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Diversity of Trichomes in *Calliandra haematocephala* Hassk. (Caesalpinioideae DC., Fabaceae Lindl.)

Sangeeta Joshi [a*], Deepa Dhami [a] and D. S. Rawat [a]

[a] Department of Biological Sciences, College of Basic Sciences and Humanities, G.B. Pant University of Agriculture and Technology, Pantnagar-263145, Udham Singh Nagar, Uttarakhand, India.

Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

Objective: High structural diversity of trichomes in the members of family Fabaceae Lindl. has been a subject of study from decades. The present study was aimed to study structural diversity of trichomes in Power Puff Tree (*Calliandra haematocephala,*Caesalpinioideae, Fabaceae) a pan-tropical ornamental tree.
Methods: Plant specimens were collected from the campus of Govind Ballabh Pant University of Agriculture and Technology Pantnagar (India) during the year 2020. Plant species was identified with the help of relevant Floras. Fresh plant material was used for vestiture and trichome study under light microscope using standard anatomical procedures. Illustrations of trichomes were drawn by using prism type camera lucida and photographs were taken at different magnifications. Standard terminology of trichome types given by payne 1978 was adopted to describe different structural types of trichomes.

**Corresponding author: E-mail: sangeetajoshi680@gmail.com;*

Results: Nine different types of glandular and non-glandular trichomes were recorded in *Calliandra haematocephala* Hassk. Four different vestiture types were recorded on the surfaces of studied parts. Trichomes were found present in all vegetative parts and some reproductive parts *viz*; peduncle, bracts, bracteole, petals while other reproductive parts like sepals, filaments, anther, ovary, style and stigma were found completely glabrous. Present study suggests that trichome morphology and distribution on different plant parts can be used to identify plant species. *Calliandra haematocephala* Hassk. can be identified in its vegetative phase of life when floral parts are lacking or fragmentary.

Keywords: Calliandra; glandular; nonglandular; trichomes; vestiture.

1. INTRODUCTION

Trichome are a kind of epidermal outgrowth or appendages of a plant surface which are of diverse forms, structures and functions [1] and have been recorded in large number of legume taxa. Trichomes may occur on all parts of a plant, persisting either throughout the lifespan of an organ, or as ephemeral. Different trichome types have been successfully used in the classification of genera and species in certain families and in the recognition of interspecific hybrids [2-4]. Trichomes are micromorphological characters which can be used to identify plant species when the plant lacks its floral structures [5,6]. *Calliandra haematocephala* Hassk. is a member of genus *Calliandra* belonging to subfamily Caesalpinioideae DC. of the family Fabaceae. *Calliandra haematocephala*is a 1-3 m high evergreen, shrub, with spreading and pendulous branches, forming a dense round head. Leaves are alternate, stipulate, petiolate, compound and bipinnate. Flowers are sessile and arranged in small, dense flower heads; watermelon pink with numerous silky stamens, fruit is a compressed legume [7]. (Fig. 1). Various pharmacological properties of leaves have been reported in *Calliandra haematocephala* such as analgesic, anticonvulsant, antipyretic, anti-ulcer and antioxidant [8,9]. Leaf extracts of *Calliandra haematocephala* have antiviral activity against RV infection *in-vitro* [9]. Leaves of *Calliandra haematocephala* have been found to be a novel source of the synthesis of zinc oxide nanoparticles and to detect the presence of hydrogen peroxide in various samples [10].

Although several macromorphological and pharmacological studies have been conducted on *Calliandra haematocephala,* detailed information of micromorphological characters like surface indumentum and trichome morphology is not well investigated. Present study was conducted to explore the structural diversity and distribution of trichomes, and vestiture types on the surfaces of all vegetative as well as reproductive parts to fill the void in information regarding micromorphology of *Calliandra haematocephala*.

