foliar epidermal anatomy was explored that can be used as important taxonomic tool and differentiation of species. Anatomical studies revealed clear cut differences in size, shapes of epidermal cells and trichome.

### Conclusion

The differences were examined in the abaxial and adaxial surfaces of the species of Solanaceae. Most of the taxonomic and anatomic features are stable in most species. It is possible that each species respond to its surrounding environment in specific way by modifying the certain characters to improve its adaptation. Statistical analysis also supports the existing classification.

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