Fig. 1. *Calliandra haematocephala* Hassk

2. MATERIALS AND METHODS

Plant specimens were collected from campus of Govind Ballabh Pant University of Agriculture and Technology Pantnagar, India. Processing of plant specimens was done following standard taxonomic procedures [11]. Plant species were identified using relevant Flora [12] and herbarium consultations at the herbaria of Botanical Survey of India, Northern Regional Center, Dehradun (BSD) and Forest Research Institute, Dehradun (DD). Voucher specimens were deposited in the herbarium of Department of Biological Sciences, Govind Ballabh Pant University of Agriculture and Technology Pantnagar, Uttarakhand, India. Fresh plant materials were used for vestiture and trichome study. Epidermal surfaces of each and every part were gently peeled off using razor blade and scraping was done wherever needed following scraping method as documented by earlier workers [13]. Peels were washed carefully and the cleared epidermal peels were preserved in 50% ethanol, stained in Safranin O and counter-stained in Alcian blue to enhance contrast. All the preparations were mounted in 25% glycerol, covered and sealed with transparent nail polish to prevent dehydration. Both adaxial and abaxial surfaces of the leaves, sepal and petals were studied. Prepared slides were examined under the light microscope in 4x, 10x, 40x, 100x objective lens and photomicrographs were obtained using Olympus MLX-B Plus microscope equipped with 1080 x 1920 megapixels cell phone camera. Exact illustrations of trichome morphology were drawn by using prism type camera lucida at 10x magnification of objective lens. All microscopic measurements were taken with the aid of an ocular micrometer inserted in the eyepiece of the microscope. These measurements were later multiplied by ocular constant with respect to the power under which they were taken. For the description of vestiture, arrangement, position of trichomes on plant surfaces, shape and size standard taxonomic terminology given by Beentje [14] and Payne [15] with slight modifications was used.

3. RESULTS AND DISCUSSION

The study of different surfaces of vegetative and reproductive parts of *Calliandra hematocephala* reveals presence of following nine types of trichomes:

Type-I) Glandular, multicellular, biseriate, colleter (MCO). These trichomes are composed of a multicellular, uniseriate stalk and two cell thick obovate head, 75-90 µm long, porrect, straight and recorded on peduncle and margin of petal (Fig. 2.G1, G2, G3; Fig. 3.A).

Type-II) Glandular, multicellular, uniseriate, brevicollate trichomes (MBR), composed of a short, unicellular stalk with multicellular secretory head, 45-60 µm long, appressed, straight and recorded only on peduncle (Fig. 2. J1, J2; Fig. 3. B).

Type-III) Non-glandular, unicellular, uniseriate, attenuate trichomes (UAT) composed of single cell with long, gradual tapering distal end, 75-600 µm long, straight and recorded on stem, leaflet margin (porrect), stipule, petiole, petiolule, rachis, abaxial surface of leaflet (spreading), adaxial surface of leaflet and bracteole (appressed) (Fig. 2. A1, A2; Fig. 3.C).

Type-IV) Non-glandular, unicellular, uniseriate, simple-acuminate (UAU) trichomes composed of single cell with thick walls and slender and sharp apex, 120-225 µm long, straight and recorded on abaxial surface of leaflet (porrect), bracts and peduncle (oriented in different directions) (Fig. 2.B1,B2; Fig. 3. E).

Type-V) Non-glandular, unicellular, uniseriate, simple-muticose (UMU) trichomes composed of a small, single cell having a blunt tip, 150-420 µm long, appressed and recorded only on peduncle (Fig. 2. H1).

Type-VI) Non-glandular, multicellular, uniseriate, simple-subulate trichomes (MSU) composed of three or more cells tapering to sharp tip, 75-420 µm long, straight and recorded on peduncle (appressed) and abaxial surface and margin of petal (porrect) (Fig. 2. C1,C2; Fig. 3.D).

Type-VII) Non-glandular, multicellular, uniseriate, acuminate with cushion trichomes (MCA) composed of a multicellular, multiseriate cushion of epidermal cells bearing long, slender cell with sharp apex, 480-600 µm long, arcuate, oriented in different directions and recorded only on peduncle (Fig. 2.D1, D2; Fig. 3.F).

Type-VIII) Non-glandular, unicellular, uniseriate, stalked-muticose trichomes (USM) composed of thick-walled single cell with a short basal stalk, oblong body parallel to surface and a blunt tip at apex, 150-270 µm long, straight, appressed and recorded on peduncle (Fig. 2. F1; Fig. 3.G).

Type-IX) Non-glandular, unicellular, uniseriate, stalked-acuminate trichomes (USA) composed of thick-walled single cell with a short basal stalk, long body parallel to the surface and a slender and sharp point at distal end, 180-270 µm long, tortuous, ascending and recorded on peduncle and bracts (Fig. 2. E1, E2; Fig. 3.H).

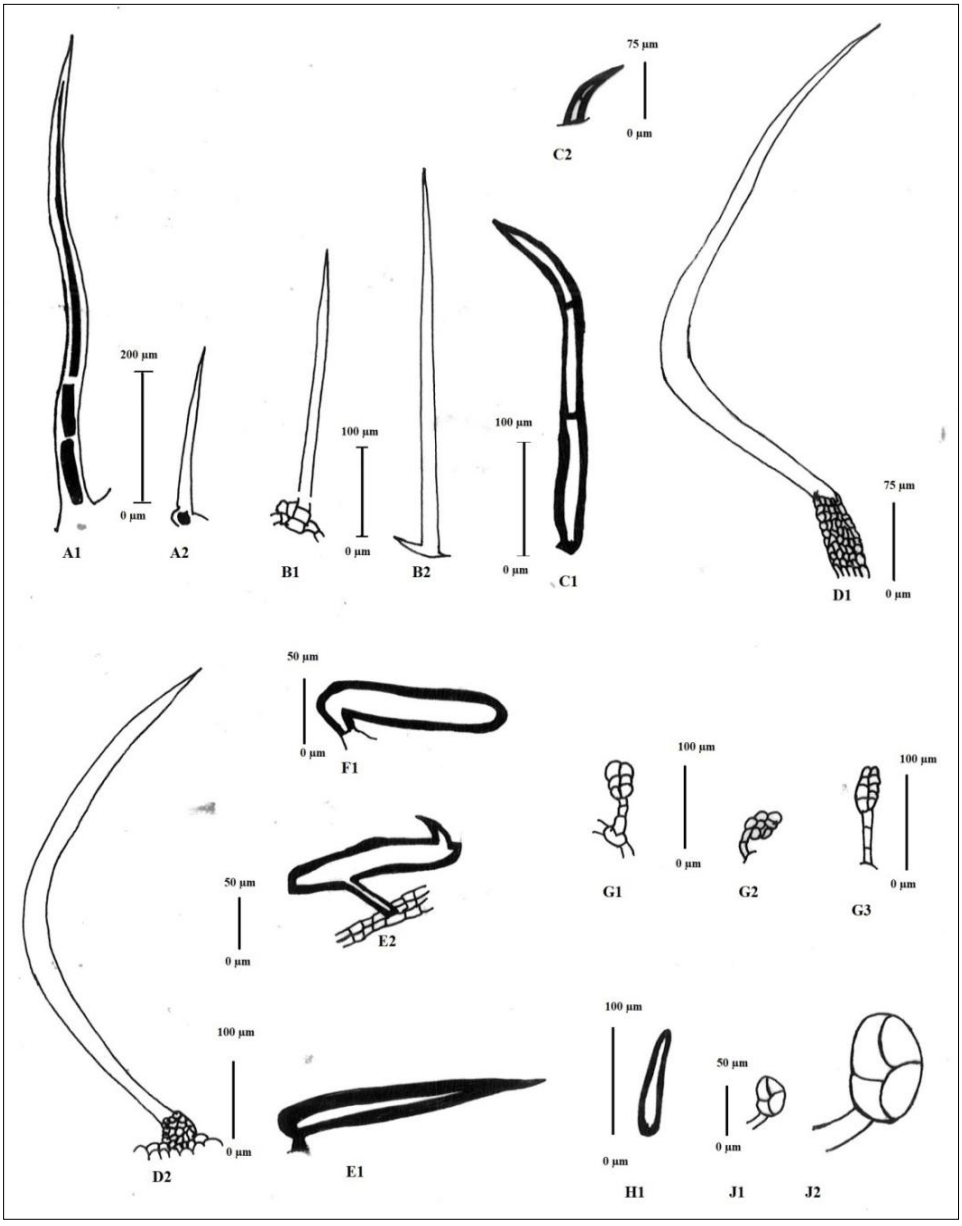

Fig. 2. Structural diversity of trichomes in *Calliandra haematocephala*. A1, A2: Non-glandular, unicellular, uniseriate, attenuate; B1, B2: Non-glandular, unicellular, uniseriate, simple-acuminate; C1, C2: Non-glandular, multicelluar, uniseriate simple-subulate; D1, D2: Non-glandular, multicellular, uniseriate, acuminate with cushion; E1, E2: Non-glandular, unicellular, uniseriate, stalked, acuminate; F1: Non-glandular, unicellular, uniseriate, stalked, muticose; G1, G2, G3: Glandular, multicellular, biseriate, colleters; H1: Non-glandular, unicellular, uniseriate, simple-muticose; J1, J2: Glandular, multicellular, uniseriate, brevicollate

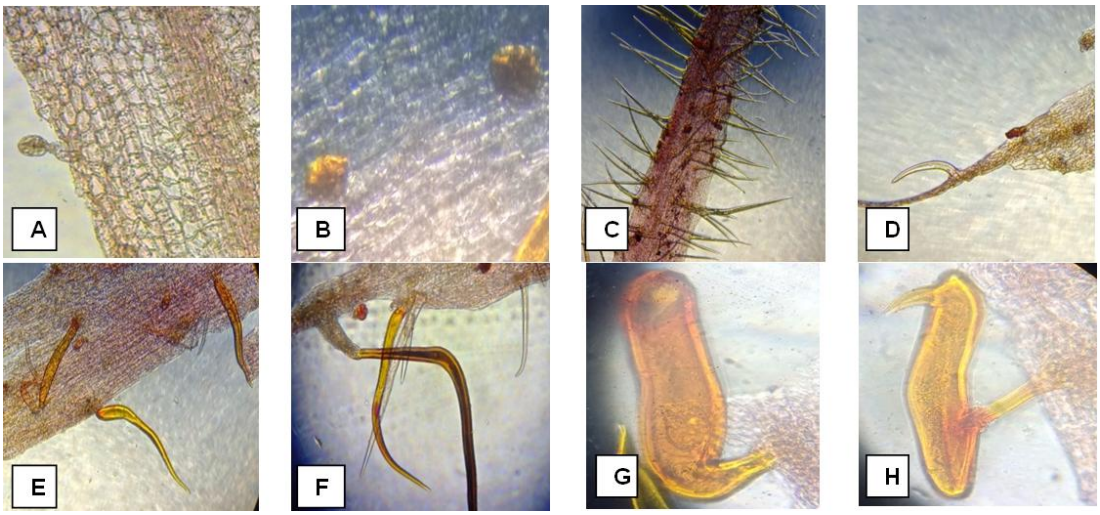

Fig. 3. Photomicrographs depicting structural diversity of trichomes. A: Glandular, multicellular, biseriate, colleters; B: Glandular, multicellular, uniseriate, brevicollate; C: Non-glandular, unicellular, uniseriate, attenuate; D: Non-glandular, multicelluar, uniseriate, simple-subulate; E: Non-glandular, unicellular, uniseriate, simple-acuminate; F: Non-glandular, multicellular, uniseriate, acuminate with cushion; G:- Non-glandular, unicellular, uniseriate, stalked- muticose; H- Non-glandular, unicellular, uniseriate, stalked-acuminate

Table 1. Vestiture type and types of trichomes on different studied parts

Plant part	Vestiture type	Types of trichomes
Stem	Puberulent	Type-III
Stipule	Puberulent	Type-III
Petiole	Puberulent	Type-III
Petiolule	Puberulent	Type-III
Leaflet (Abaxial surface)	Puberulent	Type-III, IV
Leaflet (Adaxial surface)	Puberulent	Type-III
Leaflet (Margin)	Evenly distributed	Type-III
Rachis	Lanate	Type-III
Peduncle	Strigose	Type-I, II, IV,V,VI, VII, VIII, IX
Bracts	Puberulent	Type-III, IV, IX
Bracteole	Strigose	Type-III
Sepal (Abaxial surface)	Glabrous	Absent
Petal (Abaxial surface and margin)	Puberulous & Puberulent	Type-I,VI
Filaments	Glabrous	Absent
Anther	Glabrous	Abesent
Ovary	Glabrous	Absent
Style	Glabrous	Absent
Stigma	Glabrous	Absent

The trichomes are broadly of two types-glandular and non-glandular. Non-glandular trichomes are more diversified with seven types (type-III, IV, V, VI, VII, VIII, IX) while glandular trichomes are represented by two types (type-1, II). These glandular trichomes were found restricted to peduncle and petal margin in distribution but non-glandular trichomes were more common and recorded on all the trichomatous surfaces.

Present study recorded external surfaces of all vegetative parts and some reproductive part of *Calliandra haematocephala* bearing one or more different types of glandular and non-glandular trichomes (Figs. 2 & 3). Among the studied external surfaces abaxial surface of sepals, filaments, anthers, ovary, style and stigma are completely glabrous while rest of the external surfaces (stem, stipule, petiole, petiolule, rachis, peduncle, adaxial and abaxial surfaces of

leaflets, leaflet margin, bract, bracteole, abaxial surface and margin of petals bear one or other types of trichomes. Table 1 summarizes types of trichomes present on different external surfaces. It is evident that peduncle surface bear maximum structural diversity of trichomes represented by eight types which include both glandular and non-glandular trichomes. Bract surfaces bear three types of trichomes, petal and abaxial surface bear two types oftrichomes while rest of the trichomatous surfaces bear only one type of trichomes. The type-III trichomes (Non-glandular, unicellular, uniseriate, attenuate trichomes, UAT) are the most common and widespread trichomes in this species being present on all the surfaces barring petals and peduncle.

Six different types of vestitures were recorded in this species. Visibility of different vestitures on studied external surfaces is depicted in Table 1. Puberulent type of vestiture is most common type present on eight surfaces and, interestingly, type-III trichomes mainly contribute to form this type of vestiture. Peduncle surface which bears eight different types of trichomes shows strigose vestiture.

Epidermal structures of *Calliandra hematocephala* cultivated in Egypt have also been studied earlier [7] and non-glandular, unicellular trichomes and stalked glandular trichomes on stem and leaves were reported. The present study, however, could not confirm the presence of glandular trichomes on leaves and petioles as these were recorded on only peduncle and petal margins. The complete structural diversity of trichomes (nine types) as observed in the present work was not reported earlier [7]. Leaf surfaces of 30 Fabaceae species growing in Pakistan which included *Calliandra bella* were studied [16] but the authors realized very little variations in trichomes reporting generally non-glandular trichomes which are multicellular, uniseriate, unbranched, with bulbous base and pointed tips.

4. CONCLUSION

The present study demonstrates that members of the family Fabaceae may eventually reveal a higher diversity of trichome structures if all of the external surfaces are thoroughly studied. Wide variety of trichomes which may include novel structural variations may be recorded in the future and this information will be useful in understanding taxonomic and evolutionary relationships among the taxa of this large family.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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